

EVALUATION OF NUTRITIONAL PROPERTIES OF TISSUE CULTURED SORGHUM [SORGHUM BICOLOR (L) MOENCH]

**E. G. O. Omondi¹, M. N. Makobe¹, C. A. Onyango¹, L. G. Matasyoh², M. O. Imbuga¹
and E. N. Kahangi¹**

¹Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya

²Chepkoilel University College

Email: eomondi2008@gmail.com

Abstract

Tissue culture techniques are commonly used in plants as an efficient way to propagate and store valuable genotypes. Often, some of the regenerants differ from the parental type, a phenomenon called somaclonal variability. Assessment of nutritional value variability in crops that may arise from somaclonal variability during tissue culture propagation may have a strong impact on plant breeding, conservation of genetic resources and nutrition in the areas of use. It is particularly useful in the characterization of individual cultivars, and in determining duplications in germplasm collections and for selecting parents. The *Sorghum bicolor* (L) Moench tissue culture (TC) regenerants (Seredo, Mtama 1 and El Gardam) were developed at the Jomo Kenyatta University of Agriculture and Technology towards improvement for water stress tolerance for improved food production in the ASALs in Kenya.

The study was conducted to evaluate the nutritional value of the parents and TC regenerants of *Sorghum bicolor* (L) Moench local cultivars (Seredo, Mtama 1 and El Gardam) in Kenya. For proximate composition significant ($p \leq 0.05$) differences were observed in parents and regenerants of the El-Gardam (moisture, proteins and crude fiber), Mtama 1 (proteins) and Seredo (fats and crude fiber). The mineral compositions of the parents and regenerants of the cultivars were not significantly different ($p \leq 0.05$) except for Zinc in Mtama 1 cultivar and Iron in both El-Gardam and Mtama 1 cultivars. B-vitamins showed significant differences ($p \leq 0.05$) for both thiamine and Pyridoxine in El-Gardam and Seredo. Significant variability ($p \leq 0.05$) was shown phytates content in each cultivar. The parents were observed to have significantly higher amounts of Phytates than the regenerants within all the cultivars. The study recommends Mtama 1 regenerants with low anti-nutrient appropriate for ASALs with respect to nutrient availability since anti-nutrients in sorghum have been shown to impair the bioavailability of the other nutrients to the body.

Key words: Cultivars, TC regenerants, water stress tolerance, nutritional value

1.0 Introduction

Sorghum (*Sorghum bicolor* L. Moench) is the fifth most important cereal crop in the world after wheat, rice, corn and barley (FAO, 2005). About 90% of the world's sorghum areas lie in the developing countries, mainly in Africa and Asia. This crop is primarily grown in marginal areas prone to low rainfall where other grains are unable to survive unless irrigation is available. The future of the sorghum and economy may be linked to its contribution to food security in Africa, income growth and poverty alleviation and efficient use of water in drought-prone regions in much of the developed world.

The sorghum grain quality is affected by factors such as genotype, climate, soil type, and fertilization, among others. These can affect the chemical composition and the nutritive value (Ebadi *et al.*, 2005). Starch is the main component of sorghum grain, followed by proteins, non-starch polysaccharides (NSP) and fat (BSTID-NRC, 1996). Its protein content is higher than that of corn although its nutritional protein quality is lower (Dowling *et al.*, 2002). Moreover, the high tannin content in most of the sorghum binds to protein, carbohydrates, and minerals making them unavailable for digestion in the body. Reduction of tannin levels is possible through decortication, fermentation, germination and chemical treatment according to Dicko *et al.*, 2005 and Beta *et al.*, 1999.

There is an urgent need to focus on improving crops relevant to the small farm holders and poor consumers in the developing countries of the humid and semi-arid tropics. Efficient transformation systems, both biolistic and Agrobacterium-mediated transformation, for important cereal crops such as sorghum have been established (Tingay *et al.*, 1997). Nutritional quality has been identified as priorities by major international funding programs, such as FAO, (2005) and these are protein quality and the content of vitamins and minerals (O'Kennedy *et al.*, 2006). The objective of this study was to evaluate the nutritional value and safety of the *Sorghum bicolor* (L) Moench TC regenerants compared to the parent plants.

2.0 Methodology

Seeds of three cultivars of *Sorghum bicolor* (L) Moench seeds from two growing seasons were selected from Kenya Agricultural Research Institute (KARI) Katumani, a semi-arid region in Eastern Province of Kenya. They were Seredo, Mtama 1 and El Gargam. *Sorghum bicolor* (L) Moench TC regenerants were developed towards improvement for water stress tolerance at the Jomo Kenyatta University of Agriculture and Technology. Regenerants were obtained through somatic embryogenesis on Linsmaier and Skoog's (LS) basal salts augmented with Mannitol osmoticum to simulate water stress conditions in the ASALs (Makobe *et al.*, 2006). Ground samples from the grains were used for the nutritional analysis experiment. Each experiment was carried out in duplicate.

Proximate Analysis

Moisture, protein, carbohydrates, fat, ash and crude fibre were determined according to AOAC methods specification 950.46 (AOAC, 1995).

Tannin

Analysis was done according to vanillin-hydrochloric acid method (Burns, 1963; Price *et al.*, 1978).

Phytates

HPLC method according to Camire and Clydesdale, (1982) was used in the analysis. During analysis, 50mg of each sample was utilized. Extraction was carried out with 25 ml of 3% H₂SO₄ for 30 minutes on a shaker bath (German model KS 259 basic) at medium speed for 30 min at 21°C. The slurry was filtered through fast filter paper and rinsed using a fine jet stream from a squeeze bottle, with a small volume of extracting solvent. The filtrate was then transferred to 50ml centrifuge tubes and placed in boiling water bath for 5 minutes before addition of 3ml of a FeCl₃. The tubes were heated in boiling water bath to allow for the complete precipitation of the ferric phytate complex. Centrifugation was then done at 2,500 rpm (Japan model H-2000C) for 10 minutes and the supernatant discarded. The precipitates were washed once with 30 ml distilled water, centrifuged and the supernatant discarded again. Three (3) ml of 1.5N NaOH and a few ml of distilled water were added to the contents of the tubes. The volume was then brought to approximately 30 ml with distilled water and heated in boiling water bath for 30 minutes to precipitate the ferric hydroxide. The cooled samples were centrifuged and the supernatant transferred to 50 ml volumetric flasks. The precipitate was then rinsed with 10ml distilled water, centrifuged at 2,688 X g (Japan model H-2000C) for 10 minutes and the supernatant added to the contents of the volumetric flask.

Samples of 2µl of the supernatant were injected into a HPLC (Shimazu model C-R7A plus) fitted with a 50377 RP-18 (5µl) column Cat. at an oven temperature of 30°C and RID-6A detector model. A stock solution of the standard containing 10mg/ml of sodium phytate (inositol hexaphosphoric acid C₆H₆ OPO₃Na₂)₆ + H₂O) in distilled water was prepared. Serial dilutions were made for the preparation of the standard curve. Results of the phytate content were obtained as per the calculations of Vohra *et al.*, 1965.

Phytate content (mg/g) = (y/b) × (dilution factor / weight of sample)

Where y = peak area; b = concentration

Minerals

A portion of 5gm of the samples were weighed in crucibles and transferred to hot plates in the fume hood chamber where they were charred to clear all the smoke from the carbonatious material before transferring them to the muffle furnace. The charred materials were then incinerated at 550°C until they were reduced to white

ash. The ash was cooled, 15 ml of 6N HCL added to each and transferred to 100ml volumetric flasks. Distilled water was used to top them up to the mark before mineral analysis (AOAC, 1995). Atomic Absorption Flame Emission Spectrophotometer was used for the sodium metal residue analysis of the alkali treated samples (Model A A-6200, Shimadzu, Corp., Kyoto, Japan).

Protein Digestibility

Protein digestibility was done according to the method described by Mertz *et al.*, 1984. This method involved determination of the protein content of sample before and after pepsin enzyme digestion.

The pepsin digestion involved weighing 0.2 g of ground sample that was passed through a 0.4mm screen and adding 35 ml of 0.1M phosphate buffer: pH 2 containing 1.5mg pepsin /ml. Incubation of the pepsin-sample mixture was done at 37°C for 2 h with continuous gentle shaking. The suspension was then centrifuged at 4800 X g, at temperatures of 4°C for 20 min (Centrifuge Model H-2000C, Shimadzu Corp., Kyoto, Japan). The supernatant was discarded and the residue washed with 15 ml of 0.1M phosphate buffer: pH 7 followed by centrifugation as previously done. The supernatant was again discarded and the residue washed on Whatman's No. 3 filter paper in a Buchner funnel. The filter paper containing the undigested protein residue was folded, placed in a digestion tube and dried for 2h at 80°C.

A blank was prepared and treated in the same way but without the sample. Protein content was determined using method 928.08, AOAC, 1995.

Calculation: Percentage protein digestibility = (A-B)/A

Where: A = % protein in the sample; and B = % protein factor after pepsin digest.

Group B-vitamins (Niacine, Thiamine and Pyridoxine)

A reversed-phase HPLC method by Ekinci and Kadakal (2005), modified from Cho *et al.*, (2000) was used. The sample treatment consisted of solid phase extraction (SPE) with Sep-Pak C18 (500 mg) cartridges that enabled separation of water-soluble vitamins and removed most of the interfering components. Twenty milliliters (20 ml) of water was added to 5 g of the sample. The mixture was homogenized using a homogenizer at medium speed for 1min. The homogenized samples were centrifuged for 10 min at 14×10^3 g (Centrifuge Model H-2000C Shimadzu Corp., Kyoto, Japan). The stationary phase preparation involved flushing with 10 ml methanol and 10 ml water (pH 4.2) to activate it. The homogenized and centrifuged samples were then loaded. The sample was eluted with 5 ml acidified water (pH 4.2) then 10 ml methanol at a flow rate of 1ml min⁻¹. The eluent was collected in a bottle and evaporated to dry. The residue was dissolved in mobile phase and then filtered through 0.45µm pore size filters. Approximately 20 µl of samples was injected into the HPLC column. The column elute was monitored with

a photodiode-array detector at 234 nm for thiamine, 324 nm for pyridoxine, 282 nm and 261 nm for niacin.

The vitamins were analyzed in a HPLC (Model SCL-10A, Shimadzu Corp., Kyoto, Japan) using a column of inertsil ODS 5 μ m 4.6 \times 250 mm 5LI0101Z with 0.1 mol /L KH₂PO₄ (pH 7.0)–methanol, 90:10 mobile phase (filtered through 0.45 μ m membrane and degassed by sonication), flow rate of 0.5 ml/min, a photodiode-array detector (Model Waters 2996, Waters Corp., Mailford, USA), oven temperature of 25°C, and a sample volume of 20 μ l.

Identification of compounds was achieved by comparing their retention times and UV spectra with those of standards stored in a data bank. Five different concentrations of each standard were used to prepare calibration plots for each vitamin. This was done by plotting concentration (μ g/ml) against peak area (mAU). Concentrations of the water-soluble vitamins were calculated from integrated areas of the sample and the corresponding standards.

Vitamin content (mg/g) = (y/b) \times (dilution factor / weight of sample (g) \times 1000)

Where y= is the y intercept of obtained from the standard curve of the vitamin in question, and b is the peak area of the injected sample

Statistical Analysis

Each analysis was carried out in duplicate for two growth seasons and the figures averaged. Data were subjected to the analysis of variance (ANOVA) (Snedecor and Cochran, 1987) using GenStat v.16 and Xlstat v.12. Duncan Multiple Range Test (DMRT) was used to separate the means. Significance was declared at $P \leq 0.05$.

3.0 Results and Discussion

3.1 Proximate Composition of the Sorghum Cultivars

The proximate composition of sorghum flour from the TC regenerants and parents are shown in Table 1.

Moisture content ranged from 6.4 to 8.3mg/100g and did not vary significantly ($p \leq 0.05$) between regenerants and parents of the Mtama 1 and Seredo cultivars. The protein content was found to be in the range of 12.7 to 16.8 mg/100g and varied significantly ($p \leq 0.05$) between regenerants and parents for El Gardam and Mtama 1 cultivars where the regenerants had higher values than the parents. Intercultivar comparison showed lower value for Seredo compared to the rest of the cultivars. The fat content of the flour ranged between 1.4 and 5.5mg/100g. Fat content did not vary significantly between parents and regenerants for Mtama 1 and for parents and regenerant of El Gardam cultivars but was noted to be significantly different ($p \leq 0.05$) within Seredo cultivars where the regenerants showed a higher value than the parents. Seredo regenerants (5.5 mg/100g) had the highest while Mtama parent (1.4 mg/100g) had the lowest. Carbohydrates content ranging from 68 to 73.4 mg/100g and did not vary significantly ($p \leq 0.005$) between regenerants and parents for each cultivars. El Gardam cultivar showed

lower values for intercultivar comparison of the cultivars. Crude fibre content ranged from 2.2 to 3.8mg/100g and did not vary significantly ($p \leq 0.05$) between the parents and regenerants within cultivar except for El Gardam where the parents had higher values.

The ash content was found to range between 1.5% and 2.3% El Gardam parent having the highest and Seredo parent having the lowest. The data obtained showed that the ash content did not vary significantly ($p \leq 0.05$) between regenerants and parents within and across the cultivars.

Table 1: Proximate composition of the sample raw materials (mg/100g) of the sorghum cultivars

Cultivars	Nutrients					
	Moisture %	Protein	Fat	Carbohydrates	Crude Fibre	Ash
El Gar. P	8.3 ^a ±0.1	13.8 ^b ±0.4	3.8 ^{bc} ±0.3	68.0 ^c ±0.6	3.8 ^a ±0.12	2.3 ^a ±0.2
El Gar. R	6.4 ^b ±0.4	16.8 ^a ±0.4	3.1 ^c ±0.2	68.9 ^{bc} ±0.1	2.8 ^c ±0.48	2.1 ^a ±0.5
Mta. 1 P	7.2 ^{ab} ±0.1	12.9 ^c ±0.2	1.4 ^d ±0.5	73.4 ^a ±0.44	3.0 ^{bc} ±0.42	2.1 ^a ±0.6
Mta. 1 R	6.9 ^{ab} ±0.1	14.3 ^b ±0.8	1.7 ^d ±0.2	72.3 ^{ab} ±0.17	3.2 ^b ±0.14	1.8 ^a ±0.1
Seredo P	6.4 ^b ±0.2	13.3 ^c ±0.6	4.6 ^b ±0.4	70.0 ^{ab} ±0.72	2.4 ^d ±0.21	1.5 ^a ±0.3
Seredo R	7.7 ^{ab} ±0.4	12.7 ^c ±0.3	5.5 ^a ±0.2	69.9 ^{abc} ±0.41	2.2 ^d ±0.36	2.1 ^a ±0.1
LSD	1.44	0.65	0.79	3.46	0.29	3.39
C.V%	8.3	1.9	9.7	2.0	4.1	6.9

Values are means (\pm SD). Means sharing a common superscript letter in a column are not significantly different at ($p \leq 0.05$) as assessed by Duncan's multiple range tests. S.D=Standard deviation. LSD= Least significant difference of the mean replicates. El Gar.- El Gardam, Mta. 1- Mtama 1, P-Parent, R-Regenerant

3.2 Mineral Composition

Mineral composition of the sorghum cultivars are shown in Table 2. The mineral compositions of the parents and regenerants of the cultivars were not significantly different ($p \leq 0.05$) except for Zinc in Mtama 1 cultivar and Iron in both El-Gardam and Mtama 1 cultivars. Magnesium was found to range between 0.29-2.02mg/100g, Mtama 1 parents having the lowest with El Gardam parent being the highest. The ranges for the other minerals were Copper 1.24-2.09 mg/100g, zinc 0.16-0.39 mg/100g, sodium 0.16-0.32 mg/100g, calcium 1.68-4.75 mg/100g, potassium 2.49-2.91mg/100g, Manganese 0.23-0.85 mg/100g and iron 2.73-4.08 mg/100g.

Table 2: Minerals composition (mg/100g) of the sorghum cultivars

Cultivars	Mg	Mn	Cu	Zn	Na	Fe	Ca	K
El Gar. P	2.02 ^a ±0.6	0.58 ^a ±0.2	1.35 ^a ±0.2	0.17 ^b ±0	0.25 ^a ±0.1	3.61 ^b ±0.1	4.75 ^a ±1.2	2.70 ^{ab} ±0.8
El Gar. R	1.72 ^a ±0.2	0.85 ^a ±0.1	2.09 ^a ±0.8	0.16 ^b ±0	0.21 ^a ±0	4.08 ^a ±0.2	3.23 ^{ab} ±1.9	2.52 ^b ±1.4
Mta. 1 P	0.29 ^b ±0.1	0.80 ^a ±0.6	1.24 ^a ±1	0.30 ^a ±0.1	0.32 ^a ±0.2	3.93 ^a ±1.2	3.93 ^a ±1.2	2.89 ^a ±0.7
Mta. 1 R	0.61 ^b ±0.3	0.61 ^a ±0.3	2.07 ^a ±0.9	0.16 ^b ±0.1	0.29 ^a ±0.1	3.39 ^c ±1.1	1.99 ^{ab} ±0.5	2.49 ^b ±0.4
Seredo P	0.74 ^b ±0.1	0.57 ^a ±0.1	1.58 ^a ±0.9	0.34 ^a ±0.2	0.16 ^a ±0	2.73 ^d ±0.9	1.68 ^b ±0.7	2.83 ^a ±0.7
Seredo R	0.41 ^b ±0.2	0.23 ^a ±0.1	1.81 ^a ±0.5	0.39 ^a ±0.2	0.16 ^a ±0	2.80 ^d ±1	2.22 ^{ab} ±1.1	2.91 ^a ±1.1
LSD	0.80	0.66	1.27	0.13	0.22	1.90	2.74	2.88
CV%	3.4	4.4	3.1	2.1	3.8	2.3	3.8	4.3

Values are means (± SD). Means sharing a common superscript letter in a column are not significantly different at ($p \leq 0.05$) as assessed by Duncan's multiple range tests. S.D=Standard deviation. LSD= Least significant difference of the mean replicates. El Gar.- El Gardam, Mta. 1- Mtama 1, P-Parent, R-Regenerant.

3.3 Group B-vitamins

Sorghum is an important source of B vitamins except B12. Niacin ranged between 0.65-0.99mg/100g. Mtama 1 parents showed the lowest while El Gardam parents and regenerants had the highest quantities. There were no significant ($p \leq 0.05$) differences in niacin quantities between regenerants and parents for all the cultivar. Thiamine quantities ranged between 0.14-0.94mg/100g with El Gardam parent showing the highest and Mtama 1 parent having the lowest. There were significant ($p \leq 0.05$) differences for thiamine and pyridoxine between the regenerants and parents of Seredo and El Gardam. The vitamins results are shown in Table 3.

Table 3: B-vitamins (mg/100g) in the sorghum cultivars

Cultivars	Niacin	Thiamine	Pyridoxine
El Gar. P	0.99 ^a ±0.3	0.94 ^a ±0.6	0.48 ^a ±0.1
El Gar. R	0.99 ^a ±0.2	0.43 ^b ±0.0	0.15 ^c ±0.0
Mta. 1 P	0.65 ^b ±0.1	0.14 ^c ±0.0	0.16 ^c ±0.0
Mta. 1 R	0.75 ^b ±0.1	0.22 ^c ±0.0	0.14 ^c ±0.02
Seredo R	0.95 ^a ±0.8	0.35 ^b ±0.1	0.48 ^a ±0.14
Seredo P	0.85 ^{ab} ±0.4	0.19 ^c ±0.1	0.25 ^b ±0.02
LSD	0.18	0.10	0.03
C.V%	3.9	10.3	9.9

Values are means (\pm SD). Means sharing a common superscript letter in a column are not significantly different at ($p \leq 0.05$) as assessed by Duncan's multiple range tests. S.D=Standard deviation. LSD= Least significant difference of the mean replicates. El Gar.- El Gardam, Mta. 1- Mtama 1, P-Parent, R-Regenerant.

3.4 Anti-nutrients

Anti-nutritional compounds determine the digestibility of nutrients and the absorption of minerals. Tannin content were not significantly different ($p \leq 0.05$) between the regenerants and parents for each of the cultivar and between the different cultivars. However, tannin content was generally highest in the red sorghum cultivar (Seredo) compared to the white sorghum (Mtama 1 and El Gardam). The regenerants of the Seredo and El Gardam cultivars were noted to have reduced quantities of tannin compared to their parents. There was significant difference ($p \leq 0.05$) of phytate amounts between parents and regenerants of each of the cultivars with the regenerants having low amounts compared to the parents. Phytate amounts were also noted to be generally high in Seredo compared to Mtama 1 and El Gardam and ranges from 124.3 to 351.4mg/100g. The anti-nutrients results for the sorghum varieties are shown on Table 4.

Table 4: Anti-nutrient factors (Tannin and phytates) in the sorghum cultivars

Cultivars	Tannin (%C.E)	Phytates (mg/100g)
El Gar. P	0.81 ^a ±0.1	288.9 ^c ±9.0
El Gar. R	0.70 ^a ±0.04	254.9 ^d ±3.8
Mta. 1 P	0.03 ^b ±0.01	170.0 ^e ±1.0
Mta. 1 R	0.04 ^b ±0.01	124.3 ^f ±5.2
Seredo R	0.88 ^a ±0.2	320.5 ^b ±13.3
Seredo P	2.22 ^a ±0.1	351.4 ^a ±2.6
LSD	2.07	18.11
C.V%	10.9	2.9

Values are means (\pm SD). Means sharing a common superscript letter in a column are not significantly different at ($p \leq 0.05$) as assessed by Duncan's multiple range tests. S.D=Standard deviation. LSD= Least significant difference of the mean replicates. El Gar.- El Gardam, Mta. 1- Mtama 1, P-Parent, R-Regenerant.

3.5 Protein Digestibility

Protein digestibility findings showed a range of 35.3% to 88.4% (Fig. 6). The regenerants within each of the cultivars showed higher percentages of protein digestibility compared to their parents. The lower ranges for protein digestibility were noted in the Seredo cultivars (red sorghum) and they were higher for Mtama 1 and El Gardam (white sorghum).

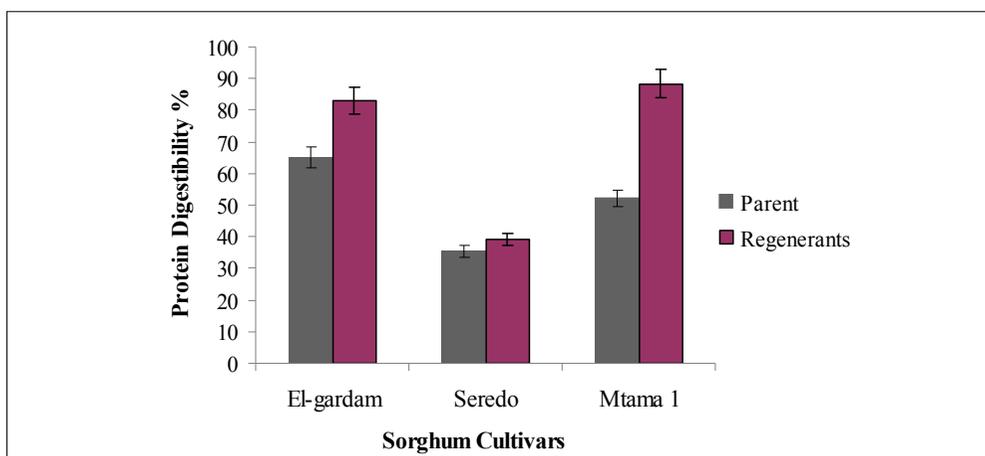


Figure 1: Protein digestibility (%) of the sorghum cultivars

4.0 Discussion

In many parts of the world, sorghum has traditionally been used in food products and various food items. It being particularly adapted to drought prone areas,

attempts are being made to improve wild type cultivars, and there is need to evaluate their nutritional quality. Sorghum has a macromolecular composition similar to that of maize and wheat (Chavan and Kadam, 1989).

The proximate compositions of the regenerants and the parents within each of the sorghum cultivars were not significantly different from each other except for moisture, protein, fats and crude fibre. El Gardama and Mtama 1 regenerants were observed to have significantly higher protein content than the parents. Protein content and composition is reported to vary due to genotype, water availability, temperature, soil fertility and environmental conditions during grain development and is usually 11-13 mg/g but sometimes higher values are reported (David and Dendy, 1995). In this study, the protein content ranged between 12.7-16.8 mg/g with El Gardam parents having the highest while Seredo regenerants had the lowest.

Fat content as a source of energy were high for the Seredo cultivar compared to the El-Gardam and Mtama 1 cultivars. Seredo regenerants (5.5 mg/100g) had the highest while Mtama 1 parent (1.4 mg/100g) had the lowest. Other studies have recorded an average of 3 mg/100g fat content (FAO, 1995) and the finding of this study fall in the same ranges. Sorghum is also good source of carbohydrates and fibre, mainly the insoluble fibre. The insoluble dietary fibre of sorghum may decrease transit time and prevent gastrointestinal problems. The finding of this study showed no significant differences in carbohydrate content within the cultivars. Crude fiber content for El Gardam and Seredo parents were observed to be significantly higher than the regenerants. This may be attributed to variation in the genotype. The values obtained for carbohydrates and fibre contents were within the range reported by Noha *et al.* (2001).

The ash content is a measure of the total amount of minerals present within a food, whereas the mineral content is a measure of the amount of specific inorganic components present within a food, such as Ca, Na, K and Cl. The ash and mineral content ranges obtained were confirmed in previous work (Noha *et al.*, 2001). The ash content within each of the cultivars did not vary significantly. The generally lower amount of magnesium in Seredo cultivar may be because divalent cations such as magnesium may be present as mineral phytate chelates (Mamiro *et al.*, 2001). Bioavailability of iron in sorghum for human subjects was found to be affected more by phytic acid than by the tannin content of the grains (Radhakrishnan and Sivaprasad, 1980).

Sorghum is an important source of B vitamins except B 12. The vitamin contents in this study were in the same ranges as those obtained by Mamoudou *et al.* 2006. El Gardam parents showed significantly higher amount than the regenerants for thiamine and pyridoxine. Seredo regenerants also showed significantly higher amount of pyridoxine and riboflavin than the parents did. The B vitamins and minerals are concentrated in the aleurone layer and germ. Removal of these

tissues by decortication produces a refined sorghum product that has lost part of these important nutrients. Vitamins are also reported to be unstable and can be lost during processing and storage (Ekinci and Kadakal, 2005).

Sorghum, like legume and oil seed meals has some limitations, due to the presence of anti-nutritional factors, such as trypsin and amylase inhibitors, phytic acid and tannin. The tannin values in this study were similar to those reported from similar studies (Beta *et al.*, 1999; Noha *et al.*, 2001). These compounds are known to interfere with protein, carbohydrates and mineral metabolism. The tannin and phytates content of dark grains is always higher than that of pale grains (Dicko *et al.*, 2006a). Red sorghum genetically has higher condensed tannin compared to white sorghum. This was confirmed in this study where the red Seredo cultivars showed higher values of tannin and phytates content compared to the white Mtama 1 and El Gardam cultivars. Tannin content did not vary significantly between parents and regenerants within each cultivar although the amount in Mtama 1 cultivar was observed to be significantly lower compared to El Gardam and Seredo cultivars. The tannin content of grain is always higher before ripening than after ripening. Tannins impart a bitter taste to the grains making them unpalatable and interfere with protein digestibility (Dicko *et al.*, 2006a). The parents were observed to have significantly higher amounts of Phytates than the regenerants within all the cultivars although the Seredo cultivar generally had the highest amount compared to the other cultivars. Higher ranges of protein digestibility for the regenerants than the parents within all the cultivars may be attributed to the Low ranges of phytates realized in this research since phytates are known to hinder digestibility of proteins. Generally, low ranges of protein digestibility in Seredo may be attributed to their high anti-nutrient content compared to the rest of the cultivars. *In vitro* studies and *in vivo* studies with livestock and laboratory animals indicate that sorghum proteins are generally less digestible than those of other cereals (Muindi and Thomke, 1981). Phytates and polyphenols such as tannins bind to both exogenous and endogenous proteins including enzymes of the digestive tract, affecting the utilization of proteins (Griffths, 1985; Asquith and Butler, 1986).

5.0 Conclusion

Variability was shown in the anti-nutrient content with Seredo showing elevated amounts than El Gardam and Mtama 1 cultivars. Mtama 1 regenerants with low anti-nutrient could be recommended for ASALs with respect nutrient availability since anti-nutrients in sorghum are shown to impair the bioavailability of the other nutrients to the body. However, methods such as fermentation, germination and fortification, as has been established in other studies by Dicko *et al.*, 2005 and Beta *et al.*, 1999, can be used to reduce the high amounts anti-nutrients in the El-Gardam and Seredo cultivars to increase their nutrient availability.

References

- Association of Official Analytical Chemists (AOAC). (1995). Official methods of analysis of AOAC international. In: Ralph H. N. (Eds). Cereal foods. AOAC international suite 500,481. Gaithersburg, Maryland 20877-2417 USA, **2(16)**, pp. 1-43.
- Beta T., Rooney L. W., Marovatsanga L. T. and Taylor J. N. (1999). Phenolic compounds and kernel characteristics of Zimbabwean sorghums. *Journal of Science and Food Agriculture*, **79**, pp. 1003-1010.
- Burns R. E. (1963). Methods of tannin analysis for forage crop evaluation. Georgia Agricultural experiment station technical bulletin, **32**, pp.1-14.
- BSTID-NRC (Board on Science and Technology for International Development-National Research Council) (1996). *Lost crops of Africa*. Academic Press, Washington DC, pp. 127-213
- Camire A. L. and Clydesdale M. (1982). Analysis of phytic acid in food by HPLC. *Journal of Food Science*, **47**, pp.576.
- Chavan J. K. and Kadam S. S. (1989). Critical reviews in food science and nutrition. *Journal of Food Science*, **28**, pp.348-400.
- Cho C. M., Ko J. H. and Cheong W. J. (2000). The effect of fermentation and drying on the water-soluble vitamin content of tahara, a traditional Turkish food. *Talanta*, **51**, pp. 799.
- David A. and Dendy V. (1995). *Sorghum and Millets Chemistry and Technology*. American Association of Cereal Chemists Press, Minnesota.
- Dicko M. H., Gruppen H., Traore A.S., Van Berkel W.J.H. and Voragen A.G.J. (2005). Evaluation of the effect of germination on phenolic compounds and antioxidant activities in sorghum varieties. *Journal of Science and Food Agriculture*, **53**, pp. 2581-2588.
- Dowling L.F., Arndt C. and Hamker B.R. (2002). Economic viability of high digestibility sorghum as food for market broiler. *Agronomy Journal*, **94**, pp.1050-1058.
- Ebadi M. R., Pourreza J., Jamalia J. E., Samie A. H. and Mirhadi S. A. (2005). Amino acid content and availability in low, medium and high tannin sorghum in grain for poultry. *International Journal Poultry Science*, **1**, pp. 27-31.
- Ekinci R. and Kadakal C. (2005). Determination of seven water-soluble vitamins in Tahara, a traditional Turkish cereal food, by high-performance liquid chromatography. *ACTA chromatographica*, **15**, pp. 121-134.
- Food and Agriculture Organization (FAO). (2005). Sorghums and millets in human nutrition. <http://faostat.fao.org/faostat/>. Date accessed December 21, 2010.

- Griffiths D. W. (1985). The inhibition of digestive enzymes by polyphenolic compounds. *Experimental Biology and Medicine*, **199**, pp. 504-516.
- Makobe M. N., Kahangi E. M., Misra A. K. and Imbuga M. O. (2006). Development of hardy sorghum cultivars for arid and semi-arid regions. *African Crop Science Journal*. **14 (4)**, pp. 297-309.
- Mamiro P. R. S., Van camp J., Mwikya S. M. and Huyghebaert A. (2001). In vitro extractability of calcium, iron and zinc in finger millet and kidney beans during processing. *Journal of Food Science*, **66**, pp. 1271-1275.
- Mamoudou H. D., Harry G., Alfred S. T., Alphons G. J. V., and Willem J. H. B. (2006). Sorghum grain as human food in Africa: relevance of content of starch and amylase activities. *African Journal of Biotechnology*, **5(5)**, pp. 384-395.
- Mertz E. T., John D. A., Kirleis A. W., Hassen M. M., Nora D. M. and Lars M. (1984). Digestibility of sorghum proteins. *Applied Biology*, **78(3)**, pp. 1333-1335.
- Muindi P. J. and Thomke S. (1981). The nutritive value for rats of high- and low-tannin sorghums treated with magadi soda. *Journal of Science Food and Agriculture*. **32**, pp. 139-145.
- Noha A. M. and Elfadil E. B. (2001). Nutritional Evaluation of Sorghum Flour (*Sorghum bicolor* L. Moench) During Processing of Injera. *International Journal of Biological and Life Sciences*, **6**, pp.2010.
- O'Kennedy M. M., Grootboom A. and Shewry P. R. (2006). Harnessing sorghum and millet biotechnology for food and health. *Journal of Cereal Science*. **44**, pp.224-235.
- Price M. L., Van Scoyoc S. W., and Butler L. G. (1978). A critical evaluation of the vanillin reactions as an assay for tannin in sorghum grain. *Journal of Agriculture and Food Chemistry*, **26**, pp. 1214-1218.
- Radhakrishnan M. R. and Sivaprasad J. (1980). Tannin content of sorghum varieties and their role in iron bioavailability. *Journal of Agriculture and Food Chemistry*, **28**: pp. 55-57.
- Snedecor G.W. and Cochran W. G., (1987). *Statistical methods*. Iowa state University Press, Iowa.
- Tingay S., McElroy D., Kalla R., Fieg S., Wang M., Thornton S. and Brettell R. (1997). Agrobacterium tumefaciens-mediated barley transformation. *The Plant Journal*, **11**, pp. 1369-1376.
- Vohra P., Gray G. A. and Kratzer F. H. (1988). Phytic acid-metal complexes. *Society for Experimental Biology and Medicine*, **120**, pp. 447-449.