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Microbial profile of smoked sardine (*Sardilella aurita*) AT smoking sites and market centres of Tema, Ghana-1

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ABSTRACT

This research was conducted at three fish smoking sites and three marketing centres in the Tema municipality, to compare the microbial load of smoked sardine (Sardinella aurita). Microbial load of smoked sardine were determined using aerobic plate count, yeast and moulds, coliform bacteria, Escherichia coli and Staphylococcus aureus counts, Salmonella typhi detections, with pH and water activity. The results indicated that the aerobic plate counts within the smoking sites ranged from 6.2×10^4 to 3.3×10^5 cfu/g; yeast and moulds from 1.1×10^2 to 9.3×10^4 cfu/g; and coliform bacteria from 0.0 to 2.1×10^4 cfu/g. For marketing centres, the values ranged from 7.2×10^4 to 4.1×10^7 cfu/g, 5.0×10^2 to 8.0×10^4 cfu/g, and 4.7 to 2.0×10^2 cfu/g for aerobic plate count, yeast and moulds and coliform bacteria respectively . E. coli was poorly represented in samples obtained from Manhean (3.1×10^2 cfu/g) and Newtown (1.8×10^4 cfu/g). Staphylococcus aureus was completely absent from all samples analysed. An isolated case of Salmonella typhi detection was made from Community 1 market samples. pH values were between 5.84 and 6.42 for all samples. Microbial counts of smoked sardine samples from smoking sites and marketing centres were significantly different ($P < 0.05$), with the latter recording higher values. Generally microbial counts for samples from the smoking sites were within acceptable limits of GSB, while those from marketing centres were not.

Keywords: Fish, Tema, *Sardinella aurita,* plate count, bacteria.

INTRODUTION

Fish and fish products are consumed as food all over the world and it provides the world's prime source of high-quality protein: Over one billion people rely on fish as their primary source of animal protein. Fish and other aquatic organisms are also processed into various food and non-food products. Fish is one of the most valuable sources of protein food which can also be used as medicine, ground into vitamins or processed into cosmetics, lubricants, varnishes, soap and margarine [1].

In Ghana and other West African countries, fish constitutes over 70% of the total animal protein intake, with marine fish accounting for nearly 80% of fish production. Large quantities of different species of fish such as sardines and the anchovies are landed during the season of glut between July and October each year, which are preserved by one of several traditional processing techniques to avoid excessive wastage [2, 3] also reported that most widely used techniques to preserve fish in Ghana are smoking, salting and sun drying, smoking of fish ranges from three hours to two days depending on the desired end product and the shelf life needed for storage and marketing of the product. Smoking also gives fish a good flavour and aroma [4].

The quality of smoked fish is essentially linked to processing and post processing procedures. The smoking, storage, handling and packaging techniques such as the use of old news prints, cement papers and polyethylene bags are all sources of contamination of fish. Fish contamination is primarily caused by microbial activities. The occurance of pathogens such as *Staphylococcus aureus, Salmonella typhimurium, Bacillus cereus* and *Clostridium botulinium* has raised major concerns among researchers since they cause food borne illnesses [5].

Recent investigation revealed unsatisfactory microbial quality of smoked *Sardinella aurita* in some smoke houses and markets centres in the Cape Coast metropolis [6]. *Sardinella* species *are* coastal, pelagic fish which prefers clear saline water with a minimum temperature below 24°C and a depth of about 350 m. These species breeds at all times of the year but with distinct peaks e.g. July - October in Ghana. Examples of *Sardinella spp.*are *S. atricauda, S. brachysoma, S. maderensis, S. hualiensis* and *Sardinella aurita* [6]

Sardinella aurita, commonly called sardines belong to the Kingdom: Animalia, Phylum: Chordata, Class: Actinopterygii, Order: Clupeiformes, Family: Clupeidae, are elongated, usually sub-cylindrical with a rounded belly, with fine and numerous lower gill rakers, usually more than 80. The standard length is 23 to 28 cm. The flanks are silvery, with a faint golden mid-lateral line, preceded by a faint golden spot behind gill opening; a distinct black spot at hind border of gill cover [7].

Fish begin to deteriorate as soon as it leaves the water. To delay spoilage, clean the fish as soon as possible and preserve. The four most popular methods of fish preservation are freezing, canning, smoking, and pickling [8].

The storage lives of good quality frozen fish are held at 0°C or lower [9]. Only a few species of fish are preserved commercially by pickling as is the process of preserving food by anaerobic fermentation in a solution of salt water to produce lactic acid, or marinating and storing it in an acid solution, usually vinegar [10].

Foods have been preserved by smoking before the dawn of recorded history. People in all cultures world the over have relied on the smoking of fish and meat products for long-term storage. The steps in the smoking process are necessary not only for safe preservation, but also to produce good flavour and aroma [11].

In all these processes, drying is of paramount importance for preservation, because it is moisture in the flesh that permits bacterial activity and spoilage. Further, the application of extracts from the smoke e.g phenols retards the development of spoilage bacteria [9]

The smoking achieves preservation by four different means: the hot smoking melts down the fat in the fish which drips away, dries the fish, it deposits compounds on the fish that inhibit the

growth of micro-organisms which cause decomposition, and hot smoking kills micro- organisms on the surface of the fish.

Food safety is an increasingly important public health issue. Governments all over the world are intensifying their efforts to improve food safety. These efforts are in response to an increasing number of food safety problems and rising consumer concerns. . In spite of the common term food poisoning, most cases are caused by a variety of pathogenic bacteria, viruses, or parasites that contaminate food, rather than chemical or natural toxins [5].

Foodborne illness usually arises from improper handling, preparation, or food storage. Good hygiene practices before, during, and after food preparation can reduce the chances of contracting an illness [12, 13]

Microorganisms are the major cause of spoilage of most seafood products. However, only a few members of the microbial community, the specific spoilage organisms (SSOs), give rise to the spoilage of fish. Some microorganisms that contaminate fish are *Escherichia coli*, *Staphylococcus aureus, Salmonella typhimurium, Bacillus cereus, shigella spp., Clostridium botulinium* etc. (Dalgaard, 2005). Examples of foods involved in outbreaks of salmonellosis are eggs, fish, poultry and other meats and raw milk [11, 14, 15]

According WHO [16] there are certain environmental conditions that must be met for microorganisms to grow and multiply and when these conditions exist they can very quickly increase in number. These conditions are, temperature, pH, time , moisture, oxygen, nutrient, water activity, etc.

MATERIALS AND METHODS

Tema, with a population of 209,000 (2005), is a city on the Atlantic Ocean coast, east of the capital city of Accra, in Greater Accra Region, in Ghana. It was originally a small fishing village which grew after the construction of a large harbour in 1961 and is now the nation's largest sea port. Tema is the nearest city to the geographical position of 0° latitude and 0° longitude.

Three smoking centres (Abonkor, Ashaman and Manhean) and three market centres (community one, community two and Newtown) all located in the Tema metropolis were selected as sampling sites.

The smoked sardine samples were purchased from three smoking sites and three marketing centres.

Readymade media, equipment and all chemicals used for the investigations were obtained from the microbiology laboratory of Food Research Institute of the Council for Scientific and Industrial Research (CSIR, Accra) and prepared according to product instructions

Sample collection

The smoked fish samples for the analysis were obtained from the three smoking sites and the three market centres. The various smoke houses and markets were visited on four (4) different occasions on weekly basis for a period of one month. All the samples purchased were kept in a thermos flask containing ice pack to arrest the growth of microbes and analyzed within four (4) hours of collection in the microbiology laboratory of Food Research Institute of the Council for Scientific and Industrial Research (CSIR, Accra)

Study Area

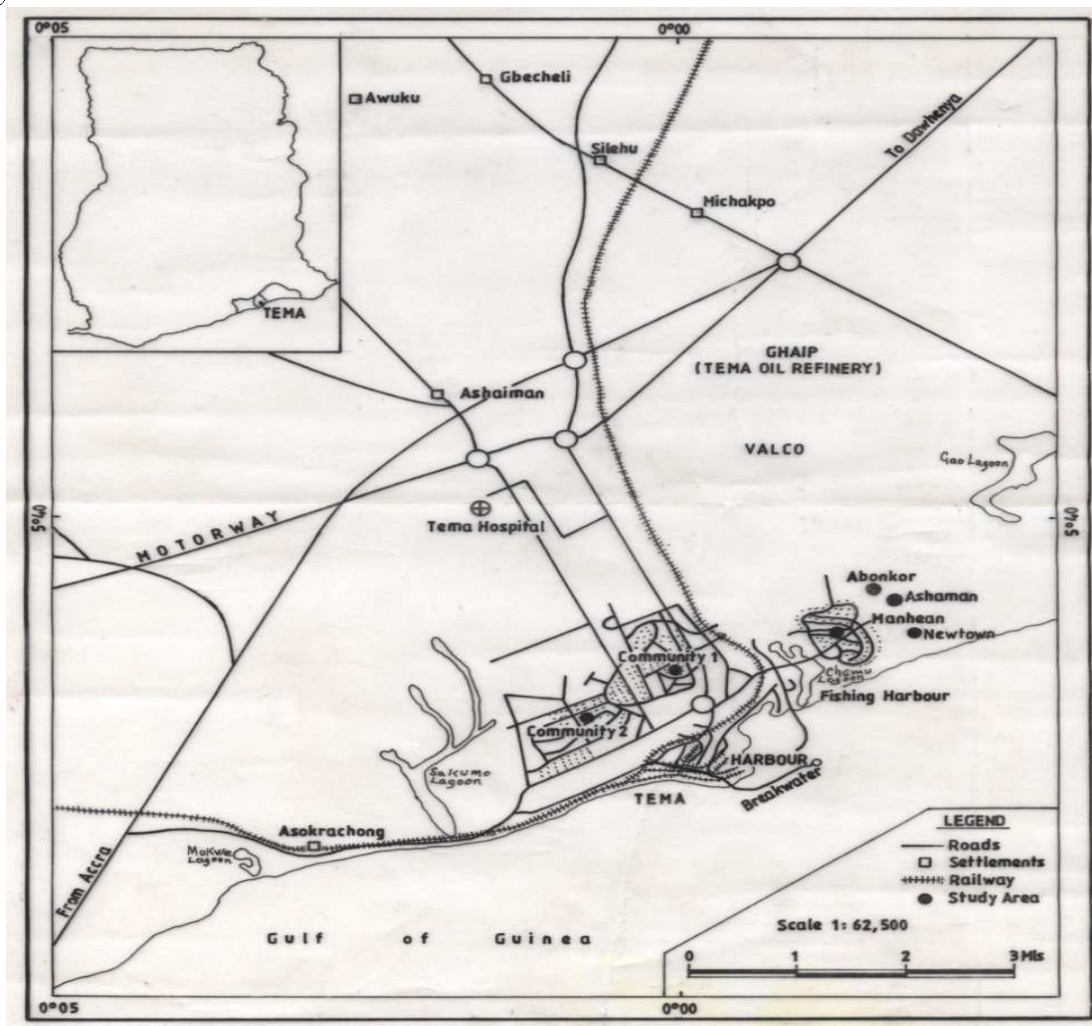

Figure 1: Map of Tema showing study area

Microbiological analysis

The samples were analysed for aerobic plate counts, coliform counts, yeast and mould count. The aerobic plate count was enumerated using the pour plate technique in a non-selective medium Plate Count Agar (PCA). The Nordic Committee of Food Analysis method was employed.

Sample Preparation: Ten (10) grams of the smoked fish was weighed into a stomacher bag and 90ml of sterile Salt Peptone Solution (SPS) as diluent was added. The content was homogenized in the stomacher for 90 seconds to obtain 1:10 dilution and serially diluted.

Reading of Plate: Plates were read after incubation, plates containing colonies between 25 and 250 were selected and counted under illuminated colony counter. The colonies were examined by microscopy, gram and catalase reactions.

Determination of Yeast and Moulds

The yeast and moulds were enumerated using the pour plate method on Oxytetracyline Glucose Yeast Extract Agar (OGYEA) medium. The International Standards Organization method (ISO.

7954, 1987E) was used for the determination. Plates containing colonies between 10 and 150 were selected and counted.

Determination of Coliform Bacteria
Coliform bacteria were determined by plating known quantities of sample on a non-selective agar (Tryptone Soy Agar – TSA) and a selective agar (Violet Red Bile Agar – VRBA) media. The method employed was the Nordic Committee of Food Analysis method. Plates with typical colonies between10-100 were selected and counted.

Confirmation: Five of the colonies from the TSA/VRBA plates were selected and sub-cultured in Brilliant Green Bile Broth medium containing Durham tube for gas formation.

Determination Thermo-Tolerant Coliform Bacteria
Thermo-tolerant coliform bacteria were determined by pour plate technique in a non-selective agar TSA and a selective agar VRBA media. NMKL.No.125 (2005) was used for the determination.

Confirmation: Five of the colonies from the TSA/VRBA plates were selected and sub-cultured in Escherichia coli Broth (EC Broth) medium containing Durham tube for gas formation. Positive tubes were tested for indole production.

Enumeration of *Staphylococcus aureus*
Determination of *Staphylococcus aureus* was done using the spread plate technique on a Baird Parker medium (BP). NMKL.No.66 was employed.

Detection of *Salmonella typhi*
The method employed was the Nordic Committee of Food Analysis method.

Biochemical tests were carried out to confirm each of the organisms Physicochemical analysis was carried out on each of the samples for the water activity and pH

Data analysis
The data from this study was analysed using MINITAB (version 16) statistical software. Statistical analysis was done using one sample t-test. Descriptive statistics, mainly means were calculated using Microsoft Excel software.

RESULTS AND DISCUSSION

3.1. Results
The microbiological analysis carried out on the smoked sardine samples were, aerobic plate count, yeast and moulds, coliform bacteria, *E. coli*, *Staphylococcus aureus* enumeration and *Salmonella* detection. The physicochemical analyses performed were water activity and pH.

Table 1 shows the results of microorganisms counts, water activity and pH values of smoked sardine samples obtained for first sampling period. *Staphylococcus aureus* and *Escherichia coli* were absent in all the samples analysed. Abonkor smoking site recorded the lowest aerobic plate count of 3.3 x 10^4cfu/g whiles the highest count of 6.9 x 10^5cfu/g was recorded for Community 2 market sample.

Table 1 Microbial count and detection (expressed in cfu/g), water activity and pH of smoked sardine obtained from smoking centres and markets in first sampling period

SAMPLING SITE	SITE CODE	AEROBIC PLATE COUNT	YEAST AND MOULDS COUNT	COLIFORM COUNT	*E. coli* COUNT	*S. aureus* COUNT	*S. typhi*	WATER ACTIVITY	pH
SMOKING	ABO	3.3×10^4	0	0	0	0	-	0.62	6.19
	ASH	1.1×10^5	2.5×10^2	0	0	0	-	0.77	6.12
	MAN	8.2×10^4	2.0×10^3	0	0	0	-	0.76	6.15
	MEAN	$\mathbf{7.5 \times 10^4}$	$\mathbf{7.5 \times 10^2}$	**0**	**0**	**0**		**0.72**	**6.15**
MARKET	COM 1	5.5×10^4	4.4×10^3	2.0×10^2	0	0	+	0.66	6.29
	COM 2	6.9×10^5	2.3×10^4	0	0	0	-	0.89	6.4
	NTM	1.9×10^5	8.6×10^4	8.0×10^1	0	0	-	0.89	6.56
	MEAN	$\mathbf{3.1 \times 10^5}$	$\mathbf{3.8 \times 10^4}$	$\mathbf{9.3 \times 10^1}$	**0**	**0**		**0.81**	**6.42**

NB: - represents absence of organism ; + represents presence of organism; ABO: Abonkor Smoking site
COM 1: Community 1 Market ; ASH: Ashaman Smoking site ; COM 2: Community 2 Market ;
MAN: Manhean Smoking site ; NTM: Newtown Market

Yeast and moulds were isolated from all samples except Abonkor and the counts ranged between 2.5×10^2 and 8.6×10^4cfu/g. Coliform bacteria counts were recorded for only Newtown market (8.0×10^1) and Community one (2.0×10^2). *Salmonella typhi* was only detected in the sample obtained from Community 1 market. The pH range for samples was 6.12 -6.56 and the water activity varied from 0.62 to 0.89

Table 2: Microbial count and detection (expressed in cfu/g), water activity and pH of smoked sardine obtained from smoking centres and markets in second sampling period.

SAMPLING SITE	SITE CODE	AEROBIC PLATE COUNT	YEAST AND MOULDS COUNT	COLIFORM COUNT	*E. coli* COUNT	*S. aureus* COUNT	*S. typhi*	WATER ACTIVITY	pH
SMOKING	ABO	7.3×10^4	0	2.6×10^4	0	0	-	0.69	6.46
	ASH	1.3×10^5	1.9×10^3	1.0×10^2	0	0	-	0.82	6.13
	MAN	5.3×10^2	0	3.6×10^4	0	0	-	0.51	6.72
	MEAN	$\mathbf{6.8 \times 10^4}$	$\mathbf{6.3 \times 10^2}$	$\mathbf{2.1 \times 10^4}$	**0**	**0**	-	**0.67**	**6.44**
MARKET	COM 1	2.0×10^5	1.3×10^4	4.0×10^2	0	0	-	0.83	6.45
	COM 2	5.2×10^4	8.0×10^3	0	0	0	-	0.65	6.13
	NTM	2.5×10^5	2.2×10^5	0	1.8×10^4	0	-	0.91	6.41
	MEAN	$\mathbf{1.7 \times 10^5}$	$\mathbf{8.0 \times 10^4}$	$\mathbf{1.3 \times 10^2}$	$\mathbf{6.0 \times 10^3}$	**0**	-	**0.8**	6.33

NB - represents absence of organism + represents presence of organism

Table 2 represents the microbial load, water activity and pH of samples analysed in the second sampling period. Aerobic plate counts were obtained for all the samples analysed. Manhean smoking site recorded lowest of 5.3×10^2cfu/g whilst Newtown market recorded highest of 2.5×10^5cfu/g. The count for yeast and moulds ranged between 1.9×10^3 and 2.2×10^5cfu/g. Abonkor and Manhean recorded no count for yeast and mould. *E. coli* was only isolated from the sample obtained from Newtown market. The lowest count for coliform bacteria was 1.0×10^2cfu/g recorded for Ashaman smoking site whilst the highest count was 3.6×10^4cfu/g for Manhean smoking site. There was no coliform bacteria count for Community two and Newtown samples. *Staphylococcus aureus* and *Salmonella typhi* were absent in all the samples analysed. The pH range of the samples was 6.13 – 6.72. 0.51– 0.91 was the water activity range for the samples.

Table 3: Microbial count and detection (expressed in cfu/g), water activity and pH of smoked sardine obtained from smoking centres and markets in third sampling period

SAMPLING SITE	SITE CODE	AEROBIC PLATE COUNT	YEAST AND MOULDS COUNT	COLIFORM COUNT	*E. coli* COUNT	*S. aureus* COUNT	*S. typhi*	WATER ACTIVITY	pH
SMOKING	ABO	5.4×10^4	0	0	0	0	-	0.7	6.4
	ASH	1.2×10^5	2.7×10^2	0	0	0	-	0.71	6.43
	MAN	1.3×10^4	5.0×10^1	0	3.1×10^2	0	-	0.69	6.17
	MEAN	$\mathbf{6.2 \times 10^4}$	$\mathbf{1.1 \times 10^2}$	**0**	$\mathbf{1.0 \times 10^2}$	**0**		**0.7**	**6.33**
MARKET	COM 1	7.4×10^5	2.5×10^3	3.0×10^2	0	0	-	0.76	5.72
	COM 2	8.3×10^5	3.5×10^4	0	0	0	-	0.81	6.23
	NTM	1.2×10^8	6.2×10^4	3.1×10^2	0	0	-	0.93	6.32
	MEAN	$\mathbf{4.1 \times 10^7}$	$\mathbf{3.3 \times 10^4}$	$\mathbf{2.0 \times 10^2}$	**0**	**0**		**0.83**	**6.09**

NB - represents absence of organism + represents presence of organism

Table 4: Microbial count and detection (expressed in cfu/g), water activity and pH of smoked sardine obtained from smoking centres and markets in fourth sampling period

SAMPLING SITE	SITE CODE	AEROBIC PLATE COUNT	YEAST AND MOULDS COUNT	COLIFORM COUNT	*E. coli* COUNT	*S. aureus* COUNT	*S. typhi*	WATER ACTIVITY	pH
SMOKING	ABO	2.3×10^2	0	0	0	0	-	0.51	5.72
	ASH	1.0×10^6	2.8×10^5	0	0	0	-	0.9	5.83
	MAN	3.3×10^3	1.0×10^2	0	0	0	-	0.6	5.97
	MEAN	$\mathbf{3.3 \times 10^5}$	$\mathbf{9.3 \times 10^4}$	**0**	**0**	**0**		**0.67**	**5.84**
MARKET	COM 1	3.8×10^3	1.4×10^3	0	0	0	-	0.63	6.11
	COM 2	2.1×10^5	7.0×10^1	0	0	0	-	0.77	5.98
	NTM	3.6×10^3	3.0×10^1	1.4×10^1	0	0	-	0.62	6.36
	MEAN	$\mathbf{7.2 \times 10^4}$	$\mathbf{5.0 \times 10^2}$	**4.7**	**0**	**0**		**0.67**	**6.15**

NB - represents absence of organism + represents presence of organism

Results of Table 3 shows microbial load, water activity and pH of samples analysed in the third week.All samples analysed had no count for *Staphylococcus aureus* and no detection of *Salmonella typhi.* Aerobic plate count ranged from 1.3 x 10^4 to 1.2 x 10^8cfu/g. Apart from Abonkor sample, all samples recorded counts for yeast and moulds and the range was 5.0 x 10^1 – 6.2 x 10^4cfu/g. Coliform bacteria count were recorded in only two samples and the count were 3.0 x 10^2 and 3.1 x 10^2cfu/g for Community one and Newtown markets respectively. Apart from Manhean sample, which recorded *E. coli* count of 3.1 x 10^2cfu/g there was no *E. coli* count for the other samples. The pH range of the samples was 5.72 – 6.43 whilst the water activity range was 0. 69 – 0.93.

Microbial load, water activity and pH of smoked sardines analysed in fourth sampling period (Table 4) revealed that there was no detection of *Salmonella typhi* in all samples. All samples recorded no count for *Staphylococcus aureus* and *E. coli*. Coliform bacteria however were isolated only from the sample obtained from Newtown market, which had 1.4 x 10^1cfu/g. There were aerobic plate counts for all samples. These ranged between 2.3 x 10^2 and 1.0 x 10^6cfu/g. Apart from Abonkor sample, yeast and moulds were isolated from all the samples and its count varied from 3.0 x 10^1 to 2.8 x 10^5.pH range for the sample was 5.72 – 6.36. The water activity range for the samples was between 0.51 and 0. 90.

DISCUSSION

The results of this investigation revealed that the microbial counts of the smoked sardine samples from the smoking sites and the marketing centres varied. Measurement of differences using t-test showed significant difference ($P < 0.05$) between the smoking sites and the marketing centres with respect to aerobic plate count, yeast and moulds, coliform bacteria and *E coli.* Comparatively, smoked sardine from the marketing centres had higher counts. Upon observation, the smoking sites had better sanitary conditions than the marketing centres. This result conforms to report by Annan [17]. Annan [17] indicated higher microbial load of smoked sardine samples obtained from the markets as compared to the smoking site in the Cape Coast Metropolis. Poor handling, packaging, transporting and storage conditions may be the probable factors for higher count obtained for the marketing centres. More so the display of smoked sardines in trays in the open markets may increase the risk of contamination. According Plahar *et al*. [18], the quality of smoked fish is essentially linked to processing and post processing procedure.

There was a significant difference ($P <0.05$) between the microbial counts obtained for smoked sardine samples within the smoking centres. This could be due to different storage procedure and conditions employed at the smoking sites, which support an earlier assertion by Plahar *et al.* [18], indicating that in Ghana smoked fish is packed and stored in baskets, basins or wooden boxes.

Within the marketing centres, there was no significant difference ($P>0.05$) in the microbial count obtained for the samples though there were marked numerical differences. The reason may be that the same handling, packaging, storage and marketing techniques for smoked sardines pertain to all the markets.

The mean aerobic plate count for the samples from the smoking sites over the four week period ranged from 6.2 x 10^4 to 3.3 x 10^5cfu/g. These values fall within the acceptable limit of Ghana Standards Board (GSB), which is1.0 x 10^6cfu/g. The mean water activity values ranged between 0.67 and 0.72 whilst the pH ranged from 5.84 to 6.44.

For the marketing centres, the mean aerobic plate count for weeks 1,2 and 4 were 3.1×10^5, 1.7×10^5 and 7.2×10^4 respectively. These are within the acceptable limit of GSB, although they are numerically greater than the counts obtained for the smoking sites. However, the aerobic plate count obtained for the third week for the marketing centre (4.1×10^7) exceeds the acceptable limit of GSB, thus making the smoked sardine unwholesome. The mean water activity values ranged from 0.67 to 0.83. The third week recorded the highest water activity of 0.83 indicating sufficient amount of water available for metabolic activities and microbial growth. Newtown market recorded a high aerobic plate count of 1.2×10^8cfu/g, a water activity of 0.93 and a pH of 6.32. This result agreed with to the findings obtained by Eyeson *et al*. [19] which indicated that an increase in aerobic plate counts for marketing centre samples was due to contamination which resulted from handling procedures and unhygienic environmental conditions.

Yeast and Moulds count for smoking sites varied between 1.1×10^2 and 9.3×10^4cfu/g. With the exception of the fourth week which recorded a mean count of 9.3×10^4cfu/g, all other counts were within the acceptable limits of GSB. The fourth week sample which had a yeast and mould count above the acceptable limit also recorded a pH of 5.84 and water activity of 0.67. According to Prescott [20], yeast and moulds require low pH and arid conditions to thrive. Thus a pH value of 5.84 and a water activity of 0.67 are adequate to support the growth of yeast and moulds, hence the high counts. Generally the yeast and mould count for the marketing centres showed levels which were unsatisfactory. Apart from the fourth sampling period sample which recorded a count of 5.0×10^2cfu/g, all other counts were above the acceptable limit of 1.0×10^4cfu/g [21].

There was no coliform bacteria count for the smoking sites except for second sampling period which recorded a value of 2.1×10^4cfu/g, above the limit specified by GSB. The mean coliform count for the marketing centres ranged from 4.7 to 2.0×10^2cfu/g. With the exception of the fourth week which recorded a low value of 4.7cfu/g, all samples obtained for the other weeks had counts above the GSB acceptable limit of 4×10^1cfu/g. This result is in agreement with report by Binta *et al*. [22]. Which indicated that unhygienic conditions of the local markets resulted in the increase of coliform bacteria counts in Kenya.

The GSB acceptable limit for *E. Coli* count in fish is 0 cfu/g. *E. coli* counts were absent in all samples except samples obtained from Manhean smoking site in the third week (3.1×10^2cfu/g) and Newtown market in the second week (1.8×10cfu/g ^{4}). These values are unsatisfactory and it indicates the presence of faecal matter in the smoked sardine samples making it unwholesome. The presence of *E. coli* also indicates inadequate control during the smoking process.

The sample obtained from Newtown market in the second week of the investigation recorded a high water activity of 0.91 and pH close to neutrality (6.44). A visual observation of this market shows poor sanitation conditions.

The investigation revealed that *Staphylococcus aureus* was completely absent in all samples analysed from both smoking and marketing centres. According to Prescott [20], *S. aureus* grows best in salty environments, hence absence of salt and other unfavourable intrinsic and extrinsic factors may have accounted for the absence of *S. aureus* in the smoked sardines and that *S. aureus* normally does not compete well with other mircoorganisms. This contradicts result obtained by Annan [17] since he isolated *S. aureus* from smoked sardine.

With the exception of the sample obtained from Community 1 market in the first sampling period, *Salmonella typhi* was not detected in any other sample. It is possible that most intrinsic and extrinsic factors of the smoked sardines could not support the growth of *S. typhi.*

CONCLUSION

The investigation conducted indicated that there was significant difference ($P < 0.05$) between the microbial counts of samples from the smoking sites and marketing centres with the latter recording higher microbial populations. In general, the levels of microbial loads in the samples from the marketing centres were unsatisfactory, making the smoked sardine unwholesome. Newtown market had the most unsatisfactory result.

Although most of the microbial counts for the smoking sites were within acceptable limits, the counts were marginal.

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