Diet-dependent trend of lipid oxidation is modified by salt addition in Iberian pigs

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Background

Lipid oxidation has been shown to be highly dependent on the feeding background of meat-producing animals (Buckley et al., 1995). The rate of lipid oxidation in muscle foods is dependent on a number of factors including the degree of lipid unsaturation (Rhee et al., 1996) and the presence of antioxidants (Monahan et al., 1992) and prooxidants (Apte and Morrisssey, 1987). Muscle contains a defensive antioxidant complex, that includes enzymes, metal binding proteins and free radical scavengers such as α-tocopherol. α-Tocopherol is concentrated in membrane fractions such as mitochondrias and microsomes (Machlin, 1984) and is known to improve oxidative stability post slaughter (Buckley et al., 1995). On the other hand, metals ions such as iron or copper, haem proteins (Apte and Morrissey, 1987) and enzymes (Asghar et al., 1991) catalyze lipid peroxidation.

Iberian pig is an autochthonous breed which is extensively fed in evergreen-oak forest in the South West of Spain. Feeding diets consist of acorns and grass principally, which provide high amounts of monounsaturated fatty acids (acorns), n-3 polyunsaturated fatty acids and α-tocopherol (grass) (Rey et al., 1997). Furthermore, exercise provide high myoglobin content.

Although meat processing generally involve salt addition there is to our knowledge a lack of information on the effect of dietary treatment on salted meat lipid oxidation.

Objetives

The objective of this study was to assess the effect of salt addition in the pattern of lipid oxidation of muscle system from pigs fed extensively or in confinement with diets containing pro/ or anti/ oxidants.

Methods

Castrated male Iberian pigs (n=125) weighing 105 kg approximately, were randomly allotted to five groups according to the type of feeding during the finish-fattening period (last 56 days prior to slaughter). A group was raised in extensive conditions with available pasture and acorns (Quercus ilex and Q. rotundifolia). The other four groups were raised in confinement and were assigned to each of the following diets: (2) Basal diet with 10 mg α-tocopherol/kg feed (BASAL); (3) Basal diet with 100 mg α-tocopherol/kg feed (B+E); (4) Basal diet with 125 mg $CuSO_4.5H_2O$ (B+Cu);(5) Basal diet with 125 mg $CuSO_4.5H_2O$ and 100 mg α-tocopherol/kg feed (B+Cu+E). Chemical analysis of feed was carried out following the AOAC (1984) methods. Fat was extracted as previously described by López-Bote et al. (1997).

Animals were slaughtered and samples from *longissimus dorsi* muscle were taken. TBARS (Thiobarbituric acid reactive substances) of representative M. *longissimus dorsi* samples were evaluated at 4°C under fluorescent light at 0,3,6 and 9 days (Salih et al., 1987). When salt was added (2%), the same procedure was used.

The extraction of muscle microsomes was achieved by differential centrifugation (Ashgar et al., 1990). The peroxidative stability of the isolated microsomes was determined by a modification of the method described by Kanner and Harel (1984). α and γ-tocopherols concentration in microsome extracts was measured as described by Rey et al. (1997).

Response data were evaluated by the GLM procedure (SAS, 1988). The comparative analysis between means were conduced using orthogonal contrast.

Results and discussions

Acorn has a high content of fat (Table 1), which is characterized by a very high content of oleic acid. This is in agreement to the literature (Rey *et al.*, 1997). Furthermore, acorn has a high content of γ-tocopherol, which had not been reported before. Grass shows a relative high proportion of C18:3 (n-3) and α-tocopherol content of the analyzed grass is relatively high and is in agreement to the range found in the literature (Rey et al., 1997).

TBARS values were measured (Fig 1) and extensive feeding Iberian pigs had significantly ($P<0.02$) higher lipid oxidation than the other groups. These results have not been reported before and it might be explained by a higher myoglobin content (Apte and Morrissey, 1987) and by a higher n-3 fatty acids concentration in tissues (López-Bote et al., 1997) than the other pigs. Groups supplemented with α-tocopherol acetate had significantly ($P<0.02$) lower lipid oxidation than the other groups as has been found before in pigs (Monahan et al., 1992). On the other hand, pigs receiving supplemented diets with copper sulphate did not show significantly higher values than the other pigs. This can be explained by the low deposition of copper in muscle tissue (Luo and Dove, 1996).

Salt-added samples showed a similar trend (Fig. 2), except the extensive feeding group that showed intermediate values. It has been found that salt can modify proteolic (Sárraga et al., 1989) and lipolitic enzymes activity (Toldra, 1992). Salt may be reduced the activity of some prooxidant enzymes that could be another factor to explain the high lipid muscle oxidation in extensive feeding pigs.

When muscle microsomes were induced to peroxidation (Fig. 3) some similar effects were observed. α-Tocopherol showed a significantly ($p<0.02$) antioxidant effect that is in agreement to other authors (Monahan et al., 1994). Also, groups supplemented with copper sulphate had a higher oxidation than the other groups, but the significative differences were not found. Nevertheless, microsomal fraction from Iberian pigs fed extensive diet had the lowest significative ($p<0.02$) oxidation after 2 hours of incubation time.

α-Tocopherol concentration in muscle microsomes from pigs supplemented with 100 mg α-tocopheryl acetate/kg feed (Fig 4) was significantly higher ($P<0.0001$) than those from pigs that received a basal diet as has been reported by Asghar et al. (1991). Extensive feeding Iberian pigs had an intermediate α-tocopherol concentration in microsome fractions that it was lower than the concentration from pigs supplemented with α-tocopherol. So the lowest oxidation of membrane extracts in pigs fed extensively cannot be attributed exclusively to differences in α-tocopherol concentration in membrane extracts (Rey et al., 1997). Furthermore, it was found a

significantly ($P<0.0001$) higher γ-tocopherol concentration than those from pigs fed mix diets. A higher antioxidant activity of γ-tocopherol has been described compared with α-tocopherol (Niki, 1986). It also possible the effect of other natural antioxidants or even antioxidant enzymes that are increased with the exercise (Ji et al., 1986).

Conclusions

-Salt addition modified the trend in muscle lipid oxidation.

-γ-tocopherol of acorns and α-tocopherol of grass resulted higher α and γ-tocopherol concentration in microsomal fraction from extensive feeding Iberian pigs, which improved the microsomal lipid stability. Possibly other natural antioxidants may be involved.

-High dietary levels of α-tocopheryl acetate improved the antioxidative status of muscle and microsomal fraction from Iberian pigs.

-Dietary copper suplementation had limited effect on the oxidative stability of muscle and microsomal fraction from Iberian pigs.

Acknowledgements

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Table 1.- Analyzed composition of experimental diets (Iberian pigs)

	Basal	B+E	B+Cu	B+E+Cu	Acorn	Grass
Dry matter	89,01	88,95	90,47	90,78	67,05	26,35
Crude protein (% DM)	13,62	13,56	12,19	13,39	4,71	13,72
Fat (% DM)	4,47	4,75	4,84	4,24	6,34	6,26
Crude fiber (% DM)	4,92	4,32	4,39	4,54	5,7	22,22
Ash (% DM)	6,92	5,08	4,80	4,89	1,7	7,31
NFE (% DM)	70,07	72,29	73,78	72,94	81,55	50,49
α-tocopherol (mg/kg DM)	9,5	125,4	21,6	108,0	20,2	171,0
γ-tocopherol (mg/kg DM)	4,2	3,3	3,4	5,5	94,8	61,0
Copper (mg/kg)	17,4	15,6	46,6	41,8	nd	nd
Fatty acids (g/100 g fatty acid)						
C16:0	24,86	24,95	24,78	25,46	12,90	18,60
C18:0	9,82	10,15	9,48	10,60	3,30	2,42
C18:1 (n-9)	31,77	31,46	32,09	31,30	67,72	11,17
C18:2 (n-6)	30,21	30,23	30,20	29,54	15,04	14,12
C18:3 (n-3)	3,33	3,21	3,46	3,09	1,04	53,69

Fig.1.- TBARS values of M. *longissimus dorsi* from Iberian pigs fed experimental diets.

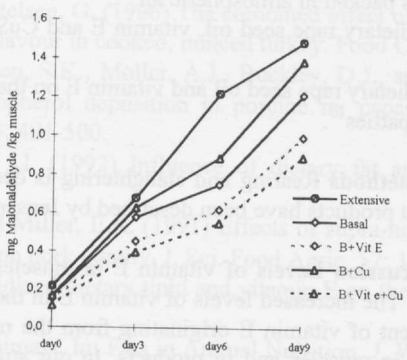

Fig.2.- TBARS values of M. *longissimus dorsi* with 2% added salt from Iberian pigs fed experimental diets.

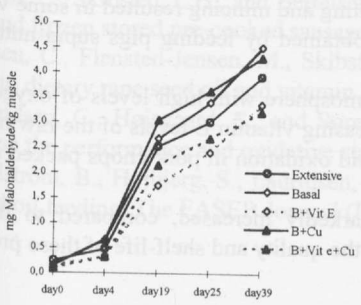

Fig.3- MetMb/hydrogen peroxide lipid peroxidation of muscle microsomes from Iberian pigs fed experimental diets.

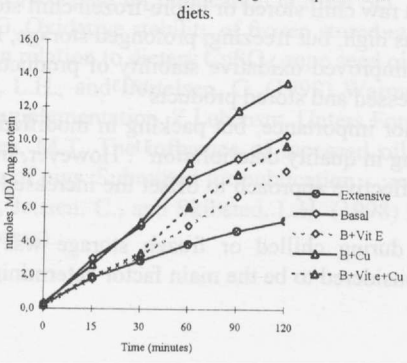

Fig.4- α and γ-tocopherol concentration of muscle microsomes from Iberian pigs fed experimental diets.