Distribution of Bacterial Isolates from Contact Surfaces of Meat Handlers in Abattoirs of Southeastern, Nigeria

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Abstract

Poor sanitation and personal hygiene practices by meat handlers in abattoirs can result in the contamination of meat by microorganisms. This study was carried out in Southeastern Nigeria to determine the bacterial isolates found in the contact surfaces of meat handlers at their workplace. Samples were collected using sterile swab sticks from 108 contact surfaces which included 36 tables, 36 hands and 36 knives of the meat handlers. The samples were taken to the laboratory for bacterial analysis. A well-structured questionnaire was also used to interview the meat handlers. Results of the bacterial isolates of the samples showed that Staphylococcus aureus was isolated in 22 (61.11%) of the 36 samples from tables, 18 (50.00%) from the hands of meat handers and 15 (41.67%) from their knives; Escherichia coli, 18 (50.00%) from tables, 9 (25.00%) from hands and 6 (16.67%) from knives; Shigella dysenteriae, 17 (47.22%) from tables, 9 (25.00%) from hands and 9 (25.00%) from knives; Klebsiella pneumonia, 17 (47.22%) from tables, 13 (36.11%) from hands and 11 (30.56%) from knives; Enterococcus faecalis, 15 (41.67%) from tables, 16 (44.44%) from hands and 12 (33.33%) from knives; Pseudomonas aeruginosa, 10 (27.78%) from tables, 9 (25.00%) from hands and 11 (30.56%) from knives. The sanitary and personal hygiene practices of the meat handlers were found to be very poor and this resulted to the high bacterial isolates found on their tables, hands and knives. It was recommended that regulatory agencies embark on routine inspection of abattoirs to ensure compliance of meat handlers to meat safety standards.

Keywords: Bacteria, Abattoir, Meat handler, Contact surface, Personal hygiene, Sanitation

Introduction

Abattoirs are establishments where livestock are killed prior to human consumption. They are also known as slaughterhouses and should be subject to inspection to ensure that the meat they produce is safe to eat[1]. Both the live animals and the carcasses are to be subjected to inspection. It is important to ensure that the animals are in good health before they are killed. Any signs of stress, injury or sickness in the part of the animals will require attention. They should not be killed yet but taken to the veterinary doctor who will take a look at it and certify that the animal is in good health before they can be slaughtered[2,3]. Inspection of the meat also continues after slaughter. This is the job of a competent and qualified inspector who will certify the safety of the meat before it is sold to the public for consumption. The carcass should be transported soon after slaughter, in a special vehicle, to a butchery or distribution center. Whatever the mode of transportation to the market, it is imperative that the vehicle is kept absolutely clean. This is because contamination can and do occur during the transportation of meat carcasses[4].

Safe meat for the consumer is linked to the butcher's shop. It is here that consumers come to buy their meat for consumption. The standard of hygiene therefore is of utmost importance to prevent contamination by microorganisms. For this reason, butcher's shops need licenses to operate, confirming that they meet all the handling specifications that ensure the safety of the meat[5]. There should be adequate working space, and the walls and floor should be constructed of durable material and should be smooth, impermeable, easily cleanable and light-colored. There should be adequate ventilation and natural light. The utensils should be clean and kept in an appropriate place. The butcher should wear a clean white gown, preferably with an apron and a white hair cover. Importantly, an approved means for the disposal of meat waste should be provided inside or outside the butchery[6].

One of the most important and common source of contamination of meat is by the meat handlers. This is because most of them do not have a good sense of hygiene[7]. The personal hygiene of the meat handlers must meet the specified standards[8] for meat safety. The health of the meat handler is also important. Any injury or bruise they may have on their fingers is a potential source of contamination. When they are not appropriately dressed with hair nets, face masks and aprons, contamination can occur. Before employment of a meat handler, medical examination is very necessary and this examination must occur periodically and routinely to ensure that their health remains in good condition as long as they continue to handle meat sold to the public[9].

Cross-contamination of microorganisms can occur during the cutting and dressing of animals and also during handling and inspection of meat. Good personal hygiene practices should prevent undue general contamination, and prevent cross-contamination with human pathogens that may cause food-borne disease[10]. When a meat handler or any other person is moving from rooms or areas containing raw meat to rooms for meat preparations, they are to wash their hands thoroughly and also change their clothes and sanitize them. Persons who come into direct or indirect contact with edible parts of animals or meat in the course of their work should maintain appropriate personal cleanliness and behavior, and should not be clinically affected by communicable agents likely to be transmitted by meat.

Materials and Methods

Sample Collection

Samples were collected using sterile swab sticks from 108 contact surfaces including 36 tables, 36 hands and 36 knives of meat handlers spread across 6 towns in southeastern Nigeria. The samples were then transported in sterile containers to the laboratory at the College of Medicine and Health Sciences, Abia State University located in Aba, Nigeria. A well-structured questionnaire was used to interview the meat handlers.

Preparation of Media and Diluents

All bacteriological media (Nutrient agar, Salmonella Shigella Agar, Mannitol Salt Agar, Campylobacter Blood Free Agar, Eosin Methylene Blue Agar and MacConkey Agar) were prepared according to manufacturer's specification. Nutrient agar was used in the isolation of heterotrophic bacteria, MacConkey Agar for faecal coliform bacteria, Eosin Methylene Blue Agar for *Escherichia coli*, Campylobacter Agar for *Campylobacter* species, Mannitol Salt Agar strictly for *Staphylococcus aureus* and Salmonella Shigella Agar for the isolation of *Salmonella* and *Shigella* species. Physiological saline used as diluents was prepared by dissolving 9.8 g of sodium chloride in 1000ml of distilled and dispensed in 90 ml and 9ml portions. Both diluents and media were sterilized in an autoclave at 121^0C for 15 minutes.

Characterization and Identification of Microbial Isolates

Microbial isolates were characterized based on cultural (colonial), microscopic and biochemical methods with reference to standard manuals. The identities of the isolates were cross-matched with reference to standard manuals for the identification of bacteria. Microorganisms that were not identified by the colonial and microscopic characteristics were further subjected to few biochemical tests.

Results

Laboratory analysis of the 108 samples from the contact surfaces of the meat handlers working at the abattoirs showed that the mean colony forming units with the Eosin Methylene Blue Agar was 1.71 x 10^6 on tables, 1.50 x 10^5 on hands and 3.09 x 10^6 on knives. With the Mannitol Salt Agar, 1.63 x 10^6 on tables, 4.10 x 10^5 on hands and 8.97 x 10^5 on knives; with Nutrient Agar, 5.47 x 10^7 on tables, 2.40 x 10^7 on hands, 1.91 x 10^7 on knives; with Salmonella-Shigella Agar, 4.48 x 10^6 on tables, 5.00 x 10^4 on hands and 2.50 x 10^4 on knives; with MacConkey Agar, 5.25 x 10^7 on tables, 5.70 x 10^6 on hands and 6.06 x 10^6 on knives; with Campylobacter Blood Free Agar, 2.16 x 10^6 on tables, 6.33 x 10^4on hands and 3.22 x 10^4 on knives (Table 1). Results of the bacterial isolates of the samples showed that *Staphylococcus aureus* was isolated in 22 (61.11%) of the 36 samples from tables, 18 (50.00%) from the hands of meat handers and 15 (41.67%) from their knives; *Escherichia coli*, 18 (50.00%) from tables, 9 (25.00%) from hands and 6 (16.67%) from knives; *Shigella dysenteriae*, 17 (47.22%) from tables, 9 (25.00%) from hands and 9 (25.00%) from knives; *Klebsiella pneumonia*, 17 (47.22%) from tables, 13 (36.11%) from hands and 11 (30.56%) from knives; *Enterococcus faecalis*, 15 (41.67%) from tables, 16 (44.44%) from hands and 12 (33.33%) from knives; *Pseudomonas aeruginosa*, 10 (27.78%) from tables, 9 (25.00%) from hands and 11 (30.56%) from knives (Table 2). Response of the meat handlers when interviewed is shown in Table 3. The table showed that 2 (5.50%) meat handlers responded "yes" to worktables being clean; 5 (13.88%) to cleaning of utensils; 6 (16.67%) to wearing proper clothing; 4 (11.11%) to wearing of hand gloves; 5 (13.88%) to routine washing of hands with soap and water; and 11 (30.56%) to being free from skin/enteric illness.

Table 1: Bacteria mean colony forming units (CFU/g) of contact surfaces

Location	EMBA	MSA	NA	SSA	MCA	CAM
Tables	1.71×10^6	1.63×10^6	5.47×10^7	4.48×10^6	5.25×10^7	2.16×10^6
Hands	1.50×10^5	4.10×10^5	2.40×10^7	5.00×10^4	5.70×10^6	6.33×10^4
Knives	3.09×10^6	8.97×10^5	1.91×10^7	2.50×10^4	6.06×10^6	3.22×10^4

EMBA- Eosin Methylene Blue Agar; SSA- Salmonella-Shigella Agar; MSA- Mannitol Salt Agar; NA- Nutrient Agar; CAM- Campylobacter Blood Free Agar; MCA- MacConkey Agar

Table 2: Distribution of bacterial isolates from contact surfaces of meat handlers

Bacteria	Table		Hand		Knife	
	n	%	n	%	n	%
Staphylococcus aureus	22	61.11	18	50.00	15	41.67
Escherichia coli	18	50.00	9	25.00	6	16.67
Shigella dysenteriae	17	47.22	9	25.00	9	25.00
Klebsiella pneumonia	17	47.22	13	36.11	11	30.56
Enterococcus faecalis	15	41.67	16	44.44	12	33.33
Pseudomonas aeruginosa	10	27.78	9	25.00	11	30.56
Salmonella enteritidis	8	22.22	0	0.00	2	5.56
Campylobacter jejuni	19	52.78	17	47.22	6	16.67
Bacillus cereus	7	19.44	11	30.56	4	11.11
Campylobacter coli	0	0.00	0	0.00	0	0.00
Serratia sp.	0	0.00	0	0.00	0	0.00
Staphylococcus saprophyticus	6	16.67	4	11.11	6	16.67
Enterobacter sp.	2	5.56	0	0.00	0	0.00

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Bacillus licheniformis	0	0.00	0	0.00	0	0.00
Micrococcus luteus	0	0.00	0	0.00	0	0.00
Micrococcus roseus	0	0.00	0	0.00	0	0.00
Bacillus subtilis	0	0.00	4	11.11	2	5.56

Table 3: Response on sanitation and hygiene measures by meat handlers

Measures for Sanitation and hygiene	**Yes**		**No**	
	n	**%**	**n**	**%**
Worktables and work surfaces are clean	2	5.50	34	94.50
Small equipment and utensils including cutting boards, knives, etc. are thoroughly cleaned	5	13.88	31	86.12
Meat handler wear proper clothing	6	16.67	30	83.33
Wearing of hand gloves	4	11.11	32	88.89
Hands are washed routinely with soap and water	5	13.88	31	86.12
Meat handlers are free from skin/enteric illnesses	11	30.56	25	69.44

Discussion

The predominant bacterial isolates found on the contact surfaces were *Staphylococcus aureus, Enterococcus faecalis*, *Escherichia coli*, *Campylobacter jejuni*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia* and *Shigella dysenteria*. *Staphylococcus aureus* was the most isolated bacteria in this study. It is a commensal and opportunistic pathogen that can cause wide spectrum of infections, from superficial skin infections to severe, and potentially fatal, invasive disease[11]. Meat handlers who had skin infections could easily contaminate the meat during cutting and processing. Cross contamination from infected equipment could also easily occur. Staphylococcal Food-Borne Disease is one of the most common Food-Borne Disease and is of major concern in public health programs worldwide[12]. A typical Food-Borne Disease caused by *S. aureus* has a rapid onset following ingestion of contaminated food. This is due to the production of one or more toxins by the bacteria during growth at permissive temperatures[13]. Symptoms include hyper salivation, nausea, vomiting, and abdominal cramping with or without diarrhea. If significant fluid is lost, physical examination may reveal signs of dehydration and hypotension. Abdominal cramps, nausea, and vomiting are the most common[14].

Klebsiella pneumoniae is another opportunistic pathogen found in the environment and in mammalian mucosal surfaces. The principal pathogenic reservoirs of infection are the gastrointestinal tract of mammals. Infection with *Klebsiella pneumoniae* occurs in the lungs, where they cause destructive changes[15]. Infected meat handlers could have contaminated the meat carcasses during cutting and processing of meat especially when they were not wearing face masks. Infection with *Klebsiella pneumoniae* can cause necrosis, inflammation, and hemorrhage within lung tissues, sometimes producing thick, bloody, mucoid sputum described as currant jelly sputum[16]. The illness typically affects middle-aged and older men with debilitating diseases such as alcoholism, diabetes, or chronic bronchopulmonary disease. This patient population is believed to have impaired respiratory host defenses[17]. Sepsis and septic shock may follow entry of organisms into the blood from a focal source.

Campylobacteriosis is an infection by *Campylobacter* bacterium commonly called *C. jejuni*. This was another common bacteria isolated in the contact surfaces. It produces an inflammatory, sometimes bloody, diarrhea or dysentery syndrome, mostly including cramps, fever and pain. The prodromal symptoms are fever, headache, and myalgia, which can be severe, lasting as long as 24 hours. After 1–5 days, typically, these are followed by diarrhea or dysentery, cramps, abdominal pain, and fever as high as 40 °C[18]. Some common routes of transmission for the disease-causing bacteria are fecal-oral and waterborne. The meat carcasses can possibly be contaminated with *C. jejuni* through flies that perch on feces and then transport them to the meat carcasses, or cross contamination from infected meat handlers. A study by Wilson and Gabriel[19] found that 97 percent of campylobacteriosis cases sampled in Lancashire were caused by bacteria typically found in livestock and in 35 percent of cases, the bacteria could be traced to cattle. *Escherichia coli* is commonly found in the lower intestine of warm-blooded organisms. Most *E. coli* strains are harmless, but some serotypes can cause serious food poisoning in their hosts[20]. *E. coli* is expelled into the environment within fecal matter. The bacterium grows massively in fresh fecal matter under aerobic conditions for 3 days, but its numbers decline slowly afterwards[21]. Fecal–oral transmission is the major route through which pathogenic strains of the bacterium cause disease. Hence, cross contamination may have occurred from the tables used for the cutting and skinning of meat or from contaminated equipment carried by vectors such as flies. Common signs and symptoms include severe abdominal cramps, diarrhea, hemorrhagic colitis, vomiting, and sometimes fever[21]. Salmonellosis is a symptomatic infection caused by *Salmonella enteritidis*. The most common symptoms are diarrhea, fever, abdominal cramps, and vomiting[22]. Most people with salmonellosis develop diarrhea, fever, vomiting, and abdominal cramps 12 to 72 hours after infection[23]. Diarrhea is often mucopurulent and bloody. In most cases, the illness lasts four to seven days, and does not require treatment. In severe cases, the *Salmonella* infection may spread from the intestines to the blood stream, and then to other body sites and can cause death, unless the person is treated promptly with antibiotics. Shimelis, *et al.*[24]studied the microbial safety in beef at selected slaughter houses and reported *E. coli* and *Salmonella* species to be common bacterial isolates with sources of contamination to include equipment, transport vehicle, cutting board and worker's hand.

The personal hygiene practices of the meat handlers was a major source of concern as the meat handlers failed to meet the basic standards of personal hygiene during the handling of meat. Most of the meat handlers do not wear proper clothing such as aprons and hair restraints when handling meat. Their fingers were dirty, some had open cuts and wounds on their hands and they did not practice regular hand washing with soap and potable water. By touching their hands on dirty contact surfaces, they can transfer microorganisms to the meat. Also by coughing and sneezing with their bare hands, microorganisms can be transferred to the meat as they handle the meat without using gloves. Several studies[25-27] have reported the occurrence of meat borne diseases attributed to the poor personal hygiene of meat handlers. Open wounds and cuts on the hands of the meat handlers can easily get infected and this can be transferred to the meat sold to the public. The cleanliness and sanitary conditions of the workplace of the meat handlers was also found to be below standard. Their work tables and work surfaces were not kept clean; there were no cleaning procedures that were kept in place for the cleaning of the premises, equipment and utensils. All these factors can be attributed as possible sources of contamination of the meat carcasses.

In conclusion, the sanitary and personal hygiene practices of the meat handlers were very poor and this resulted to the high bacterial isolates found on their tables, hands and knives. Meat handlers must adhere to proper hygiene and sanitary measures to prevent cross-contamination of microorganisms to the meat carcasses. Health regulatory agencies must embark on routine inspection of abattoirs to ensure compliance of meat handlers to meat safety standards.

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