MICROBIAL STATUS OF FRESH BEEF SAUSAGE SOLD IN TRIPOLI

Naas, H.T.¹, Garbaj, A.M.¹, Eshamah, H.L.¹ and Abolghait, S.K.*¹&²

¹Department of Food Hygiene and Control, Faculty of Veterinary Medicine, Alfateh University, Tripoli, Libya and,
²Department of Food Hygiene and Control, Faculty of Vet. Med., Suez Canal University, Ismailia, Egypt

ABSTRACT

A study on microbial quality of commercially available fresh beef sausage in Tripoli city was conducted. A total of 21 samples were investigated for total aerobic plate count, psychrotrophs, staphylococci, enterococci, coliforms and yeast & mould.

The mean of total mesophiles and psychrotrophes counts were >10⁹ cfu/g. Tested samples contained both of enterococci and total staphylococci at rate of >10⁶ cfu/g where only 5% of the tested samples contained >10⁵ cfu/g of Staphylococcus aureus. Moreover, >10⁶ MPN/g of coliforms were found in all samples. On the other hand, yeast count were >10⁷ cfu/g while only 30% of the samples contained mould at rate of >10³ cfu/g. Thus all of the tested samples contained higher mean counts of the microbial contaminants than the suggested Libyan standard specifications for fresh beef sausage. Public health importances as well as factors that may limit such high microbial counts in fresh beef sausage were discussed.

*Corresponding author
s.aboelghait@scuegypt.edu.eg

INTRODUCTION

Fresh meat and its products such as “fresh beef sausage” are easily contaminated with variety of the microorganisms. It also supports their growth if not properly handled, processed and preserved. A variety of sources including air, water, soil, feces, feed, hides, intestines, lymph nodes, processing equipment, utensils and handlers contribute to the microbial populations of the sterile muscles of healthy animals during slaughter, fabrication, and further processing and handling [1].

The bacteria normally found on the surface of meat are distributed throughout the entire product during the mincing and mixing pr-
ocess used to produce raw fresh beef sausage.

Fresh beef sausage a perishable food forms a new portion of the diet of the Libyan people.

Consumers expect wholesome meat products for consumption when handled and cooked properly. However, like any other raw food, fresh beef sausage may be contaminated during production, processing, storage and marketing with biological agents that may be hazardous to human health. Studies worldwide have shown that *Salmonella*, *E. coli*, *S. aureus*, *Listeria* spp., *Campylobacter* and other pathogenic bacteria are often present in fresh beef sausage. There is a shortage in the data concerning the prevalence of contamination with pathogen and other hygienic indicator microorganism of fresh beef sausage in Libya. This study was conducted to investigate the prevalence and levels of bacteria of public health and product spoilage concern on fresh beef sausage in Tripoli city, Libya.

**MATERIAL & METHODS**

A total of 21 fresh beef sausage samples were randomly purchased from various butchers and markets in Tripoli city. The total aerobic plate count (APC), psychrotrophic plate count, staphylococci, enterococci, yeast and mould counts were conducted using the pour-plating technique and recorded as colony forming units (cfu) per gram. Total coliforms count (TCC) was determined using Most Probable Number (MPN) technique which estimate the bacterial population density per gram.

For this purpose, a 10 g fresh beef sausage sample was transferred to a sterile plastic bag under aseptic conditions, 90 ml. of peptone water were added. then diluted to a $10^{-1}$ dilution with Peptone Water (Oxoid CM 9, UK) and stomached for 2 min by using a Stomacher (Interscience-Bag Mixer 400). Following homogenization, ten-fold serial dilutions for each sample were made in sterile peptonesalt water up to $10^{-7}$. Each of these dilutions was inoculated to specific culture media for enumeration of APC (Plate Count Agar, Oxoid CM 325, UK), staphylococci (Baird Parker Medium, Oxoid CM 275, UK), enterococci (Slanetz and Bartley Medium, Oxoid CM 377, UK), yeast and mould (Sabouraud's Dextrose Agar and Malt Extract Agar). APHA (1992).

Colonies on plates were manually counted as colony forming units per gram of sample (cfu/g). The minimum, maximum and mean of these records were tabulated and statistically analyzed to calculate the standard deviation and standard error (SE) using Microsoft excel 2003. These microorganisms were identified after isolation by Gram staining and appropriate biochemical tests. (Sharf, 1974 Speck, 1976;
For the detection of TCC, 3 fermentation tubes were set up each containing 1, 0.1, 0.01 g sample in 10 ml Lactose Broth (LB-Oxoid CM 137) as a preenrichment step. The tubes were incubated at 37°C for 24-48h. After incubation, a loop-full of the inoculum from the each tube with turbidity and gas production was transferred to Brilliant Green 2% Bile Broth (BGBB-Oxoid CM31 UK) and incubated at 37°C for 24-48h for a selective enrichment step. After incubation, BGBB tubes with turbidity and gas production were used to detect TCC. To identify E. coli, each positive BGBB tube was streaked onto Endo Agar Base (Oxoid CM 479, UK).

After incubation, colonies on the endo agar base were evaluated for E. coli and finally, presumptive E. coli colonies were chosen and subjected to IMVC (Indol, Methyl red, Voges Proskauer and Simmon Citrate tests). (Buchanan & Gibbons, 1974; and Banwart, 1979 Ramdet, 1975).

**RESULTS & DISCUSSION**

The present study revealed that the retail fresh beef sausage obtained from butchers and supermarkets in Tripoli city was contaminated with enteric and spoilage bacteria as well as yeast and mould.

The results of the microbiological study of the samples showed that the mean number of APC – represented by Mesophiles– was $2.1 \times 10^9 \pm 3.4 \times 10^8$ cfu/g (Fig. 1 and Table 1). The APC gives an indication of the gross microbial content of the food and the likelihood of spoilage if the food is not maintained under appropriate conditions. The mean total viable count usually significantly greater in processed meat products than the whole cuts & meat; that is referred to the nature of the extra processing; higher APC than whole cuts of meat.

It was observed that the APC in fresh beef sausage higher than the findings reported by earlier investigations in other geographical regions as it was reported that over 75% of 88 ground beef samples contained $10^5$ total mesophilic microorganisms per g. [2].
Table (1): Bacteriological analysis of fresh beef sausage samples N.=21

<table>
<thead>
<tr>
<th>Statistics</th>
<th>Mesophiles</th>
<th>Psychrotrophes</th>
<th>Enterococci</th>
<th>Total Staphylococci</th>
<th>Staphylococcus aureus</th>
<th>Coliforms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Min.</td>
<td>1.2x10^8</td>
<td>1.1x10^7</td>
<td>5x10^5</td>
<td>1.1x10^7</td>
<td>2x10^5</td>
<td>2.4 x10^5</td>
</tr>
<tr>
<td>Max.</td>
<td>7.4x10^9</td>
<td>5.6x10^9</td>
<td>5.9x10^7</td>
<td>8.3x10^7</td>
<td>2x10^5</td>
<td>9.2x10^6</td>
</tr>
<tr>
<td>Mean</td>
<td>2.1x10^9</td>
<td>1.5x10^7</td>
<td>9x10^5</td>
<td>6.2x10^5</td>
<td>2x10^5</td>
<td>2x10^6</td>
</tr>
<tr>
<td>S.D.</td>
<td>2x10^9</td>
<td>1.4x10^9</td>
<td>1.2x10^7</td>
<td>1.7x10^7</td>
<td>0</td>
<td>2.3x10^5</td>
</tr>
<tr>
<td>S.E.</td>
<td>3.4x10^8</td>
<td>2.4x10^8</td>
<td>2.1x10^6</td>
<td>3x10^6</td>
<td>0</td>
<td>3.9x10^5</td>
</tr>
</tbody>
</table>

The study revealed samples contained 1.5x10^9 ±2.4x10^8 cfu/g psychrotrophes and the coliforms count mean attained to 2x10^6 ±3.9x10^5 MPN/g (Table 1 and Fig. 1).

The presence of these bacteria indicates that general hygiene, especially the high level of APC and psychrotrophes counts, affects the shelf-life of fresh beef sausage.

Psychrotrophic spoilage bacteria such as *Pseudomonas* are generally not harmful but they cause food deterioration & quality loss by getting mouldy, developing a bad odor, or feeling sticky on the outside. In addition, several investigators have reported that correlation coefficients of aerobic plate, total coliforms counts, and *E. coli* counts with *Salmonella* incidence were higher [Sofos et al.,1999; Siragusa et al.,1998].
Several studies found coliforms and *E. coli* in ground beef products [2,5]. Coliform bacteria are indicator organisms for intestinal enterobacteriaceae. Indicator organisms may be employed to reflect the microbiological quality of foods relative to its shelf life or their safety from foodborne pathogens.

Microbial indicators are more often employed to assess the wholesomeness and sanitation than quality. In this study microbial contamination of fresh beef sausage with coliforms was detected in all of the samples which contained coliforms $>10^6$ MPN/g.

When microbiological testing is done, it is generally recommended that levels of generic *E. coli* also be assessed as a direct indicator of faecal contamination. Enteric pathogens such as *E. coli* O157:H7 and *Salmonella* are present sporadically and this makes routine monitoring for their presence impractical. In addition, methods for the quantitative recovery of these pathogens are lacking. In situations where the level of generic *E. coli* is increased, the risk of these pathogens being present is also considered to be increased. Therefore, the level of generic *E. coli* may be used as indicative of the extent of faecal contamination and hence an indicator of increased potential risk posed to the consumers especially when many of ground beef products have been found positive for *E. coli* O157:H7.

The ratio of levels of different pathogens as *E. coli* O157:H7 versus generic *E. coli* may not be fixed [6].

Enterococci and total staphylococci were found at rate of, $9 \times 10^6 \pm 2.1 \times 10^6$ and $6.2 \times 10^6 \pm 3 \times 10^6$ cfu/g respectively in all tested samples. However only, 5% of the samples contained $5.86 \times 10^5 \pm 1.63 \times 10^5$ cfu/g of *Staphylococcus aureus*.

In general, staphylococci are expected to exist, at least in low numbers, in all food products that are of animal origin or in those handled directly by humans, unless they are heat treated. The present study revealed that samples contained $>10^7$ cfu/g staphylococci and only one sample (5 % of the test samples) contained $2 \times 10^5$ cfu/g of *Staphylococcus aureus*. Though the mean staphylococcal count was significantly lesser. Many authors reported that coagulase positive staphylococci ranged from $10^2 - 10^4$/g and the isolation ratio was 2.3 - 16.6 % (Siriken, 2004).

Yeast and mould were estimated totally in all samples as $9 \times 10^8 \pm 2.1 \times 10^7$ cfu/g where all the tested samples contained yeast at level of $8.8 \times 10^7 \pm 2.1 \times 10^7$ while only 30% of the samples contained mould $2.3 \times 10^6 \pm 2.7 \times 10^5$ cfu/g (Table2 and Fig. 2).
Table (2): Mycological analysis of fresh beef sausage samples (N. =21)

<table>
<thead>
<tr>
<th>Statistics</th>
<th>Total Yeast and Mould cfu/g</th>
<th>Yeast cfu/g</th>
<th>Mould cfu/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Minimum</td>
<td>4.3x10^6</td>
<td>4.2x10^6</td>
<td>1x10^5</td>
</tr>
<tr>
<td>Maximum</td>
<td>5.2x10^8</td>
<td>5.2x10^8</td>
<td>4x10^6</td>
</tr>
<tr>
<td>Mean</td>
<td>9x10^8</td>
<td>8.8x10^7</td>
<td>2.3x10^6</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.2x10^8</td>
<td>1.2x10^8</td>
<td>1.5x10^6</td>
</tr>
<tr>
<td>S.E.</td>
<td>2.1x10^7</td>
<td>2.1x10^7</td>
<td>2.7x10^5</td>
</tr>
</tbody>
</table>

According to Libyan National Center for Standardization and Metrology (LNCSM, [http://www.lncsm.org.ly](http://www.lncsm.org.ly)), regarding microbiological quality of fresh beef sausage; the maximum number of APC must not exceed 10^6-10^7 (3 out of 5 samples), S. aureus 5x10^2 - x10^3 (2 out of 5 samples), while E. coli O157:H7 and Salmonella must be absent (n=5), the present study results showed very high number of APC, S. aureus and E. coli.

In conclusion, the microbiological quality of the fresh beef sausage analyzed was unsatisfactory, and the product might be an important cause of food poisoning.

Therefore, preventive measures including warning consumers of the health risks associated with eating raw fresh beef sausage and encouraging them to thoroughly cook fresh beef sausage and to adhere to safe food handling guidelines should be implemented.

In general, the microbiological quality of meat products, purchased by the consumers, is dependent on number of factors, such as the quality of the raw materials, other materials used or added during processing operations, efficacy of cooking process and whole of the sanitary measures adopted.
REFERENCES:

**American Public Health Association (1992):** Compendium of methods for the microbiological examination of foods, 3rd Ed., APHA TECHNICAL Committee on Microbiological methods for foods Washington, DC., US.

**Banwart, G. (1979):** Basic food microbiology, AVI publishing Go., Westpert, CT., US.


**Buchanan, R. and Gibbons, N. (1974):** Bergey,s manual of determinative Bacteriolog, 8th Ed., Williams& Wilkins, Baltinore, US.


**Speck, M.L. (1976):** the compendium of methods, for the microbiological examination of foods. Am. public Health Assoc., Washington, D.

الملخص العربي

الحالة الميكروبيولوجية للسجق (المرقاز) البقرى الطازج المتداول في أسواق مدينة طرابلس الغرب

هشام الطاهر النعاس، أبو بكر محمد قرباج، حنان لطفي الشماح، سعيد كمال أبوالغيط
قسم الرقابة الصحية على الأغذية، كلية الطب البيطري، جامعة القناة، طرابلس، ليبيا.
قسم الرقابة الصحية على الأغذية، كلية الطب البيطري، جامعة قناة السويس، الإسماعيلية، مصر.

تم فحص عدد 21 عينة من السجق (المرقاز) البقرى الطازج المتداول في أسواق مدينة طرابلس وذلك لغرض الفحص الميكروبيولوجي وتحديد عدد الميكروبات لتقييم جودة هذا المنتج. وحددت هذه الدراسة العدد الكلي للميكروبات المحبة لدرجة الحرارة المعتدلة والمنخفضة (بمتوسط < 10⁴)، العدد الكلي للمكورات المعوية (بمتوسط < 10⁵)، العدد الاحتمالي للمكورات العنقودية (بمتوسط < 10⁶) وكذلك العدد الكلي للفطريات والخمائر (بمتوسط < 10⁷) لكل جرام من المنتج. في حين كان عدد العينات الموجب للميكروب العنقودي الذهبي هو عينة واحدة (5%) وكان متوسط العدد لهذه العينات < 10⁵. وقد كانت معظم العينات غير مطابقة لمتطلبات المواصفة القياسية المقررة للعمل بها في ليبيا لمثل هذا المنتج. هذا وقد تمت مناقشة النواحي الصحية والعوامل التي قد تحد من تلوث المنتج بمثل هذه الأعداد من الميكروبات.