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ANTIBACTERIAL ACTIVITY OF SOME PLANT EXTRACTS ON DIFFERENT BACTERIA IN CHICKEN FILLET

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ABSTRACT

Foodborne diseases affect millions of people each year. More effective treatment methods are needed to reduce incidence of food poisoning organisms in food. In the present study, the prevalence of *S. aureus*, *E. coli* and *salmonella* was evaluated in 100 samples of chicken fillet collected from chicken processing shops in Cairo governorate. The prevalence of *S. aureus*, *E. coli* and *salmonella* was 56%, 70% and 12%, respectively. The effect of marinating chicken fillet with some medicinal plants such as extracts of Moringa, lemon and green tea against *S. aureus*, *E. coli* and *Salmonella* was studied. The 100% lemon extract showed the best antibacterial activity decreasing count of *S. aureus*, *E. coli* and *salmonella* 3, 3.5, 3-log cfu/g, respectively. On the other hand, the 6% moringa extract was better than the 4% and the 2.5% green tea extract was better than the 1% in decreasing the bacterial count. This indicates the effectiveness of such extracts in controlling bacterial growth in chicken meat and can be applied as well to other meats.

KEYWORDS: Moringa Oleifera, Lemon, Green tea, chicken, meat.

INTRODUCTION

Raw meat, particularly poultry meat, remains a source of human infection with pathogenic microorganisms. It can be easily contaminated with microorganisms because fresh meat is very suitable for microbial multiplication (**Hinton, 2000**).

Staphylococcus aureus is a significant cause of avian diseases and may thus contaminate food because of carcasses processing (**Mead and Dodd, 1990).** Enterotoxin-producing *S. aureus* is the most common cause of foodborne human illness throughout the world **(Do Carmo *et al*., 2004).**

E. coli is responsible for 25% of the infant diarrhea in developing countries **(WHO, 2000).** Shiga toxin producing *E. coli* (STEC) was first recognized as a human pathogen in 1982 in the USA when strains of serotype O157:H7 caused two outbreaks of hemorrhagic colitis **(Wells *et al.*, 1983**).

Most *Salmonella* found in poultry meat are non-host-specific and are capable of causing human food poisoning. Salmonellosis (gastroenteritis) is the most common food borne disease in human with an incubation period of 6 to 72 hours **(Behravesh, 2008)**. Bacterial food borne diseases are very expensive causing huge losses to the food industry and consumers. Despite methods to reduce or eliminate bacterial food borne pathogens, food poisoning is still a major concern **(Shan *et al.*, 2007).**

Moringa (*Moringa Oleifera*) leaf is a natural ingredient reported to be high in crude protein contents **(Bey, 2010).** Moringa leaf powder could be used as food or for medicinal and therapeutic purposes **(Anwar *et al*, 2007)**. It is used for improving wound healing, gastric ulcer, diarrhea, sore throat and cancer **(Grever, 2001)**. In many countries, Moringa leaves are used as traditional medicine for treating common illnesses **(Trees for Life, 2005).**

Lemon is an important medicinal plant of the family Rutaceae. It is cultivated mainly for its alkaloids, which have anticancer activities and the antibacterial potential in the crude extracts of different parts (viz., leaves, stem, root and flower) of Lemon against clinically significant bacterial strains has been reported **(Kawaii *et al*., 2000).**

Citrus flavonoids have a large spectrum of biological activities including antibacterial, antifungal, antidiabetic, anticancer and antiviral activities **(Burt, 2004; Ortuno *et al.*, 2006).** Lemon juices are highly useful as antibacterial agents **(Vijayakumar and Wolf-Hall, 2002).**

Green tea contains catechin and epicatechin, which have been extensively studied and shown to be effective against a wide range of food borne pathogens (**Taguri *et al.*, 2004**). There have been numerous studies on polyphenols found in green tea and their health benefits. Not only is green tea an antioxidant, but is also known for its antibacterial, antimycotic, antiviral as well as antitoxic activities against bacterial hemolysins **(Okubo *et al.*, 1989).**

Therefore, this study was planned to determine the prevalence of *S. aureus, E. coli* and *Salmonella* as food poisoning organisms in chicken fillet. Further, studying the antibacterial effects of some medicinal plants (Moringa Oleifera, Lemon and Green tea extracts) on contaminated chicken fillets.

MATERIALS AND METHODS

Collection of Samples

A total of 100 chicken breast fillet samples were collected from chicken processing shops in Cairo governorate and transported to the laboratory in ice box without delay to be examined.

Preparation of Food Homogenate

Sample preparation was applied according to **APHA (1992)**. Twenty-five grams of each examined chicken fillet samples were transferred to 225 ml of sterile buffered peptone water 0.1% then homogenized by stomacher for 2 minutes to provide a homogenate of 1/10 dil. The isolation and enumeration of *Staphylococcus aureus* (*S. aureus*) was carried out according to **FDA (2001)**, count of *Escherichia coli* (*E. coli*) was according to **APHA (1992)** and isolation of Salmonella spp. was according to **FDA (2007)**.

Preparation of Plant extracts

a. Moringa extract

According to **Bichi *et al.*, (2012)**, 10 g of Moringa Oleifera leaves powder was added to 200 ml distilled water and blended in high speed mixer for 30 minutes at 150 rpm to extract the active ingredients then filtered through Whattman filter paper and the filtrate was made up to 500 ml with distilled water in sterile beaker. This stock solution was used to formulate product at two concentrations: 4 and 6% aqueous suspension to be used in the experimental study.

b. Lemon extract

Samples were cut into halves using sterile knife and the liquid was squeezed individually in sterile container. This extract was marked as 100% concentrated juice extract. Concentration of 50% was prepared by diluting the 100% extract with right volume of sterilized distilled water (**Owhe-Ureghe *et al.*, 2010)**.

c. Green tea extract

Ten grams of ground dry green tea was added to 100 ml of distilled water and heated at 30- 40ºC for 45 min with a magnetic stirrer (DELTA Model HM- 101, Industries LTD). The mixture was then filtered with Whattman paper and the filtrate was used as green tea extract at concentrations 1 and 2.5% aqueous suspension (**Sarah *et al.*, 2010)**.

Preparation of bacterial cultures

S. aureus (ATCC 29213), *E. coli* (ATCC 8739) and *Salmonella enterica* (ATCC14028) were kindly supplied by the Department of Food Hygiene, Animal Health Research Institute, Dokki, Giza. From frozen cultures, the bacteria were activated with two successive passages in 9 ml of tryptic soy broth (TSB) (Oxoid) and incubated at 37ºC for 18 h. For each individual bacterial strain, 1 ml of the stock inoculum was added to 100 ml of TSB and incubated with shaking at 37ºC for 18 - 24 h to reach a final concentration of approximately 10^8 CFU/mL (determined by plating serial dilutions on TBX for *E. coli*, Baired Parker for *S. aureus* and XLD for *Salmonella enterica* agar). Nine ml of this inoculum was added to 25 ml of sterile saline to give approximately 10^7 CFU/mL **(Corry et al. 2007).**

Preparation of the experimental food model

Raw chicken breast fillets (boneless) were washed 10 times with distilled water and rinsed twice in sterile water, then cut into pieces of 5 cm x 5 cm using a sterile knife. The pieces were kept in sterile open petri dishes and exposed to UV (at 254 nm) for 30 min on each side and then frozen at -20°C for 24 hrs **(Valtierra-Rodriguez *el.*, 2010)**.

Inoculation of the experimental food model

Chicken breast fillet samples were divided into 4 groups as shown in Table (1).

Table (1): Experimental groups.

Groups	Treatment	A	B		C		D	
			B1	B2	C1	C2	D1	D2
Group (1)	Control group: fillets pieces were each inoculated with different concentrations of Moringa Oleifera, Lemon and Green tea extracts							
Group (2)	Fillets pieces inoculated with 100 µl of 10^7 cfu/mL of *S. aureus*							
Group (3)	fillets pieces inoculated with 100 µl of 10^7 cfu/mL of *E. coli*	no extract	4% Moringa oleifera	6% Moringa oleifera	50 % lemon extract	100% lemon extract	1 % Green tea extract	2.5 % Green tea extract
Group (4)	fillets pieces inoculated with 100 µl of 10^7 cfu/mL of *Salmonella entrica*							

The fillets pieces were incubated with different bacterial inoculum as indicated in Table (1) at room temperature for 30 min in a well aerated sterile bottle to allow bacterial attachment (**Murali *et al.*, 2012)**. Inoculated chicken fillet pieces were individually dipped in the different concentrations of the different extracts for 5 minutes, then left to dry in the laminar flow for 15 minutes. Microbial growth was assessed on the experimentally inoculated chicken fillet at zero day and every two days after incubation at 4℃ until the signs of spoilage were detected (*S. aureus* according to **FDA (2001)**, *E. coli* according to **APHA (1992)** and *Salmonella enterica* according to **FDA (2007)**). These experimental procedures were repeated three times.

Sensory evaluation

Sensory analysis (taste, color and odor) was assessed according to the descriptive sensory method of **Hedonic ASTM, (1969)** as shown in Table (2).

Table (2): Sensory evaluation of chicken fillets.

Sensory attributes description score
natural flavor, color and odor of fillet 5
No sensible change in natural flavor, color and odor 4
Sensible discoloration, Slightly sour odor and incipient rancidity in flavor 3
no natural color, moderately off-odor and off-flavor 2
Sharply sour and extremely rancid flavor/odor, extremely discolored 1

RESULTS

Prevalence of *S. aureus, E. coli* and *Salmonella* in examined chicken fillet samples

S. aureus, E. coli and *Salmonella* were recovered from 100 samples of chicken fillet with incidence rates of 56%, 70% and 12 %, respectively.

Count of *S. aureus* (log_{10} cfu/g) ranged from 1.78 to 2.54 with average of 2.10±0.03 and *E. coli* (log_{10} cfu/g) ranged from 1.30 to 6 with average of 3.03±0.19**.**

***S. aureus* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation**

Count of *S. aureus* in group 1 and group 2B1 (4%) decreased by the 2nd day of incubation. In group 2B1, *S. aureus* decreased by 0.5 log cfu/g then increased until the end of incubation. Meanwhile, in group 2B2 (6%), *S. aureus* count decreased on the 2nd day of incubation and continued till the 4th day by one log cfu/g then began to increase from the 6th day till the end of incubation (Fig. 1).

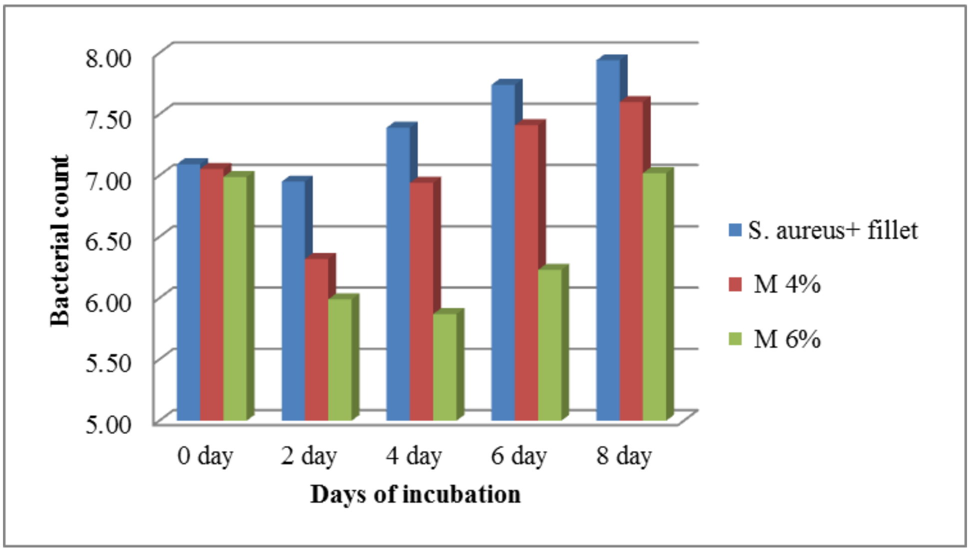

Fig (1): Mean *S. aureus* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation. M; Moringa Oleifera.

***E. coli* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation**

Count of *E. coli* as illustrated in Fig. 2, in the control group decreased by 2nd day then increased again after the 4th day till the end of incubation, while in 4% Moringa-treated fillets it decreased by the 2nd day and 4th day of incubation by 0.5 log cfu/g then remained nearly stable till the end of incubation. Moreover, in 6% Moringa-treated fillets, *E. coli* count decreased from the 2nd day of incubation until the end of the incubation by 1.5 log cfu/g.

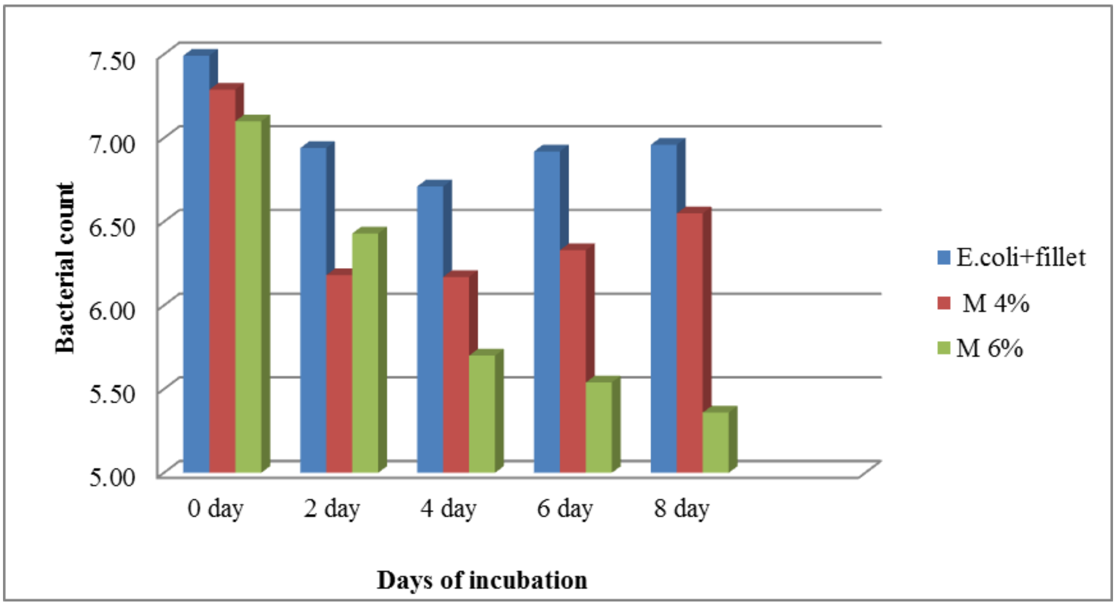

Fig (2): Mean *E. coli* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation. M; Moringa Oleifera.

***Salmonella* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation**

In Fig. 3, count of *Salmonella* in the control group showed slight decrease by the 2nd day then continued to increase from the 4th day until the end of incubation. While with 4% Moringa-treated *Salmonella*, the count decreased by the 2nd day by 0.3 log cfu/g then increased by the 4th day until the end of incubation. In contrast, the 6% Moringa decreased the *Salmonella* count from the second day till the end of incubation by one log cfu/g.

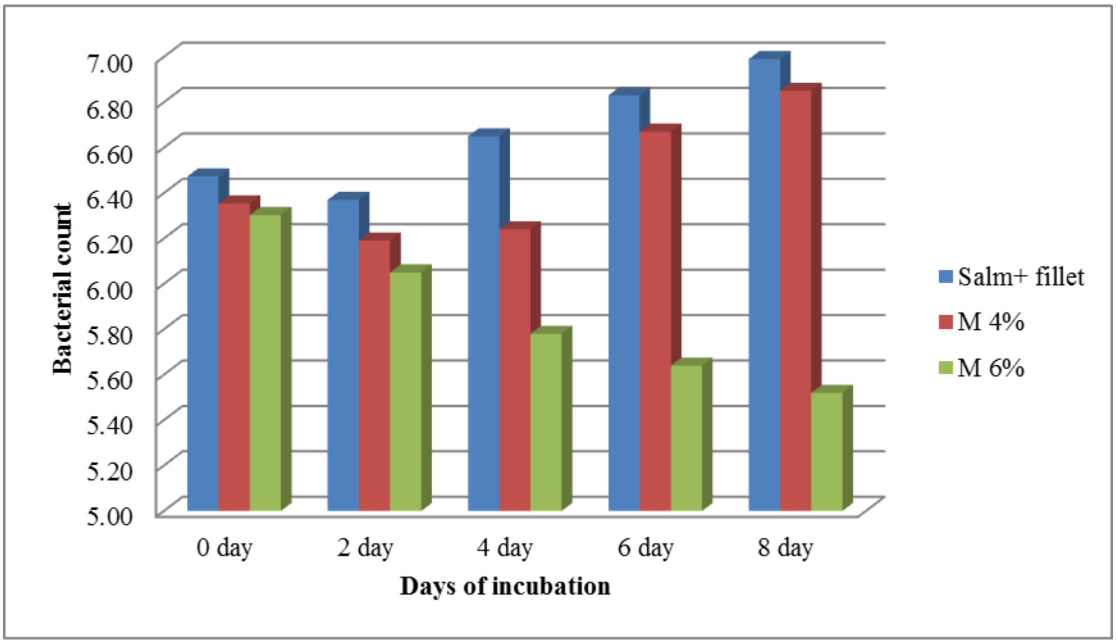

Fig (3): Mean *Salmonella* count in chicken fillet marinated with Moringa Oleifera through 8 days of incubation. Salm; *Salmonella*, M; Moringa Oleifera.

***S. aureus* count in chicken fillet marinated with lemon juice through 8 days of incubation**

Count of *S. aureus* in the control group showed slight decrease during the 2nd and 4th days of incubation then began to increase till the end of incubation. While in 50% lemon-treated, count of *S. aureus* decreased from the 2nd day of storage until the 6th day by 2 log cfu/g then slightly increased till the end of incubation. Moreover, in 100% lemon-treated, *S. aureus* count decreased 3 log cfu/g during incubation till the 6th day of incubation then slight increase was detected (Fig. 4).

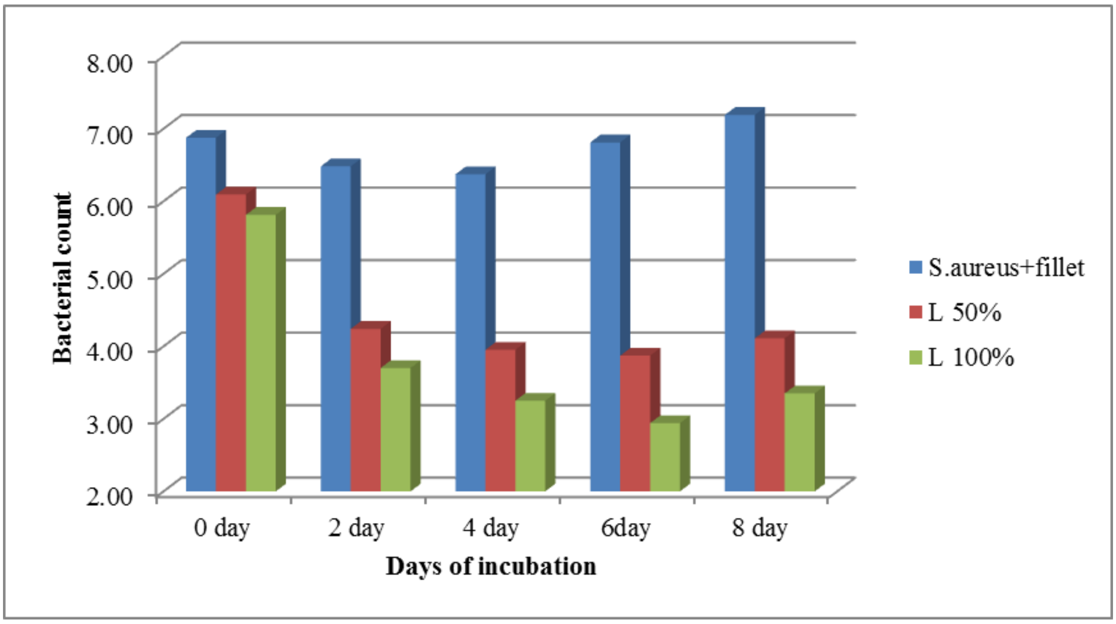

Fig (4): Mean *S. aureus* count in chicken fillet marinated with lemon juice through 8 days of incubation. L; lemon juice.

***E. coli* count in chicken fillet marinated with lemon juice through 8 days of incubation**

E. coli count in control group slightly decreased from 2nd day of storage till 4th day then increased again till the end of incubation. While with adding 50% lemon juice, count decreased during till the 6th day of incubation by about 3 log cfu/g in contrast to 3.5 log cfu/g decrease in 100% lemon treated group till also 6th day then the count increased at the 8th day of incubation as illustrated in Fig. 5.

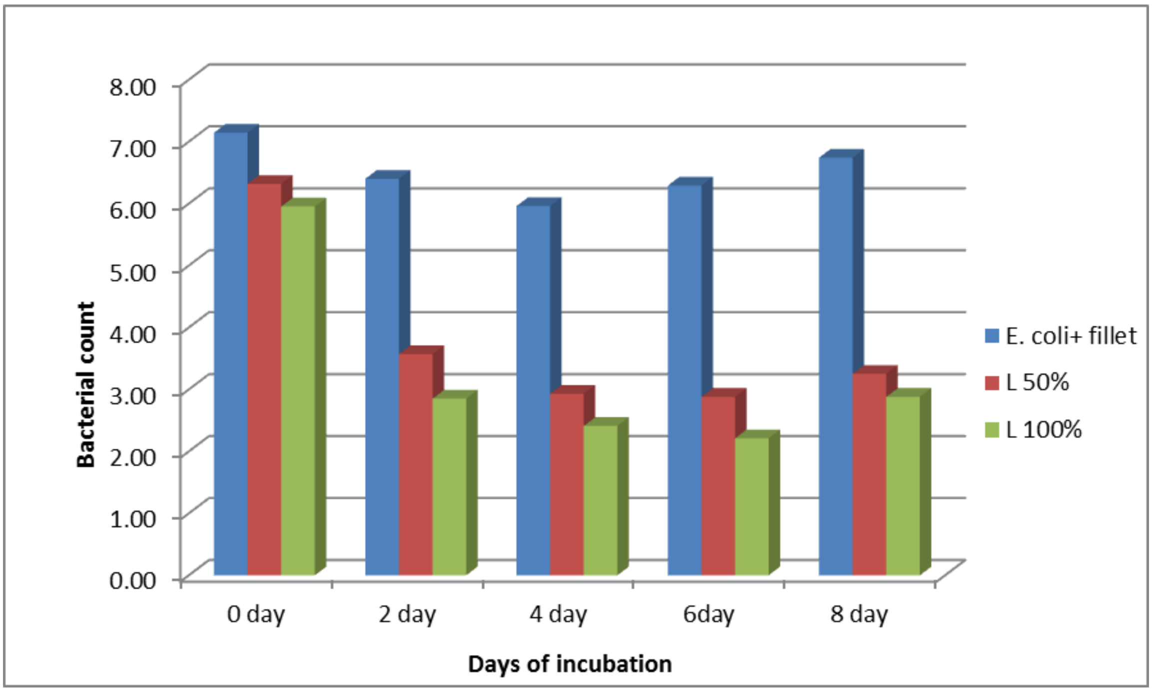

Fig (5): Mean *E. coli* count in chicken fillet marinated with lemon juice through 8 days of incubation. L; lemon juice.

***Salmonella* count in chicken fillet marinated with lemon juice through 8 days of incubation**

In Fig. 6, *Salmonella* count in the control group showed no significant decrease from the 2nd day of incubation till the 4th day then increased again till the end of incubation. While with adding 50% lemon juice, *Salmonella* decreased till the 6th day of incubation by about 2 log cfu/g. With 100% lemon juice, the count decreased by 3 log cfu/g till also the 6th day then increased till the end of incubation.

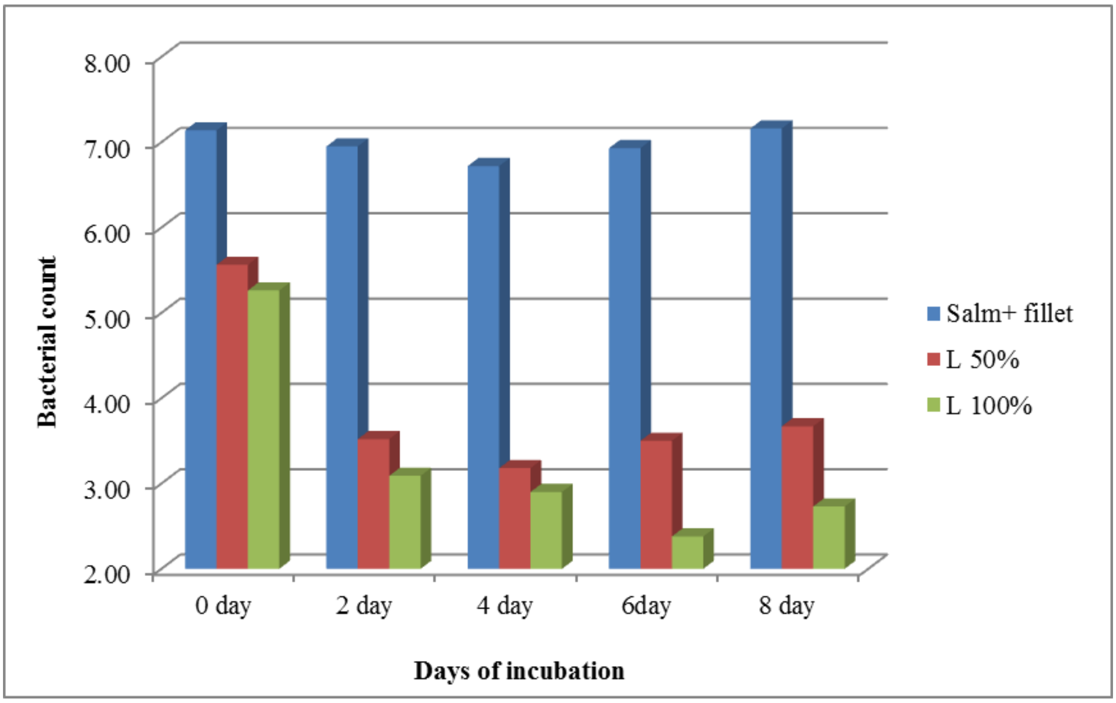

Fig (6): Mean *Salmonella* count in chicken fillet marinated with lemon juice through 8 days of incubation. Salm; *Salmonella*, L; lemon juice.

***S. aureus* count in chicken fillet marinated with green tea through 8 days of incubation**

Fig. 7 shows decrease of *S. aureus* count in control group, 1% and 2.5% green tea treated groups from the 2nd day till the 4th day then the count increased from the 6th day till the end of incubation. This decrease in 1% and 2.5% green tea were nearly similar with about 0.5 log cfu/g as compared to the control.

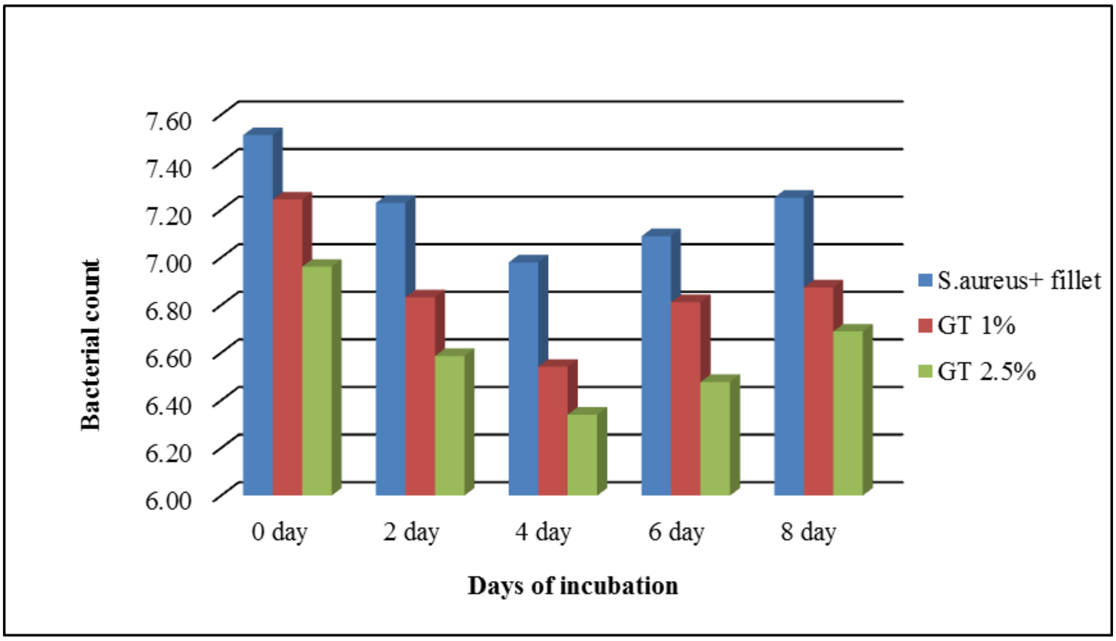

Fig (7): Mean *S. aureus* count in chicken fillet marinated with green tea through 8 days of incubation. GT; green tea.

***E. coli* count in chicken fillet marinated with green tea through 8 days of incubation**

In Fig. 8, *E. coli* count was stable in the control group on the 2nd day then increased from the 4th day till the end of incubation. *E. coli* count decreased with 2.5% green tea from the 2nd till the 4th day of incubation with about 1 log cfu/g more than the decrease with the 1% green tea treated which was 0.5 log cfu/g, then the count increased from 6th day till the end of incubation for both concentrations.

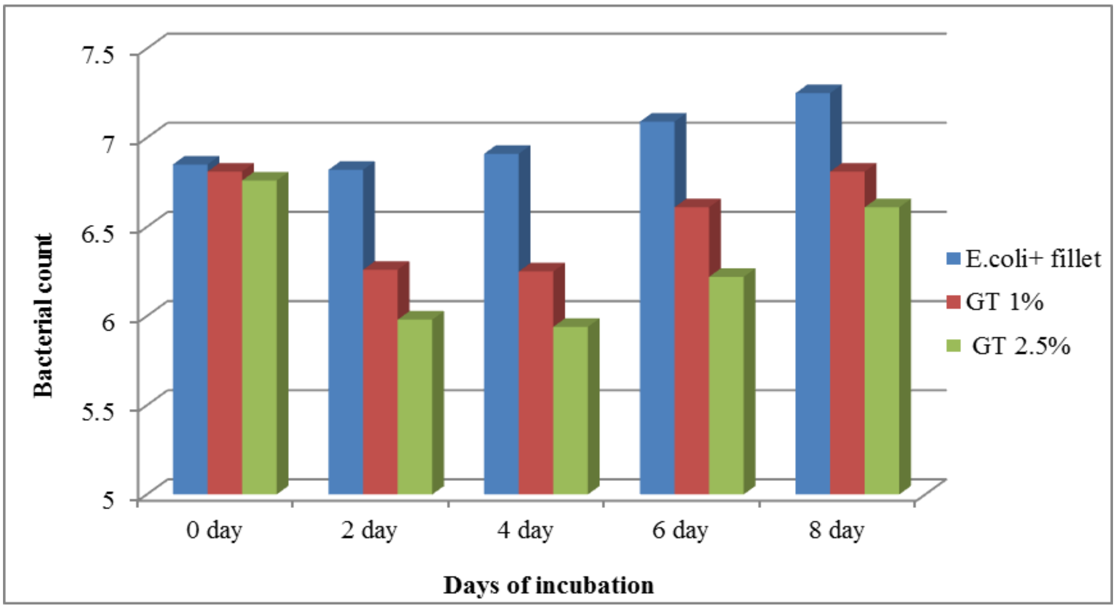

Fig (8): Mean *E. coli* count in chicken fillet marinated with green tea through 8 days of incubation. GT; green tea.

***Salmonella* count in chicken fillet marinated with green tea through 8 days of incubation**

Salmonella count in the control group showed nonsignificant decrease on the 2nd day and 4th day then increased from the 6th day till the end of incubation. While 1% green tea treated group showed decreased count of *Salmonella* by the 2nd day till the 4th day of incubation by 0.5 log cfu/g then increased thereafter till the end of incubation. Moreover, with 2.5% green tea *Salmonella* decreased from the 2nd day till the 4th day of incubation by 1 log cfu /g then the count increased thereafter till the end of incubation.

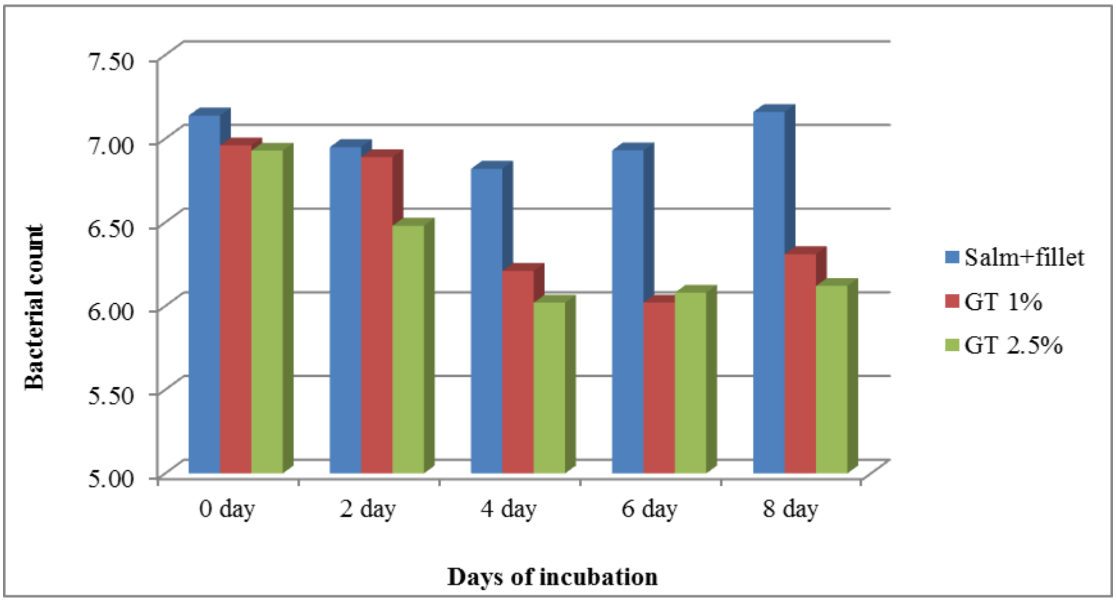

Fig (9): Mean *Salmonella* count in chicken fillet marinated with green tea through 8 days of incubation. Salm; *Salmonella*, GT; green tea.

Sensory evaluation of the chicken fillet after 8 days of incubation with different extracts

Change in the sensory characters in the control groups 2A, 3A, 4A during preservation at 4℃ began from day 2 with score (3) and day 3 with score (2) and score (1) at 6th day of incubation. Adding of different concentrations of Moringa olifera and green tea changed the fillet color to slightly greenish with score (4). On the contrary, lemon extract did not result in any color change with score (4).

DISCUSSION

Raw meat, particularly poultry meat, remains an important source of human infection with pathogenic microorganisms. It can easily be contaminated with microorganisms because fresh meat is very suitable for microbial multiplication. Poultry meat has high water activity, rich in nutrients and readily utilizable low molecular weight substances and is a source of carbon and energy by means of glucose, lactic acid, amino acids, creatines, metal and soluble phosphorus **(Hinton 2000)**.

S. aureus prevalence in chicken fillet was 56%. This is nearly similar to **Kitai *et al.*, (2005)** (65.8%), **Javadi and saeid (2011)** (65%) and **Martins *et al.*, (2013)** (62%) but higher than that obtained by **Kozaciki *et al.*, (2012)** (17.9%), **Bhaisare *et al.*, (2014)** (11.25%), **Schaumburg *et al.*, (2014)** (3%) and **Khaled *et al.*, (2015)** (3.3%). The presence of *S. aureus* in food commonly indicates contamination that may be directly introduced by workers who have skin lesions contaminated with *S. aureus*, or sneezing, coughing, or unclean hands and contaminated utensils and equipment. The hazard develops into toxin formation when raw materials and products are exposed to temperatures between 10°C and 21.1°C for more than 12 hrs or to temperatures greater than 21.1°C for more than 3 hrs **(FDA, 2001)**, so its presence indicates poor sanitation during processing.

E. coli prevalence in chicken fillet was 70%. This count was closely similar to **Bhaisare *et al.*, (2014)** (70.2%), **Odwar *et al.*, (2014)** (78%) and **Edris *et al.*, (2015)** (70%) but higher than that obtained by **Vaidya *et al.*, (2005)** (14.57%) and **Khaled *et al.*, (2015)** (10%) and lower than that obtained by **Huong *et al.*, (2009)** (100%) and **Saikia and Joshi (2010)** (98%). Presence of *E. coli* in high numbers indicates fecal or sewage pollution. This is also due to improper slaughtering techniques, contaminated surfaces and/or handling of the meat by infected food handlers **(Nel *et al.*, 2004)**.

Salmonella prevalence in chicken fillet was 12%. This is nearly similar to **Kozaciniski *et al.*, (2006)** (10.60%) and **Kozaciki *et al.*, (2012)** (7.5%). Higher results were obtained by **Thai *et al.*, (2012)** (42.9 %), **Saeed *et al.*, (2013)** (22%) and **Bhaisare *et al.*, (2014)** (33.16%). Lower results were reported by **Rabie *et al.*, (2012)** (4%), **Anju *et al.*, (2014)** (4.44%) and **Khaled *et al.*, (2015)** (3.3%). The primary reservoir of *Salmonella* is the intestinal tract of animals and birds, which contaminate the muscles and organs during slaughtering **(Paiao *et al.*, 2013)**. Poultry and poultry products are the most potential source of Salmonella-food poisoning in man **(Lynch *et al.*, 2006)** that can be transmitted to humans through handling of raw products or through consumption of undercooked poultry meat **(Kimura *et al.*, 2004)**.

Due to uncontrolled use of antibiotics and their side effects such as anaphylaxis, digestive problems, teeth and bone staining, fungal infections and photosensitivity according to the World Health Organization. And overuse of prescription antibiotics which can lead to the development of antibiotic-resistant strains of bacteria, many medical experts are taking a new look at natural, safe antibiotic alternatives.

During the last few decades, the global interest in the study of various medicinal plants has increased rapidly due to their antibacterial and antioxidant activities, low toxicity and the potential to be a cheaper alternative to costly synthetic drugs **(Chew *et al.*, 2012)**. Therefore, such plants should be investigated to better understand their properties, safety and efficiency.

The effect of Moringa Oleifera is due to the presence of some phytochemicals, but most importantly due to the activity of a short polypeptide named 4 a-Lrhamnosyloxy benzyl-isothiocyanate **(Guevara *et al.*, 1999)** and the rare combination of zeatin, quercetin, kaempferom among many other phytochemical compounds **(Bukar *et al.*, 2010)**. These compounds might be responsible for the significant decrease in the count of *S. aureus, E. coli* and *Salmonella* in the Moringa treated chicken fillet samples.

This results match those of **Hitzschky *et al.*, (2010)** who studied the antibacterial effects of aqueous extracts of Moringa seeds (*Moringa Oleifera*) at concentrations of 1:5 and 1:10 in volumes of 50, 100, 150 and 200 µL against *Staphylococcus aureus, Escherichia coli* (isolated from the organism and the aquatic environment) and *Salmonella enteritidis*. Antibacterial activity (inhibition > 13 mm) against *S. aureus* and *E. coli* isolated from the white leg shrimp was detected in aqueous extracts of Moringa but the antibacterial effects on *Salmonella enteritidis* was not demonstrated.

Adeyemi *et al.*, (2013) studied the antimicrobial effect of Moringa Oleifera on smoke-dried catfish. The treatments were the control, 1%, 2% and 3% (w/v) Moringa Oleifera Marinade. At all levels of treatment, bacterial counts for *S. aureus* and *E. coli* decreased as compared to the control samples. The 3% concentration exhibited the highest antibacterial potency. **Zeid (2014)** studied the effect of Moringa leaf powder (0.5, 0.75, 1%) in beef burger and showed that addition of Moringa at any level improved the stability characteristics and microbial quality of beef burger without affecting the sensory characteristics. **Tolba *et al.*, (2014)** studied the effects of 2, 4, 6% of Moringa Oleifera leaves extract on the microbial contamination of experimentally inoculated fresh fish fillets with 10^7cfu/g of *S. aureus.* They found that 6% Moringa was more effective as compared to 2% or 4% in reduction of *S. aureus* count where it was reduced by 1 log cfu/g within the first 5 days of the experiment.

The higher concentration of the Moringa extract (6%) showed was better than the lower concentration (4%) in decreasing the count of *S. aureus, E. coli* and *Salmonella* with average (1 log cfu/g) which helped increasing shelf life of chicken meat. This could be enhanced as well by the effects of low temperature on reducing bacterial growth.

The effect of lemon juice is mainly due to the essential oils known to penetrate the cell membrane and mitochondrial membranes, thereby increasing permeability of the organelles, which lead to ion leakage **(Raybaudi-Massilia *et al.*, 2006).** Citrus species also contain a group of flavonoids including polymethoxy flavones, flavone glycosides and limonoids that enhance antimicrobial activity **(Ladaniya, 2008)**. Citrus flavonoids have broad spectrum of biological activities including antibacterial, antifungal, anti-diabetic, anticancer and antiviral activities **(Burt, 2004, Ortuño *et al*., 2006).** The results reported here are similar to those shown by **El-Khawas *et al.*, (2015)** where lemon juice (2% citric acids) reduced *S. aureus* by 2 log cfu/g and significantly reduced *Salmonella* (2 log cfu/g) and *E. coli* (3 log cfu/g) counts. **Kumar *et al.*, (2012)** showed similarly that lemon juice inhibited the growth of *S. aureus, E. coli* and *Salmonella* with inhibition zones of 19.33±1.24, 20.0±0.81 and 21.66±0.47, respectively, but differ than **Murali *et al.*, (2012**) who reported that lemon killed *Salmonella* after 36 hrs of incubation with a 5-log reduction in growth within 1 hr of incubation.

Lemon extract at 100% concentration showed the best antibacterial activity that decreased count of *S. aureus, E. coli* and *salmonella* with 3, 3.5, 3 log cfu/g, respectively. On the other hand, 50% Lemon extract was less effective where it decreased the count of *S. aureus, E. coli* and *salmonella* with 2, 3, 2 log cfu/g, respectively.

The bacterial growth inhibitory effects of green tea are mainly related to their polyphenolic components including epicatechin, epicatechin gallate, epigallocatechin, and epigallocatechin gallate against various Gram-positive and Gram-negative bacteria **(Blanco *et al.*, 2005, Zhao *et al.*, 2002)**. Green tea leaves extract can prevent the attachment of pathogenic bacteria on the host cell membrane. Thus, green tea extract inhibits the adhesion of bacteria on host cell surface membranes and acts as a potential antiadhesive agent **(Lee et al., 2009)**. Further, Epigallocatechin gallate can reverse methicillin resistance of Methicillin resistant *S. aureus* (MRSA) by inhibiting the synthesis of Penicillin Binding Protein 2 (PBP2) **(Yam et al., 1998)**. Higher green tea extract (2.5%) was better than the 1% concentration where it decreased the count of *E. coli* and *Salmonella* with average 1 log cfu/g and of low value on *S. aureus* count. Our results are nearly similar to **Takabayashi et al., (2004)** and **Yee et al., (2002)** who reported that green tea extract did not kill *Salmonella enteritidis* in chicken fillet even after 36 hrs, even though it showed decreased total bacterial count. **Archana and Abraham (2011)** assessed the antimicrobial activity based on measuring the inhibition zones. The organisms found to be sensitive to fresh green tea extracts with zones of inhibition 6 and 14 with *E. coli* and *S. aureus*, respectively. **Maksum Radj, Rafael Adi Agustama (2013)** showed that the inhibition zone diameter of green tea extracts for S. aureus ATCC 25923 and MRSA were 18.970±0.287 mm.

The use of the Moringa and green tea extracts affected the color of the chicken fillet, while lemon extract showed no significance difference up to 4th days of storage time. Sensory characteristics remained within good levels with the three extracts. **Adeyemi *et al.*, (2013)** marinated catfish fillet with Moringa Olifera (1, 2, 3%) and sensory scores showed no significance difference among the treatments in terms of color, flavor, juiciness and over all acceptability of fish samples. **Najeeb *et al.*, (2014)** stated that the appearance, flavor, juiciness and overall acceptability value of the restructured chicken slices containing Moringa leaf powders (1%) were not affected and were safely stored without much loss in quality up to 20 days under refrigeration with lower microbial counts. **Hazra *et al*., (2012)** concluded that the use of crude extract of Moringa Oleifera leaves (1, 1.5 and 2%) had significant antioxidant and antimicrobial effects and improved the organoleptic quality by enhancing the tenderness and juiciness in cooked ground buffalo meat. **Sarah *et al*., (2010)** reported slightly bitter and greeny flavor with greenish-yellowish color for 5% and 2.5% green tea extract treatments at all sampling times. The discoloration changes of fish fillets dipped in 2.5% and 5% steadily increased with time in storage. This might be implicated to the possibly penetration of chlorophyll pigments and their subsequent interference with other biochemical active compounds in fillets of sturgeon, which caused an undesirable change in meat color. **Anna *et al.*, (2019)** showed that chicken breast muscles marinated with lemon juice improve acceptability of sensory characteristics (reduction of brightness due to acid and increase of juiciness and tenderness) compared to control.

The results here showed that there is a high level of *E. coli* isolation from chickens examined in this study indicating possible fecal pollution or sewage contamination of water used for market washing of the fillet or contamination during slaughtering, handling or processing of the meat or infected workers.

Our results indicate the effectiveness of the Moringa Oleifera, lemon and green tea extracts in association with the low temperature of incubation in controlling bacterial growth in chicken meat for days and can be applied as well to other meats. However, the overall results for all the extracts used indicated that 4 -5 days are the maximum days for storage of marinated chicken fillet at 4°C after which there will be an increase of bacterial growth. Such growth would cause fast spoilage that cannot be noticed due to the color of the extract or its odor that could be of health hazard if processed for consumption.

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