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PREVALENCE OF *ESCHERICHIA COLI* AND *STAPHYLOCOCCUS AUREUS* IN RAW BEEF SOLD IN MAKURDI, BENUE STATE, NIGERIA

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ABSTRACT

This study was undertaken to survey the Prevalence of *Escherichia coli* and *Staphylococcus aureus* in raw meat sold in Markudi, Benue state. The study was conducted on the Four (4) major markets in Makurdi namely: Wurukum, North bank, High-level and Modern markets. Forty (40) samples of meat swabs were analyzed for the presence of *E. coli* and *S. aureus*. The organisms were isolated by cultural methods using Nutrient aga, Eosine methylene Blue Agar and Cystine Lactose Electrolyte Deficient Agar. Twenty six (26) samples (65%) tested positive for Escherichia coli, while twenty one (21) samples (52.5%) had Staphylococcus aureus. Wurukum market had the highest prevalence of *both E. coli* and *S. aureus* to be 34.60% and 33.30% respectively. North bank, High-level and Modern markets had prevalence of *E. coli* to be 19.23%, 30.76%, and 15.38% respectively. The prevalence of *Staphylococcus aureus* was lowest in samples from North bank market with 14.28%. High-level and Modern markets had prevalence of *S. aureus* to be 23.80% and 28.57% respectively. These results have shown the heavy contamination of meat (beef), with *Escherichia coli* and *Staphylococcus aureus* sold in makurdi markets and this could result in severe health problems to consumers. The study recommends education of meat vendors, on proper and more hygienic methods of meat production.

Introduction

Merriam Webster Learner's dictionary defines "MEAT" as flesh of animal used as food. Meat is a valuable source of Protein, Iron, Vitamin B12, Selenium and phosphorus. It is also a good source of Niacin and Iron (Olaoye and Onilude, 2010).

In most countries, including Nigeria, fresh meat forms a significant proportion of meat intake (Olaoye and Onilude, 2010), due to its high nutrient content, it supports the growth of microorganisms (Kalalou, *et al*., 2004). The possible sources of contamination by microorganisms are through slaughtering of sick animals, washing the meat with dirty water, handling by butchers, contamination by flies, processing close to sewage or refuse dumps environment, transportation and use of contaminated equipment (Hatakka, et al., 2006). Contaminated raw meat is one of the major sources of food-borne illnesses (Bhadare et al., 2007). The risk of the transmission of zoonotic infections is also associated with contaminated meat (Hassan *et al*., 2010).

Food borne illness is a major international health problem with consequent economic reduction (Duff et al., 2003). According to Doyle and Evans (1999), food borne diseases are diseases resulting from ingestion of bacteria, toxins and cells produced by microorganisms present in food (meat). Seven pathogens have reportedly been found in animal products especially meat. They are *Esherich-*

ia coli, Listeria monocytogenes, Campylobacter jejuni, Clostridium perfringes, Salmonella sp., Toxoplasma gondii and Staphylococcus aureus. These pathogens account for approximately 3.3 to 12.3million cases of food borne illnesses and leading to about 3,900 deaths each year in the United States (Talaro 1996). Some of the microorganisms originate from the animal's intestinal tract as well as from the environment with which the animal had contact at some time before (Koutsoumanis and Sofos, 2004).

Esherichia coli and *Staphylococcus aureus* are normal flora in human but the occurrence of these organisms in food substances are indications of excessive food handling (Adamolekun and Adamolekun, 1992). Though *E.coli* bacteria are essential in healthy functioning of human and animal digestive systems, some strains of *E. coli* have become pathogenic to humans and animals. Gormley *et al*., (2011) reported about 2,429 foodborne outbreaks in England from 1992 to 2008 mostly caused by bacterial pathogen Esherichia coli O157. The Center for Disease Control in 2002, projected that about 52% of *E. coli* illnesses (gastroenteritis) were as a result of food borne transmission (Rangel *et al*., 2005).

Staphylococcus aureus is a bacterium that causes staphylococcal food poisoning, a form of gastroenteritis with rapid onset of symptoms*. S.aureus* is commonly found in the environment (soil, water and air) and is also found in the nose and on the skin of humans. (Argudin *et al*., 2010). *Staphylococcus aureus* is one of the most important agents of food poisoning in the world (Balaban and Rasooly, 2000). The primary habitats of this organism are the mucosa of the nasopharynx and the skin of humans and animals. It has also been found in water, dust and air (Casey *et al*., 2007).

Materials and methods

Study Area

Makurdi is capital of Benue State. It is located on latitude 070 41N and longitude 080 37E on the Benue state map. Makurdi is situated on the banks of the River Benue, with a total population of 297,393 (157,294 males and 140,103 females), as at 2006 Population Census (NPC 2006).

Sample Collection

Samples were obtained from vendors at Wurukum, North bank, High-level and Modern markets, which make up the major markets in Makurdi, Benue state. Forty swab samples of meat were obtained from meat vendors at the above listed markets. The samples were immediately wrapped in sterile cellophane bags to avoid contamination and then transported to the laboratory, for microbial analysis.

Sterilization of Materials

All glass wares were sterilized using hot air oven at 1700C for three hours. Glass Wire loop and inoculating needles were sterilized by flaming.

Media Preparation

All media used (Eosine Methylene Blue Agar and Cystine Electrolyte Deficient Agar) were prepared according to manufacturer's instruction.

Inoculation and Incubation

All samples were inoculated on Nutrient agar, already poured into sterile Petri dishes by streaking on the agar. The inoculated plates were incubated at 37^0C for 24 hours. Colonies observed were sub-cultured on Eosine Methylene Blue Agar (EMBA) and Cystine Lactose Electrolyte Agar (CLED).

Isolation and identification of Pure Cultures

Isolation of *Esherichia coli* was carried out on Eosine Methylene Blue Agar (EMBA) plates, which were inoculated by streaking colonies from the nutrient agar with a wire loop. The plates were incubated at 35^0c for 18- 24hours. Isolation of *Staphylococcus aureus* was carried out on sterile CLED Agar medium. The agar plates were inoculated as described previously and incubated at 350c for 48-72hours. The isolates were identified based on their cultural characteristics, morphology, Grams reaction and biochemical characteristics.

Statistical Analysis

Data was analysed using Chi- square (SPSS version 20), P value of 0.05.

Results and discussion

A total of 40 samples of meat swab (10 each), were collected from four (4) markets namely: Wurukum, North bank, High-level and Modern markets. Fig 1 gives a Graphical representation of the prevalence of E. coli and S. aureus in percentages.

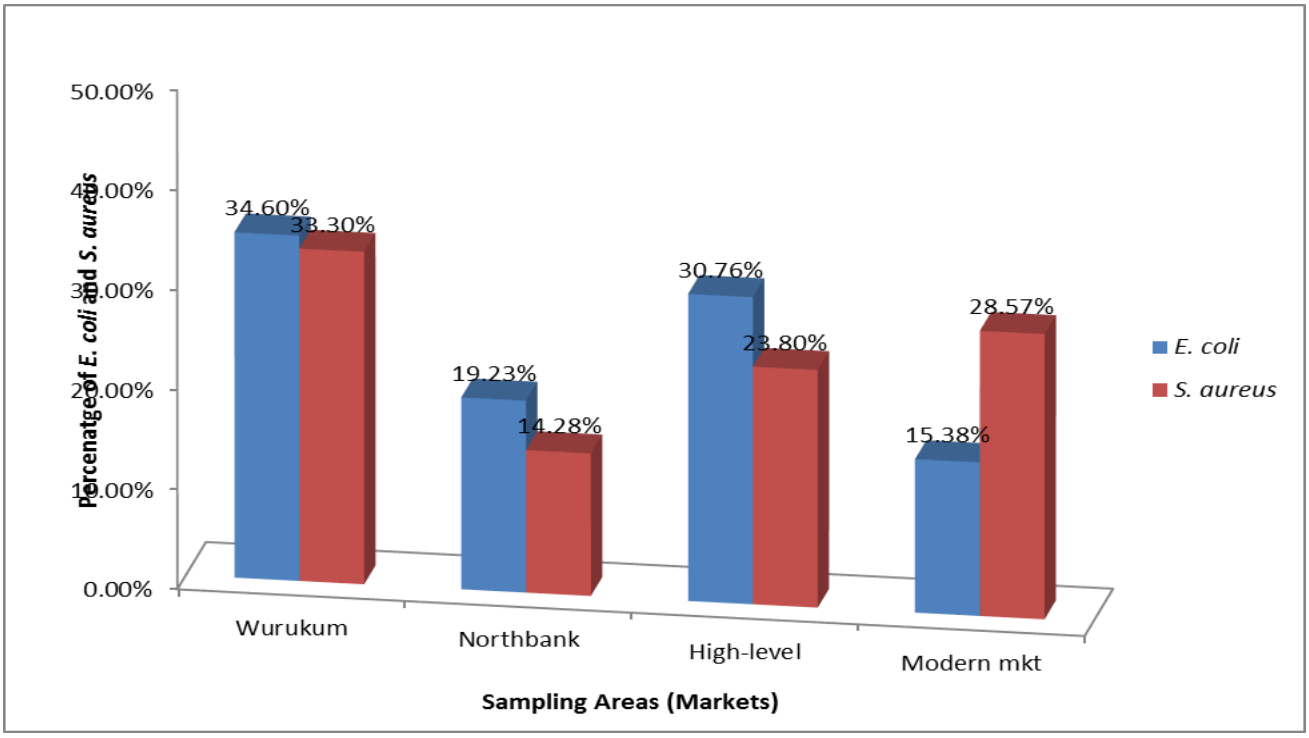

Fig 1: Prevalence of E. coli and S. aureus in Raw Meat in percentage. The prevalence of E. coli ranges from 34.60% to 15.38%, while the prevalence of S. aureus ranges from 33,30% to 14.28%.

Table 1 shows the Cultural Characteristics of E. coli Colonies on Eosine Methylene Blue Agar (EMBA). Colonies observed were pink with shiny metallic sheen, circular, opaque, low convex, with mucoid consistency.

Table1 Cultural Characteristics of *Esherichia coli* Colonies on Eosine Methylene Blue Agar

Isolate (Lab code)	Shape	Elevation	Colour	Consistency	Surface	Edge	Opacity	Organism present
W_1-W_9	Circular	Low convex	Pink	Mucoid	Shiny metallic sheen	Entire	Opaque	*E. coli*
N_1-N_5	Circular	Low convex	Pink	Mucoid	Shiny metallic sheen	Entire	Opaque	*E. coli*
H_1-H_8	Circular	Low convex	Pink	Mucoid	Shiny metallic sheen	Entire	Opaque	*E. coli*
M_1-M_4	Circular	Low convex	Pink	Mucoid	Shiny metallic Sheen	Entire	Opaque	*E. coli*

Key:

W1- W9 = (9) Isolates from Wurukum markets

N1 – N5 = (5) Isolates from Northbank markets

H1-H8 = (8) Isolates from High level markets

M1-M4 = (4) Isolates from Modern markets

Table 2: Cultural Characteristics of *Staphylococcus aureus* Colonies on CLED agar

Isolate (Lab code)	Shape	Elevation	Colour	Consistency	Surface	Edge	Opacity	Organism present
W_1-W_7	Circular	Flat	Deep yellow	Mucoid	Bright	Entire	Opaque	*S. aureus*
N_1-N_3	Circular	Flat	Deep yellow	Mucoid	Bright	Entire	Opaque	*S. aureus*
H_1-H_5	Circular	Flat	Deep yellow	Mucoid	Bright	Entire	Opaque	*S. aureus*
M_1-M_6	Circular	Flat	Deep yellow	Mucoid	Bright	Entire	Opaque	*S. aureus*

Key:

W1- W7 = (7) Isolates from Wurukum markets

N1 – N3 = (3) Isolates from Northbank markets

H1-H5 = (5) Isolates from High level markets

M1-M6 = (6) Isolates from Modern markets

These results showed the high rates of contamination of meat processed and sold at the different markets with *E. coli*, which is an indication of the presence of unacceptable levels of other pathogenic microorganisms. This contamination could be as a result of so many factors at the slaughtering and skinning points. The 65% prevalence rate in fresh beef is a clear indication of heavy contamination. This agrees with the work of Petridis *et al.,* (2006) who reported undercook ground beef implicated as a source of contamination that led to the infection of about 243 in Montana, USA. The high level of carcass contamination could have resulted from unhygienic slaughtering and meat processing done at abattoirs and slabs, where butchering of meat is mostly done on concrete floors (Umolu *et al*., 2006). Other factors include contamination from external sources like air, soil, use of non portable water and improperly washed utensils.

Contamination of the meat could also have come from internal sources like intestines content, lymph nodes, as well as cross contamination by meat handlers (Umolu *et al.,* 2006). All these could have contributed to the microbial contamination of fresh beef as reported by Umoh (2001). Meat contamination is sometimes highest during the wet season from environmental contamination by run-off water and flood by which microbes are transferred to the meat during the butchering and meat dressing (Sofos *et al.,* 1999).

This study also investigated the prevalence of *Staphylococcus aureus* in raw meat samples. The prevalence of *S. aureus* was recorded to be 52.50%. This result disagrees with a study carried out by Bakr *et al*., (2004), who reported a prevalence of 65%. The difference in result could be as a result of the diffrerence in study areas, as study by Bakr et al., (2004) was conducted in Egypt. The highest prevalence of *S. aureus* recorded was Wurukum market with 33.30% (Fig 1). This result agrees with the work carried out on beef by Inge *et al*., (2007). North bank and modern markets had prevalence of 14.28% and 28.57% respectively (Fig 1). High-level market had *Staphylococcus aureus* prevalence to be 23.80% (Fig 1), which agrees with the study carried out by karmi, (2013). There is no significant difference in the prevalence of Staphylococcus aureus among the markets, p value of 0.05.

Contamination of the meat products by *Staphylococcus aureus* could be traced to and poor hygienic and sanitary conditions of the abattoirs and markets as reported by Kluytmans *et al*., (1995). Different stages of slaughter such as scalding and chilling may affect the prevalence bacterial load of S. aureus in meat (Hansson, 2001). The high contamination of meat by *S. aureus* is the reason for the inadequate microbiological quality of meat sold in markets (Alvarez-Astorga *et al*., 2002). The high prevalence of *S. aureus* report in this study could be as a result of indirect contamination through working surfaces and knives to which the meat was exposed (Yeh *et al*., 2004).

The prevalence of *Esherichia coli* and *Staphylococcus aureus* from this report agrees with previous report by El-Gohany (1994), that foods of animal origin (meat) either cooked or uncooked were predominantly contaminated with *E. coli* and *S. aureus*.

Conclusion

This study showed that a high proportion of raw beef sold in makurdi markets, for human consumption, were contaminated with *Esherichia coli* and *Staphylococcus aureus* with variable prevalence amongst the different markets. The results showed microbial hazards of unhygienic meat processing commonly practiced in Nigeria abattoirs and markets.

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