PE4.64 Effects ewes dietary with thyme on lipid oxidation of cooked lamb meat 231.00

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Abstract— **The aim of this study was investigate if including thyme leaf in the diet of pregnant ewes has effect antioxidant on the subsequent cooked lamb meat. Thirty six Segureña sheeps were randomly assigned into three homogeneous groups. One group was fed a basal diet as control (*C*) while the diet of the other two groups was modified by substituting 3.7% (T_1) and 7.5% (T_2) of the control diet with thyme leaves (*TL*). *TBARS*, and sensory characteristics were analyzed on days 0, 2 and 4. The incorporation of thyme into the animal diet favoured the delay of lipid oxidation in cooked lamb meat samples. Cooked lamb meat from ewes fed thymus leaves showed a lower *TBARS*, rancid odour, rancid flavour and better sensorial acceptance scores. Statistically significant differences were not detected among the results obtained from the two ewe levels fed with thymus leaves.-**

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***Index Terms*— Cooked Lamb, *Thymus zygis*, lipid oxidation, sensory evaluation.**

I. INTRODUCTION

Lipid oxidation is recognized as a major quality problem by the meat industry and by consumers. Cooking procedures lead to a pronounced increase in lipid stability, since rupture of the compartmentalized cellular system by cooking facilitates the interaction of the highly unsaturated phospholipids with oxygen and catalysts [1], with subsequent formation of free radicals for propagation of oxidative reactions [2]. The short shelf-life of cooked lamb meat is one of the principal concerns for its marketing. The use of natural preservatives to increase the shelf life of meat products is a promising technology since many vegetal substances show antioxidant and antimicrobial properties. Herbs of the *Labiatae* family, particularly rosemary and thyme, which have been reported to possess substantial antioxidant activity, have been used for many centuries to improve the sensory characteristics and to extend the shelf-life of foods. In thyme the most important compounds in its essential oil (*EO*) are the phenols thymol (68.1%) and carvacrol (3.5%), which constitutes the major and more active constituents, as well as the monoterpene hydrocarbons *p*-cymene (11.2%) and γ-terpinene (4.8%) [3]. This phenolics and poly-phenolics compounds are known to have antioxidant properties, which allow them to act as reducing agents, hydrogen donors, singlet oxygen quenchers and also metal chelation properties [4]. The antioxidant effect of thyme has been showed by Tanabe *et al.* [5]. The possibility of feeding pregnant ewes diets containing thymus leaves as natural antioxidant represents a very interesting opportunity to replace synthetic antioxidant

II. MATERIALS AND METHODS

2.1. Animals and diet

Thirty six Segureña pregnant sheep were randomly assigned on the basis of their age and body condition into three homogeneous groups. Sheep in the control group were given a basal diet (*BD*) consisting of 1.3 Kg animal feed/day. The diet of the other two groups was modified by substituting 3.7%: T_1 (0.54% of Essential Oil) and 7.5%: T_2 (1.08% of Essential Oil) of the *BD* by thyme leaves (*TL*) using a pellet made from 75% barley and 25% thymus leaves. Animals given these diets were fed for 240 days, coinciding with the gestation and lactation periods in order to study the quality of the subsequent lamb meat. Sheep and lambs (9 animals per level: 27 in total) were reared at the CIFEA Research Center (Conserjería de Agricultura, Región de Murcia, Spain). The animals were weighed weekly until the lambs reached the slaughter weight of 25±2 kg. Then, the lambs were slaughtered according to Spanish regulations (RD 147/1993).

2.2. Sample preparation and experimental design

Twenty-four hours post-mortem, legs back was removed from both sides of the carcasses and vacuum packed and stored frozen (-20ºC). Legs were thawed at 4ºC overnight, cut into 1.5 cm fillets and then cooked between two heating plates (Silanos, Liscia Average, Lavastoviglie Industriali, Italy) at 160ºC for 3 minutes. The fillets reached a 72ºC internal temperature after 3 minutes, as measured by a portable T200 thermometer (Digitron Instrumentation Ltd., Merd Lane, Hertford, United Kingdom). Fillets were packaged in a transparent polystyrene tray B5-37 (Aerpack) and covered with an oxygen-permeable polypropylene film (650 cm^{-3} m^{-2} h^{-1} per 23h) with a commercial packer

(Raelma Industries Madrid Ltd., Spain). The fillets were stored at 4ºC for 0, 2 and 4 days in a display cabinet (Helkama, Finland) illuminated white fluorescent light (620 lux), simulating retail display conditions. All the measurements were made in triplicate.

2.3. *Thiobarbituric acid-reactive substances (TBARS)*

Lipid oxidation was determined at days 0, 2 and 4 of storage, and was expressed as thiobarbituric acid reactive substances (*TBARS)* (mg malonaldehyde/kg meat), as determined by Botsoglou *et al*. [6], using a UV2 spectrophotometer (Pye Unicam Ltd., Cambridge, United Kingdom).

2.4. *Sensory analysis*

A sensory analysis was made by a panel on cooked lamb meat. The panel was formed by eight persons chosen from the university community and trained according to ISO 8586-1 (1993). There were four training sessions. In the two first sessions, the colour, odour, and texture descriptors of cooked lamb meat were studied; the next two sessions were concerned with identifying, selecting and quantifying attributes to evaluate the meat. On day zero, two and four-day storage at 4ºC, cooked fillets were re-heated in an microwave, until the end point temperature inside the meat was 75-80ºC (re-heating). Sensory analysis was carried out according to ISO 4121 (2003). After cooking the samples were immediately covered with aluminium foil and they kept at 60ºC for 5 minutes maximum in a sand bath (Braun, Esplugues de Llobregat, Spain) and this temperature was maintained until it was taken for testing by the evaluation panel. Sensorial analysis was carried out according to ISO 4121 (2003). A 6-point scale was established. A linear scale of 1 (minimum intensity) to 6 (maximum) was used: 1 = non-perceivable; 2 = perceivable; 3 = slight; 4 = moderate; 5 = strong; 6 = strongly perceivable. The descriptors used were: *MC*, meaty colour; *RO*, rancid odour; *WO*, warmed-over odour; *RF*, rancid flavour; *WOF*, warmed-over flavour and *AC*, acceptance.

III. RESULTS AND DISCUSSION

3.1. *TBARS*

Table 1 shows the effects of the *TL* feeding and storage time on *TBARS* in cooked lamb meat. The *TBARS* increased with storage time significantly (P<0.05) in all the treatments. The highest values were recorded in *C* on day 21 (4.28), while *TBARS* evolved similarly in both T_1 and T_2 (3.49 and 3.8 mg MDA/kg meat respectively). This rapid accumulation of MDA in cooked samples has been showed in several earlier studies [7, 8]. There are significant differences (P <0.05) between the control and two treatments from day 2. Both T_1 and T_2 showed delayed MDA formation in cooked lamb meat stored until 4 days, emphasising the antioxidant effect of the thyme leaves. *TBARS* values for T_1 and T_2 at day 4 fell by 18 and 11% (if we compare with values obtained in *C*), respectively, as a result of *TL* dietary. The higher concentration of *TL* (7.5%) did not improve the results obtained with T_1.

The effectiveness of thyme leaves on lipid oxidation of cooked meat is due to phenolics compounds, that containing conjugated ring structures, hydroxyl group and stabilizes free radical; moreover phenolics compounds containing the carboxylic acid groups that inhibit lipid oxidation by metal chelation [9]. Direct comparison of our result with other studies is difficult since there have been limited reports on the antioxidant effects of thyme leaves *in vivo*. However, in the case of plants of the *Labiata* family, several studies has made done *in vivo*: Govaris *et al.* [10] reported that feeding with oregano oil reduced lipid oxidation in cooked turkey breast and thigh patties, compared with the control, and this effect was more effective than *post-mortem* addition. Botsoglou *et al.* [11] showed that the incorporation of dehydrated rosemary in turkey rations is an effective way to delay lipid oxidation in cooked meat during refrigerated storage.

In addition the antioxidant effect of thyme by *post-mortem* addition has been showed by Tanabe *et al*. [5]. For these authors a liquid extract of thyme incorporated on homogenized samples of porcine meat at levels ranging from 0.5% to 2.5 % w/w, showed a prevention of lipid oxidation.

3.2. *Sensory analysis*

Table 2 shows the effects of *TL* level and storage time on the sensory attributes of cooked lamb meat. The *MC* scores fell throughout storage, showing a significant (P <0.05) drop in all the treatments after day 2 of storage. At day 2 and 4 the scores of T_1 and T_2 are higher than *C* samples, showing significant differences (P <0.05) between C and T_1-T_2 only day 2. In contrast *WO* and *RO* increased during storage. *TL* samples showed lower *WO* on day 2 and 4 than *C*, so there is a slight tendency of *TL* diet to decrease the *WO* on cooked meat, but this decrease was not enough and not significant differences were found between treatments. However *RO* is affected by diet treatment. *T*here are significant differences between the two levels of *TL* in *RO* scores on day 2 and 4, showing T_2 the lowest *RO* value (1.94). As regards to flavour attributes, *RF* and *WOF* gradually rose during storage and significantly so after 2 days onwards on C and T_2, however in T_1, this increases was significant between day 0 and 2, but not between day 2 and 4. *WOF* development was more pronounced early in the storage period (day 0-2) than in the late part of storage (2-4), as also shown in other studies [12, 13]. The highest score in both attributes (*RO* & *WOF*) was found on *C* samples during all the storage. The increased in *WOF* on cooked meat during storage has been described in earlier studies. Tims & Watts [8] was first in described the term *WOF*, as an oxidized or rancid flavour that develops rapidly during the refrigerated storage of cooked meat and something is described as stale or rancid flavour. The cause of its formation is because, in meat cooked at 70-80ºC, as performed in this study, cooking leads to the disruption of muscle membrane structure and facilitates the interaction of lipid oxidation catalysts with unsaturated fatty acids, resulting in the generation of free radicals and the propagation of *WOF* [7]. This phenomenon can be retarded trough the use of aromatic plant rich in phenolics antioxidant [14]. However in our study *TL* diet treatment did not affected *WOF*. According to other studies realized on chicken meat [15], turkey [16], pork [17]and beef [14], α-tocopherol (natural antioxidant) diet did not decrease the apparition of tints related to *WOF* in meat just cooked.

The lack of effectiveness of *TL* diet on *WOF* could be due to masking of the aroma of *WOF* by the odours/flavours that describing the oxidative changes. However, diet affected rancid flavour (*RF*). There are significant differences between the two levels of thymus leaves in *RF* values on day 2 and 4. So *TL* delayed rancid flavour on cooked meat in a 27% and 25% (T_1 and T_2 respectively) at day 4 of storage, compared with *C* samples. These results are in the same line that rancid odour.

In addition, the acceptability was influenced with the *TL* level used, *AC* scores values were positively related with *TL* level (R =0.237, $P \leq 0.01$). The results suggest that the sensory acceptance in cooked lamb meat increased by the *TL*, and this acceptance was higher with high *TL* level (7.5%) used thought diet.

IV. CONCLUSION

The cooked meat of lambs from ewes fed thyme showed a lower oxidation tendency during 4 days of storage under retail display conditions. Sensory evaluation scores of cooked lamb meat indicate that meat during storage had a loss of colour, and the appearance of slight rancidity and warmed-over-flavour. All this sensory attributes contributed to that general acceptance decreased during the storage, showing *TL* diet an improvement of acceptance, because decreased rancid odour and rancid flavour mainly. No statistically significant differences were detected among the results obtained from the two ewe groups fed with thyme leaves (3.7-7.5%).

Natural feeding with thyme leaves could be an alternative to using synthetic additives in animal diets as seen from the results obtained in the present experiment.

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Table 1. Average values and standard deviations of acid reagent substances (*TBARS*: mg MDA/kg meat), in cooked lamb meat stored for 0, 2 and 4 days under retail conditions.

	Level	Day 0	Day 2	Day 4
		M ± SD	M ± SD	M ± SD
	C	0.19 ± 0.04^{z}	2.93 ± 0.24^{ay}	4.28 ± 0.22^{ax}
TBARS	T_1	0.25 ± 0.16^{z}	2.60 ± 0.28^{by}	3.49 ± 0.15^{bx}
	T_2	0.13 ± 0.01^{z}	2.76 ± 0.83^{by}	3.80 ± 0.21^{bx}

C, Control; T_1, 3.5% *TL*; T_2, 7.5% *TL*. Thiobarbituric acid reactive substances (TBARS) (mg malonaldehyde/kg meat). Means with different superscripts are significantly different (P<0.05). a, b, c: different letters within a same columns (different diet treatment) differ significantly (P <0.05). w, x, y, z: different letters within a same row (different storage day) differ significantly (P <0.05).

Table 2. Mean values and standard deviations of sensory scores in cooked lamb meat stored for 0, 2, and 4 days under retail conditions.

	Level	Day 0	Day 2	Day 4
		M ± SD	M ± SD	M ± SD
MC	*C*	4.86 ± 0.28^{x}	3.83 ± 0.29^{by}	3.52 ± 0.43^{z}
	T_1	4.80 ± 0.34^{x}	4.16 ± 0.29^{ay}	3.77 ± 0.35^{z}
	T_2	4.80 ± 0.34^{x}	4.11 ± 0.27^{ay}	3.75 ± 0.35^{z}
RO	*C*	1.02 ± 0.11^{z}	2.02 ± 0.46^{ay}	2.72 ± 0.59^{ax}
	T_1	1.00 ± 0.00^{z}	1.47 ± 0.46^{by}	2.02 ± 0.52^{bx}
	T_2	1.00 ± 0.00^{z}	1.47 ± 0.49^{by}	1.94 ± 0.56^{bx}
WO	*C*	1.08 ± 0.19^{y}	1.72 ± 0.73^{x}	2.16 ± 0.89^{x}
	T_1	1.00 ± 0.00^{y}	1.55 ± 0.51^{x}	1.91 ± 0.62^{x}
	T_2	1.00 ± 0.00^{y}	1.50 ± 0.51^{x}	1.80 ± 0.71^{x}
RF	*C*	1.00 ± 0.00^{z}	2.27 ± 0.25^{ay}	2.86 ± 0.65^{ax}
	T_1	1.00 ± 0.00^{y}	1.66 ± 0.59^{bx}	2.08 ± 0.69^{bx}
	T_2	1.02 ± 0.11^{z}	1.61 ± 0.55^{by}	2.13 ± 0.33^{bx}
WOF	*C*	1.05 ± 0.16^{y}	1.83 ± 0.70^{x}	2.11 ± 0.86^{x}
	T_1	1.02 ± 0.11^{z}	1.52 ± 0.52^{y}	2.00 ± 0.80^{x}
	T_2	1.02 ± 0.11^{y}	1.72 ± 0.57^{x}	2.05 ± 0.63^{x}
AC	*C*	4.30 ± 0.42^{x}	2.72 ± 0.49^{by}	2.02 ± 0.40^{bz}
	T_1	4.55 ± 0.41^{x}	3.36 ± 0.56^{ay}	2.77 ± 0.54^{az}
	T_2	4.63 ± 0.44^{x}	3.38 ± 0.47^{ay}	2.75 ± 0.46^{az}

C, Control; T_1, 3.5% *TL*; T_2, 7.5% *TL*.Scoring scale: (1: minimum- 6: maximum). *MC*, meaty colour; *RO*, rancid odour; *WO*, warmed-over odour; *RF*, rancid flavour; *WOF*, warmed-over flavour; *AC*, acceptance. Means with different superscripts are significantly different (P < 0.05). a, b, c: different letters within a same columns (different diet treatment) differ significantly (P <0.05). w, x, y, z: different letters within a same row (different storage day) differ significantly (P <0.05).