Dietary fat sources modulate the protective effect of vitamin E on lipid and protein oxidation in microsomal membranes from turkey muscles

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Background

Lipid oxidation is considered to be the major cause of flavour deterioration in meat and meat products during refrigerated storage (Pearson *et al.*, 1983). Moreover, lipid oxidation can raise myoglobin oxidation, that turns meat in brown colour (Anton *et al.* 1993), and protein oxidation (Mercier *et al* 1995), that can decrease functional properties (Decker *et al.*, 1992). Stabilisation of oxidative processes in meat can be obtained by vitamin E supplementation in animals diet (Monahan *et al.*, 1993).

Diet for fast growing poultry breed are generally rich in polyunsaturated fatty acids (PUFA) and the degree of unsaturation in carcass fat is thereby increased. However increasing the degree of unsaturation of the muscle membranes, by dietary manipulation, increased the oxidability of the tissues and products.

Objectives

The aim of this work is to establish the relationship between dietary fat and vitamin E status in muscle microsomal membranes, and the impact of this interaction on membrane oxidability.

Methods

36 male turkeys of BUT strain were divided in 3 groups which received, for 16 weeks, a basal diet enriched with 6% of one of the following fat sources : rapeseed or soya oil or tallow fat. Each group was divided in two subgroups which received 30 ppm (control : C) or 200 ppm (supplemented : S) of α-tocopheryl acetate (Hoffman-Laroche, France). Pectoralis major and Sartorius muscles were kept at slaughter, put in a plastic bag, stored on ice and, after 6 hours, frozen at -80°C until preparation. The microsomal membranes were obtained by differential centrifugation of muscle homogenate (Apgar & Hultin 1982). Vitamin E content was determined according to the method of Buttris and Diplock (1984). The fatty acids composition of the microsomes was determined by gas liquid chromatography (Hewlett-Packard 5890 with flame ionisation detector). Fatty acid methyl esters were prepared according to the method of Morrison and Smith (1964). The microsomes oxidation was carried out using the system : $FeCl_3$ (0.1mM) + sodium ascorbate (0.5mM) at 37°C for 5 hours. Lipid oxidation measurement were determined by the TBA-RS measurement (Lynch & Frei 1993). Protein oxidation measurement were determined by the carbonyl content (Oliver *et al.*, 1987). Data were expressed as means ± SD and student t-test was used to determine the significant differences.

Results and discussion

Table 1 shows the vitamin E content in the microsomal membranes from Pectoralis major and Sartorius muscles. Whatever the feeding mode (Tallow / Soya oil / Rapeseed oil), vitamin E content was significantly higher in membranes from animals fed the supplemented diet. The vitamin E content in membranes from supplemented animals was about 6-fold greater than in control, but with slight differences between the dietary fat source and muscles. In Pectoralis major muscle, the highest vitamin E content (supplemented) was showed in membranes from animal fed rapeseed and soya oil. At the opposite, for Sartorius muscle (supplemented) the higher vitamin E content was observed in microsomes from animal fed tallow oil. However no significant difference in vitamin E status between the two muscles was shown except for tallow fed animals. In a similar experiment on chickens, it was shown no difference in microsome α-tocopherol content, whatever the dietary fat source used in the diet (Lauridsen et al., 1997).The different fat sources influence significantly the fatty acids composition of microsomes. (Table 1) Microsomal phospholipids from animals fed soya diet, have less monounsaturated fatty acids (MUFA) and more polyunsaturated fatty acidsthan those of animals fed rapeseed oil or tallow. The differences on PUFA composition in membrane from soya fed animals were more marked on the n-6 fraction. Moreover (table1) no significant difference in fatty acids composition between the two muscles or between control and supplemented animals have been shown. Lauridsen *et al.*(1997), showed in thigh and breast membranes, a significant effect of the dietary fat source on microsomal fatty acids composition. Moreover the authors showed no significant effect of the vitamin E supplementation on the fatty acids composition.

Figure 1 shows the TBA-RS values of microsomal membranes from control and supplemented animals. At time 0, lipid oxidation of microsomal membranes from control animals were often higher than those of supplemented animals and the differences were more marked on Pectoralis major than in Sartorius muscle. The chemical induction increased highly the TBA-RS values and after a 2 hours induction maximum TBA-RS values were reached. Lipid oxidation increased significantly in control animals compared to the supplemented ones. The highest significant increase in TBA-RS was noticed in control animals fed tallow and, to a lesser extent, in control animals were fed rapeseed oil. It must be highlighted that when animals were fed soya diet, the differences in TBA-RS between control and supplemented were no significant (Figure 1). Vitamin E supplementation is efficient to lower TBA-RS values in membranes from tallow and rapeseed oil fed animals and inefficient in

membranes from soya fed animals, richer in PUFA. When olive oil was compared to tallow in animal feeding, the TBA-RS values obtained in iron-induced membranes from olive oil fed animals were higher than with tallow (Lauridsen et al., 1997).

Figure 2 shows the protein oxidation measured by the carbonyl content in microsomal membranes after an oxidation induced by a Fe^{3+}/ascorbate system. At time 0 there was no difference between control and supplemented samples whatever the feeding mode and the examined muscle. Contrary to TBA-RS values, the carbonyl content increased during the 5 hours induction, except for the soya fed animals where the maximum of protein oxidation was obtained after 2 hours. Moreover the vitamin E supplementation decreased significantly the protein oxidation in membranes from tallow and soya fed animals, while no difference between control and supplemented animals were observed for rapeseed diet for non elucidated reasons. Nevertheless, this decrease was less marked than that observed in TBA-RS values.

Conclusions

The dietary fat sources influenced the fatty acid composition of microsomal membranes in turkey Pectoralis major and in Sartorius muscles, but did not influences vitamin E status. As in meat (Mercier et al., 1998) and in homogenates (Gatellier et al., this session), in iron-induced microsomal membranes an appropriate vitamin E supplementation could lower lipid oxidation and, to a lesser extent, protein oxidation but with differences in relationship with the membranal PUFA content .

Literature cited

Anton, M., Salgues. C., Gatellier, P. et Renerre, M. (1993) Sciences des aliments. **13,** 261-274

Apgar, M.E. and Hultin, H.O. (1982) Cryobiology, **19**, 407-418.

Buttris, J.L. and Diplock, A.T. (1984). In Methods of Enzymology. ed. L. Packer, A.N. Glazer, 105, 131-138.

Decker, E.A., Xiong, Y.L., Calver, J.T., and Blanchard, S.P., (1993). Journal of Agricultural and Food Chemistry **41**, 186-189.

Lauridsen , C., Buckley, D.J., Morrisey, P.A. (1997) Meat Science, **46**, 9-22

Lynch, S.M. and Frei, B. (1993). Journal of Lipid Research **34**, 1745-1751.

Mercier, Y., Gatellier, P., and Renerre, M. (1995). In *Proc. 41th ICoMST*. San Antonio, **E10**, 562-563.

Mercier, Y., Gatellier, P., Viau, M., Remignon, H., and Renerre, M. (1998). Meat Science **48**, 301-318

Monahan, F.j., Gray, J.I., Asghar, A., Haug, A., Shi, B., Buckley, D.J. and Morrissey, P. A. (1993) Food Chemistry **46**, 1-6

Morrison W.R. and Smith L.M. (1964) Journal of Lipid Research., **5**, 600-608.

Oliver, C.N., Ahn, B.W., Moerman , E.J., Goldstein, S. and Stadman, E.R. (1987). Journal of Biological Chemistry **43**, 1339-1342.

Pearson, A.M., Gray, J.I., Wolzak, A.M. and Horenstein, N.A. (1993) Food Technology **37**, 121-129

	Pectoralis major						Sartorius					
	Tallow		Rapeseed		Soya		Tallow		Rapeseed		Soya	
	C	S	C	S	C	S	C	S	C	S	C	S
Vit. E	0.3 ± 0.1	2.9 ± 0.9	0.9 ± 0.4	4 ± 1.4	0.3 ± 0.1	3.6 ± 2.2	1.3 ± 0.6	7.4 ± 1.6	0.8 ± 0.4	3.5 ± 0.9	0.5 ± 0.3	2.2 ± 0.5
SFA	38.1±0.8	37.6±0.9	34.3±0.3	35.2±0.5	35.6±0.2	35.8±2	37.1±0.5	36.8±1.8	36.6±3.3	34.5±1.5	36.5±0.3	38.1±1.2
MUFA	25.2±0.1	25.7±1.7	27.3±0.3	23.8±0.5	18.2±0.5	16.5±0.5	25.5±0.4	25.9±1.2	32.9±1.4	28.7±2.0	18.5±0.9	17.7±1.1
PUFA	36.7±0.7	36.7±1.9	38.4±0.9	40.0±0.9	46.2±1	47.7±3.4	37.4±0.9	37.3±2.6	30.5±4.6	36.8±2.7	45±0.8	44.2±1.5
n-6	33.1±0.5	32.6±1.4	31.7±0.4	35.1±0.6	40.7±0.4	41.6±2.4	33.1±0.3	33.2±2	24.1±4.1	31.0±2.5	39.9±0.2	40.3±1.2
n-3	3.6±0.2	4.1±0.5	6.7±0.5	5.9±0.3	5.5±0.6	6.1±1	4.3±0.4	4.1±0.6	6.4±0.5	5.8±0.2	5.1±0.6	3.8±0.3

Table 1 : Vitamin E (µg/mg of protein) content and fatty acid composition (%) in microsomal membranes from Pectoralis major and Sartorius muscles from turkeys fed different fat sources and vitamin E levels.

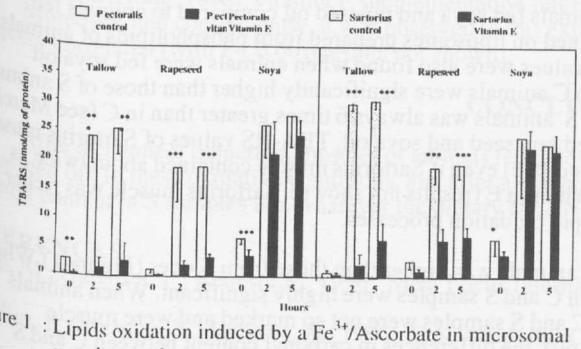

Figure 1 : Lipids oxidation induced by a Fe^{3+}/Ascorbate in microsomal membranes from turkey muscles. Values are means ± SD * p<0.05 ; ** p<0.01 ; *** p<0.001.

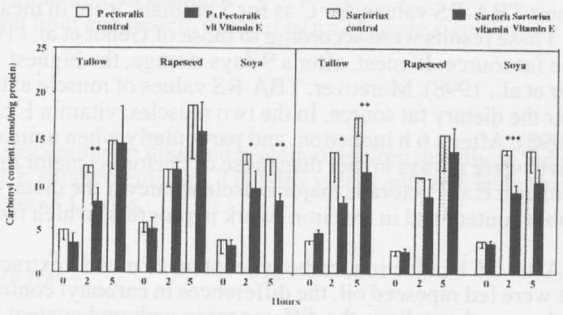

Figure 2 : Protein oxidation induced by a Fe^{3+}/Ascorbate in microsomal membranes from turkey muscles. Values are means ± SD * p<0.05 ; ** p<0.01 ; *** p<0.001.