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Production of Malt Flavoured Low-Sugar Drink from Banana *(Musa sapientum)* Fig Using Amyloglucosidase

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ABSTRACT

Banana fig from fully ripe banana, caramel and malt extract, food drink thickening agent, acetic acid and local hop extract (Alfalfa) were used in the formulation of a low-sugar malt drink. Banana fig was used as a replacement for malted barley. The pH, total titratable acidity (TTA), percentage sugar content, specific gravity and saccharification of the extracts were determined. The effects of optimization of the mashing process using industrial enzyme-amyloglucosidase were also evaluated. The pH and percentage sugar content of the banana fig extract decreased with increased mashed temperatures. Specific gravity of the banana fig extracts decreased with increased mashed temperatures, with values from 1.013 to 1.010 against mashed temperatures of 45°C and 80°C respectively. At temperatures of 70°C and above the saccharification was observed to be incomplete. However, the introduction of industrial enzyme-amyloglucosidase resulted in complete saccharification. The formulation of the malt flavoured low-sugar drink gave three samples of 6.2%, 7.4% and 11.8% sugar contents. Sensory evaluation carried out on the malt flavoured low-sugar drink with commercial Amstel malt drink showed no significant difference in taste and flavour for all the samples compared with reference sample at $p > 0.05$. But sample BS3 with 11.8% sugar content was significantly different ($p < 0.05$) from sample BS2 (7.4% sugar) and BS1 (6.2% sugar) in colour and general acceptability. All samples except BS1 with 6.2% sugar were accepted by the panellists. This malt flavoured low-sugar drink could thus help reduce health complications in conditions associated with high sugar consumption. The use of additional enzymes in combination with the amyloglucosidase could improve the extract yield, nutritional and sensory qualities of the drink.

Keywords: Low-sugar, amyloglucosidase, mashing, banana fig.

Introduction

Malt drink is a non-alcoholic, wholesome, nourishing and satisfying food drink with zero or negligible level of alcohol. Non-alcoholic drinks are additional products to beer which are produced and marketed by several breweries in Nigeria (Okon and Akpanyung, 2005). In Nigeria, malt drinks are chiefly made from malted barley (Chukwurah, 1988). Malt is a product made by limited germination of cereal grains, mostly barley, followed by drying of the grain. Conventionally, malt drink production involves the use of similar raw materials, machinery and procedure as in beer brewing. However, malt drinks are reported to be more nutritious than beer. Moreover, there are more potential customers for the malt drink than beer in view of its non-alcoholic nature (Jepsen, 1993).

The malt drinks in Nigeria contain substantial amount of reducing sugar-glucose (603.66 – 943.52 mg/dl) from the enzymic hydrolysis of starchy raw materials (barley, sorghum, maize) during mashing (Okon and Akpanyung, 2005). In most cases, sucrose is added to the formulation of these malted beverages for taste. The resultant high sugar level (up to 14%) may not be good for all consumers. Some consumers, for health purposes, do not need and do not want to consume the conventional malt

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drink because of its high sugar content. According to Okon and Akpanyung (2005), diabetic patients should exercise restraint in the consumption of malt drinks since high level of sugar in malt drinks could lead to complications in this disease condition.

This study was carried out to produce malt flavoured low-sugar drink from banana *(Musa sapientum)* using the enzyme, amyloglucosidase.

Materials and Methods

Sample collection

Fully ripe banana was sourced from a local market at Owerri in Imo State. The industrial enzyme (amyloglucosidase) was obtained from Federal University of Technology, Owerri (FUTO) while barley malt was sourced from Nigerian Breweries Plc, Aba, Abia State.

Preparation of banana "fig" and optimization of mashing conditions

Fully ripe banana was washed with tap water and hand peeled. The banana was diced into round cuts of 2 cm thickness with a sharp kitchen knife and dried between 50 – 60°C for 72 h to a moisture content of 17% in a fan-driven oven (Hot Box Gallenkamp) manufactured in England to obtain a leathery-dark brown "fig". The leathery-dark brown "fig" obtained after drying was milled to smaller particle sizes using manual grinder for 5 min to obtain the "grist". Temperature programmed mashing method (TPMM), as described by Briggs (1998), was used for the extraction. Distilled water (200 ml) at 45°C was added to 50 g of the milled banana "fig". The mash obtained was stirred continually at 45°C for 30 min; at the expiration of 30 min, the temperature of the mash was increased at the rate of 1°C/min for 25 min until it reached 70°C. Distilled water (100 ml) at 70°C was again added to the mash and the temperature of the mash was maintained at 70°C for 1 h. At this point the saccharification time was determined using iodine reagent. The mash was then allowed to cool after the expiration of 1 h and the stirrer was rinsed into the mash. The mash weight was adjusted to 450 g by addition of distilled water. After this the mash was filtered to obtain the extract using muslin cloth. The banana fig "wort" was treated with industrial enzyme-amyloglucosidase to complete the starch breakdown. Banana fig "wort" (450 g) of pH 5.33 was reduced to a constant pH of 4.5 by addition of acetic acid. Digital Jenway pH metre manufactured in England was used to monitor the pH reduction. About 2 ml of the enzyme amyloglucosidase was added to the samples at the temperature of 60°C and stirred for 30 min for the enzyme breakdown of the starch. At intervals of 2 min, the saccharification time was determined by adding a few drops of iodine reagent to the small sample in a crucible. The disappearance of the blue black colouration marked the end of complete breakdown of the starch in the samples and the time when this happened was noted as the saccharification time.

Formulation of drink

The banana "fig" extracts treated with industrial enzyme-amyloglucosidase was used for the formulation to give three different samples. In sample BS3, 100 ml of the banana "fig" extract was mixed and blended with 10% of malt extract (233.0 Brix), caramel (1%), local Hop extract (alfalfa) (1%) and 10% of food drink thickening material. In sample BS2 the same quantity of banana "fig" extract was mixed and blended with 7.5% Malt extract (174.4 0Brix), caramel (0.75%), local hop extract (alfalfa) (0.75%) and 7.5% of food drink thickening material. In sample BS1, 100 ml of banana "fig" extract was mixed and blended with 5% malt extract (116.30 Brix), caramel (0.5%), local hop extract (0.5%) and 5% food drink thickening material. After blending, the samples were filtered using muslin cloth. After the filtration, the filtrate was transferred into a 33 cl bottle, pasteurized and cooled before storing in the refrigerator prior to final sample analysis and sensory evaluation.

Proximate analysis of extract

The banana "fig" extract was analyzed for pH, total titratable acidity (TTA), specific gravity, % sugar content, extract yield and saccharificaton time. The method of Association of Official and

Analytical Chemists (AOAC, 1970) was used in the determination of the TTA. The extract yield was determined following the Institute of Brewing (IoB) (1977), while the saccharification time was evaluated following the European Brewing Convention (EBC) (1987). The % sugar was determined using hand refractometer. The Specific gravity was determined by the method described by Anderson 1970 and the pH by the method described by Vine (1981).

Sensory evaluation

Sensory evaluations were carried out on the three samples of the malt flavoured low-sugar drink with commercial Amstel malt drink and analyzed statistically using a two-way analysis of variance (ANOVA) described by Ihekoronye and Ngoddy (1985). The evaluation was based on quality parameters such as colour, taste, flavour and general acceptability. A 7-point Hedonic scale was used. Thirty (30) panellists comprising fifteen males and fifteen females, some having basic knowledge in brewing science and technology, were selected within and outside FUTO for sensory evaluation.

Result and Discussion

pH of the banana "fig" extract

The pH were 5.04, 5.00, 4.97 and 4.95 against mashed temperatures of 45, 60, 70 and 80°C respectively (Table 1). The pH of the ripe banana "fig" decreased as the mashed temperatures increased.

TTA of the banana "fig" extract

The TTA of the banana "fig" extract were 0.20 at mashed temperature of 60°C and 0.13 at 45 – 70°C range of mashed temperatures (Table 1). This could be attributed to the temperature programmed mashing method adopted (TPMM) which enhanced optimal enzyme activity that resulted in extraction of sugary extract than acidic components of the sample.

Specific gravity of the banana "fig" extract

All the specific gravity (SG) of the banana "fig" extract decreased with increased mashed temperature (Table 1). It decreased from 1.013 to 1.010 against mashed temperatures of 45°C and 80°C respectively. The decrease probably could be as a result of inactivation of the hydrolytic enzyme

Table 1: Parameters evaluated for the banana fig extract as affected by mashed temperatures

Mashed Temperature (°C)	Time (min)	pH Banana fig extract	TTA Banana fig extract
45	60	5.04	0.20
60	60	5.00	0.20
70	60	4.97	0.20
80	60	4.95	0.20
45-70*	155	4.92	0.13
Specific Gravity (20°C)		**Percentage sugar (°Brix)**	
45	60	1.0130	3.19
60	60	1.0110	3.18
70	60	1.0105	3.17
80	60	1.0100	3.17
45 – 70*	155	1.0190	5.06
	Extract yield (L°/kg)	**Saccharification time (min)**	
45	112.6		20
60	95.4		10
70	91.2		0
80	86.9		0
45 – 70*	163.6		0

* Extraction started at 45°C for 30 min, and then followed by steady increase of 1°C/min for 25 min until 70°C where the temperature was maintained for 1 h (Temperature-programmed mashing method – TPMM).

required for the conversion of the starch to sugar as temperature increased. The TPMM (45 – 70°C) adopted resulted in the marked increase in the specific gravity of the mashed samples than samples mashed at specific temperature. This implies that TPMM enhanced optimal enzyme activity than other mashed temperatures. For example, banana "fig"extract had a lower SG of 1.013 at mashed temperature of 45°C while the temperature range of 45 – 70°C gave a value of 1.019 for the TPMM (Table 1).

Percentage sugar content of the banana "fig" extract

The results of the percentage sugar content were 3.19, 3.18, 3.17 and 3.17 as against mashed temperatures of 45, 60, 70 and 80°C respectively. The percentage sugar content of the banana "fig" extract in the mashed samples decreased as the mashed temperatures increased (Table 1).

The decrease in percentage sugar content could be as a result of the inactivation of enzymes responsible for conversion of starch to sugars as mashed temperatures increased.

Extract yield of the banana "fig" extract

The TPMM gave the highest extract yield of the banana "fig" extract (Table.1). It was 163.6 litre degrees per kilogram (l°/kg). The highest extract yield obtained with this mashing method was as a result of optimal enzyme activity. Briggs (1998) stated that mashing with TPMM allows optimal enzyme activity because it allows the activities of key heat-labile enzymes in the malt, supposedly proteases and β-glucanases, as well as the actual saccharification enzymes α and β-amylase.

Saccharification time of the banana "fig" extract

The saccharification time were 20 and 10 min against mashed temperatures of 45°C and 60°C respectively. The saccharification time (time taken for the complete destruction of starch) of the mashed banana "fig" extract was longer at low mashed temperature (45°C) than medium mashed temperature (60°C) (Table 1). The longer time of saccharification could be due to the fact that mashed samples at low temperature (45°C) do not support optimal enzyme activity (α-amylase) and vice versa. Enzyme activities are affected by temperature which invariably increase with temperature until a conformational change occurs which destroys the active site and renders the enzyme inactive (Ihekoronye and Ngoddy, 1985). The saccharification time indicating partial implies that there is incomplete starch breakdown to sugars. It was partial with samples of banana "fig" extracts mashed at elevated temperature of 70°C and above. Partial saccharification could be as a result of conformational changes which occur to the hydrolytic enzymes at elevated temperature thereby rendering the enzyme inactive. There was complete saccharification for the banana "fig" extracts at 60°C. The saccharification time was 10 min for the sample. This could be as a result of optimal enzyme activity at this temperature.

Use of industrial enzyme

The introduction of industrial enzyme-amyloglucosidase increased the specific gravity from 1.0190 to 1.0318 (Table 2). This could be as a result of increased enzyme activity which hydrolysed the starch of the banana "fig" to sugar. In addition, the percentage sugar content of the extract increased with the inclusion of the enzyme-amyloglucosidase (Table 2). It increased from 5.06 to 8.0. This might be as a result of complete breakdown of starch to sugar (saccharification) by the enzyme introduced to the "wort".

The extract yield increased with the inclusion of the enzyme (Table 2). It was 163.6 (l°/kg) using TPMM before the inclusion of the enzyme but increased to 270.8 (l°/kg) after enzyme addition. The saccharification was complete with the addition of the enzyme. It was formally partial; meaning incomplete destruction of starch in the samples. The complete saccharification could be as a result of the hydrolysis of the starch retained in the "wort" by the enzyme-amyloglucosidase.

Table 2: Properties of the "wort" before and after addition of Amyloglucosidase

Parameters	No enzyme	Plus enzyme
	Banana "fig" extract	**Banana "fig" extract**
pH	5.33	4.50
TTA	0.13	–
Specific gravity (200)	1.0190	1.0318
Percentage sugar (°Brix)	5.06	8.0
Extract yield (l0/kg)	163.6	270.8
Saccharification	Partial	Complete
Mashing temperature (°C)	45 – 70	45 – 70
Mashing time (mins)	155	155

Table 3: Means sensory scores of malt flavoured low-sugar drink

Samples	Colour	Taste	Flavour	Acceptability
BS3	6.0^{b}	5.0^{a}	5.5^{a}	5.7^{b}
BS2	4.9^{a}	5.1^{a}	4.9^{a}	5.0^{a}
BS1	4.9^{a}	4.8^{a}	4.9^{a}	4.8^{a}
Commercial	5.3^{a}	5.2^{a}	5.1^{a}	5.1^{b}

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

Evaluation was done with 7-point Hedonic Scale
BS3 – about 100 ml of banana extract blended with 10% of malt extract (233.°Brix), caramel (1%), local Hop extract (alfalfa) (1%) and 10% of food drink thickening material.

BS2 – about 100 ml of banana extract blended with 7.5% Malt extract (174.4 °Brix), caramel (0.75%), local hop extract (Alfalfa) (0.75%) and 7.5% of food drink thickening material.

BS1 – about 100 ml of banana extract blended with 5% Malt extract (116.3°Brix), caramel (0.5%), local hop extract (0.5%) and 5% food drink thickening material.

Sensory evaluation

The mean sensory score of the three different products of formulated malt flavoured low-sugar drink were tabulated in Table 3. The result showed that sample BS3 (11.8% sugar) was rated highest in terms of colour with a mean score of 6.0 and significantly different with other products evaluated at $p < 0.05$. Sample BS2 (7.4% sugar), BS1 (6.2% sugar) and the reference product (Commercial-Amstel malt drink – 13% sugar) were not significantly different in terms of colour at $p < 0.05$. The high mean score of sample BS3 (11.8% sugar) for colour was attributed to the high percentage of caramel (l%) used in the recipe during the formulation of the drink. In terms of taste and flavour there were no significant differences at $p < 0.05$ in all the products formulated including the reference product. This implies that appropriate recipe was adopted during the formulation which gave products that did not differ much in taste

and flavour with the reference product. BS1 (6.2% sugar) had the lowest mean score of 4.8 for the taste while the reference product had the highest mean score of 5.2 for taste. Finally, BS3 (11.8% sugar) was rated highest in terms of general acceptability by the panellists with a mean score of 5.7 followed by the reference product with a mean score of 5.1 (Table 3). There was no significant difference at $p < 0.05$ in both products in terms of general acceptability even though they were rated differently in terms of their mean scores. BS1 (6.2% sugar) was rated lowest in almost all the attributes evaluated. In terms of colour, taste and flavour, it had mean scores of 4.9, 4.8 and 4.9 respectively while the general acceptability mean score was still the lowest with mean score of 4.8. This implies that the recipe used for the formulation was inappropriate and needs to be re-formulated so as to make it to be generally acceptable. In all the quality parameters evaluated, there was no significant difference at $p < 0.05$ for BS3 (11.8% sugar) with the reference product except for colour. This shows that BS3 (11.8% sugar) could compete well with the commercial malt drink product with improvement in its colour.

Conclusion

Low-sugar malt flavoured drink could be produced from a non-cereal based (banana "fig") using amyloglucosidase through a process that precludes malting procedures. From the result obtained, the malt flavoured low-sugar drink had similar sensory attributes with conventional malt drink. Importation of barley malt and hop extract in Nigeria for the production of malt drink may be minimized since banana and local hop (alfalfa) can be good substitutes.

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