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Original Research Article

Decontamination of broiler carcasses` skin using medicinal herbal extracts

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ABSTRACT

This study was carried out to detect the efficiency of different concentrations of medicinal herbal extract as decontaminant agent on broiler carcasses. Therefore, a total of twelve broiler carcasses treated with sumac extract (4% and 8% w/v) and rosemary extract (0.3% and 0.5% w/v) and stored at 0±1°C. All treated carcasses were sensory and microbiologically analyzed. It has been found that there were no changes in color and odor of treated carcasses, either by sumac extract (8% w/v) or rosemary extract (0.5% w/v), however, a reduction in the total bacterial count, coliforms count, *E. coli* count and *Staphylococcus* spp. count one/two log less than control samples and shelf-life of broiler carcasses was noticed and extended three to six days more than the control. From the present study, it could be concluded that sumac extract (8% w/v) and rosemary extract (0.5% w/v) are effective as broiler meat decontaminant and preservative.

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1. Introduction

Over the years, medicinal herbs physiologically contain active principles that have been exploited in traditional medicine for the treatment of various ailments as they contain antimicrobial properties (Kelmanson et al., 2000). Antimicrobial properties of herbs have been documented in ancient literature and the interest continues to the present. However, few of them have been investigated for antimicrobial activity. Medicinal value of drug plants is due to the presence of some chemical substances in plant tissues which produce antimicrobial effects. Such chemicals include alkaloids, flavanoids, glucosides, tannins, gums, resins, essential oils, fatty oils, carbon compounds, hydrogen, oxygen and nitrogen salts of some chemicals, etc.

Some studies claim that the phenolic compounds present in spices and herbs might also play a major role in their antimicrobial effects (Hara-Kudo et al., 2004). Herbs prolong the storage life of foods by preventing rancidity through their antioxidant activity or through bacteriostatic or bactericidal activity (Beuchat and Golden, 1989). There have been a number of reports of substances in each of cinnamon and clove oils that inhibit the growth of microbes such as *Salmonella* spp. (Suksrikarm, 1987). Therefore, an interesting alternative to use other chemical preservatives appear well suited to use in active packaging systems. The bacteriostatic and bactericidal effects of sumac on food-borne pathogens have been demonstrated in broth and agar media (Digrak et al., 2001). The main compounds in sumac are hydrolysable tannins and substantial amounts of flavonoids. Such property of tannins may be important for meat processors (Gulmez et al., 2006). It is well established that rosemary and sumac extracts have antibacterial properties (Sagdic and özcan, 2002).

The objective of this study is to detect *in vivo* the effectiveness of different concentrations of sumac and rosemary extracts to reduce the bacterial load on chicken broiler carcasses.

2. Materials and methods

2.1. Preparation of extracts

2.1.1. Extraction

2.1.1.1. Sumac extraction

Sumac fruit was added to sterile distilled water in a sterile bag and left at 45°C for 12 h. The bag was squeezed by hand to crush its contents. Crushed contents were filtered through cheesecloth into a sterile flask. The filtrate was concentrated and evaporated at 45°C until the solid mass was obtained. The extract was used at the same day (Gulmez et al., 2006).

2.1.1.2. Rosemary extraction

Dried plant material was milled by using hammer mill. Extraction of plant material was carried out using absolute ethanol solvent by immersion for about 3 days with intermittent shaking. Contents were filtered through a piece of gauze to remove solid plant materials then the extract was re-filled through filter paper to remove fine or colloidal particles from the extract. The enriched extract was concentrated by evaporation of the used solvent by heating in water bath at 65°C until solid mass was obtained. The solid extract was spread under shaded area until complete dryness, and then stored in the refrigerator till be used (Tandon and Ran, 2008).

2.1.2. Preparation of different concentrations of extracts

2.1.2.1. Sumac (*Rhus coriaria* L):

Sumac extract (4 and 8 % w/v) was prepared by weighting 40 and 80 grams of sumac extract. Each weight was dissolved in one liter of sterile distillated water.

2.1.2.2. Rosemary (*Rosmarinus officinalis* L)

A concentration of 0.3 and 0.5 % of (w/v) were prepared by weighting 3 and 5 grams of rosemary extract. Each weight was dissolved in one liter of sterile distillated water.

2.2. Sample collection

A total of 12 chicken broiler carcasses were collected from Beni-Suef butcher's shop then transferred to Animal Health Research Institute, Beni-Suef branch in ice-boxes. Samples were collected in sealable sterile polyethylene bag.

2.3. Sample preparation

2.3.1. Decontamination

A carcass was divided into two skinned halves; treated and control ones. Each half was considered a unit and was separately treated in sterile polyethylene bags (a concentration of extracts used for 3 units). Each bag would be slowly shaken and rotated for 10 min. Then, it will be left to complete dripping (10-15 min), transferred to a separate plastic bag and examined at zero time then stored at 0 ±1°C and examined every 3 days till spoilage.

2.3.2. Skin samples preparation

Skin sample was prepared by maceration technique as recommended by ICMSF (1978). Skin samples were separately taken from breast and thigh regions for periodical examination.

2.4. Protocol of samples preservation and examination

Samples were stored at 0 ± 1ºC, weighted and analyzed at the zero time and every three days till spoilage signs appeared.

2.5. Sensory evaluation

Sensory evaluation was carried out according to Economou et al. (2009). Acceptability as a composite of appearance, odor and flavor were estimated using nine point hedonic scale. A total of five panelist were asked to evaluate chicken breast and thigh samples (samples were boiled in water at 100°C for 15 minutes in separated area from testing area to avoid odors influencing panelist`s rating, and served warm to panelists).

2.6. Microbiological analysis

2.6.1. Aerobic Plate Count (APC)

The method was recommended by Maturin and Peeler, (2001). Ten grams of each sample was added to 90 ml of sterile peptone water to prepare decimal dilutions of 10^{-2}, 10^{-3}, 10^{-4}, and others as appropriate. Plate count agar medium was used and was promptly incubated for 48 ±2h at 35°C.

2.6.2. Enumeration of coliforms

Violet red bile agar medium (VRBA) was prepared. Counted purple-red colony was surrounded by zone of precipitated bile acids (Feng et al., 2002).

2.6.3. Enumeration of E. coli

One mL of initial dilution (10^{-1}) was used. The inoculum mixed with the Tryptone Bile X-glucuronide agar (TBX) medium to obtain evenly dispersed colonies. Inoculated plates were incubated at 37 ± 1°C for 4 ± 1 h and then at 44 ± 1°C for 21 ± 3 h. Typical colonies are blue. *E. coli* is ß-glucuronidase-positive, so grow on TBX agar at 44°C with production of typical blue colonies (ISO, 2001).

2.6.4. Enumeration of staphylococci

One hundred microliters from each dilution were aseptically transferred to the surface of duplicate sets Baird-Parker medium and spread by a sterile glass spreader. Inoculated plates were incubated at 37°C for 24 h. Suspected colonies were black in color. Plates re-incubated at 37°C for another 24 h and repeated count. *Staphylococcus* spp. count per cm^2 was calculated (APHA, 1992).

3. Results and discussion

3.1. Sensory findings

Grade of sensory acceptability for all samples (control and treated by sumac/rosemary) indicated that 60%-80% of samples from both breast and thigh were ranged between very good and excellent. The current findings revealed that there were no changes in color or taste detected by panelists (data not shown). Herbal extracts extended the shelf-life of broiler meat for three to six days more than control. Similar results were obtained by Glumez et al. (2006) and Zakariene et al. (2015). Vatansever et al. (2008) found that sumac-treated samples had natural, acceptable color and odor. Moreover, Ntzimani et al. (2010) reported that the presence of rosemary oil in cooked produced a distinct but acceptable pleasant odor and taste, well received by the panelists. From the present data, it could be concluded that sumac and rosemary extracts did not change the organoleptic properties of broiler meat.

3.2. Microbiological findings

Table (1) showed APC of sumac or rosemary-treated samples indicating that 8% sumac extract was more effective than the lower concentration (4 %) with a reduced total bacterial count one to two log less than the control. Similar findings were reported by Gulmez et al. (2006) and Vatansever et al. (2008). Such results might be attributed to the antimicrobial content of sumac. This held the view of Nasar-Abbas and Halkman (2004). In this respect,

Wetheritlt and Pala (1994) stated that various tanniniferous plants, including sumac *Rhus coriaria* L., have been known to contain naturally occurring compounds with antimicrobial activities. Concerning APC of rosemary treated samples; it was revealed that 0.5% rosemary extract was more effective than 0.3% of the same extract. This might be due to rosemary extract antimicrobial effects of phenolic components such as carvacrol, thymol, p-cymene, and γ-terpinene. This agrees with Skandamis and Nychas (2001) and Ntzimani et al. (2011). Meanwhile, Senter et al. (2000**)** considered the limit of microbiological acceptability for poultry products is of 10^7 cfu/g . Moreover, ICMSF (1978) reported that the upper limit for microbiological acceptability is 10^7 cfu/g in food (extension of chilled broiler shelf-life).

Table 1. Statistical analytical of APC of broiler's skin treated by different herbal extracts at different chilling (0±1 ºC) periods.

Extract	Zero day		3rd day		6th day		9th day		12th day		15th day	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated
	Mean ± S. E.											
Breast												
Sumac 4%	$3X10^6 \pm 2X10^6$	$9X10^5 \pm 1X10^6$	$5X10^6 \pm 2X10^6$	$1X10^6 \pm 1X10^6$	$6X10^6 \pm 2X10^6$	$2X10^6 \pm 1X10^6$	$2X10^7 \pm 1X10^7$	$3X10^6$ * $\pm 2X10^6$	-	$5X10^6 \pm 2X10^6$	-	$6X10^6 \pm 2X10^6$
Sumac 8%	$5X10^5 \pm 1X10^5$	$2X10^4$ ** $\pm 8X10^3$	$2X10^7 \pm 1X10^7$	$4X10^4$ *** $\pm 3X10^3$	$3X10^7 \pm 2X10^7$	$5X10^4$ *** $\pm 2X10^3$	$3X10^8 \pm 2X10^8$	$5X10^5$ *** $\pm 7X10^3$	-	$7X10^5 \pm 1X10^5$	-	$3X10^6 \pm 1X10^6$
Rosemary 0.3%	$5X10^5 \pm 2X10^5$	$4X10^5 \pm 1X10^5$	$3X10^7 \pm 2X10^7$	$3X10^6$ ** $\pm 2X10^6$	$4X10^7 \pm 3X10^7$	$4X10^6$ ** $\pm 2X10^6$	$5X10^7 \pm X10^7$	$5X10^6$ ** $\pm 2X10^6$	-	$7X10^6 \pm 3X10^6$	-	-
Rosemary 0.5%	$3X10^5 \pm 9X10^4$	$5X10^4$ * $\pm 2X10^4$	$4X10^6 \pm 2X10^6$	$3X10^5$ *** $\pm 3X10^4$	$6X10^6 \pm 3X10^6$	$6X10^5$ ** $\pm 2X10^4$	$5X10^7 \pm 2X10^7$	$8X10^5$ *** $\pm 2X10^5$	-	$1X10^6 \pm 3X10^5$	-	$4X10^6 \pm 9X10^5$
Thigh												
Sumac 4%	$3X10^6 \pm 2X10^6$	$1X10^6 \pm 1X10^6$	$1X10^7 \pm 4X10^6$	$2X10^6 \pm 1X10^6$	$2X10^7 \pm 7X10^6$	$3X10^6 \pm 2X10^6$	$4X10^7 \pm 1X10^7$	$4X10^6$ * $\pm 2X10^6$	-	$5X10^6 \pm 2X10^6$	-	$7X10^6 \pm 2X10^6$
Sumac 8%	$3X10^6 \pm 1X10^6$	$4X10^4$ *** $\pm 3X10^3$	$4X10^7 \pm 2X10^7$	$6X10^4$ *** $\pm 5X10^3$	$5X10^7 \pm 2X10^7$	$1X10^5$ *** $\pm 5X10^4$	$1X10^8 \pm 8X10^7$	$6X10^5$ *** $\pm 3X10^4$	-	$1X10^6 \pm 4X10^5$	-	$5X10^6 \pm 2X10^6$
Rosemary 0.3%	$7X10^5 \pm 9X10^4$	$6X10^5 \pm 9X10^4$	$3X10^7 \pm 2X10^7$	$3X10^6$ ** $\pm 2X10^6$	$4X10^7 \pm 3X10^7$	$4X10^6$ ** $\pm 2X10^6$	$8X10^7 \pm 6X10^7$	$5X10^6$ ** $\pm 2X10^6$	-	$1X10^7 \pm 5X10^6$	-	-
Rosemary 0.5%	$3X10^5 \pm 1X10^5$	$6X10^4$ ** $\pm 1X10^4$	$5X10^6 \pm 2X10^6$	$4X10^5$ ** $\pm 1X10^5$	$1X10^7 \pm 5X10^6$	$8X10^5$ ** $\pm 2X10^5$	$3X10^8 \pm 1X10^8$	$1X10^6$ *** $\pm 5X10^5$	-	$3X10^6 \pm s1X10^6$	-	$5X10^6 \pm 1X10^6$

(-): spoiled sample, * $P \leq 0.5$, ** $P \leq 0.05$, *** $P \leq 0.005$

Dealing with coliform count of rosemary or sumac extract treated samples, it was indicated that sumac extract (8% w/v) and rosemary extract (0. 5% w/v) significantly reduced the count of coliforms in treated broiler meat less than control (Table 2). Glumez et al. (2006), Vatansever et al. (2008) and Mastromatteo et al. (2009) reported similar findings. High significant differences were detected by

Dickens and Ingram (2001). Low or no significant differences were demonstrated by Machado de Melo et al. (2012) attributing to the higher phenolic content of extracts. More or less parallel findings obtained by Bursal and Koksal (2011), Machado de Melo et al. (2012) and Aliakbarlu et al. (2014). However, Quintavalla and Vicini (2002) stated that a decreased microbial growth rate might be due to the direct inactivation by contact between active agents and microorganisms. Furthermore, Gulmez et al. (2006) stated that sumac exerted an antibacterial effect against coliforms and presumptive fecal coliforms, playing an important role in decontamination and protected poultry carcasses against spoilage and pathogenic populations. From the present data, it could be summarized that sumac and rosemary extracts may act as a source of food preservative.

Table 2. Statistical analytical of coliforms of broiler's skin treated by different herbal extracts at different chilling (0±1 ºC) periods.

Extract	Zero day		3rd day		6th day		9th day		12th day		15th day	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated
	Mean ± S. E.											
Breast												
Sumac 4%	$2X10^3 \pm 1X10^3$	$2X10^3 \pm 1X10^3$	$6X10^3 \pm 6X10^2$	$5X10^3 \pm 1X10^3$	$9X10^4 \pm 3X10^3$	$1X10^4 \pm 1X10^4$	$1X10^5 \pm 4X10^4$	$2X10^4 \pm 1X10^4$	-	$2X10^4 \pm 1X10^4$	-	$6X10^4 \pm 3X10^4$
Sumac 8%	$3X10^3 \pm 2X10^3$	$2X10^2$ *** $\pm 3X10^1$	$6X10^3 \pm 6X10^2$	$1X10^3 \pm X10^2$	$6X10^4 \pm 2X10^4$	$2X10^3$ ** $\pm 8X10^2$	$8X10^4 \pm 1X10^4$	$9X10^3 \pm 8X10^3$	-	$1X10^4 \pm 9X10^3$	-	$3X10^4 \pm 3X10^4$
Rosemary 0.3%	$2X10^3 \pm 1X10^3$	$6X10^2$ * $\pm 1X10^2$	$5X10^3 \pm 2X10^3$	$2X10^3 \pm 6X10^2$	$8X10^4 \pm 3X10^3$	$6X10^4 \pm 1X10^4$	$1X10^5 \pm 3X10^3$	$7X10^4 \pm 9X10^3$	-	$1X10^5 \pm 4X10^4$	-	-
Rosemary 0.5%	$5X10^3 \pm 2X10^3$	$4X10^2$ ** $\pm 1X10^2$	$8X10^3 \pm 6X10^2$	$3X10^3 \pm 2X10^3$	$9X10^4 \pm 3X10^2$	$3X10^4$ * $\pm 2X10^4$	$1X10^5 \pm 4X10^4$	$4X10^4 \pm 3X10^4$	-	$5X10^4 \pm 3X10^4$	-	$7X10^4 \pm 4X10^4$
Thigh												
Sumac 4%	$3X10^3 \pm 2X10^3$	$2X10^3 \pm 1X10^3$	$9X10^3 \pm 5X10^2$	$5X10^3$ * $\pm 2X10^3$	$9X10^4 \pm 3X10^3$	$3X10^4$ * $\pm 2X10^4$	$2X10^5 \pm 7X10^4$	$4X10^4 \pm 2X10^4$	-	$6X10^4 \pm 2X10^4$	-	$9X10^4 \pm 2X10^4$
Sumac 8%	$8X10^2 \pm 1X10^2$	$5X10^2 \pm 6X10^1$	$6X10^3 \pm 7X10^2$	$1X10^3$ ** $\pm 4X10^2$	$8X10^4 \pm 9X10^3$	$6X10^3$ ** $\pm 5X10^3$	$9X10^4 \pm 1X10^4$	$1X10^4$ ** $\pm 1X10^4$	-	$3X10^4 \pm 2X10^4$	-	$4X10^4 \pm 3X10^4$
Rosemary 0.3%	$7X10^2 \pm 7X10^1$	$4X10^2 \pm 2X10^2$	$2X10^4 \pm 2X10^4$	$6X10^2$ *** $\pm 1X10^2$	$9X10^4 \pm 2X10^3$	$7X10^4$ * $\pm 1X10^4$	$1X10^5 \pm 3X10^3$	$8X10^4 \pm 8X10^3$	-	$2X10^5 \pm 9X10^4$	-	-
Rosemary 0.5%	$3X10^3 \pm 2X10^3$	$7X10^2$ *** $\pm 6X10^1$	$6X10^3 \pm 2X10^3$	$1X10^3$ * $\pm 3X10^2$	$9X10^4 \pm 0$	$3X10^4 \pm 3X10^4$	$1X10^5 \pm 2X10^3$	$4X10^4$ ** $\pm 1X10^3$	-	$6X10^4 \pm 3X10^4$	-	$8X10^4 \pm 4X10^4$

(-): spoiled sample, * $P \leq 0.5$, ** $P \leq 0.05$, *** $P \leq 0.005$

Fig. 1 (a, b, c, d) explained that sumac extract (8% w/v) and rosemary extract (0.5% w/v) significantly reduced count of *E. coli* in broiler meat less than the control. Similar results were reported by Glumez et al. (2006), Vatansever et al. (2008) and Mastromatteo et al. (2009). High significant differences were reported by Dickens and Ingram (2001). The present findings might be due to antibacterial effects of extracts on *E. coli* (an index organism for poultry hygiene). This come in contact with Ntzimani et al. (2010) who mentioned that samples treated with rosemary and oregano oils

showed 1.0 log CFU/g decrease compared to control samples. Therefore, extracts have *E. coli*-inhibitory effect.

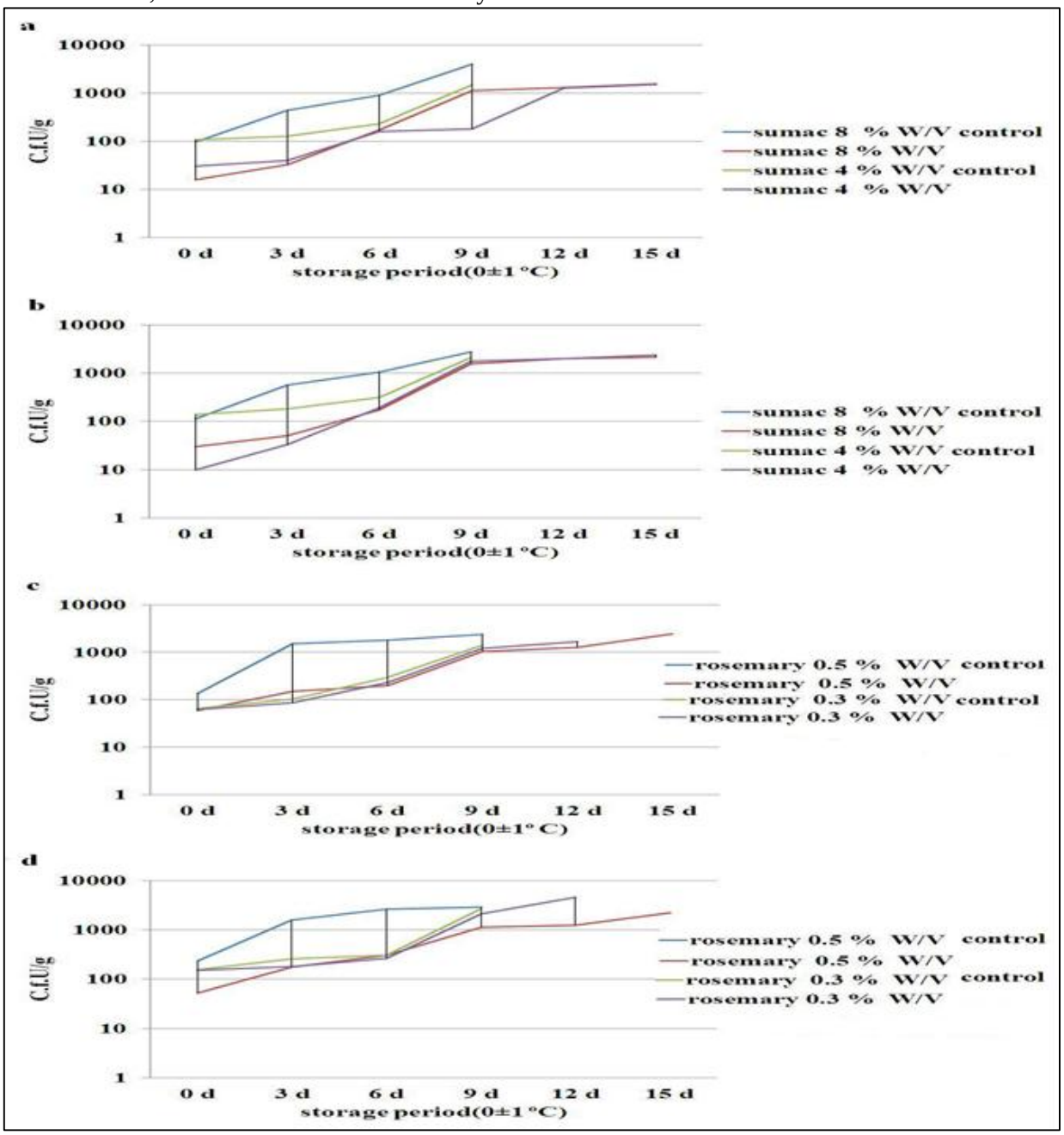

Fig. 1. *E. coli* count of broiler carcasses` skin treated by medicinal herbal extracts. a) *E. coli* count of broiler breast skin treated by sumac extract. b) *E. coli* count of broiler thigh skin treated by sumac extract. c) *E. coli* count of broiler breast skin treated by rosemary extract. d) *E. coli* count of broiler thigh skin treated by rosemary extract.

Staphylococcus spp. count of rosemary or sumac extract-treated samples revealed a significant growth inhibition (Table 3). Results referred to the use of rosemary extract might be attributed to the growth inhibition induced by its phenolic content (Witkowska et al., 2013). In this matter, Weerakkody et al. (2010) reported that ethanol or hexane extracts of oregano and rosemary strongly inhibited the growth of *S. aureus*. On the other hand, Nasar-Abbas and Halkman (2004) found a moderate

antibacterial activity for water extract of sumac against *Staph*. spp.

Table 3. Statistical analytical of *Staph*. spp. of broiler's skin treated by different herbal extracts at different chilling (0±1 °C) periods

Extract	Zero day		3rd day		6th day		9th day		12th day		15th day	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated	Control	Treated
	Mean ± S. E.											
Breast												
Sumac 4%	$4X10^4 \pm 2X10^4$	$3X10^3$ ** $\pm 1X10^3$	$5X10^4 \pm 2X10^4$	$5X10^3$ ** $\pm 1X10^3$	$8X10^4 \pm 4X10^3$	$3X10^4 \pm 2X10^4$	$9X104 \pm 7X10^3$	$4X10^4 \pm 1X10^4$	-	$5X10^4 \pm 1X10^4$	-	$7X10^4 \pm 1X10^4$
Sumac 8%	$1X10^5 \pm 1X10^5$	$1X10^2$ *** $\pm 1X10^3$	$2X10^5 \pm 1X10^5$	$6X10^2$ *** $\pm 2X10^2$	$3X10^5 \pm 2X10^5$	$5X10^3$ *** $\pm 1X10^3$	$4X10^5 \pm 2X10^5$	$2X10^4$ ** $\pm 7X10^3$	-	$5X10^4 \pm 5X10^3$	-	$8X10^4 \pm 3X10^3$
Rosemary 0.3%	$4X10^4 \pm 9X10^3$	$8X10^3$ ** $\pm 9X10^2$	$7X10^4 \pm 7X10^3$	$2X10^4 \pm 3X10^3$	$1X10^5 \pm 4X10^4$	$9X10^4 \pm 2X10^4$	$3X10^5 \pm 3X10^4$	$1X10^5 \pm 3X10^4$	-	$2X10^5 \pm 3X10^4$	-	-
Rosemary 0.5 %	$3X10^4 \pm 9X10^3$	$4X10^3$ * $\pm 1X10^3$	$7X10^4 \pm 6X10^3$	$1X10^4$ ** $\pm 7X10^3$	$9X10^4 \pm 6X10^3$	$5X10^4 \pm 2X10^4$	$2X10^5 \pm 6X10^4$	$1X10^5 \pm 3X10^4$	-	$2X10^5 \pm 3X10^4$	-	$3X10^5 \pm 2X10^4$
Thigh												
Sumac 4%	$5X10^4 \pm 2X10^4$	$5X10^3$ ** $\pm 9X10^2$	$7X10^4 \pm 2X10^4$	$7X10^3$ ** $\pm 1X10^3$	$1X10^5 \pm 2X10^4$	$4X10^4 \pm 2X10^4$	$2X10^5 \pm 3X10^4$	$5X10^4 \pm 1X10^4$	-	$1X10^5 \pm 4X10^4$	-	$2X10^5 \pm 4X10^4$
Sumac 8%	$2X10^5 \pm 1X10^5$	$6X10^2$ *** $\pm 6X10^1$	$3X10^5 \pm 2X10^5$	$1X10^3$ *** $\pm 1X10^2$	$5X10^5 \pm 4X10^5$	$1X10^4$ *** $\pm 3X10^3$	$7X10^5 \pm 6X10^5$	$4X10^4$ *** $\pm 5X10^3$	-	$6X10^4 \pm 6X10^3$	-	$7X10^4 \pm 6X10^3$
Rosemary 0.3%	$5X10^4 \pm 6X10^3$	$1X10^4 \pm 4X10^3$	$8X10^4 \pm 6X10^3$	$5X10^4 \pm 1X10^4$	$2X10^5 \pm 6X10^4$	$1X10^5 \pm 6X10^4$	$4X10^5 \pm 3X10^4$	$2X10^5 \pm 6X10^4$	-	$3X10^5 \pm 5X10^4$	-	-
Rosemary 0.5 %	$5X10^4 \pm 6X10^3$	$5X10^3$ ** $\pm 2X10^3$	$8X10^4 \pm 6X10^3$	$2X10^4 \pm 6X10^3$	$1X10^5 \pm 4X10^4$	$6X10^4 \pm 2X10^4$	$4X10^5 \pm 2X10^5$	$1X10^5$ * $\pm 3X10^4$	-	$2X10^5 \pm 4X10^4$	-	$3X10^5 \pm 5X10^4$

(-): spoiled sample, * $P \leq 0.5$, ** $P \leq 0.05$, *** $P \leq 0.005$

4. Conclusion

From the present study, it could be concluded that sumac and rosemary extracts did not change the organoleptic properties of broiler meat, reducing the total bacterial count one to two log less than control and had *E. coli* inhibitory effect. Both sumac extract (8% w/v) and rosemary extract (0.5% w/v) were effective as poultry meat decontaminant and preservative.

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