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Effect of olive leaf extract on physicochemical characteristics of Karadi ram meat during frozen storage

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Abstract

The objective of the present study was to investigate the effect of natural extract of olive leaves on physiochemical traits of Karadi ram meat stored at -18°C for 60 d. The active phenolic compounds in the olive leave were determined through chromatographic analysis. The meat slices were treated with 1, 3 and 5% olive leaves extract (OLE) and 0.01% butylatedhydroxy toluene (BHT) for 60 d. The results showed that meat samples treated with 3 and 5% OLE retained the highest ($P<0.05$) moisture and proteins contents and lowest ($P<0.05$) cholesterol concentration compared with control and 1% OLE treatment. Meat samples treated with 3 and 5% OLE had lower ($P<0.05$) cooking loss percentages and increased ($P<0.05$) water holding capacity (WHC). It was concluded that the Karadi meat maintained a better quality during cold storage for 60 d at low temperature when OLE was at added at the rate of 3 and 5% compared to BHT and control.

Keywords: Olive leaf extract; physicochemical; ram meat

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Introduction

Meat is one of the essential components of human diet. Sheep meat is an excellent source of high biological value of protein, vitamins B-complex, minerals and long-chain omega 3 polyunsaturated fatty acids (Lawrie, 2002). However, high degree of polyunsaturation accelerates the degree of oxidative process leading to deterioration in meat nutritional value in addition to flavour, texture and colour. Lipid peroxidation is a major cause of meat deterioration, resulting in discoloration and off flavour development (Gray et al., 1996; McCarthy et al., 2001; Brewer, 2007). Protein is one of the major constituent of meat which is highly susceptible to oxidative damage. Keeping in view the consumers concerns regarding safety and toxicity of synthetic antioxidants, natural sources of phenolic antioxidants and antimicrobial agents as alternative are highly recommended (Lee and Lee, 2010; Beal et al., 2011).

Nutritional treatment can be used to manipulate the stability of the meat. Many plants extracts rich in phenolic compounds have positive effect on prevention of lipid peroxidation. Plants extracts rich in phenol are good sources of preventing lipid oxidation in food products. Olive leaves are the agricultural by-product which contains polyphenol and tocopherol which have several biological activities (Paiva-Martins et al., 2009). Olive leaves have antioxidants and antimicrobial properties due to the phenolic compounds like soleuropein, tyrosol and hydroxyltyrosol (McDonald et al., 2001; Pereira et al., 2007).

The meat industry is searching for natural solution to minimise oxidative rancidity and extend the shelf-life of meat and meat products. The antioxidant activities of various plants have been recorded. Therefore, a study was designed to investigate the effect of the different concentrations of olive leaves extract and storage period on some of physiochemical characteristics of Karadi sheep meat during frozen storage at -18^{o}C for 60 days.

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Materials and Methods

Preparation and identification of active compounds of olive leaves extract

20 gram dried olive leaves powder was extracted with 400 ml 70% (v/v) ethanol for 2 h at 40°C by using shaker. The samples were 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 percent (w/w) extraction yields of plant materials were calculated. The crude extracts were kept in refrigerator (4ºC) in glass bottle (24 h) until use in the formulation. The active compounds of olive leaves extract were analyzed by high performance liquid chromatography (HPLC) according to the method described by Garcia et al. (2000).

Preparation of meat samples

The leg cuts were removed from Karadi carcass after 24 h of chilled storage at -18°C. The leg cuts were separated into lean meat, fat and bone. The external fat and heavy connective tissues were trimmed off from lean meat slices of leg cuts. The outer surfaces of streak were removed to avoid possible contamination before cutting into slices. The meat slices were randomly divided into five batches (2 kg each batch). One served as a control which was immersed into distilled water and three treatments were immersed into solutions containing 1, 3 and 5% olive leaf extract (OLE) (w/v). The fourth treatment was immersed into 95% ethanol solution containing 0.01% butylated hydroxyl toluene (BHT) (w/v) for 20 h at 4°C. After treatment, the samples were drained and divided into portions. The portions were placed on plastic foam meat trays, packed into polyethylene film and kept in a freezer at -18°C for 60 d. The physiochemical and properties of meat slices at 0, 30 and 60 d of storage time were evaluated.

Physiochemical analysis of meat samples

Moisture, crude protein and ash contents were determined by the method of proximate analysis (AOAC, 2000). The lipid contents were extracted as described by Folch et al. (1957). Water holding capacity (WHC) and pH was determined as described by Wardlaw et al. (1973) and Ibrahim et al. (2011) respectively. Cooking loss was determined according to the method of Murphy and Zerby (2004). In order to calculate the cooking loss, meat samples were weighed before and after the heat treatment. Cholesterol concentration was determined by the procedure of Rhee et al. (1982), using $FeCl_3^-$ ethylacetate and concentrated sulphuric acid as colour developing reagent.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) using SAS/STAT (2010). Duncan's multiple range test was used to find any significance difference between the mean values of each parameters (Duncan, 1955).

Results and Discussion

As a results of the HPLC chromatogram analysis of olive leaves extract three phenolic compounds isolated were Oleuropein, Olecanthal and total Tyrosol (Tyrosol + Hydroxytyrosol), and their concentration were 78.66, 4.43 and 2.92 mg/g of dried leaves respectively.

Chemical composition of meat samples

Changes of moisture, protein, fat and ash contents of meat slices among treatments during storage at -18^{o}C for 60 d are shown in Table 1. All treatments showed significant ($P<0.05$) decreased in moisture content as storage period progressed. Moisture contents increased significantly ($P<0.05$) in meat treated with 5% OLE compared with control and BHT. The loss of moisture at the end of the storage period may be due to the distortion of myofibril at cold storage that led to the poor water retention of the meat (Kandeepan and Biswas, 2007). The results showed that meat samples treated with 3 and 5% OLE have the highest protein content at any storage time compared with control and 1% OLE treatment. It was observed that the addition of 3 and 5% OLE to the meat samples exhibited a significant reduction in the fat content during frozen storage for 60 d as compared with control 1% OLE. It was observed that both 3 and 5% OLE had the lowest content of ash at the end of the storage period. These results agreed those of Ageena (2001) and Al-Dhaheri (2012) who recorded higher moisture and protein contents and lower fat of meat and minced beef during frozen storage as the period storage increased.

Cooking loss

During storage at -18^{o}C cooking loss percentages of meat samples in the control samples started to increase ($P<0.05$) rapidly after 60 d of storage (Fig. 1). The meat samples treated with 3 and 5% OLE recorded decrease ($P<0.05$) in cooking loss at the end of the storage as compared with control and 1% OLE. It is possible due to the role olive leaves extract with concentration 3 and 5% as natural antioxidant may be enough to bind water and increasing WHC which led to increase ability of meat tissues to retain water and decreasing moisture loss during storage and cooking (Arora et al., 2000). Previous researchers showed that sheep meat or beef meat treated with seed grape extract

Table 1: Changes in chemical composition of sheep meat slices treated with different concentrations of olive leaves extract and synthetic antioxidant (BHT) during storage at -18°C for 60 days

Trait (%)	Storage time (days)	Treatments				
		C	1% OLE	3% OLE	5% OLE	BHT
Moisture	0	69.10±0.06^{aA}	69.22±0.01^{aA}	71.25±0.03^{aA}	71.61±0.05^{aA}	69.13±0.01^{aA}
	30	65.14±0.07^{eB}	65.38±0.07^{dB}	67.35±0.07^{bB}	67.93±0.08^{aB}	66.03±0.03^{cB}
	60	63.46±0.20^{dC}	64.32±0.02^{cC}	66.17±0.02^{aC}	66.45±0.03^{aC}	64.63±0.05^{bC}
Protein	0	21.30±0.03^{aC}	21.72±0.04^{aC}	22.19±0.01^{aC}	22.47±0.01^{aC}	21.72±0.02^{aC}
	30	22.44±0.02^{eB}	22.61±0.03^{dB}	23.79±0.02^{aB}	23.69±0.03^{bB}	22.94±0.01^{cB}
	60	23.24±0.03^{eA}	23.45±0.02^{dA}	24.46±0.02^{bA}	24.66±0.02^{aA}	23.57±0.01^{cA}
Fat	0	6.44±0.04^{aC}	6.11±0.07^{bC}	4.12±0.04^{cC}	3.33±0.04^{dC}	6.10±0.06^{bC}
	30	8.95±0.07^{aB}	8.63±0.05^{bB}	6.10±0.04^{dB}	5.56±0.04^{eB}	7.54±0.04^{cB}
	60	9.54±0.08^{aA}	8.73±0.03^{bA}	6.56±0.03^{dA}	6.08±0.04^{eA}	8.15±0.03^{cA}
Ash	0	2.36±0.02^{aC}	2.18±0.02^{bC}	1.70±0.02^{cC}	1.71±0.02^{cC}	2.24±0.03^{bB}
	30	2.57±0.05^{abB}	2.47±0.04^{bB}	1.93±0.05^{cB}	1.85±0.04^{cB}	2.64±0.05^{aA}
	60	2.85±0.04^{aA}	2.69±0.01^{bA}	2.10±0.04^{cA}	2.03±0.05^{cA}	2.76±0.06^{abA}

Means having different small letters (within same row) and different capital letter (within same column) for each treatment are significant (P<0.05); OLE: olive leaf extract; BHT: Butylated hydroxytoluene

Table 2: Effect of different concentration of olive leaves extract and synthetic antioxidant (BHT) on pH values and water holding capacity (WHC) percentage of sheep meat slices during storage at -18°C for 60 days

Treatment	pH value ± S.E			WHC% ± S.E		
	Storage time (day)					
	0	30	60	0	30	60
C	5.89±0.01^{dA}	5.81±0.01^{bB}	5.56±0.01^{cC}	46.65±0.01^{dA}	39.58±0.01^{eB}	35.75±0.01^{eC}
1% OLE	5.91±0.01^{cA}	5.86±0.01^{bB}	5.59±0.01^{cC}	47.47±0.02^{cA}	40.68±0.01^{cB}	36.01±0.01^{cC}
3% OLE	6.15±0.01^{bA}	5.95±0.01^{aB}	5.69±0.01^{bC}	51.16±0.02^{bA}	47.80±0.02^{bB}	42.70±0.01^{bC}
5% OLE	6.21±0.01^{aA}	5.97±0.01^{aB}	5.75±0.01^{aC}	52.35±0.02^{aA}	48.05±0.04^{aB}	43.00±0.04^{aC}
BHT	5.90±0.01^{cA}	5.88±0.01^{bB}	5.58±0.01^{cC}	46.12±0.01^{eA}	40.27±0.02^{dB}	35.90±0.02^{dC}

Means having different small letters (within same column) and different capital letter (within same row) for each treatment are significant (P<0.05); OLE: olive leaf extract; BHT: Butylated hydroxytoluene; WHC: water holding capacity

(*Origanum majorano*) had less cooking loss percentages during frozen storage (Saleh, 2007; Al-Dhaheri, 2012). Mitsamoto et al. (1995) found that phenolic compounds in plant extracts stabilized cell integrity and enhanced the ability of meat tissue to retain sarcoplasmic components, which resulted in less drip and more weight retention during storage.

pH values and water holding capacity

Effect of different concentration of olive leaves extract and synthetic antioxidant (BHT) on pH values and water holding capacity of ram meat steaks stored at -18°C for 60 days are presented in Table 2. The pH of the control treatment was 5.56 at the end of the frozen storage for 60 days, whereas, pH values in meat samples treated with 3 and 5% OLE had significantly (P<0.05) higher pH at 60 days of frozen storage as compared with control samples. No significant difference appeared in pH values among meat samples treated with 1% OLE, BHT and non-treated control at the end of the frozen storage at -18°C for 60 days. Water holding capacity significantly (P<0.05) increased in all olive extract treated groups at 60 days of frozen storage at -18°C compared to non-treated control and BHT (Table 2). On the same day of frozen storage, WHC percentages in control samples and synthetic antioxidant (BHT) declined while, meat samples treated with 3 and 5% OLE had significantly (P<0.05) higher WHC percentage compared with control samples. On the other hand, all treatment showed a gradual decrease in WHC with the progressed of storage time. It was observed that meat samples treated with olive leaves extract in low concentration (1%) had lower WHC which may be due to protein loss (Offer and Trinick, 1983). Increase in pH value might be due to the large number of hydrophilic sites on meat protein, resulting in more binding of water molecules through hydrogen and ionic bonding to the hydrophilic sites of polypeptides (Hamm, 1977).

Cholesterol concentration

The results in Table 3 showed that the cholesterol concentration of the control samples gradually declined at 60 days of frozen storage. At the end of frozen storage period (day 60), meat samples treated with 3 and 5% OLE had significantly lower (P<0.05) cholesterol as compared with the control and other treatments. The results exhibited that meat slices treated with olive leaves extract at concentration of 3 and 5% OLE were more effective as a natural antioxidant than synthetic antioxidant (BHT). It is possible due to the role active phenolic compounds in olive leaves extract to protect cell membranes of meat. Therefore, low density lipoprotein stability of olive leaves extract and their phenolic compounds may be the result of their ability to scavenge free radicals produced from lipid

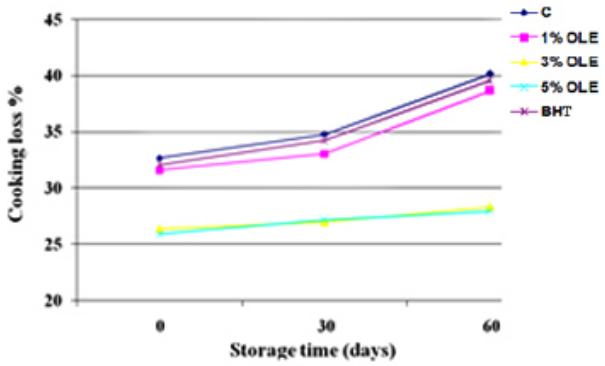

Fig. 1: Effect of different concentration of olive leaves extract and synthetic antioxidant (BHT) on cooking loss percentage of sheep meat stored at -18°C for 60 days

Table 3: Effect of different concentration of olive leaves extract and synthetic antioxidant (BHT) on cholesterol concentration of sheep meat slices during storage at -18°C for 60 days

Treatment	Cholesterol concentration (mg/100 tissue)		
	Storage time (day)		
	0	30	60
C	74.60±0.02^{aA}	73.30±0.03^{aA}	72.18±0.02^{aA}
1% OLE	73.74±0.02^{bA}	70.66±0.03^{bB}	68.56±0.02^{bC}
3% OLE	71.64±0.02^{eA}	67.57±0.01^{eB}	65.83±0.02^{eC}
5% OLE	72.16±0.02^{dA}	68.49±0.01^{dB}	66.39±0.01^{dC}
BHT	72.87±0.01^{cA}	69.18±0.03^{cB}	67.26±0.02^{cC}

Means having different small letters (within same column) and different capital letters (within same row) for each treatment are significant different (P<0.05); OLE: olive leaf extract; BHT: Butylated hydroxytoluene

oxidation, thereby, delaying lipid oxidation and reduced cholesterol concentration by providing hydroxyl groups (Shi et al., 2003). In the present study, the lack of an improvement in lipoprotein stability of meat steaks treated with low concentration (1% OLE) may imply that the antioxidant compounds present were not enough to retard lipid oxidation (Keceli and Gordon, 2002). Similar results were reported by King et al. (1998) who indicated that cholesterol concentration decreased as storage period increased. This probably is due to addition of spices that may contain phenolic compounds, which prevent lipid oxidation (Dugan, 1980).

Conclusion

It can be concluded that the Karadi meat treated with olive leaf extract at concentration of 3 and 5% are effective in maintaining the quality during frozen storage for 60 days.

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