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THE SUSCEPTIBILITY PATTERN AND EFFICACY OF COMMERCIALLY AVAILABLE DISINFECTANTS TO STAPHYLOCOCCUS AUREUS ISOLATED FROM BUTCHERS TABLE IN ABAKALIKI MEAT MARKET

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Abstract
This study investigated the susceptibility pattern and efficacy of commercially available disinfectants to Staphylococcus aureus isolated from butchers table in Abakaliki meat market. Butchers' tables were swabbed using appropriately labelled sterile cotton swab sticks. The labelled swab sticks were transported to the laboratory for immediate microbiological analysis. The susceptibility pattern of Staphylococcus aureus was determined using the modified Kirby-Bauer susceptibility test method using Dettol, Purit, Isol, and Savlon for which the percentage susceptibility at 25% and 50% concentrations were 78.6% (22), 78.6%(22), 96.4% (27), 82.1%(23) and 89.3%(25), 89.3%(25), 92.9%(26), and 78.6%(22) respectively. This indicates the occurrence of antimicrobial resistant strains of food borne Staphylococcus aureus. Effective cleaning and a safe level disinfection of butchers environment (implements, wooden slabs, and tables) as a means of safe guarding consumers' health is strongly recommended.

Keyword: *Susceptibility, Pattern, Efficacy, Disinfectants and Staphylococcus aureus*

Background to the Study

Food-borne pathogens are the leading cause of illness and death in developing countries resulting in the loss of labor force which could have contributed in the economic growth (Fratamico *et al;* 2005). *Staphylococcus aureus* are Gram positive facultative anaerobic spherical bacteria that produces a very heat stable toxin. The bacteria are borne by humans as part of body normal flora of the skin, nasal nares which are transported to food. The bacteria grow best at our body temperature and also at room temperature. *Staphylococcus aureus* can multiply rapidly in food held at room temperature and the toxin can be produced by the microorganism growing in the food. This toxin is called enterotoxin because it causes gastroenteritis or inflammation of the lining of the

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intestinal tract. (Centre for Disease Prevention and Control, 2013). Contaminated raw meat is one of the main sources of food-borne illnesses (Bhandare *et al;* 2007). The risk of the transmission of zoonotic infections is also associated with contaminated meat (Hedberg *et al;* 1992). International Food Management Agencies, especially the World Health Organizations (WHO), the Food and Agriculture Organization (FAO) and the International Hazard Analysis Critical Control Point Alliance (HACCP) have already provided guidelines to member countries about safe handling procedures such as HACCP and Good Manufacturing Practices (GMPs).

The widespread habit of raw beef consumption is a potential cause if food-borne illnesses besides the common factors such as over-crowding, poverty inadequate sanitary condition and poor general hygiene (Siddiqui *et al;* 2006). Ikeme (1990) noted that sources of surface fresh meat contamination could be from polluted air in the environment, unsterile knives, equipments and utensils, clothing and hands of personnel. Antibiotic resistance levels are also elevated among food-borne pathogens as in *staphylococcus* (Mache, 2002). Although, it is difficult to prove a direct role of drug resistance in bacteria contaminating food items with increased clinical cases of resistant infections, the presence of such bacteria in food items could play a role in the spread of antimicrobial resistance amongst food-borne pathogens (Farzana *et al;* 2009). Lack of awareness about food safety and hygiene among butchers results in food contamination, the butchers or personnel can be carriers of this pathogen who eventually transfer these food-borne pathogen to consumers (Toit *et al;* 2005).

Objective of the Study

The objective of this study is to investigate the susceptibility pattern and efficacy of commercially available disinfectants to *Staphylococcus aureus* isolated from butchers table in Abakaliki meat market. Butchers' tables were swabbed using appropriately labelled sterile cotton swab sticks.

Materials and Method

Study Area/Setting: The samples for study were obtained from meat market butchers tables. Meat market is located along gunning road intersecting Abakpa main market in Abakaliki metropolis. Meat market slaughterhouse is the largest of its kind in Abakaliki city. Abakaliki coordinates 6°20'N 8°06'E / 6.333°N 8.100°E with a total population of one hundred and forty one thousand four hundred and thirty eight (141,438) people as at 2006 population census. The propensity of food-borne pathogen contamination is high owing to the overcrowded nature of the market, as well as unchecked and bad drainage system in the market.

Sample Collection: Twenty-eight swab samples were collected at random from the butchers table using sterile swab sticks. The samples were sent to the microbiology laboratory immediately for analysis.

Bacteriological Analysis: All the samples were aseptically cultured on bacteriological culture media including nutrient agar, mannitol salt agar, and nutrient broth and the plates were incubated at 37°C for 18-24 hrs. Each of the samples was first inoculated in nutrient broth overnight at 37°C prior to subculture onto solid culture media plates as aforementioned. Suspect colonies were subcultured onto fresh culture plates to get pure cultures (Cheesbrough, 2006).

Morphological Characterization: The morphological characteristics of colonies of the pure culture growing on the media were examined as described by (Clement *et al;* 2002) with reference to their sizes, pattern of edge and margin, surface texture, elevation, consistency and colour.

Antimicrobial Susceptibility Test: Antimicrobial susceptibility test of *S. aureus* was performed on Mueller Hinton agar (oxoid, England) using antimicrobial disinfectants diluted at 50% and 25% concentration. The disinfectants used were Dettol, Savlon, Purit, Isõl diluted at 50% concentration and 25% concentration respectively. The resistance and susceptibility patterns of *S. aureus* were determined by the modified Kirby-Bauer susceptibility test method as recommended by the NCCLS (now CLSI). Morphologically identical 4-6 bacterial colonies from an overnight culture were suspended in 5ml nutrient broth and incubated for 4 hours at 37C. Turbidity of the broth culture was equilibrated to match 0.5McFarland standards. The surface of Mueller Hinton agar (oxoid, England) plate was evenly inoculated with the cultures using a sterile cotton swab stick. After this, four (4) holes were pierced through the surface of the Mueller-Hinton agar media designated "S", "P", "I", and "D", denoting savlon, purit, Isõl and Dettol respectively using a sterile cork borer. A drop of these antimicrobial disinfectants was placed in each of these holes using a Pasteur pipette. After 18-24 hours of incubation, the diameter of growth inhibition was measured and interpreted as sensitive or resistant by the NCCLS (NCCLS, 1999).

Result

The result of this research work which was carried out using twenty-eight swab samples collected from twenty-eight different butchers table at meat market in Abakaliki metropolis following routine bacteriological analytical procedure and biochemical characterization are tabulated as:

Table 1: Diameter of Inhibition (Mm) Reading at 50% Concentration of Disinfectants

Sample (S/No)		D	P	I	S
1.	1.	30mm	28mm	27mm	30mm
2.	2.	27mm	32mm	40mm	22mm
3.		31mm	28mm	25mm	28mm
4.		27mm	27mm	29mm	38mm
5.		27mm	20mm	30mm	20mm
6.		33mm	30mm	25mm	40mm
7.		R	25mm	24mm	R
8.		28mm	25mm	26mm	28mm
9.		25mm	30mm	20mm	R
10.		20mm	24mm	28mm	27mm
11.		28mm	29mm	36mm	30mm
12.		27mm	25mm	30mm	20mm
13.		30mm	31mm	35mm	33mm
14.		25mm	R	25mm	R
15.		20mm	28mm	24mm	R
16.		20mm	30mm	35mm	20mm
17.					
18.		30mm	29mm	32mm	30mm
19.		30mm	38mm	25mm	25mm
20.		30mm	33mm	29mm	R
21.		30 mm	34mm	30mm	32mm
22.		R	20mm	25mm	20mm
23.		32mm	30mm	32mm	32mm
24.		30mm	30mm	35mm	30mm
25.		25 mm	20mm	27mm	30mm
26.		28mm	20mm	27mm	20mm
27.		R	R	R	20mm
28.		25mm	R	R	R
29.		28mm	20mm	32mm	20mm

Key: D=Dettol, P=Purit, I=Isol, S=Savlon, R=Resistance

Table 2: Diameter of Inhibition (Mm) At 25% Concentration of Disinfectants

Sample (S/No)	D	P	I	S
1.	25mm	29mm	32mm	25mm
2.	24mm	20mm	32mm	24mm
3.	28mm	20mm	30mm	23mm
4.	32mm	26mm	32mm	30mm
5.	28mm	21mm	35mm	25mm
6.	30mm	26mm	32mm	30mm
7.	32mm	27mm	24mm	28mm
8.	R	24mm	32mm	28mm
9.	34mm	32mm	30mm	30mm
10.	36mm	24mm	38mm	25mm
11.	20mm	23mm	24mm	25mm
12.	R	25mm	28mm	20mm
13.	22mm	22mm	20mm	20mm
14.	R	R	25mm	20mm
15.	25mm	21mm	26mm	23mm
16.				
17.	22mm	22mm	36mm	30mm
18.	26mm	29mm	28mm	26mm
19.	28mm	R	30mm	R
20.	22mm	R	28mm	R
21.	26 mm	20mm	28mm	20mm
22.	20mm	R	22mm	20mm
23.	20mm	24mm	28mm	30mm
24.	30mm	25mm	30mm	22mm
25.	26 mm	22mm	30mm	R
26.	R	28mm	22mm	20mm
27.	R	R	25mm	R
28.	20mm	R	26mm	R
29.	R	R	R	R

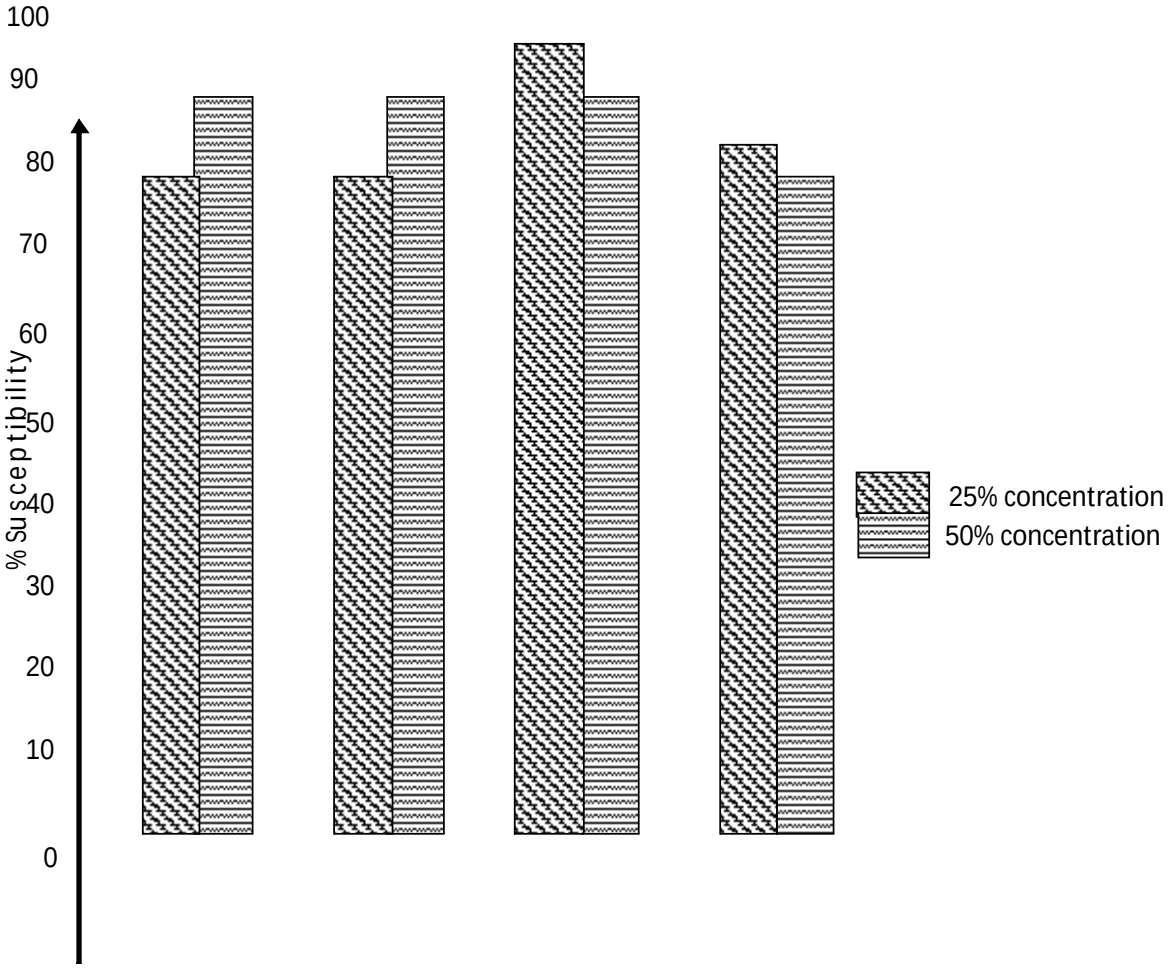
Key: D=Dettol, P=Purit, I=Isol, S=Savlon, R=Resistance

Table 3: Percentage Susceptibility at 25% & 50% Concentration of Antimicrobial Disinfectants

(%)Dilution	Dettol	Purit	Isol	Savlon
25	78.6%(22)	78.6%(22)	96.4%(27)	82.1%(23)
50	89.3%(25)	89.3%(25)	92.9%(26)	78.6%(22)

Table 4: Percentage Resistance at 25% & 50% Concentration of Disinfectants

(%)Dilution	Dettol	Purit	Isol	Savlon
25	21.4%(6)	21.4%(6)	3.6%(1)	17.9%(5)
50	10.7%(3)	10.7%(3)	7.1%(2)	21.4%(6)



Discussion, Conclusion and Recommendation

The result of this study showed that at 50% concentration of the disinfectants, *Staphylococcus aureus exhibited* a high percentage of susceptibility as against *Staphylococcus aureus susceptibility* at 25% concentration of the disinfectants. The susceptibility readings at 50% concentration of disinfectants are 89.3%(25) for Dettol, 89.3%(25) for Purit, 92.9%(26) for Isol, and 78.6%(22) for savlon while at 25% concentration of the disinfectants it reads 78.6%(22) for Dettol, 78.6%(22) for purit, 96.4%(27) for Isol, and 82.1%(23) for savlon, with Isol at its peak in both the two tests. This is in agreement with (Russell and McDonnell, 2000) that the efficacy of antimicrobial products may depend on, and vary significantly with the formulation used. However, from this study, *S. aureus* are more sensitive to Isol than the rest of the disinfectants. At 25% concentration of the disinfectants, Dettol is 21.4% resistant, Purit 21.4%, Isol is 3.6%, Savlon is Dettol is 10.7%, Purit is 10.7%, Isol is 7.1%, Savlon is 21.4%. It was observed that *Staphylococcus aureus* was least resistant to Isol at both 25% and 50%

concentration. This is followed by Dettol and Purit. The scientific committee on Emerging and Newly Identified Health risks (2009) reported that bacteria resistance arises from a mechanism causing decrease in intracellular concentrations of biocides below a threshold level that is harmful to the bacterium. McDonnell and Russell (2000) also found put that resistance is either a hereditary natural property of an organism, or acquired by mutation and acquisition of plasmids or transposons. However, this resistance is likely due to the presence of active efflux. In appropriate use of antimicrobial agent as in low dosage, short contact, or irregular application is primarily responsible for the emergence of many resistant bacteria species including staphylococci (Yilmax and Kaleta, 2009). Thus, resistance to disinfecting agents induced by sub-lethal concentrations of the active compound will significantly increase minimum inhibitory concentration values for most antimicrobials. Hogan and Smith (2008) who tested eight strains of *S. aureus* to determine if repeated in vitro exposure to sub-lethal concentrations of four commercial biocides could enhance bacterial tolerance. They found that the responses of *S. aureus* to chlorhexidine, sodium hypochlorite and iodophor were not affected by prolonged exposure to these reagents. Efflux pumps have been shown to decrease the efficacy of numerous biocides including quaternary ammonium compounds, phenolic parabens, and intercalating agents, most notably in *S. aureus* that express pumps such as QacA-D, Qach, and QacH.

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