

Protein synthesis in the skin of Merino lambs, measured by a flooding dose of $^2\text{H}_5$ -phenylalanine, using gas chromatography/mass spectrometry

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A gas chromatography/mass spectrometry (GC/MS) method for the analysis of deuterium labelled phenylalanine ($^2\text{H}_5$ -Phe) has recently been developed (1). We have adapted this method to a Hewlett Packard 5970 quadruple MS interfaced with a Hewlett Packard 5890 GC and used it to measure the effects of liveweight, diet and feed intake on fractional protein synthesis rate (FSR) in skin, using a flooding dose procedure (2).

Two groups of 12, five month old Merino lambs, weighing either 34 or 26 kg, were fed a lupin/hay diet at maintenance in individual pens for four weeks. Each group was then divided into four subgroups: they were fed isonitrogenous diets based on either canola or lupin (75% hay plus 25% canola meal or lupins) to maintain weight (M) or at 0.6 of maintenance (0.6M). These treatments were applied for 28 days and protein synthesis was measured on days 4 and 24. On days 4 and 24, a total of 0.2 g L-phenylalanine/kgW^{0.75} (containing 0.027g $^2\text{H}_5$ -Phe) dissolved in saline, was injected into the jugular vein over 10 minutes. Blood samples were drawn at 0, 5, 10, 20, 40, 70, 100 and 130 minutes from the start of the infusion, and skin biopsies were taken at 0 and 130 minutes. Sample preparation, GC/MS analysis and calculations were as described by Calder et al. (1) except that phenylethylamine was formed into a tert-butyl dimethylsilyl derivative and ionized at 70 V electron energy on MS.

	FSR on Day 4 (%/d)				FSR on Day 24 (%/d)			
	M		0.6M		M		0.6M	
	Canola	Lupin	Canola	Lupin	Canola	Lupin	Canola	Lupin
34 kg	17.6	17.9	16.1	15.2	18.0	17.2	14.7	15.9
26 kg	19.5	15.4	16.9	13.2	17.2	15.8	15.8	14.9
P < 0.05	Weight x diet, intake				Intake			

FSR of skin was 10-13% lower at 0.6M compared with the M sheep (P<0.01) at both times. Feeding the canola diet resulted in higher synthesis rates in the light lambs on day 4. There were no significant differences in FSR between the two liveweights. The results indicate that skin protein synthesis rate was responsive to feed intake and to type of protein fed.

1. Calder AG, Anderson SE, Grant I, McNurlan MA, Garlick PJ. The determination of low d5-phenylalanine enrichment (0.002-0.09 atom percent excess), after conversion to phenylethylamine, in relation to protein turnover studies by gas chromatography/electron ionization mass spectrometry. *Rapid Commun Mass Spectrom* 1992;6:421-4.
2. Nash JE, Rocha HJG, Buchan V, Calder GA, Milne E, Quirke JF, Lobley GE. The effect of acute and chronic administration of the β -agonist, cimaterol, on protein synthesis in ovine skin and muscle. *Br J Nutr* 1994;71:501-13.