

## EFFECT OF ENZYME HYDROLYSIS ON SACCHARIFICATION AND CONSUMER ACCEPTABILITY OF BANANA FIG "MALT" DRINK

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### ABSTRACT

Production of low-glycemic malt flavoured drink from non-cereal base using amyloglucosidase was carried out in this study. Banana fig was used as a replacement for malted barley. The effects of mashing methods on pH, Total Titratable acidity (TTA), sugar content, specific gravity, saccharification time as well as the yield of the extracts were investigated. The resulting drinks were evaluated using 20-member panel on a 7-point Hedonic scale. The pH of the banana fig extract decreased significantly ( $p=0.05$ ) as the mashing temperature increased. It was 5.04, 4.97 and 4.95 against mashing temperatures of 45, 65 and 80°C respectively. The %TTA did not follow a reverse trend with the pH. The specific gravity of all the "must" samples decreased ( $p>0.05$ ) with increasing mashing temperatures. It decreased from 1.013 to 1.010 against temperatures of 45 and 80°C respectively. There was a significant ( $p>0.05$ ) decrease in % sugar content of the extract as the temperature of mashing increased. Generally the sugar content of banana fig drinks ranged from 6.2 to 11.8% while the commercial malt samples had a range of 11.3 to 14%. One of the mashing methods designated as "temperature programme mashing" (TPM) method gave the highest ( $p>0.05$ ) extract yield. The saccharification of banana fig worts were partial, but introduction of exogenous enzyme resulted in their complete saccharification within 10min. There was no significant ( $p<0.05$ ) difference in taste, and flavour/aroma of all the hydrolysed banana fig samples compared to the reference sample (Amstel malt). But one of the samples designated, as BS3 was significantly different from others in terms of colour and general acceptability. Banana fig beverage proved a good substitute for the conventional high-sugar malt drinks in our market today.

**KEY WORDS:** Banana fig, Barley malt, Glycemic, amyloglucosidase, mashing

### INTRODUCTION

Malt is a product made by limited germination of cereal grains, mostly barley, followed by drying of the grain. Malt drink is a non-alcoholic, food drink. In Nigeria malt drinks are chiefly made from malted barley (Chukwurah, 1988). Conventionally malt drink production involves the use of similar raw materials, machinery and procedure as in beer brewing (Rooney, 1969). However the malt drinks are reported to be wholesome and more nutritious than beer (Okon and Akpanyung, 2005) and there are more potential customers for malt drink than beer in view of its non-alcoholic value (Jepsen, 1993).

Nevertheless, malt beverage contain substantial amount of reducing sugar (glucose) derived from the enzymic hydrolysis of starchy raw materials (barley, sorghum, and maize) during mashing (Okon and Akpanyung, 2005). Again, in most cases sucrose is added in the formulation of these malted beverages for taste purposes; giving rise to high sugar level, up to 14°Brix (Okon and Akpanyung, 2005). We do know, from recent research reports (Jenkins 2002; Miller, 2002; FAO, 1997) that high sugar products are not good for all consumers. Some consumers, for health purposes, deny themselves the luxury of vitamins and minerals found in conventional malt drink simply because of its high glycemic value. Much will be achieved if a low glycemic "malt" drink could be produced without sacrificing the taste, body and aroma of the conventional malt. Adjustment of the mashing process may be necessary in order to obtain a good wort from banana fig. This study was designed to produce a low sugar (low-glycemic) "malt" drink from banana (*Musa Sapientum*) using amyloglucosidase hydrolysis. The acceptability of the low sugar "malt" drink as an alternative for high sugar malt was also investigated. The choice of banana as a replacement for barley is because of the following reasons: (i) Overripe banana is readily available (ii) Overripe banana constitute postharvest loss. (iii) Over ripe banana does not contain as much starch as barley (iv) There is an assumption that banana will help to increase the body (bulk) of the drink due to its high content of soluble

solids.

### MATERIALS AND METHODS

#### Procurement of Material

Ripe and over-ripe banana, and local hop substitute (alfalfa) were sourced from a local market in Owerri, Nigeria, while industrial yeast (amyloglucosidase), acetic acid, caramel, malt extract, and polyanionisches cellulose polymer, ANTISOL<sup>®</sup> were collected from Food Science and Technology, Federal University of Science and Technology, Owerri, Nigeria. Barley malt was collected from Nigerian Breweries, Aba, a commercial brewing company

#### Banana fig preparation

Fully ripe and over-ripe banana fruits were washed with tap water and hand peeled separately. The peeled bananas were diced into round cuts of 2cm thickness with sharp kitchen knife and dried between 50-60°C for 72 hr to moisture content of 17% in a fan-driven oven (Hot Box Gallenkamp, England). Dark-brown leathery palates called fig, were obtained at the end of drying. The figs were stored in an air-tight container until used.

#### Extraction of wort

The leathery-dark brown fig obtained after drying was milled to a smaller particle sizes using manual grinder to obtain "grist". Four mashing methods including temperature programmed mashing (TPM) method described by (Briggs, 1998), and infusion method at three different temperatures (45, 70 and 80°C) were used. For the TPM method, 200ml distilled water at 45°C was added to 50g of milled banana fig and barley malt respectively. The mash obtained was stirred continually at 45°C for 30min. At the expiration of the 30min, the temperature of the mash was increased at the rate of 1°C/min for 25min until it reached 70°C. 100ml distilled water at 70°C was again added to the mash and the temperature of the mash was maintained at 70°C for 1hr. During this period,

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he saccharification time was determined using iodine reagent. The mash was then cooled after the expiration of the 1hr and the stirrer was rinsed into the mash. The mash weight was adjusted to 450g by the addition of distilled water. After this the mash was filtered to obtain the extract using a combination of filter cloth and paper. For the infusion method, the same ratio of water addition was used as TPM but the mixtures were left at the specified temperatures for 60min. Later, amyloglucosidase enzyme was introduced during mashing, but only TPM was used in this case. The worts were treated with amyloglucosidase to complete starch breakdown. 450g of each wort sample were adjusted to pH of 4.5 by addition of few drops of acetic acid. Few drops of the enzyme concentrate was added to the wort at temperature of 60°C and stirred for 30min for the enzyme to hydrolyse the starch. At intervals of 2 min, the saccharification time was measured by adding few drops of iodine reagent on the small quantity of the sample. The disappearance of the black blue colouration marked the end of complete breakdown of the starch in the samples and the time when this happened was noted.

#### Formulation of low-glycemic "malt" drink .

Overripe banana fig extracts treated with amyloglucosidase was used. Three products including BS1, BS2, BS3 were obtained. Overripe fig extract was chosen based on the result of chemical analysis carried out on the extract before final product formulation. After the blending, the products were filtered using a combination of filter paper and muslin cloth, and then filled into 33cl bottle. The bottled drinks were pasteurized at 65°C for 15min and cooled before storing in the refrigerator prior to final product analysis and sensory evaluation.

#### Chemical Analysis of Extract.

The worts were analyzed for pH, percentage total titratable acidity (TTA), Specific gravity, sugar content, Extract yield and saccharification time. The method of Association of Official and Analytical Chemists (AOAC, 1990) was used in the determination of TTA. Extract yield was determined following the method developed by the Institute of Brewing (IoB, 1977) while the saccharification time was evaluated following the

European Brewing Convention method (EBC, 1987). The % sugar was determined using hand refractometer, while specific gravity was determined by the method described by Anderson, (1970). Digital (Jenway, England) pH meter was used to monitor the pH of the samples.

#### Sensory evaluation

Sensory evaluation was carried on the three samples of formulated low-glycemic "malt" drink with commercial malt drink, Amstel Malta, as control. The evaluation was based on quality parameters such as colour, taste, flavour/aroma and general acceptance. A 7-point hedonic scale and twenty (20) panelist comprising of ten males and females each, some having basic knowledge in brewing science and technology were used. The experiment was a two-factor treatment in a randomized complete block design having product x3 and gender of judges x2 as sources of variation. Data collected were analyzed using the method of a two-way analysis of variance (ANOVA) procedure described by Mahony, (1986).

#### RESULTS AND DISCUSSION

Result of chemical analysis of barley and banana fig extract is shown in Table 1. The pH of banana fig decreased ( $p=0.05$ ) as the mashing temperature increased. It was 5.04, 4.97 and 4.95 against temperatures of 45, 65 and 80 °C respectively. The decrease in the pH values as the mashing temperatures increased might have been probably as a result of low rate of production of sugary extract in the mashed samples due to enzyme inactivation as the mashing temperature increased (Briggs, 1998). This implied that the mashed samples had more of acidic component with higher mashing temperature. The pH of the barley extract remained somehow constant between the mashing temperatures. The percentage total titratable acidity of the ripe banana fig extract did not follow a reverse trend with that of pH (Table 1). A similar trend was observed for barley wort. The %TTA of the overripe banana fig had the lowest % TTA of 0.05 against mashing temperature of 45°C. It could be that the TPM method enhanced production of sugary extract as a result of optimal enzyme activity.

Table 1: Effect of mashing process on some brewing parameters of banana fig compared with barley malt.

Parameters measured	Infusion at 45 °C			Infusion at 65 °C			Infusion at 80 °C			TPM		
	Ripe fig	Overripe fig	Barley	Ripe fig	Overripe fig	Barley	Ripe fig	Overripe fig	Barley	Ripe fig	Overripe fig	Barley
S. Gravity	1.0130	1.0148	1.0200	1.0110	1.0130	1.0250	1.0100	1.0130	1.0200	1.0190	1.0149	1.0200
pH	5.04	NA	3.64	5.0	4.9	3.75	3.67	4.9	3.86	4.92	5.3	3.80
%TTA	0.13	0.05	0.2	0.19	0.19	0.14	0.16	0.14	0.12	0.13	0.16	0.13
Brix	3.19	3.29	5.67	3.18	3.19	6.9	3.17	3.35	5.68	5.06	3.80	7.52
Yield (l/kg)	112.6	111.5	172.1	95.4	112.7	214.1	86.9	101.2	188.9	163.6	129.1	247.2
Saccha (min)	partial	partial	25	partial	partial	20	partial	partial	20	partial	partial	20

Data are average of three readings.

Generally, the specific gravity of banana fig extract decreased with increasing mashing temperature (Table 1). It decreased from 1.013 to 1.010 against temperatures of 45°C and 80°C respectively. The decrease might have been probably as a result of inactivation of the hydrolytic enzyme required for the conversion of starch to sugar as temperature increases. The specific gravity of the overripe fig was higher than that of banana fig. This shows that the extract from overripe fig has higher soluble solids than extract from ripe fig. Barley extract had higher specific gravity than the banana figs in all the methods of mashing used. The highest SG occurred when TPM method was used compared to the infusion method implying that

The percentage sugar content of the banana fig in the mashed samples decreased as the mashing temperature increased (Table 1). It was 3.19, 3.18 and 3.17% as against mashing temperatures of 45°C, 65°C and 80°C respectively. The decrease in % sugar content could, again be attributed to

the inactivation of enzymes responsible for conversion of starch to sugars as mashing temperatures increases. The % sugar content of ripe, overripe fig and barley were 3.18, 3.19 and 6.8 respectively. But under TPM methods they were 5.06, 3.8 and 7.52 respectively. This could be as a result of TPM adopted which enhanced the conversion of starch to sugar as a result of optimal enzyme activity.

Among the mashing methods TPM gave the highest extract yield for both banana fig and barley (Table1). The highest extract yield obtained with this method was as a result of effective enzyme activity. Briggs, (1998) stated that mashing with TPM allows optimal enzyme activity because it allows the activities of key heat-labile enzymes in the malt, supposedly proteases and  $\beta$ -glucanases as well as the actual saccharification enzymes  $\alpha$  and  $\beta$ -amylase. Generally, banana fig had lower hot water extract than barley malt. High soluble extract was due to higher solubility of the barley endosperm (Bathgate and Palmer, 1975). The low extract

yield of banana fig could be attributed to incomplete solubilization of the polymerized fig. However, the extract yield of the ripe and over ripe banana fig increased with the inclusion of the enzyme (Table 2). It was 163.6 and 129.1 l<sup>0</sup>/kg

before the inclusion of the enzyme but increased to 270.8 and 137.5 l/kg for ripe and overripe banana fig respectively after enzyme addition. The extract yield was even higher than that of barley (247.2 l/kg).

**Table 2: Effect of exogenous enzyme inclusion on brewing parameters of banana fig Extract**

Parameters	No Enzyme treatment			Enzyme treatment		
	Ripe fig	Overripe fig	Barley	Ripe fig	Overripe fig	Barley
pH	5.33	5.25	3.80	4.5	4.5	3.80
TTA (g/100g)	0.13	0.05	0.13	0.15	0.10	0.14
Specific Gravity	1.0190	1.0149	1.0290	1.0318	1.0157	1.0290
<sup>0</sup> Brix	5.06	3.08	7.52	8.0	4.0	7.52
Ext. Yield (l/kg)	163.6	129.1	247.2	270.8	137.5	247.2
Sach. (min)	20	20	20	15	10	20
Degree of Sach.	partial	partial	Complete	complete	complete	complete

Values on the same row with the same superscripts are not significant different from each other

The saccharification time (time taken for the complete destruction of the starch) of the mashed banana fig was longer at low mashing temperatures (Table 1). It was 20 and 10 minutes against mashing temperatures of 45°C and 60°C respectively. The longer time of saccharification could be due to the fact that samples mashed at low temperature do not support optimal enzyme activity ( $\alpha$ -amylase) and vice versa. Enzyme activities are temperature dependant, increases with temperature until at some elevated temperature a conformational change occurs which destroys the active site and renders the enzyme inactive (Ihekoronye and Ngoddy, 1985). Mashing above gelatinization temperatures enables adequate amylolysis. Partial saccharification indicates that there is incomplete starch breakdown to sugars. Saccharification was partial with samples of banana fig using fixed mashing temperature of 70°C and above, but full saccharification was obtained when enzyme was introduced.

The introduction of the amyloglucosidase increased the specific gravity of both the ripe and over ripe banana fig "must". It increased from 1.0190 to 1.0318 and 1.0149 to 1.0157 for ripe and overripe banana figs respectively (Table 2). This could be as a result of increased enzyme activity which hydrolyzed the starch of the banana figs to sugar. In addition, the % sugar content of the ripe and over-ripe fig increased with the inclusion of the enzyme. (Table 2). This might probably be as a result of complete saccharification of the starch to sugar by the enzyme introduced to the wort.

The mean sensory score of the different malt drinks is tabulated in Table 3. The result showed that sample BS3 was rated highest ( $P < 0.05$ ) in terms of colour with mean score of 6.0, which was significantly different from others. Sample BS2, BS1 and the reference product (Commercial-Amstel Malt Drink) were not significantly different from each other in terms of colour at  $P < 0.05$ . The high mean score of sample BS3 is attributed to the high percentage caramel colour imparted to it from the overripe fig used. In terms of taste and flavour there was no significant ( $P < 0.05$ ) difference in all the products including the reference product. This implies that an appropriate recipe was adopted during the formulation to give products that do not differ much in taste and flavour with the reference product. BS1 had the highest mean score of 5.5 for taste while the reference product had the highest mean score of 5.2 for flavour. BS3 was rated highest in terms of general acceptability by the panelist with a mean score of 5.7 followed by the reference product with a mean score of 5.1. This could be because of its high body (viscous) characteristics. There was no significant difference at  $P < 0.05$  in both products even though they were rated differently in terms of their mean scores. BS2 was rated lowest in almost all the attributes evaluated sensorily. This implied that the recipe used for the formulation of this particular sample need to be re-visited.

**Table 3: Means sensory scores of low-glycemic malt flavoured drink.**

SAMPLES	COLOUR	TASTE	FLAVOUR	ACCEPTABILITY
BS3	6.0 <sup>b</sup>	5.0 <sup>a</sup>	5.5 <sup>a</sup>	5.7 <sup>b</sup>
BS2	4.9 <sup>a</sup>	5.1 <sup>a</sup>	4.9 <sup>a</sup>	5.0 <sup>a</sup>
BS1	4.9 <sup>a</sup>	4.8 <sup>a</sup>	4.9 <sup>a</sup>	4.8 <sup>a</sup>
COMMERCIAL	5.3 <sup>a</sup>	5.2 <sup>a</sup>	5.1 <sup>a</sup>	5.1 <sup>b</sup>

Evaluation was done with 7-point Hedonic Scale.

BS3 = 11.8 % sugar BS2 = 7.4% sugar BS1 = 6.2% sugar Commercial = 13.0% sugar

Means in the same column with the same superscript are not significantly different, while those with the different superscripts are significantly different at ( $p < 0.05$ ).

## CONCLUSION

Low-glycemic malt flavoured drink could be produced from banana fig using amyloglucosidase through a process that precludes malting procedures. From the result obtained the malt drink has similar organoleptic characteristics when compared with conventional malt. The low-glycemic malt flavoured drink will go a long way to control and reduce the incidence of Type I diabetes, obesity, coronary artery disease and all other sugar related diseases. In addition, importation of barley malt and hop extract in Nigeria for production of malt drink will drastically be reduced. This is because banana fig

and local hop (Alfalfa) could be used as substitutes for production of low-glycemic malt flavoured drink.

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