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## Repeatabilities of blood plasma metabolites and their associations with leanness in genotypes showing a wide divergence in carcass composition

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### ABSTRACT

Strain differences in lean : fat partitioning were typified by a marked contrast in a lean growth index giving equal but opposite emphasis to the weights of protein and fat, in line with previous publications. Strains showed characteristic differences in blood metabolites which did not necessarily reflect genetic variations within a strain and correlated responses to within-breed selection for changes in body composition.

High repeatabilities of blood metabolites within a strain were found for pasture-fed lambs aged between 5 and 14 months for samples collected on the same day following overnight fasts of 20 vs 24 hours, but were lower for samples taken from fasted animals at different ages. Between-age within-strain repeatabilities (averaged across two trials) were moderate for plasma urea (0.24) creatinine (0.38) and non-esterified fatty acids (0.18), but low for beta-hydroxybutyrate (0.09). Similar moderate heritability estimates were found for urea and creatinine. Although standard errors were very high for this small dataset based on only 12 sires, estimates for carcass composition were in broad agreement with other more reliable published values. Also in line with other results, sire-progeny variations in urea and beta-hydroxybutyrate were more positively related to carcass protein than carcass fat, while the reverse was true for creatinine. Metabolically contrasting pairs of metabolites showed potentially useful predictive associations with sire-progeny variations in protein:fat partitioning.

### INTRODUCTION

Breeds and strains of contrasting growth and body composition show variation in plasma metabolites reflecting genetic differences in protein metabolism, fat metabolism and energy balance (Cameron, 1992). Most studies of metabolic associations with carcass composition have been on a small scale and for breeds and strains managed under controlled nutritional conditions; quantitative estimates of within-breed variations and associations are limited for pasture-fed sheep.

This paper reports results from two trials involving breeds and strains showing wide divergence in carcass composition. It examines characteristic variations in plasma urea (UREA), creatinine (CREA), beta-hydroxybutyrate (BOH) and non-esterified fatty acids (NEFA) in order to evaluate their potential usefulness to improving carcass composition by selection.

### MATERIALS AND METHODS

#### Trial 1

Eight contemporaneously reared genotypes were represented: purebred lambs from the high-fat (Sd+) and low-fat (Sd-) Southdown backfat selection lines at Massey University (Kadim *et al.*, 1989), and Texels; crossbred lambs from Romney ewes, by each of these 3 (Tex) breeds of ram; and crossbred lambs by Texel rams mated to either Sd+ or Sd- ewes.

Blood samples were collected at 6 months (March) and 14 months (November) of age from 125 pure- and crossbred lambs (10 to 27 per genotype) born in 1993 at the Tokanui Research Station. Three samples were taken on

each of these 2 occasions: immediately off-pasture at 1 pm; at 9am the following day after an overnight fast with access to water; and at 1pm that same day after continuation of the fast in woolshed pens.

Body composition was assessed in the live animal from ultrasonic scans of eye muscle width (EMW), eye muscle depth (EMD) and subcutaneous fat depth (EFD) at the 12th rib, at 11 months (May) and 14 months (November) of age using an Aloka Model SSD500 scanner with a 5 MHz transducer.

#### Trial 2

A total of 330 crossbred lambs from Romney ewes sired by Sd+, Sd- and Texel rams were allocated at random to either high or moderate feeding levels (pasture allowance) at 4 months, including cross-over levels from 9 months of age. They were slaughtered at either 4, 9, 12 or 15 months of age.

Blood samples were taken following an over-night fast: at 3 (weaning), 6 (March), 9 (May), 12 (August) and 14 (November) months of age. All surviving lambs were also scanned for EMW, EMD and EFD prior to slaughter and at 6 months of age (March). Carcass composition was assessed by chemical analyses of water, fat, ash and protein based on methods described by Kirton *et al.* (1962). Carcass eye muscle width (A), eye muscle depth (B), subcutaneous fat depth (C) and tissue depth (GR) measurements were taken using methods described by Waldron *et al.* (1992).

For both trials, jugular blood samples were collected using vacutainers, serum separated by centrifuge and metabolites analysed using standard methods and an Hitachi 717 random access autoanalyser.

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**RESULTS**

**Trial 1**

**(1) Growth and development**

Average strain differences in growth and in composition on a liveweight-adjusted basis (Table 1) were calculated from adjusted sire and dam breed means ignoring any effects of hybrid vigour, from a model also fitting birth rank and birthday.

Strain differences in live animal compositional indicators were most marked for females and for EFD (all three strains) and EMD (Sd+ vs Tex and Sd-). Moderate weight-adjusted repeatabilities were found for these traits, but a low repeatability estimate was obtained for EMW at constant liveweight.

**(2) Plasma Metabolites**

Repeatabilities (intra-class correlations within strains) were slightly higher on a logarithmic than an arithmetic scale. They were higher for metabolite concentrations in blood collected after an overnight fast than from animals bled soon after removal from pasture, especially at 6 months of age.

Average repeatabilities for fasted metabolite concentrations across both ages and both fasted samplings, and average single measurement correlations across the two ages (in brackets) were: UREA 0.64 (0.27), CREA 0.53 (0.21), BOH 0.28 (0.08), NEFA 0.32 (0.18). Thus within genotype, animals tended to rank much more similarly for measurements taken on the same day following overnight fasts of 20 vs 24 hours than they did for the same fasted measurements taken at 6 and 14 months of age.

Significant strain effects (  $P < 0.01$ ) were found for UREA and CREA, the former trait higher in Sd+ compared with Sd- and Texels (+10%), and the latter higher in Texels compared with Southdowns (+20%). There were no detectable genotype differences in repeatability estimates.

Sex and weight-adjusted genotype means (fitting separate regression lines) gave the average strain relativities shown in Table 2. Single trait correlations with live animal indicators are also presented.

Correlations were higher among genotypes than between animals within genotypes. Among genotypes, UREA showed a similar positive association with EMD and EFD; CREA, BOH and NEFA showed a negative correlation with EFD.

**TABLE 2:** Average purebred effects for blood metabolites and correlations with liveweight-adjusted ultrasonic scan dimensions from Trial 1.

	UREA	CREA (x10 <sup>2</sup> )	BOH	NEFA
<b>Strain relativities#</b>				
Sd- average*	9.79	7.72	0.42	1.18
Sd+/Sd- %	106	108	101	105
Tex/Sd- %	102	123	100	131
Significance	**	**	ns	ns
<b>Correlations among genotypes</b>				
With EMD	0.23	-0.05	-0.12	-0.05
With EFD	0.24	-0.44	-0.27	-0.38
<b>Correlations within genotypes</b>				
With EFD	-0.06	-0.02	0.12	0.01

\* in mmol/l (back-transformed from log scale); refer text for trait abbreviations

# sire and dam breed effects pooled assuming zero hybrid vigour

Relative growth rates of EFD to liveweight showed a wide range among genotypes: from 0.74 for Texels to 1.75 for Texel x Sd+ crosses. CREA and UREA together accounted for 60% of the variation among genotype x sex means for this measure of fat development.

**Trial 2**

Between animal correlations among pairs of measurements taken at the three ages were similar for each metabolite. Average repeatabilities were: CREA 0.54 (twice the value found in Trial 1), UREA 0.20 and BOH 0.09 (both similar to Trial 1 estimates).

Animal variations in carcass composition were assessed as deviations in the weights of protein, fat, water and ash from their average regressions on carcass weight across all slaughter ages in a linear model fitting (on a log scale) the significant effects of breed and sires within breed (random variate), sex and nutritional treatment (fixed effects). Sire estimates were used to derive heritabilities (from variance components) and associations among traits (from sire means). Results are summarised in Table 3.

Despite the small number of sires (12, each represented by 16 to 36 progeny) and the diverse range of genotypes and slaughter ages, heritabilities of carcass

**TABLE 1:** Average purebred effects and repeatabilities for liveweight and ultrasonic scan dimensions from Trial 1 (standard errors in brackets).

	Liveweight		Liveweight adjusted *		
	May	Nov	EMW	EMD	EFD
<b>Strain Relativities#</b>					
Sd- average	36.2 (1.0)	44.2 (1.4)	58.4 (1.1)	28.4 (0.6)	3.29 (0.32)
Sd+/Sd- %	92	90	105	114	150
Tex/Sd- %	111	103	104	105	74
Repeatabilities (Nov & May)			0.14	0.41	0.31
(unadj. for weight)		(0.66)	(0.34)	(0.50)	(0.34)

\* adjusted for age at measurement and genotype x liveweight regression effects (apart from values in final row); EMW = eye muscle width, EMD = eye muscle depth, EFD = subcutaneous fat depth

# sire and dam breed effects pooled assuming zero hybrid vigour

**TABLE 3:** Average purebred effects for carcass weight and chemical composition, heritability estimates (significance levels in brackets) and correlations among sire means from Trial 2

Trait	Sd- (kg)	Sd+/Sd-# % (signif.)	Tex/Sd-# %	h <sup>2</sup> (signif.)	Correlations among sires		
					Protein	Water	Fat
HCW	18.6	96 (***) (ave sed=1.01)	117	0.35 (***)			
Liveweight-adjusted							
Protein	3.44	89 (***)	105	0.37 (***)			
Water	10.9	92 (***)	107	0.26 (**)	0.69		
Fat	3.43	142 (***)	72	0.31 (***)	-0.84	-0.92	
Ash	0.893	92 (ns)	109	0.22 (**)	0.38	0.59	-0.55
3Prot-Fat (Lean growth index)	11.8	39 (***)	150	0.50 (***)	0.96	0.84	-0.96

# purebred effects assuming zero hybrid vigour (back transformed from log scale)

**TABLE 4:** Average purebred effects for carcass indicators and blood metabolites, heritabilities (significance levels in brackets), and between-sire correlations with lean growth index from Trial 2

Trait*	Sd-	Sd+/Sd- % (signif.)	Tex/Sd-# %	h <sup>2</sup> (signif.)	Between sire correlations with (3Prot-Fat) (Lean growth index)
<b>Carcass indicators (mm)</b>					
A	55.3	94 (*)	104	0.63 (***)	0.70
B	32.9	105 (ns)	100	0.18 (*)	0.24
C	1.92	192 (ns)	120	1.02 (***)	-0.76
GR	6.66	193 (**)	102	0.54 (***)	-0.76
<b>Metabolic Indicators (mmol/l)</b>					
UREA (4 mo)	7.19	106 (ns)	97	0.71 (***)	-0.34
UREA (6 mo)	7.59	99 (ns)	107	0.31 (**)	-0.63
UREA (9 mo)	8.19	115 (*)	125	0.20 (*)	-0.18
CREA (4 mo) (x10 <sup>2</sup> )	7.65	101 (**)	119	0.14 (*)	0.31
CREA (6 mo) (x10 <sup>2</sup> )	7.77	110 (*)	121	0.31 (**)	0.40
CREA (9 mo) (x10 <sup>2</sup> )	8.26	104 (*)	119	0.04 (ns)	0.41
BOH (4 mo)	0.36	105 (ns)	70	0.72 (***)	-0.36
BOH (6 mo)	0.31	98 (ns)	102	-ve (ns)	0.17
BOH (9 mo)	0.36	84 (*)	74	0.01 (ns)	0.12

\* refer text for trait abbreviations

# purebred effects assuming zero hybrid vigour (back transformed from log scale)

components were similar to those published by Waldron *et al.* (1992) for dissected components of 6 month old lamb carcasses. In the final row of Table 3 parameters for an index of lean growth (the excess of protein relative to fat weight), calculated giving approximate equal emphasis to residual variations in the weights of protein and fat (on a

log scale), are presented. This index highlights the major contrast among the genotypes studied. Between-sire (within breed) heritabilities, and correlations of linear carcass measurements and blood metabolites with this index are shown in Table 4. Standard errors of heritability estimates (SE) were closely described by the relationship : SE =

$0.075 + 0.38h^2$ , where  $h^2$  is the parameter estimate (i.e. for  $h^2 = 0.3$ ,  $SE = 0.19$ ).

For comparison, average heritabilities of ultrasonic measurements at 5 and 9 months of age were: EMW 0.15, EMD 0.57 and EFD 0.52.

## DISCUSSION

Strain differences in lean : fat partitioning were in line with published reports (Kadim *et al.* 1989; Clarke and Kirton, 1990; Binnie *et al.*, 1995) and are typified by the marked contrast in the lean growth index giving equal but opposite emphasis to the weights of protein and fat (Table 3), and indicating on a purebred basis almost a four-fold difference between high-fat Southdowns (low lean growth) and Texels (high lean growth), with low-fat Southdowns intermediate.

Strain differences found in blood metabolite concentrations were also similar to published estimates (e.g. Carter *et al.*, 1989; Van Maanen *et al.*, 1989; Cameron 1992). Sheep selected for low backfat thickness or high lean growth have tended to show lower blood urea and often reduced creatinine as well, although high creatinine levels are characteristic of the Texel breed. Thus strains of sheep have characteristic differences in blood metabolites which may not necessarily reflect genetic variations within a strain and correlated responses to within-breed selection for changes in body composition (Sinnott-Smith and Woolliams, 1988).

High repeatabilities of blood metabolites within a strain were found for pasture-fed lambs aged between 5 and 14 months for samples collected on the same day after an overnight fast, but were lower for samples taken from the same animal at different ages. Between-age within-strain repeatabilities (averaged across both trials) were moderate for plasma urea (0.24) and creatinine (0.38), but low for beta-hydroxybutyrate (0.09).

High sire-component heritabilities among animals within a strain (Trial 2) are also suggestive of characteristic metabolic differences among animals within a strain. Although the standard errors of these estimates were high, the estimates for carcass weight and composition were closely in line with those coming from larger, more appropriate databases. Estimates for blood metabolites also broadly agree with those published by Cameron and Cienfuegos-Rivas (1990), for pen-fed 20 week old lambs following a 31 hour fast (0.35 for UREA, CREA and BOH), with the suggestion in Trial 2 of higher estimates in younger (4-6 months) than in older lambs.

Associations with sire-progeny variations in protein : fat partitioning (lean growth), also tended to be higher for metabolites measured in young lambs. On average at 4 to 6 months of age these associations were negative for urea (-0.5) and positive for creatinine (0.35) but low and variable for beta-hydroxybutyrate. By contrast, Kuhn *et al.*, (1993), found positive associations (0.2 to 0.4) for both urea and creatinine concentrations of Romney sires at 9 and 13 months with the yearling backfat depth of their ewe progeny.

Heritabilities and between-sire associations with carcass leanness suggest that blood metabolites could be at least as useful as ultrasonic eye muscle scans for ranking animals and sires for genetic improvement of lean growth rate. This conclusion is in line with evidence from the work of Cameron and Cienfuegos-Rivas (1994), indicating complementary effects in selection indices that include metabolites with different associations with carcass lean versus carcass fat content. Ultrasonic estimates of carcass fat and eye muscle measurements, show similar complementary effects (Clarke and Rae, 1991), but also provide some opportunity to focus selection on dimensions indicative of variations in high value lean cuts rather than just overall lean : fat partitioning within the carcass.

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