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# Use of n-alkanes to estimate the intake of beef heifers on natural grassland in southern Brazil

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## Introduction

The technique of alkanes to estimate dry matter intake (DMI) by wild and domestic herbivores has advanced considerably in the last 20 years (Dove and Mayes, 2006). Alkanes are long chain, saturated hydrocarbons present in the plant cuticle. They are relatively indigestible in the gastrointestinal tract and can be recuperated in the faeces. Compared to other markers normally used (*e.g.* chromium oxide, ytterbium), it is possible to determine simultaneously the external and internal marker in a unique analysis and to estimate digestibility, faecal excretion, DMI and diet composition (Dove and Mayes 1991). This is a great advantage of this technique to support studies of plant-animal interactions in rangeland environments.

## Methods

The experiment was carried out at the experimental station of the Federal University of Rio Grande do Sul (lat 30°05'S, long 51°40'W and 46 m a.s.l.), southern Brazil, in an representative area of Campos grassland (Boldrini *et al.* 2010). Since 1986, this 31 ha experimental area has been managed under continuous grazing and different forage allowances (FA) for beef cattle. The experiment was arranged in randomized blocks with two replicates and eight experimental units (EU). Treatments were FA of 4, 8, 12, and 16 kg DM/100 kg of liveweight. These FA levels were adjusted every 28 days using the put-and-take technique. The experimental period was divided in two seasons: summer (02/16 to 02/27/2009) and spring (10/11 to 10/22/2009).

The experimental animals were 15 months old crossbred (Angus and Hereford X Nelore) heifers. Three animals per EU were dosed twice daily (at 8 am and 4 pm) with cellulose pellets containing 200 mg of dotriacontane (C<sub>32</sub>) and this was administered for 12 days in each season. From the 7<sup>th</sup> to the 12<sup>th</sup> day of dosing, faecal samples were collected per rectum simultaneously to the administration of C<sub>32</sub>. To estimate the amount of forage consumed by the animals, the simulated grazing technique was applied between the 7<sup>th</sup> and 10<sup>th</sup> day of C<sub>32</sub> administration period.

The determination of n-alkanes in forage and in faeces samples followed the methodology described by Dove and Mayes (2006) in the range of C-chain between 27 and 35.

The DMI was then estimated according to the equation of Dove and Mayes (1991); DMI were calculated from the ratio of alkane homologous pairs C<sub>31:32</sub> and C<sub>33:32</sub>, with or without correction for faecal recovery. The values of faecal recovery were: C<sub>31</sub> = 0.757; C<sub>32</sub> = 0.785; C<sub>33</sub> = 0.809 as suggested by Dove and Mayes (1991).

ANOVA was used to test the effects of fecal recovery rate (FRR), FA and its interaction with DMI calculated by the homologue pairs of C<sub>31:32</sub> and C<sub>32:33</sub>, with or without correction by FRR. We also tested the regression models (linear or quadratic) for these parameters. The analyses were performed by JMP<sup>®</sup> v.9.

## Results

Regardless of the season, the estimations of DMI from the pair C<sub>31:32</sub> were affected by faecal recovery correction ( $P < 0.0001$ ) and by FA ( $P < 0.0001$ ) in the summer. Data also showed an interaction between FRR correction and FA in spring ( $P < 0.0001$ ). For the C<sub>32:33</sub> pair, no difference was observed between the DMI data corrected and non corrected by FRR. The DMI estimations with C<sub>32:33</sub> pair were unaffected by the interaction between the factor of correction and FA.

The DMI data of heifers, estimated by the pairs of C<sub>31:32</sub> and C<sub>32:33</sub> with or without correction by FRR, and the regression analysis with FA are showed in Table 1. The DMI estimated using the pair C<sub>31:32</sub> was on average 51% lower than the DMI calculated using C<sub>31:32</sub> when corrected by the FRR, whereas the difference estimated using the pair C<sub>32:33</sub> was on average 0.4% greater than DMI using FRR. We fitted a quadratic or linear regression of DMI against FA. However, the models generated between the DMI and estimated C<sub>31:32</sub> in the summer were not significant.

The DMI data estimated using the C<sub>31:32</sub> pair was affected ( $P < 0.0001$ ) by the correction in both seasons. This indicated that alkanes of this pair were unresponsive to similar FRR, limiting its application in the DMI estimation as suggested by the theory of the double alkane (Dove and Mayes 1991). The DMI corrected data were 50% lower compared to the non-corrected data. In addition to the effect of correction by the FRR, it is important to verify the interaction effect between FRR and the forage allowance because this correction can vary dramatically. The interaction effects showed that the C<sub>32:33</sub> pair was unaffected by

**Table 1. Daily DMI (% LW) by heifers estimated with the pair of alkanes C<sub>31:32</sub> e C<sub>32:33</sub>, without (NC) or with (WC) correction by the rate of fecal recuperation and a regression analysis of the levels of forage allowance on natural grassland in southern Brazil.**

Alkane pairs	Forage allowance (% LW)				Model	P value	R <sup>2</sup>	Standard error
	4	8	12	16				
<b>Summer</b>								
C <sub>31:32</sub> NC	0.93	1.57	2.32	1.83	y=0.90+0.07x	0.1627	0.41	0.045
C <sub>31:32</sub> WC	0.49	0.86	1.38	1.06	y=0.48+0.04x	0.2022	0.41	0.031
C <sub>32:33</sub> NC	1.99	2.44	2.83	2.76	y=1.85+0.06x	0.0274	0.77	0.020
C <sub>32:33</sub> WC	2.06	2.55	2.95	2.88	y=1.92+0.07x	0.0264	0.77	0.022
<b>Spring</b>								
C <sub>31:32</sub> NC	1.92	2.29	3.06	3.11	y=1.61+0.15x	0.0003	0.94	0.168
C <sub>31:32</sub> WC	0.87	1.20	1.20	1.33	y=0.87+0.04x	0.0477	0.60	0.016
C <sub>32:33</sub> NC	1.95	2.04	2.38	3.24	y=1.48+0.14x	0.0137	0.77	0.037
C <sub>32:33</sub> WC	1.82	2.22	2.14	3.06	y=1.54+0.12x	0.0607	0.64	0.048

the isolated correction and by the interaction between the factor correction and forage allowance.

In theory, the different levels of FA determine variations in the abundance of feed which affects the DMI. Even in optimum feeding conditions like the summer period, these could vary in natural grasslands in southern Brazil and this probably promotes changes in DMI. The regression analysis between forage allowance and DMI calculated with the pair C<sub>32:33</sub> without FRR showed a good fit, whereas with the C<sub>31:32</sub> pair this response was not confirmed.

## Conclusion

The evidences of this study allowed identifying that the

alkane technique should be paired with the dosed C<sub>32</sub> as far as estimates of DMI is concerned on natural grassland in southern Brazil, is the n-alkane C<sub>33</sub>.

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