Comparative assessment of nutritive values and safety characteristics of bread sold in Bangladesh


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Abstract

Safe and good quality food products are essential to minimize human health risks and to facilitate business. The objective of this study was to assess the nutritional quality and defects of bread produced in Bangladesh. A total of sixty different types of bread sold in Bangladesh were collected by stratified random sampling and categorized as Brand shop (A), Local shop (B), and Street shop (C) bread. The samples were assessed for their nutritive values, microbiological contaminations, aflatoxins, heavy metals content, and sensory attributes. Nutrient composition and microbial quality were analyzed by using standard methods of the Association of Official Analytical Chemists (AOAC) and the American Public Health Association (APHA). High-Performance liquid chromatography method was used for aflatoxins determination. All categories of bread were low in macro and micronutrients (protein, fat, calcium, and iron) and high in sodium and alcoholic acidity. The heavy metals detected in bread (lead, cadmium, nickel and arsenic) were within the specification. Microbiological results showed that Salmonella spp. and Bacillus spp. were detected in many of the bread samples, whereas Escherichia coli and coliform were found in significant amounts in all categories of bread. Another important health concern that aflatoxins B₁ and B₂ were found in three bread samples of the local shops. In terms of sensory attributes, the brand shop bread had acquired the highest score for taste, texture, color, and overall acceptability. The study results showed that many of the bread samples were not suitable for healthy consumption, and special awareness should be taken for public health concern; it also warrants further research on safety and manufacturing practices of bread in Bangladesh.

1. Introduction

Bread, the most common form of cereal intake in many countries, has been designated the staff of life and rightly so since it provides more nutrients per gram of weight than any other single food source like meat, milk, potatoes, fruits, and vegetables (Campbell et al., 1991). Bread is a staple food in many countries around the world. In most European countries, it is the source of half of the total carbohydrate requirements, one-third of the protein requirements, and over 50% of the B-vitamins and 75% of the vitamin-E requirements (Islam and Haque, 2011). It is a good source of nutrients, such as macronutrients (carbohydrates, protein, and fat) and micronutrients (minerals and vitamins) that are all essential for human health. These values make the bread to be known as an essential food for human nutrition, and this has led all the countries throughout the world to study the composition of the bread to improve its nutritive value (Ijah et al., 2014).

In Bangladesh, the popularity of bread is increasing in both urban and suburban areas. Consumption of leavened wheat bread has increased dramatically in...
developing countries like Bangladesh because of changing food habits, increasing the working population outside the home, and urbanization. It is consumed extensively in most homes, restaurants, and hotels. The urban lifestyle is a more market dependent for food with very limited capacity for home preparation. As a consequence, the diet can be even more heavily biased towards pre-prepared and pre-cooked ready to eat food. According to the Bangladesh Bureau of Statistics (BBS) data, bread and cookie (biscuit) production were 100,305 metric tons in the year 2010-11. Over 100 manufacturers and 4,500 traditional small factories are producing bread and cookies in Bangladesh. The market size of the automated bread and cookie processing industries is estimated at approximately $56.5 million, excluding the traditional small bakery industries (Hussain and Leishman, 2013).

Nowadays, consumers have become concerned about their health and increasingly aware of the nutritional and wholesome quality of food products. There is little information on what bleaching and maturing agents are mixed with the flour other than meet bakers’ criteria, and toxicological tests may not realistically assess the dangers since chemicals are tested separately (Stauffer and Beech, 1990). The general public is uninformed about the effects of the processing that flour undergoes (Sabir and Sharef, 2013). Many international and national regulatory bodies provide standard specifications for different food products to protect consumer’s interests and help food manufacturers comply with the required formulation and processing conditions. Standards for bread stipulate basic ingredients, minimum loaf weights, solids content (not less than 62%), moisture content (maximum 38%), nutritive values and labeling of bread, and Codes of Practice during baking and post bake handling (SACN, 2012; FDA, 2019). Moreover, the addition of micronutrients such as iron (≥1.65 mg/100 g of flour), calcium (235–390 mg/100 g of flour, except self-rising flour), thiamine (≥ 0.24 mg/100 g of flour) and niacin (≥ 1.60 mg/100 g of flour) is mandatory in many developed countries like UK, USA, to restore nutrients lost during milling or bread-making process (O’Connor, 2012).

Bakery products, like many processed foods, are subject to physical, chemical, and microbiological spoilage. Many industrially produced baked goods with a surface that is essentially sterile can quickly lead to fungal, microbial contamination as a result of exposure to airborne contaminants during cooling, slicing, wrapping as well as equipment contact (Saranraj and Geetha, 2012). Well above 90% of bread were contaminated during post bake handling. Several studies reported the presence of various pathogenic microorganisms and the highest incidence of antibiotic-resistant Staphylococcus, Escherichia coli, and Bacillus spp. in beaked bread (Demissie and Natea, 2018). Also, molds such as Mucor and Rhizopus and fungi like Aspergillus and Fusarium spp. were found involving in deteriorating the bread quality. Food-borne disease outbreaks resulting from the contaminated bakery products have been reported in many countries including Australia, Europe, and the USA (NZFSA, 2007; Al-Fuad et al., 2018). However, the microbial quality and safety of bread sold in Bangladesh are still not well documented.

On the other hand, the key attributes of bread for the consumer are flavor and texture. The most important flavor compounds are formed due to the Maillard reaction and caramelization during baking. Besides the flavor attributes, the freshness, color, texture, and biting properties dramatically influence the overall perception of bread. When a loaf of bread is removed from the oven, a series of changes starts that eventually lead to deteriorating the quality of the bread (Gellynck et al., 2009). Since bread is an important part of daily food consumption, it follows that such food items should be healthy and wholesome. Therefore, the study is conducted to determine the nutritional properties and factors related to sound health, such as microbiological parameters, aflatoxins, heavy metals, and organoleptic attributes of bread. The objective also includes comparing the experimental values with the national and international standards of bread.

2. Materials and methods
2.1 Collection of samples
The most commonly consumed bread in Bangladesh is white bread. White bread samples were collected randomly from street-side shops, local shops, and brand shops. A total of twenty white bread samples were collected from each type of shop located in different areas of Dhaka city, Bangladesh, and coded as A, B, and C for the brand shop, local shop, and street shop respectively. All the bread samples were collected in the morning to get freshly prepared and immediately transferred to the laboratory of cereal technology, the Institute of Food Science and Technology (IFST), Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka-1205, Bangladesh for analysis.

2.2 Nutrient composition analysis
Nutritional compositions (moisture, crude protein, crude fat, crude fiber, and ash contents) were determined
using the method of (AOAC, 2005), while carbohydrate content was determined by difference, and energy was calculated using Atwater conversion factors. pH, alcoholic acidity, and acid insoluble ash were determined by the procedure described by Bhatt and Gupta (2015).

2.3 Minerals and heavy metals analysis

Minerals and heavy metals contents of the collected samples were determined by atom absorption spectrophotometer (AAS, Spectra AA 220, USA Varian), flame emission spectrophotometer (JENWAY, PFP7) and UV spectrophotometer (Analytikjena, SPECORD 205). The ash residue of each sample was digested with a mixture of perchloric acid and nitric acid (1:4 v/v). The samples were left to cool, and the contents were filtered through Whatman filter paper 42. Each sample solution was made up to a final volume of 25 mL with deionized water. The aliquot was used separately to determine the mineral and heavy metal contents of the bread samples following the procedure described by Juhaimi et al. (2016).

2.4 Microbiological analysis

Microbiological examination of the studied bread samples was performed to assess bacterial, mold and yeast load under laboratory conditions. Standard Plate Count (SPC), yeast and mold count, and enumeration of total coliform, E. coli and Salmonella spp. of the bread samples were examined according to the method of Downes and Ito (2001). In this regard, at first serial dilution was done as follows. Each sample of bread (25 g) was homogenized with 225 mL of sterile peptone water to prepare stock solution. Stocks were serially diluted (1:10) and the same procedure was repeated to make 10^{-4} by adding 100 µL of stock solution to 900 µL peptone water. A total of 100 µL of diluted sample was inoculated onto nutrient agar (NA) for the determination of total viable bacterial count (TVBC) following the spread plate technique. On the other hand, total coliform count, E. coli count, Salmonella spp. count and Bacillus spp. count was found using membrane Faecal Coliform (mFC) Agar, MacConkey-Sorbitol Agar, Salmonella Shigella (SS) Agar and PEMBA (Polymyxin pyruvate egg yolk mannitol bromothymol blue agar) medium respectively with 100 µL of diluted sample following streak plate technique. Besides, Rose Bengal Agar (RBA) media was used for yeast and mold count and PDA (Potato Dextrose Agar) was applied for Aspergillus spp. count following spread plate technique with that of diluted sample. Incubation was implemented at 37°C for 24-48 hrs except for RBA and PDA which were incubated at 25°C for 72-96 hrs. Cultural characteristic and biochemical tests were also carried out for further confirmation.

2.5 Aflatoxins detection

Detection and quantification of aflatoxins were performed with a High-Performance Liquid Chromatography (HPLC) (Agilent 1200 G1316A ColCom, Germany). The experimental procedure was carried out according to Saleh et al. (2009). Briefly, the dried and finely crushed bread samples were mixed with acetone and distilled water and then shaken for 30 mins. The slurry mixtures were centrifuged at 6000 rpm for 30 min. The supernatants, consisting of extracted aflatoxins, were concentrated with a concentrator (Technie, DB-3, UK) under nitrogen gas. Finally, the extracts were diluted with 300 µL (0.3 mL) mobile phase (Methanol 22.5% +Acetonitrile 22.5% + 55% Deionized water) as injecting solution. The injection volume was 30 µL, and the retention time was 15 mins.

2.6 Sensory evaluation

Sensory evaluation of the bread samples (using the attributes of taste, smell, texture and overall acceptability) was carried out by ten trained members of IFST, BCSIR, Dhaka, Bangladesh, using the hedonic scale method as described by Awasthi et al. (2012) where the score 1 represents dislike extremely, and 10 represents like extremely.

2.7 Statistical analysis

The mean and standard deviations of the triplicate analyses were calculated using GraphPad Prism 8.0 (GraphPad Software, San Diego, CA). The analysis of variance (ANOVA) was performed to determine significant differences between the means using Tukey tests.

3. Results and discussion

3.1 Nutrient composition

The results of the nutritional composition of the selected bread samples are presented in Table 1. The moisture contents of all categories of bread samples were ranged from 31.18 to 37.87%, where the moisture of the street shop bread (37.87%) was significantly higher than the local shop (31.75%) and brand shop bread (31.18%). The moisture content of food is usually used as a significant indicator of food quality, and it has a potential impact on the sensory, physical, and microbial properties of the bread (Olaoye et al., 2006). However, in Bangladesh Standards (BDS382:2016), there is no specified limit to the moisture content in bread (BSTI, 2012). Protein is an essential nutrient in the diet that plays a crucial role in the cellular maintenance, growth, repair, and functioning of the human body. According to our dietary pattern, bread is one of the major sources of...
The protein content in all categories of bread samples studied was poor in quantity and ranged from 4.67 to 5.37%, which might be due to the use of low extraction rate of refined flour in the white bread. The local shop category of bread (B) had the highest protein content (5.37%). The fat content was the highest in the brand category (A) bread (3.9%), and a similar amount was found in the local shop (B) and the street shop (C) categories of bread (4.69%). The fat content of the bread mainly depends on the addition of oil in the preparation of the dough. The ash content of the three categories of bread ranged from 1.06 to 1.43% where the street shop (C) bread had the maximum value. The ash content of bread mostly depends on the extraction rate of the flour used. In Bangladesh, there are no data available for the extraction rate of flour used in the bread. Carbohydrates are an ideal source of energy for the body because they get converted more readily into glucose and therefore expected to be high in bread. Carbohydrate was found abundant in all categories of bread within the range of 50.76 to 57.41%, where the street shop (C) category of bread had the least carbohydrate content. In Bangladesh Standards, there are no specific requirements for these proximate values of bread. The pH of the bread samples ranged from 4.92 to 5.30. The street shop (C) category of bread samples had the lowest pH value of 4.92. Only the pH values of the brand shop (A) bread samples are within the standard limits (5.30 to 6.0) of BDS 382:2016 for white bread (BSTI, 2012). The pH can influence the microbial-ecology which ultimately determines the shelf stability of the bread products (Ndife et al., 2013). Alcoholic acidity determines the shelf life of bread. It is expected to be low, and lower is the better in bread. The alcoholic acidity (0.10); however, the street shop (C) bread had the highest amount (0.25). No acid insoluble ash was detected in any category of bread samples. Acid insoluble ash indicates silica contamination.

The importance of a food item depends on its safety aspects and how much energy it can provide to a person when it is consumed by him/her. Referring to Table 1, the brand (A) and local shop (B) bread samples had a significantly higher energy value of 295 and 290 Kcal/100 g than the street shop bread samples of 263 Kcal/100 g. Regarding the proximate composition, the brand shop (A) bread showed higher compliance with the BDS382:2016 specifications.

### 3.2 Mineral composition

The results of the mineral composition of the three categories of bread samples are presented in Table 2. Calcium (Ca), Iron (Fe), Sodium (Na), and Potassium (K) were detected in all the categories of bread. The metabolic functions of minerals for life are important and have been extensively reported in many literatures (Champe and Harvey, 1994).

#### Table 2. Mineral composition of the three classes of bread samples (mg/100 g)

<table>
<thead>
<tr>
<th>Samples</th>
<th>Ca</th>
<th>Fe</th>
<th>Na</th>
<th>K</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>10.19±0.88&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.21±0.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>431.44±10.00&lt;sup&gt;a&lt;/sup&gt;</td>
<td>93.74±8.33&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>B</td>
<td>9.49±1.84&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.52±0.15&lt;sup&gt;b&lt;/sup&gt;</td>
<td>452.82±13.5&lt;sup&gt;b&lt;/sup&gt;</td>
<td>60.41±7.5&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>C</td>
<td>7.89±1.38&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.15±0.26&lt;sup&gt;a&lt;/sup&gt;</td>
<td>410.31±13.40&lt;sup&gt;a&lt;/sup&gt;</td>
<td>48.24±6.45&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are expressed as means±SD of triplicate determination. Values with different superscript within the columns are significantly different at (p<0.05). A = Brand Shop bread; B = Local shop bread; C = Street shop bread.

Fe is required in mammalian nutrition to prevent anemia and infections; enhances the body’s immune system and proper functioning of other organs of the body (Abbaspour et al., 2014). The studied bread samples contained the average Fe content ranged from 1.15 to 1.21 mg/100 g. Ca is an important constituent of body fluids and bone formation in conjunction with phosphorus (Ebuehi et al., 2007). Ca also plays a major role in nutrition, including bone formation, maintenance, and growth, tooth formation, blood clot formation, absorption of vitamin B12, and contraction of muscles (Soetan et al., 2010). All three categories of bread (Table 2) had the minimum Ca content that ranged from 7.89 to 10.19 mg/100 g. The mineral composition data (Table 2) showed that all the bread was low in Fe and Ca content. Similar results in bread have been reported by (Al-Kanhal et al., 1999). Several studies found that inadequate intake of micronutrients (minerals) has been...

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**Table 1. Nutritional composition of the three categories of bread samples**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (%)</td>
<td>31.18±1.28&lt;sup&gt;a&lt;/sup&gt;</td>
<td>31.75±1.62&lt;sup&gt;a&lt;/sup&gt;</td>
<td>37.87±1.33&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Protein (%)</td>
<td>4.67±0.72&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.37±0.58&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.72±0.98&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Fat (%)</td>
<td>5.39±0.63&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.69±0.86&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.69±0.85&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>1.06±0.09&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.07±0.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.43±0.52&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Fibre (%)</td>
<td>0.48±0.19&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.41±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.64±0.26&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Carbohydrate (%)</td>
<td>57.41±1.53&lt;sup&gt;a&lt;/sup&gt;</td>
<td>56.71±2.94&lt;sup&gt;a&lt;/sup&gt;</td>
<td>50.76±2.93&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>pH</td>
<td>5.30±0.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.99±0.22&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.92±0.19&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Alcoholic acidity (mg%)</td>
<td>0.10±0.06&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.23±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.25±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Acid Insoluble Ash (%)</td>
<td>ND&lt;sup&gt;a&lt;/sup&gt;</td>
<td>ND&lt;sup&gt;a&lt;/sup&gt;</td>
<td>ND&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Energy (kcal/100 g)</td>
<td>295±3.33&lt;sup&gt;a&lt;/sup&gt;</td>
<td>290±1.72&lt;sup&gt;a&lt;/sup&gt;</td>
<td>263±2.34&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are expressed as means±SD of triplicate determination. Values with different superscript within the rows are significantly different at (p<0.05). A = Brand Shop bread; B = Local shop bread; C = Street shop bread. ND = Not detected.
associated with severe malnutrition, increased disease conditions, and mental impairment (Alam et al., 2000). The mineral composition data showed that the mean Na content in all categories of bread ranged from 410.31 to 452.82 mg/100 g. Local shop (B) bread samples contained the highest quantity of Na which may not be suitable for hypertensive individuals. Similar results were found in bread made of white wheat flour with a sodium content of 207 to 304 mg/100 g and 217 mg/100 g, 548 mg/100 g as reported by Demirözü et al. (2003) and Ebuehi et al. (2007) respectively. Bread with reduced-sodium helps to maintain a healthy heart and circulation (Young, 2001). A maximum level of 120 mg/100 g of Na in bread is allowed (Meuser et al., 1994). The variation of Na content in the bread samples under this study might be due to the addition of different quantities of salt in the local bakeries. K content was found in the bread samples ranged from 48.24 to 93.74 mg/100 g. K and Na are minerals of concern in healthcare. The ratio of Na and K in any food item is an important factor; too much Na and less consumption of K may contribute to a high prevalence of hypertension (Saupi et al., 2009; Tanase et al., 2011).

3.3 Heavy metals contamination

The concentration of the heavy metals Lead (Pb), Cadmium (Cd), Nickel (Ni), and Arsenic (As) in the bread samples were analyzed and shown in Table 3. Pb and Cd are the most abundant heavy metals and are particularly toxic to the human body. Excessive content of these metals in food is associated with several diseases, especially of the cardiovascular, renal, nervous, and skeletal systems, and also implicated in carcinogenesis, mutagenesis, and teratogen (Magomya et al., 2013). The concentration of Pb was high in the brand shop (A) and local shop (B) bread samples (0.016 mg/kg), while it was low in the street shop bread sample C (0.014 mg/kg). The concentration of Cd, As, and Ni in the tested samples were 0.0095 - 0.001, 0.001-0.0059, and 0.03 - 0.06 mg/kg respectively. According to the BDS 382:2016 for white bread, the permissible limits of Pb, Cd, As, and Ni are 2.0, 1.0, 1.0, and 1.0 mg/kg respectively (BSTI, 2012). Although the heavy metals content of the three categories of bread samples analyzed was within the acceptable limit, but they may still have some toxic potentials with detrimental impact becoming apparent only after decades of exposure.

3.4 Microbiological contaminations

Microbiological contamination is a very serious issue for ready to eat food. Many food-borne diseases outbreak has been associated with various food-borne pathogens (Oranusi et al., 2013). Food-borne diseases are reported to be widespread in the contemporary world and responsible for about one-third of death worldwide (WHO, 2002). Although the microbial control is of great importance to the bakery industries, Bangladesh Standard still avoids the requirements of microbiological tests in bread completely. The results (Table 4) obtained for total aerobic counts were satisfactory in 90% bread samples of all categories. The total bacterial count (TBC) of the bread samples ranged from < 10 to Too Numerous to Count (TNTC) CFU/g with the highest count being recorded for street shop (C) bread (10% bread had TNTC) while the lowest counts were obtained in brand shop (A) bread (no TNTC). TBC is an indicator of the general quality of the product rather than safety. Samples having high counts of TBC may be the result of poor temperature control, improper packaging, and storage. According to the Food Standards of Australia New Zealand (FASNZ, 2001), the bacterial count of the ready to eat food above 10⁹ CFU/g are categorized as unsatisfactory and are indicative of poor hygiene or food-handling practices. The presence of Coliform and E. coli in ready to eat food is undesirable because it indicates poor hygienic conditions which have led to contamination or inadequate heat treatment (FASNZ, 2001). Coliforms and E. coli were detected in all categories of bread samples ranged from 2 to 100 CFU/g. As per the Food Standard of Australia New Zealand (FASNZ, 2001), ideally, E. coli should not be detected, and a level of <3 per gram indicates the satisfactory limit for this organism. In this study, E. coli was found above 100 CFU/g in 20% street shop (C) bread and in 10% of both brand (A) and local shop (B) bread samples which are unacceptable (FASNZ, 2001) and indicating the possible fecal contamination of food, water or food workers and poor hygienic processing practices (Oranusi et al., 2013).

![Table 3. Heavy metals contamination of the three categories of bread samples](https://example.com/table3)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Bread samples (mg/kg)</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>Maximum Limit¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pb</td>
<td>0.016±0.017</td>
<td>0.016±0.014</td>
<td>0.014±0.01</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Cd</td>
<td>0.0005±0.00</td>
<td>0.0006±0.00</td>
<td>0.001±0.00</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>As</td>
<td>0.005±0.01</td>
<td>0.001±0.00</td>
<td>0.004±0.01</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Ni</td>
<td>0.041±0.03</td>
<td>0.06±0.06</td>
<td>0.03±0.02</td>
<td>1</td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as means±SD of triplicate determination. A = Brand Shop bread; B = Local shop bread; C = Street shop bread.

Table 4. Percent distribution of microbial analysis of bread samples

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Bread samples (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A (20)</td>
</tr>
<tr>
<td><strong>Total viable count (CFU/g)</strong></td>
<td></td>
</tr>
<tr>
<td>&lt;10^3 Satisfactory</td>
<td>90(18)</td>
</tr>
<tr>
<td>10^3 Marginal</td>
<td>10(2)</td>
</tr>
<tr>
<td>&gt;10^3 unsatisfactory</td>
<td>0</td>
</tr>
<tr>
<td><strong>Yeast and mold count (CFU/g)</strong></td>
<td></td>
</tr>
<tr>
<td>&lt;10 Satisfactory</td>
<td>35(7)</td>
</tr>
<tr>
<td>10 ≤ 10^3 Marginal</td>
<td>65(13)</td>
</tr>
<tr>
<td>&gt;10^3 unsatisfactory</td>
<td>0</td>
</tr>
<tr>
<td><strong>Total Coliform count (CFU/g)</strong></td>
<td></td>
</tr>
<tr>
<td>&lt;3 Satisfactory</td>
<td>65(13)</td>
</tr>
<tr>
<td>3-100Marginal</td>
<td>25(5)</td>
</tr>
<tr>
<td>&gt;100 unsatisfactory</td>
<td>10(2)</td>
</tr>
<tr>
<td><strong>E. coli (CFU/g)</strong></td>
<td></td>
</tr>
<tr>
<td>&lt;3 Satisfactory</td>
<td>65(13)</td>
</tr>
<tr>
<td>3-100Marginal</td>
<td>25(5)</td>
</tr>
<tr>
<td>&gt;100 unsatisfactory</td>
<td>10(2)</td>
</tr>
<tr>
<td><strong>Salmonella 25ug</strong></td>
<td></td>
</tr>
<tr>
<td>Bacillus spp. (CFU/g)</td>
<td>15(3)</td>
</tr>
<tr>
<td>Aspergillus spp. (CFU/g)</td>
<td>ND</td>
</tr>
</tbody>
</table>

Figures in parentheses are the number of samples, ND = not detected

Bread mainly spoiled by mold growth due to post-baking contamination (Smith et al., 2004; Pateras, 2007). The total mold and yeast count in all categories of bread samples ranged from <1.0×10^3 to 3.6×10^4 CFU/g. The highest mold and yeast counts (3.6×10^4 CFU/g) were observed in the local bread sample. This higher level indicates potential health hazards or imminent spoilage according to Food Standards Australia New Zealand. According to the food standards, ready to eat food should be free of Salmonella as the consumption of food containing this pathogen may result in foodborne illness (FASNZ, 2001). Table 4 shows that Salmonella was found 10%, 15%, and 20% in the brand shop (A), local shop (B), and street shop (C) bread samples, respectively. Bacillus spp. was found 15% in the brand shop (A) bread samples and 10% in both local shops (B) and street shop (C) bread samples. The occurrence of Bacillus spp. in the food could be due to inappropriate processing, incomplete heating, or secondary contamination via contact with contaminated equipment and utensils (Oranusi et al., 2013). Aspergillus spp. was not detected in any studied bread sample of all categories.

3.5 Detection and quantification of aflatoxins

The aflatoxins B1, B2, G1, and G2 were analyzed in all tested samples that are summarized in Table 5. The results showed that no aflatoxin was found in any bread sample of the brand shop (A) and street shop (C) considering the retention time and detection limits (0.01 ppb) as specified for mentioned HPLC. However, aflatoxins B1 and B2 were found only in three bread samples (Figure 1) collected from the local category shops (B) which might be due to the secondary metabolism of some filamentous fungi or mold under some suitable conditions like temperature and humidity (Zain, 2011). Aflatoxin-B1 was found in one local shop bread sample with a concentration of 101.63 ppb within the specified retention time. The aflatoxin-B2 was found in another two local shop (B) test samples with a concentration of 2.21 and 5.25 ppb. Aflatoxin B toxicity as mycotoxin is closely related to hepato-carcinogenicity from Aspergillus flavus. The Food and Drug Administration (FDA) has established more specific guidelines for acceptable levels of aflatoxins in human food and animal feeds. The Codex Alimentarius Commission (1999) is considering a recommendation to establish a limit for aflatoxins in foods of 15 ppb of total aflatoxins for all foods worldwide (Saleh et al., 2009). There is no such type of limiting value specified by the Bangladesh standard. Our observed value was much higher than that of the specified limits. Now, it is clear that when aflatoxin contamination does occur at a level above the legal limit, it can lead to significant loss and may also be detrimental to health and health policy. From the processing and preservation point of view, we do agree with the opinion passed by CharmLey et al. (1995) that the extent of aflatoxins presence and concomitant economic loss is now becoming more generally recognized.

Table 5. The tabular form of the results of aflatoxin in analyzed bread samples

<table>
<thead>
<tr>
<th>Aflatoxins</th>
<th>Bread Samples A (20)</th>
<th>B (20)</th>
<th>C (20)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>Not Detected</td>
<td>Detected in one sample</td>
<td>Not Detected</td>
</tr>
<tr>
<td>B2</td>
<td>Not Detected</td>
<td>Detected in two samples</td>
<td>Not Detected</td>
</tr>
<tr>
<td>G1</td>
<td>Not Detected</td>
<td>Not Detected</td>
<td>Not Detected</td>
</tr>
<tr>
<td>G2</td>
<td>Not Detected</td>
<td>Not Detected</td>
<td>Not Detected</td>
</tr>
</tbody>
</table>

Figures in parentheses are the number of samples.

3.6 Sensory attributes

Sensory scores of all selected categories of bread samples are presented in Figure 2. The brand shop (A) samples had the maximum score for appearance (8.66), smell (8.5), taste (8.33) and texture (8.16) whereas the minimum score for appearance (5.5), smell (4.5), taste (4.5) and texture (5.17) were recorded in the street shop (C) samples. Regarding the overall acceptability of the

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Experimental samples, the maximum score (8.83) was obtained from the brand shop (A) bread samples, and a minimum score (5.03) was obtained from the street shop (C) samples. The local shop bread was obtained 7.33 scores for the overall acceptability. The sensory scores of the bread samples were significantly different (p<0.05) from each other due to the different ingredients content, baking procedures, or unique manufacturing practices (Eke et al., 2013). This observation is in conformity with the findings of Ebuehi et al. (2007).

4. Conclusion

The results of this study provide valuable information about the nutritional composition and wholesomeness of the bread sold in Bangladesh. The most consumed breakfast items, white bread, were found low in macro- and micronutrients. Bangladesh Standard Specification for white bread avoids microbiological and many other test parameters like protein, fat that are required for assessing bread quality. The Na content in all categories of bread is high which may not be suitable for hypertensive individuals. The experimental findings showed that the nutritional and microbial quality of many bread samples sold in Bangladesh is not suitable for healthy consumption. Education of the food handlers on food safety practices has become crucial, and relevant regulatory bodies in Bangladesh should establish and maintain strict hygiene conditions during bread production to safeguard the health of the consumers. It also demands further research on revising and updating the Bangladesh Standard Specification for Bread to get on with the latest industrial and technological innovations.

Conflict of interest

The authors declare that there are no conflicts of interest.

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