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ORIGINAL RESEARCH

Microbiological quality of locally manufactured bread in rural area of Bangladesh

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ABSTRACT

In Bangladesh most of local bakeries don't have Bangladesh Standards and Testing Institution (BSTI) certification and manufacture bread in non-hygienic place or with low standard raw materials. The aim of this paper is to study microbial qualities of the bread manufactured locally in rural area. These samples were analyzed for bacterial contamination, using Standard Microbiological Procedure (Morphological & Biochemical test). All sample contain huge bacterial contamination; isolated organisms were *Bacillus subtilis* and, *Bacillus cereus* in most case, but one sample contain *Staphylococcus sp.* and *E. coli* which is alarming for public health. .

KEY WORDS: *Microbiological quality, Bread quality, Public health*

Introduction

Wheat has been cultivated by man since before recorded history; Ernesto Schiaparelli, an Italian Egyptologist found bread in grave which were baked between 2435-2305 B.C. but commercial bread production by yeasts started in Ancient Egypt around 300 B.C. From the beginning bread and bakery products remaining as basic food products for human. It is a good source of nutrients, such as macronutrients (carbohydrates, protein, and fat) and micronutrients (minerals and vitamins) that are essential for human health (Malomo, Ogunmoyela *et al.* 2012). Global bread consumption raised 30.1% from 2005 through 2010, where Latin American sales grew the fastest, rising 48.5% from 2005 to 2010, followed by Africa and the Middle East (45.4%), Oceania (39.7%), Asia Pacific (37.4%) and Eastern Europe (34.4%) and it is increasing day by day according to Bakery Equipment Manufacturers and Allieds (BEMA) report (Malovany 2011). In Bangladesh, bread is second most widely consumed food product after rice and consumed rate

of bread is increasing day by day. As a results several non-registered local bakeries come in this market and manufacture bread in non-hygienic place or with low standard raw materials. Most of them don't have Bangladesh Standards and Testing Institution (BSTI) certification. In Chhatak three bakeries on 25 January 2017 (uttorpurbo24.com 2017), in Sitakunda thirty bakeries on 08 January 2017 (bd24times.com 2017), in Munshiganj fifteen bakeries on 20 June 2016 (Islam) and also in capital Dhaka four bakeries on 22 August 2016 (shadhinbangla24.com 2016) and three bakeries on 07 January 2016 (bangla.thereport24.com 2016) found guilty for using low quality raw material, toxic chemicals for color and flavors in non-hygienic conditions; are few example from several newspaper. Pathogenic microorganisms have been so far reported to contaminate a wide range of foods triggering food borne infections or intoxications including the enteric complications, abdominal pain, fever, hemorrhagic colitis, bloodstream infection, meningitis, joint infection, kidney

failure, paralysis, miscarriage, etc (Sezanur Rahman, Alamgir Kobir *et al.* 2014, Noor 2016). Indeed, the Bangladeshi people are quite prone to microbial attack due to the relatively dense population with extremely unhygienic sanitary facilities, as revealed through the propagation of various waterborne microorganisms into the surrounding environment in close proximity causing enteric diseases (Munshi, Rahman *et al.* 2012, Acharjee, Rahman *et al.* 2014, Bari and Kawasaki 2014, Marjan, Kanta Das *et al.* 2014, Alam, Feroz *et al.* 2015). Approximately 30 million people in this country are known to suffer from various food and water borne illnesses each year (Rashed 2012, Khairuzzaman, Chowdhury *et al.* 2014, Noor and Munna 2015) The aim of this study was to find bacterial contamination in bread manufactured by local bakeries in rural area (Shekpara, Jhenaidah) of Bangladesh.

Materials and Methods

Sampling

Three local bakeries Prokhor, Shahin and Makhon are selected for this study during October 2015 to February 2016 time periods. One gram of each bread sample was blended and mixed in 9ml of distilled water then, the dilution was serially made up to 1: 10,000.

Pure Culture of Bacteria

1ml of the aliquots prepared from each of the bread was transferred into Nutrient Agar containing Petri dishes for determination of cultivable microorganisms and incubated at 35°C for 24 hours. From randomly growing bacteria on this culture media we count CFU (Pollack 2011) and separate pure culture by Streak plate method into new medium for grown.

Morphological tests

All pure culture were subject to morphology test ie. Gram Staining, spore forming test, Motility test in MIU media (Food and Administration 2009).

Biochemical tests

Pure culture were subject to following biochemical test, Oxidase test, catalase test, indole production test, Methyl Red Test, Voges-Proskauer (VP) test and Glucose Fermentation Test (Kerstens and Vancanneyt 2005, Goldman and Green 2015).

Results

CFU count in sample

We found bacterial growth in all sample, average 23 CFU was recorded for 1ml sample culture after 4 times dilution of 1gm bread (Figure 1).

$$\begin{aligned} \text{Number of bacteria per ml} &= \frac{\text{CFU} \times \text{Dilution factor}}{\text{Volume of culture plate}} = \frac{23 \times 10^4}{1} \\ &= 2.3 \times 10^5 \text{ Bacteria/ml} \end{aligned}$$

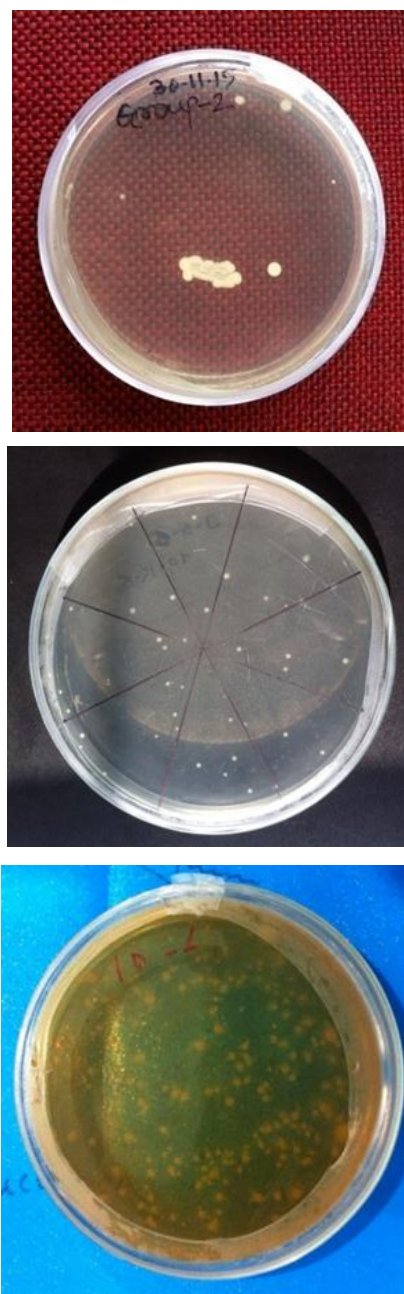


Figure 1: Bacterial culture on Media plate from different sample and different dilution factor

Bacteria identification

In this investigation, six different bacterial strains were

Table 1: Bacterial identification test for six different isolates from three different breads

Parameters	B	B1	D	D1	S	S1	
Morphology Test							
Colony features	Creamy brown	Dull gray	Yellow		Metalic	Gray	White
Cell arrangement	Rod shape	Rod shape	Clusters		Rod shape	Cocci shape	Flat, Irregular Cocci
Gram staining	+	+	+	-	+	+	
Motility	+	+	-	+	+	+	
Selective media							
Blood Agar	+	+	+	+	+	+	
Biochemical reaction							
Catalase	-	+	+	+	+	-	
Oxidase	-	+	-	-	+	-	
Indole	-	-	-	-	-	-	
Voges-Proskauer	+	+	+	-	+	+	
Methyl Red	-	-	-	+	-	-	
Glucose	+	+	+	-	+	+	
Possible Bacterial Strain	<i>Bacillus subtilis</i>	<i>Bacillus cereus</i>	<i>Staphylococcus sp</i>	<i>E. coli</i>	<i>Bacillus cereus</i>	<i>Bacillus subtilis</i>	

isolated on the basis of colony formation size, shape and color of colony from three different breads and assigned B, B1 (from Prokhor), D, D1 (from Shahin) and S, S1 (from Makhon). Then different biochemical and microbiological tests are performed with all mentioned strains (Result shown in table.1)

Discussion

The experiments performed in this research showed that huge microbiological contamination of all three samples. It's very alarming that one sample (Shahin Bread Co.) contain *E. coli* and *Staphylococcus sp* which are pathogenic for human. Other samples contained *Bacillus subtilis* and *Bacillus cereus*. Those are casual agents for Rope spoilage of bread. Pope *et.al* (Pepe, Blaiotta *et al.* 2003) isolated 61 *Bacillus* strains from ropy breads. These strains were characterized on the basis of their phenotypic and genotypic traits. All of the isolates were identified as *B. subtilis* by biochemical tests, but molecular assays revealed that besides strains of

B. subtilis, *B. licheniformis*, *B.cereus* and isolates of *B. clausii* and *B. firmus* were identified on that research (Pepe, Blaiotta *et al.* 2003). The most efficient chemicals used in bread making are propionic acid, calcium propionate, acetic acid, and calcium hydrogen phosphate can use to control contamination with *Bacillus*. A dose of 1-5 g/kg of flour leads to a delay of ropiness of 3 to 21 days (Dan 2000). The microbial isolates found were attributed to unhygienic practices, handling, health of workers and temperature as well as low moisture respectively that favored their growth/contamination. This calls for the need to legislate and enforce sanitary guidelines for bakers and retailers to curb the menace of food poison and health hazard. Proper education and public awareness should be conducted for bakery professionals.

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