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Comparison of herbage intakes estimated from *in vitro* or alkane-based digestibilities

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ABSTRACT

In 34 lactating ewes (10 fistulated at the rumen and abomasum), herbage intakes were calculated using herbage digestibilities estimated either *in vitro*, or by using the plant cuticular wax component pentatriacontane (C₃₅ alkane) as an internal marker. Alkane-based herbage intakes in intact ewes rose from 1672 g OM/d at 14d of lactation to 2798 g OM/d at 42d. In the fistulated ewes, the equivalent herbage intakes were 1736 and 2240 g OM/d respectively. Alkane-based and *in vitro*-based intakes were not significantly different at 14d of lactation in either intact or fistulated ewes. Later in lactation, alkane-based intakes were significantly higher in both groups of ewes. In the fistulated ewes, estimates of the proportion of digested OM apparently disappearing across the rumen and of the efficiency of microbial protein synthesis were calculated from abomasal flows of digesta and microbial protein. These results suggested that the alkane-based herbage intakes were the more accurate.

Keywords Intake, alkanes, *in vitro*, digesta flow, microbial protein, lactation, ewes

INTRODUCTION

In studies of herbage intake, the digestibility of consumed herbage is commonly estimated using an *in vitro* procedure (e.g. Tilley and Terry, 1963). This approach assumes that the single estimate of digestibility so obtained is applicable to all the test animals, regardless of their level of intake. By contrast, the long-chain hydrocarbons (alkanes) of plant cuticular waxes have been shown to accommodate individual differences in herbage digestibility, and have recently been used to estimate herbage intake (Mayes *et al.*, 1986; Dove *et al.*, 1989a). Dove *et al.* (1989a) compared alkane-based and *in vitro*-based estimates of herbage intake, but only had indirect evidence that the former estimates were more accurate. In the present study, intakes estimated using an *in vitro*-based and an alkane-based procedure were coupled with measurements of digesta flow and microbial protein production. From each estimate of intake, it then becomes possible to calculate the extent of rumen digestion and the efficiency of microbial protein synthesis, and thus gain insight into which is the more accurate estimate of intake.

MATERIALS AND METHODS

Estimates of herbage intake were obtained, at days 14, 28, 42 and 85 of lactation, in twenty-four lactating Greyface ewes (Border Leicester x Scottish Blackface) grazing perennial ryegrass pastures. Faecal outputs were estimated from the faecal dilution of a dose of 1g of chromium, administered once per day as Cr₂O₃. In ten lactating ewes of the same breed, fistulated at the rumen and abomasum, abomasal flows of DM and microbial protein were estimated for 3-day periods starting on days 14, 28 and 42 of lactation, using inert markers (Cr, Ru) and ³⁵Na₂SO₄ respectively. Marker infusion and digesta sampling procedures have been described elsewhere (Dove *et al.*, 1988). In these ewes, faecal output was estimated from faecal Ru concentrations. During the study, samples of ingested pasture were obtained using four Scottish Blackface male castrate sheep, fistulated at the oesophagus. Herbage digestibilities were then estimated either by a standard *in vitro* procedure (Tilley and Terry, 1963) or from the herbage and faecal levels of the cuticular wax alkane pentatriacontane (C₃₅ alkane). The faecal recovery of

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C_{35} was assumed to be 0.95, based on our previous measurements (Mayes *et al.*, 1986; Dove *et al.*, 1989b).

RESULTS AND DISCUSSION

At 14 d of lactation, the *in vitro*-based and alkane-based estimates of herbage digestibility were 0.798 ± 0.0057 and 0.829 ± 0.0195 respectively. However, this difference in digestibility did not generate significant differences in estimated herbage intake in either intact or fistulated ewes (Table 1)

TABLE 1 Alkane-based and *in vitro*-based herbage intakes in lactating ewes

	14d	Stage of lactation		
		28d	42d	85d
Intact ewes				
Alkane-based	1672	1713***	2798***	2566***
<i>In vitro</i> -based	1581	1410	2070	1911
s.e.d	111.1	31.2	57.1	51.7
Fistulates				
Alkane-based	1736	1639**	2240***	-
<i>In vitro</i> -based	1776	1539	1763	-
s.e.d	104.6	28.8	71.7	-

** $P < 0.01$; *** $P < 0.001$, paired t-test

At 28d, the *in vitro* digestibility was 0.771 ± 0.0131 , while the alkane-based digestibility was 0.815 ± 0.0057 . By 42d, the difference was more marked (*in vitro* 0.768 ± 0.0099 ; alkane 0.860 ± 0.0094), but was slightly smaller by 85d (*in vitro* 0.760 ± 0.0041 ; alkane 0.816 ± 0.0060).

These differences in estimated digestibility resulted in significant differences between the two estimates of herbage intake at 28, 42 and 85d of lactation in intact ewes (Table 1). Intakes by the fistulated ewes were essentially similar; alkane-based intakes were significantly higher at both 28d and 42d of lactation. At 42d of lactation, which might be expected to be near peak intake, the alkane-based intakes were 35.2% and 27.1% higher than the *in vitro*-based intakes, in intact and fistulated ewes respectively.

In the fistulated ewes, the digesta DM flows at 14, 28 and 42d of lactation were 1146, 1137 and 1318

g/d respectively. The equivalent microbial protein flows were 39.8, 41.7 and 51.7 g N/d. At 14d, the calculated proportions of digestible OM apparently disappearing in the rumen (OMADR) were 0.571 and 0.584 for the alkane-based and the *in vitro*-based intakes, respectively. The respective efficiencies of microbial protein production were 37.1 and 40.1 g microbial N/kg OMADR per day. Neither of these differences was significant. At 28d of lactation, the proportion of OMADR calculated from the alkane-based intake (0.568) was significantly higher than that derived from the *in vitro*-based intake (0.536; s.e.d. 0.0087, $P < 0.01$). The microbial efficiency for the alkane-based estimate (40.3 g N/kg OMADR) was similar to those at 14d but significantly less than the value for the *in vitro*-based intake (47.3; s.e.d. 2.22, $P < 0.05$). These trends were more pronounced by 42d of lactation where, compared with the *in vitro*-based estimate, the alkane-based estimate of the proportion of OMADR was significantly higher (0.581 v. 0.443; s.e.d. 0.0155, $P < 0.001$) and the microbial efficiency was significantly lower (44.3 v. 69.6 g N/kg OMADR; s.e.d. 4.53, $P < 0.01$) but consistent with earlier periods.

Over the period of lactation studied, the alkane-based herbage intakes in both intact and fistulated ewes showed a marked increase between 14d and 42d of lactation, as might be expected in lactating ewes. By contrast, *in vitro*-based intakes rose much less in intact ewes and not at all in the fistulates. Similarly, the overall mean proportion of OMADR (0.574) and the mean microbial efficiency (41.2 g N/kg OMADR) for alkane-based intakes, were similar to the respective mean values of 0.61 ± 0.049 and 37.8 ± 10.62 g N/kg OMADR cited by the Agricultural Research Council (1984). By contrast, the corresponding values for *in vitro*-based intakes either fell progressively (proportion of OMADR) or rose sharply (microbial efficiency) over the course of lactation. While a progressively increasing rate of passage during lactation could result in a lower proportion of OMADR, it seems unlikely that this could account for the apparent 74% increase, between 14d and 42d of lactation, in the efficiency of microbial protein synthesis calculated from *in vitro*-based intakes (Agricultural Research Council, 1984). Hence, from the consistency of the C_{35} alkane-based results, both during lactation and in relation to published values, we have therefore concluded that these were the more accurate estimates

of herbage intake. However, we also urge that similar studies should be conducted with more animals, in which faecal outputs are determined from the dilution of orally administered synthetic alkanes, which obviates the need for assumptions about faecal alkane recoveries (Mayes *et al.*, 1986; Dove *et al.*, 1989a).

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