

ORIGINAL ARTICLE

Ruminants

Influence of pre-grazing herbage mass on bite mass, eating behaviour, and dairy cow performance on pasture

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Abstract

Knowledge about individual daily herbage dry matter (DM) intake (DMI) helps identifying efficient dairy cows and adapting supplementation better to herbage intake and nutrient requirements of grazing dairy cows. With the aid of behavioural characteristics, raw data recorded with the RumiWatch (RW) system and processed with the RW converter 0.7.3.31 (C31), estimation of herbage DMI may be possible. First, C31, which allows differentiation of prehension bites and mastication chews, was validated through direct observation of behavioural characteristics and compared to the previous RW converter 0.7.3.11 (C11). Further, the influence of a low and high pre-grazing herbage mass (HM), with the same target herbage allowance (HA), on bite mass, DMI, number of prehension bites, and milk production was investigated. In total, 24 lactating Holstein cows were pairwise allotted to one of two HM treatments. The cows received a new pasture paddock twice per day with a daily target HA of 22 kg DM per cow/day. On average, low HM (LHM) and high HM (HHM) paddocks had an HM of 589 and 2288 kg DM/ha, respectively, above 6.7 click units (1 CU = 0.5 cm). Overall, LHM cows produced 2.7 kg/day more milk and 2.5 kg/day more energy-corrected milk, had the same herbage DMI and a similar prehension bite mass. The averaged bite mass per week was 0.49 g DM/bite (LHM) or 0.47 g DM/bite (HHM), respectively. A longer eating time (617 vs. 559 min/day) and a shorter rumination time (297 vs. 365 min/day) were observed for the LHM cows compared with the HHM cows. The validation of the RW showed similar results for C11 and C31 apart from prehension bites, where C31 showed a mean absolute deviation of 12.4%. Pre-grazing HM had no effect on relevant behavioural characteristics for prospective intake estimation, namely, bite mass and number of prehension bites.

KEYWORDS

feed intake, mastication chews, prehension bites, rumination, RumiWatch converter, validation

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1 | INTRODUCTION

Decreasing milk prices and gross margin per kg milk in recent years (Reijs et al., 2013) represent a challenge in milk production for dairy farmers. From an economic point of view, the proportion of grazed herbage in the ration might therefore increase. Dillon et al. (2005) showed a reduction of 2.5 € cents/L in milk production costs for every 10% increase in grazed grass on a dry matter (DM) basis in the diet. Gazzarin et al. (2021) found a 26% lower cost price for full grazing cows compared to a control group. This economic advantage, as well as better monitoring of intake during grazing—which could help to improve milk yield and feed efficiency—might motivate farmers to practice grazing (Oudshoorn et al., 2013).

To gain knowledge of energy intake, feed efficiency, and the nutritional status of grazing dairy cows, estimation of herbage dry matter intake (DMI) is essential. Different methods can be used to estimate herbage DMI of grazing dairy cows (Hellwing et al., 2015), for example, marker methods, methods based on animal performance, or through the pre- and post-grazing herbage mass (HM). However, herbage DMI estimation for grazing dairy cows by any of these methods is laborious, expensive, not applicable to daily practice conditions, or gives only an average intake of a group of grazing cows.

An alternative approach to estimate the herbage intake of individual dairy cows on pasture may be by aid of behavioural characteristics. Oudshoorn et al. (2013), for example, calculated grass intake by using the grazing time and bite frequency of individual animals. Theoretically, DMI estimation is based on the total number of eating chews and the corresponding bite mass. However, as bite mass on pasture shows large variation (0.28 up to 0.96 g of DM/bite) (Piña et al., 2020) and only prehension bites serve for herbage intake, it may be important to differentiate between prehension bites and mastication chews performed during eating. Several sensing devices have recently been developed for automatic recording of behavioural characteristics. According to the review of Andriamandroso et al. (2016), devices for jaw movement detection can be classified into five groups: jaw switches, pressure sensors, microphones, accelerometers, and electromyography. A combination of a pressure sensor and a triaxial accelerometer is used by the RumiWatch (RW) system (RWS; Rombach et al., 2018). Like the IGER Behaviour Recorder (Andriamandroso et al., 2016) and the acoustic method described by Laca and WallisDeVries (2000), the RWS with the RW converter 0.7.3.31 (C31) enables the detection of every jaw movement, as well as differentiation between eating chews in prehension bites and mastication chews performed during eating. Consequently, it might be possible to estimate herbage intake more precisely based on the number and mass of prehension bites. Several factors, like the animal's anatomy (including mouth and body size), as well as sward factors (such as height and bulk density) have been mentioned to influence bite mass (Rook, 2000) and even duration on pasture (Piña et al., 2020). Furthermore, bite mass is higher within the first 60 min on pasture compared to afterward (Alvarez-Hess et al., 2021).

In the present study, the first objective was to validate the new C31 through direct observations and, additionally, to compare the results with those of the previous RW converter 0.7.3.11 (C11). Purely intuitively, it would be expected that cows in high pre-grazing HM could ingest more herbage per prehension bite, per grazing bout and day compared to cows that grazed in low pre-grazing HM. Moreover, according to Pérez-Prieto et al. (2013), the effect of HM on DMI seems inconsistent under strip and rotational grazing management. Therefore, the second objective of the present study was to investigate the effects of a low and high pre-grazing HM at the same target herbage allowance (HA) on bite mass, number of prehension bites, DMI, and milk production. In addition, in view of the prospective intake estimation, the relationship between the number of ingestive bites and herbage DMI was investigated.

2 | MATERIALS AND METHODS

2.1 | Experimental design, animals and housing

The study was carried out at the Agroscope experimental farm in Posieux, Switzerland (46° 46' 1.0" N, 7° 6' 18.7" E) from August 31 until September 20, 2015 (3 weeks). All experimental procedures were in accordance with the Swiss guidelines for animal welfare and approved (No. 2015_11_FR) by the Animal Care Committee of the Canton Fribourg, Switzerland. All experimental cows passed a medical check-up. The experiment was performed as a balanced-block design with two feeding treatments. The trial lasted 21 days, with 14 days of adaptation of pasture-experienced dairy cows and a 7-day measuring period. Twenty-four Holstein and Red Holstein cows were pairwise allotted to the two treatments based on body weight (BW), milk yield, days in milk (DIM), and lactation number, whereby all parameters were equivalent. At the beginning of the experiment, cows on high HM (HHM) had an average BW of 649 (SD 63) kg, were 196 (SD 46) DIM, had on average 2.5 (SD 1.7) lactations, and produced 23.3 (SD 5.0) kg of milk/day. The cows on low HM (LHM) had an average BW of 660 (SD 42) kg, were 187 (SD 63) DIM, had on average 2.6 (SD 1.8) lactations, and produced 22.6 (SD 4.6) kg of milk/day.

Cows grazed 19 h/day, had free access to water, and were not supplemented in the barn. Both groups were kept on pasture from 0730 to 1500 h and from 1730 to 0500 h the following morning. In the meantime, cows were kept in a loose housing system, where cows were milked, sampled, and the alkane capsules were administered. The treatments consisted of two different pre-grazing HM, either 2288 kg DM/ha for HHM ($n = 14$) or 589 kg DM/ha for LHM ($n = 14$), but with the same targeted HA. The pre-grazing HM of LHM and the sward height of 10 click units (CU; measured with an electronic rising plate metre [Jenquip, Feilding, New Zealand]; 1 CU = 0.5 cm) were equivalent to the pre-grazing sward surface height recommendations for continuous stocking systems (Mosimann et al., 1999). The HM of HHM should be representative for a rotational grazing system with 1300 and 1600 kg

DM/ha (O'Donovan & Delaby, 2016) but was higher than intended, with a sward height of 18 CU (measured with an electronic rising plate metre) and an average of 2288 kg DM HM/ha ($n = 14$). During the measurement period, 14 grazing plots were used for each group, and cows were given access to a new plot after every milking.

To achieve the different pre-grazing HM, experimental grazing paddocks were cut, 10.5 (SD 1.1) days for LHM or, for HHM, 21.5 (SD 1.1) days previously, to a sward height of 8.5 (SD 0.75) CU (measured with an electronic rising plate metre). Both groups were provided with the same HA targeted at 22 kg DM/cow per day above 6.7 CU. Before each meal, HM was measured by cutting a 1 m wide stripe with a bar mower (Bucher Landtechnik AG, Niederweningen, Switzerland, Rekord 38). The pre- and post-grazing HM was cut on average above 6.7 CU, corresponding to 43 mm uncompressed sward surface height according to Mosimann et al. (1999). Between the two treatments, the lengths of the cut stripes differed (HHM: 7.4 [SD 1.0] m and LHM: 11.8 [SD 4.3] m). The weighed fresh HM of the cut strips and the analysed herbage DM content of the previous day were used to calculate the HM; thereby, the size of the paddocks were calculated and fenced off to achieve the attended HA. New paddocks were offered twice per day after each milking. Post-grazing sward height was measured after each meal with the aid of the electronic rising plate metre. The cows received no supplementation of minerals or vitamins during the experiment. On average, the paddock size, measured for each meal (twice a day) during the experiment, was 554 (SD 112) m² for HHM and 4135 (SD 3187) m² for LHM. Pastures with an age of >2 years were used. With seven samples for each treatment, LHM and HHM, the sward composition and chemical composition of herbage were ascertained. The swards were composed of different grasses: 72 (SD 4.1) % fresh matter (FM) (mainly *Lolium perenne*), legumes 26 (SD 4.4) % FM (mainly *Trifolium repens* and *T. pratense*), and forbs 2 (SD 0.63) % FM (mainly *Taraxacum officinale* and *Plantago lanceolata*). Table 1 contains information on the chemical composition of the herbage fed during the measuring period.

2.2 | Sample collection and data recording

Milk yield was measured twice daily at 0500 and 1600 h during milking in the milking parlour (Fullwood, Arnold Bertschy AG) with a Pulsameter (LMS GmbH). Furthermore, milk composition was analysed on days 2, 4 and 6 during the measuring week. Aliquot milk samples per day were made with samples from the morning and evening milking and finally stored in a tube containing a Broad-Spectrum Microtab II (Gerber Instruments AG), at 8°C for the further analysis of milk fat, protein, and lactose contents and somatic cell counts.

Individual herbage intake on pasture was estimated using the n-alkane double indicator method (Mayes et al., 1986). From 6 days before until the penultimate day of the measuring week, a gelatine capsule (HGK-17-60 sl; Capsula GmbH) was administered twice daily (at 0530 and 1630 h) to the cows. Each gelatine capsule contained 0.5 g of dotriacontane (Minakem Beuvry Production S.A.S.) on a

TABLE 1 Chemical composition of herbage with a low ($n = 14$) and a high ($n = 14$) pre-grazing herbage mass^a

| Item | LHM | | HHM | |
|--|---------|-----------------|---------|-----------------|
| | Average | SD ^b | Average | SD ^b |
| Dry matter (g/kg of wet weight) | 174 | 30.2 | 167 | 22.0 |
| Analysed nutrient and mineral composition (g/kg of DM) | | | | |
| Organic matter | 902 | 4.1 | 902 | 3.1 |
| Crude protein | 240 | 26.4 | 184 | 9.9 |
| Acid detergent fibre | 188 | 13.5 | 218 | 10.9 |
| Neutral detergent fibre | 328 | 28.2 | 359 | 17.7 |
| Crude fibre | 167 | 12.9 | 195 | 8.8 |
| Calcium | 10.6 | 1.72 | 10.4 | 1.14 |
| Phosphorus | 4.2 | 0.60 | 3.6 | 0.39 |
| Magnesium | 2.9 | 0.26 | 2.7 | 0.22 |
| Sodium | 0.5 | 0.29 | 0.5 | 0.17 |
| Potassium | 33.0 | 4.60 | 34.3 | 3.69 |
| Calculated energy supply ^c (kg of DM) | | | | |
| NE _L (MJ) ^d | 6.6 | 0.14 | 6.3 | 0.07 |
| Analysed n-alkane contents (mg/kg of DM) | | | | |
| Dotriacontane | 6.7 | 0.59 | 6.3 | 0.51 |
| Tritriacontane | 87.4 | 9.97 | 83.7 | 8.68 |

Abbreviations: DM, dry matter; HHM, high herbage mass; LHM, low herbage mass.

^aHigh and low pre-grazing herbage mass were 2288 and 589 kg DM/ha, respectively.

^bStandard deviation.

^cAccording to Agroscope (2015).

^dNet energy lactation.

carrier of 4.5 g dried fruit pomace. During the measuring week, faeces of each cow was spot-sampled indoors, after morning milking (between 0600 and 0630 h), and stored for further analysis at -20°C. The faeces samples, taken to determine the content of alkanes with or without a stimulus, were pooled for each cow and the measuring week. The herbage sampling started 24 h before faeces sampling and ended 24 h earlier. Herbage samples were collected twice daily, in the morning (0800 h) and the afternoon (1700 h) to determine the content of alkanes. The collection was carried out by mimicking the grazing behaviour of the cows on pasture. Concretely, the experimental cows were followed one by one and their grazing behaviour was observed. Subsequently, small samples of the most likely grazed herbage by each cow, in relation to the botanical composition and grazing depth, were cut with a battery grass shearer (Gardena, Husqvarna, Schweiz AG). These samples were chopped and stored at -20°C for further analysis.

For behavioural records, the cows wore the RumiWatch halter (RWH; Itin + Hoch GmbH, Liestal, Switzerland; Rombach et al., 2018) 4 days before and until the end of the measuring period. Direct

observations were performed to validate the RW converter 0.7.3.31 (C31, Itin+Hoch GmbH) output for ingestive and rumination behaviour. The observation was done on days 1, 3 and 5 during the measuring week for each cow. A total of 72 10-min observation sequences (1 measuring period \times 3 observations \times 24 cows) were generated for validation and performed by one and the same trained observer as by Rombach et al. (2018). A detailed description of the behavioural characteristics validated through observation is provided in Table 2.

2.3 | Laboratory analysis

Milk samples were analysed by a Combi-Foss FT+ (Foss). This technique uses Fourier transform infrared spectrometry (Milkoscan FT+, Foss) to detect the contents of milk fat, protein, and lactose. Fluorescence flow cytometry (Fossomatic FC200, Foss) was used to measure the number of somatic cells in the milk samples.

Fresh herbage samples were first dried at 60°C for approximately 15 h and then at 105°C for 3 h to determine the DM. The frozen herbage samples were lyophilised (Delta 1-24 LSC, Christ) and subsequently milled through a 1.0 mm screen (Brabender mill with titanium blades; Brabender GmbH and Co. KG). Afterward, lyophilised herbage subsamples were dried for 3 h at 105°C to determine DM and subsequently incinerated by 550°C until a stable mass was reached to determine the ash content (procedure 942.05; AOAC International, 1995). Mineral residues in the herbage ash were dissolved by nitric acid and analysed for Ca, P, Na, Mg and K with inductively

coupled plasma optical emission spectrometry (ICP-OES Optima 7300 DV; PerkinElmer) based on ISO (2009) method 27085. The contents of n-alkanes HC32 and HC33 (trtriacontane, C33H68) were analysed as described by Thanner et al. (2014). The N content was analysed using the Dumas method (ISO, 2008; method 16634-1) on a C/N analyser (Trumac CNS, Leco Instruments), and the results were multiplied by 6.25 to generate the crude protein (CP) content. The content of ADF (acid detergent fibre) (AOAC International, 1995; procedure 973.18), neutral detergent fibre (NDF) (AOAC International, 1995; procedure 2002.4), and crude fibre (AOAC International, 1995; procedure 978.10) for the herbage samples was analysed using a Gerhardt Fibertherm (Gerhardt GmbH & Co. KG). For NDF analysis, heat-stable amylase and sodium sulfite were added. Correction for residual ash, obtained after 2 h of incineration at 550°C, was done for ADFom and NDFom.

2.4 | Data calculation and statistical analysis

Crude fibre was used to estimate digestible organic matter for the calculation of net energy for lactation (NEL) according to Agroscope (2015).

To detect the accuracy of C11 and C31 compared to direct observation, the mean absolute deviation percent (MADP) of each behavioural characteristic was calculated (Rombach et al., 2018):

$$\text{MADP}_x = \frac{\sum_{k=1}^N |\text{vis}_x^k - \text{RW}_x^k|}{\sum_{k=1}^N |\text{vis}_x^k|} \quad (1)$$

TABLE 2 Definitions of the used behavioural characteristics

| Behaviour | Definition |
|-----------------------------|---|
| Prehension bites | Bites to gather or sever a bunch of feed (herbage) during feed intake. |
| Prehension time | Time spent for prehension bites, including interruptions between prehension bites as long as 5 s. |
| Mastication chews | Chews performed during eating for mechanical breakdown of the ingested material to finer particles. |
| Mastication chews head up | Chews performed during eating for mechanical breakdown of the ingested material to finer particles after moving the head 10 or more cm above the herbage surface. |
| Mastication chews head down | Chews performed during eating for mechanical breakdown of the ingested material to finer particles when the head is lower than 10 cm above the herbage surface. |
| Mastication time | Time spent for mastication chews including interruptions between chews of up to 5 s. |
| Mastication time head up | Time spent for mastication chews performed with the head up including interruptions between chews up to 5 s. The cow is holding the head upright. |
| Mastication time head down | Time spent for mastication chews performed with the head down including interruptions between chews of up to 5 s. The cow is holding the head lowered near the herbage sward. |
| Eating chews | Total amount of prehension bites and mastication chews during eating. |
| Eating time | Time spent for eating chews including interruptions between eating chews of up to 5 s. |
| Rumination chews | Chews with the molars during rumination for mechanical breakdown of regurgitated materials to finer particles. |
| Rumination time | Time spent for rumination chews including chewing interruptions of up to 5 s. |
| Bolus count | A regurgitated mass of cud, which is swallowed again after chewing, counted when mass of cud is regurgitated. |
| Chews per bolus | Chews performed during rumination between the regurgitation and swallowing of one bolus. |

where vis_x is the direct observed behavioural characteristic x observation ($k = 1$ to N) and RW_x is the behavioural characteristic x measured automatically with the RWH and evaluated by C11 or C31, respectively, corresponding to the direct observation sequence ($k = 1$ to N). Herbage intake calculation is based on equations proposed by Mayes et al. (1986).

Equation (2) was used to calculate the daily herbage DMI of every experimental cow.

$$\text{herbage DMI} = \frac{\frac{F_{33}}{F_{32}} \times A_{32}}{H_{33} - \frac{F_{33}}{F_{32}} \times H_{32}} \quad (2)$$

where herbage DMI represents the daily herbage DMI (kg); F_{33} and H_{33} are the concentrations of tritriacontane (C33H68; HC33) (mg/kg DM) in faeces and herbage; F_{32} and H_{32} are the concentrations of HC32 (mg/kg DM) in faeces and herbage, respectively; and A_{32} is the daily dose of HC32 (mg/day DM), administered via the gelatine capsules.

Bite mass was calculated by dividing the herbage DMI by the number of prehension bites or eating chews, respectively, performed on pasture and recorded by the RWS. For the intake rate, the herbage DMI was divided by the eating time on pasture.

For statistical analyses, Systat 13 (Version 13.0, SYSTAT Software) was used. Data for milk yield, milk composition, HA, pre-grazing HM, grazing areas, and ingestive as well as rumination activities were collected over 7 days and averaged per cow and the measuring week. These averages and the average herbage DMI estimated for 7 days by the n-alkane double indicator method were analysed with the following linear mixed model:

$$Y_{ijk} = \mu + \tau_i + P_j + \varepsilon_{ijk} \quad (3)$$

where Y_{ijk} is the response variable, μ is the overall mean, τ_i is the fixed effect of the treatment i ($i = \text{HHM, LHM}$), P_j is the random effect of the cow pair j (1,...,12), and ε_{ijk} is the random error. Not normally distributed data were transformed using logarithmic transformation. If the normal distribution was not achieved by transformation, as well as for the data of the validation of C11 and C31, the Kruskal–Wallis test was used. A p -value of ≤ 0.05 was considered significant, whereas p -values between $0.05 < p < 0.10$ were considered to indicate a trend.

3 | RESULTS

Due to technical problems with 2 of the 24 RWH, 22 of the 1-week RW records and those of the corresponding cows could be used for further evaluation. However, the milk and intake data of all cows could be used to compare the two treatments.

3.1 | Validation of the converters

Between the two converters, C11 and the refined C31, no significant differences were found based on MADP (Table 3) for

TABLE 3 Comparison of mean absolute deviation percent of converter C11 (0.7.3.11) and C31 (0.7.3.31) to direct observation (reference) ($n = 72$)

| Behaviour | C11 | C31 | SE ^a | p -value |
|--|------|------|-----------------|------------|
| Ingestive behaviour (mean absolute deviation percent) | | | | |
| Eating time ^b | 6.4 | 6.0 | 3.09 | 0.85 |
| Eating chews ^b | 9.7 | 11.2 | 2.09 | 0.17 |
| Prehension bites ^b | 40.1 | 12.4 | 4.21 | <0.001 |
| Mastication chews head down | – | 51.5 | – | – |
| Sum of mastication chews & prehension bites head down | – | 13.8 | – | – |
| Mastication & prehension time head down | – | 7.0 | – | – |
| Mastication chews head up | – | 67.7 | – | – |
| Mastication time head up | – | 73.6 | – | – |
| Ruminating behaviour (mean absolute deviation percent) | | | | |
| Rumination time ^b | 2.3 | 2.1 | 12.76 | 0.95 |
| Rumination chews ^b | 4.2 | 3.9 | 12.75 | 0.85 |
| Bolus count ^b | 8.9 | 8.9 | 11.64 | 1.00 |
| Chews per bolus ^b | 11.5 | 10.4 | 14.68 | 0.75 |

^aStandard error.

^bNonparametric test (Kruskal–Wallis) for statistical analysis.

eating time ($p = 0.85$), number of eating chews ($p = 0.17$), rumination time ($p = 0.95$), number of rumination chews ($p = 0.85$), number of rumination boluses ($p = 1.0$), or chews per bolus ($p = 0.75$). Compared to C11, C31 showed a lower MADP for the number of prehension bites ($p < 0.001$). In contrast to C11, C31 enables differentiation between mastication chews and prehension bites. Therefore, the MADP for additional behavioural characteristics, such as the number of mastication chews with head down, mastication chews and prehension bites with head down, mastication chews with head up, and time spent with these characteristics, were calculated only for C31. Compound bites (simultaneous mastication chews and prehension bites), which occur frequently according to Galli et al. (2018), are not detected as such by the C31. It is assumed that compound bites are classified as prehension bites, but this assumption has not been verified.

3.2 | Eating and rumination behaviour

Cows grazing on LHM, compared to HHM, ate longer ($p = 0.004$), performed more mastication chews with the head down ($p = 0.03$), and had a lower eating frequency ($p = 0.002$) (Table 4). In addition, compared to HHM cows, LHM cows spent more time with mastication and prehension with head down ($p = 0.003$). No

TABLE 4 Effect of low (LHM) and high (HHM) pre-grazing herbage mass^a on ingestive and rumination behaviour evaluated using C31 ($n = 22$)

| Item | LHM | HHM | SE ^b | p-value |
|---|--------|--------|-----------------|---------|
| Ingestive behaviour performed on pasture | | | | |
| Eating time (min) | 617 | 559 | 10.5 | 0.004 |
| Eating chews (n) | 42,950 | 42,220 | 941.9 | 0.60 |
| Prehension bites (n) | 32,366 | 32,244 | 1392.5 | 0.95 |
| Mastication chews head down (n) | 6087 | 4055 | 816.7 | 0.03 |
| Mastication + prehension time head down (min) | 540 | 469 | 12.3 | 0.003 |
| Mastication chews head up (n) | 4497 | 5920 | 650.4 | 0.16 |
| Mastication time head up (min) | 77.0 | 90.4 | 9.46 | 0.35 |
| Eating frequency (n /min) | 69.7 | 75.5 | 1.10 | 0.002 |
| Ruminating behaviour performed on pasture | | | | |
| Rumination time (min) | 297 | 365 | 7.1 | <0.001 |
| Rumination chews (n) | 18,436 | 23,625 | 721.9 | <0.001 |
| Rumination rate (n /min) | 68.4 | 71.4 | 1.24 | 0.08 |
| Bolus count (n) | 365 | 441 | 14.3 | 0.005 |
| Chews per bolus (n /bolus) | 50.8 | 54.1 | 1.49 | 0.15 |

Abbreviations: HHM, high herbage mass; LHM, low herbage mass.

^aHigh and low pre-grazing herbage mass were 2288 and 589 kg DM/ha, respectively.

^bStandard error.

differences were observed for the number of eating chews ($p = 0.60$), the number of prehension bites ($p = 0.95$), the number of mastication chews with head up ($p = 0.16$), and time spent with mastication with head up ($p = 0.35$). Fewer rumination chews ($p < 0.001$), a shorter rumination time ($p < 0.001$), and a lower number of boluses ($p = 0.005$) occurred for cows grazing on LHM compared with HHM. A trend for a lower rumination rate ($p = 0.08$) was observed for the LHM cows compared to the HHM cows. The number of chews ($p = 0.15$) per bolus did not differ between the two groups.

3.3 | Herbage intake and bite mass

Pre-grazing HM did not affect the herbage DMI ($p = 0.33$), bite mass ($p = 0.55$), or intake rate ($p = 0.22$).

3.4 | Relation of ingestive behaviour and daily herbage intake

Figures 1 and 2 show the correlation between herbage DMI and the performed eating chews or prehension bites, respectively. The coefficient of determination between eating chew (C31) and herbage DMI was numerically lower for HHM cows compared to LHM cows.

In contrast, a numerically higher coefficient of determination for the correlation between prehension bites evaluated by C31 and herbage DMI was found for the HHM cows compared to the LHM cows.

3.5 | Sward characteristics

During the measuring week, paddocks of the LHM treatment had, compared to HHM, a lower pre-grazing sward height ($p < 0.001$) and showed a trend for a higher post-grazing sward height ($p = 0.051$) (Table 5). However, a lower pre-grazing ($p < 0.001$) and post-grazing HM per ha ($p = 0.001$) was found in LHM compared to HHM pastures. Compared to cows grazing on HHM, LHM cows had a greater grazing area per meal ($p < 0.001$) and a greater HA per day ($p < 0.001$).

3.6 | Milk yield and milk composition

Cows grazing on LHM compared to HHM produced more milk ($p = 0.008$) and energy-corrected milk ($p = 0.03$) (Table 6). A trend for greater milk protein content ($p = 0.06$) was found for the LHM cows compared to the HHM cows. No treatment effects were seen for milk fat content ($p = 0.22$), milk lactose content ($p = 0.63$), or number of somatic cells ($p = 0.84$).

FIGURE 1 Relation of daily herbage intake and performed eating chews on pasture for low (LHM) and high herbage mass (HHM) (evaluated by C31) ($n = 20$). HHM, high herbage mass; LHM, low herbage mass

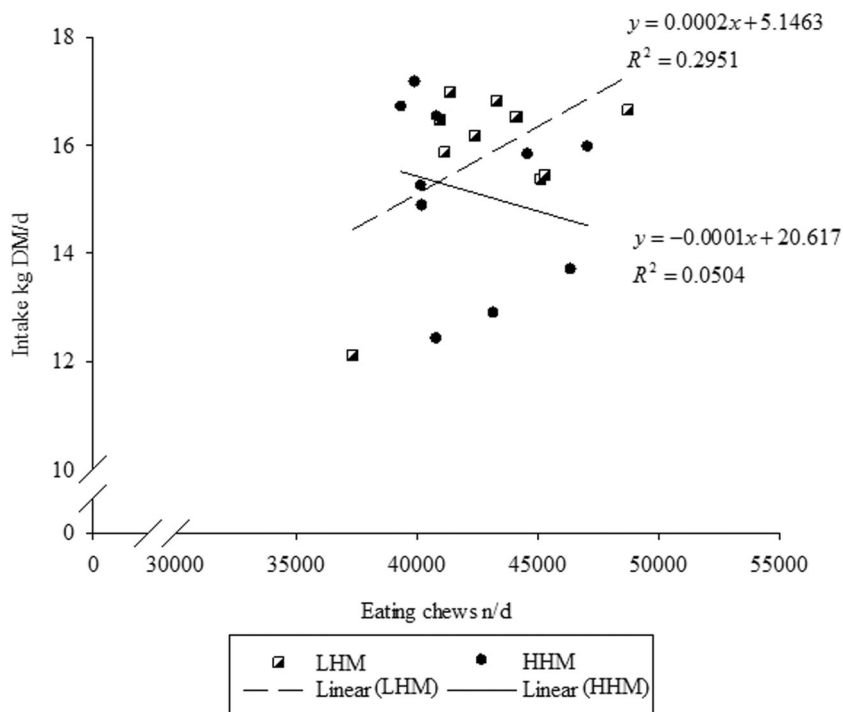
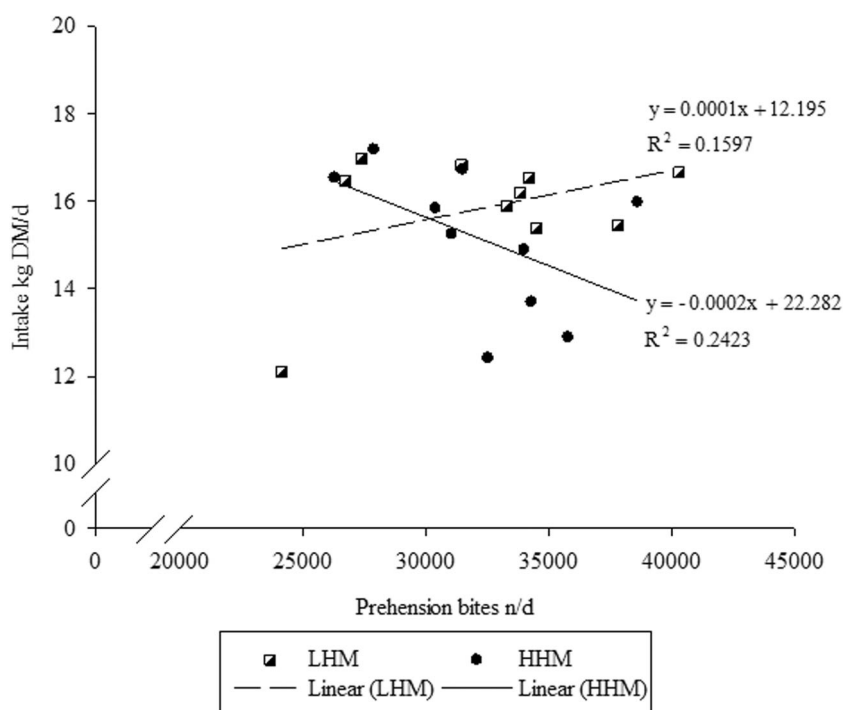


FIGURE 2 Relation of daily herbage intake and performed prehension bites on pasture for low (LHM) and high herbage mass HHM (evaluated by C31) ($n = 20$). HHM, high herbage mass; LHM, low herbage mass



4 | DISCUSSION

The present study validated a RW converter—C31—through direct observations and compared the results with those of the previous converter—C11. Another aim was to investigate the effects of a low and high pre-grazing HM, at the same target HA, on bite mass,

number of prehension bites, and milk production. Two pre-grazing HM, namely LHM and HHM, were chosen as common pre-grazing HM for continuous stocking systems and rotational grazing systems, respectively. Moreover, in view of the prospective intake estimation, the relationship between number of ingestive bites and herbage DMI was investigated.

TABLE 5 Sward characteristics of the pastures, grazed by the low (LHM) and the high (HHM) pre-grazing herbage mass^a cows (*n* = 14)

| Item | LHM | HHM | SE ^b | <i>p</i> -value |
|--|------|------|-----------------|-----------------|
| Sward characteristics | | | | |
| Pre-grazing sward height (CU ^{c/d}) | 10.2 | 18.2 | 0.10 | <0.001 |
| Post-grazing sward height (CU ^{c/d}) | 7.7 | 7.5 | 0.08 | 0.051 |
| Pre-grazing herbage mass (kg DM/ha ^{c/e}) | 589 | 2288 | 21.4 | <0.001 |
| Post-grazing herbage mass (kg DM/ha ^{c/e}) | 150 | 200 | 9.5 | 0.001 |
| Grazing area (m ² /meal ^f) | 4135 | 554 | 174.5 | <0.001 |
| Herbage allowance (kg DM day/cow ^g) | 22.8 | 21.0 | 0.27 | <0.001 |

Abbreviations: HHM, high herbage mass; LHM, low herbage mass.

^aHigh and low pre-grazing herbage mass were 2288 and 589 kg DM/ha, respectively.

^bStandard error.

^cNonparametric test (Kruskal–Wallis) for statistical analysis.

^dClick units (1 CU = 0.5 cm).

^eMeasured above 6.7 click units.

TABLE 6 Effect of low (LHM) and high (HHM) pre-grazing herbage mass^a on milk production, milk content, and herbage intake (*n* = 24)

| Item | LHM | HHM | SE ^b | <i>p</i> -value |
|---|------|------|-----------------|-----------------|
| Milk production performance | | | | |
| Milk yield (kg/day) | 25.4 | 22.7 | 1.40 | 0.008 |
| Energy-corrected milk (kg/day) | 26.6 | 24.1 | 1.39 | 0.03 |
| Fat (%) | 4.4 | 4.6 | 0.09 | 0.22 |
| Protein (%) | 3.6 | 3.4 | 0.07 | 0.06 |
| Lactose (%) | 4.6 | 4.6 | 0.04 | 0.63 |
| Cell number (1000/ml ^{c/d}) | 138 | 103 | 42.5 | 0.84 |
| Herbage DMI (kg/day) ^{e/f} | 15.6 | 15.0 | 0.43 | 0.33 |
| Prehension bite mass (g DM/bite) ^g | 0.49 | 0.47 | 0.023 | 0.55 |
| Herbage DMI rate (g/min) ^g | 25.7 | 27.2 | 0.84 | 0.22 |

Abbreviations: DMI, dry matter intake; HHM, high herbage mass; LHM, low herbage mass.

^aHigh and low pre-grazing herbage mass were 2288 and 589 kg DM/ha, respectively.

^bStandard error.

^cSomatic cell count.

^dLog-10 transformed for statistical analysis.

^eNonparametric test (Kruskal–Wallis).

^fEstimated by the *n*-alkane double indicator method.

^gNumber of eating chews and eating time were evaluated by C31.

4.1 | Validation of the converters

Like the IGER Behaviour Recorder (Ungar & Rutter, 2006) and the acoustic method described by Laca and WallisDeVries (2000), the RWS is one of few systems that permit differentiation between mastication chews and prehension bites. Ungar and Rutter (2006) distinguished bites and chews with the aid of the amplitudes, which were generated by the elongation of a graphite powder-packed silicon tube that was placed around the lower jaw. The discrimination criterion was a small subpeak that occurred only for bites after the normal peak. As the RWS showed pronounced differences in the pressure amplitudes, the approach to differentiate eating chews was to combine the pressure and acceleration signals. The additional signals of a triaxial accelerometer allow the differentiation of mastication chews from prehension bites. Therefore, the C31 converter is capable of identifying prehension bites with an MADP of 12.4% compared to direct observation, which is similar to the deviation of 11.4% measured by Laca and WallisDeVries (2000) for cattle when using the acoustic method. Although the accuracy of C31 for identifying prehension bites was sufficient from our view, improvement of the evaluation software (C31) should continuously be carried out. The MADP of 40.1% for the number of prehension bites evaluated by C11 resulted from the inability of C11 to differentiate eating chews. Although C31 identifies prehension bites, the MADP for mastication during eating was large, due to the inability of the triaxial accelerometer to differentiate mastication during eating from mouth movements during idling or grooming. However, the sum of mastication chews and prehension bites with the head down and their duration showed an acceptable MADP of lower than 15%. Besides the different MADP for the number of prehension bites, no other differences between the two converters were found. The missing differences between the two converters could be explained by the similarity of the decision trees. The differentiation between mastication chews and prehension bites follows after differentiation between eating and rumination.

Rombach et al. (2018) observed a similar MADP for the comparison between C11 and direct observation; for number of boluses (9%), chews per bolus (14%), eating chews (11%), rumination chews (6%), and time spent ruminating (4%) and eating (7%). Employing the RWS, Werner et al. (2018) found deviations similar to those of visual observation for eating chews (7% and 10%) and rumination chews (8% and 9%). Ruuska et al. (2016) observed with the RWS an overestimation of 18% for eating time of stall-fed cows, compared to continuous observation over 12 h. Differences to our validation may appear due to the different environments (pasture vs. tie-stall and loose housing), feeds (pasture herbage vs. grass silage and TMR), the longer observation duration (10 min vs. 12 h), and the converter version (C31 vs. 0.7.0.0). The evaluation with the converter 0.7.0.0 used by Ruuska et al. (2016) is based exclusively on pressure raw data compared to the pressure and accelerometer raw data used by C31. Furthermore, the accuracy of the observations has to be considered as well, as discussed by Rombach et al. (2018). A deviation of 7% for rumination time and 5% for eating time between direct

observation and the IGER Behaviour Recorder was published by Rutter et al. (1997). This is higher than the MADP of 2.1% for C31 and 2.3% for C11 for rumination time, and similar to the MADP of 6.4% for C31 and 6% for C11 for eating time measured in our investigation.

4.2 | Eating and rumination behaviour

In contrast to our study, Pérez-Prieto et al. (2013) found a lower intake rate of 30.7 compared to 34.4 g DM/min when cows grazed on pastures with a low HM. The larger difference in the HM between the groups in their experiment may be a major reason for the difference to our investigation. A higher HM might enable grazing dairy cows to ingest more feed per bite and, therefore, achieve a higher intake rate.

In our study, the LHM cows spent more time eating than the HHM cows, 617 versus 559 min/day. Much shorter eating times (530 or 485 min/day) but still longer for cows on swards with a lower pre-grazing HM, were reported by Pérez-Prieto et al. (2013). This discrepancy in eating time to our study might be explained by a greater herbage intake rate of the cows in the investigation performed by Pérez-Prieto and Delagarde (2012), enabling cows to meet their nutritional needs within a shorter period. Nevertheless, the longer duration of eating time of 58 min/day of the LHM cows in our study is similar to the 45 min/day detected by Pérez-Prieto and Delagarde (2012). Furthermore, the increase in eating time on pasture of 8.3 min/day per 1 cm lower pre-grazing sward height is comparable to the 10.8 min/day and 11.7 min/day measured by others (Gibb et al., 1997; Pulido & Leaver, 2001). In contrast to the longer eating time, the number of eating chews and prehension bites did not differ between LHM and HHM. The same findings occurred in another study (Stakelum & Dillon, 2004). The longer eating time of the LHM cows at an equal number of eating chews and prehension bites might be explained by the increased time needed to find the appropriate herbage to be picked up. On the other hand, the HHM cows had the possibility to graze on small paddocks with a high HM.

With regard to a preliminary herbage intake estimation aided by behavioural characteristics, the differentiation of mastication chews from prehension bites is necessary and might improve the estimation (Laca & WallisDeVries, 2000), as only prehension bites serve for feed intake. Already Rook et al. (1994) and Gibb et al. (1997) mentioned prehension bites and mastication chews during eating. In the present study, a proportion of 24% (HHM) to 25% (LHM) for mastication chews during eating was found. Comparable results were reported by Chacon et al. (1976) with 21 to 25% for non-lactating Jersey cows.

Rumination chews and time, as well as the number of boluses, were higher when cows grazed in HHM. Pérez-Prieto et al. (2013) also observed a considerable increase of rumination time from 370 min/day to 457 min/day when cows grazed on swards with a higher HM. Beltrán et al. (2019) found no differences in rumination time when cows had access to higher or lower HM. Missing differences in ADF and NDF (between the groups of Beltrán et al.,

[2019]) might be the reason for a missing difference in rumination time. Therefore, higher ADF and NDF intake of the HHM group, in the present study, may partially explain this observation, as an overall higher intake of physically effective fibre increases rumination time (Beauchemin & Yang, 2005).

Although the eating and rumination times were significantly different between the HM treatments, the daily sums of these activities were quite similar at 914 (LHM) and 924 min (HHM), respectively. As in our study for grazing dairy cows, other authors have found a negative relationship between eating and rumination time in the barn (Schirrmann et al., 2012) and on pasture (Rook, 2000). Beauchemin (2018) called this a compensatory relationship.

4.3 | Herbage intake and bite mass

The literature involves contrasting results regarding the effect of HM on herbage DMI at the same HA. For example, Curran et al. (2010), McEvoy et al. (2009), and our study measured HM above 40, 40, and 43 mm, respectively, and found no effect. Wims et al. (2010) measured HM above 35.5 mm and reported a negative effect. Wales et al. (1999) measured HM above ground level and found a positive effect. This inconsistent pattern can be explained by the sward height above which the HA was measured. Pérez-Prieto and Delagarde (2012) showed that the effect of HM on DMI was positive, null, or negative when HA was calculated at ground level, 2.5 cm, or 5 cm above ground level, respectively. In order to reproduce the effects of HM correctly and comprehensively, HA should be fully available to dairy cows. Therefore, HA may be calculated from a level equal to the defoliation limit of dairy cows. Pérez-Prieto and Delagarde (2012) reported that cows were unable to graze below 2 to 3 cm measured with a herbometer (AGRO-Systèmes); this corresponds to 3.4–4.9 cm above ground level according to the folding rule method transformed based on Mosimann et al. (1999).

In our study, the cows had the same average prehension bite mass in both treatments. This is in contrast to Stakelum and Dillon (2004), who found a higher prehension bite mass for cows grazing on swards with a higher HM and a height of 31.6 cm compared to a lower HM and a height of 17.8 cm (Hill Farm research organisation sward stick), respectively. Gibb et al. (1997) showed, in comparison to our study, a lower average bite mass for cows grazing on swards with a height of 5 cm compared to 7 cm. Alvarez-Hess et al. (2021) showed a lower (i.e., 0.38 g DM/bite) and a higher bite mass (i.e., 0.62 g DM/bite), depending on the HA; therefore, cows with high HA had a greater bite mass. Differences in HA, and a missing differentiation of mastication and prehension bites to calculate the bite mass, might be a reason for these contrary results. A greater number of mastication chews would decrease the bite mass and vice versa. Laca and WallisDeVries (2000) already mentioned that differentiation between mastication chews and prehension bites is an important issue in intake estimation for grazing cattle. Prehension bite masses similar to those in our study (i.e., 0.42 g DM/bite) were

observed by Prendiville et al. (2010) for grazing Holstein-Friesian cows.

4.4 | Relation of ingestive behaviour and daily herbage intake

It was expected and confirmed that prehension bites correlate much better to herbage DMI than the total number of eating chews. Admittedly, prehension bites or eating chews alone would not be adequate predictors for herbage DMI. However, the highest coefficient of determination was found between herbage DMI and the number of eating chews performed by the LHM cows. This could be caused by the more regular herbage structure of the LHM pastures, which might have favoured a low bite mass variation. The overall low coefficient of determination within and between herbage DMI and the number of eating chews or prehension bites may, in part, be related to the within animal variation in the bite mass. Bite mass and the number of bites are the most variable components of feeding behaviour and are influenced by an enormous range of factors (Andriamandroso et al., 2016). Overall, a large range of the averaged prehension bite mass per cow on a weekly basis, between 0.36 and 0.63 g DM/prehension bite, and in the bite mass, between 0.26 and 0.41 g DM/eating chew, was found in our investigation. A similar range between 0.33 and 0.74 g DM/bite has been presented by Barrett et al. (2001), Alvarez-Hess et al. (2021), and Penning and Rutter (2004).

Eating chews or prehension bites explained only a minor part of the variation of herbage DMI, namely 5%–30% and 16%–24%, respectively. For reliable herbage DMI estimation, characteristics beyond behavioural ones are thus needed. Halachmi et al. (2016) concluded that an extensive range of factors potentially affect voluntary feed intake, and showed that incorporating feeding behaviour into existing DMI models improved estimation accuracy by 1.3 kg/cow per day. Therefore, additional animal-, environmental-, and feed-related factors affecting herbage DMI should be considered in prospective models for reliable herbage DMI estimation of grazing dairy cows.

4.5 | Sward characteristics

The HA was compared on a DM basis. Due to the duration of DM determination, the DM values for fresh herbage of the previous day had to be used for HA or paddock area calculations (see Section 2.2). This delay resulted in a 7.9% lower pre-grazing HA for the HHM cows (1.8 kg DM/day per cow) in comparison to the LHM cows. Curran et al. (2010) observed with a difference in HA of 5 kg DM/day per animal an increased milk yield of 0.5 kg/day, 0.14 percentage units in milk protein concentration, and 1.9 kg DMI/day when cows had grazed on pastures with an HM of 2400 kg DM/ha. Therefore, the effect of the differences in the pre-grazing HA in our study may be of minor importance for the interpretation of the results, as the

differences in the HA were less than a third of those used by Curran et al. (2010). Furthermore, both treatments in our study had a HA above 20 kg DM herbage per animal and day, which is less restrictive than the 15 kg DM per day and animal used by Curran et al. (2010).

4.6 | Milk yield and milk composition

Cows grazing on LHM produced more milk and energy-corrected milk compared to cows grazing on HHM, which was primarily caused by the higher NEL and CP content of the ingested herbage. The deterioration in the quality of high swards is a consequence of advanced maturity (Stakelum & Dillon, 2004), the accompanying increase of the ADF content, and the decrease of CP and NEL content of herbage. Also, Curran et al. (2010) and Muñoz et al. (2016) have reported increased milk production by cows grazing on low compared to high HM swards, due to the higher nutritive value of herbage in low HM swards. Beltrán et al. (2019) found no or minor differences in milk yields at different HM even though the cows showed no difference in DMI as in the present study. Differences might be due to greater differences in the chemical composition of herbage fed to HHM and LHM cows in the present study. Further, McEvoy et al. (2009) studied the effects of different HM over the whole grazing season, and they reported milk yield was not different between HM, though solid-corrected milk yield differed.

To cross-check the milk yield and DMI results, calculation of the milk production potentials (results not shown) was carried out. Based on the DMI results and the herbage NEL content of the present study as well as the energy maintenance requirements of the cows (estimated by means of the BW, Agroscope, 2015) and the energy content of 1 kg ECM (3.14 MJ NEL/kg ECM; Agroscope, 2015) the milk production potential were calculated. The milk production potential showed noticeable differences to the measured ECM in the present study. In both treatments, LHM and HHM, the differences were about the same. According to our evaluation, the milk quantity determination as well as the analysis of the milk components might be omitted as a possible cause for the difference. Another cause for the differences could be the mobilisation of body tissue, which would not be expected in cows with 200 DIM to this extent, even under pasture conditions. Furthermore, the use of the surplus protein in the herbage as an additional source of energy by the dairy cows could be a possible explanation. Finally, an under-estimation of herbage intake with the marker method might also be a possible cause, although this method performed well with dairy cows fed fresh herbage in the barn (Kaufmann et al., 2011).

Cows in LHM also showed a trend toward higher milk protein content, which was probably due to the improved energy supply, compared to HHM cows. Muñoz et al. (2016), applying a large difference between HM treatments (>1600 kg DM/ha), also found a trend for higher milk protein content. In contrast, other studies (Curran et al., 2010; McEvoy et al., 2009) with a lower difference between HM treatments observed no effect of HM on milk protein content. In the current study, although more milk was produced and a

trend for higher milk protein content was found with LHM, the high protein concentration of herbage in LHM may have adversely affected nitrogen use efficiency compared to HHM.

5 | CONCLUSIONS

The evaluation software C31 enables a differentiation between eating chews in mastication chews and prehension bites, whereby the error for the estimation of the number mastication chews is large. No further differences in accuracy between C11 and C31 were observed for grazing dairy cows. Within the given framework, HM had no influence on average daily bite mass, herbage DMI, or number of prehension bites as investigated by other studies; this may be due to the differing sward heights above which the HA is measured. Finally, eating chews or prehension bites alone were insufficient to estimate herbage DMI and explained only a minor part of herbage DMI variation. However, including these behavioural characteristics in more sophisticated DMI models might improve the accuracy of a preliminary DMI estimation and has to be examined in further studies.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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