Effect of olive leaves extract on some physical and chemical traits of broiler meat during refrigeration storage

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Abstract:

This study was carried out in High Education lab., Animal Science Department, College of Agricultural Science, University of Sulaimani during June to July, 2016 to examine the effect of two concentration of natural olive leaves extract (OLE) 2 and 4% v/v, by application of two methods (spray and immersion) on some physical (water holding capacity and cooking loss), and chemical (thiobarbituric acid and free fatty acid) characteristics of broiler meat during refrigerate storage (4±1°C) for 0, 2, 5 and 7 days. The treatments included: T_1: (control), T_2: 2% OLE spray, T_3: 2% OLE immersion, T_4: 4% OLE spray and T_5: 4% OLE immersion. Water holding capacity (WHC) and cooking loss (CL) percentages showed significantly differences ($p \leq 0.01$) among treatments after 5 and 7 days storage, the highest WHC value (43.500%) recorded in T_5 after 5 days storage when sample was treated with 4% OLE emersion compared with control treatment which recorded the lowest WHC value (34.200%) after 7 days storage. For CL after 5 days storage T_4 recorded lower value (38.170%) compared to T_2 (42.920%), whereas after 7 days storage T_5 recorded lower CL value (42.380%) compared to T_1(48.015%) and T_2 (46.285%). Thiobarbituric acid (TBA) value decreased significantly ($p \leq 0.01$) with using of OLE after 7 days storage, and the highest value recorded in T_1 (1.410 mg malonaldehyde/kg). Also there were a positive effect of OLE on free fatty acid (FFA) value after 2, 5 and 7 days of storage , after 2 days storage T_5 recorded lest FFA value (0.381%), while the highest value recorded in T_1 (0.925%). Also after 5 and 7 days storage OLE treatments recorded lower FFA value compared to T_1 which recorded the highest value (1.150%) after 7 days storage.

These results suggested that using of olive leaves extract as natural antioxidant to improve broiler meat quality characteristics and extend shelf-life during refrigerate storage, which may have implications of meat processors.

Key words: Olive leaves; broiler meat; refrigeration storage, physical and chemical meat traits.

تأثير استخدام مستخلص اوراق الزيتون في بعض الصفات الفيزيائية والكيميائية للحوم الدجاج المخزونة بدرجة حرارة التبريد

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مدرس

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المستخلص:

أجريت هذه الدراسة في مختبر الدراسات العليا، قسم العلوم الحيوانية، كلية العلوم الزراعية، جامعة السليمانية خلال شهري حزيران وتموز2016 لتحديد تأثير تركيزين مختلفين لمستخلص اوراق الزيتون الطبيعي (2و4 % حجم/حجم) بتطبيق طريقتين (الرش برذاذ والغمر) في بعض الخصائص الفيزيائية (قابلية حمل الماء والفقد عند الطبخ) والكيميائية (حامض الثايوباربوتيرك والاحماض الدهنية الحرة) للحم الدجاج المخزونة بدرجة حرارة التبريد (4±1°م) لمدة 0 ، 2، 5 و 7 ايام.تضمنت المعاملات: المعاملة الاولى (السيطرة)، المعاملة الثانية: اللحوم المعاملة برذاذ مستخلص اوراق الزيتون بتركيز 2%، المعاملة الثالثة: اللحوم المغمورة بمستخلص اوراق الزيتون بتركيز 2%، المعاملة الرابعة: اللحوم المعاملة برذاذ مستخلص اوراق الزيتون بتركيز 4% و المعاملة الخامسة: اللحوم المغمورة بمستخلص اوراق الزيتون بتركيز 4%. اظهرت قابلية حمل الماء والفقد عند الطبخ فروقات معنوية (P≤0.01) مابين المعاملات بعد فترة خزن 5 و 7 ايام، اعلى قيمة لقابلية حمل الماء (43,500 %) سجلت في المعاملة الخامسة بعد 5 ايام من التخزين، بينما سجلت اقل قيمة (34,200%) في المعاملة الاولى بعد فترة 7 ايام من الخزن. اما لنسبة الفقد عند الطبخ، بعد فترة الخزن لمدة 5 ايام اظهرت المعاملة الرابعة اقل قيمة (38,170 %) مقارنة بالمعاملة الثانية (42,920 %)، لكن بعد فترة خزن 7 ايام سجلت المعاملة الخامسة اقل قيمة للفقد عند الطبخ (42,380 %) مقارنة بالمعاملة الاولى (48,015 %) والمعاملة الثانية (46,285 %). انخفضت قيم حامض الثايوباربوتيرك معنوياً(P≤0.01) مع استخدام مستخلص اوراق الزيتون بعد فترة خزن 7 ايام، حيث سجلت اعلى قيمة في المعاملة الاولى(1,410 ملغم مالونالديهايد/ كغم). لوحظت ايضاً تأثير ايجابي لأستخدام مستخلص اوراق الزيتون في قيم الاحماض الدهنية الحرة بعد فترات خزن 2، 5 و 7 ايام، بعد فترة خزن 2 يوم اظهرت المعاملة الخامسة اقل قيمة (0,381%)، بينما اعلى قيمة سجلت في المعاملة الاولى (0,925 %)، لوحظت ايضاً بعد فترة خزن 5 و 7 ايام اللحوم المعاملة بمستخلص اوراق الزيتون سجلت اقل قيم لنسبة الاحماض الدهنية الحرة مقارنةً بالمعاملة الاولى والتي سجلت اعلى قيمة (1,150 %) بعد فترة خزن 7 ايام.

من النتائج يمكن الاقتراح بأمكانية استخدام مستخلص اوراق الزيتون كمضاد اكسدة طبيعي لتحسين الصفات النوعية للحوم الدجاج . واطالة فترة خزنها بالتبريد والتي يمكن استخدامه بعمليات تصنيع اللحوم.

Introduction:

Poultry meat proves to be fits consumer demand as essential part of non – vegetarian diet for its rich nutrients source, low fat content with a high unsaturation fatty acids degree and low levels of sodium and cholesterol. Poultry meat also provide bioactive substances which has useful effects on consumer health, like conjugated linoleic acid (CLA), vitamins, antioxidants and a balanced ratio of poly unsaturated fatty acids, therefore some time it considered as "functional foods" (4, 9).

Meat quality is the key criterion of meat and meat product evaluation, which susceptible to quality deterioration and shelf life impact directly on quality changes, a rapid quality deterioration is observed in meat stored improperly (6). This may result in adversely changes in meat sensory and spoilage unacceptable to consumers. The short shelf life of refrigerate stored poultry meat is due to its composition such as microflora, pigment and unsaturated lipid contents (19). Compared to red meat, poultry meat is characterized by a higher content of unsaturated fatty acid (such as arachidonic and linoleic acid) that are especially susceptible to oxidation processes, as well as by the presence of specific micro-organisms which may freely proliferate under typical cold storage conditions (4^{o}C) (13).

The production of healthy food is an integrated primary goal, which contributes to the health and consumer well-being, which leads to developing numerous methods for preservation meat against lipid oxidation. Growing consumers concerns about synthetic antioxidant because of their safety and potential toxicology have pressed the food industry to find alternative natural sources of phenolic antioxidants (22, 15, 14, 5). Generally, Olive leaves extracts are a good source of several natural antioxidants including Oleuropeoside compounds such as Oleuropein and Verbascoside, and flavonoid compounds such as Luteolin, Luteolin-7-glucoside, apigenin-7-glucoside, diosmetin, diosmetin-7-glucoside, rutin and catechin, and simple phenolic compounds such as tyrosol, hydroxyl tyrosol,vanillin, vanillic acid and caffeic acid (12).

The aim of this study is investigate the effect of concentrations of 2 and 4%of olive leaf extract in two methods (immersion and spray) during refrigerate periods (0, 2, 5 and 7 days) on some physio-chemical qualities of broiler meat.

Material and methods:

Broiler chicken was purchased from the local market, after slaughtered the breast had cut and removed from the carcasses, cutting into approximately 2×2 cm slices. According to method of Skerget et al. in 2005 (22), 20 gm of dried olive leaves powder was extracted with 400 ml of 70% (v/v) ethanol for 2 hr at 40°C, then centrifuged

at 5000 rpm for 15 min., ethanol was evaporated by a rotary evaporation at 40°C. The remaining aqueous solution was dried in air oven at 40°C and the concentrations of 2 and 4% v/v were prepared. Meat samples were dipped and sprayed with pre-chilled OLE solutions up to 24 hr, meat slices were divided into five treatments (T1: control, T2: 2%OLE spraying, T3: 2% OLE immersion, T4: 4% OLE spraying and T5: 4% OLE immersion). The portions were kept under refrigeration condition (4±1°C) for 0, 2, 5 and 7 days, evaluated for some physical and chemical evaluation.

Water holding capacity (WHC)

Water holding capacity (WHC) was determined according to Wardlaw *et al.* in 1973 (24). 20gm of minced muscle sample was placed in centrifuge tube containing 30ml of 0.6M NaCl and was stirred with glass rod for 1 min. The tube was kept at refrigeration temperature (4˚C) for 15 min, stirred again and centrifuged at 2806.1 xg (4˚C) for 15 min. The supernatant was measured and amount of water retained by samples and expressed in percentage. The WHC was reported as ml of 0.6 M NaCl per 100g of muscle according to the following formula:

$$\text{WHC \%} = \frac{\text{initial solution weight - final solution weight}}{\text{sample weight (gm)}} \times 100$$

Cooking loss

Cooking loss was determined according to Murphy and Zerby in 2004(18). Muscle samples (20gm) were placed in an open aluminum boxes and cooked for 8.5 min in oven pre-heated to 176˚C to an internal temperature of 70˚C. After cooking, the samples were dried with a paper towel. Each sample was cooled for 30 min, cooking weight was measured. The cooking loss was calculated by the following formula:

$$\text{Cooking loss\%} = \frac{\text{Raw sample weight – cooked sample weight}}{\text{Raw sample weight (gm)}} \times 100$$

Thiobarbituric acid (TBA) value

The TBA values were determined according to the method described by Witte *et al*. in 1970(25). Twenty grams of the muscle were blended with 50ml of cold solution containing 20% trichloroacetic acid (TCA) in 2M phosphoric acid. The resulting slurry was transferred quantitatively to a 100ml volumetric flask with 40ml distilled water. The sample was diluted to 100ml with distilled water and homogenized by shaking. A 50ml portion was filtered through Whatman No.1 filter paper. Five ml of filtrate was transferred to a test tube followed by 5ml of fresh thiobarbituric acid (TBA) (0.005M in distilled water). The blank was prepared by mixting 5ml of distilled water with 5ml of TBA. The tubes stoppered and the solution mixed and kept in the dark for 15-17 hr at room temperature to develop the colour reaction. The absorbance was read at 530 nm by using spectrophotometer (Shimdzu, Japan). The TBA value was expressed as mg malonaldehyde (MDA)/kg muscle, and calculated by multiplying the absorbance (A) by 5.2 factor as follows:

TBA value (mg MDA/kg muscle) = $A_{530} \times 5.2$

Free fatty acids (FFA)

FFA was analyzed as method described by Egan et al in 1981(8). 100 gm of homogenized with 250 ml of chloroform, blend the mixture for 2-3 min. and filter it immediately through a large filter paper. Then re-filter it through a paper containing a small amount of anhydrous sodium sulphate, twenty five ml of 95% ethanol neutralized with drops of 0.1 N NaOH after adding phenolphthalein. The solution was added to 25 ml of the filtered above and the mixture tittered with 0.1 N NaOH until the pink colour persists for 15 seconds. The FFA calculates as oleic acid as percentage of the sample.

Statistical analysis

The statistical analysis system (SAS Institute, 2010). General linear model (GLM) with SAS program(21), Factorial Complete Randomized Design (CRD) was used to study the effect of treatments on studied traits. Duncans multiple range test (7) was used to determine significant differences among means.

Results and discussion:

Water holding capacity (WHC)

Table (1) showed significant differences ($p \leq 0.01$) among all the treatments during periods of storage in WHC. T_1 was differ significantly ($p \leq 0.01$) compared with each of T_2 , T_3, T_4 and T_5 at 5 and 7 days storage. After 5 days storage, T_5 was differ significantly ($p \leq 0.01$) compared to T_2. The results showed at storage after 5 days that T_5 the highest ability of WHC (43.500%), while T_1 appeared the lowest ability of WHC (34.200%) after 7 days storage.

These results may be probably due to that meat samples treated with 2% and 4% OLE protected the declined in the pH, binding more water molecules through hydrogen and ionic bonding to the large number of hydrophilic sites of meat polypeptides with pH increasing (11).

There were a significant differences ($p \leq 0.01$) among periods for each treatment , all treatments showed a gradual decrease in WHC as storage time was progressed.

Table 1: Effect of different concentration of olive leaves extract on WHC percentage of broiler breast meat during refrigerate storage (Mean±S.E).

Treat.	Storage period (days)			
	0	2	5	7
T_1 (Control)	^{a} 50.750±0.450 ^{A}	^{a} 47.200±0.898 ^{B}	^{a} 38.600±0.408 ^{C}	^{a} 34.200±0.816 ^{D}
T_2 (2% OLE spray)	^{a} 49.600±0.490 ^{A}	^{a} 45.850±1.102 ^{B}	^{b} 41.150±0.857 ^{C}	^{b} 38.600±0.245 ^{D}
T_3 (2% OLE emersion)	^{a} 50.250±0.857 ^{A}	^{a} 45.550±0.776 ^{B}	^{bc} 41.950±0.613 ^{C}	^{b} 37.650±0.449 ^{D}
T_4 (4% OLE spray)	^{a} 50.250±0.040 ^{A}	^{a} 46.450±0.613 ^{B}	^{bc} 43.300±0.327 ^{C}	^{b} 39.400±0.245 ^{D}
T_5 (4% OLE emersion)	^{a} 48.700±0.408 ^{A}	^{a} 47.650±0.367 ^{A}	^{c} 43.500±0.245 ^{B}	^{b} 39.550±0.286 ^{C}

-Means having different small letters (abc) among treatments for each period (column) are significantly different ($p \leq 0.01$).

-Means having different capital letters (ABCD) among periods for each treatment (row) are significantly different ($p \leq 0.01$).

Cooking loss:

The results presented in Table (2) showed significant differences ($p \leq 0.01$) between treatments after 5 and 7 days of storage. T_4 recorded significantly lower cooking loss value (38.170%) compared to T_2 (42.920%) at 5 days storage. Whereas T_5 recorded the lowest cooking loss value (42.380%) as compared with T_1 (48.015%) and T_2 (46.285%) after 7 days storage, this results similar to that obtained for water holding capacity (Table 1) and may be associated to the role of olive leaves extract in T_4 and T_5 as a natural antioxidant which bind with more water and increasing meat tissues ability to retain water, led to more water holding capacity with decreasing moisture loss during storage and cooking conditions (3), and to the phenolic compounds in plant extracts stabilized cell integrity and enhanced the ability of meat tissue to retain sarcoplasmic components, which resulted in less drip loss and more weight retention during storage (17).

There were also significant differences ($p \leq 0.01$) in cooking loss percentage in different periods for all treatments. The results showed increasing in cooking loss percentage with extending storage period, which are agreed with Wang in 2000(23). Also Al-Haju in 2005(1) was recorded that increasing in cooking loss associated with advancing in storage periods. Therefore in our study showed that the highest percentage of cooking loss recorded in T_1 as compared to other treatments.

Table 2: Effect of different concentration of olive leaves extract on Cooking loss percentage of broiler breast meat during refrigerate storage (Mean±S.E).

Treat.	Storage period (days)			
	0	2	5	7
T_1 (Control)	^{a}32.025±0.592 ^{A}	^{a}34.025±0.135 ^{A}	^{ab}41.845±0.061 ^{B}	^{a}48.015±0.763 ^{C}
T_2 (2% OLE spray)	^{a}33.170±0.841 ^{A}	^{a}36.580±2.344 ^{A}	^{bd}42.920±0.098 ^{B}	^{a}46.285±0.576 ^{B}
T_3 (2% OLE emersion)	^{a}33.260±0.229 ^{A}	^{a}38.002±1.972 ^{B}	^{ab}41.275±1.613 ^{B}	^{ab}45.955±0.200 ^{C}
T_4 (4% OLE spray)	^{a}33.880±0.196 ^{A}	^{a}36.080±1.861 ^{AB}	^{ac}38.170±0.743 ^{B}	^{ab}45.325±0.290 ^{C}
T_5 (4% OLE emersion)	^{a}33.240±0.188 ^{A}	^{a}37.055±0.510 ^{AB}	^{abc}39.305±0.020 ^{BC}	^{b}42.380±0.710 ^{C}

-Means having different small letters (abc) among treatments for each period (column) are significantly different ($p \leq 0.01$).

-Means having different capital letters (ABCD) among periods for each treatment (row) are significantly different ($p \leq 0.01$).

Thiobarbituric acid (TBA)

The results in Table (3) showed significant differences ($p \leq 0.01$) among treatments at the same periods in TBA value, samples treated with OLE T_2 T_3, T_4 and T_5 recorded significantly ($p \leq 0.01$) lower TBA value which were 0.999, 1.065, 0.912 and 0.908 mg MDA/kg muscle respectively when compared to T_1 (1.410) after 7 days storage period. This result agreed with that obtained on beef meat about TBA (10), which showed higher antioxidant activities were recorded with addition of olive leaves extract. This may be attributed to the antioxidant activity against lipid oxidation of phenolic compounds within olive leaves extract (14), It is assumed that inhibition of lipid oxidation and hydrogen donor ability is enhanced with the increasing amount of hydroxyl groups within phenolic structures present in olive leaves extract mainly Oleuropein and hydroxyl tyrosol(16).

TBA values of the same treatment increase with extending storage periods (Table 3).

Table 3: Effect of different concentration of olive leaves extract on TBA value (mg malonaldehyde MDA/kg) of broiler breast meat during refrigerate storage (Mean±S.E).

Treat.	Storage period (days)			
	0	2	5	7
T_1 (Control)	^{a}0.392±0.022 ^{A}	^{a}0.715±0.006 ^{B}	^{a}0.840±0.017 ^{B}	^{a}1.410±0.318 ^{C}
T_2 (2% OLE spray)	^{a}0.345±0.046 ^{A}	^{a}0.536±0.016 ^{AB}	^{a}0.720±0.003 ^{BC}	^{b}0.999±0.001 ^{C}
T_3 (2% OLE emersion)	^{a}0.315±0.014 ^{A}	^{a}0.663±0.018 ^{B}	^{a}0.747±0.298 ^{B}	^{b}1.065±0.028 ^{C}
T_4 (4% OLE spray)	^{a}0.388±0.018 ^{A}	^{a}0.562±0.009 ^{A}	^{a}0.614±0.002 ^{AB}	^{b}0.912±0.055 ^{B}
T_5 (4% OLE emersion)	^{a}0.345±0.019 ^{A}	^{a}0.535±0.018 ^{A}	^{a}0.615±0.002 ^{AB}	^{b}0.908±0.002 ^{B}

-Means having different small letters (abc) among treatments for each period (column) are significantly different ($p \leq 0.01$).

-Means having different capital letters (ABCD) among periods for each treatment (row) are significantly different ($p \leq 0.01$).

Free fatty acids (FFA)

Results presented in Table (4) showed significant differences ($p \leq 0.01$) among treatments in FFA percentage. After 2 days storage, T_5 recorded significantly lower FFA value (0.381%) compared to T_4 (0.450%), and all the treatments T_2, T_3, T_4 and T_5 showed significantly lower FFA value which were 0.445, 0.435, 0.450 and 0.381% respectively when compared to T_1 (0.925%). After 5 and 7 days storage the samples which treated with OLE T_2, T_3, T_4 and T_5 showed lower value of FFA as compared with T_1, which recorded the highest FFA value (1.150%) after 7 days storage. This result may be interrelating with lipid oxidation and have been proposed it has a pro-oxidant effect on lipids (20). Therefore, the lipolytic enzymes action from larger bacterial count on lipid leading to increase in free fatty acids releasing, which contribute to the generation of undesirable meat aroma and flavor (2).

FFA values were increase at storage period for each treatment, results after 7 days storage showed the highest value compared to other periods.

Table 4: Effect of different concentration of olive leaves extract on FFA value (%) of broiler breast meat during refrigerate storage (Mean±S.E).

Treat.	Storage period (days)			
	0	2	5	7
T_1 (Control)	$^{a}0.225\pm0.004^{A}$	$^{a}0.925\pm0.004^{B}$	$^{a}0.106\pm0.003^{C}$	$^{a}1.150\pm0.041^{D}$
T_2 (2% OLE spray)	$^{a}0.235\pm0.004^{A}$	$^{bc}0.445\pm0.004^{B}$	$^{b}0.711\pm0.001^{C}$	$^{b}0.986\pm0.004^{D}$
T_3 (2% OLE emersion)	$^{a}0.230\pm0.008^{A}$	$^{bc}0.435\pm0.004^{B}$	$^{c}0.515\pm0.004^{C}$	$^{b}0.995\pm0.003^{D}$
T_4 (4% OLE spray)	$^{a}0.231\pm0.001^{A}$	$^{b}0.450\pm0.008^{B}$	$^{c}0.486\pm0.359^{B}$	$^{b}0.995\pm0.005^{C}$
T_5 (4% OLE emersion)	$^{a}0.235\pm0.004^{A}$	$^{c}0.381\pm0.050^{B}$	$^{c}0.505\pm0.012^{C}$	$^{b}0.989\pm0.006^{D}$

-Means having different small letters (abc) among treatments for each period (column) are significantly different ($p \leq 0.01$).

-Means having different capital letters (ABCD) among periods for each treatment (row) are significantly different ($p \leq 0.01$).

Conclusion

From the results obtained in this study, it can be concluded that using of olive leaves extract at concentrations of 2% or 4% as natural antioxidant to improve quality characteristics and extended shelf-life of broiler meats during refrigerate storage, which may have implications for meat processors.

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