Physiological and biochemical characterisation of four South African varieties of Bambara groundnut (Vigna subterranea (L.) Verdc.)

Deshni Pillay
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Introduction

Bambara groundnut (*Vigna subterranea* (L.) Verdc.) is an indigenous African crop, grown and consumed throughout the continent, fulfilling the protein portion of the diets of many of its inhabitants (Heller and Mushonga, 1997). It is especially suitable in sub-Saharan Africa where it is cultivated in traditional farming systems, intercropped with other root and/or cereal crops. In South Africa, it is cultivated in the northern and eastern parts of the country, at subsistence level (Swanevelder, 1998). Lack of seed, unsuitable varieties, pod losses during harvesting and superstition related to traditional preferences and practices (Swanevelder, 1998) has contributed to the limited cultivation of this pulse in South Africa. A major constraint is the selection of varieties suitable for a specific area and according to the requirements of the different farmers. This crop was chosen for study because it was phenotypically diverse, highyielding, widely but not intensively cultivated, important for food security, applicable to low-input cultivation practices and suited to diverse climatic conditions.

Seeds of bambara groundnut exhibit a high phenotypic diversity and varieties selected for cultivation are usually dependent on the seed-coat colour and pattern, as this is often linked to palatability and growth potential. Seed-coat colour is attributed to the presence and amount of phenolic derivatives (Werker, 1997; Beninger et al., 1998), which have reported antimicrobial activity (Nicholson and Hammerschmidt, 1992) and disease resistance (Higuera and Murthy, 1987; Pakela, 2003), as well as adverse tastes, poor digestibility (Aw and Swanson, 1985) and decreased nutritive protein value (Krygier et al., 1982).

Other seed physiology aspects that also need consideration when selecting varieties for planting and consumption are the germination potential, seedling growth and emergence. Sreeramulu (1983) showed that germination time of bambara groundnut varies between 7-15 d, depending on temperature, soil type and water availability. For wild forms, however, germination is erratic and can take up to a month after sowing (Hepper, 1970). Old seeds can result in low germination and stunted seedlings (Sreeramulu, 1983), whilst soaking seeds and temperatures between 16.7°C and 39.5°C are optimal for germination (Massawe et al., 1999; Kocabas et al., 1999; Massawe et al., 2003). Thus germination potential is a crucial measure of plant establishment and performance in the field.

Furthermore physiological studies have shown that growth and development of bambara groundnut is greatly influenced by external factors like photoperiod (Linnemann, 1993; Linnemann et al., 1995), temperature
(Linnemann and Craufurd, 1994; Brink et al., 2000) and soil type (Uguru and Ezeh, 1997; Suwanprasert and Sinsawat, 2002). Knowing the physiological and biochemical characteristics can assist commercial and subsistence farmers when selecting varieties for cultivation and consumption. Disease resistance, large yields and even stand are some requirements of commercial farmers whilst phenotypic diversity, quick cooking, high palatability and low phenolic content are more important to subsistence farmers.

For this study selected advanced varieties (SB 1-1, SB 7-1 and SB 20-2/A) and one local variety (MBP 51) were compared to evaluate if different farmers needs are satisfied. The suitability of these varieties was evaluated by germination tests, quantifying the phenolic content and growth performance between soil types using emergence, plant height, plant weight and leaf number/plant as parameters, then relating them to the seed size and colour. A further aim of the investigation was to correlate phenotypic, physiological and biochemical characteristics of the four selected varieties.

**Materials and methods**

**Bambara groundnut varieties**

Four varieties of bambara groundnut were selected. Three of these, SB1-1, SB7-1 and SB20-2A (Cilliers and Swanevelder, 2002), were provided by the ARC-Grain Crops Institute (Potchefstroom, South Africa). The fourth variety MBP 51 was purchased from a local seed producer, EcoLink (Nelspruit, South Africa). The former three varieties were developed for use nationally and the latter variety has been developed locally. The seed material from the ARC-Grain Crops Institute has been described phenotypically (Cilliers and Swanevelder, 2002). Differences in seed size were expressed in the 100-seed weight parameter of each variety.
Physiological and biochemical characterisation of four South African varieties of Bambara groundnut

Fig 1: The four varieties of bambara groundnut selected for this study, a = SB 20-2A, b = SB 1-1, c = MBP 51 and d = S B7-1.

Physiological characterisation

Experiment 1: Germination potential and seedling growth

The germination tests were conducted according to guidelines for Vigna unguiculata (L.) Walp., (ISTA, 1999) with some changes. Evaluations were performed at 8 and 10 d, constituting the first and final evaluations respectively, since at the recommended 5 d the number of germinated seeds was negligible.

Four replicates of 100 seeds of each variety of bambara groundnut were selected for the seed germination and seedling growth study. Seeds were surface disinfected for seed surface mycoflora, placed on germination paper, rolled-up and incubated at 25°C for 10 d. All seeds were evaluated after 8 and 10 d for the first and the final ratings, respectively.

For each of the varieties, the percentage germination was determined by recording the number of normal, abnormal, diseased and non-germinated seeds in each replicate. Diseased seeds observed on the 8-day evaluation were removed. Epicotyl and radicle lengths were measured to establish differences in seedling potential amongst the varieties. Final seedling measurements were made with a ruler at the end of 10 d only. Seedling growth was quantified by measuring the radicle length with a length of 10 mm implying germination (Garcia-Huidobro et al., 1982).

Only results of the normally germinated seeds at the final evaluation were used for analysis (ISTA, 1999).
Experiment 2: Effect of soil type on the emergence, growth and development of four varieties of bambara groundnut

A randomised block factorial experimental design, with four varieties and two soil types, and replicated three times was used. The experiment was planted in a glasshouse at the University of Pretoria Research Farm (Hilcrest, Pretoria). The average daylight hours, for Pretoria, between April and July are 11.5 and 10.7 h, respectively (Buys, 1978). Average temperature was between 23-25 °C.

Eighteen seeds/variety were planted, with a seeding rate of three seeds/pot (20 cm diameter) and at a depth of 3 cm. Loam topsoil was amended with river sand (Conradie Organics, Pretoria) or potting soil (Just Nature, Pretoria) in a 1:2 ratio, constituting soil type A and soil type B respectively. Watering was every two days, just sufficient to keep the soil moist and no additional fertilizers were used.

Three plants/variety were randomly selected at 30-day intervals, for the planned trial period (120 d), to monitor plant growth and development. Observations included the emergence percentage, plant height, plant weight and number of leaves/plant. Emergence was observed 10 days after sowing (DAS).

The trial was repeated at the ARC Research Farm (Roodeplaat, Pretoria). Both the trials were terminated at 90 DAS, as a high incidence of powdery mildew occurred. While the University trial was completely destroyed the Roodeplaat trial recovered and was evaluated at 120 DAS. These results, however, were not included in the analysis.

Biochemical characterisation

Experiment 3: Extraction, quantification and separation of various phenolics from the bambara groundnut varieties

Sample preparation: One hundred seeds per variety were selected and soaked overnight at room temperature, in 100 ml of distilled water. After 24 h, the seed coats, endosperms and embryos were separated, frozen in liquid nitrogen and freeze dried. The samples were lyophilised and ground into a powder with a mortar and pestle. The fine powder was sieved (5 mm) to ensure homogeneity.

Extraction of phenolics: The obtained powder (0.050 g) was exhaustively extracted with 1.0 ml of solvent (methanol:acetone:water, 7:7:1, v:v:v), according to the methods used by Regnier (1994). The suspension was homogenised for 1 min and then shaken for 1 h at 150 rpm before being centrifuged at 12000 x g for 5 min. After centrifugation the supernatant was removed and saved. The remaining pellet was rehomogenised, shaken and centrifuged as above. The two supernatants were combined. This procedure
was repeated four times, to ensure at least 90% extraction of the total soluble phenolics. The resulting supernatant was reduced under vacuum and adjusted to 1.0 ml with distilled water. The remaining precipitate was dried at 56 °C for 24 h and stored. This procedure resulted in two replicates of four tubes, which were used for further analysis.

**a) Total soluble phenolic acids:** Four tubes were used for the determination of total soluble phenolics. The prepared aliquoted samples of supernatants were concentrated under vacuum to 1.0 ml as above and used to determine the total soluble phenolic content with the Folin-Ciocalteu reagent.

**b) Extraction of non-conjugated phenolic acids:** The same four tubes were used for the determination of non-conjugated phenolic acids and ester-bound phenolics. The prepared aliquoted samples of supernatants was concentrated to 500 µl and trifluoroacetic acid (TFA, 2 %) was added to each sample to acidify the solution, before extraction with 1.0 ml anhydrous diethyl ether (Regnier, 1994). The combined extracts were dried under vacuum and the resulting precipitate was resuspended in 100 µl methanol. This solution was used to determine the non-conjugated phenolics with the Folin-Ciocalteu reagent.

**c) Extraction of ester-bound phenolic acids:** Ammonium sulphate (20% w/v) was added to the remaining solution and followed by ethyl acetate (20% v/v); the supernatant was removed and left to evaporate. The process was repeated five times to ensure maximum extraction of esters. The combined extracts were dried under vacuum and the resulting precipitate was resuspended in 100 µl methanol (Regnier, 1994). This solution was used to determine the ester-bound phenolics with the Folin-Ciocalteu reagent.

**d) Conjugated phenolic acids:** After removing the non-conjugated and ester-bound phenolics, from the total pool of soluble phenolics, the remaining portion constituted the soluble conjugated phenolic derivatives.

**Folin-Ciocalteau assay:** The method used to determine the total soluble phenolic content and the non-conjugated phenolic content, is based on the reduction of the phospho-molybdenum/phospho-tungstate present in the Folin-Ciocalteau reagent (Swain and Hillis, 1959). The concentrated supernatants were diluted three times with distilled water and 5 µl was added to 25 µl of 50 % (v/v) Folin-Ciocalteau, 170 µl distilled water, and mixed. After 3 min, 0.5 ml of saturated aqueous sodium carbonate was added, mixed and incubated at 40°C for 30 min. For the blank control 5 µl of distilled water was added instead of the supernatant. Gallic acid was used as a phenolic standard to construct a standard curve ranging from 0 to 400 µg. The concentration of phenols in the various extracts was calculated from the standard curve and expressed as mg gallic acid g⁻¹ dry weight.

**Extraction of cell wall-bound phenolics:** The fine seed coat powder (0.050 g) was extracted four times with 1.0 ml 70 % acetone. The combined supernatants were dried under vacuum, resuspended in 500 µl methanol and stored at 4°C for further analysis.
Thin layer chromatography (TLC) analysis: The cell wall-bound phenolics (proanthocyanidins) were isolated by preparative TLC. The crude extract (5 µl) of each seed coat of the four varieties was chromatographed on TLC aluminium plates (20 cm x 20 cm), (Merck, silica gel 60F_{254}) and glass plates (20 cm x 20 cm), with a solvent system of butanol: acetic acid: water (4:1:5), as described by Markham (1982). The plates were placed in a glass column with the prepared solvent and left to migrate. The cell wall-bound phenolics were viewed under a dual-wavelength (260/340 nm) UV lamp to distinguish the separated compounds, according to the R_{f} values of the prepared standards.

Data analysis and experimental design

All statistical analyses were performed using GenStat (2000) program. Analysis of variance (ANOVA) was performed for the normally germinated seeds, in experiment 1, for the final evaluation (10 d) only. A least significant difference (of P < 0.05) was accepted as significant (Snedecor and Cochran, 1980).

Experiment 2 was designed as a randomised complete block design with six replicates, testing for differences between four varieties and two soil types (i.e. eight treatment combinations), at two locations. ANOVA was used to test for differences between the main effects of varieties and soil types, as well as the variety soil type interaction for each locality. Then a combined ANOVA was run to test for treatment-locality interaction, which was not significant (P > 0.05), meaning that the eight treatments responded similarly at both localities. Therefore the two localities were compared as replicates and pooled for the statistical analysis. The data were acceptably normal with homogeneous treatment variances. Treatment means were separated using Fischers’ protected t-test and least significant difference (LSD) at the 5% level of significance (Snedecor and Cochran, 1980).

Results

Germination percentage, seed size and colour

Differences in the seed size (Table 1) colour and pattern (Fig. 1) were observed. The analysis indicated no statistical difference (P > 0.05) for germination percentage (Fig. 2) amongst the varieties for the normally germinated seed category, at both evaluations (8 and 10 d). There were significant differences amongst the varieties with regards to seedling length (Fig. 3). Seedlings of MBP 51 where significantly longer (7.4 cm) for mean epicotyl length. Similarly for mean radicle length, MBP 51 had the highest length (8.09 cm) and was similar to SB 1-1 (6.44 cm) and SB 7-1 (6.95 cm) whilst SB 20-2A had the lowest radicle length (4.47 cm) with significant differences to the other three varieties (Fig. 3).
Table 1: Summary table of the physiological and biochemical characteristics of the four bambara groundnut varieties investigated *Means within a row not followed by the same symbol are significantly different (P< 0.05)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>SB 1-1</th>
<th>SB 7-1</th>
<th>SB 20-2A</th>
<th>MBP 51</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seed coat colour</td>
<td>Brown, with red speckles</td>
<td>Dark red</td>
<td>Cream</td>
<td>Brown</td>
</tr>
<tr>
<td>100-seed weight (g)</td>
<td>52.03</td>
<td>44.04</td>
<td>52.98</td>
<td>24.98</td>
</tr>
<tr>
<td>Germination percentage (%)</td>
<td>Mean of normally germinated seeds</td>
<td>45.25*</td>
<td>44.50</td>
<td>33.50a</td>
</tr>
<tr>
<td>10 day evaluation</td>
<td>Mean of normally germinated seeds, at 10 d</td>
<td>45.25*</td>
<td>44.50</td>
<td>33.50a</td>
</tr>
<tr>
<td>Epicotyl</td>
<td>4.78*</td>
<td>5.65ab</td>
<td>5.11ab</td>
<td>7.40</td>
</tr>
<tr>
<td>Radicle</td>
<td>6.44d</td>
<td>6.95cd</td>
<td>4.47</td>
<td>8.09cd</td>
</tr>
<tr>
<td>Emergence percentage (%)</td>
<td>Pooled results over 2 locations</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>soil type A</td>
<td>58.10*</td>
<td>68.60a</td>
<td>78.30a</td>
<td>68.30a</td>
</tr>
<tr>
<td>soil type B</td>
<td>76.00a</td>
<td>71.50a</td>
<td>79.90a</td>
<td>74.00a</td>
</tr>
<tr>
<td>Total soluble phenolics (mg gallic acid/g dry weight) in seed coat</td>
<td>30.40*</td>
<td>39.20ab</td>
<td>3.50</td>
<td>50.10a</td>
</tr>
<tr>
<td>Non-conjugated phenolics (mg gallic acid/g dry weight) in seed coat</td>
<td>0.89a</td>
<td>0.71a</td>
<td>0.15</td>
<td>1.88</td>
</tr>
<tr>
<td>Ester-bound phenolics (mg gallic acid/g dry weight) in seed coat</td>
<td>0.52a</td>
<td>0.78a</td>
<td>0.08</td>
<td>1.68</td>
</tr>
</tbody>
</table>
Fig 2: Mean percentage of normally germinated seeds, of four varieties of bambara groundnut for the first (8 d) and final (10 d) evaluations. *Each value is a mean percentage of four lots of 25 seeds, replicated four times. Means within a column not followed by the same letter are significantly different (P< 0.05)

Fig 3: Mean epicotyl and radicle lengths of normally germinated seedlings of four varieties of bambara groundnut, at the final evaluation (10 d). *Each value is a mean percentage of four lots of 25 seeds, replicated four times. Means within a column not followed by the same letter are significantly different (P< 0.05)
Emergence, growth and development of the bambara groundnut varieties

No significant difference was observed amongst the varieties for the emergence in the two soil types (Fig. 4). Results for the growth studies indicate that there was no significant difference for the variety soil type interaction tested, for plant height and plant weight parameters at 30, 60 and 90 DAS (Table 2). A significant difference was observed for leaf number at 60 DAS only, whilst for the other two evaluations (30 and 90 DAS), there was no significant difference for this parameter.

Table 2: Summary table of the characteristics of each of the four varieties of bambara groundnut investigated *Means within a column not followed by the same symbol are significantly different (P< 0.05)

<table>
<thead>
<tr>
<th>Variable</th>
<th>30 d</th>
<th>60 d</th>
<th>90 d</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Soiltype A</td>
<td>Soiltype B</td>
<td>Soiltype A</td>
</tr>
<tr>
<td>Plant height (cm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SB 1-1</td>
<td>21.6*a</td>
<td>22.7a</td>
<td>25.5*b</td>
</tr>
<tr>
<td>SB 7-1</td>
<td>23.1a</td>
<td>22.4a</td>
<td>23.5b</td>
</tr>
<tr>
<td>SB 20-2A</td>
<td>25.4a</td>
<td>23.8a</td>
<td>25.4b</td>
</tr>
<tr>
<td>MBP 51</td>
<td>21.8a</td>
<td>22.3a</td>
<td>23.3b</td>
</tr>
<tr>
<td>Plant weight (g)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SB 1-1</td>
<td>12.7a</td>
<td>11.0b</td>
<td>21.1b</td>
</tr>
<tr>
<td>SB 7-1</td>
<td>13.0a</td>
<td>11.1b</td>
<td>19.1b</td>
</tr>
<tr>
<td>SB 20-2A</td>
<td>11.4a</td>
<td>10.7b</td>
<td>16.6b</td>
</tr>
<tr>
<td>MBP 51</td>
<td>8.1a</td>
<td>8.2b</td>
<td>12.9b</td>
</tr>
<tr>
<td>Leaf number/plant</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SB 1-1</td>
<td>4a</td>
<td>4a</td>
<td>7q</td>
</tr>
<tr>
<td>SB 7-1</td>
<td>4a</td>
<td>4a</td>
<td>6rs</td>
</tr>
<tr>
<td>SB 20-2A</td>
<td>4a</td>
<td>4a</td>
<td>7u</td>
</tr>
<tr>
<td>MBP 51</td>
<td>4a</td>
<td>4a</td>
<td>6rs</td>
</tr>
</tbody>
</table>
Phenolic content in the different bambara groundnut varieties

There was a highly significant difference amongst the varieties for the amounts of soluble and cell wall-bound phenolics in the seed coat only (Fig. 5a). The total soluble pool of phenolics comprised of non-conjugated, ester-bound and conjugated phenolics (Table 1). The embryo and endosperm components of the seeds had negligible amounts of these phenolic derivatives (Fig. 5b and 5c) and were not analysed further. TLC separation of the crude extracts of the cell wall-bound phenolics for the respective varieties indicated that three phenolic compounds were associated with SB 20-2A only (Fig. 6a and b), compound 1 ($R_f = 0.34$), compound 2 ($R_f = 0.78$) and compound 3 ($R_f = 0.86$).
Fig 5: Total soluble phenolics in equivalent mg gallic acid/g dry weight, of the four varieties (SB 1-1, SB 7-1, SB 20-2A and MBP 51) of bambara groundnut, in (a) seed coat, (b) embryo and (c) endosperm
Figure 6a

Figure 6b

**Discussion**

**Correlating germination and phenolic content to seed phenotype**

In this study there was no significant difference observed for the germination study implying that these varieties can be selected according to the specific
preferences of farmers and not only on their phenotypic characters (seed size and colour). Light coloured seeds are usually desirable because of their reputation of being highly palatable and easy to digest, whilst dark coloured seed are promoted for their disease resistance. Studies on cowpeas (Pakela, 2003), a close relative of bambara groundnut, have reported increased disease resistance for dark coloured and patterned seeds relative to lighter ones. Accordingly farmers are encouraged to plant dark coloured seeds for better crop performance. However, darker seeds have higher phenolic derivatives especially in the seed coats, as reported for cowpeas (Pakela, 2003), dry beans (Beninger and Hosfield, 1999), legumes species (Sosulski and Dabrowski, 1984), selected brassica seeds (Simbaya et al., 1995) and shown in this study. These phenolic derivatives contribute to adverse tastes, poor digestibility (Aw and Swanson, 1985) and decreased nutritive protein value (Krygier et al., 1982).

But farmers, especially subsistence ones, prefer cultivating light coloured varieties that are more palatable than darker ones. Since variety SB 20-2A performed equally well to the other varieties it can be recommended for cultivation, to suit the needs of individual farmers.

Seed physiological factors like seed size and seedling growth haven been shown to affect germination and crop performance (Sreeramu, 1983), especially important since bambara groundnut can have poor field establishment due to low germination and seedling growth (Massawe et al., 1999; Kocaba et al., 1999; Massawe et al., 2003). These studies also indicated that germination percentage was dependent on genotype, temperature, seed size and age. The latter two parameters exhibited a directly proportional relationship to growth potential in the previous studies whilst the results in this study indicated an inverse relationship between seed size and seedling length, for the varieties selected. MBP 51 seeds with the longest seedlings, weighed the least as compared to SB 1-1, SB 7-1 and SB 20-2A, which all had higher 100-seed weights. The latter varieties had similar mean epicotyl lengths that differed significantly from MBP 51. For radicle length a significant difference was only shown in SB 20-2A which had the shortest mean length.

Seed size and colour are therefore not good indicators of a variety’s genotype and growth potential, since significant differences existed between SB 1-1, SB 7-1 and SB 20-2A, which are all advanced varieties, and MBP 51 that was developed by a local seed producer. Our results have shown that although phenolic content and seed size are important from a seed pathological perspective these traits are not applicable when selecting seeds for consumption purposes. The phenolic investigation clearly elucidated that dark coloured varieties contained significantly larger amounts of total soluble phenolics, as expressed by the non-conjugated, ester-bound and conjugated phenolics in the seed coats only. The cream variety contained negligible amounts of soluble phenolic derivatives investigated, in the seed coat, embryo
and endosperm. This thus implies that SB 20-2A would be more palatable and suitable for cultivation by subsistence farmers, as this is their primary requirement.

**Emergence, growth and development of bambara varieties in two soil types**

Bambara can perform successfully in a variety of soil conditions but has optimal yield in vertisol (Uguru and Ezeh, 1997) with some varieties being sensitive to saline and calcareous soils (Suwanprasert and Sinsawat, 2002). Studies (Sreeramulu, 1983) on emergence indicated that crop performance is directly affected by genotype, temperature, seed size and age. This study contrasted with our study since there were no significant differences amongst the selected varieties between the soil types. In this study soil type A, a mixture of loam topsoil and river sand and soil type B a mixture of loam topsoil and potting soil served as a nutrient poor medium and a nutrient rich medium respectively. From the emergence percentages calculated all varieties performed well in both soil types, suggesting that these varieties are well adapted for subsistence farmers that often plant in areas unsuitable for cultivation of economically important crops. The results obtained for the plant growth analysis and development studies show that soil type and variety cannot be used as the only distinguishing parameters when selecting varieties for cultivation.

The significant difference in leaf number at 60 DAS only can be attributed to the different growth phases in plants. Bambara groundnuts usually mature between 90-120 DAS, depending on variety, temperature, photoperiod and water potential (Swanevelder, 1998). Vegetative growth should therefore increase during this crucial period as the plants starts to initiate pod formation. Food energy reserves would be increasing and physiologically the plants are photosynthesizing more. The increase in leaves would facilitate these processes and can be attributed to the differences observed at this stage.

**Linking biochemical, physiological and phenotypic characteristics of the varieties**

The phenolic content is an important characteristic of seeds, with regsrds to consumer preferences and food quality. Phenolics quantified were soluble (non-conjugated, ester-bound and conjugated phenolics) and cell wall-bound phenolics. Varieties SB 1-1, SB 7-1 and MBP 51, contained higher quantities of the former phenolics than variety SB 20-2A, which correlated with the pigmentation differences observed in the seed coats. The non-conjugated and ester-bound phenolics form only a minor part in the pool of total soluble phenolics extracted, whilst the remaining portion comprises of conjugated
phenolics. As previously reported for cowpeas (Pakela, 2003), a higher concentration of soluble phenolics was present in the seed coat than the endosperm and embryo. The compounds associated with the cell wall-bound phenolics included gallic acid, cafferic acid and para-coumaric acids which were isolated from SB 20-2A only. These results correlate with similar research by Beninger and colleagues (1998) on dry bean varieties, who did not detect any cell-walled phenolics in dark pigmented seeds.

The results suggest that in the seed coats of bambara groundnuts, the phenolics acids predominately exist in soluble conjugated forms and not bound to the cell-walls. These conjugated phenolics are responsible for the palatability in seeds by attributing a bitter taste. This thus can contribute to the varieties selected for cultivation amongst farmers, as varieties with light coloured seed coats are preferred for consumption. Since variety SB 20-2A has a lower soluble conjugated phenolic content and it performed similarly to the dark pigmented varieties for the germination, emergence and growth parameters, our results indicate that it can still be recommended to farmers for cultivation, as there is no strong evidence to support that darker coloured seeds perform better under the same conditions.

**Conclusions**

Commercial farmers and small-scale farmers have different requirements when choosing varieties for cultivation. For farmers, the colour of the seed coat can clearly distinguish phenolic content relating to palatability, and seed size will determine the seed potential in germination, emergence, growth and development. Since there was no significant difference in the germination, emergence and growth potential of SB 20-2A, relative to the three other dark coloured varieties, it can be recommended for cultivation amongst farmers. Furthermore there was a significantly higher concentration of soluble phenolics in the seed coats of the former varieties than the latter one. Selecting varieties using phenotype as the only criterion can be misleading as this study showed that larger and dark colour seeds did not correlate to better growth performance. Instead distinctions can be made amongst the genotypes of the advanced varieties (SB 1-1, SB 7-1 and SB 20-2A) and the locally produced one (MBP 51). The bambara groundnut varieties used in this study, performed equally well in germination and emergence tests and were conducive to cultivation in both soil types, which are desirable characteristics, especially to subsistence farmers. Finally these characteristics investigated further substantiate the versatility of bambara groundnut especially to resource-poor farmers growing for subsistence use.
Acknowledgements

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References


