ANNUAL REPORT

Trees and Crops for the Future, TC4F

Trees and Crops for the Future - TC4F - develops knowledge on sustainable plant production and plant based product development within agricultural and boreal forest systems with the main objective to support the development of a new circular bioeconomy in Sweden.



TREES FOR THE FUTURE CROPS FOR THE FUTURE



Trees and crops for the future, TC4F Annual report 2022

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Trees and Crops for the Future-TC4F

Trees and Crops for the Future – TC4F – is a strategic research program that develops knowledge on sustainable plant production and plant-based products of forestry and agricultural systems. TC4F's main objective is to provide fundamental research that generates knowledge to support the transition towards a sustainable circular bioeconomy.

The TC4F research program focuses on two plant production systems, trees (T4F) and crops (C4F). Within the third phase (2021-2025) of the program, the primary objective is to support knowledge contributing to the development of a sustainable circular bio-economy, utilizing the unique and common aspects of forests and agricultural systems. The transitioning from fossil derived products (fuels, materials and chemicals) towards biobased alternatives along the circular bio-based economy path is more important now than ever, with the increasing global threats from climate change, pandemics, migration and regional conflict.

National preparedness to secure availability of food, fuel, material and chemicals have become an important issue on the agenda. Fossil-based emissions are the primary driver of climate change, which may have irreversible impacts on domestic forest and crop production in the future. Thus, developing forest and crop systems that simultaneously sequester and store carbon, and supply the economy with alternatives to fossil intensive products is an imperative.

Secondly, given that Sweden is not a fossilbased product producing country itself, a circular economy based on domestically produced biobased products will help reduce geo-political risks, for example disruptions in global supply chains. The transition to a carbon neutral bio-based economy is bold in concept, and requires new and cutting edge science to deliver the knowledge that will allow forest and crop production to be maximized in the face of ongoing climate change. This requires basic and applied research focused on enhancing ecosystem carbon storage, productivity, linkages between production and soil communities, as well as research regarding plant-based products.

In 2022, the TC4F program continued to push numerous research boundaries to address these goals, including:



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Trees for the future (T4F):

• Establishment of two new national scale forest field experiments to quantify how mixed species forests and forest genetic enhancement can improve forest productivity, resilience, and carbon storage.

• Initiation of two new research projects focused on forest genetics and breeding.

• Initiation of two new research projects aimed at better understanding how forest compositional choices affect production and carbon sequestration.

• Initiation of two new research projects focused on forest fertility, and soil carbon storage.

• Publication of 49 new scientific manuscripts addressing the above themes.

• Mentorship of 24 junior scientists (PhD students and Post Doc researchers). A total of 26 researchers recieve part of their salary from T4F.

• Ca. 100 milj SEK were received from other funding sources for project deriving from T4F.

Crops for the future (C4F):

 Five C4F projects ended in 2022, within the topics of plant-based biostimulants, domestication of Lepidium campestre, regulation of carbon flow, insect pheromone production in plants and alternative starch qualities

Michael Gundale Program Leader T4F Göran Ericsson Dekan SFAK • Six new C4F projects were started in the research areas of medium chain fatty acids, potato tuber sink and starch development, wax esters, starch properties, induced mutations for desired traits and genetic background for quality traits

• Six new PhD-students/post-docs were started within the five above mentioned projects

• A large number of new projects were funded based on research carried out in C4F based projects, as an example, a total of 25 milj SEK was received in 2022 from Formas, Vinnova and Energimyndigheten for projects related to the Plant Protein Factory.

• A total of 18 scientific peer-review papers have been published..

Two PhD-students defended their theses

Our research plan, and scientific output covers a broad range of scale, from genes, individual plants, as well as whole agricultural and forested landscapes and products thereof. Our current planning phase (Phase III), will continue to deliver urgently needed knowledge for development of sustainable and resilient land management systems and plant based products for the future, which will contribute to a strong and vital circular biobased society.

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Bridging Basic Research to Application in TC4F

The research program TC4F takes fundamental research to application in many different ways. Here, some examples are illustrated.

Theme 1: "Breeding for the future" uses genomic research for applied tree breeding, developing more efficient and directed breeding technologies.

Theme 2: "Future forest composition - Tree species for the future" Future climate change demands new forest management tools that fit with the new climate. For this, preparation for the use of exotic tree species and increased use of underused native trees are an obvious path.

Theme 3: "Plant-soil interactions and soil carbon stockst" An important goal of this theme is to understand how a wide range of forestry activities influence soil carbon stocks through influencing carbon inputs from vegetation and the soil microbiome composition and activity.

Program part **C4F** develops many plant related products, for example superabsorbent materials from wheat protein and plant based oils and fuels. *Text Anke Carius,*

Illustration Daria Chrobok and Anke Carius



T4F

Breeding for the Future

AIM: Tree production and health



T4F: Genetics and Breeding

To ensure maintained or improved productivity, improvements to feedstock quality and resilience to changing climate conditions requires rapid genetic improvement, which cannot be achieved using traditional breeding approaches with >30 year breeding cycles. Coupled to this, there is a pressing need to understand ecosystem functions and how these will be affected by climate change, including how soil microbial communities will respond and how those responses will affect interaction with other species, such as how the economics of carbon-for-nitrogen exchange between ectomycorrhizal fungi (EMF) and Norway spruce and Scots pine will be altered. Developing such understanding requires forming links from molecules through to the ecosystem, employing genetic and genomics approaches, and integrating these to the landscape scale. A fundamental requirement for this on both the breeding and microbial community sides is the need for genomic tools including genome assemblies, population genetics data, suitable analysis pipelines, and user-friendly tools for utilising and exploring the data.

Project Updates

O Quantified the effect of different nitrogen sources on autumn senescence (Fataftah et al. 2022).

o Preparations began for a new aspen common garden experiments focused on assessing natural variation and evaluating genetic modifications (GMO).

o Defined a new light protection mechanism in conifer needles (Bag et al, submitted).

o Finalized a multi-year/multi-genotype/ multi-omics study on autumn senescence in aspen (Lihavainen et al, submitted).

o We have applied the long non-coding RNA pipeline developed during 2021 to datasets from aspen (leaf and wood) and Norway spruce (drought, cold, seasonal variation, wood and needle development) to identify and rank-order lincRNAs for downstream characterization.

o We performed a new Norway spruce drought experiment to perform RNA-Seq, sRNA-Seq and epigeneomics assays (ATAC-Seq, ChIP-Seq). o Completed gene annotation of Norway spruce.

o Increased bioinformatics capacity by recruitment of a new facility member (Aman Zare).

o Close collaboration in genomic selection in KAW funded project is continuing. An update of the results from the earlier genomic selection research in Scots pine together with radiata pine was collated in book chapter.

o Optimizing genetic gains and diversity in long term is crucial in terms of maintaining variation for future in tree breeding programs.

o Project for different breeding scenarios using simulations in Norway Spruce and Scots pine was initiated.

o We monitored genetic composition in nine seed crops from six pine and spruce seed orchards, finding distinct differences in the rate of background pollen contamination and genetic diversity between pine and spruce. o Background pollen contamination in pine crops varied from 87% at young orchard age to 12% at mature age, whereas this rate ranged between 27%–4% in spruce crops.

o Substantial variance in parental contribution was observed in all orchards with 30%–50% parents contributing to 80% of the progeny.

o Selfing was low (2%–6%) in all seed crops. Compared to natural stands, orchard crops had slightly lower heterozygosity but no strong signal of inbreeding.

o Examined the frost hardiness variation in seed orchard crops intended for the most northern climate zone. We found that seedlings produced by mating among orchard genotypes were less hardy than expected while the opposite was observed in seedlings sired by external pollen. Our results suggest that parent genotype origins, mating composition and maternal epigenetic effect could all influence the climate adaptation of orchard crops.

Societal value

Forestry in Sweden faces increasing pressures from climate change, management policies and the need for biodiversity maintenance and ecosystem services such as carbon sequestration and storage. These often-conflicting pressures must be balanced against the need to maintain economic viability of the industry. Forests serve important societal and cultural roles and the influence of changes in forestry practise must be balanced against these needs. One of the most pressing issues is the need to develop, either through breeding or genome engineering, climate resilient trees for replanting of forests. This theme is developing resources to enable both approaches and to further deploy the developed tools in breeding programs. The genomic resources we are establishing are essential to enabling genomic based selection, a breeding approach that massively reduces the breeding cycle period by enabling selection of elite or desirable trees at the early seedling stage using only genetic marker information.



Map indicating the pine (A) and spruce (B) orchards and natural stands included in the study. Observed heterozygosity (Ho), inbreeding coefficient (FIS) are shown in the middle panel, variation in frost damage of pine orchard crops is shown on the right panel.

Text by T4F coordinator group: Vaughan Hurry, Nathaniel Street, Mari Suontama, Urban Nilsson and Michael Gundale Illustration: Daria Chrobok, Nat Street

Photos: by authors



Future forest composition

AIM: Use of exotic tree species & underused native species



T4F: Future forest composition – Tree species for the future

With climate change the need for adaptations of forest management has been addressed by the forest sector and forest agency. One strategy is to increase the proportion of underused native trees, another to increase knowledge on the potential of non-native new tree species. Besides tree species choice in plantations, changing silvicultural systems must also be further investigated, i.e. using clear-cut free forestry and natural regeneration under shelterwoods. Since many of the above-mentioned management tools are poorly represented in field experiments and operational forestry, models and simulation studies need to complement empirical data when they are evaluated. In addition, climate change will make current empirical models less accurate because future climate will not be the same as when data for the models were collected. Therefore, new growth models need to be developed to capture the effects of a changing climate on tree growth and damage.

Project Updates

The two most interesting tree species to introduce as supplement for Scots pine and Norway spruce are Silver birch and Siberian larch (also known as Russian larch). The reasons why they are especially interesting are because of their good potential growth and their classification as native tree species by the Swedish Forest Agency. During 2022, T4F together with the Forest Agency, investigated the practical use of Siberian larch in northern Sweden.

T4F also collaborated with the research program Trees For Me which aims at increasing the use of Silver birch substantially but also the use of the fast growing broadleaves European aspen, hybrid aspen and poplar.

Non-native tree species are restricted by certification rules today and can currently be regarded to be of less interest. However, climate change may result in a different view on non-native tree species and in T4F the research on potentially new tree species is regarded as a strategy for future adaptations to climate change in the Swedish forestry. T4F collaborated with Skogforsk in a project with the aim to analyze a 40-year-old tree species experiment in northern Sweden with eight native and non-native tree species. Exotic tree species have also been addressed in several excursions that we have contributed to. When introducing new tree species, future damage is of special interest. Therefore, a post-doc has been examining possible damage to the three larch species commonly used in Sweden. The postdoc program will result in a review of Russian, European and hybrid larch potential and a study of resistance biology of the different larch species including the role of plant secondary metabolites in defense of larch canker disease (Lachmellula willkommii).

Regeneration of Scots pine in northern Sweden is currently very problematic. The term multi-damaged forests is used to describe a situation with damage from browsing, fungi, and insects. We have, with a simulation study, shown that current regeneration results will result in significant production losses and significant reduced effects of improved genetic material. We have established a regeneration experiment on nine locations in northern Sweden where novel scarification methods are tested in combination with tree species and fertilization at planting. We will continue to establish regeneration experiments during the coming years.

Mixed species forest will most certainly become more common in the future. Together with researchers from LUKE research institute, Finland, we have analyzed the current status of Swedish and Finnish forests and evaluated growth models for mixed species stands. The next step is to make a simulation study where various levels of mixed species stands are evaluated on a landscape level.

A possible effect of climate change is the increased frequency of dry years similar to what we experienced during 2018. It is possible that forest management need to adapt to shortage of water for tree-growth. We have examined Norway spruce and Scots pine growing side by side in randomized experiments to study tree species differences on the effects of dry years on tree growth. Another study has been on request from Södra skogsägarna, on sites where they had found sudden pine needle casts during recent years. Here we have demonstrated how the dry summer 2018 probably shifted an opportune fungus to be a pathogen on Scots pine.

We will establish a nationwide mixed species experiment on 10 locations from Halland in the south to Västerbotten in the north. During 2022, five sites have been selected and seedlings have been cultivated for the planting on five sites during spring 2023. The last five sites will be established during spring 2024.

Societal value

Multi-damaged forests in northern Sweden have been investigated together with a group of practitioners and researchers led by the Forest Agency. We have participated in more than 20 excursions and seminars when T4F-research has been communicated to the forest sector. T4Freserachers have been interviewed in several forest magazines making the research visible for Swedish forest owners and many more that read forest magazines. The impact of T4F-research can be seen on the demand for Silver birch and Siberian larch seedlings. Because of great interest in alternative tree-species there are absolutely no seedlings available of these two tree-species for the planting season of 2023. All available seedlings sold out early in the autumn. It is of course not only research and communication from T4F that has inspired this interest, but we believe that it has contributed significantly.

Damage to forests is a major threat to future forest production. Together with the forest damage center (FDC), researchers from T4F have been involved in several projects aiming at reducing the impact of future damage. Examples are constructing models for wind-damage and damage by fire. We have also been involved in examining methods for reducing damage by browsing with the application of repellants.



Much of the research is done in close collaboration with the forest sector ensuring that research results can find their way to fast implementation in practical forest management.

Text by T4F coordinator group: Vaughan Hurry, Nathaniel Street, Mari Suontama, Urban Nilsson and Michael Gundale, Illustration: Daria Chrobok, Photos: Anke Carius





T4F: Tree-Soil Interactions and Forest C Stocks

The third major objective and theme of T4F is to understand the impact of tree, stand and landscape level forest composition choices on landscape carbon balance, which includes consideration of tree biomass growth, as well as impacts on soil biodiversity and soil carbon stocks. Climate change, causing both soil warming and changing soil water availability, is expected to have a strong impact on soil carbon stocks by altering both the quantity and quality of carbon inputs to soil, and by impacting the composition and activity of the soil microbiome. The soil microbiome serves as a very important control on soil carbon by controlling the decomposition of soil organic matter. The regulation of soil organic matter dynamics not only influences soil C stocks, but also influences tree growth through their nutrient recycling activities that supply trees with nutrients. The net impacts of forest production and management on soil and ecosystem biodiversity, and carbon and nitrogen exchange remains debated, and thus unravelling the direct and indirect effects of forest composition and forest management activities on soil biodiversity and soil carbon turnover remains a critical frontier in understanding how forestry can be used most effectively to achieve carbon neutrality goals.

For this reason, the research priority for this theme within T4F is to study the impact of forest management, including the new genetic and compositional approaches described in Theme 1 and 2, on plant-soil microbiome interactions, soil carbon cycling processes, and their contribution to net ecosystem biodiversity and carbon exchange. These frontier research questions need new tools if they are to be answered, and within T4F we have not only been implementing existing technologies, such as DNA amplicon sequencing to assess soil microbial diversity, but have also been playing a leading role in developing new methodologies, such as soil microbiome-tree metatranscriptomics, soil dialysis and establishing an "artificial root" system to study root-soil microbiome communication mechanisms. Milestones 2022

o In 2022 we published a break-through study where we analyzed in detail the seasonal transcriptional activity of spruce roots and demonstrated how this was coordinated with the transcriptional behavior of the root-associated ectomycorrhizal community – and how this

- coordination is changed under altered nutrient availability (Law et al, 2022 PNAS).
- o In 2022 we completed a study describing the recruitment of the root microbiome by Scots pine and Norway spruce seedlings following outplanting into clear cuts, describing the legacy effect of nursery production, and the positive effects of adding small amounts or organic, but not inorganic, nitrogen to the outplanted seedlings (Schneider et al, in prep).
- o We showed that diffusive sucrose efflux from microdialysis probes increased in soils amended with N-rich litter. Our study confirms that microdialysis allows time-sensitive insight into the dynamic interactions of carbon and N in the rhizosphere (Buckley et al 2022).
- o In a parallel study we show that microdialysis can be used to study growth response of the soil microbial community to the release of carbon into the rhizosphere in the form of sucrose. This shows the potential of this technique in unravelling plant microbial interactions from a plant root signaling perspective. This was visualized with scanning electron microscopy



and the stimulated fungi were identified with DNA sequencing. The uniqueness of using microdialysis to study microbial interactions is the possibility to not only release a compound of interest, but to also follow the resulting effect on microbial community composition and activity. (Buckley et al in prep).

Initiatives for 2022

o A new post doc (David Castro) was recruited to study carbon and nitrogen transfer between tree and fungi in the field. David will also collaborate on projects to investigate the effect of exotic forest plantations on soil microbial biodiversity and carbon sequestration, and also on the effect of monocultures vs mixed species forests on soil biodiversity and carbon storage

o A new post doc (Andreas Schneider) will investigate long term effects of nitrogen fertilization on root associated fungal activity and the subsequent changes to this community after fertilization is stopped. This project will investigate whether there are any long term effects (up to 30 years after ceased fertilization) on fungal community activity that might affect soil carbon accumulation and plant nitrogen availability. In many studies the effects of fertilization on soil microbial composition is transient, but there are few studies looking at the effects on soils and soil microbes after fertilization is stopped.

o A new post doc (Clydecia Spitzer) will evaluate foliar and root growth traits across 4 native and 8 exotic tree species. The project aims to evaluate whether leaf and root acquisition strategies are consistently related to each other, and which traits can best explain soil carbon accumulation rates. The work will be fundamental for understanding how species selection impact productivity and carbon sequestration, and the relationship of these two ecosystem services to each other.

Societal value

While forest production has long been valued, society increasingly values forests for other ecosystem services, including carbon sequestration. For society to make informed decisions about synergies and trade-offs between production and whole ecosystem carbon sequestration (which includes soil C), we need a detailed mechanistic understanding of management and utilization affect soil microbial communities, both function and diversity, and in turn how this impacts soil carbon stability and accumulation. This detailed understanding will inform us about the fundamental relationship between aboveground production, and soil carbon accumulation rates, which is needed to inform this discussion.

The metagenomics, metatranscriptomics and soil dialysis protocols we have established within the T4F program have been successfully deployed in the field to study forest ecosystem-level responses. Within T4F, these tools are now increasingly being deployed to understand how the macro above ground impacts of forestry activity, which we can all observe, impact on micro below ground diversity and the soil and ecosystem processes that depend on this highly diverse community.

Text by T4F coordinator group: Vaughan Hurry, Nathaniel Street, Mari Suontama, Urban Nilsson and Michael Gundale Illustration: Daria Chrobok, Simon Law Photos: Anke Carius

T4F Field Experiments:

In 2022, T4F initiated two new national level experiments to address important issues in Swedish Forestry, including an experiment focused on forest genetics, and an experiment focused on mixed species forests. Both genetics and forest mixtures are potential management strategies to make forests more resilient to climate change or promote climate change mitigation by enhancing carbon capture.

Regarding genetics, breeding programs over the past half century have actively selected for trees with higher growth rates. Whether these genetic improvements to growth also lead to increased stand level carbon stocks, including soil carbon pools, remains unknown. To address this knowledge gap, we are establishing an experiment that represents a gradient of genetic improvement. The experiment will focus on Norway Spruce (Picea abies), and will focus on four different levels of genetic improvement (% growth increase), including: 1) unimproved trees (0%), 2) the first generation seed orchard material (10% improved), 3) second generation seed-orchard material (15% improved), and 4) the cross-control material from the latest breeding cycle (25-30% improved).

The experiment will be set up in each of three sites, including Västerbotten, Dalarna and Västergötland. The seed material that will be planted at each location will come from the specific breeding zones for which the genetic improvements have been made. The experiment will consist of 16 plots in each trial, including 240 trees per plot, total 3840 trees per site and 11 520 trees in total across the three sites. In 2022, sites for the experiment were selected and clear-felled. Nursery seedling of each improvement level were also initiated in the greenhouse. In the spring, 2023 plots will be established consisting of the different seedlings types. Regarding mixed species forests, researchers and society have debated the potential benefits of mixed species forest management, as an alternative to traditional mono-culture forestry. Compared to monocultures, several positive outcomes of mixed species stands have been identified, including increased biodiversity, water quality, esthetical and recreational values and reduced stand vulnerability to pest and pathogens.

Map over experimental sites throughout Sweden:

Mixed Forests

Two sites in Vindeln, one site in Siljansfors, Asa and Tönnersjöheden each.

The genetic experiment

is set up in Vindeln, Siljansfors and Remninsgtorp.

To address this knowledge gap, T4F began the installation of a new experiment in 2022 including experimental mixtures and monocultures of 4 different species, at 5 locations spanning a range of site conditions in all regions of Sweden. Tree-species that will be included in the experiment include Norway spruce, Scots pine, birch, and Siberian larch. In the experiment, plots with monocultures of each species will be compared with six two-species mixtures and one four-species mixture. In addition, we will establish monocultures and mixtures with two initial spacings, 1.5 x 1.5 m and 2.2 x 2.2 m, to understand how positive effects of species mixing might change with increasing competition intensity.

These treatments sum up to 22 plots in total on each of the 10 sites. In 2022, the first 5 sites were selected, including Småland, Halland, Dalarna, and 2 locations in Västerbotton. Sites were clear-felled, and planting will occur early in the growing season, 2023.

The experiment will provide a unique tool that currently is missing from the Swedish forest research infrastructure to evaluate species mixing effects on a wide range of ecosystem processes and properties, such as productivity, carbon sequestration, and biodiversity.

Text by T4F coordinator group: Vaughan Hurry, Nathaniel Street, Mari Suontama, Urban Nilsson and Michael Gundale Photo: Mari Suontama, Anke Carius

Aman Zare How curious are you, actually? - The Bioinformatics Platform UPSCb

Having worked with insects during his PhD, and later with human and mice data. Aman has been amazed by the complexity of plant genomes, even small plants that we see as cute little flowers can have massive genomes. Today, at the UPSCb Aman supports researchers with all kinds of bioinformatic tasks ranging from simple biological quality assessment, finding differentially expressed genes, enrichment analysis, and single cell analysis. Within T4F he is assisting with the huge task of assembling the spruce genome by TSSeq data analysis, independently of their model oraanism.

How is data typically processed at the UPSCb?

First check-up for transcriptional data: Was the sequencing deep enough? The first assessment of the data is whether the sequencing produced enough reads and whether there are anomalies in the dataset, for example batch effects or hints of uneven conditions durina sequencing. The number of reads depends on what kind of analysis the researcher would like to perform and for each one, a certain minimum number of reads should be achieved. The price for sequencing depends highly on the number of reads produced. Nowadays, most sequencing services are guite experienced, and it rarely happens that a dataset turns out to not be sequenced deeply enough, however, it needs to be checked, and the researcher will be informed.

Pre-Processing

For transcriptional analysis, ribosomal RNA should be filtered out. Adapter sequences can be left in the reads, and they should be

trimmed as well. As the sequencing proceeds, the later reads can be of poorer quality, and should be checked. Then the data set is cleaned up for these sequencing anomalies, it can be mapped to the initial genome. Main Processing At this stage, the quality of the data is more thoroughly assessed to see if the distribution of the data follows the expected pattern matching the analytical methodology. To compare datasets within an experiment, some normalization to internal controls is needed in order to account for technical variations After normalization, it is then possible to merge the biological replicates, or to consider them as separate biological replicates

Most of the time, people are primarily interested in identifying differentially expressed genes among their sample groups of biological replicates. However, deeper analyses can be rewarding:

- Geneontology generation
- Enrichment analysis
- Genes-of-interest network analysis
- Find underlying biological pathways

Some colleagues work on machine learning with focus on networks to try to predict pathways and unknown mechanisms.

Aman is the go-to person for anyone to analyse datasets, the bioinformatics platform help with bioinformatic work at upsc quality of data produced that the experiment was designed properly. This basically means that enough biological and technical replicates were included, that sequencing is "deep" enough, and that the data produced will be comparable. Even though co-workers in the bioinformatics platform have chosen to lay their focus on bioinformatics, they do have a biological education as a base and are fully competent to discuss experimental setups and recommend strategies and solutions if provided with information about what researchers are trying to find.

It is common, for example, that researchers want to compare expression levels when a particular abiotic condition is varied, such as temperature. The bioinformatics platform can then help to decide how many biological replicates should be included. Including too few biological replicates is the most common issue occurring with data in the pre-assessment. Of course, everyone is aware that more replicates are also more expensive, however it is good to keep in mind, that the most expensive data is that which cannot be analysed or is inconclusive.

etc.

Aman Zare: Studied molecular biology at Umeå University J Did Master's and PhD in Molecular biology Postdoc at Karolinska in Hong Kong in bioinformatics: Ming Wai Lau Centre for Reparative In PhD, he worked mostly with the fruit fly Drosophila melanogaster and in Hong Kong he Worked mostly with human and mice data. Started in August last year at the UPSC bioinformatics platform (UPSCb) and this was when Aman started to work with plant data Aman is the go-to person for anyone who needs

The UPSCb bioinformatics facility is currently expanding and hiring more personnel to fulfil the need created by the growing interest of researchers to have their data analysed by experts and also, as sequencing is getting more affordable, the increasing number of datasets to be analysed.

For this purpose, UPSCb offers packages that research groups can subscribe to that fall into the broad categories of long- or short-term support.

Often, PIs nowadays do not only want the data to be analysed professionally, but they also want to understand how the analysis is done, and they also want their PhD students and postdocs to understand so that they can learn to do at least some basic analysis themselves later on. The UPSCb platform has prepared a lot of templates and instructions and they will support young researchers throughout the process; even some tutoring can be involved.

The UPSCb offers Slack Communication, as it often works better than "email-chat" to communicate within the project (Slack is a chatbased collaboration tool, an app, that emphasizes communication), so it is easy to ask questions! In other cases, a PI can choose a complete handsoff data analysis service, which then will be provided by the experts in the UPSCb platform. After the project, the platform can give recommendations about future questions and new setups, for example which genes they would see most likely to respond to targeting, if a metabolomic analysis could be interesting or if there are more bioinformatics tools that they can use on their data.

At the end of a project, the platform can provide figures and material and methods for the bioinformatics part of the manuscript, and can also help with the writing of discussions, just as one might expect in any good collaboration.

Fig. 1: A closer look at step 4 a/b The actual analysis of the data: Image by https://raw.githubusercontent.com This map shows the processing of data at a bioinformatics facility in thechnical terms. The stops are named with methodologies used, stages are discribed in the legend, as they also occur in the main text.

Step 1: Consult with UPSCb before creating data!

> Fig 2: A simple roadmap about how a researcher should proceed if they would like to use the bioinformatics platform.

Step 6: Crawl deeper down the rabbit hole.

Step 1: Consult with UPSCb how to proceed for deeper analysis, new directions and insight or completely new approaches.

There are always new things to be discovered in biology, so the question really is: How curious are you? How deep do you want to go down the rabbit hole?

Aman himself has always been curious about the geographical diversity of trees, both longitudinally and latitudinally, for example differences between Sweden and Norway. Aman has plans to join a field work group to sample Aspen roots on widely distributed sites all the way to Mo i Rana in Norway. So, Aman hopes to come full circle on the data produced from this trip by having the chance to enrich the analysis of it with his bioinformatic skillset.

Text and illustration: Anke Carius

Adding depth to forestry research:Clydecia SpitzerCorrelation between above and below ground tree traits.

Many people have seen a meme recently circulating on social media showing two significantly different sized carrots, but with inverted amount of foliage. A plant biologist could react to a few things in this image. First of all: Is it realistic that the size of the root in relation to the leaves can vary so much in a single species? If you have ever grown carrots in your own garden, you may know that it can actually be difficult to tell how big a carrot will be just by looking at the green shoot. This is the basic challenge in plant biology, to see fundamental patterns in nature through a noisy background of random variation.

Root traits in trees have become a hot research topic in forestry research, not only when determining how much carbon a forest stores, but also from an ecological perspective as to how the development and production of roots affects symbiotic partnerships with soil fungi (i.e. mycorrhiza). While root-shoot trait correlations would be relatively easy to investigate with carrots, it is still debated how well above-and belowground characteristics relate to each other in trees. An extraordinary young

> Photo 1: Clydecia Spitzer Illustration: Carrot meme, starecat.com

postdoc has now set out to provide us a better understanding of what is going on below ground in the forest, and whether these

> processes can be revealed through simple measurements of above ground traits. Clydecia Spitzer grew up in Guyana in South America, a country that has been significantly exposed to severe climate change effects. Therefore, she has made it her life's goal to contribute to research focused on carbon sequestration, since it is a main tool to mitigate climate change. During her PhD work, Clydecia in comparing root traits to above ground traits in Swedish Sub-arctic

tundra, in order to help better understand how sub-arctic environments are responding to climate change. She has now started her post doc project on root traits of forest trees.

Photo 2: (above). Pinus sylvestris seedling. Belowground biomass can often be much larger than aboveground biomass.

SUCCESS It's not always what you see

Previous publications have stated that around 20% of the tree biomass is accounted for in roots, however this depends on many different factors in the environment. Little is known regarding how easily measured aboveground traits in trees, such as leaf morphology, leaf nutrients, and height, actually correlate to the underground traits of the roots, and how these relationships affect carbon sequestration. It will be very valuable to find out which traits are related to each other, and which can be completely independent of each other. It is obvious that many of the root traits will be influenced by the soil conditions they grow in. Abiotic factors such as water and nutrient availability, temperature, and soil properties will influence both root traits and above ground traits.

The microbial community and mycorrhizal partners are key biotic interactions in forest soils that can determine how well trees grow. As trees can have a very strong influence on the soil community that develops, Clydecia is very interested in how soil communities conditioned by the previous stand rotation influences the next generation of seedlings.

Photo 3: (below)

Birch grown with soil conditioned by various tree species.

In crop farming, it is very common and useful to follow a certain crop rotation to prevent pests and soil nutrient depletion, but would a rotation of tree species in a forest be equally useful, or are trees so well suited for specific site conditions, that these soil community effects do not matter? Do these microbial interactions have any consequences for mixed forests? These are questions Clydecia is addressing in her research.

Currently, Clydecia is gathering and analysing data for all relevant native and exotic trees species in Sweden, using a series of experimental field trials established in the 1980's. It is already somewhat established that the thickness of tree roots corresponds to the amount of associated mycorrhizal fungi. However, there are a range of other root traits that have not yet been considered, such as a trees ability to produce exudates, or their growth form, such as their surface area per unit root length (referred to as the specific root length). Clydecia will reveal whether these root trait relationships across many tree species can be easily predicted by analysing simple leaf traits, such as the ratio foliar carbon to nitrogen, or the ratio of leaf surface area to mass. She will further evaluate whether trees exhibit coordination of traits across the whole tree, spanning from leaves to roots to soil microbes, which may help predict how different tree species control soil carbon accumulation and sequestration rates.

During her first year as a post doc, Clydecia has compiled a huge quantity of data on foliar and root traits for 12 different native and exotic tree species in Sweden. She has also performed a large greenhouse experiment to test how tree species impacts on soils influence the next generation of seedlings. Her research will make a significant contribution to better set up forestry for oncoming challenges associated with climate change and climate change mitigation.

Text and illustration: Anke Carius, Clvdecia Spitzer, Michael Gundale Pictures: Clydecia Spitzer

Photo 4: Sampling Abies sibirica needles for trait measurements

Photo 4: Plant-soil feedback experiment with four tree species

REPORTS

C4F- Crops for the Future

C4F program has been functioning as a research platform since its start, supporting a large number of projects, connected to other large projects. During 2022, about half of the projects were finished and new projects have been initiated, in which the involvement of young researchers (PhD students, postdoc and project leaders) has been further highlighted among the other important criteria. The overall progress of the research within C4F has been in principle as planned in 2022.

As a research platform, C4F has been supporting a number of research projects, which have connected to other large projects funded by other funding agencies. During phase 3 of the program, the update of projects have become more frequent, which facilitates implementation of timely projects dealing with current research of high interest for sustainable agriculture production.

During 2022, five projects were finished, of which one project has been prolonged for finishing PhD education. Five new projects have been initiated. All projects have involved young researchers (PhD student or postdoc/researcher) and two new project leaders with the docent competence have joined C4F. The overall progress of research within C4F has gone according to plan in 2022. A number of peer-reviewed articles of high quality have been published, while a number of other manuscripts have been submitted or in the pipeline for publication. Additionally, a number of new complementary research projects have been received from Nova Nordic Foundation, FORMAS, VR, etc. partially due to support from the C4F program. The annual C4F workshop was held on 29 Nov. in Malmö.

The plant model protein systems and the Cd stress projects progressed very well. New results on the protein rich systems have been generated and are in the pipeline to be complemented, finalized and submitted for publishing. We are soon about to finalize the Cd stress study which used advanced imaging to map plant stress, and a manuscript on the results will be submitted in coming weeks. The plant protein fractionation project has progressed very well. We finalized and submitted the paper on greenhouse gas emissions from biobased diapers during the spring 2022 and it was published in the very beginning of 2023. A range of new projects, based on activities related to this project or other work by the post-doc also started during 2022 (GreenLeaFood funded by Formas, Green2Feed funded by energimyndigheten, BSRC funded by energimyndigheten, Sensory project funded by Formas, HMMA project funded by Vinnova, a second C4F project, Mistra Food Future). As a result, the post-doc will leave this project and work in other projects, and we are currently recruiting a new post-doc for the present project.

The green diapers superabsorbents project was started in 2022 and it has progressed well since the start. A new post-doc (Anna-Lovisa Nynäs) was recruited during the spring 2022. She has focused on the fractionation procedure of the proteins from green biomass in order to get a higher protein yield that can then be used for absorbent materials. Therefore, protein fractionation has been carried out in the Plant Protein Factory, pulp has been collected and various fractionation methods are being evaluated in the lab to understand opportunities to increase protein extraction from the pulp. In parallel, a manuscript has been written, evaluating the path of the nitrogen in the green biomass and in what form it ends up in the various fractions. For the sugar beet project, the lab work of the PhD-student has been finalized during 2022. Thus a study on understanding the physiological background of the use of biostimulants, a study on the effects of biostimulants combined with drought stress, and a study on effects of the use of biostimulants in field cultivation of sugar beet was carried out and finished. The student has started to compile the results into five manuscripts of which the first one was published in 2022. The student will defend his thesis in 2023.

For the legume project, papers on mixed faba bean gels, 3D-printing of faba bean-based materials and foaming properties of protein nanofibrils from mung bean has been published. A new PhD Student will start to work on this project in early 2023.

Perspectives of the genome analysis and evolution of oil accumulation in Cyperus tubers is in preparation for a manuscript. A manuscript is in preparation for genome edited potato with redirected carbon flow from modifying promoters of transcription factors. The sink involvement of plastid starch synthase has been investigated in potato, a previously unknown gene duplication was characterized and the involvement of the enzyme in starch reserve structure was for the first time determined. A manuscript is about to be submitted.

Fig. 1. (Left) Plant height and (right) root diameter of sugar beet treated with equal nitrogen content from PBB and NS, eight weeks after planting. (Figure By Okanlawon Lekan Jolayemi)

The results on wax esters in the pheromone production project was published and a novo application was granted to Kamil Demski. Integration of new precursors into oil has successfully been shown. A proof of concept for the technology was published. Work has now focused on gene mining of Lindera species (which store oil composed of medium chain fatty acids) with the perspective of gene editing in Camelina.

The efficient protoplast-based genome editing protocols for field cress and rapeseed established within the program have enabled generation of transgene-free mutants of the target crops by using CRISPR/Cas9. A large number of CRISPS-edited mutation lines of both species have been generated for improving the seedcake and oil qualities, and some of them have been evaluated and some are still under evaluation. Manuscripts about some of the results are under preparation and one manuscript with improved oil quality in field cress was accepted. The PhD student Sjur Sandgrind working on field cress within the program defended his thesis in the end of 2022.

After finishing her PhD defence in October 2021, Xue continued to work with the publication of her work in the beginning of 2022. PhD student Shishanthi has completed and published one review article and worked on revising one paper about starch branching in barley. Regarding the autophagy project, genetic engineering, molecular and cell biology experiments were performed using the model plant Arabidopsis, by our team members in Uppsala campus SLU. Further elucidation of our findings and their transferability onto crop species will be carried out in synergistic collaboration with our colleagues at the Department of Plant Breeding in Alnarp.

Research outcomes and associated outreach activities deal with new knowledge and information on novel potential uses of plant oils, proteins, starches and other compounds from side streams, which can be used as food, feed and industrial applications. The program contributed to generation of novel plant materials for further breeding or direct uses in product quality research and future potential applications, emerging of new research areas such as bio-based composites for food and non-food uses, possibility of crop improvement by regulating autophagy process, and renewable sources of plant produced insect pheromones for pest management. SLU Grogrund has continued to support novel projects in 2022 and some of them are connected to the C4F projects.

Detailed research findings and progress

Protein-based biostimulants (PBBs) are derived from the hydrolysis of protein-rich raw materials of plant and/or animal origins, usually byproducts or wastes from agro-industries. The active ingredients produced by hydrolysis have the capacity to influence physiological and metabolic processes in plants, leading to enhanced growth, nutrient and water-use efficiency, tolerance to abiotic and biotic stresses, and improved crop yield and quality. Two examples of PBBs (hydrolyzed wheat gluten and potato protein) were shown to have an effect on the early growth of three sugar beet varieties. Both PBBs had a significant stimulating effect on early sugar beet growth and development (Fig.1). The PBBs can be further developed into superabsorbent polymers (SAPs). To conclude, PBBs/SAPs developed from agro-industrial wastes have the potential for sustainably supplying water and nutrients in agricultural systems and for enhancing plant growth and development over a substantial period.

The possible uses of green biomass as a source for local production of protein suitable for human food and animal feed has received increased attention lately. Amino acids are the dominating type of nitrogen in all the fractions. Levels of nitrate and nitrite were generally low but differed between biomass sources and their fractions with the highest levels of nitrate in hemp biomass and fractions. Both RuBisCO and other types of proteins present in the green biomass sources were found in all fractions of all biomass sources, which was initially washed using the facility in the Plant Protein Factory in Alnarp (Fig. 2). Essential and non-essential amino acids correlated strongly with the total nitrogen content indicating an equal share of those to the total nitrogen in all fractions. The highest nitrogen content among fractions was found for the green protein fraction and the white protein fraction, although the lack of correlation with the protein content determined by HPLC, indicate a breakdown of proteins during the fractionation process. The pulp fraction was found suitable as feed for ruminants, green protein and green juice as feed for pigs, green protein as feed for chicken and white protein as food for humans as related to their amino acid composition. However, some biomass sources e.g. hemp, showed high levels of nitrate that are not suitabe to be useful as feed and food. To conclude, the majority of the biomass sources evaluated here, has the potential in a biorefinery concept to be used as feed and food, although, content of antinutritional components, feasibility of the process, and uses of the brown juice have to be further evaluated before final industrial applications are possible.

Fig.2 The first part of the process in the pilot facility, whe processed. (Photo by Anna-Lovisa Nynäs).

Superabsorbents contribute the highest CO₂ emission share in a biobased diaper. Recycling of reagents for production of biobased superabsorbents reduces CO₂ emission by 50%. Only a few biobased diapers resulted in lower CO₂ emissions than fossil-based ones. Biodegradable diapers are superior from a circular perspective. Biobased superabsorbents are currently on a low TRL and need to be developed.

Fig.2 The first part of the process in the pilot facility, where the material is fed into a washer before being further

Faba bean based edible inks for food 3D printing have been successfully produced. The effect of starch/protein and fiber/protein ratio on texture and microstructure of mixed faba bean gels has been investigated. At high starch concentrations, replacing starch with protein resulted in lower viscosity and weaker gels. However, at high protein concentrations, replacing protein with starch/fiber can create inhomogeneities in the protein network resulting in weaker gels (Fig. 3).

Fig. 3. Visual overview of studies performed on faba bean mixed gel systems. Starting from the left with the extraction of fractions from faba beans before combining with water in different ratios and heating to create gels that are characterized by microscopy and large and small deformations. (Figure by Mathias Johansson)

Improved protocol of how to produce protein nanofibrils (PNFs) from mung bean, how they are affected by food-related pH and how they can improve foam stability when compared to non-fibrillated protein from the same source was published. The result showed that when adding salt during protein extraction a more pure protein isolate could be produced that formed longer PNFs than observed before when using a protein isolate extracted with only isoelectric precipitation. The curved morphology of the PNFs was affected by increased pH, the morphology remained at pH 7 but the PNFs were a bit shorter. Curved PNFs from mung bean form more stable foams that have non-fibrillated protein even at low concentrations as 1 mg/mL.

The major progress in the plant model protein systems studied towards industrial enduses included testing of the innovative processing of two protein rich systems and comparing preprocessed materials versus control ones. We observed clear differences between the variants differing in genetical make-up which showed different functional properties of the materials. New results obtained in the area of yet unexplored diverse genetic background having legumes, which might be very helpful in designing new protein-rich foods (Fig. 4). We also obtained new results on industrial wheat that was exposed to drought and Cd stresses in controlled conditions and we were able for the first time in nondestructive manner monitor the stress impact on plant roots.

cessed (left) and not processed (right). (Photos by Faraz Muneer).

This is a promising approach to better understand the climate and Cd stress impact on wheat development and finding strategies to deal with the current climate change and Cd accumulation problems.

The work on CRISPR/Cas9-edited starch in potato has resulted in one manuscript that was recently accepted for publication. An important finding was that the enthalpy of gelatinisation and retrogradation was favored by amylopectin branching density. PhD student Shishanthi is compiling data for another paper about potato starch with simultaneous mutations in SBE and GBSS genes. We are also planning the last part of her thesis work that will focus on material properties that will be carried out in collaboration with a group at KTH. Shishanthi has completed and published a review article about starchbased blends and composites for bioplastics applications to obtain a good basis for the design of the material characterizations.

Fig. 4. Foaming capacity of different legume samples after the innovative processing; a) faba bean, b) pea, pre-pro-

In carbon allocation, two articles were published. One article deals with proteomics in relation to oil in Cyperus that showed yellow nutsedge tubers to group with oilseeds and another is about oil induction in wheat endosperm with seed X-ray imaging. A manuscript on WRINKLED1 overexpression in rice was submitted. Five additional manuscripts are in various stages of preparation, on evolution of oil accumulation in Cyperus and on heterologous characterization of transcription factor interactions promoting oil accumulation.

In pheromones three manuscripts were published. One manuscript validated the whole production process of pheromone precursors (Fig. 5) from field trial as oil constituents including their processing and evidenced application in pest control.

Fig. 5. Rapid assembly of pheromone biosynthetic pathway in Nicotiana benthamiana for the production and release of moth sex pheromone components. The Cauliflower mosaic virus 35S promoter (p35S) and Octopine Synthase gene terminator (tOCS) have been used to regulate gene expression in plants. ATF1 has also been controlled by trichome specific promoter pCYP71D16. (a) Step-wise metabolic engineering strategy for leaf-based pheromone production of (Z)-11-hexadecenol, (Z)-11-hexadecenal and (Z)-11-hexadecenyl acetate. (Figure by Yihan Xia)

Efficient protoplast regeneration protocols developed by us for genome editing by CRISPR/ Cas9 for field cress and rapeseed are routinely used in our lab for generating transgene-free edited lines on the target genes. We have now generated more edited lines of rapeseed in order to have more of the 12 transporter (GTR) genes mutated simultaneously in the edited lines for effectively reducing the seed glucosinolate (GSL) level and have further evaluated some of the edited GTR lines in both species. Due to the difference in ploidy level, it is much easier to get homozygous lines for field cress than rapeseed. Screening for homozygous lines of rapeseed with editions in different target genes is still ongoing (Fig. 6).

The results on field cress showed that knocking out of GTR1 or GTR2 gene resulted in significantly reduced GSL contents in the seed of the mutants and preliminary results showed a stunted phenotype with a reduced seed yield. A number of mutation lines targeting on the genes regulating the phytic acid content in rapeseed have been obtained which are grown in biotron for obtaining homozygous lines. Knocking out of important genes affecting oil fatty acid profile in field cress resulted in significant increase in oleic acid level, but significantly reduced erucic acid level and the manuscript on this work was accepted for publishing.

Fig. 6. CRISPR-edited mutation lines of rapeseed grown in biotron (Photo by Li-Hua Zhu)

Timothy material was collected from a large-scale field trial (Fig. 7), started 2020, with 266 varieties to assess variation in forage quality parameters within and across seasons and geographic locations (Svalöv, Lövsta and Rödbäcksdalen). The experiments have been harvested three times (early, mid and late summer) and all harvested material have been collected, dried and shipped to Lantmännen Lantbruk AB. The samples have been ground and prepared for analyses by Lantmännen. At the moment, the first batch of samples are subjected near-infrared reflectance spectroscopy (NIRS) analysis by Lantmännen.

The results will tentatively be available later on regarding forage guality parameters - crude

protein and fat content, total fibre content, cellulose and lignin content, sugar content, digestibility of organic matter, metabolic energy and net energy for lactation.

In the autophagy project,

we made a massive progress in revealing 1) time-resolved organ-specific dynamics of plant autophagy under three different types of stress (autophagy-inducing drug treatment, mimicking amino acid starvation; depletion of nitrogen or carbon).

2) We demonstrated that indeed plant autophagy is fine tuned to physiology of individual organs.

3) We demonstrated that autophagic activity previously considered as non-selective (i.e. "bulk") has selectivity that allows protection of essential cellular components even under severe stressinducing conditions. This finding will be crucial to further elucidate previously hidden functions of a number of cellular components.

4) We have established a state of the art method that allowed us to isolate intact plant vacuoles together with trapped cargo delivered there by autophagy for degradation. We secured funding at the EPIC-XS facility (European facility for proteomics) to perform quantitative LC-MS analysis of the proteins contained within the purified autophagic cargo. Our samples are currently being processed at the facility in Ghent, Belgium.

5) We have discovered one of the core regulatory steps being significantly different in plant autophagy when compared to animal and fungal autophagy.

Fig. 7. Field trial of timothy (Photo by Pär Ingvarsson)

This finding opens a new perspective on how plant autophagy might have co-evolved with plant endomembrane trafficking system.

6) We have established transgenic material and initial protocols required for further elucidation of how plant autophagy regulates population of organelles controlling response to oxidative stress, peroxisomes (Fig. 8). This project is run by Florentine Ballhaus, who was hired as a PhD student in our group on the April 1st, 2022.

7) We have also initiated the part of the project related to the potential role of autophagy in microspore embryogenesis. In our previous study we identified new small organic molecules specifically regulating plant autophagy and now have established a plan on testing those compounds for their capacity to enhance efficiency of microspore embryogenesis in calcitrant cultivars of *Brassica napus*. This work is planned in collaboration with our colleagues at the Department of Plant Breeding in Alnarp and from other European institutions.

Fig. 8. Pexophagy, selective degradation of peroxisomes by autophagy. Confocal microscopy image of epidermal root cells expressing marker for autophagic structures (magenta) and marker for peroxisomes (green). Arabidopsis plants were subjected to salt stress that triggered proliferation of peroxisomes, organelles helping plants to cope with various types of abiotic stresses, including high salt. After the stress trigger was removed, plant cells decreased the population of peroxisomes by degrading the surplus of these organelles via selective autophagy, i.e pexophagy. White arrows point to autophagic structures engulfing overabundant peroxisomes to deliver them for degradation. Understanding how plant cells maintain the population of peroxisomes in various cell types will be an essential step in developing plants more tolerant to high soil salinity, elevated air temperatures and increased sun light.

In what way the research has contributed to social benefit

Within C4F program, some projects are closely connected to or have been transferred to UDIs or EIPs, one way to transfer TC4F knowledge into product-based projects, thus benefiting to the society. Whereas some other projects have potential significant social benefits, thus benefiting the society in long-run.

Opportunities to produce proteins for food, feed and material locally from green biomass will have a direct and strong social impact.

The use of biostimulants is seen as a novel and creative way to promote plant growth and replace part of the chemical input to agriculture. If successful biostimulants are produced and used in agriculture, this will contribute to novel job opportunities and also reduce the risks with using chemical inputs.

The research produced for the innovatively processed protein systems is contributing to society in different new ways to improve legume use in new protein-rich food applications. A popular science report about the research on Cd stress impact on the Swedish wheat has been available on-line for public.

The starch research aims to improve product quality in food as well as non-food applications. Our research on retrogradation can in the long run reduce food waste by prolonging product shelf life.

Good knowledge about the relationships between genetics, structure and properties is also vital for designing functional and sustainable materials that may be used in future food packages.

Insect pheromones for pest management is non-toxic and produced from a renewable resource contrasting to pesticides. Camelina derived pheromone blends have been proven to work in the field. Faba beans, oat, peas, rapeseed, field cress, potato are Swedish crops with a good nutritional profile, which can be a good plant based protein alternative to soy-products and gluten. Legumes and field cress can help with nitrogen fixation or prevent from nutrient leaching when incorporated into an intercropping system, which will thus enrich the soil and reduce over fertilization.

Novel CRISPR-edited lines of oilseed crops with improved oil and protein or seedcake qualities would in long-run contribute to increased plant oil production and improved seedcake quality as a source of high value protein for food, feed and industrial uses, and consequently reducing the fossil use and benefiting the human health and environment.

Diapers has a high carbon foot-print and if more sustainable alternatives can be developed, this will have a huge impact on the society.

How does C4F take basic research to application

The majority of the C4F projects are more orientated in applied research, in which we make our great efforts on transferring the knowledge obtained from basic research in oil, protein and starch as well as material science into potential food, feed and industrial applications in one way or another. Some examples are as followings:

1. The work on biostimulant uses on sugarbeet span from basic research on mechanisms in plants to application in novel biostimulants being used in agriculture.

2. Understanding the chemical background of superabsorbent capacity of functionalized proteins require basic research while the development of diapers for children or old people takes the project out to the reality.

3. We try to understand underlying mechanisms that determines protein extractability in green leaf, which is basic science. The results will though be used for production of local plant protein for food and feed which is a direct application of the research.

4. Wheat gluten quality for breeding climate stable wheat

5. Cd stress in wheat-management strategies

6. By understanding the structure and properties of the different fractions; protein , starch and fibre new foods can be developed and optimised. Mixtures will also be valuable to show how pure fractions is needed. Previous knowledge on the structure and behaviour of the faba bean fractions was used in the bio-ink development in the 3D-printing project.

7. Starch is a useful part in a composite material where it can provide oxygen barrier functionality and something else gives strength and water resistance. Starch with increased amylose content has nutritional benefits since it has more slow carbohydrates that potentially can decrease our insulin response and thereby reduce the risk to develop type II diabetes.

8. Pheromones from a plant production source for pest management is a good example of going from basic science on specific pheromone compounds and their genetic background in e.g. moths to applications in production and pest management. In 2022 the whole chain from biosynthesis to production via extraction and final pest interruption was shown under field conditions and published in Nature Sustainability. 9. Identified target genes in model species or the same species from basic research have been used in improving target traits in oil crops by CRISPR/Cas9.

10. CRISPR/Cas9 edited plant material was studied for bio-material applications

Vice program leader Li-Hua Zhu

C4F RESEARCHER PORTRAIT Faraz Muneer A protein researcher contributing to the Swedish food transition

Already as a master student Faraz Muneer got interested in how left over materials from plants can be used instead of simply ending up as waste and also industrial by-products of plant processing industries. Today he is doing research on the properties of locally produced proteins sources to produce highly nutritious food products.

In his research Faraz explores the plant materials that are wasted in agricultural systems, for example, when broccoli is harvested, around 70 percent of the plant is normally not used. He and his colleagues take these materials and extract

proteins from it. They also get fibers and so-called brown juice as side products in the process. This brown juice can be turned into food, feed, bioplastics, biogas and fertilizers, Faraz Muneer explains. While different proteins can be used for different things, it is also possible to modify the properties of proteins, either by processing them after the harvest or by doing plant breeding to make the plants produce the desired proteins. The

focus of Faraz' team is to improve the processing of proteins, to get more proteins out, improve the

functionality of the proteins and the efficiency of the whole process. They can for example add mechanical energy to break the proteins apart and make them more useable. This research is important since the more proteins we can get

from our own fields in Sweden, the less we must import from countries far away.

More than 50 percent of the food consumed here is imported. The war in Ukraine and COVID-19 have affected the food supply chain, the food prices are increasing, and we need a longterm strategy for our food production. This has triggered Faraz to look at new alternatives. How can the local production of food in Sweden be

increased, and how can research contribute to that.

Developing new plant-based materials instead of producing synthetic materials is also a step towards a more sustainable production. For example, by developing super absorbing materials from plants the synthetic materials that are used for example in diapers can be replaced. In recent years Faraz Muneer has experienced a personal development as a researcher, going from mostly being interested in the basics of protein research to become more interested in the bigger picture. Looking more broadly at science, questions that interest him now are:

"How can we improve humanity and the problems we are facing now? What can be solved by research? How can we integrate different factors when we are doing research? "

Facts

Faraz Muneer came from Pakistan to Sweden in 2009 to study at SLU. As a PhD student here he used plant proteins to make bioplastics. As a researcher he explores how proteins from different plants can be used as food and animal feed. He also study how plants can be used to produce absorbent materials for the medical sector or hygiene products. He finds out how the proteins function and how their structure impact the properties of food or materials. He is into sports and spend a lot of his spare time playing cricket in a club in Malmö.

Author: Lisa Beste

Image Credits: Faraz Muneer and artverau via pixabay

TC4F publications and activities 2022

The five institutions involved in TC4F have published 49 articles in T4F and 18 in C4F in peer-reviewed scientific journals. Read here how many have been involved with supervision of students, teaching, received other grants and contri buted to popular scientific activities. For T4F, the information is now presented via institution instead of per theme as the themes according to the program plan involve several institutions.

Authors marked in **bold** represent researchers that have been financed by, or are associated to, TC4F.

T4F - Department of Forest Ecology and Management

Scientific publications

During 2022 Department of Forest Ecology and Management has published 18 peer reviewed scientific articles in international journals. Authors marked in bold represents researchers that have been financed by, or are associated to, the research program.

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17. Scharn R, Negri IS, **Sundqvist MK**, Løkken JO, Bacon CD, Antonelli A, et al. Limited decadal growth of mountain birch saplings has minor impact on surrounding tundra vegetation. Ecology and Evolution. 2022;12(6):e9028.

18. Senior JK, **Gundale MJ**, Iason GR, Whitham TG, Axelsson EP. Progeny selection for enhanced forest growth alters soil communities and processes. Ecosphere. 2022;13(2):e3943.

19. **Stangl ZR**, Tarvainen L, Wallin G, Marshall JD. Limits to photosynthesis: seasonal shifts in supply and demand for CO2 in Scots pine. New Phytologist. 2022;233(3):1108-20.

Other funding that has been received partially or fully due to the TC4F research

T4F support led to a successful FORMAS proposal to S. Jämtgård, titled "Kan priming-effekten i marken öka växters tillväxt under ökad CO2 genom att lindra näringsbegränsningar?"

T4F support led to a successful FORMAS proposal to M.J. Gundale titled, "Evaluating trade-offs between productivity, climate impacts, and biodiversity in Swedish forests.

Education

a) PhD theses, MSc theses, Bachelor theses:

Theresa Ibanez, Wildfire in Swedish boreal forests, January 4, 2022 (Supervisors M-C Nilsson and M.J. Gundale).

b) Supervision and teaching (include supervision of finished and on-going students, include teaching and organization of courses):

Vincent Buness, PhD Student, Starting 2022 (Supervisors M.J. Gundale and M.K. Sundqvist).

Gaya ten Kate, Masters Student, Starting 2022 (60 ECT) (Supervisors M.K. Sundqvist and Z. R. Stangl.)

Forest Ecology (Undergraduate Course #SG0220), Course Organizer, M.K. Sundqvist. Teachers, M.K. Sundqvist, M.J. Gundale, Marie-Charlotte Nilsson and Z. R. Stangl.

Forest Ecosystem Ecology (MSc. Course #). Teachers, M.J. Gundale, M-C. Nilsson.

Forest Management (MSc. Course #). Teachers, M.J. Gundale

Skoglig ekologi och botanik (Undergraduate Course #, BI1382), Teacher: M.K. Sundqvist

Skogsekosystemets kem<mark>iska</mark> grunder (Undergraduate Course, SG0210. Teacher S Jämtgård).

Tyra Tornberg, Masters student, Autumn 2022 (30 ECTS) (Supervisor Nils Henriksson)

Marcus Björs, Bachelor student Spring 2022 (15 ECTS)

(Supervisor Nils Henriksson)

Personnel		
Nam	Gender	Part of full time
	& Position	financed by TC4F
Michael Gundale	M, Professor	0%
Sandra Jämtgå <mark>rd</mark>	F, Ass.Professor	0%
Zsofia Stangl	F, Researcher	25%
Maja Sundqvist	F, Researcher	25%
Clydecia Spitzer	F, Post Doc	<mark>100</mark> %
Morgan Karlsson	M, Technician	40%
Ilse van Duuren	F, Technician	40%
Torgny Näsholm	M, Professor	0%
Marie-Charlotte Nilsson	F, Professor	0%
Nils Henriksson	M, Researcher	0%
Lina Nilsson	F, Technician	50%
Andreas Schneider	M, Post Doc	100%
Shun Haseqawa	M, Post Doc	8%

T4F - Department of Plant Physiology, Umeå Universitet and Skogforsk

Scientific publications

During 2022 Department of Plant Physiology has published 13 peer reviewed scientific articles in international journals. Authors marked in bold represents researchers that have been financed by, or are associated to, the research program.

Borthakur D, Busov V, Cao XH, Du Q, Gailing O, Isik F, et al. Current status and trends in forest genomics. Forestry Research. 2022;2(1).

Heuchel A, Hall D, Zhao W, Gao J, Wennström U, 2. Wang X-R. Genetic diversity and background pollen contamination in Norway spruce and Scots pine seed orchard crops. Forestry Research. 2022;2(1).

Guo J-F, Wang B, Liu Z-L, Mao J-F, Wang X-R, Zhao 3 W. Low genetic diversity and population connectivity fuel vulnerability to climate change for the Tertiary relict pine Pinus bungeana. Journal of Systematics and Evolution. 2022.

Xu J, Luo H, Zhou S-S, Jiao S-O, Jia K-H, Nie S, et al. 4. UV-B and UV-C radiation trigger both common and distinctive signal perceptions and transmissions in Pinus tabuliformis Carr. Tree Physiology. 2022;42(8):1587-600.

Calleja-Rodríguez A, Klápště J, Dungey H, Graham 5. N, Ismael A, García-Gil MR, et al. Genomic Selection in Scots (Pinus Sylvestris) and Radiata (Pinus Radiata) Pines. In: De La Torre AR, editor. The Pine Genomes. Cham: Springer International Publishing; 2022. p. 233-50.

Akhter S, Westrin KJ, Zivi N, Nordal V, Kretzschmar 6. WW, Delhomme N, et al. Cone-setting in spruce is regulated by conserved elements of the age-dependent flowering pathway. New Phytologist. 2022;236(5):1951-63.

Curci PL, Zhang J, Mähler N, Sevfferth C, Mannap-7. peruma C, Diels T, et al. Identification of growth regulators using cross-species network analysis in plants. Plant physiology. 2022;190(4):2350-65.

Liu S, Zhang L, Sang Y, Lai Q, Zhang X, Jia C, et al. Demographic History and Natural Selection Shape Patterns of Deleterious Mutation Load and Barriers to Introgression across Populus Genome. Molecular Biology and Evolution. 2022;39(2).

9 Ranjan A, Perrone I, Alallag S, Singh R, Rigal A, Brunoni F, et al. Molecular basis of differential adventitious rooting competence in poplar genotypes. Journal of Experimental Botany. 2022;73(12):4046-64.

Vergara A, Haas JC, Aro T, Stachula P, Street 10. NR, Hurry V. Norway spruce deploys tissue-specific responses during acclimation to cold. Plant Cell Environ. 2022;45(2):427-45.

Arshad R, Saccon F, **Bag P**, Biswas A, Calvaruso C, 11. Bhatti AF, et al. A kaleidoscope of photosynthetic antenna proteins and their emerging roles. Plant Physiology. 2022;189(3):1204-19.

Fataftah N, Edlund E, Lihavainen J, Bag P, Björkén 12. L, Näsholm T, et al. Nitrate fertilization may delay autumn leaf senescence, while amino acid treatments do not. Physiologia Plantarum. 2022;174(3):e13690.

Boussardon C, Bag P, Juvany M, Šimura J, Ljung K, 13. Jansson S, et al. The RPN12a proteasome subunit is essential for the multiple hormonal homeostasis controlling the progression of leaf senescence. Communications Biology. 2022;5(1):1043.

Personnel **Department of Plant Physiology Gender & Position** Name Stefan Jansson M, Professor Kathryn Robinson F, Researcher

Pushan Bag M, PhD student Sanchali Nanda F, PhD student Jenna Lihavainen F, Postdoc Nazeer Fataftah M, postdoc Nathaniel Street M, Professor

0%

Part of full time financed by TC4F	
0 %	
50 %	
0 %	
0 %	
0 %	
0 %	

Elena van Zalen	F, PhD student	100%
Camilla Canovi	F, PhD student	100 %
Sara Westmann	F, PhD student	0 %
Teitur Kalman	M, PhD student	20 %
Chanaka Mannapperuma	M, Postdoc	0 %
Aman Zare	M, Research engineer	100 %

Department of Ecology and Environmental Science

(iao-Ru Wang	F, Professor	0 %
lade Bruxaux	F, Postdoc	30 %
David Hall	M, Researcher	0 %
Wei Zhao	M, Researcher	0 %
Alisa Kravtsova	F, PhD student	0 %

Skoaforsk

-	
Mari Suontama	F, Researcher
Sara Abrahamsson	F, Researcher
Jon Ahlinder	M, Researcher
David Hall	M, Researcher
Torgny Persson	M, Researcher
Ulfstand Wennström	M, Researcher

POPULAR SCIENTIFIC PUBLICATIONS (REPORTS ETC)

Jansson S (2022) Fotosyntesen och skogsdebattens önsketänkanden. I Antologin "SKOGENS VÄRDEN – forskares reflektioner, Mittuniversitetet s 42-43. https://www.miun. se/ontentassets/5eadb642622a45c189d35963c14fcb68/ skogens-varden.pdf

Gensaxen kan bidra till EU:s gröna giv – om lagen ändras. Intervju i Tidningen Curie 11/5 2022 https://www. tidningencurie.se/nyheter/2022/05/11/gensaxen-kanbidra-till-eus-grona-giv-om-lagen-andras/

Bergkvist E, Polfjärd J, Eriksson D, Jansson S, Sundström J (2022). Beslut brådskar – EU måste tillåta gensax-grödor. Ny teknik 26/5 2022. https://www.nyteknik.se/crispr-opinion/ beslut-bradskar-eu-maste-tillata-gensax-grodor/118307

Jansson S (2022). Participation in panel debate at Symposium "European response to environmental and health crises: what role for foundations? French Academy of Sciences, Paris 8/6 2022

Vaughan H (2022) Gene-edited tomato offers new plantbased source of vitamin D. Interview in New Scientist 23/5 2022. https://www.newscientist.com/article/2321469gene-edited-tomato-offers-new-plant-based-source-ofvitamin-d/amp/

Varför slår aspens löv ut så sent? Intervju i SR förmiddag i P4 Dalarna 23/5 2022

Obminska A (2022). Så kan träd fånga in mer koldioxid. Intervju i Ny teknik 24/8 2022. https://www.nyteknik.se/ premium/sa-kan-trad-fanga-in-mer-koldioxid-7035404

Jansson S, Bag P (2022) Barrträdens skydd. Bi-lagan 2/2022 8-9. https://bioresurs.uu.se/wp-content/uploads/2022/09/ bilagan2022 2.pdf

von Bothmer R, Días Carrasco O, Fagerström T, Jansson S, Ortega-Klose, F, Ortiz R, Ángel Sánches M (2022). PARA UNA AGRICULTURA SOSTENIBLE (Book) ISBN 978-956-404-859-8. PARA UNA AGRICULTURA SOSTENIBLE - ChileBIO

Klarin G (2022) Kvickare fotosyntes ökade skörden med 33 procent. Intervju i SR Vetenskapsradion 10/10 2022. https://sverigesradio.se/artikel/kvickare-fotosyntes-okadeskorden-med-33-procent

Jansson S (2022). Kan och bör vi genförbättra grödor? Föredrag Umeå Pensionärsuniversitet 25/10 2022

Jansson S (2022). CRISPR/Cas9 i växtforskning och växtförädling. Fika efter en forskare, Umeå universitet, Väven, Umeå 29/10 2022. https://www.umu.se/forskning/ nyheter-och-arrangemang/popularvetenskapligaarrangemang/fika-efter-en-forskare/stefan-janssoncrisprcas9-i-vaxtforskning-och-vaxtforadling/

Jansson S (2022). Hur skall 2020-talets genetik komma till samhällets nytta? Lecture at symposium "Vad vi kan – och inte kan!" Fysiografiska sällskapet, Lund 23/11 2022

Ingvarsson P, Jansson S, Nilsson O, Sundström J (2022). Skogsforskare redovisar visst intressekonflikter. DN Debatt 24/11 2022. https://www.dn.se/debatt/skogsforskare-redovisar-visst-intressekonflikter/

Jansson S (2022) What are NGT and how are they regulated today? New genomic techniques and their future in the EU. Webinar Brussels 6/12 2022. https://tillsammansieuropa. eu/sv/event/new-genomic-techniques-and-their-future-in-the-eu-2022-12-06-1114/register

POPULAR SCIENTIFIC PRESENTATIONS

Abrahamsson, S. Westin, J. Kunskapartikel Skogforsk, 2022. Genomisk selektion på gran. https://www.skogforsk.se/ kunskap/kunskapsbanken/2022/genomisk-selektion-pagran/

Hall, D., Wennström, U. Kunskapartikel Skogforsk, 2022. Genetisk diversitet och bakgrundspollinering i tall- och granfröodlingar. https://www.skogforsk.se/kunskap/kunskapsbanken/2022/genetisk-diversitet-och-bakgrundspollinering-i-tall--och-granfroodlingar/.

Hall, D., Wennström, U., Kroon, J. Kunskapartikel Skogforsk, 2022. Stort genutbyte mellan tallar i norra Europa. https://www.skogforsk.se/kunskap/kunskapsbanken/2022/stort-genutbyte-mellan-tallar-i-norra-europa/.

Suontama, M. Calleja-Rodriguez, A. Persson, T. Kunskapartikel Skogforsk, 2022. De genetiska egenskaperna står sig över tid. https://www.skogforsk.se/kunskap/kunskapsbanken/2022/De_genetiska_egenskaperna_star_sig_over_tid/.

Suontama, M. 2022. Skogsträdsförädling med nya utvecklade metoder möjliggör snabbare anpassning till framtidens klimat. Skogens värden. Dnr: MIUN 2022/1393. ISBN: 978-91-89341-70-8.

Collaboration with industry and/or other parts of society

There are ongoing interactions with the forestry industry including Holman Skog AB and Stora Enso.

Investments in research infrastructure

Employed Aman Zare as a member of the UPSC bioinformatics facility.

EDUCATION

PHD THESES, MSC THESES, BACHELOR THESES SUPERVISION AND TEACHING DEPARTMENT OF PLANT PHYSIOLOGY

a) PhD theses, MSc theses, Bachelor theses PhD: Andreas Schneider. Perturbance and stimulation: Using nitrogen addition and high-throughput sequencing to study fungal communities in boreal forests. June 2022. Main supervisor Street, N.

b) Supervision and teaching (include supervision of finished and on-going students, include teaching and organization of courses)

Xiao-Ru Wang. Main supervisor for PhD-candidate Alisa Heuchel. Tentative thesis title: Seed orchard genetics and climate adaptation. Expected date for dissertation: Nov. 2023.

Xiao-Ru Wang. Main supervisor for PhD-candidate Bea Andersson. Tentative thesis title: The distribution of fitness effect of new mutations. Expected date for dissertation: Nov. 2023.

Street N. Main supervisor for PhD-candidate Camila Canova. Tentative title: The role and origin on long non-coding RNAs in conifer genomes. Expected defence date for dissertation: June 2025.

Street N. Main supervisor for PhD-candidate Elena van Zalen. Tentative title: Comparative genomics of conifers. Expected defence date for dissertation: Nov 2025.

Researchers in T4F program at Skogforsk have been involved in teaching courses and excursions at SLU and Umeå University as well as involved in Alica Heucel's and Tuuli Aro's PhD research.

T4F - Department of Forest Genetics and Plant Physiology

During 2022 the Department of Forest Genetics and Plant Physiology has published 5 peer reviewed scientific articles in international journals. Authors marked in bold represents researchers that have been financed by, or are associated to, the research program.

Scientific publications

1. Aghbolaghi MA, Dedicova B, Sharifzadeh F, Omidi M, **Egertsdotter U**. Plants. 2022;11(22):3122.

2. **Castro D**, Concha C, Jamett F, Ibáñez C, Hurry V. Soil Microbiome Influences on Seedling Establishment and Growth of Prosopis chilensis and Prosopis tamarugo from Northern Chile. Plants. 2022;11(20):2717.

3. Law SR, Serrano AR, Daguerre Y, Sundh J, Schneider AN, Stangl ZR, et al. Metatranscriptomics captures dynamic shifts in mycorrhizal coordination in boreal forests. Proceedings of the National Academy of Sciences. 2022;119(26):e2118852119.

4. Nielsen UB, Hansen CB, Hansen U, Johansen VK, **Egertsdotter U.** Accumulated effects of factors determining plant development from somatic embryos of Abies nordmanniana and Abies bornmuelleriana. Frontiers in Plant Science. 2022;13.

5. Vergara A, Haas JC, Aro T, Stachula P, Street NR, Hurry V. Norway spruce deploys tissue-specific responses during acclimation to cold. Plant Cell Environ. 2022;45(2):427-45.

Joint Efforts and Collaborations

Field experiments underway by researchers supported by T4F are involved in ongoing collaborations with HolmenSkog (David Castro), StoraEnso (Tuuli Aro) and Skogforsk (Tuuli Aro). The SE lab continues to research requirements for spruce embryo developmental processes to improve the SE developmental success rates at all steps, from initiation to plant growth in the field with wide application for the forest industry. The spruce SE transformation platform performs transformations as a service primarily, for the UPSC community, but also has international collaborations. The focus for the platform's internal activities is to develop the technology for efficient CRISPR/Cas transformation.

Personnel

Name	Gender & Position	Part of full time financed by TC4F
Vaughan Hurry	M, Professor	0%
Annika Nordin	F, Professor	0%
Ulrika Egertsdotter	F, Professor	25%
Tuuli Aro	F, PhD student	50 %
Tinkara Bizjak	F, PhD student	25%
Sofie Johansson	F, Researcher	50 %
David Castro	M, Postdoc	100%
Simon Law	M, Postdoc	15%

Eduaction

a) PhD theses, MSc theses, Bachelor theses Castro, D. (2022). Who comes first? Implications of the plant-microbiome-soil continuum feedback on plant performance. Acta Universitatis Agriculturae Sueciae. ISBN: 978-91-7760-929-2. https://res.slu.se/id/publ/116835

b) Supervision and teaching (include supervision of finished and on-going students, include teaching and organization of courses)

Hurry, V. Main supervisor for PhD-candidate Tuuli Aro. Tentative title: Genetic variation in frost tolerance in Norway spruce. Expected date for dissertation: December 2024

Nordin, A. Main supervisor for PhD-candidate Tinkara Bizjak. Tentative title: Boreal forest microbial communities in connection to nitrogen. Expected dissertation: June 2025.

Vaughan Hurry, teacher - Plant biology for future forests (SG0242 30061)

Vaughan Hurry, teacher - Trädbiologi, genetik och evolution (BI1383 HT2022)

Vaughan Hurry, teacher - Animal & Plant Physiology (UmU 5MO101)

Vaughan Hurry, teacher – Writing science (UmU, graduate course)

T4F - Southern Swedish Forest Research Centre

During 2022 the Southern Swedish Forest Research Centre has published 13 peer reviewed scientific articles in international journals. Authors marked in bold represents researchers that have been financed by, or are associated to, the research program.

Scientific publications

1. Aldea J, Ruiz-Peinado R, del Río M, Pretzsch H, Heym M, Brazaitis G, et al. Timing and duration of drought modulate tree growth response in pure and mixed stands of Scots pine and Norway spruce. Journal of Ecology. 2022;110(11):2673-83.

2. Ara M, Berglund M, Fahlvik N, Johansson U, **Nilsson** U. Pre-Commercial Thinning Increases the Profitability of Norway Spruce Monoculture and Supports Norway Spruce–Birch Mixture over Full Rotations. Forests. 2022;13(8):1156.

3. Ara M, Barbeito I, Kalén C, **Nilsson U**. Regeneration failure of Scots pine changes the species composition of young forests. Scandinavian Journal of Forest Research. 2022;37(1):14-22.

4. **Goude M, Nilsson U,** Mason E, Vico G. Comparing basal area growth models for Norway spruce and Scots pine dominated stands2022.

5. **Goude M, Nilsson U**, Mason E, Vico G. Using hybrid modelling to predict basal area and evaluate effects of climate change on growth of Norway spruce and Scots pine stands. Scandinavian Journal of Forest Research. 2022;37(1):59-73.

6. Jonsson A, Elfving B, Hjelm K, Lämås T, **Nilsson U.** Will intensity of forest regeneration measures improve volume production and economy? Scandinavian Journal of Forest Research. 2022;37(3):200-12.

7. Liziniewicz M, Barbeito I, Zvirgzdins A, Stener L-G, Niemistö P, Fahlvik N, et al. Production of genetically improved silver birch plantations in southern and central Sweden2022.

8. Marčiulynas A, **Marčiulynienė D**, Mishcherikova V, Franić I, Lynikienė J, Gedminas A, et al. High Variability of Fungal Communities Associated with the Functional Tissues and Rhizosphere Soil of Picea abies in the Southern Baltics. Forests. 2022;13(7):1103.

9. **Marčiulynienė D**, Marčiulynas A, Mishcherikova V, Lynikienė J, Gedminas A, Franic I, et al. Principal Drivers of Fungal Communities Associated with Needles, Shoots, Roots and Adjacent Soil of Pinus sylvestris. Journal of Fungi. 2022;8(10):1112.

10. Mazziotta A, Lundström J, Forsell N, Moor H, Eggers J, **Subramanian N**, et al. More future synergies and less trade-offs between forest ecosystem services with natural climate solutions instead of bioeconomy solutions. Global Change Biology. 2022;28(21):6333-48.

11. Mensah AA, **Holmström E**, Nyström K, **Nilsson U**. Modelling potential yield capacity in conifers using Swedish long-term experiments. Forest Ecology and Management. 2022;512:120162.

12. **Ogana FN**, Sjödin F, Holmström E, Fries C, **Nilsson U.** Effect of Aspect-Slope on the Growth of Conifers in a Harsh Boreal Climate of Northwest Sweden. Forests. 2022;13(2):301.

13. Persson M, Trubins R, Eriksson LO, Bergh J, Sonesson J, **Holmström E**. Precision thinning – a comparison of optimal stand-level and pixel-level thinning. Scandinavian Journal of Forest Research. 2022;37(2):99-108.

Personnel

Name

Gender & Position Part of full time

financed by TC4F

Jorge Aldea Michelle Cleary Benjamin Forsmark Martin Goude Karin Hjelm Emma Holmström Diana Marciulyniene Magnus Mossberg Urban Nilsson Friday Ogana Narayanan Subramanian Andis Zvirgzdins Postdoc Researcher, Docent Postdoc Researcher Researcher, Docent Reseracher, Docent Postdoc Programmer Professor Postdoc Researcher PhD-student

C4F - Crops for the Future

Scientific publications

1. Andreasson E, Kieu N, Zahid MA, Carlsen F, Lenman M, **Sandgrind S**, et al. Invited Mini-Review Research Topic: Utilization of Protoplasts to Facilitate Gene Editing in Plants: Schemes for In Vitro Shoot Regeneration From Tissues and Protoplasts of Potato and Rapeseed: Implications of Bioengineering Such as Gene Editing of Broad-Leaved Plants. Frontiers in Genome Editing. 2022;4:780004.

2. Bettelli MA, Capezza AJ, Nilsson F, **Johansson E**, Olsson RT, Hedenqvist MS. Sustainable Wheat Protein Biofoams: Dry Upscalable Extrusion at Low Temperature. Biomacromolecules. 2022;23(12):5116-26.

3. **Demski K**, Ding B-J, Wang H-L, Tran TNT, Durrett TP, **Lager I**, **et al.** Manufacturing specialized wax esters in plants. Metabolic Engineering. 2022;72:391-402.

4. Fei M, Jin Y, Hu J, Dotsenko G, Ruan Y, Liu C, **et al.** Achieving of high-diet-fiber barley via managing fructan hydrolysis. Scientific Reports. 2022;12(1):19151.

5. **Jayarathna S, Andersson M,** Andersson R. Recent Advances in Starch-Based Blends and Composites for Bioplastics Applications. Polymers [Internet]. 2022; 14(21).

6. **Johansson M**, Johansson D, Ström A, Rydén J, **Nilsson K**, Karlsson J, et al. Effect of starch and fibre on faba bean protein gel characteristics. Food Hydrocolloids. 2022;131:107741.

7. **Johansson M, Nilsson K**, Knab F, **Langton M**. Faba Bean Fractions for 3D Printing of Protein-, Starch- and Fibre-Rich Foods. Processes [Internet]. 2022; 10(3).

8. **Jolayemi OL**, Malik AH, Ekblad T, Fredlund K, **Olsson ME, Johansson E**. Protein-Based Biostimulants to Enhance Plant Growth—State-of-the-Art and Future Direction with Sugar Beet as an Example. Agronomy. 2022;12(12):3211. 9. **Lama S**, Vallenback P, Hall SA, Kuzmenkova M, **Kuktaite R**. Prolonged heat and drought versus cool climate on the Swedish spring wheat breeding lines: Impact on the gluten protein quality and grain microstructure. Food and Energy Security. 2022;11(2):e376.

10. **Lan Y**, Chawade A, **Kuktaite R**, Johansson E. Climate Change Impact on Wheat Performance—Effects on Vigour, Plant Traits and Yield from Early and Late Drought Stress in Diverse Lines. International Journal of Molecular Sciences. 2022;23(6):3333.

11. **Markgren J**, Rasheed F, Hedenqvist MS, Skepö M, **Johansson E**. Clustering and cross-linking of the wheat storage protein α -gliadin: A combined experimental and theoretical approach. International Journal of Biological Macromolecules. 2022;211:592-615.

12. **Muneer F**, Hedenqvist MS, Hall S, **Kuktaite R**. Innovative Green Way to Design Biobased Electrospun Fibers from Wheat Gluten and These Fibers' Potential as Absorbents of Biofluids. ACS Environmental Au. 2022;2(3):232-41.

13. Niemeyer PW, Irisarri I, Scholz P, Schmitt K, Valerius O, Braus GH, **et al**. A seed-like proteome in oil-rich tubers. The Plant Journal. 2022;112(2):518-34.

14. **Nilsson K**, Sandström C, Özeren HD, Vilaplana F, Hedenqvist M, **Langton M**. Physiochemical and thermal characterisation of faba bean starch. Journal of Food Measurement and Characterization. 2022;16(6):4470-85.

15. **Snell P**, Wilkinson M, Taylor GJ, Hall S, **Sharma S**, Sirijovski N, **et al**. Characterisation of Grains and Flour Fractions from Field Grown Transgenic Oil-Accumulating Wheat Expressing Oat WRI1. Plants. 2022;11(7):889.

16. Statkevičjūtė G, Liatukas Ž, Cesevičienė J, Jaškūnė K, Armonienė R, **Kuktaite R**, et al. Impact of Combined Drought and Heat Stress and Nitrogen on Winter Wheat Productivity and End-Use Quality. Agronomy. 2022;12(6):1452. 17. Wang H-L, Ding B-J, Dai J-Q, Nazarenus TJ, Borges R, Mafra-Neto A, **et al.** Insect pest management with sex pheromone precursors from engineered oilseed plants. Nature Sustainability. 2022;5(11):981-90.

18. Xia Y-H, Ding B-J, Dong S-L, Wang H-L, **Hofvander P**, Löfstedt C. Release of moth pheromone compounds from Nicotiana benthamiana upon transient expression of heterologous biosynthetic genes. BMC Biology. 2022;20(1):80.

Popular scientific publications (reports etc)

https://www.vinnova.se/globalassets/mikrosajter/storskalig-forskningsinfrastruktur/dokument/slutrapporter-ensidingar/2020-00830_slu-phd-lan-yuzhou-.docx.pdf

Interviews and presence in media

- Interview about Climate smart wheat in Livsmedel i focus (link is under request of the journalist).

- De tar genvägen via växter för att lura insekter (2022) https://sverigesradio.se/artikel/de-tar-genvagen-via-vaxter-for-att-lura-insekter

Scientific presentations

- Muneer F. 2022. Applications of plant proteins in materials and food. Oral presentation, C4F workshop, Malmö, Sweden, November 29.

- Johansson, M., Johansson, D., Ström, A., Rydén, J., Nilsson, K., Karlsson, J., ... & Langton, M. 2022, Effect of starch and fibre on faba bean protein gel characteristics. Poster presentation at the conference Healthy, Safe, and Sustainable Foods of the Future, DTU, Copenhagen, Denmark, October 13th.

- Johansson, M., Johansson, D., Ström, A., Rydén, J., Nilsson, K., Karlsson, J., ... & Langton, M. 2022, Effect of starch and fibre on faba bean protein gel characteristics. Poster at the conference Data Driven Food Research & Innovation (Food Science Sweden), online, March 15th. - Johansson, M., Nilsson, K., Knab, F., & Langton, M. 2022. Faba Bean Fractions for 3D Printing of Protein-, Starch- and Fibre-Rich Foods. Presentation at the SLU lunch seminar series "Värt att veta", online February 3rd, 2022. Video recording available at: https://play.slu.se/media/ Worth+KnowingA+Faba+bean+fractions+for+3D-printing+of+protein-+and+fibre-rich+foods/0_3721h6v1

- Holla S., Bozhkov P.V., Schumacher K, Minina E.A. 2022. "Plant-specific role of cysteine protease atg4 in autophagy". A talk at the 5th Plant Proteases Conference in Ljubljana, Slovenia September.

- Ballhaus F., Hicks G.H., Sabljić I., Bozhkov P.V., Dauphinee A.N. 2022 . "Unravelling plant autophagy mechanisms using chemical genetics". A speed talk and poster presentation at the Conference of the Scandinavian Plant Physiology Society in Longyearbyen, Svalbard in August/ September.

- Sandgrind S., Li X., Ivarson E., Wang E-S., Guan R., Kanagarajan S., and Zhu L.-H. 2022. CRISPR/Cas9 gene editing of the novel oil and cover crop, Lepidium camestre, for improvement of oil quality. Oral presentation. 25th International Symposium on Plant Lipids. July 10th-15th, 2022, Grenoble, France.

- Moss O., Li X., Kanagarajan S., Guan R., Ivarson E. and Zhu L-H. 2022. Genome editing of rapeseed by CRISPR/ Cas9 for reducing phytic acid content. Poster presentation. 25th International Symposium on Plant Lipids. July 10th-15th, 2022, Grenoble, France.

- Sandgrind S., Li X., Ivarson E., Wang E-S., Guan R., Kanagarajan S., and Zhu L.-H. 2022. Domestication of the potential novel oilseed crop Lepidium campestre for the Nordic climate by gene editing. Oral presentation. The SPPS conference 2022. 30th of August - 2nd of September. Longyearbyen, Svalbard, Norway. - Moss O., Li X., Kanagarajan S., Guan R., Ivarson E. and Zhu L-H. 2022. Genome Editing of Rapeseed by CRISPR/ Cas9 for Reducing Phytic Acid Content. Poster presentation. The SPPS conference 2022. 30th of August - 2nd of September. Longyearbyen, Svalbard, Norway.

- Moss O. 2022. Genome editing of rapeseed for reducing antinutritional compounds in the seed. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Sharma S. 2022. Carbon channelling of storage compounds in sink tissues. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Kuktaite R. 2022. Climate impact on the gluten polymers evaluated by a proteomic Approach. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Jolayemi O.L. 2022. Effect of equal N from PBB and chemical source on growth and physiology of sugar beet. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Jayarathna S. 2022. New starch for novel applications. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Lan Y. 2022. The impact of Cd and drought on wheat and study root architecture using neutron computed tomography. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Demskil K. 2022. Replacing Pesticides by Developing an Oilseed Platform for Insect Pheromone Production. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Klara Nilsson and Mathias Johansson have presented their work on 3D printing at the lunch seminar series "Värt att veta" at SLU https://play.slu.se/media/Worth+Knowing-A+Faba+bean+fractions+for+3D-printing+of+protein-+and+fibre-rich+foods/0_3721h6v1 - Jolayemi O.L. 2022 Comparative effect of equal N content from PBB and NS on agronomy, physiology and metabolism of sugar beet. Presentation at C4F annual workshop 29 November.

- Sandgrind S. 2022. Genome editing of Lepidium campestre using CRISPR/Cas9. Oral Presentation at C4F annual workshop 29 November

- Mathias Johansson. 2022. Legume based gels – texture and microstructure. Oral Presentation at C4F annual workshop 29 November

- Anna-Lovisa Nynäs. 2022. Presentation of the pilot facility at the PlantLink day in Alnarp, 5th of October.

- Anna-Lovisa Nynäs. 2022. Proteins from green biomass – Food, feed and superabsorbents. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Chala G.B. 2022. Assessing forage quality parameters in Timothy to enable breeding of high-quality forage with minimal environmental impact. Oral presentation, C4F workshop, November 29, Malmö, Sweden.

- Minina E. A. 2022. Recruiting plant autophagy for developing smart crops. . Oral presentation, C4F workshop, November 29, Malmö, Sweden.

Collaboration with industry or other parts of society

- Lantmännen
- Gasum
- Oriflame
- Grönsaksmästarna
- Region Skåne
- Lilla Harrie Valskvarn
- Ly Lyckeby Stärkelse AB
- Orkla
- Havredals Biodevelop AB
- RISE
- KTH
- Chalmers Kl
- Sveriges Stärkelseproducenter Förening
- DLF Beet Seed AB
- Kalmar Ölands Trädgårdsprodukter Findus

- FoodHills and ISCA Technologies
- Lund University
- Gunnarshögs <mark>Gård AB</mark>
- Syngenta
- Planta LLC
- SLU Grogrund a number of research projects
- connected to the C4F program
- Nelson Seed

Other funding that has been received partially or fully due to the TC4F research

- Novo Nordisk Foundation, Synthetic Wax Esters from Plants - A Green Source of the Pivotal Feedstock. 2.5 mil DKK, Kamil Demski main applicant

- Formas, Breeding oilseed rape (Brassica napus L.) for durable disease resistance and improved seed cake nutritional quality, 3 mil. SEK, Selvaraju Kanagarajan and Lihua Zhu, main and co-applicants.

- Formas, Unlocking plant protein and CRISPR potato starch novel uses: Design of fibre-based absorbents with tailored functionalities. Ramune Kuktaite, main applicant.

- VR, Increasing food security and decreasing malnutrition via innovation agro-platforms, crop breeding, and novel food processing of common beans in Uganda. 2.99 mil. Ramune Kuktaite, main applicant.

- Formas, Nutritious, tasty and health-promoting novel wheat products in research infrastructure, 6 mil, Eva Johansson, main applicant.

Formas, A strong pulse - resilient Swedish grain legume supply from field and factory to fork. 5.99 mil. Maud Langton, main applicant.

Education

a) PhD theses, MSc theses, Bachelor theses

Herneke, Anja. (Female) 2022. Doctoral thesis: Plant protein nanofibrils: characterising properties for future food. Uppsala: Sveriges lantbruksuniversitet. Acta Universitatis Agriculturae Sueciae, 2022:44. ISBN: 978-91-7760-963-6. Swedish University of Agricultural Sciences.

Sjur Sandgrind (Male) 2022. Doctoral thesis: Genome editing of oilseed species by CRISPR/Cas9 for trait improvement.

Lomma: Sveriges lantbruksuniversitet. Acta Universitatis Agriculturae Sueciae, 2022:77. ISBN: 978-91-8046-030-9, eISBN: 978-91-8046-031-6. Swedish University of Agricultural Sciences.

Elander, Pernilla (Female) 2022. Doctoral thesis: Living through hard times: Dispose of or sequester? Plant subcellular strategies for stress resilience. Uppsala: Acta Universitatis Agriculturae Sueciae, 2022:60. ISBN: 978-91-7760-995-7, eISBN: 978-91-7760-996-4. SLU. (https://res.slu.se/id/publ/119473). Swedish University of Agricultural Sciences.

Evelyn Elizabeth Villanueva Gutierrez (Female). 2022. Doctoral thesis: Bolivian tomatoes: genetic diversity, quality traits and value chains. Lomma: Sveriges lantbruksuniversitet. Acta Universitatis Agriculturae Sueciae, 2022:62. ISBN: 978-91-8046-000-2, elSBN: 978-91-8046-001-9.

b) Supervision and teaching (include supervision of finished and on-going students, include teaching and organization of courses)

LMuneer, Faraz. Main supervisor for Intern-student Vandamme, Lilou. Title: Processing of faba bean and pea protein to improve their functional properties for food application. 31st Oct to 23rd December, 2022.

Kuktaite, Ramune. Grogrund Workshop organizer and teacher "Product quality in PLS0089 Workshop Series II – Methods in Modern Plant Breeding", Dec 2, 2022, Alnarp, Sweden.

Kuktaite, Ramune. Teaching at the course "Plant biology for breeding and protection (Bl1296), Alnarp.

Herneke, Anja. Teaching at the course "Food Technology", (LV0112) 15 ECTS, Uppsala

Johansson, Mathias. Lab supervisor at the course Food Chemistry and Food Physics (LVO110), 15 ECTS, Uppsala

Langton, Maud Main supervisor for PhD-student Herneke, Anja. Title: Functionalization of nanofibers from plant based proteins

Langton, Maud Main supervisor for PhD-student Johansson, Mathias. Title: Legume based Gels – Microstructure and Texture

Hofvander, Per. Supervisor for PostDoc Shrikant Sharma

Hofvander, Per. Supervisor for PostDoc Kamil Demski

Zhu, Li-Hua. Supervisor for PhD candidate Oliver Moss. Tentative title: Improvement of seedcake quality of rapeseed for high quality food and food uses. Expected date for dissertation: 2025.

Roger Andersson. Main supervisor for PhD-candidate Shishanthi Jayarathna. Tentative title: New starch for novel applications. Expected date for dissertation: January, 2024.

Kanagarajan, Selvaraju. Co-supervisor for PhD candidate Sjur Sandgrind. Tentative title: Genome editing of oil crops. Expected date for dissertation: 2022.

Kanagarajan, Selvaraju. Supervisor for PhD candidate Oliver Moss. Tentative title: Improvement of seedcake quality of rapeseed for high quality food and food uses. Expected date for dissertation: 2025.

Kuktaite, Ramune. Main supervisor for PhD-candidate Sbatie, Lama. Tentative title: Wheat quality in a varying climate. Expected date for dissertation: June, 2023.

Minina, EA. Course organizer and teacher at the course "Real Time Quantitative PCR – theory, experimental design and data analysis", (PNS0215), 3.5 ECTS, SLU.

Roger Andersson. Course organizer and teaching at the course "Plant food science", (LV0113), 15 ECTS, SLU.

Hofvander, Per. Teaching at the course "Applied Plant Biotechnology" (BI1344), Alnarp.

Hofvander, Per. Teaching at the course "Sustainable plant production" (BI1295), Alnarp

Sharma, Shrikant. Teaching at the course "Växters kemi och biokemi" (KE0070), Alnarp.

Grimberg, Åsa. Teaching at the course "Växtförädling och växtfysiologi" (BI1367), Alnarp.

Grimberg, Åsa. Teaching at the course "Odling och kvalitet" (TD0010), Alnarp.

Grimberg, Åsa. Teaching at the course "Advanced plant breeding and genetic resources" (BI1345), Alnarp.

Grimberg, Åsa. Teaching at the course "Växters kemi och biokemi" (KE0070), Alnarp.

Andersson, Mariette. Teaching at the course "Advanced plant breeding and genetic resources" (BI1345), Alnarp.

Lager, Ida. Course leader and teaching at the course "Växters kemi och biokemi" (KE0070), Alnarp

Zhu, Li-Hua. Course organiser and teaching at the course "Advanced Plant Breeding and Genetic Resources" (BI1345), Alnarp

Zhu, Li-Hua. Course organiser and teaching at the course "Applied Plant Biotechnology" (BI1344), Alnarp.

Alyona Minina. Main supervisor for PhD-candidate Holla, Sanjana. Tentative title: Revealing the dynamics of plant autophagy. Expected date for dissertation: September 2023.

Alyona Minina. Main supervisor for PhD-candidate Ballhaus Florentine. Tentative title: Membrane-bound and membraneless organelles in plant stress response. Expected date for dissertation: September 2026.

Alyona Minina. Main supervisor for project student Kjelstrom, Jarl. Title: Use of Fungal bioluminescence pathway as a reporter for plant autophagy. 15 ECTS.

Alyona Minina. Lecture on advanced microscopy methods for the Masters degree course at Uppsala University "Genetic and Molecular Plant Science" (15 ECTS), September-October 2022

Alyona Minina. Organizer and teacher of the qPCR course (3.5 HEC, P000008 F0027) for the Organism Biology PhD School. SLU, Uppsala. November-December 2022

Florentine Ballhaus. Lab and seminar teacher for the Masters degree course at Uppsala University "Genetic and Molecular Plant Science" (15 ECTS), September-October 2022

Zhu, Li-Hua. Supervisor for PhD candidate Sjur Sandgren. Tentative title: Genome editing of oil crops. Expected date for dissertation: 2022.

Zhu, Li-Hua. Supervisor for PhD candidate Oliver Moss. Tentative title: Improvement of seedcake quality of rapeseed for high quality food and food uses. Expected date for dissertation: 2025.

C4F- Crops for the Future, Personnel

Name	Gender & Position	Part of full time financed by TC4F
Eva Johansson	F, Professor	15%
Li-Hua Zhu	F, Professor	10%
Maud Langton	F, Professor	0
Roger Andersson	M, Professor	0
Pär Ingvarsson	M, Professor	0
Anne-Maj Gustavsson	F, Docent	0
Anna Westerbergh	F, Docent	0
Thomas Prade	M, Docent	0
Galia Zamaratskaia	F, Ass. Prof.	0
Mariette Andersson	F, Researcher	0
Ramune Kuktaite	F, Researcher	0
Åsa Grimberg	F, Researcher	0
lda Lager	F, Researcher	0
Selvaraju Kana <mark>g</mark> arajan	M, Researcher	8%
Alyona Minina	F, ass. Prof.	35%
Per Hofvander	M, Researcher	0
Girma Bedada Chala	M, Researcher	25
Sjur Sandgrind	M, PhD student	50%
Faraz Muneer	M, postdoc	100%
Kamil Demski	M, Postdoc	40%
Shrikant Sharma	M, Postdoc	40%
Adrian Dauphinee	M, Postdoc	0%
Florentine Ballhaus	F, PhD student	35%
Anna-Lovisa Nynäs	F, PhD student	50%
Oliver Moss	M, PhD student	50%
Anja Herneke	F, PhD student	0
Sanjana Holla	F, PhD student	0
Sbatie Lama	F, PhD student	0
Shishanthi Jayarathna	F, PhD student	30%
Mathias Johansson	M, PhD student	0
Lan Yuzhou	M, PhD student	25%
Lekan Jolayemi	M, PhD student	50%
Linda Öhlund (Lantmännen)	F, Foragebreeder	0
Xueyuan Li	M, Research	0

*Researchers listed with 0% have received financing from TC4F earlier which resulted in projects with independent financing.

TC4F Economy 2022

In 2022, TC4F received 28,8 mio SEK of funding which were distributed according to the budget of which 88% were used. The surplus was caused by delays in recruitment due to the Covid-19 pandemic.

	SLU	UmU	Skogforsk	Total
Distributed Funds (tkr)				
Coordination	2969			2969
Plant Physiology (UMU)		5100		5 100
Forest Genetics and Plant Physiology	4 454*	A		4 454
Southern Swedish Forest	4 454*			4 454
Forest Ecology and Management	<mark>3</mark> 504			3 504
Wildlife, Fish and Environmen <mark>tal Studies</mark>	<mark>0</mark> *			0
Skogforsk			1 100	1 100
C4F (LTV)	7194			7194
TOTAL	22 57 <mark>5</mark>	5 100	1 100	28 775
Costs, spent funds (tkr)				
Coordination	846			846
Plant Physiology (UMU <mark>)</mark>		5100		5100
Forest Genetics and Plant Physiology	1 947			1 947
Southern Swedish F <mark>orest</mark>	5 065			5 046
Forest Ecology and Management	<mark>3 9</mark> 46			3 946
Wildlife, Fish and Environmental Studies	<mark>73</mark> 1			731
Skogforsk			1 100	1100
C4F (LTV)	6 709			6 709
Total	19 246	5100	<mark>1 1</mark> 00	25 449
PROFIT T4F	2 <mark>845</mark>	0	0	
PROFIT C4F	48 <mark>4</mark>			
Total PROFIT	3 329	0	0	3329

*assigned postdoc grants

