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Pre-slaughter stress and lipoperoxidation: protective effect of vitamin E and plant extracts rich in polyphenols given to finishing cattle.

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Introduction

Stress of animals can induce oxidative stress (Chirase et al., 2004) and, consequently, increases production of free radicals. During rearing, animal stress may be detrimental for reproduction and growing performances and, at slaughter, for the quality of meat products due to lipid and protein peroxidation (Aurousseau, 2002). Supplementation of ruminant diets with oil seeds rich in n-3 polyunsaturated fatty acids (PUFA) represents a rapid and efficient way to improve the nutritional quality of products. However, PUFA are susceptible to peroxidation. Our previous studies on dairy cows and on stressed sheep fed on a n-3 PUFA-rich diet showed that the simultaneous oral administration of lipophilic vitamin E (vit E) and hydrophilic plant extracts rich in polyphenols (PERP) reduced plasma lipoperoxidation (Gobert et al., 2008; Durand et al. unpublished data). The present study aimed at evaluating the protective effects of vit E and PERP against plasma lipoperoxidation in finishing cows given a n-3 PUFA-rich diet and submitted to emotional and physical pre-slaughter stress.

Materials and Methods

Thirty-two Normand cull cows were given a straw (30%) and concentrate (70%)-based diet supplemented with lipids (40g oil/kg diet DM) provided by extruded linseeds (C group, n=16), or the same lipids associated with vit E (155 IU/kg) and PERP (7 g/kg diet DM) (EP group, n=16) during their 100d finishing period. Eight cows from each feeding group were slaughtered under minimized stress (stress-) conditions (5min transport from the cattle shed to the abattoir accompanied by a conspecific in the lorry). Eight cows from each feeding group were submitted to additional (stress+) physical exercise (15 min transport, 28 min walking in a unknown environment accompanied by 2 purposely noisy stockpersons, and again 15 min transport) and to psychological stress (non accompanied by a conspecific). Blood samples were collected immediately after stunning (before bleeding) by venepuncture. Stress levels were evaluated by plasma levels of cortisol, glucose and non-esterified fatty acids (NEFA). Plasma α -tocopherol (vit E) was measured by HPLC-UV spectrophotometry and lipoperoxidation intensity by plasma malondialdehyde (MDA) as described by Gobert et al. (2008). Plasma free-hydroxynonenal (4-HNE) and free-hydroxyhexenal (4-HHE), specific markers of n-6 and n-3 PUFA peroxidations respectively, were determined by GC-MS. All data were submitted to ANOVA analysis using the Statistical Analysis System software (SAS Institute, Cary, USA, 2000).

Results and Discussion

Plasma cortisol level (Table 1) was 1.8-fold higher in the stress+ cows ($P=0.007$; average 106.8 ng/mL) than in the stress- cows. This level increase was lower compared to those observed in sheep driven with the aid of shepherd dogs (x6.7, Durand et al., unpublished

data). Plasma glucose and NEFA (Table 1) were significantly higher in the stress+ cows (+8 and +81% respectively, $P=0.01$), probably due to increased energy needs to sustain physical exercise.

Table 1: Effect of diets (C vs EP) and pre-slaughter treatments (Stress - vs Stress +) on stress, lipoperoxidation biomarkers and antioxidant status in plasma collected just after slaughter.

Diets	C		EP		SEM	P-values		
Treatments	Stress -	Stress +	Stress -	Stress +		Antioxidant	Stress	Antioxidant x stress
Cortisol (ng/mL)	72.11	105.26	47.50	108.37	17.31	ns	0.007	ns
Glucose (mg/dL)	72.88	82.21	73.24	75.99	2.47	ns	0.01	ns
NEFA (mmol/L)	0.14	0.25	0.13	0.24	0.04	ns	0.008	ns
MDA (μ g/mL)	0.06	0.07	0.06	0.05	0.02	ns	ns	ns
HNE (ng/mL)	3.50	2.58	1.95	2.16	0.57	0.07	ns	ns
HHE (ng/mL)	0.45	0.42	0.38	0.34	0.06	ns	ns	ns
Vit E (μ g/mL)	3.17	2.91	7.75	8.77	0.73	<0.0001	ns	ns

Intensity of plasma lipoperoxidation (Table 1) was not increased in the stress+ group in contrast in sheep submitted to larger stressing conditions (Durand et al, unpublished data). Moreover, plasma HNE concentrations (Table 1) tended to decrease in plasma of cows given dietary vit E and PERP ($P=0.07$). Thus the addition of the two antioxidant sources had a protective action against lipoperoxidation, as reported in plasma of dairy cows (Gobert et al. 2008) and sheep (Durand et al. unpublished data). The protective action was observed in all cows, whether submitted to minimized or added pre-slaughter stress. Plasma vit E (Table 1) was not decreased by pre-slaughter stress and showed relatively high concentration in EP diet (up to 8.3 μ g/mL). When vit E exerts its antioxidant activity, it is transformed and consequently, plasma levels decreased. The lack of stress conditions on both plasma peroxidized products and vit E levels, was thus coherent.

Conclusions

Although physical exercise during pre-slaughter stress increased the use of energy reserves in cows, this did not favour plasma lipoperoxidation in contrast to earlier observations on plasma of sheep. Independently of the pre-slaughter stress conditions used in our study, the addition of vit E and PERP to the diet tends to protect plasma lipids against peroxidation in cows as earlier reported in sheep.

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