Original article

Some microbiological and nutritional properties of Borde and Shamita, traditional Ethiopian fermented beverages

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Abstract: Borde and Shamita are two popular fermeated beverages of thick consistency drunk in the southern part of Ethiopia. They are prepared from maize and barely, respectively. The pH values of borde and shamita were 4.1 and 4.2, respectively and counts of aerobic mesophilic bacteria and lactic acid bacteria were high in both products (around 109 cfu/ml). The counts of Enterobacteriaceae was around 106 cfu/ml whereas yeast count ranged between 107 and 108 cfu/ml for both products. In all counts variations within samples were markedly low (coefficient of variation, 5-11 %). Both beverages had comparable protein and fat contents. A third of the borde was soluble. Compared to the raw ingredients, fermentation has resulted in increase in protein, fat and ash contents of the finished products.[Ethiop. J. Health Dev. 1995;9(1):105-110]

Introduction

In many parts of Africa, villagers prepare fermented beverages from maize, sorghum, millet, barley or from various mixtures of these cereals. There is some information on the fermentation of a variety of African beverages such as Pito, Burkutu and Obiolor from Nigeria (1-3), Kaffir or Bantu beer from southern Africa (4), Merissa from Sudan (5), .Busaa from Kenya (6) and Tella from Ethiopia (7).

A variety of fermented cereal beverages are produced in the different parts of Ethiopia. These consist of different varieties of Tella, Borde, Shamita, Korefe, and others. The microbiology of Tella has been reported very recently (7) and short descriptions of the other products are found elsewhere (8) .

Horde and shamita are among the important and popular fermented beverages consumed in the southern regions of Ethiopia. Horde is produced by fermenting maize whereas barley is the major ingredient for shamita production. The beverages are thick in consistency and serve as meal replacements for most people
who cannot afford a reasonable meal. In most open markets in southern Ethiopia, horde and shamita are available for purchase.

For shamita preparation, barley is dehulled, roasted and ground. To 100 kg barley flour, three kg of salt, nine kg of ground linseed and small amount of spices consisting of black cumin (Nigella sativa), Ethiopian caraway (Trachyspermum ammi), false cardamom (Aframomum korarima) and Ocimum sp. are added. Ground linseed is believed to ensure thick consistency of the product. A single preparation is usually made by thoroughly

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Table 1: **counts of the various microbial groups in shamita and borde**

	pH	log (cfu/ml)			
		AMC[1]	Enterobacteriaceae	Yeasts	LAB[2]
Shamita					
Mean	4.2	9.00	6.38	7.01	8.92
SD	0.02	0.63	0.60	0.78	0.67
%CV	2.1	7.0	9.4	11.1	7.5
Borde					
Mean	4.1	9.13	6.37	8.27	8.95
SD	0.15	0.75	0.72	0.49	0.49
%CV	3.7	8.4	11.3	5.9	5.4

[1]AMC, Aerobic mesophilic count
[2]LAB, Lactic acid bacteria

mixing 25 kg of the above ingredients with 50 liters of water. One liter of shamita from a previous preparation is added as a starter . Mixing is usually done in the evenings and the product is ready for consumption the following morning. At this stage about 200 9 of ground bird's-eye chili (Capsicum minimum) is added to the product and then immediately served. It is consumed while at an active stage of
fermentation as noted by constant gas production. The product turns too sour four hours after being ready for consumption. A laborer usually consumes about three to four liters of shamita and a liter costs about USD 0.15.

Borde is prepared mainly from maize. Twenty five kg of maize flour is soaked in excess water and then deeply roasted in a hot flat metal pan. After cooling for about 30 min, about 250 9 of malt is thoroughly mixed into it. This is put into a large clay container and further blended in about 30 litres of boiling water. At this stage, 10 kg of ground barley whipped in hot water is added to it and allowed to ferment overnight. The addition of ground barley is believed to be important in gas production. About one liter of borde from a previous fermentation is usually added as starter. In the morning, the whole fermenting mixture is filtered using wire sieves and the filterate is served for consumption. Depending on the preference of consumers, ground chili (Capsicum minimum) may be added to it at 1 serving. Horde is a very popular meal replacement which is consumed

while at an active stage of fermentation. An average worker consumes about three liters of borde in I the morning. This is enough to keep him for I most of the day without any additional meal.

Information on microbiological and biochemical properties of traditional fermented beverages in Ethiopia is very scanty. The purpose of this work is, therefore, to evaluate the microbiological and biochemical quality of two fermented beverages as made available to the consumer in an open market in Awassa, Southern Ethiopia.

Methods

Source and Collection of Samples; This study was carried out in Awassa, a town located 275 km south of Addis Ababa. It has a population of about 63,000. A total of 30 samples each of borde and shamita were collected at random from open-market vendors on different sampling days. Microbiological analysis was conducted within two hours of collection.

Microbiological analysis; Twenty-five ml of borde or shamita were sampled aseptically at 12 or 24 hr intervals. They were separately diluted in 225 mI of sterile water and processed for the following microbiological tests. Aerobic mesophilic bacteria were

Table 2: **Distribution (%) of dominant microorganisms in Shamita and borde**

Beverage type	Mean AMC*	Number of isolates	% of isolates					
			I	II	III	IV	V	VI
Shamita	$1.0x10^9$	420	66.1	16.5	6.7	5.7	2.2	2.0
Borde	$8.9x10^8$	370	31.4	25.2	30.3	3.6	9.5	

*AMC, Aerobic mesophilic count (log cfu/ml) I Bacillus II Lactobacillus III Micrococcus
IV Staphylococcus V Acinetobacter VI Streptococcus

analyzed after further dilution of samples in sterile water. Volumes of 0.1 ml of appropriate dilutions were spread-plated in duplicate on pre-dried surfaces of Plate Count Agar (PC; Merck). Colonies were counted
after incubation at 30 to 32 °C for 48 hrs. For the enumeration of Enterobacteriaceae volumes of 0.1 mI of appropriate dilutions were spread plated in duplicate on pre-dried surfaces of Violet Red Bile Glucose (VRBG) Agar (Oxoid) plates. The plates were incubated at 30 to 32 OC for 24 h. purple red colonies were counted as members of Enterobacteriaceae. For counting lactic acid bacteria, volumes of 0.1 mI of appropriate dilutions were spread plated in duplicate on pre-dried surfaces of de man, Rogosa sharpe (MRS) agar (Oxoid) plates.

Colonies were counted after incubation in an anaerobic jar (Oxoid) at 32 °C for 48 h. For the enumeration of yeasts and mo.lds, volumes of 0.1 ml of appropriate dilutions were spread plated

in duplicate on pre-dried surfaces of chloramphenicol-bromophenol-blueagar (CBB) consisting of yeast extract, 5.0; glucose, 20.0; chloramphenicol, 0. 1; bromophenol blue, 0.01; agar, 15; pH, 6.0 to 6.4 (g/1 in distilled water). Yeast colonies were counted after incubating the plates at 25-27 °C for 5 days. Flora assessment was done as follows. After colony counting, 10-15 colonies were selected at random from countable PC agar plates. The
sub-cultures were further purified by repeated plating and differentiated into various bacterial groups by the following characteristics. Phase- contrast microscopy was used to examine cell shape and grouping, presence or absence of endospores and motility. Gram reaction was determined using the KOH test of Gregersen (9). Cytochrome oxidase was tested by the method of Kavacs (10). Catalase test was made with 3% (v/v) H2O2 solution. Glucose metabolism was investigated by the O/F test of Hugh & Leifson (11).

Biochemical Analyses: pH was measured by aseptically placing the electrode of a digital pH meter in the samples. Moisture content was determined by drying a sample to constant weight in a ventilated thermostatic oven at 70 °C. Borde and shamita samples were freeze dried and stored at 4 °C until they were further analysed for protein, fat and ash. To determine total protein content, two 9 sample was digested with 15 mI concentrated sulphuric acid for 45 min at 410 °C. A solution of sodium hydroxide -sodium thiosulphate was
added to the digest and distilled into a solution of boric acid and titrated with 0.2N hydrochloric acid according to AOAC 981.10 (12). Protein percentage was calculated by multiplying %N by 6.25. For fat determination a two gm sample was solubilized in alcohol (2 ml) and hydrolyzed with 10 ml concentrated hydrochloric acid at 70-80 °C for 40 min. The hydrolyzed fat was extracted with petroleum ether. The ether was evaporated from the extract and the fat was dried to constant weight at 100 °C for 90 min according to AOAC 922.06 (12). Ash was determined by igniting a five gm sample in furnace at 550 °C to constant weight according to AOAC 923.1(12).

Protein availability was estimated by the in vitro disappearance of dr)' matter after treating a sample with papain as described by Kazanas and Fields (13).

Table 3: Protein, fat and ash content of horde andshamita as determined by Agren and Gibson (17). N.D. notdetermined

	% protein		% fat	%ash
	total	soluble		
Roasted				
Maize	8.7	N.D	4.6	1.6
Borde	9.55	3.31	6.88	3.66
Roasted				
Barely	9.0	N.D	1.9	2.1
Shameta	10.37	3.46	6.85	5.92

Results

Borde and shamita had low pH values($\leq$4.2) with insignificant variation within samples (Coefficient of variation, CY, <4%). Counts of aerobic mesophilic bacteria and lactic acid bacteria

were also high (around 109 cfu/ml) for both products. Counts of Enterobacteriaceae was over 106 cfu/ml in both products but yeast counts were slightly higher in horde samples. In all counts variations within samples were markedly low (CY, 5- 11%) (Table 1).

A total of 790 isolates were obtained from countable PC plates in this study. The aerobic mesophilic flora was dominated by a variety of bacterial genera (Table 2). Bacillus spp. heavily dominated the microflora in shamita. The major genera that dominated horde were Bacillus, Micrococcus and Lactobacillus spp. The mean moisture contents of horde and shamita were 86.43% and 81.03%, respectively. Shamita had slightly higher content of protein and ash than horde (Table 3). No marked difference was observed in their fat content. Over a third of the total protein in both fermented beverages was soluble.

Discussion

Horde and shamita could be considered as acidic beverages. The low pH observed 'in horde and shamita is in agreement with other observations tnade during fermentation of traditional beverages in Sudan (5) and Nigeria (3). The low pH value obtained in a short fermentation time showed that sufficient fermentable sugar was available and the lactic acid bacteria involved were strong acid producers.

Despite the high yeast counts, both beverages are known to have low alcohol content due to the short fermentation time. The products are desired to have low alcohol content and, to this effect, are usually consumed within two to three hours. A similar non-alcoholic fermented beverage was reported from Nigeria (3). Longer holding could render the products too alcoholic for meal replacements.

The high amount of gas in the fermented products was indicative of the activity of yeasts and other gas producing microorganisms. Lactic acid bacteria and yeasts are important in the fermentation of various African fermented beverages or foods made from cereals (1 ,3,6, 14,15). The count of Enterobacteriaceae in both products was in the order of 106 cfu/ml and these could contribute to acid production in the early stages but could be inactivated at the final pH level. Some members of Enterobacteriaceae were reported to be involved in the fermentation of African maize beverages or fO<His (6,14).

The aerobic microflora of shamita was markedly dominated by bacillus spp whereas that of horde was dominated by bacillus and micrococcus spp. In the presence of sufficient numbers of lactic acid bacteria, Bacillus species may not contribute to the fermentation of the products. They could not multiplyat pH levels as low as 4. However, in the initial stages of fermentation, micrococci may acidify the flower-and-water paste or bacillus may grow, producing lactic acid, gas, alcohol, acetoin and small amounts of esters and aromatic compounds (16). The dominance of bacillus spp. in Shamita might be due to the addition of an assortment of spices both before the initiation of the fermentation and at its completion. A variety of spices are known to contain a high load of Bacillus spores. Bacillus sp. were reported to be actively involved in the fermentation of Obiolor, a Nigerian acidic non-alcoholic fermented beverage (3).

The high microbial load in horde and shamita could make the products good sources of microbial protein and this might contribute to their role as meal replacements. Some members of Enterobacteriaceae are reported to
synthesize some vitamins while the lactic acid bacteria and yeasts could be responsible for production of lactic acid and flavour components, respectively (14). , The protein contents of roasted Ethiopian barley and maize, which are used in the production of shamita and horde, are 9.0% and 8.7 % , respectively (17) .The fermented products appeared to have higher protein content. A marked increase was also noted in fat and ash content in the fermented products.

The increment in the various nutrients could be due to the proliferation and action of microorganisms during fermentation. Similar increase in crude protein and fat was also observed during the traditional fermentation of maize and sorghum in Nigeria (18). Heat treatment of the grains could also encourage microbial protein production as noted by Abasiekong (18).

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