

Microbial and Physicochemical Characterization of Maize and Wheat Flour from a Milling Company, Lesotho

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Abstract

Maize meal flour and wheat flour are the most widely consumed cereals in Lesotho. The present study was undertaken to investigate the physicochemical and microbiological quality of special, super and extra super brands of maize meal flour; cake, white wheat and whole brown wheat brands of wheat flour produced by a mill company in Maseru, Lesotho. Three samples of each flour were collected on each of five sampling trips during January to May 2012, making a total of 90 samples, and were analysed physico-chemically for ash, fat, protein, moisture content and pH. Microbiologically, total plate count, total coliforms, *Bacillus cereus*, *Salmonella* spp, *E. coli*, yeasts and moulds were also determined. A series of biochemical tests were done to characterize the isolated micro-flora. Moisture content ($9.89 \pm 0.76\%$ to $13.31 \pm 0.64\%$) and pH (5.83 ± 0.06 to 6.40 ± 0.12) of the flours were within recommended legal limits. However, fat content (33% (n=90)) in maize samples exceeded the maximum recommended limit. The fat content ($2.00 \pm 0.08\%$ to $5.29 \pm 0.10\%$) of all maize meal brands was significantly higher ($P < 0.05$) than that of wheat flour ($0.90 \pm 0.03\%$ to $1.87 \pm 0.02\%$). Protein content in maize meal was significantly lower than that of wheat flour, ranging from $7.01 \pm 0.91\%$ to $9.64 \pm 0.09\%$. Ash content for the samples ranged from 0.29-1.21%. Statistically, bacterial counts in the different brands of flours showed significant differences ($p < 0.05$). In a more qualitative assessment, microbes of the genera *Escherichia*, *Klebsiella*, *Aerobacter*, *Enterobacter*, *Salmonella*, *Bacillus*, *Aspergillus*, *Penicillium*, *Fusarium*, *Candida*, *Saccharomyces* and *Rhodotorula* were identified in the flours. Pathogens (*Bacillus cereus* and *Salmonella* spp) and indicator organisms (coliforms and *E. coli*) were above World Food Programme (WFP) recommended limits. This study highlighted the need for stringent cleaning and sanitation regimes to be fully implemented in such companies for consumer safety and public health protection.

Key words: Maize, Wheat, Microbiological, Milling Company

Introduction

Maize meal flour and wheat flour constitute a large part of the daily diet in both rural and urban population of Lesotho. The flour contains high proportions of carbohydrates (starch), and to a lesser extent minerals, fats and proteins (Batool et al., 2012). Out of the three major food crops (maize, wheat and sorghum) grown in Lesotho, maize is by far the largest cultivated crop - on average by 70-80% of the total population (Cownie, 1993).

However, maize and wheat from local farmers is not sufficient to feed the people in Lesotho, so milling companies supplement with grain imports from South Africa. Mukeere and Dradri (2006) reported that about 95% of maize and wheat used by milling companies in Lesotho is imported from the region, making the grain prone to contamination during the transportation, storage and handling processes.

In addition, due to the blending of cereal grain from different sources to produce flour, the physico-chemical characteristics of the flours produced in Lesotho is not documented. Lesotho has two major companies which produce maize and wheat flour at commercial scale and the companies use WFP standards to ascertain quality.

Although flour is generally regarded as a safe product due to its low water activity, a variety of pathogenic and non-pathogenic microorganisms contaminate the flour during processing (Berghofer et al., 2003 and ICMSF, 1998). Pathogens that contaminate flour survive for extended periods and produce toxins, even though their growth is retarded under low moisture conditions. Moisture content of flour ranges from 11-14% (Batool et al., 2012) but the stipulated limit is 15% (WFP, 2012). Above the limit, flour is susceptible to microbial attack which causes food poisoning and off-flavors (Shobha et al., 2011; Andah, 1976). Consequently, the storage conditions after milling and packaging are very important for the quality of the flour since they affect the shelf life and safety of the consumer.

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The quality of cereals and postharvest handling practices of the produce play an important role in the shelf life of the flour (Berghofer et al., 2003). Cereals can be contaminated with mycotoxin producing fungi and pathogenic bacteria during crop growth, pre-harvesting, post-harvesting, drying, transportation and storage (Setamou et al., 1997). Contamination adversely affects cereal yield and the quality and nutritional value of the produced flour. Mould growth is the most common cause of microbial spoilage and deterioration in the quality of cereal grain and flour during storage. Reports showed that mycotoxin producing fungi (*Apergillus spp*, *Penicillium spp*, *Fusarium spp*) and pathogenic bacteria (*Salmonella spp*, *Bacillus cereus*) can contaminate flour including its products, and the contamination levels are influenced by climatic conditions during cereal ripening and harvesting (Blackburn, 2006; Richer et al., 1993).

Refined maize meal flour and cake flour are produced (from the endosperm) by removing the outer skin of the seed kernels. The removal of the outer skin of the grain (dehulling) during milling reduces the microbial load, ash protein and fat content (Fandohan et al., 2006). This has a profound effect on the physico-chemical and microbiological quality of the flour. Flour with low ash content (biomass) – like extra super maize meal (refined) and cake flour – have reduced fat and protein content and concomitantly low microbial load (Sperber, 2007). Flour with high fat content can develop off-flavors during long periods of storage due to fatty acid oxidation especially when exposed to oxygen (Sewald and DeVries, 2002). Although wheat and maize grains are subjected to vigorous cleaning processes, not all microorganisms and toxins are removed from the final flour product because microorganisms penetrate the kernel of the grain during growth and storage (Bolade, 2009).

The maize meal flour produced by flour milling companies in Lesotho includes special (from undehulled maize), super (from partially dehulled maize) and extra super (from completely dehulled) maize meal. On the other hand, three types of wheat flour are produced which are whole (brown) wheat flour, white flour and cake flour. The high protein content flour –whole wheat flour – is used to produce crusty bread while lower protein content flour- cake flour- is used to produce softer products such as cakes, cookies and pie crusts.

Flours can be contaminated by spoilage microorganisms along the food chain and reduces the shelf life and safety of the consumer. In Lesotho there is insufficient information documented about the contaminants in flours produced by milling companies, subsequently no information is available to mitigate problems in case of food borne disease outbreak.

The main purpose of this study was to determine the microbiological and physico-chemical quality of flours produced by a milling company in Maseru, Lesotho and whether they comply with recommended legal standards

(WFP). This will provide information useful for formulating mitigation measures against food borne and related diseases.

Materials and Methods

Sample collection A total of 90 freshly milled maize and wheat flour samples were collected from a Flour Milling Company located in Maseru, Lesotho. The company produces three brands maize flour namely special, super and extra super maize meal and three brands wheat flour comprising of cake, white and whole (brown) wheat flour. One samples of each brand was collected each week for five consecutive weeks. Each sample was stored in a sterile sealed polythene bad with labels. Samples (1000 g) were collected from January to May 2013. The samples were then taken to the microbiology laboratory and stored in a refrigerator at 4 °C prior to analysis.

Physico-chemical Analyses Flour samples were analysed for ash, protein, fat, moisture and pH according to the method described in AACC (2000).

Microbiological Analyses. For microbiological analysis, flour (20 g) was suspended in 80 ml of sterile 0.1% peptone water (Oxoid CM009) and homogenized for 2 min in a sterile stomacher bag. Serial dilutions (10⁻¹ to 10⁻⁵) were prepared and plated in triplicate into each specific medium. Aliquots, 0.1 ml and 1 ml, of each dilution were used for spread plating and pour plating respectively, into the various media (AMPH, 2001). To enumeration total bacteria count (TPC), standard method agar (Acumedia 7157A) was used, incubated at 37 °C for 24 h (pour plating). Coliforms were enumerated on Violet Red Bile Agar (VBRA) (Oxoid CM0107) media, incubated at 37 °C for 24 - 48 h (pour plating). Different representative colonies based on cultural characteristics were inoculated into nutrient broth (Merck HG000C42) at 37 °C over night and streaked into nutrient slant for further analysis (Morton, 2001). Yeast and mould counts were enumerated on Potato Dextrose Agar (PDA) (Merck HG00C100) with chloramphenicol (2%) (spread plating). The plates were incubated at 25 °C for 7 days (Batool et al., 2012).

To enumerate *Salmonella spp*, flour samples (25 g each) were suspended for 2 min in 225 ml lactose broth (Merck HG00C97) and homogenized in a sterile stomacher bag and then pre-enriched for 24 h at 35-37 °C. There after, 1 ml of the pre-enriched culture was transferred to Selenite Cysteine (SC) (Merck 1.07709) broth and incubated for 18-24 h at 35-37 °C (Wallace et al.,2001). Concurrently, 0.1 ml of the pre-enriched culture was transferred into 10 ml of Heketonc Enteric (HE) agar (Acumedia 7138) plates and incubated for 24 h at 35 °C. *Bacillus cereus* was enumerated using *Bacillus cereus* agar (Oxoid CM0617) which was incubated at 37 °C for 24 h (Aydin et al., 2009) and further biochemical tests (gram staining,

Voges Proskauer reaction, gelatin hydrolysis, nitrate reduction, tyrosine degradation and lysozyme test) were done for identification of the strains.

Screening for E. coli strains The different colonies from VRBA plates were picked and inoculated on the Eosin-methylene blue (EMB) agar (Merck 1.01342). The plates were incubated at 37 °C for 24 h. Strains that exhibited green metallic sheen on EMB agar were identified as E. coli. Coliforms that were dark red colonies were further streaked on fresh VRBA for identification. Isolates were purified by repeated streaking on appropriate media and conventional biochemical tests using IMViC test, Indole (I), Methyl red (M), Voges Proskauer (V), and Citrate (C), was done as described by Stanier et al. (1987). Other biochemical tests performed included Gram reaction, Catalase formation, carbohydrate fermentation (glucose, lactose and sucrose) (Harrigan, 1998) and motility test (ICMSF, 1998).

Identification and characterization of Fungi Different isolates of yeasts and molds were transferred from Potato Dextrose Agar (PDA) to Dichloran Rose Bengal Chlorophenicol (DRBC) agar (Oxoid CM 727) and Sabourad Dextrose Agar (SDA) (Oxoid CM0139) incubated at 25 °C for 7 days for further analysis. The isolates were purified by sub-culturing and thereafter grouped according to their culturing and morphological features. Cotton blue (lactophenol stain) was used to identify unknown fungi (Cheesbrough, 2000). The identification was achieved by placing a drop of the stain on a clean slide with the aid of a mounting needle. Subsequently, a small portion of the mycelium from the fungal culture was picked, placed and evenly spread on a slide. A cover slip was gently applied with little pressure to eliminate air bubbles. The preparation was mounted and observed using a (Carl Zeiss primo star, Germany) light microscope (mag ×1000).

STATISTICAL ANALYSIS Triplicate determinations were carried out and standard errors were calculated for all results. All data collected were analyzed using one way analysis of variance (ANOVA) to determine significant differences ($p < 0.05$) among the means. All statistical tests were carried out using the “SPSS”16.0 package.

Results

Physicochemical composition of wheat flour and maize meal flour samples The physico-chemical characteristics of different types of maize meal flour and wheat flour obtained from the milling company in Maseru are depicted in Table 1.

Table 1. Physicochemical parameters of wheat flour and maize meal flour samples (n=90) obtained from the milling company in Maseru.

Sample	pH	% Moisture	% Ash	% Fat	% Protein
<i>Special maize meal</i>	6.30 ±0.05 ^a	12.3 3±0.34 ^a	1.21 ±0.11 ^a	5.29 ±0.10 ^a	9.64 ±0.09 ^a
<i>Super maize meal</i>	6.15 ±0.11 ^a	12.11 ±0.53 ^a	0.48 ±0.03 ^b	4.10 ±1.10 ^a	8.77 ±0.15 ^a
<i>Extra super maize meal</i>	6.01 ±0.08 ^a	11.90 ±0.63 ^a	0.29 ±0.008 ^b	2.00 ±0.08 ^b	7.01 ±0.91 ^b
<i>Cake flour</i>	5.8 3±0.06 ^a	9.8 9±0.76 _b	0.5 4±0.07 ^b	0.9 0±0.03 ^c	6.5 2±0.13 ^b
<i>White flour</i>	6.05 ±0.08 ^a	11.97 ±0.92 ^a	0.65 ±0.09 ^b	1.06 ±0.03 ^c	11.50 ±0.64 ^c
<i>Whole (brown) wheat flour</i>	6.40 ±0.12 ^a	13.31 ±0.64 ^a	0.71 ±0.18 ^b	1.8 7±0.02 ^b	13.76 ±0.55 ^c

*The values shown in Table 1 are mean values of the five different batches ± standard error.

*Means in the column with different superscripts are significantly different at 95% confidence limit ($P < 0.05$).

*Maximum acceptable limits (WFP 2012): Fat 2%; Moisture 14-15%; Ash 3%

*Minimum acceptable limits (WFP 2012): Protein 7-8%

The values of the physico-chemical parameters varied with batches and types of the flours collected. The pH and % moisture content of the products ranged from 5.83±0.06 to 6.40±0.12 and 9.89±0.76 to 13.31±0.64 respectively. The highest moisture contents in both maize meal flour (12.33±0.34%) and wheat flour (13.31±0.64) were found in dehulled flour samples with high percentage ash contents. Cake flour had the lowest protein content (6.52±0.13%) while whole wheat flour had the highest (13.76±0.55%). However, protein content in maize meal flour (7.01±0.91% to 9.64±0.09%) was significantly lower ($P < 0.05$) than that of wheat flour (6.52±0.13% to 13.76±0.55%). The fat content of maize meal flour (2.00±0.08% to 5.29±0.10%) was significantly higher ($P < 0.05$) than that of wheat flour (0.90±0.03% to 1.87±0.02%). Ash content for the samples (maize meal flour and wheat flour) ranged from 0.29 to 1.21%. The highest (1.21%) was found in whole maize (special maize meal flour). There was a significant difference in the physicochemical content (% moisture, % ash, % fat and % protein content) of the different types (brands) of

flour samples at $P < 0.05$. There were no significant differences in pH among the different samples.

Microbiological analysis of the flour products A variety of microorganisms were found contaminating the flour samples and these were bacteria, fungi and molds. The isolated fungi were of the genus *Aspergillus*, *Candida*, *Fusarium*, *Saccharomyces*, *Penicillium* and *Rhodotorula*; (Figure 1 and 2) while bacteria isolated belonged to the Enterobacteriaceae family (*Escherichia*, *Klebsiela*, *Enterobacter* and *Aerobacter*), *Bacillus* spp and *Salmonella* spp.

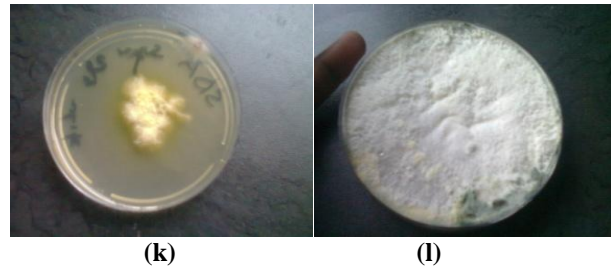
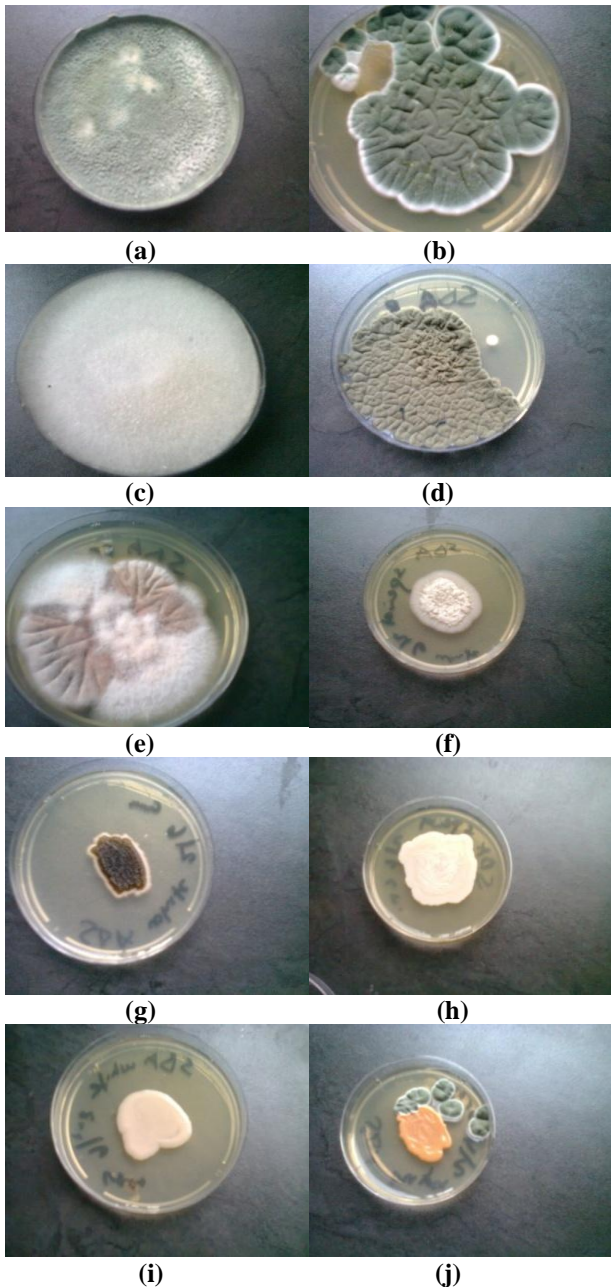


Figure 1. Different colonies of frequently isolated molds and yeasts on Sabourad Dextrose agar (a) *Penicillium*, (b) *Penicillium*, (c) *Penicillium*, (d) *Fusarium* (e) *Aspergillus*, (f) *Fusarium*, (g) *Aspergillus*, (h) *Candida*, (i) *Saccharomyces*, (j) *Rhodotorula* , (k) *Aspergillus*, (l) *Aspergillus*.

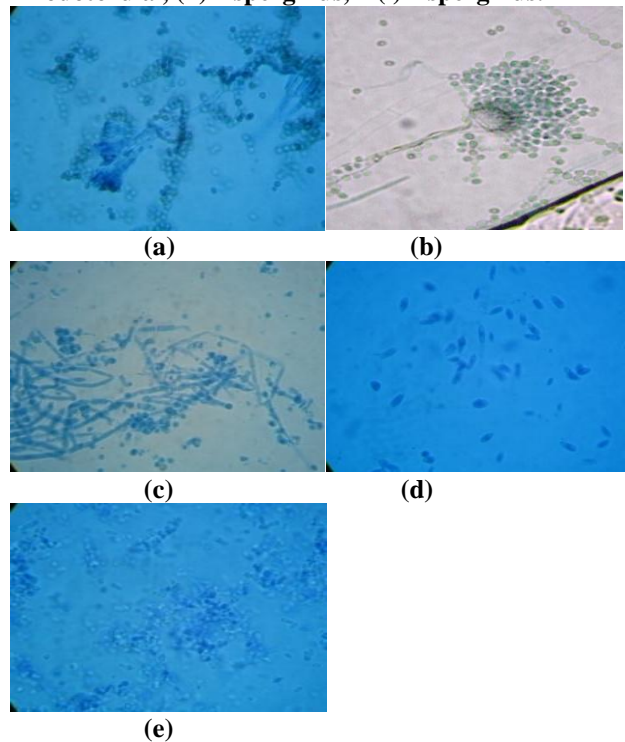


Figure 2. Microscopic morphology of fungi identified (a) *Aspergillus*, (b) *Penicillium*, (c) *Fusarium*, (d) *Saccharomyces*, (e) *Candida*.

Yeast and Moulds ranged from $\log 1.04 \pm 0.55$ cfu/g in cake flour to $\log 3.90 \pm 0.76$ cfu/g in whole (brown) wheat flour. Moulds with high isolation frequency belonged to the genera *Aspergillus* (33%) and *Penicillium* (25%) (Figure 3).

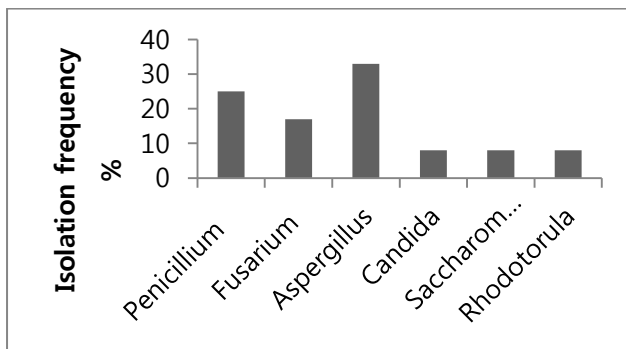


Figure 3. Isolation frequency of different genera of fungi spp in both maize meal flour and wheat flour

The level of contamination among the batches of each brand varied but did not differ significantly. However, contamination among the different types of the flour samples differed significantly ($P < 0.05$) (Table 2).

All samples analysed had levels of *E. coli* (log mean cfu/g) ranging from 3.53 ± 0.67 to 4.71 ± 0.58 with the highest counts found in special maize meal Batch No 2 (5.62 ± 0.34). For all samples, mean total coliforms (3.02 ± 0.56 for extra supper maize meal to log 3.73 ± 0.44 for whole brown flour) and mean total plate counts (3.71 ± 0.46 for extra supper maize meal to 5.95 ± 0.35 for special maize meal) were above legal limits (log 2 cfu/g) set by WFP. *Salmonella* spp were detected in 13.3 % of the samples ($n=90$) and *Bacillus cereus* counts were more than log 2 cfu/g in 90% ($n=30$) of the samples. Increase in % ash content was directly proportional to increase in microbial content of the samples (Figure 4).

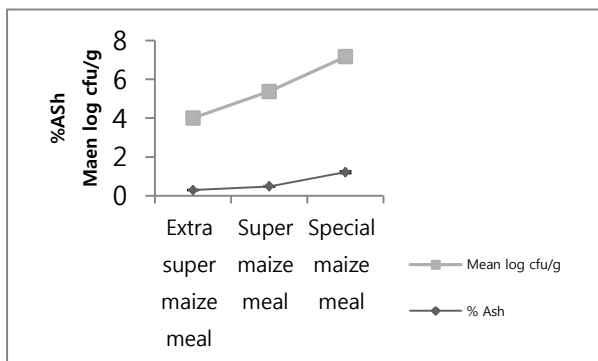


Fig 4. Relationship between ash content and total plate count of maize meal flour

Discussion

Physicochemical composition of wheat flour and maize meal flour samples Physico-chemical characteristics of maize and wheat flour are the major determinants of consumer acceptability and safety. Moisture is a very important parameter when taking into account the quality of flour and acceptability of flour products. It affects the shelf life and microbial growth during storage (Batool et al., 2012; Aydin et al., 2009;

ICMSF, 1998). According to WFP (2012) moisture content of maize meal flour and wheat flour should not exceed 15.5%. Results of the present study showed that the moisture content of between 9.89 ± 0.76 and 13.31 ± 0.64 of the flours was within recommended limits. In similar studies with wheat flour, Batool et al., 2012; Akpe et al., 2010 and Aydin et al., 2009 observed moisture content with the range 9-13%. Mahmood (2004) reported that moisture content of wheat and maize varieties depended largely on the genetic makeup and is also influenced by the agronomic and climatic conditions. Although flour has low water activity (Eyles et al., 1989), high moisture content more than the legal limit (15%), supports growth and toxin production by contaminating microorganisms. Different moisture content and pH values correlated with the level of microbial growth found on each product. Microorganisms have different moisture content and pH optima for their growth. Most bacteria grow best at about pH 7 and poorly at pH below 4 (ICMSF, 1998).

Yeasts and molds therefore thrive better in low pH food products where bacteria cannot compete (Hendrich and Bryant, 2006). The pH of the flours ranged from 5.0-6.5 (close to neutral) and the predominant microbes were bacteria, and this agreed with the findings of Hendrich and Bryant, 2006. The most contaminated products were whole wheat flour (moisture content- $13.31 \pm 0.64\%$) and special maize meal flour (moisture content- 12.33 ± 0.34) which had higher moisture content and pH values. Flour is susceptible to spoilage especially when stored improperly (in humid conditions that encourage moisture absorption) or for too long. Under such conditions, flours have been shown to develop off flavors that could result in a low quality product when used for baking (deMan, 1990). Protein content plays a very important role especially in the texture and palatability of the final product of wheat flour (deMan, 1990). To ensure good quality of the final product, the minimum requirement of protein in maize meal flour is 8.0% and maximum limit for fat is 3.0% (WFP, 2012). Maize meal samples (33% ($n=45$)) had fat content which exceeded the maximum legal limit. High fat content triggers rancidity during storage (Shobha et al., 2011; Andah, 1976), giving rise to off-flavor in the baked or cooked product from the flour.

Although protein content in wheat flour decreases during storage (Sur et al., 1993), it is a very important parameter when considering the quality of the wheat and the stability of the flour products. The results of the current study corresponded to studies by Ekinic and Unal (2003) and Aydin et al., (2009), with protein contents of wheat flour ranging from 7- 13.5. High protein content in maize meal, above 8% dry weight, and above 11% in wheat flour is an indication of good quality flour. With the exception of cake flour, all the flours were of good quality in terms of protein content. Ash content is an indication of bran in wheat or husks in maize (Ekinic and

Unal, 2003). It also indicates the level of mineral composition in the flour. The maximum recommended limit for ash in flour is 3% (WFP, 2012). All the samples had percentage ash content which was within the recommended limits and the results were agreed with results of other researchers (Batoool et al., 2012; Aydin et al., 2009; Bolade, 2009). Whole maize meal flour and wheat (brown) flour had the highest pH, Moisture, Ash, Fat and Protein content. The study showed that physico-chemical parameters of the flours decreased with increase in the level of dehulling of maize and wheat grains. This also reduced microbial load in the flour samples (Figure 4).

Microbiological analysis of the flour products Products of maize meal flour and wheat flour constitute a large part of the daily Lesotho diet in the form of bread and papa (thick paste porridge). Flour is known to be a safe commodity due to its low water activity, but microbial isolation and enumeration in this study revealed presence of important populations of microorganisms in higher counts than recommended legal limits. Research has shown that very few cases have implicated maize and wheat flour product to food borne disease outbreaks (Aran and Eke, 2005; Batoool et al., 2012). Studies on the microbiological composition of maize meal and wheat flour in Lesotho are very valuable from the view point of health risk assessment from the consumption of flour. Aran and Eke (2005); Batoool et al. (2012) and Akpe et al. (2010) found similar results of microbial contamination of flour. They further characterized the fungi and bacteria to species level and the predominant and frequently isolated fungi and bacteria were *Aspergillus niger* (48.4 %) and *Klebsiella pneumoniae* respectively. In the current study *Aspergillus* (33%) and *Penicillium* (25%) were frequently isolated from the flours (Figure 3). Fungi in maize and wheat flours constitute a divergent group of microorganisms consisting of several species. The level of contamination of flour by yeasts and moulds is of paramount importance when considering the quality and safety of food. The maximum legal limit for fungi in flour is log 5 cfu/g (WFP). Yeasts and moulds from the results (Table 2) were within recommended limits. Higher level of fungi more than the legal limits deteriorates the quality of food and causes food borne diseases. Food decomposition occurs due to the presence of moulds and many species of moulds are known to produce mycotoxins (Bullerman, 1997). Diseases caused by mycotoxin producing fungi include gangrenous form of ergotism and cancer of the reproductive system in women and children (Payne, 1992). Some species of yeasts and moulds are known to infect and cause diseases in humans (Bullerman, 1997). *Fusarium* spp. are a major (Ryan and Ray, 2004) and that is why its presence in processed food indicates the ineffectiveness of food processing and some strain are harmful to humans and cause foodborne illness. Berghofer et al. (2003) and Eyles

problem in agriculture as they contaminate cereals prior to harvesting (Eyles et al., 1989).

High isolation frequency of *Aspergillus* and *Penicillium* (Figure 4) from the flour samples can be attributed to poor handling of the raw material along the food chain. Storage moulds such as *Aspergillus* and *Penicillium*, and the soil moulds; *Fusarium* spp., penetrate well into the kernel of grain and produce toxins which can be difficult to remove during food processing. Therefore, the absence of microscopic fungi mycelium does not indicate that a product does not contain mycotoxins. On the contrary, the presence of toxin producing fungi in food products is not a ground to assume that they contain mycotoxins. The identified fungi are important in food safety as they have been reported to produce mycotoxins which have varying implications for health and the economy especially in developing countries like Lesotho (Akpe et al., 2010).

All flours analysed had levels of *E. coli* higher than WFP legal limits (log 1 cfu/g). Studies done in Australia and Pakistan (Aydin et al., 2009; Berghofer et al., 2003) showed results similar with results of the present study of total plate and coliform counts ranging from 102 to 105 cfu/g. Coliform count, TPC and *E. coli* are important as they give an indication of the hygienic properties of the food. TPC and coliform count indicate effectiveness and efficiency of the food chain process and gives information regarding the shelf life and organoleptic changes during storage of the food stuff (Batoool et al., 2012). Higher Coliform count, TPC and *E. coli* counts more than the legal limits indicate poor sanitation and/or problems with the process control and handling of the raw materials and their products. However, unlike yeast and mould counts the significance of coliform count, TPC and *E. coli* counts for raw products which are processed before consumption in the flour is largely determined by the intended use. *Escherichia coli* O1157: H7 can cause fatal illness when present at levels as low as 10 per gram of food (WFP, 2012). According to WFP (2012) food standards, *Salmonella* spp. should not be detected in 25g of food samples. WFP (2012) reports state that *Salmonella* spp. in food stuff accounts for more than 50 % of all food poisoning cases. A research done by Richter et al. (1993) reported very low number of *Salmonella* spp. (1.32% (n=3040)) in wheat flour. *Salmonella* spp. causes diseases such as typhoid, paratyphoid and food poisoning. Certain strains of *Bacillus cereus* are known to cause food poisoning. The *Bacillus cereus* counts were above the recommended limit of log 2 cfu/g. The bacteria is found in stools and is common in soil, hence food such as cereals often contain these organisms during pre- and post harvesting stages. These organisms can produce spores and can survive heat processing et al. (1989) reported very low *B. cereus* levels in flour (0.3MNP/g and 4.2% n=142).

Relationship between ash and microbial count Flours with low bran or ash content typically have low microbial

content (Sperber, 2007) due to the milling process (dehulling) which concentrate in excess of 90% of total microorganisms present onto the maize husk and wheat bran. Extra super maize meal flour had the least ash content and microbial count. As cereal grain layers are separated, surface-adhering contaminants are concentrated in the bran or husks and removed from the maize and wheat germ and pollard (Sperber, 2007). The inner semolina fraction contains low microbial counts, and it is the cleanest end product of the milling process. Removal of bran in wheat and husks in maize reduces the ash (biomass) of the final product concomitantly reducing the microbial content, thus improving the safety of the product. However, essential minerals and nutrients are lost as the ash content of maize and wheat grains is reduced but this can be solved by food fortification. High microbial count found in the study might be due to microorganisms already present on the cereal grains from which the flour was obtained, the method of milling and the milling machine used. Ottogalli and Galli (1979); Berghofer et al. (2003), and Aydin et al. (2009) found bacteria on cereal grains and in flour mill products.

Water used during the milling processes is also a source of contamination. Residue build-up in milling machines could also constitute a significant source of microbiological contamination (Berghofer et al., 2003) thereby jeopardizing the safety and health of the consumer.

This study showed that maize meal flour and wheat flour produced by a milling company in Maseru, Lesotho was highly contaminated with different microorganisms. Higher levels, more than legal limits of *E. coli*, *B. cereus* and total coliforms in the flour compromise the safety, storage and organoleptic characteristics of the final product. Presence of *Salmonella* spp in 13.3% of the samples rendered the flour unsafe for human consumption. Fungi contamination was within legal limits (below log 5 cfu/g), making the flours safe from mycotoxin poisoning. The content of protein, fat and ash in the flour was high enough to supply the daily requirements in the diet. However, more stringent measures in terms of microbial contamination need to be put in place by the milling company to protect consumers from food borne and related illnesses.

Table 2. Microbial analysis (log₁₀ cfu/g) of the different flour samples.

<i>Sample and Batch N^o</i>	Total plate count	Total coliforms	<i>Bacillus cereus</i>	<i>Salmonella spp</i>	<i>Esherichia coli</i>	Yeasts and Moulds
<i>Special maize meal</i>						
1	5.35±0.02	3.41±0.07	4.11±0.95	1.51±0.34	4.10±0.55	2.78±0.94
2	6.50±0.71	3.91±0.78	3.90±0.08	ND	5.62±0.34	3.12±0.34
3	6.12±0.11	3.56±0.49	3.90±0.94	0.13±0.09	5.33±0.99	3.00±0.67
4	5.90±0.90	3.76±0.19	4.00±0.57	ND	4.00±0.07	2.93±0.09
5	5.89±0.03	3.40±0.12	3.90±1.10	ND	4.49±0.93	3.11±0.75
Log mean (x ± Sx)	5.95±0.35^a	3.61±0.33^a	3.96±0.73^a	0.82±0.22^a	4.71±0.58^a	2.98±0.56^a
<i>Super maize meal</i>						
1	5.15±0.32	3.51±1.07	4.10±0.09	ND	3.11±0.35	3.11±0.06
2	5.30±0.51	3.21±0.97	3.90±0.78	0.15±0.007	3.82±0.49	3.26±0.48
3	5.10±0.91	3.26±0.56	4.01±0.44	ND	4.13±0.75	3.77±0.76
4	4.00±0.70	3.00±0.35	3.20±0.66	ND	3.92±0.77	2.07±0.56
5	4.89±0.93	3.27±0.16	4.09±0.10	ND	3.61±0.25	2.00±0.25
Log mean (x ± Sx)	4.89±0.67^b	3.25±0.62^a	3.86±0.41^a	0.15±0.007^b	3.72±0.52^b	2.84±0.42^a
<i>Extra super maize meal</i>						
1	4.15±0.05	2.70±0.67	3.45±0.34	ND	3.10±0.15	2.48±0.49
2	4.50±0.61	2.48±0.06	2.30±0.31	ND	3.22±0.04	2.78±0.66
3	3.62±0.21	3.08±0.79	3.72±0.79	ND	3.03±0.79	2.70±0.52
4	3.07±0.99	3.89±0.90	2.17±0.11	ND	2.80±0.57	2.11±0.08
5	3.19±0.43	2.97±0.36	1.29±0.28	ND	2.99±0.74	2.30±0.97
Log mean (x ± Sx)	3.71±0.46^c	3.02±0.56^a	2.59±0.37^b	ND	3.03±0.46^b	2.47±0.54^a
<i>Cake</i>						

<i>flour</i>						
1	4.35±0.78	3.61±0.12	5.11±0.05	ND	3.23±0.86	1.04±0.55
2	4.27±0.63	3.51±0.78	4.15±0.08	ND	4.12±1.30	2.11±0.77
3	4.14±0.19	3.26±0.89	4.14±1.04	ND	3.11±0.19	2.31±0.97
4	4.90±0.21	3.23±0.15	3.99±0.50	ND	3.41±0.90	2.03±0.59
5	4.88±0.08	3.08±0.32	4.05±1.10	ND	3.80±0.08	2.47±0.45
Log mean (x ± Sx)	4.51±0.38^b	3.33±0.45^a	4.29±0.55^c	ND	3.53±0.67^b	1.99±0.67^b
<i>White flour</i>						
1	4.44±0.89	3.51±1.09	4.21±0.05	ND	4.02±0.59	2.78±0.71
2	4.84±0.79	3.31±0.06	4.06±0.89	ND	3.64±0.84	1.12±0.04
3	5.17±0.14	3.36±0.19	4.37±0.04	ND	3.15±0.97	2.00±0.18
4	4.08±0.77	3.36±0.17	4.00±0.17	ND	4.07±0.67	2.93±0.05
5	4.80±0.09	3.30±0.56	4.50±1.11	ND	4.21±0.43	2.11±0.55
Log mean (x ± Sx)	4.57±0.54^b	3.37±0.41^a	4.23±0.45^c	ND	3.82±0.70^b	2.19±0.31^a
<i>Whole (brown) wheat flour</i>						
1	4.51±0.17	3.54±0.78	4.16±0.33	ND	3.67±0.62	1.78±0.04
2	5.00±0.61	3.98±0.07	4.00±1.32	0.07±0.01	4.18±0.66	2.22±0.27
3	5.72±0.18	3.23±1.06	4.09±0.94	ND	4.27±0.09	3.90±0.76
4	3.89±0.65	4.04±0.19	5.04±0.88	ND	4.01±0.12	2.13±0.09
5	4.79±0.54	3.86±0.12	5.07±1.56	ND	4.45±0.31	3.85±0.54
Log mean (x ± Sx)	4.78±0.43^b	3.73±0.44^a	4.47±1.01^a	0.07±0.01^c	4.12±0.36^a	2.78±0.34^a

*The values shown in Table 2 are mean values ± standard error.

*Log means values in the column with different superscripts are significantly different at 95% confidence limit (P<0.05).

*ND: Not detected

*Maximum acceptable limits (WFP 2012): TPC log 5 cfu/g; Coliform log 2 cfu/g; *Salmonella spp.* 0 per 25 g; *E. coli* log 1 cfu/g; *Bacillus cereus* log 2 cfu/g and Fungi log 3 cfu/g

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