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FATTY AND LEAN PIGLETS ESTABLISH EARLY DIFFERENCIATION IN DNA, RNA AND PROTEIN RETAIN: COMPARATIVE TRIAL ON ALENTEJANO BREED *vs.* CROSSBRED PIGLETS

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ABSTRACT

The aim of this trial was to compare protein, DNA and RNA concentrations in muscle samples from Alentejano piglets (AL) versus Large-White Landrace crossbred piglets (C), since DNA-RNA interrelations and the relations of each parameter with the protein, can serve as indicators for the number of muscle cells and the ability to synthesize protein. The Alentejano swine breed carcasses are characterized by offering a high fat/muscle ratio, consequently it is important to determine the reasons of the relatively small proportion of muscle in the carcass. Samples were taken from *longissimus dors*i muscle (LD) of 6 Alentejano piglets (AL) and 6 crossbred piglets (C) slaughtered at 21 days of age. DNA, RNA and protein concentrations were determined. The LD samples from AL piglets showed lower concentration in the DNA compared with C piglets (131.87 ± 10.50 mg/100 g *vs.* 169.38 ± 10.50 mg/100 g; $P < 0.05$). However, the LD from AL piglets presented greater concentration of RNA (180.50 ± 11.25 mg/100 g *vs.* 134.00 ± 11.25 mg/100 g; $P \leq 0.01$) and protein (22.41 ± 0.83 *vs.* 18.91 ± 0.83 g/100g; $P \leq 0.01$) when compared to the LD from C piglets. The RNA/ADN and Protein/DNA relations were also significantly higher ($P < 0.01$) in samples of muscle from AL piglets. These results indicate a lower number of muscle cells in the LD of AL piglets however, the situation reversed when capacity for synthesis was considered.

Key words: Alentejano swine breed / piglets / *longissimus dorsi* / DNA / RNA / proteins.

1 INTRODUCTION

According to several authors (Suzuki and Cassens, 1980 and Swatland, 1973) the number of muscle cells in pigs is stabilized at the time of birth, and postnatal muscle mass growth takes place, essentially, by hypertrophy of myofibrils, *i.e.* during intrauterine life a hyperplasic process, while after birth a hypertrophic process would prevail. So, whereas the DNA concentration can be an indicator of the number of nuclei, the muscle protein/DNA ratio can predict the size of cells and the RNA/DNA ratio has been pointed as an indicator of the ability of protein synthesis by nucleus, as proposed by Hoffman *et al.* (1983). Also the ratio RNA/protein has been used in other studies (Attaix *et al.,* 1988; Canario *et al.*, 2007) to evaluate the capacity for protein synthesis. Both fat/muscle and muscle/bone ratio in pig carcasses of different breeds, can find justification in the levels of these parameters and a relationship can be established between them. Local breeds, among which the Alentejano pig can be included, have been appreciated primarily by specific characteristics of the processed products. However, gradually, the fresh meat market begins to awaken interest; the small amount of muscle on carcasses has been pointed to, as an obstacle to the pursuit of this activity. In this paper, we investigate, by comparing Alentejano with an improved genotype (LW × LR), the ability for protein synthesis and the genetic predisposition of the Alentejano breed in terms of number and size of muscle, based on assumptions recommended by the previously mentioned authors.

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2 MATERIALS AND METHODS

Six purebred Alentejano (AL) piglets and six crossbred piglets Large-White × Landrace (C) were used. Shortly after birth, 3 AL piglets and 3 C piglets were cross fostered to C and AL sows, respectively, while the other six (full siblings) remained with their natural mothers. At the age of 21 days the adopted and non-adopted full sibling piglets of each genotype were slaughtered and samples (5–6 g) of the *Longissimus dorsi* muscle (LD) were taken. Those samples were immediately frozen in liquid nitrogen and then stored at −20 °C until the analysis. The laboratory tests comprised the following LD determinations:

DNA: determined by the method proposed by Labarca *et al.* (1979) briefly, this method is based on a DNA extraction by solubilising the tissue in EDTA and quantification using a spectrofluorometer,

RNA: determined by the methodology proposed by Munro *et al.* (1969), after the separation of DNA, RNA and proteins (these last two by alkaline digestion), RNA is determined by spectrophotometry.

Proteins: the method used was proposed by Lowry *et al.* (1951) and is based on the colorimetric measurement of a complex formed after the reduction of the Folin phenol by proteins.

Statistical analysis. The data were subjected to analysis of variance using the mixed model (maximum likehood, Harvey, 1993). The mathematical model used was as following:

$$Y_{ijk} = \mu + Gen_i + Trat_j + (Gen \times Trat)_k + \varepsilon_{ijk}$$

where:
Y_{ijk} is observed value for the parameters analysed,
μ is common constant average,
Gen_i is fixed effect of the *i* th genotype (i = 1, 2),
$Trat_j$ is fixed effect of the type of milk ingested *j* th (j = 1, 2),
$(Gen \times Trat)_k$ is fixed effect of the interaction between genotype *i* and type of milk *j*,
ε_{ijk} residual error

Table 1: *Concentrations of DNA, RNA, protein and RNA/DNA and Protein/DNA relationships in the longissimus dorsi muscle (LD) of Alentejano (AL) and crossbred (C) piglets at 21 days of age (least squares mean ± standard error of mean)*

	AL (n = 6)	C (n = 6)	SL
DNA (mg/100 g)	131.87 ± 10.50	169.38 ± 10.50	P < 0.05
RNA (mg/100 g)	180.50 ± 11.25	134.00 ± 11.25	P ≤ 0.01
Protein (g/100 g tissue)	22.41 ± 0.83	18.91 ± 0.83	P ≤ 0.01
RNA/Protein (×1000)	8.05 ± 0.53	7.14 ± 0.53	ns
RNA/DNA	1.38 ± 0.10	0.82 ± 0.10	P < 0.01
Protein/DNA	171.28 ± 10.99	116.03 ± 10.99	P < 0.01

SL– Significance level; ns – not significant

3 RESULTS

There were no significant differences in piglets' weight at 21 days of age independently of genotype and type of milk ingested. The concentrations of DNA, RNA and protein in LD were significantly influenced by the genotype (Table 1) but were not affected neither by the type of milk nor the interaction between genotype and type of milk.

According to data in Table 1, we can see that AL piglets compared to C piglets presented, in the *longissimus dorsi* muscle: (i) significantly lower concentrations of DNA and (ii) significantly higher RNA and protein concentrations, and iii) significantly higher RNA/DNA and Protein/DNA ratios .

4 DISCUSSION AND CONCLUSIONS

The results obtained in this trial showed lower DNA concentration in LD muscle of AL piglets which may indicate lower number of muscle cells, when compared to C piglets. However, as indicated by Hoffman *et al.* (1983), a higher protein/DNA ratio suggests AL piglets have larger LD muscle cells than C piglets. Both the RNA/DNA and RNA/Protein ratios were higher in AL than C piglets, indicating that AL piglets have higher protein synthesis capacity at this age. Hoffman *et al.* (1983) have compared (at 110 days of gestation) foetuses from a fat type breed (Ossabaw) with a lean type breed (Yorkshire) and they also determined higher DNA and lower RNA concentrations in skeletal muscle of the lean type Yorkshire swine breed. The authors have interpreted these results as being indicators of greater protein synthesis in foetus cell of Ossabaw pigs despite the fact that these piglets apparently had lower number of muscle cells. Although hereby presented results have been obtained at an early stage of life of the piglets, we can suspect that lower lean meat content of AL pig carcases after finishing/fattening period, might be linked more to the lower number of muscle cells, than to the ability of this breed to synthesize protein, although the traditional feeding handling, management and production system are not favourable to lean tissue accretion during life long growing. Rehfeldt and Kuhn (2006) indicated that hypertrophy of muscle cells is enhanced when its number is lower, therefore the limit of muscle cells growth is attained sooner and the energy is canalized sooner to fat deposition, which also helps, at least partially, to explain the higher fat

content of AL pigs carcasses when compared with lean type genotypes at the same age (Serra *et al*., 1998).

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