Conference Proceedings

The XIIth International Silage Conference

Silage Production in relation to animal performance, animal health, meat and milk quality

July 5-7, 1999
Uppsala Sweden

SWEDISH UNIVERSITY OF AGRICULTURAL SCIENCES
SLU

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Contents

Welcome

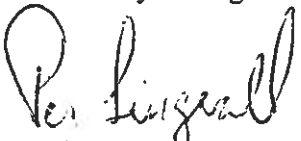
Welcome to Sweden and to the Swedish University of Agricultural Sciences in Uppsala. 220 delagates from all over the world, including a number of leading scientists presenting papers, posters and prepared to run six work shops are attending the

XIITH INTERNATIONAL SILAGE CONFERENCE.

Swedish silage scientests have been invited to attend many of which have also participated in 10 of the 11 previous Silage Conferences held at 2 to 3-yearly intervals from 1972 to 1993. When we were asked to arrange the XIIth conference we were delighted and felt very honoured, but also felt a little anxious over being responsible for this old, established institution.

A small country such as Sweden has limited resources for research work and has to rely on collaboration and support. Our contact with institutes and scientific colleages all over the world is necassary to follow such a broad topic as **Silage production and Utilization**. The friendly attitudes and great support we have had during the preparation for these days at our University both from scietific colleagues and many international companies, has encourage us and finally made it possible to complete all the arrangements. Our thanks to all of you.

Today the members of the community are more and more interested in food production to secure their own welfare, but they also want to know more about how we feed and treat our animals. Silage is a very important part of the feed ration which can influence both the health of the animals and the food beeing produced. Looking at the topics with high listed in EU`s Fifth Framework everyone must realize that we have to broaden the silage topics that we study and also involve scientests from other related research areas. That is why we chose the theme **Silage Production in relation to Animal performance, Animal health and meat and milk quality.** Jackie Merry gave us a clever **LOGO** in Wales – we kept it. Thank You Jackie. Roger Wilkins gave us some good advice when planning this conference and trying to activate everyone concerned. The final outcome of the conference will be dependent upon the efforts of every delegate. We look forward to seeing you all, both during in the presentations and discussions and informally during social events. **WELCOME.**



Per Lingvall, on behalf of the orginising committee.

Conference organisers

Organising Committee

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Prof. Sven Lindgren, National Food Administration, Uppsala

Dr Michael Murphy, Dept of Animal Nutrition and Management, SLU, Uppsala

Prof. Hans Wiktorsson, Dept of Animal Nutrition and Management,SLU, Uppsala

Conference Secretariat

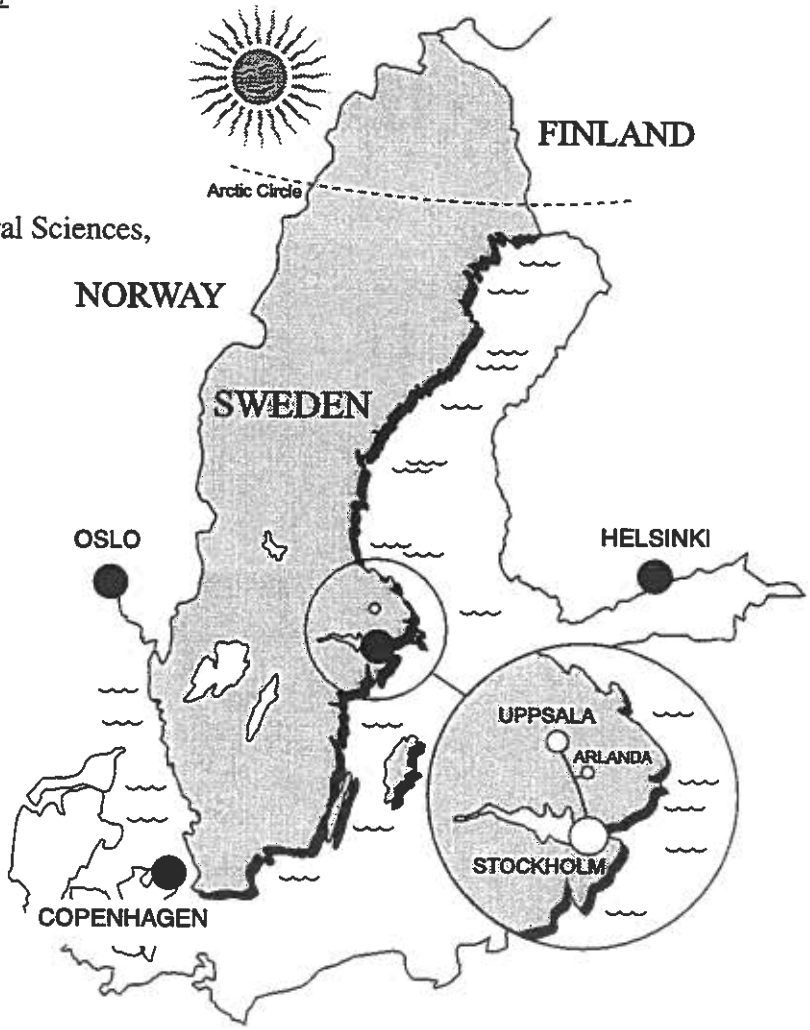
SLU Conference Service, Uppsala

Conference web site

(information available about one year after the conference):
www.service.slu.se/conference/silage/

Conference Venue

The Ultuna campus of SLU,
the Swedish University of Agricultural Sciences,
Uppsala, Sweden



The conference organisers wish to thank organisations presented on the following pages for the generous contributions and financial support

Medipharm

The Municipality of Uppsala

The Swedish Council for Forestry and Agricultural Research (SJFR)

SJFR is an organisation under the Swedish Ministry of Agriculture and was founded in1945.

The Council encourages and supports scientifically significant research in forestry, agriculture, veterinary medicine, food, fish, horticulture and in reindeer husbandry.

The research is of importance for involved industries and for those concerned with environmental issues related to forestry, agriculture and fisheries. The research aims to lay the fundament for production of nutritious, safe and healthy food and to support animal health and welfare. Furthermore, the research aims to support the creation of functional and beautiful parks and gardens in our cities.

SJFR is mainly working with projects but is also financing salaries for scientists, travels, conferences, equipment etc. Every year the council receives approximately 600 applications that are reviewed by scientific committees. The committees for *Plants, Animals, Biogeochemistry, Ecology* and *Economics & Society* consist of researchers who assess research projects´ scientific merits. The committees for *Forestry, Agriculture, Horticulture, Fish, Food and Reindeer husbandry* consist of researchers and representatives who judge both the projects´ scientific merits and their significance for their respective industries.

For more information phone (+46) 8 5454 1260, E-mail sjfr@sjfr.se or our website www.sjfr.se

Svalöf Weibull AB

Svalöf Weibull (SW) is one of the largest plant breeding and seed groups in Europe. SW markets agricultural seed and services in some forty countries and has subsidiaries, associated companies and plant b reeding stations in fifteen countries.

Annual sales are about 110 million Euro of which more than 15 million Euro is reinvested in plant breeding. In addition, SW holds 15% of the shares in BASF Plant Science (BPS), where the investment in plant biotechnology R&D will be 500 million Euro over the next 10 years.

SW is to 60% owned by the Swedish Farmers Supply and Crop Marketing Association (SLR) and to 40% by BASF Aktiengesellschaft.

Svalöf Weibull is breeding most of the crops grown in Sweden and temperate zone of the world We are breeding almost all the fodder crops grown in Sweden. Important breeding goals for fodder crops as grasses and legumes: winter hardiness for very cold conditions, adapted to long day conditions, faster regrowth of the crop, adapted to two or three (or four) cuts a year.

Svalöf Weibull is committed to continuous research and development programs to improve the productivity of our hay and silage seed mixtures. We currently market 33 different seed mixtures for hay and pasture in Sweden, suitable for all different micro-environmental conditions. Through our research and development we contribute to and support Sweden's leading position in milk production in Europe.

A short presentation of the Academy

The task of the Royal Swedish Academy of Agriculture and Forestry is to promote agriculture and forestry and their related fields with the support of science and practical experience and in the interest of society.The Crown Prince in 1811, later King Carl XIV Johan, initiated the foundation of an Agricultural Academy in Sweden. He was probably thinking along the lines of the French Academy of Agriculture.On 28 December 1811, the Royal Swedish Academy of Agriculture was founded by Royal Decree. The Crown Prince became the highest representative of the Academy. Many contempo-rary persons of importance became members.In the late 1940ís the idea arose of widening the role of forestry within the Academy. This was introduced in 1956, when the Academy was reorganised and the name was changed to the Royal Swedish Academy of Agriculture and Forestry (KSLA).Over the years the Academy has received several donations, donated to provide funding of research grants, scholarships for study trips abroad and awards that are presented to deserving representatives of forestry and agriculture, both in the practical field as well as in the theoretical.Today the Royal Swedish Academy of Agriculture and Forestry consists of three sections - a General, an Agricultural and a Forestry Section. The activities are led by the Academy's boards, elected standing committees etc. through assemblies, conferences and seminars.

Alfa Laval Agri

Alfa Laval Agri is a full service supplier to dairy farmers. The company markets equipment and complete systems for milk production and animal husbandry. Service and sales of a wide range of accessories are also key aspects of the operations. In roughage feeding Alfa Laval Agri offers mixer wagons and rail suspended feed wagons as well as silage additives.

The Alfa Laval Agri worldwide distribution network, totals 55 market companies, 2 500 dealers, 2 800 mobile shops and 1 600 service vans, serving about one million customers.

Nordiska djurskyddsföreningen i Uppsala

The Nordic Animal Protection Society in Uppsala works within the County of Uppsala. The society mainly works for information and education. During the last years it has started education of children from 6 to 19 years with different material and leaders between ages. At the Swedish Univ of Agrc. Sciences it promotes studies in relation to animal welfare and animal behaivor, specific for pet animals by sponsering scientific papers and stimulating students to arrange seminars for the public. It also produces small information leaflets and arranges open days in town centres.

Kverneland

Kverneland is one of the world's leading manufactures and distributors of agricultural implements for small and large farming operations, as wells as big farms. Founded more than 100 years ago, Kverneland offers the most extensive and complete product range in the market.

The Kverneland group of companies embraces fourteen production companies, four in Germany, three in France, two in Holland, two in Norway and one each in Denmark, Italy and the U.K. Products are sold through Kverneland's own sales companies in fifteen countries and through local importers in all other markets. The Kverneland group manufactures a complete range of machines comprising three main product lines: Arables, grass and potatoes. Kverneland has 3.800 employers and a turnover of 4 billions NOK.

Efficient processes advanced control systems and raw materials of the right quality are prerequisites for maintaining our position in the worlds market. Kverneland made an early commitment to quality in term, of both functionality and durability.

Today, it all comes down to higher yields, lower operating costs and co-existence with nature. Our goal is to solve the farming problems not only of today but also of the future.

Ab Hanson & Möhring

Ab Hanson & Möhring is an independent privately owned trading company. It is a member of the Salinity Group and was founded already in 1830. Ever since those early days the company has been trading with salt. Together with sales and trading of other bulkproducts this is still the principal business at Hanson & Möhring. Over the years other business areas have developed such as fertilizers, charcoal and chemicals.

Ab Hanson & Möhring has a special department for agricultural products. The Agro department manufactures and markets saltlicks for animals (cattle, horses and wild ruminants). The SP Salt Lick is very well known and is currently No. 1 on the Swedish market. Smaller quantities are exported to Denmark, Germany, France and Japan.
The Agrodepartment had been working with silage additives for some 30 years and has a solid knowhow regarding this area of farming. Development and research within this specialized niche is done in close collaboration with the Swedish University of Agricultural Sciences. The latest result of this cooperation is a new and very effective silage additive called **Kofasil ULTRA®** well on its way of taking a leading position on the Swedish market.

TRIOPLAST

Trioplast is one of Europe's leading manufacturers of high quality stretch film for silage wrapping and wide polythene film for bunker silos. Our well-known stretch films with brand names such as Triowrap®, Agrispinflex® and Tenospin®, has been developed together with the Swedish University of Agricultural Sciences to meat the demands of our customers.

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World Society for the Protection of Animals (WSPA)

Svenska Djurskyddsföreningen – The Swedish Animal Protection Society

is working with information and aducation about the life of wild and domestic animals to protect them against abnormal treatment, stress and bad utilization. The society also finances research projects in relation to the welfare of domestic and wild animals. It supports advice to young people and educates leaders of this activities. The society works over all Sweden.

svenskmjölk
SWEDISH DAIRY ASSOCIATION

The Swedish Dairy Association is the joint organisation for Swedish dairy farmers and the Swedish dairy industry, combining all available knowledge in these fields. The organisation is owned by somewhat more than 13,600 dairy farmers through their eight dairy co-operatives, eleven livestock co-operatives, two semen prodcing companies, and nine breed societies.

The Swedish Dairy Association can be described as a long chain of knowledge from the consumer, all the way to the cow. By collection, analysis, and dissemination of knowledge within this chain, we can efficiently look after and create a public opinion in favour of the spheres of interest of dairy farmers and the dairy industry. An important task of the Swedish Dairy Association is to strengthen the belief in the future in the mind of our principals.

Which are the key development areas? Based on the combined chain of knowledge, it is essential to focus on influencing *public opinion and dairy policy*; to gather knowledge available today and to carry on *research* for the knowledge of tomorrow; to vigorously promote the competitiveness and *profitability* of Swedish dairy production; to collect and custom-make *advisory* services requested by the dairy producers; and, as *healthy animals* make the foundation of the whole process, to co-ordinate all knowledge within the field of veterinary medicine.

TRIMA

Trima AB based in Bergsjö, Sweden design, manufacture and market front loaders with comprehensive range of implements and accesories for all tractor makes. The principal market is the agricultural sector.
Trima´s objective is to translate ideas into practical solutions in which the driver, the tractor and the implement work as a unit, During the last decade Trima has established itself as a world leader in the frontloader sector. Export sales account for over 80 % of production and are managed directly from Sweden and wholly-owned subsidiaries.

Neste Oxo AB, a part of the Finnish group Neste Chemicals, serves the industry with important intermediates such as, aldehydes, alcohols and acids. Manufacturing facilities are located in Sweden and Belgium, where plants utilise the latest technology to meet high demands for product quality and environmental safety.

Neste Oxo has, thanks to its strong raw material integration in aldehydes, an important role to play in this interesting market.

Late 1996 Neste Oxo inaugurated a new multipurpose acid plant in Stenungsund, Sweden. 2-Ethylhexanoic acid and Propionic acid are produced here in a world scale site.

Neste Oxo AB employee some 300 people, turnover is 1.3 billion SEK. The production facilities in Sweden are certified to ISO 14001 standard.

Neste Oxo AB, SE 444 84 Stenungsund, Sweden. Phone + 46 303 728 600

Final Programme

Monday 5.7 1999

07.50	Bus departure, see time table	
08.00	Registration and set-up posters. Refreshments	
09.00	Welcome - Introduction	*T. Rosswall*, the rector of SLU
SESSION 1.	**Chairman P.G. Knutsson, Sweden**	**Lecture hall = A (Aula)**
09.30 - 10.30	The future role of silage in sustainable animal production *R. Wilkins*, UK	
10.30 - 11.00	Coffee /tea	
11.00 - 11.45	Relationships between silage based diets and feed conversion *E. Burstedt - M. Murphy*, Sweden	
11.45 - 12.30	Can HACCP principles be applied for silage safety? *S. Lindgren*, Sweden	
12.30 - 13.30	Lunch	
SESSION 2.	**Chairman M.K. Woolford, UK**	
13.30 - 16.30	Poster session I. incl. 60 minutes discussion (coffee /tea available 15.00-15.45) Room D,E, F and G. Discussion in room A.	
SESSION 3.	**Chairman B. Everitt, Sweden**	**A**
16.30 - 17.15	Silage and health *J.M. Wilkinson*, UK	
17.15 - 18.00	The relationship between silage quality and food quality *S. Sivelä*, Finland	
18.30 - 21.30	Welcome Barbeque at Ultuna	

Tuesday 6.7 1999

07.50	Bus departure, see time table
SESSION 4.	**Chairman M. Mo, Norway**
08.30 - 11.00	Poster session II. incl. 60 minutes discussion (coffee /tea available 9.30-10.00) Posters in relation to workshops B, C and D. Rooms D, E, F, G. Discussion room A
SESSION 5.	
11.00 - 15.00	(lunch 12.00 - 13.00)

Workshops (coffee/tea available 14.30-15.00)	*Chairman / secretary*	Room
A. Regulation of silage fermentation	*F. Weissbach - T.Pauly*	A
B. Protein utilization of silage	*R. E. Muck - G. Broderick*	J
C. Methods to predict feeding value of silage-based diets	*C. Thomas - M. Murphy*	O
D. Design and planning of silage experiments	*O'Kiely - Rammer - Seeger*	H
E. Determination and control of aerobic instability	*G.Pahlow - F. Driehuis*	K
F. Silage feeding in relation to food quality and animal health.	*Sivilä - Lindgren - Merry - Jakobsson.*	L
15.00 - 16.30 Summary - discussion	**Chairman R. Jones, U.K**	**A**

19.00 - 23.00 Conference Dinner at Västmanland-Dala Student Nation

Wednesday 7.7 1999

07.50 Bus departure, see time table
Accompanying persons are welcome to join the programme.

SESSION 6. Chairman H. Wiktorsson, Sweden **A**

08.30 - 09.30 Conclusions - adressing the next conference
K. Bolsen - M. Wilkinson

09.30 - 10.00 Coffee/tea

10.00 - 11.00 Presentation of LEGSIL
R. Wilkins - G. Pahlow

11.00 - 16.00 Kungsängen Research Centre - Open day
Bus transport provided

Kungsängen Research Centre

Group I = A Takes the bus for demonstration of LEGSIL crops
M.Tuvesson - M. Halling

Group II = B Visit to research activities in nutrition, feeding and management of cattle
J. Bertilsson - M. Murphy

Group III = C Demonstration of silage research and facilities

a) Storage stability - demonstration of methodology
T. Pauly / J. Andersson

b) Optimal ensiling of chopped forage - Collaboration with producers
C. Rammer / A. Engqvist

c) Optimal ensiling technology of bales
Collaboration with producers and researchers
D. Slottner / P. Lingvall.

Demonstration of heat stress, gas production, plastic quality factors = permeation and leakage, point preasure by loading, film colour etc.

d) Regulation of density in silage
R. Nylund

12.30 -13.30 Buffé lunch in the machine store

13.30 -14.30 Group I = B; Group II = C; Group III = A.

14.45 -15.45 Group I = C; Group II = A; Group III = B.

16.00 Bus to Stockholm Arlanda airport leaves Kungsängen

Main papers

The future role of silage in sustainable animal production

R J WILKINS[1], L SYRJÄLÄ-QVIST[2] & K K BOLSEN[3]
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[2]*University of Helsinki, Department of Animal Science, P O Box 28 00014 Helsinki, Finland*
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Abstract

Although silage currently makes a major contribution to ruminant feeding in Europe, its contribution on a global basis is much less. It is anticipated that there will be increased competition, particularly in Europe, with other feeds, especially grazed grass and grain. Opportunities for reducing the costs of silage production are discussed and the importance of constraining machinery and storage costs is stressed. The nutritive value of silage limits the contribution it can make to systems with very high levels of animal production, but approaches to restricting fermentation and protein breakdown in the silo will increase silage intake and protein utilisation and thus potential silage contribution. The present and prospective situation of silage is discussed for four regions – Nordic Europe, North West Europe, North America and South and East Asia. It is suggested that total silage production will fall in North West Europe and that in both the Nordic area and North West Europe there will be reduction in grass silage, with increase in silage made from other crops. Maize and lucerne will continue to be the major silage crops in North America and there will be an increasing need to improve preservation efficiency and nutritive value of these silages. In South and East Asia technological packages are being developed which may lead to substantial increase in silage making.

Introduction

Since silage making was introduced into Europe and North America in the mid 19th Century the technique has been widely advocated for the conservation of grass and other forage crops. This has arisen principally because of the potential to ensile wet crops and thus avoid the need for prolonged periods of dry weather required for hay making. Silage making was not, however, widely adopted for another Century, when the coincidence of progress in mechanisation, the availability of polythene sheeting to maintain anaerobic conditions and effective additives to control silage fermentation combined to produce a streamlined and reliable method for the production of silage of high feeding value.

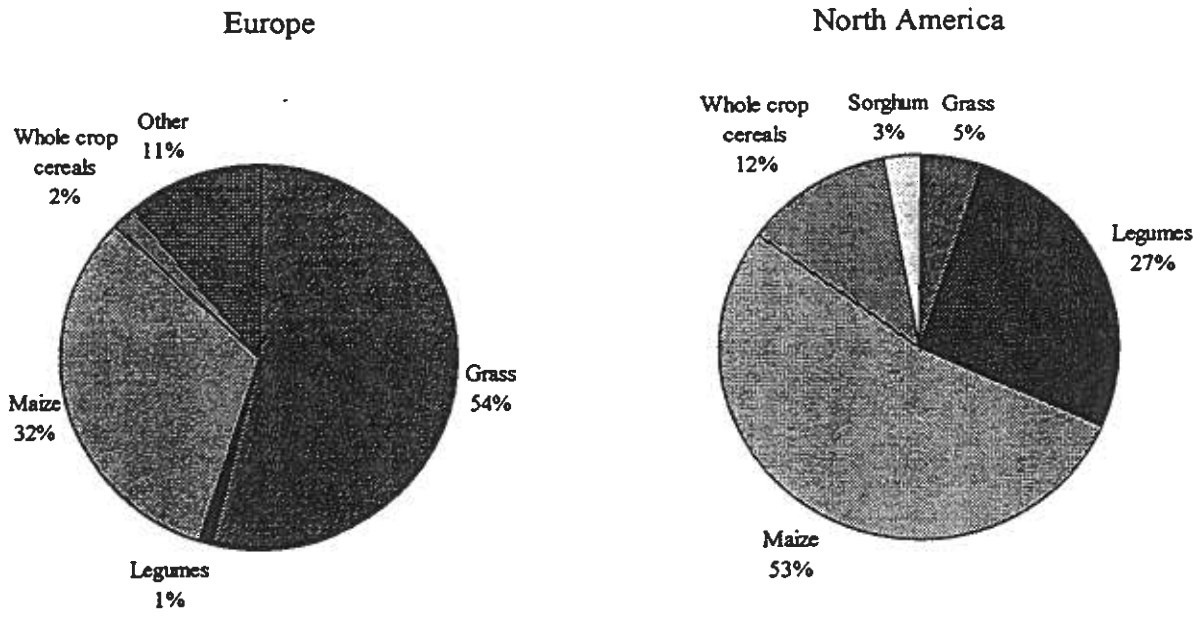
In the UK, silage output increased from only 0.35m t in 1947 to 46m t in 1987 (Brassley, 1996). Wilkinson and Bolsen (1996) indicated a total of some 199m t of silage dry matter (DM) produced in Europe and North America compared with 316m t of hay DM (Table 1). The proportion of conservation as silage was 0.5 in Europe compared with 0.22 in North America. Global estimates of silage making do not appear to have been made, but would be at least 250m t DM. Although silages contribute 10-25% of total feed nutrients to ruminants in Western Europe (calculated from Lee, 1988), on a world basis the proportion of nutrients derived from silage is only around 2%. The production of silage DM is low compared with estimated global availability of 900m t DM of cereal residues and straw (Kossila, 1984).

Table 1. *Estimated quantities of hay and silage produced in Europe and North America, 1994 (m t DM) (from Wilkinson and Bolsen, 1996)*

	Hay	Silage
Europe	152	152
North America	164	47
	316	199

A wide range of crops and systems are used for silage making. Figure 1 summarises the crops used in Europe and North America, emphasising the predominant use of grass in Europe and maize in North America. Silage making is generally a feature of intensive systems for milk and meat production. At a global level the major role is to provide feed in winter when crop growth rates are below those required to satisfy animal requirements. There is though important use of silage, particularly in North America, in storage feeding systems with animals housed and fed silage throughout the year. The third possible role of silage to supply feed at times when crop growth is limited by moisture shortage is, as yet, of less importance.

Figure 1. *Proportions of different crops harvested for silage in Europe and North America, 1994 (based on areas of crop harvested) (adapted from Wilkinson and Bolsen, 1996)*



The advantage of the reduced dependence of ensiling on good weather conditions has already been noted. There are other important advantages, particularly with annual crops. Much higher yields of nutrients per ha can be obtained from maize, sorghum and other cereals when they are harvested as whole crops rather than for grain. The flexibility in harvest dates means that decisions can be made late in the growing season as to whether a crop is taken for silage rather than grain, but more importantly early harvest for silage may facilitate improved cropping systems and rotations, with early harvesting allowing summer (e.g. maize) and winter (e.g. wheat) crops to be seeded sooner. With all crops, the ease of mechanisation from harvesting to feeding is a major advantage, particularly with more industrialised farming systems.

Against these advantages, silage is disadvantaged because of its high moisture content and lower nutrient density (per unit of fresh matter) than many other feeds. The high moisture content and need for anaerobic conditions necessitate more sophisticated storage conditions than for hay and other dried products, whilst both moisture content and nutrient density factors reduce the feasibility of transport, so that silage is normally produced and stored close to where it is to be fed.

This overview indicates that although silage is an important feed, there are substantial opportunities for further increase in silage making to replace other feeds in areas where silage is already an important source of nutrients and also potential for much greater use of silage in climatic situations in which it is currently of little importance. In some areas, however, increased competition with grains, by-products and grazed grasses may substantially reduce the use of silage, particularly grass silage, as discussed later in this paper.

This paper will firstly discuss limitations to the use of silage and opportunities for improving technology and then appraise critical factors for the future of silage in four regions of the world.

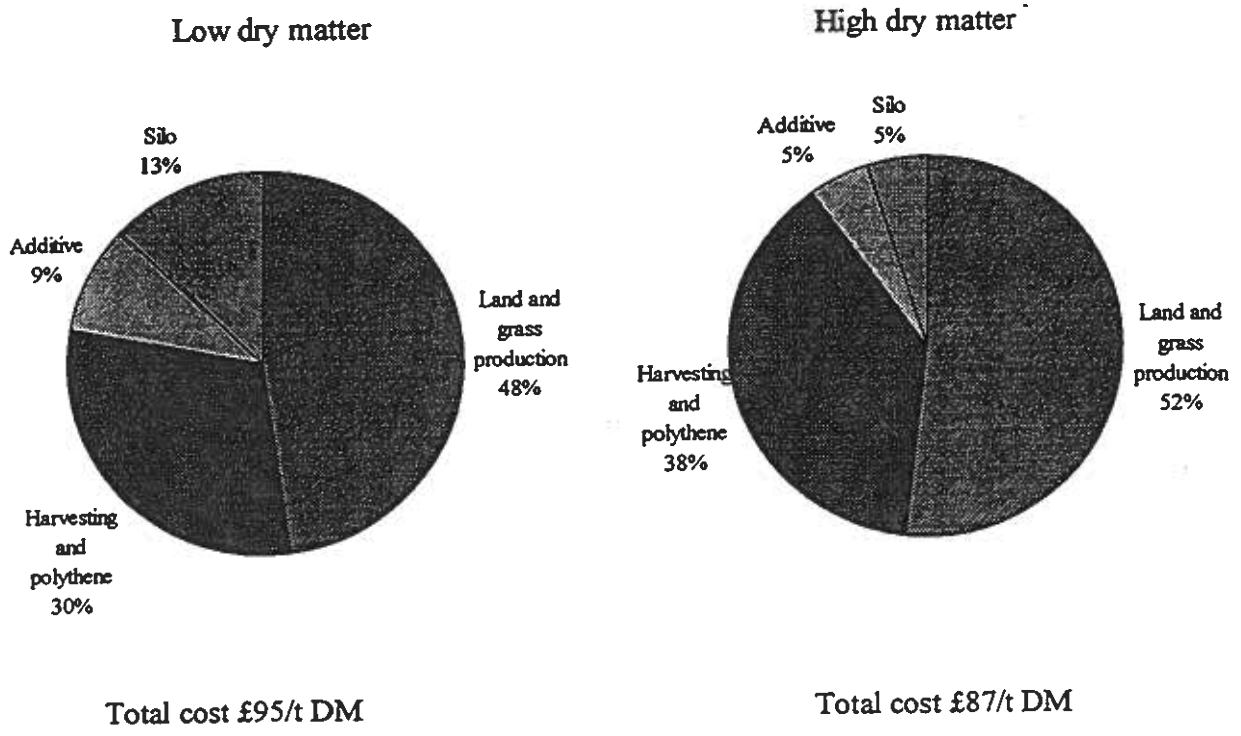
Limitations to silage and opportunities for improvement

This section considers limitations that may arise from cost or resource requirements, restrictions to feeding value and constraints to protect the environment.

Costs and resource requirements. Figure 2 indicates the different elements of the cost of producing silage DM as assessed by Mayne *et al.* (1998) for Northern Ireland. The cost of grass production represents about half of total cost, making grass silage a more expensive source of nutrients than grazing. Although zero grazing will still incur harvesting costs, the lower level of DM losses with zero grazing and absence of costs for additive and silo again mean that silage is more costly than zero grazing. Whilst silage is still a cheaper source of nutrients than grain, the difference is now much less than 20-40 years ago. In Finland, for instance, the production costs per feed unit are 0.9 FIM for grazing, 1.5 FIM for silage and 1.7 FIM for grain.

Costs per tonne of silage DM may be reduced by increasing the output of silage per hectare - through increased crop yields or reduced losses during the ensiling process - or by reducing the inputs in crop production or in silage making or storage.

Figure 2. *Cost of production of low and high DM silage in 3-cut system in Northern Ireland (adapted from Mayne et al., 1998)*



Increases in silage yield will reduce the costs per tonne for land rental and for crop establishment and harvesting. Thus use of high yielding varieties or species and optimising growth conditions may reduce costs per tonne of silage, although in some cases increasing costs per hectare. Cost reduction by lowering inputs in crop production can be achieved by use of long-term rather than short-term grassland, improved utilisation of fertilisers and organic manures and, in some cases, by substitution of grass by other forages, such as legumes, maize and cereals. There are good opportunities for substantial reductions in fertiliser inputs to grass without reducing yields, and at the same time reducing environmental losses of N compounds (Wilkins, 1996), and for the replacement of grass with forage legumes in production systems (Hopkins *et al.*, 1994). Such alterations in costs of crop production will improve the competitive position of silage in relation to concentrate feeds, but not in relation to grazing and zero grazing, because they will also benefit from the reduced costs.

Efficient sealing of silos will reduce costs per tonne of silage by lowering in silo losses. The largest source of loss during ensiling generally arises from aerobic respiration of plant tissues and of micro-organisms. Whilst there are possibilities for treatment of crops at ensiling to stop plant respiration and for biological or chemical additives to limit microbial respiration, it is probable that the main approach to limit losses will continue to be good management techniques for rapid filling, sealing and emptying of silos. Big bale techniques have demonstrated the

possibilities for practical application of these principles to produce silage with low levels of loss from aerobic processes. Losses also arise from fermentation and effluent and from physical loss of plant fragments in the field. The first two of these sources of loss are, of course, greater in wet than dry silage as demonstrated in Table 2. Even with wet crops substantial reduction in loss may result from the use of additives to limit total fermentation (or reduce heterolactic fermentation), or the incorporation of absorbent materials in the silo. Such approaches are, however, only likely to have a marginal affect on the cost of production of silage DM as a halving of losses from say 20% to 10%, even if achieved without cost, will only reduce costs of production/t of silage DM by 10%. However, impact on feeding value, as discussed later, may be far greater than this.

Table 2. Losses of dry matter (%) from unwilted and wilted silages in Eurowilt experiments (from Zimmer and Wilkins, 1984)

	Unwilted	Wilted
Field	2.5	8.0
Effluent	3.2	0
Other in silo losses	12.9	8.5
	18.6	16.5

Harvesting and storage represent about half of total costs, so that reductions in these items will have a large effect on the competitive position of silage. The cost of machinery has increased with increases in throughput capacity, with physical conditioning following cutting and with reductions in target chop lengths. Storage costs have, in many countries, increased because of tighter legislation to avoid risks from silage effluent contamination. These factors have increased the attraction of big bale systems which may involve less length reduction and avoid costly silo structures, but at a cost of more polythene/t silage stored, and increase in labour and feeding costs. Nevertheless such systems are now well established in much of the world and have many attractions for silage production with relatively low initial and subsequent costs. There is also likely to be further development of technology to improve baler throughputs, increase bulk density and improve wrapping efficiency. Radical developments in precision-chop harvesters, on the other hand, are less likely. With all machine systems a key factor for cost reduction is to achieve the highest possible throughput during the life of the machine. Obvious approaches to achieve this objective are larger farms, machinery sharing, contractor use of machinery, and use of multipurpose machines (e.g. big balers) reviewed and discussed in the Australian situation by Kaiser and Evans (1997).

Limitation of feeding value

Many experiments have been carried out over the last three decades on animal production from silage-based diets. The results have been variable with the main limiting factors when using

grass silage being low intake, inefficient utilisation of energy and low and unbalanced supply of amino acids.

Generally silage intake has been found to be lower than that of fresh forage and hay (Cushnahan and Gordon, 1995), although similar intakes of silage and fresh forage (Cushnahan and Mayne, 1995) and silage and barn-dried hay (Huhtanen, 1993) have been recorded. The intake of silage will be limited by the characteristics of the crop prior to ensiling in relation to its composition and digestibility, but it is also influenced by changes occurring during fermentation and storage, as modified by factors such as DM content, chop length and use of additives. There is a clear increase in DM intake with wilted compared with direct cut silage, but this approach to improve silage feeding value is limited, as there are only minor or even negative effects on milk yields (Ettala *et al.*, 1982; Bertilsson, 1987; Gordon, 1987). Digestibility and utilisation in wilted silage may be lowered through contamination with ash (Gordon, 1981; Bertilsson, 1990) and by increased passage rate (Tuori *et al.*, 1993).

Despite the complexity of factors limiting intake, there has been recent progress in improving intake prediction and thus the reliability with which silage feeding systems can be planned. Nousiainen and Hellämäki (1997) have developed an intake index for grass silage based on digestibility (D-value) and fermentation products (Table 3). In two major studies in the UK, near infra red spectroscopy (NIRS) provided precise estimates of intake of grass silages by cattle (Steen *et al.*, 1998), dairy cows and lambs (Offer *et al.*, 1998), with interesting possibilities of undertaking NIRS on fresh rather than dried samples.

Table 3. *The effect of D-value and fermentation quality on the intake index of grass silage (from Nousiainen and Hellämäki, 1997)*

D-value	Acids[a] g/kg DM	Ammonia g/kg N	Intake Index
72	50	30	110
72	80	50	105
72	120	80	98
72	120	120	95
69	50	30	105
69	80	50	100
69	120	120	90
64	50	30	97
64	80	50	92
64	120	80	85
64	120	120	82

[a] Lactic acid + volatile fatty acids.

Limitations to nutrient intake mean that silage alone cannot satisfy the requirement of high producing dairy cows or intensively growing cattle and sheep without supplementation. The

chemical composition and nutritive value of silage vary widely and thus the appropriate balance of nutrients in the supplements will also need to vary. Grass silage is usually fed with energy and protein supplements. Energy supplements have generally decreased silage DM intake but increased total DM intake. In a literature review, Ryhänen *et al.* (1996) found a mean substitution rate (decrease in silage DM intake per increase in concentrate DM intake) of 0.39, although substitution rates increase with increase in concentrate intake and with the extent of fermentation in the silo and are lower with low digestibility silage (Thomas and Thomas, 1989; Huhtanen, 1998). Substitution rate is not clearly affected by replacement of starch with fibre-based concentrates (Thomas and Thomas, 1989), but it is reduced with concentrates of high crude protein (CP) content (Aston *et al.*, 1998).

Silage nitrogen is degraded during fermentation to peptides, amino acids and ammonia. This soluble nitrogen fraction is rapidly degraded in the rumen, with negative effects on silage nitrogen utilisation (Syrjälä, 1972; Chamberlain *et al.*, 1985). Diets containing a large proportion of silage have been shown to have a low efficiency of microbial protein synthesis in the rumen compared with grass on hay-based diets (Thomas and Thomas, 1985), resulting in inadequate post-ruminal flows of protein. The protein value of restrictively fermented silage was, however, not inferior to hay (Huhtanen, 1998). Nevertheless many experiments have shown substantial responses in milk output to protein supplements, even where silages have a CP content of c. 160 g/kg (Chamberlain *et al.*, 1989; Thomas and Thomas, 1989; Tuori, 1992). Aston *et al.* (1994) reported linear increase in milk yield and protein as supplement CP content was increased from 120 to 240 g/kg DM. The increased intake of silage DM probably results from more efficient cell wall digestion (Oldham, 1984), increased microbial protein synthesis in the rumen and a better balance of amino acids. The duodenal digesta from silage fed animals appears to be deficient in certain individual amino acids, in particular methionine and lysine (Thomas and Chamberlain, 1982) or histidine (Vanhatalo *et al.*, 1997). Therefore it should be possible to replicate the effect of protein supplements by supply of individual amino acids.

It is unfortunate that although supplements high in CP content may increase silage intake and milk yields, such a feeding strategy is likely to result in a large increase in nitrogen loss in faeces and urine, with risks of increased loss to the environment (Figure 3). A number of approaches to improve protein supply are more promising. The possible use of individual amino acids has already been noted. Protein breakdown and water soluble carbohydrate (WSC) fermentation may be restricted by use of high rates of chemical additives, as demonstrated by Huhtanen (1998). Increases in true protein in silage may result from use of bacterial inocula, as demonstrated by Jones *et al.* (1996), and some forages may have natural protection to protein breakdown in the silo, as shown for red clover and lotus by Albrecht and Muck (1991). A further approach, appropriate in some regions, may be the feeding of mixtures of high CP grass silage with silages from maize or whole crop cereals with a wider energy:protein ratio, as demonstrated by Phipps *et al.* (1995).

Although approaches such as these may be used to improve silage feeding value, increases in the genetic merit of dairy cows mean that limitations to feeding value are likely to become an increasing constraint to reliance on high quantities of silage in the feeding of the highest yielding animals.

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watercourse pollution, studies by Foy and Kirk (1995) showed that major deterioration in river water quality in Northern Ireland coincided with the silage-making season, indicating the inadequacy of the precautions that were being taken at that time to prevent pollution, when the predominant silage making method was direct cutting of unwilted grass silage.

Whilst it is possible to collect effluent and then use this as a fertiliser or as a feed, as discussed by Offer *et al.*, (1991), the most appropriate approach is generally to restrict production by increasing the DM content of the ensiled material. This can be achieved by using naturally dry crops such as mature maize or whole-crop cereals, by the mixture of dry and wet crops or by the addition of absorbent materials, but the best approach is likely generally to be field wilting. There has been progress in developing machinery for macerating (or super-conditioning) crops in the field to increase wilting rates, with such techniques being particularly effective with lucerne (Savoie *et al.*, 1993), but with the need to constrain costs of silage making it will be necessary to carefully examine the cost benefit of techniques to increase wilting rate and simpler techniques involving crop spreading to maximise interception of radiation may well be more appropriate (Bosma, 1991).

Future of Silage in Different Regions

Nordic Europe

Although the growing period, or the period when the average daily temperatures are permanently over +5^{o}C, is short in Nordic countries compared with the situation in other European countries there are differences between the north, with a growing period of only 100-110 days, compared with 210-220 days for Denmark in the south of the Nordic area. The grazing period is therefore about three months in the north and about six months in the south. Despite the short growing season, maximum rates of grass growth are higher in the north than the south, associated with day lengths up to 24 hours. This gives total annual yields of approaching 10 t DM/ha up to the latitude of the Arctic Circle and, in the drier areas, good conditions for efficient grass utilisation.

Grass forages have traditionally an important role in Nordic countries. In Norway about 58 % of the arable land is grassland. In Sweden, Finland and Denmark the corresponding portions of grassland are 36 %, 33 % and 16 %, respectively. In Iceland 98% of the fields are in grass production (Nordic Statistical Yearbook 1998).

Due to the short grazing period and long indoor feeding period in the Nordic countries, conserved forages play an important role in feeding of ruminants. From the 1960's grass silage making became more and more common at the expense of hay production. The diet of the cows today is dominated by silage, particularly in Finland and Norway. Although there might be some advantages of including hay in the ration, the cost of maintaining two forage conservation methods on the farm is too high. Up to 1990 more hay than silage DM was produced in Sweden, but during the last ten years big bale silage has increased considerably at the expense of haymaking. In Denmark grass is still the major crop for silage, accounting for about half of the total silage production (Mo, 1995). During the last 15 years, however, the ensiling of maize and other whole crops (barley, wheat) have increased. In Iceland hay is still the dominant roughage for winter feeding although ensiling, especially of big bales, increased enormously in 1990's.

Grassland is suited so well to the northern parts of Europe that it is very difficult to find a competitor for grass silage, expect in Denmark and South Sweden. Many attempts have been made to grow maize north of the 60° latitude, but the results have generally been poor. In recent years whole crop silage from barley, wheat and oat has been of much interest for farmers in northern areas of Europe.

The production of whole crop silage (excluding maize) is geographically possible at about the same latitudes as grass silage production. Besides quite high yields (very often over 10 t/ha), the reliability of yield is high. Being annual crops, they are not exposed to the winter damage which may severely reduce grass yields. Furthermore, harvesting equipment already present on farms for grass silage can be used for whole crop silage and there are environmental advantages. There is a lower requirement for N fertiliser for whole crop, good opportunities for use of animal manures and a DM content at harvest sufficiently high to prevent effluent production.

Many experiments have compared grass silage and whole crop silage in cattle feeding (e.g. Phipps *et al.*, 1995). The lower digestibility of whole crop silage restricts its use especially in the feeding of high producing dairy cows, but it can substitute for about 20 % of grass silage without decrease in milk production (Jaakkola and Joki-Tokola, 1999).

In addition to competition with whole crop cereals, there is the prospect of increased competition for grass silage from cereal grain. At present the cost of nutrients in silage is lower than that in grain, as noted earlier. Changes in CAP support in Agenda 2000 will however reduce the price of grain and whole crop silage in relation to grassland feeds. In view of this increased competition, it will be particularly important to take actions to lower the production costs and to improve nutritive value if grass silage is to maintain its current dominant position in the Nordic countries.

North West Europe

This region has high annual grass production, but with limited herbage growth during winter. The predominant herbages are grasses and grass-legume mixtures and swards are generally grazed during the main growing season with silage being the principal method of grass conservation.

In NW Europe, there was a large increase in silage making from about 1970 to 1990, with silage succeeding hay as the main method of conservation, and an increase in the total amount of conserved forage. This was largely at the expense of grazing and Wilkins (1992) noted that the proportion of grass in the UK that was conserved rather than grazed increased from 28% to 47% over that period The importance of maize silage also increased markedly in the 1990's.

The position of grass silage in NW Europe has been increasingly challenged in recent years with a trend for reduction in the price of animal products, reductions in the price of grain, increases in the yields and reliability of maize production and increases in the costs of silage harvesting and storage. As noted earlier, the costs of supplying nutrients in conserved grass are markedly higher than in grazing, so that the trend to increase silage at the expense of grazing was increasing production costs. There has, particularly in Ireland and UK, been increased attention in the last

five years to extending the grazing season and reducing the period of reliance on conserved forage. This trend is likely to continue along with progress in breeding grasses with longer growing seasons and the use, in some areas, of crops such as kale and fodder roots that can be grazed during the winter. Extension of the grazing season will, however, be constrained by risk of poaching on wet, poorly-drained soils, the low rates of grass growth that can be sustained with low radiation inputs in winter and high losses which occur when grazing is deferred to a time substantially after the period of growth (Wilkins, 1995a).

There will still be substantial production of grass (or grass-clover) silage from areas used principally for grazing, through the removal of herbage that is in excess of the requirements for grazing during peak periods of growth. This will be leafy material of high quality, because of the need for frequent harvesting to give rapid regrowth and ensure continuity of supply of high quality material for grazing. In some systems this may provide sufficient material to bridge the gap between the grazing seasons, but in areas with more prolonged winter periods or heavy soils there will be some requirement for crops grown specifically for conservation.

In many areas maize will be the outstanding candidate, with maize silage being a good complement for grass silage (Phipps *et al.*, 1995) because of its high content of readily available carbohydrates. New varieties of Italian and hybrid ryegrasses may have characteristics similar to maize with high ratio of digestible carbohydrate to CP. With all grass silages there will be a strong case for adopting techniques to preserve WSC during ensiling in order to improve overall nutrient utilisation, through systems involving severe wilting or use of additives to restrict fermentation. There may also be opportunities for breeding new grass varieties with reduced rates of protein breakdown in the silo and the rumen (Wilkins, 1995b). Forage legumes may become more important in future production systems, seeking to capitalise on the high nutritive value of legumes and low production costs. The high intake of well-preserved forage legumes is well recognised, but there is increasing evidence of retention of true protein in the silo with red clover (Jones *et al.*, 1993) and protection of protein during ensiling and in the rumen with lotus and sainfoin. There are, substantial problems in achieving high crop yields with lotus and sainfoin, but red clover is well adapted to much of NW Europe and is particularly attractive in organic production systems, which are likely to increase markedly in importance.

It will be important to constrain the costs of silage production, because of increased competition from cereal growing. Thus reduction in machinery and silo costs will be particularly important. It is probable that the total use of conserved forage will fall and that an increased proportion of silage will be produced as a 'by-product' from grazing and from maize, with some increase in use of red clover. Silages will be made from wilted crops, in order to restrict effluent production in these areas of high livestock density and environmental sensitivity.

North America

Although dealing specifically with North America, most of the comments apply also to the southern part of South America, where there has been considerable recent increase in silage making. There have been considerable improvements in silage making on farms in the past 6-8 years. Many dairy and beef cattle producers have initiated significant changes to improve their silage programmes. These include: shorter chop lengths, field-wilting forages to the correct DM

content, proper sizing of the silo's width and height, improved maize and/or sorghum hybrids, faster removal rates during the feedout phase, more effective sealing and the use of bacterial inoculants (Waybright, 1997; Bolsen *et al.*, 1998).

However, poor silages are still often made, despite in many cases, the availability of appropriate technology. These arise from (i) delayed filling (often 7-10 days to fill), (ii) ensiling forages at either too low (lucerne, ryegrass and clover) or too high (some maize and sorghum hybrids) DM contents, (iii) poor chopping (lucerne, grass and whole-crop small-grain cereals, (iv) slow removal during feed out, (v) uneven fermentation (this may largely be solved by the use of a bacterial inoculant). Inadequate sealing is a major source of loss and requires further attention. Horizontal silos are commonly left unprotected, with losses in the top 1 m typically in the range 40-80% (Bolsen *et al.*, 1993). In addition to this direct loss, inclusion of some top spoilage in the diet will reduce DM intake and digestibility significantly (Whitlock and Bolsen, unpublished data). Whilst materials such as limestone, sawdust and molasses have been evaluated as surface covers, a properly weighted polythene sheet is still the most effective method to reduce DM and nutrient losses (Holmes, University of Wisconsin, Madison, personal communication).

Major problems which still require improved technical solutions are aerobic deterioration of maize silage, improved wilting rates for grasses and forage legumes and methods for improving grain utilisation in maize silage.

There have been recent advances in relation to wilting rates and improved grain utilisation. For grass and lucerne, disc mowers have increased mowing capacity compared to cutterbars, and the installation of conditioning devices on mower-conditioners, such as crushing rolls or plastic brushes, have increased the drying rate of forages by 20 to 40%. More intensive mechanical conditioning through a series of serrated rolls (mat-making or super conditioning) has field-dried lucerne up to two to three times faster compared to field-wilting in a conventional windrow (Savoie *et al.*, 1993).

For harvesting whole-plant maize, machines with an on-board kernel processor have drawn more and more attention, especially from dairy farmers in North America (Shinner, 1997). The kernel processor helps to minimise the risks associated with hybrids that increase rapidly in DM content with maturity. The harvest window is extended and the utilisation of the energy and fibre components of the silage is improved. Cows fed the kernel-processed maize silage had fewer kernels in the faeces (0.9 *vs.* 3.1%), higher milk production and improved milk protein and lactose values compared to cows fed the unprocessed maize silage diet (Harrison *et al.*, 1997). Young *et al.* (1998) ensiled whole-plant maize at 80 to 90% milk-line and reported that growing cattle fed kernel-processed silage gained 9% faster and 6% more efficiently than those fed unprocessed silage. The relationships between kernel processing, maize hybrid, milk-line score and chop length need further clarification, particularly as processing affects preservation efficiency, aerobic stability and starch and fibre digestibilities (Satter and Muck, Dairy Forage Research Center, Madison, personal communication).

Looking to the future, there are clearly opportunities for further improving the efficiency of silage making on farms through better management and use of relatively low-cost technology that is already available. Maize and lucerne are likely to remain the major ensiled forages and

there will be an increasing premium on improvement in the nutritive value of these silages (Kung and Muck, 1997). High yield, high energy concentration and low cost are key features in the importance of maize in this region, both in intensive systems and as a complement to pasture-based dairy and beef production. Whilst there is much current increase in bag and bale silages, particularly to ensile 'extra' silage, it is doubtful whether these systems will continue to gain in popularity.

South and East Asia

One of the major limitations to livestock production in the dry-humid and humid tropics is the uneven supply of pasture forage during the year. Most of the tropics have differentiated wet and dry seasons, and there is commonly a surplus of forage during the wet season and a shortage during the dry. In many systems the surplus remains as standing hay to be consumed during the dry season. This material is of very low quality and is used inefficiently. Systems with more intensive land use may have very high reliance on straw and other crop by-products.

Ensiling has commonly been advocated as the approach for improving systems particularly as it is very difficult to make hay of high quality because of extremely high rainfall during the period of rapid herbage growth. Until recent years, however, silage has had very little impact in this area. Difficulties have arisen from tropical grasses and legumes typically having higher concentrations of cell wall components and lower levels of fermentable carbohydrates than their temperate counterparts (Catchpoole and Henzell, 1971; Jarrige *et al.*, 1982), low levels of available mechanisation, limited availability and high cost of materials for effective sealing of silos and low prices for animal products. There are, however, now opportunities for increased use of silage making, particularly in areas where human population densities permit some production of specialist forage crops or regions where surplus herbage can be cut from common lands. The development of markets for milk in, for instance, Thailand and India, with requirement for regular supply of feeds has increased the case for considering silage production.

Appropriate silage making techniques are outlined by Bolsen and Faylon (1994) and by Tripathi *et al.*(1995). They have recently been introduced in several countries in South and East Asia regardless of the size of farm and type of livestock programme. It is important to be able to chop the cut forage in order to achieve successful fermentation and storage. This may be done effectively with stationary choppers and programmes are currently in hand to encourage farmers to form cooperatives and share equipment needed to make silage from their own surplus forage. Polythene sheets and polythene bags are much more readily available for producing sealed silos, and with increased milk prices inputs such as these may be more readily considered in production systems. An interesting approach in Thailand is the development of a 'small bag' silage technology in which forage will be harvested by hand, chopped and filled by hand into a bag containing 10-30 kg of forage, that is then compressed and sealed (Nakamanee, 1997). Each bag would provide silage for only a few cow days, thus avoiding risks of deterioration when silos are opened for long periods in hot weather conditions. Innovative approaches of this type are required to fit into smallholder farming systems.

Bolsen and Faylon (1994) draw attention to a wide range of plant products and by-products which may be used to improve the ensilability and nutritive value of tropical silages. These

include molasses, cereal brans, citrus pulp and other fruit processing residues. There are also possibilities for making silage from by-products such as sugar cane bagasse and oil palm fronds (Zainal, University Putra Malaysia, Serdang, personal communication), but these are likely to be low quality maintenance feeds.

We believe that silage making will increase in this region, but care must be taken to develop appropriate technologies and to look at the way in which silages can best complement other feeds that may be available. Silage from special forage crops is likely to be one of the most expensive sources of nutrients (other than grain), and so the achievement of good preservation and high quality is particularly important.

Conclusions

Silage will continue to have a substantial role in animal production systems in temperate areas of the world to supply energy, protein and fibre to housed animals during periods of low crop growth. There will, however, be increased competition as a feed source with grazing and grain, particularly in areas like NW Europe with a relatively long growing season and where silage making is already well established. There will be a need for efficient use of good technology and inputs in order to constrain the costs of silage production. Cooperation and contractor use of machinery will be particularly important in achieving cost reduction. It is probable that there will be some replacement of grass silage with silages made from maize, whole crop cereals and forage legumes. In some temperate and tropical areas where silage currently supplies only a small proportion of nutrients (e.g. Australia, New Zealand and Latin America) there will probably, in contrast, be increases in silage production, to capitalise on advantages of silage technology in producing more even seasonal supply of nutrients from grassland and forage. This expansion is being encouraged by progress in technology, increase in milk and meat prices and increase in consumption of animal food products.

The potential of silage to contribute to tropical and sub-tropical animal production is, as yet, largely unrealised. The opportunities are greatest in areas which are seasonally dry, but with limitation of water supply from irrigation. Appropriate technologies for making silage from forages and by-products are emerging, but there will be a continued requirement to harvest material of reasonable feeding value and to maintain that feeding value and reduce losses during storage. There are opportunities for cooperative use of chopping machines and for storage in small well-sealed plastic silos.

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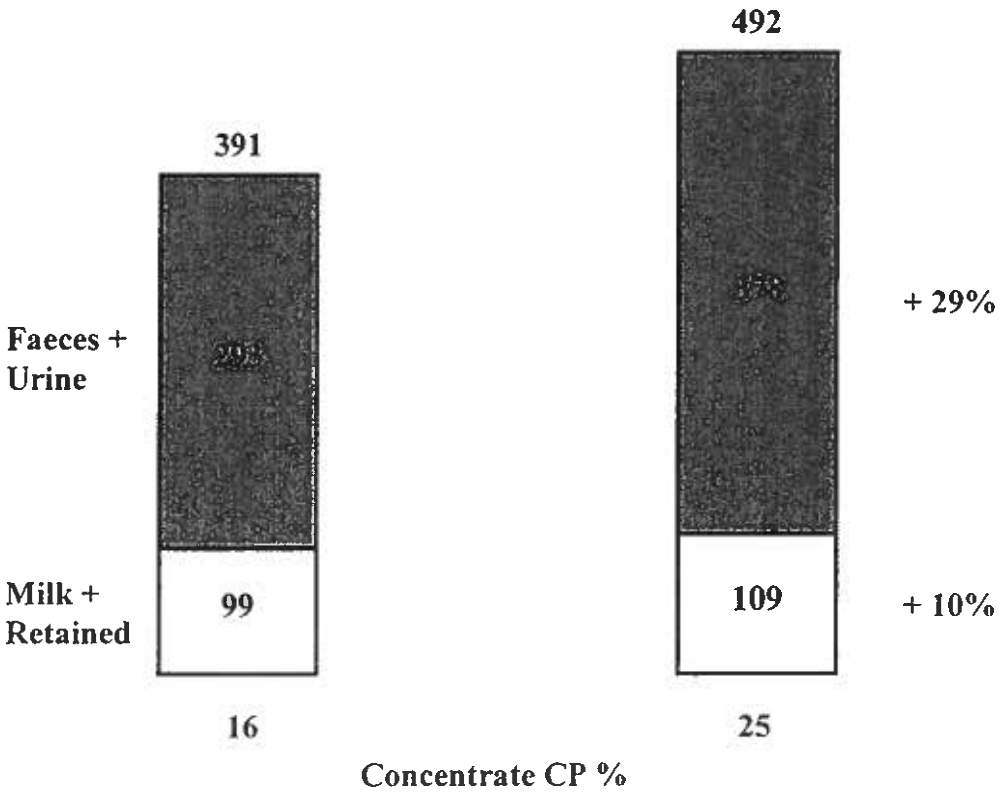
Figure3. *Effect of increasing CP % in concentrate fed with ad lib. grass silage on N in milk and excreta (g/day) (from Aston et al., 1998)*



Environmental Issues

There has over the last two decades been increased concern for the environmental impact of animal production systems. This section will concentrate on the risks from silage effluent contaminating watercourses, although there may also be adverse effects from leaching of N and P from land growing crops for silage, adverse effects of micro-organisms in silage (and deteriorated silage) on human and animal health (discussed by Platt, 1999) and concerns and for the aesthetic impact of silos and feeding structures. Efforts must be made to develop plastic recycling programmes for continued acceptance of silage in production systems (see Negra and Rogers, 1997).

The high biological oxygen demand of silage effluent is well recognised. This has led to legislation in many countries not only to prevent water contamination, but also for new permanent silos to be designed to tight specifications to ensure effluent collection, thus resulting in a massive increase in the cost of silos, even if they are to be used only with dry crops. In the UK, restrictions on bale silage and field stacks is less severe. Despite legislation to restrict

RELATIONSHIPS BETWEEN SILAGE BASED DIETS AND FEED CONVERSION

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Introduction

The feeding value of silage depends largely on its effect on intake but also on its importance for the metabolism and feed efficiency of the animal fed the silage. The importance of a high feeding value has increased along with a continuously enhanced genetic production potential. Furthermore the proportion of the herbage conserved as silage has increased in many countries during the last decades. A major reason for that increase is a desire to lessen the dependence on weather conditions for forage preservation. Ensiling also enhances the possibilities for mechanisation and a higher utilization of grassland productivity than haymaking. The quality of silage can, however, vary within a wide range and is to a high degree related to management at ensiling. A good management gives a forage of high feeding value, while suboptimal procedures result in a low quality or even unusable feed.
The objective of this paper is to discuss factors affecting utilization of diets based on silage from grass and legumes crops in animal production with cattle.

Voluntary silage intake

Several feed related factors are suggested to affect silage intake. It has been shown that intake of silage is generally lower than the intake of the corresponding fresh forage. In a great number of comparisons with sheep the reduction in intake of direct cut silage varied between 1 to 64% and was on average 30% (Demarquilly and Dulphy, 1977). Later results presentated by Keady et al (1996) indicate that the difference between the ensiled and the fresh forage may be smaller for cattle than for sheep. Futhermore Cushnahan et al (1993) concluded that data from sheep was not a reliable predictor of intake for cattle. The production potential of the animals used to measure the ingestability may also affect the magnitude of the difference as well as if silage DM is corrected for losses of volatile compounds or not. Animals with a low energy requirement are probably less sensitive than animals with a high requirement. As will be discussed below silage intake can vary to a great deal due to the fermentation quality of the silage. This could influence the results from comparisons between silage and fresh forage. The continues improvements in techniques for producing silage should, however, imply that the reduction of intake by ensiling the crop could be less today than it was a couple of decades ago.

Silage quality factors

The fermentation process during ensiling alters the chemical composition that can affect both the nutritive value and the intake of the forage. The fermentation characterised by a utilization of soluble carbohydrates and proteins results in an enrichment of the less digestible fractions of the forage. The depression in silage intake compared to the fresh crop is often related to the fermentation end products.

Due to the modification of the composition that occurs in the fermentation process the prediction of silage intake is generally more difficult than with unfermented forages. Many

models have been developed with a varying degree of accuracy. Of the most frequently used parameters for measuring fermentation quality pH *per se* generally seems to be of minor importance. Ammonia as proportion of total N has been negatively correlated to intake of silage as well as the content of total acids or volatile fatty acids in the silage (Rook and Gill,1990; Huhtanen,1993). The effect of ammonia is probably indirect through its correlation to other fermentation products acting as the causal agent (Rook and Gill,1990).

Silages with different characteristics will result in different rumen fermentations and utlimately in differences in the metabolic responses of the cow. Chamberlain and Choung (1993) demonstrated this with differences between fermented and restricted fermented silages. Lactate levels in the silage have been shown to affect both DM intake (Huhtanen, 1993) and in product qualities (Chamberlain and Choung, 1993). While these fermentation characteristics are important there are several other characteristics which are also influencing intake and performance.

Lactate is rapidly metabolised in the rumen but with a low ATP yield which may be a limitation for microbial processes in the rumen (Chamberlain and Choung, 1993; Huhtanen (1993). Even in silages with high lactate concentrations less than 10% of rumen digestible OM in a lactating cow will come from lactate. While this may be substantial it is most likely too small to elicit a response that can be accurately measured in vivo under varied conditions. A 5% decrease in MPS from 19 g to 18 g microbial N kg^{-1} digested OM would reduce microbial protein flow to the duodenum by 50 - 60 g d^{-1} which might be difficult to demonstrate in lactating cows. However, lactate might be influencing rumen metabolism indirectly by affecting the microbial ecology. High intakes of lactate from silage are often associated with high intakes of starch rich supplements to the lactating cow which also can alter rumen ecology. The effects must be related to the microbial ecology.

The degradation rate of the fiber fraction has been shown to be highly variable. This functional characteristic may be as important as the chemical composition. It will influence passage rate and thereby also influence intake. It would appear to also influence the VFA composition. Other important factors influencing rumen function and intake are functional gravity, lignification and ratio between hemicellulosa and cellulosa.

VFA composition in the rumen

The VFA composition found in the rumen of cows consuming grass silage based diets differs from that found in diets based on maize or alfalfa silage. Often the butyrate levels are generally higher with grass silage diets and there is less propionate when the diets are supplemented with grain. This is especially true in Sweden, Norway and Finland(Murphy et al. 1999; Randby, 1998; Jaakkola and Huhtanen, 1993) but not so prevalent in Denmark (Stensig et al. 1998).

Table 1.

	Molar percent of total VFA			*OM Intake*	*Concentrate*
	Acetate	Propionate	Butyrate	kg	% of diet DM
Sweden[1]					
High roughage	63	19	15	16,0	30
Low Roughage	62	19	17	15,7	70
Finland[1]					
High roughage	65	17	14	6,2	25
Mid Roughage	64	16	16	6,4	37
Low Roughage	60	17	17	6,3	75
Norway[1]					
60% Roughage	65	20	12	16	40
Denmark[1]					
High roughage	60	22	13	14	38
Low Roughage	56	27	13	15	48

[1] See text for references.

One of the main causes for this difference could be the rate of fiber digestibility. This might also explain why there is a difference in Denmark. The rumen degradable NDF (RD NDF)in red clover, orchard grass, timothy and perennial ryegrass harvested at recommended maturity was 32, 51, 40 and 45, respectively (Murphy, unpublished). This recommended maturity corresponding to stages 50 and 52 of Simon and Park is a little later than Maturity 2 in the study of Hoffman et al. (1993). Nevertheless at Maturity 2 they reported rumen degradable NDF percentages of 19, 40, 28 and 50 for red clover, orchard grass, timothy and perennial ryegrass, respectively. Thus with the exception of ryegrass grasses grown in northern climates had a greater degradability even at a later maturity. At Maturity 3 RD NDF for ryegrass was only 35% as reported by Hoffman et al. The lignin fraction was approximately 1 percentage unit higher in the American study. It could be additionally seen in the Swedish study that there was a decrease in the RD NDF with the same species grown in different locations. The RD NDF fraction differed by about 5 percentage units between Uppsala and the far south of Sweden when harvested at the same stage of maturity.

In Dutch experiments with silages harvested at two different stages of maturity de Visser et al. (1998b) reported that the molar percentages of propionate and butyrate were 18 and 13%, respectively and these did not alter with stage of maturity or increasing starch levels in the diet. Even though starch decreased the degradation rate there was no interaction between starch and stage of maturity. In contrast Friggens et al. (1998) presented equations to correlate diet chemical composition and ruminal VFA proportions but only one silage was included as the basal diet which was a major limitation.

During approximately a two week interval of the second harvest the NDF content of a mixed sward increased from 47% of DM to 52% while the RD NDF decreased from 49% to 48%. The same development was seen in the first harvest (Murphy, unpublished). Even though the metabolizable energy decreased from 10.1 to 9.7 MJ per kg DM this was probably due more to a decrease in cell contents, especially protein, than to a decrease in fiber quality. de Visser et al. (1998a) also reported no significant changes in NDF degradation rates between perennial ryegrass silages harvested after 21 days or 36 days of regrowth even though NDF content increased from 39% to 46% of DM. As expected and noted in several trials the rumen pool sizes of NDF tend to increase with increasing maturity of grass silages.

This has been both accompanied by a decrease in digestibility (Jaakkola et al. 1993) and with no changes in digestibility or degradation rate (Van Vuuren et al. 1999; de Visser et al. 1998a). The last situation then would simply be due to the higher NDF content in the grass harvested at a later date. Other reports from Holland indicate a more profound effect of stage of maturity on the rate of degradation (Bosch et al., 1992a, Bosch et al. 1992b)). Unfortunately those silages were not from the same harvest or even from the same year. Even in those trials, the propionate concentration was relatively low (16 - 17.5%). The rate of passage increased with stage of maturity and this was due to changes in the specific gravity.

Animal performance

When comparing animal production from diets with or without silage the result is dependent on the proportion of the forage in the diet and if ad libitum feeding is applied or not. Ad libitum feeding enables an assessment of the feeding value but renders it more difficult to separate effects of intake from that of the nutritive value. The animal response at ad libitum feeding could also be related to the production potential of the animal. For instance a cow with a low merit for milk production is expected to give less response at increased intake than than the high merit cow. Measuring feeding value with low yielding animals could thus give misleading results. Therefore when discussing feed conversion of silage based diets it would be appropriate to discriminate between situations where silage is fed ad libitum and when it is fed in restricted amounts.

Silage versus hay

Performed comparisons between silage and hay based diets have often resulted in higher production and/or a better feed efficiency with silage. In a review of experiments in Norway Presthegge (1959) and Saue (1966) found higher milk produktion with silage when forage feeding was restricted. A higher feed efficiency with silage diets are also reported by Stone et al.(1960) and Waldo (1977) but not by Murdoch and Rook (1963).

In a series of experiments with restrictedly fed cows by Burstedt(1978) and Bertilsson and Burstedt (1984) the production results were consistently in favour of silage. In the experiment by Bertilsson and Burstedt the combined effect of harvesting time and conservation method was examined. The silage was direct cut with formic acid as an additive. From the data it seems that the difference between hay and silage depends on stage of maturity at harvest or that the maturity at harvest is less important for silage provided a good fermentation quality (Table 1). That also means that the advantage of silage was smaller at an early than at a later harvest. Of the measurements performed in the experiments the higher gross energy content especially at late harvest and a higher digestibility of organic matter of silage could explain part of the differences obtained. Results from more recent experiments seems to support the opinion that well preserved silage will give a better animal performance than hay from the same crop (Åkerlind et al.,1999; Andrieu et al., paper presented at this conference).

Table 2. Intake and milk production in experiments with restricted feeding of hay or unwilted silage harvested early or late. Bertilsson and Burstedt, 1984.

Reference	Treatment	Intake, kg DM/d. Forage	conc.	Production kg FCM/d. Rel. Fig.
Bertilsson, 1983[1)]				
Expt. 1 (45 cows)	Hay early	8.8	6.8	100
Lact. w. 2-35	Silage early	8.7	7.1	104
	Silage late	8.6	7.1	101
Expt. 2 (48 cows)	Hay early	8.7	6.0	100
Lact. w. 2-40	Hay late	8.5	5.3	91
	Silage late	8.8	6.3	105
Expt. 3 (38 cows)	Hay early	7.7	6.9	100
Lact. w. 2-40	Hay late	7.7	6.4	92

Wilting

The increased intake of silage generally obtained by wilting has not always resulted in a corresponding increase in animal performance (Rohr and Thomas, 1984). Often the effect has been small or even negative. The variation in the response to wilting obtained in different experiments is probably related to weather conditions at wilting. In the Eurowilt project (Rohr and Thomas,1984) it was also shown that a poor performance with wilted silage was related to a high ash content of the silage, indicating suboptimal wilting conditions. The effect of wilting on animal production may also depend on nutrient supply from the supplements fed together with the silage. Other factors influencing the response are level of intake and the ability of animals used in the experiments to respond to an increased intake. At intake levels exceeding the production potential of the animals the marginal response is expected to be poor due to the law of diminishing returns. It could thus be argued that restricted feeding should by applied when meaasuring effects on nutritive value. In experiments with early lacating cows no differences in daily yields of fat, protein and milk were found (Honig et al.,1984; Bertilsson, 1990). The results indicate almost identical nutritional values of wilted and unwilted silages. Both silages had a good fermentation quality. Dry matter content was 13 to 27 percent units higher in the wilted silage The proportion of forage in the diets varied between 51 to 59%. These results illustrate the dependence on wilting conditions and that no general conclusion about the effects of wilting on feeding value can be done.

Effect of supplements

It is well known that concentrate feeding generally depresses intake of ad libitum fed silage. The rate at which concentrate substitute silage varies videly due to the quality of the feedstuffs used. The substitution effect is often ascribed to a decreased digestibility of fiber with an increased level of concentrate. This should imply that substitution rate (SR) is related

to level of concentrate feeding. Results from different experiments are, however, somewhat conflicting, which might be explained by too low levels of concentrate to disturb the fiberolytic capacity of the rumen microbes. Misleading values of SR could also be obtained if the increased concentrate feeding results in a higher production and as a consequence a higher intake capacity. However, if the concentrate feeding results in a higher energy balance of the animal this should according to Faverdin et al. (1991) give an increase in SR and thus counteract the effect of the increased appetite. The depressing effect on silage intake of concentrate feeding seems to be related to the ingestibility of the silage. SR is thus expected to increase with increased intake of the silage when fed alone (Thomas, 1987). A high SR should thus be obtained with a well fermented highly digestible silage.

The effect of concentrate composition and substitution rate has been investigated in several experiments. The most consistent effect on the intake of silage is found the with protein content in the supplement. Thus Castle (1982) concluded from his own experiments and experiments by others that feeding oilseed meal or increasing the protein content of the concentrate had a positive effect on silage intake. In more recent studies with rapeseed meal Tuori (1992) found a significant increase in silage intake when crude protein content of the diet increased.

It has been suggested that preformed amino acids and/or peptides would enhance microbial protein synthesis especially in starch rich diets as the amyolytic bacteria have relatively large requirements for peptides (Russell et al., 1983). In the Cornell model the peptide concentration in the rumen modulates microbial protein synthesis. It has also been generally assumed that the levels of amino acids and peptides are very low in the rumen.

In vitro experiments have both confirmed positive effects on feed degradation and microbial protein synthesis (MPS) of including amino acids and peptides (Oh et al., 1999, Griswold et al., 1996; Hoover et al., 1996) and refuted the positive effects (Jones et al. 1998; Soto et al., 1994). In vivo experiments generally have shown no extra value of preformed peptides and amino acids on MPS (Oh et al.,1999; Wallace, 1999) or only with a rapidly fermentable energy source (Chikunya et al. 1996). Aspects which have to be considered when judging the benefits of peptides include the effects of the N source on digestion, the recycling of microbial protein in the rumen (Morrison and Mackie, 1996) and which bacterial populations need to be stimulated (Wallace, 1999).

Most of the data concerning protein degradation in the rumen is based on in sacco studies. As protein is calculated to be digested as soon as it leaves the nylon bag the degradation of larger peptides and amino acids to ammonia has been overestimated. Experiments in Norway with infused amino acids (Volden et al.,1998) showed a large escape of amino acids from the rumen. In vitro (Eriksson, 1999) and in vivo trials (Murphy, unpublished) in Sweden have shown that with both grass and alfalfa silage the concentration of amino acids in the rumen is between 5 and 15 mmol l^{-1}. The concentration of peptides in the rumen is more difficult to assess and which size of peptides which would be influential for microbial protein synthesis is not well documented. Also chemical alterations to the peptide chain can render them inaccessible for the rumen bacteria (Wallace, 1999). However, these compounds would still be largely available to the animal in the small intestines and could make a sizeable contribution to the protein supply. Thus it is reasonable to assume that modern protein evaluation systems based on in sacco results underestimate the flow of feed peptides and amino acids and thereby underestimate the protein value of the silages.

Rumen bacteria in silage based diets

A major determinant of the nutritive value and intake potential of silage is rumen fiber degradability. Many strategies designed to alter rumen fermentation are based on an assumed microbial ecology that is perhaps dominant in North American diets. Studies on the microbial ecology of Swedish cows (Van Gylswyk, 1990; Van Gylswyk and Murphy, 1993) have shown while some aspects such as the dominance of *Prevotella ruminicola* are similar there exist other more profound differences in other aspects. Most interesting with respect to the VFA composition and the higher intake of NDF (as % of LW) (Bertilsson, unpublished) are the differences in fiberolytic and cellulolytic bacteria.

Percentages of the cellulolytic bacteria species in lactating cows fed silage and hay diets are presented in Table XX (Van Gylswyk and Murphy, 1993). The total numbers of bacteria expressed per g of rumen contents differed more in response to the type and amount of supplements in the diet than to the type of forage (Van Gylswyk and Murphy, 1993). *Butyrivibrio fibrisolvens* was the dominant cellulolytic organism with hay diets but on silage diets *Ruminococcus sp.* were dominant. In addition *Eubacterium cellulosolvens* was always around 20% of the cellulolytic population. This bacteria has only previously been reported as being important in Dutch cows. These differences between hay and silage diets were significant but the distribution differences within forage preservation type were not.

Table 3. Proportions (% of total colonies examined) of cellulolytic bacteria resembling *Butyrivibrio fibrisolvens, Eubacterium cellulosolvens, Ruminococcus albus* and *R. Flavefaciens* in hay and silage diets supplemented with various concentrates.

Organism	***B. fibrisolvens***	***E. cellulosolvens***	***R. albus***	***R. flavefaciens***
Diet	In % of total colonies examined			
Hay rations				
Low roughage	60	20	14	6
High roughage	74	15	6	5
High sugar	75	22	2	2
Silage rations				
Low roughage	15	29	34	22
High roughage	32	15	24	29

In typical North American studies *Fibrobacter succinogenes* is the dominating cellulolytic organism. For both hay and silage with various supplements the typical cellulolytic bacteria differed from North American studies indicating that the difference most likely was related to the type of forage. Our studies signify not only differences between North America and Sweden but are notable also because they indicate a difference in rumen metabolism between hay and silage diets. Thus one possible explanation to production differences between hay and silage diets may be in the bacterial population. *B. Fibrisolvens* is only weakly cellulolytic and the shift in populations to the *Ruminococcus sp.* which are stronger cellulolytic bateria might be significant in fiber degradation and end-product formation. Other noteworthy observations are the absence of *Streptococcus bovis* which is also different from North American studies in which *S. bovis* is found to be important in rumens with high lactate levels (Russell and Hespell, 1981; Russell et al., 1979). Other differences include lactate metabolism where the major lactate utilising bacteria were *Sarcina sp.* (Van Gylswyk et al., 1992). If these ecologies

are typical only for Swedish cows or are related to the type of forage and are similar in other northern European cows is not possible to ascertain as comparative studies in other Scandinavian countries are lacking.

Conclusions

Ensiling is in many respects superior to hay making and has successively increased in importance as the method for conserving forage in many countries. Silage has in several studies given a higher animal production and permits a better utilization of grassland productivity than conservation as hay. Our studies on rumen ecology indicate differences in rumen metabolism between hay and silage, which could be one explanation for differences in production. Wilting has more or less consistently resulted in an increased intake, while effects on production has been variable. However experimental results indicate that the nutritive value is not affected if wilting is performed under good weather conditions and with a proper technique.

Silage from grasses grown in northern climates are characterised by a high fiber degradability and will give a high proportion of butyrate and a relatively low content of propionate in the rumen. The high rate of fiber degradation should have a positive effect on intake and might in that respect be as important as the chemical composition of the silage.

Effects of composition of the concentrate fed together with silage has varied between different experiments. The most consistent effect has been found with protein content in the supplement. The positive response has often been explained by an increased fiber degradation. However experimental data are not unequivocal and it is possible that different mechanisms are involved depending on the fermentation quality of the silage.

In future research the functional characteristics of the silage must be better defined and included in the evaluation of the results.

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Can HACCP Principles be Applied for Silage Safety?

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Introduction

Silage-making is a convenient method of preserving different varieties of crops and by-products for animal feed purposes. The basic technology relies on a rapid lactic acid fermentation in sufficient concentrations which in combination with anaerobicity suppresses pathogenic and spoilage organisms and preserves the feed until needed.

Though simple in theory, many factors exist which affect the safety and quality of silage. An uncontrolled growth of micro-organisms can affect animal health and cause nutritional losses, silage heating and accumulation of organisms which affect safety at a later stage and reduce the shelf-life of food produced from milk and meat from silage-fed animals. Factors affecting silage fermentation and storage include the composition of the crop, harvest, wilting, additives and silage system including loading, pressing and covering.

Information on managerial options to be taken in order to improve silage safety is not systematic. Precautions to be taken are usually based on technical treatments and on the application of different additives.

This overview aims to inform on matters regarding safe food production according to Hazard Analysis and Critical Control Points (HACCP) principles and to relate these to silage products.

Food safety aspects

Risk Analysis and Food Safety

Health hazards caused by food-borne micro-organisms are regarded as unacceptably high in developed and developing countries. International organisations such as FAO/WHO, Codex Alimentarus and EU have taken different initiatives in order to improve food safety. Precautions taken in order to ensure public health protection have to be based on Risk Analysis, which is widely recognised as the fundamental methodology underlying the development of

food safety. It is composed of three separate but integrated elements, namely Risk Assessment, Risk Management and Risk Communication (Anon. 1995). Risk Assessment is a key element in assuring that sound science is used to underpin management decisions on potential options (Anon. 1997a, Anon. 1998). Data for the Risk Assessment process are obtained from epidemiological sources, the medical and veterinary medical fields and from food science. The scientific steps in the evaluation are hazard identification, exposure assessment, hazard characterisation and risk characterisation. The outcome of the Risk Assessment process may include a ranking of the hazards. Risk Management is the process of weighting policy alternatives in the light of the results from the Risk Assessment and, if required, selecting and implementing appropriate control options, including regulatory measures. Risk Communication is the interaction between all interested parties, including the consumer, on matters related to the risk. It is generally accepted that transparency should be applied in all steps of the Risk Analysis process.

The Risk Analysis process helps the regulatory administrations and also other interested parties to focus on the major causes of problems and on options to be taken in order to control these problems.

HACCP and Food Safety

Traditional applications of Good Management Practices (GMP), Good Hygienic Practices (GHP) and Good Agricultural Practices (GAP) are used for production of safe and wholesome food products. These practices include measures to be taken in order to produce food with an acceptable level of hygiene in the end-product.

The term HACCP (Hazard Analysis and Critical Control Points) has been established in recommendations from EU and Codex to be applied for safe food production (Anon. 1997b). HACCP was initially formulated in the NASA space programme of 1971 for providing food for the astronauts (Anon. 1997b).

The HACCP system is a scientific and systematic way to control food safety and it focuses primarily on prevention rather than relying on end-product testing. It can be applied throughout the food chain from primary production to the consumer. The application of HACCP clearly defines the responsibility of the producers and the retail system for the safety of food. The HACCP system consists of seven principles including Hazard Analysis, determination of Critical Control Points and establishment of critical limits. The Hazard Analysis has to identify hazards which need to be eliminated, reduced to an acceptable level or controlled at an accept-

able level. In many cases, acceptable limits, which have to be met by the producer, are set on microbial contaminants. Microbial criteria used for these limits have usually been established by legal authorities.

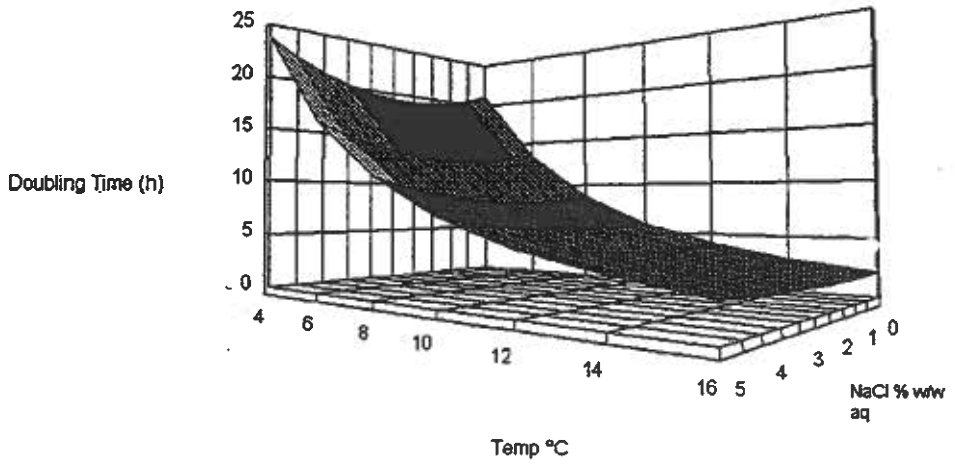
Control measures for each hazard have to be established and points for its control have to be defined. Critical limits for each Critical Control Point have to be set. Criteria used for critical limits include measurement of temperature, time, pH, A_w and additives.

In general, HACCP is intended for food safety but the concept can also be applied to other aspects of food quality. The application of HACCP at farm level has been suggested in a report by Newman (1997).

Predictive modelling

Predictive microbiology is a relative new discipline in work on food safety. By the use of data simulation, growth parameters are used as variables in relation to microbial growth for a fast and convenient system for the estimation of safety and shelf-life. Systems have been established and applied within EU and USA for calculation of microbial growth under different conditions (Overview, 1997). Models exist for organisms such as Salmonella, *Clostridium botulinum, Listeria monocytogenes* and *Campylobacter*. Growth parameters in the models concern temperature, atmosphere, pH, acid and salt content and A_w (Figure 1). These models have to be validated through practical trials and can be used for estimation of safety and shelf-life.

Figure 1. Predictive model indicating doubling time for *Listeria monocytogenes* in relation to temperature and concentration of NaCl.



Microbial hazards in silage

Considerable numbers of micro-organisms occur on fresh forage used for silage-making. A proportion of this flora is epiphytic, while others exist on the plant surface due to contamination with organisms originating in the soil or in manure used for fertilisation. Some detrimental micro-organisms such as clostridia, moulds and yeasts enter the silage crop through contamination with residual material from previous silage products. However, only a minor number of the organisms contaminating the fresh crop are either involved in silage fermentation or become a hazard.

Microbial activities restricting the quality of silage or known to cause health hazards occur during three different phases: (i) the fermentation phase, (ii) the storage phase, and (iii) the out-take and feeding phase. The accumulation of these organisms is either the result of a slow and incomplete fermentation or of aerobic deterioration during storage. The initiation of some of these activities may occur in the field (Weinberg and Muck, 1996).

Accumulation of pathogens

Toxic metabolites, infectious organisms and allergenic spores are health hazards associated with silage making (Woolford, 1990; Lindgren, 1991; McDonald et al., 1991). Microbial toxins are of either bacterial or fungal origin. The outer membrane of G^- bacteria contains the endotoxin Lipid A. Whether this toxin is a hazard during feeding of ruminants has not been demonstrated. Injection of it will cause well-defined symptoms such as fever, reduced rumen function and increased production of prostaglandin (Östling and Lindgren, 1991).

The neurotoxin produced by *Clostridium botulinum* is another example of a bacterial toxin known to cause problems for silage-fed animals (McDonald et al., 1991). Mycotoxins like Aflatoxins, Ochratoxins, Trichothcenes and Zearalenone produced by the genera *Aspergillus, Penicillium* and *Fusarium* have been observed in silage. The biological effects of these toxins can be neurotoxic, oestrogenic, hepatotoxic, nephrotoxic and carcinogenic (Oldenburg, 1991).

Infectious bacteria such as *Listeria monocytogenes* and strains of *E coli* and *Klebsiella pneumonie* can survive and grow under bad silage conditions. The enterobacteria are involved in intestinal disorders and mastitis and strains of *L. monocytogenes* are responsible for a number of diseases in animals including spontaneous abortion (Fenlon et al., 1989; McDonald et al., 1991; Grant et al.. 1995).

Spores

Endospores of saccharolytic and proteolytic clostridia can develop under bad silage conditions and spores of the saccharolytic *C. tyrobutyricum* are regarded as the major spoiler of hard cheese (Bergère and Accolas, 1985). These spores accumulate in silage and enter the milk via faecal contamination of udders. The spoilage of cheeses is characterised by blowing due to the production of hydrogen gas and to aromatic changes caused by butyric acid . .

Microbial heating

The initial heating of silage during loading is caused by an enzymatic aerobic respiration of sugar in the crop in the presence of oxygen. This initial temperature increase is expected to be restricted, since the quantity of oxygen entering with the crop is low (Table 1). However, the initial heating is often much higher than the 1-3 °C reported in the table. This indicates an un-controlled supply of oxygen due to slow loading or due to transportation of the crop into the silo with high pressure air.

Microbial heating is common during storage and feeding. By assimilating sugars or lactate during storage in non-airtight silos, yeasts such as *Hansenula, Candida* and *Saccharomyces* increase the temperature. Above 40-50 C° the yeasts are inactivated and there is a shift in the flora towards strains of the genus *Bacillus* with which temperatures above 80 C° can be reached (Lindgren et al.,1985; Jonsson, 1989).

Table 1. Theoretical calculation of respiratory heating during silage loading in relation to DM content. The calculation is based on $1m^3$ silage.

DM $g\ kg^{-1}$	*Estimated content per* m^3		*Amount of oxygen*		*Theoretical calculation of amount of energy released*	*Theoretical calculation of temperature increase*
	Water	*Air*	*Litres*	*Moles*		
200	400 l	500 l	90 l	4	457 kcal	1.1 °C
300	280 l	620 l	112 l	5	571 kcal	2.0 °C
400	208 l	700 l	126 l	5.6	640 kcal	3.2 °C

Lactate degradation

Aerobic assimilation of lactate in the silage by a variety of organisms such as moulds, yeasts and *Bacillus* reduces the conservation potential (Lindgren et al., 1985) and acetic acid bacteria can aerobically convert lactate to acetate (Spoelstra et al., 1987). This lays the basis for storage instability. Lactate can also be anaerobically degraded to butyric acid by the activity of *Clostridium tyrobutyricum* (McDonald et al., 1991) and to acetic acid in a secondary metabolism by *Lactobacillus plantarum* (Lindgren et al., 1990).

Changes in chemical composition

Biochemical processes occurring in the silage crop from harvest and further on during storage and feeding will change the composition of the forage. Plant enzymes affect the initial respiratory activity, proteolytic enzymes initiate a degradation of proteins and polysaccharide degrading enzymes attack cellulose, hemicellulose and fructans (Henderson, 1991). This enzymatic degradation in combination with microbial activities by lactic acid bacteria, clostridia, enterobacteria, Bacillus, yeasts and moulds will cause considerable changes in the composition of the forage. Microbial end-products such as ammonia, amines, organic acids, alcohol, esters, manitol, peptides etc. affect the taste and digestibility of the silage. Together with the heat production and effluent losses, these activities reduce the content of energy. Zimmer (1980) calculated energy losses due to microbial activities and plant respiration. He reported that the main losses are caused by aerobic deterioration during storage and unloading. Unfavourable conditions during these periods may result in a loss of up to 25% of the energy.

Accumulation of end-products is often used as a quality indicator for silage. Lactate is the main indicator of a suitable fermentation process. The importance of acetic acid can be disputed. This substance has a good antagonistic effect against moulds and yeasts. The acid is produced by heterofermenters in the initial phase and during an anaerobic or aerobic degradation of lactate at a later stage. Butyric acid is an indicator of the growth of saccharolytic clostridia. However, the number of spores is usually not well correlated to the content of the acid (Jonsson, 1989). Accumulation of ammonia is the main characteristic of silage spoilage. Ammonia is the end product of microbial deamination of amino-acids. It is mainly Enterobacteriaceae and proteolytic clostridia which are involved in this process (McDonald et al., 1991).

Control factors in silage

The major antagonism in silage against detrimental organisms is caused by a rapid acid production in combination with anaerobic conditions (McDonald et al. 1991). A reduction in water activity (A_w) may have an additional synergistic effect on the antibiosis (Seale, 1986).

The production of fermentation end-products is mainly caused by lactic acid bacteria with an accumulation of lactate and, to a smaller extent, acetate. However, end-products such as carbon dioxide, hydrogen peroxide, diacetyl and bacteriocins may be involved (Lindgren and Dobrogosz, 1990). A prerequisite for the initiation of a proper fermentation is that the silage crop contains a sufficient amount of lactic acid bacteria in combination with fermentable sugars and other nutrients and that the temperature is suitable.

Slow and incomplete fermentation in silage favours the growth of enterobacteria and clostridia (Woolford, 1990). The enterobacteria compete with LAB for carbohydrates thereby causing a decrease in lactic acid. If the carbohydrate content is depleted, enterobacteria and proteolytic clostridia initiate ammonia production. This seriously compromises the quality of the silage (Seale, 1986).

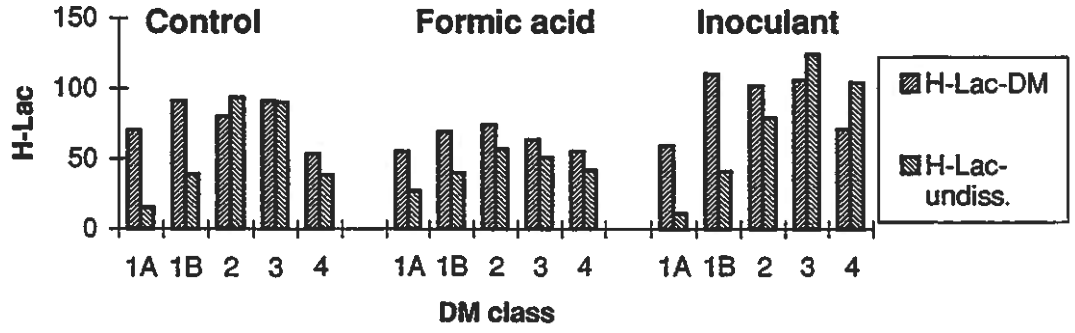
In silage literature, pH is still used as the major quality criterion indicating the preservation potential. However, a more realistic indicator is the concentration of undissociated organic acids. Many weak acids in their undissociated form can penetrate the cell membrane and accumulate within the cell cytoplasm. The undissociated molecules dissociate and release protons which lower the pH. This activity affects the proton pump which regulates the internal pH. Intracellular acidification then results in a loss of viability or cell destruction (Kashket,1987).

The concentration of acids in silage is expressed per kilo dry matter (DM). With this expression there is no understanding of the potential of preservation caused by the acid. This can easily be exemplified by comparing a well-preserved silage to the same silage to which 20% extra water has been added. This addition does not change the relation of organic acids to DM, but acid concentration in the water phase is reduced. Subsequently the antimicrobial activity and the storage stability are diminished in the silage.

Usually *L. plantarum* produces concentrations of undissociated lactic acid around 100mmol l^{-1} and calculation of the final concentration of undissociated lactic acid l^{-1} H_2O in silage from

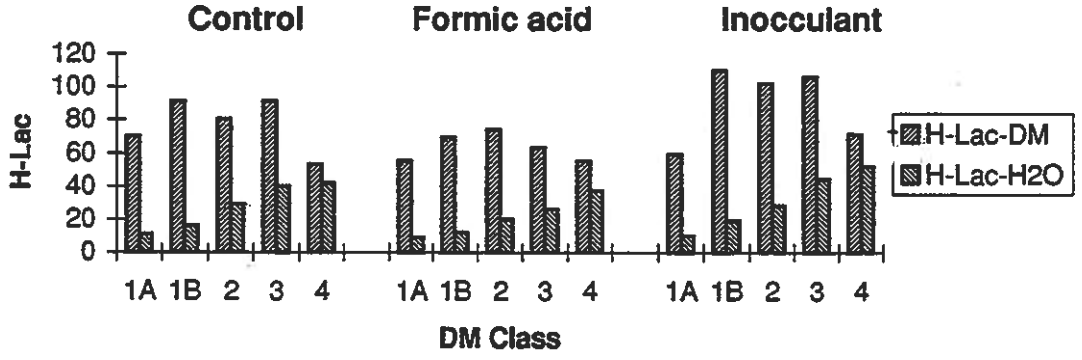
data presented at the Eurobac conference (1990) indicates levels varying from 15- 124 mmol (Figure 2).

Figure 2. Calculation of concentration of total lactic acid (g kg^{-1} DM) and undissociated lactic acid (mmol l^{-1} H_2O) from the Eurobac data (Eurobac Conference, 1990). 1A, DM<180g kg^{-1},WSC<15g kg^{-1}; 1B, DM<180g kg^{-1},WSC>15g kg^{-1}; 2, DM 180 -250g kg^{-1}; 3, DM 250-350g kg^{-1}; 4, DM> 350g kg^{-1}. (Number of samples for each staple 10< n >40).



The lowest levels of undissociated acid were observed at low DM values. Addition of formic acid also reduced the concentration of undissociated lactic acid at all DM contents. The content of lactic acid expressed as a part of the DM does not indicate reliable differences in the fermentation pattern and this content varied between 55-120 g kg^{-1}. The result also shows the importance of the WSC content for the fermentation process at low DM values.

Figure 3. Content of lactic acid related to DM (g kg^{-1}) respective water phase (g l^{-1}). For legends see Fig.2



A comparison of the content of lactic acid in the DM and water phases, respectively, indicates limited differences in the amount of lactic acid in DM between the different DM classes. Calculation of lactic acid in the water phase indicates a constant increase between the different DM classes (Fig. 3).

The sensitivity to the acid condition varies among micro-organisms (Baird-Parker, 1980). Enterobacteria, clostridia and *Listeria* were found to be inhibited at values ten times lower than the level produced by the lactic acid organisms (Östling and Lindgren, 1993). The inhibitory activity is usual constant for the undissociated acid within a pH interval from 4.0-5.5. Yeasts and moulds are not sensitive to the acid conditions existing in silage and maintenance of anaerobic conditions in the silo is the most important factor in the prevention of their growth. Members of both groups can also use lactate in an aerobic assimilation, which reduces storage properties (Lindgren et al. 1985). Acetic acid is reported to be more inhibitory to yeasts and moulds than lactic acid (Baird-Parker, 1980).

Wilting of silage crops has conflicting effects on the success of silage-making. Wilting reduces the amount of water to be transported to the silo and the production of effluents in the silo (Bosma, 1991). Wilting is also important in order to increase the concentration of WSC in the liquid phase of the silage crop. Subsequently the preserving potential is higher due to the increase in the concentration of acids in this phase. Availability of water for microbial growth is related to water activity (A_w) and wilting has a selective effect on the growth of contaminating micro-organisms since the demand of water differs among micro-organisms (Lindgren, 1991). A decrease in A_w retards the competition from the Enterobacteria and the sporulation of clostridia (Jonsson, 1989). The extrapolation of DM content to A_w has been demonstrated by Weissbach (1996). Yeasts and moulds can in general grow at water activities where bacteria do not grow and wilting improves the growth conditions for these organisms. Wilting above the press water level will, however, restrict the distribution of sugars and acids in the water phase and facilitate the transport of oxygen through the material.

Additives

Additives are used to improve silage safety. They include selective inhibitors such as acids and salts, and stimulants such as inoculants, fermentable carbohydrates and enzymes. Combinations of the two types also exist. Reviews on the effect of additives on the silage process have been

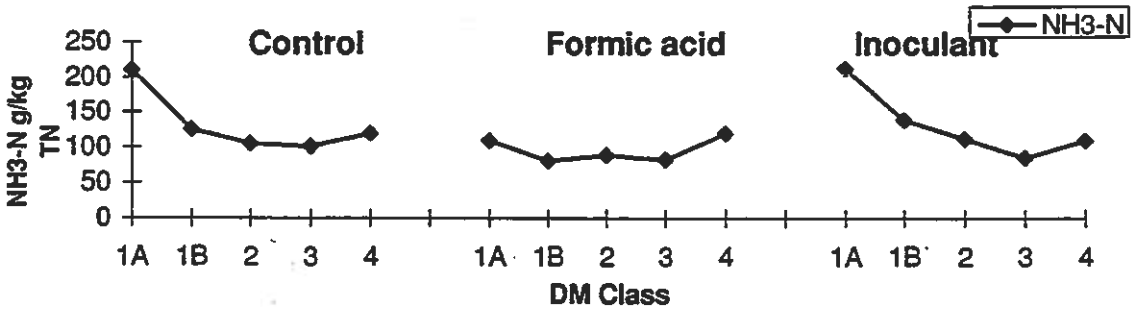
reported during recent decades (McDonald,1981; Seale, 1986; Spoelstra, 1991; Henderson, 1993; Weissbach, 1996; Weinberg and Muck, 1996). In general each additive has a specific impact on the process. Inorganic acids were first to be used to lower the initial pH and thereby retard microbial competitors to the lactic flora. Today, formic acid is frequently used for this purpose. By combining hexamine and nitrite, a specific clostridium inhibitory effect in silage is obtained. Inoculants are added in order to supplement the crop with a fast-growing lactic flora for a rapid fermentation. Pahlow and Honig (1993) reported that bacterial inoculants are most successful at DM intervals between 20-35% DM. Similar observations could be found in the data collected for the Eurobac Conference (Eurobac, 1990).

At low DM levels, the content of fermentable carbohydrates is low and addition of enzymes and sugars increases this content. The rational effect of addition of acids to silage with DM levels above 35% can be questioned. At such levels, the distribution of acid in the silage is not effective and the acid may stimulate mycotoxin production at high DM levels (Pettersson et al., 1989).

The information on experiments comparing additives is voluminous but not systematically developed as seen in the proceedings of the International Silage conferences in Dublin and Aberystwyth.

There is a need for the development of predictive models. At the Eurobac meeting Zimmer (1990), summarised the results of treatments with formic acid and inoculants and compared the results to controls for different DM classes. The results were obtained from a number of different laboratories in Europe. The compliation of the results indicated an improvement in

Figure 4. Variation in ammonium content in relation to DM content and treatment. For legends see Figure 2.



silage quality expressed as ammonium as a result of addition of formic acid at low DM densities (Figure 4). The results also showed a general improvement in quality at higher DM values. At DM values below 180g $kg^{-1,}$ the initial content of WSC was important for the quality in the control silage and the inoculated silage (Figures 2 and 4). Increasing the DM content above 35 g kg^{-1} slightly reduced the ammonia content for all treatments.

Aerobic instability is a fundamental problem during storage and feeding. No efficient additives have been reported as having an important impact on this problem. Addition of benzoic and propionic acids may to some extent improve the stability. Most results reported for the traditional inoculants are negative. Some well-fermented silage prepared by inoculation and with high levels of lactic acid and remaining sugars seem on the contrary to be subjected to a fast aerobic spoilage (Weinberg and Muck, 1996). Recent results show an increase in the content of acetic acid after inoculation with *Lactobacillus buchneri* which improved the storage stability (Dreihuis et al., 1996). Inoculation with Lactobacillus species has also been reported to inhibit the growth and aflatoxin production of *Aspergillus flavus* (Gourma and Bullerman, 1995).

Quality parameters

Criteria have been established to indicate the quality of silage. These include estimation of pH, ammonia, dry matter (DM), water soluble carbohydrates (WSC), lactic acid, butyric acid and acetic acid, and microbial testing for enterobacteria, spores of saccharolytic clostridia and lactic acid bacteria, yeasts and moulds. These criteria indicate different aspects of deterioration of the silage. The importance of criteria is to provide values indicating safe storage or acceptable levels of spoilage and/or accumulated hazardous micro-organisms. These values are necessary for the practical development of silage technology in respect of different crops, silage conditions, silage systems and additives. If HACCP principles are intended to be established, criteria give important information on conditions to be achieved. Since the identity of silage is strongly diversified as a matter of crop composition, conditions during silage making, silage system and additives, it is difficult to set general critical limits.

The relevance of some criteria commonly used in silage literature might be disputed. This refers to pH as an indicator of preserving conditions, lactic acid in DM as an indicator of the fermentation activity and WSC in DM as an indicator of the probability for a safe fermentation. The concentration of butyric acid and the content of clostridia spores are not well correlated. A

more reliable indicator of the quality would be to express the concentration of WSC and lactic acid in the liquid phase. Microbial activities affecting the quality occur in the water phase. The preservation potential in a silage relies on the content of undissociated acids and to a minor degree on the pH. Spores of clostridia exist in silage even at low concentrations of butyric acid and are absent in silage with a high butyric acid content.

HACCP for silage

It is evident from the above overview that silage-making is subjected to detrimental processes affecting animal health, the content of nutrition and energy and the quality and safety of foods produced from silage-fed animals. The total economic and health-related consequences of these effects are difficult to estimate.

The basis for safe silage-making has to rely on the application of Good Agricultural Practices (GAP). These practices depend on precautions to be taken during fertilisation of the growing crops with manure, preventing contamination of the crop with soil during cutting , wilting of low density crops, fast loading and precautions during loading to avoid contamination of the fresh crop with material in and around the silo and the use of silo systems which do not allow the entry of oxygen.

Compared to production of safe food, the complexity of silage-making is extensive and, even with the application of GAP, conditions exist which strongly interfere with the production of safe silage. Some of these conditions depend on the weather, low WSC, low level of a fermentable flora etc. These conditions are beyond the control of the silage producer.

A general statement among stake-holders is that food safety starts at the farm and the expression "from stable to table" is linked to food safety issues. Problems caused as a result of feeding animals with bad quality silage will affect animal health, as well as food safety. The application of HACCP principles in combination with GAP may be a way to improve silage safety. Arimi et al. (1997) stressed the importance of farm-based HACCP programmes for controlling Listeria.

A prerequisite for the introduction of a HACCP-based control system is the identification of normally-occurring hazards and critical control points for these hazards. In this respect the use of additives is the most reliable way to achieve control at a specific step. However, existing information does not present a summarised comprehensive picture of the effect of silage conditions and additives on the outcome of a silage. Available data on effects are fragmented and not systematically expressed. Before HACCP plans are elaborated for the silage process, pre-

dictive models have to be established, taking into account the diversity of factors affecting the outcome. Models have to summarise existing knowledge and indicate the probable outcome of precautions to be taken.

Conclusion

Silage-making relies on a complex system where different factors may interfere.

Good Agricultural Practices have to be applied as the basis for safe production.

Application of principles for HACCP will give an additional tool to improve silage safety.

For HACCP-based control systems for silage making, the silage process has to be expressed in predictive models which indicate the outcome of precautions taken in respect of quality criteria.

There seems to be a need to revise some of the criteria used for indicating silage quality.

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Silage and health

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1. Summary

The process of preserving crops by fermentation in silos is under the control of the farmer to a much lesser degree compared to the level of control by the manufacturer over the production of other fermented foods, such as cheese and yoghurt. Additives designed to direct the extent and pattern of the fermentation are relatively unpopular in most countries, and their use is not guaranteed to remove the risk of undesirable components in silage. Hazards to health associated with silage fall into three categories: i) undesirable micro-organisms e.g. *Listeria, enterobacteria, clostridia* and moulds; ii) undesirable chemicals, e.g. toxins, and iii) excess acidity and other metabolic disorders. In some regions of Europe, the production of silage is discouraged or prohibited because of the risk of undesirable microbes, but the principal risk in these areas is that of the secondary fermentation of cheese made from milk contaminated by bacterial spores from silage, rather than direct hazards to human or to animal health. With the possible exception of high dry matter silage conserved in large bales, respiratory hazards to humans and to animals from moulds and their spores generally are less from silage than hay. Much less is known about the epidemiology of diseases that may be linked to undesirable chemicals and excess acidity in silage. Therefore, research is needed to define epidemiologically and mechanistically the risks to animal and human health from silages contaminated with pathogenic bacteria and mycotoxins, and to understand more completely the relationships between the physical and chemical compositions of silage and metabolic disorders in animals.

2. Introduction

Links between the food given to animals, their health, and the real and potential risks to human health have increased in importance recently, particularly in the United Kingdom following the epidemic of bovine spongiform encephalopathy (BSE).

Consumers require reassurance that the food they consume is free of contamination by hazardous microbes or chemicals. Recent feedingstuffs legislation in the UK (Marsden and Nelson, 1999) was introduced as a direct result of the BSE epidemic and may herald further legislation in Europe and elsewhere to reduce the risk of the contamination of human food, particularly that derived from livestock.

Silage making involves harvesting grass and other forage crops by mechanical harvester, usually with considerable reduction in the size of particles which accelerates the release of plant cell contents. The harvested materials are then placed in a silo, or compacted in bales, which is sealed to produce anaerobic conditions. The silo is opened after several weeks or months and the silage is removed for use as food for ruminant animals. Preservation of the crop is achieved by the conversion of plant water-soluble carbohydrates (WSC) glucose, fructose, fructans and sucrose to organic acids, principally lactic acid. There is also a variable degree of degradation of proteins to

amino acids and other nitrogenous compounds. The biochemical and microbiological reactions in the silo have been reviewed in detail by Woolford (1984) and by McDonald *et al.* (1991).

The process of preserving crops by fermentation in silos is under the control of the farmer to a much lesser degree compared to the level of control by the manufacturer over the production of other fermented foods, such as cheese and yoghurt. Additives designed to direct the extent and pattern of the fermentation are relatively unpopular in most countries (Wilkinson, et al., 1996), and their use is not guaranteed to remove the risk of undesirable components in silage. Surveys have revealed a huge range of preservation quality in silages, with significant proportions of unstable and badly preserved silages (Weissbach, 1996, Haigh, 1996 a,b). Clearly, there is room for improvement, both in the application of existing knowledge and also in our understanding of the severity of the biological and economic problems that can arise when silage preservation is of low quality.

The total quantity of silage produced annually in Western Europe appears to be relatively static (Wilkinson, et al., 1996), but underlying changes in the technology of silage production may have significant implications for both human and animal health. For example, on the negative side, the increased popularity of forage maize may lead to greater incidence of mycotoxicoses (Lepon, 1990; Oldenburg, 1991); more baled silage may lead to a greater incidence of listeriosis (Fenlon, 1988); and reduced use of effective additives may increase the incidence of poorly preserved silages which present greater risk of growth of potentially pathogenic enterobacteria (Ostling and Lindgren, 1995) and greater risk of metabolic disorders from ammonia, amines, and other degraded nitrogenous compounds (Howie, 1988). Greater use of silage in the nutrition of horses may be related to a increase in the number of clinical cases of botulism (Roberts, 1988; Whitlock and Buckley, 1997).

On the positive side, higher dry matter silages produced by the implementation of techniques for the rapid removal of water from leafy forage crops in the field prior to ensiling (Wright *et al.*, 1997; Wilkinson *et al.*, 1999) may be associated with a reduction in the incidence of lameness in dairy cattle (Clarkson, *et al.*, 1993; Joules, 1993). Reduced levels of fertiliser nitrogen may be reflected in reduced risk of nitrate poisoning (Hill, 1999) and of reproductive problems associated with elevated levels of ammonia in blood (Robinson, 1996, McEvoy, *et al.*, 1997).

In this paper the potential hazards to animal and human health from the ensilage process are evaluated, and recommendations are made for future research into silage and health. Hazards to health associated with silage fall into three categories: i) undesirable micro-organisms, e.g., *Listeria, enterobacteria, clostridia* and moulds; ii) undesirable chemicals, e.g., toxins, and iii) excess acidity and other metabolic disorders. These three categories are discussed in the following sections.

3. Undesirable micro-organisms

Undesirable micro-organisms can enter the silo by several routes, including soil and in particular livestock waste which comtains high numbers of potential pathogens (Mawdsley *et al.*, 1995). As livestock waste is applied to grassland destined for ensiling, this process provides a route for the transfer of pathogens to animals, if they survive on herbage, enter the silo with the harvested crop and survive the period of ensilage.

Most research on undesirabel micro-organisms in silage has been focussed on bacteria and fungi, as described below. But recently Merry *et al.* (1997) highlighted the fact that protozoal pathogens such as *Cryptosporidium parvum*, which is sometimes detected in livestock waste, can survive fairly adverse conditions in the silo. This factor has implications for both human and animal health as the

organism forms a very resistant oocyst which is likely to survive on forage for considerable periods of time.

3.1 Enterobacteria

The role of enterobacteria, otherwise known as coliform organisms, in the silage process has been reviewed by Woolford (1984), McDonald *et al.* (1991), and Lindgren (1991). Spoelstra (1987) and McDonald *et al.* (1991) describe the role of enterobacteria in the reduction of nitrate to ammonia via nitrite in the early phase of the ensiling process, with the production of nitrous oxide. The fermentation of glucose by enterobacteria produces a range of end products including lactic acid, formic acid, acetic acid, ethanol and 2,3 butanediol (McDonald *et al.*, 1991).

The most important organism in this group from the viewpoint of the risk to health is *Escherichia coli* species, which can cause acute diarrhoea and death (Cooke, 1993). An important risk factor in the contamination of crops with *E. coli* and related enterobacteria is the application of animal manures to grassland prior to harvesting crops for ensilage (Davies *et al.*, 1996). Whether or not the enterobacteria dominate the epiphytic microflora on the crop at harvest is probably irrelevant. Thus, Rammer *et al.* (1996) were unable to demonstrate that the repeated application of cattle manure as slurry to a clover/grass ley had a major impact on the fermentation quality of silages made from the treated pasture. In earlier work, they found that applications of farmyard manure as solid material resulted in reduced silage quality, with elevated pH, increased concentrations of ammonia -N, and butyric acid, with high numbers of *Bacillus* and *Clostridium* spores (Rammer *et al.*, 1994).

Ostling and Lindgren (1995) studied the effect of inoculation of grass with enterobacteria on fermentation and production of endotoxin. They found that additions of relatively large numbers of enterobacteria (10^6 and 10^8 g^{-1} fresh matter) resulted in a slower rate of lactic acid production and an increase in ammonia-N in the silage. Endotoxin concentration was 6 to 7 μg g^{-1} fresh matter, with no change in concentration during 125 days of ensilement. An increase in the number of gram negative bacteria from 10^7 to 10^9 during storage overnight resulted in an increase in endotoxin from 6 to 97μg g^{-1} fresh matter (Ostling, unpublished data).

The most important factors likely to determine the proliferation of enterobacteria in silage are the conditions within the silo from the time of entry of the crop. If conditions are favourable to the growth of enterobacteria, then animals consuming the silage may suffer from intestinal disorders and mastitis (Lindgren, 1991). Slow fermentations favour the growth of enterobacteria (Woolford, 1984). A rapid decrease in the pH of ensiled crops has an inhibitory effect on the number of enterobacteria, which compete with the lactic acid bacteria for available carbohydrate in the early phase of the ensiling process and yet produce less acid (Lindgren, 1991, McDonald *et al.*, 1991, Mawdsley *et al.*, 1995). The minimum inhibitory concentration for enterobacteria is between 6 and 10 mM of undissociated lactic acid, some 10 times lower than the total quantity of undissociated lactic acid (100 mM) in a normal silage of pH 4.2 containing 22 g total lactic acid/kg fresh weight (Lindgren, 1991).

The key factor determining the extent to which enterobacteria are likely to develop in silages is the **rate** of lactic acid production in relation to the amount and availability of WSC in the crop fresh weight. There is no information in the literature to confirm this point, but circumstantial evidence suggests that the development of enterobacteria in silages may well be common and extensive. Because lactic acid is the predominant component of crop acidification in the primary phase of ensilage (Bolsen, *et al.*, 1995), some indication as to the probability of enterobacterial development can be gained from studying the rate of acidification in crops during the first 72 hours of ensilage. Weissbach (1996) investigated this point in relation to the rate of decrease in the pH of more than

100 different crops in the first 72 hours of ensiling. Crop dry matter (DM) concentrations ranged from 150 to 600 g/kg. The mean decrease in pH was 0.8 (s.e.±0.7), with a range from zero to 2.2 units. This work was undertaken at a relatively high temperature (25°C); at lower temperatures, the rate of decrease in pH would be expected to be significantly slower. Leguminous crops have a higher buffering capacity than graminaceous crops and as a result the rate of decrease in pH is relatively slow in ensiled legumes (McDonald et al., 1991). Merry (personal communication) observed that enterobacteria persisted for more than 14 days in baled red clover silage.

The pathogenic coliform of most concern is *E. coli* Ø157:H7. Research is needed to establish the factors that determine the growth and survival of this organism in silage, so that farmers can be told how to avoid the predisposing conditions by altering crop and silage management.

3.2 *Listeria*

Listeria monocytogenes is a common bacterium in soil and vegetation that causes disease in a wide range of animals and also in humans, usually those with suppressed immunocompetence (Fenlon, 1988; McDonald *et al.*, 1991). Most of the research on *Listeria* in silage has been concentrated on baled silage because of the association of listeriosis with mouldy baled silage (Fenlon, 1988).

It is important to remember that about 0.3 of the total weight of a large round bale occurs in the outer 10 cm . This layer is usually the wettest because of condensation of moisture on the polyethylene wrapping; it is also at the greatest risk of air ingress from incomplete sealing at harvest or via holes in the wrapping that can occur subsequently. Fenlon (1988) described the aerated outer layer of round bales as a selective culture medium for *Listeria.*

The characteristics of contaminated silage implicated in confirmed outbreaks of listeriosis are i) a very high pH and ii) a high population of *L.monocytogenes*, which can exceed 10^6 per gram of fresh silage (Table 1).

Table 1. Characteristics of silages given to sheep suffering from confirmed signs of listeriosis.
From Fenlon (1988)

Farm	L.monocytogenes.gram silage fresh weight^{-1}	pH of contaminated silage
A	1.5×10^2	5.5 to 9.1
B	2.0×10^3	7.8 to 9.1
C (i)	$>10^6$	7.3 to 7.6
C (ii)	1.0×10^3	8.6 to 8.7

The critical factor in the growth of *L. monocytogenes* is the presence of oxygen. Thus, a close association between aerobically-deteriorated wet silage and the growth of *L. monocytogenes* might be expected. However, Fenlon and Wilson (1991) showed that growth of *Listeria innocua* (non pathogenic) could occur in the laboratory under anaerobic conditions if the pH was 5.0 or above and water activity was 0.98 or above. Thus, the outermost layer of the bale, and also the layer of silage just below the top sheet of bunker and clamp silos might contain significant populations of *Listeria,* even if oxygen penetration into the material is limited> This is especially likely if the crop is wet at harvest and if the sheet or wrap is heated subsequently to cause condensation of water at the surface silage/polyethylene interface.

The growth of *Listeria* in silage probably is inhibited below pH 5.0 (McDonald *et al.*, 1991). Undissociated lactic acid was particularly effective in inhibiting the growth of *L.monocytogenes* (Ostling and Lindgren, 1993). However, Fenlon and Wilson (1991) stated that *Listeria* can survive relatively low pH conditions for long periods of time, especially if trace levels of oxygen are present. These organisms can provide a focus for growth when conditions become more favourable; for example, when the silo is opened and the silage is oxygenated.

It is difficult to separate contaminated silage from uncontaminated material, especially in the case of baled material because it is put as the whole bale into a ring or rectangular feeder after unwrapping. Prevention of growth of *Listeria* is most likely to be effective if the DM content of the silage is relatively high and attention is paid to ensuring that the seal is complete and protected from damage throughout the storage period.

3.3 *Clostridia*

Research into the role of *Clostridiium* species in silage has been dominated by studies of their adverse effects on the preservation quality of silage. These anaerobic bacteria are responsible for the secondary fermentation of glucose and lactic acid to butyric acid by saccharolytic species such as *C. butyricum* and for proteolysis by species such as *C. perfringens* (see reviews by Woolford, 1984; McDonald *et al.*, 1991; Weissbach, 1996).

The direct effects of clostridial fermentations on animal and human health are less evident than those on silage compositional quality. The combined effects of loss of digestible energy during the secondary fermentation of lactic acid to butyric acid (McDonald, *et al.*, 1973), coupled with reduced intake by the animal (Wilkins *et al.*, 1971; Demarquilly, 1973; Steen *et al.*, 1995) might result in clinical signs of acetonaemia in high-yielding dairy cows in early lactation. (Howie, 1988).

Proteolytic clostridial activity in silage can result in extensive degradation of plant proteins to ammonia (McDonald *et al.*, 1991), which can have adverse consequences on animal health. Chamberlain and Choung (1993) suggested that elevated levels of ammonia in peripheral blood, resulting from absorption of ammonia from the rumen following meals of silage of high total nitrogen content, high ammonia nitrogen content, or both, might exceed the ability of the liver to detoxify ammonia via the urea cycle. The consequences of high blood ammonia on animal health are discussed in Section 4.2 below.

Clostridium botulinum is the organism of greatest concern with regard to health. Distinct types of botulinum toxin (Types B, C and D) are pathogenic to animals (Wilson and Miles, 1975; Roberts, 1988). It is surprising that relatively few incidents of botulism have been related directly to silage (Roberts, 1988), which suggests that if *C. botulinum* is present in the crop at the time of ensiling, the numbers are probably small, and its development, even in poorly preserved silage, is relatively slow.

The risk of botulism is increased considerably if the silage is contaminated with mammalian or avian remains, and botulism has been reported in cattle given silage made from pasture to which poultry manure had been applied (Smart *et al.*, 1987). However, the greatest risk of clinical botulism is probably in horses given contaminated baled silage (Ricketts *et al.*, 1984). Horses and other non-ruminants may be more liable to botulism than ruminants, possibly because of detoxification of botulinum toxins by the rumen microflora.

Very little work has been undertaken to establish the factors affecting the growth of *C. botulinum* in silage that is not contaminated with dead animals or birds. Given the ubiquitous nature of the organism, it could develop in wet silages in which the pH was maintained above 4.6 (Roberts, 1988).

3.4 Fungi

The growth of fungi in silages poses two hazards to health - first, a challenge from airborne spores to the sensitive mucosal surfaces of the lungs and respiratory passages, and second, metabolic disorders associated with the ingestion of mycotoxins. In this section, the effects of airborne spores on health are discussed; while the effects of mycotoxins on health are considered in Section 4.3.

Clarke (1988) reviewed the effects of mouldy hay and silage on animal health. The principal animal at risk of respiratory disease is the horse, though humans are also at risk of respiratory challenges if they are exposed continually to high airborne concentrations of spores, for example, when unloading roofed or tower silos by hand in restricted air spaces. Respiratory disease in the racehorse, where maximal lung capacity is crucial to successful running, can be disastrous. Silage products, based on carefully sealed mould-free minibales, have been developed especially for horses. These products probably are the first animal feeds to be sold with a mycological assessment and a certificate of good hygienic quality.

Aspergillus fumigatus, *Actinomycetes,* and *Penicillium* species are associated with mouldy hay that has undergone aeration and heating (Gregory *et al.*, 1963). Possibly, similar groups of organisms develop in mouldy silage once the pH has risen to 6 or above following the growth of yeasts and aerobic bacteria, which metabolise the acids in silage (Woolford, 1984). Recently, *Penicillium roqueforti* has been shown to be responsible for the aerobic deterioration of silage (Weissbach, 1996; Auerbach, *et al.*,1998). The fungus can develop in acidic conditions at low levels of oxygen and its spores can survive for long periods; it also can produce toxins including roquefortin C and patulin (see Section 4.3)..

When a group of 35 beef cattle was provided with mouldy grass haylage (600g DM kg^{-1}) as bedding, the animals became lame within 24 hours. The lameness was diagnosed as digital dermatitis (J. Hill, personal communication), indicating rapid infection of the feet by fungi in the haylage.

4. Undesirable chemicals

4.1 Excess nitrate

The role of nitrate in silage has been reviewed by Hill (1999) and by Weissbach *et al.* (1993). Although nitrate can have a positive effect on the fermentation, stimulating the production of acetic acid instead of ethanol in heterolactic fermentations and inhibiting the development of clostridia, excess nitrate can be detrimental to the fermentation process if large amounts are reduced fully in the silo to ammonia, which acts as an alkali to produce ammonium salts of the fermentation acids and reduces the rate of acidification.

A particular risk to health is the production of toxic oxides of nitrogen in the early phase of ensilage. The reduction of nitrate to nitric oxide (NO), a colourless gas, is followed by its oxidation on exposure to air to nitrogen dioxide (NO_2), a yellow-reddish-brown gas with an irritating odour. Nitric oxide and nitrogen dioxide react with water to form nitrous acid (HNO_2) and nitric acid (HNO_3), respectively. These gases and acids cause respiratory irritation by destroying the

membranes of the respiratory tract. The hazard, known as "silo-fillers' disease" has been recognised for many years (Lowry and Schuman, 1956). A survey in Minnesota, USA, revealed that 42% of 322 silages made in towers contained nitrogen dioxide in concentrations considered hazardous to human health (Scaletti *et al.*, 1960).

It is unusual for livestock to be exposed to oxides and acids of nitrogen, unless they are kept in confinement immediately adjacent to a silo. O'Kiely et al (1999) described one such situation in Ireland in which 10 calves and the farmer tending them suffered severe respiratory distress 24 hours after an adjacent unroofed bunker silo had been filled with wilted first-cut grass and sealed. The quick action of the farmer, who released the calves into an open yard as soon as symptoms of respiratory distress were experienced, averted a disaster. Six calves recovered quickly, but four required veterinary treatment for several days.

The ensiled crop had received 34,000 litres of cattle manure as slurry.ha^{-1} in February 1998, followed by 140 kg fertiliser nitrogen.ha^{-1} in two applications in mid-February and mid-March 1998. The herbage was ensiled on 24 May 1998. Samples of forage, taken 4 days after ensiling, were seen to be intensely yellow in colour and had the characteristics shown in Table 2. Apart from the very high crude protein content, the most unusual feature of the yellow-coloured forage was its extremely low pH, suggesting contamination with nitric acid, which had severely restricted the fermentation and most likely hydrolysed a significant proportion of potentially digestible dry matter. The authors concluded that if animals are housed close to large silos, they must be inspected very often in the first few days after the silo is filled.

Nitrate toxicity from silage is a potential risk to animal health. However, studies of the processes in the rumen, reviewed by Hill (1999), lead to the conclusion that the production of methaemoglobin in the presence of high concentrations of nitrate in the liver of ruminants given silage is unlikely, because i) the level of ingestion of nitrate from silage is likely to be relatively low and ii) nitrate probably is reduced in the rumen to ammonia before absorption into blood.

Table 2. **Characteristics of yellow forage, four days after ensiling, presumed to be contaminated with nitric acid.**
From O'Kiely et al. (1999)

Characteristic	Colour of ensiled forage	
	Normal (green)	Yellow
Dry matter (g.kg^{-1})	214	194
pH	4.3	1.1
Nitrate (mg.litre)	400	501
Crude protein (g.kg DM^{-1})	193	294
Ammonia-N (g.kg total N^{-1})	94	31
Lactic acid (g.kg DM^{-1})	66	9
Acetic acid (g.kg DM^{-1})	21	8
Propionic acid (g.kg DM^{-1})	3	3
Butyric acid (g.kg DM^{-1})	0.2	0.3
Ethanol (g.kg DM^{-1})	6	18
Buffering capacity (mEq.kg DM^{-1})	686	97
Digestibility of DM in vitro (g.kg^{-1})	700	573

4.2 Excess ammonia

Circumstantial evidence, reviewed by Ferguson and Chalupa (1989), Robinson (1996) and Chamberlain and Wilkinson (1996), points strongly to a negative effect of excessive amounts of rumen-degraded protein on fertility. The major challenge to animal health from excess ammonia in silage is to the liver, where the ammonia is detoxified to urea prior to its excretion in urine. If animals consume silage containing relatively high levels of ammonia together with other degraded nitrogenous compounds (e.g. amines, amino acids), then surges of absorbed ammonia probably will occur in the liver via portal blood. If the liver cannot fully metabolise the ammonia to urea, then elevated levels of ammonia are likely to occur in peripheral blood. McEvoy et al (1997) concluded that elevated levels of plasma ammonia were related directly to the death of developing embryos in sheep. Surprisingly, several of the embryos that survived grew into oversized lambs, suggesting that excess ammonia might upregulate embryo development. These results have clear implications for animals given diets containing high levels of silage of high nitrogen content, and highlight the importance of a) restricting the production of ammonia in the silo and b) balancing silages with adequate readily fermentable energy to maximise the conversion of ammonia to microbial protein in the rumen.

Howie (1988) suggested that excess ammonia in the rumen might result in elevated concentrations in the large intestine. In such situations, not all the ammonia in the rumen is absorbed through the rumen wall, and some leaves the rumen in the digesta. This ammonia could act as an irritant to the large intestine and result in more rapid bowel movement and looser faeces. More rapid passage of digesta through the intestines may also reduce the absorption of magnesium and other essential elements.

Excess ammonia in silage also might be linked to lameness in animals. If ammonia, or amines, or associated endotoxins are predisposing factors to inflammation, then a link to the laminitis syndrome in animals may be established. At present, the evidence is circumstantial. Bazeley and Pinsent (1984) reported an apparent link between acute laminitis in dairy cattle and relatively high concentrations of ammonia in the silages given to the affected animals. However, this link may, however, reflect reduced voluntary intake of poorly preserved silage and increased proportions of concentrates in the diet. Further epidemiological studies are needed to confirm a relationship between excess ammonia in silage and lameness in animals (see Section 6).

4.3 Mycotoxins

Mycotoxins are the products of fungal metabolism. They can be found in silage and any animal feed that has deteriorated during storage. Gotlieb (1997) stated that fungi with the capacity to produce toxins have three critical environmental requirements: a) temperature above freezing point, b) moisture above 200 $g.kg^{-1}$, and c) oxygen. With regard to silage, the key factor is oxygen which, if present, can promote mycotoxin production during the storage period. Oldenburg (1991) highlighted the wide range of water activity, temperature, and pH values at which most filamentous fungi can grow.

The genera *Fusarium, Aspergillus* and *Penicillium* are considered to be the most important producers of mycotoxins in silage (Oldenburg, 1991). *Fusarium* species are common plant pathogens, particularly in forage maize. *Aspergillus* and *Penicillium* species are most likely to be found in silages that have undergone aerobic spoilage. One of the most common mycotoxins is deoxynivalenol ((DON) or vomitoxin, which is produced by *Fusarium* (Whitlow and Hagler, 1997). This toxin is used as a marker to indicate the possible presence of other mycotoxins in the feed. A concentration of 3 to 5 mg $DON.kg^{-1}$ may indicate a potential health problem. Other mycotoxins of

concern are aflatoxin, T-2 toxin, zearalenone, moniliformin, ochratoxin, roquefortin C, and patulin (Oldenburg, 1991; Whitlow and Hagler, 1997).

Mycotoxins exert detrimental effects on the animal through three primary mechanisms: i) alterations of nutrient content, absorption, and metabolism; ii) changes in endocrine and neuroendocrine functions; and iii) suppression of the immune system (CAST, 1989). The signs of mycotoxicosis in animals are reduced feed consumption, decreased animal performance, poor fertility, and increased incidence and severity of disease (Whitlow and Hagler, 1997).

Seglar (1997) described several case studies in the USA where mycotoxicosis was implicated as a possible cause of dairy cattle ill health. Mycotoxins may not always be to blame for poor animal performance, and in view of the multiplicity of potential toxins, it is sometimes impossible to make a definitive diagnosis of the cause of the disease. But, as Oldenburg (1991) stressed, mycotoxins are serious hazards to both animals and humans, and, therefore, setting levels in silages that are safe or tolerable also is impossible.

The eyespot disease of forage maize (*Kabatiella zeae*) was encountered in UK for the first time in 1998. There appears to be no information on possible health hazards to animals from this fungus, but farmers have reported reduced feed consumption from crops that were harvested and ensiled after being killed rapidly and prematurely by the disease. Similarly, in the USA, there is evidence that the contamination of ensiled forage maize by mycotoxins is greater when the crop is exposed to frost prior to harvest (Gotlieb, 1997). The drier, frosted (dead) material is less easy to consolidate in the silo, allowing more oxygen to be entrapped within the mass of the ensiled crop, which, in turn, allows fungal growth to continue for a longer period of time. Fungi also may produce more toxins when stressed by cold weather, limited oxygen supply, or both.

Neotyphodium endophyte infection of perennial ryegrass (*Lolium perenne*) and the associated production of the indole alkaloid lolitrem B has been studied in relation to "ryegrass staggers", a neurological disorder in grazing animals (Oldenburg, 1997). There is no evidence to indicate whether or not the fungus or its toxin can survive the ensiling process, but the possibility of survival cannot be ruled out, especially in high dry matter material ensiled with little consolidation in large bales.

The question arises as to the extent to which the mixed microbial population in the rumen might be capable of metabolising toxins that can poison the animal. Weimer (1998) describes examples of microbial metabolism of plant toxins in the rumen , including the deacetylation of tricothene mycotoxins (e.g. DON and T-2 toxin) by *Butyrivibrio fibrisolvens*, albeit *in vitro*. The principle of successful detoxification, in the silo or in the rumen, is that the compound must be present in concentrations that provide a selective advantage to the organism. Thus, the infection of crops (or silages) by fungi is unlikely to result in microbial metabolism of toxins, unless the infection is significant, uniform, and continuous. Technologies for efficient crop conservation and legislation in animal feed storage are designed to reduce the frequency and severity of fungal infections and toxin production. Thus, when mycotoxin contamination of silage does occur, microbial metabolism of the toxins is less likely, rather than more likely.

5. Excess acidity

Wilkinson (1988) discussed the crucial role of salivary bicarbonate in buffering the acidity of silage and acids produced in the rumen. Because of the buffering system in ruminants clinical signs of acidosis are not common and attempts to relate the voluntary intake of silage to its acidity have met

with mixed degrees of success (Thomas and Thomas, 1985). Very little work has been carried out to determine the factors that affect the production of saliva in ruminants given different types of feed. Bailey (1958) demonstrated that saliva production per unit of feed consumed was three times greater for dried grass and hay than for silage, suggesting that perhaps length of fibre, rather than pH or buffering capacity, influences saliva production. In consequence, animals given short-chopped silage are likely to produce less saliva than those given the same silage as unchopped material and therefore may develop sub-clinical acidosis.

Cushnahan *et al.* (1996) reported a decrease in the voluntary intake of well-preserved grass silage as the period of ensilement increased from zero to 52 weeks (Table 3). The final pH of the silage was 3.7. The authors noted changes in eating patterns in addition to changes in composition of the silage, but period of ensilement had no effect on digestibility. These results support other circumstantial evidence of possible progressive depletion of buffering capacity in animals given silage diets for long periods of time, to which they respond by increased consumption of supplementary mineral salts and a preference for sources of long fibre such as straw. In more extreme situations, the animal may resort to drinking the urine of other animals and by spitting out regurgitated boli of food during rumination. In clinical acidosis, feed intake and rumination may cease totally for short periods of time.

Table 3 **Effect of period of ensilement on characteristics of grass silage and voluntary intake by dairy cows**
From Cushnahan *et al.* (1996)

	Period of ensilement (weeks)				
	0	3	9	52	s.e.m
Dry matter ($g.kg^{-1}$)	174	176	185	182	4.1
pH	5.3	4.0	3.8	3.7	0.07***
Total N ($g.kg\ DM^{-1}$)	24.6	25.3	25.0	24.6	1.44
Soluble N ($g.\ kg\ DM^{-1}$)	4.5	14.2	15.2	15.5	0.91***
Ammonia N ($g.kg\ total\ N^{-1}$)	11.3	55.7	70.9	94.3	2.46***
Lactic acid ($g.kg\ DM^{-1}$)	18.8	92.1	99.2	106.7	10.56***
n-butyric acid ($g.kg\ DM^{-1}$)	0	1.0	4.2	2.6	0.97*
Intake of DM ($kg.d^{-1}$)	15.3	14.5	13.3	9.9	0.43***

6. Epidemiology of silage and health

Considering the importance of silage as a feed for livestock, it is surprising that more research apparently has not been carried out to establish the scale of the possible health risks associated with the deficiencies in silage discussed in the earlier sections of this review. In one epidemiological study (Clarkson et al,, 1993) a relationship between the dry matter content of grass silage and lameness in dairy cows was described. The relationship was not good, but at least it indicated a possible area for further, more detailed, study.

Epidemiological studies are needed to establish whether or not silage is a significant source of hazardous organisms or chemicals in the food chain. Considering what might go wrong, it is remarkable that silage is not implicated more frequently in animal and human diseases. Perhaps nobody has investigated silage as the possible cause of disease. In some areas, the risk to the food

chain from clostridial fermentations is well-recognised, and silage making is either discouraged (e,g, in the Auvergne region of France) or prohibited by law (e.g. in the Emmental region of Switzerland and the Parma region of Italy). However, the risk in these regions is not principally to human health, but to the economic loss associated with the secondary blowing of cheeses during their maturation as a consequence of clostridial spores from silage infecting both milk and cheese (Wilkinson, *et al.*, 1996).

If new legislation is introduced to protect the human population from contaminated food, it follows that greater attention must be given to the hygienic preservation of silage. If coordinated epidemiological studies are undertaken across Europe with the objective of defining the hygienic status of silage and the risk factors associated with the occurrence of unhygienic silages, then a good start will have been made in providing farmers with better guidance so that poor quality silage is avoided and good health of animals and humans is safeguarded.

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Table 2. Chemical composition of forage pea silage after 90 days ensiling (all values g/kg DM unless otherwise stated)

	10 weeks		12 weeks		14 weeks			Significance†		
	Control	Inoc	Control	Inoc	Control	Inoc	s.e.d	H	I	H x I
DM (g/kg)	268	276	297	293	280	288	22.6			
pH	4.71	3.77	4.02	3.73	4.08	3.63	0.145	**	***	*
NH_3-N (g/kg N)	110.8	34.6	61.3	48.5	62.2	42.1	9.42	*	***	**
Crude protein	228	227	200	199	204	199	6.62	***		
NDF	484	455	417	424	436	406	13.29	***	*	
Starch	19.2	23.2	74.8	71.6	124.9	170.1	15.41	***		

†H = Harvesting Date; I = Inoculant Treatment; * = $p<0.05$; ** = $p<0.01$; *** = $p<0.001$

The chemical composition of the silages at 90 days is summarised in Table 2. The DM contents of the silages were similar for all treatments. The control silage from the first harvest had the highest pH and ammonia content. These undesirable fermentation characteristics may be linked to the low WSC content and high buffering capacity of the forage. Both increasing the maturity of the crop at harvest and the application of an inoculant significantly lowered the pH of the silages, with the inoculant having greatest impact on the 10 week growth forage. Although the crude protein contents of the silages from the later harvests were significantly lower than that of the 10 week growth silage they are still greater than that reported by Potts (1982). The starch contents of the 12 week and 14 week growth silages were three and five times greater than that of the 10 week growth silage respectively.

Conclusions

In this study the optimum growth stage for harvesting and ensiling forage peas was 205 (flat pod) which occurred at 12 weeks growth and gave optimum sugar substrate and low buffering for a satisfactory fermentation. It is essential to harvest peas before crop lodging for maximum return of DM and CP yield.

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Distribution and activity of microbial inoculants applied at reduced volumes with the Pioneer Appli-Pro™ Application Systems

Introduction

Certain microbial silage inoculants, used under the proper conditions, are effective in improving silage preservation and giving enhanced animal performance. The uniform distribution of the microbial inoculants is necessary to ensure the forage is adequately preserved during ensiling.

The Appli-Pro™ Application System is an integrated system consisting of a precision liquid applicator and a highly concentrated microbial inoculant. The system allows the application of Pioneer® brand microbial inoculants in a reduced volume, eliminating the need for frequent interruptions in harvesting for reloading of water or inoculant.

We have conducted a series of studies to assess the dispersal of microbial inoculant organisms when applied in a typical commercial volume of 2.0 l/t or in a reduced volume of 0.25 l/t. During the course of the study, comparisons were made between two types of commercial applicators for application of the inoculant at normal and reduced volumes.

Method and Materials

Texas Red* sulfonyl chloride was used as a fluorescent reporter molecule to label the cell surface of *Enterococcus faecium.* Bacterial cells were treated with Texas Red sulfonyl chloride to form a stable sulfonamide linkage resulting in intensely fluorescent cells which could easily be distinguished from autofluorescent plant debris or epiphytic microbes by flow cytometry.

In the U.S. orchard grass was cut with a mower, raked into wind rows and wilted to a dry matter of 35%. The forage was chopped with a self-propelled forage chopper and inoculated at $1x10^6$ labeled cells/g forage with the Appli-Pro™ applicator at rates of 2.0 l or 0.25 l/t. Application was directly into the throat of the chopper. Forage was deposited in rows in the field and ten equally spaced samples were collected for analysis. Duplicate trials were performed on first and third cut crops.

Enumeration of the labeled cells was done by flow cytometry at 544 nm using a Helium-Neon laser. The number of labeled cells per gram of sample was calculated with reference to an internal standard of Florosphere latex beads.

Distribution of non-labeled inoculant organisms and silage fermentation was done in Europe with rye grass forage at a dry matter of 16%. Forage was precision chopped and inoculated with the Appli-Pro applicator at a rate of 0.25 l/t or with an Ag-Spray applicator at a rate of 2.0 l/t. Both applicators were calibrated to deliver $1x10^5$ CFU/g Pioneer brand 1188 inoculant per gram forage.

Ten equally spaced samples were collected and inoculant organisms were enumerated by dilution plate count techniques using modified MRS agar.

Eighteen PVC model silos were prepared for each treatment and monitored for pH decline with triplicate openings at 0, 1, 3, 7, 45, and 90 days.

Results

Trials conducted on orchard grass allowed the recovery of 60% and 25% of the applied organism from first and third cut, respectively. More variability was observed with the reduced volume (0.25 l/t) than with the standard commercial volume (2.0 l/t).

Frequency distributions pooled from the two studies suggested that the distributions at both volumes were similar. Analysis of the mean deviation indicated that the distributions were statistically identical between volumes and studies.

Studies of the distribution of inoculant organisms on rye grass showed similar results. Traditional plate count methods resulted in distributions which were statistically identical and within the normal variation for recovery of organisms.

Fermentation profile studies suggest that there is a more rapid fermentation in the inoculated forage when compared to control but no difference between application volumes and applicators.

Conclusions

The Appli-Pro applicator can accurately deliver inoculant organisms in a reduced volume of liquid. The distribution of organisms applied by the Appli-Pro applicator is similar in both the standard commercial volume (2 l/t) and the reduced volume (0.25 l/t).

Comparison of the commercial Ag-Spray applicator and the Appli-Pro Applicator system suggest a similar level of precision in the delivery of inoculant organisms. There appears to be no effect of delivery volume reduction on the distribution organisms or in the fermentation of the ensiled forage.

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Ensiling during difficult conditions of two direct cut forages, with different botanical composition

Introduction

Small amounts of red clover in a ley may change the conservation characteristics of the forage compared to a pure grass ley. The aim of the experiment was to describe the effects of different conservation treatments depending on botanical composition in the ley, during difficult weather conditions.

Materials and methods

Timothy *(Phleum pratense)* in pure stands and a mixture of timothy (800 g kg^{-1} DM) and red clover *(Trifolium pratense)* (200 g kg^{-1} DM) were grown during the rainy summer of 1998 at Röbäcksdalen research farm (63°35´N, 20° 45´ E). The crops were analysed for dry matter (DM), buffering capacity, crude protein (CP), water soluble carbohydrates (WSC), neutral detergent fibre (NDF), rumen degradable organic matter (RDOM) and ash. Conservation was carried out in small plastic silos with five different treatments: no additive (Control), formic acid (Formic), lactic acid bacteria and enzymes (Siloferm Plus™, Medipharm AB) and molasses (LAB-A + mol), lactic acid bacteria and enzymes (Feedtech™, Alfa Laval Agri) (LAB-B), lactic acid bacteria, enzymes and sodium benzoate (Feedtech II™, Alfa Laval Agri AB) (LAB-C + benz). Each silo was filled with 10 kg of fresh material. The silos were opened after 90 days and samples were taken from each silo. One sample was analysed fresh for DM, pH, WSC, lactic acid, acetic acid, butyric acid, 2-3 butanediol, NH_3-N and ethanol. Another sample was dried at 60 °C during 48 hr and analysed according to the Cornell net carbohydrate and protein model (Sniffen *et al*, 1992). The results were used for calculating the feed carbohydrate and protein fractions; sugars (CA), starch and pectin's (CBI), available structural cell wall (CBII), indigestible fiber (CC), non protein nitrogen (buffer soluble) (PA), soluble protein (buffer soluble) (PBI), rumen degradable protein (PBII), slowly degradable protein (PBIII) and fiber bound protein (PC) (Sniffen *et al,* 1992). Chemical composition and feed fractions of the silages were statistically analysed as two randomised block experiments, one for each crop.

Results

Results from analyses of the crops and silages are presented in Table 2, 3 and 4. The calculated feed carbohydrate and protein fractions are presented in Table 1.

Table 1. Carbohydrate and protein fractions (Sniffen et al, 1992) in the silages (g kg^{-1} DM)

Treatment	Crop	n	CA	CBI	CBII	CC	PA	PBI	PBII	PBIII	PC
Control	Timothy	3	191^{a}	21.3^{a}	453^{a}	84.8^{ab}	55.5^{b}	0	31.4^{a}	6.3^{a}	7.6^{a}
Formic	Timothy	3	161^{a}	17.9^{a}	477^{a}	81.6^{ab}	42.8^{a}	0	41.1^{b}	19^{b}	6.0^{a}
LAB-A + mol	Timothy	3	193^{a}	21.5^{a}	443^{a}	62.4^{a}	79.7^{c}	0	28.9^{a}	6.3^{a}	7.0^{a}
LAB-B	Timothy	3	172^{a}	19.1^{a}	469^{a}	92.8^{b}	56.6^{b}	0	27.0^{a}	7.0^{a}	7.0^{a}
LAB-C+ benz	Timothy	3	154^{a}	17.1^{a}	457^{a}	106^{b}	58.5^{b}	0	29.1^{a}	7.0^{a}	7.0^{a}
All	Timothy	15	175	19.5	460	86	59	0	32	9.0	6.9
Control	Clover/Timothy	3	211^{a}	23.4^{a}	366^{bc}	84.8^{a}	76.1^{b}	0	43.8^{a}	12^{a}	9.3^{a}
Formic	Clover/Timothy	3	195^{a}	21.6^{a}	383^{c}	90.4^{a}	38.4^{a}	0	45.2^{a}	58^{b}	9.3^{a}
LAB-A + mol	Clover/Timothy	3	281^{b}	31.2^{b}	268^{a}	97.6^{a}	87.5^{c}	0	47.2^{a}	12^{a}	9.0^{a}
LAB-B	Clover/Timothy	3	207^{a}	23.0^{a}	302^{ab}	137^{a}	77.0^{bc}	0	45.9^{a}	14^{a}	8.6^{a}
LAB-C+ benz	Clover/Timothy	3	228^{a}	25.4^{a}	301^{ab}	128^{a}	73.5^{b}	0	47.1^{a}	11^{a}	7.3^{a}
All	Clover/Timothy	15	224	24.9	324	107	70,5	0	45.8	22	8.7

Means of the same crop in each column with the same superscript do not differ significantly($p>0.05$).

Table 2. Chemical composition (g kg^{-1} DM) and RDOM of the two crops

Crop	DM g kg^{-1}	CP	WSC	NDF	RDOM %	Ash	Buffering cap meqv(100 g DM)$^{-1}$
Timothy	199	91	135	536	83.8	90	20
Clover/Timothy	142	137	132	432	88.7	97	35

Table 3. Chemical contents of the silages (g kg^{-1} DM)

Treatment	Crop	n	CP	ADF-CP	Soluble CP (%)	NDF-CP	ADF	NDF	NSC*	Ash	Lignin	Crude fat
Control	Timothy	3	101^{ab}	7.7^{a}	55.0^{b}	14.0^{a}	390^{b}	546^{a}	212^{a}	105^{a}	35.3^{ab}	43.3^{a}
Formic	Timothy	3	109^{b}	6.0^{a}	39.3^{a}	25.0^{b}	407^{b}	564^{a}	199^{a}	112^{a}	34.0^{ab}	41.7^{a}
LAB-A + mol	Timothy	3	122^{c}	7.0^{a}	65.3^{c}	13.3^{a}	351^{a}	512^{a}	283^{b}	116^{a}	26.0^{a}	43.0^{a}
LAB-B	Timothy	3	98^{a}	7.0^{a}	58.0^{b}	14.0^{a}	403^{b}	569^{a}	208^{a}	106^{a}	38.7^{b}	42.7^{a}
LAB-C+ benz	Timothy	3	102^{b}	7.0^{a}	57.7	14.0^{a}	410^{b}	573^{a}	234^{ab}	113^{a}	44.3^{b}	49.7^{b}
All	Timothy	15	*106*	*6.9*	*55*	*16.0*	*392*	*552*	*194*	*109*	*35.6*	*43*
Control	Clover/Timothy	3	141^{a}	9.3^{a}	54.0^{b}	21.0^{a}	358^{b}	460^{b}	235^{a}	110^{a}	35.3^{a}	63.3^{ab}
Formic	Clover/Timothy	3	151^{ab}	9.3^{a}	25.3^{a}	77.5^{b}	335^{b}	483^{b}	216^{a}	103^{a}	37.7^{a}	54.3^{a}
LAB-A + mol	Clover/Timothy	3	156^{b}	9.0^{a}	56.0^{b}	21.5^{a}	291^{a}	375^{a}	312^{b}	112^{a}	40.7^{a}	53.3^{a}
LAB-B	Clover/Timothy	3	146^{ab}	8.6^{a}	52.6^{b}	23.3^{a}	355^{b}	448^{b}	230^{a}	117^{a}	51.3^{a}	67.0^{b}
LAB-C+ benz	Clover/Timothy	3	139^{a}	7.3^{a}	52.6^{b}	19.0^{a}	337^{b}	437^{b}	264^{a}	101^{a}	58.7^{a}	67.0^{b}
All	Clover/Timothy	15	*147*	*8.7*	*48*	*30.5*	*335*	*440*	*249*	*110*	*44.5*^{a}	*61*

Means of the same crop in each column with the same superscript do not differ significantly (p>0.05).

Table 4. Chemical contents of the silages (g kg^{-1} DM).

Treatment	Crop	n	DM g kg^{-1}	NH_3-N (%)	WSC	pH	Lactic acid	Acetic acid	Butyric acid	2,3-Butanediol	Ethanol
Control	Timothy	3	185^{a}	6.4^{b}	21^{ab}	3.67^{a}	106^{b}	17.8^{a}	$< 1.0^{a}$	< 1.0	10.0^{a}
Formic	Timothy	3	188^{a}	4.5^{a}	16^{a}	3.88^{a}	56^{a}	18.0^{a}	12.7^{a}	< 1.0	19.0^{b}
LAB-A + mol	Timothy	3	191^{a}	5.6^{ab}	27^{ab}	3.77^{a}	116^{b}	11.4^{a}	$< 1.0^{a}$	< 1.0	58.0^{b}
LAB-B	Timothy	3	186^{a}	6.7^{b}	29^{b}	3.68^{a}	106^{b}	17.5^{a}	$< 1.0^{a}$	< 1.0	9.6^{a}
LAB-C+ benz	Timothy	3	183^{a}	7.3^{b}	25^{ab}	3.67^{a}	111^{b}	18.2^{a}	$< 1.0^{a}$	< 1.0	9.4^{a}
All	Timothy	15	186	6.1	23	3.73	99	16.5	2.5	< 1.0	21.6
Control	Clover/Timothy	3	126^{a}	7.4^{b}	4^{a}	3.92^{a}	160^{c}	34.1^{c}	0.5^{a}	< 1.0	8.2^{a}
Formic	Clover/Timothy	3	146^{b}	3.1^{a}	33^{b}	3.84^{a}	97^{a}	24.3^{b}	3.2^{a}	< 1.0	7.9^{a}
LAB-A + mol	Clover/Timothy	3	166^{c}	3.7^{a}	30^{b}	3.84^{a}	128^{b}	13.7^{a}	$< 1.0^{a}$	< 1.0	36.0^{b}
LAB-B	Clover/Timothy	3	130^{ab}	7.3^{b}	4^{a}	3.94^{a}	153^{bc}	34.6^{c}	$< 1.0^{a}$	< 1.0	8.6^{a}
LAB-C+ benz	Clover/Timothy	3	139^{ab}	6.8^{b}	5^{a}	3.90^{a}	148^{bc}	31.1^{c}	$< 1.0^{a}$	< 1.0	7.9^{a}
All	Clover/Timothy	15	141	5.6	15	3.89	137	27.5	0.7	< 1.0	13.8

Means of the same crop in each column with the same superscript do not differ significantly (p>0.05).

Conclusions

The mixed crop with timothy and red clover responded differently to the treatments compared to the pure timothy crop. Treatments with addition of molasses resulted in a high level of ethanol. All treatments resulted in well preserved silages with different fermentation characteristics.

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Effect of stage of growth and field wilting on the characteristics of fermentation of Sainfoin *(Onobrychis viciifolia).*

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Introduction

Sainfoin *(Onobrychis viciifolia) is* an under-exploited leguminous crop grown in the United Kingdom and other areas of Northern Europe. The crop has several attributes that make it particularly attractive to animal production, the presence of condensed tannins which reduces the risk of bloat in the grazing ruminant (ENTEC, 1998), the possible role of condensed tannins in increasing the deposition of protein into muscle (ENTEC, 1998) and a low degree of leaf shatter during the production of hay. The experiment reported in this paper examines the changes in chemical composition and nutritive value of the crop cut at three stages of growth and the impact of field wilting on the ensilability of the crop.

Materials and method

Sainfoin *(Onobrychis viciifolia* var. Cotswold Common) was sown in late March 1997 into a prepared seed bed at a rate of 86 kg seed (unhulled) per hectare. The soil at the site was a gravelly silt (Bengeo Complex: pH 7.3, P 2.6 mgll, K 46 mg/1 and Mg 112 mg/1). No fertiliser was used after the crop was sown and the plots were hand-weeded. The crop was harvested at three stages of growth (inflorescence emergence - April 27 (E), half bloom - May 11 (HB) and full bloom - May 27 (FB)). The crop was chopped (rotary guillotine) and ensiled immediately after harvest or wilted for 16 h. The various forages were ensiled in triplicate for each treatment in one litre laboratory silos without any additives. After 90 days of storage, the silos were opened and silages analysed for chemical composition. Data of chemical composition were analysed statistically using analysis of variance techniques with date of cut and period of wilting being the independent variables.

Results and discussion

The yields of DM were 3.1, 3.7 and 3.9 t DM/ha at E, HB and FB stages of growth respectively. The level of production represented a single cutting cycle and it acceptable to cut sainfoin at least twice during one growing season. Yields of 7 to 9 t DM/ha have been recorded elsewhere in the United Kingdom (Hill, 1998). The chemical composition of sainfoin cut and harvested immediately or wilted for 16 hours are given in Table 1. Increasing maturity of the crop increased the concentration of NDF ($p<0.05$) and reduced the buffering capacity ($p<0.01$). Wilting tended to increase the concentration of cell wall components and WSC but buffering capacity of the crop at various stages of growth was reduced. The buffering capacity of sainfoin is lower than lucerne but higher than grass. The chemical composition and nutritive value of ensiled sainfoin are in Table 2. There were significant increases in the concentrations of DM ($p<0.05$), NDF ($p<0.01$), pH ($p<0.001$) and amino-N ($p<0.01$) with increasing crop maturity and period of wilting. The increase in NDF may have influenced the decline in DOMD with increasing crop maturity. Increasing crop maturity decreased the concentration of fermentation acids significantly only if the crop was wilted (lactic acid $p<0.05$). Butyric acid was observed in silages made from unwilted or immature (E) wilted sainfoin. This may well reflect the impact of high buffering capacity, low sugar content and high moisture content prior to ensiling. Wilting sainfoin to dry matter contents greater than 300 g DM/kg promoted high quality fermentation and inhibit micro-organisms detrimental to the fermentation.

Table 1. Chemical composition of sainfoin (g/kg DM unless stated) prior to ensiling.

	Harvested immediately			Wilted for 16 h			s.e.d.
	IE	HB	FB	IE	HB	FB	
DM (g/kg)	234	266	274	279	328	402	11.7
DOMD	622	608	595	612	600	587	9.3
NDF	507	520	549	518	544	568	13.6
ADF	321	338	341	336	341	348	11.4
Lignin	27	34	31	32	35	34	4.5
WSC	84	76	68	89	79	78	9.5
CP	236	207	198	221	199	194	8.6
BC (mE/kg DM)	539	482	467	503	450	422	22.5

Table 2. Chemical composition and nutritive value of sainfoin silage (g/kg DM unless stated)

	Harvested immediately			Wilted for 16 h			s.e.d.
	IE	HB	FB	IE	HB	FB	
DM (g/kg)	244	262	282	284	322	416	19.6
NDF	517	531	551	506	538	561	11.6
ADF	347	344	350	344	359	357	9.9
Lignin	31	27	36	30	31	33	3.6
CP	204	191	190	196	186	181	6.4
DOMD	600	587	577	605	592	579	8.6
pH	4.1	4.23	4.55	4.4	4.65	5.01	0.55
Ammonia-N (g/kg total N)	109	86	92	96	86	82	11.6
WSC	19	22	18	14	12	15	3.6
Lactic acid	64	55	54	58	39	34	9.7
Ethanol	4.1	4.3	3.6	2.2	1.1	3.4	2.6
Acetic acid	22	20	23	16	15	13	4.8
Butyric acid	3.1	1.7	1.1	1.1	0.1	<0.1	0.12

Conclusions

The pattern of fermentation was improved by field wilting reducing the concentration of butyric acid and ammonia-N. Yield of DOMD per hectare was maximum at full bloom. The increase in yield of DOMD was only 0.035 between HB and FB but the increase between E and FB was 0.167. If field wilting is not feasible, it is likely the crop should be treated with an additive (as with other legumes for silage) to circumvent the effect of high buffering capacity.

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Effects of wilting and application of a bacterial inoculant on the fermentation characteristics of lupin silage.

Introduction

Lupin grain is used extensively in North America and Australia as a concentrated protein supplement for livestock. Whilst research by Milford (1994) has shown that *Lupinus albus* has the potential to grow well in many areas of the UK as a seed crop, there is little information in the literature as to its potential as a high protein forage crop. The aim of this study was to evaluate the production potential of forage lupins under UK conditions, and to evaluate their fermentation characteristics when ensiled with and without an inoculant.

Materials and Methods

Lupinus albus (cv. Amiga) was sown on the 15 May 1998 on land previously prepared by applying farmyard manure at 25 t/ha. A compound fertiliser was applied to the seedbed at a rate of 60 kg N, 24 kg P_2O_5, 42 kg K_2O/ha, and the lupins sown at a rate of 129 kg/ha. No herbicide was applied during the establishment period. The crop was harvested at a height of 100mm on the 8 October 1998. Half the crop was immediately chopped (< 30mm) through a forage harvester and ensiled in laboratory scale silos (1kg). The remaining crop was artificially wilted for a period of 24 h using a forced draught cold air drier before being chopped and ensiled as described above. Treatments imposed were unwilted or wilted forage either untreated (Control) or inoculated (Inoc.) with 10^6 CFU/g FM of *Lactobacillus plantarum* (Ecosyl Bio-Products, UK). Four replicated mini silos of each treatment were incubated for a period of 90 days.

Results

The lupin forage was at growth development stage 4.7 (pod colour brown) at harvesting. The chemical composition and yield of the pre-ensiled lupin crop are shown in Table 1. Production of lupins as a forage crop was high at more than 9t DM/ha, and almost twice that previously reported by Wiatrak (1996). The lupin crop was low in dry matter (DM) and artificially wilting for 24 hrs only succeeded in raising the DM content by 15 g/kg, presumably as a result of the thick nature of the stems and the lack of mechanical conditioning. The crude protein content of both the unwilted and wilted crop was in excess of 185 g/kg, with no significant difference observed between drying treatments. The soluble sugar content (WSC) of the forages was also unaffected by crop drying, and was in excess of 153 g/kg DM. The

Table 1. *Production and chemical composition of unwilted and wilted lupin forage (g/kg DM unless otherwise stated)*

	Unwilted	Wilted	s.e.d.
Dry matter (g/kg)	161	175	6.5
Crude protein	190	185	5.5
ADF	360	358	13.3
NDF	458	435	13.4
WSC	165	153	17.5
Buffering capacity (meq/kg)	304	338	25.0
Ash	49.6	54.4	3.70
Yield (kg DM/ha)	9239	---	---

buffering capacity of the lupin crop was similar to that of grass species, and considerably lower than that normally expected for leguminous crops.

The chemical composition of the control and inoculant-treated lupin silages from unwilted and wilted forage are shown in Table 2. All silage treatments fermented well with pH values less than 4.45. Although no significant difference in pH was observed between drying treatments, additive treatment had a highly significant effect, with inoculant application resulting in more extensive acidification. Extensive protein degradation was evident, with ammonia-N contents exceeding 100 g/kg N for all treatments except the inoculated wilted silage (73 g/kg N).

Table 2. *Mean composition of lupin silage after 90 days ensiling (all values g/kg DM unless otherwise stated)*

	Unwilted		Wilted			Significance†	
	Control	Inoc.	Control	Inoc.	s.e.d	D	A
DM (g/kg)	150.1	147.9	183.3	173.2	4.64	***	ns
pH	4.42	3.96	4.45	3.87	0.071	ns	***
Crude protein	208.6	198.8	234.3	218.8	4.21	***	**
NH_3-N (g/kg N)	177.0	130.5	131.0	73.0	9.59	***	***
ADF	459.8	442.6	443.5	433.4	14.18	ns	ns
NDF	544.2	528.8	500.1	482.3	20.11	**	ns
WSC	7.8	10.3	7.8	13.0	0.96	ns	***

†D = Drying treatment (unwilted or wilted); A = Additive treatment
** = $p<0.01$; *** = $p<0.001$; ns = not significant
There were no Drying treatment x Additive treatment interactions

Conclusions

The relatively high WSC content and low buffering capacity of a lupin forage crop at growth stage 4.7 ensured good preservation, while inoculant additive treatment provided a more rapid and extensive acidification. Further work to determine the nutritive value of lupin forage is now required since the high crude protein content of this crop suggests lupin silage has potential as a protein supplement for high-energy grass or maize crops.

Acknowledgements

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The Potential of By-Products From The Food Industry Ensiled Together With Poultry Manure for Ruminants

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INTRODUCTION

Food industry by-products (pomaces and pulps) and poultry excreta are valuable alternative feed resources for both small and large ruminants (Jayasuriya 1985). There are indications that combinations of poultry excreta with sugar beet pulp or apple or tomato pomaces meet the physiological and nutritional requirements of ruminants (Muller 1980, Karabulut 1997). The aim of this study was to investigate the feeding value and growth potential for fattening lambs of silages produced from apple pomace (AP), tomato pomace (TP) and sugar beet pulp (SBP) each mixed with poultry manure from cage layers (CLM).

MATERIALS AND METHODS

AP, TP and SBP were ensiled in fibreglass silos after mixing CLM at a 70/30 ratio on DM basis. 3% molasses (dry weight basis) was used as a silage additive. Silages were fed to fattening lambs for 56 days in order to determine feed values in vivo. Silages were offered ad libitum with a concentrate supplementation equivalent to 2% of individual live weights (LW). The concentrate contained 143 g/kg DCP and 10.6 MJ/kg ME.

RESULTS AND DISCUSSION

The chemical composition and digestibilities of the silages are shown in Table 1 and data from the digestion trial in Table 2. Digestibilities of AP + CLM, TP + CLM and SBP + CLM silages were higher than in corresponding silages without CLM (Bartocci et al. 1980). CLM appeared to have a high digestibility and that affected digestibilities of the mixed silages positively. All mixed silages were classified as "good quality" according to the Flieg Score System. Pathogens such as enterobacteria, yeast and mould were not detected in the silages. Lambs fed on SBP + CLM or AP + CLM showed significantly higher LW gains than lambs on TP + CLM silage ($P<0.05$).

CONCLUSIONS

Good quality silages with a high feed value were produced by ensiling AP, TP or SBP mixed with CLM.
LW gains of fattening lambs were satisfactory particularly with SBP + CLM and AP + CLM silages.

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Workshop A
Regulation of silage fermentation

Poster abstracts

Use of a genetic algorithm to improve inoculants for grass silage

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INTRODUCTION

Genetic algorithms are 'adaptive' computer programmes which operate in a manner that mimics the evolutionary process in living organisms. Individuals within genetic algorithm populations are represented as a string of 'genes', with each gene corresponding to an experimental parameter and the level of each parameter represented as a different 'allele'. The initial population is randomly generated and the 'fitness' of individuals assessed experimentally. The best (fittest) are selected to produce the next generation by the processes of mutation and recombination. In this way, progressively fitter individuals are produced.

The selection of improved combinations of silage inoculants is a novel application of genetic algorithms. A 'proof of principle' experiment was conducted in which a genetic algorithm was used in combination with laboratory-based, short-term ensilage experiments to select the level(s) of inoculant, enzyme and sugar to use in the treatment of grass silage.

MATERIALS AND METHODS

Five generations of treatments were carried out, each containing 50 different individuals. Each individual was an additive treatment made up of 8 ingredients at a level between 0 and 5 (as defined in Table 1).

Table 1. Silage additives and application levels used in genetic algorithm experiment

	Additive	Units	Level 1	Level 2	Level 3	Level 4	Level 5
1	Freeze-dried *Lactobacillus plantarum*	$CFUg^{-1}FW$	10^3	10^4	10^5	10^6	10^7
2	Freeze dried *Pediococcus pentosaceus*	$CFUg^{-1}FW$	10^3	10^4	10^5	10^6	10^7
3	Freeze dried *Lactobacillus buchneri*	$CFUg^{-1}FW$	10^3	10^4	10^5	10^6	10^7
4	Fresh culture *L. plantarum*	$CFUg^{-1}FW$	10^3	10^4	10^5	10^6	10^7
5	Fresh culture *P. pentosaceus*	$CFUg^{-1}FW$	10^3	10^4	10^5	10^6	10^7
6	β-glucanase enzyme	• $g\ kg^{-1}$ FW	0.1	0.25	0.5	1.0	3.0
7	xylanase enzyme	• $g\ kg^{-1}$ FW	0.1	0.25	0.5	1.0	3.0
8	sugar fructose:glucose (60:40)	% of silage DM	0.1	0.5	1	2.5	5

FW= fresh weight, DM = dry matter

A mixed grass sward was mown after 4 weeks growth. Two adjacent plots were used with cuts staggered at 2-week intervals between July and September 1998. Chopped herbage (100g) was inoculated with 2 ml of liquid (as a fine spray), packed into small laboratory silos and ensiled for 2 days. The resultant silage was analysed for lactate, pH and amino acids. A fitness value was assigned to each of the treatments using the equation: 1 /{1+ (pH wt x[pH/control pH])+(lac wt /[lac/control lac])+(AAwt x[AA/controlAA])+cost/40}

The three parameters pH, lactate (lac) and amino acids (AA) were weighted (wt) 0.25, 0.40 and 0.35 respectively. A relative cost factor was included to prevent the genetic algorithm selecting unrealistic levels of additive. After each generation the genetic algorithm programme selected treatments for the next generation. The two selections which gave the highest fitness values in generation 5 were used to treat grass, alongside a commercial inoculant (containing freeze dried *L.plantarum* and *P.pentosaceus* each at 10^5 $CFUg^{-1}FW$ plus 1 $\mu g\ kg^{-1}$ FW of an enzyme mixture) and an uninoculated control. The inoculated and control herbages were ensiled in triplicate and opened at intervals of 1, 4, 14 and 100 days.

RESULTS AND DISCUSSION

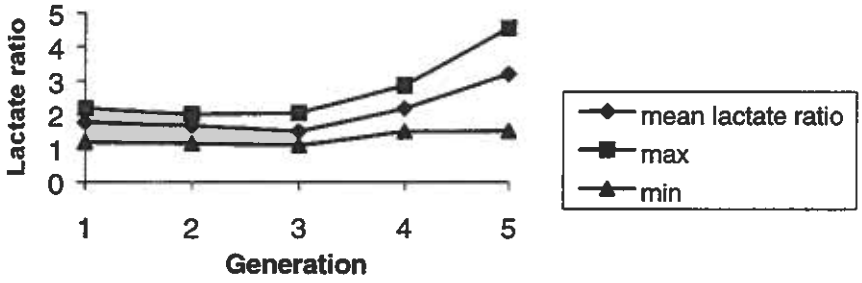
The mean pH, lactate, amino acid content and fitness values of the fifty silage samples in the 5 generations are shown in Table 2. Silages made over the course of the five generations would be expected to deteriorate

in quality due to the seasonal decline in the water soluble carbohydrate content of the herbage (which decreased from 177 to 111 g kg^{-1} DM during the course of the 5 generations). However, despite the fact that the herbage ensiling potential decreased, the quality of the silage produced was maintained throughout. The improvement in silage quality over the five generations is illustrated in Figure 1 by the change in lactate accumulation relative to the control silage.

Table 2. Silage fitness parameters at each generation

Parameters (mean values)	Generation 1	Generation 2	Generation 3	Generation 4	Generation 5
pH	3.83	3.66	3.56	3.73	3.73
Lactate (g kg^{-1} DM)	70.3	77.1	72.2	78.5	68.4
Amino acid (mol kg^{-1}DM)	0.24	0.26	0.27	0.32	0.40
Fitness	0.119	0.108	0.106	0.113	0.122

Figure 1. Ratio of lactate (treated/control) at each generation of genetic algorithm experiment



The fermentation characteristics of silages (mean, s.d.) inoculated with the two best treatments from generation 5 are shown in Table 3. Both inoculants gave a faster rate of pH decline and lactate accumulation than either a control silage or one treated with a commercial inoculant. Less amino acid accumulated in the two combinations selected by the genetic algorithm indicating lower levels of protein breakdown in the silage.

Table 3. Fermentation characteristics of grass silage over 100 days

		Inoculant 1	Inoculant 2	Commercial inoculant	Control
PH	Day 1	4.26 (0.01)	3.92 (0.02)	5.40 (0.03)	5.87 (0.04)
	4	3.53 (0.02)	3.54 (0.02)	3.71 (0.02)	3.91 (0.02)
	14	3.43 (0.01)	3.45 (0.01)	3.62 (0.02)	3.62 (0.02)
	100	3.52 (0.01)	3.52 (0.05)	3.64 (0.03)	3.59 (0.01)
Total lactate	Day 1	45.9 (4.3)	57.0 (3.5)	13.6 (0.9)	7.30 (2.6)
(g kg-1DM)	4	116.4 (18.6)	118.4 (11.9)	100.5 (16.3)	62.5 (13.0)
	14	91.1 (22.8)	114.6 (41.5)	103.1 (13.9)	90.3 (5.0)
	100	159.7 (1.0)	152.3 (15.2)	150.4 (3.3)	150.8 (4.8)
Amino acid	Day 1	0.16 (0.01)	0.09 (0.02)	0.19 (0.02)	0.21 (0.02)
(mol kg-1 DM)	4	0.23 (0.04)	0.26 (0.05)	0.29 (0.04)	0.36 (0.02)
	14	0.32 (0.01)	0.31 (0.02)	0.35 (0.05)	0.39 (0.02)
	100	0.14 (0.02)	0.22 (0.03)	0.29 (0.08)	0.29 (0.04)

CONCLUSIONS

The results of this study show that a genetic algorithm can be used to improve the design of silage inoculants, providing a logical way of optimising the composition of the inoculant. A short-term ensilage period was successfully used in the evaluation of treatments. The 'fittest' combinations selected by the genetic algorithm produced silages with improved chemical characteristics compared to those produced from an uninoculated silage or one which was inoculated with a commercially available product.

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The effect of variety, cut, moisture level, and microbial inoculant on the terminal pH of ensiled alfalfa.

Introduction

Small-scale vacuum packets are a useful model for ensiling small quantities of forage for experimental purposes. Using this model, we determined that there are differences between alfalfa varieties in the pH they attain when ensiled, and that these differences were maintained across years and cuts. To facilitate the selection of alfalfa genotypes for improved ensiling qualities, it was important to determine which management practices affect variety differences.. To look at these effects, we ensiled seven different alfalfa varieties representing a wide range of germplasm at three different moistures, over three cuts, with two different microbial treatments, utilizing small-scale vacuum packets.

Methods and Materials

For each alfalfa variety tested, 1 meter of plant material was harvested from the middle row of a test plot. The harvested forage was cut into 3-cm lengths before chopping with a food processor. Fifty grams of this material were placed on a paper plate and wilted to a predetermined dry matter (25, 35, or 45%). Each sample was inoculated with the appropriate microbial treatment (Pioneer® Brand 1174 silage inoculant (1174), or an experimental inoculant (ST)) or left untreated (Control). The forage was mixed and ensiled in polyethylene vacuum packets. The packets were kept at room temperature for 28 days, at which time the packets were opened and the pH of the contents determined. Three test plots per variety were sampled, with three reps per plot for each treatment. The same test plots were sampled during the 1st, 3rd, and 4th cuttings.

Results

Differences between varieties, dry matters, cuts, and microbial treatments were highly significant ($P<0.001$). Varieties 1 and 6 had the lowest terminal pH, while varieties 3 and 7 had the highest. Variety had the least interaction with the other factors; the rankings of varieties from best to worst remained constant regardless of dry matter, cut, or microbial treatment. The 45% dry matter silage averaged the lowest terminal pH while the 25% dry matter silage had the highest pH. The alfalfa-specific inoculant (ST) produced silage with a lower pH than 1174 ($P<0.001$) while the 1174 inoculated silage had lower pH than uninoculated Control ($P<0.001$). Cut had the biggest influence on pH ($3<1<4$) and significantly interacted with variety, dry matter, and microbial treatment ($P<0.001$).

Summary

There are significant differences between alfalfa varieties in their ability to generate pH low enough to ensile successfully. Management factors such as dry matter, microbial treatment, and cut significantly influence the terminal pH of ensiled alfalfa.

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Effects of harvesting date and application of a bacterial inoculant on the fermentation characteristics of field bean silage.

Introduction

The field bean is a high yielding short-term crop that has potential as a high protein forage crop. Previous work (Faulkner, 1985) has shown that field beans can produce heavy crops of forage without any requirement for nitrogenous fertiliser. However, there is limited information available in the literature on the ensiling potential of field beans. The aim of this study was to compare the yield and chemical composition of field beans harvested at three different stages of growth, and ensiled with and without an inoculant.

Materials and Methods

Field beans (*Vicia faba*) were drilled on 29 April 1998 at a seed rate of 280 kg/ha. Compound fertiliser (P_2O_5, K_2O) was applied to the seedbed to achieve phosphate and potash indices of 3. A pre-emergent herbicide, Opogard 500SC (terbutryn + terbuthylazine), was applied at a rate of 2.8 l/ha on the 2 May. In addition, a fungicide, C-Flo 2 (carbendazim) was sprayed at a rate of 1litre/ha on the 1 July and again on the 25 July, and an insecticide, Aphox, (pirimicarb), was applied at a rate of 280 g/ha on the 16 July. The crop was cut at three different growth stages; first pod set (204), pods fully formed (205) and pod fill (207), which corresponded to 10, 12 and 14 weeks growth respectively. The forage was harvested at a height of 100 mm using a Haldrup plot harvester. Samples of the forage as harvested were analysed for dry matter (DM) and water soluble carbohydrate (WSC) contents and buffering capacity. The forage was then artificially wilted for a period of 24 h using a forced draught cold air drier, before being chopped and ensiled in 10 kg silos. Half the forage was ensiled untreated (Control) whilst the other half was treated with an inoculant (Live System; Genus Ltd, UK) applied at a rate of 10^6 CFU (*Lactobacillus plantarum*) per gram fresh matter (FM). Three replicated mini silos of each treatment were incubated for a period of 90 days. The silos were then opened and representative sub-samples analysed for DM, ammonia nitrogen (NH_3-N), crude protein (CP) (N x 6.25), neutral detergent fibre (NDF) and starch contents, and pH.

Results and Discussion

The yield and key chemical characteristics of the freshly cut crop are shown in Table 1. The forage yield of field beans increased significantly between harvest dates, with the yield at 14 weeks more than double that at 10 weeks. Whilst there was also a significant increase in DM

Table 1. *Production and chemical composition of field beans cut at different growth stages*

	10 weeks	12 weeks	14 weeks	s.e.d	Sign.[†]
Dry matter (g/kg)	120.9	134.5	153.1	5.22	**
WSC (g/kg DM)	186.0	157.3	147.0	13.17	
Buffering capacity (meq/kg)	350	312	339	31.4	
Yield (kg DM/ha)	3698	5167	7760	238.3	***

[†] ** = $p<0.01$; *** = $p<0.001$

with maturity, the DM content of the crop at all three growth stages was comparatively low. The buffering capacity of the crop was broadly similar to that of grass, and lower than that for other legume crops (Fychan *et al.*, 1998).

Table 2. *Chemical composition of field bean silage after 90 days ensiling (all values g/kg DM unless otherwise stated)*

	10 weeks		12 weeks		14 weeks			Significance[†]	
	Control	Inoc	Control	Inoc	Control	Inoc	s.e.d	H	I
DM (g/kg)	165	169	216	218	197	199	6.1	***	
pH	4.18	3.96	3.91	3.70	3.83	3.57	0.070	***	***
NH_3-N (g/kg N)	98.6	67.1	79.4	54.2	81.7	47.1	6.67	**	***
Crude protein	225	222	205	198	205	202	9.7	*	
NDF	401	405	446	438	436	417	17.3	*	
Starch	12.1	13.8	34.6	37.8	80.4	77.8	7.15	***	

[†]H = Harvesting Date; I = Additive Treatment; * = $p<0.05$; ** = $p<0.01$; *** = $p<0.001$
There were no significant Harvesting Date x Inoculation Treatment interactions

The field beans were difficult to wilt, and although harvesting date significantly influenced the DM content of the 90 day silages (Table 2) the values recorded were comparatively low for all treatments. Both delaying the harvesting date and the use of an inoculant lowered the pH of the resultant silage. The ammonia content was also reduced by the application of an inoculant, and was significantly lower for silage prepared from forage cut at 12 and 14 weeks growth than for silage made from the first cut. However the crude protein content was highest for the 10 weeks growth silage.

Conclusions

In this study delaying the harvest until 14 weeks growth (pod fill – growth stage 207) gave the highest DM and CP yields and also a higher starch content compared to the earlier harvesting periods. Whilst a satisfactory fermentation was achieved for all treatments, the pH and ammonia results indicate an improved fermentation of field beans can be achieved by applying an inoculant. Controlling diseases and insect infestations in the field bean crop required multiple applications of fungicide and insecticide.

Acknowledgements

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Effects of harvesting date and application of a bacterial inoculant on the fermentation characteristics of forage pea silage.

Introduction

There is a renewed interest in the use of forage peas as a high yielding short term crop with potential for a high crude protein content. Previous work (Potts, 1980) has shown that stage of growth at harvest has considerable effect on DM yield. However, there is limited information available in the literature on the ensiling potential of forage peas. The aim of this study was to compare the yield and chemical composition of forage peas harvested at three different stages of growth, and ensiled with and without an inoculant.

Materials and Methods

Forage peas (*Pisum sativum*) were drilled on 29 April 1998 at a seed rate of 212 kg/ha. A compound fertiliser (P_2O_5, K_2O) was applied to the seedbed to achieve phosphate and potash indices of 3. A pre-emergent spray, Senate (terbutryn + trietazine), was applied at a rate of 4 l/ha on the 2 May. The crop was cut at three different growth stages; first pod set (204), flat pod (205) and pod fill (207), which corresponded to 10, 12 and 14 weeks growth respectively. The forage was harvested at a height of 100 mm using a Haldrup plot harvester. Samples of the forage as harvested were analysed for dry matter (DM) and water soluble carbohydrate (WSC) contents and buffering capacity. The forage was then artificially wilted for a period of 24 h using a forced draught cold air drier before being chopped and ensiled in 10 kg silos. Half the forage was ensiled untreated (Control) whilst the other half was treated with an inoculant (Live System; Genus Ltd, UK) applied at a rate of 10^6 CFU (*Lactobacillus plantarum*) per gram fresh matter (FM). Three replicated mini silos of each treatment were incubated for a period of 90 days. The silos were then opened and representative sub-samples analysed for DM, ammonia nitrogen (NH_3-N), crude protein (CP) (N x 6.25), neutral detergent fibre (NDF) and starch contents, and pH.

Results and Discussion

The yield and key chemical characteristics of the freshly cut forage pea crop are shown in Table 1. There was a trend for increasing DM yield up to 12 week growth, however, the crop had lodged by 14 week growth and resulted in a DM yield similar to the 10 week growth. The DM content of the forage was low at 10 and 12 weeks growth, but had increased significantly by 14 weeks growth, possibly due to the onset of senescence. The crop at 10 weeks growth had a lower WSC content and higher buffering capacity than the later cuts.

Table 1. *Production and chemical composition of forage peas cut at different growth stages*

	10 weeks	12 weeks	14 weeks	s.e.d	Sign.†
Dry matter (g/kg)	152	154	206	10.0	*
WSC (g/kg DM)	156	238	211	21.1	*
Buffering capacity (meq/kg)	276	223	219	19.9	
Yield (kg DM/ha)	5593	6172	5596	279.1	

† * = $p<0.05$

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Table 1. Chemical composition and digestibilities of silages, % (DM basis)

	DM	OM	CP	EE	CF	NFE	CA	ME,MJ/kgDM
AP+CLM	28.1	85.5	18.5	3.3	25.1	38.6	14.5	8.7
SBP+CLM	34.1	79.8	24.3	1.5	18.6	35.5	20.2	9.8
TP+CLM	25.6	72.3	16.7	3.4	31.8	20.4	27.7	6.3
Digestibilities								
AP+CLM	55	62	61	45	25	92	-	-
SBP+CLM	76	78	78	74	74	86	-	-
TP+CLM	52	54	66	78	25	83	-	-

DM= dry matter; OM= organic matters; CP= crude protein; EE= ether extracts; CF= crude fibre; NFE= nitrogen free extract; CA= crude ash; ME= metabolizable energy.

Table 2. Results from the feeding trial with fattening lambs

	Control**	AP+CLM	SBP+CLM	TP+CLM	Sem	Sig
Total liveweight gain, kg	16.2^{a}	14.8^{a}	15.1^{a}	10.3^{b}	0.674	*
Average daily liveweight gain, g	289^{a}	265^{a}	269^{a}	184^{b}	11.859	*
Average daily silage DM consumption, g	1279^{a}	1211^{ab}	1116^{ab}	1040^{b}	39.582	*
Feed efficiency, kg	4.42^{a}	4.57^{a}	4.15^{a}	5.64^{b}	0.203	*

** Concentrate feed mixture was offered ad libitum

a,b= means with different supercripts, within rows, differ significantly ($P<0.05$)

sem= standart error mean, significance tested at $P<0.05$ (*)

Ensiled liquid potato by-product for dairy cows in early lactation

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By-products from the human food industry often provide valuable nutrients for ruminant livestock at low cost. Liquid potato feed (LPF) is one by-product, produced from a short burst of high pressure steam which ruptures the outer cells of the potato, thus providing a 'skinned' potato for processed foods. LPF has a high proportion of potato skin giving it a higher protein content than potato in general, however, the LPF also has a high water content with a dry matter (DM) of only 120 g/kg. The LPF is delivered to farm in bulk tanker and stored in an enclosed silo.

In this trial the effects of including ensiled LPF as a partial replacement for concentrates in a mixed ration based on grass and maize silage were studied with dairy cows in early lactation. Twenty-four cows were randomly assigned (balanced for calving date, parity, liveweight and previous milk yield) to either a treatment (POT) or a control (CON) group. Both groups were initially offered a common diet to allow covariance adjustments to be made if the inital parameters were not identical. The diets were then altered gradually over a two-week period until the diets offered in Period 1 (POT = 50 kg LPF cow^{-1} day^{-1}) were achieved. After 20 days the diets were adjusted to the Period 2 specification 1 (POT = 30 kg LPF cow^{-1} day^{-1}) and offered for 46 days (Table 1). All diets were formulated to be identical in metabolisable energy (11.7 MJ ME kg DM^{-1}) and crude protein (160 g kg DM^{-1}). Minerals were included at 100 g cow^{-1} day^{-1} and 4 kg cow^{-1} day^{-1} of 18% dairy compound was given in the parlour.

The cows were housed in a shed which comprised cubicles with a feed barrier at the front of each cubicle and a dunging passage with access to water troughs behind the stalls,. This system enabled the cows to eat when in the cubicle area. Group feed intake and water consumption were recorded daily, with daily recording of individual milk yields. Individual samples of milk were taken once per week from consecutive afternoon and morning milkings and for analysis of fat, protein and somatic cell count. Faecal pH and dry matter were assessed for each animal at 14-day intervals. Cow liveweights were measured and condition scores were assessed at the beginning and end of each period.

The dry matter intake (DMI) of cows given the control diet was higher in both periods than for those given the POT diets by 7.1 and 4.5 % respectively (Table 2). However, the total intake of water was relatively constant, with the cows given the CON diet drinking more water from the water trough than those given the POT diet. Yields of milk, milk fat and protein were lower for the POT group than for the CON group in Period 1 ($p<0.05$) (Table 3). However, in Period 2 only the milk fat and milk yield were significantly lower ($p<0.05$). There were no significant differences between the two groups in faecal pH, faecal dry matter content, liveweight or condition score.

Table 1 Diet composition, proportion of total dry matter (fresh matter)

	Pre-Experiment	Period 1		Period 2	
		POT	CON	POT	CON
Grasss silage	0.227	0.407	0.437	0.394	0.422
	(0.256)	(0.354)	(0.716)	(0.442)	(0.695)
Maize silage	0.267	-	-	-	-
	(0.262)	-	-	-	-
Liquid Potato Feed	0.144	0.328	-	0.222	-
	(0.279)	(0.574)	-	(0.430)	-
Wet maize gluten	0.155	0.019	0.047	0.018	0.054
feed	(0.146)	(0.016)	(0.063)	(0.016)	(0.070)
Wheat	0.152	0.050	0.228	0.108	0.139
	(0.043)	(0.012)	(0.102)	(0.036)	(0.090)
Soyabean meal	0.054	0.050	0.101	0.086	0.091
	(0.014)	(0.012)	(0.042)	(0.026)	(0.040)
Citrus pulp	-	0.115	0.154	0.117	0.185
	-	(0.026)	(0.063)	(0.034)	(0.080)
Straw	-	0.025	0.028	0.050	0.053
	-	(0.006)	(0.011)	(0.014)	(0.023)

Table 2 Voluntary intakes of feed DM (DMI), feed fresh weight (FWI) and water, mean (s.d)

		Period 1		Period 2	
		POT	CON	POT	CON
Total DMI	kg day^{-1}	19.4 (0.7)	20.9 (0.6)	19.8 (0.9)	20.7 (1.0)
Total FWI	kg day^{-1}	78.7 (3.1)	46.9 (1.3)	63.7 (2.6)	47.3 (2.9)
Water Drunk	kg day^{-1}	46.4 (5.6)	75.8 (4.4)	61.5 (5.8)	77.9 (6.1)
Total intake	kg day^{-1}	125.2 (6.6)	122.7 (5.1)	125.2 (6.6)	125.1 (7.1)

Table 3 Covariance adjusted milk yield and milk composition.

		Period 1				Period 2			
		POT	CON	s.e.m	sig.	POT	CON	s.e.m	sig.
Milk Yield	kg day^{-1}	31.7	34.6	0.72	**	30.2	31.8	0.98	NS
Milk Fat	g kg^{-1}	40.0	47.6	1.74	**	41.4	47.5	1.98	*
Milk Protein	g kg^{-1}	30.9	31.3	0.42	NS	32.5	32.6	0.46	NS
Yield of Milk Fat	kg day^{-1}	1.3	1.66	0.08	**	1.27	1.57	0.09	*
Yield of Milk Protein	kg day^{-1}	0.99	1.10	0.04	*	0.99	1.08	0.05	NS
Somatic cell count	'000 cell	100	313	133	NS	123	297	117	NS

It is concluded that the relatively wet POT diet in Period 1 (248 g DM kg^{-1}) was not able to support milk yield at a comparable level to the CON diet (438 g DM kg^{-1}). The mechanism for this effect is not clear, but it could be due to: i) the fresh weight of the diet (i.e. a physical limitation to intake), ii) a nutritional inadequacy or iii) antinutritional factor(s) in the LPF. In Period 2 the POT diet (291 g DM kg^{-1}) was able to support milk production at a similar level to that of the control group (424 g DM kg^{-1}), indicating that ensiled LPF was an adequate replacement for concentrates up to a maximum inclusion between 30 and 50 kg LPF cow^{-1} day^{-1}.

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Nutritive values and fermentation characteristics of barley silage varieties.

The major forage sources fed to cattle in the province of Alberta, Canada are alfalfa and small grain cereal silages. Although alfalfa is an excellent forage, environmental conditions do not always favour its growth. Small grain cereals can be grown in a wide range of climatic and soil conditions, are annual crops, and are not subject to winter kill. Improvements in forage yields and in the efficiency of utilization of forage are a result of varietal selection and improved forage conservation techniques. The yield potential of many cereal grain varieties has been determined, but their ability to sustain milk production has not been well documented. Good quality forage is essential to achieve adequate digestible energy intake in high producing dairy cows. Forage quality is determined using various criteria, such as CP, NDF, and ADF content. Alfalfa silage tends to be higher in protein and lower in NDF content than cereal silages, thus the feeding value of alfalfa silage is considered superior to cereal silages. The objective of this study was to determine the nutritive quality of three barley varieties harvested as whole crop barley silage. Barley silages were evaluated relative to alfalfa silage.

The barley cultivars were two row Seebe , semi-dwarf Duke and Lacombe which were harvested, wilted and ensiled in plastic bags, at the University of Alberta Research Station. The crops were harvested at the soft dough stage and the Seebe cultivar reached the dough state a week later than the Duke and Lacombe cultivars. On the day of harvest samples were taken and pooled for analysis. After ensiling samples were taken from each silo bag at 2 wk intervals over an 8 wk period and stored at -20 °C until analyzed. The results indicated that DM of Seebe at ensiling and subsequent sampling was higher ($P< 0.0\ 5$) than observed for Duke and Lacombe. Silage DM was not influenced by time post-ensiling. The levels of acetic and propionic acids in Duke silage were greater ($P<0.05$) than observed for the other silages. Lactic acid concentration (g/kg DM) in silages varied from 83.4 (Lacombe) to 90.3 (Seebe). The concentration of this acid at the end of 4 and 8 weeks of ensiling was 95.2 and 89.1 g/kg DM, respectively. CP in the original forage was higher for Duke than Seebe and Lacombe. However, the CP contents of silages were not different. The level of ADIN in the three silages was similar and represented about 15.5% of total N content. The NH3-N concentration were 145.4, 143.5 and 133.2 g/kg of total N for Duke, Lacombe and Seebe silages. NDF contents of fresh forages and silages for Duke, Lacombe, and Seebe were 501, 567, 537 (at time of ensiling), and 507, 541, 507 g/kg DM, respectively. ADF levels in Duke (275), Lacombe (306) and Seebe (289 g/kg DM) silages were similar to that observed at harvesting.

Silages prepared from whole crop barley varieties were evaluated for rumen DM degradation kinetics. High and medium quality alfalfa silages were included for comparison purposes. Three ruminal cannulated, non-lactation Holstein cows were utilized. Fresh silages were incubated in situ for 0, 1, 2, 4, 8, 16, 24, 48, 72, 96, and 144 h. Rate of DM degradation was similar among the barley silages and medium quality alfalfa silage. However, high quality alfalfa silage showed a higher ($P<0.05$) rate of DM degradation than barley or medium quality alfalfa silage. Silage protein was highly soluble (78.3%) and degradable (87.1%). The rate of CP degradation of alfalfa silages was more than twice that observed for barley silages. The CP

disappearance was different among silages up to 16 h of rumen incubations, but after 16 h of rumen incubation, no statistically significant differences were observed.

Four Holstein heifers (546 ± 7.07 kg) cannulated in the rumen and duodenum were fed a diet containing 20% concentrate and 80% of each of the test silage (Duke, Lacombe, Seebe, and alfalfa) in a 4 × 4 Latin square design experiment and each period lasted 21 days. The concentrate portion of the TMR was based on rolled-barley, trace-mineralized salts, and vitamins. Feed intake and rumen fermentation characteristics including pH, VFA, lactic acid, ammonia N and microbial protein production were measured. The DMI was higher for heifers fed alfalfa silages (9.6 kg/d) compared to heifers fed Lacombe silage (6.4 kg/d), but did not differ for other dietary treatments. Heifers fed Seebe had a numerically higher DMI than heifers fed Duke and Lacombe silages (8.8, 7.0, and 6.4 kg/d, respectively). Heifers fed alfalfa silage had a lower total rumen fill than heifers fed barley silages. Rumen turnover time was numerically higher for heifers fed Duke (21.5 h) and Lacombe (21.9 h) silage compared to heifers fed Seebe (17.3 h) and alfalfa silage (16.0 h). Mean pH of ruminal fluid was higher for heifers fed Lacombe silage (6.71) than for heifers fed Seebe silage (6.57), and no differences were observed for the other dietary treatments. Mean ruminal concentration of ammonia N was not affected by dietary treatment. Total ruminal VFA concentrations were higher for cows fed alfalfa silage than for cows fed Lacombe silage, and no differences were observed among the other treatments. The contribution of individual VFA to the total VFA concentration was influenced by silage source. The whole crop barley silage varieties influenced DMI and rumen fermentation; possibly by their NDF content and digestion characteristics of the fibre. Cows fed barley silage diets had a lower N intake (171 g/d) than cows fed alfalfa (271 ± 23.5 g/d, $P < 0.05$), reflecting the higher N content of alfalfa silage. Total N and non-ammonia N flow at the duodenum were higher ($P < 0.05$) for cows fed alfalfa compared to cows fed Lacombe, but did not differ among cows fed other dietary treatments. Dietary treatments had no effect on ruminal bacterial yields and ammonia and dietary residual N reaching the duodenum.

Twenty early-lactation Holstein cows were blocked according to parity (8 second lactation and 12 >second lactation), date of calving, and milk yield. Cows were assigned to 4 dietary treatments following a 2-wk covariate period and were fed the test diets (Duke, Lacombe, Seebe, and alfalfa silage) for 12-wk. Diets contained 50% concentrate and 50% forage on a DM basis and were fed once daily as a TMR. The DMI of cows averaged 3.03% of body weight and cows fed Lacombe silage had lower DMI (21.4 kg/d) than cows fed Duke (22.0 kg/d) or Seebe (22.7 kg/d). Milk, 4% FCM and milk component yields were not affected by silage type and production responses were greater for cows with a parity greater than two than for cows with a parity of two (37.5 vs. 33.7 kg/d). Milk and 4% FCM yield were 37.0, 33.5; 32.8, 31.4; 34.7, 34.1; and 37.8, 35.4 kg/d for Duke, Lacombe, Seebe, and alfalfa silage, respectively. Milk fat and lactose content were not affected by diet, but milk protein content was lower for cows fed Duke silage (3.06%) than for cows fed Seebe (3.28%) and Lacombe silage (3.32%).

Our research has demonstrated that there are differences among whole crop barley varieties in terms of ensiling fermentation characteristics, ruminal fermentation characteristics, digestion, and utilization by dairy cows, but the animal has a tremendous ability to modulate these differences and maintain relatively constant performance.

Effect of the epiphytic lactic bacteria on the conservation of grass mixture pasture silage

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Introduction

The conservation of a crop as silage is often related to its ensilability in terms of its concentration in water soluble carbohydrates (WSC) and its buffering capacity (BC). However, even with a good ensilability, the conservation is variable. This could be related to a low population of epiphytic lactic bacteria (LAB) as well as the diversity of LAB on the crop when harvest. The aim of this study was to relate the conservation of a crop to the epiphytic LAB population.

Materials and methods

Forages from a grass mixture pasture were harvested three times during the 1996 season. The first and the second harvest were cut during the first growth cycle at vegetative stage and fully heading stage respectively. The third harvest was the regrowth of the crop harvested at vegetative stage at the first growth cycle. Dry matter (DM), WSC and BC of the forage ensiled are shown in table 1. Forages were harvested without wilting with a forage harvester. 15 kg of chopped forages untreated or treated with a commercial inoculant (*Lactobacillus plantarum*, *Pediococcus acidilactici*, 1 x 10^5 UFC g^{-1} FM) were put into pails lined with sterile plastic bag. Each replicate correspond to a different load from the field. The silos were opened after 60 days of incubation. For microbiological analysis, 20 g of chopped forage was homogenised in 200 ml of peptone water (2 g l^{-1}) as described by Östling and Lindgren (1995). LAB were enumerated on MRS (BBL) agar supplemented with cycloheximide (0,4 g l^{-1}). For each harvest, 150 colonies were used to established the epiphytic LAB population. LAB were further characterised to the genera according to Sharpe (1979).

Table 1. Chemical characteristics related to ensilability of the grass mixture pasture

Harvest	DM g kg^{-1}	WSC g kg^{-1} DM	BC meq kg^{-1} DM
1	211	83	415
2	277	82	334
3	281	74	369

Results

MRS allowed growth of an important proportion of Gram negative bacteria. Therefore, real epiphytic LAB population was lower than what was enumerated on MRS (table 2). Numbers of LAB varied during the season being lowest at the first harvest and highest at the second harvest (table 2). However, quality conservation, as measured by butyric acid and ammonia content, was lowest in harvest 2 (table 3). Among epiphytic LAB, homofermentative bacteria were dominant at each harvest but the proportion was lower at the first harvest with 63% compared to 83% for other harvests (table 2). Consequently, content of acetic acid was low in every harvest probably reflecting the high proportion of epiphytic homofermentative LAB. Nevertheless, the highest proportion of heterofermentative LAB dominated by *Leuconostoc* was observed in harvest 1 where we also observed highest concentration of acetic acid (table 3). For all harvests, inoculation increased the lactic acid content, decreased the pH and ameliorated other chemical parameters probably because the epiphytic LAB population was

not very efficient. However, the butyric acid content was not completely eliminated suggesting that other factors such as sugar content or type of sugar was limiting.

Table 2. Epiphytic lactic acid bacteria on the grass mixture pasture to be ensiled.

Harvest	Total CFU on MRS g^{-1} FM	LAB (% of total CFU)	Other	Epiphytic LAB g^{-1} FM	Population of epiphytic LAB
1	1.3×10^4	63	38	8.3×10^3	17%, ho *Lactobacillus*, 40%, ho cocci, 6%, *Pediococcus*, 31%, *Leuconostoc*, 6%, he *Lactobacillus*
2	2.5×10^5	69	31	1.7×10^5	18%, ho *Lactobacillus*, 56%, ho cocci, 9%, *Pediococcus*, 9%, *Leuconostoc*, 8%, he *Lactobacillus*
3	6.4×10^4	76	24	4.9×10^4	35%, ho *Lactobacillus*, 48%, ho cocci, 5%, *Leuconostoc*, 12% he *Lactobacillus*

FM= fresh matter, ho= homofermentative, he= heterofermentative

Table 3. Chemical composition of grass mixture pasture silages.

Harvest	PH	Lactic	Acetic	n-Butyric	Ethanol	$N\text{-}NH_3$	Soluble-N
		g kg^{-1} DM				(% total N)	
1- U	4.4	77.5	10.8	3.7	6.9	5.9	50.8
1- I	4.1	101.7	9.6	1.9	4.6	5.0	48.8
2- U	4.6	44.4	4.9	13.2	7.9	9.0	53.3
2- I	4.2	68.2	5.0	8.3	6.4	7.7	51.4
3- U	5.3	32.9	4.4	4.4	7.3	9.4	53.2
3- I	4.8	45.8	6.8	3.9	5.7	9.6	50.6

U= untreated; I= inoculated.

Conclusions

In this trial, quality conservation could not be related only to the number of epiphytic LAB. The composition and the efficiency of the epiphytic LAB was important. To ensure high quality conservation of silage, good ensilability of the crop is also necessary.

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Fermentation quality of silage made from alfalfa, grass or an alfalfa-grass mixture

Introduction

The main aim of this work was to investigate the effect of the addition of grass to alfalfa, resp. the comparison of fermentation quality of grass, grass-alfalfa mix and alfalfa silages. It is universally known that alfalfa has bad ensiling qualities with a high nitrogen and a low WSC (water soluble carbohydrate) content. The restriction of two procceses, respiration (Fujita at al., 1995) and proteolyses (Takahashi, 1998) is important. In the Czech Republic this problem is solved mainly by using biological silage additives. An alternative way is to grow alfalfa in mixture with grass. This work follows the research papers Takahashi (1998) and Loucka et al. (1995, 1998) and creates a linking part in The Czech-Japanese project ME 194.

Material and Methods

The following plants were used for testing: a ryegrass-fescue hybrid var. Perun (*Lolium multiflorum* L. x *Festuca pratensis* Huds.), alfalfa var. Bobrava (*Medicago sativa* L.) and their mixture (29 % of grass). The plants were grown in the same field (0.5 ha) under the same agrotechnical and bioclimatic conditions. The first harvest of grass, grass-alfalfa mixture and alfalfa was preserved in special tube silos (10 litres of volume) without additives. The field wilting of grass took 20 to 22 hours and the field wilting of grass-alfalfa mixture and alfalfa 26 to 28 hours. The tube silos (each experiment was repeated 8 times) were stored at a temperature of 25 °C. Silage samples were analyzed after 3 months of fermentation. For the estimation of the nutritive value and fermentation parameters, routine analytical techniques were employed.

Results

Alfalfa needs a longer time for wilting than grass. In spite of the fact that alfalfa was wilted 6 hours longer than grass, the grass had a significantly higher content of dry matter than alfalfa.
The content of crude protein of alfalfa was twice as high and the content of WSC only about 1/3 lower than grass. According to the crude fibre content both grass and alfalfa was cut at the optimum stage of maturity.

Table 1: Nutritive values of fresh forages

		Grass	Grass-alfalfa mixture	Alfalfa
Dry matter	[g/kg]	378 a	279 b	250 b
CP	[g/kg DM]	101 a	182 b	201 b
ADF	[g/kg DM]	304 a	307 a	276 a
NDF	[g/kg DM]	526 a	407 ab	334 b
PDI	[g/kg DM]	61 a	93 b	92 b
NEL	[MJ/kg DM]	6.7 a	5.8 ab	5.3 b
NEG	[MJ/kg DM]	6.9 a	5.7 ab	5.2 b
WSC	[g/kg DM]	90 a	73 ab	61 b

When alfalfa is ensiled without additives and at a low DM content (25 %), it is not possible to expect a good fermentation quality (pH 4.8, LA/VFA 1.2 %, proteolyses 10.2 %). With the mixture of grass and alfalfa (29 % grass) we found a significantly higher level of pH 4.4, LA/VFA 2.2 %, proteolyses 5.4 % than with alfalfa alone.

Table 2: Silage parameters

	Grass	Grass-alfalfa mixture	Alfalfa
Dry matter [g/kg]	362 a	255 b	241 b
CP [g/kg DM]	107 a	185 b	207 b
ADF [g/kg DM]	327 a	342 a	343 a
NDF [g/kg DM]	530 a	424 ab	361 b
PDI [g/kg DM]	60 a	75 b	75 b
NEL [MJ/kg DM]	5.9 a	5.2 ab	4.8 b
NEG [MJ/kg DM]	5.8 a	4.9 ab	4.5 b
..... pH			
Lactic acid [%]	1.4 a	1.5 a	1.8 b
Acetic acid [%]	0.4 a	0.5 a	1.1 b
Lactic acid/VFA [%]	3.1 a	2.2 a	1.2 b
Proteolyses [%]	2.5 a	5.4 a	10.2 b

Mixture (29 % grass + 71 % alfalfa), CP = crude protein, ADF = Acid Detergent Fiber, NDF = Neutral Detergent Fiber , PDI = Protein digestibility at the intestine, NEL = Net Energy for Lactation, NEG = Net Energy for Growth, WSC = Water Soluble Carbohydrates, VFA = Volatile Fatty Acids.

Conclusions

The fermentation quality of the silage was better when alfalfa was ensiled together with grass (29 % grass) compared with alfalfa alone.

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Key words: silage, alfalfa, grass

Technical Considerations in Extraction of Lactic Acid from Silage

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Keywords: Silage, Lactic Acid, Extraction, Electrodialysis, Yields

1. INTRODUCTION

The emergence of new markets for lactic acid application namely, biodegradable thermoplastics, has intensified the search for cost effective production systems. Agricultural residues of plant origin constitute a biomass resource that can be used as a low cost substrate for solid state fermentations. The spontaneous lactic acid fermentation of lignocellulosic residues during silaging processes makes the resultant silage a strong potential source of lactic acid. With substrate costs contributing immensely towards conventional production costs, the extraction of lactic acid from silage becomes an interesting proposition both from an economic and environmental point of view. However, due to the heterogenous nature of silage microflora, the selective recovery and concentration of lactic acid requires delicate unit operations to produce lactic acid of acceptable quality as dictated by the user industries. Investigations into lactic acid extraction, concentration, and purification are reported on in this paper. The influence of process yields and economics on scale up is briefly highlighted.

2. EXPERIMENTAL

The various preparations of whole corn and grass silage used in this work were sourced from surrounding farmers in the province of Lower Austria. Deionised water at ambient temperature was the extracting solvent, at solvent to silage ratios of 2:1 and 3:1. The extractions were conducted in laboratory scale batches. Manual press filtration was used to recover the crude lactic acid extract. The particulate extract was clarified by either centrifugation or ultrafiltration before recovering and partially purifying the lactic acid by monopolar electrodialysis. In one variation, the clarified extract was first vacuum concentrated at 90°C before electrodialysis. The electrodialysis unit was acquired from Eurodia Industrie, France. The ED stack had 8 cell pairs with an effective membrane area of 2 dm^2/ sheet. The maximum limiting current density recommended for the stack is 450 Am^{-2}. Analytical determination of lactic acid, acetic acid, propionic acid, sugars and alcohols was done by HPLC (HP 1050C) using a Merck polyspher OA KC column and RI (HP1047 A) detectors.

3. RESULTS AND DISCUSSION

3.1. Extraction of Silage

Extractions were conducted at ambient temperature reflecting positively on the technical and economical aspects of lactic acid extraction from silage. Extract lactic acid (LA) concentration ranged from 6g/l to 14g/l and the yields ranged widely from 40 to 96gLA/kg dry matter(DM) of silage. More concentrated extracts (40 g/l lactic acid at 63gLA/kgDM) have been obtained with mechanical pressing without solvent addition (Steinmüller and Eibensteiner, 1994). The lactic

acid concentrations varied depending on source indicating the need to standardise the ensiling processes if a standard extraction protocol is to be commercially practical. Higher solvent extraction volumes resulted in dilute lactic acid extracts which therefore require energy to concentrate. Proper mixing and contacting of the solvent and silage is however important to ensure consistent lactic acid yields. Press filtration of the hydrated silage gives a very crude extract with suspended plant mucilage. Extract (volume) recoveries from the manual press filter ranged from 60% and 73%. Higher volume recoveries of upto 90% can be achieved with hydraulic presses. However, beyond 90% the extraction of gummy mucilage becomes predominant presenting purification problems. Super critical fluid extraction (SFE) trials by Danner (1998) gave 13gLA/kg DM lucerne/barley silage, a low yield attributed to poor silage preparation. Although the SFE extract is relatively clean, the process is complicated, deemed capital intensive, and therefore difficult to scale up.

3.2. Treatment of Crude Extract

Centrifugation did not remove all the particulate and colloidal material which rendered the extract chemically unstable. To clarify the extract further, an adsorption filtration step using activated charcoal was incorporated into the process. This treatment significantly reduced dissolved colour and finely suspended particules retaining 80 to 90% of the desired organic acids. However, complete colour removal may prove costly and this step may be omitted for lactates destined for road deicing and as animal feed supplement. Ultrafiltration with tubular membranes yielded a stable particulate free permeate with a consistent colour and recovery yields averaged a satisfactory 80% leaving behind a concentrate with colloidal particulate material.

3.3. Electrodialysis (ED) of Pretreated Extract

Current efficiencies (CE) ranged from 40% to 60% for the ultrafiltered extract indicating a significant improvement from the 30% which was observed with centrifuged extract. Electrodialysing the extract straight after the centrifugation step presents operational problems from membrane fouling by dissolved organic macromolecules. This phenomenon not only reduces the efficiency of the ED, but also results in an increased demand for thorough and frequent membrane cleaning regimes both of which significantly reduce the operating life span of the membranes. Current efficiencies of 50% are common in conventional large scale ED applications although laboratory scale units can reach CE of upto 90%. Efficiencies with silage extract could be enhanced further by a (costly) ion exchange step to lower the level of cations and anions which compete with lactates for transportation. Centrifugation could be considered as a pretreatment step to minimise membrane fouling and channel blocking during the ultrafiltration. Concentration of the extract before electrodialysis did not significantly enhance performance.

4. CONCLUSIONS

The technical feasibility and economics of lactic acid extraction from silage could be improved by increasing lactate yields through standardised ensiling processes to ensure high and consistent output productivities. The use of additives namely, lactic acid bacteria inoculants and hydrolytic enzymes to enhance the fermentation should be promoted to make silage a competitive source of lactic acid for use in animal feeds and as a road deicing agents among other applications.

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Silage quality is affected by lactic acid bacteria strain combinations and dosage

Introduction

Silage quality is influenced not only by the composition of the harvested crop and by silage management but also by the ensiling characteristics of the fermenting lactic acid bacteria (LAB). Since epiphytic LAB vary considerably in number and ensiling characteristics, it is helpful to use specifically chosen silage inoculants as starter cultures. In combination with good silage management, these inoculants can result in silages with reduced losses and higher feed value.

Over several years, we have screened a worldwide collection of LAB for a range of different ensiling characteristics within our silage inoculant development program. Our screening has shown that there are significant differences in the characteristics of these organisms.

Method and Materials

The ability to reduce pH was determined by incubating single strains in a crop-specific, plant extract broth. After 6 h of incubation at 37°C the terminal pH was measured.

Osmotolerance was estimated by growing LAB strains in modified MRS with 100 g KCl/l. This would correspond to approximately 45% DM of an ensiled crop. After 3 days of incubation at 37°C the CFU/ml were estimated by serial dilution on MRS plates.

Substrate utilization was determined using the API50 CH or Biolog GP system. Cells were grown overnight on the appropriate media, harvested, washed and resuspended in minimal media. Wells were inoculated according to the manufacturer's directions, incubated at 37 C, and scored daily for substrate utilization and growth.

For evaluating the effect of single strains, strain combinations, and inoculation rate on aerobic stability or *in-vitro* dry matter digestibility the crop was ensiled in 2.8 l PVC silos and stored at 20°C for 45 to 120 days. Uninoculated, ensiled material was used as negative control silage. Silos were opened and samples taken for further analysis. during the fermentation process.

Aerobic stability was determined by placing approximately 1 kg silage in a plastic lined polystyrene container with a buried thermocouple. Containers were placed in a constant temperature incubator at 27 C and the temperature of the silage was recorded every three hours for seven days by an automated datalogger.

In-vitro dry matter digestibility was measured using a modification of the Tilley-Terry method. Dried silage samples (500 mg) were incubated with strained rumen fluid and the digestion of the sample was determined by measuring the water displaced by gas produced during the fermentation. Water displaced was recorded every two hours for 24 hours to estimate the digestion rate. The slope of the log phase was calculated and expressed as percent of a standard. The standard was used throughout all tests to control for variability in rumen fluid composition.

Results

In a series of tests under laboratory conditions with 235 strains, we observed differences between strains in their ability to lower pH. The majority of strains were able to decrease the pH to pH 4.5. Only a minor number of strains could lower the pH to pH 4.1 in the respective plant extract.

Strains of LAB, even within the same species, exhibit considerable diversity in their ability to utilize sugars. Some strains can metabolise a variety of substrates while other are only capable of utilizing a limited subset. It appears from our testing that the utilization patterns may be inducible and highly dependent upon the previous substrates available for growth.

Growth of LAB strains on media with decreased a_w indicated that there is a difference in osmotolerance between strains. Strains having a higher osmotolerance should perform better in high DM silages.

Silage inoculated with single or combinations of LAB had different aerobic stability and *in-vitro* digestibility. The differences did not appear to be additive and could not be predicted from performance of the individual strains alone. Different strain combination had specific, optimum dose levels that could be determined in dose titration studies.

Conclusions

In our product development work for new silage inoculants, differences between strains of LAB were observed. In a series of tests under laboratory conditions we observed differences between the ability of strains to lower pH and to grow at low a_w. Sugar utilization can be variable between strains of the same species and can change depending upon the previous growth substrate. It was further demonstrated that silages inoculated with different strains and combinations resulted in different aerobic stability and *in-vitro* digestibility.

From these results it is clear that LAB are a diverse group of organisms and that only a few possess suitable properties for use in commercial silage inoculants.

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Acidic or alkaline preservation of whole-plant cereals

It is shown that conservation of whole-plant cereals, such as whole-plant oats, is possible either by lactic acid fermentation or by chemical conservation with urea. Ammonia released from urea by spontaneous hydrolysis causes an alkaline reaction and acts against anaerobic deterioration (butyric acid fermentation) as well as aerobic spoilage (moulds and yeast). Furthermore, it can be expected that ammonia will improve silage digestibility and offers additional nitrogen for protein synthesis by rumen microorganisms.

Material and Methods

Whole-plant oats was harvested at late dough stage at a dry matter concentration (DM) of about 600 g kg^{-1} and subsequently ensiled in small bunker silos (6 m^3). For comparison of both procedures, treatments were as follows:
(A) without additives, (B) KOFASIL LIQUID (3 l t^{-1} herbage), (C) urea (30 kg t^{-1} herbage) and (D) urea (30 kg t^{-1} herbage) and heating to 60 °C for one month.
Heating was included to simulate the typical increase in temperature which normally occurs by urea treatment in big silos. Subsequent to a 6 month storage period, silages were fed to sheep and beef cattle (steers) for determination of digestibility, energetic feeding value and voluntary feed intake.

Results

All silages were well preserved and neither contained butyric acid nor mould. Results on fermentation pattern, nutrient composition, digestibility and energy concentration are given in Table 1. Urea treatment improved digestibility of organic matter (DOM) and concentration of metabolizable energy (ME).

Table 1: Fermentation pattern and feeding value of acidic and alkaline preserved whole-plant oat silages for sheep
(crops harvested at late dough stage, digestibility measured in sheep)

	Treatment			
	A without additives	B Kofasil liquid (3 l t^{-1})	C urea (30 kg t^{-1})	D urea (30 kg t^{-1}) (heated to 60 °C)
DM [g kg^{-1}]	661	661	624	684
pH	6.09	6.12	8.82	8.13
Fermentation pattern [g kg^{-1} DM]				
Lactic acid	8.0	8.6	1.1	1.5
Acetic acid	6.8	12.1	19.1	21.4
Butyric acid	0	0	0	0
NH_3	0.68	0.57	20.0	31.3
Urea	-	-	0.64	0.58
Crude protein	99	130	226	215
DOM [%]	59.3	61.1	64.9	70.8
ME [MJ kg^{-1} DM]	8.37	8.84	9.30	10.01

Analysis for aerobic stability proved the two alkaline silages (treatment C and D) to be very stable, whereas the acidic silages (treatment A and B) and mixtures of grass silage and alkaline silages deteriorated within few days upon exposure to air.

In contrast to cattle, alkaline silages could be fed as sole feedstuff to sheep. Admixing grass silage, however, resulted in a good acceptance of all whole-plant oat silages by cattle. Data on digestibility, energy concentration and voluntary feed intake of these mixtures determined with cattle (400 kg live weight) are shown in Table 2. The total silage diet contained 35-37 % whole-plant oat silage on a DM basis. Urea treatment enhanced DOM and ME concentration of these mixtures in trials with cattle. Additionally, an increase in voluntary feed intake was observed.

Table 2: Feeding value and voluntary feed intake of mixtures of grass and whole-plant oat silages by cattle

	Treatment			
	A	B	C	D
DM [g kg^{-1}]	425	407	401	422
DM proportion of whole-plant oat silage in mixture [%]	37.2	35.9	35.3	36.9
Crude protein [g kg^{-1} DM]	130	134	176	168
DOM [%]	71.5	71.6	74.5	74.0
ME [MJ kg^{-1} DM]	10.06	10.06	10.47	10.38
Daily DM intake [kg $animal^{-1}$]				
grass silage[1)]	5.25	5.36	5.57	5.65
whole-plant oat silage	3.11	3.00	3.04	3.30
mixture	8.36	8.36	8.61	8.95
Daily energy intake [MJ ME $animal^{-1}$]	84.10	84.10	90.15	92.90

[1)] grass silage: DM [g kg^{-1}] = 273; DOM [%] = 80.1; CP [g kg^{-1} DM] = 147; ME [MJ kg^{-1} DM] = 11.19

Conclusions

Summarising the results, both acidic and alkaline preservation of whole-plant cereals is possible. However, urea treatment can additionally increase digestibility and, admixed to acidic silage prior to feeding, potentially improve voluntary feed intake.

Key words: whole-plant cereal silages, whole-plant oat silages, acidic and alkaline preservation, urea, energetic feeding value, voluntary feed intake, sheep, cattle.

Solid v liquid urea -ammonium sulphate as additives for ensiled forage maize

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A liquid solution of urea-ammonium sulphate may be used as an additive for ensiled forage maize with similar effects on the fermentation process as that produced by the addition of liquid urea solution alone (Wilkinson and Selmer-Olsen, 1999). But the quantities of liquid to be added are considerably greater than those used in the treatment of grass crops. For example, at 5 kg urea per tonne of fresh crop weight (about 15 kg per tonne of DM, the amount needed to raise the crude protein equivalent of the silage to about 140g/kg DM), the amount of urea solution added is almost 12 litres per tonne fresh crop weight. If ammonium sulphate is added at 1.5 kg/tonne fresh weight to rectify the decrease in total sulphur (S), and additional 3 litres of 40% m/m solution is required.

The development of a solid preparation of urea-ammonium sulphate in feed-grade prilled form introduces the possibility of reducing substantially the quantity of additive to be applied. This experiment was undertaken to test the hypothesis that solid and liquid preparations of urea-ammonium sulphate have similar effects on the fermentation and nutritional composition of ensiled forage maize.

Forage maize (cv Nancis) was harvested by Claas self-propelled forage harvester at the Leeds University Farm on 21 October 1997. Average length of chopping was 10 mm. Immediately after harvest, the crop was weighed into lots of 10 kg fresh weight and each lot was spread out evenly in a thin layer on a clean concrete floor. Additives were applied to the crop at the levels indicated below by sprinkling (solid) or by spraying via an air-pressurised Hozelock 2 hand sprayer of 1.25 litres capacity(liquid). The liquid treatments were U1AS and U2AS as in Wilkinson and Selmer-Olsen (1999) i.e. 2.5 kg urea/tonne FW + 1.5 kg AS/t FW (U1AS) or 5 kg urea/tonne FW + 1.5 kg AS/t FW (U2AS). The solid treatments were 2.5 or 5.0 kg/t of a prill containing 70% by weight of urea and 30% by weight of ammonium sulphate. Thus the total quantities of additive applied were lower for the solid urea-ammonium sulphate than for the liquid urea-ammonium sulphate, by about 1.5 kg/tonne FW.

The results are presented in Table 1 as the main effects of solid v liquid urea-ammonium sulphate. The only significant interaction between physical form and level of addition was for water soluble carbohydrate content (WSC) which was lower ($P<0.05$) for the solid additive (5.8 g/kg DM) than for the liquid additive (7.8 g/kg DM) at the lower level of addition only.

There was no effect of physical form on dry matter content, or on the concentration of lactic acid in the silage. pH was lower, and the contents of acetic acid and ethanol higher for silages made with liquid urea-ammonium sulphate than for those made with the solid additive. Differences between silages made with liquid or solid additives in the contents of starch, WSC and NDF were generally small. Neutral cellulase digestibility (NCD) was higher ($P<0.01$) for the silages made with the solid than those made with the liquid additives, suggesting that possibly there was less fermentation loss in the silages made with the solid urea-ammonium sulphate than with the liquid additive.

A possible explanation for the higher concentrations of acetic acid and ethanol and the lower values for NCD in the silages made with the liquid additive was that the fermentation was buffered at the outset to a greater extent by the liquid than by the solid additive, but this hypothesis requires confirmation in further work.

Table 1 **Effect of physical form of urea -ammonium sulphate on fermentation and nutritive value of ensiled forage maize (g/kg DM unless otherwise stated)**

	Liquid	Solid	s.e.m.	Sig.
DM[1] (g/kg)	306	302	1.9	**NS**
pH	4.08	4.24	0.02	***
Lactic acid	41.0	40.0	0.9	**NS**
Acetic acid	9.0	3.7	0.9	**
Ethanol	34.4	31.1	0.9	*
Buffering capacity (meq/kgDM)	551	443	17.6	**
NCD[2]	718	755	6.4	**
Starch	110	135	14.3	**NS**
WSC	7.0	5.4	0.4	*
NDF[3]	339	329	7.6	**NS**
Total N	21.0	19.2	0.78	**NS**
TSN[4] (g/kg total N)	547	580	15.2	**NS**
NH_3N (g/kg total N)	72.1	64.1	2.8	**NS**

[1] Corrected for volatiles [2] Neutral cellulase digestibility
[3] Neutral detergent fibre [4] Total soluble N

It is concluded that urea-ammonium sulphate may be added to forage maize in both liquid and solid forms. On the basis of this experimnet the solid form apprears to have significant advantages over the liquid form, both in terms of the lower bulk of material to be applied per tonne of crop and also in terms of improved crop preservation.

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Influence of DM content on silage fermentation of sulla (*Hedysarum coronarium* L.) cut at two stages of maturity.

Key words: *Hedysarum coronarium* L., silage, wilting, conservation quality.

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Introduction

Sulla (*Hedysarum coronarium* L.) is a short-lived legume that grows well in clay and calcareous, low fertility soils in semiarid Mediterranean environments. It is used for pasture, because of its bloat-safe characteristics due to condensed tannins, or stored mainly as hay. The conservation of this legume as silage may be of particular interest because of its high sugar content, DM yield and good nutritional value in the early stages. The presence of condensed tannins can greatly reduce the N transformation in ensiled forage that results from plant and microbial enzymatic activities (Albrecht and Muck, 1991).

The objective of this study was to assess the effect of wilting on the conservation quality of sulla silages at two maturity stages.

Materials and Methods

A first cutting of sulla cv. Grimaldi was grown near Ancona (43°N lat.) and harvested at early bud (5th May) and early flowering stages (19th May) with a DM yield of 6 and 8 t/ha, respectively. The herbage was ensiled in laboratory silos (2-litre jars) prepared with either fresh or wilted material (4 cm theoretical chop length) with three replications at increasing DM levels ranging from 96 to 454 g/kg FM at early bud stage and from 124 to 450 g/kg FM at early flowering stage. The silos were maintained at 22 ± 2°C and opened after 130 days. The herbage samples were analysed for the ensilability characteristics and *in vitro* organic matter digestibility (OMD, Tilley and Terry method), and the silage for the conservation quality and DM losses. The fermentation DM losses were calculated by multiplying the weight loss (by gases) by 1.4.

Results and Discussion

The herbage at cutting differed for the two stages as far as the DM content (96 and 124 g/kg FM), WSC content (176 and 114 g/kg DM at early bud and early flowering, respectively), BC (389 and 350 meq/kg DM), TN (35 and 30 g/kg DM) and OMD (740 and 650 g/kg OM) are concerned. The higher moisture content of the herbage at early bud almost doubled the wilting time required to reach a DM content of 450 g/kg FM in comparison to the early flowering stage (3.5 *vs.* 2 days of field wilting). The pH at cutting was very similar in the two stages (mean value 5.6). The mean nitrate content was about 0.02 g/kg FM, resulting to be lower than the value (0.1 g/kg FM) considered to be useful to prevent clostridial activity during fermentation (Wieringa, 1966). During wilting, the WSC concentration in the plant water increased with increasing DM content from 19 to 106 g/l at early bud and from 16 to 56 g/l at early flowering, following linear trends (fig. 1).

The fermentation characteristics of the silages are given in Table 1. At the early bud stage the silages with DM content lower than 348 g/kg FM underwent a clostridial fermentation, as shown by the butyric, propionic, and isovaleric acid production, while, at early flowering, the silage fermentation was already very good at 243 g/kg FM. The NH_3-N content was always lower than 100

g/kg TN, except for the direct cut silage at the early bud stage. As observed by others authors (e.g. Albrecht and Muck, 1991) these low values, together with the relatively low NPN content (in comparison to lucerne silages wilted to similar DM content), would suggest that protein was protected against degradation during the ensiling process even in clostridial silages. The more intense the clostridial fermentation, the higher were the DM losses. The DM losses decreased with increasing wilting level showing different linear trends over the two stages of maturity (Fig. 2).

Table 1. Composition and DM loss of silages at early bud and early flowering stages (mean values, g/kg on DM basis except for DM on fresh weight and NH_3-N and NPN on TN).

Stage	Early bud							Early flowering					
DM	97	137	209	228	286	348	457	120	184	243	307	335	456
pH	5.0	4.3	4.9	4.3	4.6	4.8	5.5	3.8	3.9	4.0	4.4	4.5	4.8
Lactic acid	0	79	15	53	8	22	13	70	71	43	19	14	8
Acetic acid	63	9	7	7	7	7	3	13	14	15	12	11	7
Propionic acid	22	4	5	2	0.2	0	0	0.1	0	0	0	0	0
Butyric acid	76	32	30	21	7	0.3	0	2	1	0	0	0	0
Isovaleric acid	0.1	0	23	18	19	5	0	0.2	1	0.3	0.7	1.2	0.8
Ethanol	16	16	18	14	12	6	7	16	11	7	5	5	3
NH_3-N	145	47	91	97	72	55	48	50	71	60	92	59	47
NPN	490	550	550	560	550	540	520	590	510	530	480	470	350
DM loss	184	126	153	116	102	50	33	83	64	48	45	39	29

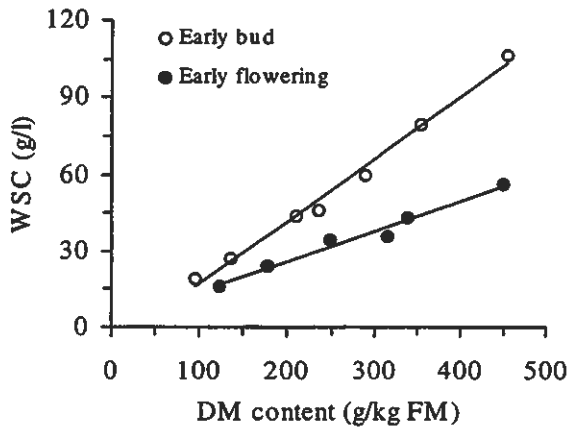

Figure 1. Evolution of WSC in the plant water (g/l) during wilting.

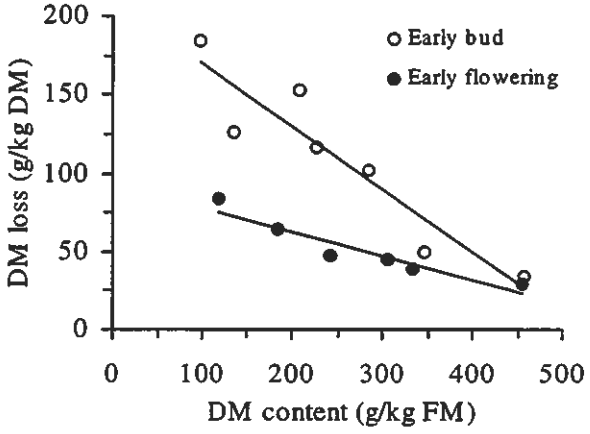

Figure 2. Relationship between the DM loss and DM content in sulla silages.

Conclusions

At early bud stage, with an high OMD, the sulla herbage in lab-silos underwent a good fermentation when ensiled at a wilting level of at least 350 g DM/kg FM. At early flowering stage, characterised by higher yield and lower OMD, a very good conservation quality was already obtained at a wilting level of 250 g DM/kg FM. The too long wilting time at early bud stage could be responsible for the bad fermentation pattern in silages with DM content lower than 350 g/kg FM. Mechanical conditioning and tedding could be useful to efficiently shorten the drying period of sulla, legume that is characterised by high moisture content and high stem diameter.

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Additives in ensiling whole crop barley

Introduction

High dry matter (DM) yields and a pressure to decrease production costs of forages favour the production of whole crop silage. Feeding value and ensiling characteristics of whole crop are dependent on crop DM content and the proportion of grain and straw in the whole crop, which changes with advancing maturity. This study was conducted to evaluate the effects of crop maturity and silage additives on the fermentation quality of whole crop barley silage.

Materials and methods

Whole crop barley was chopped with a laboratory chopper and ensiled in 10 l capacity silos at three stages of maturity: milk stage (I), dough stage (II) and yellow ripeness (III). The contents of DM, water soluble carbohydrates (WSC) and starch of the whole crop material with advancing maturity were 329, 389 and 483 g/kg, 238, 128 and 60 g/kg DM and 138, 221, 298 g/kg DM, respectively. The additives, applied as a solution were: no additive (control; C), urea 15 g/kg feed dry matter (U15), urea 30 g/kg DM (U30), urea 45 g/kg DM (U45), formic acid 4 l/t feed (FA) and a mixture (2:1) of FA and propionic acid 4 l/t feed (FP). Duplicate silos were opened after 180 d for chemical composition and fermentation characteristic assessments.

Results

The content of WSC decreased but that of starch increased in the whole crop silage with advancing maturity (Table 1). All treatments resulted in satisfactory fermentation at the earliest stage of maturity. Practically no urea was found in urea-treated silages indicating complete hydrolysation at all application levels, due to a relatively high moisture content at all crop maturity stages. Inclusion of urea led to a strong lactic acid fermentation and a simultaneous high pH at earlier maturity stages probably due to a high buffering capacity afforded by high ammonia concentrations. At the yellow-ripeness stage urea produced the most acceptable silage fermentation achieved even at the lowest application rate of urea. However, urea treated silages were more prone to aerobic deterioration than other silages. Ensiling without an additive or with acid additives ensured good fermentation in the silages at the two earliest maturity stages i.e. when the DM content of the material was less than 400 g/kg. Acids were not effective in controlling clostridia spores or butyric acid fermentation at the yellow-ripeness stage. Except for lower pH with FA, differences in silage fermentation between the two acid based additives were negligible. Inclusion of propionic acid did not reduce the amount of yeasts and moulds in the silage.

Conclusions

In the present study, whole crop with DM content less than 400 g/kg was ensiled satisfactorily without an additive. However, by using acid additives in-silo fermentation and breakdown of protein can be further restricted. At the yellow ripeness stage urea produced good quality silage even at an application rate of 15 g/kg crop DM. Aerobic deterioration in urea treated silages may, however, be considered a risk in conditions where DM content of crop at yellow-ripeness is often less than 500 g/kg.

Table 1. Effect of additives on chemical composition and fermentation quality of whole crop silages at three different stages of maturity

Additive...	C	U15	U30	U45	FA	FP	SEM	Comparison a) L	Q	C	SEM	Comparison b) 1	2	3
Maturity I														
DM, g/kg	324	315	315	321	317	310	3.4	NS	NS	NS	2.4	*	NS	o
pH	3.70	4.12	4.39	4.61	3.51	3.75	0.036	***	*	NS	0.032	o	***	**
In DM, g/kg														
Lactate	86	117	152	191	60	55	3.7	***	NS	NS	2.4	*	***	NS
Butyrate	0.0	0.0	0.6	1.1	0.2	0.1	0.04	***	***	*	0.08	NS	NS	NS
WSC	18	31	13	12	30	107	4.0	NS	NS	*	25.1	NS	NS	o
Starch	112	95	89	71	105	134	3.9	***	NS	NS	3.9	NS	**	**
N	14.8	22.1	29.5	36.8	14.3	15.4	0.40	***	NS	NS	0.42	**	***	NS
In N, g/kg														
NH_3-N	75	409	565	649	28	32	7.3	***	***	*	2.7	***	***	NS
Soluble N	814	866	910	923	716	754	12.1	***	NS	NS	21.0	NS	**	NS
Maturity II														
DM, g/kg	378	368	367	378	377	371	4.0	NS	o	NS	3.8	NS	NS	NS
pH	3.91	4.53	4.93	5.59	3.65	4.19	0.102	***	NS	NS	0.046	***	***	***
In DM, g/kg														
Lactate	50	77	112	131	34	4	4.1	***	NS	NS	1.8	*	***	***
Butyrate	2.9	25.5	11.7	0.5	0.4	0.1	0.55	***	***	***	0.28	***	NS	NS
WSC	98	57	41	40	61	211	34.8	NS	NS	NS	34.7	NS	o	*
Starch	229	215	212	226	231	245	16.3	NS	NS	NS	11.2	NS	NS	NS
N	12.9	21.0	28.9	34.4	12.5	12.8	0.15	***	***	*	0.13	***	***	NS
In N, g/kg														
NH_3-N	78	457	468	690	35	37	83.5	**	NS	NS	13.1	***	***	NS
Soluble N	758	811	866	882	685	657	16.3	***	NS	NS	20.8	NS	***	NS
Maturity III														
DM, g/kg	478	471	486	481	458	469	4.1	NS	NS	o	5.8	NS	NS	NS
pH	4.92	5.42	6.53	8.31	4.73	4.67	0.074	***	***	NS	0.038	NS	***	NS
In DM, g/kg														
Lactate	15	45	59	39	20	21	0.1	***	***	*	0.9	***	***	NS
Butyrate	6.4	1.2	0.2	0.9	7.5	7.5	0.48	***	***	NS	0.21	*	***	NS
WSC	42.8	31.6	18.2	14.8	27.2	31.5	2.06	***	NS	NS	2.99	*	NS	NS
Starch	296	317	328	332	315	356	32.7	NS	NS	NS	29.7	NS	NS	NS
N	13.4	20.3	26.2	31.4	13.3	13.7	0.37	***	o	NS	0.36	***	***	NS
In N, g/kg														
NH_3-N	69	407	515	539	74	70	13.4	***	***	o	7.7	***	***	NS
Soluble N	523	673	744	787	537	550	16.6	***	*	NS	14.1	*	***	NS

DM= dry matter; WSC=water soluble carbohydrates. Statistical comparisons: a) linear (L), quadratic (Q) and cubic (C) effects of urea; b) C vs. U15+FA+FP (1), U15 vs. FA+FP (2), FA vs. FP (3). (At maturity II treatment U45 instead of U15 was used in the statitistical comparison b). Statistical significance: NS not significant; o $P<0.10$; * $P<0.05$; ** $P<0.01$; *** $P<0.001$.

Urea-ammonium sulphate as an additive for ensiled forage maize

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Urea has been used for many years as an additive to increase the total nitrogen content of ensiled forage maize. Following addition to the crop at harvest, a proportion of the urea may be expected to be degraded in the silo by microbial urease to ammonia and carbon dioxide (Muck and Kung, 1997), with an increase in pH and a possible reduction in the number of lactic acid bacteria in the early phase of ensiling. As an acid salt which might liberate sulphuric acid in an acid environment, ammonium sulphate may restrict the total extent of fermentation, reduce the extent to which urea is hydrolysed to ammonia, and decrease the proportion of volatile fatty acids (VFA) in the ensiled product. Further, the sulphur in ammonium sulphate (0.28 by weight) might prove useful in rectifying any N:S imbalance in the rumen.

The experiment reported here was designed to test the hypothesis that urea-ammonium sulphate produces a higher proportion of lactic acid, and a lower proportion of ammonia-N (NH_3-N) in ensiled forage maize compared to the addition of urea alone.

Forage maize (cv Nancis) was harvested by Claas self-propelled forage harvester at the Leeds University Farm on 21 October 1997. Average length of chopping was 10 mm. Immediately after harvest, the crop was weighed into lots of 10 kg fresh weight and each lot was spread out evenly in a thin layer on a clean concrete floor. Additives were applied to the crop at the levels indicated below by spraying via an air-pressurised Hozelock 2 hand sprayer of 1.25 litres capacity. The treatments were: **CON** - no additive; **U1** - 2.5 kg urea /tonne FW, added as a 38.5% m/m solution; **U2** - 5 kg urea /tonne FW added as 38.5% m/m; **AS** - 1.5 kg of ammonium sulphate/t FW added as a 40% m/m solution; **U1AS** - 2.5 kg U/t + 1.5 kg AS/t; **U2AS** - 5.0 kg U/t + 1.5 kg AS/t.

Three replicate lots were treated separately for each treatment. Each lot of forage was packed as tightly as possible into polythene containers lined with two polythene bags, each of which was tied individually with string before an airtight polythene lid was placed on the top of the container. Once filled, each container was weighed before being stored in an unheated building for 107 days before being opened, mixed and sampled for analysis.

Values for pH were low for all treatments, indicating a stable fermentation dominated by lactic acid (Table 1). The presence of acetic acid indicated a heterolactic fermentation and there were also trace levels of acetaldehyde and mannitol in the silages, confirming this pattern of fermentation (McDonald *et al.*, 1991).

Addition of urea alone was reflected in little change in either the pattern or the extent of fermentation compared to the untreated silage, except for a reduction in acetic acid ($P<0.05$) and an increase in ethanol ($P<0.001$), only in the silage made with the higher level of urea (U2). Addition of ammonium sulphate alone (AS) resulted in lower lactic acid and increased ethanol ($P<0.001$) compared to the untreated silage. Addition of urea-ammonium sulphate was reflected in no significant changes in fermentation compared to untreated silage.

Table 1 **Effects of liquid urea, ammonium sulphate and urea-ammonium sulphate on the fermentation of ensiled forage maize (all values g/kg DM unless otherwise stated)**

	CON	U1	U2	AS	U1AS	U2AS	s.e.m
DM (g/kg)[1]	309.3^{ac}	300.0^{bcd}	300.7^{bd}	298.3^{b}	304.3^{cd}	301.0^{bd}	1.19
pH	4.11^{bc}	4.30^{a}	4.21^{ab}	4.16^{b}	4.07^{bc}	4.07^{bc}	0.034
Lactic	42.7^{ac}	39.7^{ad}	40.4^{ad}	37.5^{bd}	40.8^{ad}	41.2^{ad}	1.55
Acetic	9.00^{a}	9.40^{a}	3.30^{b}	8.73^{a}	9.03^{a}	7.87^{a}	1.13
Total VFA	10.0^{b}	10.4^{b}	4.43^{c}	9.73^{b}	10.0^{b}	8.87^{b}	1.16
Ethanol	28.9^{e}	31.6^{de}	33.0^{de}	37.7^{ac}	32.0^{de}	36.0^{bc}	1.55
Buffering capacity (meq/kg DM)	508^{bc}	454^{e}	477^{ce}	530^{b}	547^{bd}	545^{bd}	17.6

[1] Corrected for volatiles. Means with different superscripts are different ($P<0.05$)

Table 2 **Effects of liquid urea, ammonium sulphate and urea-ammonium sulphate on nitrogenous components and total sulphur (S) in ensiled forage maize**

	CON	U1	U2	AS	U1AS	U2AS	s.e.m
Total N (g/kg DM)	13.9^{e}	18.4^{b}	23.0^{a}	15.6^{d}	19.2^{b}	23.2^{a}	0.37
Urea N (mg/kg DM)	5.77^{c}	108.2^{b}	194.9^{a}	13.5^{c}	93.3^{b}	183.3^{a}	9.30
TSN (g/kg total N)	409^{f}	513^{d}	560^{ac}	445^{e}	542^{bc}	606^{a}	8.79
NH_3-N g/kg total N	38.1^{g}	62.6^{d}	54.9^{e}	100.1^{a}	80.2^{b}	72.4^{c}	1.45
Total S g/kg DM	1.51^{c}	0.99^{d}	0.82^{e}	1.91^{a}	1.49^{c}	1.68^{b}	0.05

Means with different superscripts are different ($P<0.05$). TSN = Total soluble N

Addition of U and AS to forage maize increased the concentration of total N ($P<0.05$). Addition of UAS did not increase total N compared to the same level of U (Table 2). UAS compared to U gave similar levels of urea N and higher (P,0.05) proportions of total N as NH_3N. The proportion of total soluble N in the total N was higher ($P<0.05$) for UAS than for U at the lower level of addition only. NH_3-N concentrations were lower for UAS than for AS, and lower at the higher level of U than at the lower level of U ($P<0.001$), indicating lower levels of urea hydrolysis to ammonia at higher levels of addition of N than at lower levels. NH_3-N concentrations were higher for UAS than for U ($P<0.05$) at both levels of addition of U. Total S was lower for U1 and U2 compared to CON, similar for U1AS and CON, and higher than CON for AS and U2AS. Differences between treatments in neutral cellulase digestibility, ash, starch and residual water-soluble carbohydrates were small.

It is concluded that urea-ammonium sulphate increased both total N and total S in maize silage, and had similar effects on the pattern and extent of fermentation to those produced by the addition of urea alone.

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Influence of inoculation and pre-wilting of extensively used grass on silage quality

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Introduction

In order to preserve diversity of the landscapes and for a sustainable agriculture a graded cultivation intensity is necessary. That's why along with more or less intensively used meadows, one part of the meadows has to be managed extensively. That means that the grass is grown without or with only small amounts of N fertilizer and/or harvested at late stages of maturity. But reducing the intensity of grassland farming, increases the risk of badly fermented silages and high butyric acid contents (Weissbach, 1996; Wyss and Vogel 1995). The aim of this trial was to investigate the influence of pre-wilting and of inoculation of extensively used forage on silage quality.

Material and Methods

As material we used a special forage mixture, which is in Switzerland intended for species rich meadows. The main grasses in this mixture are *Arrhenatherum elatius, Trisetum flavescens* and *Festuca rubra* as well as *Lotus corniculatus* and different herbs.
Forage of the first (21 June) and second (16 August) cut was short chopped and ensiled in laboratory silos with a volume of 1.5 litres. Two silos per treatment were filled.
For both cuts grass was ensiled with three different dry matter levels. These were 31, 39 and 45 % for the first and 34, 42 and 51 % dry matter for the second cut.
In addition to pre-wilting, a part of the grass was treated with an inoculant containing *Lactobacillus plantarum* (Bactensil Plus). This inoculant was applied with water.
The laboratory silos were stored at room temperature (approx. 20° C) and weighed regulary to measure gaseous losses. Dry matter (DM), ash, crude protein, crude fibre and sugar were determined at filling. After a storage period of 153 days, the silos were opened and again, nutrient contents in addition to fermentation parameters were analysed.

Results and Discussion

The nutrient contents of the green material per kg DM of the first cut were: 62 g ashes, 65 g crude protein, 364 g crude fibre and 69 g sugar. For the second cut the contents were: 85 g ashes, 109 g crude protein, 281 g crude fibre and 81 g sugar. The crude fibre content of the first cut was relatively high.
The untreated control silage of the first cut, ensiled with a DM-content of 31 %, showed a high pH-value, a high butyric acid content, a small lactic acid content and a high ammonia-N proportion (Table 1). According to the DLG evaluation scheme, this silage attained only a score of 14 points out of a maximum of 100. Under these conditions, that is to say with increasing pre-wilting degree, the butyric acid content and the ammonia-N proportion were reduced. The scores of these silages were 24 and 62. That means, that the quality was a little improved from bad to mediocre. Also the gaseous losses decreased with increasing DM-content.
For the grass of the first cut the silage additive was very efficient and improved the quality of the silages of all three DM-contents. These silages had lower pH-values, no butyric acid, lower ammonia-N proportions, higher lactic acid contents and lower gaseous losses than the untreated silages. All silages with inoculant attained the maximum of 100 scores.

For the second cut the untreated silages had a better quality in comparison to the first cut. The butyric acid content decreased with increasing DM-content form 5 to 0 g and the silages attained scores between 85 and 95.
Still under these conditions the inoculation improved the fermentation quality. The treated silages showed lower pH-values, lower ammonia-N proportions and lower gaseous losses in comparison to the untreated silages. The silages scored again 100 points. Driehuis et al. (1996) also observed an improvement of the fermentation quality in silages with DM-contents between 40 and 55 %, when treated with an inoculant.

Table 1. Fermentation parameters and silage quality of the silage at different dry matter contents and with or without inoculation

Treatment	Cut	DM %	pH	Lactic acid g/kg DM	Acetic acid g/kg DM	Butyric acid g/kg DM	NH_3-N as % of total N	Gaseous losses %	DLG scores
control	1	28.3	5.2	2	18	28	21.7	10.5	14
inoculant	1	30.0	3.9	59	7	0	4.9	3.7	100
control	1	35.9	5.5	0	9	18	16.0	9.3	24
inoculant	1	37.5	4.0	48	6	0	6.2	3.2	100
control	1	42.8	4.9	0	4	7	12.0	6.9	62
inoculant	1	44.6	4.0	31	4	0	5.4	2.1	100
control	2	32.5	4.5	35	21	5	8.6	5.1	87
inoculant	2	32.4	4.1	58	6	0	3.7	3.6	100
control	2	40.1	4.8	17	9	3	9.0	5.4	85
inoculant	2	40.7	4.1	46	6	0	3.4	2.5	100
control	2	48.7	4.8	10	9	0	7.0	4.6	95
inoculant	2	49.6	4.1	37	6	0	3.6	1.8	100

Conclusions

The results of this trial confirm, that the silage quality of extensively used grass, especially from the first cut, is bad. With pre-wilting the fermentation quality can be improved a little. Under these conditions the addition of an inoculant showed a good efficacy. The question is, whether it is economically efficient to use silage additives with this forage or if, the weather permitting, haymaking is the adequate conservation method.

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Role of *Streptomyces achromogenes ISP5028* in the fermentation of poor quality grass for silage.

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Introduction

It is generally accepted that silage quality depends on the quality of grass and the pattern of fermentation in silo. Grass low in soluble carbohydrate (the substrate for lactic acid production) can have a slow rate of acidification during conservation resulting in inadequate restriction in undesirable microbial activity and secondary fermentation of lactate by Clostridia. Cell-wall degrading enzyme additives (based on cellulases and hemicellulases) have been examined by many authors however, extracellular peroxidases have not been investigated as potential additives for ensiling of poor quality grass. The aim of the experiment reported here was to investigate the role of *Streptomyces achromogenes ISP5028* (an actinomycete producing high levels of extracellular peroxidases) in the fermentation of poor quality grass for silage production.

Material and methods

Perennial ryegrass was swathed and conditioned on 9 May 1998, and harvested on 10 May 1998 using a precision-chop forage harvester 18 hours after swathing. Harvested grass was ensiled (four replicates per treatment) in 5 kg plastic bag silos. Harvested grass was either inoculated prior to ensiling with 10^6/ml spores of *Streptomyces achromogenes ISP 5028* per kg fresh-weight of grass (suspended in 20 ml of water) or ensiled without further treatment (control silage). The silos were opened after 1, 2, 3, 5, 10, 20, 30 and 60 days and analysed for DM, pH, soluble carbohydrates, ammonia-N, fermentation acids (lactic, acetic, isomeric forms of butyric, valeric and caproic acids and ethanol), electrometric titration (Offer *et al.*, 1993), NDF, ADF, crude hemicellulose (NDF-ADF) and estimated DOMD. Chemical compositional data were analysed by two-way analysis of variance. Spores were isolated from the ensiled grass after each silo sampling using the method of Shirling and Gottlieb (1966) and viable CFU were counted for all silages except those of 60 days.

Results and Discussion

The grass prior to ensiling had a high moisture content (792 g/kg), low levels of soluble carbohydrate (208 g/kg DM) and moderate levels of N (29.6 g/kg DM). After 60 days of ensiling, evidence of poor fermentation characteristics was observed for both silages by the decomposition of lactic acid to butyric acid (secondary fermentation; Table 1). There were significant increases (both $p<0.01$) in the concentrations of the various isomeric forms of valeric and caproic acids. The reasons for the elevated concentrations of longer chain volatile fatty acids is not fully understood but may reflect an increased availability of carbohydrates derived from sugar-phenyl propanoid cross-linked compounds altering the pattern and composition of end products after fermentation. The concentrations of NDF, ADF and hemicellulose as well as the estimated DOMD of the control and inoculated silages are in Table 1 . A significant decrease in the concentration of NDF ($p<0.01$) but no changes in the concentration of ADF or crude hemicellulose were observed in the inoculated silages after 30 days of ensiling: No significant changes in cell wall components were observed in the control silages after 60 days of ensiling. This observation could be explained by the presence of extracellular peroxidases effecting ester linked phenyl-propanoid compounds in the hemicellulose fraction of the cell wall rather than ether linked compounds of the core-lignin fraction. Low levels of spores of *Streptomyces achromogenes ISP 5028* were isolated from the control silages (maximum viable spore: 1.68×10^8 spores/g fresh-weight after 10 days of ensiling). The pattern of growth of *Streptomyces achromogenes ISP 5028* was different in the inoculated silages with a peak number after 10 days of ensiling. Growth of similar actinomycetes has been observed in conditions similar to

those observed in silage system (pH<5, micro-aerophilic environments) and the production of extracellular peroxidases continues, albeit at a lower level, at pH of below 5 (Rob et al., 1997).

Table 1. Chemical composition of silages (g/kg DM unless stated) after 60 days storage.

	Control	Treated	s.e.d
Corrected DM (g/kg)	235	236	11.4
NDF	512	448	13.5**
ADF	295	288	12.6
Crude hemicellulose	217	160	14.1**
pH	4.12	4.02	0.49
Ammonia-N (g/kg total N)	87	77	10.3
Nitrogen	25.1	26.4	1.77
Amino-N (g/kg total soluble N)	725	775	12.4*
Lactic acid	78.1	101.7	11.5**
Acetic acid	16.0	21.7	6.65
Propionic acid	0	0.12	0.11
Butyric acid	0.32	0.97	0.07**
Valeric acid	0.12	1.02	0.03**
Caproic acid	0	0.21	0.06*

Conclusions

The results from this study indicate that actinomycetes that produce extracellular peroxidases such as *Streptomyces achromogenes ISP 5028* may have a role in acting as an additive to improve the pattern of fermentation of poor quality grass for silage production. The potential risk of challenge to the respiratory system of the animal by spores of actinomycetes may however influence the use of such organisms in practice.

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Fatty acid pattern of Austrian maize silage varieties before and after ensiling

Introduction and object of study

Green maize and the corn itself fresh and after ensiling is the basis of fattening cattle, pig and poultry production in Austria (. The crude fat content and the fatty acid pattern of maize plays an important role for the body fat quality especially in pig production. Maize is well known for its high content in linoleic acid. Anaerobic lactic bacteria fermentation (see poster Grajewski et al.) and its influence on the fatty acid pattern is yet not investigated well.

The aim of this study was to get information if there are differences before and after the fermentation of maize products and to show the differences in fatty acid pattern of different varieties such as *Barbara* and *Jaspe*.

Material & methods

Typical Austrian maize varieties such as *Barbara* and *Jaspe* were harvested in September and silaged as whole silage maize (WPS). The same types were harvested later on as corn cob mix silages (CCM) and as whole grain silage (WGS) in the end of october 1998.

The fermentation took place under controlled conditions at the BAL Gumpenstein in Styria and was analyzed at beginning of december. Containers with 250 l of volume with 100 cm filling height were used. Fatty acid pattern was determined by GC at the Institute of Nutrition. Fat was extracted by cold solution of dichlormethane/ methanol (2/1 v/v) before transmethylation was performed.

Results

Table 1 shows the results of *Jaspe* maize before and after fermentation

Fatty acids %	WPS before	WPS after	CCM before	CCM after	WGS before	WGS after
palmitic	10.4	9.5	10.6	9.1	9.2	8.9
stearic	2.4	2.2	2.3	2.2	1.9	1.9
oleic	29.2	30.6	35.9	35.4	32.7	33.0
linoleic	51.3	52.1	49.2	51.5	53.9	54.0
linolenic	5.9	4.9	1.5	1.3	1.4	1.3

Acids below 1.0 % are deleted

Fatty acids are listed in % of total fatty acids

Table 2 shows the results of *Barbara* maize before and after silage fermentation

Fatty acids %	WPS before	WPS after	CCM before	CCM after	WGS before	WGS after
palmitic	10.9	9.1	10.0	8.9	9.0	9.1
stearic	2.4	2.0	2.3	2.2	2.1	2.2
oleic	19.3	19.6	23.7	23.8	22.7	22.9
linoleic	55.7	61.4	60.7	59.6	63.2	62.3
linolenic	8.7	3.8	1.5	1.3	1.4	1.4

About 10 % higher contents of oleic acid are observed in the variety *Jaspe*. The trend was opposite in the linoleic acid contents.
Small differences are seen before and after the fermentation within one commodity.
The linolenic acid was found less in the fermented stuff than in the fresh material.

Conclusions
Clear differences in the fatty acid pattern are observed between the varieties *Barbara* and *Jaspe*. The fatty acid pattern of un- and fermented maize materials of the same variety differs little and only in the linolenic acid content.

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The effect of inoculant treatment of alternative crop forages on silage quality and *in vitro* rumen function

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Introduction

The predominant winter feed for UK livestock is grass silage. However, the protein status of this feed means that concentrate supplements must be fed to maintain adequate levels of production. With the current emphasis on reducing inputs of supplementary concentrates, alternative ensiled forages of high protein content are being examined (Fraser *et al.*1999). When feeding ruminants predominantly forage-based diets the efficiency of rumen function is of paramount importance as the rumen microbial population provide a substantial proportion (> 70 %) of the amino acids that are available for absorption from the small intestine. The aim of this work was to compare the ensiling potential of three alternative forages and to assess their relative merits in terms of efficiency of rumen function and in particular microbial protein synthesis when fed as sole diets to *in vitro* rumen continuous cultures.

Materials and Methods

Kale (*cv.* Pinfold and Keeper) was cut at the 17 week growth stage and the lucerne (*cv.* Vertus) and clover (*cv.* Merviot) were second re-growths. Forages were wilted for 48 h and baled either untreated or after application of an inoculant (Live System, Genus, Crewe; *Lactobacillus plantarum* 10^6 CFU/g FM) for kale and red clover, whilst lucerne was only ensiled untreated. After 90 d of ensiling, bales were cored and samples taken for chemical compositional analysis (DM, pH, WSC, starch, lactic acid, ammonia, nitrogen and total and free amino acid contents) and to provide samples for *in vitro* rumen function studies. The rumen simulation technique (Rusitec) described by Czerkawski and Breckenridge (1977) was used to study rumen function. Each experiment (3 replicate runs) compared alternative forage silages which were fed unsupplemented at 15 g DM/day to fermentation vessels infused with artificial saliva to give a liquid outflow rate of 2.9 %/h. Measurements included the outflow of ammonia and VFA's, DM digestibility and efficiency of microbial protein synthesis using N_{15} as the marker for microbial N.

Results

There were marked contrasts in the composition of the silages prepared from different forages, particularly in the N and energy fractions (Table 1). Both untreated and inoculated kale silages had low pH values and high lactic acid concentrations, with only minor differences between other constituents. Inoculated red clover silage had a lower pH and lower concentrations of ammonia and free amino acids (AA), with 12 % higher protein AA content than the untreated silage. Both red clover silages contained the lowest concentrations of protein breakdown products and residual WSC and starch. In contrast, kale silages had the lowest protein AA concentrations and the highest concentrations of protein breakdown products, but the highest concentrations of WSC and starch. Untreated lucerne silage was intermediate in terms of silage quality with lower total AA content than red clover and lower starch and WSC contents than kale. The results for rumen function parameters are shown in Table 2. No significant differences were found between any of the rumen function values for inoculated compared to untreated kale silage. However, for red clover silages increases in rumen microbial growth efficiencies were obtained with inoculant addition. Digestibility values for both clover silages

were similar but higher daily production of VFA's with the inoculated silage suggested a relationship with the higher residual WSC and starch levels in this silage.

Table 1. Composition of 90 d silages

	Kale		Lucerne	Red Clover	
	Untreated	Inoculated	Untreated	Untreated	inoculated
DM (g/kg FM)	141.4	145.3	352.2	268.2	303.8
pH	4.06	3.99	5.28	5.26	4.52
WSC (g/kg DM)	8.0	8.9	26.9	7.3	10.7
Starch (g/kg DM)	77.0	75.2	16.1	16.7	17.2
Lactate (g/kg DM)	162.2	172.4	12.8	39.4	70.5
Total N (g/kg DM)	19.03	18.83	33.17	36.83	35.30
Ammonia-N (g/kg N)	84.97	88.30	49.52	93.64	60.50
Total AA (moles/kg N)	42.95	43.60	48.24	50.41	60.11
Protein AA (moles/kg N)	23.03	24.61	24.51	36.45	41.46
Free AA (moles/kg N)	19.92	18.99	22.01	13.81	13.54

AA = amino acid; Protein AA = Total – free AA

Efficiency of microbial protein synthesis was also greater with the inoculated red clover silage. Kale silage had a lower daily ammonia-N outflow and microbial-N synthesis value, but higher daily outflow of VFA's than for the clover silages. This was probably related to the higher energy but lower protein content of these silages. Lucerne silage had the lowest value for efficiency of microbial protein synthesis and the highest ammonia-N outflow.

Table 2 Daily outflows of ammonia and VFA's, efficiency of microbial protein synthesis and digestibility values from Rusitec continuous cultures fed alternative crop silages

	Kale		Lucerne	Red Clover	
	Untreated	Inoculated	Untreated	Untreated	Inoculated
% DMD	84.3 (1.85)	84.1 (1.80)	67.8 (0.31)	78.6 (1.49)	77.3 (0.73)
VFA[b]	64.8 (1.12)	67.9 (3.78)	50.8 (1.46)	56.7 (5.80)	60.1 (3.70)
Ammonia-N[b]	3.8 (0.90)	4.0 (0.24)	13.5 (1.28)	12.4 (0.51)	10.7 (0.27)
Microbial-N[a]	7.9 (0.62)	7.7 (0.44)	6.1 (0.45)	8.2 (0.41)	9.3 (0.92)

a = g/kg OM apparently digested; b = mmol /d; SEM values are shown in parentheses

Conclusions

Inoculation of red clover had a positive effect on silage quality, particularly the protein fractions, and this corresponded with the highest *in vitro* value for efficiency of microbial protein synthesis. No comparable effects were observed with inoculated kale silage, which had the lowest protein content, but rumen microbial growth efficiency was higher than for lucerne silage. This may be related to the higher levels of rapidly digestible energy (starch and WSC) in kale silage that enabled more efficient utilisation of protein breakdown products.

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The prevention of alcoholic fermentation in high dry matter grass silage

Introduction

Ethanol is a common, usually minor fermentation product in silages. However, Driehuis and Van Wikselaar (1996) found that ethanol was the main fermentation product in about 20% of the grass silages produced from wilted grass. Ethanol represented up to 90% of the total of fermentation products in these silages, indicating that an alcoholic rather than a lactic acid fermentation had occurred. Yeasts are probably responsible for this phenomenon. Here we show that ethanol fermentation in high dry matter grass silage can be prevented by the addition of a bacterial inoculant or by maceration of the grass prior to wilting.

Materials and Methods

Grasses (mainly *Lolium perenne*) from regular permanent pastures were used in this study. Grass was cut with a disc mower, spread and wilted for 6-30 h before ensiling. Ensiling was in airtight 1-l jars (0.4 kg per jar), which were stored at 20°C. At different times after ensiling, duplicate silages were opened and analysed for weight loss and pH. In the experiment in which the effects of maceration and inoculation were investigated, the grass was divided in two portions. One portion was processed through an experimental macerator. Macerated and untreated grass were wilted for 24 h. Before ensiling, the untreated grass was divided in two portions. One portion was sprayed with a suspension of an inoculant (Silage Inoculant 1188, Pioneer Hi-Bred International), providing 10^5 colony forming units/g. The other portion of untreated grass and the macerated grass were sprayed with demineralised water.

Results

Fig. 1a shows the pH in the course of the ensilage of two grass crops with a similar DM content. One crop développed into a normal silage with lactic acid as the main fermentation product ('lactic acid silage'), whereas the other, for unknown reasons, developed into a silage with ethanol as the main fermentation product ('ethanol silage'). The pH of the ethanol silage dropped much slower than that of the lactic acid silage and remained above 6.0 for 60 days. A sharp increase in DM loss of ethanol silages was detected between 14 and 60 days of ensilage (Fig. 1b), probably reflecting the carbon dioxide that is produced in ethanol fermentation. The composition of the silages is shown in Table 1. Both silages were well preserved, as indicated by the low ammonia-N contents and the absence of butyric acid.

The influence of maceration of grass and addition of an inoculant on ethanol fermentation could be investigated in an experiment in which the untreated grass by chance developed into an ethanol silage. Both the maceration and the inoculant treatment increased the rate and extent of pH decline and prevented excessive ethanol fermentation (Fig. 2, Table 1). With respect to the rate and extent of pH decline, the inoculant treatment was more effective than the maceration treatment. Inoculant-treated silages contained a higher content of lactic acid and a lower content of acetic acid than silages made from macerated grass.

Conclusions

Ethanol fermentation in high dry matter grass silages starts about 14 days after ensiling and leads to high DM losses. The low contents of ammonia-N and butyric acid in ethanol silages indicated that protein-degrading microorganisms such as enterobacteria and clostridia did not develop to large numbers. Treatments that stimulate lactic acid fermentation (maceration of grass, addition of a silage inoculant) were found to prevent ethanol fermentation.

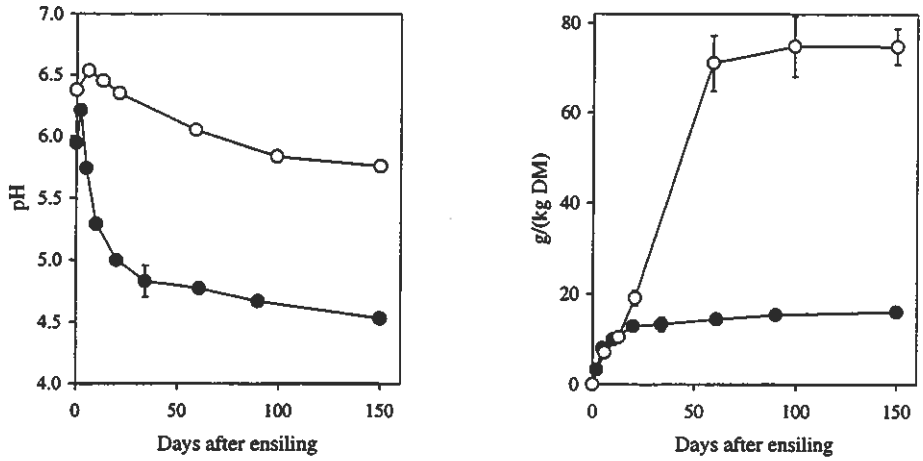

Figure 1. Silage pH (a) and DM loss (b) in the course of ensilage of grass crops developing into ethanol silage (○) or lactic acid silage (●) (DM at ensiling 475 and 425 g/kg, resp.).

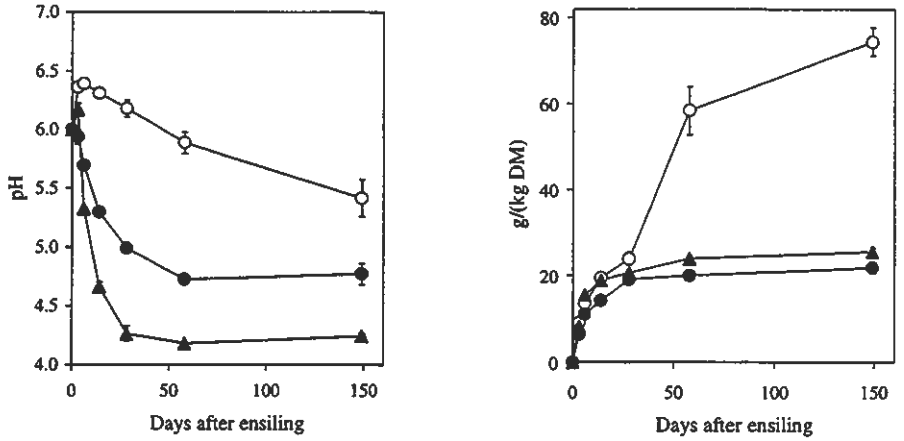

Figure 2. Silage pH (a) and DM loss (b) in the course of ensilage of untreated grass (○), macerated grass (●) and inoculated grass (▲) (DM at ensiling 454, 492 and 454 g/ kg, resp).

Table 1. Composition of the silages shown in Fig. 1 and 2 after 150 days ensilage.

	pH	Lactic acid (	Acetic acid	Etha-nol g/kg DM	DM loss	Sugars)	Ammo-nia-N (g/kg N)
Silages shown in Fig. 1							
Ethanol silage	5.84	1.8	6.4	57.0	69.9	112	6.8
Lactic acid silage	4.53	32.3	11.2	6.6	16.5	49	8.2
Silages shown in Fig. 2							
Untreated	5.41	10.1	4.3	63.0	82.8	82	10.7
Maceration	4.77	18.7	14.9	8.7	22.6	162	10.8
Inoculation	4.24	44.5	6.2	13.1	26.4	162	8.1

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The effect of harvest time on fermentation and fungal growth of two ensiled maize varietes in Austria.

Object of study:
Fresh and ensiled maize harvested in various forms is in our area the basic fodder in animal feeding. Especially in the case of feeding doses for cattle, ensiled maize may be given in the same percentage as it is in the case of pigs. So, a stock-breeder has to cope with the same problems of mould fungi. That is why the proper time of harvesting and technique of ensiling decide about the nutritive value and quality of silages. The content of fat and fatty acids, especially linoleic acid of which high quantities are contained in the whole plant , is the reason for the high energy content of maize. The pattern of fatty acids before and after ensiling is presented in another paper (see poster of Boehm et al.).
The aim of the experiment was to determine the changes in selected nutrients, quality and the general microflora of two typical varieties of Austrian maize (Jaspe and Barbara) dependent on the time and form of ensiling. The whole plant (WPS) was ensiled at the beginning and at the end of September. Corn cob mix silages (CCM) were made at the end of September and at the end of October, and whole grain silage (WGS) only at the end of October. Ensiling was carried out at BAL Gumpenstein in Styria (pilot scale silos). pH was controlled continueusly, and at the beginning of December, samples of all silages were analysed. Fungi, yeasts and moulds was determined according to the PN-R-64791/12/94.

Table 1. The fermentation quality maize .

Type of silage	Date of (ensiling) harvest	Maize Variety	Dry matter (g)	pH	Sugars (g) after ensiling	NH_4-N (mg)	Lactic acid (g)	Acetic acid (g)
						per kg dry matter		
WPS	02.09.98	Jaspe	262,2	3,45	16,10	440,5	75,7	12,0
	02.09.98	Barbara	279,8	3,56	15,65	421,0	58,0	16,3
	21.09.98	Jaspe	326,3	3,58	24,70	548,5	75,7	22,6
	21.09.98	Barbara	345,8	3,72	15,75	452,0	58,7	12,5
CCM	21.09.98	Jaspe	617,5	3,78	6,92	384,0	22,5	3,3
	21.09.98	Barbara	660,3	3,76	11,65	391,0	18,7	4,3
	29.10.98	Jaspe	706,3	5,18	7,56	124,5	13,6	1,2
	29.10.98	Barbara	727,7	5,82	14,05	77,5	10,1	1,5
WGS	29.10.98	Jaspe	728,2	6,38	9,36	19,5	9,8	1,0
	29.10.98	Barbara	743,4	6,32	15,10	22,5	10,2	0,9

Conclusions:
Significant differences in theof dry matter content was observed between Jaspe and Barbara varieties. Barbara had higher content of DM. Also the variety – specific differences in the sugar content were detected that did not affeetlactic and acetic acids concentrations in CCM and WGS. In silages made at the end of October, the pH was too high and the concentration of organic acids too low.

Table 2. Microbiological evaluation of Jaspe and Barbara before and after ensiling.

Parameter	Maize Variety	WPS		CCM		WGS	
		before	after	before	after	before	after
Aerobic Mesophilic Bacteria (CFU/g)	Jaspe	$1x10^8$	$8x10^4$	$7x10^6$	$3x10^5$	$6x10^4$	$4x10^5$
	Barbara	$9x10^7$	$1x10^4$	$3x10^5$	$1x10^5$	$4x10^5$	$3x10^6$
Fungi yeast and moulds (CFU/g)	Jaspe	$2x10^7$	$1x10^4$	$1x10^6$	$1x10^2$	$3x10^5$	$4x_5^{10^5}$
	Barbara	$9x10^6$	$9x10^2$	$1x10^6$	$3x10^3$	$9x10^4$	$4x10^4$

In whole grain silages a mean count of $4x\ 10^5$/g CFU moulds was isolated which indicated that the silage was unstable and oxygen was present during the fermentation process. Moulds of the Penicillium spp. type were dominant and they arew quickly in the samples. Also Aspergillus spp. and Monascus spp. were isolated. In the whole plant silage and CCM the presence of mould was lower. The general number of fungi (including yeast), was significantly reduced in the process of ensiling the whole plant and CCM (both varieties). This result was not achieved when whole grain was ensiledwhich proves that it is difficult to obtain stable and good silage of whole maize grain containing more than 70% DM and harvested at the end of October.

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Intensive Conditioning of Forage - effects on fermentation and silage quality

Background

Results from several independent studies in pilot and laboratory scales, have shown that intensive mechanical treatment of forage at mowing can be a successful way to improve the fermentation characteristics of non-chopped forage (Muck *et al.*, 1989; Walther, 1991). Since the plant juice becomes more readily available for lactic acid bacteria, comparable or even better fermentation than for precision-chopped forage have been achieved. Today, mowers with new types of conditioners, developed to give the crop a more severe treatment, are commercially available. The present study was undertaken in order to elucidate whether the promising fermentation results reported with intensive conditioning, can now be utilised by farmers in practical silage- making.

Method

Two harvesting methods were compared:

- Intensive conditioning (IC) in combination with a loader wagon
- Traditional system (TS) with a mower conditioner in combination with a precision chopper.

The aim was to illustrate the effect of intensive conditioning on forage ensilability.

Two similar experiments were performed with a grass-dominated crop cut with an interval of about two weeks. The forage was wilted in swaths to 27-30 % DM in the first experiment and to 35-39 % in the second. During silo filling, samples were taken for chemical and microbial analyses. The degree of conditioning of the forage was quantified by means of conductivity measurements (Koegel *et al.*, 1995). The forage was ensiled in small laboratory silos containing two litres. Two silos per treatment were opened after 2, 6, 30 and 100 days of storage, and silages were analysed with respect to DM-content, ammonia-N and fermentation products. Microbial analyses were made only on the silages stored for 100 days.

Results

Conductivity measurements on forage *directly after mowing* showed a very small difference in degree of conditioning between the intensive and conventional treatment. *After harvesting*, the conductivity of precision-chopped forage was 2-3 times higher than that of intensively conditioned forage harvested with a loader wagon. In both experiments, higher LAB levels were found in the precision-chopped forage.

Table 1. Fresh forage at ensiling

	Expt 1		Expt 2	
	IC	TS	IC	TS
Conductivity index	8	22	12	24
LAB, log CFU/g	3.0	4.9	3.0	4.5
DM, %	30.1	26.8	38.6	35.3

In both experiments the pH-decline was considerably faster in the chopped than in the intensively conditioned forage. This was probably due to the higher degree of conditioning in the chopped forage, which made nutrients more readily available for lactic acid bacteria. Furthermore, the final NH_3-N levels were lower in the chopped forage. The microbial analyses showed an overall good hygienic quality in all the silages stored for 100 days.

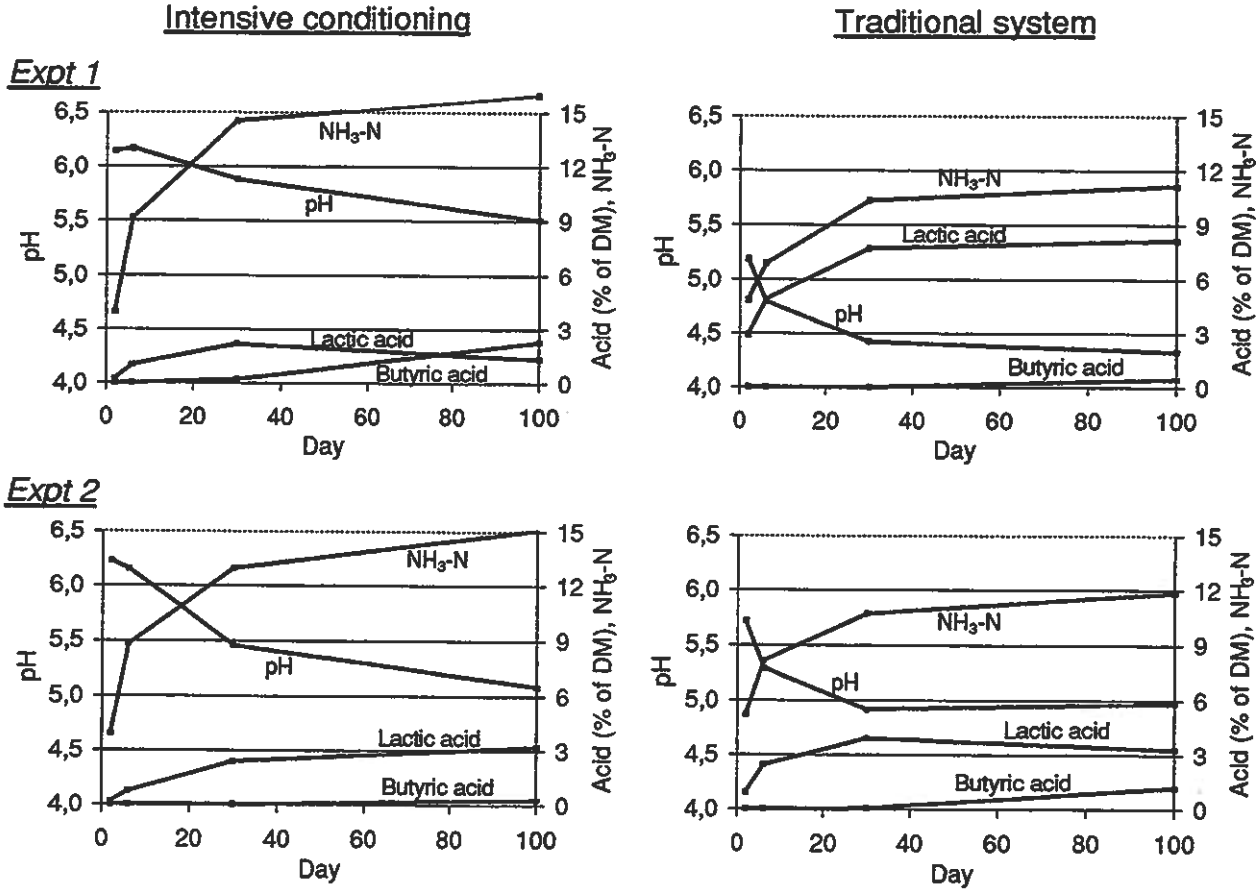

Fig 1. Course of fermentation

Based on the results from this study, the mechanical treatment of forage in the intensive conditioner can not be regarded as sufficient to improve ensilability to any appreciable extent.

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An evaluation of newly selected bacterial strains as additives for maize silage.

Introduction The application of biological silage additives to maize can often produce variable results, particularly in terms of aerobic stability. Three studies were conducted to determine the effect of newly selected bacterial strains on the aerobic stability, dry matter (DM) loss during ensiling, fermentation characteristics and nutritive value of maize silage.

Methods Whole crop maize was harvested at 393, 290 and 380 g DM/kg for studies 1 (1996), 2 and 3 (1997) respectively. Freeze-dried bacterial inoculants were hydrated and applied at 2 l/t forage to achieve 100,000 CFU/g maize. Propionic acid was included in study 1 as a positive control.

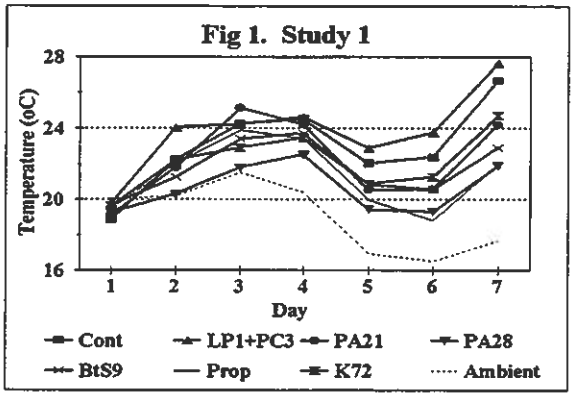
Study 1	Control, *Lactobacillus plantarum* (LP1) + *Pediococcus pentosaceus* (PC3), *L. plantarum* (PA21), *L. plantarum* (PA28), *Bacillus thuringiensis* (BtS9), Propionic acid (5 l/t) and *P. pentosaceus* (K72)
Study 2	Control and *L. plantarum* (PA28)
Study 3	Control, *L. plantarum* (PA28), *L. plantarum* (PA28) + *L. plantarum* (K270), *L. plantarum* (K270) + *P. pentosaceous* (PP202) and *P. pentosaceous* (K72)

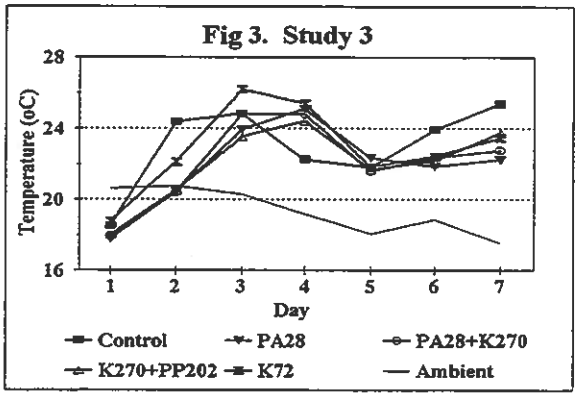
Control and treated forage was ensiled in laboratory scale silos (2.5 kg) with 4, 5 and 5 replicates/treatment for studies 1-3, respectively.

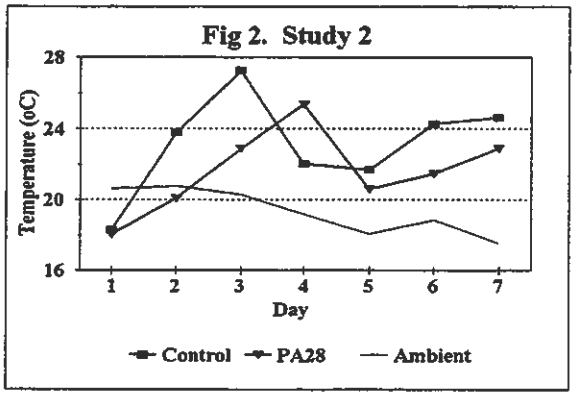
Results Fourty five days after ensiling the DM loss, silage fermentation characteristics and nutritive value were determined. Aerobic stability (Fig. 1-3) was assessed using the temperature profile of silage stored in insulated boxes for 7 days after opening. Data were analysed using an analysis of variance for a completely randomised design in Genstat 5.

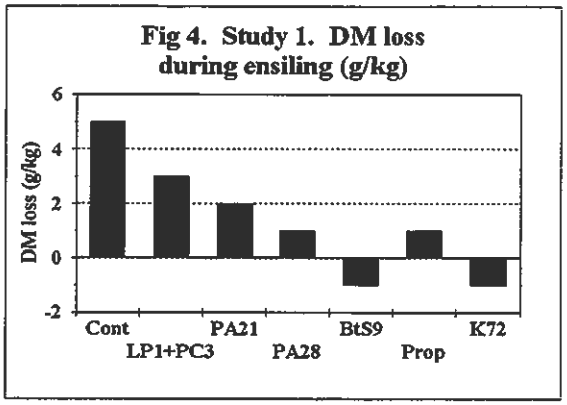
Several inoculant treatments included in studies 1, 2 and 3 improved aerobic stability. In study 1 (Fig. 1), temperatures of PA28 and propionic acid treated silages were lower than the control silage on day 6 ($P<0.05$), and by day 7 all treated silages, except LP1 + PC3, had lower temperatures than the control silage ($P<0.05$, except K72 $P>0.05$). Treatment with PA28 resulted in lower ($P>0.05$) temperatures than the propionic treated silage on days 1-5 and day 7. In study 2, silage treated with PA28 had lower temperatures than control silage on days 1 and 2 ($P<0.05$), and showed an increased number of hours to 2°C rise above ambient temperature (19 and 43 hours for PA28 and the control, respectively) ($P<0.05$). When compared to the respective controls, cumulative temperature in study 2 was lower with PA28 during the first 48 hours after opening ($P<0.01$), and in study 3 with PA28 + K270 during the first 24 hours after opening ($P<0.05$).

In study 1, the DM loss during ensiling in treated silages were consistently lower than the control (Fig. 4) (P>0.05, except BtS9 and K72 P<0.05). Also in study 1, the metabolisable energy content of the inoculated silages were consistently higher than the control (Fig. 5) (P>0.05, except PA21 P<0.05). Overall, the application of the inoculants did not effect ammonia nitrogen, pH, lactic or acetic acid, ethanol, crude protein, starch or acid detergent fibre content (P>0.05), all of which were within the expected range. Levels of propionic acid and butyric acid were negligible (< 4 g/kg CDM).

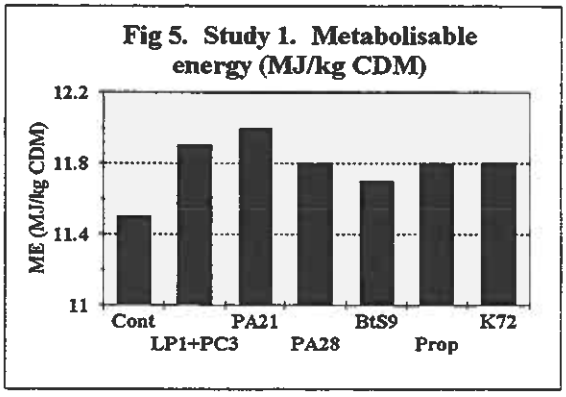








Conclusions It can be concluded that the application of newly selected bacterial strains in studies 1, 2 and 3 improved aerobic stability, although the mechanism by which this occurred is not fully understood. Effects on DM loss during ensiling, fermentation characteristics and nutritive value of maize silage were limited.



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Possibilities of ensiling paddy straw and agro-industrial byproducts for rearing crossbred calves in the tropics

Attempts were made to make test silage based on paddy straw, molasses, cotton-ginning trash, malt sprouts with hulls supplemented with minerals, which thereby increase the palatability of paddy straw and may meet the nutrient requirements for maintanance and growth in growing calves and open a way for the utilization of certain agro-industrial byproducts and wastes viz. Malt sprouts with hulls and cotton ginning trash.

For conducting feeding trial, approximately 10 000 kg silage (wet weight) was prepared using the following formula.

FORMULA FOR 10 000 kg SILAGE

Paddy straw	1000 kg
Molasses	333 kg
Cotton ginning trash	1333 kg
Malt sprout with hulls	1666 kg
Water	5666 kg
Supplemented with minerals	
Sodium sulphate	9.00 kg
Calcium phospate	6.00 kg
Calcium carbonate	20.50 kg

The pit was opened after 6.5 months of sealing.

Twelve male crossbred calves of approximately two years of age and similar live weight were divided into two comparable groups of six calves in each in a randomised block design. The first group of calves (control group) were maintained on *Ad libitum* barseem (*Trifolium alexandrinum*) hay feeding, while the experimental group was kept on *Ad libitum* silage feeding. In addition to this, 1 kg green barseem was also fed to each calf per day so as to meet the carotene requirement.

At the time of silage feeding, extra amount of molasses (6.67% of wet weight of silage) was added so as to prevent the animals from selective feeding of silage ingredients. The experimental feeding was carried out for about 39 days. During the last 7 days of which a digestion and balance trial involving quantitative collection of faeces and urine was conducted, so as to study nutrient
utilization. The silage suffers from having high insoluble ash, poor digestibility of protein as indicated by low DCP values and does not provide sufficient energy and protein to meet the maintance and growth requirements. The effective feeding value (79.63) indicate a fair palatability of silage but voluntary consumption of silage by the animal could not provide the optimum nutrients as per NRC (1971) recomendations and it is not possible to rear cross - bred calves solely on such type of silage without any supplementation of concentrate mixture.

Workshop B
Protein utilization of silage

Poster abstracts

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Nitrogen utilisation by lambs offered ensiled legumes.

Introduction

Ensiled grass is presently the major source of winter feed for dairy cows and other livestock in the UK. However, the quality and quantity of protein in grass silage tends to limit its use for milk production, which means that bought-in concentrate supplements must be fed to maintain high production levels. Whilst forage legumes appear to have potential as home-grown, protein-rich feeds for livestock (Frame *et al.*, 1998), there is very little information available regarding their nutritive value when ensiled. The aim of this study was to compare the nitrogen retention of lambs offered ensiled red clover, lucerne, lotus and sainfoin.

Materials and Methods

Silage was prepared from first-cut sainfoin (cv. Sombourne) and second-cut red clover (cv. Merviot), lucerne (cv. Vertus) and lotus (cv. Leo). The sainfoin and red clover crops were cut at the late flower growth stage, the lucerne crop at the early flower growth stage and the lotus crop at the late bud growth stage. All the crops were cut using a crimper conditioner disc mower and left in swaths to wilt for 48 h. The crops were then baled using a fixed chamber round baler, before being wrapped in 6 layers of 750 mm wide film. No additive treatments were used. Three bales of each forage were used during the experiment, with each bale fed for 7 days. In each case, the entire bale was chopped using a commercial big bale chopper, and the chopped material stored in baskets in a cold store maintained at a temperature of 4°C. A straight-through design was chosen, with each experimental silage offered to 6 Suffolk cross wether lambs aged 10 months, housed in metabolism crates. Following a 14 day adaptation period, data were collected over a 7 day measurement period. Throughout the experiment the forages were offered *ad libitum*, with feeding levels designed to ensure 10 – 15 % refusals each day. During the measurement week the amounts and dry matter (DM) contents of feed offered and feed refused were recorded daily. A bulked sub-sample of each silage 'as offered' was analysed for total nitrogen (TN) (expressed as crude protein (CP); TN x 6.25), ammonia, lactate, water soluble carbohydrates (WSC), gross energy (GE), neutral detergent fibre (NDF) and pH. Urine and faecal output from each animal was measured daily, and sub-samples were retained. Sub-samples of feed refused, urine and faeces were bulked on an individual animal basis, before undergoing TN determinations.

Results

The chemical composition of each of the silages is given in Table 1. The relatively high pHs (> 4.36) indicate a restricted fermentation in all crops, presumably as a result of the moderately high DM contents. However, the lactate content of the red clover silage was more than twice that recorded in the other silages, and

Table 1: Composition of the silages offered (all values g/kg DM unless otherwise stated).

	Red clover	Lucerne	Lotus	Sainfoin
DM (g/kg)	286	313	325	292
pH	4.67	5.05	4.63	4.36
NH_3-N (g/kg TN)	106	103	83	5
CP	207	183	218	121
Lactate	54.2	21.4	16.0	26.4
WSC	15.4	6.7	49.9	29.3
GE (MJ/kgDM)	19.1	19.0	19.2	18.7
NDF	339	481	329	404

suggests this crop underwent a homolactic fermentation. With exception of sainfoin the legumes silages had comparatively high crude protein contents in excess of 183 g/kg DM. Extensive proteolysis was apparent in the red clover and lucerne silages, with ammonia-N contents exceeding 100 g/kg TN. The low ammonia content in sainfoin probably reflects the low protein status of the crop at harvesting.

Nitrogen (N) intake was highest on the red clover and lotus silages (Table 2). Although most of the N in the sainfoin silage appeared to be in an indigestible form, N digestibility was around 0.7 for the other three silages. The highest N loss in urine, 62 % of N intake, was recorded for lambs offered the lucerne silage. Whilst the N loss in urine was lower for the lambs offered the red clover and lotus silages, it still accounted for 40 - 50 % of N intake. These differences in N intake, N loss in faeces and N loss in urine led to statistically significant differences in the amount of N retained by each group of lambs, with the highest and lowest N balances recorded for lotus and sainfoin silages respectively.

Table 2: N utilisation by lambs offered ensiled legumes.

Forage:	Red clover	Lucerne	Lotus	Sainfoin	s.e.d.	F
N intake (g N/d)	51 ^{a}	39 ^{b}	55 ^{a}	24 ^{c}	3.2	***
N output in faeces (g N/d)	16 ^{a}	12 ^{b}	15 ^{a}	20 ^{c}	1.4	***
N output in urine (g N/d)	24 ^{a}	24 ^{a}	24 ^{a}	7 ^{b}	2.1	***
N balance	11 ^{a}	3 ^{b}	16 ^{c}	-3 ^{d}	1.9	***

F = Forage Type; $n = 24$; *** = $p<0.001$.
Values in the same row with different superscripts are significantly different ($p<0.05$)

Conclusions

N intake was comparatively high for the lambs offered the red clover, lucerne and lotus silages, confirming these crops as high-protein forages. However, further research is required to formulate feeding strategies that maximise the efficiency with which this N is utilised. For example, the substantial loss of N in urine indicates an inefficient capture and uptake of rumen degradable N, and suggests that there is scope to improve N retention by feeding these crops as mixtures in combination with high energy forages. The forages offered during the current experiment were prepared without additives, and it is possible that if a biological inoculant were applied proteolysis would be reduced, resulting in improved N retention.

Acknowledgements

This work was funded by MAFF and the Milk Development Council.

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Characterizing Protolytic Inhibition in Red Clover Silage

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Introduction

A major problem in silage production is the extensive protein degradation that occurs during the ensiling process. Native proteases degrade 44 to 87% of the plant's cytoplasmic protein into ammonia, amino acids and small peptides resulting in economic losses of up to $94.4 billion per year (based on 1994 U.S. silage production, predominately corn and alfalfa). Although red clover lacks many agronomic traits of alfalfa, silage produced from red clover has higher total protein available to the animal at feed out. Reduced proteolysis in red clover is not due to differences in the inherent proteolytic activity or protein composition compared to alfalfa. The improved true protein content of red clover silage appears to be due to the presence of a soluble polyphenol oxidase (PPO) and soluble polyphenols that inactivate hydrolytic enzymes upon exposure of cellular contents to oxygen. We have been characterizing the red clover system to gain an understanding of the molecular mechanisms underlying the observed decrease in proteolytic activity in red clover silage.

Methods

Immature leaves (0.2-0.3g fresh weight) of greenhouse grown red clover were harvested, placed in a 2 mL screw cap microfuge tube along with two glass beads, quick frozen in liquid nitrogen, and ground in the frozen state using a Mini-Bead Beater™ *. Samples were ground for 30 sec., refrozen in liquid nitrogen and ground again for 30 sec. Three grinding-freezing cycles were sufficient for complete homogenization. Polyphenol oxidase activity was measured using *O*-diphenol substrates and 2-nitro-5-thiobenzoic acid (Esterbauer et al. 1977). Proteolytic activity was measured using ninhydrin reagent (Moore, 1968).

Results

Extracts of red clover contain high levels of PPO activity (Fig. 1), as measured by the production of caffeoquinone (9 to 13.3 μmoles caffeoquinone min^{-1} g^{-1} FW). It appears that the caffeate ion (free acid group) is a preferred substrate compared to other polyphenols (e.g., chlorogenic acid, CGA and protocatechuic acid, ProCA). The phenyl propanoid side chain is critical for proper binding to the red clover PPO based on the low activity (20-30 fold less) using ProCA as a substrate. Chlorogenic acid, a caffeoyl ester conjugate, also has lower activity but not as severe. We have not been able to isolate other caffeic acid conjugates identified in red clover, clovamide and phasic acid, to determine if there is a higher or lower specificity for these molecules. There appears to be some genetic variation in total extractable PPO activity depending upon the red clover clone (see Fig. 1 for a comparison of two clones).

As red clover extracts are incubated in the presence of oxygen there is a rapid browning and a decrease in caffeic acid and its conjugates (Fig. 2). This general browning and more specific loss of caffeic acid (decrease light absorbance at 300 to 320 nm, Fig. 2) coincides with decreases in proteolytic activity. Using model proteases, representing both serine (trypsin) and cysteine (papain) classes of proteases, we have shown that red clover extract plus caffeic acid inhibits their proteolytic activity (Fig. 3). There was significant loss of proteolytic activity when enzyme preparations were pre-incubated (20 min.) with red clover extracts and caffeic acid. The loss of activity in our model system is similar to the loss of proteolytic activity seen in red clover juice and in alfalfa juice mixed with red clover extracts (Fig. 4).

Conclusions

It seems clear that the loss of proteolytic activity in red clover silage is due to the reaction of PPO generated caffeoquinones with specific proteases. There is the possibility of a synergistic role of other red clover phenolics in the rapid and effective inhibition of proteases and this is currently under investigation. We are also characterizing the major proteases in alfalfa and red clover to determine which ones are most susceptible to caffeoquinone inhibition and their role in protein degradation.

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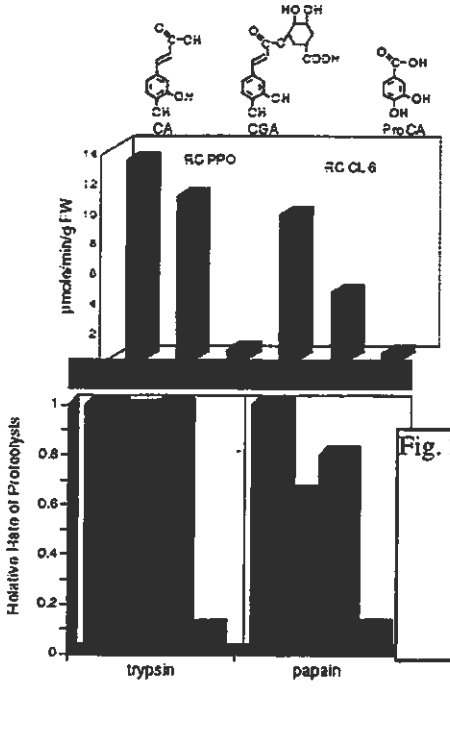

Fig. 1 Red clover PPO activity using three different polyphenols

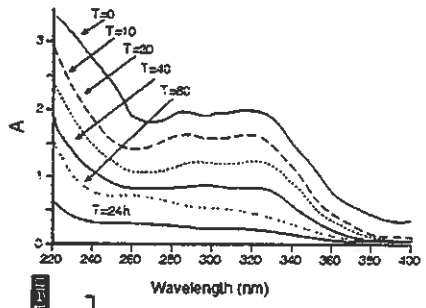

Fig. 3 Inhibition of trypsin and papain hydrolysis of BSA (20 mg mL^{-1}) using red clover extracts and caffeic acid. No add.= protease with BSA; Boil. RC= protease + BSA + boiled red clover ext.+ caffeic acid; CA= protease + BSA + caffeic acid; RC PPO= protease + caffeic acid + red clover extract.

Fig 4 Protolysis in alfalfa and
Alf = untreated alfalfa e
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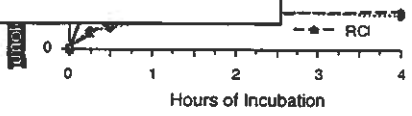
mixture of untreated al
extracts.



*Mention of a particular trademark or brand does not constitute an endorsement by the USDA-ARS.

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Effects of two additives and harvesting methods of wilted silage and concentrate level on milk production

Introduction

Ensiling wilted grass has increased in Finland during the last years. About a third of silage is wilted and roughly half of that is ensiled into big bales. Wilted silage is susceptible to aerobic deterioration and moulding. Propionic and benzoic acids have been found to possess antimicrobi- al properties against yeasts and moulds. After joining the EU it was forecast that the proportion of concentrate would increase in dairy cow diets in Finland due to a fall in concentrate prices. The objective of this experiment was to assess the effects of wilted precision chopped bunker and big bale silage ensiled either with formic acid or a mixture of formic, propionic and benzoic acid and two levels of concentrate supplementation (40 or 60% of DM) on milk production.

Materials and methods

The silages were made from a first-cut meadow fescue-timothy (*Festuca pratensis-Phleum pratense*) sward. Grass was cut with a mower-conditioner and harvested either, after 8-20 h wilting using a precision-chop forage harvester and ensiled in two bunker silos or baled into big bales after 24 h wilting, and wrapped with 50% overlapping and six layers of plastic film.The purpose was to wilt precision-chopped to 30% and big bale grass to 35-40% DM content. Both silages were ensiled either with formic acid (FA) applied 5 l/t as AIV 2 (800 g/kg formic acid and 20 g/kg orthophosphoric acid) or with an acid mixture 5 l/t (560 g/kg formic acid, 300 g/kg propionic acid, 90 g/kg benzoic acid and 50 g/kg stabiliser (FPBA), Valio Ltd, Farm Services). Forty-eight Finnish Ayrshire cows were used in a continuous design with a 2 (harvesting methods) x 2 (additives) x 2 (concentrate levels) factorial arrangement of experimental treatments. The experimental period of ten weeks was preceded by a 2-week covariance period. Silages were offered *ad libitum* with either 9 or 16 kg/d of concentrates throughout the experiment. Cows consumed only 15 kg/d on the average at higher concentrate level. The concentrate contained cereals and food industry by-products (crude protein 174, ether extract 47, starch 273, NDF 317 g/kg DM, ME 12.2 MJ/ kg DM). Feed digestibility was measured using four wethers and used to calculate metabolisable energy (ME) content. ME intake and utilisation was also calculated based on total diet digestibility for individual cows, determined during the sixth experimental week using acid insoluble ash as an internal marker. Data was subjected to analysis of covariance. Interactions between the main effects were not significant and therefore only main treatment effects are presented.

Results and discussion

Dry matter content of big bale silages was higher due to a longer wilting time and therefore the pH was higher and fermentation acid content lower than that of precision chopped silage. Fermentation quality of silages fed was good, and between additives there were only small differences (Table 1). FA-silage was more prone to aerobic deterioration manifested by warming and moulding before feeding during an exceptionally warm spring season, and twice (9.0 *vs.* 4.5 %) as much was discarded compared with FPBA-silage. About a quarter of big bales had moderate molding with half of the bales having only minor spots of moulds concentrated on the surface of the bales. Following fungal species were found: *Arthrinium phaeospermum*, *Scopulariopsis brevicaulis*, *Fusarium culmorum*, *Fusarium tricinctum*, *Penicillium* spp., *Mucor* and *Actinomycetes*.

Table 1. Silage chemical composition

	Dry matter	Crude protein	NDF	pH	WSC	Lactic acid	Acetic acid	Prop. acid	Butyr. acid	Valer. acid	Eth-anol	NH_4-N	Soluble N	ME
	g/kg	--g/kg DM--			----------g/kg DM----------							--g/kg N--		MJ/kg DM
Precision chopped bunker silages														
FA	305	162	555	4.63	97	24	13	0.7	1.9	1.4	5	51	604	10.6
FPBA	318	161	560	4.55	112	30	14	3.9	0.5	0.3	3	40	577	10.4
Big bale silages														
FA	423	162	572	4.90	102	7	8	0.1	0.0	0.9	4	34	587	10.7
FPBA	385	165	581	4.91	112	8	8	3.1	0.1	1.0	5	40	616	10.5

No significant differences were found between harvesting methods or additives on silage intake, milk and protein yield and protein content of milk, or in the efficiency of ME utilisation (Table 2). Milk fat content and yield were higher in milk produced with precision chopped than with big bale silage. There was a tendency (P=0.18) for lower milk and protein yields with FPBA-silage partly due to a lower digestibility of diet compared with FA-silage (OM 0.726 *vs.* 0.735). Sensory quality of milk was slightly lower with bunker than bale silage and for FA- than FPBA-silage possibly as a result of silage warming. Increasing concentrate supplementation from 9 to 15 kg/d, reduced silage intake with a substitution rate of 0.52, but increased milk, fat and protein yield and protein content. Milk yield response to concentrate was only 0.4 kg milk per kg concentrate increase due to a reduction in silage intake and total diet digestibility, particularly for fibre (OM 0.735 *vs.* 0.725, NDF 0.650 *vs.* 0.593) and also increased partitioning of energy towards live weight gain. Efficiency of energy utilisation for milk production or dietary protein for milk protein output was not affected by the level of concentrate supplementation.

Table 2. Effect of harvesting method, silage additive and concentrate level on milk production

Silage	Harvesting		Silage Additive		Concentrate		SEM	Significance		
	Prec. chop	Big bale	FA	FPBA	9 kg	15 kg		Har-vest	Addi-tive	Concen-trate
Intake (kg DM/d)										
Silage	10.8	10.8	10.8	10.8	12.2	9.4	0.15			***
Concentrate	10.5	10.7	10.6	10.6	7.9	13.3	0.12			
Yield										
Milk, kg/d	30.4	30.6	31.0	30.0	29.3	31.6	0.55			**
Fat, g/d	1306	1241	1286	1261	1238	1309	23.2	*		*
Protein "	959	954	972	942	912	1002	15.6			***
Milk composition										
Fat, g/kg	42.9	40.8	41.5	42.2	42.2	41.5	0.52	**		
Protein "	31.6	31.4	31.4	31.6	31.2	31.8	0.16			*
Smell & taste[1)]	4.02	4.14	4.02	4.14	4.07	4.09				
Feed efficiency										
ME utilisation[2)]	0.60	0.59	0.59	0.60	0.60	0.59	0.010			
Milk/feed prot.	0.27	0.26	0.27	0.26	0.27	0.26	0.003		*	
Live weight, kg	582	582	579	585	576	589	3.0			**
change kg/d	0.05	0.14	0.05	0.14	0.04	0.16	0.051			P=0.11

Statistical significance: * P 0.05; ** P<0.01, *** P<0.001. [1)] Scale 1-5 points.
[2)] Calculated from the intake of digestible OM determined in cows accounting live weight change

Conclusions

Animal performance of wilted precision chopped bunker silage and big bale silage were similar, when their quality was good. Additives did not prevent aerobic deterioration or molding of silage but a mixture of formic, propionic and benzoic acid decreased these parameters compared to formic acid. Production responses to higher concentrate feeding were small and the profitability of this feeding strategy depends on the on-farm feed prices.

The effect of applying different additives on the *in situ* rumen degradation of nitrogen and neutral detergent fibre in pea-wheat bi-crop silages.

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Introduction A major limitation to the efficient utilization of grass silage by ruminants is the asynchronous release of silage nitrogen and dietary energy in the rumen. This is because the high volatility of the ammonia in the silage is often not matched by the supply of a readily fermentable energy source such that microbial protein synthesis is sub-optimal. Additive treatment of silage can improve their fermentation and reduce this problem. Similarly, the use of pea-wheat bi-crop silages may overcome this problem because of a postulated synchronous supply of readily fermentable energy from the wheat and volatile nitrogen from the peas. However, little is known about the suitability of conserving pea-wheat bi-crops with currently available additives. Therefore this study evaluated the effect of different additives on the *in situ* degradation of nitrogen (N) and neutral detergent fibre (NDF) in pea-wheat silages.

Materials and Methods Pea-wheat (75:25; peas:wheat) bi-crops were harvested at growth stages 207 (full pod) and 72 (early milk) respectively for peas and wheat when the combined DM content was 301 g/kg. The bi-crops were treated with lactic acid bacteria based inoculants [*Lactobacillus buchneri* (LB; 10^5 CFU/g fresh weight; FW) or *Lactobacillus plantarum* (LP; 10^6 CFU/g FW)], quebracho tannins (QT; 16 g/kg FW) and formic acid (FA; 2.5 g/kg FW). Each treatment was replicated 6 times in laboratory silos made of polyethylene bags. The silage pH was measured directly from the silage juice after 112 days of ensilage, while chemical composition and rumen degradability of N and NDF were measured on freeze-dried samples. The model of McDonald (1981; $D = A+B(1-e^{-c(t-t_L)})$, was used to describe the degradation parameters, where D = degradation after t hours, A = the washing loss, B = the potentially degradable insoluble fraction, A+B = potential degradability, c = the fractional rate of degradation, and t_L = lag time i.e. the time before the commencement of degradation of B. The effective degradability (ED, McDonald, 1981) was calculated using a constant fractional outflow rate (k) of 0.04/h

Results All the silages appeared to be well fermented and had an average pH of 4.0. The mean silage crude protein and NDF contents were 144 ± 3.40 (SD) and 521 ± 33.2(SD) g/kg DM respectively. The *in situ* N and NDF degradation profiles of the bi-crops are presented in Figure 1, while the degradation parameters are shown in Table 1. QT and FA reduced ($P<0.01$) the A, ED and A+B fractions of both N and NDF compared to the other treatments. In comparison with the control, QT also reduced ($P>0.05$) the rates of degradation of N and NDF whereas, FA increased ($P<0.05$) the rates. QT was also more effective than LB and LP at reducing ($P<0.01$) the A, ED and c values and increasing ($P<0.05$) the B and A+B values. In comparison with the control silage, LB decreased

(P<0.05) the B and A+B values and increased (P<0.05) the c value. In contrast, LP decreased the A value for both N and NDF, decreased A+B value for N and ED value for NDF and increased the c value for N only.

Figure 1: *In situ* rumen degradability of nitrogen (N) and neutral detergent fibre (NDF)

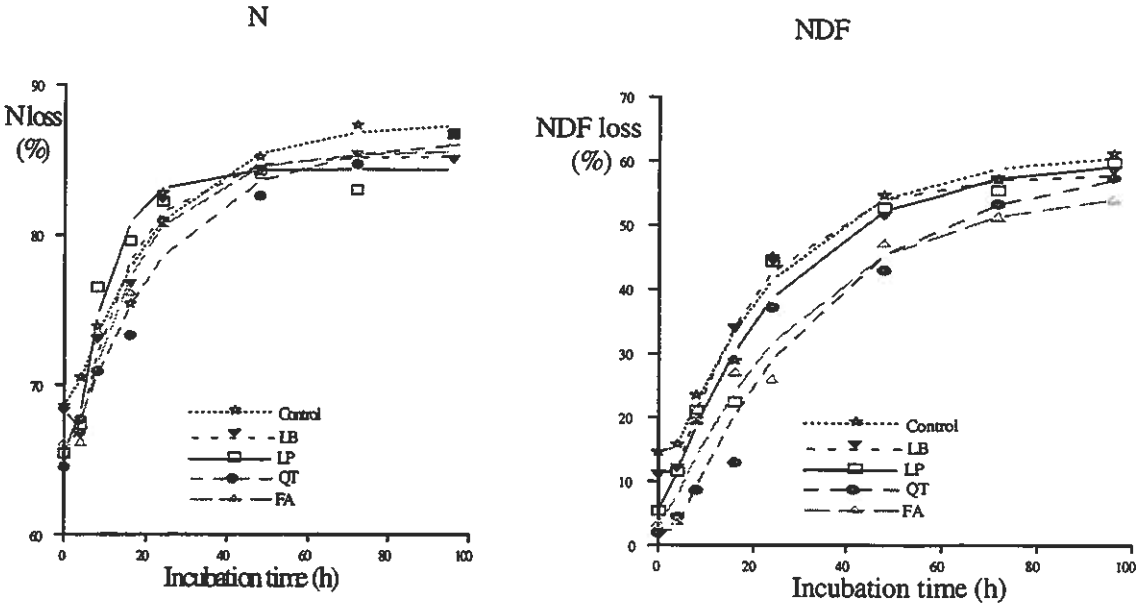

Table 1: *In situ* rumen nitrogen and neutral detergent fibre degradation characteristics of silages treated with different additives

	Nitrogen (%)					Neutral detergent fibre (%)				
Silage	A	B	A+B	ED	c	A	B	A+B	ED	c
Control	68.6	19.2	87.8	78.5	0.052	14.6	48.2	62.9	35.4	0.039
LB	68.2	16.7	84.9	77.8	0.091	11.2	47.3	58.5	34.8	0.053
LP	65.4	19.4	84.9	78.6	0.129	5.4	55.7	61.0	32.3	0.041
QT	64.5	21.9	86.4	75.6	0.043	2.0	59.8	61.8	23.8	0.030
FA	64.4	21.4	85.8	76.7	0.073	3.1	52.6	55.7	26.4	0.036
SED	0.32	0.60	0.54	0.44	0.009	0.82	1.47	1.49	0.72	0.006
Prob.	**	**	**	**	**	**	**	**	**	*

LB: *Lactobacillus buchneri*; LP: *Lactobacillus plantarum*; QT: quebracho tannins; FA: Formic acid; A: zero time washing loss; B: potentially degradable insoluble material; A+B: potential degradation; ED: effective degradability at k=0.04/h (where k: fractional outflow rate); c: rate of degradation (%/h) of B; SED: standard error of difference between means; **: P<0.01; *; P<0.05.

Conclusions Three of the additives used here (LP, QT and FA) appeared to exert an intra-ruminal protein-sparing effect. However, only QT slowed down the rate of degradation of N and NDF without reducing the extent of degradation. These effects may be due to the protein and carbohydrate binding properties of tannins. The slow degradation of the silage protein achieved with QT would probably be effecting a more synchronous supply of N and energy to the rumen and thus enhance the efficiency of microbial protein supply.

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The gastro intestinal tract (GIT) disappearance of amino acids in silages treated with tannins, formic acid or formaldehyde

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Introduction

Formaldehyde can be used to reduce proteolysis in silage. However, the recognition of the potentially carcinogenic effects of formaldehyde (Sigma-Aldrich, 1988) has made it less attractive. Tannins may be a less toxic alternative to formaldehyde. However, questions remain regarding the effect of low silage pH on the complexes between tannins and proteins and the release of protein by the dissociation of tannin-protein complexes post-ruminally. This study compares the effects of treatment of silages with tannins, formic acid or formaldehyde on the disappearance of amino acids in the gastro intestinal tract of dairy cows.

Materials and methods

Chopped first cut perennial ryegrass was ensiled in laboratory silos. The tannins (mimosa [MT] or quebracho [QT]) were added at a rate of 50 g/kg DM, while the formaldehyde (F) and formic acid (FA) were added at a rate of 12.5 g/kg DM. The additives were added in 20 ml aliquots/kg fresh weight. The treatments were control (C), C+FA, C+F, C+MT, C+QT, C+F+FA, C+MT+FA and C+QT+FA. Each treatment was replicated 4 times and the silos were opened after 49 days of ensiling and representatively sampled. Pooled samples were freeze-dried and ground to pass through 3.2 or 1 mm apertures for use in the GIT digestibility study in dairy cows and amino acid analysis respectively. The GIT digestibilities of amino acids were studied using the mobile bag technique (Hvelplund *et al.*, 1992). Amino acids in the silage samples and in the residual materials following incubation in the rumen, pepsin-HCl hydrolysis and passage through the intestine were measured using the modified high performance liquid chromatography method described by Salawu *et al.* (1997).

Results and discussion

The patterns of apparent disappearance of the amino acids in the GIT (Fig. 1) did not differ between the control silage and all the additive-containing silages. The only exception was for arginine (Table 1), where a significantly ($P<0.05$) higher proportion of the amino acid in silages treated with additives disappeared in the intestine. As can be seen from Table 1, individual differences also occurred between the amino acids in their actual disappearance in the intestine of the rumen undegradable true protein. These differences in disappearance between the amino acids may relate to differences in their affinity or the affinity of their side chain for the tannins or formaldehyde at both the low silage pH and the near neutral rumen pH. The proportion of rumen undegradable amino acids that were lost in the intestine was higher ($P<0.01$) in silages containing a

Fig. 1: The disappearance of total amino acid (true protein) from mobile bags in the rumen, pepsin-HCl solution and intestine

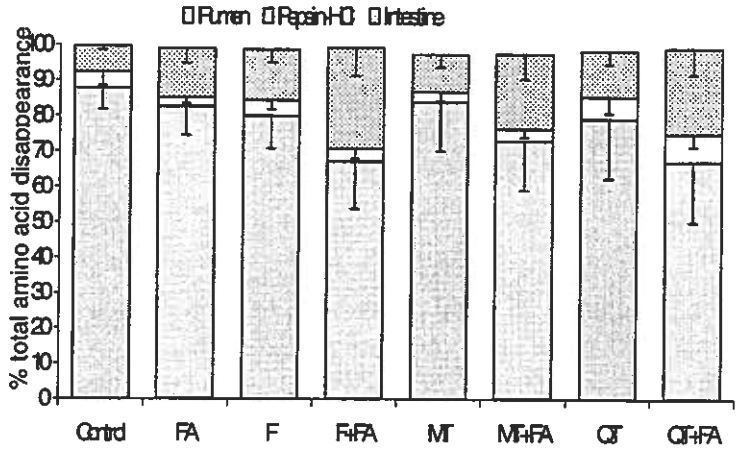

Table 1: Percentage (%) disappearance of amino acids in the intestine

Silage	Aspartate	Serine	Arginine	Methionine	Phenyl alanine
C	7.41	8.30	24.4	7.12	7.17
FA	14.6	15.8	35.9	5.92	16.7
F	13.7	18.0	36.9	7.32	17.8
MT	10.2	14.3	56.9	3.58	12.3
QT	13.4	17.0	58.9	5.20	13.1
F+FA	33.1	36.7	59.9	14.1	32.4
MT+FA	20.8	31.1	60.9	11.0	24.0
QT+FA	23.3	33.1	88.4	8.03	28.5
SED	0.97	1.11	2.53	0.78	1.13
Prob.	**	**	**	**	**

SED: standard error of difference between means;
**: P<0.01; %: percentage of the total disappearance

combination of tannins or formaldehyde with formic acid. This high effect may be due to the combined effect of the tannins or formaldehyde (protein binding) and formic acid (pH reduction) during the ensilage period. The differences between the tannins in their effects may be due to differences in their chemical structure, molecular weight and degree of polymerization (Reed, 1995).

Conclusion

Tannins may be used in a manner similar to that of formaldehyde to reduce proteolysis during ensilage and to shift the site of digestion of silage true protein from the rumen to the intestine where they can be digested and the amino acids absorbed with greater efficiencies. Further work to establish the effect of type and concentration of tannin on silage fermentation and quality are required.

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Effect of three levels of maceration on lucerne silage, nitrogen fractions, rumen degradability and voluntary intake.

Keywords: Conditioning, maceration, lucerne (alfalfa), silage, nitrogen, intake, degradability.

Introduction. Maceration is an intensive mechanical treatment, usually applied at the time of mowing. Fresh forage is fed through finely corrugated steel rolls turning at differential speeds, under a tight clearance (typically 1 mm between rolls). In previous studies, maceration has been shown to increase the field drying rate, the count of lactic acid bacteria at ensiling and the rate of lactic acid formation (or pH decline). Maceration has improved voluntary intake and ruminal degradability in some instances (usually under good conservation conditions) while, in other instances (usually under poor conservation conditions, i.e. prolonged wilting, rainfall), maceration has caused increased respiration loss and no benefit in animal production.

Object of study. Maceration can be applied at various levels of intensity, thereby producing various changes in the field drying rate, losses, silage fermentation and animal response. The object of this study was to compare three levels of maceration (light, intermediate, intense) in terms of forage quality, nitrogen fractions, ruminal degradability and sheep voluntary intake.

Materials and methods. Lucerne was mowed at three dates: (1) June 14, 1996 at 10% bloom in Lennoxville (southern Québec); (2) June 10, 1997 at early vegetative stage (EV) in Deschambault (central Québec); (3) June 24, 1997 at late vegetative stage (LV) in Deschambault. In 1996, four conditioning treatments were applied: 0x, one passage between two rubber rolls in a commercial mower-conditioner (the control); 1x, one passage through three macerating rolls turning at 850, 1040 and 2600 rpm and integrated in a self-propelled mower (prototype described by Savoie et al., 1999, Appl. Eng. Agr., in press); 2x, two passages through the same three macerating rolls by picking up the once-macerated windrow; 3x, three passages through the three macerating rolls. In 1996, the windrows were left in the field during 45 h under poor to moderate drying conditions (0.2, 2.8 and 0.2 mm of rain d^{-1} and 20.4, 18.3 and 15.3°C average temperature on June 14, 15 and 16, respectively). The wilted windrows were harvested and chopped at 9.5 mm theoretical length of cut. The four conditioned forages were stored at a wet density of 400 kg m^{-3} in 200-L drums. The 1996 silages were fed to 24 male sheep (6 sheep per treatment); voluntary intake was measured during the sixth week and forage digestibility was measured during the seventh week, by total feces collection. In 1997, four conditioning treatments were also applied: 0x, no conditioning (the control); 1x, 2x and 3x were the same maceration treatments as in 1996 but applied in the laboratory with a 3-roll macerator identical to the one used in the field. In 1997, windrows were wilted partly outside, partly inside to avoid rain. The wilting time was 22 h for EV lucerne (31.6°C average temperature) and 12 h for LV lucerne (25.4°C). In 1997, wilted forage was chopped at 4 mm and stored in mini-silos (4 L); there was no sheep feeding trial. In both years, all silages were analysed for chemical composition, silage characteristics, nitrogen fractions and ruminal degradability by the nylon bag technique in fistulated cows (procedures are described in detail by Agbossamey et al., 1998, Can. J. Anim. Sci. 78:399-405). Nitrogen was fractionated according to the Cornell protein system. Rumen degradability was reported as the effective degradability of dry matter assuming a 2% h^{-1} average rate of passage ($EDDM_2$).

Results. Experimental data are reported in Table 1. The dry matter (DM) was not significantly affected by maceration in 1996 (45 h wilting in rainy conditions) and in 1997 LV (short 12 h wilt). However maceration significantly increased DM (65.1 vs 44.5%) in 1997 EV (22 h wilt in hot weather). The crude protein (CP) was generally not affected by maceration. Neutral detergent fibre (NDF) increased significantly in 1996 and 1997 EV, probably due to leaching and respiration losses. The 1997 LV had less increase of NDF due to a short wilt without rain or excessively hot weather. Silage characteristics in 1996 indicated good conservation of 0x and 1x silages but very poor conservation of 3x silage, probably due to soil contamination as a result of two additional pickup operations. The 1997 EV silages had relatively high values of DM and correspondingly high pH. The 1997 LV silages had slightly lower pH and higher lactic acid after maceration. The nitrogen

fractions indicated that maceration reduced proteolysis with a lower fraction A (the soluble non-protein N). Most of the preserved protein was in the form of B_2 and B_3, i.e. moderately and slowly degradable protein. The effective degradability of DM ($EDDM_2$) from fistulated cows was similar at 0x and 1x, but tended to decrease at 2x and 3x. The voluntary dry matter intake (DMI) of the 1996 silages fed to sheep increased with maceration, from 2.31% of body weight (BW) d^{-1} at 0x, to 2.88% at 1x and 2.76% at 2x. The digestibility of silage in sheep was the same at 0x and 1x but decreased at 2x and 3x.

Table 1. Chemical composition, silage characteristics, nitrogen fractions and animal response after four mechanical treatments applied to lucerne at mowing: control conditioning (0x), maceration once (1x), maceration twice (2x), maceration thrice (3x).

Item	1996 Field Study				1997 Lab.; Early Vegetative				1997 Lab.; Late Vegetative			
	0x	1x	2x	3x	0x	1x	2x	3x	0x	1x	2x	3x
Chemical composition (% of DM, except DM)												
DM	34.6	28.0	38.3	33.7	44.5	62.4	67.3	65.6	33.0	38.8	35.1	29.8
CP	18.1	19.8	17.5	18.1	21.9	20.1	20.2	20.5	20.3	19.8	20.3	19.6
NDF	46.8	50.1	51.2	55.6	32.9	38.4	39.5	39.4	42.3	43.9	43.2	45.6
ADF	41.4	41.8	43.6	46.9	28.6	31.1	31.4	31.1	37.3	39.0	38.9	40.6
Ash	11.3	11.3	13.8	13.3	12.1	12.5	13.7	13.6	10.9	11.5	12.7	12.8
Silage characteristics (% of DM, except pH)												
pH	4.5	4.7	5.0	7.1	5.6	5.5	5.7	5.6	5.1	4.8	4.9	4.9
Lactic acid	5.71	4.53	4.03	0.07	2.49	1.45	1.01	1.27	3.45	4.47	3.73	4.62
Acetic acid	2.92	4.80	3.46	3.27	0.81	0.72	0.54	0.70	3.32	2.30	3.51	3.66
Propionic acid	0.07	0.13	0.10	0.98	0.04	0.09	0.11	0.16	0.23	0.05	0.12	0.10
Butyric acid	0.01	0.03	0.87	3.89	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Nitrogen fractions (% of total N)												
A	54.7	46.3	42.7	36.5	69.2	34.7	24.9	38.2	64.0	60.8	62.0	58.9
B_1	1.7	1.7	1.7	7.2	2.1	4.0	5.5	2.6	2.6	1.7	2.4	2.9
B_2	31.9	36.7	38.6	40.1	22.9	45.7	48.2	41.6	26.4	30.2	28.7	30.9
B_3	4.2	7.6	8.1	6.8	1.8	10.1	15.6	12.5	1.0	1.3	1.6	1.0
C	7.5	7.7	8.9	9.4	4.0	5.5	5.8	5.1	5.9	6.0	5.3	6.2
Animal response												
$EDDM_2$ (%)	65.6	65.5	63.8	58.6	70.6	67.4	66.1	64.9	60.9	61.4	60.3	61.5
DMI (% BW d^{-1})	2.31	2.88	2.76	2.41								
Dig. (%)	60.8	60.1	56.2	46.7								

Conclusions. Maceration of fresh lucerne can accelerate field drying and increase dry matter under good drying conditions. However, prolonged wilting under rain or high temperature increases the fibre content and reduces the rumen degradability and the digestibility of macerated lucerne. Voluntary intake is significantly increased after light maceration of lucerne silage fed to sheep. Light maceration (three rolls once) can improve protein quality by reducing proteolysis and increasing the fraction of slowly degradable protein but intense maceration (three rolls thrice) is not recommended in conditions of frequent rain or prolonged wilting.

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Digestibility and nitrogen balance in cattle offered silages made with different species and strains of lactic acid bacteria*

Introduction. Although there are many published examples of improvements in silage nutritive value following inoculation with homofermentative lactic acid bacteria at ensiling, few of these experiments were conducted under the conditions of inadequate to marginal fermentable carbohydrate supply that frequently prevail in Ireland. Fitzsimons *et al.* (1992) examined the potential of strains of *Pediococcus acidilactici* isolated from Irish silages or obtained from bacterial collections from different laboratories to utilise a range of substrates and to decrease pH during ensilage. They identified *Pediococcus* spp. (G 24) as capable of causing a more rapid drop in pH at the commencement of silage fermentation, when applied to grass of adequate fermentatable carbohydrate content and ensiled in laboratory silos. Duffner (1993) and Duffner *et al.* (1994) demonstrated the ability of *Lactobacillus plantarum* (DCU 101), isolated from an Irish silage and added at ensiling to grass of adequate fermentable carbohydrate content, to increase the rate or extent of pH decline. The objectives of the present experiment were to determine if the effects of *Pediococcus* spp. (G 24) and *L. plantarum* (DCU 101) when used separately or in combination were manifest on a farm-scale with grass of low fermentable carbohydrate content, and if digestibility and nitrogen balance in cattle subsequently fed the silage were improved.

Materials and Methods. Grass was ensiled from the first regrowth of a *Lolium perenne* sward in mid-July. Alternate pairs of loads of grass were ensiled following treatment with: (A) no additive, (B) *L. plantarum* (Ecosyl; Zeneca BioProducts and Fine Chemicals Ltd.; Control) at 3.2 l/t, (C) *L. plantarum* (DCU 101) @ 3.1 l/t, (D) *Pediococcus* spp. (G 24) @ 3.3 l/t or (E) *L. plantarum* + *Pediococcus* spp. (DCU 101 + G 24) @ 3.3 l/t. The target application rate for the bacteria added in treatments (B), (C) and (D) was 10^6 colony forming units (CFU)/g grass, while the target for each bacteria in treatment (E) was 5 x 10^5 CFU/g. Following 47 days storage, silages were offered *ad libitum* to crossbred continental heifers (417 (s.d. 23.4) kg mean initial liveweight; 14/treatment) and supplemented with 2 kg concentrates per head daily for 112 days. Simultaneously, *in vivo* coefficients for the digestibility of the various dietary fractions, together with nitrogen balance and the concentrations of blood metabolites, were determined using Friesian steers (425 (s.d. 32.3) kg mean initial liveweight) in a 5 (additive treatment) x 5 (period) Latin Square design experiment. Each period was of 28 days duration. For the final 10 days of each period, each steer was offered the appropriate silage at 0.9 of *ad libitum* intake and the quantity of concentrates offered was adjusted to a similar forage:concentrate ratio to the heifers. On day 18 of each period, blood samples were collected via jugular catheter 0.5 hours prior to feeding and 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 5.5, 7.5, 11.5, 15.5 and 23.5 hours post-feeding. Faeces and urine collection took place between days 21 to 28.

Results. Mean (s.d.) composition of the grass at ensiling was dry matter (DM) 138 (18.2) g/kg, *in vitro* DM digestibility 746 (20.7) g/kg, water soluble carbohydrates (WSC) 9 (3.7) g/l aqueous phase and buffering capacity 522 (38.1) mEq/kg DM. Total lactic acid bacteria on untreated grass were 1.1 x 10^6 CFU/g. The recovery rates of edible silage DM were 650, 700, 710, 720 and 760 kg/t DM ensiled for treatments (A) to (E), respectively. Mean silage DM intake by the heifers was 5.97, 5.91, 5.60, 5.85 and 5.87 (s.e.m. 0.135; $P>0.05$) kg/day for treatments (A) to (E), respectively, with corresponding carcass gains of 478, 473, 485, 483 and 506 (s.e.m. 18.6; $P>0.05$) g/day. The mean (s.d.) composition of the concentrates offered to the steers was DM 849 (4.2) g/kg, crude protein 146 (12.1) g/kg DM, ash 32 (1.6) g/kg DM, crude oil 27 (0.5) g/kg DM, crude fibre 58 (17.1) g/kg DM, neutral detergent fibre 263 (24.3) g/kg DM and acid detergent fibre 48 (7.7) g/kg DM. The mean composition of the silages offered to the steers is summarised in Table 1, and feed intake, dietary *in vivo* digestibility, nitrogen balance and blood composition are summarised in Table 2. Although

all silages underwent a lactic acid dominant primary fermentation, a secondary fermentation proceeded throughout feedout, with pH values and concentrations of acetic acid and ammonia-N progressively increasing (data not shown).

Table 1. Mean (s.d.) composition of silages offered to the steers in the *in vivo* digestibility and nitrogen balance experiment.

	No additive	*L. plantarum* (Control)	*L. plantarum* (DCU 101)	*Pediococcus* spp. (G 24)	*L.p.* + *P.* spp. (DCU 101 + G 24)
Dry matter[1]	170 (7.0)	163 (4.6)	168 (19.0)	165 (9.1)	168 (4.2)
Crude protein[2]	151 (2.5)	161 (12.5)	157 (8.4)	155 (8.7)	157 (8.7)
in vitro DMD[1]	699 (24.0)	666 (8.2)	668 (19.9)	646 (43.9)	668 (23.6)
NDF[2]	650 (34.0)	619 (52.6)	617 (71.9)	565 (44.2)	587 (21.5)
ADF[2]	366 (18.9)	354 (7.8)	358 (12.1)	374 (7.5)	368 (11.1)
Ash[2]	93 (4.0)	90 (2.1)	92 (3.6)	98 (3.8)	99 (12.2)
pH	4.30 (0.638)	4.22 (0.084)	4.22 (0.311)	4.52 (0.217)	4.26 (0.378)

[1]=g/kg; [2]= g/kg DM

Table 2. Silage additive treatment effects on *in-vivo* digestibility, nitrogen balance and blood composition.

	No additive	*L. plantarum* (Control)	*L. plantarum* (DCU101)	*Ped. spp* (G24)	*L. p.* + *P. spp.* (DCU101+G24)	s.e.m.	Sig
Silage DMI (kg/day)	4.88	5.03	5.01	4.98	5.02	0.173	NS
Concentrate DMI (kg/day)	1.56	1.61	1.61	1.54	1.57	0.054	NS
Total DMI (kg/day)	6.44	6.64	6.61	6.51	6.59	0.205	NS
Digestibility (g/kg)							
Dry matter	751	763	752	740	741	11.1	NS
Organic matter	769	783	772	760	776	10.4	NS
Nitrogen	693	706	689	677	677	16.4	NS
NDF	734	734	731	733	724	14.0	NS
ADF	696	695	684	684	675	13.2	NS
DOMD	706	724	713	698	698	9.2	NS
Nitrogen balance (g/day)							
Total N intake	154	157	158	161	157	4.7	NS
Digestible N intake	107	112	109	109	106	5.3	NS
Urine N	59	58	65	47	73	8.7	NS
N retention	48	53	44	62	33	11.4	NS
Blood composition (mmol/litre)							
β-hydroxybutyrate	0.39	0.49	0.46	0.47	0.44	0.072	NS
Glucose	4.32	4.27	4.25	4.29	4.30	0.066	NS
Non-esterified fatty acids	0.10	0.08	0.08	0.11	0.10	0.010	NS
Urea	3.80	3.84	3.38	3.82	3.75	0.234	NS

Conclusions. The crop used contained insufficient fermentable carbohydrate to sustain the dominance of a lactic acid primary fermentation. Under these conditions, none of the four bacterial inoculant treatments improved *in vivo* digestibility or nitrogen balance by steers, or altered the concentration of blood metabolites.

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Comparison of the effects of dietary nitrogen supplements on the intake and milk production of Finnish Ayrshire dairy cows fed grass silage-based diets.

A policy of being self-sufficient with respect to the production of protein sources to meet the protein requirements of dairy cows has lead to Finnish dairy cow rations traditionally being formulated from high crude protein (CP) silages supplemented with cereal based concentrates. Production of high CP content silages has been achieved using a combination of early cutting and high application rates of N fertilizer. The current experiment was designed to challenge this strategy by evaluating intake and milk production responses to increases in dietary N content derived from silage, urea, wheat gluten or rapeseed.

MATERIALS and METHODS

Two second-cut silages were prepared from grass swards containing Timothy (*Phleum pratense*) and Meadow Fescue (*Festuca pratensis*) fertilized with either 50 or 100 kg N/ha. Grass used to produced each silage was cut, wilted for 6h, picked up using a precision-chop forage harvester and preserved with a formic acid based additive, applied at a rate of 5.1 l/t. A basal concentrate (C) was formulated from (g/kg on an air-dry basis) barley (307), oats (460), molassed sugar beet pulp (200) and a proprietary vitamin and mineral mixture (33). Three additional isonitrogenous concentrates were prepared by replacement (g/kg) with urea (14.4), wheat gluten ((WG) 57.2) or heat-moisture treated rapeseed cake ((RSC) 188). The level of inclusion of additional N to concentrate C was designed to be equivalent to the differences in predicted N intake between silages.

Sixteen Finnish Ayrshire dairy cows (mean live weight 583 kg), were used to evaluate experimental diets according to a cyclic change-over design, with four 28 day experimental periods and a 2 (silages) x 4 (concentrates) factorial arrangement of treatments.

Cows were offered silage *ad libitum* and 10 kg/d of concentrates fed as two equal meals at 05.30 and 16.30 h. Feed intake and milk production was measured during the last 7 days of each experimental period. Milk composition was determined from samples collected over four consecutive milkings.

RESULTS

Silage prepared from grass fertilized with 50 kg N/ ha (S_{50}) had the following fermentation characteristics, pH 4.4, ammonia-N 39.1 (g/kg total N), lactic acid 15.2 (g/kg DM), total volatile fatty acids 10 (g/kg DM) and residual sugar 149 (g/kg DM). Respective values for silage prepared from grass fertilized with 100 kg N/ha (S_{100}) were 4.4, 63.2, 27.6, 14.5 and 93, respectively. Increasing N fertilizer application resulted in CP contents of 120 and 148 g/kg DM, for S_{50} and S_{100}, respectively.

The chemical composition of concentrates measured using standard procedures is shown in Table 1. Mean effects of silage and concentrate supplements on nutrient intake and milk production are presented in Table 2. Increases in silage N content failed to stimulate an increase in silage or total diet DM intake, and resulted in mean marginal daily decreases in milk and milk protein yield of 0.2 kg and 21 g, respectively. In contrast, increases in dietary N content derived from urea, WG or RSC resulted in increases in milk yield of 0.5, 1.2 and 3.2 kg/d, respectively. Furthermore, these sources resulted in respective marginal milk protein responses of 0.036, 0.167 and 0.220 g/g CP, demonstrating that utilization of dietary N for milk protein synthesis was improved when additional N was derived from true protein sources rather than urea. Dietary inclusion of additional N as RSC elicited the highest milk production responses driven by significant increases in DM intake.

CONCLUSIONS

Inefficient utilization of additional dietary N, indicates that application of N fertilizer should be based on grass sward N requirements, rather than as a means of increasing the CP content of grass silage-based dairy cow rations. Dietary inclusion of RSC resulted in the highest milk production responses, which irrespective of silage CP content were equivalent to increases in milk yield of 1.7 kg and milk protein output of 62 g / kg RSC inclusion.

TABLE 1. Concentrate chemical composition.

	Concentrate			
	C	Urea	WG	RSC
Dry matter (g/kg)	888	887	890	892
Composition of dry matter				
Organic matter	926	927	930	927
Crude protein	121	161	162	171
Neutral detergent fibre	266	268	253	283
Acid detergent fibre	113	116	110	139
Ether extract	29.6	29.8	28.4	41.7
Metabolisable energy [1] (MJ/kg DM)	11.9	11.8	12.1	12.1

[1] Calculated based on published ME values for individual ingredients (Tuori *et al.* 1996)

TABLE 2. Mean treatment effects on nutrient intake and milk production

	Silage		Concentrate				SEM [1]	Orthogonal contrasts [2]			
	S_{50}	S_{100}	C	U	WG	RSC		C1	C2	C3	C4
Intake											
Silage DM (kg/d)	12.5	12.6	12.2	12.6	12.4	13.0	0.09		**		*
Total DM (kg/d)	21.4	21.0	20.9	21.3	20.8	21.7	0.15				*
Total OM (kg/d)	19.5	19.2	19.1	19.4	19.0	19.8	0.13				*
Crude protein (g/d)	2863	3152	2700	3085	3012	3233	22.0	***	***		***
AAT (g/d) [3]	1901	1875	1824	1839	1890	2000	14.0		**	***	**
ME (MJ/d) [4]	243	236	237	239	236	247	1.70	*			*
Yield											
Milk (kg/d)	26.9	26.7	25.6	26.1	26.8	28.8	0.19		***	***	***
ECM (kg/d)	29.7	29.2	28.1	29.0	29.5	31.3	0.25		***	*	**
Fat (g/d)	1291	1268	1225	1278	1278	1337	15.5		*		
Protein (g/d)	893	872	837	851	889	954	6.68		***	***	***
Lactose (g/d)	1316	1301	1249	1273	1308	1406	10.9		***	***	***
LWT (kg)	595	597	594	595	596	599	1.86				
LWT change (kg/d)	-0.21	-0.25	-0.18	-0.23	-0.14	-0.36	0.06				

[1] SEM refers to standard error of silage means, corresponding values for concentrate means are 1.41 times greater. [2] Orthogonal contrasts; C1 (S_{50} vs. S_{100}), C2 (C vs. U, WG and RSC), C3 (U vs. WG and RSC) and C4 (WG vs. RSC). Significant differences at $P < 0.05$. $P < 0.01$ and $P < 0.001$ levels are indicated by *, ** and ***, respectively. [3] Calculated based on published values (Tuori *et al.* 1996). [4] Calculated based on predicted ME for silage based on OM digestibility (MAFF 1975) and published (Tuori *et al.* 1996) values for concentrates.

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Water-soluble browning products in heated herbages and silages of grasses

Key words: Forage grasses, Browning, Maillard

Introduction

When forages are ensiled and preserved, their protein quality often decreases due to the Maillard reaction (Van Soest 1982). This important reaction is a reaction of amino groups and reducing sugars, promoted by heating and produces browning polymeric products, melanoidins, in the late stage. The quality reduction of protein or the degree of protein denaturation has been evaluated on the basis of the nitrogen content of acid detergent-insoluble material. However, the heated and fermented forages also contain a considerable amount of soluble Maillard reaction products that could affect nitrogen utilization by ruminants (Van Soest & Mason 1991). The soluble Maillard reaction products are lost in laboratory tests based on solublility with enzymes or rumen fluid and in nylon bags and therefore little information is available on the chemical properties and nutritional and physiological effects of the soluble Maillard rection products. In the present work we isolated and characerized soluble browning products from heated herbages and silages of grasses.

Materials and methods

Timothy (*Phleum pratense* L.), perennial ryegrass (*Lolium perenne* L.), *Lolium* x *Festuca* hybrid derivative and quackgrass (*Agropyron repens* (L.) Beauv) were grown as pure swards and cut at the booting or heading stage. Round baled silages were made from wilted herbages of timothy and hybrid derivative.

Each fresh herbage of the grasses was heated in an air-tight can at 60°C for 24 h and then extracted with 0.1M sodium acetate solution with occasional stirring for 24 h at room temp. The acetate solution was obtained by filtration and applied to a reversed phase cartridge column pre-washed with ethanol and water. Material retained by the cartridge column was washed with water and then eluted successively with 20% ethanol (20% fraction) and 60% ethanol (60% fraction). The eluate was concentrated *in vacuo* and lyophilized. The soluble browning products in the round baled silages were also isolated by the same method as above.

Model Maillard browning products (GGH) were prepared by heating a mixture of D-glucose and glycine (1M : 1M) dissolved in 0.1M sodium hydrogen carbonate solution at 95°C for 24 h. The resultant brown solution was placed in cellulose tubing (MW>12000) and dialyzed against water. The nondialyzate was condensed *in vacuo* and lyophilized.

Results and discussion

Most part of the soluble browning products in the acetate solution was retained by the

reversed phase cartridge column for all the heated herbages and silages. The yields of the 20% and 60% fractions from the heated herbages ranged from 17.5 to 46.2 g kg^{-1} DM and from 5.8 to 9.5 g kg^{-1} DM, respectively. For the silage samples the 20% and 60% fractions were obtained in yields of 8.8-11.5 g kg^{-1} DM and 4.4-5.2 g kg^{-1} DM, respectively. Most part of GGH was also retained by the reversed phase cartridge column and eluted into the 20% fraction. The nitrogen contents of the 20% fractions from the heated herbages were 21.7-46.9 g kg^{-1} DM and those of the 60% fractions were 25.0-34.1 g kg^{-1} DM. GGH had the highest nitrogen content (60.1 g kg^{-1} DM).

All the soluble browning fractions showed general absorption spectra having no absorption maximum in the visible region (Fig. 1). When compared for the 20% fraction, the absorption coefficients (450 nm) were in the order GGH>>the silage samples≥the heated herbage samples, indicating more advanced Maillard browning in GGH. For both herbage and silage samples the 20% and 60% fractions showed quite similar IR spectra, in which some absorption bands characteristic of GGH, model melanoidin, were observed (Fig. 2).

Conclusion

The soluble browning products of heated herbages and silages of grasses can be isolated and fractionated by using the reversed phase cartridge column. The elemental and spectroscopic analyses indicated that the soluble browning products isolated were melanoidins or melanoidin-like substances.

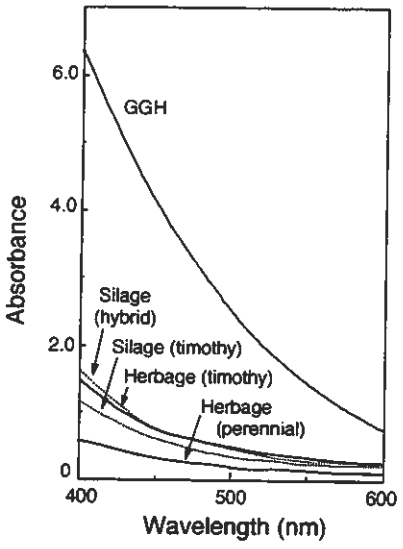

Fig. 1. Absorption spectra of GGH and the 20% fractions from heated herbages and silages.

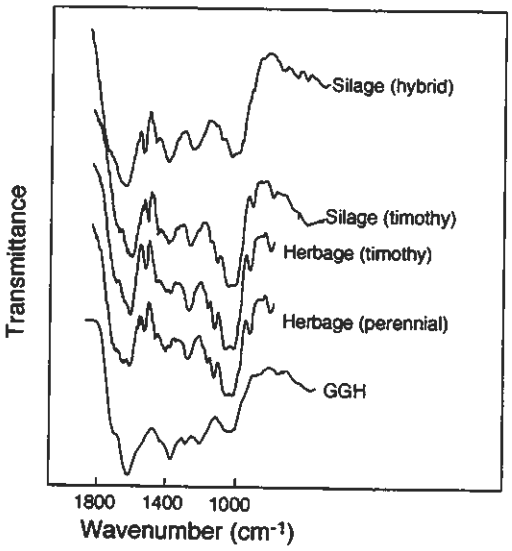

Fig. 2. IR spectra of GGH and the 20% fractions from heated herbages and silages.

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The influence of N-fertilization and botanical composition of the sward on the nutritive quality of silage.

Key words: botanical composition, nutritive value, N-fertilization, silage

The aim of this study was to determine the influence of two factors: botanical composition (of ensiled sward) and the levels of mineral N-fertilization on fermentation parameters and chemical composition of the resulting silage. During the years 1997-1998 studies were conducted on the nutritive value of silage. Four grass mixtures were ensiled. N fertilizer was applied in three doses: 120 kg, 180 kg and 240 kg per ha (N-120, N-180, N-240). The grass was ensilaged in laboratory silos. The silage were analyzed for dry matter (DM) content, pH, NH_3-N of total N and fatty acids: lactic, acetic and butyric by the Lepper method. Their value was presented in scores according to the Flieg-Zimmer scale. The silages were analyzed for: dry matter content, crude protein, crude fibre, crude ash and fat level in relation to dry matter by the use of NIRS technique. The concentration of net energy for lactation (NEL) in the silages was calculated using digestible coefficients.
No differences among grass mixtures were found in the nutritive value of silages. All silages, had, in spite of the botanical composition similar concentrations of nutritive components which indicated similar fermentation processes. N fertilization influenced the nutritive value of silage. Increased doses of mineral nitrogen increased the crude protein content and decreased the content of crude fibre and nitrogen-free extracts in the silage DM. DM level in the silages depended on N-fertilization and varied from 24,4 % (N-120) to 33,6% (N-180). The level of N fertilization also influenced fermentation parameters. With increasing N-doses, decreasing of lactic acid and increasing of butyric acid contents were observed. Silage from grass mixtures treated with low N doses had higher Flieg-Zimmer scores than silages treated with high N-doses. The botanical composition did neither influence the concentration of nutritive parameters nor the fermentation parameters of silages. However, higher doses of mineral N increased concentrations of butyric acid and decreased lactic acid in silages.

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Effects of nitrogen fertilisation of grass on fermentation in untreated and formic acid treated silage

Introduction

Both the extent and pattern of silage fermentation may vary considerably due to the characteristics of ensiled material. Plant species and agronomic measures have their own contribution to the ensiling process. The purpose of the present experiment was to assess how an increasing rate of nitrogen (N) fertilisation modifies the composition of grass and subsequent in silo fermentation of untreated or formic acid treated silage.

Materials and methods

A timothy grass (*Phleum pratense*) was fertilised with 0, 50, 100 and 150 kg N/ha. Grass was cut, wilted and chopped with a laboratory chopper. The four grasses were ensiled in laboratory silos of 0.13 l capacity using four additive treatments as follows: untreated control (C), formic acid (FAa) solution equal to 4 litres of pure FA/t grass, and FA based additives FAb (formic acid 550, ammonium formate 240, propionic acid 50, benzoic acid 10 and ethyl benzoate 10 g/kg, Kemira Chemicals Oy) and FAc (formic acid 734, ammonium formate 56 and ethyl benzoate 25 g/kg, Kemira Chemicals Oy) both 5 litres/t grass. Five replicate silos (80 g grass/silo) for each treatment were made. Silos were opened after 90 days for chemical analysis (two silos) and for the measurement of aerobic stability assessed using the temperature rise method (3 silos). In order to assess fermentation losses, total gas production during the ensiling period was measured from the silos. The results were tested by analysis of variance. Treatment effects were further separated using orthogonal contrasts for the following comparisons: untreated vs. FA additives, FAa vs. FAb + FAc, FAb vs. FAc, linear (L), quadratic (Q) and cubic (C) effect of N fertilisation rate, and the interactions between the main effects. The statistical significances are referred to in the text. Differences between the three FA (FAa, FAb, FAc) treatments were minor, and therefore treatment means are presented in this abstract only for C and FAa silages.

Results

Increasing N fertilisation resulted in a reduced dry matter (DM) and water soluble carbohydrate (WSC) concentrations and in an increased crude protein and nitrate concentration in grass (Table 1). During the ensiling process gas production was lower in FA silages than in untreated silages indicating lower fermentation losses (Table 2). The use of high rates of N resulted in a linear decrease in gas production in untreated silages but in FA silages production remained fairly constant (interaction $P<0.001$). Consequently, there were distinct differences in the fermentation quality of untreated silages due to fertilisation, whereas in FA silages differences were much smaller (Table 2). The pH value in untreated silages increased from 3.93 to 4.65 with the increasing N fertilisation rate but remained close to 4 in all FA silages (interaction $P<0.01$). Reflecting the changes in grass the WSC concentration in silages decreased with increasing fertilisation rate. Lactic acid concentration was higher in untreated silages, and it decreased with the increasing rate of N fertilisation, whereas in FA silages no differences were observed (interaction $P<0.01$). N fertilisation changed untreated silage fermentation towards a more heterofermentative type with higher contents of acetic and propionic acid and higher proportion of ammonia N. However, at the same time butyric acid concentration decreased from a very high value to zero, most probably due to increased nitrate concentrations. Experimental treatments had only negligible effects on the aerobic stability of silages during the first seven days after opening the silos.

Conclusions

N fertilisation of grass clearly modifies the fermentation process in untreated silage. When unfertilised grass was ensiled butyric acid concentration was very high while the values for other quality criteria were good. Nitrogen fertilisation prevents clostridial fermentation due to nitrate but also appears to lead to a heterofermentative type of fermentation and lower quality of untreated silage. This could be explained at least partly by the lower WSC concentration and higher grass buffering capacity. The use of formic acid based additives exceeds the effects of N fertilisation and the quality of silage was good irrespective of N fertilisation.

Table 1. Grass chemical composition.

	N-fertilisation, kg N/ha			
	0	50	100	150
Dry matter, g/kg	286	274	244	250
In dry matter, g/kg				
Ash	54	64	67	67
Crude protein	92	138	149	170
Water soluble carbohydrates	135	83	55	51
Nitrate	<0.2	0.3	3.9	4.8
Soluble N, g/kg N	393	299	336	349
Buffering capacity, meq/kg DM	239	297	321	304

Table 2. Silage fermentation quality.

Additive	Untreated				Formic acid			
N-fertilisation, kg N/ha	0	50	100	150	0	50	100	150
Dry matter, g/kg	291	280	242	252	294	271	248	255
pH	3.93	4.20	4.18	4.65	3.72	3.74	3.82	3.90
In dry matter, g/kg								
WSC	35	10	5	5	49	21	16	16
Lactic acid	64	60	50	43	34	39	35	32
Acetic acid	10	11	28	28	17	16	9	10
Propionic acid	0.0	0.2	0.5	2.2	0.0	0.0	0.0	0.0
Butyric acid	13.6	13.0	0.3	0.0	0.0	0.0	0.0	0.0
Isovaleric acid	0.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0
Total acids	88	85	79	73	51	55	44	42
Ethanol	11.5	10.0	5.5	7.8	4.6	5.7	6.0	5.1
Ammonia N, g/kg N	64	72	115	109	19	22	23	26
Soluble N, g/kg N	732	704	754	730	585	578	688	642
Gas, ml/80 g grass	527	645	266	138	166	79	139	141

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Development of timothy during progressing growth and subsequent nutritional implications

Maturity of grass at harvest determines the upper limit of nutritional quality of silage at feeding. With progressing development of plants, DM yield increases, but its digestibility and content of nutrients decrease. The digestibility of grasses is high in the northern areas of grass production, but the daily decline is fast especially during the primary growth due to reproductive development of the plants. The changes are not necessarily linear but are controlled by environment (e.g. weather).

Timing of the first harvest of grass is therefore an important decision on a dairy farm, which largely dictates the potential milk yield and/or need of concentrate supplementation. In an unpublished literature review (n=54), an increase of 0.01 in D-value increased silage dry matter intake by 0.155 kg/d, milk production by 0.30 kg/d and milk protein content by 0.12 g/kg.

In Finland Valio Ltd. collects field samples around the country and analyses them to assist farmers in optimizing the harvest time of grass. The objective of this study was to develop this service. The material is simultaneously used to develop laboratory methods and to model grass development.

Changes in timothy during primary growth

The development of timothy (*Phleum pratense*) was documented by sampling a second year timothy-meadow fescue ley twice a week in 1996, 1997 and 1998 in Jokioinen (61 °N), Finland. The fields were fertilized with 100 kg N/ha in the spring. Timothy was manually separated from the samples and divided into leaves and stems, which were analysed for ash, crude protein (CP) and organic matter digestibility (OMD) by an *in vitro* cellulase method.

The decrease in OMD of timothy was clear with progressing maturity (Fig. 1a). The variability in the results decreased, when OMD was expressed as a function of cumulative temperature (above 5 °C) from the onset of growth in spring (Fig. 1b). The CP content, which has traditionally been used as an indicator of optimal harvest time, proved to be less accurate (Fig. 1c). The decrease in OMD is closely connected with decreased proportion of leaves in the plant (Fig. 1d). The development was somewhat curvilinear because of a slow decrease of OMD early in the spring, and the development also levelled in the end of the sampling period. During the practically important phase, the decline of OMD was rather linear.

In vivo digestibility and microbial protein synthesis in the rumen

Each year 4 to 6 pilot scale silages were prepared at one week intervals from the same leys as described earlier. The grass was cut with a flail harvester and ensiled unwilted with 4 l formic acid per tonne of fresh grass. This preservation method was chosen to minimize the variability in quality, which could be caused by different wilting conditions and fermentation type. The silages were fed to adult wether sheep as a sole feed at maintenance level in 3 separate Latin square experiments. The digestibility of the silages was determined using total faecal collection, and synthesis of microbial protein in the rumen using excretion of urinary purine derivatives.

The decline in OMD was an average 0.0064 units per day. During 1996 and 1997 the decline was quite linear, but in 1998 there was some curvilinearity as the decline between harvests 2 and 3 was 0.0120 units, and between harvests 3 and 4 only 0.0036 units per day. The curvilinearity was decreased when OMD was expressed as a function of cumulative temperature (Fig. 2). Curvilinear development has been observed in our earlier studies as well emphasizing the need of good methods in choosing the time of harvest.

The production of microbial nitrogen (MN) in the rumen per kg DM ingested was on average 11.8 g. Production of MN increased with increasing OMD of silage (Fig. 3; non-significant in 1998). When MN production was expressed per kg digestible OM, it was not affected by silage OMD and was on average 18.3 g.

Conclusions

There seems to be potential to improve the estimation of nutritional quality (presented best by OMD) of grass. Advisory service utilizing environmental data connected with few calibration samples could be created. Modelling grass development is needed to improve the control of quality and quantity of feeds harvested. Ultimately also the regrowth(s) should be covered.

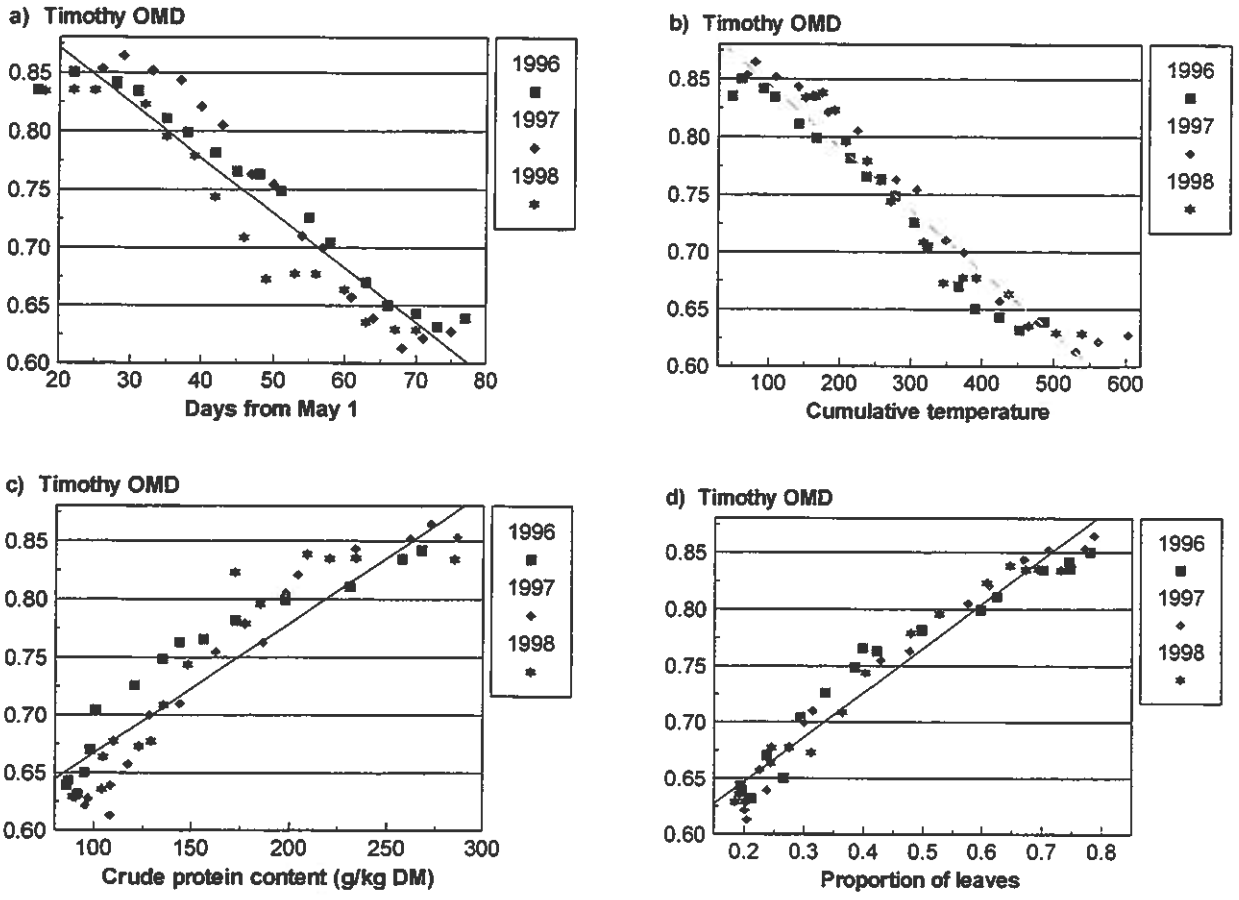

Figure 1. Timothy organic matter digestibility (OMD) as a function of date (a), cumulative temperature (b), crude protein content (c) and proportion of leaves (d).

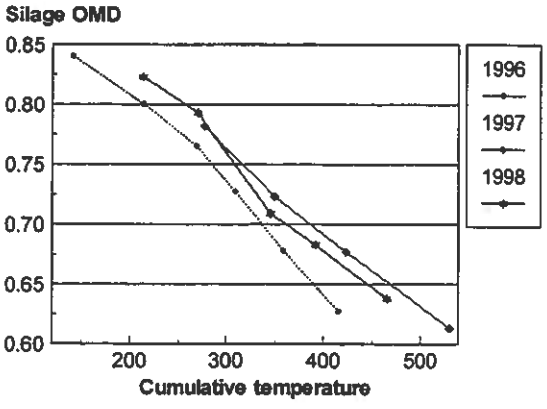

Figure 2. The OMD of silages as a function of cumulative temperature.

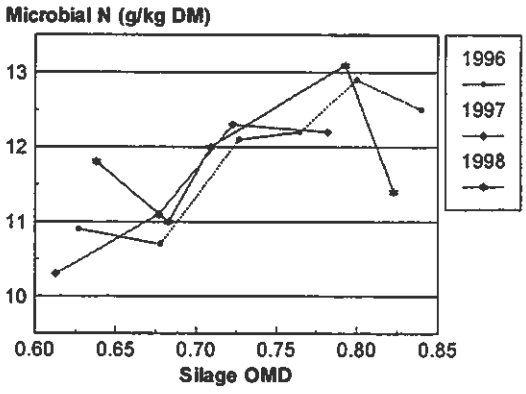

Figure 3. The effect of silage digestibility on MN production in the rumen.

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A rapid and economical technique for predicting free amino acid content in legume silage.

Introduction

It is well established that extensive degradation of protein occurs during ensilage. Conventional analyses currently used to evaluate silage quality do not quantify the extent of protein hydrolysis. Free amino acid concentration gives a measure of protein breakdown and is generally quantified in silages by carrying out full amino acid analysis which is both time consuming and expensive. We have devised a rapid method based on a modified ninhydrin assay for determining free amino acid levels in silage. Here we describe a study on the effects of bioadditives on proteolysis during ensilage of red clover and lucerne forages using this technique.

Material and methods

Second cuts of lucerne (cv. Vertus) and red clover (cv. Merviot) were harvested in July, 1998. Crops were wilted to approximately 35% and chopped with a precision chop forage harvester. Forages were then ensiled in 1kg silos with the following treatments; (a) control, (b) inoculant [*Lactobacillus plantarum* 10^6cfu /g FM (Live System, Genus, Crewe, UK)], (c) sugar [50:50 glucose:fructose, 14g/kg FM] and (d) combined inoculant and sugar. Duplicate silos were opened for each treatment on days 2 and 4 and triplicate silos on days 15 and 90. Samples were analysed for pH, WSC, nitrogen, and ammonia. Free amino acid content was determined by a ninhydrin colorimetric assay based on the method of Rosen (1957). The original assay was devised for quantifying amino groups following separation by column chromatography. Our current assay is based on analysing crude silage extracts with this technique. We have established that this technique gives a good prediction of free amino acid content in silages produced from a range of forages when compared with data obtained with an amino acid analyser after correction for ammonia content ($r^2 = 0.87$) see Figure 1.

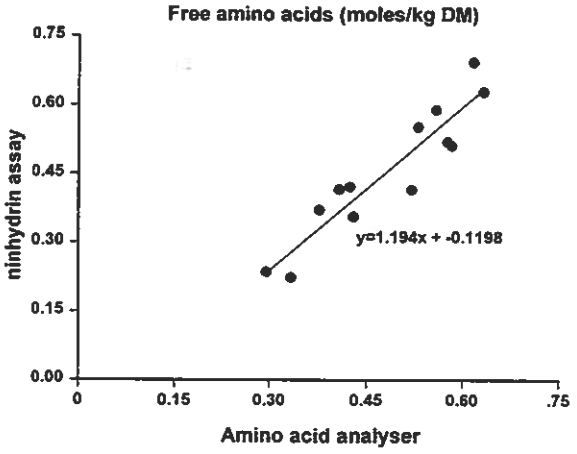
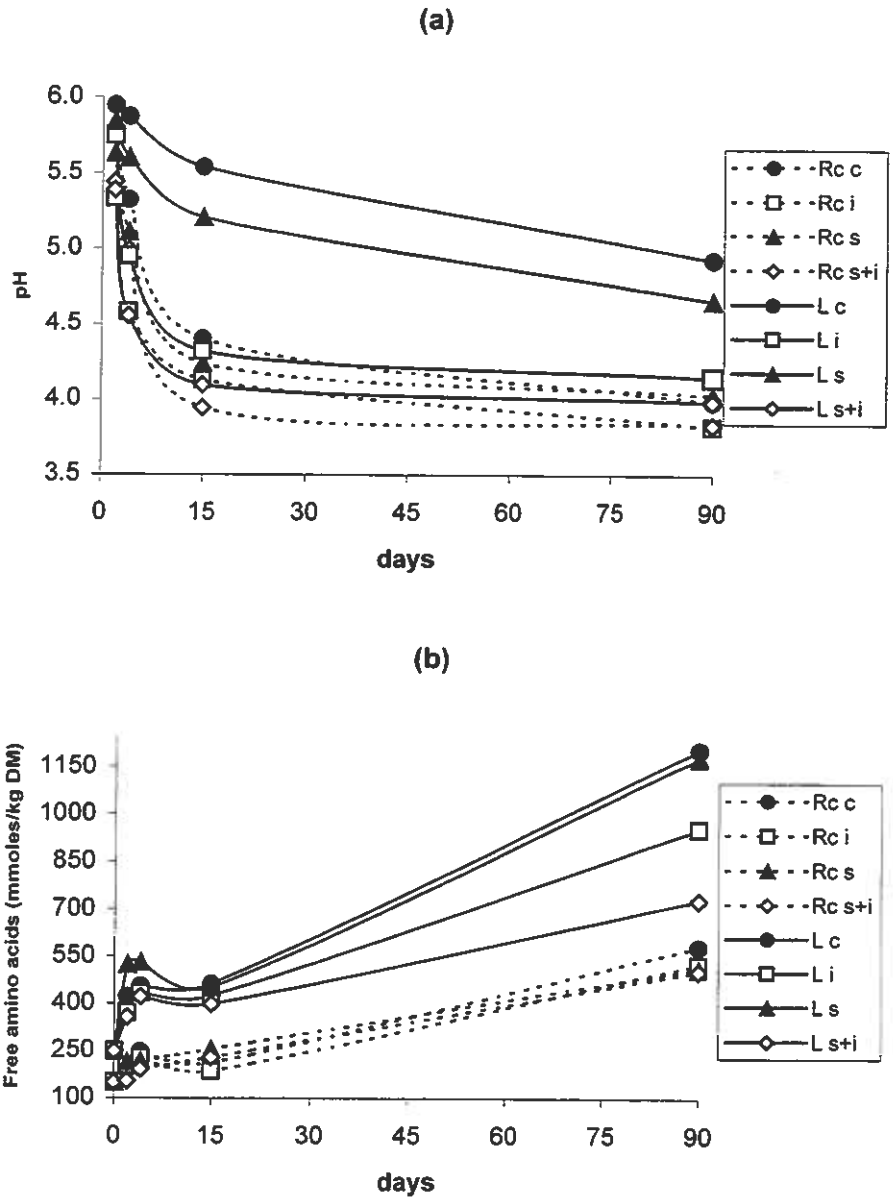


Figure 1. Comparison of free amino acid content determined by an amino acid analyser and the ninhydrin technique.

Results

Red clover silages and inoculated lucerne silages were well preserved with low pH and ammonia-N contents (< 30g/kg N). Changes in pH and free amino acid content during ensilage of red clover and lucerne are presented in Figure 2. Treatment with inoculant had a marked effect on the rate of pH decrease in lucerne silages and to a lesser extent red clover silages. This rate of decrease was accelerated when inoculant was applied in combination with soluble sugar. All red clover silages and inoculated lucerne silages had stabilised at a low pH (<4.5) by day 15. The predicted free amino acid content of lucerne silages was considerably higher than that of red clover silages throughout the period of ensiling. Predicted free amino acids accumulated rapidly during the first 2 days of ensilage but little increase was observed between days 2 and 15. All silages showed a marked net increase between days 15 and 90, irrespective of treatment. Whilst treatment effects were minimal with red clover silages, inoculant and combined sugar and inoculant treatments greatly reduced free amino acid levels in lucerne silages

Figure2. Changes in (a) pH and (b) predicted free amino acid content during ensilage of lucerne (L) and red clover (Rc) with and without additives (c = control, i = inoculant, s = sugar and s + i =sugar + inoculant).



Conclusion

Both treatment and forage effects on rates of proteolysis were observed using a ninhydrin based colorimetric assay. Addition of inoculant had a positive effect on silage fermentation charcteristics and silage quality. Treatment with sugar alone showed no benefit however it enhanced the effect of inoculant on lucerne suggesting that substrate supply was limiting. Changes in free amino acid content during ensilage showed a biphasic pattern with a rapid initial increase which stabilised by day 4 and a further increase after day 15 which occurred irrespective of forage or treatment. Initial rates of increase may have been due to combined plant and microbial protease activity whilst increases observed during the later phase were probably due to microbial activity although further study is required to elucidate the relative contributions of these activities to protein breakdown.

Rosen H. (1957) *Archives of Biochemistry and Biophysics* **67**, 10-15

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Protein content of a range of ensiled legumes

Introduction

Silage forms the major portion of the forage component of ruminant winter diets in the UK. Crops currently ensiled such as grass and maize produce silages have a low protein content and a high concentration of low molecular weight protein breakdown products. This can result in inefficient use of nitrogen by the ruminant with poor nitrogen retention and consequent losses to the environment in faeces and urine. The objective of this study was to examine the ensiling potential of a range of high protein legumes and establish how the extent of protein degradation is influenced by crop species and treatment.

Material and methods

Lotus (cv. Leo), lucerne (cv. Vertus) and red clover (cv. Merviot) were sown in May 1997. Regrowths were cut in September 1997. Lotus and lucerne were cut at the late bud growth stage and red clover at the early flower growth stage. Crops were cut using a mower with crimper conditioner and wilted over 48 hours. Forages were ensiled untreated or inoculated (*Lactobacillus plantarum* 10^6 cfu/g FM [Live System, Genus, Crewe, UK]) Bales were made with a Wolvo mini round baler and wrapped in six layers of film. Cored samples were taken from bales following 90 days of ensiling and these were analysed for pH, ammonia, nitrogen, WSC, starch, total and free amino acid content.

Results

The mean composition of all silages is presented in Table 1. All three crops benefited from application of inoculant with reduced pH and ammonia content and higher lactic acid and protein contents ($p<0.01$). Untreated lotus silages were poorly fermented with a low lactic acid content and a high WSC content relative to inoculated lotus silages. Ammonia contents were also low in these silages compared with untreated lucerne and red clover silages. Inoculated red clover silages had a notably higher ammonia content than that of inoculated lotus and lucerne silages. Both treated and untreated lucerne silages contained high levels of protein breakdown products (ammonia + free amino acids) compared with red clover and lotus silages.

Table 1. Composition of 90 day silages (g/kg DM unless otherwise stated)

	Lotus		Lucerne		Red clover		sed	sig.		
	u	i	u	i	u	i		Crop	Treat	C x T
DM (g/Kg)	325	312	324	396	279	271	18.1	***	NS	*
pH	5.14	4.35	5.56	4.51	5.34	4.51	0.186	NS	***	NS
WSC	42.7	22.8	17.1	19.5	10.4	9.5	2.90	***	**	***
Starch	16.9	16.2	19.3	19.1	13.3	13.4	4.91	NS	NS	NS
Lactic acid	9.4	34.5	26.5	43.9	56.3	77.5	8.35	***	**	NS
Acetic acid	5.6	6.2	15.6	12.9	15.5	21.2	5.67	*	NS	NS
Nitrogen	36.7	36.3	35.5	36.7	37.3	36.1	0.68	NS	NS	NS
Ammonia N[a]	26.2	19.1	61.3	17.6	78.2	58.4	6.02	***	***	**
Total AA[b]	47.1	49.3	46.7	55.2	44.0	48.1	1.11	**	***	*
Protein AA[bc]	30.9	34.0	27.1	32.9	30.6	34.8	1.21	*	**	NS
Ammonia+Free AA[b]	18.1	16.7	24.0	23.7	19.0	17.4	0.43	***	**	NS

U= untreated, i= inoculated, AA= amino acids, [a] = g/kg N, [b] = moles/kg N, protein AA = total amino acids – free amino acids.

Table 2 shows the concentrations of amino acids which have been found to be limiting in rumen microbial protein for ruminant animal production (Ragland-Gray, 1997).

Table 2. Concentrations of limiting amino acids in 90 day silages (moles/kg N)

	Lotus		Lucerne		Red clover		sed	sig.		
	u	i	u	i	u	i		Crop	Treat.	C x T
Methionine	0.49	0.49	0.48	0.47	0.41	0.44	0.066	NS	NS	NS
Threonine	2.15	2.24	2.22	3.11	1.95	2.34	0.147	**	**	*
Valine	2.78	2.89	3.45	3.50	3.09	3.17	0.194	*	NS	NS
Isoleucine	2.01	2.10	2.47	2.44	2.24	2.31	0.134	*	NS	NS
Leucine	3.46	3.63	3.74	3.95	3.77	3.88	0.124	*	NS	NS
Phenylalanine	1.79	1.88	2.08	2.07	1.90	1.93	0.067	*	NS	NS
Histidine	0.82	0.87	0.89	0.98	0.74	0.81	0.059	*	NS	NS
Lysine	2.23	2.44	1.60	2.63	1.65	2.14	0.184	NS	**	NS
Arginine	1.36	1.67	0.90	1.72	1.03	1.36	0.107	*	***	*

U= untreated, i=inoculated.

All silages had a similar profile of limiting amino acids. Methionine was uniformly low in all silages irrespective of crop or treatment. Losses of threonine, lysine and arginine were significantly reduced by inoculant treatment ($p<0.01$). This was most evident in lucerne silages where the respective concentrations of these amino acids were 40, 64 and 90% greater after treatment with inoculant. Lysine and arginine contents were relatively high in untreated lotus silages compared with untreated lucerne and red clover silages.

Conclusion

The extent of protein breakdown during ensilage is dependent on crop and treatment. Inoculant treatment had a marked effect on the composition of lotus, lucerne and red clover silages. A beneficial effect was observed on protein composition both in terms of protein content and levels of limiting amino acids. The very notable effect of inoculant treatment on threonine, lysine and arginine content in lucerne silages reflects a similar effect on ammonia content. These amino acids have been demonstrated to limit nitrogen retention in ruminants and furthermore lysine is considered to be limiting in terms of milk yield. Other workers have demonstrated that arginine and lysine have a relatively high rumen escape value when administered in an unprotected form (Velle *et al.* 1997). This suggests that their concentration in silage may be of nutritional significance. The less extensive protein breakdown observed in lotus and red clover silages, compared with lucerne silages, shows a relationship with nitrogen retention studies in sheep where more favourable nitrogen balances were observed with similar silages (Fraser *et al.* 1999). Further study is needed to elucidate the nutritional significance of the observed changes in concentrations of individual amino acids in silage.

Fraser M. *et al.* (1999). Nitrogen utilisation by lambs offered ensiled legumes. In *Proceedings for the XIIth International Silage Conference,* Uppsala, Sweden.
Ragland-Gray K.K. *et al.*(1997). *Journal of Animal Science*, 75, 3038-3045.
Velle W. *et al.* (1997). *Journal of Dairy Science*, **80,** 3325-3332.

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Workshop C
Methods to predict feeding value of silage based diets

Poster abstracts

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Nutritive and feeding value of conserved forages according to harvest and conservation methods : hay, silage, round bale wrapping.

INTRODUCTION

The nutritive value and DM intake of conserved forages are determined primarily by the quality of fresh forage at cutting. The milk production covered by forages increases by 0.4 kg for each point of OMd. The harvest and conservation methods have also an influence on the animals performance to a lesser extent. Hay-making reduces organic matter digestibility (OMd) and often also volontary intake, especially in conditions of bad weather and protracted field drying. This also holds true for wrapped round bales. Ensiling hardly reduces OMd, if at all, but results in a decrease, sometimes dramatic, of volontary intake and above all of protein value when conditions are bad, especially with uncontrolled butyric fermentation. Direct-cut or slightly wilted silage, made with a precision-chop harvester and preserved with formic acid, is nearly always of excellent quality, provided ensiling conditions are good.

MATERIALS AND METHODS

Different harvest methods have been compared at INRA (Orcival experimental farm) during four years. In each comparison, conserved forages were prepared in good meteorological conditions with the same forage (Upland, natural grassland) mown together and harvest immediatly with a precision-chop harvester for the direct-cut (DS), or wilted during two days for wrapped round bales (WRB) and drying on the field three days for hay (H). The direct-cut silage was preserved with a mixture formic acid (2/3)/formalin (1/3) applied at 3.7 l/t .
After six months of storage, the conserved forages were offered to lactating dairy cows (Holstein Friesan) after 10 weeks of lactation with the same level of concentrate and minerals in each comparison. The cows were divided in treatment groups (twelve animals) assigned on the basis of lactation number, calving date, weight, milk production and the same voluntary forage intake during the pre-experimental period. The experimental periods were 12 weeks long.

RESULTS AND DISCUSSION

For the three trials, Organic Matter digestibilty (OMd) averaged 0.656 and 0.644 and intakes were 13.3 and 13.0 kg DM (differences not significant) with the direct-cut silages and the wrapped round bales respectively. The FCM yield was 2 kg/day/cow higher with direct-cut silage than with wrapped round bales ($P<0.05$).
Comparing direct-cut silages and hays (n=3) similar results were observed. The OMd (0.656 and 0.642) and forage intake (12.7 and 12.5 kg DM) were not significantly different, but the FCM was also higher with direct-cut silages (+2 kg/day/cow).
The efficiency of hays and wrapped round bales were very similar as OMd, forage intake and FCM, for the three trials.
As a result milk yield of cows fed with DS were higher than with cows fed either WRB or H and liveweight gain of WRB and H groups were negative in most cases. Thus, the efficiency of direct-cut silages for milk production should be the highest. Similar effects have been observed with dairy cows in an experimental design of the three conserved forages (JP Andrieu, C Demarquilly, J Rouel - Ann. Zootech (1995) 44, Suppl, 371). This effect is not yet explained. Different digestive forage- concentrate interactions might be involved according to the type of forages as well as the nature and the amount and/or the metabolic efficiences of digestion end products.

The chemical composition of the forages (g/kg DM, unless stated otherwise) and performance of lactating dairy cows.

Treatment	DS	WRB	DS	H	WRB	H
Number trials	n=3		n=3		n=3	
Chemical composition						
DM	233	615	230	860	661	859
OM	905	912	896	902	912	908
CP	143	145	143	150	135	139
CF	316	329	310	295	315	300
PH	3.89	5.40	3.93		5.68	
Ammonia N (%N)	6.2	6.3	7.0		5.5	
Soluble N (%N)	44.7	40.2	43.6		35.8	
Lactic acid (g/kg DM)	50.2	3.4	47.6		2.5	
Acetic acid (g/kg DM)	15.8	3.7	18.5		2.0	
Butyric acid (g/kg DM)	2.8	0.4	3.4		0.3	
Alcohols (g/kg DM)	15.0	2.0	13.3		1.3	
OM digestibililty (%)	65.6	64.4	65.6	64.2	61.7	61.7
Animal performance						
DM intake (kg/day)						
Forage	13.3	13.0	13.0	12.7	12.7	12.5
Concentrate	5.1	5.0	5.4	5.5	5.7	5.8
4% FCM, kg	23.2^a	21.2^b	22.6^a	20.5^b	21.5	21.4
Milk fat (%)	39.2	39.8	39.3	39.6	40.1	39.6
Milk protein %	29.6	30.0	29.4	30.4	30.0	30.1
Liveweight gain (g/day)	+ 3	- 50	0	+ 6	- 80	- 40

In each trial, values with differing superscripts differ at 5% level.

CONCLUSION

There is an effect of the methods of conserving forage on the performance of stock, but this effect is smaller than that of digestibility at cutting. The choice of the harvest method must also considerate the meteorogical conditions with the success probability and cost. For each harvest method, a perfect mastership is required to obtain a good conservation. As a result the crop strategy (vegetation stages, wilted time) should be different.

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Effect of replacing lucerne silage with red clover silage in the diets of lactating dairy cows.

INTRODUCTION

Lucerne (*Medicago sativa*) is a major, high protein forage fed to dairy cows. However, during ensiling, as much as 60% of the CP in lucerne silage (LS) may be broken down to NPN; high levels of NPN in LS depress protein efficiency in lactating dairy cows (Nagel & Broderick, 1992). Polyphenol oxidase, an enzyme system in red clover (*Trifolium pratense*), converts phenols present in the plant into quinones; quinones react rapidly with forage proteins in the silo (Jones et al., 1995). Thus, red clover silage (RCS) typically has much lower NPN than LS (Albrecht & Muck, 1991). The objective of these two trials was to determine the relative feeding value of RCS and LS for lactating dairy cows.

MATERIALS AND METHODS

Red clover and lucerne were field wilted, chopped and ensiled in concrete stave silos. Four diets were fed in Trial 1: two containing 60% DM from LS or RCS plus 36% DM from ground high moisture corn (HMC), and two containing the same ingredients except that low-solubles fish meal replaced 3% of the DM from HMC. No attempt was made to equalize dietary CP. Twenty multiparous (four fitted with ruminal cannulae) and eight primiparous cows were randomly assigned to replicated 4X4 Latin squares. Diets were fed for 3-wk periods before switching (total 12 wk); yield and intake data were analyzed from wk 2 and 3 of each period. In Trial 2, red clover, lucerne, and a mixture of lucerne and red clover planted together, were field wilted, chopped and ensiled in concrete stave silos. Three diets were fed containing 60% DM from one forage, 32 to 36% DM from unground HMC, plus sufficient soybean meal to give about 16.5% CP. Twenty-one multiparous cows (three fitted with ruminal cannulae) were randomly assigned to replicated 3X3 Latin squares. Diets were fed for 4-wk periods before switching (total 12 wk); yield and intake data were analyzed from wk 3 and 4 of each period. Apparent DM and NDF digestibilities were estimated using indigestible ADF as an internal marker in fecal grab samples. Blood and ruminal sampling were done on the last day of each period in both trials.

RESULTS AND DISCUSSION

The LS fed in Trial 1 averaged 7 percentage units higher in CP than RCS; thus, RCS diets had about 4 percentage units less CP. The two forages contained similar amounts of NDF and ADF. In previous trials, RCS had only 1.5 percentage units less CP (DM basis) than LS containing equal NDF. As expected, RCS had less NPN: 31 versus 49% of total N. Intake of DM was lower on the two RCS diets and was not influenced by fish meal addition (Table 1). Without fish meal, milk yield was 1.5 kg/d greater on LS than on RCS, but there were no differences between these two diets in secretion of fat, protein and SNF (Table 1). Fish meal feeding resulted in similar responses in milk, protein and SNF yields on both forages; protein yield increased 70 g/day with fish meal. Feed efficiency (milk : DM intake) was greater on RCS than on LS; feeding fish meal removed differences between RCS and LS diets. This suggested that energy availability on RCS was greater than that on LS. Reduced blood and milk urea and ruminal ammonia on RCS were confounded by the lower CP contents of RCS diets.

Unlike Trial 1, CP levels of LS and RCS fed in Trial 2 were more like those observed in earlier studies where RCS contained 1 to 2 percentage units less CP than LS. Silage CP contents altered over the course of the trial--LS declined and RCS increased from levels used in diet formulation. Thus, the LS diet was slightly under (16.1% CP) and the RCS diet slightly over (16.8% CP) the target of 16.5% CP; the LS + RCS diet contained 16.4% CP. As expected, RCS had less NPN: RCS was 19% and LS + RCS was 23% lower in NPN than LS. The RCS and LS + RCS forages were, respectively, 2.3 and 4.2 percentage units lower in NDF than LS. Based on chemical composition, LS + RCS was more like RCS than LS. Intake of DM was lower on the RCS and LS + RCS diets, but apparent digestibilities of DM and NDF were substantially greater than on the LS diet (Table 2). There were no differences in yield of milk and milk components among the three diets (Table 2). Similar milk yields at lower DM intakes resulted in higher feed efficiencies on the RCS and LS + RCS diets. Although milk urea and ruminal ammonia were not different among diets, concentrations of blood urea tended to be lower on the RCS and LS + RCS (Table 2), despite similar levels of dietary CP. This suggested that efficiency of CP utilization was improved on the two diets containing RCS.

Table 1. Effect of feeding forage as Lucerne silage (LS) or red clover silage (RCS), with or without supplemental fish meal (FM), on performance of lactating dairy cows (Trial 1).

Item	LS	RCS	LS + FM	RCS + FM	SEM	$P > F^2$
DM intake, kg/d	24.2[a]	21.6[b]	24.1[ab]	22.4[b]	0.6	< 0.01
Weight gain, kg/d	0.16	0.48	0.34	0.48	0.12	0.21
Milk yield, kg/d	33.4[b]	31.9[c]	34.9[a]	33.7[b]	0.3	< 0.01
Fat, kg/d	1.20	1.12	1.20	1.14	0.03	0.19
Protein, kg/d	0.95[b]	0.92[b]	1.02[a]	0.99[a]	0.01	< 0.01
SNF, kg/d	2.74[bc]	2.68[c]	2.89[a]	2.84[ab]	0.04	< 0.01
Milk : DM intake	1.38[b]	1.47[a]	1.45[a]	1.51[a]	0.02	< 0.01
Blood urea, mg N/dL	17.8[b]	7.9[c]	20.9[a]	10.7[c]	0.3	< 0.01
Milk urea, mg N/dL	17.7[b]	7.2[d]	21.3[a]	10.7[c]	0.3	< 0.01
Ruminal pH	6.02	6.06	6.15	6.06	0.04	0.29
Ruminal ammonia, mM	16.1[a]	4.2[b]	17.4[a]	4.9[b]	0.6	< 0.01

[a,b,c]Means within the same row without a common superscript differ ($P < 0.05$).
[1]SEM = Standard error of the mean.
[2]Probability of a significant effect of diet.

Table 2. Effect of feeding forage as lucerne silage (LS), red clover silage (RCS) or a mixture of LS and RCS (grown together) on performance of lactating dairy cows (Trial 2).

Item	LS	RCS	LS + RCS	SEM	$P > F^2$
DM intake, kg/d	25.5a	23.0[b]	24.2[b]	0.4	0.01
Weight gain, kg/d	0.38	0.03	0.29	0.15	0.29
DM digestibility, %	56.3[c]	64.1[b]	67.6[a]	0.9	< 0.01
NDF digestibility, %	42.7[b]	49.9[a]	51.3[a]	0.5	< 0.01
Milk yield, kg/d	32.0	32.7	33.6	0.6	0.74
Fat, kg/d	1.08	1.12	1.21	0.04	0.82
Protein, kg/d	0.98	0.99	1.02	0.02	0.73
SNF, kg/d	2.75	2.81	2.90	0.06	0.75
Milk : DM intake	1.27[b]	1.43[a]	1.40[a]	0.04	0.05
Blood urea, mg N/dL	13.13[a]	12.89[ab]	12.40[b]	0.39	0.05
Milk urea, mg N/dL	9.92	10.46	9.60	0.40	0.29
Ruminal pH	6.15	6.05	6.15	0.05	0.23
Ruminal ammonia, mM	9.30	8.56	6.94	0.95	0.22

[a,b,c]Means within the same row without a common superscript differ ($P < 0.05$).
[1]SEM = Standard error of the mean.
[2]Probability of a significant effect of diet.

CONCLUSIONS

Intake of DM was depressed more than 2 kg/d on diets containing RCS compared to diets containing LS. Although there was lower milk yield on RCS than LS in Trial 1, milk yield was not different in Trial 2. There were no differences in yield of fat, protein or SNF due to forage source in either trial. Response to supplemental bypass protein (fish meal) was similar for both LS and RCS, suggesting that protein status limited utilization of both forages. Fiber and DM digestibility were greater on RCS. Higher milk yield per unit DM intake and depressed blood urea on diets containing RCS suggested energy and CP utilization were more efficient than on diets containing LS.

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Effect of Feeding Mixed Forage Diets on Milk Production.

Introduction

High intakes of forage are required to sustain good levels of milk production in winter. Intake of grass silage as the sole forage is often limited. Partial replacement of grass silage with good quality maize silage has increased forage intake and milk production. However, in areas not suitable for growing maize, other feeds need to be considered. These could include a limited amount of very high quality grass silage (~800gDMD/kg), wet by-product feeds e.g. ensiled pressed sugar beet pulp, or autumn saved pasture for feeding in early winter.

Materials and Methods

For 8 weeks in early lactation autumn calved cows were fed on a standard grass silage, which was harvested in late May, as the sole forage (S), or it was partially replaced with other feeds, e.g. a very high quality grass silage which was either unwilted (U) or wilted (W) prior to ensiling, or ensiled pressed sugar beet pulp (P), or autumn saved grass (G). Silage S was offered *ad libitum* to all cows (10 cows/diet) while the other feeds were fed at a fixed level of 5 kg DM/day on top of silage S. Supplement P contained 0.5 kg DM soyabean meal and 4.5 kg DM pressed pulp. The autumn saved grass (G) was cut daily and fed indoor. All cows received a standard concentrate supplement containing 180 g CP and 11.2 MJ ME/ kg fresh weight at 6 kg /cow/ day.

Results

The composition of the silages and other feeds are shown in Table 1. Silage S was high in DM digestibility compared with most first cut silages. Silage U and W were highly digestible, as planned, but were only marginally better than silage S. The digestibility of P was higher than that of the silages. The DM content of the grass was generally low and variable, and its DM digestibility declined over time (757-693 g/kg) as the amount of dead material increased.

Intake of silage S was considerably reduced (37-50%) by feeding the other supplements (Table 2). Total forage intake was not increased by feeding silage U or W, due to complete substitution (0.98 and 1.05, respectively) for silage S. Forage intake was increased by 12% and 8% by feeding P or G with S, with high substitution rates (0.77 and 0.82, respectively). Feeding silage U or W with S significantly increased milk yield (1.7-1.9 kg/day) but yield of fat and protein was not significantly increased as fat and protein concentrations tended to decline. Feeding grass (G) with S significantly increased milk yield (1.5 kg/day) and protein yield (+68 g/day) but not fat yield, and had no effect on milk composition. Feeding pressed pulp (P) had the greatest effect on milk yield (2.7 kg/day) and yield of fat and protein (+199 g/day) and improved milk protein concentration (+1.3 g/kg). Cow weights, liveweight gain and body condition score were similar for all treatments.

Conclusions

Despite the high digestibility of the standard silage (S), feeding other feeds with silage S increased milk yield on all mixed forage diets and increased yield of fat and protein on the SP and SG diets. Feeding pressed pulp resulted in the highest intake of forage and the highest level of milk production. Feeding grass in late autumn also improved milk production and achieved a considerable saving (37%) in silage intake. A greater difference in forage intake and in milk

production would be expected from the mixed forage diets if the digestibility of the standard silage was closer to normal first cut silage (~700gDMD/kg).

Table 1. Chemical composition of the silages, pressed pulp, grass and concentrates.

	Standard silage (S)	Unwilted silage (U)	Wilted silage (W)	Pressed pulp (P)	Fresh grass (G)	Concs.	Soyabean meal
Dry matter (g/kg)	199	184	288	216	132	872	861
Ash (g/kg DM)	86	92	96	74	107	61	59
C. Protein (g/kg DM)	146	169	183	123	302	217	540
NDF (g/kg DM)	475	434	412	461	471	133	77
ADF (g/kg DM)	305	289	258	245	252	104	52
WSC (g/kg DM)	22	24	69	76	59		
Invitro DMD (g/kg)	754	781	785	826	725	894*	924*
pH	3.62	3.82	4.14	3.80			
NH3-N (g/kg Total N)	95	88	69	33			
Lactic acid (g/kg DM)	129	105	60	41			

* Determined by neutral cellulase gammanase digestion method

Table 2. Effect of mixed forage diets on feed intake and on milk production

Treatment	S	SU	SW	SP	SG	sem	sig
Feed intake (kg DM/d)							
Standard silage	10.3	5..3	5.2	6.4	6.5	0.37	***
Other forage	-	5.1	4.9	5.0	4.7	0.024	***
Total forage	10.3	10.4	10.1	11.5	11.1	0.36	*
Total DMI	15.6	15.7	15.3	16.7	16.4	0.36	*
Production							
Milk (kg/d)	21.9	23.8	23.6	24.6	23.4	0.42	**
Fat (g/d)	970	1025	1006	1047	1018	28.6	NS
Protein (g/d)	706	742	741	827	774	18.6	**
Fat + Protein (g/d)	1676	1767	1745	1875	1790	44.5	*
Composition							
Fat (g/kg)	45.0	43.5	41.9	42.9	42.8	1.03	NS
Protein (g/kg)	32.9	31.5	31.0	34.2	32.4	0.58	**
Final live weight (kg)	562	559	559	554	568	14.7	NS
Liveweight gain (kg/d)	0.50	0.47	0.20	0.48	0.42	0.09	NS

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Effect of a biological additive on silage fermentation, digestibility, ruminal degradability, intake and performance of lactating dairy cattle in Galicia (NW Spain)

Key words: inoculant, silage quality, nutritive value, milk production

Introduction and object of the study

Rainfall occurrence during herbage harvest for silage in spring is fairly common in galician conditions. About 30% of silages made in dairy farms are additive-treated, half of them with formic acid. In spite of its proven efficacy in difficult ensiling situations when dosed and applied correctly, this additive is not well accepted by farmers, because of its corrosiveness and handling difficulties. Recently, biological additives are gaining popularity, although its effectivity in farm practise is uncertain, particularly under poor ensiling conditions.

In this experiment an inoculant additive, selected from a previous screening test carried out at the CIAM using laboratory silos, is compared with formic acid and a control without additive when applied to low-dry matter herbage with the object of investigate its mode of action on silage fermentation quality, nutritive value and dairy performance.

Material and methods

A first cut of a mixed sward (12 ha, predominantly perennial ryegrass) was directly ensiled using a precission-chop harvester, on 4 and 5 May 1996, in alternate loadings treated with a bacterial inoculant/enzyme preparation (I: Equiplant-Plus/Lalsil-PS®, Lallemand, France), at 7.1 g t^{-1}; formic acid (F: 850 g kg^{-1}) at 3.0 l t^{-1} and no additive (C: control), under rainy weather conditions. The treated and control silages were stored in unroofed trench silos (two per treatment). Additionally, twelve laboratory silos per treatment were made from core-sampled herbage, taken from trench silos at each trailer unloading. Sampling dates were 1, 3, 7, 14, 32 and 56 days post ensiling and eleven months later during the feeding experiment.

The three silages were offered *ad libitum* plus 7 kg $head^{-1}$ d^{-1} of a barley/soyabean meal concentrate (20% CP) to 24 lactating Friesian/Holstein cows in a change-over design with two experimental periods of four weeks each. A standard silage was used during the pre and post-experimental periods of 25 days. Individual silage intakes were recorded daily four days a week during the experiment using Calan-Broadbent doors. *In vivo* digestibility of the silages was measured using five sheep per treatment. Silage ruminal degradability was determined on three rumen-fistulated, non-lactating cows fed at maintenance with a similar diet to that used in the feeding experiment.

Results

The mean dry matter (DM) and water-soluble carbohydrates (WSC) contents of control herbage were 172 and 22.8 g kg^{-1} (fresh basis) respectively. The lactic acid concentrations after ensiling increased at a higher rate, and pH drop was faster for inoculant-treated silages than for the other two treatments, particularly when compared to formic acid silages. On day 56, respective values for I, F and C silages were; pH 3.88, 4.14 and 4.16 (s.e. 0.08); lactic acid 117.2, 65.5 and 86.0 (s.e. 18.9) g kg^{-1} DM; ammonia-N 54.9, 56.5 and 77.1 (s.e. 4.1) g (kg total N)$^{-1}$ and WSC 10.1, 7.2 and 5.9 (s.e. 0.5) g kg^{-1} DM respectively, as an average of laboratory and farm silos. Silo

type and interaction additive x silo type were not significant for those variables. Effluent output, measured in laboratory silos, was increased by formic treatment, with mean values of 66.8, 153.6 and 60.2 (s.e. 13.0) ml (kg herbage ensiled)$^{-1}$ for treatments I, F and C, respectively. Inclement weather in autumn and winter caused poor storage conditions, particularly during the feeding experiment. Analysis of the samples taken directly from trough along the control periods of the trial showed lower values of pH, butyric acid and ammonia-N of inoculated silage compared with control.

Digestibility of organic matter (OMD) of the silage was not affected by additive treatments. Control silage showed higher nitrogen (N) ruminal degradability than treated silages, whilst silage DM degradability was increased by the inoculant.

No significant differences were detected among treatments in silage DM intake, milk yield and milk composition, although absolute values of milk yield were lower for control silage, showing formic acid silage a trend towards higher milk fat content. Treated silages significantly increased daily yield of milk fat (F: +10%, I: +8.5%) and protein (F: +3%, I: +3.9%) with respect to control silage.

Table 1.- Chemical analysis and nutritive value of silages. Dairy performance

	Control	Formic	Inoculant	s.e.m.[1]	Significance.[2]
Oven DM (g/kg)	178.6	174.0	183.1	0.513	NS
Crude Protein (g/kg DM)	132.1	136.5	144.7	0.340	+
Acid Detergent Fiber (g/kg DM)	357.4	361.1	337.2	0.686	+
pH	4.35	4.39	4.14	0.079	+
Butyric Acid (g/kg DM)	13.0	7.40	5.38	0.926	+
Ammonia-N (g/kg total N)	97.2	72.0	57.8	0.812	*
OMD(%)	67.10	67.24	67.91	0.91	NS
N Degradability (%)	81.22	80.38	79.31	0.269	*
DM Degradability (%)	58.02	58.60	60.30	0.232	*
Silage DM intake (kg/day)	9.48	9.94	9.40	0.429	NS
Milk production (kg/day)	21.9	22.5	22.7	0.322	NS
Milk Fat (%)	3.21	3.40	3.26	0.073	NS
Milk Protein (%)	2.77	2.75	2.72	0.024	NS
Fat yield (kg/day)	0.69	0.77	0.75	0.025	*
Protein yield (kg/day)	0.60	0.62	0.62	0.018	*

[1] standard error of the mean [2] NS (non significant); + ($p<0.10$); * ($p<0.05$)

Conclusions:

It is concluded that the inoculant additive tested proved to be effective in improving fermentation quality of silage from low-dry matter herbage compared with a control without additive and, to a lesser extent, to formic acid. Additionally, inoculant reduced ruminal N degradability of silage and increased, at a similar level than formic acid, daily yields of milk fat and protein.

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The effect of wilting and ammonium tetraformiat (ATF) additive on big bale second cut grass fermentation and subsequent lamb feeding intake and performance

INTRODUCTION

In recent years, there has been a considerable increase in big bale silage made from second cut grass in Iceland. The variability of the silage quality seems to be considerable. By wilting the grass or by using additives it is, however, possible to stabilize the fermentation conditions and silage quality (McDonald et. al 1991). The present investigation was undertaken to test the effect of ammonium tetraformiat (ATF) additive on fermentation quality of second cut grass with and without wilting in big bales. The fodder was also tested in lamb feeding intake trials where growth performance of the animals was measured.

MATERIALS AND METHODS

Silage was made from a second-cut permanent field (dominating species *Poa pratensis*) cut on 24. August and baled on 24. and 25. August 1998. Four treatments were investigated:

- I O - direct harvested, without additive
- I ATF - direct harvested, 3.2 l/t ATF*
- II O - wilted, without additive
- II ATF - wilted, 3.8 l/t ATF

* GrasAAT™ ammonium tetraformiat -64.5% formic acid

During the wilting period (II) the grass was tedded three times. The weather conditions for wilting can be characterized as favourable i.e no precipitation during the field wilting period. The grass was baled with a flex-chamber round baler (VERMEER) and wrapped with 6 layers of plastic film. The bales were kept outdoor during the storage period.

Chemical analysis. Samples were taken from each bale at harvest and again after the ensiling period. The oven-dry DM values (60°C 24h) of the silage were not corrected for volatile products. The dry matter digestibility (DMD) was determined in vitro with the pepsin-cellulase method (Jones & Hayward 1975). Carbohydrates (the water soluble sugars, glucose, fructose and sucrose, WSC) and fermentation products were determined enzymatically (Boehringer Mannheim 1989) and crude protein with classical Kjeldahl method (MAFF 1986).

Animal feeding trial. Fourty lambs, 5 months old, were housed in four pens and offered silage ad libitum during a period of 48 days. The bales were grouped in 6 blocks (4 treatm. x 6 bales). The silage from each block (one bale) lasted for 7-9 days. Water and minerals were on offer at all times. The lambs were weighed each week. After the feeding period they were slaughtered and the carcass weight and the dressing out percentage determined.

Statistical analysis. t-test was used to examine the significance of the differences between the treatments using samples and DM-intake values from each 7-9 days period (block) as paired replicates.

RESULTS

The effects of the additive treatment and wilting (DM-level) on silage composition and the animal performance are presented in table 1. All the silages were rather well preserved - without notable fungal contamination or butyric acid odor. By wilting the material up to 42-43% DM the fermentation was significantly restricted, as indicated by the high WSC-level in the silage. The wilting increased the silage intake and the liveweight gain of the lambs. The treatment with ATF significantly decreased pH at both DM-levels, and the concentration of ethanol and the degradation of WSC at the lower DM-level. The additive treatment increased the silage intake at the lower DM-

level too. At the higher DM-level, however, neither the fermentation products nor the animal response, were significantly affected by the additive treatment.

Table 1. *Effects of wilting and ammonium tetraformiat (ATF) on the measured characteristics of silage made from second cut grass in big bales and results from lamb feeding intake trials.*

	I O	I ATF	II O	II ATF	Significance		
	a	b	c	d	a:b	c:d	ab:cd
Herbage composition							
Dry matter, g kg^{-1}	268	276	428	433	NS	NS	* * *
Buff. capacity (mEq kg^{-1}DM)	262	256	212	229	NS	NS	* * *
Silage composition							
DM, g kg^{-1}	246	253	416	420	NS	NS	* * *
pH	5.44	5.12	5.90	5.52	* *	* *	NS
Ammonia N, g kg^{-1} tot.N	102	92	51	48	NS	NS	* * *
Lactic acid, g kg^{-1} DM	15.9	14.2	3.5	2.8	NS	NS	* *
Acetic acid, –	5.9	5.6	3.1	2.6	NS	NS	NS
Ethanol, –	18.5	13.6	6.0	4.5	* *	NS	* * *
WSC, g kg^{-1} DM	21.1	54.9	101.2	105.3	*	NS	* * *
DMD- (in vitro) in silage at feeding/in grass at baling - ratio	0.97	1.01	1.00	1.01	NS	NS	NS
Silage intake, kg DM day^{-1}	0.78	0.87	0.92	0.95	* *	NS	* *
Weight gain, g day^{-1}	114	134	155	160	NS	NS	* *
Carcass weight, kg	12.3	13.3	13.4	13.7	NS	NS	NS
Dressing percentage	40.2	41.5	40.8	41.7	NS	NS	NS

The DM-intake of the silages showed the strongest correlation with the WSC content of all fermentation parameters studied (r=0.73, p=0.0001), confirming earlier results with first cut grass (Guðmundsson 1994). The WSC-content of the silages (y) was significantly correlated with the DM-content of the silage (x):

without ATF $y = 0.46 x - 8.96$ $r^2 = 0.93$ p=0.0001

with ATF $y = 0.32 x - 2.88$ $r^2 = 0.74$ p=0.0003

Therefore, the need for additive treatment with ATF of second cut grass seems to be limited to the DM-level up to x = 43% (where y1 = y2).

CONCLUSIONS

In conserving second cut grass in big bales, the emphasis should be put on harvesting techniques which restrict degradation of the WSC in the silage, by wilting under favourable weather conditions, and ATF-treatment of herbage with 40-43% DM.

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The effects of feeding AIV-2 and AIV-3 treated silages on the growth and efficiency of nutrient utilization of fattening bulls

Key words: silage, fattening bulls, chemical additives, metabolism, rumen.

Object of study. The objectives of this study were to determine the intake of AIV-2 and AIV-3 silages, the effects on the growth rate, fermentative processes in the rumen, nutrient digestibility and health of animals.

Introduction. The consumption and digestibility of silage depends on its quality, and the quality of silage depends on the silage making technology (W. Holmes, 1989). Formic acid and salts of formic acid restricts silage fermentation and DM losses (P. McDonald et al., 1991). That greatly affects nutrient digestibility, metabolism in the rumen availability of protein and other nutrients.

Materials and methods. A trial (98 days) was conducted at the Lithuanian Institute of Animal Science with 18 Lithuanian Black-and-White bulls allotted to three analogous groups by age (17-18 mo.), weight (400.6-402.0 kg) and weight gain. The bulls were maintained loose in individual pens and watered automatically. The diets were balanced according to the feeding standards. The bulls in Groups 1 (control), 2 and 3 (experimental) were fed, respectively, silage of ordinary fermentation *ad libitum*, AIV-2 (80% of formic acid, 2% of ortophosphoric acid, anti-corrosion compounds) treated (6 l/t) silage and AIV-3 (70% of ammonia tetraformiat, anti-corrosion compounds and blue dye-stuffs) treated (7 l/t) silage. High-quality silage was made in trench silos with a capacity of 50 tonnes from first cutting clover-timothy grass. The animals in all groups were additionally fed compound feed consisting of 88% of barley meal, 9% of soybean meal and 3% of mineral-vitamin mix and molasses.

Physiological studies carried out in the course of the trial included nutrient digestibility *in vivo*, microbiological and biochemical analyses of the rumen contents.

Results. The trial results indicated that average daily intakes of AIV-2 and AIV-3 treated silages were, respectively, 28.07 and 29.50 kg or by 1.87 and 0.69 kg less in comparison with untreated silage intakes (29.54 kg). Animals in all groups consumed the same daily amount of concentrated feed (3.0 kg) and molasses (0.57 kg). However, the digestion of silage treated with chemical additives was higher (Table 1). Protein digestibility was higher, respectively, by 5.56 and 5.71% and that of nitrogen-free extract by 10.61 and 5.47% compared with untreated silage. AIV-2 and AIV-3 treatment of silage increased the energy value of 1 kg dry matter by 0.28 and 0.19 MJ, and the daily contributions of metabolizable energy for animals in experimental groups were, respectively, by 3.82 and 4.06 MJ higher. Besides, intakes of digestible protein were by 28.66 and 41.48 g higher, too, in comparison with the control group.

Table 1. Silage digestibility (*in vivo*), %

Nutrients	Groups		
	Control 1	Experimental 2	Experimental 3
Dry matter	59.17±0.68	68.65±0.75	61.33±0.50
Organic matter	67.67±1.52	71.55±0.86	68.11±0.36
Protein	56.53±1.54	62.09±0.74**	62.24±0.47*
Fat	73.26±3.20	67.44±8.07	64.93±2.60
Fibre	70.47±0.47	71.62±1.03	70.68±0.74
Nitrogen free extract	63.70±1.45	74.31±0.87***	69.17±0.56**

*P < 0.05; **P < 0.025; ***P < 0.01.

The growth rate of bulls in all three groups was high, and the average daily gain was from 1.149 to 1.197 kg. The average daily gain of bulls fed untreated silage was 1.149 kg and that of bulls fed AIV-2 and AIV-3 treated silages was 1.178 and 1.197 kg, respectively. Thus, the growth rate of bulls fed AIV-2 and AIV-3 treated silages was, respectively, by 2.51 and 4.13% higher.

The treated silage has influenced microflora activity in the rumen. At the end of the trial, infusoria count and pH value increased, respectively, by 8.99 and 48.44% and 0.13 and 0.3 units, while VFA concentration decreased by 1.25 and 2.42 mmol/100 ml for AIV-2 and AIV-3 groups. AIV-3 treated silage was beneficial to protein synthesis in the rumen. At the end of the trial, the levels of total nitrogen, protein nitrogen, non-protein nitrogen and ammonia nitrogen were, respectively, by 6.58, 4.2, 2.27 and 0.59 mg/100 ml higher compared with the control group (Table 2).

Table 2. Microbiological and biochemical indicators of the rumen contents

Item	Group	At the end of pre-trial	At the end of trial
pH	1 (control)	6.63±0.14	6.59±0.06
	2 (experimental)	6.44±0.07	6.72±0.12
	3 (experimental)	6.87±0.07	6.89±0.18
Infusoria count thous.ml	1 (control)	277.8±111.9	285.7±32.8
	2 (experimental)	335.4±71.97	311.4±38.1
	3 (experimental)	393.6±84.01	424.1±126.0
Total VFA, mg/100 ml	1 (control)	10.14±0.38	12.11±0.25
	2 (experimental)	11.06±0.39	10.86±0.97
	3 (experimental)	9.69±0.95	9.69±1.28
Total nitrogen, mg/100 ml	1 (control)	100.45±12.38	103.76±9.94
	2 (experimental)	104.96±14.43	100.79±2.12
	3 (experimental)	100.92±13.85	110.34±9.81
Protein nitrogen, mg/100 ml	1 (control)	89.62±9.48	93.55±7.31
	2 (experimental)	93.95±12.12	90.94±1.52
	3 (experimental)	90.49±10.47	97.75±6.75
Non-protein nitrogen, mg/100 ml	1 (control)	10.83±2.92	9.32±2.63
	2 (experimental)	11.01±2.31	9.85±0.64
	3 (experimental)	10.43±3.37	12.59±3.10
Ammonia nitrogen, mg/100 ml	1 (control)	6.25±1.77	6.53±1.78
	2 (experimental)	7.93±0.79	6.34±0.33
	3 (experimental)	6.25±0.95	7.12±1.68

Conclusions

〈 Silage treated with chemical additives had a positive influence on the growth of animals. The daily gains of bulls fed AIV-2 and AIV-3 treated silages were respectively by 2.51 and 4.13% higher.

〈 The AIV-2 and AIV-3 affected rumen metabolism. At the end of the trial, infusoria count was by 8.99 and 48.44% higher and VFA concentration in the rumen by 1.25 and 2.42 mmol/100 ml lower in comparison with the untreated silage. The chemical additives had no negative effect on the animal health, and all blood values corresponded to the physiological norm.

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Use of macro in situ bag technique to evaluate the effect of maturity and mechanical processing on DM digestibility of corn silage

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Key Words: corn silage, macro in situ, maturity, mechanical processing

INTRODUCTION

The in situ technique has become a common method to estimate the digestibility of feedstuffs. Typically a feedstuff is incubated after it has been dried and ground to 1 to 2 mm. Doggett (1998) showed that drying and grinding of corn silage to 1 or 2 mm obscured differences due to maturity (1/2 milkline vs blackline) when using the in situ method. In addition, with the advent of mechanical processing of corn silages, differences due to processing would also be eliminated if corn silage was further processed to 1 to 2 mm prior to in situ incubations. Therefore, we have adapted the use of a "macro in situ" technique (Doggett, 1998) to estimate the ruminal digestibility of specific fractions of corn silage. The objective of this study was to evaluate the effect of maturity, mechanical processing, and hybrid on the rate and extent of DM digestibility as estimated by the macro in situ bag technique.

MATERIALS AND METHODS

The macro in situ bag technique was used to estimate the ruminal DM digestibility of 18 corn silages. Approximately 20 grams on a DM basis of wet unground corn silage was measured into the macro in situ bags. This differs from the conventional in situ method where approximately 4 grams of dried and finely ground corn silage is weighed into a nylon bag. The dimensions of the nylon macro in situ bags were 30 cm by 35 cm, whereas the conventional size of an in situ bag is 10 cm by 20 cm. For this study, corn silage was harvested during two consecutive years with and with out mechanical processing at a range of maturities. The first year one hybrid (Pioneer® hybrid 3845) was harvested at hard dough, 1/3 milkline, and 2/3 milkline, and the second year two hybrids (Pioneer® hybrids 3845 and Quanta) were harvested at 1/3 milkline, 2/3 milkline, and blackline (physiological maturity). All of the corn silage treatments were incubated in the rumen for 8, 16, 24, 48, and 96 hours. The first year each corn silage treatment was incubated in four different cows, and the second year each treatment was incubated in three cows. The instantly soluble portion of the corn silage was measured by soaking duplicate macro in situ bags with each corn silage treatment in cold water for 10 to 15 minutes. Dry matter disappearance and rate of disappearance were estimated for each corn silage treatment after subtracting out the instantly soluble portion of the corn silage. Rate of disappearance of the potentially degradable DM was calculated as the natural log of the disappearance of the potentially degradable insoluble fraction regressed against time.

RESULTS

Dry matter disappearance tended to be greater for processed corn silage at all time points at the advanced maturities (2/3 milkline and blackline). The earlier maturities had more variable results. Both hybrids harvested at 1/3 milkline the second year had a lower DM disappearance at 8 and 96 hours. Rate of DM disappearance for processed corn silage harvested at 1/3 and 2/3 milkline (year 1) and blackline (hybrid 3845 – year 2) had rates that were slightly slower than unprocessed silage.

This can be attributed to processed silage having greater DM disappearance at the earlier time points and similar DM disappearance at the later time points.

Table 1. Effect of mechanical processing of corn silage harvested at varying maturities on DM disappearance.

Pioneer® Hybrid 3845 – Harvested Fall 1996

	Hard Dough		1/3 Milkline		2/3 Milkline	
	Proc	Unproc	Proc	Unproc	Proc	Unproc
DM disappearance						
Instantly Soluble	25.7	22.0	20.8	21.8	17.9	19.6
8 h	14.6	8.1	18.3	8.6	11.9	8.4
16 h	23.8	16.2	22.7	15.9	21.6	15.4
24 h	30.6	22.2	34.6	26.0	30.5	25.9
48 h	48.6	44.7	50.1	48.2	46.3	45.2
96 h	58.3	55.7	61.4	59.3	59.0	58.0
Disappearance rate	3.81	3.76	3.49	3.92	3.30	3.50

Pioneer® Hybrid 3845 – Harvested Fall 1997

	1/3 Milkline		2/3 Milkline		Blackline	
	Proc	Unproc	Proc	Unproc	Proc	Unproc
DM disappearance						
Instantly Soluble	31.6	28.9	14.8	19.9	19.9	25.6
8 h	9.5	12.2	11.2	7.6	7.2	5.1
16 h	22.0	21.5	26.0	17.7	21.5	19.7
24 h	28.6	36.2	26.9	29.1	31.6	26.1
48 h	42.6	42.4	45.1	35.5	38.2	38.0
96 h	50.4	56.7	58.5	53.8	57.1	55.1
Disappearance rate	4.12	2.84	3.04	2.27	2.29	2.57

Pioneer® Hybrid Quanta – Harvested Fall 1997

	1/3 Milkline		2/3 Milkline		Blackline	
	Proc	Unproc	Proc	Unproc	Proc	Unproc
DM disappearance						
Instantly Soluble	37.4	31.5	33.8	31.0	34.6	30.7
8 h	4.7	8.4	10.0	6.3	3.8	3.7
16 h	14.4	19.9	18.3	11.9	20.3	14.5
24 h	34.8	31.0	31.3	11.3	29.5	19.6
48 h	47.3	39.9	41.6	30.1	41.9	34.5
96 h	54.4	58.0	58.9	52.8	56.3	48.3
Disappearance rate	4.99	2.46	2.62	1.82	3.13	2.90

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The effect of altering the amount of corn and grass acreage on whole farm economics and nutrient management – using a computer simulation model (DAFOSYM)

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Key Words: corn silage, grass silage, simulation, nutrient management, economics

INTRODUCTION

Environmental concerns have become more of an issue in recent years, and dairy operations are beginning to have strict manure management regulations imposed upon them in the United States and Europe. Farm management practices that are economically favorable are not necessarily environmentally friendly. Therefore, it would be beneficial to evaluate how a new management practice would effect the economics and nutrient balance of the whole farm prior to adoption of the practice. A dynamic computer model (DAFOSYM – Dairy Forage System Model) has been developed to evaluate whole farm economics and nutrient balances. The objective of this study was to determine how the ratio of grass silage and corn silage acreage affected profitability and nutrient balance on large and medium size dairy farms in the Pacific Northwest.

MATERIALS AND METHODS

Two commercial dairies (medium and large) in Western Washington were selected to collect information about the whole farm to input into DAFOSYM. The data collected included information about farm size, cropping, grazing practices, machinery, tillage and planting, harvesting, feed storage, animals, milking facilities, manure storage, and economic parameters related to the dairy operation. Seattle weather data were used to simulate crop growth on the respective farms. The amount of acreage planted in corn was altered from 1/3 to 2/3 of the total acreage for each farm, with the remaining acreage planted in grass. Simulations were conducted to evaluate differences in nitrogen flow and economic parameters over a period of 25 years.

RESULTS

The medium size dairy operation consisted of 95 hectare. The farm milked 427 cows, and the rolling herd average was 10,455 kg of milk/cow/year. The diet consisted of alfalfa hay, grass silage, corn silage, and two purchased grain mixes. The grass fields were harvested five times per year for wilted grass silage. The grass fields were irrigated with about 41 cm of water, and 75% of the manure produced annually was applied to the grass acreage. No commercial fertilizer was applied. Corn was harvested for silage at approximately 1/3 to ½ milkline stage of maturity. The corn was irrigated with 8 to 10 cm of water, and approximately 25% of the manure produced annually was applied to the corn acreage. Commercial fertilizer was applied once post planting at a rate of 14 kg N/hectare. Manure solids were removed using a separator system. The liquid portion of the manure was stored in a lagoon and applied to the crops in the spring and summer, and the solids were applied to crops in the spring, late summer, and fall. The large dairy operation consisted of 231 hectares. The farm milked 994 cows, and the rolling herd average was held constant in the simulations at 10,449 kg of milk/cow/year. In general, the diet ingredients consisted of alfalfa hay, corn silage, grass silage, corn grain, whole cottonseed, canola meal, Megalac®, whole potatoes, distiller grains, barley/beet pulp/molasses mixture, grain screening pellets, and a mineral and vitamin package. The grass acreage was harvested six times per year for wilted grass silage. The grass fields were irrigated with about 18 cm of water annually, and about 68% of the manure produced annually was applied to the grass acreage. Commercial fertilizer was applied at a rate of 54 kg N/hectare three times per

growing season. Corn silage was harvested in late September. The corn was irrigated with approximately 18 cm of water annually, and about 32% of the manure produced annually was applied to the corn. Commercial fertilizer was applied once pre-planting and once post planting at a rate of approximately 62 kg N/hectare. This farm used a flush system for manure. The solids were separated from the liquid and stored on a concrete slab. The liquid was stored in lagoons. The separated solids, bedded solids, slurry, and liquid manure were applied to crops in the spring, summer, and fall.

Table 1. Feed utilization, economic parameters, and nitrogen available, used, and lost to the environment for a 25 year analysis.

	Med 1/3 Corn	Med 2/3 Corn	Med % Change	Large 1/3 Corn	Large 2/3 Corn	Large % Change
Grass silage production (ton DM)	665	436	-52.5	1649	605	-172.6
Corn silage production (ton DM)	335	666	+98.8	1006	2110	+109.7
Milk production (kg/cow/year)	10,455	10,452	-0.03	10,449	10,449	-----
Net return per mature cow ($)	1685	1733	+2.8	1799	1796	-0.2
Return to mgmt/unpaid factors ($)	318,304	339,044	+6.5	534,658	531,238	-0.6
N from manure and fertilizer (kg)	61,572	59,797	-3.0	163,474	168,211	-2.9
N removed in crops (kg)	22,821	22,350	-2.1	60,536	56,563	-7.0
N lost to atmosphere (kg)	30,552	29,197	-4.6	79,085	80,695	-2.0
N lost to ground water (kg)	2436	2964	+21.7	12,272	13,081	+6.6
Unused soil N (kg)	5762	5286	-9.0	11,581	17,871	+54.3
Soil N used over that available (%)	74	73	-1.4	72	65	-10.8

The simulations with 1/3 versus 2/3 of the total acreage planted in corn predicted very different outcomes between the large and medium size farms. Results from the medium farm simulations, where the level of milk production was not maximized in the model, indicated that it would be economically beneficial to plant 2/3 of the total acreage in corn primarily because there was a reduction in feed expenses (7.3%). Nitrogen flow differed between simulations with more N being leached in the ground water (22%) and a slight reduction in the amount of soil N used over that available when 2/3 of the acreage was planted in corn. There was not an economical advantage to increasing corn acreage when simulations were done holding milk yields constant on the large farm. There was a reduction in purchased feed costs, however total feed costs, fertilizer costs, and labor costs increased as corn acreage increased from 1/3 to 2/3 of the total acreage. Therefore, the net return per mature cow and return to management and unpaid factors was similar between simulations. Unused soil N increased (54%) when corn acreage was increased. This can be attributed to the greater application rates of N fertilizer and to less N being removed by the corn compared to the grass crop.

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Determination of fibre content and digestibility of silage

Introduction. The digestibility of organic matter is one of the most important characteristics of the quality of silage. The determining of organic matter by classic *in vivo* methods requires much time and labour, therefore different research institutions have tried to improve various *in vitro* methods for determining digestibility. Comparison of grass feed digestibility determined *in vivo* and *in vitro* by DAISY II ANKOM's analyser showed a high correlation between them (r=0.960) (O. Kärt et al, 1997). Due to that in our investigation *in vitro* method was used. Since this method takes 3 days, simpler and appropriate methods for evaluating organic matter digestibility have been tried to find out.

Digestibility of silage is mostly affected by its fibre content. Different fibre fractions are determined - crude fibre by Weende system, neutral detergent fibre (NDF) and acid detergent fibre (ADF) by Van Soest system. Our aim was to study which fibre fraction was best correlated with organic matter digestibility and find shorter but rather precise method appropriate for evaluating the digestibility of silage. For that purpose we compared *in vitro* method by Tilley and Terry (1963) that is preferred in laboratories and in correlation with organic matter digestibility *in vivo* (Borba, Ribeiro, 1996) in the case of grass feeds, with the other methods.

Material and methods. In 1997/98 156 samples of silage's prepared of grass in different development stage were analysed in the Institute of Animal Science of the Estonian Agricultural University. During determining NDF, ADF, degradability and *in vitro* digestibility by ANKOM's analyser we followed the instructions given by the firm ANKOM. Also the standards of NDF and ADF, α-amylase (ANKOM Crop - #FAA), filter bags (ANKOM Crop - #F57) and reagents (solutions A and B) from the same firm were used. The results were statistically processed by programme Excel 97 and on the basis of calculated regression equations the quality of silage samples analysed in the laboratory in 1997/98 were evaluated.

Results and Discussion. The highest correlation occurred between OMD and ADF ($r = -0.914$; $P < 0.001$) and between OMD and crude fibre ($r = 0.906$; $P < 0.001$). Correlation between NDF and OMD was lower but also statistically significant ($r = -0.764$; $P < 0.01$) (Table 1).

We compared OMD gained by different methods with digestibility determined by *in vitro* filter bags method, analysing 156 silage samples. Correlation was highest between digestibility determined by *in vitro* filter bags method and digestibility calculated by degradability ($r = 0.946$; $P < 0.001$), also between *in vitro* digestibility and digestibility calculated by ADF ($r = 0.914$; $P < 0.001$). There was a weaker correlation between digestibility calculated by NDF and *in vitro* digestibility ($r = 0.766$; $P < 0.01$) and digestibility calculated by crude fibre and *in vitro* digestibility ($r = 0.594$; $P < 0.05$). Consequently, OMD of the feed should be calculated by its ADF content or dry matter degradability.

Conclusions. Of all fibre fractions ADF and crude fibre are in best correlation with feed digestibility. The digestibility of silage prepared of grass at its early development stage is better than of that prepared at later stages. The organic matter digestibility of silage prepared of graminaceous at ear formation stage was 69.6%, at early flowering 65.5% and at full flowering 58.4%. OMD of silage prepared of clover-rich grass (75% red clover) at flower bud formation stage was 72.1%, at early flowering 66.8% and at full flowering 59.4%.

Table 1. Fibre content and comparison of digestibility of silage determined by different methods

Raw material and development stages of silage	CF in DM %	NDF in DM %	ADF in DM %	OMD *in vitro* %	OMD * %	OMD ** %	OMD *** %	OMD **** %
Graminaceous silage:								
ear formation n=16	25.2	51.0	31.0	69.6	70.5	73.7	75.1	66.9
early flowering n=20	29.5	56.3	34.3	65.5	65.5	70.8	73.3	62.5
flowering n=7	32.7	62.5	39.6	58.4	57.4	62.2	71.6	57.4
Graminaceous-rich silage (25% clover):								
ear formation n=6	28.2	55.7	33.9	70.1	66.0	75.9	73.9	63.0
early flowering n=19	32.4	59.0	37.2	65.6	61.1	68.7	71.7	60.3
flowering n=5	34.8	66.8	41.3	53.3	54.8	54.7	70.5	53.8
Silage of clover and graminaceous (50% clover):								
blossom bud formation n=22	26.01	46.4	30.8	69.7	70.9	69.9	76.8	70.7
flowering n=16	31.5	57.6	36.4	63.3	62.3	66.3	74.5	61.4
Clover-rich silage (75% clover):								
blossom bud formation n=13	24.7	44.5	30.7	72.1	71.1	76.1	79.1	72.2
early flowering n=12	27.1	47.4	34.5	66.8	65.3	67.2	78.3	69.8
flowering n=13	32.9	56.5	39.4	59.4	57.7	62.6	76.4	62.3
Clover-rich silage (90% clover):								
early flowering n=3	26.5	42.8	31.2	65.7	70.3	66.1	78.6	73.6
flowering n=2	33.1	56.9	40.4	58.9	56.1	58.6	76.4	62.0
Galega:								
flowering n=2	25.1	48.4	31.2	73.9	70.3	84.4	79.0	69.0

*OMD = 118 - (0.153 x ADF g/kg) (Tilley and Terry, 1963)
**OMD = 6.51 + 1.24 x degradability *in vitro*) (Tilley and Terry, 1963)
***OMD red clover = 87.3 - 0.33 CF (Kivimäe, 1959)
***OMD timothy = 88.6 - 0.52 CF (Kivimäe, 1959)
****OMD = 109 - (0.0826 x NDF) (Borba and Ribeiro, 1996)

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Voluntary intake and digestibility of oil palm fronds silage.

OBJECT OF STUDY

Oil palm frond (OPF) is one of the most abundant agricultural by-products in Malaysia. Almost all of pruned fronds are discarded in the plantation. OPF is expected to become a promising feed resource for Ruminants. Several preservation methods of OPF have been shown, such as drying, forming to pellet, silage and so on. However, ensiling may be the most suitable method to preserve the OPF at low cost. Thus, this study was carried out to elucidate the voluntary intake and digestibility of four different types of processed OPF, such as silage, NaOH treated silage, drying and pellets.

MATERIAL AND METHODS

The OPF was chopped with an engine driven chopper at the length of approximately 1cm. The chopped materials were subjected to four treatments as follows: 1.OPF silage (SILAGE) where chopped OPF was stocked in 100L drums for 1 month.2.NaOH treated OPF silage (N-SILAGE) where chopped OPF was stocked in 100L drums after mixing with 15kg of 10% NaOH solution and 100kg of fresh OPF.3. Dry OPF (DRY) where OPF was chopped twice and drying. 4.OPF pellet (PELLET) which involved drying, grinding and compacted to pellets of 12mm in diameter.

Voluntary dry matter intake and digestibility of four kinds of processed OPF at *ad libitum* feeding were measured with total feces collection method using 16 Kedah-Kelantan (KK) cross yearling heifers weighing approximately 160 kg. The experimental duration consisted of an initial 6 days adjustment period and 3 days measurement period. Each processed OPF was mixed with the basal ration, which was consisted of 50% palm kernel cake, 20% palm oil mill effluents, 16% tapioca waste (dry), 10% rice bran, 2% mineral vitamin mixture, 1% salt and 1% urea. The digestibility of the basal ration was 69.5%. Each processed OPF was mixed with the basal ration at 25, 40 and 60% on dry matter basis, respectively. These mixed rations were fed to animals *ad libitum*. The animals were offered the mixed ration in two equal feeds a day (9 a.m. and 4 p.m.).

RESULTS

The pH of SILAGE and N-SILAGE were 3.62 and 5.74.All types of OPF had almost same CP, EE and NDF contents except for high ASH content of N-SILAGE (Table 1). Voluntary dry matter intakes of SILAGE, N-SILAGE and DRY with basal ration were lower than those of PELLET with basal ration. On the other hand, the digestibility of SILAGE, N-SILAGE and DRY *per se* were higher than those of PELLET *per se*. Therefore, digestible dry matter intake of SILAGE and DRY *per se* were almost similar to those of PELLET *per se*. On the other hand, the digestible dry matter intake of N-SILAGE *per se* was higher than those of other processed OPF (Table 2).

CONCLUSIONS

Voluntary dry matter intake and digestibility of ensiling OPF were the same as the value of dry OPF. In case of NaOH treatment was introduced the ensiling process, the treated OPF silage had higher digestible dry matter intake than the other processed OPF.

Grinding (forming to pellet) increased the voluntary intake of OPF. However, the low digestibility spoiled the high intake. Then, digestible dry matter intake of OPF pellet was almost the same as those of dry OPF and OPF silage.

Prediction of the feeding value of grass silage from analysis of herbage at the point of ensiling

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Introduction

Grass silage forms the basal diet of the vast majority of dairy and beef cattle in Ireland and the United Kingdom during the winter feeding period. The low intake characteristics of grass silage-based diets have long been recognised as a major limitation to milk and beef production during this period. Recent developments in feed characterisation of grass silage (Park *et al.*, 1997) have facilitated considerable improvements in prediction of silage feeding value. However, this information would be of considerably greater value in practice if predictions of silage feeding value could be determined from analysis of herbage prior to ensiling. Consequently, the aim of the present study was to examine the effects of management of the sward prior to harvesting and herbage composition at ensiling on the feeding value of the resultant silages.

Material and Methods

Twenty-five plots, each 10 x 1.5 m, were laid out in three replicate randomised blocks on a predominantly perennial ryegrass sward. The plots received either 72, 96, 120, 144 or 168 kg N/ha on 24 March. Herbage from the plots was harvested on either 10, 17, 24 or 31 May or 7 June. The herbage from the 75 treatments was ensiled in 225 silos (6 kg capacity) either untreated (U) or treated with either formic acid (F) (850 g/kg) at the rate of 3 ml/kg, or an inoculant (I) (Ecosyl, Zeneca Bio-Products) at the rate of 3 ml/kg. Following a 176 day fermentation period the silos were opened and sampled for the determination of potential dry matter (DM) intake and digestible organic matter in the DM (D-value) using NIRS as described by Park *et al.* (1997). Predicted metabolisable energy (ME) intake was estimated for a 500 kg steer from a combination of potential DM intake and D-value, with ME being determined as silage D-value x 0.16. For each additive, the correlation between individual silage variables and herbage composition was identified by stepwise multiple linear regression analysis. Regression analysis was also undertaken to examine the relationship between additive treatment, harvest date and level of fertiliser N application, with harvest date and N fertiliser fitted as polynomials up to degree 3.

Results

There were large variations in the chemical composition of the herbage at ensiling and in the resultant silages. For example herbage pH, buffering capacity (BC) and concentrations of DM, crude protein (CP), neutral detergent fibre (NDF), water soluble carbohydrate (WSC) and nitrate N varied from 5.77 to 6.31, 211 to 302 mEq/kg DM, 150 to 207 g/kg, 77 to 170 g/kg DM, 516 to 625 g/kg DM, 185 to 295 g/kg DM and 114 to 523 mg/kg DM respectively. Silage pH, D-value, potential DM intake and concentration of ammonia N varied from 3.70 to 4.54, 624 to 780 g/kg DM, 65.2 to 94.1 g/kg $W^{0.75}$, 34 to 125 g/kg total N respectively.

There was no significant relationship between ammonia N content of the untreated silage and herbage pH and the concentrations of acid detergent fibre (ADF), NDF, WSC, nitrate, acid detergent lignin (ADL), cellulose or hemicellulose. Ammonia N concentration of the untreated silages was negatively correlated with herbage DM content and yield, and positively correlated with herbage BC and the concentration of CP, acid insoluble nitrogen (AIN), ash and true protein (TP). Ammonia N concentration of the formic acid-treated silages was positively correlated with herbage ash, nitrate and ADL concentrations and negatively correlated with herbage WSC concentration. For the inoculant-treated silage,

ammonia N concentration was negatively correlated with herbage WSC concentration and positively correlated with ash, NDF, ADL and hemicellulose concentrations.

There was no relationship between silage D-value or predicted ME intake and herbage WSC, nitrate or cellulose concentrations. Silage D-value and predicted ME intake were positively related to TP, AIN, CP (R^2 values greater than 0.70), pH, NDF, hemicellulose (R^2 values of 0.50 to 0.70), BC, ash and ADF (R^2 values of 0.28 to 0.50) and negatively correlated with herbage yield (R^2 values of 0.88 to 0.92), and concentrations of ADL (R^2 values of 0.39 to 0.51) and DM concentrations (R^2 values of 0.30 to 0.41).

Further analysis of the relationship between the chemical composition of the herbage at ensiling and additive treatment on the potential ME intake of the resultant silages, when offered to 500 kg steers, identified positive relationships which are best described by the following relationships:

MEI for U = 78.6 – 0.0042 (yield) – 0.076 (NDF) + 0.349 (DM) + 2.596 (PN) – 0.0221 (nitrate) (R^2 = 0.94) Equation 1

MEI for F = -4.1 + 2.557 (PN) – 0.0036 (yield) + 0.264 (DM) + 7.28 (pH) (R^2 = 0.93) Equation 2

MEI for I = 75.2 – 0.0053 (yield) – 0.087 (Hemi) + 2.401 (PN) + 0.2715 (DM) – 0.0143 nitrate + 0.493 (EE) (R^2 = 0.94) Equation 3

where: MEI = metabolisable energy intake (MJ/day); yield = DM yield at harvest (kg DM/ha); NDF = neutral detergent fibre (g/kg DM); DM = dry matter (g/kg); PN = protein N (g/kg DM); nitrate = mg/kg DM; Hemi = hemicellulose (g/kg DM); EE = ether extract (g/kg DM), pH = pH of grass at ensiling.

There were significant ($P<0.05$) additive by harvest date by level of N fertiliser interactions for predicted silage feeding value which could be described by the following equations:

MEI for U = 121.2 – 0.97 (D) – 0.040 (N) – 0.0033 (DxN) Equation 4

MEI for F = 110.8 – 0.93 (D) + 0.047 (N) – 0.0033 (DxN) Equation 5

MEI for I = 126.8 – 1.31 (D) – 0.022 (N) – 0.0033 (DxN) (R^2 = 0.96***) Equation 6

where: D = day delay in harvest after 10 May and N = nitrogen fertiliser (kg/ha).

These relationships indicate that the response to inoculant treatment improved as the level of fertiliser N increased within each harvest date, however the response to inoculant treatment decreased with delay in harvesting. Regardless of cutting date, formic acid treatment tended to increase predicted ME intake when 144 or 168 kg N was applied/ha, but had no effect at lower levels of fertiliser N application.

Conclusions

It is concluded that silage feeding value, determined as potential ME intake of silage, was strongly correlated with herbage yield, and protein and fibre fractions in herbage. Herbage yield and the concentrations of NDF, TP, DM and nitrate provided the best fit relationship with potential ME intake of the untreated silages (R^2 of relationship = 0.94). Delaying harvesting had the most detrimental effect on silage feed value, which was partially overcome by reducing level of N fertiliser application and choice of additive use.

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Estimation of dry matter intake in *ad libitum* silage feeding

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Determination of dry matter intake plays an important role in *ad libitum* silage feeding as it helps to balance dairy cows' rations and provide them with all essential nutrients. Lot of factors affect dry matter intake (Van Vuuren et al., 1995) and many of them have been taken into consideration in different regression equations (Holter et al., 1997; Rayburn and Fox, 1993). The purpose of our study was to explain how the amount of compound feed added to the ration and the quality of silage affect silage dry matter intake when silage is fed *ad libitum.*

Material and methods. Two experiments in which silages of different quality were used, were carried out with four lactating cows in 4x4 Latin squares. In the first experiment the cows were fed graminaceous-rich (75% graminaceous + 25% clover) pre-wilted silage which DM content was 49.4% and its crude protein, crude fibre and metabolizable energy content in DM was 11.48%; 33.06% and 8.64MJ/kg, respectively. Silage used in the second experiment was prepared from pre-wilted clover which DM content was 27.26% and the content of crude protein, crude fibre and metabolizable energy in DM was 17.38%; 22.25% and 9.25 MJ/kg, respectively. No preservatives were used during ensiling. In addition to silage, the cows were fed compound feed prepared from barley and soybean meal that covered 25, 40, 55 or 70% of the cows' energy need. A proportion of barley and soybean meal in the compound feed was calculated so that the protein content of the ration was 15%.

Results and discussion. Effect of compound feed on silage intake, milk yield and composition are concisely given in table 1.

Table 1. Effect of compound feed on silage intake, milk composition and yield

Items	Compound feed proportion to energy need %			
	25 a	40 b	55 c	70 d
Dry matter intake kg	17.6	18.9	19.4	19.9
silage intake kg	15.1^{cd}	13.9^{cd}	12.1^{ab}	11.7^{ab}
concentrate intake kg	2.5	5.0	7.3	8.2
Energy intake MJ	161.1^{bcd}	187.7^{a}	200.4^{a}	209.3^{a}
Dry matter intake per 100 kg life weight %	3.27	3.55	3.62	3.72
In ration dry matter:				
crude fibre %	24.8	23.4	21.5	20.4
metabolizable energy MJ/kg	9.2^{bcd}	9.9^{ad}	10.4^{a}	10.5^{ab}
FCM production kg	19.7	21.3	22.2	21.7
Milk composition:				
fat %	4.43	4.39	4.34	4.36
protein %	3.22	3.29	3.35	3.37
lactose %	4.64	4.72	4.73	4.71
urea mg/l	305^{cd}	289	275^{a}	274^{a}

a, b, c, d - $P < 0.05$

The daily intake of silage DM ranged from 15.1 to 11.7 kg, according to the proportion of compound feed in the ration. Each supplementary kg of compound feed DM in the ration reduced silage DM intake by 0.597 kg per day ($r = -0.586$, $p < 0.001$). As the energy content of a compound feed DM is higher than that of a silage, a higher proportion of compound feed in the ration increased energy intake ($p < 0.05$). Analysis of data revealed that statistically significant, although not very high correlation existed between silage DM intake and total ration DM intake ($r = 0.412$, $p < 0.05$), metabolizable energy content of silage and its DM intake ($r = 0.434$, $p < 0.05$) and metabolizable energy content of silage and total ration DM intake ($r = 0.394$, $p < 0.05$). Negative correlation between silage crude fibre content and silage DM intake was weak and statistically insignificant ($r = -0.131$, $p > 0.05$). DM intake and the number of days in milk were also insignificantly correlated ($r = -0.276$, $p > 0.05$).

Correlation between silage crude fibre content and silage intake was quite low despite the fact that cell wall composition and the rate of their ruminal hydrolysis are the main factors that determine ruminal fill and consequently DM intake as well. The results of an investigation carried out by Beauchemin (1996) also showed that cell wall, particularly NDF content of feeds does not correlate adequately with DM intake. This can presumably be resulted from the composition of the ration and its content of easily hydrolysing carbohydrates. Recent researches by De Visser et al. (1998) and Van Vuuren et al. (1999) suggest that the amount of supplementary starch in the ration affects cell wall digestibility to a large extent. The results of our experiment also indicated that silage intake was affected more by starch-rich barley meal than by silage composition and nutritive value.

Conclusions. When silage is fed *ad libitum* the DM intake is more affected by silage ME content than by its CF content. Each supplementary kg of barley based compound feed DM in the ration reduced silage DM intake of dairy cows by 0.597 kg per day.

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The effect of stage of ensiling on nutritional value and yield of triticale, oats, oat/peas and oat/vetch in the Western Cape.

INTRODUCTION

The optimal stage of ensiling for wheat is the soft dough stage. In the Western Cape triticale and oats are often harvested for silage in the boot stage to obtain the highest nutritional quality. The question is, how much of the potential yield of nutrients is lost by this practice. The objective of the study was to determine the effect of harvesting stage on nutritional value and yield of triticale, oats, oats/vetch and oats/peas.

MATERIALS AND METHODS

Four different silage crops/ crop combinations were be planted on the 30 th of April 1998 in a randomized block design with four replicates of 0.5 ha per crop. The pH (KCl) of the soil was 5.5, phosphorous 42 ppm and potassium 230 ppm. Before planting 175 kg of 3:2:0 was applied per hectare. Triticale, oats, oats/vetch and oats\peas were planted with a 12 metre Amason planter. Oats (cv Sederberg) was planted at 120 kg ha^{-1}, triticale (cv Rex) was planted at 150 kg ha^{-1}, oats (cv Sederberg 60 kg ha^{-1}) mixed with peas (cv Glenroy 50 kg ha^{-1}) and oats (cv Sederberg 60 kg ha^{-1}) mixed with vetch (cv Max 30 kg ha^{-1}). Seven weeks after planting 125 kg Kysan (Limestone ammonium nitrate plus sulphur, 27% N, 3.7% Ca and 3.5% S) was applied per hectare. When the crops reached the boot stage on the 31st of August, the yield and quality were determined every week until the soft dough stage was reached. Yield was determined by cutting 10 quadrants of 0.25 m on each of the experimental plots. Samples were weighed and dried at 60°C for 72 hours. The dried samples were milled though a 1mm sieve and chemically analysed. The *in vitro* organic matter digestibility (IVOMD) was determined according to Tilley and Terry (1963), crude protein (CP) by the Kjeldahl method and neutral detergent fibre (NDF) according to Van Soest *et al.*, (1991).

RESULTS AND DISCUSSION

The crude protein content of triticale, oats, oat/pea and oat/vetch was 13.8%, 12.1%, 14.3% and 13.7% at the boot and 6.5%, 6.9%, 6.9% and 7.3% at the soft dough stage respectively. The TDN content of triticale, oats, oat/pea and oat/vetch was 73.5%, 74.1%, 72.7% and 73.6% at the boot and 68.7%, 67.7%, 66.9% and 66.2% at the soft dough stage respectively. The highest protein and TDN content for all crops were obtained at the boot stage. The dry matter yield of triticale, oats, oat/pea and oat/vetch was 5.1, 5.1, 4.7, and 4.8 tonne DM ha^{-1} at the boot and 11.1, 9.6, 9.3 and 9.9 tonne DM ha^{-1} at the soft dough stage respectively. The highest yield of DM and TDN for all crops was obtained at the soft dough stage. The dry matter content of of triticale, oats, oat/pea and oat/vetch was 18.1%, 18.2%, 16.3% and 16.2% at the boot and 41.3%, 33.1%, 34.2% and 32.1% at the soft dough stage respectively. Direct harvesting with a flail silage harvester is possible at the soft dough stage. Wilting is required if the crops are harvested at the boot stage.

The yield and nutritional value of triticale is given in Table 1. The highest nutritional value was

obtained at the boot stage while the highest yield of tonne of DM and total digestible nutrient (TDN) content was obtained at the soft dough stage.

TABLE 1. The effect of cutting date on the composition and yield of triticale (cv. Rex).

Stage	Boot		Flowering				Soft dough			
Date	31Jul	7Aug	14Aug	21Aug	28Aug	4Sept	11Sept	18Sept	28Sept	SEM
Composition (% of DM)										
DM %	18.1^{a}	19.3^{a}	22.8^{b}	25.3^{c}	29.8^{d}	33.6^{e}	38.8^{f}	41.3^{g}	48.5^{h}	0.58
Ash%	7.6^{a}	7.0^{b}	6.1^{c}	5.5^{d}	5.1^{e}	4.6^{f}	4.4^{fg}	4.2^{fg}	4.0^{g}	0.13
CP%	13.8^{a}	12.4^{b}	11.0^{c}	9.8^{d}	8.6^{e}	7.4^{f}	7.1^{fg}	6.5^{g}	5.3^{h}	0.28
TDN% 73.5^{a}	68.8^{b}	68.6^{b}	67.4^{bc}	64.7^{d}	66.6^{cd}	65.0^{de}	68.7^{b}	64.2^{d}		0.64
NDF %	48.6^{a}	63.1^{b}	54.9^{ab}	54.9^{ab}	52.1^{a}	53.0^{a}	52.5^{a}	49.0^{a}	55.6^{ab}	2.86
Yield (tonne ha^{-1})										
Wet material	28.4^{abc}	30.9^{a}	31.3^{a}	29.7^{ab}	31.0^{a}	25.8^{cd}	25.8^{cd}	26.9^{ab}	22.6^{d}	1.2
DM	5.14^{a}	5.96^{a}	7.11^{b}	7.48^{b}	9.19^{cd}	8.67^{c}	10.0^{d}	11.1^{e}	10.9^{e}	0.31
CP	0.71^{ab}	0.74^{b}	0.78^{b}	0.73^{ab}	0.79^{b}	0.65^{bc}	0.71^{ab}	0.73^{ab}	0.583^{c}	0.03
TDN	3.71^{a}	4.10^{a}	4.88^{b}	5.05^{b}	5.94^{cd}	5.77^{c}	6.50^{de}	7.64^{f}	7.02^{ef}	0.23

abcdef Values in the same row with no common superscript differ significantly ($P< 0.05$)
SEM = Standard error of means, CP = Crude protein, TDN = Total digestible nutrients, NDF = Neutral detergent fibre.

CONCLUSIONS

The highest protein and energy value of triticale, oats, oat/pea and oat/vetch was obtained by harvesting at the boot stage. However, harvesting at the boot stage will result in up to 50% loss of potential yield of dry matter and total digestible nutrients per hectare . The soft dough stage is recommended as optimal stage for ensiling as this will result in the highest yield of total digestible nutrients per hectare

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Feeding value and ensilability of two sorghum varieties under tropical conditions

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Keywords: sorghum, parameters of feeding value, rapid fermentation test, LAB inoculants

INTRODUCTION

Limiting factors of performance-oriented cattle husbandry in the tropics and subtropics are the urgent seasonal deficiency of fodder, the lack of fodder with high crude protein or energy content and the scarce availability of concentrates. A precondition to guarantee the continual fodder supply for the livestock during the arid season, too, is the conservation of higher quality forage while silage making is of special importance in the humid tropics.
The aim of this study was to investigate feeding values and ensilabilities of two newly bred sorghum species during the period relevant for green fodder use (between the 60th to 100th day of vegetation).

MATERIAL AND METHODS

Experiments were carried out in 1996 at the Universidad Central de las Villas, Santa Clara, Cuba. The examined plant materials were sorghum varieties C.I.A.P.-2 (high tannin) and U.D.G.-110 (low tannin). A total of 5 plant samples were taken in 10 days intervals beginning at the 60th day of vegetation. A cellulase method according to FRIEDEL AND POPPE (1990) was used to determine the nutritive value and to estimate the digestibility of the organic matter. In order to estimate ensilability, the „sugar-buffer capacity-quotient" (S/BC) and a biological test on ensilability („rapid fermentation test") as described by PIEPER et al. (1989) were employed to evaluate the variants „without additives", „addition of sugar", „addition of LAB inoculants" and „addition of sugar and LAB in combination". Silage prepared in plastic bags in the variants „direct-cut (fresh) without additives", „fresh with addition of LAB", „wilted without additives" and „wilted with addition of LAB" was used to verify the results of the methods explained first.

RESULTS

- Due to the increasing percentage of the panicle on the whole plant (approx. 40 % at dough stage), the digestibility of the organic matter (OMD) and the energetic feeding value rose with advancing maturity. 69.2 % OMD (5.7 MJ NEL/kg DM) for C.I.A.P.-2 and 68.5 % OMD (5.6 MJ NEL/kg DM) for U.D.G.-110 were detected at vegetation day 100. At this stage the OMD of the panicles were 80.6 % (C.I.A.P.-2) and 83.9 % (U.D.G.-110).
- The plant material ensiled in plastic bags showed less digestibility than the fresh plants, especially during the late cutting dates (day 90 and 100) and more distinctly for U.D.G.-110.
- The crude protein content of the whole plants decreased throughout the vegetation period and reached 4-5 % of the DM at dough stage.
- The S/BC-quotient showed values above 3.0 for both sorghum varieties from vegetation day 80 onward.
- The biological test on ensilability led to the following results:
  - Natural ensilability of C.I.A.P.-2 was estimated as „good" only at dough stage and of U.D.G.-110 from early milky seed stage to early dough stage. Later growth stages were not evaluated.

- Addition of sugar prevented re-increasing of pH-values.
- Combined treatment of sugar and LAB gave stable silages from the flush stage.

- The data obtained from the material ensiled in plastic bags indicated that, for all variants with C.I.A.P.-2, the pH did not fall below the DM-dependent critical value, especially at the early cutting dates. Thus, a stable silage could not be prepared before the emergence of panicles. Silage made of U.D.G.-110 were of high quality at all sampling times.
  Addition of LAB always resulted in declining pH-values.

CONCLUSIONS

- Digestibility of sorghum at dough stage corresponds to that of maize for silage in Europe. Thus for dairy cattle, sorghum silage could be high energy fodder with relatively high starch contents (cutting height should be used to influence digestibility and the energetic feeding value). One problem is the low crude protein content at this growth stage.
- The energetic potential of direct-cut sorghum can be used effectively for cattle feeding only during a short period of time each year. Preservation of the plant material with lactic acid fermentation is suggested. Losses caused from silage effluents are not expected due to the high DM content at this stage.
- Different methods for the evaluation of ensilability predicted stable silage at cutting dates around dough stage. This cutting date is most favourable with respect to the energetic nutritive value *and* ensilability. Further, the high tannin content of C.I.A.P.-2 had no negative effects on the fermentation process (biological test on ensilability) at dough stage.
- The use of certificated LAB inoculants is recommended for the preparation of sorghum silage.
- Sorghum can be ensiled at dough stage without additional sugar.

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Effect of dry matter content of whole crop wheat on feed intake, digestibility and milk production.

Objective. The aim was to examine the effect of dry matter content (DM) of whole crop wheat (WCW) on feed intake and digestibility and milk production of dairy cows.

Materials and methods. At week 4 of lactation 40 Holstein/Friesian cows were allocated to treatment. All cows received forage *ad libitum* (2:1 DM ratio of WCW and grass silage) and 10 kg fwt/d of concentrate (260 g CP/kg DM and 12.8 MJ ME/kg DM). The four forage were:

1. **Low DM** Fermented low DM WCW + grass silage
2. **High DM** Fermented high DM WCW + grass silage
3. **High DM/Additive*** Fermented high DM WCW (with *L. buchneri*) + grass silage
4. **Urea-treated** Urea treated (4%) WCW + grass silage

* *Lactobacillus buchneri* (5.4×10^6 cfu)

Low, High DM and urea-treated (4% of DM) WCW were harvested on 1, 22 and 30 July 1996. Treatment effects on diet digestibility and nitrogen excretion were measured by total collection of faeces and urine from four lactating cows in a 4 x 4 Latin square experiment. The diets and treatments were the same as those used in the applied feeding trial.

Results. Grass silage was well preserved but was only of moderate energy value. As the DM content of WCW increased, NDF content decreased while starch and predicted ME value increased. The urea-treated crop fermentation was restricted and its CP content was markedly higher than other forages. Urea-treated forage had the highest predicted ME value.

Forage composition.

Composition (g/kg CDM*)	Grass	Low DM	High DM	High DM/Add.	Urea
CDM (g/kg)	307	301	511	494	584
CP	141	116	104	110	199
NDF	601	610	522	524	414
Starch	0	32	170	170	306
Predicted ME (MJ/kg DM)	10.7	9.6	10.4	10.0	12.4
Lactic acid	54	54	19	17	4
Acetic acid	14	16	9	4	9
pH	3.95	3.98	5.03	4.63	7.30
Ammonia N (g/kg of total-N)	45	65	74	62	247

* CDM = silage dry matter corrected for volatile components

- **Feed intake and milk production.** The lowest and highest forage intakes were recorded on Low DM and Urea-treated, with intermediate values for the two High DM treatments. Intake for the Urea-treated crop was higher ($P<0.05$) than for fermented crops. Although forage intakes for the two High DM treatments were substantially higher when compared with Low DM, differences were not significant. There was no effect of additive. The lowest and highest milk yields were recorded for cows on Low DM and Urea-treated respectively, with intermediate values for High DM and High DM/Add. The difference between Low DM and Urea approached significance. There were no significant treatment effects for either content or yield of milk fat. The lowest and highest values for milk fat yield were recorded for Low DM and Urea-treated, respectively, with intermediate values noted for the two High DM treatments. The highest milk protein content was recorded for Urea-treated and the difference approached significance. There was no treatment effect of additive and no differences noted

when comparing the untreated Low DM and untreated High DM. The yield of milk protein produced by Urea-treatment was higher ($P<0.05$) compared Low DM, with intermediate values recorded for the two High DM treatments.

Feed intake and milk production

	Low DM	High DM	High DM/Add.	Urea	SED
Forage DM I (kg/d)	10.9	11.7	11.7	13.7	0.56
Total DM1 (kg/d)	19.7	20.5	20.5	22.5	0.56
Milk yield (kg/d)	27.9	28.5	28.8	29.2	0.75
Fat (g/kg)	47.7	46.3	46.6	47.4	1.24
Protein (g/kg)	33.0	32.9	32.2	34.2	0.63
Fat yield (g/d)	1307	1317	1350	1376	40.6
Protein yield (g/d)	910	935	926	992	19.9

- **Diet digestibility and nitrogen excretion** The digestibility of all fractions except starch was lower for High DM than for Low DM but with Urea-treated only the digestibility of starch and energy were lower. The additive had no effect. With increasing crop maturity, the ME concentration (MJ ME/kg CDM) of the diets fell. With the Urea-treated, nitrogen excretion in faeces and urine was increased by 15% and 48% compared with High DM.

Diet digestibility (%) and ME concentration

	Low DM	High DM	High DM/Add.	Urea	SED
CDM	68.0	65.7	66.6	67.6	0.48
Organic matter	70.2	67.9	68.8	69.5	0.57
Energy	68.8	66.2	67.3	67.1	0.59
NDF	56.8	51.6	53.3	59.7	1.63
Starch	96.4	96.6	96.7	92.6	0.54
Crude protein	68.7	63.6	64.9	67.3	0.85
ME (MJ/kg CDM)	11.3	10.9	11.0	10.7	-

Discussion. The composition of the three forms of WCW produced for this study showed a number of now well established differences. According to laboratory-based estimates, the predicted ME value of the WCW increased from 9.6 to 12.4 MJ/kg CDM as DM content increased. However, the results of the digestibility study with lactating cows show the opposite trend with a fall from 10.4 to 9.0 MJ/kg CDM. A similar trend has been found when the same crops were evaluated in sheep and highlights the problems of predicting accurately the ME content of WCW.

Conclusions. Increasing DM content from 300 to 500 g/kg in fermented WCW resulted in a small increase in both forage DM intake and milk yield. With fermented WCW DM content did not affect either milk composition or yield of milk constituents. Although there were only modest differences in animal performance between early and late harvested fermented WCW the higher DM yield/ha that occurs at the later harvest make it the more attractive crop for farmers to produce. When compared with high DM fermented WCW, the use of urea-treated WCW resulted in a large and significant increase in forage intake but only a small non-significant increase in milk yield because of the reduced ME value of the urea-treated crop. There was an increase in both content and yield of milk protein when compared with high DM fermented WCW. Whether these increases are sufficiently large to persuade farmers to use urea to conserve WCW waits to be seen. Present laboratory methods for predicting the ME value of WCW appear to be unreliable and may seriously overestimate the value of more mature crops.

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Effect of increasing levels of a formic acid based additive on *ad libitum* intake of grass silage and the production of milk and meat.

When wet crops are used for silage production, formic acid based additives are normally recommended to ensure good fermentation. When crop dry matter (DM) exceeds approx. 300 g/kg, acid based additives are not regarded to be necessary, because extensive butyric acid- and acetic acid fermentation normally do not occur. However, acid based additives restrict the extent of fermentation even at DM concentrations above 300 g/kg. The objective of this study was to evaluate the effect of increasing doses of a formic acid based additive under conditions where untreated silage was expected to be of good fermentation quality.

MATERIALS AND METHODS

After drought, a second cut sward consisting mainly of timothy, was direct cut at 300 g/kg DM (per kg crop DM: 106 g CP, 60 g sugar) using a precision chop harvester, on 13-21 of August 1997. The crop was either untreated or treated with 4 or 8 L/t of GrasAAT (Hydro Nutrition, Oslo, Norway; 645 g/kg of formic acid, 60 g/kg of NH_3, 15 g/kg of corrosion inhibitors (for skin and metal) plus caramel colour). The crop was ensiled in tower silos of 92 m^3. The measured application rates were 0, 4.14 and 8.19 L/t for the three respective treatments.

Each silage was offered *ad libitum* to 10 Norwegian Red dairy cows (NRF) in mid lactation during 9 weeks following a 4-week preliminary period. Concentrates were offered at the same level to the three groups of cows. The cows received 100 g/d of a mineral and vitamin mix. Fifteen steers (NRF), on average 17 months old, were used in a cross-over trial with 3 periods, each of 5 weeks. The three silages were fed *ad libitum* without concentrates, but with 50 g/d of a mineral and vitamin mix.

RESULTS

The control silage contained butyric acid at 10.0 g/kg DM and NH_3-N at100 g/kg of total N, but was otherwise well fermented (Table 1). The silage added 4 L/t was subject to the most extensive lactic- and acetic acid fermentation, but butyric acid fermentation was totally

Table 1. Chemical composition of the silages

	GrasAAT treatment, L/tonne		
	0	4	8
DM, g/kg	294	297	304
pH	4.37	4.14	4.37
NH_3-N, g/kg TN	100	98	114
NH_3-N, g/kg TN (corrected)	100	66	51
g/kg DM:			
CP	116	117	120
True protein	48.9	58.7	63.3
Sugar	60.6	54.9	74.8
NDF	546	548	550
Lactic acid	59.0	61.9	32.4
Formic acid	0.2	5.2	9.8
Acetic acid	9.8	15.2	12.7
Propionic acid	0	0	0.1
Butyric acid	10.0	0	2.2
Ethanol	8.0	6.4	5.7

Table 2. Feed intake and yield of the dairy cows

	GrasAAT treatment, L/tonne				
	0	4	8	SEM	p
Silage, kg DM	11.6	11.3	12.2	0.47	NS
Silage, kg DM/100 kg BW	2.01[a]	2.05[ab]	2.24[b]	0.07	0.06
Concentrates, kg DM	5.65	5.67	5.67		
Milk, kg	21.6[a]	21.2[a]	23.1[b]	0.43	0.02
ECM[1], kg	21.8[a]	21.3[a]	23.4[b]	0.49	0.02
Milk fat, g/kg	41.9	41.2	41.9	0.5	NS
Milk protein, g/kg	31.8[a]	32.3[ab]	32.6[b]	0.2	0.07
Lactose, g/kg	46.0	45.6	46.0	0.2	NS
Milk taste[2]	3.74[a]	4.01[b]	3.87[ab]	0.08	0.09
FFA, m.eq./L	0.83[a]	0.98[b]	0.77[a]	0.04	0.002
Milk urea, mM	3.16[a]	3.46[b]	3.57[b]	0.07	0.002
Milk acetone, mM	0.09[a]	0.06[b]	0.08[a]	0.01	0.004
BW at the beginning of expt.	581	556	545	17	NS
BW change, g/d	106	80	251	79	NS

[1] Energy-corrected milk
[2] Five-point scale for milk aroma and taste where 1 = poor quality milk, and 5 = high quality milk

Table 3. Silage intake and body weight gain of the steers

	GrasAAT treatment, L/tonne				
	0	4	8	SEM	p
Silage, kg DM	8,5	8,2	8,7	0,22	NS
Silage, kg DM/100 kg BW	1,80	1,78	1,89	0,05	NS
Initial weight, kg	459,1	453,1	450,6	2,5	0,06
Final weight, kg	480,1	475,1	475,9	2,6	NS
BW gain, g/d	749	785	901	70	NS

inhibited, and the silage obtained a lower pH than the other two silages. The highest application rate (8 L/t) restricted silage fermentation, but did not inhibit production of a small amount of butyric acid.

Silage intake and body weight gain were highest on silage treated with 8 L/t in experiments both with dairy cows (Table 2) and steers (Table 3). Cows receiving silage treated with 8 L/t produced highest milk yield in kg, kg ECM, g fat, g protein, and g lactose. Differences between control silage and silage treated with 4 L/t were small and insignificant, except for milk composition. The milk protein concentration increased with increasing application rate.

CONCLUSIONS

Treatment with 4 L/t of GrasAAT produced a well fermented silage with low pH and no butyric acid, but did not restrict lactic and acetic acid fermentation. A restricted silage fermentation, which was obtained by the addition of 8 L/t, was of significantly greater importance for silage intake, and meat and milk production, than the absence of the undesirable butyric acid as obtained by the silage added 4 L/t. Extensive butyric acid fermentation in wet silage is by no doubt detrimental to feed intake and animal production. This experiment suggests that the disadvantages of a moderate concentration of butyric acid may be of less importance for silage intake and the quantity of milk and meat produced in silage of high compared with low DM concentration. The milk quality (i.e. the concentration of spores of Clostridium spp., the organoleptic quality, milk acetone concentration), however, may be negatively affected by a small or moderate butyric acid concentration in the silage.

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Influence of different grass forage making technologies on feed composition, quality and milk production in dairy cows.

Key words: grass silage, chemical additives, quality, milk production.

Introduction
The development of feeding practices which utilize the silage of high quality is important for the improvement of both the milk production and the profitability of the farms in Latvia. *The aim of the studies* was to determine the effect of wilting of herbage prior to ensiling and the use of additives AIV-2; AIV-3 and AIV-2000 on the fermentation characteristics, nutritive value of grass silage and subsequent milk production. The investigations were carried out by collaboration between the Research Centre "Sigra" and Kemira Chemicals OY.

Material and methods
The silages were made from the grass /clover sward (dominated by timothy *Phleum pratense* and red clover *Trifolium pratense*) in the first decade of June.
Experiment No.1
The following treatments were used:
-unwilted with an additive AIV-3 (6 l ton^{-1})
-wilted with no additive
-wilted with an additive AIV-2 (5 l ton^{-1}).
Forage was harvested with chopper TUPLA JUNKAR - 170 and ensiled into plastic covered silos. For the feeding trial 3 analogous groups of Latvian brown cows (10 cows in the group) were formed. The duration of the trial was 90 days. Experimental diets involved a comparison between 3 types of silage. Silage was offered ad libitum. The cows received 2 kg of hay and 300 g of concentrate per litre of milk.
Experiment No.2
Two types of big bale silage were made:
-with no additive
-with an additive AIV-2000 (5 l ton^{-1}).
The forage was harvested with baler SIPMA Z - 279.
The feeding trial was carried out with 2 analogous groups each containing 10 cows. The cows of the control group received untreated silage, the cows of the experimental group - silage with an additive AIV-2000. Silage was fed ad libitum. The cows of the both groups received 2.2 kg of hay and 4.3 kg of concentrates.
For silages the fermentation quality and chemical composition were determined. The feed intake and milk yield were recorded daily. Milk samples were analysed once a week.

Results
Experiment No.1
Unwilted silage contained 230 g DM kg^{-1}, but in wilted silages the content of DM was 300...320 g kg^{-1}. The quality of silages was influenced by treatments used. The

content of CP was higher in unwilted silage with AIV-3 (156 g kg^{-1} DM) than in the both wilted silages (133...134 g kg^{-1} DM). The concentration of organic acids in unwilted silage was 119 g kg^{-1} DM. In the both wilted silages the extent of fermentation was lower - 87...97 g of organic acids kg^{-1}DM. The use of additive AIV-2 increased the proportion of lactic acid in wilted silage (84 % from the sum of acids) in comparison with wilted silage with no additive (58 %). Silage intake averaged 9.5 kg DM per day for unwilted silage with AIV-3 compared with 9.4 kg DM per day for wilted silage with no additive and 10.2 kg DM per day for wilted silage with AIV-2. The milk yield of cows fed different silages was similar (15.5...15.7 kg 4 % FCM per day). The diet containing wilted silage with no additive was the most economic.

Experiment No.2

DM content of big bale silage was 240...256 g kg^{-1}. Silage with no additive contained 162 g CP kg^{-1} DM, but in silage with AIV-2000 the content of CP was 174 g kg^{-1} DM. The use of an additive AIV-2000 decreased the concentration of organic acids (126 g kg^{-1} DM vs 98 g kg^{-1} DM) and eliminated the development of clostridial fermentation. Silage with an additive contained 4 times more sugars than untreated silage. The better quality of silage with AIV-2000 resulted in higher DM intake in comparison with untreated silage (10 kg DM per day vs 8.1 kg DM per day) and higher milk output (15.8 kg 4 % FCM per day vs 14.8 kg 4 % FCM per day).

Conclusions

Use of additives AIV-2 and AIV-3 and wilting resulted in the improved fermentation quality of conventional grass/clover silage. Wilting and use of an additive AIV-2 increased silage DM intake by 9 % with no effect on the milk yield. Use of an additive AIV-2000 increased the content of crude protein in big bale silage by 7 % and decreased the concentration of organic acids by 22 %. Cows offered big bale silage with AIV-2000 produced by 7 % more 4 % FCM per day in comparison with untreated big bale silage.

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Effect of nitrate contents in maize silage on ruminal methane emission and energy metabolism in sheep

Methane is a potent greenhouse gas which is expected to contribute about 15% of global warming. Livestock and their wastes contribute significant amount (Takahashi et al., 1997). Several forage crops, especially maize accumulate high nitrate. In general, ensiling tends to decrease nitrate content of the materials, but sometimes the significant amount still remains in good fermented silage. Heavy fertilization including manure is a primary factor of most nitrate-nitrite poisoning. Small amount (less than 0.1% nitrogen equivalent in dry matter basis) of nitrate can be reduced in an assimilatory manner to ammonia without any accumulation of harmful nitrite. However, reduction of nitrate alters the redox potential in the rumen fluid and causes a decrease in methanogenesis and molar proportions of propionic and butyric(Takahashi et al., 1993; Takahashi et al., 1998.). Respiratory trials were conducted to evaluate the quantitative effect of nitrate contained in maize silage on methane emission and energy metabolism in sheep with ventilated chamber (Takahashi et al., 1983). Experimental maize silage which contain different contents of nitrate were collected from six steel upright silos ensiled at almost same period in different dairy farmers. According to the nitrate content, low nitrate silage (L: 0.01% in dry matter basis) and moderate nitrate silage (M: 0.10% in dry mater basis) were prepared from the collected materials. Additionally, to evaluate the effect of high nitrate (H) 8.5g of sodium nitrate meal-1 head-1 was administered into the rumen via fistula under feeding condition of moderate nitrate silage. With the lapse of day, a marked increase in methaemoglobin formation and decline of volatile fatty acids (VFA) production occurred in high nitrate condition (H). In consequence, oxygen consumption, carbon dioxide production and metabolic rate were depressed gradually. Fig.1 shows the suppression of rumen methanogenesis in sheep received high nitrate (H) was amplified with passage of day. No remarkable changes in hemoglobin and VFA were observed in sheep fed moderate nitrate silage (M). Total rumen methanogenesis, however, decreased by 22 % in moderate nitrate silage (M), whereas the depression was 36 % in high nitrate condition.
Therefore, except for the difficulties in controlling nitrate content in feed, 0.1 % NO3-N (100% dry matter basis) in maize silage may become a natural manipulator to control methane emmission from ruminants without any nitrate hazard.

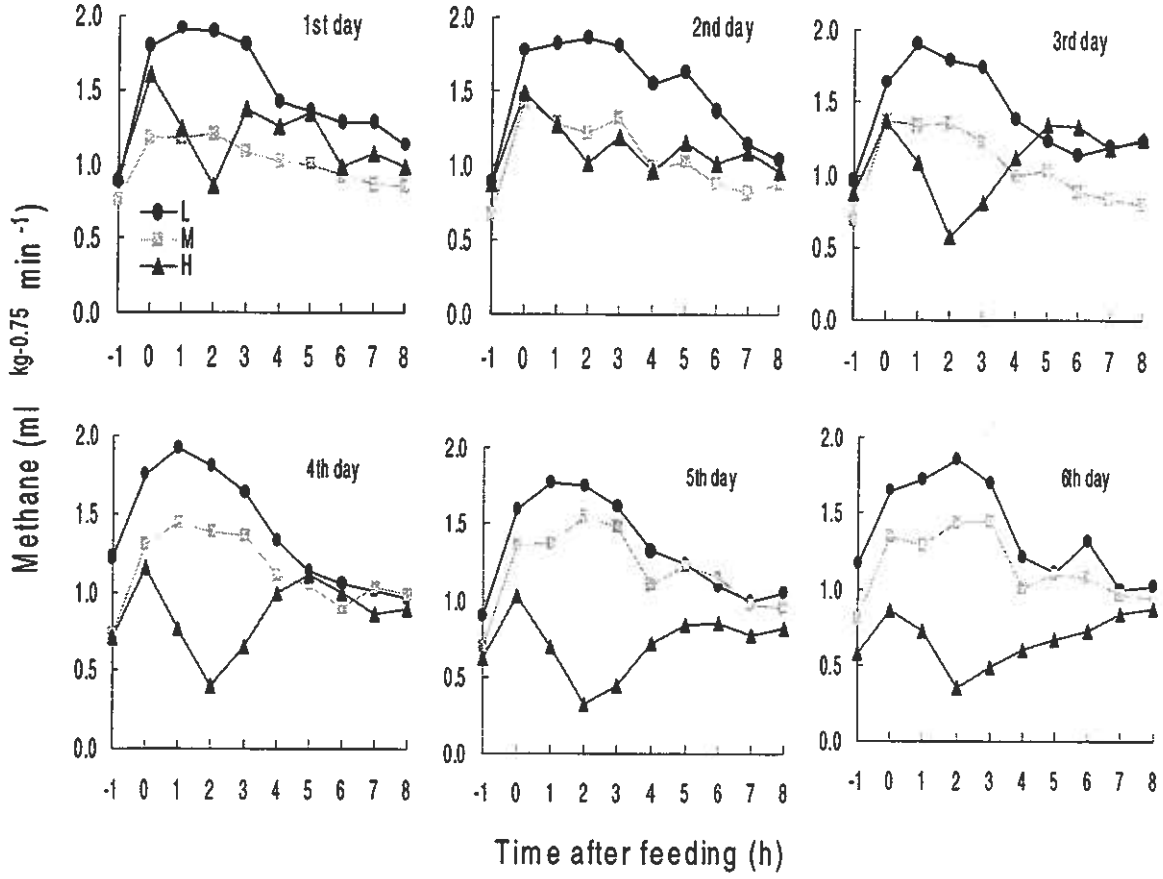

Fig.1. Change in methane emission form sheep fed maize silage contained nitrate.

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Keywords: maize silage, nitrate, methane emission, greenhouse gas, energy metabolism

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Influence of AIV-2000 treated red clover-grass silage on feed intake and milk yield

INTRODUCTION

The silage quality, losses during fermentation and storage stability are greatly influenced by using additives. Therefore, the animal performance can also be influenced. Several authors have reported a positive response of formic acid application on silage intake (Castle & Watson, 1974; Parker & Grawshaw, 1982). Martinsson (1992) reported 9% higher dry matter (DM) intake with formic acid- treated silage and the milk yield tended to increase because of the better nutrient supply.

The aim of this experiment was to study the effect of AIV-2000 treated silage on the performance of dairy cows. AIV-2000 is based on formic acid and contains in addition ammonium formate and esters of benzoic acid. It is expected that AIV-2000 is more effective and less corrosive compared to other AIV additives.

MATERIALS AND METHODS

Two silage clamps of 12 t each were ensiled in this study. Silages were made without using additive (untreated) and with using AIV-2000. The additive was applied at an application rate of 5 litre ton^{-1} fresh matter (FM). The silage material was red clover-grass mixture (about 50% red clover and 50% grass on FM basis) harvested on the 26th of June. Herbage was cut with a mower conditioner and wilted in the field for half day. Thereafter the herbage was chopped with a precision chop-harvester at 5 cm chop-length and ensiled in a clamp. The average chemical composition of silage material was as follows: dry matter (DM) 231 g/kg, crude protein (CP) 148 g/kg DM, crude fibre (CF) 277 g/kg DM, crude ash 53 g/kg DM, water soluble carbohydrates (WSC) 102 g/kg DM.

The feeding trial was conducted (at Laboratory of feeds) from Jan. 12 to March 15, 1998. Four similar Friesian dairy cows in their second lactation were chosen (average milk production 24 kg/day). A switch-back design was applied with four cows, two treatments and three experimental periods. Each experimental period lasted for 21 days, of which the first two weeks were the preliminary period and the last week the data recording. Silage was fed *ad libitum*; minerals and vitamins were given according to the animal requirements; common salt and water were freely available. The concentrate was supplied according to average milk production level – 7 kg per cow per day. The following calculations were made: dry matter intake (DMI), milk yield, concentration of fat, protein and urea in milk and changes in liveweight.

RESULTS AND DISCUSSION

The results are presented in Table 1. In general, adding AIV-2000 resulted in better silage quality. Intake of silage DM varied from 11.2 to 13.4 kg/day within feeding trial. The average intake of untreated silage was 12 kg DM/day. A significant higher intake 4.2% was obtained with additive treated silage ($p<0.001$). The higher intake can be explained by better silage quality because AIV-2000 treated silage did not contain butyric acid. Furthermore, AIV treated silage was less acidic and contained more residual sugars which obviously affected the DM intake.

Milk yield varied from 20.3 to 28.5 kg/day and ECM yield from 18.2 to 33.2 kg/day. A slightly higher milk yield (0.4 kg) was obtained with additive treated silage at the probability level $p<0.11$. The average ECM yield of untreated silage was 26.0 kg/day. When additive treated silage was fed, the average ECM yield increased by 0.6 kg. However, the results were not significant. The increase in milk yield was obviously the result of higher intake. A part of the forage energy was also stored in cow's body tissues. When untreated silage was fed, the live weight of the dairy

cows remained the same. The live weight increased with AIV silage, most likely due to higher intake.

The milk composition was similar in both treatments. There were no significant differences in milk fat- or protein concentrations. The concentration of urea indicated sufficient protein and energy supply in the diet of the cows.

Table 1. The chemical and microbiological composition of silage. Treatment effect of silage on performance of dairy cow.

Parameter	Untreated	Additive treated	$LSD_{0.05}$	Significance
Dry matter (DM), g/kg	241	252		
Crude protein (CP), g/kg DM	140	138		
Crude fibre (CF), g/kg DM	276	267		
Sugars (WSC), g/kg DM	25	81		
pH	3.8	3.9	0.3	NS
Ammonia N/total N, %	8.1	7.3	2.9	NS
Acetic acid, g/kg DM	20	17	6.6	NS
Butyric acid, g/kg DM	4.7	0	4.7	*
Ethanol, g/kg DM	13	10	5.2	NS
Clost. spores, /g FM	300	700		
Yeasts, $x10^3$ cfu/g FM	2800	116		
Moulds, x 10^3 cfu/g FM	2.6	0.5		
Milk yield, kg/cow day	25.1	25.5	0.51	$p<0.11$
ECM yield, kg/cow day	26.0	26.6	1.17	NS
Milk composition:				
fat, g/kg	42.2	42.3	0.24	NS
protein, g/kg	30.7	30.5	0.06	NS
urea, mg/dl	24.9	26.3		
DM intake:				
silage, kg/cow day	12.0	12.5	0.12	***
concentrates, kg/cow day	7.0	7.0		
Live weight, kg	546	553		
Changes in weight, kg/day	0.02	0.3		

ECM – energy corrected milk, NS – not significant, ***$p<0.001$; **$p<0.01$; *$p<0.05$

$LSD_{0.05}$ – least significant difference at the 5% probability level (silage data n=3; feeding n=28)

CONCLUSIONS

Application of AIV-2000 to red clover-grass mixture resulted in better silage quality compared to untreated silage. First of all secondary fermentation was avoided and the development of moulds and yeasts was slightly inhibited. AIV treated silage resulted in higher intake of dairy cow. Milk yield tended to increase in relation to the increased intake.

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Comparison of nutritive value of clamped and baled grass silage

Object of the study
The aim of this study was to investigate the influence of harvest technology on the nutritive value of silage for heifers

Key words: silage, big bale, clamp, nutritive value, heifer, daily gain

Introduction
The roughages obtained from the grasslands in Poland are primarily harvested and preserved as field-dried hay. The losses of the nutritive components may reach up 50% (Zastawny, 1993). One possibility of improving the feed quality might ensiling pre-wilted sward. One of the newest methods of preservation in Poland is ensiling in big cylindrical bales. The purpose of this study was to determine whether the ensiling of pre-wilted grass with different technologies results in different fermentation and nutritive qualities.

Material and methods
The studies on the nutritive value of grass silage were conducted during the years 1993-1995. The experiment was carried out at Falenty Experimental Station near Warsaw on 9 ha meadow. The silage was made according to two methods: in clamps and in big cylindrical bales (about 400 kg) wrapped with 4 layers of plastic film. The fresh grass was pre-wilted to a dry matter concentration of approximately 40%.
During the feed experiment the silage were sampled and analysed for DM, pH and fatty acids: (lactic, acetic and butyric). These values were used to calculate scores acc. to the Flieg-Zimmer scale (Zimmer, 1966). The chemical composition of feed samples was determined and the net energy for lactation (NEL) was calculated using digestible coefficients by Podkówka (1978). Silages were analysed for: DM, crude protein, crude fibre, crude ash and fat level in relation to dry matter by the use of NIRS technique (Podkówka, 1993).
During 100 days of feed experiment clamp silage were fed to 10 heifers (200 kg) and round bale silage to another 10 heifers. The heifers were fed ad libitum. The daily feed intakes and refusals were recorded. Live weights were determined at the beginning, in the middle and at the end of the study.

Results
The results of chemical analyses of the silage are shown in Table 1. The mean DM content of the silage from clamp and big bale was 477 and 474 g kg^{-1}, respectively. The fresh material contained 182 g kg^{-1} total protein and 281 g kg^{-1} crude fibre. Both silage had similar crude protein and crude fibre contents. The NEL value of feeds varied between 6,26 MJ kg^{-1} DM for clamp silage and 6,28 MJ kg^{-1} DM for big bale silage. The silage assessed acc. to the Flieg-Zimmer scale was good. Both silages had 70 scores. No butyric acid was found in any silage. The pH value in clamp silage was 5,1 and in big bale silage 5,2.

Table 1. Chemical composition (g kg^{-1} DM) of feeds and fermentation parameters of the ensiled material

Examined parameters	Green material	Clamp Silage	Big bale Silage	$LSD^{0.05}$
Dry matter	246	477	474	-
Crude protein	182	183	177	-
Crude fibre	281	268	267	-
Crude ash	96	108	103	-
Crude fat	34	25	20	-
Nitrogen-free extracts	407	416	425	-
Net energy lactation (MJ kg^{-1}DM)	6,37	6,26	6,28	-
Acids in FM of silage (%)				
- lactic	-	2,52	2,61	0,27
- acetic	-	1,71	1,93	0,49
- butyric	-	0,00	0,00	0,00
PH	-	5,1	5,2	0,22
Flieg-Zimmer scores	-	70	70	7,81
Quality acc. to Flieg-Zimmer	-	Good	Good	-

Both silage were preserved well and were readily eaten by heifers. Daily intakes were higher than 6 kg DM of silage (Table 2). The overall live weight gains between two silages were not significantly different. The mean daily gains were 0,608 and 0,551 kg d^{-1} for big bales silage and clamp silage, respectively.

Table 2. Feed intakes (kg DM d^{-1}) and mean daily gains fattening heifers (kg d^{-1})

Feed	Feed intake (kg DM d^{-1})	Daily gains (kg)
Clamp silage	6,25	0,551
Big bale silage	6,19	0,608
$LSD^{0,05}$	0,56	0,080

Conclusions

The fermentation quality of the both silage was good. Both silages had a high nutritive value. The overall live weight gains of heifers fed on the two silages were not significantly different.

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Effects of processing whole-plant maize silage on growth performance and nutrient digestibility in feedlot cattle.

Summary

Sixty heifers and 12 steers were used to evaluate the effects of mechanically processing (crushing the kernels of) whole-plant maize silage on feedlot performance and nutrient digestibility. The three treatments were: preensiled processed, postensiled processed, and nonprocessed maize silages. Heifers fed the processed maize silages grew faster and were more efficient than those fed nonprocessed silage. Steers consuming the two processed silage diets had numerically higher DM, OM, NDF, and ADF digestibilities and significantly higher starch digestibilities than those fed the nonprocessed silage diet. These data suggest that processing whole-plant maize before or after ensiling has a positive effect on both rate and efficiency of gain and nutrient utilization, particularly when the kernels approach the black layer stage of maturity.

(Key Words: Mechanically Processed, Maize Silage, Growing Cattle, Feedlot.)

Introduction

Maize is the most important silage crop used in growing cattle diets throughout the High Plains region of the USA. It has been suggested recently that processing the whole-plant through a forage harvester equipped with an on-board kernel processor could improve growth performance and nutrient digestibility in feedlot cattle. The objective of this study was to evaluate processing maize silage either at the time of harvest or when it is removed from the silo prior to feeding in high-silage diets for feedlot cattle.

Experimental Procedures

Sixty crossbred heifers (avg. initial wt., 268 kg) were used in an 80-day growth trial. The three maize silage treatments were: preensiled processed (PRE), postensiled processed (POST), and nonprocessed (control). Each diet contained 90% of the appropriate silage and 10% supplement (DM basis). The maize hybrid was Pioneer 3394, which was grown under irrigation during the summer of 1996. The six-row, self-propelled forage harvester (CLAAS Jaguar 880) was equipped with an in-line kernel processor. The whole-plant maize was in the 90% milkline stage of maturity, contained 360 g kg^{-1} DM, and was chopped to a 10 mm particle length. The postensiled processed silage was put through a stationary Roskamp roller mill immediately prior to feeding.

Nutrient digestibilities of the three maize silage diets were determined using 12 ruminally cannulated, yearly steers (avg. wt., 330 kg) in a 21-day metabolism trial. Each diet was fed once daily ad libitum to four steers.

Results and Discussion

The results of the two trials are shown in Table 1. Heifers fed the PRE silage had the highest DM intake; those fed the POST silage, the lowest. Heifers receiving the PRE or POST silage diets had higher avg. daily gains than those receiving the control silage diet. Feed efficiency (feed/gain) also was significantly improved by processing, either PRE or POST ensiling.

Steers fed the two processed maize silage diets had numerical improved DM, OM, NDF, and ADF disappearances versus those fed the control silage diet. Starch disappearance was highest for the POST silage diet; lowest, for the control silage diet.

The slight improvement in feed efficiency and greater starch disappearance observed for the POST processed silage versus the PRE processed silage were likely due to an increase in surface area of the kernel and more starch granules exposed to ruminal degradation in the POST processed maize silage. Although all kernels were disrupted in both processed maize silages, those in the POST silage had a more "flake-like" appearance.

Table 1. Effect of processing whole-plant maize silage on growth performance and nutrient digestibilities (g kg^{-1}).

Silage	Daily DM intake, kg	Avg. daily gain, kg	Feed/kg of gain, kg**	Starch	DM	OM	NDF	ADF
PRE	9.62^{x}	1.46^{a}	6.6^{a}	949^{b}	757	775	594	544
POST	9.07^{y}	1.42^{a}	6.4^{a}	967^{a}	755	767	576	546
Control	9.35^{xy}	1.33^{b}	7.0^{b}	931^{c}	747	763	557	542

** 100% DM basis.

[a,b,c] Means within a column with different superscripts differ ($P < 0.05$).

[x,y] Means within a column with different superscripts differ ($P < 0.10$).

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An attempt to screen the extent of fermentation in silage on farms in Västerbotten County and its effects on the carbohydrate and protein fractions

Methods

Farmers with higher energy values than 10 MJ ME in analysed silages were asked via mail about harvesting methods, storing and if they had noticed any special effects on the dairy cows. Key factors I looked for were dry matter content and additives. Farms with interesting silages were then visited. With the help of pH-papers, Clinitest for sugars, and Merckuquant test for ammonium nitrogen a group of silages with signs of unrestricted fermentation and another group of restricted fermented silages were selected. 28 silages were then sampled and analysed for energy (ME) with the in vitro VOS method and crude protein. Rumen degradable protein was measured with the modified S.Griseus in vitro method. Except this soluble protein, protein broken into ammonium nitrogen and protein bound to AFD-fibre were analysed. The carbohydrate fraction was analysed as ADF- and NDF-fibre, sugars, lactic acid, ethanol and volatile fatty acids as butyric, acetic and propionic acids.

Results

The variation from 104 to 184 grams crude protein and from 471 to 594 grams NDF-fibres per kg dry matter reflects different stages of maturity at harvest. The ADF-fibres varied from 277 to 377 grams.
The analysis showed clearly that in practise the fermentation process results in very different amounts of fermentation products in silage and affects on the protein fraction. In the Nordic AAT- system the rumen degradable part of the protein is set to 80 %. But the variation I found was between 52 to 81 percent degradable crude protein. The variation in the soluble part of the crude protein was still greater, from 32 to 73 percent. In spite of the high solubility between 1 to 10 % of the crude protein was broken into ammonium nitrogen. On the other hand between 3 to 11 % of the crude protein had become bound to ADF-fibres. The ADF-bound protein is a not digestible part of the rumen by pass fraction. Corrected for this the variation in the utilisable rumen by pass protein was between 16 to 39 percent of the crude protein.

Due to different dry matter content, use of formic acid and other factors the fermentation process had resulted in between 22 to 128 grams of different fermentation products per kg dry matter. Of these between 15 to 56 percent were contributed by a not wished fermentation of ethanol, acetic, butyric and propionic acids. Formic acid ranged from 0 to 19 gram per kg dry matter. In the most fermented silages just 5 grams of sugars were left in the dry matter but as much as 160 grams in another silage with restricted fermentation. Of the calculated NSC fraction, between 10 to 81% consisted of different fermentation products.

Discussion

The use of standard factors for AAT calculations results in the same value as long as different silages have the same energy content (ME). But considering the big variation in fermentation these analysis illustrate, that may not be true in practise.

Because of the included fermentation products, NSC calculated as a difference can give a false picture of the balance between nonstructural carbohydrates and soluble protein in the

rumen. But except this, analysis used in the CNCPS -model give more information about the quality of fibres and protein. Therefor such analysis and the CNCPS guidelines could be a better base for ration formulations than the standard analysis of crude protein and NDF.

With the big variation in silage fermentation it is obvious that the real feeding and milk output could be different than expected from rations based upon standard analysis. Calculations on data from a Swedish feeding trial with different additives gave very strong but negative correlation between acetic acid in the silage and feed intake. The sum of volatile fatty acids and ethanol in percent of the total amount of fermentation products gave a still stronger negative correlation with the feed intake. Finnish experiences are that volatile fatty acids and ammonium nitrogen have a strong negative impact on the feed intake. Such effects should be considered in ration formulations. But perhaps it is not necessary to distinguish all the different fermentation products. Therefor it perhaps would be possible to implement the Finnish titration routine for measuring the extent of fermentation in Sweden too. Except better possibilities to predict feed intake and balance rations it could give a better understanding of how to achieve a well preserved silage.

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Performance responses and partitioning of nutrients by Hereford x Friesian steers fed on grass silage based diets

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INTRODUCTION

Cattle fed on grass silage generally have higher fat:protein ratios than those fed other forage-based diets. This has been shown to be at least partly due to impaired protein accretion (Gill *et al.*, 1987) and increased fat accretion (Greathead *et al.*, 1996). However, other studies have found variable responses, either no effect (Steen and Moore, 1988) or increased carcass fat (Lonsdale, 1976) in comparison to dried grass diets. Differences between these studies include silage quality and quantity of ingested nitrogen and energy. However, since the partitioning of nutrients between fat and lean deposition is also driven by an animals intrinsic growth plan (which is a function of age, genotype and its physiological state), as well as nutritional status, other factors may be important. An experiment was designed to investigate the effect of diet (based on silage alone or supplemented with additional energy and/or protein) and stage of development on aspects of animal performance and the partitioning of nutrients between fat and lean deposition.

MATERIAL AND METHODS

Ninety two Hereford x Friesian steers were allocated on liveweight to one of 4 dietary treatments; grass silage fed either alone (diet S) or supplemented with fishmeal (diet FM; 150 g/kg silage DM intake fed at equal ME intake to silage) or forage-concentrate (F:C) diets of silage and a barley/soya concentrate (80:20) at ratios of 70:30 or 30:70 (on a DM basis). These diets were selected to investigate the effects of supplying additional protein at constant ME (FM diet), or extra energy and protein (70:30 F:C diet), the latter such that it was non-limiting for growth (30:70 F:C diet). Eight animals were slaughtered at the start of the trial to determine initial carcass chemical composition. Of the remaining 21 animals per group, 3 were slaughtered at liveweights ranging between 250 and 550 kg, at 50 kg intervals. Animals were individually fed and diets were offered *ad libitum* (except for diet FM see above) along with 100 g/d of a commercial premix. At slaughter, individual fat depots were dissected (omental, mesenteric, perirenal) and half carcasses were minced for the determination of carcass fat and protein. Intake and liveweight gains were analysed by regression and ANOVA using initial liveweight as a covariate. General regression was used to test the effects of diet and stage of development and interactions on all other measured parameters. The main dietary effects are reported in this paper.

RESULTS AND DISCUSSION

The composition of the silage was 271.9g freeze dry matter/kg, 26.5 g total-N/kg DM, 2.4 g ammonia-N/kg DM, 98.3g water soluble carbohydrates/kg DM, 11.8 MJ ME/kg DM and a pH of 3.8. The total-N and ME were 31.1 and 108.4 g/kg DM and 13.5 and 14.1 MJ/kg DM for the concentrates and fishmeal, respectively. DM intakes increased ($P < 0.001$) with increasing level of concentrates and this generated the expected differences in both energy and nitrogen intake (Table 1). Liveweight gains were increased by approximately 10, 20 and 32% on the FM, 70:30 and 30:70 F:C diets, respectively, compared to the silage alone. Consequently, animals fed on these diets reached target weight more quickly than animals fed silage. For example, 30:70 F:C fed animals achieved the average slaughter weight of 400 kg on average 70 days before those fed on silage. Average carcass weights were heavier on concentrate diets but composition was similar across diets. Rates of carcass protein gain were higher on FM and concentrate diets compared to the silage ($P < 0.001$). In contrast, carcass fat gains were similar between silage and FM but were much

Table 1 Effect of diet on animal performance and carcass composition

	Diets					
	Silage	**Fishmeal**	**70:30 F:C**	**30:70 F:C**	**S.e.**	**P**
Total DM intake (kg/d)	4.92	4.89	5.49	6.87	0.125	0.001
Energy intake (MJ/d)	58.1	58.8	67.5	89.2	1.54	0.001
Nitrogen intake (kg/d)	130.4	180.6	152.7	203.9	3.82	0.001
Liveweight gain (kg/d)	0.93	1.02	1.12	1.22	0.039	0.001
Feed conversion efficiency (kg gain/kg feed * 100)	19.7	25.4	20.6	19.3	0.54	0.001
Av. liveweight at slaughter (kg)	396	398	411	397	29.9	NS
Carcass weight (kg)	202.8	198.9	207.6	208.3	0.011*	0.004
Killing-out pecentage (%)	0.53	0.52	0.54	0.54	0.004	0.002
Days on trial **	272	250	230	202	0.029*	0.094
Carcass protein (kg)	35.8	35.9	35.8	35.9	0.015*	NS
Carcass fat (kg)	41.8	39.8	45.8	48.7	0.039*	NS
Carcass protein gain (g/d)	79.3	86.0	94.4	107.4	2.83	0.001
Carcass fat gain (g/d)	128.9	131.6	174.8	211.7	6.90	0.001
Fat:protein gain ratio	1.17	1.11	1.28	1.36	0.042*	0.06
Total non-carcass fat (kg)	23.2	22.2	27.4	30.7	0.031*	0.008
Total fat (kg)	65.2	62.2	73.5	79.6	0.033*	0.098

* s.e. expressed on log scale ** number of days on trial to reach the average liveweight at slaughter; NS = not significant.

higher on the 70:30 and 30:70 F:C diets ($P < 0.001$). This resulted in similar fat:protein gain ratios between silage and FM fed animals but this ratio was approximately 15% higher on the concentrate diets ($P = 0.06$). Carcass fat and total non-carcass fat (sum of the individual dissected depots) in animals fed concentrate diets was higher than those fed on silage or FM. The silage quality (as reflected in ME and total-N) was good which contributed to high intakes and liveweight gains. The effect of fishmeal supplementation was small in contrast to Gill *et al.* (1987) working with an average quality silage and suggests that in this experiment the animal's requirement for protein deposition was met from the silage alone. Supplementing with additional concentrates reduced days on trial, but the animals achieved the same amounts of carcass protein but deposited more fat.

CONCLUSIONS

Supplementation of good quality silage with protein and/or energy has little effect on carcass protein deposition although it reduced time taken to achieve the same carcass weight, by 22-70 days. Supplementation of silage with concentrates increased fat deposition. The requirement to supplement silage with additional concentrates is a balance between cost effectiveness, time taken to achieve a particular slaughter weight and the risk of increased levels of body fat.

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Effects of maturity of corn silage on digestibilities, passage rates, and milk production of lactating cow.

Introduction

Corn is used as the primary forage for dairy cattle rations in northern area of Japan, so determining the most suitable hybrid at which to plant corn is important in silage production and milk production. The differences of maturities among corn varieties affect silage quality and nutritive value, and may affect dry matter (DM) intake, digestibilities, passage rates and milk production of lactating cow. The experiment was to evaluate the effect of two different maturing hybrid corn for use as silage in the diets of dairy cows on DMI, total tract nutrient digestion, passage rates of digesta, and milk production and composition.

Material and Methods

Two different maturing varieties of whole plant corn were used in the experiment. Early maturing hybrid (Noruda; 83 d, Minesota rating) and medium maturing hybrid (Pioneer 3795; 95 d, Minesota rating) were planted and harvested at same time, and stored in stack silo.

Twelve Holstein cows (MBW:680kg) were started on experiment 7 to 10 wk postpartum in a cross over design with two 3-wk periods. Cows were assigned to one of two treatment sequences: 1) early maturing hybrid (EH) or 2) medium maturing hybrid (MH).

Cows were fed either corn silage of early maturing hybrid or silage of medium maturing hybrid *ad libitum* intake with supplemental concentrates and timothy-red clover silage to meet the nutrient requirements.

Apparent digestibilities were determined using ADL as an internal marker. Passage rates were estimated from fecal excretion curves of Cobalt (Co), Dysprosium (Dy), Ytterbium (Yb), Lanthanum (La), Celium (Ce), and Samarium (Sm) as the markers for liquid, corn silage (Corn), timothy-red clover silage (Grass), beet pulp (BP), Soybean meal (SBM), and compound feed (CF), respectively, on the basis of two compartment model of Grovum and Williams (1973, Br. J. Nutr., 30, 313-329). The markers were prepared and dosed by the procedures outlined Moore et al (1992, J. Anim. Sci., 70 3528-3540) with a modification.

Results

EH was harvested at soft dough stage (24.6% DM) and MH was harvested at early dent (27.3% DM). EH contained 7.5% crude protein (CP), 43.9% Neutral detergent fiber

(NDF) and 25.3% Acid detergent fiber (ADF), and MH contained 7.6% CP, 47.7% NDF, and 30.7% ADF. The effluent production from MH silage (213 kg/ton) was greater than that from EH silage (110 kg/ton). Fermentative quality of MH silage was similar to that of EH silage.

The DMI of cows fed EH silage was higher than that of cows fed MH silage (10.0 kg/day vs 9.1 kg/day), and concentrates of cows fed EH treatment was lower than that of MH treatment (10.3 kg/day vs 11.1 kg/day). There was not a significant difference in total feed intake of cows fed between EH and MH treatments.

Apparent digesibilities of DM, CP, and NDF were similar between treatments. Significant differences were not found in ruminal passage rates (k_1) of liquid and solids except SBM between treatments. Total mean retention time (TMRT) of Dy applied to corn silage averaged longer 4.0 hour for MH diets than for EH diets.

No differences were observed for milk yield and milk composition of cows between treatments.

Conclusion

Results of the experiment indicated that the maturity of corn strongly affected chemical composition, effluent losses, and nutritive value of silage. DMI of cows fed EH silage was greater than MH silage , probably because of more rapid digestion and passage in total digestive tract. Our date suggest that it is advantageous to plant the early maturing hybrid corn for use as silage in the diets of lactating cows in northern area of Japan .

Key word: Corn silage, Milk production, Passage rate

Table 1. Ruminal passage rate (k_1) and total mean retention time (TMRT) in lactating cows fed different maturing corn silages [1)]

	k_1 (%/hour)		TMRT (hour)	
	EH	MH	EH	MH
Co (Liquid)	6.8	6.7	23.4	23.9
Dy (Corn silage)	4.6	4.3	36.2 [b]	40.2 [a]
Yb (Timothy- red clover silage)	4.4	4.5	38.2	39.3
La (Beet pulp)	4.8	4.9	37.8 [b]	41.5 [a]
Ce (Soybean meal)	5.3 [a]	4.5 [b]	32.4	35.1
Sm (Compound feed)	5.0	5.2	32.4	33.3

1) EH = Early maturing hybrid , MH= medium maturing hybrid.

2) SEM=Standard error of mean.

3) [a, b] Means in same row with different superscripts differ ($P<0.05$)

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The effects of stage of maturity at harvest and kernel processing on the nutrient digestibility of maize silage.

Summary

Twelve ruminally cannulated crossbred steers were used to evaluate the effects of advancing stage of maturity and kernel processing of whole-plant maize silage on nutrient digestibility. The six silage diets were: 50 percent milk-line processed or unprocessed, 80 percent milkline processed or unprocessed, and 7 days post black-layer processed or unprocessed. Steers consuming the 80 percent milkline and 7 days post black-layer processed diets had numerically higher DM and OM digestibilities, and all processed diets had numerically higher starch digestibilities. However, the three processed diets did have numerically lower fiber digestibilities (NDF and/or ADF). Steers consuming the 80 percent milkline diets had numerically higher nutrient disappearances than those fed the diets containing the other two stages of maturity. Yield samples taken at each of the three harvests showed that whole-plant DM and grain yields increased with advancing maturity. The data indicate that harvesting at the 80 percent milkline stage of maturity and processing the whole-plant maximized DM yield and nutrient utilization.

(Key Words: Mechanically Processed, Maize Silage, Stage of Maturity, Growing Cattle, Feedlot.)

Introduction

Improving the digestibility of whole-plant maize (both the stover and grain) would have a positive impact on growing cattle performance. It has been suggested that using a kernel processor on the forage harvester could improve nutrient digestibility. The objective of this study was to evaluate the effect of stage of kernel maturity at harvest and processing on the utilization of high-silage diets by growing cattle.

Experimental Procedures

The nutrient digestibilities of six maize silage diets were determined using 12 ruminally cannulated, yearling steers in a Latin square metabolism study. The steers were housed in a climate-controlled barn where they were tethered in individual tie stalls. The 21-day periods consisted of four phases: a 10-day diet adaptation, an 8-day total fecal collection (two, 4-day periods), a 2-day ruminal fermentation, and a 1-day ruminal evacuation.

Pioneer 3394 maize hybrid was grown under irrigation during the summer of 1997. A three-row, self propelled precision cut forage harvester (FieldQueen) was used to harvest the whole-plant at the three stages of maturity, which were 50 and 80 percent milkline and 7-days post black-layer

and the DM contents were 320, 380, and 420 g kg^{-1}, respectively. The forage was chopped to a 10 mm particle length and four, 1.2 H 1.8 m concrete pilot-scale silos were filled at each harvest date. Two silos were filled with chopped forage that was put through a stationary kernel processor (Roskamp roller mill), and two silos were filled without further processing. At each stage of maturity, three 18 m sections of whole-plant maize were hand-harvested and separated into stover and grain portions, which were dried and weighed for yield determinations.

Results and Discussion

The preensiled stover increased in contents of DM, CP, NDF, and ADF as maturity advanced, and whole-plant DM yield and the proportion of grain in the whole-plant also increased with advancing maturity (data not shown).

The effects of stage of maturity and processing whole-plant maize silage on nutrient digestibilities are shown in Table 1. Steers consuming the 80 percent milkline and 7-days post black-layer processed silage diets had numerically higher DM and OM digestibilities, and all processed silage diets had numerically higher starch digestibilities. However, the three processed silage diets did have numerically lower fiber digestibilities (NDF and/or ADF). The 50 and 80 percent milkline silage diets had numerically higher DM, OM, CP, NDF, and ADF disappearances than did the respective 7-days post black-layer silage diets.

The minimal improvement in starch disappearance observed in the processed silages were likely due to an increased surface area of the kernel and more starch granules exposed to ruminal fermentation compared to the unprocessed maize silage diets. The slight negative impact of processing on fiber digestion could be due to a carbohydrate effect on ruminal bacteria activity.

Table 1. Nutrient digestibilities (g kg^{-1}) of the six maize silage diets.

	50% milkline		80% milkline		7-days post black-layer		
Item	P	U	P	U	P	U	SE
DM	729^{a}	738^{a}	743^{a}	739^{a}	718^{ab}	702^{b}	9.7
OM	747^{a}	762^{a}	770^{a}	762^{a}	735^{ab}	723^{b}	9.7
Starch	964^{a}	946^{b}	964^{a}	940^{b}	949^{ab}	933^{c}	6.8
CP	719^{c}	781^{a}	805^{a}	769^{ab}	714^{c}	741^{b}	14.0
NDF	504^{c}	535^{b}	541^{a}	558^{a}	502^{c}	512^{bc}	12.0
ADF	486^{bc}	537^{a}	523^{ab}	533^{a}	466^{cd}	455^{d}	18.0

^{a,b,c,d} Means within a row with different superscripts differ ($P < 0.05$).

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Effect of addition of *Acremonium* cellulase on cell wall constituents and *in vitro* dry matter digestibility of silages

Key Words: Cellulase, *in vitro* dry matter digestibility, Organic cell wall, Silage

Introduction

It is reported that new Cellulase (CEL) derived from *Acremonium* may be used as a silage additive to stimulate silage fermentation by promoting lactic acid bacteria with adequate amounts of water-soluble carbohydrate (WSC) resulting from the hydrolysis of cellulose (Ataku *et al.*, 1997; Aniwaru *et al.*, 1997, 1998). At present, however, there are insufficient data about the effect of the cellulase on cell wall constituents and *in vitro* dry matter digestibility (IVDMD). The purpose of this report was, therefore, to demonstrate the effect of CEL on contents of organic cell wall (OCW) and its both high (Oa) and low (Ob) digestible fractions, and IVDMD in the silages of alfalfa and timothy.

Materials and Methods

Alfalfa (cv. Euver, 21.3% DM, 16.5% CP, 8.0% WSC, and 49.4% NDF) and timothy (cv. Hokusen, 21.5% DM, 8.4% CP, 8.4% WSC, and 68.0% NDF) were ensiled in laboratory silos. Four levels of the CEL (Meiji Seika Ltd.; 0, 0.005, 0.01, and 0,02% to alfalfa and 0, 0.006, 0.012, and 0.024% to timothy) were added to each forage. The silos were opened after 50 d and the OCW, Oa and Ob contents (Abe *et al.*, 1979) and IVDMD were determined.

Results

The OCW, Oa and Ob contents and in vitro dry matter digestibility of the silages are shown in Table 1. Cell wall constituents were declined in all silages treated with additional CEL. Moreover, in timothy silages with 0.024% CEL and alfalfa 0.02% CEL, decrease of the Oa was highest, while decrease of the Ob lowest. For both forages, 0.02% or more than 0.02% CEL appears to bo the excessive addition, because of hydrolysis of Oa stimulated and hydrolysis of Ob depressed.

In timothy silages, IVDMD were increased significantly by additions of 0.006% and 0.012% CEL, but decreased significantly by 0.024%. In all alfalfa silages treated with CEL, IVDMD decreased significantly.

Relationships among the cell wall constituents and IVDMD of silages are shown in Table 2. There was a positive relationship between Oa and IVDMD in both forages. This study further provided additional information that Oa is responsible for the changes of IVDMD in the CEL-treated silage.

Conclusions

These results suggested that cell wall constituents were reduced by addition of CEL, and that Oa was responsible for the changes of IVDMD in the CEL-treated silage.

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Table 1. Cell wall constituents and *in vitro* dry matter digestibility of the silages.

	Cellulase Addition (%)	OCW	Ob	Oa	IVDMD
		(%DM)			(%)
Timothy	0	70.1^{A}	56.2^{Aa}	13.9^{Aa}	78.3^{B}
	0.006	62.0^{B}	50.6^{Bb}	11.4^{Ab}	80.1^{A}
	0.012	58.3^{C}	47.1^{Bc}	11.1^{Ab}	79.2^{AB}
	0.024	57.8^{C}	51.0^{ABb}	6.8^{Bc}	73.1^{C}
Alfalfa	0	50.3^{Aa}	41.2^{a}	9.1^{a}	57.8^{Aa}
	0.005	47.0^{ABb}	38.7^{b}	8.3^{a}	54.2^{Bbc}
	0.010	47.0^{ABb}	38.5^{b}	8.5^{a}	54.8^{ABb}
	0.020	46.0^{Bb}	40.1^{ab}	6.0^{b}	52.2^{Bc}

OCW: Organic cell wall, Ob: low digestibile cell wall, Oa: high digestibile cell wall, IVDMD: *in vitro* dry matter digestibility.
Means with the same material with different superscripts letters are different; $^{A\ B\ C}$ $P<0.01$, $^{a\ b\ c}$ $P<0.05$.

Table 2. Relationships among the cell wall constituents and IVDMD of the silages.

		OCW	Ob	Oa
Timothy	Ob	0.87**		
	Oa	0.78*	0.38	
	IVDMD	0.34	-0.11	0.78*
Alfalfa	Ob	0.67		
	Oa	0.73*	-0.03	
	IVDMD	0.93**	0.52	0.78*

OCW, Ob, Oa and IVDMD: See Table 1, ** $P<0.01$, * $P<0.05$.

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Near Infrared Transmission Study for Fresh Grass Silage Analysis

The potential of fresh grass silage analysis by Near Infrared Transmission (NIT) was studied using an Infratec 1265 Meat Analyzer. Global calibration equations were calculated by modified partial least square (MPLS) regression analysis for various constituents in a spectral library of 774 grass silage samples from North German farms ranging from 15% to 80% corrected dry matter. The calibration and cross validation statistics for dry matter (DMc), pH, lactic acid (LA), acetic acid (AA), butyric acid (BA) and to a lesser extent ammonia nitrogen (NH_3-N) and ammonia nitrogen as a proportion of total nitrogen (NH3-N/ N_{tot}%) indicated an analytical potential for substituting the hitherto used sensory method for silage quality assessment (Table 1).

Table 1: Calibration and Cross Validation Statistics of Global Calibration Equations for Dry Matter, pH, Lactic Acid, Acetic Acid, Butyric Acid, Ammonia Nitrogen Content and Ammonia Nitrogen Proportion of Total Nitrogen

		Dry $Matter_c$	pH	LA*	AA*	BA*	NH_3-N*	NH_3-N/ $_{tot}$ (%)
Calibration	RSQ	0.94	0.79	0.81	0.72	0.81	0.62	0.67
	SEC	2.71	0.22	0.95	0.50	0.65	0.10	2.43
Cross Validation	1-VR	0.93	0.77	0.80	0.71	0.79	0.60	0.65
	SECV	2.79	0.23	0.98	0.51	0.68	0.11	2.49

*Dry Matter based

Independent validation of these equations was performed on two smaller sets of grass silage samples (GSNITVAL and GSGC9899) from comparable geographical background and comparable sources. The calibration and cross validation results were confirmed in so far as satisfactory standard errors of prediction for pH and fermentation products were achieved (Table 2).

In order to test whether a further increase in the accuracy of predicition could be achieved by a new calibration approach capable of accommodating non linearities in data sets was tried. In this case the local calibration procedure newly developed by SHENK and WESTERHAUS was employed by first individually comparing the spectra of the two validation sets to the 774 grass silage samples of the spectral library. After the selection of 75 calibration samples of high spectral similarity with each test sample each of these was then predicted by a specifically calculated calibration equation. However, overall standard errors of prediction of these local calibrations for the fermentation products in the two validation sets generally showed no consistent improvement in prediction accuracy over the conventional global calibrations. In contrast to this, the accuracy of predicting DMc by the local calibration approach resulted in a statistically significant improvement in the degree of fit between reference and predicted data. As a matter of fact, the local calibration approach reduced the prediction error of DMc from 3.8% to 1.5%.

Table 2: Standard Error of Prediction of Global and Local Calibration Equations for Dry Matter, pH, Lactic Acid, Acetic Acid, Butyric Acid, Ammonia Nitrogen Content and Ammonia Nitrogen Proportion of Total Nitrogen

	GSNITVAL		*GSGC98*	
	Global	Local	Global	Local
pH	0.26	0.33	0.22	0.27
LA*	1.2	1.7	1.4	2.4
AA*	0.41	0.36	0.39	0.51
BA*	0.63	0.77	0.55	0.53
NH_3-N*	0.09	0.09	0.05	0.09
NH_3-N/ N_{tot} (%)	2.7	3.0	1.7	3.2

*Dry Matter based

A spectrochemical interpretation of these results is tentatively given: Generally the absorption pattern of fresh grass silages in the near infrared is dominated by OH-absorptions typical for water which seem to give rise to a non linear relationship between NIR absorptions and water content over the enormous range in DMc studied here. Under these conditions spectral similarity between library and test samples is primarily a function of DMc content. This explains why the local calibration approach results in an improvement of prediction accuracy for DMc over the conventional global calibration. In contrast to this, all the fermentation products assessed here have much less influence on the spectra and thus on the calculation of spectral similarity between library and test samples. As a consequence, the local calibration approach does not improve the accuracy of assessing these constituents.

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Nutritional evaluation of a new fodder for maintenance feeding of adult buffaloes. I. Ensiled paddy straw and potato haulm.

Attempts were made to develop a simple and economic technology, where sophisticated machinery or the use of chemicals are not involved , which can be adopted by the poor and illiterate farmers. In this new technology, paddy straw (*Oryzasativa L*) was ensiled with potato haulm (*Solanum Tuberosum*) in 1:5 ratio and supplemented with 3.0 % molasses.

Potato haulm was harvested and chaffed, mixed with paddy straw in the ratio of 5:1 and supplemented with 3.0 % molasses. About 6000 kg of silage was prepared in a Katchha silopit, using 5000 kg potato haulm, 1000 kg paddy straw and 180 kg molasses. The silopit was opened after 104 days of ensiling the original material. The representative silage sample was drawn and evaluated as per Breirem and Ulvesli (1960) recommendations.

The silage made during the current study contained an appreciable amount of crude protein (10.21%), lactic acid (2.38%), TVFA (2.24%). NH4 - N as percent of total nitrogen was only 2.16% and pH was 5.16. Based on these important chemical parameters this silage was graded as "TOP QUALITY SILAGE."

The data related to relative intake (RI) of dry matter and nutritive value index (NVI) clearly indicate that silage fed to the animals was quite palatable and acceptable and was 72% as good as standard barseem hay fed to the control group buffaloes.

The data of the present experiment indicate that adult nonproducing buffaloes could be reared successfuly if consumption of ensiled paddy straw and potato haulm is about 18 kg per head per day. The required amount of energy and protein could be met through this amount of silage. However the systematic pesticide residue in the ensiled material should be below 2 ppm.

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The Effect of a Commercial Silage Inoculant (Pioneer® brand 1188) on Animal Performance

Introduction:

Favourable bacteria supplied as a tool for silage making have been commercially available in Europe now for some 20 years. The initial appearance of such inoculants was greeted with a great deal of sceptism as to their actual value and worth. Such was the interest in the product concept of silage inoculation that the Eurobac Group was established and in 1986 a meeting was held in Uppsala to review the then currently available data. The conclusions of this meeting now reviewed with the benefit of some 13 years of additional research data proves interesting. Development in research tools has enabled not only additional efficacy studies to be conducted, but in addition, detailed investigation (13) has occurred into the microbial ecology of using bacterial inoculants. This has led to the development of more targeted and robust products. The objective of this paper is to present an overview of efficacy studies that have been performed with one such commercialised product, the results of which contradict some of the original Eurobac conclusions.

Materials and Methods:

A range of typical European forages (grass silage with a dry matter range of 18 – 48%, and maize silage of 28 – 37% dry matter) were ensiled using good experimental techniques to ensure identical pre-ensiling material across treatments for each experiment. In every study, untreated silage was compared to material inoculated with Pioneer® brand 1188, applied according to manufacturer's instruction to provide 10^5 CFU/g forage. Animal feeding studies were performed following recommended protocols for total collection sheep digestibility trials, beef performance evaluations or dairy feeding studies.

Results:

Type of Trial	Crop Type	Crop %DM range	Control	1188	Difference	P<	References
			Digestibility Org. Matter (%)				
Wether digestibility	maize	37	74,0[a]	77,3[b]	3,3	0,01	2
	grass	19-48	70,6[a]	72,3[b]	1,7	0,01	3, 4, 5, 6, 7, 9,11
			Average daily Gain (g/day)				
Beef feeding	maize	37	1254[a]	1329[b]	75	0,06	2
	grass	17-49	795[a]	877[b]	81	0,01	4, 8, 10
			Milk Yield (kg/day)				
Dairy feeding	maize	28-37	22,9[a]	23,8[b]	0,9	0,05	1
	grass	18-48	23,4[a]	24,5[b]	1,1	0,05	9, 12

a,b different superscripts indicate significant difference

Conclusion:

One of the biggest criticisms that can be levelled at the Eurobac conclusions is that they were based on summarising data across all products, rather than assessing individual inoculants on their own merits. Each product's efficacy characteristics will be different, the variation depending upon the original selection criteria used and subsequent bacteria chosen, in addition to the final product's viability. Indeed, through the use of targeted screening tools in product development, it has been demonstrated through animal feeding studies that it is possible to produce an inoculant that is efficacious across a range of ensiling conditions. This is somewhat at odds to the original Eurobac conclusions, perhaps it would now be better to conclude that ;

" The ensiling conditions across which an inoculant will work are determined by the selection criteria used during product development ".

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The Nutritive Value and Aerobic Stability of Round Baled Silages from Grass-alfalfa Mixed Crop Ensiled with Additives Containing Formic Acid

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INTRODUCTION

Grass- legume have a high nutritive value but are difficult to ensile. Wilting and baling these materials can make good silage, but this silage is very susceptible to the influence of outside factors which may lead to the development of undesired organism, especially moulds. Formic acid as well as other organic acids and their salts facilitate ensiling of grasses and legumes. They can also limit the development and activity of moulds.

MATERIALS AND METHODS

Silages were made from mixtures of grasses and alfalfa (lucerne 79%, cocksfoot 16%, timothy 5%). The experimental material was obtained from plots from the first cut, at the phase of earing of grasses and budding of alfalfa. After 12 h. wilting the plant material was ensiled using a Sipma baler and wrapped with four layers of stretch film. Two experimental treatments were applied to the fresh forage: a) silage preparation containing 55%formic acid, 24% ammonia formate, 5% propionic acid, 1% benzoic acid and 1% esters of benzoic acid - „KemiSile 2000" (manufactured and delivered by Kemira Chemical Oys - Finland) b) an untreated control without additive. The preparation was applied during harvesting (4l/t fresh material). Six bales from each treatments were analysed (one bale was one replicate). The bales were opened after 60 days and samples were collected using a special silage core. The following parameters were determined in the silages: basic nutrients, fermentation products , aerobic stability estimated by temperature measurements, changes in chemical composition after 7 days exposure at 20°C and as well as quantities of E. coli and Clostridium bacteria, yeasts and moulds found in silages at the moment of sampling .

RESULTS AND DISCUSSION

The examined silages were characterised by a high protein concentration (approximately 200g/kg DM, similar to the initial material) and by a low concentration of ammonia nitrogen (2.5 - 2.7g/kg DM). Furthermore, the experimental silages were found to contain, moderate concentrations of lactic acid (46 - 48 g/kg DM) and low concentrations of butyric (3.2 – 4.1 g/kg DM). The applied additive reduced the content of butyric acid and increased quantities of soluble sugars which remained in the silage. No significant changes in chemical composition of the experimental silages were found after 7 days exposure to air, although the amount of lactic acid decreased, while the amount of ammonia and butyric acid increased and pH rose. However, the above changes were smaller in samples without additive, temperature was found to increase from 20.7 to 24,6° C from 5th day of exposure onwards. Silage samples containing the additive were found to contain 6 times lower levels of Clostridium bacteria and 2.5 times smaller quantities of moulds. The additive did not affect the number of yeast, while the number E. coli was higher.

CONCLUSIONS

Chemical additives containing formic, propionic and benzoic acid improved the quality and stability of mixed crop silages preserved in bales. Simultaneously, the application of this additive reduced the number of moulds compared with untreated bales.

Table 1. Chemical composition of silages from wilted grass-legumes mixture (bales)

	with additive		without additive	
	after opening	after 7 days	after opening	after 7 days
dry matter g/k	332	381	338	380
crude protein g/kgDM	202	201	187	178
crude fiber g/kg DM	267	275	260	268
NH_3 N g/kg DM	2.6	2.7	2.6	3.9
lactic acid g/kg DM	49	47	46	44
formic acid g/kg DM	9	8	-	-
acetic acid g/kg DM	23,54	26	26	29
butyric acid g/kg DM	3.2	3.6	4.1	4.3
WSC g/kg DM	38	30	29	26
pH	5,16	5,77	5,37	6,54

Figure 1. Changes of temperature in silages during 7 days exposure to the air

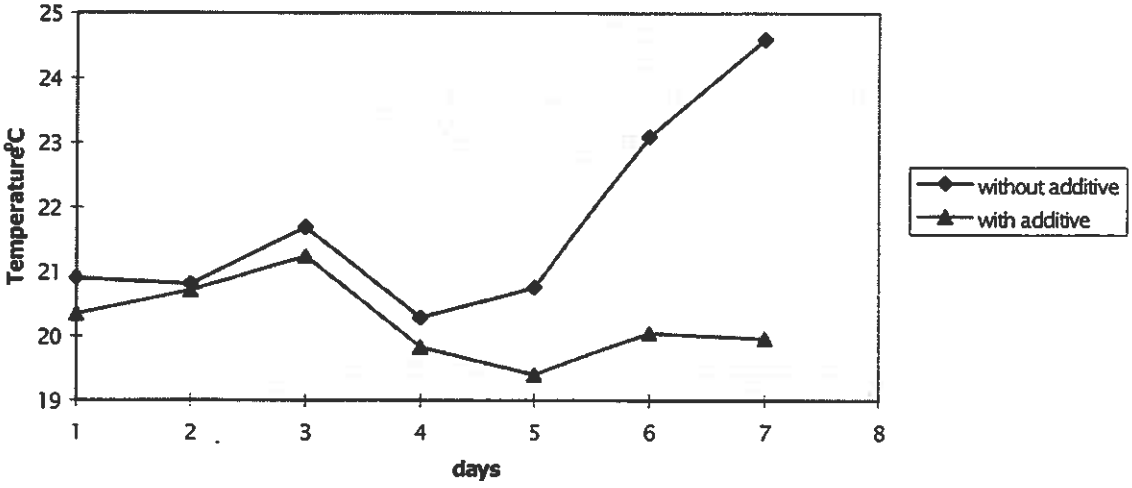

Table2. Hygenic value of silages from grass-legumes mixture

Microorganism	c.f.u./ g FM	
	with additive	without additive
E. coli	$1.8*10^4$	$1.4*10^4$
Clostridium	$0.7*10^5$	$4.5*10^5$
yeasts	$1.3*10^7$	$1.1*10^7$
moulds	$0.2*10^4$	$0.4*10^4$

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Effect of long-term high temperature on spores of *Clostridium tyrobutyricum*

Key words: *Clostridia tyrobutyricum*, spores, temperature, Farmyard manure, compost

Object of the study

The aim of the study was to investigate if temperatures possible to obtain during the composting process affects clostridial spores. The hypotheses was that spores exposed to high temperatures (50 - 70°C) of long duration, significantly will be reduced.

Introduction

Cattle manure makes a valuable resource in herbage production, but the use of uncomposted farmyard manure increases the hygienic risks. Manure particles picked up at harvest by the chopper, follows the herbage into the silo, and causes poor quality silage rich in clostridial spores (Rammer and Lingvall 1997; Rammer et al 1994). Besides being detrimental to the silage process, spores of *C. tyrobutyricum* may contaminate milk and, being able to survive pasteurisation at the dairy, cause 'late blow' in hard cheeses.

Optimal conditions for *C tyrobutyricum* is at 37°C, pH 5.8 and water activity over 0.94. Growth can occur between 10 to 42 °C and within a pH interval of 4.7 to 7.3 (Bergére and Hermier 1970). In vegetative state clostridia are as sensitive as other organisms for high temperatures, but in spore forms they are resistant to both high temperatures and desiccation.

Out growth of vegetative cells from spores are inhibited at temperatures above 42°C. But the spores can also to some extent be affected by high temperatures, and spores of clostridium are considered to be relatively heat sensitive. Thermal death of spores, however, is markedly dependent on the composition and pH of the suspending medium (Russell 1982). Depending on pH and composition of medium, Bergére and Hermier (1970) showed that ninety percent of the spores of *C tyrobutyricum* could be killed at 80°C for 110 to 260 minutes. Accordingly, if the spores are exposed for sufficiently long-term, there is a possibility that the number of clostridia could be reduced by temperatures possible to reach in a carefully made compost.

Material and methods

20 g-samples of well mixed farmyard manure with initially 1.3×10^3 colony forming units (cfu) g^{-1} manure (unspecified clostridial spores), were infected with spores of *Clostridium tyrobutyricum* (10^4 cfu g^{-1}). The manure samples were stored aerobically or anaerobically (BBL Gaspack system) in an incubator. Bottles with reinforced clostridial medium (RCM), inoculated with 10^4 spores ml^{-1}, were used as control. The temperature of the incubator was regulated manually to resemble temperature changes normally achieved in a compost. Starting at 20°C, the temperature reached 71°C after 10 days and was thereafter gradually decreased during 90 days. Optimal temperature for clostridial growth (37°C) was achieved after four days. Samples of manure and RCM media were taken out from the incubator after 30, 60 and 90 days and the number of clostridial spores was enumerated according to Jonsson (1990).

Results

The numbers of viable clostridial spores recovered in the manure samples and in the RCM media at different times are presented in Table 1. Only in aerobically stored manure stored for 30 days were viable spores found. In anaerobically stored manure no spores were found and also not in any of the bottles of RCM. No spores were detected after 60 and 90 days, neither in the manure nor in the RCM, independently of storage method. Some growth, however, had probably occurred as the medium in the bottles was turbid.

Table 1. The number of viable clostridial spores (log cfu g^{-1}) enumerated in manure and RCM (reinforced clostridial medium) at the experimental start (0) and after aerobic or anaerobic storage for 30, 60 or 90 days. Mean values and (standard deviation)

	Manure; stored		RCM; stored	
Sampling day	**Aerobically**	**Anaerobically**	**Aerobically**	**Anaerobically**
0; natural	3.1 (0.0)	3.1 (0.0)	0	0
0; inoculated	4.0 (0.0)	4.0 (0.0)	4.0 (0.0)	4.0 (0.0)
30	2.2 (1.9)	<2.0	<2.0	<2.0
60	<2.0	<2.0	<2.0	<2.0
90	<2.0	<2.0	<2.0	<2.0

Conclusions

- The result of this lab-scale experiment showed that spore reduction owing to long-term high temperature could occur.
- A spore reduction due to heat killing might be a possible effect of the composting process.
- To prove a more general application of this assumption, however, further investigations in full-scale compost experiments must be performed.

Acknowledgement

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Management factors preventing growth of clostridia in silage

Key words: Conditioning, wilting, additive, film colour, film layers, silage quality, clostridia

The object of work

The aim of this trial was to study the effects on hygienic quality of:

- Degree of wilting (300 and 400 g/kg).
- Variation in water activity within the swath, by comparing conditioned and unconditioned swaths.
- Heat stress and oxygen permeation using black and white film in 6 or 10 layers.
- Additives (untreated control and Kofasil Ultra).

Introduction

Spores of clostridia can cause problems in cheese making and this has resulted in a reduction of payment if the milk contain too many spores at delivery. Wilting is an efficient strategy to reduce the risk of clostridia growth in silage. In Sweden, the recommendation is to wilt in the swath, without tedding, to reduce the risk of soil contamination. This practice has, however, some drawbacks. The drying will be slower than if the crop is spread over a larger surface and the surface of the swath will dry faster than the bottom. If the crop is ensiled in big bales, or after use of self-loading wagons, there might be pockets of lower DM than the average of the crop. A low DM content, particularly when combined with increased temperature and oxygen permeation, can cause an environment suitable for clostridia growth.

Materials and methods

The crop was a first cut of a grass dominated mixed clover / grass ley, fertilised, in the spring, with 20 ton farmyard manure/ha. The weather conditions during harvest were not suitable for wilting and therefore the crop was ensiled 1 or 2 days after mowing.

Temperature in the bales was measured using thermocouples, placed on the bale surface, under the film, and approximately 10 cm into the bale. The temperatures were measured every minute and the mean temperature for each hour was calculated.

The additive used was Kofasil Ultra, 4-5 l/ton fresh matter. Kofasil Ultra is an additive containing hexamine, sodiumpropionate, sodiumnitrate and sodiumbenzoate. It was sprayed over the swath in front of the pick-up. To ensure a correct application rate, the container with Kofasil Ultra was weighed after each bale and the flow was adjusted when necessary.

Samples from the silage were taken with a sample corer (ø 4 cm) at three consecutive depths from the surface: 0 - 10 cm, 10 - 20 cm and 20 - 30 cm, at 6 different points on the southern side of the bale. These samples were analysed depthwise to determine the results on fermentation of depth within each bale.

Results

The planned DM-contents were not achieved, due to humid weather conditions. The DM after 1 or 2 days wilting were 263 (SD=15,9, n=56) and 344 (SD=38,6, n=32) g/kg respectively. These DM contents were not enough to inhibit the growth of clostridia. Due to the weather, the harvest was delayed, which resulted in a mature crop of low WSC concentrations, which often is difficult to ensile.

Table 1. Results of silage analysis from the different treatments. Mean values only. (n=3)

Day wilt	Conditioned	Additive	Film Layers	Colour	DM g/kg	pH	NH_3 g/kg N	g/kg DM WSC	Lact	Eth	Butyr	Spores log CFU/g
1	YES	CONT.	6	WHITE	260	4.34	97,1	4,7	56,5	8,9	2,6	3,4
1	YES	CONT.	6	BLACK	252	4.43	105,9	3,6	53,6	10,5	6,5	5.2
1	YES	KOFASIL	6	WHITE	249	4.51	64,0	11,35	54,1	5,0	<0,3	<2
1	YES	KOFASIL	6	BLACK	264	4.66	52,7	21,80	45,0	3,9	<0,3	<2
1	NO	CONT.	6	WHITE	234	4.86	154,4	5,2	31,3	14,3	16,8	6.0
1	NO	CONT.	6	BLACK	250	4.72	154,0	2,3	29,7	11,0	17,4	6.2
1	NO	KOFASIL	6	WHITE	242	4.72	87,3	23,37	42,3	5,0	<0,3	<2
1	NO	KOFASIL	6	BLACK	258	4.56	66,2	29,30	44,0	4,7	<0,3	<2
1	NO	CONT.	10	WHITE	256	4.86	143,5	2,0	34,9	14,5	13,5	5.6
1	NO	CONT.	10	BLACK	241	4.43	143,5	1,8	51,0	11,7	9,8	4.7
1	NO	KOFASIL	10	WHITE	261	4.66	61,1	23,99	46,0	4,8	<0,3	<2
1	NO	KOFASIL	10	BLACK	252	4.42	65,3	18,70	50,8	5,7	<0,3	3.6
2	YES	CONT.	6	WHITE	290	4.77	153,3	6,2	41,5	12,5	9,4	6.0
2	YES	CONT.	6	BLACK	317	4.90	121,4	10,27	20,1	7,7	10,3	4.9
2	YES	KOFASIL	6	WHITE	318	4.75	58,5	30,79	32,9	4,5	<0,3	2,5
2	YES	KOFASIL	6	BLACK	333	4.66	69,9	32,42	34,9	3,6	<0,3	<2
2	NO	CONT.	6	WHITE	342	4.98	110,0	22,47	20,4	13,1	5,7	5.6
2	NO	CONT.	6	BLACK	339	4.94	126,3	15,50	23,3	12,0	12,2	6.4
2	NO	KOFASIL	6	WHITE	334	4.85	71,3	47,03	29,0	4,3	<0,3	<2
2	NO	KOFASIL	6	BLACK	381	5.14	41,1	56,52	18,0	3,4	<0,3	<2

Conditioning was in several cases positive, probably due to a higher DM-content in the bottom of the swath, which made it possible to limit clostridia growth. Effects of conditioning were more pronounced in combination with 1 days wilting than if the crop was wilted two days. The effect could have been more pronounced in combination with two days wilting if the crop had been conditioned day 1 instead of day 2.

Mean temperatures were 3°C higher on the surface and 4°C higher at 10 cm in black bales compared to white. The differences in hygienic quality were small and not consistent. Higher ambient temperatures and more sunshine could have resulted in larger differences between film colours. Increased number of layers did not have much effect.

Differences between depths were difficult to evaluate, since this is dependent on the conditions in the swath such as variations in the field as well as an increased risk of oxygen leakage and / or higher temperature closer to the surface.

The effect of the additive was clear and positive. Only bales treated with Kofasil Ultra have fulfilled the requirements of well-fermented silage. Bales without Kofasil Ultra contained large numbers of spores, and were often rich in butyric acid and ammonia.

Conclusions

- **To avoid clostridia growth in bale silage made from a crop treated with farmyard manure, an appropriate silage additive is required.**
- **Conditioning of the crop limited clostridia growth in several cases.**
- **The application of Kofasil Ultra inhibited clostridia growth very efficiently.**
- **Differences in hygienic quality between bales with different film colours were small.**

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Combinations of biological and chemical silage additives

Key words: *Clostridium tyrobutyricum*, yeast, silage, biocontrol, lactic acid bacteria, sodium benzoate

Object of the study
The aim of the study was to investigate the inhibitory effect on growth of clostridia and yeast in silage by two lactic acid bacterial (LAB) strains in combination with a commercial biological silage additive. The goal was to improve the commercial additive, through combinations of LAB with or without sodium benzoate.

Introduction
Silage production often relies on the fermentation ability of the naturally occurring LAB. Numbers of epiphytic LAB are, however, usually small on crops. To ensure a sufficient number of fast growing LAB, inoculants consisting of LAB suited for ensiling purposes (with or without enzymes) may be added to the crop. Provided the concentrations of readily available carbohydrates of crops are sufficiently high, LAB inoculants results in a rapid pH drop and a homolactic fermentation. To improve bacterial inoculants, emphasis should be made to find LAB strains that inhibit clostridial growth and prolong the storage stability of silage. Addition of a commonly used food preservative, sodium benzoate, also could be useful.

Material and methods
Five experiments were carried out in 1997 in order to evaluate silage additives based on LAB under different conditions. Combinations of different LAB-strains and chemical products that might encourage lactic acid fermentation, inhibit clostridial growth and increase storage stability of the silage were used. Two of the LAB-strains tested in the study had *in vitro* proved to repress clostridial growth (Sr3.54) and yeast-growth (Si3), respectively.

Mineral fertilised grass swards and clover-grass swards were harvested at first cut and second cut. The grass swards were direct cut and precision chopped. Clover-grass swards were both direct cut and wilted, and either precision chopped or long cut. Before application of additives, all crops were infected with a suspension containing clostridia (*Clostridium tyrobutyricum*, $3x10^2$ (g FM)$^{-1}$), yeast (*Pichia anomala* and *Candida lambica*, 10^2 (g FM)$^{-1}$) and mould (*Penicillium roqueforti*, $0.2x10^2$ (g FM)$^{-1}$. The commercial product Lactisil 100®, consisting of two strains of *Lactobacillus plantarum* and two strains of *Pediococcus acidilacti* plus cellulase (Medipharm) added at a rate of 10^5 bacteria (g FM)$^{-1}$, was used as a base in all the inoculant treatments. Untreated herbage and herbage treated with formic acid (FA) were used as controls. Treatments with Lactisil 100® supplemented with Sr3.54 (*Lactococcus*) at two rates (10^5 or 10^4) or Si3 (10^5;) or with Sr3.54 (10^5) and 200 or 400 g sodium benzoate (t FM)$^{-1}$ were tested. Treated herbage was filled in experimental silos (nine per treatment) to a density of 150 kg DM m^{-3}, and three of the silos were opened after 3, 8 and 100 days of fermentation. Silages were analysed both chemically and microbiologically, and aerobic stability were measured.

Results
The DM concentrations at ensiling of the different experiments varied between 190 to 288 g kg^{-1} in direct cut crops and between 350 to 394 g kg^{-1} in wilted crops. The concentration of WSC was approximately 85 g (kg DM)$^{-1}$, or 25 g (kg H_2O)$^{-1}$ in direct cut crops and 50 g (kg H_2O)$^{-1}$ in wilted crops at first cut. At second cut the concentration of WSC was 14 g (kg H_2O)$^{-1}$ in clover-grass crop and 30 g (kg H_2O)$^{-1}$ in grass crop. The buffering capacity of the crops ranged between 248 and 343 mekv OH (kg DM)$^{-1}$.

Silage from direct cut grass sward at first cut (250 g DM) was significantly improved by inoculation of Lactisil 100® plus Sr3.54, while further addition of sodium benzoate reduced the

clostridial effect of the inoculant. At second cut, the clostridial activity instead to some extent was inhibited by the sodium benzoate in the silage from direct cut grass sward (290 g DM).

At first cut, all types of LAB inoculants encouraged homolactic fermentation and inhibited clostridial activity in silages from direct cut (210 g DM) precision chopped clover-grass swards, while FA reduced silage quality. If wilted, differences between treatments were small. FA restricted fermentation and LAB inoculants encouraged lactic acid fermentation. At second cut, all silages from direct cut (190 g DM) precision chopped clover-grass swards were of high quality, and treatments with LAB inoculants only slightly improved silage quality.

Ensiling of long cut clover-grass sward was difficult. Control silage from both direct cut and wilted crops were of poor quality. The additives improved chemical quality of silages to some extent, but the clostridial activity was not repressed, specially not in the wilted material.

Table 1. Number of clostridial spores (log cfu (g FM)$^{-1}$) in silages made from precision chopped, direct cut grass (A; 25% DM and E; 29% DM) or clover/grass crops (B_1; 21% DM and D; 19% DM), precision chopped wilted clover/grass crops (B_2; 39% DM), long cut unwilted clover/grass crops (C_1; 21% DM) and long cut wilted clover/grass crops (C_2; 35% DM).

Treatment	A	B_1	B_2	C_1	C_2	D	E
Control	3.3	<2.0	<2.0	5.9	5.6	<2.0	4.8
FA	5.5	5.0	2.5	3.5	5.3	<2.0	4.7
Lactisil A	2.4	<2.0	<2.0	2.7	5.9	<2.0	4.3
Lactisil B	2.3	<2.0	<2.0	5.4	6.6	<2.0	3.8
Lactisil A 200NB	6.5	<2.0	<2.0	4.7	5.0	<2.0	4.2
Lactisil A 400NB	5.8	2.2	<2.0	3.8	5.1	<2.0	4.5
Lactisil A Si	-	-	-	-	-	<2.0	3.2
$LSD^{0.05}$	*2.13*	-	-	*0.83*	*0.57*	-	*2.22*

Table 2. Storage stability (number of days with no CO_2 production) of silages made from precision chopped, direct cut grass (A; 25% DM and E; 29% DM) or clover/grass crops (B_1; 21% DM and D; 19% DM), precision chopped wilted clover/grass crops crops (B_2; 39% DM), long cut unwilted clover/grass crops (C_1; 21% DM) and long cut wilted clover/grass crops (C_2; 35% DM).

Treatment	A	B_1	B_2	C_1	C_2	D	E
Control	5	3	4	1	1	5	4
FA	>6	>6	5	4	1	>6	4
Lactisil A	4	4	1	4	5	5	3
Lactisil B	4	4	2	5	>	5	4
Lactisil A 200NB	>6	5	4	6	>6	6	2
Lactisil A 400NB	>6	>6	5	>6	>6	6	4
Lactisil A Si	-	-	-	-	-	6	1

Conclusions

- Chemical quality of silages were improved by the use of all biological silage additives.
- Addition of Sr3.54 repressed clostridial growth in silage from precision chopped crops.
- In long cut prewilted silages the number of clostridial spores was not effectively reduced by any additive.
- Storage stability of inoculated silages was prolonged by the addition of sodium benzoate.
- Inoculation of LAB usually encouraged lactic acid fermentation and addition of formic acid restricted fermentation process.

Acknowledgements

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Workshop D
Design and planning of silage experiments

Poster abstracts

Effect of method of conditioning and field wilting on field losses, chemical composition and feeding value of grass silage made in upland United Kingdom.

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Introduction

The majority of the pastures in upland areas overlay highly organic soils or peat. In wet weather, these soil conditions can contaminate grass at mowing and at harvest. Kasper *et al.* (1997) showed that conditioning of grass with nylon brushes increased the rate of water loss from swaths compared to conventional systems under good environmental conditions. Little work has been done with nylon brush conditioners in upland areas of Northern Europe.

Material and methods

The site chosen for the experiment was Malham in North Yorkshire (530 metres above sea level). An improved upland sward (0.65 *Lolium perenne*, 0.2 *Phleum pratense*, 0.1 *Poa trivialis*, 0.05 *Trifolium repens, Festuca* sp. and others) was mown on 3 July 1997 between 14.30 and 15.30 h using one of two mower systems; a Vicon AM 2400 HPC (disc mower with nylon brush roller and ribbed steel conditioning drum, spreading the grass automatically to 1.0 of ground area; treatment HPS) or JF CMT 245 (disc mower with steel V-spoke conditoner and multi-crimp 3 drum system; treatment JF. The swath treatment for the HPC system was achieved by the addition of swath boards (HPW). Treatments JF and HPW had swath widths of 1.2 m. The herbage from each treatment was assessed for DM, WSC, buffering capacity (BC) and ammonia at mowing, and after 6, 17, 21 and 24 h of field wilting. Losses of fresh-weight and DM during wilting were assessed using the method of Wilkinson *et al.* (1996). The grass from all treatments was harvested using a Claas 695 self-propelled forage harvester. The silages were made in triplicate in 0.300 m^3 barrel silos, consolidated during filling by treading and stored for 115 days. Silage was offered *ad libitum* to weaned Mule ewe lambs (6 per treatment, mean live-weight 26.3 kg) for 13 days followed by 6 days of measurement of voluntary intake and digestibility.

Results and Discussion

The greatest change in DM was observed after HPS treatment; indicating that nylon brush conditioning is only effective if the water can leave the herbage mass (Table 1). No difference between JF and HPW were observed for water loss (28.2 and 29.1 g/kg respectively) but grass spread and conditioned showed a threefold increase in loss of water (84.8 g/kg. The level of WSC declined ($p<0.001$) during wilting, but method of conditioning had little effect on the loss of WSC. The concentration of ammonia-N in the grass increased with period of wilting ($p<0.01$) with the highest concentration observed in HPW (19.5 g ammonia-N/kg total N) after 24 h. This indicated proteolysis had occurred in the swath. The low level of ammonia in HPS may reflect the greater exposure of the crop to radiation during wilting of the spread grass. The main differences in chemical composition of the silages were related to the loss of nutrients during field wilting. The lower digestibility (*in vitro*) of HPW reflected the lower concentration of WSC and the higher level of NDF in the silage compared to JF or HPS silages. Proteolysis in the swath prior to harvest in HPW reduced the concentration of amino-N ($p<0.01$), increased the concentration of ammonia-N ($p<0.001$) and accelerated the process of secondary fermentation (an increase in the concentration of butyric acid ($p<0.001$)). Not surprisingly, the voluntary intake of silage HPW was significantly lower ($p<0.001$) than JF or HPS. No difference in the digestibility of DOM was observed between the three silages but a signifcant decrease in the intake of DOM ($p<0.05$) was observed with HPW compared to the other two treatments.

Table 1. Chemical composition of grass (g/kg DM unless stated).

	Conditioner	Duration of wilting (hours)					s.e.d.	
		At mowing	6	17	21	24		
DM (g DM/kg)	JF	135	142	162	182	189	2.52	Cond.**
	HPW		137	156	171	197		Wilt ***
	HPS		159	169	194	209		C x W^{ns}
WSC	JF	301	241	191	179	157	13.16	Cond.^{ns}
	HPW		275	207	185	154		Wilt ***
	HPS		267	188	170	157		C x W^{ns}
BC (mE/kg DM)	JF	349	350	326	312	273	11.25	Cond.*
	HPW		357	333	321	285		Wilt ***
	HPS		328	329	291	294		C x W*
Ammonia (g/kg total N)	JF	8.9	2.2	3.1	6.2	8	3.12	Cond.**
	HPW	19.5	2.1	5.7	9.6	19.5		Wilt ***
	HPS	9.8	2.7	4.2	7.5	9.5		C x W^{ns}

Table 2. Chemical composition (g/kg DM unless stated) and feeding values of silages.

	Conditioning system			s.e.d.	Signif.
	JF	HPW	HPS		
DM content (g DM/kg)	203	208	215	6.26	ns
DOMD *in vitro*	669	663	670	2.97	ns
Total ash	74	73	73	2.62	ns
NDF	488	501	486	6.38	***
Crude protein	119	119	117	4.89	ns
Ammonia - N (g/kg total N)	77	118	74	3.67	***
Amino - N (g/kg total soluble N)	505	479	534	8.01	***
WSC	31	18	34	10.64	ns
pH	3.95	4.52	3.97	0.04	***
Lactic acid	124	82	118	12.28	***
Acetic acid	33	46	34	4.65	***
Butyric acid	1	26	2	6.24	ns
Intake of DM (g/kg LW/day)	27.3	18.2	29.7	3.22	*
Digestibility of DOM	660	641	657	7.22	ns
Intake of DOM (g/kg LW/day)	18	11.7	19.5	3.24	*

Conclusions

Rapid drying of cut herbage was not achieved in this experiment and therefore little restriction on fermentation was observed. Spreading of grass may prevent the formation of ammonia in the swath prior to ensiling and nylon brush conditioning systems may condition grass too much leading to rapid nutrient loss and proteolysis in the swath prior to ensiling.

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Efficiency of wilting of temperate grass crops using high performance conditioning.

Introduction

There are strong environmental and economical arguments in favour of high dry matter silage as wilting to over 30% dry matter content virtually eliminates risk of pollution from silage effluent. In temperate climates rapid wilting of crops is difficult and often leads to high crop dry matter losses (Lamond 1988). Improvements were observed in drying efficiencies of crops wilted in small trays following mowing with flail or conditioner mower compared to unconditioned by Wright (1996). This paper is concerned with the efficiency of field scale wilting of grass crops using a new high performance conditioner mower (HPC) compared with conventional flail conditioner mower (FL) and mowing followed by tedding (FLT).

Materials and Methods

Three 0.8 ha blocks of primary growth of hybrid ryegrass cv Augusta were mown from the same field on 18 May 1994. Three different mower treatments were used; a flail mower conditioner (JF Farer), the same mower conditioner followed by spreading (Fahr Centipede) so that the crop covered 100% of the ground area, and a high performance conditioning mower (HPC, Vicon, UK). This latter mower has integral metal and nylon rollers rotating at high speed to provide extensive conditioning, and a series of deflector plates attached to the rear of the mower for immediate even spreading of the crop over 100% of ground area. Yield of grass at mowing was 3.2 t DM/ha. Speed of mowing was kept constant at 8 km/h for all treatments. Following mowing the grass was wilted for 8 hours. The weather was dull in the morning but moderately sunny in the afternoon, with net solar energy during the day reaching 7.6 MJ/m^2. The average wind speed was 6.7km/h. Three random samples of mown grass each from an area of $0.5m^2$ were taken from within each of the three 0.8 ha wilted treatments at 0930, 1100, 1300, 1500 and 1730 hours. All treatments were raked into rows at 1730 hours and harvested without additive by a precision-chop forage harvester, transported by trailer and ensiled in covered bunker silos. After a 90 day ensiling period the silos were sampled (6 replicate samples per treatment) to their full depth using a 50 mm diameter motorised corer. The data for field wilting (n=45) were statistically analysed using ANOVA, with a treatment structure of Wilting System x Period of wilt.

Results

Significant increases of 11 % and 72 % in the wilting rate (g/kg/h) of the FLT and HPC treatments respectively compared to the FL were recorded during the 8 h period, with significant increases from these treatments appearing after 3.5 h (Table 1a). Significantly higher residual sugars (WSC) and digestibility (DOMD) were observed in the HPC treatment indicating a more rapid wilt with less nutrient losses (Table 1b). As expected significant differences were observed in silage DM content between treatments (Table 1c), and the higher DM content in the HPC treatment appeared to give a restricted fermentation with higher pH values. A significantly lower acetic acid content observed in the HPC treatment would indicate a more homolactic fermentation than the other treatments.

Table 1 (a). *Changes in dry matter content (g/kg FM) of crops mown with high performance conditioner (HPC), flail conditioner (FL) or flail conditioner and tedding (FLT) during an 8h wilting period.*

Time of day	HPC	FL	FLT	s.e.d	Prob.
0930	174	160	171	6.7	
1100	187	166	190	9.6	
1300	220	169	219	11.5	**
1500	255	190	228	19.1	*
1730	267	214	231	6.7	***

(b) *Chemical composition of crops (g/kg DM) after 8 h wilting period.*

	HPC	FL	FLT	s.e.d	Prob.
CP	155	149	157	3.2	
WSC	201	198	185	4.3	*
DOMD	690	678	679	2.8	*

(c) *Chemical composition of silages prepared from the different pre-harvesting treatments (g/kg DM, unless otherwise stated).*

	HPC	FL	FLT	s.e.d.	Prob.
DM (g/kg)	237	177	213	5.0	***
pH	4.35	4.29	4.18	0.05	**
Ammonia-N (g/kg N)	76.5	76.7	65.1	3.60	**
ME (MJ/kg DM)	11.7	11.4	11.6	0.13	
Crude Protein	183.4	182.9	172.1	3.70	*
Lactic acid	79.0	84.9	83.1	6.30	
Acetic acid	27.3	53.9	40.1	6.80	**

Prob = statistical significance at probability levels, * P<0.05, ** P<0.01, *** P<0.001.

Conclusions

Efficiency of DM wilt was 11.6, 7.5 and 6.8 g/kg/h for the HPC, FLT and FL treatments respectively. Stable silages were obtained from all treatments, however HPC treatment gave improved homolactic fermentation with lower acetic acid content.

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Storage control and handling of damp feeds sealed with plastic film

Key words: Silage, wrapped bales, big bag, CO_2, gas pressure, tightness, valve, vacuum lifting, sampler.

Object of study

To present an environmentally-friendly system of improving the overall quality of forage in wrapped bales.

Materials/System

Automatic Release and Back Valve (ARBV)

Silage, air tight storage in silos, bags and bales, CO_2-storage, as well as Modified Atmospheres (MA) and Controlled Atmospheres (CA) are all storage techniques that produce CO_2. Large amounts of gas are produced in the beginning of the storage period, specially for silage. This gas builds up an over- pressure in the silo or bale and will leak through the plastic seal to the surrounding atmosphere. Releasing this gas at low over-pressure as well as avoiding new air (oxygen) to penetrate plastic films (round and square bales, big bags and clamps) is the primary working principle of the *Automatic Release and Back Valve (ARBV).*

Measuring of Tightness

Assuming that the film-sealed storage system is tight enough, the gas pressure inside the packages is higher than of the ambient air during daytime and lower during nighttime. This is due to changes in temperature (sunshine and global radiation). This phenomenon facilitates the second principle of the *ARBV* through measuring the differences in pressure with a pressure gauge. If no difference is measured, this indicates that the bale is not tight and oxygen gets into the package every night, thus increasing forage losses. The tightness of a silo or bale can be quantified by means of the Variable Pressure Test, which determines the "tightness time". In bales an under-pressure of at least -200 Pa is created. The "tightness time" expresses the period of time (s) for the pressure to change from -200 to -150 Pa. The longer the tightness time, the tighter the package. This way the quality of the contents can be documented. *MätHinken* is a "measuring bucket" with a complete set of tools for measuring the tightness.

Vacuum Lifter

Wrapped bales are very easily damaged during handling. A *vacuum lifter* which increases the possibility to reduce handling damages has been developed. The principle is a big cap, used together with the *ARBV*, which produces under-pressure inside the bale. In this way the lifting force is independent of the film itself, i.e. air-pressure underneath the bale presses the load against the lifting cup. Lifting directly after wrapping evacuates some oxygen and leads to better laminated films. Hence, the lifting equipment also aids the preservation process. Hopefully this equipment will be possible to sell later this year (1999).

Sampler (STICKIT)

The fourth principle of the system connected to the *ARBV* is the small, manual sampler *STICKIT* for roughages. This sampler gives small, fast, well-cut and easy-to-post samples for analysis both before and after wrapping. The *ARBV* is used to reseal the wrapped bale after sampling. The sampler, only 16 mm in diameter, 250 mm long and weighing 150 g, can easily be put it in the pocket. Thus, *STICKIT* is more convenient than other currently used methods.

Results/Conclusion

- The use of ARBV results in tighter bales with better hygienic quality and lower losses. This can be seen in Table 1 which shows the results of a study made in Kungsängen in 1995-1996 using bales with whole crop silage. The sample consists of 65 bales with valves and 60 bales without valves.
- There is better tightness after vacuum sucking. This is indicated by tests performed during the development of the vacuum lifter. Table 2 shows the difference in tightness time before and after performing vacuum sucking (to a level of - 4 kPa). Improvement of the tightness is probably due to the layers of film that are better laminated against each other. Moreover, tightness measurements after loading, transporting and unloading after handling with vacuum-lifting also indicated better "tightness time" compared with corresponding handling with a mechanical bale handler.

Table 1. Silage analysis from whole crop silage in 125 bales. Mean values and results of t-tests.

	g/kg	log C F U / g			g/kg DM				
	DM	Clostridia	Yeasts	pH	Crude protein	WSC	Lactic acid	Butane-diol	Butyric acid
65 bales with valve	303	2,6	2,3	4,5	105	15	45	4	7
60 bales without valve	313	2,9	2,8	4,6	102	14	39	5	8
Difference		*0,3*	*0,4*	*0,1*	*3,0*	*0,9*	*6,3*	*1,1*	*0,9*
Standard deviation 65 bales with valve	19	1,2	1,0	0,4	7,7	7,5	11,0	1,9	5,2
Standard deviation 60 bales without valve	21	1,2	0,7	0,4	7,6	6,0	14,2	2,5	4,4
Best significance, %		*77,0*	*99,4* sign**	*79,1*	*97,4* sign*	*55,7*	*99,4* sign**	*99,3* sign**	*73,1*

Table 2. Tightness before and after evacuating with vacuum lifter (to -4 kPa)

Bale no.	Before evacuating Tightness time, s	After evacuating Tightness time, s	After re-sealing bales with holes Tightness time, s
1	39	>>60	
2	42	>>60	
3	5	6	>60
4	21	58	
5	25	123	
6	33	>>60	
7	25	>>60	
8	18	42	
9	60	>>60	
10	<10	<10	>50
11	<10	<10	28
12	28	>60	
13	<10	<10	55
14	24	>60	
15	36	>60	

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Impact of ensiling on cell wall carbohydrates assessed by chemical analysis and ruminal fibrolytic enzyme activity

Object of study

During the storage in silo, soluble carbohydrates and nitrogenous compounds may be extensively fermented to modify the utilisation of forages. The process might also affect the cell walls of ensiled forages, because plant enzymes, microbial activity and acidity could degrade a part of structural carbohydrates. The effect on cell walls, however, has not been fully understood, whereas cell wall digestion would be a primary concern in animals fed forage-based diets.

In this experiment we studied the effects of wilting and formic acid treatment on the composition and the digestion of cell walls of Italian ryegrass silage. Changes in cell walls were assessed by detergent fibre and monosaccharide components, and the digestibility was evaluated by fibrolytic enzyme activities after *in vitro* incubation with rumen microorganisms.

Materials and methods

First growth of Italian ryegrass (*Lolium multiflorum* Lam) was harvested at early heading stage. The herbage was chopped into 13 mm length and ensiled directly or after being wilted to targeted DM contents of 300, 400 and 500 g kg^{-1}, respectively. Formic acid was added to the high moisture wilted herbage (DM 300 g kg^{-1}) at rates of 2, 4 and 8 g kg^{-1}, respectively. Silos (1capacity) were stored for 45 d at ambient temperature.

The contents of neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to the conventional procedures. Starch-free NDF was subjected to acid hydrolysis and the hydrolysate was used for HPLC analysis of cell wall monosaccharides.

In vitro incubation was carried out at 39°C for 24 h using a mixture (1:4) of rumen fluid and artificial saliva. Fibrolytic enzymes were extracted by ultrasonic treatment and then used for assays of carboxymethylcellulase (CMCase) and xylanase activities.

Results and discussion

Fermentation was effectively restricted as the moisture content decreased or the rate of added formic acid increased. The most vigorous fermentation was found in direct cut silage; the pH value was 3.96 and the organic acids were 200 g kg^{-1} DM. The highest level of formic acid

almost completely inhibited the acid production (only 18.6 g kg^{-1} DM total acids in the silage). The proportion of lactate to total acids was over 0.8 and no butyrate was found in any silages.

The contents of NDF, ADF and ADL were lower in silages than those in fresh material and the reduction of NDF was related to enhanced organic acid production. Compared with cellulose, hemicellulose was more susceptible to ensilage and the reduction averaged 0.25 of initial content based on detergent analysis. Monosaccharides analysis indicated that arabinose was more affected than xylose and cellulose. The reduction of xylose and arabinose was not related to the extent of fermentation, while that of cellulose was decreased as the level of DM increased. The content of cellulose was always higher when determined by the detergent system than by monosaccharides analysis, while the effect of ensilage was similar regardless of analytical procedure.

Both CMCase and xylanase activities were lower in silages incubated *in vitro* than in the fresh material, and the reduction was greater with xylanase than CMCase. The extent of fermentation did not affect xylanase activity either in naturally-fermented or acid-treated silage, while the effect of wilting was quadratic on CMCase activity.

It can be concluded that the ensiling process may lower the cell wall digestibility of Italian ryegrass silage, although the effect appeared to be more pronounced when judged by detergent analysis. The extent of fermentation may have only a minor influence on the cell walls of silages, while the effect on monosaccharide components was variable.

Results of naturally fermented Italian ryegrass silage

Item	Direct cut	Wilted			SE	Contrast
		HM	MM	LM		
pH	3.96	4.27	4.51	4.71	0.06	L
Total acids (g kg^{-1} DM)	200	101	80.8	61.0	10.6	L,Q
Hemicellulose (g kg^{-1} DM)	138	137	138	151	9.32	NS
Cellulose (g kg^{-1} DM)	249	255	262	269	4.47	L
Xylose (g kg^{-1} DM)	95.6	105	101	105	3.89	NS
Arabinose (g kg^{-1} DM)	10.8	15.4	14.3	15.3	1.78	NS
Cell wall glucose (g kg^{-1} DM)	219	222	235	250	4.29	L
CMCase (mol RS g^{-1} DM h^{-1})	48.0	43.6	44.9	46.2	0.55	Q
Xylanase (mol RS g^{-1} DM h^{-1})	152	145	144	146	4.93	NS

HM, high moisture; MM, medium moisture; LM, low moisture; RS, reducing sugar; L, linear effect; Q, quadratic effect.

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Certification of silage additives in Germany by DLG.

Introduction

The DLG approval scheme for silage additives has proved its importance for practical farmers and industry since its introduction in a revised form 8 years ago.

It gives the farmer an overview of the increasing and diversifying market and makes it possible for him to select the additives according to the specific requirements on his farm. The annual control of the product by off-the-shelf samples guarantees that the product has not been changed.

For Industry the impartial evaluation is an important argument in merchandising the product.

Nevertheless, based on the experience of the past few years it has been necessary to modify the evaluation frame in some points, especially the more precise description and definition of the four target crop groups for the category "improving fermentation".

The test scheme

The testing scheme has to take into account that there are at the moment three different *"aims of action"*, directly related to the ensiling process which cannot all be covered by one single additive.

I Improving the fermentation process
II Improving aerobic stability
III Reducing effluent.

Group I has to be split up further for different *target crop groups*, requiring different additive approaches.

To better describe the fermentability of the forages, the "fermentation coefficient" (FC), defined by WEISSBACH and HONIG (1996) is used. It combines substrate availability, buffering capacity and DM content of the crop in one figure.

IA Forages, *difficult to ensile:*
insufficient fermentation substrate and/or too low in dry matter;
characterised by FC <35

IB Forages, *moderately difficult or easy to ensile in the lower DM range*:
grasses, legumes, maize, whole crop cereals with sufficient fermentable substrate characterised by FC ≥35 and DM ≤35 %

IC Forages, *moderately difficult or easy to ensile in the higher DM range*:
grasses, legumes, maize, whole crop cereals with sufficient fermentable substrate characterised by FC ≥35 and DM >35 % up to about 50 % (insufficient water availability limits efficacy above this DM content)

ID *Special Forages*
Forages requiring special additive effects (as fodder beets, pulps, sugar beet pulps) or forages for which additives are specially designed

In addition to the approval in at least one of the groups I to III *nutritional* and *other effects* can also be claimed and may be attained in two further groups:

IV Improving nutritive value and animal performance
V Additional effects

Group IV is split up into 3 subcatagories

IVA Improving feed intake
IVB Improving digestibility
IVC Improving performance, stated according to animal species or type of production, e.g. milk production or fattening sheep or cattle

Group V has one subcatagory till now:
VA Prevention of clostridial development in the silage.

Further categories may follow if they are urgently required.

The testing procedure

The DLG Quality Seal is managed by the DLG Certification Unit (DLG-CU) which is accredited according to standard EN 45011 since 1996. To run the necessary tests the DLG-CU co-operates with laboratories which satisfy the requirements of the standard EN 45001. To be awarded with the DLG Quality Seal first has the composition of the additive to be described, especially the active ingredients. Bacterial strains must be registered. Secondly the correctness of the declared composition has to be proved by an analytical test. Thirdly the additive must have shown an improvement compared to an untreated control in five independent experiments with different forages of the claimed target group. For group I the parameters to be judged are:

- course of fermentation
- fermentation pattern
- losses.

Tests are normally carried out on laboratory scale with three replicates over a 90 up to 120 days ensiling period. Other experiments, meeting the standards of sound scientific practice, may be accepted as well. This is especially valid for groups IV and V. To be awarded in category IV additives must have shown an improvement to an untreated control in at least three independent experiments with animals. The experimental design, the length of the test period, the number of animals and the statistical analysis must allow for the exclusion of other effects except caused by the additives. The degree of improvement has to be significant for practical conditions (e.g. at least plus 5 % feed intake, plus 1,5 % organic matter digestibility, plus 1,0 kg milk yield or plus 5 % weight gain).

The decision on the approval is made by a review committee of seven experts. All data provided are strictly confidential, especially in cases, where the additive has not been approved.

Response

Since its revision in 1991 the evaluation scheme has found high response. By January 1999 50 brands/formulations, provided by 21 producers or distributors, have been awarded with the DLG Quality Seal for Additives. Many of them are acknowledged for two or more aims of action. The availability of approved silage agents according to the different aims of action is shown in table 1.

Table 1 DLG approved silage agents according to aim of action.

Category		A	B	C	D
I	Improving fermentation	4	40	33	-
II	Improving aerobic stability	9			
III	Reducing effluent	-			
		feed intake	digestibility	milk yield	weight gain
IV	Improving nutritive value	10	20	9	8
V	Additional effects Inhibition of clostridia	3			

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Methods for studying the ecology of lactic acid bacteria populations in whole plant corn silage.

Introduction

Silage is produced by the induced fermentation of various crops by lactic acid bacteria (LAB). Since the number and type of epiphytic lactic acid bacteria can vary widely by crop and location, silage inoculants containing LAB are frequently applied to forage to improve fermentation. In the development of forage inoculants, it would be useful to understand the interactions between microbial populations in silage in order to understand why certain epiphytic or inoculant LAB dominate a fermentation and produce either good or poor silage.

Carbohydrate fermentation can vary among species of LAB as well as different strains of the same species. We have developed a procedure for surveying the dominant lactic acid bacteria in silage by using physiological differences to make a tentative identification, followed by DNA fingerprinting to identify individual strains. By knowing the dominant population in a silage sample we hope to understand how different strains affect fermentation and aerobic stability. This method can also be used to monitor the presence of inoculated strains.

Materials and Methods

Field trials were done in 1995 and 1996 at the Pioneer Livestock Nutrition Center (Polk City, IA) with whole plant corn ensiled in PVC mini-silos. The same location and the same hybrid were used for both experiments. Silage was either treated with Pioneer® Brand 1132 silage inoculant or left untreated (control). Silos were opened at intervals and pH, bacterial and yeast counts, and aerobic stability were determined. Colonies (96) from each treatment were randomly picked from MRS plates and stored frozen in MRS/glycerol in microtiter plates for later analysis. Each colony was subsequently tested for gas production in MRS broth, cell morphology, and acid production in alfalfa and whole plant corn extract broth. In addition, approximately 32 isolates from each treatment were randomly selected for DNA analysis.

Initial classification of strains was based on gas production in MRS broth and cell morphology. Acid production (pH) of each strain in alfalfa extract broth and whole plant corn (WPC) extract broth was normalized to a standard strain of *Lactobacillus plantarum*. When normalized pH values in alfalfa broth were plotted against normalized pH values in WPC broth, isolates of the same genus and species clustered with type strains on the scatter plots and allowed tentative identification to genus (and sometimes to species). Identification of individual strains was done by restriction analysis of total DNA digested with EcoR1. Patterns of restriction fragments were scanned and profiles analyzed with GelCompar software (Applied Maths). Strains of

the same genus and species often clustered together, and were considered identical if their profiles had a greater than 90% similarity.

Results

We surveyed the LAB populations in whole plant corn (WPC) silage in 1995 and 1996 after 0, 2, 4 and 90 days ensiling. The populations changed during the ensiling period and were different for each of the 2 years of sampling, even though the location and hybrid were the same. While the numbers of lactic acid bacteria were comparable between years in terms of CFU/g, the composition of the populations was different between the two years. Homofermentative strains were dominant in the 1995 forage and heterofermentative strains were dominant in the 1996 forage. In 1995, product strains from Pioneer brand 1132 silage inoculant dominated the fermentation and could be detected 90 days after fermentation. In 1996, product strains were not dominant in the fermentation, although they could be detected in the silage. Many different strains were identified at each sampling time, and at least 10 different LAB species belonging to at least 5 different genera were identified. Within each species, multiple strains could be identified by DNA profiles. In these two trials, there was not a clear relationship between a particular bacterial population and either pH or aerobic stability.

Conclusions

We have developed methods to identify LAB strains in silage and follow them over time. While the method is time-consuming, we found that it was useful for surveying population shifts over time and for tracking our inoculant product strains. It was clear from this work that it is not enough to look at LAB populations simply in terms of CFU/g on MRS plates, since microbial populations can be completely different between samples with the same counts. We may be able to correlate such information on LAB populations with silage characteristics in order to select better inoculant product strains. Further work is needed to develop more rapid methods for looking at changes microbial populations in silage, and to extend the methods to non-lactic acid bacteria, yeast, and mold.

Selection and Application of Excellent Lactic Acid Bacteria for Silage Preparation

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The fermentation heating in the silage was consistently correlated with microorganism development and plant respiration. The temperature rose rapidly only in the early stage of the ensiling processes and could reach above 45•. In addition the growth of most lactic acid bacteria (LAB) would be inhibited by the high-temperature conditions and the resulting silage will be of poor quality. Therefore, the type and size of LAB in the silage environment may be need to be controlled. In the present study, three LAB strains isolated from forage crops were selected for ability to grow at high temperature and at low pH conditions, and their characterization and application for silage preparation were studied.

MATERIALS AND METHODS

Alfalfa (*Medicaco sativa*) and Italian ryegrass (*Lolium multiflorum*) were harvested at the flowering stage in August 1996. Silages were prepared using a small scale system of silage fermentation (Cai *et al.* 1998). Three LAB strains, *Pediococcus acidilactici* LA3 and LA 35, *P. pentosaceus* LS 5 and SL 1 (*Lactobacillus casei*, isolated from a commercial inoculant) were used as additives. The inoculum size of LAB was $1.0x10^5$ cfu g^{-1} of fresh matter basis. The silos were kept at 25• and 48•, and the chemical composition and fermentation loss of the silages were analysed after 60 d of storage.

RESULTS AND DISCUSSIONS

Three isolates were gram-positive and catalase-negative tetrad cocci that did not produce gas from glucose, formed approximately equal quantities of L(+) and D(-) lactic acid and were able to grow at low-pH (3.5) and high-temperature (>45•) conditions. When stored at 25•, all LAB-inoculated silages were well preserved, and exhibited significantly ($P<0.05$) reduced fermentation losses than their control in alfalfa and Italian ryegrass silages. When stored at 48•, strains LA 3 and LA 35-inoculated silages were also well preserved, with a significantly ($P<0.05$) lower pH, butyric acid, ammonia nitrogen content, gas production and DM loss, and significantly ($P<0.05$) higher lactate content than the control, but LS 5 and SL 1-inoculated silages were of poor quality and showed similar levels of these components to control silages in the two kinds of silage.

TABLE 1. Fermentation quality of silage ensiled at 25• or 48• for 60 d*

	Alfalfa silage					Italian ryegrass silage				
	Ut	LA 3	LA 35	LS 5	SL 1	Ut	LA 3	LA 35	LS 5	SL 1
25•										
pH	4.85	4.40	4.20	4.50	4.15	4.60	4.15	4.14	4.20	4.00
DM (% FM)	19.62	20.16	20.21	20.20	20.24	20.70	21.20	20.50	20.34	20.80
Lactic acid (% FM)	0.42B	0.94A	1.01A	0.89A	1.05A	0.85B	1.49A	1.65A	1.42A	1.60A
Acetic acid (% FM)	0.60	0.50	0.50	0.55	0.53	0.21	0.20	0.27	0.18	0.25
Butyric acid (% FM)	0.63A	0.06B	nd	0.10B	nd	0.1A	0.04B	0.02B	0.06B	0.01B
Propionic acid (%FM)	0.23	0.00	nd	0.00	nd	0.02	0.02	0.01	0.01	nd
Ammonia-N (% DM)	0.66A	0.45B	0.38B	0.46B	0.40B	0.47A	0.28B	0.32B	0.36AB	0.25B
48•										
pH	5.10	4.76	4.37	5.10	5.00	4.82	3.77	3.85	4.85	4.80
DM (% FM)	19.33	19.87	20.00	19.96	19.30	20.50	22.20	21.60	20.15	20.40
Lactic acid (% FM)	0.31B	0.74A	0.80A	0.35B	0.38B	0.50B	1.03A	0.98A	0.53B	0.62B
Acetic acid (% FM)	0.29	0.20	0.18	0.30	0.27	0.15	0.10	0.13	0.12	0.20
Butyric acid (%FM)	0.78A	0.13B	0.12B	0.70A	0.75A	0.45A	0.02B	0.04B	0.31A	0.39A
Propionic acid (% FM)	0.02	nd	0.01	0.02	nd	0.02	nd	0.02	0.01	0.02
Ammonia-N (% DM)	0.72A	0.56B	0.48B	0.78A	0.75A	0.32A	0.17B	0.19B	0.27A	0.30A

*Values are means of three silage samples. Means in the same silage row with different superscripts are significantly different ($P<0.05$). Ut, Control; DM, dry matter; FM, fresh matter; nd, not detected.

The strains LA 3, LA 35, LS 5 and SL 1 used in this study were homofermentative LAB which could grow well at 25• and at low-pH (3.5) conditions. Inoculation with these LAB at 25• may result in beneficial effects by improving silage quality. The strains LA 3 and LA 35 could grow at 50•, but strains LS 5 and SL 1 did not grow at this temperature and may die above 45•. The strains LA 3 and LA 35 improved silage quality at the high-temperatures. While strains LS 5 and SL 1 were unable to grow and ferment WSC to produce sufficient lactic acid resulting in the pH value of silage not palling to less than 4.2, and so allowing the butyric acid fermentation by clostridia to occur.

The results confirmed that *P. acidilactici* LA 3 and LA 35 were considered suitable as potential silage inoculats, and they were more effective in improving silage quality than *P. pentosaceus* LS 5 and inoculant strain *L. casei* under high-temperature (48•) conditions.

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In-Vitro-Test System for the Evaluation of Fermentation Characteristics of Plant Material and for Examining the Efficiency of Biological and Chemical Additives

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keywords: silage-making, lactic acid bacteria, rapid fermentation test, osmotic tolerance

INTRODUCTION

The occurring difficulties with the production of well-preserved silage lead to a wide use of chemical and biological additives. In practical silage-making biological additives are frequently used based on lyophilized bacteria. While using these additives the variability of the fermentation conditions must be considered. Some of these conditions are simulated by the introduced tests. They show the possibility to investigate the interactions between

- plants with different contents and varieties of fermentation substrates and secondary plant contents,
- different and changing epiphytic micro-organisms of plants and
- effects of silage additives under different fermentation conditions.

MATERIAL AND METHODS

The represented tests are rapid methods, developed according to the rapid fermentation test by Pieper *et al.* (1996). The aim of this test is to assess the silage potential of herbage with the help of aqueous plant mixtures. The principle of the used method is the measure of the pH-value in aqueous suspension after a certain time regime and the analysis of the fermentation products in the filtrates of incubation mixtures. This method is very simple and rapid in its use and shows good reproductive results. Positive reasons for the use of these aqueous mixtures as a model for silage-making are:

- The process of acidification under aqueous conditions is caused by lactic-acid production in the first place.
- It is possible to work under anaerobic conditions because of the little penetration depth and solubility of oxygen in water.
- An increased osmotic pressure (can be simulated by the addition of osmotic effective salts similar to the wilting of herbage).
- The composition of the aqueous plant mixtures can be changed by additives.
- The fermentation temperature can be varied.

The single tests make it possible to give statements about the following problems according to practical silage-making:

1. determination of the acidification activity of the native epiphytic micro-organisms,
2. determination of the efficiency of lactic-acid bacteria inoculants and their use under different fermentation conditions (supply of fermentation substrates, temperature, water availability, native epiphytic micro-organisms),
3. determination of the acid-tolerance of lactic-acid bacteria (LAB),
4. occurrence of plant specific contents inhibiting fermentation,
5. amount of water soluble carbohydrates (WSC) and buffering capacity of plant material,
6. increase of extraction of fermentable substrates by additives (e.g. amylase, cellulase),
7. efficiency of biological and chemical additives for the improvement of anaerobic/aerobic stability.

Results are shown by examples, which deal with the evaluation of silage characteristics of herbage and with the examination of the efficiency of biological additives during reduced water activity (Test 2). The used herbage were silage maize (37% DM) and alfalfa (17% DM) with an addition of 2% saccharose. 50g of the herbage were mashed, homogenised and mixed with 200ml distilled water and differently concentrated NaCl solutes (0,44 mol; 0,87 mol; 1,30 mol; 1,73 mol - including the plant water). After that homogenised samples were incubated at 35 degrees Celsius. The pH-values were measured after 0, 14, 18, 22, 26, 38 and 46 hours and after that the lactic acids, volatile fatty acids and ethanol were determined. The native epiphytic micro-organisms (untreated control) and two LAB were examined (A / B 10^6 cfu/g FM).

RESULTS AND DISCUSSION

Table 1 shows the pH-value after 46 hours and the corresponding contents of lactic acids. Electrolytic solutes influence the degree of dissociation of organic acids. That is why it is necessary for the examination of fermentation efficiency to consider not only the pH-value but also the produced acids.

Table 1. Influence of the water activity on the metabolic efficiency of LAB.

pH-value after 46 h and lactic acid production (% DM) - pH / lactic acid					
	0 mol NaCl	**0,44 mol NaCl**	**0,87 mol NaCl**	**1,30 mol NaCl**	**1,73 mol NaCl**
maize					
untreated control	3,5 / 5,8	3,3 / 5,6	3,5 / 3,1	4,3 / 0,8	4,9 / 0,0
inoculant A	3,3 / 9,5	3,0 / 8,9	3,1 / 6,5	3,6 / 2,7	4,9 / 0,0
inoculant B	3,3 / 8,4	3,1 / 8,4	3,1 / 6,0	3,3 / 4,1	4,8 / 0,0
alfalfa + 2% saccharose					
untreated control	3,8 / 13,5	3,7 / 12,7	4,3 / 5,2	5,0 / 0,0	5,0 / 0,0
inoculant A	3,7 / 16,5	3,5 / 15,5	3,5 / 13,0	5,0 / 0,3	5,0 / 0,0
inoculant B	3,6 / 15,7	3,5 / 15,7	3,5 / 14,3	3,9 / 7,7	5,0 / 0,0

The following statements are made:

1. The native LAB and the silage potential from various herbage are different.
2. The metabolic efficiency of LAB is decisively influenced by the water activity.
3. A higher lactic-acid production was achieved by the inoculated treatments.
4. While reducing water activity the lactic production was clearly reduced, too.
5. In the 1,73 mol NaCl solute an acidification did not take place.
6. The inoculant B was under hight osmotic pressure more effective than the inoculant A.

CONCLUSIONS

The achieved results are not always new but similar to those found in the literature.
Because of the logic of these results it can be concluded that the efficiency of this test system is able to solve the named aims.

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Design of silage experiments to study fermentation kinetics

Introduction

Silage of good end-point quality is required if we are to reduce concentrate inputs and optimise the nutrition of silage-fed ruminants. However, to understand the processes that determine the end-point quality we need to study the kinetics of fermentation characteristics that play a role in determining quality parameters. As fermentation progresses, with or without the application of appropriate additives, the fermentation characteristics (*e.g.* pH, lactic acid VFA concentrations) and quality parameters (digestibility and ammonia - N, carbohydrate and protein content) follow various patterns. In order to model these profiles we need sufficient degrees of freedom to fit an empirical or deterministic model and test its lack-of-fit. The statistical design of a given silage experiment must generate sufficient replication in order to clearly define each phase of the fermentation profile and thus facilitate modelling. If the relevant model is known an optimum design can be identified (Box and Lucas, 1959), but this is rarely the case and we must cater for models with variable numbers of parameters. The ensilage process has three main phases (a) the lag-phase where the initial pH of the forage mass barely changes (b) a rapid decline in pH as a consequence of exponential microbial (mainly lactic acid bacteria) growth with lactic acid and VFA production, which is modified by forage factors such as buffering capacity, and (c) the stable pH and lactic acid levels that give an indication of final quality. Quality indicators such as ammonia, protein and carbohydrate fractions etc. also change and attain their asymptotic levels. We have used an example of inoculant treated silage to demonstrate fermentation progression in terms of pH decline and increase in the concentration of lactic acid (Merry, Dhanoa and Theodorou, 1995).

Materials and Methods

A first re-growth of perennial ryegrass was mown, chopped and treated with a strain of *Lactobacillus plantarum* at an inoculation rate of 10^6 colony forming units g^{-1} fresh matter and used to prepare 21 replicate silages (approximately 1 kg) in glass laboratory silos. Three replicates were destructively sampled after 0.7, 1.5, 2.5, 5.5 and 14 days of incubation at 20-25°C and representative samples taken to prepare water extracts for the measurement of pH values and lactic acid concentrations.

Results

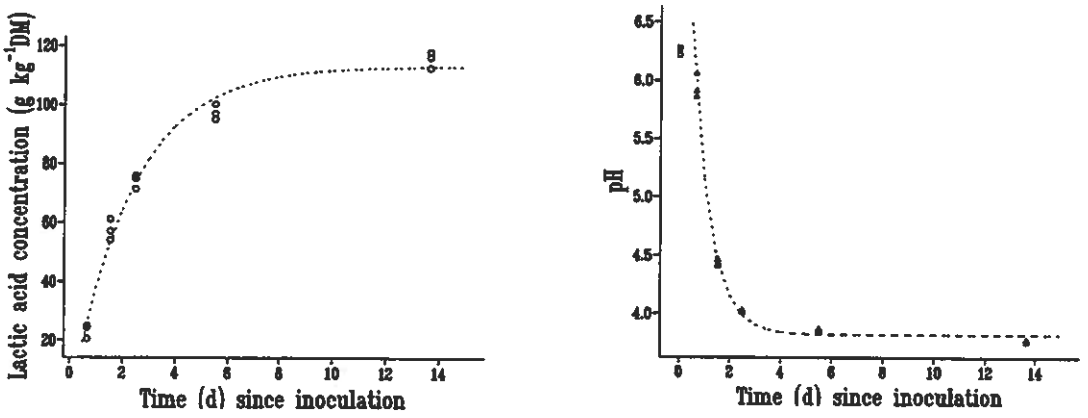
In Figures 1a and 1b respectively the decline in pH and rise in lactic acid concentration, in inoculant treated silage is shown. The starting herbage pH of 6.26 [□] was reduced to a final pH of 3.8 within one week. There is evidence of a short lag phase but insufficient sampling times do not allow its identification during modelling. Similarly, the rise in lactic acid concentration followed a well-defined pattern. The profiles for pH and lactic acid can be modelled using the Mitscherlich or simple exponential model:

$$y = a + b\,(1 - e^{-ct})$$

Where c is a rate parameter, t is time, a and b the model parameters and y is any quality indicator.

Fig. 1 Changes in lactic acid concentration and pH during ensilage of inoculant treated perennial ryegrass.

a) b)



The R^2 values for pH and lactic acid for the fit of the above equation were 0.997 and 0.978 respectively. In the case of lactic acid the lack-of-fit test showed significant departure of mean values from the fitted curve, mainly due to very small variability among replicate silos.

In this study there were only 4 degrees of freedom available to model these patterns, making it difficult to attempt more complex modelling *e.g.* growth functions. In our experience we need to plan for four parameter models. With the 2 or 3 degrees of freedom needed for lack-of-fit, we should aim for 6 or 7 total degrees of freedom for the fitting of a model. If resources are limited then priority should be in favour of maximum possible time points even if replication has to be reduced at each opening time. For example, in the present case we might have benefited from more opening times by reducing replication from 3 to 2 at each time point. After the fit of an appropriate model the lack-of-fit variation may be pooled with replication (or pure error) variation to provide more error degrees of freedom for the calculation of confidence intervals of estimates of model parameters. Alternatively, re-sampling method of Bootstrap (Efron and Tibshirani, 1993) may be used to construct confidence intervals.

Conclusions

We have found that kinetic modelling as used in this study helps to understand the mode of action of experimental silage treatments. The end-points may be the same but the speed and manner of attaining them gives considerably more information about the potential activity of the additive or inoculant under study.

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The effects of timing of biological additive application on wilted grass silage

Introduction

Micro-organisms growing as epiphytes on grass forage are greatly responsible for fermentation processes occurring during ensiling. Once cut and chopped the amount of lactic acid bacteria, enterobacteria, yeasts and clostridia rapidly increases. The time between harvesting and ensiling gives epiphytes an advantage over bacterial inoculants, particularly if grass is wilted. The dominant growth of natural epiphytes during early ensiling may also have an important role in aerobic deterioration. This study evaluated the application method of biological additive (simultaneously during cutting, chopping or ensiling) and extent of wilting on silage quality and aerobic stability.

Materials and methods

A series of two *Phleum pratense-Festuca pratensis* ensiling experiments were conducted on August 7 1996 (I) and June 17 1997 (II) in 400 l cylindrical silos for 126 and 153 days, respectively. In both cases the application of inoculant (I, *Lactobacillus plantarum*, $5*10^6$ cfu/g, Valio Ltd, Finland) was made in triplicate silos in three different ways; directly on the cut sward (IC), during precision-chop harvesting (IH) and in the silo (IS). Grass preserved with either none or a formic acid based additive (FA) applied at a rate of 5 l/t (800 g/kg CHOOH) during silo filling were used as controls. Forage was wilted for 3 h in Exp.I (dry matter content (DM) 279 g/kg) and either for 3 h (DM 230 g/kg) and for 6 h (DM 274 g/kg) in Exp.II. Chemical composition and microbiological parameters were assessed from both the raw material and silage.

Results

Due to prolonged sunshine and warm weather in the first experiment, grass DM after 3 h wilting was much higher than in the second. Success of ensiling in Exp.I was also influenced by a higher amount of water soluble carbohydrates (WSC) (122 vs. 62 g/kg DM). The effect of wilting time was negligible, and had only a minor effect on the proportion of soluble nitrogen and WSC content. Reflecting the extent of fermentation, the concentration of WSC was higher, and lactic acid lower in FA silage than other silages, the effect being more profound with higher DM in Exp.II (interaction $P<0.05$) (Table 1). However, larger differences were observed between inoculant application techniques. The concentration of ammonia in FA silage was lower than that of untreated or IS silage but was higher than in IC and IH. Inoculant clearly decreased silage pH in IC and IH as compared with untreated silage. With a lower DM content in Exp.II pH was the same in untreated and IS silages (interaction $P<0.05$). The quality of IS was poorer also in terms of higher amounts of propionic and butyric acid as compared with other I silages, especially in low DM silage ($P<0.05$). This was associated with higher numbers of clostridias and yeasts (Table 2). Formic acid treated silage was found the most stable in terms of aerobic stability.

Conclusions

Lactobacterial inoculant produced a better quality silage compared to untreated material, but only when it was applied soon after cutting or during chopping. If inoculation was conducted during silo filling, the natural grass epiphytes maintained an advantage over the bacterial

inoculant resulting in untreated and inoculated silages relatively equal in terms of fermentation quality.

Table 1. Silage fermentation quality

	DM g/kg	pH	Ethanol g/kg DM	WSC g/kg DM	Lac.acid g/kg DM	Ac.acid g/kg DM	But.acid g/kg DM	Amm.N g/kg N
Experiment I								
NA	262	4.24	10.3	22.6	93.5	26.8	0.09	59
FA	265	4.17	9.6	50.1	66.0	23.6	0.14	50
IC	292	4.14	6.3	38.5	105.7	17.8	0.08	33
IH	274	4.12	5.9	32.3	99.7	19.2	0.10	33
IS	260	4.20	9.6	28.7	102.0	24.9	0.09	57
SEM	1.5	0.014	1.36	4.92	5.32	1.38	0.04	2.49
Experiment II								
3 h wilting								
NA	225	4.47	7.6	15.3	63.6	19.5	10.3	108
FA	231	4.09	8.9	29.6	74.5	12.8	1.2	71
IC	240	4.12	3.8	21.2	94.2	15.3	0.0	59
IH	220	4.08	3.9	25.1	92.7	13.8	0.0	53
IS	232	4.46	6.9	15.6	71.3	15.5	11.4	94
6 h wilting								
NA	252	4.12	5.7	31.0	84.3	15.1	1.0	71
FA	253	4.02	6.5	52.4	49.0	12.4	0.0	52
IC	329	4.04	2.8	27.5	87.2	10.6	0.0	32
IH	254	3.97	3.8	30.2	92.8	12.2	0.0	44
IS	257	4.00	4.3	28.7	90.3	14.3	0.3	59
SEM	7.2	0.078	0.52	3.13	6.81	0.97	2.52	5.6

Table 2. Silage microbiological quality in Exp.II, ^{10}log cfu/g, except clostridia ^{10}log MPN/g silage

	3 h wilting					6 h wilting					
	NA	FA	IC	IH	IS	NA	FA	IC	IH	IS	SEM
Coliforms	0.95	0.95	0.95	0.95	0.95	3.47	0.95	0.95	0.95	0.95	0.252
Yeast	3.28	0.95	1.50	3.42	1.50	4.00	1.79	2.08	2.40	3.87	0.285
Molds	1.33	2.29	2.89	2.16	1.89	1.75	2.06	2.45	2.00	1.22	0.174
Clostridia	4.04	3.42	1.43	2.51	4.04	2.81	0.54	0.48	1.04	2.88	0.316
LAB	8.01	8.62	6.70	6.75	7.57	8.33	8.68	5.89	5.79	8.41	0.250
HFLAB	7.68	8.62	4.58	6.05	7.34	8.28	8.68	4.00	5.24	8.29	0.414
Aer.spores	3.33	2.42	2.07	1.30	3.02	2.65	1.45	2.05	1.39	1.63	0.191

NA= No additive; FA= formic acid; IC= on the cut sward inoculated; IH= precision cut harvesting inoculated; IS= silo inoculated; DM= dry matter; WSC= water soluble carbohydrates; LAB= Lactic acid bacteria; HFLAB= Heterofermentative lactic acid bacteria.

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Correlation between colour and temperature of LDPE stretch film used in silage bales.

Key words: Bale, carbon dioxide, colour, LDPE, oxygen, permeation, reflectance, silage, solar irradiation, spectroscopy, stretchfilm, temperature, UV-VIS-NIR.

The use of bale silage is increasing due to rational handling and cost effectiveness. In order to give high quality silage it is very important to restrict unwanted micro-biological activities to very low levels. Carbon dioxide producing processes can be suppressed by high partial pressure of CO_2 inside the bales. Aerobic processes are, of course, suppressed by low oxygen pressure. Consequently, the wrapped bale should be as gas tight as possible. The temperature of the stretchfilm should, therefore, not be too high, because high temperatures lead to high permeation rates of oxygen and carbon dioxide through the film and a consequently increasing micro-biological activity inside the bale. Micro-biological processes are normally, of course, also promoted by moderately increased temperatures.

Object of study

In this investigation we have studied the correlation between the fraction of the total solar energy reflected by differently coloured LDPE stretch films and the bale surface temperatures as well as the temperatures inside the bales.

The solar reflectance, R_s, is defined as the fraction of the total solar irradiation that is reflected by a body - in this case LDPE films. R_s is calculated from the obtained spectra according to ASTM E 891, Table 4 for air mass 1.5 [1]. This air mass is corresponding to the conditions in northern Europe.

Material & methods

The measurements were performed with an UV-VIS-NIR-spectrophotometer equipped with an integrating sphere. Spectra between 200 and 2500 nm were obtained on a single layer of film. The samples were placed at the exit port of the sphere i.e. at the rear of the sphere. The side of the films not covered by tackifier was directed towards the light source of the spectrophotometer. Behind the samples a radiation trap (a cavity having a black internal surface) was placed in order to absorb all light, which was not absorbed or reflected by the sample. It is assumed, that in a real bale, the light that is not reflected or absorbed by the stretchfilm is absorbed by the silage inside the bale.

The bale surface temperatures were measured with an IR-camera, while the temperatures inside the bales were measured using a thermocouple of copper-constantan. The spectroscopic measurements were performed on a single film. The bales, however, were wrapped with six layers of LDPE film. The nominal film thickness was 25 µm. Four different films were investigated concerning solar reflectance and bale surface temperature and internal bale temperature. The colours of the films were white, light green, dark green, and black. The values of the solar reflectance, R_s, obtained were 31, 28, 13, and 4,4%, respectively. The bale surface temperatures measured a clear summer day at 12 a.m. were 32, 35, 46, and 53 °C,

respectively. The surface temperatures were measured by the IR-camera in those parts of the bales which were directed to the south.

Results

As expected the temperature of the white film was lower than the black one. The temperature difference is quite pronounced - more than 20 °C. Between the white and the black films one finds the light and dark green films regarding the R_s values. If the R_s values found are plotted as a function of bale surface temperature a very nice linear correlation results. A similar linear correlation is even found for the mean temperature during a whole week measured approximately 10 cm inside the bale.

The measurements above show, that the film ability to reflect the solar irradiation is a very important factor in determining the temperature of the film. Moreover, the fact that there is also a good correlation with the temperature at approximately 10 cm below the surface confirms further the importance of the films ability to reflect solar irradiation. It can be noticed that the temperature in the bale with the white film exceeds the ambient temperature with only a few degrees. The bale with the black film has, under the same period, been up to 25 °C warmer than the ambient.

As mentioned above, the temperature influences the permeation rate of gases for a film. In most cases there is an Arrhenius relationship, i.e. an exponential relation, between permeation rate and temperature. The increase in permeation rates of O_2 and CO_2 through LDPE films are estimated for the increase in temperature caused by the absorption of solar energy. Input data besides temperatures used in the calculations are taken from ref. [2, 3]. No consideration has been taken to possible effects of additives such as tackifier, carbon black or titanium dioxide. The relative difference in permeation for O_2 and CO_2 between a white and a black LDPE film exposed to the weather condition a clear summer day, as above, at 9 a.m. was found to be 50% and 40%, respectively. The temperature difference was 7.4 °C. The difference in permeation rate at 12 a.m. the same clear day was 190% and 140 %, respectively. The temperature difference was in this case 21.4 °C. The ambient temperature at 12 a.m. was 23,6 °C.

Conclusions

The examples above show that differences in ability to reflect solar irradiation have a pronounced influence on gas tightness, especially for parts, which are directly exposed to sun light. In the calculations above no consideration has been taken to the tightness of the overlapping joints of the films.

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Inducing aerobic instability in laboratory scale silages

Key words:
Silage, aerobic stability, heating, yeasts, silage additives, screening method, ensiling technology.

Objective

To develop a method to increase the frequency of obtaining aerobically unstable silages on a laboratory scale. The screening and efficacy testing of silage additives, which are designed to "improve aerobic stability", generally requires the provision of an aerobically unstable silage as negative control. This is unlikely to occur, if an ideal anaerobic ensiling technique is adapted. Different protocols for length of storage, degree of compaction as well as frequency and duration of defined air infusion have been investigated, from which the following recommendation was derived. It is currently being operated and further tested for the approval of silage additives in Germany.

Materials and Methods

High dry matter (DM) grass and maize (approx. 450 g DM kg^{-1} and approx. 300 g DM kg^{-1} respectively) were ensiled in either glass (1.5 l) or PVC (2.8 l capacity) laboratory silos for a storage period of 7 weeks. Highly compacted silages, corresponding to a porous volume of 4.5 liter kg DM^{-1} were compared to silos containing only 75% of this amount, in order to ease the gas flow when half of the silos were aerated after 4 and 6 weeks respectively. This was achieved by opening 6 mm holes (8 mm, PVC) in the lid and the bottom of the mini silos for 24 hours. Aerobic stability after unloading the silos was determined by recording the temperature rise of a silage sample (equivalent to 100 g DM) in an insulated container 4 times a day during one week (Honig, 1986: Grass and Forage Reports 1990, 76-82, 1990, Swedish University of Agricultural Sciences).

Results

Without air infusion about 90 % of the silages remained stable for almost the entire recording period and the degree of silo fill had little effect (Table 1). By controlled infusion of air twice during storage silages could be produced, which started to deteriorate within 3 to 4 days upon exposure to air. However, this was only achieved with a high degree of probability in the loosely filled silos. In the well-compacted silages the initiation of a facultative aerobic spoilage flora of lactate assimilating yeasts was avoided owing to restricted gas exchange and oxygen supply (Figure 1). Consequently, a reliable, early start to heating processes was not induced in these silages. Whilst the described method of challenging aerobic deterioration can be used very satisfactorily with grass crops, a high proportion of the maize silages proved to be very unstable in one out of three experimental years and deteriorated immediately after opening. This was probably due to a particularly responsive flora of spoilage yeasts.

Conclusion

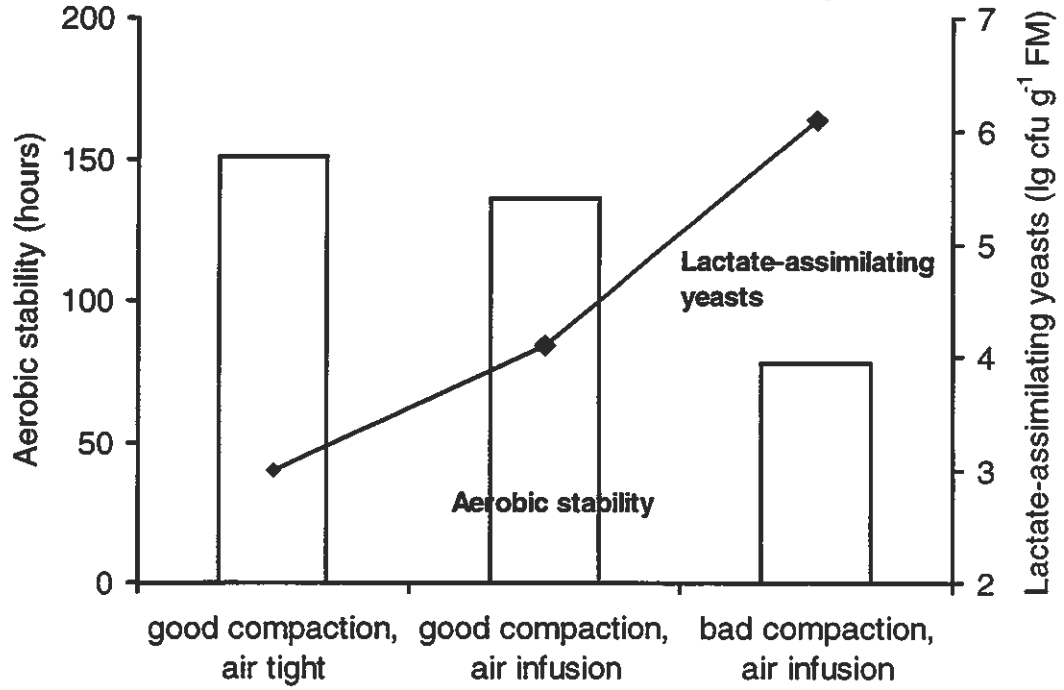
A simple and reproducible method has been developed and successfully tested in numerous ensiling experiments over three years. It reliably gives untreated controls of considerably reduced aerobic stability in high DM grass silages and allows the evaluation of appropriate, stabilising silage additives with a higher probability of utilisable experiments. For respective investigations in maize silages, an attenuated air treatment has to be considered and properly defined, which does not overcharge potential test products and mask differences which may otherwise have been observed.

Table 1 Effects of compaction and air infusion on the aerobic stability of maize silages

Compaction	Air influence	Gas losses	Aerobic stability	Aerobic DM loss
	(+/-)*	(g kg^{-1} DM)	(Hours)	(g kg^{-1} DM)
Good	-	50.1	158	3.9
Bad	-	52.2	152	7.3
Good	+	55.3	153	15.0
Bad	+	68.4	91	52.0

* (+/-) = with or without air infusion

Fig.1 Effects of compaction and air infusion on the aerobic stability of grass silages



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Different application methods of a bacterial inoculant and its influence on silage fermentation

Key words: Lactic acid bacteria, inoculant, application technique, silage quality

Object of the study

The aim of this pilot experiment was to investigate if application methods of an commercial inoculant of lactic acid bacteria affects the silage fermentation. The hypothesis was that an early application of the inoculant would have a positive effect on silage fermentation.

Introduction

Silage fermentation relies on the accumulation of organic acids, which are formed through the activity of homofermentative and heterofermentative lactic acid bacteria (LAB) under anaerobic conditions. The numbers of naturally occurring LAB on crop, however, are usually small in comparence with other organisms of the epiphytic microflora. To facilitate and ensure a successful silage process, silage additives may be added to the crop at ensiling. Additives are usually divided into two groups; inhibitors (e.g. organic acids) and stimulants (e.g. inoculants), and the additives are usually applied to the herbage in the chopper.

The objective of inoculants is to provide the crop with a sufficient number of fast growing LAB, thereby ensuring a rapid acidification to restrict the activity of undesirable organisms. The main advantages with inoculants are that they are harmless to handle and not corrosive to the machinery. They can also be used in organic farming without restrictions. If the concentration of readily available substrates in the original crop is insufficient, however, the bacteria in an inoculant will not be able to produce enough lactic acid to lower pH to an acceptable level (Merry et al 1997).

In Sweden, silage additives based on organic acids traditionally have been dominating, and inoculants have only slowly been accepted by the farmers. Some objections against inoculants are that the bacterial solution have to be prepared before use, and that the durability of the prepared solution is limited. The earlier the inoculants could be applied in the mechanised systems of forage, the easier for the farmer to plan and organise the application work. Müller et al (1993) had shown that the epiphytic LAB not only survive, but also multiplies during prewilting. If the organisms of the inoculant also survive in swath during the prewilting period and even start to multiply, application in the mower conditioner may be a more adaptable method. Hence, the inoculated LAB can earlier start competing with the other epiphytic organisms. Problems with uneven distribution of the additive in long cut material could also be overcomed by this way.

Material and methods

A clover/grass sward was cut with a mower conditioner (Kverneland 347) at third cut 1998. Additives in the form of lactic acid bacteria (Lactisil ®; *Lactobacillus plantarum, Pediococcus acidilacti* (10^5 (g FM)$^{-1}$) was applied in front of the conditioner (MC; front) or immediately after the conditioner (MC; after). The crops were allowed to prewilt for approximately 30 h before it were chopped with a precision chop harvester (Taarup SE 2100). Prewilted crops harvested with precision chop harvester with (CP) or without application of LAB in the chopper were used as control. The experiment was replicated in two blocks. The precision chopped herbages of all treatments were filled in experimental silos (3 steel silos (25 L); 3 glass silos (1.7 L) / treatment and block), and stored at constant temperature (25°C). After three days the glass silos were opened and the initial pH decrease was measured. After 100 days of storage the steel silos were opened and the silage sampled for chemical analyses.

Result

At ensiling the dry matter concentration of silage crop was approximately 326 g kg^{-1}. The crops contained on average 189 g crude protein (CP) and 92 g water soluble carbohydrate (WSC) (kg $DM)^{-1}$, while the buffering capacity was approximately 413 mekv OH (kg $DM)^{-1}$.
The results of the pH determinations after three and 100 days are presented in table 1. Significant effect of LAB independently of application technique was noticed. The initial pH decrease was fastest in silages where the LAB inoculant was applied in front of the mower conditioner.

Table 1. The pH of silages after three and 100 days of fermentation. Inoculant of LAB applied in the precision chop harvester (PC), in front of the mower conditioner (MC; Front) or after the mower conditioner (MC; after)

Days of		LAB application			Mean	
Storage	Untreated	PC	MC; front	MC; after	values	LSD
3	4.83	4.53	4.46	4.58	4.60	0.043
100	4.39	4.25	4.20	4.22	4.26	0.020

Chemical composition of silages after 100 days of storage are presented in table 2. All silages were of high quality. Significant effect from LAB, with a more homolactic fermentation, was seen independently of application technique. Application in front of the mower conditioner tended to give the best silage quality.

Table 2. Chemical composition of silages after 100 days of fermentation (n=6). Inoculant of LAB applied in the precision chop harvester (PC), in front of the mower conditioner (MC; Front) or after the mower conditioner (MC; after).

		LAB application			Mean	
	Untreated	PC	MC; front	MC; after	value	LSD
DM; g kg^{-1}	328	335	313	284	315	15.2
NH_3-N, g (kg tot-N)$^{-1}$	79.2	67.4	65.1	68.6	70.1	3.24
Lactic acid; g (kg DM)$^{-1}$	110	100	104	111	106	5.8
Acetic acid; g (kg DM)$^{-1}$	27.7	13.7	12.9	22.2	19.1	1.64
2,3-Butanediol; g (kg DM)$^{-1}$	3.2	2.8	2.7	2.0	2.7	0.71
Succinic acid; g (kg DM)$^{-1}$	13.2	6.2	6.7	7.5	8.4	0.95
Ethanol; g (kg DM)$^{-1}$	3.9	1.5	1.3	1.7	2.1	0.55

Conclusion

- Silage quality was positively affected by LAB inoculation
- The positive effect of the LAB inoculant was not reduced after prewilting on field for 30 h.
- Application of the LAB inoculant in front of the mower conditioner slightly improved silage quality compared with other treatments.
- More basic studies on the topic are required

Acknowledgements

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Assessing Fermentation Quality in Grass and Legume Silages: Interlaboratory Comparison

Analytical methods of feed testing are regularily compared in collaborative interlaboratory studies to support government and intergovernmental regulations especially in the field of traded feeds by organisations such as the Association of Official Analytical Chemists and its national and international equivalents. In this respect, presumably because of their lesser relevance in commerce, forage analytical methods are much less studied collaboratively. This is why an international scientific activity like the currently running EU funded project LEGSIL on legume silages for milk production requires collaborative testing of methods.

In order to ascertain the comparability of silage quality data obtained in the LEGSIL participant labs in Finland and Germany 12 silages (4 legume silages prepared without previous wilting at the University of Helsinki from Goats rue, *Galega orientalis* and 8 grass silages from farms in Finland and Northern Germany in the dry matter range from about 20% to 65% DM) were divided into 0.5 kg to 1.0kg subsamples and distributed in the frozen state to the three labs. At each of these labs the currently used methods for sample preparation, extraction and quantification were employed on this set of silages. The following parameters were assessed : Dry matter content (DM) was determined by oven drying and expressed as DM corrected for volatile losses. The pH was determined by pH-electrode and the ammonia-N-content was determined by ammonia sensitive electrode and related to the total N content of the sample. The fermentation acids (lactic acid (LA), acetic acid (AA), propionic acid (PA) and butyric acid (BA) were all determined by gas chromatography and expressed as a proportion of dry matter.

Comparison of Silage Quality Methodology at VALIO, FAL and HY

	DM	pH	LA	AA	PA	BA	NH_3
r^2 (FAL:Valio)	**0.99**	**0.98**	**0.97**	**0.99**	**0.95**	**0.03**	**0.83**
r^2 (FAL:HY)	**0.99**	**0.98**	**0.96**	**0.99**	**0.82**	**0.52**	**0.95**
r^2 (Valio:HY)	**1.00**	**0.99**	**0.99**	**0.99**	**0.93**	**1.00**	**0.93**
Sy_x (FAL:Valio)	**1.40**	**0.09**	**0.60**	**0.18**	**0.05**	**0.27**	**1.46**
Sy_x (FAL:HY)	**1.87**	**0.09**	**0.71**	**0.20**	**0.09**	**0.26**	**0.79**
Sy_x (Valio:HY)	**0.68**	**0.06**	**0.30**	**0.13**	**0.04**	**0.02**	**0.97**

DM = Dry Matter; LA = Lactic Acid; AA = Acetic Acid; PA = Propionic Acid; BA = Butyric Acid; NH_3 = Ammonia-N Fraction of total N

The results of the statistical evaluation of the resulting data are given in the table above. In those silage quality parameters where the sample set exhibited a high degree of variation, the coefficients of determination demonstrated a very high degree of correspondence between the

three labs (DM, pH, LA and PA). Near zero contents of PA and BA form the underlying cause of lower coefficients of determination between labs. The standard errors of predicting the results of one lab from those of another ($s_{y\text{-}x}$) are generally highly satisfactory. The correspondence between the two Finish labs was nearly always better than that between either of them and the German lab.

The general consequence to be drawn from this interlaboratory comparison is one of high standardisation and comparability achieved without prior adaptation and modification of the routine procedures of the participating labs.

Workshop E
Determination and control of aerobic instability

Poster abstracts

Developing a technology for ensiling forage crops for small-holder cattle owners in developing countries

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In developing countries, such as in the semi-arid tropics in Africa, many small cattle farms comprise a few milking cows only (2-5). In the dry winter season which lasts for 6 months, the cows don't produce any milk and even starve from lack of forage. On the other hand, in the rainy summer, there is a surplus of natural grasses and forage crops which can be preserved for the dry season. Conserving forages from the rainy to the dry season would support continuous milk production throughout the year and might have a significant impact on the standard of living in these areas. Hay making is uncertain due to late rainfall and harvest at a late maturation stage that avoids the rain would result in a decrease in forage quality. Therefore, the best option is to harvest the crops at peak nutritional value and to preserve them by ensiling. However, the small-holder cattle owners in these areas cannot afford the large investments required for conventional ensiling. An alternative method could be ensiling in small units, in re-usable plastic bags (10-20 kg) that can be prepared manually by the farmer and his family and can be used one at a time according to need.
The purpose of the current work was to evaluate the potential of ensiling of forage crops in plastic bags which are not completely air tight. We hypothesized that in the presence of low concentrations of oxygen, acetic acid and other volatile fatty acids are produced during the ensiling fermentation, and these inhibit the development of yeasts and moulds in the bags.
In the first experiment, four types of plastic bags were filled with chopped maize, and sealed with tape. Internal gas samples were taken six times during 63 days to study fermentation dynamics. In the second experiment, wheat was ensiled in polyester/polyethylene (PET/PE) bags for 6 months. These bags proved to be the best in the first experiment with maize. Three bags were sampled every month. Internal gas composition was determined before sampling the last bags.
Oxygen concentration in bags of all types did not exceed 10%. Carbon dioxide concentration in the PET/PE bags was around 25% in the first experiment (day 63) and around 17% in the second. Visual and olfactory appraisal revealed that both maize and wheat silages were of good quality. Lactic and acetic acid concentrations were 3-4% and 1.5-2.0%, respectively. Yeast and mould counts did not exceed 10^5 CFU g^{-1} in most samples.
These results indicate that it is possible to obtain high-quality silage in small plastic bags. The results reveal that the bags must not be completely impermeable to air in order to obtain good silage.

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The effect of applying an inoculant containing *L buchneri* to high dry matter ryegrass swards ensiled in wrapped, round bales

Introduction

The ensilage of grass in wrapped big bales now accounts for 18% of grass conserved in the United Kingdom. Big bale silage is often prone to aerobic deterioration due to a restricted fermentation, lack of consolidation, entrapment of air, and/or damage to the wrap which allows survival of moulds and yeasts. The use of additives in big bale silage has improved the resulting fermentation but many of the additives available give little protection against subsequent aerobic spoilage.

An organism (*Lactobacillus buchneri*), has been shown to produce aerobically stable silages when incorporated with a range of high dry matter forages (Driehuis et al, 1996). This experiment examines the effect of inoculating high dry matter grass with 2 biological inoculant silage additives, Biotal ASG 15 (Brand Names Axcool/Biocool) and ASG 55, both containing *L. buchneria*, for ensiling high dry matter grass in wrapped big bales.

Materials and Methods

A predominantly perennial ryegrass sward was cut on 16 July 1996 using a Taarup 307 disc mower/conditioner. The swaths were allowed to wilt *in situ* for 48 hours to achieve a target dry matter of 400 g/kg. The resulting herbage was then sampled for chemical analysis prior to harvest with a fixed chamber Krone KR 125 round baler. The application of the 2 test additives was via a dribble bar attached immediately in front of the pick up reel to achieve a target application rate of 4 l of inoculant per tonne of fresh herbage.

Each bale was weighed, individually wrapped with four layers of polythene film and stacked outside in a single layer, on a polythene sheet. Following 120 days storage, the bale wrap was removed and the bale reweighed. Each bale was then given a visual score for surface moulding and cored for chemical analysis and aerobic stability assessment. A further aseptic sample was taken for microbial assessment. Following the aerobic stability assessment, samples were again taken for microbial assessment.

Results And Discussion

Adverse weather during early July delayed cutting, resulting in a more mature crop. Climatic conditions following cutting allowed a good wilt to be achieved. Mean bale weight was 343 kg at a DM of 463 g kg^{-1} and soluble sugar content of 89 g kg^{-1} DM. The actual additive rate applied was 5.54 and 5.28 l $tonne^{-1}$ for ASG 15 and ASG 55 respectively.

At opening, weight change was extremely variable but all bales increased in fresh weight by an average 14.4 kg indicating an ingress of rainwater through the layers of wrap. Visible moulding was generally slight and any mould present was confined to the bale surface i.e. were not penetrating. Dry matter contents at opening were generally similar to the DM content of the grass at ensiling.

The chemical composition of resulting silages is given in Table 1. The high fibre content and relatively low protein content reflects the maturity of the crop. Both additives contained specific hydrolytic polysaccharase enzymes to increase the availability of soluble carbohydrates. For ASG 15 there was a significant increase in water soluble carbohydrate content ($P<0.01$). The fermentation characteristics suggests that for high dry matter, big bale silage a good fermentation was achieved for all treatments. The inoculant treatments significantly increased acetic acid ($P<0.01$) and reduced butyric acid ($P<0.05$) and ethanol ($P<0.01$) when compared to the untreated control. The inoculants also reduced the ratio of Lactic to Acetic acid ($P<0.05$) which normally infers a more heterolactic fermentation. However, levels of Caproic and Valeric acids were too low to be detected. Hence the silages appear to have undergone a controlled fermentation resulting in both Lactic and Acetic acid..

Table 1 Chemical Composition of Big Bale Silage

	Control	ASG 15	ASG 55	SEM	d.f.	CV
Oven Dry Matter (g/kg)	445	505	485	9.94	40	8.0 ***
On a DM basis (g/kg DM)						
Neutral Detergent Fibre	728	731	732	2.86	40	1.5
Water Soluble Carbohydrate	36.3	47.1	36.2	2.58	40	25.0 **
Organic Matter Digestibility	584	575	582	5.56	40	3.7
Total Ash	51	50	50	1.13	40	8.7
Crude Protein	130	131	129	2.60	40	7.7
Ammonia N as % Total N †	8.1	6.3	6.5	0.11	40	16.5
pH ‡	5.6	5.3	5.2	0.008	40	4.2 *
Lactic acid	40.1	43.0	46.5	5.10	40	45.8
Acetic acid	17.6	26.1	28.2	2.39	40	38.6 **
Ratio of Lactic to Acetic ‡	2.71	1.64	1.70	0.045	40	66.8 *
Propionic acid	7.4	9.0	9.6	1.07	40	47.7
N-Butyric acid †	33.4	15.5	17.4	0.40	40	34.7 *
Ethanol	52.8	32.2	38.9	3.90	40	36.6 **

† means reported have been back-transformed from logarithmic transformation
‡ means reported have been back-transformed from square root transformation.
SEMs refer to transformed data.

The microbial analysis on the fresh samples were supportive of the chemical analysis in which all silages had generally undergone a lactic fermentation. Counts of yeasts, moulds and enterobacteria were relatively low but variation was large. There was a significant ($P<0.001$) increase in lactic acid bacteria numbers with inoculant treatment. From the data it would appear the increase in acetic acid seen in the chemical analyses can be attributed to *L. buchneri.*

During aeration, the untreated silages achieved higher temperature lifts than either of the 2 inoculants. Following 7 days aeration there was little deterioration with treated silages (Table 2). Moulds increased slightly but remained relatively low. Lactic acid bacteria numbers remained static. In contrast, the untreated silages showed marked increases in yeasts, moulds and enterobacteria. Thus after the 7 days aeration, the untreated silages contained significantly higher counts of yeasts ($P<0.001$) and enterobacteria ($P<0.05$). In addition they contained significantly lower counts of lactic acid bacteria.

Table 2 Microbial Counts (log count of colony forming units/g) for silage following 7 days aeration

	Control	ASG 15	ASG 55	SEM	d.f.	CV%
Yeasts @ 25°C	4.89	1.52	2.50	0.435	40	56.7 ***
Moulds @ 25°C	4.15	3.07	3.18	0.477	40	53.3
Lactic acid bacteria	6.81	8.36	8.36	0.342	40	16.9 **
Enterobacteria @ 30°C ‡	3.30	1.82	1.65	0.035	40	33.5 *
Clostridia	5.35	4.77	5.25	0.284	40	21.5

‡ means reported have been back-transformed from negative reciprocal transformation
SEMs refer to transformed data.

Conclusions

The addition of ASG 15 and ASG 55 to mature, high dry matter grass, ensiled in wrapped big bales resulted in an improved fermentation as measured by pH, lactic acid and ammonium N content, and reduced variability between bales. The results were more pronounced for ASG 55.

The addition of ASG 15 and ASG 55 also reduced visible moulding on the bale surface at opening and improved aerobic stability. Again, the results were more pronounced for ASG 55.

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Lactobacillus buchneri improves aerobic stability of laboratory and farm scale whole crop maize silage but does not affect feed intake and milk production of dairy cows

Introduction

Aerobic spoilage of silages is manifested (1) as heating during the feeding period and (2) as spoilage of surface layers. Aerobic spoilage generates losses and reduces the hygienic quality and (sometimes) palatability. Aerobic spoilage is usually initiated by yeasts that oxidise the preserving acids. Thereafter, also other microorganisms (e.g. moulds, bacilli, *Listeria* monocytogenes) start to proliferate. Silage additives aiming to improve aerobic stability should therefore inhibit the growth of spoilage yeasts. Previous studies have shown that the use of *Lactobacillus buchneri* as a silage inoculant reduces the growth and survival of yeasts and improves the aerobic stability of different silage crops (Driehuis *et al.* 1996, 1999). Conversion of lactic acid to acetic acid and 1,2-propanediol is an important underlying principle of this effect (Oude Elferink *et al.* 1999). Here we show the results of a study on the effect of a *L. buchneri* containing inoculant on the fermentation characteristics and aerobic stability of whole crop maize silage ensiled under laboratory and farm conditions and on feed intake and milk production of high yielding dairy cows.

Materials and Methods

An 8-ha plot of maize was harvested at 19 September 1997. Half of the material was treated with an inoculant containing *L. buchneri* and a blend of polysaccharolytic enzymes (Biotal Ltd, Cardiff, UK), the other half was not treated. The treatment provided 10^5 cfu/g of *L. buchneri*. The maize was ensiled in two 100-t bunker silos. During filling samples were taken from each load. The bulked samples were used to make laboratory silages in 1-l airtight jars (0.5 kg per jar) and double-layered polyethylene bags (5 kg per bag). Laboratory silages (duplicates) were analysed after 84 days storage at 20°C. The farm silages were opened 80 days after ensiling and unloaded at a rate of 2 m per week. Triplicate samples from the silo faces were analysed 36 and 56 days after opening. Aerobic stability was determined by incubation of silage samples in polystyrene containers with perforated lids at 20°C and monitoring temperature and defined as the time needed to increase the temperature to 21°C.

Treated and untreated silage were compared in a trial with 64 dairy cows (Holstein-Friesian x Dutch-Friesian) kept in a loose housing system. The ration of the animals consisted of maize silage, grass silage, ground maize ears and concentrates (providing 33, 30, 9 and 29%, respectively, of the total net energy intake). The ration was given ad libitum as a total mixed ration. The experimental design was a randomised block experiment with 2 groups of 32 animals, lasting 6 weeks. During week 1 to 3, both groups received untreated maize silage in their ration. During week 4 to 6, one group received untreated silage, the other group treated silage. Feed intake was determined per group. Feed refusals were weighed 3 times per week. Milk production was determined daily. Milk composition was determined once per week in week 1 to 5 and twice in week 6. Results were statistically analysed using Anova. Results of week 1 to 3 were used as covariates for the comparison of results of week 6.

Results

Ensiling in jars simulated ensiling under optimal conditions. Ensiling in bags simulated ensiling under 'air-stress' conditions (simulating surface layer material). With both silo types, *L. buchneri*-treated silages had higher acetic acid and 1,2-propanediol contents and lower

lactic acid contents and slightly increased pH and dry matter (DM) loss values (Table 1). *L. buchneri*-treated silages had lower yeast and higher lactic acid bacteria (LAB) counts and a higher aerobic stability. The aerobic stability improving effect of *L. buchneri* was most pronounced in the silages ensiled in bags. Similar responses were detected in the farm silages (Table 1). However, the effect of *L. buchneri* on yeast counts and aerobic stability was clearly less pronounced than in laboratory silages. 1,2-Propanediol was lower in the treated farm silages than in the treated laboratory silages, suggesting a lower activity of *L. buchneri*. A lower silage temperature during the storage period might be an explanation for this. Yeast counts were very high in the farm silages and, consequently, aerobic stability values were low. This suggests that sealing or silage density may have been non-optimal. Total feed intake and milk, milk fat and milk protein production of cows receiving untreated or *L. buchneri*-treated maize silage were 20.8 and 20.4 kg DM/d, 38.8 and 38.8 kg/d, 1638 and 1639 g/d and 1258 and 1284 g/d, respectively. There were no statistically significant differences between the treatments.

Conclusions

The present study showed that inoculation with *L. buchneri* improves the aerobic stability of whole crop maize silage. The efficacy of *L. buchneri* was higher under laboratory conditions than under farm conditions. No effects of *L. buchneri* on feed intake and performance of dairy cows were observed.

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Table 1. Composition and aerobic stability of untreated and *L. buchneri*-treated maize silage.

	pH	Lactic acid (	Acetic acid g/kg DM	1,2-Pro-panediol	DM loss)	Yeasts (log cfu/g)	LAB	Aerobic stability (h)
Lab. silages (jars)								
Untreated	3.86	56.7	15.7	0.0	12.8	2.2	7.9	245
Treated	3.91*	49.7*	19.2	5.7*	16.3*	2.0	9.0*	>336*
Lab. silages (bags)								
Untreated	3.93	38.3	20.3	0.0	23.3	5.4	8.6	30
Treated	4.08*	31.8*	28.6*	5.3*	27.7*	2.1*	9.6*	293*
Farm silages								
Untreated	3.88	47.7	12.2	0.0	nd	7.1	7.7	9
Treated	3.92	47.3	18.5*	3.8*	nd	5.7*	8.7*	41*

*Means of untreated and treated silage are significantly different ($P<0.05$); nd, not determined

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Lactobacillus buchneri can improve the aerobic stability of silage via a novel fermentation pathway: the anaerobic degradation of lactic acid to acetic acid and 1,2-propanediol.

Introduction

Aerobic spoilage of maize silage is a well known problem, which is generally initiated by yeasts. Silage additives aiming to improve the silage quality should therefore not only aid to a rapid silage acidification, but also prevent growth of spoilage yeasts. Silage additives based on *Lactobacillus buchneri* can effectively inhibit growth and activity of spoilage yeasts, and have been used to improve the aerobic stability of silage. Here we show that this inhibition of spoilage yeasts is probably mainly due to the capability of *L. buchneri* to ferment lactic acid to acetic acid and 1,2-propanediol.

Methods

Whole crop maize (37% DM) was treated with water or *L. buchneri* ($1x10^6$ cfu/g) and ensiled in double layered polyethylene bags (5 kg/bag). Two silages per treatment were opened 7, 14. 28, 56, 84, and 137 days after ensiling and analyzed for chemical composition (HPLC + GC), yeast numbers (Malt Extract Agar, pH 3.5), *Lactobacillus* numbers (Rogosa agar, pH 5.4), and aerobic stability. The aerobic stability was determined by measuring temperature increase in silage samples incubated in polystyrene containers with perforated lids at ambient temperature. The aerobic stability was defined as the time needed to increase temperature 1°C above ambient.

Results and Discussion

The effect of *L. buchneri* on the fermentation pattern, microflora, and aerobic stability of whole crop maize silage is shown in fig. 1. During the first month of ensiling there is little difference between the untreated and *L. buchneri* treated maize silage. However, after one month of ensiling the fermentation pattern of the treated silage starts to differ from that of the control silage, because the lactic acid in the silage is being degraded to acetic acid and 1,2-propanediol. Simultaneously, yeast numbers start to decline and the aerobic stability starts to improve. After 4.5 months of ensiling almost all lactic acid in the treated silage has been degraded, while yeasts have almost disappeared, and the silage has obtained a very high aerobic stability. These results indicate that *L. buchneri* is capable of improving the aerobic stability of silage by inhibition of yeasts. An important underlying principle of this effect seems to be the anaerobic degradation of lactic acid to acetic acid and 1,2-propanediol. The stoichiometry of this novel fermentation reaction was studied in pure cultures of *L. buch*eneri (at pH 4). The proposed fermentation pathway is depicted in fig. 2. Recent experiments (not shown) have indicated that the *L. buchneri* also produces other, yet unidentified metabolites with antifungal activity. To which extent these metabolites can help to prevent aerobic spoilage in silage is not yet clear.

Conclusion

Silage inoculation with *L. buchneri* inhibits yeast growth and improves the aerobic stability of the silage. This seems mainly due to the capability of *L. buchneri* to ferment lactic acid to acetic acid and 1,2-propanediol.

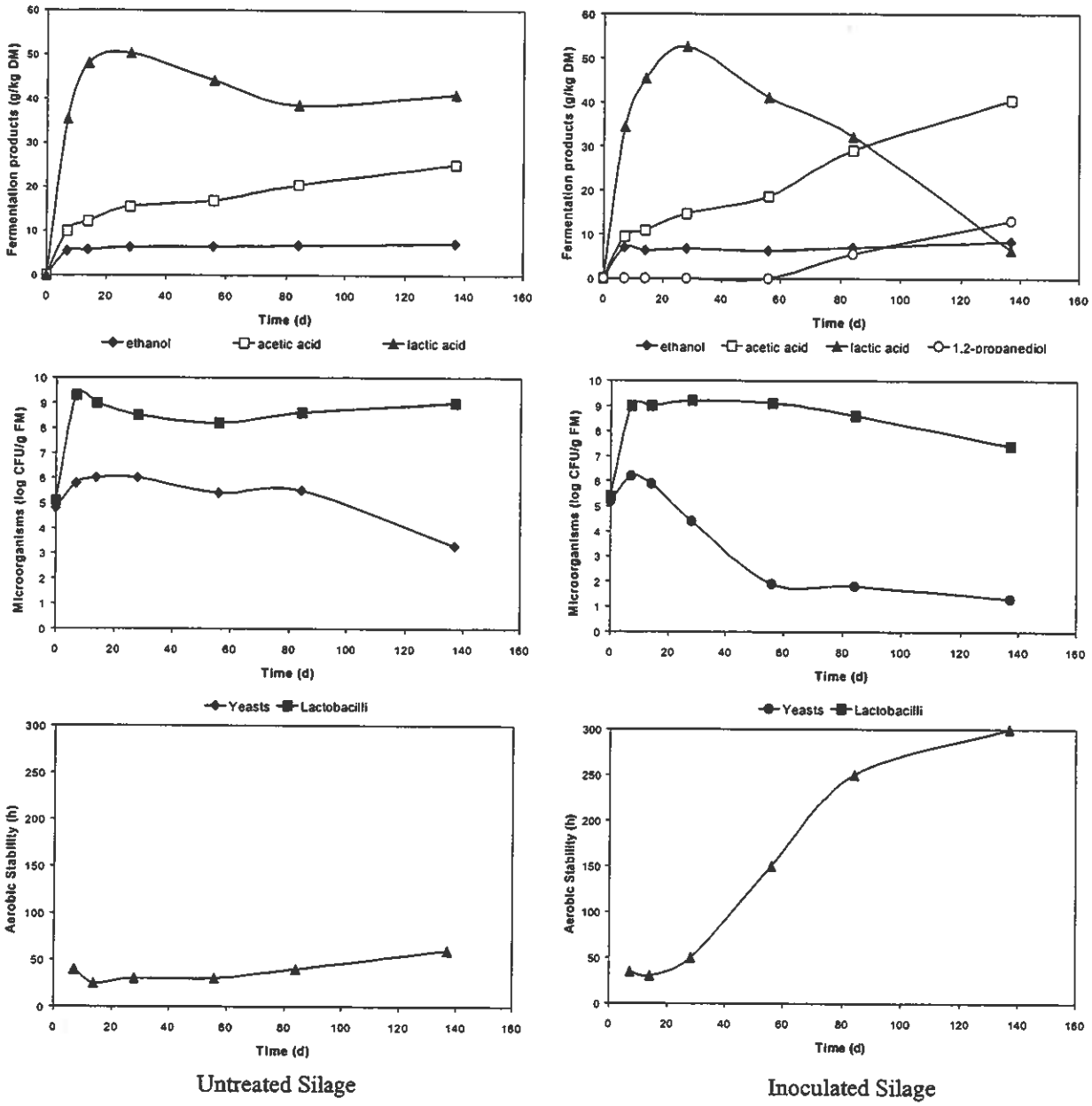

Figure 1: The effect of *L. buchneri* inoculation on silage fermentation, microflora, and aerobic stability of maize silage followed in time.

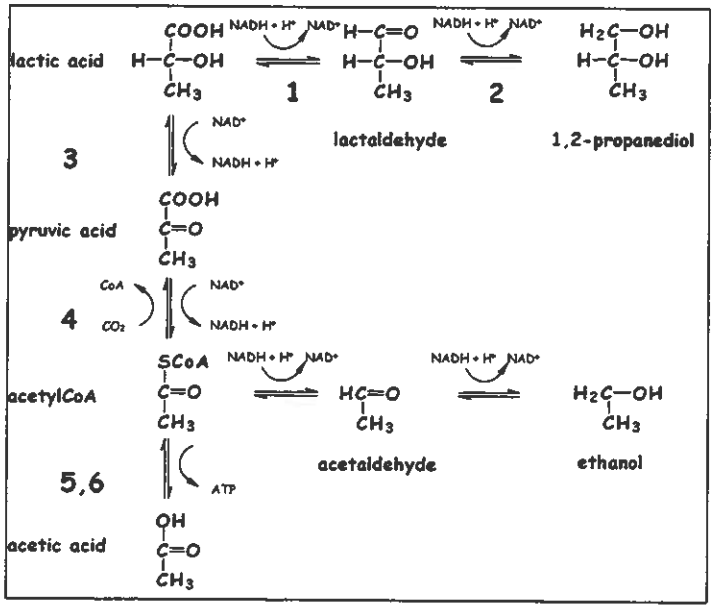

Figure 2: Anaerobic degradation pathway of lactic acid in *L. buchneri.*

The Effect of Applying Lactic Acid Bacterial Inoculants at Ensiling on the Fermentation and Aerobic Stability of Whole Crop Wheat Silage

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INTRODUCTION

Wheat for silage is popular in many areas because it can be grown at a wide range of climatic and soil conditions. However, one of the major problems encountered in preserving whole crop wheat is the stability at feed-out. This paper presents data about the effect of silage additives containing different kinds of lactic acid bacteria (inoculants) on the fermentation and aerobic stability of whole crop wheat silage.

MATERIALS AND METHODS

Whole crop wheat (*Triticum spp*) was harvested at the milk ripening stage (368g/kg DM) and chopped to *ca* 1.5cm (with a Wintersteigner^{R} chopper, Austria), treated and ensiled in 1.5l glass jars (Weck^{R}, Germany) equipped with a lid that enables gas release only. The jars were stored at 25 °C ± 2 °C for 65 days before opening. At the end of the ensiling period the silages were sampled for biochemical analysis (Weinberg *et al.* 1995) and subjected to an aerobic stability test lasting 5 days in a system developed by Ashbell *et al.* (1991).

The following treatments were used: control (no additives), and two different inoculants. Inoculant A 1188 (Pioneer, USA), containing *Lactobacillus plantarum* and *Enterecoccus faecium* and Inoculant B Biomax LP (CHR Hansen's, Denmark), containing *Lactobacillus pentosus*. Inoculants were applied as follows: on day of the experiment, 0.5g of the inoculum powder was suspended in 20ml tap-water and sprayed over a 1x4m area, followed by thorough mixing. Thus *ca* $0.5x10^{6}$ cfu/g^{-1} were applied.

All fermentation and aerobic stability data were analysed using analysis of variance (ANOVA) procedures (SAS Institute Inc., SAS/STAT release 6.03, 1988).

RESULTS AND DISCUSSION

The analysis of the fresh wheat is given Table 1. The biochemical composition of silages at silo opening are shown in Table 2. Both untreated and treated silages were well preserved as would be expected with a carbohydrate rich crop such as wheat. Treatment with the two bacterial inoculants significantly improved fermentation, with reduced pH ($p< 0.01$) and volatile fatty acids levels ($p<0.001$), and an increased lactic acid level ($p<0.01$) and lactic acid : volatile fatty acids ratio level ($p<0.001$). The water-soluble carbohydrates levels were lower in the treated material, suggesting a more extensive fermentation in these silages.

The aerobic stability data (Table 3) showed that the inoculated silages spoiled upon aerobic exposure faster than the control. This was evident from intensive CO_2 production and development of moulds. Aerobic deterioration of inoculated silages were associated with high levels of residual water-soluble carbohydrates and lactic acid and lack of volatile fatty acids. There was no yeast and moulds in any of the silages prior to exposure, except for the control in which low numbers of yeasts were detected. Therefore, the silages were expected to be stable upon aerobic exposure. However, after 5 days exposure, mould numbers increased more so in the treated silages. The control silages had also high yeast counts. The inoculated silages were unstable.

L. pentosus was tested because it is known to ferment pentose sugars to lactic and acetic acids. Wheat is rich in hemicellulose which is partially hydrolyzed to pentose sugars during ensiling, and this should have resulted in high levels of acetic acid which inhibits yeasts and moulds. However, our results did not show that trend.

There was no significant difference between the two types of lactic acid bacterial inoculants from the point of biochemical composition and aerobic stability assessments of silages.

CONCLUSIONS

Inoculations gave rise to a more extensive fermentation with significant improvements in pH, volatile fatty acids, lactic acid and lactic acid : volatile fatty acids ratio. Both inoculants do not look promising in protecting whole crop wheat silage upon aerobic exposure. Other types of inoculants which will protect small grain silages upon aerobic exposure should be tested.

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Table1. Biochemical and microbiological composition of the wheat as ensiled.

DM (g/kg)	368.0
pH	6.66
Ash (g/kg DM)	93.0
Crude protein (g/kg DM)	138.0
WSC (g/kg DM)	52.0
Lactic acid bacteria (log_{10} cfu/g)	NF
Yeasts (log_{10} cfu/g)	NF
Moulds (log_{10} cfu/g)	NF

DM= dry matter, WSC= water-soluble carbohydrates, NF= not found

Table 2. Biochemical composition of silage

	Control	Inoculant A	Inoculant B	Sem	Sig
DM (g/kg)	335.3^b	347.3^a	353.3^a	1.45	***
pH	4.35^a	3.87^b	3.89^b	0.071	**
WSC (g/kg DM)	42.7	26.0	24.7	5.20	NS
Lactic acid (g/kg DM)	7.5^b	35.3^a	28.1^a	3.31	**
Acetic acid (g/kg DM)	5.8	4.3	4.7	0.68	NS
Propionic acid (g/kg DM)	6.0^a	0.0^b	0.0^b	0.20	***
Butyric acid (g/kg DM)	1.5	0.0	0.0	0.62	NS
Total VFA (g/kg DM)	13.3^a	4.3^b	4.7^b	0.28	***
Lactic acid : VFA ratio	0.6^b	8.2^a	6.0^a	0.391	***

VFA= volatile fatty acids, sem= standard error mean, significance tested at $p<0.05$ (*), $p<0.01$ (**) and $p<0.001$ (***)

Table 3. Results of the aerobic stability test

	Control	Inoc. A	Inoc. B	Sem	Sig
pH after exposure	4.31^a	3.96^b	4.03^b	0.072	*
CO_2 production (g/kg DM)	3.87	8.85	9.85	3.468	NS
Yeast counts before exposure (log_{10} cfu/g)	3.4	NF	NF	- -	- -
Mould counts before exposure (log_{10} cfu/g)	NF	NF	NF	- -	- -
Yeast counts after exposure (log_{10} cfu/g)	7.3	NF	NF	- -	- -
Mould counts after exposure (log_{10} cfu/g)	5.5	7.6	7.6	- -	- -

Effect of *Lactobacillus sp.* and *Enterococcus sp.* on Silaging and Aerobic Stability

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Keywords: silage, aerobic stability, *Lactobacillus, Enterococcus*, lactic acid bacteria

Objectives

To improve the aerobic stablility and general quality characteristics of silages through a silage starter culture containing a mixture of *Lactobacillus sp.* and *Enterococcus sp.*.

Introduction

A high quality silage is characterised by a low pH, a low content of ammonia-N and butyric acid. The aerobic stability is also an important factor in practice, especially when silage silos are left open for sometime before feeding. Many silages immediately start to deteriorate by exposure to air. The activity of spoilage microorganisms leads not only to nutritive losses resulting from an oxidation of the lactic acid and the watersoluble carbohydrates, it can further end up in formation of mycotoxins which can be potentially lethal. By addition of special starter cultures the products of silage fermentation can be influenced.

Experiment and Methods

ENSILING PROCESS

By use of sterile material for ensiling the fermentation process is initiated only by the bacterias from the inoculum additive. This enables the investigation of the effects of these microorganisms on the fermentation pattern. As material for testing the inocula two different substrates were prepared. Ground maize kernel with a DM of about 630 $g*kg^{-1}$ was radioactive irradiated (cobalt) and a part of it was mixed with natural material to get a concentration of lactic acid bacteria (LAB) of about 10^2 g^{-1} fresh mass (FM). Silages were made from material inoculated with the starter culture as well as uninoculated for a control (sterile control - SC, mixed control - MC; sterile inoculated - SI, mixed inoculated - MI). The natural microflora in the mixed silage material therefore competes with the added inoculum LAB for nutrients during the fermentation. The experiment was planned for laboratory silos in a scale of 6.5 L. The compressing of the silage was done with a pneumatic press. To get information at different fermentation stages for each sampling day (day 8 and 39) 2 silos were prepared.

ANALYTICAL DETERMINATIONS

An extract was made by use of a laboratory homogeniser. Distilled water was added to the samples in a relation of silage material FM to distilled water of 1 to 5. From the aqueous solution all the chemical analyses were done.

Sugars and organic acids were analysed by HPLC (HP 1050C, Column: Merck Polyspher OA KC and RI Detector - HP1047). For the measurement of ammonia-N, a distillation method was applied.

AEROBIC STABILITY

To determine aerobic stability of the silages, a method by Honig (1990) was modified and implemented. It is based on monitoring temperature which rises due to increased microbial activity of samples exposed to air.

Results and Discussion

In silage SI the sugars were converted relatively slowly mainly into lactic acid (see Table 1). In all other samples even in the sterile control (SC) more acetic acid could be found. Higher ammonia-N values could be observed in the silages mixed with the non-radiated maize material. This confirms, that the naturally occurring microorganisms in the maize material are responsible for formation of increased ammonia-N and acetic acid during the fermentation process.

Also the fermentation losses at day 39 in silage SI were 1,2 g/kg FM lower than in silage MI.

Already at day 39 an improvement of the aerobic stability due to inoculation with the starting culture from 30 to 50 h could be reached. This results can be seen in Table 2 by comparing the mixed control (MC) with the mixed inoculated silage (MI).

Tab 1: Fermentation pattern till day 39

	Silage	0 days	8 days	39 days
Glucose [g kg-1DM]	SC	11,2	13,8	15,5
	MC	11,2	3,1	1,6
	SI	11,2	10,4	0,1
	MI	11,2	2,0	0,4
Lactic acid [g kg-1DM]	SC	1,9	4,0	4,0
	MC	1,9	8,0	10,2
	SI	1,9	11,6	19,1
	MI	1,9	11,1	12,3
Acetic Acid [g kg-1DM]	SC	0,7	1,5	1,5
	MC	0,7	3,1	3,7
	SI	0,7	1,0	0,9
	MI	0,7	3,9	3,6
Ammonium-N [g kg-1DM]	SC	0,15	0,26	0,40
	MC	0,15	0,29	0,49
	SI	0,15	0,27	0,41
	MI	0,15	0,32	0,46
pH	SC	5,44	5,17	4,79
	MC	5,44	4,47	4,00
	SI	5,44	4,29	3,53
	MI	5,44	4,31	3,87

Tab. 2: Aerobic stability of the maize silages after 39 days of fermentation

Silages	SC	MC	SI	MI
time stable in h	25	30	44	50

Conclusions

In summery it can be stated that the silage starter culture improves silage quality and the aerobic stability. The main effects are the increased production of lactic acid, a better conversion of sugars and a lower pH value, which are responsible for a better aerobic stability.

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Inoculation with *Lactobacillus buchneri* improves the aerobic stability of barley silage.

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INTRODUCTION

Poor packing, leaky silos, and insufficient removal of silage during feeding can result in poor aerobic stability of silages. Buffered propionic acid-based products and ammonia have been used to some success but propionic acid is costly and ammonia is difficult to handle. Microbial inoculation is relatively inexpensive and safe to work with, and inoculation with homolactic acid bacteria has improved silage quality. However, aerobic stability has often been worse with homolactic fermentations. *Lactobacillus buchneri* (Lb) has been used specifically to improve the aerobic stability of silages. Research has shown that Lb can improved the aerobic stability of corn silage (Driehuis et al., 1996; Ranjit et al., 1998). The present study was designed to determine the effect of varying doses of Lb, a commerical inoculant (homolactic acid bacteria and *Propionibacteria*), and a buffered propionic acid-based product on the fermentation and aerobic stability of barley silage. Carry-over effects of the treated barley silage on the aerobic stability of total mixed ration was also determined.

MATERIALS AND METHODS

Whole plant barley (39.4% DM) was ensiled in triplicate 20-L laboratory silos after the following treatments: 1) Untreated; 2) 1 x 10^5 cfu of Lb/g of fresh forage (LB); 3) 5 x 10^5 cfu Lb/g (LB5); 4) 1 x 10^6 cfu Lb/g (LB10); 5) Biotal Plus (BP; *Lactobacillus, Pediococcus* and *Propionibacterium*), 1.1 x 10^5 cfu/g (BP); and 6) TMR-Mate™ (a buffered propionic acid-based product containing buffered propionic acid, acetic acid, benzoic acid, and citric acid; 56% active ingredients; Cargill, Inc., Minneapolis, MN), 2 g/kg of fresh forage. After 69 days, silos were opened and the contents of each silo thoroughly mixed. Silage was analyzed for products of fermentation. Four kilograms each of silage was returned to its original silo and incubated aerobically at 22^{0}C. In addition, a total mixed ration (TMR) was made from each silage that contained 30% (DM basis) barley silage, 30% alfalfa silage (untreated), and 40% of a dairy concentrate. Four kilograms of the TMR were placed in silos to aerobically deteriorate. Aerobic stability was defined as the number of hours before a 2^{0}C rise in temperature.

RESULTS AND DISCUSSION

The composition of silage after 69 days of fermentation is show in Table 1. Dry matter recovery after ensiling was greatest for silage treated with a mixed microbial inoculant (BP) and least for silage treated with the intermediate amount of Lb (LB5). Inoculation with BP also resulted in reduced ($P < 0.05$) concentrations of ethanol, ammonia, butyric acid, and water soluble carbohydrates but increased ($P < 0.05$) concentration of lactic acid when compared to untreated silage. However, BP had no effect on propionic acid concentration suggesting that added *Propionibacteria* were ineffective.

Silages treated with Lb had markedly greater ($P < 0.05$) concentrations of acetic and propionic acids when compared to untreated silage. The concentration of lactate in silages treated with LB5 and LB10 were numercially, but not statistically lower than in untreated silage but LB silages had greater ethanol concentrations than did untreated silage. Inoculation with LB resulted in lower ($P < 0.05$) concentrations of residual water-soluble carbohydrate concentrations than in untreated silage.

All silages had extremely low (< 1,000 cfu/g) numbers of yeast. When exposed to air, control silage was stable for 377 hours (Table 2). Treating silage with increasing doses of Lb improved aerobic stability of silages. Silage treated with the highest level of Lb (LB10) was stable for more than 760 hours. The aerobic stability of silage treated with BP was numercically the lowest

among the treatments (155 hours). Silage treated with a buffered propionic acid additive (TM) was stable for 621 hours.

When barley silages were mixed into a TMR with alfalfa silage (5.91 log cfu yeast/g) and a dairy pellet, the stability of the TMR was less than of silage alone (Table 2). Improvements in aerobic stability from LB were small and not as dramatic as in the silage alone.

CONCLUSIONS

Treating silage with an additive containing homolactic acid bacteria and *Propionibacteria* improved silage fermentation but not the aerobic stability of silage. Inoculation with LB resulted in more acetate and propionate production resulting in improvements in aerobic stability. Treating silage with a buffered propionic acid preservative also improved the aerobic stability of barley silage.

Table 1. Chemical and microbial composition of barley silage after 69 days of ensiling.

Treatment	DM	DMR[1]	pH	Yeast	Acetate	Propionate	Butyrate	Lactate	Ethanol	NH_3-N[2]	WSC[3]
	%	%		log cfu/g	% of DM						
Control	35.7^{cd}	92.9^{ab}	4.70^{a}	2.89	1.27^{f}	0.06^{d}	0.64^{a}	6.03^{b}	0.83^{b}	0.314^{a}	1.06^{a}
LB	34.6^{d}	91.8^{bc}	4.46^{b}	2.36	4.08^{c}	0.77^{a}	0.02^{b}	6.40^{b}	1.37^{a}	0.256^{ab}	0.33^{c}
LB5	37.3^{b}	89.5^{c}	4.46^{b}	2.01	4.45^{b}	0.45^{c}	0.02^{b}	5.19^{b}	1.52^{a}	0.230^{ab}	0.29^{c}
LB10	36.6^{bc}	92.6^{ab}	4.46^{b}	1.18	4.77^{a}	0.61^{b}	0.01^{b}	5.54^{b}	1.56^{a}	0.260^{ab}	0.33^{c}
BP	39.5^{a}	95.2^{a}	4.12^{d}	2.42	1.85^{e}	0.01^{d}	0.01^{b}	9.20^{a}	0.32^{c}	0.193^{b}	0.71^{b}
TMR-mate	37.7^{b}	91.7^{bc}	4.32^{c}	2.77	2.55^{d}	0.39^{c}	0.03^{b}	9.39^{a}	0.52^{bc}	0.303^{a}	0.43^{c}
SE	0.2	0.6	0.03	2.56	0.02	0.03	0.03	0.41	0.09	0.019	0.03

[a, b, c, d, e, f]Means in columns with unlike superscript differ ($P < 0.05$).

[1]DM recovery.

[2]Ammonia nitrogen.

[3]Water soluble carbohydrates.

Table 2. The aerobic stability (hours) of silage and total mixed ration (TMR).

Item	Control	LB	LB5	LB10	BP	TMR-mate	SE
Silage	377^{bc}	588^{ab}	683^{a}	$>760^{a}$	155^{c}	621^{ab}	63
TMR	39^{ab}	41^{ab}	46^{a}	48^{a}	35^{b}	41^{ab}	2

[a, b, c]Means in rows with unlike superscript differ ($P < 0.05$).

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The effect of sodium benzoate based additives on quality and storage stability of whole crop silage

INTRODUCTION

Whole crop silage is usually made of grain or legume grain mixtures, harvested at dough stage of grain. Whole crops of barley, wheat and oats are normally easily ensiled but the silage quality and aerobic stability are often unsatisfactory. The reason may be that the material remains coarser than grass, resulting in conditions that the incoming oxygen is quickly distributed within the silo. Therefore lactic acid fermentation is delayed and heating processes may start in the silage. Due do this and several other factors clostridial fermentation may easily occur as well. Several research workers (Weisbach, F. et al., 1988; Andersen, P. et al., 1987; Lomstein, E. et al., 1992) studied the ensiling of whole crop cereals and concluded that the risk that clostridial fermentation may occur in such silage is high if no additives are used.

The aim of this study was to explain the effect of sodium benzoate based additives on the quality and storage stability of whole crop silage.

MATERIALS AND METHODS

A pilot-silage trial was carried out at silage laboratory. The silage crops used was barley, oats and pea mixture, harvested at the dough stage of barley and milky stage of oat grain (cut on 5 August). The silage crop contained about 65% of barley, 25% of oats and 10 % of peas. The chemical composition of silage material was as follows: dry matter (DM) 302 g kg^{-1}, crude protein (CP) 103 g kg^{-1} DM, crude fibre (CF) 305 g kg^{-1} DM, crude ash 62 g kg^{-1} DM and water soluble carbohydrates (WSC) 82 g kg^{-1} DM.

The mixture was cut with a scythe, thereafter chopped in a chopper to 4-8 cm chop-length and ensiled in 3 litre glass jars. Chemical additives used were Superben and Niben, each added at application rate of 5 l ton^{-1} fresh matter (FM). Both additives were also used in combination with biological additive Silomeister (2.5 + 2.5 l ton^{-1} FM). Superben is based on sodium benzoate and hexamine. Niben is based on sodium benzoate and sodium nitrite. A total of 5 treatments with 3 replicates were ensiled. Glass jars were sealed with 4 layers of plastic film (0,025 mm thickness) to imitate conditions in big bale silage. Silos were stored at 18-25 ° C for 120 days.

RESULTS AND DISCUSSION

The results of the chemical and microbiological analyses are presented in Table 1. The results expressively showed how low silage quality might be obtained when no additives were used. In untreated silage the concentration of butyric acid was extremely high. With other accompanying negative fermentation products such silage is not likely to be consumed by ruminants. Considering relatively high WSC concentration of crop a good fermentation quality was expected. However, clostridia, enterobacteria or other detrimental microorganisms developed and initiated proteolytic processes in the silage.

The use of additives considerably improved fermentation. When Superben was used the silage quality still remained unsatisfactory. A good silage quality with minimal losses was only obtained with Niben. Both chemical additives, particularly Niben, were more effective when used in combination with the inoculant. Added lactic acid bacteria (LAB) probably created a faster fermentation and thereby a faster drop in pH.

All treatments had a good aerobic stability (> 7 days, data not shown). However, that is not relevant in clostridial fermented silage. Such silage with high volatile fatty acid (VFA) concentration is usually stable.

Table 1. The effect of additive treatment on quality of whole crop silage (barley/oat/pea =65/25/10).

Analyses	Untreated control	Superben (5 l/ton FM)	Niben	Nib.+Silo. (2.5 + 2.5 l/ton FM)	Sup.+Silo.	$LSD_{0.05}$
Dry matter, g/kg	284	272	286	292	287	
pH	5.2	4.6	4.2	3.7	3.7	0.1
Ammonia-N, % of TN	21.8	15.2	8.5	8.5	14.0	3.4
Acetic acid, g/kg DM	13	12	14	13	13	4.8
Butyric acid, g/kg DM	49	14	0	0	0	5.8
Ethanol, g/kg DM	21	19	5	9	13	2.8
Butanediole, g kg DM	9	2	0	1	2	1.2
Clost. spores, /g FM	8100	1800	120	6	450	
Moulds, cfu/ g FM	13500	1600	120	40	45	
DM losses, g/kg DM	28	12	5	5	13	3.4

[a] Least significant difference at the 5% probability level (n=3).

CONCLUSIONS

The results showed that when making silage from whole crop cereals and fermentation conditions are difficult, such as in big bale, a high silage quality cannot be guaranteed without using effective additives. Both additives, Superben and Niben are based on sodium benzoate and improved fermentation. Niben was more effective against clostridial activities. The use of Niben resulted in good quality silage with minimal dry matter losses. Niben was also effective in combination with the biological additive Silomeister.

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Comparison of homofermentative and heterofermentative Lactobacillus Strains as Silage Inoculum to improve Aerobic Stability

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Key words: aerobic stability, silage, lactic acid bacteria, homofermentative, heterofermentative

Introduction

Various products for improving silage fermentation are present on the market. Most of these commercially available inoculates are based on homofermentative lactic acid bacteria strains (LAB). The main advantage of homofermentative strains is their ability for fast and efficient lactic acid production which results in a rapid decrease in pH. This does not necessarily relate to aerobic stability of the silage during the unloading (feeding) period.
Therefore a comparison of homofermentative and heterofermentative LAB as silage inoculum and their effect on the fermentation process and aerobic stability is investigated in this study.

Materials and Methods

SCREENING OF BACTERIAL CULTURES
Different grass silage provided by farmers from the area of Lower Austria, were used for the isolation of LAB strains. The isolated strains where cultured on MRS-medium under aerobic conditions. HPLC (HP 1050C, Column: Merck Polyspher OA KC and RI Detector - HP1047) was used for analysing the fermentation products.
Two strains (HO 268, HO 147) which mainly produced lactic acid were selected as homofermentative bacteria. As heterofermentative LAB two strains (HE 101, HE 219) were chosen, which produced lactic acid, acetic acid and some ethanol. The selected bacteria were fermented, harvested, lyophilised and used as silage inoculum in the ensiling experiments.
ENSILING PROCESS, ANALYTICS AND AEROBIC STABILITY
Chopped maize plants were treated with either one of the lyophylisated organisms in a concentration of about $5*10^7$ LAB/g FM (fresh mass) and one lot was ensiled untreated as a control. All samples were ensiled in duplicate. There were performed silos in a laboratory scale with 1 l (day 1, 4 and 7) and 6.5 l (day 32). The chemical analyses were performed on an aqueous extract of the silage by HPLC on day 1, 4, 7 and 32. Evaluation of the aerobic stability of the silage samples at day 32 was done using a method by Honig (1990).

Results and Discussion

The addition of different LAB strains as an inoculum for silage production could improve the decrease in pH in all inoculated silages compared to the control up to day 7. The relatively high pH-value at day 7 in the control might be due to the low amounts of lactic acid and the significantly high prduction of ethanol. At day 32 the pH-value of the control silage was decreased very rapidly and reached the same level as the inoculated silages except the sample HE 219, where the pH was increasing. This is due to the shift from lactic acid to acetic acid which have different pK_a values (pK_a of lactic acid is 3,87, pK_a of acetic acid is 4,76).

Some differences in the concentration of lactic acid were found as expected. The concentration of lactic acid was obviously higher in silage inoculated with LAB compared to the control silage at day 7. One exception was the HO 147 which was not able to convert all the sugar. At day 32 the highest amount of lactic acid was found in the silage treated with the homofermentative strain HO 268. An interesting point is the decrease of lactic acid and increase of acetic acid concentrations in silage HE 219 from day 7 to day 32. High amounts of acetic acid were also found in HE 101 (Table 1). However, alomost no difference in the concentration of propionic acid was observed.

Tab. 1: pH-value and production of organic acids in maize silage in laboratory scale when inoculated with different LAB.

		organism				
PARAMETERS	time [day]	C	HO 268	HO 147	HE 101	HE 219
GLUCOSE [g/kg]	1	21,7	26,1	32,0	33,9	31,7
	4	2,6	0,7	13,0	3,1	4,1
	7	0,3	0,4	5,5	0,5	0,4
	32	0,1	0,7	0,2	0	0,1
LACTIC ACID [g/kg]	1	6,3	11,1	3,9	1,3	3,1
	4	17,3	40,4	19,8	25,0	22,1
	7	23,7	45,5	21,5	28,4	24,9
	32	39,7	44,2	35,1	33,7	3,3
ACETIC ACID [g/kg]	1	3,8	2,9	2,7	1,0	2,0
	4	9,0	5,5	11,3	10,9	11,7
	7	10,6	7,1	10,4	12,5	15,0
	32	11,3	9,3	12,2	28,3	51,0
PROPIONIC ACID [g/kg]	1	0,9	0,8	0,7	0,3	0,5
	4	0,6	1,1	0,6	0,3	0,5
	7	0,6	1,1	0,6	0,4	0,5
	32	0,4	1,4	0,7	0,5	1,0
ETHANOL [g/kg]	1	3,3	2,4	1,8	1,3	1,4
	4	10,6	9,2	3,8	7,1	4,7
	7	14,4	9,4	3,7	8,2	5,4
	32	11,2	7,6	3,6	7,7	7,2
pH-value	1	5,43	5,64	5,34	5,64	5,46
	4	4,22	3,92	3,99	3,92	4,02
	7	4,13	3,93	3,92	3,93	3,95
	32	3,74	3,85	3,87	3,73	4,11
AEROBIC STABILITY [d]	32	18	14	15	40	70

C...control, HO 268...homofermentative strain, HO 147...homofermentative strain, HE 101...heterofermentative strain, HE 219...heterofermentative strain

The aerobic stability on day 32 was 70 h for HE 219, which was the highest, and 40 h for HE 101. The control silo had an aerobic stability of 18 h. The stability of HO 268 was only 14 h, and the one of HO 147 was 15 h .

Conclusion

A high concentration of lactic acid did not manifest a positive effect on the aerobic stability. The best aerobic stability was found in the silage with the highest concentration of acetic acid (HE 219 and HE 101). The concentration of acetic acid in the control silage is compareable with the concentration in the silages inoculated with homofermentative strains (HO 268 and HO 147). Therefore acetic acid seems to be an important factor influencing the aerobic stability of silage.

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Factors affecting bunker silo densities.

Introduction

Attaining a high density in a silo is important for two primary reasons. Firstly and most importantly, density and dry matter content determine the porosity of the silage. Porosity, in turn, sets the rate at which air can move into the silo and subsequently the amount of spoilage that can occur during storage and feedout. Secondly, the higher the density the greater the capacity of the silo. Thus, higher densities generally reduce the annual cost of storage per ton of crop by both increasing the amount of crop entering the silo and reducing crop losses during storage. The factors affecting density in bunker and pile silos are not well understood. General recommendations have been to spread the crop in 15 cm layers and pack continuously with heavy, single-wheeled tractors. In a survey of lucerne silage in 25 bunker silos, Ruppel et al. (1995) found that tractor weight and packing time (min/t as fed or min/m^2) were the most important factors affecting density. However, both factors only explained a small fraction of the variation observed, and layer thickness was not measured. The objectives in our study were to measure density in a wider range of bunker silos and correlate those densities with filling practices.

Methods

Twenty collaborating county extension agents in Wisconsin measured densities in over 160 bunker silos containing either maize or haycrop (largely lucerne) silage. Density was measured with a 5-cm diameter corer, taking cores at approximately chest height at four locations across the silage face. Core depth, distance from the top and distance from the floor were recorded. Cores and a grab sample were express mailed to the Center for determination of weight, dry matter content and particle size distribution. A survey was filled out for each silo sampled. Information requested from farmers included: number of packing tractors, tractor weight, number of tires per tractor, tire pressure, tire condition, number of drive wheels, silage delivery rate, packing time per day, harvest time per day, filling time, filling technique, initial layer thickness, silo dimensions, maximum silage height, crop, crop maturity, and theoretical length of cut. These factors were then correlated with measured dry matter densities.

Results

The range of densities and dry matter contents observed in haycrop and maize silages are shown in Table 1. Ranges of dry matter densities were similar for both haycrop and maize silages. Densities on the low end suggested little packing whereas the highest densities were in the range observed in tower silos. Average dry matter densities were slightly higher than a recommended minimum density of 225 kg/m^3.

Densities were positively correlated with the height of silage above the core, indicating the effect of self-compaction in bunkers. To put densities on a common basis, all densities were adjusted to the median depth below the surface (2.16 m) using eq. 15 of Pitt (1983) and assuming a compressibility of 1.5 x 10^{-5} Pa^{-1}. Adjusted dry matter density was positively correlated with average packing tractor weight (W; kg), packing time (T; number of tractors x h/t as fed), and dry matter content (D; g/kg). Density was inversely correlated with the initial

depth of the crop when spread in the silo (L; cm). These factors combined together as follows:

$$\frac{W}{L}\sqrt{T \cdot D}$$

explained 18% of the variation (Fig. 1). Use of rear duals or all duals on packing tractors as shown in Fig. 1 had little effect on density. Other factors such as tire pressure, crop and average particle size were not significantly correlated with density. Thus the low r^2 of the four-parameter packing factor probably reflects variability in accurately estimating parameters such as initial depth of the crop and packing time per tonne rather than missing factors important to determining density.

One practical issue raised in the study was packing time relative to crop delivery rate to the silo. Packing time per ton was highest (1 to 4 min/t as fed) under low delivery rates (<30 t as fed/h) and generally declined with increasing delivery rate. Packing times were consistently less than 1 min/t as fed at delivery rates above 60 t/h in our survey. These results suggest that farmers using contractors for harvesting their silage crops probably will need to pay particular attention to spreading the crop in a thin layer and would benefit from using several packing tractors simultaneously.

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Table 1. Summary of core samples collected from 168 bunker silos.

	Haycrop Silage (87 silos)			Maize Silage (81 silos)		
Characteristic	Average	Range	Std. Dev.	Average	Range	Std. Dev.
Dry Matter, %	42	24-67	9.50	34	25-46	4.80
Wet Density, kg/m^3	590	210-980	175	690	370-960	133
Dry Density, kg/m^3	237	106-434	61	232	125-378	46
Avg. Particle Size, mm	11.7	6.9-31.2	3.8	10.9	7.1-17.3	2.0

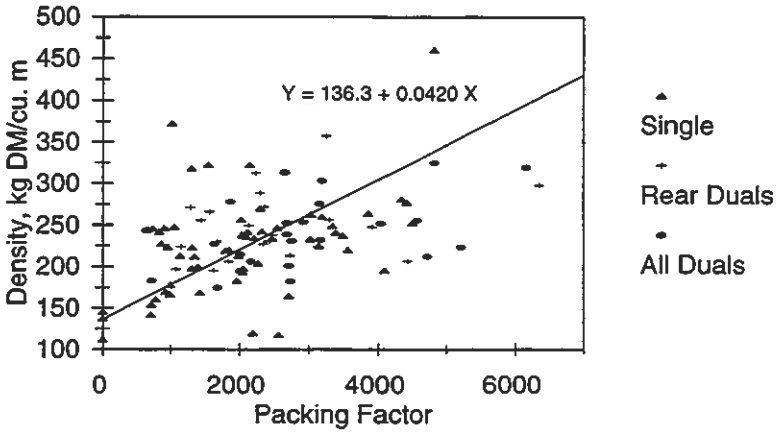

Fig. 1. Adjusted dry matter density as related to the packing factor (W/L√TD) and use of duals on packing tractors.

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The effect of fermentation quality on the aerobic stability of direct cut or slightly prewilted grass silage

Introduction

The exposure of silage to air after opening of the silo and during feeding is occasionally accompanied by aerobic deterioration of silage, resulting in economic losses due to DM loss and poorer feeding quality. Aerobic deterioration caused by the proliferation of microflora, which has remained dormant under anaerobic conditions, is characterised by rise in temperature and pH, and oxidation of the fermentation end-products (Bolsen et al. 1996). The ability to estimate the risk for aerobic spoilage of grass silage according to standard fermentation quality is still very uncertain, although it could be a valuable tool on commercial farms. The aim of this work was to study the effect of silage additives on aerobic stability and find out microbiological and chemical parameters which may predict the aerobic deterioration of grass silage.

Methods

Data (N = 338) used in this study was collected from 11 silage experiments done during the years 1994-1997. All silages were made from timothy (*Phléum pratense*) and meadow fescue (*Festuca pratensis*) mixed herbage. Direct cut (4 trials) or slightly prewilted grass (7 trials) was ensiled in laboratory scale (5-15 kg) or small experimental scale (500-1000 kg) silos. The treatments were divided into three groups: control (C), formic acid (FA, 4 g/kg) and lactic acid bacteria inoculation (I, 10^6 cfu/g). Microbiological and chemical composition and ten-day aerobic stability after at least 90 days of fermentation of the silages were determined according to Rauramaa et al. 1989. The chemical and microbiological quality and aerobic stability were tabulated across the treatments. On the second stage the samples were classified into three groups: aerobically stable (Σ°C<10), slightly deteriorated (Σ°C 10-20) and deteriorated (Σ°C >20) after 5 days of air exposure. The quality parameters were then calculated across these three groups. The results were calculated with least squares analysis of variance (MINITAB/GLM-procedure) using treatment or stability group and number of experiment as factors in the linear model.

Results and discussion

On average, all the treatments resulted in relatively good fermentation quality in the respective silages (table 1). Typical and significant differences existed in pH and in ammonia-N, organic acid and residual sugar content between C, FA and I silages. The results show however, that a good fermentation quality of silage during the silo-opening does not always prevent aerobic deterioration. I silages were most sensitive to aerobic spoilage and FA silages were most stable during air exposure, C silages being intermediate. The grouping of silages according to stability revealed that low pH, ammonia-N and residual sugar content and higher lactic acid content were significantly associated with poorer aerobic stability after silo-opening. The microbiological quality of silages didn't differ significantly between the stability groups, although there was a very slight tendency that deteriorated silages contained more yeasts and molds than stable silages. According to this data it can be simplified that good quality I and C silages may be aerobically instable, but FA prevents spoilage. However, this data set contained only quite low DM silages (mean DM 198 g/kg, range 128-314). Consequently, these results may not be applicable to more heavily prewilted silages.

Conclusions

In conclusion, well fermented low-DM or slightly prewilted silages preserved with inoculates or no additives seemed to be most prone to aerobic spoilage. It is difficult to find out parameters which specifically predict aerobic instability or stability. More work is needed to understand the aerobic spoilage of prewilted silages.

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Table 1. The effect of grass treatment on fermentation quality and aerobic stability of silages

		Treatment[2) 3)]				Significance
		C	FA	I	SEM	
N of samples		57	156	107	320	
DM	g/kg	216	222	218	36.2	ns
pH		4.2	4.1	4.0	0.48	**<0.001**
Ammonia-N	g/total N	84	48	64	23.7	**<0.001**
Ethanol	g/kg DM	18	16	15	14.7	ns
Acetic acid	g/kg DM	33	22	23	21.9	**<0.001**
Propionic acid	g/kg DM	2	1	1	3.2	**0.010**
Butyric acid	g/kg DM	2	1	0.2	6.2	**0.071**
Lactic acid	g/kg DM	72	59	78	39.5	**<0.001**
VFA/Lactic acid		1.3	0.6	0.6	2.33	**0.032**
Residual WSC	g/kg DM	15	35	20	38.8	**<0.001**
Enterobacteria	log cfu/g	0.97	1.23	1.18	1.564	ns
Yeasts	log cfu /g	1.76	2.28	2.21	2.541	ns
Moulds	log cfu /g	1.54	1.63	1.53	1.788	ns
Lactobacilli	log cfu /g	7.79	7.84	7.56	1.274	ns
Heterof. lactobacilli		6.67	6.90	5.64	2.520	**0.012**
Clostridia spores	log mpn/g	2.52	2.36	2.36	1.973	ns
Aerobic stability after[1)]						
3 days	Σ°C	3.3	2.1	5.4	5.47	**<0.001**
5 days	Σ°C	9.7	4.5	13.0	10.58	**<0.001**
7 days	Σ°C	17.3	8.2	21.5	15.84	**<0.001**
9 days	Σ°C	29.8	17.6	36.4	23.87	**<0.001**

[1)] Determined as sum of temperature rise (sample-ambient temperature)
[2)] Grass treatments; C = control, FA = formic acid (4 g/kg), I = lactic acid bacteria inoculation (10^6 cfu/g)
[3)] Results presented as LS-means

Table 2. Fermentation quality of silages according to aerobic deterioration group.

		Deterioration group[1) 2)]				Significance
		<10 Σ°C	10-20 Σ°C	> 20 Σ°C	SEM	
N of samples		192	79	49		
DM	g/kg	223	214	209	2.1	**<0.001**
pH		4.2	4.0	3.9	0.31	**<0.001**
Ammonia-N	g/total N	59	73	55	3.0	**0.001**
Ethanol	g/kg DM	17	16	15	6.8	ns
Acetic acid	g/kg DM	26	26	23	20.2	ns
Propionic acid	g/kg DM	1.2	1.3	1.1	2.36	ns
Butyric acid	g/kg DM	0.9	1.6	1.1	4.10	ns
Lactic acid	g/kg DM	62	80	79	29.0	**<0.001**
VFA/lactic acid		0.82	0.80	0.68	1.368	ns
WSC	g/kg DM	31	17	14	34.5	**<0.001**
Enterobacteria	log cfu/g	1.13	1.13	1.38	1.449	ns
Yeasts	log cfu/g	2.01	2.44	2.31	2.781	ns
Moulds	log cfu/g	1.54	1.62	1.79	1.759	ns
Lactobacilli	log cfu/g	7.82	7.56	7.81	1.265	ns
Heterof. lactobacilli	log cfu/g	6.75	6.39	6.10	2.212	ns
Clostridia spores	log mpn/g	2.55	2.03	2.20	1.710	ns

[1)] Deterioration groups; aerobically stable (Σ°C<10), slightly deteriorated (Σ°C 10-20) and deteriorated (Σ°C >20)
[2)] Results presented as LS-means

Aerobic stability of pea-wheat bi-crop silages treated with different additives

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Introduction

The high buffering capacity and low water soluble carbohydrate content of legumes makes them difficult to conserve as silage (Cussen *et al.*, 1995). Legume silages are therefore less stable when exposed to air. Although different additives are being used for improving the fermentation and quality of grass and whole crop silages, little is known of their suitability for bi-crop silages. These studies compares the effect of using microbial, formic acid, quebracho tannins or a sulphite-salt based additives on the aerobic stability of pea-wheat bi-crop silages.

Materials and methods

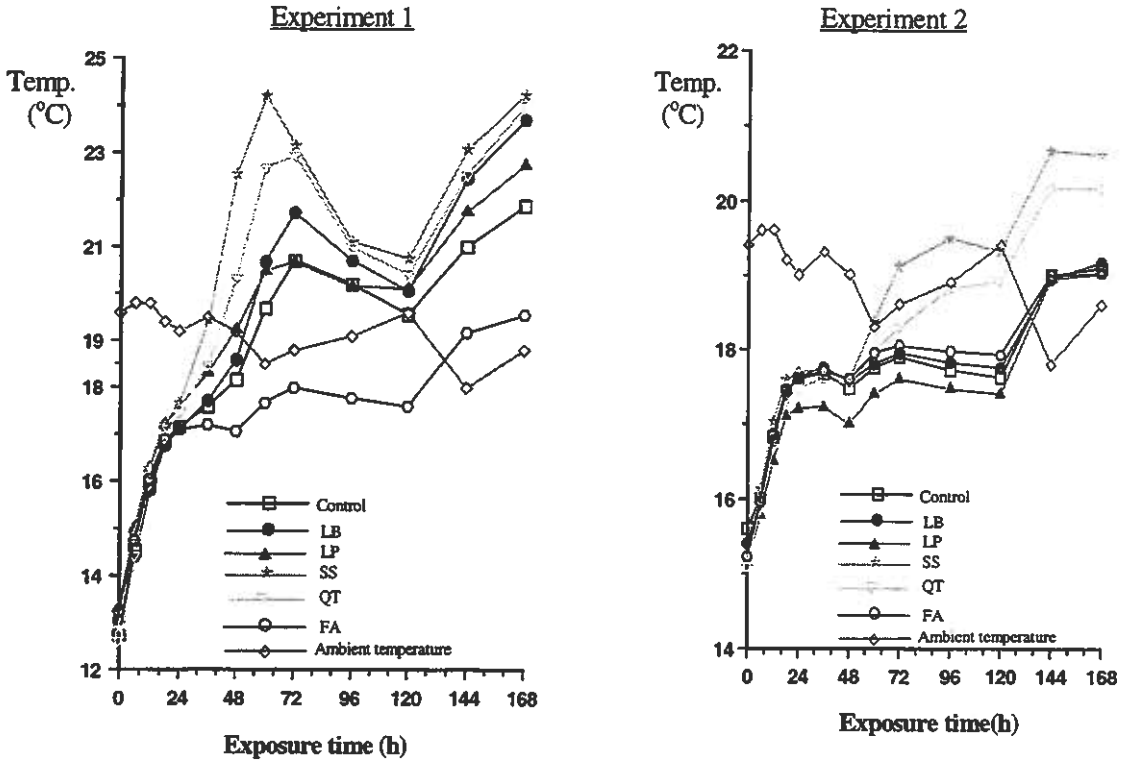
Two studies were conducted with pea-wheat bi-crop silages. The ratio of peas to wheat was 3:1 in experiment 1 and 1:3 in experiment 2. Both ratios of bi-crops were harvested at 320 g dry matter (DM)/kg when, the peas and wheat had reached growth stages 209 (yellow wrinkle pods) and 79 (Late milk to early dough stage) respectively. The bi-crops were treated with two lactic acid based bacterial inoculants (*Lactobacillus buchneri* [LB; 10^5 CFU/g fresh weight; FW] or *Lactobacillus plantarum* [LP; 10^6 CFU/g FW]), sulphite salts (SS; 1ml sulphite solution/kg FW), quebracho tannins (QT; 16 g/kg FW) and formic acid (FA; 2.5g/kg FW). The silages are prepared in laboratory silos made of polyethylene bags and each treatment was replicated 6 times. Aerobic stability was measured by changes in silage temperature following exposure to air for 7 days.

Results and discussion

The silages from the 2 experiments appeared to ferment well. The overall average pH was 4.0 and 4.1 in experiments 1 and 2 respectively. Irrespective of additive treatment, all the silages in experiment 1 with the high pea ratio were less stable than those in experiment 2 (Figure 1). Except for the FA-treated silage that was stable for over 5 days in experiment 1, the temperature of all the other silages rose above the ambient temperature within 2 days. In experiment 2, only SS-treated silages had temperature rise that was above ambient within 3 days. Other silages including the control were aerobically stable for 5 days and none reached a temperature higher than 25 °C after 7 days of exposure to air. In experiment 2, LP-treated silage had the best aerobic stability. Formic acid and LP may have delayed the onset of aerobic deterioration because of a postulated rapid reduction of pH during fermentation and maintenance the same when exposed to air. Such rapid reduction in pH may suppress the buffering effect of the legumes and delays the growth of undesirable microbes like yeasts, moulds and *Bacillus* spp. that are responsible for aerobic deterioration in silage. Surprisingly, in both experiments, control silages were more stable than LB, SS or QT-treated silages. Although SS has been reported to enhance aerobic stability of grass and maize silage

(Hill, *et al* 1996), its' ineffectiveness in this study may indicate that it is less suitable for pea-wheat silages.

Figure 1: Aerobic stability of silages treated with different additives



Conclusions

These studies showed that additive treatment of pea-wheat bi-crop silages may not be necessary for producing a stable product provided good fermentation is ensured by rapid silo filling and good consolidation. Of the additives evaluated, FA and LP appeared to be the best for preventing aerobic spoilage in pea-wheat bi-crops. The results also showed that higher levels of legumes like peas in a bi-crop mixture might reduce the aerobic stability of cereal-legume bi-crop silages.

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Influence of pre-wilting degree on aerobic stability of grass silages

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Introduction

Aerobic deterioration of silages is one of the main problems in silage making. Over the last years the silage quality was improved by reducing the bad fermentations. But by preventing butyric acid fermentation and restricting acetic acid formation the risk of aerobically unstable silages increase (Weissbach, 1996; Wyss, 1995). Furthermore, with increasing pre-wilting degree, it is more difficult to consolidate the grass and to reduce the gas exchange. In this way air infiltration, which is the predominant cause of aerobic instability, is facilitated. The objective of the present study was to investigate the influence of the pre-wilting degree on the silage quality and especially on the aerobic stability.

Material and Methods

In 1998 grass of a third and fourth cut was ensiled with five different dry matter contents. The dry matter contents varied between 22 and 68 % for the third cut and all treatments were ensiled the same day. For the fourth cut the DM varied between 29 and 68 % and here the highest pre-wilting degree was ensiled one day later. As material we used forage of a sown meadow, which contained ryegrass, white and red clover. The proportion of clover was about 60 %. The grass was short chopped and ensiled in polyethylene bags with a volume of 30 litres. Two silos per treatment were filled.

Dry matter (DM), ash, crude protein, crude fibre and sugar were determined. After a storage period of 90 days, the silos were opened and again, nutrient contents in addition to fermentation parameters were analysed at filling. Furthermore, aerobic stability was measured by placing silage samples in containers with holes and by monitoring the temperature. Four repetitions were filled per treatment and silo. The containers were stored at room temperature (approx. 20° C). Aerobic stability was defined as the period needed to increase the temperature to 1° C above ambient. In addition to temperature, pH-values were analysed after 3, 7, 10 and 14 days.

Results and Discussion

The nutrient contents of the fresh grass of the third cut were: 111 g ash, 188 g crude protein, 220 g crude fibre and 72 g sugar per kg DM. For the fourth cut the contents were: 96 g ash, 195 g crude protein, 192 g crude fibre and 104 g sugar. The consolidation varied between 114 and 200 kg DM per m^3. The treatments with the lowest pre-wilting degree had the lowest consolidation per kg DM. The highest consolidation was found at about 50 % DM. According to Honig (1994) a consolidation around 250 to 300 kg DM/m^3 is needed to limit air infusion.

The intensity of the fermentation is strongly influenced by the pre-wilting degree. With increasing DM-content higher pH-values were observed and the sum of acetic and propionic acid as well as the ammonia-N proportion decreased (Table 1). No or only small contents of butyric acid were found (< 2 g/kg DM). The silages with about 30 % DM had the highest lactic acid contents. No acid lactic was determined in silages with DM-contents higher than 50 %. Silages with the lowest DM-contents had the lowest sugar contents. According to the DLG evaluation scheme the silages with higher pre-wilting degrees showed lower scores. The high pH-values and the low acetic acid contents were responsible for this result. The question is, whether this scheme is suitable for judging the quality of silages with high DM-contents.

After opening the silos yeasts were also determined. The colony forming units of yeast varied between 10^2 and 10^5 per g. In general, silages with higher DM-contents showed a higher infection. Concerning aerobic stability, the silages were more unstable with increasing DM-content. An exception was the silage with the highest DM-content for the fourth cut. Here one repetition didn't become warm and the other only after 160 hours, but the temperature rise wasn't very high. Silages with very high DM-contents dry very fast, so that there isn't enough humidity for the development of the yeasts. The highest temperature rise was observed in the silages with about 50 % DM. The rise of the temperature caused a pH-riseThe high sugar content, the low acetic and propionic acid content and the low compaction are the main factors.

Table 1. DM-content, sugar content, fermentation parameters and aerobic stability of the silages of the third and fourth cut (average of two repetitions per prewilting degree)

DM	pH	Sugar	NH_3-N as % of	LA	AA+PA	DLG	Aerobic	Max.
%		g/kg DM	total N	g/kg DM	g/kg DM	scores	stability hours	Temp. diff. °C
21.4	4.8	12	13	69	24	77	226	3.5
31.2	4.9	15	11	104	16	77	211	4.9
42.5	5.2	56	5	69	8	63	208	5.9
55.3	5.6	97	3	0	1	50	113	14.0
67.6	5.6	96	2	0	1	49	121	4.3
27.8	4.8	13	11	196	27	77	198	7.5
39.0	5.2	54	7	109	12	65	175	9.1
47.1	5.6	91	4	21	7	54	152	16.7
54.5	5.7	93	3	0	2	48	122	6.2
65.8	5.7	91	2	0	1	46	292	1.4

LA: lactic acid; AA+PA: acetic and propionic acid
Temp. diff.: Difference of temperature to the ambient temperature

Conclusions

The results of these trials show that the grass silages with higher pre-wilting degree are aerobically more unstable in comparison to the silages with lower DM-contents. The high sugar content, as a result of a less intensive fermentation, the low acetic acid content, the yeasts as well as the consolidation, which wasn't high enough for the highest pre-wilting, degrees, are the main reasons for this results.

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Aerobic Stability of Silage Treated with Lactic Acid Bacteria

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The preservation of forage crops as silage depends upon the production of sufficient acid to inhibit activity of undesirable microorganisms under anaerobic conditions. When the silo is opened, aerobic conditions prevail at feeding time, the silage is subject to aerobic microbial growth and is therefore potentially unstable. Therefore, the prevision of aerobic deterioration of silage is important. In this experiment, tow strains of lactic acid bacteria (LAB) originally isolated from forage crops were used as silage additives, and their effect on silage fermentation and aerobic stability were examined.

MATERIALS AND METHODS

Alfalfa (*Medicaco sativa*) at flowering stage, Italian ryegrass (*Lolium multiflorum*) at flowering stage and sorghum (*Sorghum bicolor*) at milk stage were obtained from an experimental field at the National Grassland Research Institute (Tochigi, Japan) on June 1995 to October 1996. Silage were prepared using a small scale system of silage fermentation (Cai *et al.* 1997). Two LAB strains, *Lactobacillus casei* FG 1 and *L. plantarum* FG 10 isolated from forages were used as additives at 1.0×10^5 cfu/g of fresh matter (FM). The silos were kept at 25• and opened after 40 d of storage. The silage quality and changes in the counts of yeasts, pH and the contents of water soluble carbohydrates (WSC) and lactic acid were examined at 1, 3, 5 and 7 d of aerobic incubation at 25•.

RESULTS AND DISCUSSIONS

The strains FG 1 and FG 10 treatments in each of three silages were well preserved, which had significantly lower pH values, contents of butyric acid, propionic acid and ammonia nitrogen, gas production and dry matter loss, whereas significantly higher contents of residual WSC and lactic acid than the respective control silage. As shown in Table 1, yeast counts were high in all LAB treated silages and they increased rapidly during the aerobic exposure. As a results, these silages that spoiled upon aerobic

exposure faster than the respective controls. Most yeasts isolated from deteriorated silages showed high tolerance to lactic acid but low tolerance to butyric acid, and they were able to grow at low pH conditions and assimilate lactic acid.

TABLE 1. Changes in yeasts, water soluble carbohydrates, lactic acid, and pH during aerobic exposure of silage.[1]

Treatment	Yeast				Lactic acid				pH			
	1	3	5	7	1	3	5	7	1	3	5	7
	-- (log cfu/g of FM) --				-- (g/kg of FM)--							
Alfalfa[2]												
Control	ND[3]	ND	ND	3.0[b]	12.8[b]	12.6[a]	12.7[a]	11.4[a]	5.5	5.6	5.5[b]	5.7[b]
FG 1	4.3	5.3	8.8	8.1[a]	17.5[a]	9.5[b]	4.5[b]	3.4[c]	5.2	6.2	6.8[a]	6.3[a]
FG 10	3.8	6.7	9.0	9.3[a]	18.9[a]	10.3[b]	6.8[b]	6.8[b]	5.6	5.8	6.4[a]	6.6[a]
Italian ryegrass												
Control	2.3[b]	2.7[b]	3.5[b]	7.2[b]	30.6[b]	30.2	29.3[a]	25.5[a]	5.0	5.0[b]	5.1[b]	5.5[b]
FG 1	5.8[a]	7.8[a]	9.8[a]	8.5[ab]	36.8[a]	30.4	16.7[b]	8.8[b]	4.8	5.6[a]	6.2[a]	6.3[a]
FG 10	5.2[a]	8.2[a]	8.5[a]	9.3[a]	35.7[a]	32.1	15.2[b]	6.2[b]	5.1	5.8[a]	6.0[a]	6.3[a]
Sorghum												
Control	3.5[b]	4.4[b]	7.8	8.3	38.8	30.4	32.5[a]	20.2[a]	4.3	4.6[b]	5.0[b]	5.6[b]
FG 1	6.0[a]	10.2[a]	8.2	8.3	43.5	36.7	10.2[b]	4.2[b]	4.4	5.8[a]	6.7[a]	6.5[a]
FG 10	6.3[a]	9.8[a]	9.5	8.5	45.6	28.9	7.5[c]	3.6[b]	4.0	6.2[a]	6.4[a]	6.4[a]

[abc]Means in the same row within a silage type with different superscripts differ (P<0.05). [1]Values are means of three silage samples. [2]Fresh matter. [3]FG 1, *L. casei*; FG 10, *L. plantarum*. [4]Not detected.

The strains FG 1 and FG 10 used in this study were homofermentative lactobacilli and they are able to grow at low pH conditions. Inoculation with these strains may result in beneficial effects by promoting the propagation of LAB and by inhibiting the growth of clostridia and aerobic bacteria, as well as by decreasing the fermentation loss and improving silage quality. Most yeast strains isolated from deteriorated silage had a high tolerance to lactic acid, but low tolerance to butyric acid. These yeasts were able to grow at low pH conditions and utilize lactic acid and WSC for growth, but are inhibited by low concentrations of butyric acid and propionic acid. Results showed that the yeasts would grow vigorously after the opening of the silo and lead to the aerobic deterioration in the LAB treated silages. The relatively high level of butyric acid and propionic acid produced in the control silages could explain the great stability observed in these silage. The results confirmed that *L. casei* and *L. plantarum* improved fermentation quality but did not inhibit the growth of yeast and prevent aerobic deterioration of the silage.

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Aerobic instability - Effects and possibilities for its prevention.

Key words:
Aerobic stability, heating, losses, energy concentration, ensiling technology, additives

Situation

Increasing yield potential of ruminants and the tendency to feed silage all year round demand top quality silage of good stability after unloading. Whereas the fermentation quality has been improved markedly over the years, aerobic instability during feedout causes even growing problems, due to reduced palatability as well as energy content of the silage.

The microbial reasons for aerobic spoilage are well established. In all types of forage crops lactate assimilating yeasts can initiate instability upon exposure to air. In maize acetic acid bacteria can also play a complementary role in aerobic deterioration.

The degree of negative effects on forage quality and losses which occur under the influence of different technological factors are discussed.

Effects

Intensive aerobic decomposition is expressed by increasing heat development in the silage, marked losses of highly soluble nutrients and thus a strong reduction of the net energy content. The ratio between losses of DM and net energy ranges from 1 : 1.4 to 1 : 1.7.

In table 1 the effect of three important factors is shown:

- Stability of the silage under influence of air as influenced by fermentation quality. Higher stability gives the farmer more time during feed out.
- Depth of penetration of air as a consequence of the quality of filling and unloading technique. The looser the material the deeper air ingress is to be expected.
- Progression of the unloading face through the silo as an effect of the adaptation of silo width to herd size, resulting in progression speeds of below 0,5 m up to greater than 3 m/week.

Table 1: Temperature rise, reduction of net energy concentration and net energy losses at different storage and unloading conditions

Weekly progression in silo (m)	Air penetration depth (m)	Temperature rise above ambience at unloading (°C)			Reduction of Net Energy Lactation (MJ/kg DM)			Losses Net Energy Lactation (%)		
		Stability of silage under air influence, days								
		1	3	7	1	3	7	1	3	7
1	1	23	16	0	0,4	0,16	0	16	8	0
	2	27	27	27	1,0	0,9	0,5	38	34	21
2	1	11	4	0	0,07	0	0	4	1	0
	2	23	16	0	0,35	0,16	0	16	8	0
3	1	4	0	0	0	0	0	1	0	0
	2	14	5	0	0,13	0,05	0	7	2	0

The data for temperature rise refer to silages of around 35 % DM content in medium sized silos. According to varying insulation conditions due to type of forage and silo size they may be lower or still higher.

The data given in the table point out the great influence of all three factors. A stability potential above three days provides a marked reduction of loss and spoilage risk. The same applies, if the progression speed of the silage face exceeds one m/week , and the penetration depth of the air remains at one meter or less.

Prevention

There are two main measures which can be taken to prevent excessive spoilage due to aerobic instability: Application of a "perfect" ensiling technology and the use of adequate additives.

- Ensiling technology
  - Ensuring sufficient consolidation, especially in view of continually increasing harvesting capacities. According to possible air flow DM densities of 200 to 300 kg/m^3 which is dependent on forage type and DM content should be achieved.
  - Precision chop, especially at higher DM contents to improve compactability.
  - Provision of rapid effective sealing. Intermediate covering is advisable if breaks occur during the filling process.
- Improving aerobic stability by adequate silage additives, which are available in several preparations now. Results of the research centre Bredstedt are shown in Table 2. A reliable effect is only to be expected if the additive is applied before or during ensiling.

  Restricting the treatment to the top layer of 0,5 to 1 m may reduce the risk of insufficient consolidation in this upper part of the silo, while the bottom part is protected by the better compaction of the forage.

Table 2: Improvement of aerobic stability by different additives compared to an untreated control

Crop	Preparation	Dose rate	Number of experiments	Improvement, (days)
Grass	Heterofermentative LAB	10^5 cfu/g FM	1	2,9
Maize	Heterofermentative LAB	10^5 cfu/g FM	5	3,7
	Benzoate/propionate	0,4 % of FM	2	5,9
	Formate/propionate	0,4 % of FM	1	4,7
	Urea	0,2 % of FM	2	4,2
Whole crop cereals	Heterofermentative LAB	10^5 cfu/g FM	1	2,1
	Urea	0,2 % of FM	1	6,6

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Silage additives in big round-bale silage production

Keywords: Big-bale silage, silage additives, silage quality, losses, storage stability, clostridia, yeast, mould,

Object of the study
The aim of the study was to evaluate the effect of three different additives on bale silage quality. The effect of increasing the amount of liquid added together with the additive, without increasing the amount of active substance or organisms was also investigated.

Introduction
The flexibility of big-bale silage systems is obvious. To ensure high quality silage of the long cut material, however, prewilting to a rather high DM concentration is advised (Jonsson et al 1989, Beaulieau *et al* 1993, Lingvall & Lättemäe 1999). In bales of slightly wilted material, high numbers of clostridia spores often are noticed. To facilitate silage fermentation and prevent clostridial growth, silage additives usually are added to crops at baling. In the long cut material, as in big bales, an even distribution of additives is difficult to achieve. Hence, the positive effect of additives often is limited (Jonsson *et al* 1989). As fermentation commonly is restricted in baled silage, storage stability of opened bales also is low. Addition of additives which could inhibit clostridia in baled silage and prolong storage stability of opened bales, would further improve this silage system (Lingvall & Lättemäe 1999).

Material and Methods
The influence of different silage additives on the quality of first cut bale silage was studied in a randomised block trial. The additives were added on the swath, in front of the pick-up of the baler (MF 828; flex chamber), according to following plan:
1. Control (Untreated control); **2. Kofasil** (Kofasil Ultra; 4 litre (tonne FM)$^{-1}$); **3. Lact 4** (Lactisil 200; 5x10^5(g FM)$^{-1}$; in 4 litre water (tonne FM)$^{-1}$); **4. Lact 8** (Lactisil 200; 5x10^5(g FM)$^{-1}$; in 8 litre water (tonne FM)$^{-1}$); **5. Lact 400 4** (Lactilis 200 + 400 g Sodium Benzoate; 4 litre water (tonne FM)$^{-}$); **6. Lact 400 8** (Lactilis 200 + 400 g Sodium Benzoate; 8 litre water (tonne FM)$^{-1}$); **7. Promyr 4** (Promyr (formic acid, propionic acid and ammonia); 4 litre (tonne FM)$^{-1}$); **8. Promyr 8** (Promyr; 4 litre (tonne FM)$^{-1}$ + 4 litre water (tonne FM)$^{-1}$).

From all bales, samples of silage crops for chemical and microbiological analyses were cored out. The bales were weighed, measured and wrapped with 6 layers of a certified white stretch film (25 µ thickness ; 750 mm width; 60 - 70% pre stretching). The bales were stored outdoors, fenced in, and placed standing on the gable-end. After 100 - 120 days of storage, the stretch film was removed, and surfaces of the bales were inspected concerning mould and yeast. The bales were weighed and samples were cored out. Silages were analysed chemically (DM, pH, ammonia-N, VFA, lactic acid, ethanol) and microbiologically (clostridia spores, mould, yeast). The storage stability of silages were determined by measuring the CO_2 production.

Results
The DM concentration of silage crop at ensiling was approximately 320 g (kg FM)$^{-1}$. The concentration of crude protein (CP) was approximately 164 g (kg DM)$^{-1}$, and the buffering capacity 332 g (kg DM)$^{-1}$. The concentration of WSC was low (63 (kg DM)$^{-1}$).

The chemical composition and the number of clostridia spores of silages are presented in Table 1. Losses and storage stability of silages are presented in Table 2. Silage quality was improved by all silage additives. The concentration of butyric acid was measured but, was only found in untreated silages (2.5 g (kg DM)$^{-1}$.

Table 1. Chemical composition and number of clostridia spores of round-bale silages treated with different silage additives (n = 6)

Treatment	pH	NH_3-N g (kg tot-N)$^{-1}$	WSC g (kg DM)$^{-1}$	Lactic acid, g (kg DM)$^{-1}$	Acetic acid, g (kg DM)$^{-1}$	Ethanol g (kg DM)$^{-1}$	Clostridia cfu (g FM)$^{-1}$
Control	4.8	50	4.2	16.3	5.2	6.3	2.7
Kofasil	4.6	42	9.0	19.7	5.0	3.6	<2.0
Lact 4	4.4	42	3.3	26.7	6.0	4.9	<2.0
Lact 8	4.5	41	3.7	24.5	5.4	5.5	3.1
Lact 400 4	4.5	44	2.8	23.7	6.0	5.5	2.6
Lact 400 8	4.5	46	3.0	24.7	5.7	5.0	<2.0
Promyr 4	4.7	47	7.3	15.2	4.2	5.1	3.4
Promyr 8	4.7	47	13.2	17.8	4.8	5.0	2.1
Mean values	4.6	45	5.8	21.1	5.3	5.1	
LSD	0.2	0.8	4.3	4.5	0.9	1.3	

Table 2. Losses of DM, calculated nutrient losses (kg weight loss x 2.5 = kg WSC), and storage stability (days of prevented CO_2 production) of round-bale silages treated with different silage additives (n = 6)

Treatment	DM g (kg)$^{-1}$	Bale weight kg	Bale density kg DM	Weight losses kg (bale)$^{-1}$	Nutritive losses kg (tonne)$^{-1}$	Storage stability Days
Control	303	661	118	10.3	39	0.5
Kofasil	309	660	119	6.4	24	6.0
Lact 4	302	671	118	8.1	30	2.0
Lact 8	311	672	120	8.8	30	1.5
Lact 400 4	296	659	112	7.6	29	2.5
Lact 400 8	311	670	121	7.3	27	6.5
Promyr 4	309	661	119	7.8	29	1.0
Promyr 8	301	652	112	7.8	29	1.5

Conclusions

- All the silage additives improved the chemical quality of the round-bale silage
- Kofasil Ultra, Lactisil 4 litre (tonne)$^{-1}$ and Lactisil with sodium benzoate in 8 litre of water reduced the number of spores below the detection limit
- Storage stability was improved by Kofasil Ultra and Lactisil with sodium benzoate in 8 litre of water

Acknowledgements

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The Nutritive Value and Aerobic Stability Round Baled Silages from Grass-alfalfa Mixed Crop Ensiled with Additives Containing Formic Acid

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INTRODUCTION
Grass legumes have a high value but are difficulty to ensiled. Wilting and balling these material allow to obtain good silage, but this silage is very susceptible to the influence of outside factors which may lead to the development of unwelcome organism especially moulds. Formic acid as well other organic acid and their salts facilitates ensiling of grasses and legumes. They can also limit the development and activity of moulds.

MATERIALS AND METHODS
Silages were made from mixtures grasses and alfalfa (lucerne 79%, cocksfoot 16%, timothy 5%). The experimental material was obtained from plots from the first cut, at the pfase of earing of grasses and budding of alfalfa. After 12 h. wilting the plant material was ensiled using Sipma baler and wrapped with four layers of plastic. Two experimental treatments were adopted in the experiment: with aplication silage preparation containing 55%formic acid, 24% ammonia formate, 5% propionic acid, 1% benzoic acid and 1% esters of benzoic acid - „KemiSile 2000" (manufactured and delivered by Kemira Chemical Oys - Finland) and without any additive. The preparation was applied during harvesting in the amount 4l/t fresh material. Six bales from each treatments were analysed (one bale was one replicate). The bales were opened after 60 days and samples were collected using special silage core. The following parameters were determined in analysed silages: basic nutrients, final products of fermentation, stability in aerobic condition estimated by temperature measurements, changes in chemical composition after 7 days exposure in 20 ° C and as well as quantities of E. Coli and Clostridium bacteria, yeasts and moulds found in silages at the moment of sampling .

RESULTS AND DISCUSSION
The examined mixed crop silages were characterised by high protein concentration (appromiximately 200g/kg DM) which was similar to the initial material as well as by low concentration of ammonia nitrogen (from 2,5 to 2,7g/kg DM). Furthermore, the experimental silages were found to contain, moderate concentration (46 - 48 g/kg DM) and low concentration of butyric (3,17 - 4,10 g/kg DM). The applied additive reduced the content of butyric acid and increased quantities of soluble sugars which remained in silage. No significant changes in chemical composition of experimental silages were found after 7 days exposition, although the amount of lactic acid decreased, while the amount of ammonia and butyric acid increased, also pH was found to rise. However the above changes were smaller in samples without the examined additive, temperature was found to increase from 27,7 to 24,59° C from 5th day of exposure onwards. Silage samples containing the additive were found to contain 6 times lower levels of Clostridium bacteria and 2,5 times smaller quantities of moulds. The additive did not affect the number of yeast, while the number E. Coli was higher.

CONCLUSIONS
Chemical additives containing formic acid and propionic acid and benzoic improve the quality and stability of mixed crop silages in bales. Simultaneously, the application of such additives reduces the number of moulds in them.

Table 1. Chemical composition of silages from wilted grass-legumes mixture (bales)

	with additive		without additive	
	after opening	after 7 days	after opening	after 7 days
dry matter g/k	332,95	380,55	338,00	379,52
crude protein g/kgDM	202,46	201,43	186,76	177,89
crude fiber g/kg DM	267,11	274,64	259,87	268,12
N-NH_3 g/kg DM	2,55	2,72	2,58	3,85
lactic acid g/kg DM	48,89	46,96	45,88	44,19
formic acid g/kg DM	8,99	7,69	-	-
acetic acid g/kg DM	23,54	25,86	25,93	28,73
propionic acid g/kg DM	2,30	2,35	2,13	2,36
butyric acid g/kg DM	3,17	3,61	4,10	4,29
WSC g/kg DM	38,16	30,12	28,64	25,67
pH	5,16	5,77	5,37	6,54

Graph 1. Changes of temperature in silages during 7 days exposure to the air

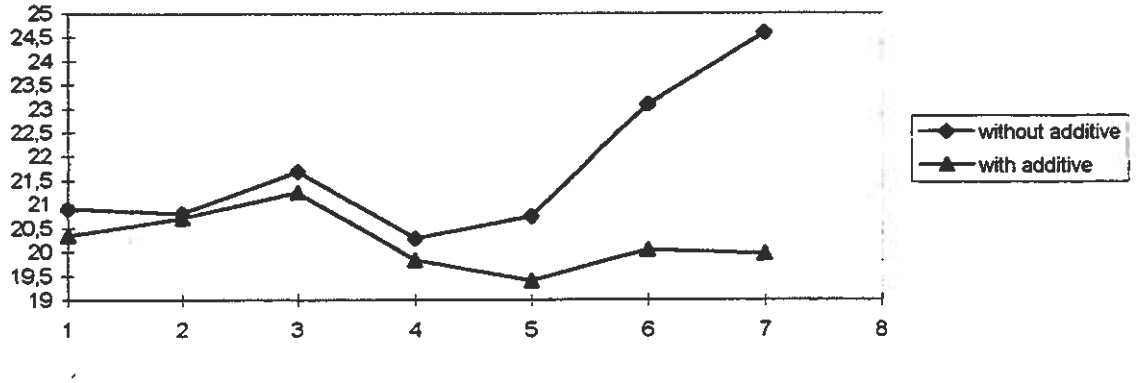

Table2. Hygenic value of silages from grass-legumes mixture

Microorganism	c.f.u. in 1g FM	
	with additive	without additive
E. coli	$17,5*10^3$	$13,60*10^3$
Clostridium	$0,67*10^5$	$4,48*10^5$
yeasts	$125,33*10^5$	$110,08*10^5$
moulds	$0,17*10^4$	$0,43*10^4$

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Effect of additives on fermentation quality and aerobic deterioration of grass silage.

Introduction

When the silo is opened, the silage will be exposed to aerobic conditions. The phenomenon being called aerobic deterioration is often caused by the growth of aerobic microorganisms. It has been demonstrated that aerobic deterioration of silage results in temperature and pH increases, DM losses and surface mould growth. The aerobic deterioration can be prevented by propionic acid and higher level of volatile fatty acids. Additives based on formic acid or lactic acid bacteria are used to improve the fermentation quality of silage on many dairy farms. Although a few workers report that the formic acid or bacterial inoculum inhibit aerobic dererioration, little is known about the effect of these additives. Prevoius reports indicated their useful effect on grass silage quality. The present study carried out to determine the effect of these additives on aerobic deterioation of grass silage.

Materials and Methods

1) Silage preparation:

First cut timothy (Phleum pratense L.;heading stage), orchardgrass (Dactylis glomerata L.;heading stage) and alfalfa (Medicago sativa L.,early blooming stage) were pre-wilted indoors for ÇQdays. The grass was cut into about 2-3cm length before ensiling and ensiled in laboratory silos (capacity 1l) without any additives and with formic acid (Sibest, a product conatining 85% formic acid manufactured by Daisel Chemical Industries Ltd.), bacterial inoculant which contained a single strain of the organism Lactobacillus casei subsp. rhamnosus (Snow Lact L, a product manufactured by Snow Brand Seed Co., Ltd.) or a mixture of bacterial inoculant (Snow Lact L) and cellulase preparation derived from Trichoderma viride (Meicelase, a product manufactured by Snow Brand Seed Co., Ltd.). The glass bottle silos were kept at 25Åé for 30 days in a room, and five replicates per treatment were used for chemical and microbiological analysis.

2) Aerobic deterioration:

After the ensilage of 30 days, the silos were opened and the silages were packed in containers of expanded polystyrene (capacity 1.5l). These containers were kept at 26Åé for 7 days in an incubators, surface of the silages being exposed to air. Temperatures of the center of silages in containers were monitored by thermisters. Three replicates per treatment were used for chemical and microbiological analysis, but two replicates were used for measurement of temperaure. 3) Chemical analysis: Dry matter contents of grass and silages were determined by oven-drying at 135Åé for 2 hours. Lactic acid content was determined by the colorimetric method of Barker and Summerson. Volatile basic nitrogen (VBN) content was measured by steam distillation. Volatile fatty acids (VFA) content was measured by a gaschromatography. 4) Microbiological analysis: The plating media were GYP-calcium carbonate agar for lactic acid bacteria, bouillon agar for aerobic bacteria and YM agar containing 0.5% lactic acid for moulds and yeasts. The plates for lactic acid bacterial and aerobic bacterial counts were incubated for 5 days at 35Åé and plates for moulds and yeasts counts were incubated for 6 days at 25Åé. The viable colonies were counted by dilution plate method.

Results

1) Fermentation quality and viable counts of silages: The fermentation quality of the untreated silages of timothy, orchardgrass and alfalfa silages were characterized by high content of butyric acid
(1.68-6.28%DM) and low V score (2.4-60.4). The fermentation quality of formic acid-treated silages of timothy, orchardgrass and alfalfa silages were better than that of the untreated silage. The formic acid treatment was markedly effective for the improvement of the fermentation quality of silages in all species of grass. The fermentation quality of the inoculant-treated timothy and orchardgrass silages were good, while the fermentation quality of alfalfa silage was not improved with the inoculant treatment. The inoculant and enzyme-treated timothy and orchardgrass silages were well preserved, with a low pH value and high lactic acid and low butyric acid contents. However, alfalfa silage showed low lactic acid production and high butyric acid and VBN productions.

2) Aerobic deterioration: The untreated timothy and orchardgrass silages showed no distinct temperature rise after opening the silos. The temperature rise in the formic acid-treated timothy and orchardgrass silages started ÇPto 2 days after the onset of aerobic exposure. The maximum temperature of these silages were high. The inoculant-treated or inoculant and enzyme-treated timothy and orchardgrass silages showed a slight temperature rise. In alfalfa silage, the untreated and formic acid-treated silages showed a distinct temperature rise. The temperature rise in these silages started 3 to 4 days after the onset of aerobic exposure. The pH of the formic acid-treated timothy and orchardgrass silages tend to increase 7 day after the onset of aerobic exposure. The pH of the inoculant and enzyme-treated orchardgrass silage was higher than that of the untreated silage. After the 7-day aerobic exposure period, the yeast and mould numbers tend to increase in the formic acid-treated timothy and orchardgrass silages. The yeast numbers of these silages were 108-1010 cfu/gDM and the mould numbers were 107-108 cfu/gDM. Moreover, the aerobic bacterial numbers of these silages showed an increase compared with the silages at the time of opening. In alfalfa silage, the counts of aerobic bacteria, yeast and mould in the formic acid-treated silages showed an increase compared with the silage at the time of opening.

Conclusions

In this study, the formic acid-treated silages after the 7-day aerobic exposure period were characterized by low lactic and acetic acids contents, high VBN (%T-N) and high pH. The counts of yeasts and moulds in these silages increased 104-105 to 108-1010 cfu/gDM and 103 to 107-108 cfu/gDM respectively during aerobic exposure period. High counts of yeasts and moulds played a significant role in the aerobic deterioration of these silages. The inoculant or inoculant and enzyme-treated orchardgrass silages after the 7-day aerobic exposure period were characterized by low lactic and acetic acids contents, and the counts of yeasts and moulds increased 103 to 108 cfu/gDM and <102 to 103-105 cfu/gDM during aerobic exposure period. However, the temperature rise of these silages were slight. These results confirm that though formic acid or inoculant or inoculant and enzyme treatments improve the fermentation quality of timothy and orchardgrass silages, formic acid is not to prevent the aerobic deterioration of these silages. However, inoculant or inoculant and enzyme are able to prolong the aerobic stability of silages.

Key words: Additives, Aerobic deterioration, Enzyme, Fermentation quality,
Formic acid, Grass silage, Inoculant.

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Improvement of silage quality by innovative covering system

Introduction. The introduction of the practice of ensiling forage by farms devoted to animal husbandry, and particularly of silage corn has contributed to the reduction in the cost of feed and consequently to an increase in profits. Over the years as a result of the ever increasing application of this practice a number of factors have been noted which influence the final quality of the product. These are now mainly well know even if not always fully controlled in practice.
Silage making is a natural method of preservation which makes use of the microbes normally present on the forage to transform the sugars in the plant into organic acids. These, by lowering the pH of the mass, prevent other types of fermentation which are anormalous to the objective which is to preserve the forage with minimum losses and to modify its characteristics as little as possible.
Having considered the factors which are involved in the biochemistry of the fermentation a number of procedures have been proposed to obtain an optimum result, for some of the factors there has been greater practical application than for others which have remained unchanged over a period of time because they have been considered well know, difficult to change or of little interest. Amongst the latter is the manner of covering silos which is of primary importance to obtain a good result for the ensiling process.
The importance of correct and rapid covering immediately after having completed the loading of the forage in the silo derives from the need to create an anaerobic atmosphere so as to aid the development of lactic bacteria which are mainly responsible for the acidification which renders the silage stable over a period of time.
In order to exploit the phenomenon of natural acidification by the creation of appropriate conditions for the ensiling process, avoiding the use of any additive in the forage, reducing the cost of production of silage by means of good practice and the reduction of wastage, it was decided to test an innovative method of covering which should have the best characteristics for the conservation of forage by means of silage making.

Materials and methods. Two identical bunker silos situated next to each other with central common wall were prepared with an overall capacity of 420 tons of whole chopped silage corn at 32.5% DM content of first growth of the same variety. Harvesting and filling of the silos was carried out in the same day in a continuous operation using the normal procedures. On completion of filling, one of the silos was covered with commonly available white polyethylene sheet 200 μm thick; the other silo was covered with plastic film denominated "Silostop" of much lower thickness (45μm) and having a marked barrier effect towards oxygen and other suitable characteristics compared to other traditional polyethylene sheet as shown in Table 1. In order to assure efficacious impermeability conditions, in both silos the sheets were spread deep down on the silo walls and also on the concrete floor for about 1 meter before filling. On the top two silos prepared as described was spread a second sheet of the same polyethylene as used on the first silo. Subsequently tyres were spread over the entire surface to apply weight to the sheets and keep them in good contact with the forage below. The lower edges of sheets were brought together and closed applying other weights according to the normal practice in the area. Both silos were covered in the same way. The difference was only the type of plastic sheet in contact with vegetable material.

After the fermentation period the silo covered with the standard sheet was opened and once the cutting face had been set back and was fully exposed, a series of samples were selected vertically through the silage and mixed to obtain an average result. The resultant sample was sent for analysis. After having collected three such samples, the silo covered with virtually impermeable film was opened and the same procedure was carried out.

Results and conclusions. The mean results of the tests carried out on the samples selected of "Silostop" vs. standard sheet showed that whereas the figures relating to chemical composition as protein, NDF and amides can be considered not different, those which identify better conservation and hence a higher quality of silage (pH, lactic acid, NH_3-N and total N) relating to the samples deriving from the silo covered with virtually impermeable thin film (Table 2).

Moreover it was noted that the average thickness of the wasted material in the upper surface layer in contact with the cover was clearly noticeable in the control silo, particularly in the corners along the side. Whereas thanks to the good adherence and impermeability to oxygen, which are decisive factors in favouring fermentation, it was never produced in the silo covered with impermeable thin film.

In view of the positive result of the test and to examine the possibility of reducing the deterioration of the film either from inherent causes or external damage and to simplify the ensiling process and also that of daily silage removal a number of other silos have been prepared and are under observation. For these a combination has been used of virtually impermeable thin film and woven fabric not susceptible to UV radiation, which is particularly robust and reusable, weighed down with woven sacks of the same material which once filled with sand or gravel are laid along the sides of the silos. These sacks effectively replace in a practical and hygienic manner all other types of inert matter which have so far been used, at the same time preventing deterioration of the silage mass.

The principal objective of this system is to improve the quality of the silage and to reduce the unit costs. The economies obtained derive from a better quality of silage, reduction in wastage, and reduction in the use of labour in the production and removal of the silage. The reduction in damage to the environment should not be ignored which derives from relinquishing the use of certain inert materials which were previously used for covering.

Table 1: Characteristics of "Silostop" compared with a polyethylene sheet (same thickness)

Test	Silostop	Polyethylene
Thickness (μm)	45	45
Longitudinal breaking load (N/mm^2)	38	22
Transversal breaking load (N/mm^2)	30	20
Longitudinal expansion at break point (%)	300	280
Transversal expansion at break point (%)	310	350
Permeability to O_2 at 85%R.H., 23°C ($cm^3/m^2/24h$)	100	4000
Permeability to O_2 at 85%R.H., 50°C ($cm^3/m^2/24h$)	500	12000

Table 2: Chemical composition and characteristics of fermentation of two experimental batches

	Silostop	Polyethylene
Dry matter (%)	32.3	32.6
Crude protein (% of DM)	23.5	22.7
NDF (% of DM)	46.5	46.2
Amides (% of DM)	24.1	24.2
pH	3.78	3.97
Lactic acid (% of DM)	3.67	3.18
$N\text{-}NH_3$ (% total N)	5.33	6.12

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The effect of ambient temperature on the rate and extent of aerobic deterioration in maize silages, and the subsequent effects on *in vitro* fermentation characteristics assessed using the Reading Pressure Technique (RPT).

Introduction Aerobic deterioration of silage at feed-out can be difficult to avoid, particularly when silage is used as a buffer feed during the summer. The current study evaluates the effect of ambient temperature on the aerobic deterioration of maize silage, and the subsequent effects on *in vitro* fermentation characteristics.

Methods Four types of maize silage (Ms1-Ms4, 268, 308, 337 and 364 g DM/kg, respectively) were frozen. At 7 day intervals a representative sample of each silage was thawed, aerated and placed in insulated boxes (4 replicates) in a controlled environment maintained at 15, 25 or 35 °C. Aerobic stability was assessed from silage temperature, and silage was sampled at the end of the measurement period to determine DM loss, pH and count of total yeasts and moulds (TYM) (Table 1). An *in vitro* gas based feed evaluation system (RPT) (Mauricio *et al.*, 1999) was used to measure fermentation profiles, and therefore describe the rate and extent of degradation during a 96 hour incubation period, of fresh and deteriorated silage. Data were fitted using the model of France *et al.* (1993) to calculate the time to half asymptote (T/2), combined fractional rate/h (μ) at T/2 and cumulative gas production at 12 and 96 hours. DOMD was measured at 96 hours.

Results **Table 1. Measurements of aerobic deterioration**

Silage	Ambient (°C)	Hours to 2°C rise above ambient	Mean temp (oC)	DM loss (g/kg)	pH	TYM $Log_e(X+1)$ (CFG/g)
	15	168	12.9	20	3.25	18.3
Ms1	25	165	18.4	48	3.47	23.1
	35	93	34.4	117	5.62	32.1
	15	152	14.1	22	3.32	18.8
Ms2	25	165	18.4	48	3.25	22.2
	35	116	32.9	95	5.19	32.4
	15	160	12.7	7	3.26	16.6
Ms3	25	165	18.3	11	3.16	15.9
	35	101	34.0	134	6.09	26.6
	15	160	13.7	63	3.69	30.9
Ms4	25	82	25.2	89	5.72	31.1
	35	36	33.4	162	7.00	25.4
s.e.d		**7.9**	**1.17**	**17.8**	**0.156**	**3.22**

Increasing ambient temperature from ≤ 25 °C to 35 °C had a negative effect on aerobic stability, reducing the hours to a 2°C rise above ambient ($P<0.05$), and increasing mean

temperature (P<0.05), DM loss (P<0.05, except Ms2 P>0.05), silage pH (P<0.05) and TYM (P<0.05, except Ms4 P>0.05). Maize harvested at 364 g DM/kg (Ms4) was less stable than the remaining silages, and this was particularly evident as ambient temperature increased.

Table 2. *In vitro* gas production and DOMD.

Silage	Ambient (°C)	T/2 (h)	Fractional rate/h μ at T/2	Cumulative gas (ml) 12h	96h	DOMD
	Fresh	10.57	0.078	143	259	0.705
	15	10.39	0.075	149	268	0.681
Ms1	**25**	10.83	0.070	136	253	0.699
	35	13.25	0.063	112	243	0.673
	Fresh	9.46	0.084	160	269	0.697
	15	10.97	0.070	137	257	0.686
Ms2	**25**	9.69	0.076	150	258	0.702
	35	11.93	0.065	125	249	0.679
	Fresh	9.30	0.089	167	275	0.733
	15	9.54	0.083	164	277	0.743
Ms3	**25**	9.18	0.081	169	282	0.735
	35	10.17	0.072	148	264	0.725
	Fresh	8.92	0.093	183	293	0.715
	15	10.09	0.082	163	284	0.724
Ms4	**25**	10.10	0.071	155	275	0.687
	35	10.67	0.071	148	272	0.725

Fermentation profiles detected changes during the initial stages of incubation that can not be detected using traditional end-point analysis. Silage deterioration at 35 °C reduced the readily degradable substrate, reflected in reduced gas production and rates of degradation, and decreased μ at T/2. This effect is modified by stage of maturity at harvest.

Conclusions The results of this study indicate that ambient temperature has a marked influence on the rate and extent of aerobic deterioration in maize silage, which subsequently altered fermentation profiles during a 96-hour incubation period. The RPT offers a rapid method with which to identify the extent of aerobic deterioration and implications of feeding material of this type.

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Workshop F
Silage feeding in relation to food quality and animal health

Poster abstracts

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Effluent production from grass ensiled in round big bales wrapped with 4 layers of plastic film

Introduction

During 1990-1995, big-bale silage accounted for 18% of total silage made in England and Wales. It is generally accepted that with clamp silage, materials ensiled with DM contents above 280g/kg produce little or no effluent (Bastiman 1976). Limited data (Jones and Jones, 1995) on effluent from wrapped big bale silage has indicated that low DM material gives comparable production levels as similar material ensiled in a clamp, although more recent data (Fychan and Jones, 1996) suggest that total effluent flow from wet (200g /kg DM) big bale silage is double that from bunker grass silage ensiled at the same DM content.

This experiment examines the effects of dry matter content at baling and effects of season of harvest on effluent from big bales stored under cover to establish the potential pollution risk from such systems.

Materials and Method

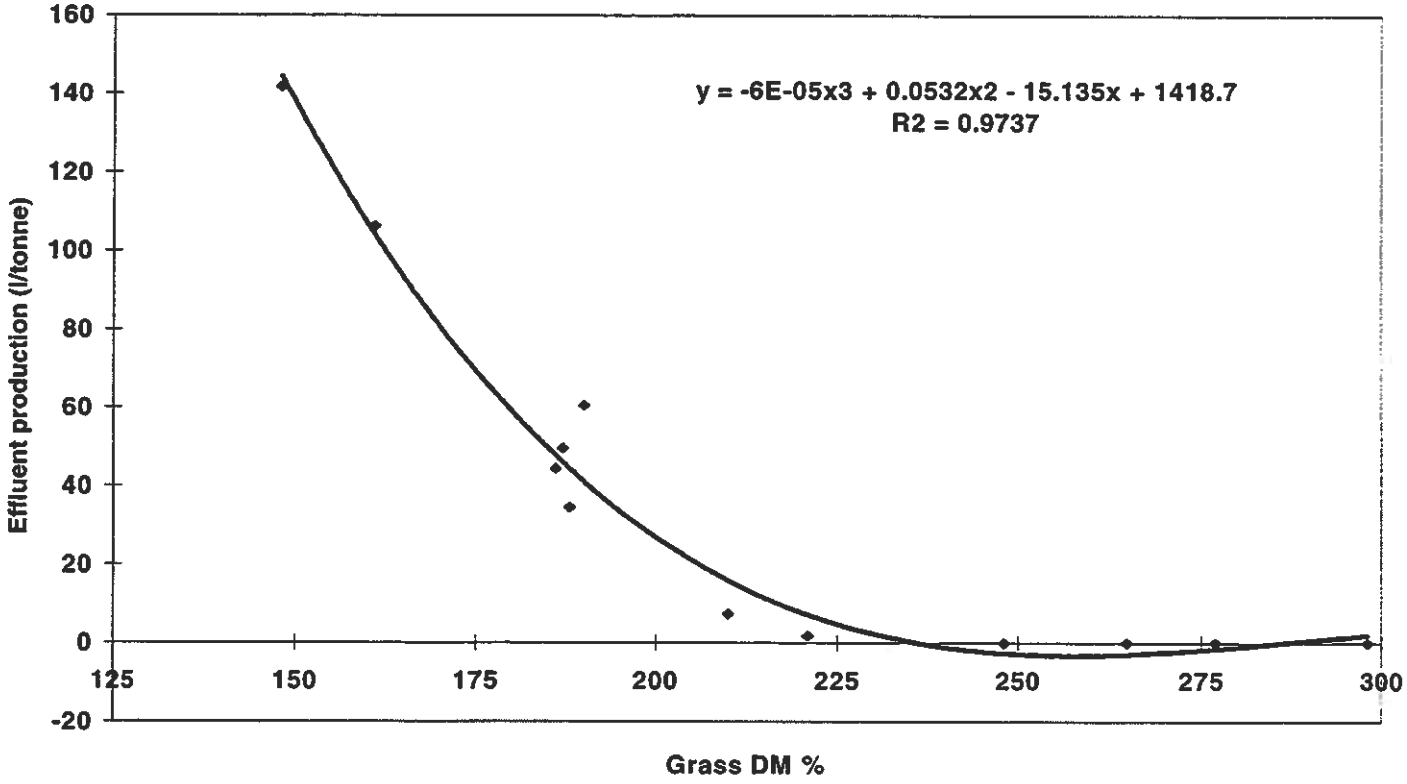
Predominantly perennial ryegrass swards were cut with a disc mower/conditioner in May, July, September and October 1997 and wilted within the resulting swath to attain target Dry Matter contents of 150, 200 and 250g/kg. The resulting herbage was subsequently harvested using a Krone KR 125 fixed chamber baler and wrapped, using a Kverneland 7515 wrapper, with 4 complete layers of black, 750mm wide film applied as a 50% overlap. For each dry matter treatment at each cut, fifteen bales were produced, in three replicates of 5 bales, giving a total of 180 bales. Each group of five wrapped bales was stacked two high under cover on raised, slightly inclined boards fitted with gutters which directed all escaping effluent to a collection vessel. Individual bales are weighed into and out of store and analysed for dry matter content.

Effluent collected during the storage period was sampled and analysed for dry matter, BOD, pH, water soluble carbohydrate and ammonia nitrogen. Bales were opened a minimum of 56 days after ensiling and any effluent released at opening was again collected, sampled and analysed.

Results

Figure 1 shows the relationship obtained between grass dry matter at ensiling and effluent produced.

Figure 1 Effect of grass DM on effluent flow from big bale silage



The close correlation of the data shows that seasonality has little effect and that the effluent production from wrapped big bales is similar in quantity to that predicted from clamp silage. Effluent flow pattern was also similar to clamp silage with a peak flow being achieved at days 3 to 5.

The effects of grass dry matter at ensiling on bale weight change and effluent production are shown in Table 1. Dry matter content ranged from 148 to 298 g /kg. Effluent analyses were inconsistent but there was a tendency for total solids, pH, ammonia nitrogen and BOD in effluent to increase with increasing dry matter of grass at baling and to increase during storage, i.e. the less effluent produced, the more concentrated it became.

Table 1 Effect of grass DM content at ensiling on bale weight change and effluent production

		No wilt	20 hr wilt	34 hr wilt	SEM (df)	CV
Cut 1 - May	DM at ensiling (g/kg)	148	187	186	4.41 (28)	9.8 ***
	Weight loss during storage (kg/tonne)					
	Fresh	155.9	63.7	51.8	9.19 (28)	39.3 ***
	Dry matter	150	121	131	22.37 (28)	64.6
	Effluent (l/tonne grass ensiled)					
	During storage	119.3	45.4	41.8	9.23 (4)	23.2 **
	At Opening	11.5	7.7	7.3	1.86 (28)	81.6
Cut 2 - July	DM at ensiling (g/kg)	210	248	277	3.7 (28)	5.8 ***
	Weight loss during storage (kg/tonne)					
	Fresh	42.8	28.9	30.6	2.05 (28)	23.2 ***
	Dry matter	143	105	148	21.1 (28)	61.7
	Effluent (l/tonne grass ensiled)					
	During storage	7.33	0	0	-	-
	At Opening	6.38	0.07	0	-	-
Cut 3 - Sept	DM at ensiling (g/kg)	161	188	221	4.4 (28)	9.0 ***
	Weight loss during storage (kg/tonne)					
	Fresh	130.8	68.7	51.4	9.49 (28)	43.9 ***
	Dry matter	130	121	120	21.5 (28)	67.3
	Effluent (l/tonne grass ensiled)					
	During storage	106.19	34.52	1.78	5.11 (4)	18.6 ***
	At Opening	10.05	8.23	1.83	1.14 (28)	65.9 ***
Cut 4 - Oct	DM at ensiling (g/kg)	190	265	298	4.68 (28)	7.2 ***
	Weight loss during storage (kg/tonne)					
	Fresh	35.3	9.0	9.4	2.97 (28)	64.2 ***
	Dry matter	85	110	50	17.59 (28)	83.2 **
	Effluent (l/tonne grass ensiled)					
	During storage	20.1	0	0	-	-
	At Opening	2.05	0	0	-	-

Conclusions

The experiment has established a close relationship between grass dry matter at ensiling and subsequent effluent production. Ensiling grass in wrapped big bales at a dry matter content in excess of 240 g/kg will minimise any pollution risk.

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The influence of the number of layers of film cover and film colour on silage preservation, gas composition and mould growth on big bale silage

INTRODUCTION

Baled silage is a significant forage conservation system in Ireland, being used on 82% of all silage-making farms, and accounting for 32% of the forage area conserved as silage[1]. The barrier to gas movement which the stretch film forms is of particular importance in ensuring satisfactory silage preservation. When preserved in round bales, 50% of the forage is within a 17 cm distance of the film barrier. In Ireland, standard wrapping practice is to use 4 layers of 25 µm black film. The objective of the trial described here was to determine the effects of using different levels of film cover and different film colours, on silage preservation, bale gas composition and mould growth on baled silage.

MATERIALS AND METHODS

Three different levels of film cover (nominally 2, 4 and 6 layers) and five different film colours (black, clear, green, light green and white) were applied to bales in a 3 x 5 factorial design with 6 replications per treatment. The grass ensiled was a first-harvest, predominantly perennial ryegrass sward which was wilted to a dry matter concentration of approximately 300 g/kg. The grass was baled with a 1.2 m x 1.2 m chopper baler (Krone KR 130). The bales were transported to the storage area where they were wrapped with either 2, 4 or 6 layers of each of the five films (IP Europe). The bales were placed singly in a net-protected storage area. Concentrations of O_2, N_2 and CO_2 were measured at five different times during the storage period. Samples were taken from a bale sampling port placed in one of the flat ends of the bales approximately 20 cm from the top of the bale. Samples were taken with 20 ml syringes for subsequent analysis in a gas chromatograph fitted with Molecular sieve 5A and Poropak Q columns[2]. Following a 9-month storage period, the bales were cored for silage analysis, and the extent of mould development on the bale surface was estimated.

RESULTS AND DISCUSSION

Silage composition and mould assessment data for the main effects are given in Tables 1 and 2. There were no significant interactions recorded. The level of film cover had a significant effect on silage digestibility, pH and ammonia-N concentration. The most significant effect was on mould development, however, where the 2 layer cover resulted in high levels of mould development and 6 layers of cover resulted in less mould. Interestingly, film colour did not influence any of the measured silage or mould parameters significantly. It is possible that the prevailing temperate climate did not necessitate the use of reflective films.

The gas composition results for the main effect of layers are presented in Figs. 1, 2 and 3. Significant differences between treatments were recorded for all gases at all times except for the final O_2 reading (Day 275). Oxygen was present in all samples at low levels, with the 2-layer treatment having the higher concentration at four of the sampling times. The initial high concentrations of CO_2, early in ensilage, declined significantly over the bale storage period. N_2 levels showed a substantial increase over the storage period, with substantial treatment effects being evident. It is possible that the increasing N_2 levels recorded may be a better indicator of air entry than O_2 levels as oxygen entering the bale may quickly be respired. Film colour did not have a significant effect on gaseous composition.

Table 1. The effect of film cover (no. of layers) on silage composition and mould growth

No. of layers	DM (g/kg)	DMD (g/kg)	pH	Crude protein (g/kg DM)	Lactic acid (g/kg DM)	NH_3N (g/kg N)	Visible mould (% area)
2	301	763	4.9	140	27	100	21.5
4	310	764	4.7	137	28	86	1.7
6	316	775	4.7	137	27	84	0.7
SEM	*6.45*	*3.51*	*0.04*	*1.75*	*0.92*	*4.35*	*2.1*
Signif.	NS	*	**	NS	NS	*	***

Table 2. The effect of film colour on silage composition and mould growth

Colour	DM (g/kg)	DMD (g/kg)	pH	Crude protein (g/kg DM)	Lactic acid (g/kg DM)	NH_3N (g/kg N)	Visible mould (% area)
Black	304	768	4.7	139	27	85	7.7
Clear	313	763	4.7	136	27	96	8.7
Green	310	762	4.9	138	26	92	9.3
Light green	310	773	4.8	138	28	86	5.2
White	308	772	4.7	139	28	90	9.0
SEM	*8.33*	*4.53*	*0.05*	*2.26*	*1.19*	*5.61*	*2.7*
Signif.	NS	NS	NS	NS	NS	NS	NS

Fig. 1: O_2 levels, 2 vs 4 vs 6 layers

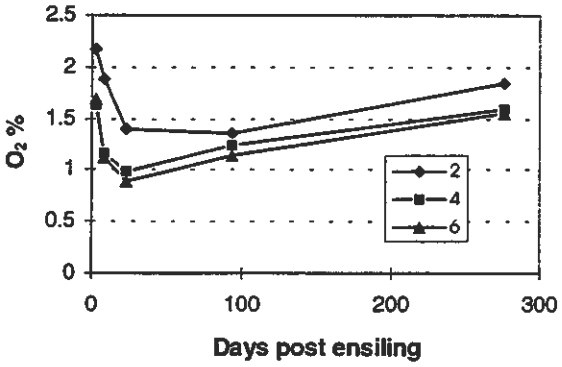


Fig. 2: N_2 levels, 2 vs 4 vs 6 layers

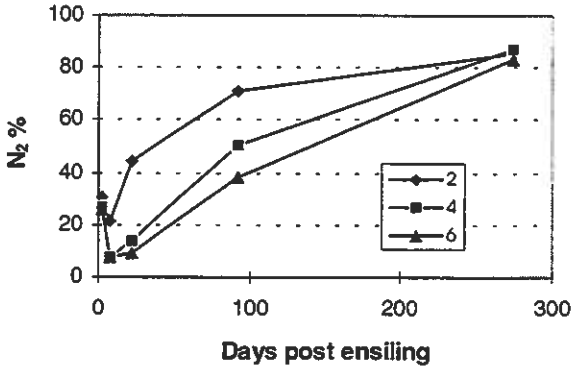
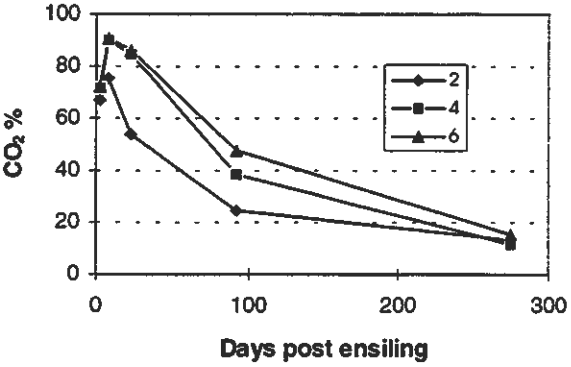


Fig. 3: CO_2 levels, 2 vs 4 vs 6 layers



CONCLUSIONS

The level of film cover on bales significantly affected silage preservation, mould development and gaseous composition of wrapped silage bales. The measurement of gaseous composition may prove a useful technique for assessing the permeability of the stretch film cover.

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Carcass structure and meat quality of dairy-beef fattening bulls fed silage treated with chemical additives

Key words: silage, fattening bulls, chemical additives, dressing percentage, muscle pH, water binding capacity, cooking losses, protein value index.

Object of study. The aim of our study was to determine the growth rate, dressing percentage and meat quality of dairy-beef fattening bulls fed AIV-3 and AIV-10 treated silage in comparison with feeding untreated silage.

Introduction. The dressing percentage, morphological composition, quality and nutritional value of cattle carcasses depends on the balance of feeding (M. Vestergaards, et al., 1996). The feeding system, assortment and quality of feeds, and various feed additives have the greatest influence on the dressing percentage of cattle carcasses (L. Kalm, et al., 1991).

Materials and methods. In 1996-97, a 160-day-trial was carried out with 18 Lithuanian Black-and-White bulls allotted to three analogous (by age, weight and weight gain) groups of six bulls each. The diets were balanced according to the feeding standards. The bulls in Groups 1 (control), 2 and 3 (experimental) were fed, respectively, untreated silage *ad libitum*, AIV-3 (70% of ammonia tetraformiat, anti-corosion compounds and blue dye-stuffs) treated (7 l/t) grass and AIV-10 (77.5% formic acid, 2% of ortophosphotic acid, 3% of ethyl benzoate, anti-corosion compounds, and blue dye-stuffs) (6 l/t) grass. AIV-type chemical additives are produces in Finland. High-quality silage was made in trench silos of 50 tonnes using first cutting clover-timothy grass. The animals in all groups were additionally fed compound feed consisting of 85% of barley meal, 12% soybean meal and 3% of mineral-vitamin mix.

At the end of the trial, three bulls from each group were slaughtered for control investigation. Chemical composition of meat and physicochemical indicators were analysed at the Analytical Laboratory of the Institute of Animal Science.

Results. The trial indicated that each animal consumed daily on average 23.66 kg of untreated silage, 23.69 kg of AIV-3 treated silage and 23.05 kg of AIV-10 treated silage, and the same amount (2.83 kg) of compound feeds. The levels of metabolizable energy obtained were, respectively, 89.79, 92.71 and 93.73 MJ and those of digestible protein, respectively, 763.01, 875.4 and 971.69 g.

The growth rate of bulls in all groups was high. The average daily gain in Group 1 was 1.172 kg, in Group 2 - 1.211 kg and in Group 3 – 1.249 kg. Thus, the daily gains of bulls fed AIV-3 and AIV-10 treated silages were, respectively, by 3.37 and 6.57% higher.

The control slaughter results indicated that the yield of abdominal fat was by 0.45% ($P < 0.05$) and dressing percentage by 1.19% ($P < 0.05$) higher for bulls fed AIV-3 treated silage. AIV-10 treated silage had no significant effect on these traits (Table 1).

Table 1. Control slaughter data

Item	Groups		
	Control 1	Experimental 2	Experimental 3
Finish weight, kg	585.0±22.91	556.67±3.33	550.0±20.21
Hot carcass weight, kg	303.93±11.17	293.33±2.61	286.53±10.52
Hot carcass yield, %	51.97±0.33	52.69±0.17	52.10±0.11
Yield of abdominal cavity fat, %	1.62±0.12	2.07±0.14*	1.93±0.12
Yield of carcass and abdominal cavity fat, %	53.58±0.36	54.77±0.18*	54.04±0.24

*$P < 0.05$.

AIV-3 and AIV-10 treated silages had no significant influence on the morphological composition of carcass (meat : bone : tendon ratio in half carcass).
The contents of dry matter, protein and fat in the *musculus longissimus dorsi* of bulls fed AIV-3 (Group 2) and AIV-10 (Group 3) treated silages were higher by, respectively, 0.45 and 0.82% ($P < 0.025$), 0.35 and 0.6% ($P < 0.05$) and 0.15 and 0.18% ($P < 0.05$) (Table 2).

Table 2. Chemical composition (%) of *musculus longissimus dorsi*

Item	Groups		
	Control 1	Experimental 2	Experimental 3
Dry matter	24.62±0.14	25.07±0.34	25.44±0.23**
Protein	21.08±0.15	21.43±0.15	21.68±0.23*
Fat	2.34±0.07	2.49±0.4	2.52±0.09*
Ash	1.05±0.02	1.05±0.01	1.09±0.01

*$P < 0.05$; **$P < 0.025$.

The analysis of the chemical composition of ground meat indicated that there were no significant differences between the groups. There was a tendency for fat content increase in AIV-3 group and fat content decrease in AIV-10 group.
The long dorsal muscle pH and water binding capacity were by, respectively, 0.92 ($P < 0.005$) and 0.75 units and 3.71 and 1.88% higher for Group 2 (AIV-3) and Group 3 (AIV-10), while cooking losses were by 7.28 ($P < 0.005$) and 3.71% lower. The levels of tryptophan in AIV-3 and AIV-10 groups were by 22.86 ($P < 0.01$) and 25.63 mg / 100 g higher compared with the control group. The protein value index in those groups was also by 23.45 and 10.31% higher (Table 3).

Table 3. Physical and chemical characteristics of *musculus longissimus dorsi*

Item	Groups		
	Control 1	Experimental 2	Experimental 3
Meat pH	5.60±0.05	6.52±0.17****	6.35±0.3
Water binding capacity, %	63.64±1.61	67.35±2.56	65.52±5.0
Cooking losses, %	48.86±0.38	41.58±0.51****	45.15±2.0
Triptophan, mg/100 g	286.87±2.25	309.73±1.32***	312.50±4.5
Oxyprolin, mg/100 g	75.41±8.02	65.57±5.79	74.61±7.0
Protein value index	3.88±0.38	4.79±0.38	4.28±0.31

***$P < 0.01$; ****$P < 0.005$.

Conclusions

ÑWeight gains bulls fed AIV-3 and AIV-10 treated silages were higher, respectively, by 3.37 and 6.57%. The dressing percentage in AIV-3 group was by 1.19% higher in comparicon with the control group.
ÑThe content of dry matter, protein and fat in the *musculus longissimus dorsi* of bulls fed AIV-3 and AIV-10 treated silages were higher.
ÑFeeding AIV-3 and AIV-10 treated silages increased water binding capacity and pH of *musculus longissimus dorsi* and decreased meat cooking losses.

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Fat colour and the quality of meat from beef cattle offered grass silage or maize silage-based diets [+]

Introduction. Grass silage is the predominant forage in the diet of Irish beef cattle finished indoors. There is opportunity in certain parts of Ireland to grow and ensile maize as a cattle feedstuff. If there are differences in the appearance or sensory quality of beef due to the source of dietary forage, the ability of Irish beef exporters to service existing or new markets might be enhanced. As there is little information available on the relative influence of grass silage and maize silage on meat quality, the objective of this study was to determine the impact on fat colour and selected meat quality attributes of substitution of grass silage with maize silage in the diet of finishing beef cattle.

Materials and Methods. Forty-five continental crossbred heifers were offered *ad libitum*, grass silage (GS), a mixture of 500 g GS and 500 g maize silage (MS)/kg dry matter (DM) or MS in a randomised block design. Animals also received 3 kg concentrates (310 g citrus pulp, 460 g barley, 160 g soyabean, 50 g molasses and 20 g mineral/vitamin mixture/kg) daily. After 167 days all animals were slaughtered. The weight of the kidney+channel fat depot was recorded and a sample was stored at 4°C for 48 h prior to colour analysis (Strange et al., 1974). Subcutaneous fat colour was visually assessed 1h post-mortem and categorised as white (score = 1), yellow (score = 3) or intermediate (score = 2). Samples of subcutaneous fat and the longissimus thoracis et lumborum (LTL) muscle were removed 24 h post-mortem for immediate assessment of colour of fat, assessment of lean colour after 14 days (Strange et al., 1974), assessment of drip loss from LTL at 2 days post-mortem (Honikel, 1987), assessment of Warner Bratzler shear force (Shackelford et al., 1994) for LTL after 2, 7 and 14 days ageing and for sensory perception of LTL after 14 days ageing (AMSA, 1978).

Results. The GS had mean (s.d.), 180 (4.2) g DM/kg, 166 (7.1) g crude protein/kg DM and 753 (16.0) g digestible DM/kg. The corresponding values for MS were 297 (7.6), 105 (2.5) and 802 (14.3). Animal performance data are summarised in Table 1 and fat colour and meat attributes are summarised in Table 2.

Table 1. Animal performance.

	Maize silage (g/kg dry matter)				
	0	500	1000	s.e.d.	Significance[1]
Initial bodyweight (kg)	443	442	443	1.2	NS
Bodyweight gain (g/day)	845	907	1015	57.8	L**
Carcass weight (kg)	324	327	341	5.5	L**
Carcass weight gain (g/day)	653	678	756	31.6	L**
Kidney+channel fat (kg)	13.8	14.0	13.9	1.17	NS

[1] L is the linear effect of maize silage inclusion level.

Table 2. Attributes of meat quality

	Maize silage (g/kg dry matter)				
	0	500	1000	s.e.d.	Significance[1]
Subcutaneous fat colour					
- visual assessment	2.60	2.37	1.30	0.209	L***,Q*
Hunter "b" value	17.0	17.0	13.5	0.53	L**,Q**
Kidney+channel fat colour					
Hunter "b" value	27.0	24.6	20.7	1.25	L***
Longissimus thoracis et lumborum					
Colour					
Hunter "L" value	35.7	36.9	36.8	0.60	NS
Hunter "a" value	16.5	16.2	16.2	0.42	NS
Hunter "b" value	9.6	9.6	9.7	0.26	NS
pH_{48}	5.55	5.54	5.52	0.015	L*
Drip loss (%)	3.53	3.68	4.00	0.392	NS
Shear force (kg) - 2 days	6.85	6.18	6.62	0.601	NS
- 7 days	4.26	4.72	4.50	0.418	NS
- 14 days	3.64	3.62	3.64	0.265	NS
Tenderness[2]	5.69	5.69	5.47	0.301	NS
Juiciness	4.75	4.86	4.91	0.271	NS
Flavour	4.24	4.24	4.00	0.178	NS
Firmness	4.97	5.05	5.26	0.216	NS
Texture	3.69	3.84	3.69	0.179	NS
Chewiness	3.23	3.00	3.32	0.213	NS
Acceptability	3.90	3.99	3.82	0.191	NS

[1]L and Q are linear and quadratic effects of maize silage inclusion level, respectively.
[2]For tenderness, 1 = extremely tough, 8 extremely tender; for juiciness, 1 = extremely dry, 8 = extremely juicy; for flavour, 1 = very poor, 6 = extremely good; for firmness, 1 = extremely mushy, 8 extremely firm, for texture, 1 = very poor, 6 = extremely good; for chewiness, 1 = not chewy, 6 = extremely chewy; for acceptability, 1 = not acceptable, 6 = extremely acceptable

Conclusion. Under the conditions of this experiment, substitution of grass silage with maize silage in the diet of finishing heifers resulted in whiter fat but had no effect on the sensory perception of meat quality.

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Effects of *Escherichia coli* 0157:H7 added to grass at ensiling on the early stages of silage fermentation

Introduction. Enterobacteria are frequently present on grass at ensiling where contamination with animal manure or soil has occurred. These gram negative, non-sporing, rod-shaped and facultatively anaerobic bacteria ferment carbohydrates to short-chain organic acids (especially acetic acid) or butanediol. They have weak proteolytic activity but can deaminate and decarboxylate some amino acids. Most species can reduce nitrate via nitrite to ammonia, and some can also produce nitrous oxide. Growth limiting factors for enterobacteria include pH <4.5 to 5.0, a_w <0.95 and temperature <8°C. From among the enterobacteria, *E. coli* is particularly important because of the virulence of some strains towards livestock and humans. The objectives of the present experiment were to trace the development of a specific non-infectious strain of *E. coli* 0157:H7 in the early stages of silage fermentation, to determine its influence on the fermentation profile and to determine the effects on *E. coli* 0157:H7 numbers of altering the ensilage conditions.

Materials and Methods. A leafy regrowth of unwilted permanent grass was harvested in mid September and precision-chopped. Sixty samples of 6 kg grass were randomly assigned to the following additive treatments: (a) no additive, (b) *E. coli* 0157:H7, (c) formic acid (850 g/kg; 3 ml/kg grass), and (d) *E. coli* 0157:H7 plus formic acid (i.e. (b) + (c)). A non-infectious strain of *E. coli* 0157:H7 was used, and was applied at $\log_{10}$ 4.5 colony forming units/g forage. Additives were applied manually before ensiling in laboratory silos. Three silos per treatment were opened after 0, 1, 2, 3 and 10 days ensilage, and subjected to chemical and microbiological analyses.

Results. Forage microbiological and chemical analysis data are summarised in Table 1. Mean (s.d.) forage dry matter digestibility at ensiling was 722 (14.1) g/kg. Silage made without additive underwent a rapid, lactic acid dominant fermentation during the 10 days of ensilage. Formic acid restricted fermentation, reducing (P<0.001) the concentration of lactic acid, acetic acid and ammonia-N, as well as pH and buffering capacity, and increasing (P<0.001) the concentration of water soluble carbohydrates and nitrate. Inoculation of forage did not alter (P>0.05) silage fermentation. In the absence of formic acid, counts of Enterobacteria (which were high on day 0) and *E. coli* 0157:H7 decreased rapidly, and were absent after 10 days ensilage. Formic acid increased (P<0.001) the initial rate of decrease in Enterobacteria counts, but resulted in higher (P<0.001) counts on day 10. However, these latter values were relatively low and appeared to be decreasing. Similarly, formic acid increased the initial rate of decline in counts of *E. coli* 0157:H7, but by day 10 was associated with a higher count than when formic acid was not applied.

Conclusions. Grass ensiled with or without formic acid provided contrasting ensiling conditions. Addition of *E. coli* 0157:H7 to grass at ensiling did not influence the fermentation profile up to day 10, either in the presence or absence of formic acid. Altering the ensilage conditions by using formic acid changed the survival pattern of *E. coli* 0157:H7 and of Enterobacteria. Both Enterobacteria and the inoculated *E. coli* 0157:H7 rapidly decreased to low levels or disappeared within the time-frame studied.

Table 1. Forage chemical and microbiological composition during the early stages of fermentation.

	Dry matter (g/kg)	Crude protein (g/kg DM)	pH	Ammonia-N (g/kg N)	Lactic acid (g/kg DM)	Acetic (g/kg DM)	Prop. (g/kg DM)	Butyric (g/kg DM)	Total VFA (g/kg DM)	WSC (g/kg DM)	B.cap. (mEq/kg DM)	Nitrate (mg/l)	EB	E.c.
Day 0														
No additive	185	150	6.27	29	5	5	0.08	0	5	62	430	54	8.12	0
E. coli (E.c.)	181	154	6.37	31	4	5	0	0	5	59	434	92	7.77	4.57
Formic acid (FA)	185	149	4.63	16	3	2	0.03	0	2	71	445	143	7.62	0
E.c. + F.A.	190	146	4.57	13	3	1	0	0.35	1	66	419	167	7.26	4.50
Day 1														
No additive	179	166	5.13	28	17	16	0	0.1	17	43	468	122	8.15	0
E. coli	170	158	5.07	32	17	19	0.17	0	19	41	461	132	7.67	4.89
Formic acid	188	142	4.47	15	5	2	0	0	2	69	419	128	6.18	0
E.c. + F.A.	186	143	4.53	15	4	2	0	0	2	78	433	123	6.11	3.64
Day 2														
No additive	181	146	4.90	38	36	12	0	0	12	39	490	13	7.21	0
E. coli	178	147	4.90	42	40	12	0	0	12	40	494	13	6.37	3.64
Formic acid	182	148	4.47	21	11	3	0	0	3	72	460	39	5.95	0
E.c. + F.A.	184	144	4.40	22	9	3	0	0	3	75	436	30	6.02	3.05
Day 3														
No additive	188	144	4.70	42	38	11	0	0	11	34	556	0	4.16	0
E. coli	190	147	4.70	42	40	13	0	0	13	36	549	3	4.91	2.79
Formic acid	192	143	4.37	23	10	3	0	0	3	72	476	69	4.12	0
E.c. + F.A.	192	137	4.40	22	11	3	0	0	3	70	469	37	4.61	2.59
Day 10														
No additive	179	143	4.30	54	69	16	0	0	16	29	622	3	0	0
E. coli	180	148	4.23	56	73	16	0	0	16	30	636	1	0	0
Formic acid	195	138	4.23	32	22	5	0	0	5	68	476	37	1.36	0
E.c. + F.A.	188	147	4.33	30	19	4	0	0	4	72	486	62	1.39	0.54
SEM (3-way interaction)	4.0	3.9	0.061	1.8	2.1	0.6	0.04	0.05	0.6	3.1	12.7	16.3	0.520	0.192
Significance Day (D)	**	**	***	***	***	***	NS	**	***	***	***	***	***	***
Formic (F)	***	***	***	***	***	***	NS	*	***	***	***	***	NS	***
E. coli (E)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*
D x F	NS	*	***	**	***	***	NS	**	***	***	***	*	NS	***
D x E	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	**	*
F x E	NS	NS	NS	NS	NS	*	NS	NS	*	NS	NS	NS	NS	*
D x F x E	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*

EB = Enterobacteria (log_{10} colony forming units (CFU)/g) recovered on VRBGA; E.c. = *E. coli* 0157:H7 (log_{10} CFU/g) recovered on TSA/SMAC

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Effect of ethanol in silage on milk quality and composition

The problems with milk tainted by feed have increased in Norway during the past few years. In Ireland, high ethanol concentrations were found in silage from herds with feed-tainted milk (P.A.M. Rogers 1993, pers. comm.). The main objective of this study was to examine whether ethanol, in concentrations that could be found in well fermented silage, could impart feed-flavour to milk.

MATERIALS AND METHODS

A crossover study using 24 dairy cows was carried out in February and March 1997. Two weeks prior to the study, the cows were assigned to six blocks of four similar cows with regard to parity, days postpartum, silage intake, milk yield and composition, and BW. One cow from each block was allocated to each of four groups, so that the groups were as equal as possible. Throughout the study two groups were offered barley treated with propionic acid and two groups ensiled barley. All cows received well fermented grass silage *ad libitum*, a restricted amount of a protein concentrate, and 100 g of a mineral and vitamin supplement. Chemical composition of the feeds are given in Table 1. During a 9-d period, one of the two groups of cows fed each barley type were supplemented with 600 g daily of pure ethanol. During a 5-d transition period no ethanol was fed, and thereafter the last two groups of cows were fed ethanol during a second 9-d period. The ethanol was fed in a solution containing 205.5 g/kg of ethanol, and was poured on and mixed into the grass silage ration of each cow. Grass silage and ethanol were fed at 7.00, 13.00 and 15.30 h. The cows were milked at 6.00 to 7.00 h and at 15.30 to 16.30 h. Milk samples were taken from morning and evening milk on day 2 and 9 during each period and judged for taste and aroma by a sensory panel of three persons at the Norwegian Institute for Food and Environmental Analyses.
In a study in October 1997, duplicate milk samples from 33 cows were used to evaluate the taste and aroma of milk to which 0.25 g/kg of ethanol was added.

Table 1. Chemical composition of the feeds

	Grass silage	Propionic acid treated barley	Ensiled barley	Protein concentrate
DM, g/kg	245	669	618	885
pH	4.01	4.09	4.50	
NH_3-N, g/kg TN	78	8	19	
g/kg DM:				
OM	933	984	980	896
CP	171	115	126	380
True protein	97	91	96	337
Starch		562	571	172
Sugar	19	20	18	
Crude fat	38	25	28	64
NDF	510	269	224	151
Lactic acid	61	0	12	
Formic acid	12	0	0	
Acetic acid	16	0	2	
Propionic acid	0	34	0	
Butyric acid	0	0	0	
Ethanol	3	0	7	

RESULTS

Milk yield tended to decrease when ethanol was fed, but due to increased fat and protein concentrations, the yield of ECM increased by 0.9 kg. The concentrations of lactose and urea in the milk decreased, the concentrations of acetone and ethanol increased, the concentration of FFA tended to increase, whereas α-tocopherol-concentrations were unaffected (Table 2). The milk obtained poorer scores for aroma and taste when ethanol was fed, and feed flavour was the dominant off-flavour. The effect of ethanol on milk composition was in general smaller for cows fed barley treated with propionic acid than for cows fed ensiled barley.

Table 2. Feed intake and milk yield over the two periods, and average milk composition for day two and nine

	Cows fed propionic acid treated barley (n=12)				Cows fed ensiled barley (n=12)				All cows (n=24)			
	Con-trol	Etha-nol	SEM	*P*	Con-trol	Etha-nol	SEM	*P*	Con-trol	Etha-nol	SEM	*P*
Silage, kg DM	10.3	10.1	0.12	NS	10.5	10.4	0.12	NS	10.4	10.3	0.08	NS
Barley, kg DM	6.15	6.12	0.018		6.07	6.08	0.009		6.11	6.10	0.020	
Conc., kg DM	2.01	2.03	0.011		1.99	1.89	0.045		2.00	1.96	0.025	
Ethanol, kg DM	0	0.566	0.003		0	0.571	0.002		0	0.569	0.002	
Milk, kg	27.2	27.2	0.20	NS	26.6	25.9	0.20	0.02	26.9	26.5	0.14	0.08
ECM,[1] kg	26.8	27.6	0.18	0.02	26.3	27.2	0.17	0.004	26.5	27.4	0.12	<0.001
Fat, g/kg	38.1	40.3	0.28	<0.001	39.5	43.5	0.34	<0.001	38.8	41.9	0.26	<0.001
Protein, g/kg	33.9	34.3	0.10	0.01	33.4	35.1	0.16	<0.001	33.7	34.7	0.13	<0.001
Lactose, g/kg	46.4	46.0	0.06	0.005	45.7	45.3	0.08	0.005	46.0	45.7	0.05	<0.001
Urea, mM	5.80	5.47	0.053	0.002	6.09	5.74	0.061	0.003	5.94	5.60	0.038	<0.001
Acetone, mM	0.03	0.04	0.001	<0.001	0.04	0.09	0.006	<0.001	0.03	0.07	0.005	<0.001
α-toc,[2] mg/kg	0.95	0.89	0.038	NS	0.84	0.84	0.052	NS	0.89	0.87	0.031	NS
Ethanol, g/kg	<0.01	0.04	0.004	<0.001	<0.01	0.07	0.008	<0.001	<0.01	0.05	0.005	<0.001
Milk taste[3]	4.04	3.74	0.082	0.03	3.78	3.15	0.100	0.002	3.91	3.44	0.067	<0.001
FFA, m.eq./L	0.72	0.75	0.035	NS	0.65	0.74	0.023	0.03	0.69	0.74	0.020	0.07

[1] Energy-corrected milk

[2] α-tocopherol (Vitamin E)

[3] Five-point scale for milk aroma and taste where 1 = poor quality milk, and 5 = high quality milk

DISCUSSION AND CONCLUSIONS

Forages with a high initial concentration of water-soluble carbohydrates are most susceptible to ethanol fermentation, especially if lactic acid fermentation is depressed (e.g., in silages treated with a small quantity of formic acid). Ethanol fermentation may occur in silage with low as well as with high DM concentrations (Driehuis and van Wikselaar 1996).

The physiological and biochemical pathways responsible for inducing off flavour in milk when cows ingest ethanol are not fully understood. Ethanol transmitted from the feed to the milk could not explain the observed off-flavour in the milk. The fact that silages which are classified as well fermented, and having a pleasant appearance and aroma, still may contain ethanol in such amounts that it may impart feed flavour in milk, is nevertheless of considerable practical importance. Precautions should be taken to avoid extensive ethanol fermentation in grass silage and other feeds that are to be fed to dairy cows.

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A new fungal problem on big-bale silage in Ireland

The problem

While mold colonies of microfungi such as *Penicillium* and *Fusarium* are commonly seen on conventional and on big-bale silage, macrofungi are rarely encountered. However, within recent years a mushroom growth has been appearing with increasing frequency on big bales in Ireland. The mushroom is *Schizophyllum commune*, a gilled bracket fungus (phylum Basidiomycota). The fungus is best known as a white-rot fungus with a worldwide distribution. Its small bracket-like fruit-bodies are commonly seen on fallen branches in deciduous woodland, especially in warmer climates. It also occurs in temperate regions, on wood and a variety of other substrata (Cooke, 1961). While usually considered a saprotroph, it is pathogenic on a variety of plant hosts and is increasingly being reported as a human pathogen (Sigler *et al.*, 1997).

Schizophyllum on big-bale silage in Ireland

The first recorded occurrence of *S. commune* on big-bale silage in Ireland was in October 1990, in Co. Leitrim. In November 1991 the fungus was found on big bales in Co. Tipperary, 150km from the first location. Interestingly, Webster (1991) reported finding *Schizophyllum* on silage in Devon, England in October 1990. The Irish finds were of particular mycological interest because *Schizophyllum* had been recorded only once before in Ireland, 150 years previously on wooden beams at Cork Harbour. By 1995 agricultural interest in *Schizophyllum* intensified as more big bales at various locations were found to be colonised by the fungus. A recent countrywide survey conducted by the authors showed that *Schizophyllum* is now of common occurrence on big bales throughout Ireland. In January 1999 collections of big bales were inspected along six routes, each *c*a 150km long and traversing regions of different agricultural enterprise. Fifty farms were visited along each route and bales were examined for the presence and extent of *Schizophyllum,* in addition to other bale parameters. The fungus was recorded present on 159 of 300 farms (53%), varying in incidence from 20 to 80% of farms, depending on geographical region. The extent of occurrence within a collection of bales also varied, from one or a few bales affected to >75% on some farms. The fungus occurred on silage from diverse sward types, cut at different times during summer and autumn and wilted to varying degrees. Both black and white-wrapped bales were affected and the fungus grew regardless of bale orientation during storage. Correspondence with researchers in Britain and other countries does not indicate that *Schizophyllum* is a problem on baled silage in Europe or elsewhere.

Features of *Schizophyllum* on big bales

Schizophyllum can develop in bales within four to six weeks of wrapping. From August onwards, depending on bale age, *Schizophyllum* is readily visible on the plastic surface of affected bales. Having emerged through the polythene wrap the fungus is first evident as

small white growths which eventually expand and develop into gilled bracket mushrooms, often in dense overlapping clusters 10 to 15cm in diameter. More extensive growths to *ca* 50cm wide have been seen. The individual mature fruit-bodies (basidiocarps) are tough and elastic, grey-white to brown in colour, fan-shaped with very short stipes and a tomentose/downy wrinkled upper surface. Cap margins are furrowed, scalloped and incurved. The gills are peach/pink when young and spread out radially from where the fruit-bodies are attached to the bale. With age the gills become brown to grey in colour. A characteristic feature of the gills is the manner in which they appear to split, hence the common name for the fungus - 'split gill'. By springtime, older *Schizophyllum* specimens are often shrivelled, brittle and are readily dislodged. A green algal growth may be present on the surface of such specimens.

Schizophyllum within bales

Before emergence through the polythene, the occurrence of *Schizophyllum* in a bale is indicated by the presence of small firm bumps under the polythene surface. The fungus may occur in any position on a bale but most often on the curved sides and shoulders. When the plastic covering is removed from a bale, mats of white to tan-coloured mycelium can be seen on the silage surface, usually accompanied by masses of dense rubbery differentiating tissue. Mechanical pressure exerted by the enlarging fungal mass causes a stretching of the polythene wrap and its eventual penetration or splitting by the fungus. Within bales, *Schizophyllum* colonies ramify widely and deeply. The diffuse white mycelium is not macroscopically distinguishable from other fungi but microscopically can be identified by the presence of clamp connections and numerous minute peg-like projections along the hyphae. The fungus is readily grown in culture on malt extract medium supplemented with chloramphenicol and chlortetracycline (both at 0.15mg ml^{-1}) to inhibit bacterial growth. Most isolates form fruit-bodies within 4 to 8 days. Other species of *Schizophyllum* are known but only *S. commune* has been found on bales.

Consequences of *Schizophyllum* on big-bale silage

Within the past decade *Schizophyllum* has established rapidly in its new niche on big-bale silage and is now widespread in Ireland. It has not been found on conventional pit silage and is of rare occurrence in Irish woodlands. Its growth on silage causes loss of feedstuff and also, through air ingress, facilitates growth of aerobic molds and bacteria with consequent health risks to livestock and humans. *Schizophyllum* can be a human pathogen which gives further ground for concern. Farmers are advised not to offer affected bales to livestock. Meanwhile, both field and laboratory experiments are in progress to determine what factors predispose big-bale silage to growth of *Schizophyllum commune* and how it might be controlled.

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Maize silage in Sweden

Maize area

South- Sweden represents the northernmost cultivation zone for maize in Europe. During the last fifteen years the maize crop rather constantly has covered about 2 000 hectares. The new EU subventions for maize crop might increase the interest for this cultivation also in Sweden. Furthermore, variants with better cultivation qualities have appeared in recent years.

Harvest in late autumn

Normally, the maize harvest will be performed during the first part of October. The farmer wants a high dry matter content (ca 30% DM) as well as a high proportion of cobs in his silage. Therefore, he often choses to wait with the harvest until frost has dried the crop. Advisers often recommend this system.

However, in years with bad climatic conditions, like in 1998, the harvest will take place during rainy or frosty periods in late October or November. These late harvests may result in silage with bad hygienic quality and lowered feeding values.

Penicillium roqueforti — a problem

Normally, maize is easily ensiled and reaches low pH-values rather quickly. Therefore, many farmers do not use additives in the silo. However, the fungus *Penicillium roqueforti* is able to grow at low pH-values and in a nearly anaerobic environment.

Especially in late-cut maize silage, *Penicillium roqueforti* has appeared quite frequently, in some herds accompanied by high somatic cell counts in the milk. This may cause apprehension for problems in production of high quality milk. Therefore, a field-study on hygienic quality in maize silage is running in Scania during the winter 1998/99.

Actual study

In this study 40 dairy farms are followed, 20 of them with and the other 20 without maize silage. Information is collected around the following facts.

- Maize cultivation methods, i.e. variety, amount of seed, date of sowing, fertilisers, irrigation, plant protection
- Data about silo preparation, harvest technique, additives, silo covering, harvest date

- Samples are taken on all types of silage on the farms for hygienic evaluation
- Data around the somatic cell counts in tank milk during the last 15 months

The intention is to find measures in cultivation and harvesting, which can be used as advises to the farmer in order to secure a high quality in maize silage during Swedish climatic conditions.

Preliminary results

During previous years, intensive extension service was given to some of the experimental herds, due to high cell counts in milk related to quality problems in maize silage. The year 1998 was an extremely bad year with rain in the most part of the growth and harvest seasons. This was expected to result in great microbial stress in forage silages.

However, preliminary results from our study indicate, that well educated farmers were successful in silage making from both maize and grass, even during these bad circumstances. Many farmers in both groups, lacking the intensive extension mentioned above, produced silages (maize and/or grass) of worse hygienic quality. The fungi *Penicillium roqueforti* was the dominating type of mould.

Important factors, except for extreme weather, influencing the maize silage quality seem to be as follows.

- Sowing date. The plant must have a chance to reach dough maturity
- Use of varieties, keeping green and fresh until harvesting. These give better chopping and compaction in the silo
- Use of additives (formic acid/propionic acid)
- Harvest time. Avoid late harvesting and don't wait for frost in order to increase DM content. Late cuts in November - December reduce nutritive and hygienic values
- Extension of farmers

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Microbiological status in tower silos

Background and methods

To get a general idea of the hygienic status and cleaning needs in steel tower silos for silage, a survey was carried out in 1997. During spring, tower silos on ten farms were inspected. Both silage and feed residues in various parts of the silos were sampled and examined for microbial growth. The mould *Penicillium roqueforti,* which is suspected of suppressing the immune response in cows and thereby increasing the incidence of mastitis, received special attention.

Results

Feed residues with extensive growth of aerobic micro-organisms were found in all silos, especially on the filler-unloader and in the upper parts of the silo. In all silos *P. roqueforti* was isolated from samples taken from either the silo walls, filler-unloader or upper part of the tower. It was also recovered in all but one of the samples taken from the silage surface. *P. roqueforti* was isolated from samples taken at a depth of 25–35 cm below the silage surface in five of the ten silos.

In general, the higher up in the silo samples were taken, the larger were the amounts of yeast, moulds and aerobic bacteria, fig 1. The amount of *P. Roqueforti* classified on a scale 0-4 showed the same general increase with height in silo, fig

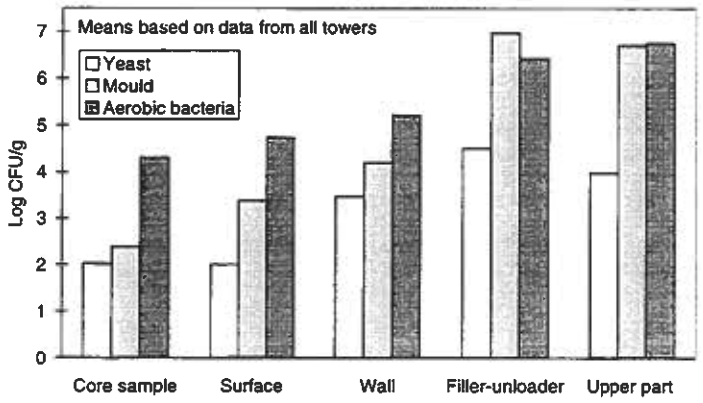

Figure 1.

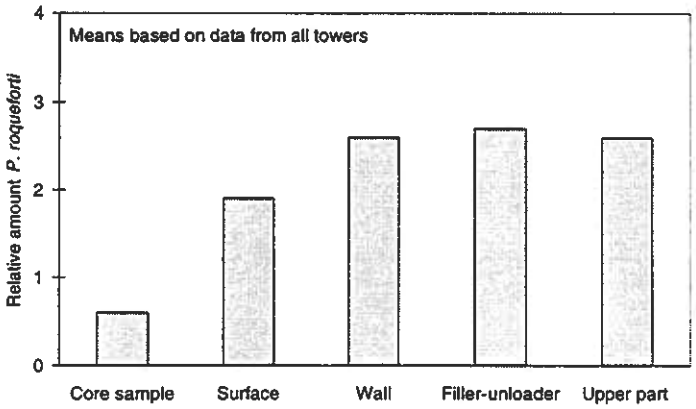

Figure 2.

2.

Conclusions

The study showed that *P. roqueforti* is common in tower silos and can be found in large amounts, especially in feed residues. The regular occurrence of *P. roqueforti* in the surface layer of silage is most likely due to spore dissemination from upper parts of the tower and filler-unloader in response to vibrations and air movements that occur during unloading.

Ongoing studies

To elucidate the extent to which a re-infection of the silage surface can be prevented or reduced by removing old feed residues, a full-scale experiment was started in 1998 in which the effects of different cleaning procedures were assessed.

The study included three treatments (two silos per treatment):

A Control (no cleaning operations)
B Cleaning with a heavy jet of water
C As in B, supplemented with a chemical disinfection

Cleaning and disinfection were performed in empty towers in late May 1998. During the subsequent feeding period all silos will be sampled on three occasions, using a scheme identical to the one applied in the survey. This phase of the work had not been completed as of March 1999.