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Bacteriological Evaluation of Some Smoked Meat, chicken and Fish Products

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A B S T R A C T

A grand total of ninety random samples of smoked products were collected from different local supermarkets and classified into 30 samples of both of smoked fish (smoked tuna and herring) (15 of each), samples of smoked poultry (smoked chicken and turkey) (15 of each) and samples of smoked meat (smoked rose beef and salami) (15 of each), these products were subjected to determine aerobic plate count(APC), coliform count and isolate *Staph aureus*. Results revealed that smoked tuna contained higher mean of APC (cfu/g) ($1.62 \times 10^8 \pm 7.24 \times 10^7$), while the lower results reported in smoked chicken ($2.75 \times 10^5 \pm 6.59 \times 10^4$). For total coliform count (cfu/g) smoked turkey had the higher mean value ($5.25 \times 10^7 \pm 16.09 \times 10^6$), while smoked herring had the lower mean value ($1.81 \times 10^4 \pm 83 \times 10^2$). Total *Staphylococci* count (cfu/g) was higher in smoked turkey ($11.06 \times 10^7 \pm 4.25 \times 10^7$) and lower in smoked chicken ($23 \times 10^3 \pm 6.5 \times 10^3$).

Keywords: Smoked tuna, Smoked chicken, Smoked turkey, *Staph aureus,* coliform.

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1. INTRODUCTION

Smoking is one of the oldest methods of preserving fish or any other meat for that matter long before there were no refrigerators and freezers (Moshood et al., 2012). The main purpose of smoking food products is that it not only act as colouring and flavouring agent but also has antibacterial and antioxidative proprieties (Davidson, 2001). Drying effects during smoking, together with the antioxidant and bacteriostatic effects of the smoke, allow smoked products to extend shelf-life (Eyo, 2001). Therefore, microbiological examination of food and environmental samples is generally recommended to validate and verify the efficiency of foods safety and quality control (International Commission on Microbiological Specification for Foods (ICMSF), 2011). Although smoking and its antimicrobial properties, common food-borne pathogens could contaminate smoked products and cause illness in human consumer such as *Staphylococcus aureus* (Abd El-Shahid, 2015). *Staphylococcus aureus* produces a wide variety of toxins including staphylococcal enterotoxins with demonstrated emetic activity, *staphylococcus aureus* toxins are a major cause of food poisoning, which typically occurs after ingestion of different foods. Symptoms are of rapid onset and include nausea and violent vomiting, with or without diarrhea. The illness is usually self-limiting and only occasionally severe enough to warrant hospitalization (Argudin et al., 2010).

The aim of this work was to perform trend analysis to reveal probable gaps and shortcomings in monitoring of microbiological contamination of smoked preparations which included smoked meat products (smoked salami and smoked rose beef), smoked poultry products (smoked chicken and smoked turkey) and smoked fish products (smoked herring and smoked tuna) focusing on the following determinations: determination of aerobic plate count, determination of total *coliform* count, determination of total *staphylococci* count, and isolation and identification of *Staphylococcus aureus*.

2. MATERIAL AND METHODS

2.1. Collection of Samples:

A grand total of ninety random samples of smoked products were collected from different local supermarkets and classified into 30 samples of both of smoked fish (smoked tuna and herring) (15 of each), samples of smoked poultry (smoked chicken and turkey) (15 of each) and samples of smoked meat (smoked rose beef and salami) (15 of each). The collected samples were kept in separate plastic bags and transferred directly to the laboratory of Food Hygiene, Animal Health Research Institute, Tanta Branch, in an insulated ice box under complete aseptic conditions without undue delay to be subjected for bacteriological examination

2.2. Preparation of samples according to American Public Health Association (APHA) (1992).

2.3. Determination of total aerobic plate count "APC" according to (APHA, 1992):

plate count agar media at 37ºC for 48 hrs. Total aerobic plate count (APC) /g was calculated on plates containing 30-300 colonies and each count was recorded separately.

2.4. Determination of total staphylococci counts " according to International Commission on Microbiological Specification for Foods (ICMSF) (1996).

2.5. Isolation of suspected S. aureus isolates (ICMSF 1996)

A loopful of the original homogenate was spread on the surface of Baired parker agar plates, and incubated at at 37ºC for 48 hrs. suspected colonies of *Staph. aureus* which appear as black and shiny with narrow white margins and surrounded by a clear zone extending into the opaque medium, were counted as *Staph. aureus* and subjected to further identification.

2.5.1. identification of isolated st. aureus:

Morphological examination by: Staining according to Cruickshank et al. (1975). Motility test according to ICMSF (1996); Deoxyribonuclease test (DN-ase) and Thermostable nuclease test (TN-ase) according to Lachica et al. (1992). Biochemical identification: Catalase activity test according to MacFaddin (1976). Detection of hemolysis according to Bailey and Scott (1978). Mannitol test according to Bailey and Scott (1978). Coagulase test according to American Public Health Association (APHA) (1992). Deoxyribonuclease test (DN-ase) and Thermostable nuclease test (TN-ase) according to Lachica et al. (1992).

2.6. Statistical analysis:

The data was statistically treated by using SPSS program for windows (Version 16) (SPSS Inc. Chicago, IL and USA) (Steel and Torrie, 1980).

3. RESULTS:

Concerning smoked herring, table (1) revealed that the aerobic plate count (cfu/g) was 2.96 $x10^7$ ± 1.65 $x10^7$while the minimum was 1$x10^4$ cfu/g and the maximum was1.72 $x10^8$. Concerning smoked tuna, it was noticed in Table (1) that the aerobic plate count (cfu/g) of smoked tuna were 1.62 $x10^8$ ± 7.24 $x10^7$ with maximum value 1 $x10^9$ and minimum value 5 $x10^6$. Table (1) showed that the aerobic plate count (cfu/gm) of smoked salami was 8.36 $x10^6$ ±2.72 $x10^6$with maximum value 3 $x10^7$and minimum value 1 $x10^6$. Concerning smoked rose beef, it was noticed in Table (1) that the aerobic plate count(cfu/g) were 1.36 $x10^7$ ± 4.66 $x10^6$ with maximum value 5 $x10^7$and minimum value 1 $x10^6$. Regarding smoked chicken, the results in table (1) showed that the aerobic plate count (cfu /gm) of smoked chicken were 2.75 $x10^5$ ± 6.59 $x10^4$ cfu/g and the minimum count was 5 $x10^4$ cfu/g while the maximum count was 7 $x10^5$ cfu/g. Concerning smoked turkey, in Table (1) it was noticed that the APC were 3.13 $x10^7$ ± 5.15 $x10^6$ with maximum value 4.5 $x10^7$ and minimum value 2 $x10^7$. The results in table (2) showed total coliform count (cfu/g) in smoked herring was 1.81 $x10^4$± 83 $x10^2$ with a minimum of 4 $x10^3$and maximum of 1 $x10^5$. the total coliform count of smoked tuna was (cfu/g) 8.82 $x10^6$±1.36 $x10^6$, where the minimum was 2 $x10^7$ and maximum was 16 $x10^5$ in table 2. Total coliform count(cfu/g) in smoked salami such product was 4.72 $x10^6$±11.9 $x10^5$where the minimum was 11 $x10^4$and maximum was 15 $x10^6$. The total coliform count in rose beef (cfu/g) 4 $x10^4$± 1 $x10^4$ where the minimum was 3 $x10^4$ and maximum was 5 $x10^4$. While total coliform count in the smoked chicken products were 7.16 $x10^5$±5.45 $x10^5$cfu/g where the minimum was 2 $x10^3$cfu/g and the maximum was 6 $x10^6$cfu/g. While total coliform count in smoked turkey products were 5.25 $x10^7$± 16.09 $x10^6$ cfu/g where the minimum was 3 $x10^7$ cfu/g and the maximum was 1 $x10^8$ cfu/g. While the results in table (3) showed total Staphylococci count in smoked herring (cfu/g) was 18 $x10^5$±15 $x10^5$with minimum count 4 $x10^3$and maximum count 1.97 $x10^7$. While the total Staphylococci count (cfu/g) as showed in table (3) was 39.5 $x10^5$±88 $x10^4$ with minimum count 7 $x10^5$ and maximum count 73 $x10^5$. While the total Staphylococci count(cfu/g) in smoked salami was 31.7 $x10^5$±14 $x10^5$with minimum count 1 $x10^5$and maximum count 15 $x10^6$. While the total Staphylococci count (cfu/g) in rose beef was 1.62 $x10^8$ ± 7.24 $x10^7$ with minimum count 5 $x10^6$ and maximum count 1 $x10^9$

Total Staphylococci count was 23 $x10^3$±6.5 $x10^3$ cfu/g in the examined samples of smoked chicken products. Total Staphylococci count was 11.06 $x10^7$±4.25 $x10^7$cfu/gin the examined samples of smoked turkey as illustrated in table (3) and with 3 x10^8 cfu/g as a maximum and 1.5 x10^6 cfu/g as minimum. The results in table (4) and table (5) showed that *S.aureus* isolated from 66.6% of the examined smoked herring samples. where 53.3% were coagulase positive and 13.3% were coagulase negative. *staph aureus was* isolated from 26.6% of the examined smoked tuna samples of smoked tuna. Where 20% were coagulase positive and 6 % were coagulase negative, *staph aureus* was isolated from 20. % of the examined smoked salami samples. Where 6% were coagulase positive and 13.3%were coagulase negative. *staph aureus* failed to be isolated of any samples in smoked rose beef. *staph aureus* was isolated from 60% 0f samples of smoked chicken, 40% were coagulase positive while 20% were coagulase negative showed that *S.aureus* were isolated from 13.3% 0f samples of smoked turkey and all were coagulase positive.

Table (1): Statistical analytical results and acceptability of APC (cfu/g) of the examined samples of smoked products (n=15)

Smoked Products	Min.	Max.	Mean ± S.E	Accepted samples*		Unaccepted samples*	
				No.	%	No.	%
Smoked Herring	1x10^4	1.72 x10^8	2.96 x10^7 ± 1.65 x10^7	8	53.3	7	46.6
Smoked Salami	1 x10^6	3 x10^7	8.36 x10^6 ±2.72 x10^6	5	33.3	10	66.7
Smoked Rose Beef	1 x10^6	5 x10^7	1.36 x10^7 ± 4.66 x10^6	2	13.3	13	86.7
Smoked Tuna	5 x10^6	1 x10^9	1.62 x10^8 ± 7.24 x10^7	1	6	14	94
Smoked Chicken	5 x10^4	7 x10^5	2.75 x10^5 ± 6.59 x10^4	4	26.6	11	73.4
Smoked Turkey	2 x10^7	4.5 x10^7	3.13 x10^7 ± 5.15 x10^6	11	73.3	4	26.7

*= Permissible Limit should not exceed 10^5 in smoked fish, 10^4 in smoked poultry products and 10^4 in smoked meat according to EOS (2005). Means within a column followed by different letters showed high significant difference *(P < 0.05).*

Table (2): Statistical analytical results and acceptability of total coliform count (cfu/g) of the examined samples of smoked products (n=15)

Smoked Products	Min.	Max.	Mean ± S.E	Accepted samples*		Unaccepted samples*	
				No.	%	No.	%
Smoked Herring	4 x10^3	1 x10^5	1.81 x10^4± 83 x10^{2b}	6	40	9	60
Smoked Salami	11 x10^4	15 x10^6	4.72 x10^6±11.9 x10^{5b}	4	26.7	11	73.3
Smoked Rose Beef	3 x10^4	5 x10^4	4 x10^4± 1 x10^{4b}	11	73.3	4	26.7
Smoked Tuna	16 x10^5	2 x10^7	8.82 x10^6±1.36 x10^{6b}	2	13.3	13	86.7
Smoked Chicken	2 x10^3	6 x10^6	7.16 x10^5±5.45 x10^{5b}	6	40	9	60
Smoked Turkey	3 x10^7	1 x10^8	5.25 x10^7±16.09x10^{6a}	11	73.3	4	26.7

*= Permissible Limit should not exceed 10 in smoked fish, 10^2in smoked poultry and 10^2 in smoked meat cfu/g according to EOS (2005). Means within a column followed by different letters showed high significant difference *(P < 0.05).*

Table (3): Statistical analytical results and acceptability of total *Staphylococci* count (cfu/g) of the examined samples of smoked products (n=15)

Smoked Products	Min.	Max.	Mean ± S.E	Accepted samples*		Unaccepted samples*	
				No.	%	No.	%
Smoked Herring	4 x10^3	1.97 x10^7	18 x10^5±15 x10^{5b}	8	53.3	7	46.7
Smoked Salami	1 x10^5	15 x10^6	31.7 x10^5±14 x10^{5b}	5	33.3	10	66.7
Smoked Rose Beef	1 x10^6	6 x10^6	3 x10^6±10.8 x10^{5b}	9	60	6	40
Smoked Tuna	7 x10^5	73 x10^5	39.5 x10^5±88 x10^{4b}	5	33.3	10	66.7
Smoked Chicken	1 x10^3	6.5 x10^4	23 x10^3±6.5 x10^{3b}	3	20	12	80
Smoked Turkey	1.5 x10^6	3 x10^8	11.06 x10^7±4.25 x10^{7a}	7	46.6	8	53.4

*= Permissible Limit should not exceed _ 0 in all smoked products cfu/g according to EOS (2005)
Means within a column followed by different letters showed significant difference *(P < 0.05).*

Table (4) incidence of *S.aureus* isolated from the examined samples of smoked products. (n=15)

Smoked Products	Positive Samples	
	No.	%
Smoked Herring	10	66.6
Smoked Salami	3	20
Smoked Rose Beef	0	0
Smoked Tuna	4	26.6
Smoked Chicken	9	60
Smoked Turkey	2	13.3

Table (5) incidence of coagulase positive from isolated *S.aureus* strains isolated from the examined samples of smoked products (n=15)

Smoked Products	Coagulase Positive Samples		Coagulase negative Samples	
	No.	%	No.	%
Smoked Herring	8	53.3	2	13.5
Smoked Salami	1	6	2	13.3
Smoked Rose Beef	0	0	0	0
Smoked Tuna	3	20	1	6
Smoked Chicken	6	40	3	20
Smoked Turkey	2	13.3	0	0

4. DISCUSSION

Each food type should be carefully evaluated through risk assessment to determine the potential hazards and their significance to consumers. When a food is repeatedly implicated as a vehicle in food borne disease outbreaks, application of microbiological criteria may be useful (Micheal and Larry, 2007). The traditional method of examining microbiological safety, storage, stability, and sanitary quality of food is to test a representative portion or samples of the final product for the presence of certain pathogens, the number or level of certain pathogens (e.g., *Staphylococcus aureus*), different microbial groups (e.g., aerobic plate counts), and indicator bacteria (e.g., coliforms are used as an indicator of sanitation) per gram or milliliter of product (Silva, 2002). In this study, the smoked samples (smoked fish, meat and chicken products) were evaluated bacteriologically through determination of total bacterial count, coliform counts and the prevalence of the target food borne pathogen *staphylococcus aureus.*

Smoking of fish is one of the most ancient processing technologies for centuries used for preservation for centuries and is still widely used for this purpose among several communities in the third world where up to 70% of the catch is smoked for preservation (Ward, 1995). Concerning smoked herring, table (1) revealed that the aerobic plate count (cfu/g) was 2.96 x10^7 ± 1.65 x10^7while the minimum was 1x10^4 cfu/g and the maximum was1.72 x10^8.lower results were recorded by El_Shater (1994) and EL Sayed (1995), who recorded a mean value of 1.53 × 10^4/g with a minimum value of 1 × 10^2 and maximum value of 1.8 × 10^8/g. higher results were reported by Hammad (1985) for total bacterial count of smoked eel. The same author proved that, the smoking process reduced the total bacterial count (cfu/gm) from 2.9 ×10^5 in fresh eel to 1x10^3 after smoking.

Concerning smoked tuna, it was noticed in Table (1) that the aerobic plate count (cfu/g) of smoked tuna were 1.62 x10^8 ± 7.24 x10^7 with maximum value 1 x10^9 and minimum value 5 x10^6. Lower results were obtained by Meigy et al. (2013) and Ahmed (2015). Table (1) showed that the aerobic plate count (cfu/gm) of smoked salami was 8.36 x10^6 ±2.72 x$10^6$with maximum value 3 x$10^7$and minimum value 1 x10^6.

Concerning smoked rose beef, it was noticed in Table (1) that the aerobic plate count(cfu/g) were 1.36 x10^7 ± 4.66 x10^6 with maximum value 5 x$10^7$and minimum value 1 x10^6. Regarding smoked chicken, the results in table (1) showed that the aerobic plate count (cfu /gm) of smoked chicken were 2.75 x10^5 ± 6.59 x10^4 cfu/g and the minimum count was 5 x10^4 cfu/g while the maximum count was 7 x10^5 cfu/g. Higher value was reported by Awad (1997). Concerning smoked turkey, in Table (1) it was noticed that the APC were 3.13 x10^7 ± 5.15 x10^6 with maximum value 4.5 x10^7 and minimum value 2 x10^7. The results in table (2) showed total coliform count (cfu/g) in smoked herring was 1.81 x10^4± 83 x10^2 with a minimum of 4 x$10^3$and maximum of 1 x10^5. lower results were recorded by Gbogbolomo (2012) coliform count ranged from zero to 2 $\times 10^3$cfu/gm. the total coliform count of smoked tuna was (cfu/g) 8.82 x10^6±1.36 x10^6, where the minimum was 2 x10^7 and maximum was 16 x10^5 in table 2. Lower result was obtained by Nyarko et al. (2011).

Total coliform count(cfu/g) in smoked salami such product was 4.72 x10^6±11.9 x$10^5$where the minimum was 11 x$10^4$and maximum was 15 x10^6. The total coliform count in rose beef (cfu/g) 4 x10^4± 1 x10^4 where the minimum was 3 x10^4 and maximum was 5 x10^4. While total coliform count in the smoked chicken products were 7.16 x10^5±5.45 x10^5cfu/g where the minimum was 2 x10^3cfu/g and the maximum was 6 x10^6cfu/g. While total coliform count in smoked turkey products were 5.25 x10^7± 16.09 x10^6 cfu/g where the minimum was 3 x10^7 cfu/g and the maximum was 1 x10^8 cfu/g. While the results in table (3) showed total Staphylococci count in smoked herring (cfu/g) was 18 x10^5±15 x$10^5$with minimum count 4 x$10^3$and maximum count 1.97 x10^7. Lower results were recorded by Adegunwa et al. (2013). While the total Staphylococci count (cfu/g) as showed in table (3) was 39.5 x10^5±88 x10^4 with minimum count 7 x10^5 and maximum count 73 x10^5.Lower results were obtained by Jeyasanta et al. (2015). While the total Staphylococci count(cfu/g) in smoked salami was 31.7 x10^5±14 x$10^5$with minimum count 1 x$10^5$and maximum count 15 x10^6. While the total Staphylococci count (cfu/g) in rose beef was 1.62 x10^8 ± 7.24 x10^7 with minimum count 5 x10^6 and maximum count 1 x10^9

Total Staphylococci count was 23 x10^3±6.5 x10^3 cfu/g in the examined samples of smoked chicken products. Total Staphylococci count was 11.06 x10^7±4.25 x10^7cfu/gin the examined samples of smoked turkey as illustrated in table (3) and with 3 x10^8 cfu/g as a maximum and 1.5 x10^6 cfu/g as minimum. The results in table (4) and table (5) showed that *S.aureus* isolated from 66.6% of the examined smoked herring samples. where 53.3% were coagulase positive and 13.3% were coagulase negative. lower result was obtained by Abou Youssef (2014). who found *S.aureus* in 36% of the examined samples. *staph aureus was* isolated from 26.6% of the examined smoked tuna samples of smoked tuna. Where 20% were coagulase positive and 6 % were coagulase negative, nearly similar result was obtained by Abolagba and Uwagbai (2011), while lower result was obtained by Nyarko et al. (2011). *staph aureus* was isolated from 20. % of the examined smoked salami samples. where 6% were coagulase positive and 13.3%were coagulase negative. *staph aureus* failed to be isolated of any samples in smoked rose beef. Nearly similar results were obtained by Millard (1999). *staph aureus* was isolated from 60% 0f samples of smoked chicken, 40% were coagulase positive while 20% were coagulase negative lower result was obtained by Awad (1997) who showed that *S.aureus* were isolated from 13.3% 0f samples of smoked turkey and all were coagulase positive. Nearly similar results were obtained by Awad (1997). It is commonly suggested that microorganisms can enter meat preparation like sausages from meat, spices, and other ingredients, as well as from processing environment, equipment, and handlers that can have a significant impact on the microbiological status of the end-products. In general, heating during technological processing of meat products is an effective tool to reduce microbial counts of end-products (Güngör and Gökoģlu, 2010).

Poor hygienic practices in food processing plants may result in the contamination of food products with pathogens causing a serious risk for human health. Moreover, the complete elimination of pathogens from food processing environments is a difficult task, in part because bacteria can attach to food contact surfaces and form biofilms where they survive even after cleaning and disinfection (Wang et al., 2012). The safety of food must be assured by a preventative approach based on the application of a Hazard Analysis Critical Control Point (HACCP) at all stages of food chain. The HACCP system is a structured approach for identifying hazards and defining and implementing systems of adequate control. Risk-based programs have been proved successful in achieving hazard control to the extent required for consumer protection. Microbiological examination of food and environmental samples is generally recommended to validate and verify the efficiency of foods safety and quality control.

In conclusion, the process of smoking must be done under complete hygienic condition in order to minimize the risk of high bacterial load to become safe for consumer.

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