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The breath-test for sheep: a possible means of identifying lean genotypes

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ABSTRACT

Calorimetry apparatus was used to measure CO₂ production and oxygen consumption in 2 Romney and 2 Southdown ewes. Each pair consisted of a long-lean ewe and of a short-fat ewe as determined from previous records of height, length, weight and ultrasonic back fat thickness. The respiratory exchange ratio (RER) was calculated as the ratio of CO₂ produced/d to O₂ consumed/d. When the 4 ewes were fed, the RER remained about 1, characteristic of the oxidation of glucose and short chain fatty acids. When the ewes were fasted for 4 to 5 days the RER declined in a straight linear fashion with time. This is consistent with the mobilisation of long chain fatty acids. The ewes of fatter genotypes exhibited significantly steeper regressions of RER on time.

Keywords Sheep; fat; lean; fasted; calorimetry; respiratory exchange ratio

INTRODUCTION

The number of lamb carcasses graded F (overfat) in the 1980-81 season was 308,414 (0.99%) and the MF (overfat mutton) grade contained 201,349 (2.85%) (New Zealand Meat Producers Board 59th Annual Report 1981). However a survey of British wholesaler and retailers indicated that they had a strong preference for lambs with a subcutaneous fat cover over the rib eye area of no more than 4 mm (Frazer, 1982). Frazer stated that if we were to fully meet this requirement within our 1982 export production, then around 25% would not qualify.

The present experiment was part of a study set up to investigate factors controlling lipid metabolism in sheep to discover differences in lipid metabolism between genetically fat and lean ewes, with a view to developing methods for identifying genetically superior sheep for use in breeding programmes.

A previous calorimetry experiment showed that metabolic rates of the experimental sheep did not differ on a metabolic body weight basis (LW¹ kg⁻⁰.⁷⁵), so that if there were real differences in their ability to produce adipose tissue (which had not then been determined) they had to be due to differences in the efficiency of utilisation of energy for maintenance and/or anabolism. Differences in the partitioning of energy between maintenance and growth may also have been involved. It was thought that perhaps the BMR was lower for the fat animals leaving more energy available for growth; or perhaps the lean sheep responded to periods of feed deficit by mobilising fat more quickly or in larger amounts than did the fat ones.

MATERIALS AND METHODS

Four 4-tooth (3.5 yr) ewes consisting of 2 Romney and 2 Southdown were selected from the Massey-DSIR research flock on the basis of weight-corrected back fat

<table>
<thead>
<tr>
<th>TABLE 1</th>
<th>Phenotypic Characteristics of the ewes.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breed</td>
<td>Ewe No.</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>Southdown</td>
<td>56</td>
</tr>
<tr>
<td>Southdown</td>
<td>73</td>
</tr>
<tr>
<td>Romney</td>
<td>313</td>
</tr>
<tr>
<td>Romney</td>
<td>323</td>
</tr>
</tbody>
</table>

a The mean value for fat depth assessed ultrasonically between ribs 12 and 13 on both sides.
b The percent deviation for the actual fat depth or body length from the value predicted using regression equations of log fat depth or log body length against log live weight for the Southdown flock (42 ewes) or the Romney flock (21 ewes).
c Length measured by calipers from the front of the brisket to a point on the pin bones (Tuber ischii).
thickness and body length with the objective of obtaining a long lean and a short fat ewe of each breed.

The sheep were weighed and their back fat thicknesses measured ultrasonically (Goeden et al., 1980). Similar values were obtained when the sheep were re-measured 2 months later (Peterson, 1983). It was considered that these measurements indicated the existence of differences in fatness between the 2 ewes of each breed which might have a genetic basis (Table 1). The experiment was a reversal or switchback trial (Brandt, 1938) in which sheep were alternately fasted and fed for 4 or 5 days while in the calorimeters. Only 2 sheep could be monitored at once so 1 sheep of each breed was 'fasted' while the other was fed, then after a restabilisation period of at least 10 days on the following diet the treatments were reversed.

The sheep were fed $1.25 \times$ maintenance (M) energy requirements, on the basis of LWT kg$^{-0.73}$ with $1 \times M$ as nuts and $0.25 \times M$ as hay. The 'fasted' sheep were not in fact starved but fed $0.25 \times M$ i.e. one-fifth of the normal ration in the same proportions.

<table>
<thead>
<tr>
<th>Period</th>
<th>Chamber 1</th>
<th>Chamber 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>323 Fasted</td>
<td>313 Fed</td>
</tr>
<tr>
<td>2</td>
<td>73 Fasted</td>
<td>56 Fed</td>
</tr>
<tr>
<td>3</td>
<td>73 Fed</td>
<td>56 Fasted</td>
</tr>
<tr>
<td>4</td>
<td>313 Fasted</td>
<td>323 Fed</td>
</tr>
</tbody>
</table>

Indirect calorimetry equipment (Holmes and McLean, 1974) was used to measure $O_2$ consumption and $CO_2$ production by the sheep and the ratio of $CO_2$ production to $O_2$ consumption (RER) was calculated. Since ruminants utilise acetate and glucose as their major substrates for energy storage and oxidation in the fed state (Bassett, 1975) the RER characteristic of the fed state should be about 1.0 since the RER associated with the complete oxidation of either glucose or acetate is 1.0. The oxidation of long chain fatty acids is associated with an RER of about 0.7, the RER decreasing as the length of the fatty acid increases (Table 3).

Assuming that lipolysis results in the mobilisation and oxidation of long chain fatty acids, the RER of a fasted animal should move towards 0.7. With this in mind the RER was calculated to express the fasting response.

Weights and back fat thicknesses were recorded before and after the trial.

### RESULTS

The RER of the fed sheep remained about 1 with a mean of $1.066 \pm 0.032$. The fasted sheep had a lower mean RER value of $0.950 \pm 0.074 \,(P<0.025, \text{Fig. 1}).$ The RER's of the fasted sheep declined with time, while those of the fed sheep did not. The slopes of individual regression lines of fed sheep did not differ ($P>0.25$) from the slope of a common regression line for the fed sheep. The slopes of individual regression lines of fasted sheep differed ($P<0.005$) but nevertheless a common regression line for fasted sheep was derived and found to have a different slope ($P<0.005$) from that of the fed sheep.

### Table 3 Theoretical respiratory exchange ratios for the oxidation of glucose and fatty acids.

<table>
<thead>
<tr>
<th>Oxidation of glucose</th>
<th>$CO_2/O_2$</th>
<th>RER</th>
</tr>
</thead>
<tbody>
<tr>
<td>$n =$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 acetate</td>
<td>6/6</td>
<td>1.0</td>
</tr>
<tr>
<td>3 propionic</td>
<td>6/7</td>
<td>0.86</td>
</tr>
<tr>
<td>4 butyric</td>
<td>4/5</td>
<td>0.80</td>
</tr>
<tr>
<td>5 valeric</td>
<td>10/13</td>
<td>0.77</td>
</tr>
<tr>
<td>•</td>
<td></td>
<td></td>
</tr>
<tr>
<td>16 palmitic</td>
<td>16/23</td>
<td>0.696</td>
</tr>
<tr>
<td>18 stearic</td>
<td>18/26</td>
<td>0.692</td>
</tr>
</tbody>
</table>

### FIG. 1. Slopes of regressions of RER on time for 4 ewes when underfed or when fed.

Southdown ewe 56 (fat)  
Southdown ewe 73 (lean)  
Romney ewe 313 (fat)  
Romney ewe 323 (lean)
The RER response to fasting was a straight-line relationship with correlation coefficients of -1.000, -0.987, -0.888 and -0.991. Correlations in the fed sheep were lower (0.934, 0.941, -0.191, and 0.475).

The fat group exhibited steeper regression slopes when fasted than the lean group ($P=0.002$). Within the fat group, Ewe 56 had a steeper regression slope ($P=0.03$) than Ewe 313. No differences were detected in weight or back fat thickness over the trial.

**DISCUSSION**

As was expected, the RER's of the fed sheep remained about 1, while the values for fasted sheep declined in a straight line with time. Since the RER's of fasted sheep did not stabilise at a lower level but were still falling linearly, it was assumed that a steady state of fasting metabolism was not reached and so the BMR could not be calculated. Extrapolation of the regressions indicated that 3 or more extra days of fasting may have been needed to reach a steady state of fasting metabolism in the lean sheep, assuming the responses remained linear and straight, and that an RER of about 0.7 is characteristic of fasting metabolism.

Modyanov (1967) starved sheep for 144 h and found that the heat production decreased until the end of the third day, then became somewhat stable and remained at the same level subsequently. The non-protein RER after 72 h of starvation was 0.72 which Modyanov said was typical of fat combustion. The fact that the sheep in the present experiment did not fall below 0.8 after 4 or 5 days can most likely be attributed to their intake of a quarter of their maintenance requirements daily compared to Modyanov's sheep which were starved. Nevertheless it indicates that the sheep in the present study did not reach a basal metabolic state.

Annison et al. (1967) reported RER for fed sheep of 1.03 and for 24 h fasted sheep of 0.94 but pointed out that these values were more than usually difficult to interpret in ruminants where a significant proportion of the total CO$_2$ output arises by anaerobic fermentation in the rumen.

The 2 groups of ewes differed in their response to fasting as measured by RER. The fat pair had steeper regression slopes which possibly indicated a greater rate of mobilisation of fat i.e., a more accelerated lipolysis than the lean sheep. Note that this does not indicate an earlier response.

Sidhu et al. (1973) found that basal lipolysis increased with fatness in homogenates of lamb adipose tissue and postulated that factors affecting deposition of fat compensate for increased lipolysis.

No conclusions can be made regarding BMR since it was not determined. Nevertheless it is evident that the response to fasting was an extremely straight linear decrease in RER with respect to time, which was steeper in the fat ewes, especially in the Southdown ewe 56.

It is hoped that the RER response can be used to accurately identify superior genotypes with respect to lean carcass characteristics.

**REFERENCES**


