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Effect of *Satureja khuzestanica* Essential Oils on Postmortem pH and Antioxidative Potential of Breast Muscle from Heat Stressed Broiler Chicken

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ABSTRACT

The aim of this study was to evaluate the effect of *Satureja khuzestanica* essential oils on antioxidative potential of breast muscle from heat stressed broiler chicken. A total of 720 one dayold Arian broiler chicks broiler chicks were randomly assigned to 6 groups (6 replicates of 20 birds each) to evaluate the effects of administration of *Satureja khuzestanica* essential oils (SKEO) through drinking water on immune postmortem pH and antioxidative potential of breast muscle from heat stressed broiler chicken The birds were continuously received drinking water treated with Tween 80 (Cont+; 500 ppm) and SKEO at 0 (Cont-), 200, 300, 400 and 500 ppm as six experimental treatments. Breast muscle early as well as ultimate pH values at 42 days of age were non significantly lower for the birds received SKEO through drinking water at doses greater than 200 ppm. Addition of SKEO to drinking water at doses higher than 200 ppm significantly decreased the amount of TBARS values in breast muscle ($p < 0.05$). Breast muscle catalase activity in SKEO received groups were significantly higher than the control groups ($p < 0.05$). It was concluded that supplementation of drinking water with SKEO at dosed higher 200 ppm enhance the antioxidative potential of breast muscle from heat stressed broiler chicken.

Key words: Antioxidative potential, breast muscle, pH, *Satureja khuzestanica*

INTRODUCTION

High ambient temperature is a problem in many parts of the world. the lipid oxidation is a major problem in meat of the heat stressed broiler chicken. It can result in the production of off-flavours and odours, increased drip losses and lower consumer acceptability. Therefore, research was continued to enhance the quality of poultry meat fat by supplementation of poultry diets with copper (Pesti and Bakalli, 1996), tocopherols (Ponte *et al.*, 2008), green pastures (O'Sullivan *et al.*, 2004), dehydrated forage (Ponte *et al.*, 2008), bacteria (Salma *et al.*, 2007; Yang *et al.*, 2010), algal sources (Rymer *et al.*, 2010) and many other methods. Consumer interests in organic and natural poultry products, at the same time, encouraged the poultry nutritionists to consider phytogenic products for manipulation of poultry fat composition. Phytogenic products are, in addition, well known to have hypocholesterolemic effects and antioxidant potencies and they are able to increase poultry meat oxidative stability. Among many herbal spices or extracts examined, essential oils of onion and garlic (Sklan *et al.*, 1992; Konjufca *et al.*, 1997), thyme (Case *et al.*, 1995; Lee *et al.*, 2004a, b), turmeric (Honda *et al.*, 2006; Sugiharto *et al.*, 2011) and oregano (Brenes and Roura, 2010) exhibited superior hypocholesterolemic and antioxidative effects in chicken.

Satureja khuzistanica Jamzad, is a medical plant well-known for its remedial properties in traditional medicine. This plant is cultivated and produced in mass for commercial purposes by Lorestan Medicinal Plant Laboratory, Khorraman, Khorramabad, Iran, since 2006 (Hadian *et al.*, 2011). The aerial parts of the plant collectively contain up to 3% of essential oils which it spectacularly riches in carvacrol (up to 94%) (Khosravinia and Salehnia, 2012). Carvacrol was described as a phenolic, caustic and bitter tasting compound with good stability (Agricultural Research Service (ARS), 2002) which demonstrates significant antioxidant (Cuppett and Hall, 1998) properties. Accordingly, it has been reported that essential oils of *Satureja khuzistanica* (SkEO) has antioxidant effect (Abdollahi *et al.*, 2003; Radonic and Miles, 2003). This study was undertaken to examine the effect of *Satureja khuzistanica* essential oils (SkEO) on antioxidative potential of breast muscle from heat stressed broiler chicken.

MATERIALS AND METHODS

In this experimental, 720 one day-old Arian broiler chicks were obtained from a commercial hatchery and housed in a concrete floor, cross-ventilated windowless shed. Chopped barley stalks top dressed with 2 cm wood shavings were used as bedding material. The chicks were randomly assigned to 36 pens (100×180 cm) arranged in 6 rows (blocks/replicates) in parallel with the longitudinal wall of the shed at flocking density of 12 birds per m². Corn and soybean meal based super starter (24.28% CP and 2962 kcal ME/kg, 1 to 7 day), starter (21.15% CP and 2880 kcal ME/kg, 7 to 21 d), grower (18.82% CP and 2952 kcal ME/kg, 22 to 35 d) and finisher (17.63% CP and 2993 kcal ME/kg, 36 to 42 day) diets and water were provided for ad libitum consumption throughout the experimental period. The shed was equipped with wet pad-and-fan cooling system to decline the ambient temperature. Nonetheless, average temperature during day and night hours were ranged from 32 to 35 and 28 to 30°C during 21 to 42 days. Therefore, from 21 days of age the birds were exposed to seasonal extreme ambient temperatures. The effect of six experimental treatments consisted of supplementation of drinking water with 0 (control-), 200, 300, 400 and 500 ppm SkEO or 500 ppm Polysorbate-80 (control+) were examined in 6 replicates of 20 birds each. Polysorbate -80 is an emulsifier which it was used to disperse SkEO in water at 1:1 ratio (v/v). At the close of day 42 of age, one male from each treatment were randomly selected. The birds were weighed and then slaughtered and processed manually. A 10 g sample of chicken breast muscle (major pectoralis) After measuring the initial and final pH (24 h) preparation and was maintained -80°C temperature. During the test, the samples from freezing outside and manually with phosphate buffer 0.1 molar (with pH = 7.4) were homogenized with liquid nitrogen. Then centrifuged (10 min at 3000 rpm), the solids settle and the supernatant was removed for biochemical experiments. The solution to the biochemical test again at was maintained -80°C temperature.

TBARS analyses: The amount of lipid per oxidation was indicated by the content of thiobarbituric acid reactive substances (TBARS) in the breast muscle samples. Breast samples were thawed and manually homogenized in cold phosphate buffer (pH 7.4, containing 5 mM EDTA) and debris were removed by centrifugation at 3,500 g for 10 min. Supernatants were recovered and used for lipid peroxidation assessment. Tissue TBARS determined by following the production of thiobarbituric acid reactive substances as described previously (Subbarao *et al.*, 1990). Briefly, 40 µL of homogenate was added to 40 µL of 0.9% NaCl and 40 µL of deionized H₂O, resulting in a total reaction volume of 120 µL. The reaction was incubated at 37°C for 20 min and stopped by the

addition of 600 µL of cold 0.8 M hydrochloride acid, containing 12.5% trichloroacetic acid. Following the addition of 780 µL of 1% TBA, the reaction was boiled for 20 min and then cooled at 4°C for 1 h. In order to measure the amount of TBARS produced by the homogenate, the cooled reaction was spun at 1,500 g in a microcentrifuge for 20 min and the absorbance of the supernatant was spectrophotometrically read at 532 nm, using an extinction coefficient of 1.56 9 105 mol⁻¹ Cm. The blanks for all of the TBARS assays contained an additional 40 µL of 0.9% NaCl instead of homogenate as just described. TBARS results were expressed as mmol per milligram of tissue protein (mmol mg⁻¹ protein).

Statistical analysis: The statistical model used to analyze the collected data was:

$$Y_{ijk} = \mu + SkEO_i + B_k + e_{ijk}$$

where, Y_{ijk} is the dependent variable, μ is the general mean, $SkEO_i$ is the fixed effect of $SkEO$ ($i = 6$; control+and 0, 200, 300, 400, 500 ppm $SkEO$), B_k is the random effect of block ($j = 6$; 1, 2, 3, 4, 5 and 6) and e_{ijk} is the residual error. The data were analyzed using PROC MIXED of SAS 9.1 (SAS, 2003). The LSD test was used for multiple treatment comparisons using the LSMEANS statement of SAS 9.1 (SAS, 2003) with letter grouping obtained using the SAS pdmix 800 macro (Saxton, 1998). For the different statistical tests, significance was declared at $p = 0.05$. The REG procedure of SAS 9.1 was used to provide regression models for assessment of relation between $SkEO$ and water consumption.

RESULTS AND DISCUSSION

Breast muscle early as well as ultimate pH values at 42 days of age were non significantly lower for the birds received $SkEO$ through drinking water at doses greater than 200 ppm (Table 1). Addition of $SkEO$ into drinking water had significant effect on Weight breast of the birds ($p < 0.05$; Table 1). The mean of Weight breast for the birds received 400 ppm during 1 to 42 was greater than control group (Table 1). The continual incorporation of $SkEO$ in drinking water at levels greater than 200 µL L⁻¹ significantly decreased TBARS in breast muscle of broiler chickens at day 42 of age ($p < 0.05$; Fig. 1). breast muscle catalase activity in $SkEO$ received groups were significantly higher than the control groups ($p < 0.05$; Fig. 2). Lipid oxidation is a major problem in poultry meat due to the high content of polyunsaturated fatty acid (PUFA); thus, methods that are

Table 1: Effect of essential oils of *Satureja khuzistanica* (SKEO) on pH and weight breast (g) in broiler chicken up to 42 days of age

SKEO	Weight breast (g)	pH ₁₅	pH _u
CON-	424±17.484 ^{ab}	6.41±0.20 ^a	5.80±0.05 ^a
CON+	424±20.21 ^{ab}	6.14±0.19 ^a	5.77±0.06 ^a
200	395±25.38 ^b	6.24±0.16 ^a	5.81±0.04 ^a
300	426±23.57 ^{ab}	5.98±0.16 ^a	5.78±0.03 ^a
400	449±11.43 ^a	6.07±0.11 ^a	5.80±0.06 ^a
500	438±4.87 ^{ab}	6.05±0.14 ^a	5.76±0.04 ^a
SEM ³	7.57	0.07	0.02
$p < 0.05$	0.9751	0.4663	0.4496

¹Control+; The birds received drinking water supplemented with 500 ppm polysorbate-80 throughout the trial and Control-; The birds received drinking water with no additive, ²Standard error for overall mean, ^{ab}Means within a row without a common superscript differ significantly ($p < 0.05$)

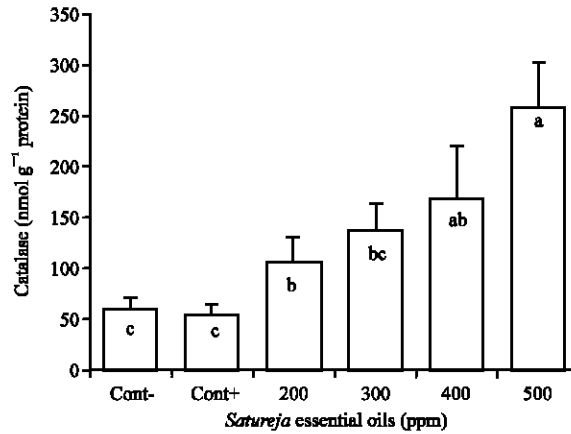


Fig. 1: Effect of *Satureja khuzestanica* essential oils on the amount of Thiobarbituric acid-reactive substances (TBARS) in breast muscle of broiler chicks at 42 days of age

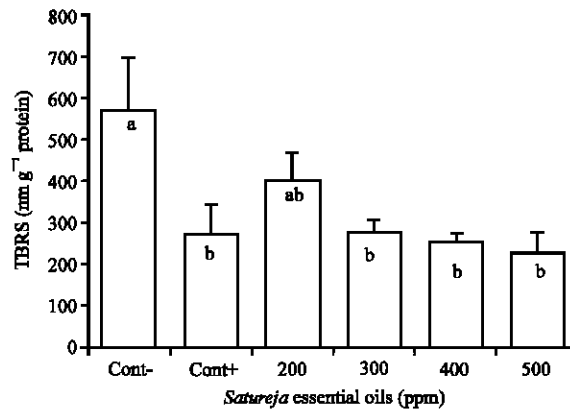


Fig. 2: Effect of *Satureja khuzestanica* essential oils on the breast muscle catalase activity of broiler chicks at 42 days of age

effective, safe and low cost for controlling poultry product stability are extremely important to the muscle food industry. To increase the storage stability of the processed meat, antioxidant compounds, such as butylated hydroxy toluene, butylated hydroxyl anisole and tocopherols, are being used. Recently, the use of natural dietary antioxidants has been advocated (Kang *et al.*, 1998, 2001) for stabilizing poultry feed and products. The essential oils from *Satureja khuzistanica* (SkEO) was described as a natural product riched in carvacrol so that almost all properties of these oils could be credited by carvacrol features.

TBARS measures malonaldehyde content in meat and is the most frequently used test to determine lipid oxidation. TBARS is positively related to the warmed-over flavor and hexanal content in meat (Shu *et al.*, 1995). In this experiment, administration of SkEO at doses >200 $\mu\text{L L}^{-1}$ into drinking water up to 42 days of age significantly decreased the TBARS in breast muscle. This effect can be attributed to the antioxidant properties of carvacrol (Abdollahi *et al.*, 2003, Abdollahi, M., A. Salehnia, S.H.R. Mort), as the major constituent in SkEO. Among many herbal spices or extracts examined, essential oils of onion and garlic (Sklan *et al.*, 1992;

Konjufca *et al.*, 1997), thyme Case *et al.* (1995), Lee *et al.* (2004a, b), turmeric (Honda *et al.*, 2006; Sugiharto *et al.*, 2011) and oregano (Brenes and Roura, 2010) exhibited superior hypocholesterolemic and antioxidative effects in chicken. Average catalase activity (per unit per mg protein) in breast muscle, was significantly increased for birds receiving SKEO ($p < 0.05$). Increased activity of this enzyme in muscle chest, had a clear linear relationship with the amount of SKEO in the water (Fig. 2). Thus, this results indicated the potential role of antioxidant SKEO. It has been shown that phenolic compounds present in the essential oils, increase the catalase activity, which in turn neutralizes hydrogen peroxide and converts lipid hydroperoxides to non-toxic material (Fki *et al.*, 2005). Therefore, finding of the present study, in concord with the conclusion made by Lee *et al.* (2003) and Ciftci *et al.* (2005) and Windisch *et al.* (2008) confirm the assumption that phytogetic extracts improve the antioxidative potential of breast muscle broiler chicken does not seem to be justified in general.

CONCLUSION

The results of this experiment led to the conclusion that the application of SkEO through drinking water may be feasible to enhance the lipid stability and antioxidative potential of breast muscle from heat stressed broiler chicken.

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