EFFECT OF BODY FATNESS ON BLOOD METABOLITES AND INSULIN INSENSITIVITY IN ADULT SHEEP

Blood glucose, free fatty acids, insulin, thyroxine and free thyroxine were measured in fat (90 kg) and thin (55 kg) adult sheep at four levels of feed intake. Differences were observed between fatness levels in blood metabolites and hormones and the possibility of insulin resistance occurring in fat ruminants is discussed.

Key words: Ruminant, insulin, glucose metabolism, obese

[Effets de l'engraissement sur les métabolites du sang et la sensibilité à l'insuline des moutons adultes.]
Titre abrégé: Métabolites du sang chez les ovins maigres et gras.
On a dosé le glucose, les acides gras libres, l'insuline, la thyroxine et la thyroxine libre dans le sang de moutons adultes gras (90 kg) et maigres (55 kg) à quatre indices de consommation. On a noté que la concentration des métabolites du sang et des hormones variait avec l'engraissement de l'animal; on examine la possibilité d'une résistance des ovins gras à l'insuline.

Mots clés: Ruminants, insuline, métabolisme du glucose, obésité

Overfeeding of dairy cows during the dry period with resulting body fatness has become more prevalent since dairy management has emphasized high energy diets and group feeding. At parturition, fat cows appear to be more susceptible to metabolic disorders such as ketosis and milk fever, digestive and infectious diseases and reproductive disturbances (Morrow 1976). Fat cows are more susceptible to the added stress of disease and frequently die despite treatment. The metabolic effects of obesity have been studied extensively in humans, pigs and rats (e.g. Lavau et al. 1979; Wangsness et al. 1981) but little work has been done on the effects of body fatness on metabolism in ruminants. Since fatness usually is a result of chronic overfeeding, it is often difficult to separate the effect of feeding level from the effect of body fatness.

The purpose of this paper is primarily to examine differences in blood concentrations of glucose, free fatty acids (FFA), insulin, thyroxine and free thyroxine (free T_4) in adult sheep at two levels of fatness, and which were fed at four levels of intake. Quantitative measurements of energy and glucose metabolism are reported elsewhere (McNiven 1984a,b).

Six adult, crossbred wethers initially weighing 60 kg were randomly allotted to two groups, fat and thin, and fed at two levels for 4–5 mo to obtain body weights of about 90 and 55 kg, respectively. At both levels of feeding the diet consisted of equal proportions of maize and grass hay. The animals received sufficient digestible protein, minerals and vitamins according to published standards (National Academy of Sciences-National Research Council 1975). The animals were fed at 0730 and 1930 h. For both fatness groups, balance (7 days) and respiration trials (3 days) were made (McNiven 1984a) and blood samples were taken, at each of four levels of metabolizable energy (ME) intake: high, 17 MJ/day; intermediate, 10 MJ/day; low, 6 MJ/day and fasting (day 5 of fast). The intermediate level always preceded fasting measurements. Five jugular venous blood samples were collected via an indwelling catheter at 0715, 1115, 1515, 1815 and 2215 h for each intake level and sheep. Plasma glucose was determined spectrophotometrically by reduction of ferricyanide, and FFA by titration. Serum insulin (Poznanski and Poznanski 1969), thyroxine (Moss et al. 1978) and free T_4 (Amerlex RIA kit, Amersham, England) concentrations were determined by radioimmunoassay. Effects of feeding

level and time of sampling were considered in the statistical model. Data were analyzed using General Linear Models (SAS Institute Inc. 1982).

Table 1 gives the mean values of blood metabolites and hormones for each body fatness and feeding level. Plasma glucose concentrations for the two fatness levels appeared to react differently to reduction in ME intake although when the effects of ME intake and time of sampling were considered, the mean values for the two fatness levels were not significantly different. Plasma FFA ($P<0.05$) and serum insulin ($P<0.01$) concentrations were significantly higher for the fat sheep. There were large variations in the serum thyroxine concentrations and the overall mean for the fat sheep was significantly higher ($P<0.05$) than for the thin sheep. No effect of feeding

level was observed but this may have been masked by changes in the proportions of free and bound thyroxine. Fat sheep had higher concentrations of free T_4 on all feeding levels and these differences were significant ($P<0001$). Feeding level had a significant effect on free T_4 concentrations. Although there was no consistent change in free T_4 concentration with decreasing feeding level, the levels at fasting were significantly lower ($P<0.05$) for both the fat and thin sheep.

Table 2 shows correlation coefficients within sheep and significance levels for the relationships between energy intake and balance, and blood metabolite and hormone levels for the thin and fat sheep. Glucose, FFA and insulin were significantly correlated for both the thin and the fat sheep. In

Table 1. Least square means of blood metabolites and hormones, body weight, energy intake and balance (three sheep per group)

	Feeding level				Least square mean ± SE	Statistical significance		
	High	Inter-mediate	Low	Fast		Fatness level	Feeding level	Time
Body weight (kg)								
Thin	61	59	53	55				
Fat	88	85	83	82				
ME intake (MJ/day)								
Thin	16.8	10.2	6.7	0				
Fat	16.9	9.9	6.4	0				
Energy balance (MJ/day)								
Thin	+4.3	+0.5	−1.0	−7.1				
Fat	+2.8	−1.7	−4.3	−8.6				
Glucose (mmol)								
Thin	3.90	3.85	3.70	3.59	3.76±0.05	NS	***	NS
Fat	3.72	4.07	3.90	3.73	3.86±0.05			
FFA (meq/L)								
Thin	0.52	0.60	0.82	1.28	0.81±0.04	*	***	NS
Fat	0.71	0.64	0.80	1.50	0.91±0.04			
Insulin (ng/mL)								
Thin	0.58	0.53	0.58	0.29	0.50±0.03	**	***	NS
Fat	0.51	0.74	0.70	0.47	0.61±0.02			
Thyroxine (μg/L)								
Thin	0.53	0.55	0.56	0.59	0.56±0.02	*	NS	NS
Fat	0.69	0.57	0.62	0.63	0.63±0.02			
Free thyroxine (ng/L)								
Thin	0.21	0.21	0.30	0.19	0.23±0.01	***	***	***
Fat	0.32	0.28	0.35	0.24	0.30±0.01			

*,**,***$P<0.05$, $P<0.01$, and $P<0.001$, respectively; NS = not significant.

Table 2. Correlation coefficients within sheep

	n	ME intake	Energy balance	Glucose	FFA‡	Insulin	Thyroxine
				Thin sheep			
ME intake	12†						
Energy balance	12	0.982***					
Glucose	60	0.450	0.445				
FFA‡	60	−0.771**	−0.779**	−0.542***			
Insulin	55	0.218	0.238	0.307*	−0.520***		
Thyroxine	60	−0.072	−0.145	−0.166	0.049	0.403**	
Free T_4	60	0.113	0.142	0.286*	−0.314*	0.462***	0.418***
				Fat sheep			
ME intake	12†						
Energy balance	12	0.886***					
Glucose	60	−0.135	0.021				
FFA‡	60	−0.534	−0.641	−0.245*			
Insulin	60	0.117	0.083	0.263*	−0.308**		
Thyroxine	60	0.186	0.046	0.193	0.005	−0.034	
Free T_4	60	0.291	0.280	0.138	−0.016	0.208	0.363**

†Mean values of five observations used for correlations with ME intake and energy balance only.
‡Free fatty acids.
*,**,***$P<0.05$, $P<0.01$, $P<0.001$, respectively.

all cases, the correlation coefficients were higher in the thin sheep.

The thin sheep appeared to react as expected to the decreasing feed intake to fasting by decreasing plasma glucose, increasing plasma FFA concentrations and by maintaining a constant insulin concentration when fed but by decreasing it greatly ($P<0.001$) when fasted. In contrast, the fat sheep had slightly increased plasma glucose concentration when feed intake was decreased, and at fasting the glucose concentration was the same as the highest intake level. FFA concentration followed a similar inverse pattern but, at fasting, the FFA concentration rose to a very high level. Insulin concentration followed the same pattern as the glucose concentration, and at fasting, the insulin concentration was the same as at the highest intake level.

In general, glucose reserves in the body are limited, and at fasting when dietary precursors to gluconeogenesis cease to be absorbed, the body attempts to spare glucose in several ways. Insulin concentration falls, which decreases peripheral uptake of glucose. Reesterification in adipose tissue is depressed, thus decreasing the requirement for glucose and α-glycerophosphate. Mobilization of FFA from adipose tissue is increased, and since utilization of FFA is proportional to the concentration in plasma (Jackson et al. 1968) then FFA will be the preferred substrate and will, therefore, spare glucose to some extent. The thin sheep in the present study showed a typical decrease in glucose and insulin concentrations and an increase in FFA at fasting.

Insulin resistance has generally been defined in monogastrics as hyperinsulinemia, measured as increased fasting insulin and (or) increased response to stimulation, together with normal or increased blood glucose (Wangsness et al. 1981). Insulin resistance in large adipocytes is well known and, in fact, enlarging adipocytes have been extensively used as models of developing insulin resistance (e.g. Digirolamo 1981). Wangsness et al. (1981) and Cote et al. (1982) demonstrated insulin resistance and differences in glucose turnover in obese pigs when compared with normal, lean pigs.

In monogastric animals, acquired insulin resistance in adipocytes does not appear to be primarily linked to alterations in insulin

binding or in hexose transport abnormalities. The most likely explanation for the insulin resistance in large adipocytes resides in the limitation of glucose utilization by certain metabolic pathways (e.g. fatty acid synthesis, glucose oxidation) and in the loss of a dynamic adaptation of these pathways to glucose availability (Digirolamo 1981). Hence, an increase in glucose uptake due to the presence of insulin will be restricted by the limited capacity of the cells to metabolize glucose (Richardson and Czech 1978).

Insulin stimulates fatty acid synthesis and glucose utilization in ruminant adipose tissue, but the effects are generally smaller than those observed in monogastrics (Vernon 1980). This poorer response to insulin may be due to the low capacity for glucose metabolism in adipose tissue arising from the inability to use glucose for fatty acid synthesis and a low capacity for glucose oxidation other than via the pentose phosphate pathway.

Altered glucose metabolism was demonstrated in goats following administration of pharmacological doses of nicotinic acid (Thornton and Schultz 1980). Plasma glucose and insulin were elevated, glucose tolerance was reduced and there was an apparent resistance to insulin action.

The extent to which an excess of body fat affects glucose metabolism, as it does in monogastrics, is not clear. For the fat sheep in the present experiment, recycling rates of glucose, as a proportion of glucose utilization rate (McNiven 1984b) increased threefold from the high to the intermediate and low levels and fivefold from high to fasting. This could be a result of a decreased sensitivity to insulin. For the thin sheep, the proportion of glucose recycled for the three fed levels was similar and low and then increased threefold for the fasting level. Pool size and glucose space were significantly smaller ($P<0.001$) in the fat sheep compared with the thin. Although blood glucose concentrations for the fat sheep at fasting were temporarily maintained within the normal range by recycling, the lesser amount of glucose in the body would probably be rapidly depleted upon continued fasting or if there were a greater drain of glucose from the body than for maintenance alone.

It is evident that the presence of additional fat tissue in the body has a definite effect on insulin, glucose and FFA concentrations in ruminants. In fact, this fat tissue may contribute directly to the increased susceptibility of some animals to metabolic disorders.

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