
COUNCIL FOR SCIENTIFIC & INDUSTRIAL RESEARCH

FOOD RESEARCH INSTITUTE



BREWING AND MALTING CHARACTERISTICS OF SELECTED SORGHUM VARIETIES FROM NORTHERN GHANA

by

Gayin, J., Manful, J.T. and Toku, P.

Food Research Institute, CSIR, Ghana

September, 2005

Table of Contents

	Summary-----	2
	List of Tables-----	3
1.0	Introduction-----	5
2.0	Materials and Methods-----	6
2.1	Physical Quality Evaluation-----	6
2.2	1000 grain Weight -----	6
2.3	%Moisture -----	6
2.4	Chemical Analysis-----	7
2.4.1	Aflatoxins -----	7
2.4.2	Crude fat and Nitrogen -----	7
2.5	Microbiological Analysis -----	7
2.6	Determination of Germination Capacity-----	7
2.7	Sorghum Malt -----	8
2.8	Malt flour -----	8
2.9	Alpha-Amylase-----	8
2.9.1	Principle-----	9
2.9.2	Assay Procedure-----	9
2.9.3	Calculation of Activity-----	10
3.0	Results and Discussion-----	11
4.0	Conclusion-----	14
	Appendix 1-----	15

List of Tables

Summary

Eight identified land races of sorghum suspected to have similar brewing

Table 1: Some physical quality characteristics of 8 sorghum varieties They

Table 2: Aflatoxin detection levels of 8 sorghum varieties They

Table 3: Mould/yeast counts & dominant flora of 8 sorghum varieties

Table 4: Germination count & alpha-amylase activity of 8 sorghum varieties were

examined. The level of malt α -amylase activity was also assessed. The moisture contents of all the samples fell within the desired range of 9.0 to 10.0% for whole sorghum brewing. Weevil damaged kernels were detected in four of the samples. YAAK/14/01, MANKAROGA and SBI SORSORG-E11 had greater weevil damaged kernels than the desired value of 4.0%. SBI SORSORG-E11 had the highest germination with 90.0% and YAAK/14/01 having the least. The total aflatoxin levels found were between 0.00% and 2.18%. For most of the varieties the various forms of aflatoxins were not detected. The *Fraifa*, *YaaKpaaji* and *Mankaroga* varieties did not show encouraging germination counts and α -amylase activity. The high values of germination and α -amylase indicate that the "Sorsorg" varieties are comparable to *Kapaaki* in terms of mashing properties. *Mankaroga*, *YaaKpaaji* and *Fraifa* show poor mashing and brewing properties, however it is recommended these three "Sorsorg" varieties go through further analysis to ascertain their mashing and brewing potentials.

Summary

Some identified land races of sorghum suspected to have similar brewing properties to *Kapaala* were studied for their malting characteristics. They were *Frafra*, *Yaakpaaji*, *Mankaraga*, *Kapaala*, *SBL9-Sarsorg-E8*, *Sarsorg-M 16*, *SBL2-Sarsorg-E11* and *Belkowige*.

Their physical and chemical qualities and germination capacities were determined. The level of malt α -amylase activity was also assessed. The moisture contents of all the samples fell within the desired range of 9.0% - 11.0% for whole sorghum brewing. Weevil damaged kernels were detected in four of the samples. *YAAKPAAJI*, *MANKARAGA* and *SBL2SARSORG-E11* had greater weevil damaged kernels than the desired range of <2%. *SBL2SARSORG-E11* had the biggest grain size with *BELKOWIGE* having the least. The total nitrogen levels found were between 1.68% and 2.18%. For most of the varieties the various forms of aflatoxins were not detected. The *Frafra*, *Yaakpaaji* and *Mankaraga* varieties did not show encouraging germination counts and alpha-amylase activity. The high values of germination and α -amylase indicate that all the "Sarsorg" varieties are comparable to *kapaala* in terms of malting properties. *Mankaraga*, *Yaakpaji* and *Frafra* showed poor malting and brewing properties. However it is recommended these three "Sarsorg" varieties go through further analysis to ascertain their mashing and brewing potentials.

1.0 Introduction

Malt is cereal grain, usually barley, which has been germinated for a limited period of time, and then dried. Several cereals can be satisfactorily malted but barley usually gives the least technical difficulty. For the production of African native beers, a variety of cereals especially sorghum are malted. More often at a time that it is considered appropriate, the Malster arrests germination by drying the grain in a stream of warm air. During germination, the food store or the endosperm of the grain is partly degraded by enzymes that attack cell walls, starch granules and the protein matrix. Essentially malt is therefore the degraded endosperm and the attendant enzymes able to complete the degradation. When relatively cool air is used for drying, the malt is pale in colour and very rich in enzymes. With greater drying temperatures, especially early in the drying process, the malt is darker in colour and the enzymic content is depleted.

At a review meeting of all AgSSIP Sorghum and Millet projects in March, 2005, it has come up that some identified land races of sorghum may have similar brewing properties comparable to well known varieties like *Kapaala*. The objective of this work is to investigate the malting characteristics of eight sorghum varieties.

2.1 %Moisture

An electronic moisture analyzer (Sartorius MA 45) was used to find the precise moisture content of the air-dried flour. The moisture content was taken by first positioning the sample pan and then taring the analyzer. The sample was then spread evenly on the pan and the drying program

2.0 Materials and Methods

Eight (8) Sorghum varieties were received from Dr. I.D.K Atokple of the Savanna Agricultural Research Institute, Tamale. The varieties were;

- Frafra,
- Yaakpaaji
- Mankaraga
- Kapaala
- SBL9-Sarsorg-E8
- Sarsorg-M 16
- SBL2-Sarsorg-E11
- Belkowitz

2.1 Physical Quality Evaluation

Sample divider was used to sample a known weight of each of the samples for physical quality evaluation. Parameters looked out for included level of broken, organic matter, immature grains, stones and weevil damaged grains.

2.2 1000g Weight

One thousand whole and healthy grains of each sample were manually counted and weighed on an analytical balance

2.3 %Moisture

An electronic moisture analyzer (Sartorius MA 45) was used to find the precise moisture content of the sorghum flour. The moisture content was taken by first positioning the sample can and then tarring the analyzer. 2g sample was then spread evenly on the can and the drying program started.

2.4 CHEMICAL ANALYSIS

2.4.1 Aflatoxins

Quantitative analysis of aflatoxins was based on Pons W.A. (1979) JAOAC 62, 586-594. The detection limits of the procedure used are:

Aflatoxin B₁ and B₂ ⇒ 0.04µg/kg

Aflatoxin G₁ and G₂ ⇒ 0.06µg/kg

2.4.2 Crude Fat & Nitrogen

Crude fat was analyzed using AOAC method 984.13 (2000) 17th Edition whilst Nitrogen was by AOAC method 920.39 C (2000) 17th Edition.

2.5 Microbiological Analysis

Moulds and yeast count was done using International Standards Organization (ISO) Method 7954 (1999).

2.6 Determination of Germination Capacity

Germination test was carried out by selecting seeds at random and cleaned. Unfilled seeds were discarded. 200 grains of each of the sorghum varieties were counted and weighed. The grains were washed and rinsed with distilled water. The grains were steeped for 24 hours in water, the water was drained and the grains evenly distributed in a compact single layer on petri dishes over laid with moistened filter paper. The petri dishes were covered and placed in the dark at room temperature for germination. Moisture supply was maintained by occasionally adding drops of water to

the filter paper. After 72 and 120 hours, germinated seeds considered to be viable were counted and presented as percentage viable seed.

2.7 Sorghum Malts

In all the malting procedure the first step was steeping the grains in 0.1N $\text{Ca}(\text{OH})_2$ for 8 hours after which grains were drained and washed with water. A period of 4 hours was allowed for air rest, followed by a second steeping in 0.5% formaldehyde for 8 hours. The grains were drained and washed with water before germination at 30°C for five days. Kilning was carried out at 55°C for 24 h.

2.8 Malt flours

20g sample of malt was milled and passed through a 0.5mm screen. 0.5g malt flour was then accurately weighed into a 100ml volumetric flask. To the volumetric flask solutions of 1% sodium chloride plus 0.02% calcium chloride plus 0.02% sodium azide was added and adjusted to volume. About 15-20minutes was allowed for the enzyme to extract at room temperature, with occasional stirring. An aliquot of the solution was filtered through a Whatman GF/A glass fibre filter paper, or centrifuged at 1,000g for ten minutes. 0.5ml of the filtrate was then diluted with 9.5ml of Extraction Buffer Solution. Activity was assayed within two hours, (Megazyme Alpha-Amylase Assay Procedure, ICC Standard No. 303).

2.9 Alpha-Amylase

The level of endogenous α -amylase in cereal grains and products significantly affects the industrial exploitation of these commodities. In the brewing industry, the level of malt α -amylase is a key quality parameter.

2.9.1 Principle

The Ceralpha procedure (employing Amylase HR reagent) for the assay of α -amylase, employs as substrate, the defined oligosaccharide "non-reducing-end blocked p-nitrophenyl maltoheptaoside" (BPNPG7) in the presence of excess levels of a thermo stable α -glucosidase. (which has no action on the native substrate due to the presence of the "blocking group"). On hydrolysis of the oligosaccharide by endo-acting α -amylase, the excess quantities of α -glucosidase present in the mixture give instantaneous and quantitative hydrolysis of the p-nitrophenyl maltosaccharide fragment to glucose and free p-nitrophenol. The assay format is shown in Appendix 1.

Essentially, an aliquot of a cereal flour extract or fermentation broth is incubated with substrate mixture under defined conditions, and the reaction is terminated (and colour developed) by the addition of a weak alkaline solution. The absorbance at 400nm is measured and this relates directly to the level of α -amylase in the sample analyzed. A standard error of less than 5% is achieved routinely and the assay is absolutely specific for α -amylase.

2.8.2 Assay Procedure

0.2ml aliquots of Amylase HR Reagent solution (unbuffered) were dispensed into glass test tubes and pre-incubated the tubes and contents at 40°C for 5 minutes. The extract was pre-incubated at 40°C for 5 minutes. To each tube containing Amylase HR Reagent solution (0.2ml) 0.2ml aliquots of pre-equilibrated malt extract was added directly to the bottom of the tube. The mix was then incubated at 40°C for exactly

20 minutes (from time of addition). At the end of the 20 minutes incubation period, exactly 3ml of stopping reagent was added and contents stirred vigorously. The absorbance of the solutions and the reaction blank were read at 400nm against distilled water, (Megazyme Alpha-Amylase Assay Procedure, ICC Standard No. 303).

2.9.3 Calculation of Activity

One Unit of activity is defined as the amount of enzyme, in the presence of excess thermo-stable α -glucosidase, required to release one micromole of p-nitro-phenol from BPNPG7 in one minute under the defined assay conditions, and is termed a **Ceralpha Unit**.

For Malt:

Units (CU)/g of milled malt:

$$= \frac{\Delta E_{400}}{10} \times \frac{3.4}{0.2} \times \frac{1}{18.1} \times \frac{100}{0.5} \times 20$$

$$= \Delta E_{400} \times 376$$

Where:

ΔE_{400} = Absorbance (reaction) - Absorbance (blank)

Incubation Time = 10 minutes

Total Volume in Cell = 3.4ml

Aliquot Assayed = 0.2ml

ϵ_{mM} of p-nitrophenol (at 400nm) in 1% tri-sodium phosphate =

3.0 RESULTS AND DISCUSSION

Physical Qualities

Table1: Some physical quality characteristics of 8 sorghum varieties

Sample	1000- kernel wt	%Moisture	%brokens	%weevil damaged	%immature
FRAFRA	25.15	8.70	0.00	0.00	0.00
YAAKPAAJI	26.21	8.75	0.00	2.13	0.00
MANKARAGA	26.23	8.68	0.00	2.64	0.00
KAPAALA	27.48	10.79	0.00	0.46	0.00
SBL9SARSORG-E8	29.28	9.28	1.36	1.31	0.14
SBL2SARSORG-E11	30.34	8.41	0.00	10.5	0.00
SARSORG M 16	28.47	9.31	0.05	0.00	0.00
BELKO WIGE	24.21	8.17	0.00	0.00	0.00

All the varieties were matured and mostly whole grains. Apart from *SBL9SARSORG-E8* which had 1.36% broken kernels all the other varieties had no brokens. In spite of this, the 1.36% still falls within acceptable level of broken kernels (<2%) for whole sorghum brewing. Moisture content of all varieties was below 10%, which is a good moisture level for grain storage. The moisture contents of all the samples fell within the desired range of 9.0% - 11.0% for whole sorghum brewing. Maximum recommended storage moisture contents for aerated shelled sorghum grain to be stored for up to 1 year is 14.0% or 13.0% if it is to be stored more than 1 year. Moisture levels below 12 to 13 percent will prevent mould formation.

Weevil damaged kernels were detected in four of the samples. However the level of 10.5% for *SBL2SARSORG-E11* was too high. This suggests

that sample may not have been stored under prescribed conditions or may have had some problems in the field. *YAAKPAAJI*, *MANKARAGA* and *SBL2SARSORG-E11* had greater weevil damaged kernels than the desired range of <2%. *SBL2SARSORG-E11* had the biggest grain size with *BELKO WIGE* having the least. The total nitrogen levels found were between 1.68% and 2.18%. The operational range for sorghum is from 1.5% to 2.2. Fat levels of less than 4% which is within operational rang was found in all eight samples. For most of the varieties the various forms of aflatoxins were not detected. Aflatoxins are poisonous, carcinogenic byproducts of the growth of several species of the mould fungus *Aspergillus*. These byproducts are produced as the fungi grow in feed grains, processed feed and food products.

The fungi (mold) that produce aflatoxins can infect important food and feed crops before, during and after harvest. These fungi, especially *Aspergillus flavus* and *Aspergillus parasiticus*, are normal soil-borne inhabitants in our environment, growing on both living and decaying plant matter. The major products in which aflatoxins are produced include corn grain, soybeans, dry beans, cottonseed, grain sorghum, wheat, peanuts and tree seeds. Four different aflatoxins, B1, B2, G1 and G2, have been identified with B1 being the most toxic, carcinogenic and most prevalent. In a few cases where there was detection the levels were very low to pose any potential threat to health. For human consumption, a level, below 20 ppb is enforced (Mirocha and Christensen 1982 and Pitt, 1989b).

SARSORG M 16

7.2×10^3

Fisarium spp.
Aspergillus spp

BELKO WIGE

1.1×10^4

Fisarium spp.

Table 2: Aflatoxin detection levels of 8 sorghum varieties

variety	N ₂ (g/100g)	Fat (g/100g)	Aflatoxins (µg/kg)				Total Aflatoxins
			B ₁	B ₂	G ₁	G ₂	
Frafra	1.77	1.65	ND*	ND	ND	ND	N/D ⁺
Yaakpaaji	1.88	1.13	1.14	0.05	ND	ND	1.19
Mankaraga	1.92	1.90	0.3	ND	ND	ND	0.30
Kapaala	1.68	3.23	ND	ND	ND	ND	N/D
SBL9-Sarsorg-E8	2.04	2.78	ND	ND	ND	ND	N/D
Sarsorg-M 16	1.91	3.55	ND	ND	ND	ND	N/D
SBL2-Sarsorg-E11	2.12	3.76	ND	ND	ND	ND	N/D
Belkowitz	2.18	3.43	0.04	ND	ND	ND	0.04

ND* - Not detected

N/D⁺ - None detected

Detection Limits of procedure used are: Aflatoxin B₁ and B₂ = 0.04µg/kg, Aflatoxin G₁ and G₂ = 0.06µg/kg

Table 3: mould/yeast counts & dominant flora of 8 sorghum varieties

Sample	Mould and Yeast Count (cfu/g)	Dominant Flora
FRAFRA	2.6x10 ⁴	<i>Fusarium</i> spp.
YAAKPAAJI	9x10 ³	<i>Fusarium</i> spp. <i>Fusarium</i> spp. <i>Penicillium</i> spp.
MANKARAGA	9x10 ³	<i>Aspergillus</i> spp
KAPAALA	950	<i>Fusarium</i> spp. <i>Fusarium</i> spp.
SBL9SARSORG-E8	4.9x10 ³	<i>Aspergillus</i> spp
SBL2SARSORG-E11	7x10 ³	<i>Fusarium</i> spp. <i>Fusarium</i> spp.
SARSORG M 16	2.2x10 ³	<i>Aspergillus</i> spp
BELKO WIGE	3.3x10 ³	<i>Fusarium</i> spp.

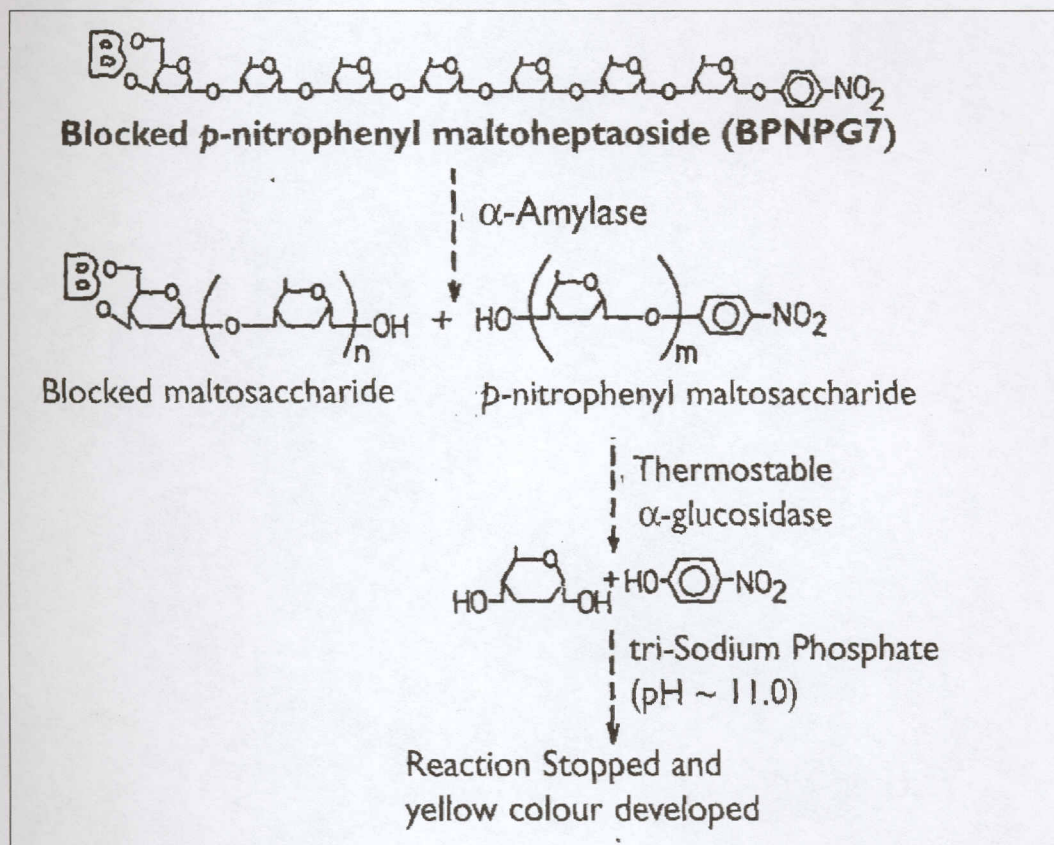
Table 4: Germination count & alpha-amylase activity of 8 sorghum varieties

Sample	Germination count (%)	α -Amylase DU (dry wt.)
FRAFRA	6	1
YAAKPAAJI	5	4
MANKARAGA	36	14
KAPAALA	89	39
SBL9SARSORG-E8	89	48
SBL2SARSORG-E11	90	46
SARSORG M 16	89	36
BELKO WIGE	60	28

The Frafra, Yaakpaaji and Mankaraga varieties did not show encouraging germination counts and alpha-amylase activity. The high values of germination and α -amylase indicate that all the "Sarsorg" varieties are comparable to kapaala in terms of malting properties. Belkowige showed a bit of promising results, but the rest of the varieties namely mankaraga, Yaakpaji and Frafra showed poor malting and brewing properties.

4.0 Conclusion

The *SBL9SARSORG-E8*, *SBL2SARSORG-E11*, *SARSORG M 16* varieties showed encouraging germination counts and Alpha-Amylase activity. The high values of germination and α -amylase activity indicate that the two varieties are comparable to *kapaala* in terms of potential malting properties. However it is recommended these three "Sarsorg" varieties go through further analysis to ascertain their mashing and brewing potentials.



Scheme 1: Theoretical basis of the Ceralpha α -amylase assay procedure

Immediately α -amylase cleaves a bond within the blocked p-nitrophenyl maltosaccharide substrate, the non-blocked reaction product containing the p-nitrophenyl substituent is instantly cleaved to glucose and free p-nitrophenol by the excess quantities of thermo stable α -glucosidase which are integral parts of the substrate mixture, and free p-nitrophenol is released. The reaction is terminated and the phenolate colour is developed on addition of tri-sodium phosphate (pH ~11.0).