ROSEMARY, ROMEGRANATE, GRAPE SEED, AND HYDROXYTYROSOL EXTRACTS AS A USEFUL STRATEGY TO EXTEND THE SHELF LIFE OF PORK PATTIES

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I. INTRODUCTION

In the last twenty years, numerous studies have been conducted to find out the antioxidant capacity of various extracts to prevent or reduce these pathways of action to reduce oxidative damage in meat [1]. This is why, in the case of meat products such as hamburgers, the main preservative used is sodium metabisulphite, an additive with antimicrobial and colour-stabilising properties. However, these sulphites can trigger allergies in people with a metabolic disorder of sulphite oxidase, involved in carbohydrate metabolism and cases of adverse reactions in asthmatic populations, manifesting in the form of dermatitis, urticaria, angioderma, and even anaphylactic shock [2]. In the present study, the antioxidant activity of several samples of pork patties exogenously enriched by incorporating natural extracts obtained from Rosmarinus officinalis, Punica granatum, Vitis vinifera seed and hydroxytyrosol (from Olea europaea), which act as preservatives and total substitutes for traditional sulphites, was analysed.

II. MATERIALS AND METHODS

Burger elaboration. To prepare the hamburgers, the minority components were mixed in 6 different containers. For that, six groups (6 batches per group) of pork patties with different formulation were established: Control (with no additives), Commercial formula (with SO_2), and 400 ppm of different extracts: Rosemary, Grape seed, Pomegranate and Hydroxytyrosol. Once the 6 samples of approximately 1 kg were obtained after mixing and separated into 80 g moulded portions, they were placed in disinfected trays and covered with cling film. Five samples were obtained for each day of analysis, up to a total of 5 days, making a total of 150 samples to be refrigerated at 4ºC for 10 days. The selected outputs during the shelf-life period to be carried out were performed on days 0, 3, 7 and 10. Colour meat was assessed by using a colorimeter during the storage and colour variations were studied. pH, TBARS, and Protein oxidation (thiol concentration) were analyzed. The concentration of thiol groups was determined spectrophotometrically after derivatisation by Ellman's reagent, 5, 5- Dithiobis (2-nitrobenzoic acid) (DTNB) [4]. The analysis was carried out at 0, 3, 7, and 10 days in triplicate. Data were statically analyzed with the statistical package SPSS 15.0 (Statistical Package for the Social Science for Window (IBM, Armonk, New York, USA). A value of $P < 0.05$ was considered statistically significant.

III. RESULTS AND DISCUSSION

With regards to lipid oxidation, at the end of the storage time, the control sample shows a wide difference, being much more oxidised than the rest. The rosemary and hydroxytyrosol samples show the lowest level of lipid oxidation, differing from the grape seed extract sample and the control sample. The commercial formula and the pomegranate sample have values close to the average of the set, but well below the control sample. Regarding protein oxidation, at day 10, there are statistically significant differences between the commercial formula, characterised

by the presence of sulphites and ascorbate, and grape seed, which also has the lowest value of free thiol groups, an indicator of higher oxidation, and sample with hydroxytyrosol. The commercial formula also maintains a more moderate decrease, maintaining a lower protein oxidation compared to the rest of the samples, reaching a minimum of around 5.89 for sample 4 at day 10 and 9 for sample 6 at day 10. In addition, the results are similar with respect to the rosemary extract since it obtains levels of TBARs much lower than the control hamburger and slightly lower than the sample where ascorbate has been used. These results can be compared to recent research carried out by the same research group, which has demonstrated the effectiveness of hydroxytyrosol as a preservative agent in meat products [5]. In reference to colour, the mean value of *a**, representing the reddish shades, generally decreases over the time period for all samples. Isolating the case of the commercial formula with sulphites, the rest of the samples have been in similar ranges in terms of *a** shade, with hydroxytyrosol differing from the rest, with a lower value.

Table 1. Effects of the addition of the different extracts in the pork patties on lipid oxidation (MDA per kg of sample) and the protein oxidation (mg thiol groups/g sample) during the 10 days of refrigeration at 4ºC.

Sample	Day 0			Day 3			Day 7			Day 10		
Lipid oxidation												
	M	SD	P-value	M	SD	P-value	M	SD	P-value	M	SD	P-value
Control	1.22	0.29	P< 0.001	1.43	0.14	P< 0.001	2.41	0.07	P< 0.001	5.38	0.68	P< 0.001
Commercial Formula	0.63	0.02		0.90	0.16		0.97	0.01		1.66	0.02	
Rosemary	0.64	0.01		0.65	0.19		1.20	0.16	P< 0.01	1.40	0.02	P< 0.01
Grape Seed	0.62	0.03		0.61	0.02		1.54	0.20	P< 0.01	2.72	0.46	P< 0.01
Pomegranate	0.72	0.14		0.65	0.11		1.92	0.04	P< 0.01	2.16	0.00	P< 0.01
Hydroxytyrosol	0.77	0.17		0.58	0.03		1.31	0.24	P< 0.01	1.31	0.10	P< 0.01
Protein oxidation												
	M	SD	P-value	M	SD	P-value	M	SD	P-value	M	SD	P-value
Control	43.4	0.68	P< 0.01	17.5	3.05	P < 0.001	8.41	2.45	P< 0.001	18.1	4.43	P< 0.01
Commercial Formula	51.2	3.27		60.5	2.26		24.9	0.59		21.7	5.26	
Rosemary	43.1	6.92	P< 0.01	36.1	0.01	P< 0.001	7.69	2.93	P< 0.001	15.5	2.41	P< 0.01
Grape Seed	55.1	2.06		21.5	0.01	P< 0.001	17.3	1.71	P< 0.001	5.89	0.33	P< 0.001
Pomegranate	32.1	5.78	P< 0.001	19.8	0.15	P< 0.001	16.1	0.33	P< 0.001	13.9	1.02	P< 0.01
Hydroxytyrosol	44.1	3.59	P< 0.01	6.41	0.17	P< 0.001	11.0	0.61	P< 0.001	9.02	0.90	P< 0.001

M: Mean; SD: Standard Deviation; *P-value<0.01; **P-value<0.001.

IV. CONCLUSION

The presence of extracts such as hydroxytyrosol or rosemary led to an improvement in lipid oxidation parameters compared to the commercial formulation, while grape seed and pomegranate extracts did not. The commercial formulation, however, did manage to maintain better protein oxidation parameters compared to the formulas with natural extracts, but with little difference compared to the samples with rosemary and pomegranate. In general, hydroxytyrosol or rosemary acted on the major causes of raw meat deterioration: redness loss, protein, and lipid oxidation.

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