

AFS 2009087/10308

Bacteriological Assessment and Hygienic Standard of Food Canteens In Kwara State Polytechnic, Ilorin, Nigeria

A. T. Ajao¹ and T. G. Atere²

¹Department of Biology, Kwara State Polytechnic, Ilorin, Nigeria.

²Department of Chemistry, Kwara State Polytechnic, Ilorin, Nigeria.

Corresponding author E-mail: ajaoabdullahi@yahoo.com, Tel: +2348060276738

(Received August 1, 2009)

ABSTRACT: This study reports the status of hygiene standard and bacteriological quality of food provided at the Kwara state Polytechnic, food canteens. Six canteens were randomly selected for the study. Ten assay visits were carried out and total bacterial counts from various environment surfaces, palms of the food handlers and various denominations of money from food sellers were determined using Agar Syringe Method containing nutrient agar for the evaluation of hygiene quality index. Food quality was also determined by testing for total viable counts, coliform, *Salmonella*, *Pseudomonas* and *Staphylococcus* counts. The sensitivity of the bacterial isolates to some antibiotics was also evaluated. Result of the study indicated that clean plates, walls tables and serving counters have high hygiene quality indices of 75%, 91%, 62% and 65% respectively. While floor, hand towels, apron, money and hand swabs have low hygiene quality indices of 53%, 36%, 56%, 46% and 40% respectively. Pudding and stew samples were of low microbial and compare to water and vegetable. Coliforms (10.8%), *Salmonella* sp (11.0%), *Enterobacter* sp (10.3%) and *Bacillus* sp (12.5%) were mostly occurred during the sampling period. The isolates were sensitive to some antibiotics while some were resistant. This study suggest that there is need to improve on hygiene practices in Kwara state polytechnic canteens in order to prevent or reduce as the case may be the incidence of food borne disease.

Key words: Hygiene, Canteen, Bacteriological assessment.

Introduction

Biological contaminants such as bacteria viruses, fungi, protozoa and helminthes constitute the major cause of food borne disease with varying degrees of severity ranging from mild indisposition to chronic or life-threatening illness or both. In developing countries such contaminant are responsible for food borne diseases such as cholera, campylobacteriosis *E.coli* gastroenteritis, salmonellosis, shigellosis, typhoid fever, brucellosis, amoebiasis and poliomyelitis (Edema *et al.*, 2005)

Aerobic colony counts, coliform and enterococci enumeration are useful and most often used means of assessing overall sanitation in the environments of food service establishment. (Collins, 1964; Jay, 1978; Moyo and Baudi, 2004).

The presence of indicator organisms, pathogens or high bacterial counts in food stuffs, food contact surfaces, equipment and utensils provides a direct and relevant measure of cleaning efficiency and hygiene. (Clark, 1965; Moyo and Baudi, 2004). However, when a contamination of food by a pathogen occurs in a canteens, restaurants, fast-food services and cafeterias as a result of failure to observe proper sanitation, improper cooling of foods, cross-contamination and long interval between preparation and consumption, a large number of people over a wide area will be affected. (Bean *et al.*, 1990).

Effective cleaning is of prime importance since it not only removes gross contamination but also any residues that could support the subsequent survivals and growth of microorganisms. (Clark,1965). As a result of inadequate cleaning micro organisms may persists on utensil and work surfaces and build in numbers (Fraizer and Westhoff,1988). The main objectives of sanitations is to minimize the access of microorganisms in food from various sources at all stages of handling (Marrioat,1989; Cords and Dychdala,1993).

Even though epidemiological evidence on outbreak of food borne disease is scarce, there are indications that foods could be contaminated to unsafe levels at the points of consumption with air flora and other microorganisms from handlers, equipments/utensils and the raw food materials. (Edema *et al.*,2008).

Effective hygiene control through bacteriological testing is vital to ensure acceptable levels of contamination and avoid adverse human health consequences of food borne illness. (Moyo and Baudi, 2004)

Kwara State Polytechnic, Ilorin canteens are commercial cantering establishment than services the campus populace both the students and lecturers.. Incidence of diarrhoea and abdominal pains are mostly prevalent at the school clinic following eating at the school canteens, such cases are presumed to be "microbial food poisoning" even though, no clinical or laboratory finding are provided. Therefore, The hygiene standards of the food preparation areas, utensils as well as the personal hygiene practice of some of the kitchen personnel are questionable

The present investigation concentrates on the status of hygiene standard of food canteen and microbiological quality of food provided. To asses hygiene within canteen kitchens and to determine the incidence of bacterial pathogen in the catering kitchen environment, hence to determine the antibiotics sensitivity of the isolates.

Materials and Methods

Environmental sampling for the detection and enumeration of microorganisms was carried out in the Kwara State Polytechnic, canteens using the Agar syringe method (Jay, 1987; Moyo and Baudi, 2004). Nutrient Agar syringe swabs were used for the determination of total viable counts, MacConkey Agar syringe swabs, Mannitol Salt Agar syringe for the qualitative determination of indicator organism and/or suspected pathogenic bacteria on predefined surface and for food handler swabs and money from the sellers.

Environmental surfaces sampled in this study include the following areas: Palm swabs of food handlers, hand towel, clean plates, walls, floor, small utensils, table, water, food (Pudding), stew and apron from any four randomly selected kitchen personnel. However sampling was done around 12:00noon after major cleaning and disinfections operations were completed at Kwara state polytechnic canteens

Air Sampling

Fifteen plates (3 plates for each type of medium) were exposed to the air at 12:00noon for 10mins in every assay visit. The exposure plates were incubated at 37⁰c for 24 hours and the developing colonies were counted and identified.

Agar Syringe Swab Preparation

The ends were cut from the needle ends of hypodermic syringes to create hollow cylinders of approximately 3.5mm in diameters. These were autoclaved at 121⁰c for 15mins. Sterile syringe cylinder were filled with agar medium incorporating a disinfectant quencher/neutralizer. Tween 80 and left to set and wrapped with Aluminum foil before used. Each of the sterile syringe cylinder contained different selective medium. Swabbing was carried out following method of (Jay, 1987; Moyo and Baudi, 2004).

Five slices of inoculated media from each predefined area were placed in each petri-dish and labelled accordingly. Incubation was done at 37⁰c for 18-24hours resultant colonies on each agar slice were counted.

Food Sample collection: Pudding, stew, water, vegetable and rice sold in Kwara State Polytechnic, canteen were sampled to assess the nature and degree of bacterial contamination. Sampling was conducted dividing each food container into four quadrants and a portion of each food from each quadrant was randomly collected. Total viable counts (TVC), coliforms, *Staphylococci*, *Streptococci*, *Salmonella*, faecal coliforms and *Pseudomonas sp* were used to determine the food quality

Food homogenate preparation

Each food sample was analyzed separately. The samples were cut into small pieces with a sterile scalpel in a mortar or sterile jar. About 10g of representative sample of each food specimen was weighed out and suspended in 90ml of peptone water and thoroughly mixed. This represented a dilution of 10^{-1} . 1ml of 10^{-1} dilution was pipette and serially diluted up to 10^{-6} . Inoculum of about 0.2ml of the selected dilution was spread on triplicate plates using glass spreader (Fawole and Oso 2001); (Oranusi *et al*; 2003; Moyo and Baudi 2004).

This technique was used for the enumeration of total viable counts, coliforms, *Staphylococcus* counts, *Salmonella*, faecal coliform and *Pseudomonas* (Okonko *et. al.*, 2009). On nutrient Agar, Samonella-Shigellar Agar (Oxoid), MacConkey Agar, Blood Agar, Eosin Methylene blue (EMB) agar and Mannitol salt agar. All cultures were incubated at 37°C and 44°C. Pure culture obtained were characterized using convention methods (Buchanan and Gibbons, 1974; Cowan and Steel, 1985).

Determination of *Staphylococcus aureus*

The suspected *staphylococcus* colonies were observed from blood agar plates and Mannitol salt agar plates. Presumptive *S. aureus* colonies were picked and a glass slide coagulase test carried out for the definitive identification of *S. aureus*.

Determination of *Streptococci*

Blood Agar plates were examined for typical streptococci either (α -hemolytic (green zone) or (β -haemolytic (clear zone). Suspected colonies were picked and inoculated onto Aesculin agar test for Aesculin hydrolysis and catalase test.

Data Analysis

Average bacterial counts from nutrients agar slices for each sampled area were used to calculate the overall hygiene quality indices indicative of the cleaning and disinfection efficacy for the Kwara state polytechnic canteen the environmental grade scale in Table 1 was used to calculate the scores. The overall hygiene indices were calculated using the following formula

$$\text{Hygiene quality index (\%)} = \frac{1 - \text{Sum of Score grade for } n \text{ observation}}{5 \times n \text{ Observation}}$$

Food handlers' hand swab average bacterial counts were used qualitatively to calculate a hygiene quality index for the kitchen personnel for the entire assessment period (Moyo and Baudi 2004).

Table 1: Environment hygiene grade scale for a 3.5mm diameter agar syringe

Growth On media	Score
Negative	0
Slight growth (10<colonies)	1
Little growth (10-30 colonies)	2
Moderate growth (up to 80 colonies)	3
Heavy growth (over 80 colonies)	4
Lawn (<200 colonies)	5

Source: Thornton, 1974, Moyo and Bauch; 2004

Sensitivity test of bacteria isolates to antibiotics

The disc diffusion method was used commercial antibiotics disc of known concentration were placed on Mueller-Hinton agar previously seeded with the bacteria isolates, the plates plated were examined for zones of inhibitions around the antibiotics disks and measured in mm (Uzeh and Adeniji, 2006).

Results

Table 2 Major bacterial isolates and overall hygiene quality indices from environmental surfaces and hand swabs from kitchen personnel for the entire sampling period.

Area	Bacterial isolate	Quality index (100%)
Floor	Pseudomonas, Enterobacter, coliform	53
Clean Plates	Coliforms, Pseudomonas	75
Walls	Streptococcus, Enterobacter	36
Hand towels		91
Apron	Staphylococcus, Pseudomonas, Coliforms	56
Table	Streptococci, Coliform, Pseudomonas	62
Money		46
Serving counter	Staphylococcus Streptococci E.coli	40
Hand swab	Pseudomonas, Staphylococcus, streptococci, E.coli, Coliform	65
	Coliform, Staphylococcus, Streptococcus	
	Streptococci, staphylococcus, Coliform, E.coli	
	Enterobacter	

Table 3: Frequency of occurrence of isolates for the entire sampling period.

Bacterial Isolate	Frequency	A	B	C	D	E	F	%
<i>E. Coli</i>	31	1	4	3	6	9	8	17%
<i>Salmonella</i> sp	11		3	1	4	2	1	6.0%
<i>Staphylococcus</i> sp	32	4	7	5	5	7	4	17.5%
<i>Enterobacter</i> sp	32	7	4			5	4	10.9%
<i>Micrococcus</i> sp	7	-	-	1	3	2	1	3.8%
<i>Bacillus</i> sp	28	6	4	4	3	5	6	15.3%
<i>Streptococcus</i> sp	35	8	4	6	4	8	5	19.2%
<i>Serratia</i> sp	4	1	-	-	1	1	2	2.1%
<i>Klebsiella</i> sp	11	2	2	1	3	2	1	6.0%
<i>Proteus</i> sp	3	-	-	1	-	2	-	1.6%
TOTAL	182							

TABLE 4: Bacterial Loads of Food Samples for Different Canteens in Kwara State Polytechnic, Ilorin

Bacteria	Pudding cfu/ml	Stew cfu/ml	Vegetable cfu/ml	Water cfu/ml	Rice cfu/ml
TVC	7.3×10^3	4.1×10^2	8.2×10^5	94×10^5	7.1×10^2
<i>Staphylococcus</i> sp	2.7×10^3	0.2×10^2	6.5×10^4	5.6×10^5	0.32×10^2
<i>Pseudomonas</i> sp	-	-	2.5×10^2	3.4×10^3	-
Feecal coliform	-	-	4.3×10^2	4.0×10^2	-
Coliform counts	0.7×10^2	0.12×10^2	7.6×10^1	6.3×10^4	0.11×10^2
<i>Salmonella</i> sp	0.2×10^2	-	0.4×10^2	0.68×10^3	-

Keys: Counts are mean of triplicates samples TVC = Total Viable Count

TABLE 5: Antibacterial Activity of Some Antibiotics Against The Isolates.

Isolate	CIP	OFL	ERY	GEN	PEN	STR	CHL
Pseudomonas	18±0.32	27±0.65	-	-	-	12±0.21	19±0.12
Staphylococcus	14±0.31	21±0.51	11±0.0	7 ±0.51	16±0.11	5 ±0.67	28±0.36
Escherichia coli	8±0.12	5±0.42	-	-	-	14±0.25	22±0.16
Salmonella sp	27±0.72	21±0.87	13±0.23	16±0.43	-	-	-
Klebsiella sp	-	5.3±0.48	17±0.69	-	-	13±0.79	11±0.26
Streptococcus sp	8±0.11	13±0.46	28±0.31	-	20±0.23	14±0.13	28±0.34

Result are the means of diameter value ± standard deviation.

KEY: CIP Ciprofloxacin

OFL: Ofloxacin

ERY: Erythromycin

GEN: Gentamycin

STRP: Streptomycin: No zone of inhibition

TABLE 6: Airflora of Food Canteen in Kwara State Polytechnic, Ilorin for the Entire Sampling Period.

Location	Aerobic mesophilic bacteria	Salmonella sp	Staphylococcus SP	Pseudomonas SP	Streptococcus SP	Enterobacter
A	16±2.11	14±4.10	13±3.4	16±4.16	11±1.12	17±3.72
B	22±3.43	8±0.83	18±5.42	11±3.96	18±6.00	13±3.37
C	16±2.42	6±0.42	18±5.42	11±3.96	18±6.00	13±3.37
D	21±4.13	2±0.14	13±3.31	7±1.71	21±6.86	18±1.12
E	32±6.83	9±0.0	21±6.42	13±7.31	12±2.36	15±3.23
F	23±4.01	5.5±0.24	17±3.6	10±1.15	18±5.31	15±86

Key: counts are means of triplicates samples

Discussion

In this research work all palms swabs, money, apron, hand towel sampled harbored coliforms, *Staphylococcus* and *Pseudomonas* with poor quality index of 40%, 46% 56% and 36% respectively (Table 2) while *Streptococcus*, *E.coli* and *Bacillus* spp were mostly occurred in the air of canteen during the sampling period in 19.2%, 17.5% 17% and 15.3% respectively (Table 3). Also most of the water and

vegetable sampled harbored coliforms, *Salmonella* and *Pseudomonas* (Table 4) Total Plate Counts (TVC) was used to measure the general bacterial load of the food sample and is useful tools in monitoring food process and the results may reflect the hygienic level of food handling and retail storage (Collins *et al.*, 1989).antibacterial activity of some antibiotics against the isolates were represented in (Table 5),while air flora of the food canteens were shown in (Table 6).

Food borne illness can be prevented by good hygiene practice during the preparation of food (Scott, 2002). To prevent occurrence of food borne, illness it is therefore, important to ensure that foods sold are safe and hygienic.

However the pathogen isolated in this present study are similar to micro organisms reported by Bankole *et al.*, (2004, 2005) and (Okonko *et al.*, 2008).In a similar study where all palms food vendors and hotel operators harboured *Staphylococcus aureus*. hotel operators among the food vendors sampled were reported to have harboured the least types of micro organisms. (Olawale *et al.*,2005) reported nine bacterial genera and two fungi in a similar study.

The quality indices for the hand towel, apron, table and money were very low, which are characterized by high number and many kinds of primary contaminant bacteria as a result of poor hygienic practice (Fraizer and Westhoff, 1988). For proper handling of food it is required that hand towel, apron and palms of the kitchen personnel should be given special added attention of through and regular cleaning as soon as possible after daily work or having more than one hand towel and apron which evidence was not being given by the cleaning staff. The *E. coli* *Pseudomonas* *Staphylococcus* and coliform isolated from the surfaces of sampled money could probably have come from the raw food materials, water, apron, dust and palms of the customers. *E.coli*, *Pseudomonas*, Coliform and *Staphylococcus* isolated from the hand swabs of the kitchens personnel, money, serving counter and apron is an evidence of cross contamination.

The presence of indicator and other organisms examined in this study is of special concern and perhaps the greatest danger associated with the water for food processing and drinking purposes (Edema *et al.*, 2001; Okonko *et al.*, 2008).

Isolation of *Staphylococcus aureus* and *Salmonella* spp in this study is of practical impact. It is an evidence of poor sanitary conditions and lack of inadequate portable water (Ibekwe *et al.*, 2008).

According to Eley (1992) the presence of total coliforms and *E.coli* in food may indicate faecal contamination which could be due to insufficient cooking cross contamination and contaminated ingredients, or possibly from the kitchen personnel

Qualitative hand swab results showed that a high fraction of the personnel's hands were contaminated by coliform. *E.coli* and *Enterobacter* even though the source of those contaminants was not determined they are highly indicative of inadequate hand sanitations. (Thatcher and clark 1973). The *staphylococcus* spp and *Streptococcus* isolated although they are normal commensal on humans reflect improper hygiene practice such as poking fingers into the nose Eley 1992.

It was observed that their was no hand sanitizer (soap or otherwise) at each of the hand wash basins for personnel to wash their hand with after using the toilet or handling food and/or handling raw foods such as meat. A common practice is to dry their hands after washing with their apron, these garments could then probably serve as sources of further contamination this was also reported in a similar study by (Moyo and Baudi 2004) in the similar study.

E.coli and *Pseudomonas* sp was resistant to the antibiotics used in this study. It was however sensitive to gentamycin in agreement with (Uzeh *et al.*, 2006)

(Okeke *et al.*, 2000) reported in the antibiotic resistance of *E.coli* isolated from Nigerian Students. *Salmonella* and *Staphylococcus* were mostly sensitive to the tested antibiotics among the five isolates investigated. They were sensitive to ciprofloxacin, ofloxacin, erythromycin and chloramphenicol. All the isolates were sensitive to chloramphenicol. Efflux pumps mechanism may be possibly to have accounted for the multi drug resistant of *E.coli* and *Pseudomonas* in agreement with (Precott *et al.*, 2004) from this investigation, it is therefore suggested that preventive measures should be recommended to the food seller. These are possible preventive measures:

- i. Food handlers must maintain high personal hygiene including washing of their hands, apron and hand towel
- ii. Food handlers with skin infection or lesions and cuts on their hands should not work in food canteen, restaurant and food preparation areas.
- iii. Payment section should be different from where to collect food.

References

- Bean, N, Griffin, P.M Giulding JS and Ivey CB (1990): Food borne disease outbreak 5-year summary 1983-1987. *J. Food Prof*; 53: 711.
- Bibek, Ray (2004): Fundamental food microbiology CRC Press, Boca Ration London New York Washington DC.
- Buchanan, R.E. and N.E Gibbson (1974): Bergy's Manual of Determinative Bacteriological. 8th ed. The Williams and Wilkins Co. Bathmore.
- Cords B.R and Dychdala G.R (1971): Sanitizer: halogens surface active agents and peroxide in Antimicrobial in foods Davidson, P.M and Branen A.L. Edsu Marcel. Dekker. New York
- Collins et al (1989): Microbiology methods 6th edition Butter worth & co (Publisher) Ltd.
- Clark, D.S. (1965): Method of Estimating the Bacteria population of surfaces. *Canadian J. Microbiol* 11:407-13
- Collins, C.H (1964): Microbial methods. Butter worth and company, London
- Cowan S.I Steel K.J (1985): Manual for the identification of Medical Bacteria Cambridge University Press. Pp 45-60.
- Edema MO, Atagese AO, Idowu AO (2005): Microbial quality of micro wave processed foods in the book of abstract of the 29th annual conferences and general meeting on Microbes as agents of sustainable development, organized by Nigeria society for Microbiology (NSM), UNAAB, Abeokuta from 6 – 10th November 2005.
- Edema M.O Osho A.T and Diala C.I (2008): evaluation of microbial hazards associated with the processing of suya (a grilled meat product. *Scientific Research and essay* Vol 3(12): 621-626.
- Fraizer W.C and D.C Westhoff (1998): Food microbiology 4th edition . Mc Graw –Hill, Singapore.
- Fawole M.O. OSO B A (2001): Laboratory manual of Microbiology: Revised Edition spectrum books Ltd. Ibadan pp. 127
- Ibekwe AC Okono I.O Onunkwo AU, Dunbraye E; Babalola E.T Onoja B,A (2008): Baseline salmoneall agglutinin titres in apparently healthy freshmen in Awka, South Eastern Nigerian *Sci Res Essay* 3(9): 225-230.
- Jay, M.J (1987): Modern Food Technology Microbiology Van Nostrand Company, New-York. Okonko IO, Ogunjobi AA, Adejoye OD Ogunnusiita Olosogba MC (2008) Comparative studies and microbial risk assessment of different water samples used for processing frozen seas foods in Ijora Olopa, Lagos state, Nigeria. *Africa J.Biotechnol.* 7(16):2902-2907.
- Okonko I.O Donbraye E and Babatunde , SOI (2009): Microbiological quantity sea food Processors and water used in two different sea processing plants in Nigeria. *Electronic Journal of environmental, Agricultural and food chemistry*, 8(8): 621:629.
- Oranusi, S.U. Umoh V J; Kwaga JKP (2003): Hazard and critical control points of Kunun-zaki, a non-alcoholic beverage in Northern Nigeria. *Food Microbial* 20:127:132.
- Thatcher F.S and D S Clark (1973): Micro organism in foods. Their significance and methods of enumerations. University of Toronto, Toronto
- Marriot N. G. (1958): Principles of food sanitation, Van Nostrand Reinhold, New York. Pp 71,101.
- Moyo, D.Z and I Baudi (2004): A Bacteriological Assessment of the cleaning and Disinfection efficacy at the Midland State University Canteen, Zimbabwe, *Pakistan Journals of Biological Sciences* 7(11): 1996-2001.
- Uzeh R.E. Ohenhem R.E and Adeniji O.O (2006): Bacterial Contamination of Tisre-suya, A Nigerian meat product. *Pakistan Journal of Nutrition* 5(5): 458-460.