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CHANGES IN THE MOLECULAR SPECIES OF PHOSPHATIDYL ETHANOLAMINE AND PHOSPHATIDYL CHOLINE OF TURKEY *PECTORALIS* MUSCLE DURING COOKING AND SUBSEQUENT REFRIGERATED STORAGE

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INTRODUCTION

Warmed-over Flavour (WOF) is the term used to describe rapid development of off-flavour in cooked meat during refrigerated storage. This problem is attributed to phospholipid oxidation (Gray and Pearson, 1987 ; Ngah *et al.*, 1993). However phospholipid fraction is a complex mixture of several classes composed of a large number of molecular species. Each molecular species is defined by its polar head, its two fatty chains and the position of the fatty chains on the glycerol and the oxidative stability of the different molecular species should be very different as related to the molecule structure. So it is of interest to follow changes in the molecular species amounts of phospholipids during processing to understand the role of the different parts of phospholipid molecules in their degradation. Recent advances in analysis of phospholipid molecular species allow their rapid separation using reversed phase HPLC system and their precise quantification with a light scattering detector (Ngah *et al.*, 1994).

The objective of this study was to investigate changes in the molecular species of the phosphatidyl ethanolamine (PE) and the phosphatidyl choline (PC) of turkey *Pectoralis* during cooking and subsequent refrigerated storage.

MATERIAL AND METHODS

Pectoralis samples of 6 male turkeys were cooked for fifteen minutes according to an industrial process (steam cooking, 80% relative humidity). The internal temperature of the samples was approximately 75°C at the end of the process. Meat samples were analysed raw and cooked after 0, 7, 14, and 21 days of storage under vacuum at 4°C.

Lipids were extracted from 10 g of meat as described by Folch *et al.* (1957). Phospholipid content was estimated by the phosphorus measurement in total lipid extracts according to the method of Bartlett (1959). Phosphatidyl ethanolamine (PE) and phosphatidyl choline (PC) were purified from total lipid extracts in two steps. First, phospholipids were separated from triglycerides on silica cartridges (Juaneda and Rocquelin, 1985). Then PE and PC were purified from total phospholipid fraction by semi preparative normal phase HPLC. Phospholipids (2 mg) were injected on a column (10 x 300 mm, lichrospher Si 60 5 µm) and they were separated into 5 classes according to the procedure described by Ngah *et al.* (1994). PE and PC were collected and separated into molecular species by reversed-phase HPLC with a RP 18 column (4.5 x 250 mm, superspher 4 µm) according to the method of Ngah *et al.* (1994). The molecular species were quantified with a light scattering detector. The results are expressed in mg on the basis of 100 g of raw meat.

Data were subjected to a one way variance analysis (Snedecor and Cochran, 1981). The fixed effect model included 5 levels : raw meat, cooked meat stored 0, 7, 14 and 21 days.

RESULTS AND DISCUSSION.

In turkey *Pectoralis*, the phospholipids accounted for 0.60g/100g of raw meat and were composed of 60% of PC and 28% of PE. These results are in agreement with those of Acosta *et al.* (1966). Cooking induced no significant change in the total phospholipid and the PC contents but a marked reduction in PE content was observed (-32%). This observation confirms the previous studies which have underlined that PE is the phospholipid the most sensitive to degradation during cooking (Gandemer, 1990 ; Ngah *et al.*, 1993). No significant changes was observed in total phospholipids and in both PE and PC during the subsequent storage of cooked meat (Tables 1 and 2).

The PC contained mainly di acyl molecular species (82%) and only a small amount of alkenyl acyl ones (18%). The most abundant di acyl molecular species were 16:0/18:2 n-6 (35%) and 16:0/18:1 (31%) whereas the main alkenyl acyl one, 16:0A/18:1, accounted for less than 8.6% (Table 1). On the contrary, the di acyl and alkenyl acyl molecular species of PE were present in a equal amount. PE is formed of six main molecular species : 18:0/20:4 n-6 (26%), 18:0/18:2 n-6 (18%), 18:0A/20:4 n-6 (17%), 16:0A/18:2 n-6 (12%), 16:0A/22:6 n-3 (11%), 16:0A/20:4 n-6 (10%)(Table 2). As expected, no significant change in PC molecular species occurred during cooking except for 16:0A/18:1 (-28%). In contrast, all the PE molecular species amounts were reduced during cooking. They were lost in the same proportion (-30% to -35%) whatever their fatty acid unsaturation and the type of molecular species (alkenyl acyl or di acyl) (Table 2). This result doesn't support the widespread hypothesis that PE is the phospholipid the most sensitive to oxidation during cooking because it contains a high proportion of long chain polyunsaturated fatty acids. Thus, in our experimental conditions, the molecular species with 20:4 n-6 and 22:6 n-3 were destroyed to the same extent that the ones containing 18:1 or 18:2 n-6. This study also points out that the PE alkenyl acyl molecular species aren't more sensitive to heat than the di acyl ones. This observation is in contradiction with the results published by Fogerty *et al.* (1989) who showed a large destruction of alkenyl acyl phospholipids during cooking of various kinds of meat. The common part of PE molecular species is the amino group of the polar head which can be involved in non enzymatic browning reactions during meat cooking (Whitfield, 1992). This work strongly supports the hypothesis that the main factor involved in PE alteration during meat cooking should be the presence of an amino group in the polar head.

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Table 1 : Changes in the PC molecular species of turkey *Pectoralis* muscle during cooking and subsequent refrigerated storage (mg/100g of raw meat).

Molecular species	Raw	Cooked meat storage time (days)			
		0	7	14	21
16:0/20:4 n-6	2.9	3.4	3.2	2.7	3.4
16:0/18:2 n-6	129	148	136	144	150
16:0/18:1	115	113	121	118	111
18:0/18:2 n-6	49 b	57 ab	55 ab	55 ab	61 a
18:0/18:1	10	11	11	11	11
Di acyl	**305**	**331**	**326**	**331**	**336**
16A/20:4 n-6	15	17	16	16	18
16A/18:2 n-6	19 a	13 b	18 ab	18 ab	21 a
16A/18:1	32 a	23 b	25 b	24 b	26 b
18A/18:2 n-6	1.5	1.1	1.3	1.1	0.9
Alkenyl acyl	**68**	**54**	**59**	**59**	**66**
PC	373	385	386	390	402

Each value is the mean of six samples. Means with the same letter are not significantly different. at 5 % level.

Table 2 : Changes in the PE molecular species of turkey *Pectoralis* muscle during cooking and subsequent refrigerated storage (mg/100g raw meat)

Molecular species	Raw	Cooked meat storage time (days)			
		0	7	14	21
16:0/20:4 n-6	1.2	1.0	0.9	1.0	0.9
16:0/18:1	7 a	5 b	4 b	5 ab	6 ab
18:0/20:4 n-6	44 a	30 b	30 b	29 b	32 b
18:0/18:2 n-6	31 a	21 b	20 b	13 b	17 b
18:0/18:1	1.0	0.7	0.6	0.5	0.7
Di acyl	**84 a**	**57 b**	**55 b**	**50 b**	**56 b**
16A/20:4 n-6	17 a	13 b	12 b	14 b	14 b
16A/18:2 n-6	20 a	14 b	13 b	13 b	16 b
16A/22:6 n-3	18 a	12 b	10 b	12 b	11 b
18A/20:4 n-6	28 a	18 b	21 b	25 b	26 b
18A/18:2 n-6	3.3a	2.1b	2.0b	1.3b	2.1b
Alkenyl acyl	**85 a**	**57 b**	**58 b**	**63 b**	**69 b**
PE	169 a	115b	114 b	113 b	124 b

Each value is the mean of six samples. Means with the same letter are not significantly different at 5 % level.