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Variation in total body adipose and adipose partitioning in maternal sheep estimated using computed tomography scanning

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Abstract

In sheep, decreasing adipose or fat is desirable in prime lambs; however, adipose also has a role in the productive capacity of the maternal ewe. There are three main depots of adipose within the body of a live animal: sub-cutaneous, intermuscular and internal (or visceral). To understand if differences exist between animals in adipose deposition and partitioning, a study was undertaken using 37 New Zealand composite-breed maternal ewes, scanned using computed tomography on three occasions from nine through to 17 months of age. The images produced were processed to estimate the weight of subcutaneous, intermuscular and visceral adipose (along with lean and bone), with the weights then expressed as a percentage of total adipose to investigate partitioning. The raw average total body adipose remained proportionate across the three scans at approximately 32% of total empty body weight. The resulting data were analysed using a REML variance components analysis in Genstat. The repeatability of fat distribution across the three scans was high, ranging from 0.77 for proportion of intermuscular adipose through to 0.83 for proportion of subcutaneous adipose. The component σ^2_{Animal} was significant for all traits indicating between animal variation in partitioning of adipose between the depots within individual animals. Cumulatively, these results demonstrate that there is variability in adipose deposition and partitioning in New Zealand sheep. If under genetic control, variability that could be exploited to optimise adipose to best meet the requirements of prime lamb production and maternal ewes.

Keywords: Computed tomography scanning; adipose; sheep

Introduction

Adipose tissue or fat, is considered an antagonist trait in sheep, with increased amounts undesirable in prime lambs, but adipose reserves are a desirable attribute in pasture-run maternal ewes in New Zealand. In hill-country environments, feed quality and quantity varies during the production year, generally, in a predictable fashion. Adipose tissue is a buffer for production animals. During times of excess feed or energy intake adipose tissue is deposited which can then later be mobilised when a restricted feed intake cannot fulfil maintenance and production requirements (Lambe et al. 2003; Walkom et al. 2014).

Adipose tissue in the live animal is commonly assigned into carcass and non-carcass depots. The non-carcass depot is adipose laid down around body organs (e.g., kidney adipose) and is referred to as visceral or internal adipose. The carcass depot is further divided in to sub-cutaneous and intermuscular adipose depots. Whilst there is a further form of carcass adipose, intramuscular adipose, its levels cannot be estimated in live sheep. Two animals may have the same total proportion of adipose in their body, but the partitioning can be significantly different. In dairy cows, very little adipose tissue is laid down subcutaneously, with a higher proportion laid down as visceral adipose, whereas, in beef cows a much larger proportion of adipose can potentially be laid down subcutaneously (Berg & Butterfield 1976). The mobilisation of adipose is different between the depots and some provide energy more readily than others (Lambe et al. 2005). Currently there is a lack of data that accurately describes the adipose levels of New Zealand maternal sheep of various genetic types, both in terms of total adipose, but also how the adipose is partitioned throughout the body. Overseas evidence would suggest that animals with Finnish Landrace genetics may preferentially lay down non-carcass compared with carcass adipose (McClelland & Russel 1972), but there have been no comparative studies carried out in New Zealand.

A data set has been generated as part of a pilot study investigating feed efficiency in New Zealand maternal sheep, which included repeat full-body computed tomography scanning of 37 ewes at three different times from the age of nine months through to 17 months of age. The resulting data set enables a preliminary insight in to variability in total adipose levels, partitioning of the adipose and the repeatability of both in New Zealand maternal sheep, which is reported in this paper.

Materials and methods

Animals

Permission for this study was granted from the AgResearch Invermay Ethics Committee (Ethics Numbers 13257, 13270 and 13456). Thirty-seven maternal composite-breed ewes were used as described by Johnson et al. (2015). The ewes were surplus stock from the Central Progeny Test and were, therefore, from a variety of breeds as described by McLean et al. (2006). As part of the study the animals were scanned by computed tomography (CT) on three occasions: CT Scan 1 = July 2014, aged nine months; CT Scan 2 = September 2014, aged 12 months; CT Scan 3 = February 2015, aged 17 months.

Computed tomography scanning

In preparation for scanning, sheep were sedated with Acetylpromazine (Ace10) given by intramuscular injection (1ml/50kg body weight). The animals were scanned in a prone position using a custom built cradle (made of a 400mm diameter PVC pipe which has been halved with three webbing straps for restraint) with hind legs extended caudally and forelegs under the chest. The animal was scanned using X-ray computed tomography (CT; GE LightSpeed 5.X Pro16 GE Healthcare, Australia) using the procedures described by Jopson et al. (1997) with a starting position before the 2nd or 3rd cervical vertebrae and an end position at the distal femur/proximal tibia. Images were collected at 30 mm intervals, with a 450 mm field of view and 5 mm slice thickness. A total of 30-32 images were collected for each animal. The images were initially processed to remove body organs. Subsequently, areas of adipose, lean and bone were calculated in each image using AUTOCAT (Jopson et al. 1995). The tissues were separated into the three tissue types according to their Hounsfield units (HU). HU value ranges were 40 - 115, 116 - 200, 201 – 255 HU for adipose, lean and bone respectively. Tissue area (count of pixels) from each image was numerically integrated and multiplied by the distance between images to estimate the tissue volume for subcutaneous, intermuscular and visceral adipose depots as well as total lean and total bone (Gundersen et al. 1988). Average pixel density was determined by weighting the average density in the individual images by the pixel area in each image. Pixel density was converted to physical density using the relationship shown between HU value and density by Fullerton (1980) which provided the conversion from a volume to a weight.

Images were isolated in the location of the 12th and 13th rib to obtain measurements reflecting the ultrasound measurement of adipose over the *M. longissimus* (C).

Data analysis

Adipose traits generated were the absolute weights of subcutaneous, intermuscular and visceral adipose, which were summed together to provide an estimate of weight of total adipose. The weight of total adipose was then divided by the total carcass weight plus internal fat (sum of all measured lean, adipose and bone (excluding body organs, skin and gutfill)) to estimate total adipose percentage. The partitioning of adipose within the body was also calculated for each depot as a percentage of total adipose and as the ratio of subcutaneous:visceral adipose. The resulting data were analysed using a REML variance components analysis in Genstat. The models fitted included fitting Scan_Number as a fixed effect and Animal_ID and Animal_ID*Scan_Number as random effects which provide variance component estimates for animal (σ^2_{Animal}) and repeat (σ^2_{Repeat}) respectively. The variance component estimates were used to calculate the repeatability of each trait with repeatability $= \sigma^2_{animal}/(\sigma^2_{animal}+\sigma^2_{Repeat})$.

The allometric growth coefficients (Huxley 1932) for the adipose depots were used to describe patterns of growth. The allometric equation takes the form:

$Ln(Y_i) = ln(a) + b(lnX) + ln(e_i)$; where: Y_i = adipose depot; X = weight of the live animal; a = the intercept; b = ratio of relative growth rates or coefficient of allometry; e_i = multiplicative error; ln = natural logarithm. If b = 1, growth is considered isometric indicating similar relative growth rates for X and Y during the period of time measured; if b ≠ 1, growth is considered heterogonic, with values greater than one indicating that X is growing faster than Y.

Results

The summary statistics for the three scan dates are in Table 1. The overall weight of the animals increased over the course of the three scans which took place over an eight-month period from when the animals were nine months old. The total weight of adipose also increased over this time, however, this was nearly proportionate to the overall gain in empty body weight, with little variation in the proportion of total adipose across the scans. There was almost a two-fold difference between the extreme animals for total weight of adipose and the subcutaneous:visceral adipose ratio. The estimates of adipose depth C increased with each successive scan.

The variance component estimates generated using REML variance components analysis are in Table 2. The component was significant for all traits. The repeatability of the traits across the three scans which was ranged from 0.62 for the Adipose Depth C measurement through to 0.89 for proportion of subcutaneous adipose.

The allometric growth coefficients were 1.3 ± 0.19 (0.9 - 1.7) for total adipose; 1.6 ± 0.25 (0.9 -2.0) for SCF; 1.4 ± 0.2 (0.9 – 1.9) for VISF; and 1.1 ± 0.16 (0.7 – 1.4) for INTF.

Discussion

Adipose tissue reserves are a critical attribute of maternal ewes and are phenotypically and genetically associated with improved maternal production, although the size of the effects are sometimes small (Lambe et al. 2005; Kenyon et al. 2014; Walkom et al. 2015). Conversely, adipose levels in prime lamb needs to be controlled to achieve consumer acceptance of the product. Adipose reserves in live sheep can be estimated subjectively using body condition scoring (Kenyon et al. 2014), or objectively through the use of ultrasound to predict the amount of adipose over the *M. longissimus* muscle. Whilst the later measurement has been shown to be related to total body composition (Young et al. 1996) and is useful as a low-cost measurement, it is not able to predict adipose deposition across the different depots. In the study described here, the authors were able to CT scan the full body of study animals, which allowed estimates of total carcass adipose, and the distribution of adipose in the body to be estimated.

In this study, a high repeatability was observed across the three scans, in particular for the proportion of total adipose and partitioning thereof as assessed by the CT, with the repeatability of the C measurement lower. Based on ultrasound measures of fatness in Australian ewes, Walkom et al. (2014) observed moderate phenotypic correlations among different adipose measures across the production

Table 1 Summary data collected for adipose traits measured using computed tomography scanning for a cohort of 37 ewes scanned on three occasions from nine to 17 months of age. Mean ± standard deviation (Min – Max).

	Scan and Age at Scanning		
	Scan 1 – 9 Months	Scan 2 – 12 Months	Scan 3 – 17 Months
Carcass + Internal Fat Weight (kg)[1]	20.8 ± 2.03 (17.6 - 25.3)	28.4 ± 2.89 (22.9 - 34.0)	31.1 ± 3.30 (23.1 - 36.8)
Subcutaneous Adipose (kg)	2.5 ± 0.59 (1.3 - 3.9)	3.2 ± 0.78 (1.8 - 4.7)	3.8 ± 0.98 (2.1 - 6.1)
Intermuscular Adipose (kg)	2.0 ± 0.39 (1.2 - 3.1)	2.4 ± 0.45 (1.5 - 3.3)	2.9 ± 0.55 (1.9 - 4.0)
Visceral Adipose (kg)	2.6 ± 0.51 (1.7 - 3.9)	3.1 ± 0.69 (1.8 - 4.6)	4.1 ± 0.86 (2.9 - 5.8)
Total Adipose (kg)[2]	7.1 ± 1.39 (4.2 - 10.3)	8.7 ± 1.83 (5.2 - 11.8)	10.8 ± 2.27 (7.1 - 15.3)
Percentage Adipose (%)[3]	34.1 ± 5.48 (21.3 - 43.2)	30.7 ± 4.97 (18.8 - 39.6)	34.7 ± 5.33 (23.0 - 44.9)
Percentage Subcutaneous Adipose (%)[4]	34.5 ± 2.9 (26.8 - 40.5)	36.2 ± 2.7 (28.5 - 42.5)	35.0 ± 2.7 (27.9 - 40.5)
Percentage Intermusuclar Adipose (%)[4]	28.5 ± 2.4 (23.1 - 33.8)	27.8 ± 2.0 (24.6 - 31.5)	27.1 ± 2.0 (23.0 - 32.0)
Percentage Visceral Adipose (%)[4]	36.9 ± 2.7 (31.3 - 42.7)	36.0 ± 2.7 (30.6 - 41.6)	37.9 ± 2.8 (32.9 - 44.1)
Subcutaneous:Visceral Weight	0.94 ± 0.14 (0.68 - 1.29)	1.02 ± 0.14 (0.71 - 1.39)	0.93 ± 0.13 (0.66 - 1.23)
Adipose depth over *M. longissimus* (C - mm)	1.9 ± 1.24 (0.4 – 5.9)	2.6 ± 1.31 (0.4 – 5.6)	3.2 ± 1.7 (0.4 – 7.0)

[1]Sum of all measured lean, adipose and bone (excluding body organs, skin and gutfill)
[2]Sum of subcutaneous, intermuscular and visceral adipose weights
[3]Total Adipose/(Carcass + Internal Fat Weight)*100
[4] Weight of Adipose Within Depot/Total Adipose*100

Table 2 Variance component estimates for adipose traits measured using computed tomography scanning for a cohort of 37 ewes scanned on three occasions from nine to 17 months of age

	σ^2_{Animal}		(σ^2_{Repeat})		
	Component	Std Err	Component	Std Err	Repeatability
Percentage Total Adipose (%)[1]	6.4	1.68	1.6	0.27	0.80
Percentage Subcutaneous Adipose (%)[2]	1.8	0.46	0.38	0.06	0.83
Percentage Intermuscular Adipose (%)[2]	0.39	0.10	0.12	0.02	0.77
Percentage Visceral Adipose (%)[2]	1.0	0.26	0.26	0.04	0.79
Subcutaneous:Visceral Weight	0.02	0.005	0.003	0.0007	0.86
Adipose Depth C (mm)	1.1	0.34	0.90	0.15	0.56

[1]Total Adipose/Empty Body Weight*100; where total adipose is the sum of subcutaneous, intermuscular and visceral adipose weights and empty body weight is sum of all measured lean, adipose and bone (excluding body organs, skin and gutfill)
[2]Weight of Adipose Within Depot/Total Adipose*100

cycle of a ewe, but with much higher, close to unity, genetic correlations, and concluded that a single measurement could be made to reflect overall levels of fatness.

The allometric growth coefficients observed were consistent with the work of Kirton et al. (1972) in that the intermuscular adipose depot had the smallest co-efficient. Because it was above one, it did indicate that it was still a growing depot, but just not at the same rate as the other two depots. Subcutaneous adipose had the highest co-efficient, which is consistent with the general consensus that this depot is the last of the major adipose depots to be laid down.

For all traits assessed, 'animal' was significant within the REML analysis, indicating significant between animal variation in the proportion of the body weight comprised of adipose, and the partitioning of that adipose. The consistency of such observations is supported by the high repeatability's, which suggests that animals that were leaner or fatter as growing lambs, continued to be leaner or fatter respectively leading in to their first mating at 18 months of age. Whether or not these differences are due to genetic differences between animals could not be investigated in this study, as the resource was not structured to enable sire (genetic) variation to be investigated. However, in an additional study (Johnson et al. 2016), a larger data set is being generated which has been designed to investigate whether or not the between animal variation observed is heritability, and therefore the result of genetic differences. As described in the introduction, there is limited literature describing body adipose variation for New Zealand's current maternal sheep population, but that there could be significant differences due to genetic differences is supported by historical New Zealand data (Kirton et al. 1972) and overseas data (McClelland & Russel 1972).

The importance of understanding variation in body adipose distribution was highlighted by Lambe et al. (2003) who observed differences in how labile different adipose depots were, concluding that visceral adipose was the most labile, followed by subcutaneous adipose with intermuscular adipose the least labile. Thus, there may be advantages in identifying ewes that do lay down more of their adipose as visceral adipose as it will potentially be better able to be utilised as energy reserve that can easily be replenished when excess energy become available, although this would need to be confirmed in New Zealand production systems. It would additionally suit prime-lamb production systems where excess carcass adipose is not desired.

Conclusion

This study has provided evidence of significant between-animal variation in the total amount of, and partitioning of adipose between New Zealand ewes derived from maternal breeds, but that additionally they are repeatable traits for an individual animal. The results support the need for a more comprehensive data set from which genetic variation in the traits can be investigated; as adequate liable adipose reserves in appropriate depots offer the potential to optimise adipose to meet the requirements of prime lamb production and maternal ewes simultaneously.

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