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Size and number variation of adipocytes during the growth of Rasa Aragonesa lambs

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SUMMARY - The size and number of adipocytes of different fat depots was studied in 45 male lambs of the Rasa Aragonesa breed, distributed in three groups of 15 lambs (G12, G24, G36), slaughtered respectively at 11.7 ± 0.67, 24.5 ± 0.57 and 35.8 ± 1.74 kg live weight (LW) and at an age of 32 ± 5, 89 ± 8 and 123 ± 8 days. G12 lambs were slaughtered on the day of weaning, G24 and G36 lambs were weaned respectively at 16.2 ± 1.32 and 18.3 ± 2.46 kg LW and were fed from then onwards on concentrated fodder and barley straw *ad libitum* until they were slaughtered. The results obtained show that there is a significant increase in the amount of fat deposited in three internal (omental, mesenteric and kidney knob and channel) fat depots as the LW at slaughter increases (P<0.01). A similar phenomenon occurs in the size of the adipocytes, hypertrophy being more evident between 24 and 36 kg (P<0.01) than between 12 and 24 kg, except in the intermuscular fat depot, where the differences were between the two most extreme weights (P<0.01). The number of adipocytes in the omental and mesenteric fat depots increased significantly during the period of the study (P<0.01), while no variation occurred in the kidney knob and channel fat depot during the same period.

Key words: Lambs, growth, fattening, adipocytes.

RESUME - *"Variation de la taille et du nombre des adipocytes pendant la croissance des agneaux Rasa Aragonesa". On a étudié la taille et le nombre des adipocytes de différents tissus adipeux chez 45 agneaux mâles de race Rasa Aragonesa, distribués en 3 groupes de 15 agneaux (G12, G24, G36), abattus respectivement à 11,7 ± 0,67; 24,5 ± 0,57 et 35,8 ± 1,74 kg de poids vif (PV) et 32 ± 5; 89 ± 8 y 123 ± 8 jours d'âge. Les agneaux du G12 ont été abattus le jour du sevrage; ceux du G24 et du G36 ont été sevrés respectivement à 16,2 ± 1,32 y 18,3 ± 2,46 kg de PV, et ont eu à leur disposition de l'aliment concentré et de la paille d'orge "ad libitum" jusqu'au le moment de l'abattage. Les résultats obtenus montrent une augmentation significative de la quantité de graisse deposée dans les trois dépôts gras internes (omental, mésentérique, pelvico-rénal) au fur et à mesure de l'augmentation du PV d'abattage (P<0,01). Des résultats semblables ont été trouvés avec la taille des adipocytes car elle augmente avec le PV, l'hypertrophie étant plus marquée entre 24 et 36 kg (P<0,01) qu' entre 12 et 24 kg, sauf dans le tissu intermusculaire dans lequel les différences ont été entre les deux poids extrêmes (P<0,01). Le nombre des adipocytes dans les tissus omental et mésentérique a augmenté significativement pendant la période étudiée (P<0,01), tandis que dans le tissu pelvico-rénal il n'y a pas eu de variation pendant cette période.*

Mots-clés : *Agneaux, croissance, engraissement, adipocytes.*

Introduction

The development of fat depots during the growth of the lambs is due to two phenomena: hypertrophy (increase in the size of adipocytes) and hyperplasia (increase in the number of adipocytes) (Vernon, 1986). Therefore, knowledge of the evolution of these two parameters is essential in order to explain the process.

In each fat depot one and/or the other phenomenon occurs with greater or lesser intensity, depending on the breed, sex, age, development stage and exploitation system of the animals (Hood, 1982; Vernon, 1986).

In this work we studied the size and number variations of adipocytes from different fat depots during the growth of Rasa Aragonesa lambs.

Material and methods

Forty-five male Rasa Aragonesa lambs were used, distributed in three groups of 15 lambs (G12, G24, G36), slaughtered respectively at 11.7 ± 0.67, 24.5 ± 0.57 and 35.8 ± 1.74 kg live weight (LW) and at an age of 32 ± 5, 89 ± 8 and 123 ± 8 days. G12 lambs were slaughtered on the day of weaning, G24 and G36 lambs were weaned respectively at 16.2 ± 1.32 and 18.3 ± 2.46 kg LW and were fed from then onwards on concentrated and barley straw *ad libitum* until they were slaughtered.

On the day of the slaughter fat samples (0.5 g) from the greater omentum, omental (OMF); the medium rectus, mesenteric (MSF); fat adjacent to the right kidney, kidney knob and channel (KKCF); tail head, subcutaneous (SCF) and sternal fat, intermuscular (IMF) were obtained immediately after exsanguination. The fat from the OMF and MSF depots was separated and weighed. The digestive content of each animal was determined (0.86 ± 0.285, 3.11 ± 0.343 and 3.83 ± 0.986 kg respectively for groups G12, G24 and G36). The carcasses were kept in a cold chamber at 4ºC for 24 hours and then the KKCF fat was separated and weighed. Back fat thickness (BFT) was measured at the intersection 4 cm from the spine and 4 cm behind the bottom rib (Colomer *et al.*, 1988).

The adipocytes fixation was made with 2% osmium tetroxide (Hirsch and Gallian, 1968). The samples were then treated with urea (8M) to liberate fixed adipocytes from the connective tissue matrix (Etherton *et al.*, 1977) and were then filtered through a nylon sieve and collected in filtering membrane (pores of 0.45 µm diameter). The images of the preparations were recorded in a computer equipped with an image analysis program (Biocom, 1992) to measure the diameter of the adipocytes. Above 200 cells, randomly chosen, were measured for each sample. The cellular volume was calculated assuming that adipocytes form is spherical.

The number of adipocytes in the OMF, MSF, and KKCF depots was calculated on the basis of the chemical fat content of the separated tissue (Soxhlet's method) and the chemical fat density value (d=0.915 g/cc) (Keys and Brozek, 1953).

Three groups of lambs were compared for each fat depot by means of ANOVAs for independent groups; the comparison between the different depots in each group of lambs was made via ANOVAs for repeated measures. The Fisher test was used to contrast the measurements in both cases.

Results

Table 1 shows the results corresponding to the amount of fat (absolute value and refered to empty LW), chemical fat, size (diameter and volume) and the number of adipocytes in the fat depot studied (OMF, MSF, KKCF, SCF, IMF) depending on the LW at the moment of slaughter (G12, G24, G36).

The Table indicates a significant increase in the amount of fat in the 3 internal fat depots (OMF, MSF, KKCF) as the slaughter LW increases ($P<0.01$). When the amount of fat refers to empty LW, it is maintained in the OMF and MSF depots and decreases in the KKCF depot by between 12 and 24 kg per LW at slaughter ($P<0.05$); a significant increase occurs on the 3 depots ($P<0.01$) between 24 and 36 kg LW, although there are no differences between 12 and 36-kg LW lambs in the KKCF depot.

The adipocytes size increases with slaughter LW, this hypertrophy being more accentuated between 24 and 36 kg ($P<0.01$) than between 12 and 24 kg, except in the IMF depot where the differences were between the extreme weights ($P<0.01$). As far as numbers are concerned, a distinction can be made between the OMF and MSF depots, where there is a continuous increase in the number of adipocytes ($P<0.01$), and the KKCF, where no variation occurred during the study period.

Table 1. Characteristics of omental (OMF), mesenteric (MSF), kidney knob and channel (KKCF), subcutaneous (SCF) and intermuscular (IMF) fat depots in male Rasa Aragonesa lambs of 11.7 (G12), 24.5 (G24) and 35.8 (G36) kg of slaughter weight. Comparison between groups for each depot

	OMF			MSF			KKCF			SCF			IMF		
	G12	G24	G36	G12	G24	G36	G12	G24	G36	G12	G24	G36	G12	G24	G36
Fat (g)	123.5^{A}	282.2^{B}	731.0^{C}	98.8^{A}	233.7^{B}	551.1^{C}	156.5^{aA}	239.5^{bA}	498.4^{B}						
Fat/empty LW (g/kg)	11.4^{A}	13.2^{A}	22.9^{B}	9.1^{A}	10.9^{A}	17.2^{B}	14.4^{a}	11.2^{bA}	15.6^{B}						
Chemical fat (g/kg)	806.2^{A}	811.6^{A}	854.2^{B}	518.8^{A}	564.7^{A}	698.7^{B}	880.7	887.3	896.6						
Adipocyte diameter (μm)	59.7^{aA}	66.4^{bA}	81.7^{B}	56.6^{A}	63.7^{B}	75.2^{C}	49.0^{A}	54.3^{A}	73.6^{B}	57.5^{A}	62.0^{A}	79.1^{B}	55.5^{A}	60.1	62.3^{B}
Adipocyte volume (pl)	130.8^{aA}	188.1^{bA}	343.3^{B}	110.6^{A}	159.9^{B}	264.7^{C}	72.2^{A}	98.9^{A}	249.9^{B}	118.4^{A}	155.4^{A}	333.9^{B}	103.9^{A}	139.7	160.7^{B}
No. of adipocytes (10^{7})	85.7^{A}	143.5^{B}	210.8^{C}	54.5^{A}	98.7^{B}	162.9^{C}	233.8	257.4	206.1						

Differences of lower case letters: $P \leq 0.05$; capital letters: $P \leq 0.01$; no letters: non significant differences

Table 2 shows the results for the 3 groups of lambs (G12, G24, G36) in terms of amount of fat, diameter and number of adipocytes for the fat depot studied (OMF, MSF, KKCF, SCF, IMF). This chart shows that at 12 kg LW the depot which presents most fat is the KKCF ($P<0.01$) followed by the OMF and the MSF, the difference between these two being significant ($P<0.05$). At 24 kg LW more fat is deposited in the OMF than in the KKCF ($P<0.05$), a situation which is accentuated at 36 kg LW ($P<0.01$).

Furthermore, the smallest adipocytes correspond to the KKCF at 12 and 24 kg and to the IMF at 36 kg LW while the largest are generally those in the OMF at the three moments of sacrifice.

Finally, it was observed that the KKCF depot is the one with the largest number of adipocytes at 12 and 24 kg LW ($P<0.01$). Nevertheless, at 36 kg LW the number of adipocytes does not differ in the three depots studied.

Discussion and conclusions

The increase in fat quantity with the increase in LW is a widely reported phenomenon in lambs during their growing and fattening phase, and occurs earlier in non-improved rustic breeds (Kempster, 1980-81).

The higher increase in the adipocytes volume above 24 kg LW (except for the IMF depot) (Table 1) indicates that the animals enter a phase of greater lipogenic activity from that weight upwards, this being due to hyperplasia and hypertrophy in the OMF and MSF depots, while this is only due to hypertrophy in the KKCF depot. This indicates that this depot shows earlier differentiation of adipocytes than the MSF and the OMF. Nouguès and Vézinhet (1977) reached the same conclusion with the Merino d'Arles breed. Equally, the results in Table 2 confirm that the KKCF depot matures earlier in its fat deposition than the other 2 internal depots studied (12 kg, $P<0.01$) while this occurs later in the OMF (36 kg, $P<0.01$). These results coincide with those of Wood *et al.* (1980) in British breeds.

The observed decrease in the amount of fat/kg of empty LW stored in the KKCF depot between 12 and 24 kg LW ($P<0.05$) suggests that the depots which develop earliest tend to mobilize a higher amount of fat during the phase of the so-called "weaning crisis" or, in other words, those depots which jointly show both hyperplasia or hypertrophy (OMF, MSF) would not be so affected by this "crisis" as those where only hypertrophy is present (KKCF). In young goats the KKCF depot is also the one where, after weaning, fat is mobilized at an early stage and with greater intensity (Bas *et al.*, 1985).

It may be interesting to note that the greatest deposition of internal fat at high weights takes place in the OMF (Table 2), as this fat does not depreciate the carcass because it is removed at the time of slaughter, although it does have a negative effect on commercial and zootechnical performance, the latter being reflected in an increase in the conversion index of the food. It could probably be claimed that the OMF is one of the most active depots, together with the SCF, throughout the growth and fattening process, given that the latter shows large adipocyte size at the end of the study period, in addition to a considerable increase in the back fat thickness between 12 and 36 kg LW (2.0 ± 1.0 *vs* 5.2 ± 2.2 mm; $P<0.01$).

To conclude, the results obtained partly justify the slaughter of Rasa Aragonesa lambs between 18 and 24 kg LW in the form of *"ternasco"* (Designation of Origin Rules and Regulations, BOE (Official State Bulletin) of 5/10/92) to avoid excessive fattening of the animals.

Table 2. Comparison between omental (OMF), mesenteric (MSF), kidney knob and channel (KKCF), subcutaneous (SCF) and intermuscular (IMF) fat depots in male Rasa Aragonesa lambs of 11.7 (G12), 24.5 (G24) and 35.8 (G36) kg of slaughter weight

	G12			G24			G36		
	Fat (g)	Ø. adip. (µm)	No. adip. (10^7)	Fat (g)	Ø. adip. (µm)	No. adip. (10^7)	Fat (g)	Ø. adip. (µm)	No. adip. (10^7)
OMF	123.5^{aA}	59.7^{a}	85.7^{A}	282.2^{a}	66.4^{a}	143.5^{A}	731.0^{A}	81.7^{a}	210.8
MSF	98.8^{bA}	56.6^{b}	54.5^{A}	233.7^{b}	63.7^{ab}	98.7^{A}	551.1^{B}	75.2^{bc}	162.9
KKCF	156.5^{C}	49.0^{c}	233.8^{B}	239.5^{b}	54.3^{c}	257.4^{B}	498.4^{B}	73.6^{c}	206.1
SCF		57.5^{ab}			62.0^{b}			79.1^{ab}	
IMF		55.5^{b}			60.1^{b}			62.3^{d}	

Differences of lower case letters: $P \leq 0.05$; capital letters: $P \leq 0.01$; no letters: non significant differences

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