

Research Article

Microbiological Quality of Meat in Butcher Shops in Gelemso Town, West Harerge Zone, Ethiopia

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This study has been undertaken to investigate bacteriological meat quality sold in shops of Gelemso town in west harerge zone, Ethiopia. Samples of kitfo and gored-gored were collected from five different meat shops. The microbial quality of raw meat sold in five most popular meat shops in the of Gelemso Town was determined to ascertain its safety. The meat samples were taken from Butcher A, Butcher B, Butcher C, Butcher D and Butcher E. A total of 10 raw meat samples were collected in two successive visits. Raw samples from Butcher B had the highest mean total aerobic bacterial count of 3.3×10^6 cfu/g for kitfo and 2.8×10^6 cfu/g for gored-gored. The lowest counts were obtained from Butcher D (7.8×10^5 cfu/g and 7×10^5 cfu/g) for Kitifo and Gored-gored were recorded respectively. The highest mean fecal counts (2.0×10^6 cfu/g for kitfo and 1.7×10^6 cfu/g for gored-gored) were obtained also from Butcher B shop. From Butcher D shop was seen the lowest fecal coliform bacterial counts of 4.3×10^5 cfu/g for Kitifo and 4×10^5 cfu/g for gored-gored. Bacterial species isolated from the samples were mostly Gram-positive rods and Gram-positive spherical bacteria in clumps. Some Gram-negative rods were also seen some of which may be fecal coliforms. In this study, it was observed that all samples collected from butcher shops detected a significant count of spoilage microbes. Hence, adequate sanitary measures should be taken from production to consumption stages.

Keywords: Abattoir; Bacterial Load; Butcher Shops; Gelemso Town; West Harerge

Abbreviations

CDCP: Centre for Disease Control and Prevention; CFUs: Colony Forming Units; FAO: Food and Agricultural Organization; HACCP: Hazard Analysis Critical Control Point; NIAID: National Institute of Allergy and Infectious Diseases; TPC: Total Plate Count; TVC: Total Viable Count; WHO: World Health Organization.

Introduction

Food-borne diseases remain the most significant food safety hazards worldwide associated with beef (Maripandi and Al-Salamah, 2010) and resulting from ingestion of bacteria, toxins, and cells produced by microorganisms present in food (Clarence et al., 2009). Food and Agricultural Organization (FAO) of the United Nations and the World Health Organization (WHO) state that illness due to contaminated food is perhaps the most widespread health problem and important cause of reduced economic productivity (Käferstein, 2003). In developing countries particularly in Africa because of prevailing poor food handling and sanitation practices, inadequate food safety regulation, weak regulatory systems, lack of financial resources to invest in safer equipment and lack of education for food-handlers (WHO, 2004).

Meat and meat products are highly perishable commodities and hence, they should be properly stored, processed, and packed and distributed in order to prevent microbial growth (Heetun et al., 2015). The level of microorganisms present in meat products can be

reduced only when they are further processed (Jay et al., 2005).

Contaminated raw meat is one of the major sources of food borne illnesses (Bhandare et al., 2007). Meat provides suitable media for growth of spoilage and pathogenic microorganisms. Health status of butcher shop workers, cloths and knives, wooden boards, and weighing scales can act as a source of microbial contamination (Abebe et al., 2019; Ali et al., 2010). A great diversity of microbes inhabits fresh meat generally, but different types may become dominant depending on pH, composition textures, storage temperature, and transportation means of raw meat (Adu-Gyamfi et al., 2012)

Pathogens, such as *Aeromonas hydrophila*, *Bacillus cereus*, *Campylobacter jejuni*, *Clostridium perfringens*, *Escherichia coli*, *Listeria monocytogenese*, *Salmonella spp.*, *Staphylococcus aureus* and *Yersinia enterocolitica* can also grow and cause illness either by multiplication in the human body (food infection), producing toxins (food intoxication) or multiplying and releasing toxins in the body (food toxico-infection). The presence of pathogens in the food supply is considered to be undesirable as they are the major cause of gastrointestinal disease throughout the world. (Hotee, 2011).

Different microbes get introduced at each stage of meat processing after slaughtering, and these tend to contaminate the meat (Ebel et al., 2004; Sumner et al., 2003). The presence of pathogenic microbes is distressing on the hygienic quality of meat. Hence, the microbiological quality of meat and meat products can be judged by the hygienic quality. Further, the microbial contamination of food

can occur by unhygienic food handling (Tachbele *et al.*, 2006). Food consumers also comprise a link in the chain of food-borne bacterial illnesses with inappropriate storage and cooking of meat and meat products (Sachindra *et al.*, 2005; Koza inski *et al.*, 2006; Tachbele *et al.*, 2006). Pathogens such as

B. cereus, *C. jejuni*, *E. coli*, *L. monocytogenes*, *S. aureus*, *Y. enterocolitica* are known to produce food-borne infections and intoxications in humans (Elmal and Yaman, 2005; Tachbele *et al.*, 2006).

In Ethiopia, the consumption of raw meat has associated with cultural practices and widespread raw beef consumption habit that can be a potential source for food borne illnesses. Raw meat is available in open-air local retail shops without appropriate temperature control and purchased by households and served at restaurants as raw, slightly cooked or well cooked (Siddiqui *et al.*, 2006). Meat handling and processing practices implemented in some butcher shops at Gelemso may provide a chance in which many spoilage microorganisms can easily grow on it and cause spoilage and food-borne disease.

However, there is no information on raw beef microbial quality status in the the study area . Hence, this study addresses microbial quality status of raw beef from Gelemso town butcher shops and forwarding necessary intervention in study area.

Therefore, the present study aims with the following objectives:

General Objective

- To evaluate bacteriological quality of meat for kitfo and gored gored sold in butcher shops of Gelemso town.

Specific Objective

- To identify the status of bacteriological contamination in the meat sample from butcher shops
- To identify the types of bacteria, present in the meat samples

Literature Review

Microorganisms Found in Meat

The bacteriological condition of carcass meat is highly dependent on the manner, in which meat animals are reared, slaughtered, and processed (Osama and Gehan, 2011). The presence of high mean values of microbial load of scrapings from meat stalls was reported in Ibadan metropolis, Nigeria (Fasanmi *et al.*, 2010). A Microbiological status of fresh beef cuts, environmental sources, in a red meat and Fecal coli-form Contamination of Beef Carcasses during the Slaughtering Process reported $1.2 \times 10^1 - 6.3 \times 10^1$ cfu/g and $3.16 \times 10^1 - 8.9 \times 10^1$ cfu/g for faecal coliform count in fresh beef samples respectively (Stopforth *et al.*, 2006 and Yalcin *et al.*, 2001). It is important that only relatively clean animals be presented for slaughtering, since it is extremely difficult to obtain clean meat from dirty animals. The presence of fecal coliforms is indicative of fecal contamination and of the potential presence of enteric pathogens especially bacterial pathogens (Anon, 2003). Therefore, the cleanliness of livestock depends on husbandry, weather and climate, methods of transport and holding conditions at the abattoir. Cattle from feedlots may carry more fecal bacteria and less soil organisms (Biswas *et al.*, 2011). Although the muscle tissues of animals are free of micro-organisms; it can easily contaminate

with both pathogenic and non- pathogenic microorganisms at the time of slaughter under poor processing conditions. In addition, the high nutritive value of meat makes it an ideal medium for bacterial growth (Prescott *et al.* 2002). Some apparently healthy animals may harbor various micro- organisms in the liver, kidneys, lymph nodes and spleen. These microorganisms and those from contamination through slaughtering can migrate to the skeletal muscles via the circulatory system (Marriot, 2004). In general, the micro-flora of meat will be that of the feedlot which are on the external surfaces of the animal contaminating the meat by direct contact through air, water, soil, manure and the hands and tools of the workers (Unc and Goss, 2004). The healthy inner part of meats has been reported to contain few or no micro- organisms, although they have been found in lymph nodes, bone marrow, and even flesh (Okonko *et al.*, 2010). Staphylococci, Streptococci, Clostridia and Salmonella, have been isolated from the lymph nodes of red-meat animals. The important contamination, however, comes from external sources during bleeding, handling, and processing. During bleeding, skinning, and cutting, the main sources of micro-organisms are the exterior of the animal (hide, hooves and hair) and the intestinal tract (Selvan *et al.*, 2007). Comminuted meats such as ground beef invariably have higher numbers of microorganisms than non-comminuted meats such as steaks (Salihu *et al.*, 2010). Commercial ground meats generally consist of trimmings from various cuts. These pieces have been handled excessively and consequently normally contain more micro-organisms than meat cuts such as steaks. Ground meat also provides a greater surface area, which itself accounts in part for the increased flora. This greater surface area of ground meat favors the growth of aerobic bacteria, the usual low-temperature spoilage flora. One heavily contaminated piece of meat is sufficient to contaminate others, as well as the entire lot, as they pass through the grinder. This heavily contaminated portion is often in the form of lymph nodes, which are generally embedded in fat. These organs have been shown to contain high numbers of micro-organisms and account in part for hamburger meats having a generally higher total count than ground beef (Salihu *et al.*, 2010).

Source Microbial Contamination of Raw Meat

Meat carcasses are highly contaminated by contact with unhygienic surfaces, by personnel and airborne organisms, will remain a possibility in all operations during the subsequent history of the meat (Adzitey *et al.*, 2011). These will include chilling, freezing, processing, cutting, packaging, transport, sale, and domestic handling, although some sources of contamination are obviously removed when the carcasses leave the slaughter floor (Biswas *et al.*, 2011).

Bacteriological quality of raw meat product is strongly influenced by the prevailing hygiene condition during their production and handling (Osama, 2011). The carcass of a healthy animal slaughtered for meat and held in a refrigerated room is likely to have only minimal surface bacteriological contamination while the inner tissues are sterile. After chilling, further processing of carcasses can result in raw meat product contamination. When carcasses and cuts are subsequently handled through the food distribution channels, they are subjected to an increasing number of micro-organisms from the cut surfaces (Okonko *et al.*, 2010). Contamination subsequently occurs by the introduction of micro- organisms on the meat surfaces in operations performed during cutting, processing, storage, and

distribution of meat (Clarence et al., 2009). However, if the meat is kept clean by preventing contamination through dirty hands, clothing, equipment and facilities and the meat is kept cold and covered, there will be little or no contamination by micro-organisms whether bacteria, yeasts, moulds, viruses, or protozoa (Osama, 2011). An employee who does not follow sanitary practices are the largest contamination source of raw meat that they meet spoilage and pathogenic micro-organisms through work and other parts of the environment while their hands, hair, nose and mouth, harbor microorganisms that can be transferred to raw meat during cutting, processing, packaging, and selling by touching, breathing, coughing, or sneezing (Biswas et al., 2011). Therefore, in the prevention of meat contamination, personal hygiene plays an important role as there are as many as 200 different species of microorganisms on a healthy human body (Featherstone, 2003).

Consequences of Food-Borne Diseases from Microorganisms

Infectious diseases spread through food or beverages are a common, distressing and sometimes life-threatening problem for millions of people around the world (NIAID, 2002). The U.S. Centre for Disease Control and Prevention (CDCP) estimated that, 76 million people suffer from food-borne illnesses each year in the United States (CDCP, 2008). This accounts for the 325,000 hospitalizations and more than 5,000 deaths. Cheleleka recorded one of the highest ever numbers of food poisoning cases in 1990 (Kimani, 2001). A total number of more than 200,000 outpatients were treated at government hospitals. Food-borne diseases especially those caused by pathogenic organisms, remain a serious problem in all countries and are extremely costly to treat (Duff et al., 2003). Health experts estimates that the yearly cost of all food-borne diseases in the United States is \$5 to \$6 billion in direct medical expenses and lost productivity (NIAID, 2002). In developing countries, where the problem of diarrheal disease is far greater, the effect on economic activity and development can only be far more severe (Adams et al., 1999). Diarrhea is a feature of most of the food-borne diseases and up to 70 percent of all episodes of diarrhea may result from ingestion of contaminated food and water (Adams et al., 1999). World Health Organization (WHO) reported that 50 million children under 5 years get diarrheal diseases each year due to contaminated water and foodstuff (Tavakoli and Riazipour, 2008). In most cases, food is not contaminated intentionally, but rather due to carelessness or insufficient education or training in food safety. Meat consumption without sanitary care may cause diseases such as tuberculosis, salmonellosis and cysticercosis in consumers (Azevedo and Bankuti, 2003).

Quality Control in the Supply Chain of Meat

To enable risks involved to be estimated and appropriate measures to be taken, analysis of the slaughtering process must be implemented by collection of abattoirs - specific microbiological monitoring data, in accordance with Hazard Analysis Critical Control Point, (HACCP) principles (Zweifel et al., 2005). A regular microbiological examination of carcasses allows reliable conclusions to be drawn about long-term hygienic conditions in slaughterhouses and butcheries. The microbiological control includes testing of the whole production chain; with samples taken from production and consumption.

Microbiological Analysis

Bacteria are microscopic organism which is not seen by naked eyes. However, the number of bacterial in each sample are required to know in certain aspects such as butcheries, disease investigation etc. Because of this, a variety of methods have been developed for the enumeration of microorganisms (including bacteria) like direct microscopic counts, membrane filtration and viable plate counts (Jacquelyn G., Black 1999). Among these methods of enumeration, viable plate counts are being used most frequency to measure bacterial populations. Viable plate counts only determine (measure) the number of viable bacterial found in the samples, unlike the microscopic counts which cannot distinguish live from dead cells. However, viable plate count takes sometimes colonies to grow (Gerard J. Tortora, 2003). Before doing plate counts, serial dilutions required to overcome colony forming units (CFU) are too numerous to count. The ideal of CFU must be <300 colonies on agar plate. This is because of when original bacterial sample inoculated without serial dilution. It is hard to count more than 300 colonies on an agar plate (Kathleen Talaro, 1993). After incubation the colonies were counted to determine the colony forming unit per centimeter square cfu/cm², all the visible colonies are calculated and represented as CFU.

Materials and Methods

Study Area

Study was conducted in different raw meat sold shops of Gelemso Town (Figure 1). Gelemso located distance to 404 km from Addis Ababa capital city of Ethiopia and 75 km to the south of zonal town, Chiro. It has found in latitude 8.8131°N and longitude 40.5203°E and an average altitude of 1814.38 Meters (5952.69 Feet) above sea level and its temperature range from maximum 25-30°C and minimum 7-20°C and annual rain fall range from 670-1000mm (HAO, 2017).

Study Design and Period

An experimental laboratory study was carried out to evaluate bacterial quality of raw meat sold in butcher shops of Gelemso Town from March 25-May 25 / 2021.

Sample Preparation and Inoculation

Representative samples were taken aseptically using sterile moistened cotton covered swabs and the swabbed sample was immersed into the test tube containing approximately 10 ml buffered peptone water. The test tubes were labeled respectively and transported to the microbiological laboratory with an icebox. Then the samples were cultured onto macConkey agar, Manitol salt agar, blood agar, brilliant green agar and incubated at 37°C for 24-48hrs [19]. Colony morphology on the plates was observed and colony Sub-culturing was done to obtain pure colonies for biochemical tests for Total coli form, Fecal coli form, Microbial load, Sanitary condition of shops.

Source of Samples

A total of five major meat shops where most people prefer to buy beef were selected for this study. The meat shops selected were coded as Butcher A, Butcher B, Butcher C, Butcher D and Butcher E. A total of 20 raw meat samples were collected in four successive visits. The meat samples for kitfo and gored-gored were collected into sterile plastic bags and transported under 4°C in a Coleman box filled with ice to prevent further contamination. The samples were analyzed

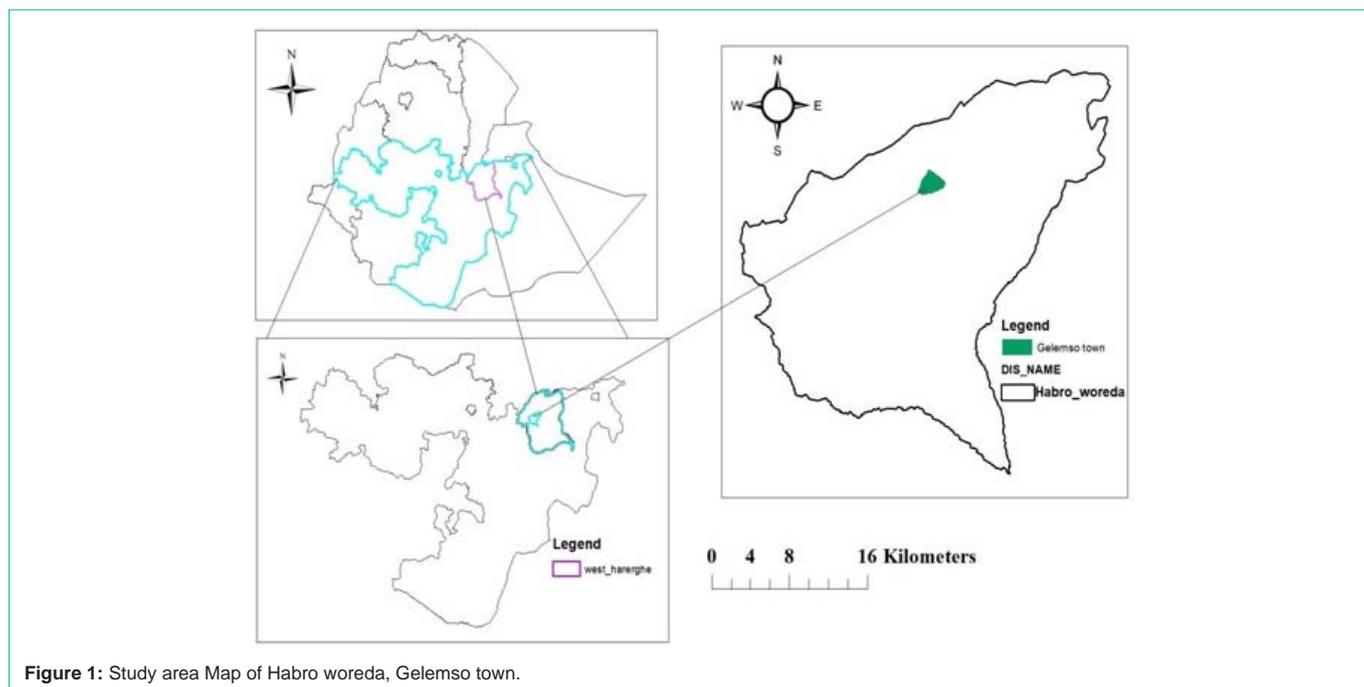


Figure 1: Study area Map of Habro woreda, Gelemso town.

immediately upon arrival in the laboratory.

Sample Analysis and Presentation

After the sample collected the microbial load was analyzed in Hirna regional veterinary laboratory. In laboratory, 1g of raw meat samples were transferred into 9 ml of water in a test tube for 10^{-1} dilution and shaken vigorously for 1 minute. Autoclaved 1-gram meats in 9 ml water served as negative controls. Decimal serial dilutions were made by transferring 1 ml of the above samples into 9 ml distilled water up to 10^{-5} . Then, 0.1 ml of 10^{-4} and 10^{-5} dilution was taken and poured onto sterile petri dish that contain solidified 25 ml of plate count agar (PCA) or MacConkey agar. This was then spread thoroughly by using flamed bent rod and incubated at 37°C for 24 -8 hours. After incubation the colonies was counted as (cfu/plate/g). Finally, the data, tables, was used for comparison of the selected raw meat samples with standard recommended guideline of microbial load of raw meat per dish. Before starting raw meat collection consent form permission was obtained from Environmental Health Department to Gelemso Town food service office, then Gelemso Town raw meat sellers.

Result

From five different shops of raw meat (Kitifo) sold, the highest bacterial count (3.3×10^6 cfu/g) was recorded in butcher B shop and the lowest (7.8×10^5 cfu/g) in butcher A shops and also Butcher B shop was with highest bacterial count (2.8×10^6 cfu/g) for Gored-gored and the lowest bacterial count (7×10^5 cfu/g) recorded in the butcher A shop (Table 1).

From five different shops of raw meat (Kitifo), the highest fecal bacteria count (2.0×10^6 cfu/g) was recorded in B butcher shop and the lowest (4.3×10^5 cfu/g) in A butcher meat shops and the B butcher shop was the highest fecal bacterial count (1.725×10^6 cfu/g) for Gored-gored, and the lowest (4.0×10^5 cfu/g) in A butcher shop respectively (Table 2).

Table 1: Mean number of aerobic bacterial colonies of Kitfo and Gored-gored samples from different raw meat shops in Gelemso Town.

Coded Name of Butcher Shop	Mean Number of Bacterial Colony (CFU/g) on Plate Count Agar		
	Kitfo	Gored-gored	Control
A	7.8×10^5	7×10^5	0
B	3.3×10^6	2.8×10^6	0
C	2.4×10^6	1.9×10^6	0
D	1.9×10^6	1.3×10^6	0
E	1.5×10^6	9.0×10^5	0

Table 2: Mean number of fecal bacterial colonies from different raw meat shops in Gelemso Town.

Name of Coded Meat Shop	Mean Number of Fecal Bacterial Colony (CFU/g) on MacConkey Agar		
	Kitfo	Gored-gored	Control
A	4.3×10^5	4.0×10^5	0
B	2.0×10^6	1.725×10^6	0
C	1.8×10^6	1.3×10^6	0
D	1.4×10^6	9.0×10^5	0
E	1.0×10^6	8.5×10^5	0

Different bacterial types were seen under the microscope. Gram-positive rods and Gram- positive clumped spherical bacteria, probably *Staphylococcus*, were the most numerous (Table 3). Gram-positive spherical bacteria in pairs and Gram-positive chained spherical bacteria, probably *Streptococcus* were the second in number seen. Less numerous were Gram-negative rods, which may include fecal coliform bacteria that grew on MacConkey agar.

The study revealed that the total coliform count of bacterial contaminants were grown in all meat samples of Kitifo and Gored-gored collected from butchery shops (A,B,C,D and E) (Table 1). The

Table 3: Types of bacteria isolated from the different raw meat shops.

Name of Coded Meat Shop	Type of Bacteria
A	G(+) cocci in chains, G(+) cocci in clumps, G(+) cocci in pairs, G(+) rods.
B	G(+) cocci in chains, G(-) rods
C	G(+) cocci in chains, G(+) cocci in pairs, G(+) rods
D	G(+) cocci in clumps, G(+) cocci in chains, G(+) rods
E	C(+) cocci in clumps, G(+) cocci in chains, G(+) rods, G(-) rods

G(+), Grams-positive; G(-), Gram-negative

highest mean values of microbial load (3.325×10^6 cfu/g and 1.825×10^6 cfu/g) were found on meat samples from B and (2.425×10^6 cfu/g and 1.85×10^6 cfu/g) were observed on meat samples from C butchery shop in the Kitifo and Gored gored respectively. This might be due to high exposure to dusts from the environment and poor hygienic condition. The lower mean values of microbial load (7.75×10^5 cfu/g and 7×10^5 cfu/g) were found on meat samples from A butchery shop analyzed in the Kitifo and Gored gored respectively.

The current findings were also in agreement with Fasanmi et al. that reported the presence of high mean values of microbial load of scrapings from meat stalls in Ibadan metropolis, Nigeria. Similarly, different genera of Gram negative and Gram-positive *bacteria* spp. were isolated in this study. The bacteria isolated were identified as *E. coli*, other coliform, *Staphylococcus* spp., *Streptococcus* spp. and *bacillus* spp. *bacteria*.

The presence of these organisms in raw meats might be the result of poor hygienic and sanitary practices employed in the slaughtering, processing, packaging, transporting storing of fresh meats. Although the muscle tissues of animals are free of micro-organisms; it can easily contaminate with both pathogenic and non-pathogenic microorganisms at the time of slaughter under poor processing conditions. In addition, the high nutritive value of meat makes it an ideal medium for bacterial growth (Prescott et al 2002).

From the results obtained, fresh meats sample were contaminated with high level of *E. coli*, other coli-form bacteria, *Staphylococcus* spp., *Streptococcus* spp. that agrees with Fasanmi et al. and Clarence et al. report which stated that these organisms are the main sources of contamination. Therefore, high total coliform obtained in this study might be attributed to poor hygienic condition and product handling practices leading to contamination.

The presence of fecal coliform is an indicator of poor sanitary condition in the butcher shops since these microorganisms originate from fecal microbiota. The result revealed the highest mean values for fecal coli-form on meat samples from B followed with D, C, E and A butchery Shops. The mean fecal coli-form count in raw meat (Kitifo) were 1.975×10^6 cfu/g, 1.775×10^6 cfu/g, 1.375×10^6 cfu/g, 1×10^6 cfu/g and 4.25×10^5 cfu/g in the B, C, D, E and A butchery Shops respectively. This finding figure is much higher than Eisel et al. and Stopforth et al. that reported 2.5×10^1 - 1.58×10^3 cfu/g and 1.2×10^1 - 6.3×10^1 cfu/g for fecal coli-form count in fresh beef samples, respectively. The sources of contamination here may come from the intestinal contents, slaughter process, through meat handling and transportation to the market, Cross contamination from tables, knives and weighing scale to other beefs is also possible. And also, the prevalence of fecal coli-form count in the raw meat (Gore-gored) were 1.725×10^6 cfu/g,

1.275×10^6 cfu/g, 9×10^5 cfu/g, 8.5×10^5 cfu/g and 4×10^5 cfu/g in the B, C, D, E and A butchery Shops respectively. This result shows that microbial contamination of Gored-gored is lower than that of Kitifo. This is probably due to the lower surface area of Gored-gored, which is not comfortable for growth and multiplication of bacteria as on Kitifo.

Even though the microbial load in Gored-gored is lower than Kitifo; it become higher than the research that reported the mean fecal coli-form counts of the different sites of the carcasses were 3.16×10^1 - 8.9×10^1 cfu/cm (Yalcin et al., 2001).

This is probable due to the shop is located next to a busy street with vehicles blowing dust with all sorts of contaminants on the meat, knives, wood cutting blocks, weighing balances and on meat chopping tables the butchers themselves pay little concern on their personal hygiene serve the meats with dirty hands and clothing. There were no storage facilities located within the vicinity. The presence of fecal coliforms is indicative of fecal contamination and of the potential presence of enteric pathogens especially bacterial pathogens (Anon, 2003). Thus, the above microbial load shows (Table 2) indicates the presence of health threat full microbe on the meat produced in Gelemso town. To avoid such threat meat products should be fried with optimum cooking temperature.

Conclusion and Recommendation

The meat and meat products being rich in several nutrients are easily attacked by microbes, which are widely are prevalent in our environment. The presence of the organisms in meat and meat products is the major concern in the preparation of high quality foods that is safe to the consumer.

From the study, high level of bacterial contamination from the different meat sources due to poor personal hygiene and environmental contamination. The place where raw meats are kept, use of open housing during selling might be the possible source for the occurrence of contamination of gram positive and gram-negative bacteria. If measures are not put in place, there may be a possible outbreak of food poisoning and or food borne infections due to consumption of the contaminated meat. This may lead to serious economic and public health problem.

Based on findings the following points are recommended: -

- Meat inspections should be strengthened by veterinary professionals in the town before and after slaughtering and before the meat is distributed to the public.
- Good personal hygiene and meat handling practices should be adhered strictly by butchers and those selling the meat.

- Awareness creation to butcher shop workers regarding meat hygiene is essential.
- Further investigation should be carried out the value of chain of Meat Handling Practices in Butcher Shops and Abattoir to point out the avenues for microbial meat contamination.

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