

**Microbiological Analysis of Beef Meat collected at different hours of the day  
in Ekpoma Town Market**

**P.I. OKOH**

**Department of Animal Science, Faculty of Agriculture, Ambrose Alli University Ekpoma,  
Edo State, Nigeria.**

**Corresponding Author's Email: Paulokoh75@gmail.com**

**Abstract**

A study on the microbiological analysis of beef meat collected at different hours of the day in Ekpoma market was carried out. 1Kg of meat sample was purchased from the market at the different times (8am, 12noon and 5pm) respectively. A part was cut into ten samples of ten grams each, which served as replicates. They were put in a clean polythene bag, labeled accordingly, and taken to the laboratory for microbiological analysis. Ten grams of the meat samples were weighed and homogenized into 90mls of sterile distilled de-ionized water, using a sterile warring blender and ten folds dilution of the homogenates was made using sterile pipettes. Mean counts of total proteolytic bacteria count (TPC), total viable count (TVC), Coliforms, *Salmonella* spp, *Pseudomonas* organisms and *Bacillus* spp, were all determined in the collected samples. Results from the study showed that total proteolytic bacteria count (TPC) was significantly ( $P < 0.05$ ) higher in samples collected at 5pm having 7.867, compared with those collected at 12noon and 8am having 6.050 and 5.267  $\log_{10}$ CFU/g respectively. Total viable count (TVC) were significantly ( $P < 0.05$ ) lesser at 8am having 4.517, compared with 12noon and 5pm which recorded 5.520 and 7.723  $\log_{10}$ CFU/g respectively. Also, coliforms counts were significantly ( $P < 0.05$ ) higher at 5pm when compared at 12noon and 8am, while *Pseudomonas* count recorded significantly ( $P < 0.05$ ) lesser value at 8am having 1.193, compared with 12noon and 5pm which had 2.500 and 3.557  $\log_{10}$  CFU/g respectively. Total *Bacillus* counts also recorded a significantly ( $P < 0.05$ ) lesser values of 1.100 at 8am compared with 1.823 at 12noon and 3.030 at 5pm, while those of *Salmonella* spp. recorded significantly ( $P < 0.05$ ) higher value of 3.030 at 5pm compared with 3.283 and 2.937  $\log_{10}$ CFU/g recorded at 12noon and 8am respectively. This shows that microbiological analysis of beef meat were higher as the time (hours) of the day progressed. **In conclusion**, meat should be bought from the market in the early hours (8am) of the day.

**Key words:** Beef, Microbial analysis, Time(hours), Market.

## Introduction

Meat has long been known for its nutritive composition, which could explain why it is being consumed by many people worldwide. In many developing countries especially Nigeria, meat is widely consumed as source of protein. It is either eaten cooked or processed into other forms to avoid associated spoilage (Olaoye and Nilude, 2010).

However, animal proteins such as meat and meat products as well as fish and fishery products are generally regarded as a high risk commodity to infection and toxication (Sulley, 2006). The food borne infection and consequent illnesses are some of the major international challenges that lead to high mortality and economic loss (CDS, 2008).

A great diversity of microbes inhabit fresh meat generally but different types may become dominant depending on the pH, composition, texture, storage temperature and transportation means of the raw meat (Ercoliniet *al.*, 2006 and Adu – Gyamfiet *al.*, 2012). Basically, two types of microbial contaminants can be expected in meat, i.e. spoilage bacteria, which are those microorganisms that spoil the product and render it unfit for human consumption, and pathogenic bacteria, which are the microorganisms that produce diseases.

Major spoilage organisms in raw meat include *Pseudomonas* spp., others may include *Shewanella*, *Bronchothrix*, and members of the *Enterobacteriaceae*. While, pathogenic microbes may include *Salmonella* spp., *Campylobacter jejuni*, *Bacillus cereus*, *Clostridium perfringes* and *C.botulinum*, *Yerisiaenterocolitica*, *Escherichia coli*, *Staphilococcus aureus* and to some extent, *Listeria monocytogenes*.

There are also reports (Turtura, 1991) on *Coliforms* spp identified in meat to be *C. freundii*, *E.coli*, *Enagglomeram* and less frequent strains are of the genera *Klebsiella*, *Shigellasonnie* and *Proteus*. While *E. coli*, and *S. aureus* are normal flora in humans and animals and their presence in foods are indications of excessive human handling (Clarence *et al.*, 2009).

Doyle (2007), reported that the microbial quality of ground meat analyzed was unsatisfactory and the product was an important cause of food poisoning. Similarly, Hassan *et al.*, (2010) reported that irrespective of the site of collection of beef, the bacterial count was high in samples incubated at room temperature (20 – 25<sup>0</sup>C) as compared with those incubated at 33<sup>0</sup>C and at refrigeration temperature (4 – 7<sup>0</sup>C). They further reported that differences between mean values of viable bacterial count per gram of beef samples collected from slaughter house and the meat shops were quite significant, as the samples from meat shops showed a higher mean viable

counts per gram than the meat samples from slaughter house examined under the same condition. In line with this report, Ahmad *et al.*, (2013), recorded various counts in aerobic plate count (APC), *E. coli* count, *Staphylococcus aureus* count and *Salmonella* detection. Mean APC's of beef, sheep and goat meat from abattoirs (5.35, 5.42 and 4.84 log<sup>10</sup> CFU/cm<sup>2</sup> respectively) were significantly (P<0.05) lower as compared to APC values of retail outlets (7.15, 6.92 and 6.62 log<sup>10</sup> CFU/cm<sup>2</sup> respectively). Mean *E.coli* counts for beef, sheep and goat meat from abattoirs and retail outlet were 2.81, 2.94, 2.64 and 2.78, 2.86, 1.94 log<sup>10</sup> CFU/cm<sup>2</sup> respectively, while mean *S. aureus* counts were 2.76, 2.91, 2.90 and 2.96, 2.80, 3.07 log<sup>10</sup> CFU/cm<sup>2</sup> respectively.

It is generally recognized that the most significant food borne hazards from fresh meat are bacteria which can cause disease in humans. Bacteria cannot be seen by the naked eyes, they cannot be detected at post-mortem inspection. The production of visually clean meat, monitored by visual inspection, is an important starting point for meat safety, but visual inspection can detect only gross faecal and other contamination. Although this gives a useful indication of the microbiological status of fresh meat, it is only by undertaking further testing that the presence and/or number of bacteria present on the surface of carcass meat or in processed meat can be assessed objectively (Nouichi and Hamdi, 2009). Consequent upon this, this study sorts to provide research information on the microbiological analysis of beef meat collected at different hours of day in Ekpoma town market.

## **Materials and Methods**

### **Study Area:**

This study was conducted in Ekpoma town. Ekpoma lies on Longitude 6.07°E and Latitude 6.75°N. It has a prevailing tropical climate with annual rainfall of about 1500 – 2000mm. The vegetation in this region represents an interface between the tropical rainforest and derived savannah (Fredrick *et al.*, 2007).

### **Samples collections:**

Meat sample of 1Kg was purchased from the open market at the different times (8am, 12noon and 5pm) respectively. A part was cut into ten samples of **ten grams each**, which served as replicates. They were collected in a clean polythene bag, labeled accordingly and then taken to Animal Science Laboratory for Microbiological analysis.

**Samples preparation:**

Ten grams of meat samples were weighed and homogenized into 90mls of sterile distilled water using a sterile warring blender. Ten folds dilution of the homogenates were made, using sterile pipettes, as described by the methods of Fawole and Oso, (2001).

**Microbiological Analysis:**

Mean counts of total viable proteolytic bacteria count (TPC), total viable count (TVC), Coliforms, *Salmonella*spp, *Pseudomonas* organisms and *Bacillus*spp, were all determined in the collected samples.

- Total proteolytic bacteria count (TPC) was enumerated on Nutrient Agar (NA) incubated at 35<sup>0</sup>C for 48hours.
- Mean counts of total viable organisms (TVC) was determined by the method described by APHA, (1984).
- Coliforms count was performed on MacConkey Agar containing bile salt incubated at 37<sup>0</sup>C for 48 hours.
- *Pseudomonas*organisms was detected by the aid of centrimide agar for each sample.
- *Bacillus* spp. was enumerated using diluted solution of 10<sup>-1</sup>and 10<sup>-2</sup>and heated to 80<sup>0</sup>C for 10 mins.
- *Salmonella* presence was detected by pre-enrichment of meat samples in lactose broth and tetra-thionate broth, while final detection on Bismuth Sulphide agar recommended by WHO procedures.

**Identification of Isolates**

Pure colonies were obtained by repeating streaking in the media and were characterized based on biochemical tests. The biochemically characterized isolates were identified according to Bergey's Manual of Determinative Bacteriology (Holt *et al.*, 1994).

**Experimental Design**

The design for the experiment was a completely randomized design (CRD), one way analysis of variance.

**Statistical Analysis**

All data were subjected to analysis of variance (ANOVA) using the XL Statistical Programme for windows (SAS, 2004).

### Results and Discussions

Results on the microbiological analysis of beef meat collected at different hours are presented in Table 1 below.

**Table 1: Means of microbial quality of beef meat collected at different time(hours)**

Microbial Analysis (log <sub>10</sub> CFU/g)	Time (Hours)			SEM
	8am	12noon	5pm	
Total proteolytic bacteria count (TPC)	5.267 <sup>b</sup>	6.050 <sup>b</sup>	7.867 <sup>a</sup>	0.494
Total Viable count (TVC)	4.517 <sup>c</sup>	5.520 <sup>b</sup>	7.723 <sup>a</sup>	0.219
Coliforms count	2.410 <sup>b</sup>	2.750 <sup>b</sup>	4.750 <sup>a</sup>	0.441
<i>Pseudomonas</i> count	1.193 <sup>c</sup>	2.500 <sup>b</sup>	3.557 <sup>a</sup>	0.270
<i>Bacillus</i> count	1.100 <sup>c</sup>	1.823 <sup>b</sup>	3.030 <sup>a</sup>	0.149
<i>Salmonella</i> spp. count	2.937 <sup>b</sup>	3.283 <sup>b</sup>	3.030 <sup>a</sup>	0.172

abc: Means with similar superscripts along rows are not significantly (P>0.05) different. SEM: Standard errors of means

Results from table (1) above showed that the total proteolytic bacteria count (TPC) analysed from beef meat were significantly (P<0.05) higher in samples collected in the evening (5pm) having 7.867, compared with those collected in the afternoon (12noon) and morning (8am) having 6.050 and 5.267 counts respectively. This shows that the longer meat stays in the market, the higher its microbial load. This result followed similar findings of Bradeaba and Sivakumaar (2013), where higher values of microbial load of meat were observed as time progresses. Also, results of TPC recorded in this study were almost similar to values reported by Ahmad *et al.*, (2013), for mean APCs of beef, sheep and goat meat from abattoirs having 5.35, 5.42 and 4.84 log<sub>10</sub> CFU/cm<sup>2</sup> respectively, which were significantly (P<0.05) lower as compared to APCs values of meat from retail outlets having 7.15, 6.92 and 6.62 log<sub>10</sub> CFU/cm<sup>2</sup> respectively.

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The total viable count (TVC) analysed from meat samples were significantly ( $P < 0.05$ ) lowest at 8am having 4.517, compared with 12noon and 5pm which recorded 5.520 and 7.723 counts respectively. This result was in agreement with the report of Bradeaba and Sivakumaar (2013), where beef showed high general viable count as against mutton and pork which showed comparatively low general population count.

Results of coliforms count in this study were significantly ( $P < 0.05$ ) higher at 5pm having 4.750 count, compared with samples analysed at 12noon and 8am, which recorded 2.750 and 2.410 counts respectively. While those of *Pseudomonas* count recorded significantly ( $P < 0.05$ ) lower value at 8am having 1.193, compared with 12noon and 5pm which had 2.500 and 3.557 counts respectively.

Total *Bacillus* counts also recorded significantly ( $P < 0.05$ ) lower values of 1.100 at 8am compared with 1.823 at 12noon and 3.030 at 5pm; an indication that meat sold at the early hours of the day (8am) tends to have least *Bacillus* spp., while results of *Salmonella* spp. analysed in meat samples recorded significantly ( $P < 0.05$ ) higher value of 3.030 at 5pm compared with 3.283 and 2.937 counts recorded at 12noon and 8am respectively. Beef meat is usually sold in the market on a platform that is exposed, which can be very unhygienic and promotes microbial infection. This is responsible for the level of microbes (especially those of bacteria) obtained in this study. The results of higher microbial analysis recorded in this study as time (hours) of the day progressed, were in line with the reports of Okoh *et al.*, (2019) on the effect of cooking methods on the microbiological load of beef collected at different time (hours) of the day in Ekpoma market.

### Conclusion and Recommendation

From this study, microbiological analysis of beef meat increased as time (hours) of the day progressed, which implies that the longer meat stays when exposed on a platform in the market, the higher the microbial load. It is therefore recommended that meat should be bought from the market in the early hours (8am) of the day, just after arrival from the abattoir.

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