Effect of forage type on swallowed bolus mass and a method for counting swallows in dairy cattle

N. Norbu,1,2* P. S. Alvarez-Hess,1,3 B. J. Leury,2,3 M. L. Douglas,1 M. M. Wright,1 S. R. O. Williams,1 A. L. Thomson,1 V. M. Russo,1,2 M. C. Hannah,1 W. J. Wales,1,3 and M. J. Auldist1,3

1Agriculture Victoria Research, Ellinbank, Victoria 3821, Australia
2School of Agriculture and Food, Faculty of Veterinary and Agricultural Sciences, The University of Melbourne, Parkville, Victoria 3010, Australia
3Centre for Agricultural Innovation, School of Agriculture and Food, Faculty of Veterinary and Agricultural Sciences, The University of Melbourne, Parkville, Victoria 3010, Australia

ABSTRACT

Dry matter intake (DMI) is a primary determinant of milk production in grazing dairy cows and an ability to measure the DMI of individual cows would allow herd managers to formulate supplementary rations that consider the amount of nutrients ingested from grass. The 2 related aims of this experiment were to define the mean number of swallowed boli and mass of the swallowed boli in Holstein-Friesian dairy cattle offered a variety of forages commonly fed in the dairy industry of southeastern Australia, and to evaluate 2 indirect methods for counting the number of swallows. Twelve ruminally-fistulated, lactating Holstein-Friesian cows were randomly assigned to 3 replicated 4 × 4 Latin square designs and offered 4 forages: fresh chicory (FC), fresh perennial ryegrass (RP), alfalfa hay (AH), and perennial ryegrass silage (RS). The experiment was conducted over 28 d with each of 4 periods consisting of 7 d with 3 d of measurement. Forage diets were offered to individual cows following the partial evacuation of the rumen. The first 20 min after forage was offered constituted the measurement period, during which all swallowed bolus were manually captured by samplers who placed their hand through the ruminal fistula and over the cardia entrance of the rumen of each cow. Concurrently, microphones and video cameras were used for the indirect measurement of swallows. The average swallowed bolus mass overall was 17.4 g dry matter (DM) per bolus with the lowest mass observed in cows offered FC (8.9 g DM/bolus), followed by RP (14.9 g DM/bolus), compared with cows offered AH (23.6 g DM/bolus) and RS (22.3 g DM/bolus). The swallowing rate was greater in cows offered FC (78 swallows/20 min) than in cows offered RP, AH, and RS (62.3 swallows/20 min). The audio recording method showed greater concordance (Lin’s concordance correlation coefficient = 0.90) with the physical capturing of the bolus through the rumen, than the video recording method did (Lin’s concordance correlation coefficient = 0.54). It is concluded that the mass of the swallowed bolus is related to forage type and that using a microphone attached to the cow’s forehead can provide an accurate measure of the number of swallows when verified against the actual number of swallows counted by manual interception of the bolus at the rumen cardia. Key words: bite mass, bite rate, dry matter intake, feeding behavior, herbage intake

INTRODUCTION

Dairying systems in southeastern Australia, New Zealand, Ireland and other parts of the world are based around the use of grazed pasture as the main source of nutrients owing to its inherent low cost. As in other feeding systems, total dry matter intake DMI is a primary determinant of milk production in dairy cows but varies widely between cows within a herd even when the opportunity to consume feed is similar (Wright et al., 2019). An ability to measure the DMI from the different components of the diets of individual cows would allow herd managers to formulate supplementary rations for cows that take into account the amount of nutrients ingested from the different forages. It would also allow more accurate identification of cows that are efficient, or inefficient, at converting feed into milk, information that could then be used in breeding programs targeting efficient cows.

Measuring the intake of grazing cows, however, is problematic due to the free-ranging nature of pasture grazing systems (Mayes and Dove, 2000). Measuring the intake of conserved forage in individual cows is similarly challenging when cows are group fed in the paddock. Some techniques exist for measuring the
forage DMI of individual cows, most notably those using indigestible markers such as n-alkanes or chromic oxide, but these methods are labor-intensive, time-consuming, and involve complex and expensive chemical analyses (Dillon, 1993; Wright et al., 2019). More recently, on-cow sensors that quantify various aspects of feeding behavior have been developed and their suitability for DMI prediction has been assessed (Rombach et al., 2018). For example, researchers have investigated the use of jaw movement recorders that quantify the number of prehension bites for use in predicting pasture DM of dairy cows, with or without the concurrent measurement of factors affecting the amount of DM prehended in each bite (bite mass; Alvarez-Hess et al., 2021). Moreover, other parameters such as feeding and chewing time have also been assessed for their suitability for predicting DMI (Pahl et al., 2016).

An approach to estimating the individual DMI of cows consuming forage that could possibly be used as an alternative to these techniques, or in conjunction with them, may be to count the number of times a cow swallows feed during a defined time period. Intuitively, the DMI of a cow is a product of the mass of DM contained within each swallowed bolus and the number of swallows a cow takes. Before such a concept could be used in a future method for measuring DMI, however, 2 main things are required. The first is to define the mass of DM contained with each swallowed bolus (bolus mass), how consistent this mass is during a feeding session and the way in which it varies with forage type. Some previous research, for example, has shown that the species of forage, and whether it is fresh or conserved, can affect the amount of DM swallowed in each bolus (Gill et al., 1966; Stuth and Angell, 1982; Minnee et al., 2019). Next, a method for counting the number of swallows remotely is required. Such a method does not currently exist and there are limited data in the literature.

The current experiment, therefore, had 2 related aims, both of which contribute toward the future objective of developing a method for estimating DMI by counting swallows in cattle. The first aim was to define the mean mass of the swallowed bolus in Holstein-Friesian dairy cattle offered a variety of forages commonly fed in the dairy industry of southeastern Australia. The second aim was to evaluate 2 indirect methods for counting the number of swallows, namely video observation and on-cow microphones, against the actual number of swallows counted by manual interception of the bolus at the rumen cardia. We hypothesized that swallowed bolus mass would be affected by forage type and that both indirect methods for counting swallows would be equally concordant with the number of swallows counted by manual interception of the bolus at the rumen cardia.

MATERIALS AND METHODS

The experiment was conducted at Agriculture Victoria Research’s Ellinbank Centre in Victoria, Australia (38°14′S, 145°56′E). All procedures were conducted in accordance with the Australian Code of Practice for the Care and Use of Animals for Scientific Purposes (NHMRC, 2013). Approval to conduct the experiments was obtained from the Department of Jobs, Precincts and Regions Agricultural Research and Extension Animal Ethics Committee.

Cows

The experiment used 12 ruminally-fistulated, lactating Holstein-Friesian dairy cows that had calved in late winter/early spring (July to August 2020). At the start of the experiment, cows had a body weight of 679 ± 71 (mean ± SD) kg and a mean BCS of 4.1 ± 0.2 on the 8-point scale Earle (1976). Cows’ mean number of lactations were 6.0 ± 2.4 and were 216 ± 105 DIM, and had mean milk yields of 24.8 ± 4.8 kg milk/cow per d.

Before the experiment, and on nonmeasurement days during the experiment, all cows were managed with the main research herd, which received ~6 kg of DM of a 50:50 mix of wheat and barley grain, ~10 kg of DM of perennial RS and ~4 kg of DM of perennial ryegrass pasture per cow per day, each offered in 2 equal portions per day, either during (grain) or after milking (silage and pasture).

Experimental Design and Treatments

Each of the 12 cows received each of 4 treatments over 4 wk in a replicated Latin square experiment. The 4 treatment diets were fresh chicory (FC), fresh perennial ryegrass pasture (RP), alfalfa hay (AH), and perennial ryegrass silage (RS). Each cow was treated and measured only once per week and always on the same day each week. By the end of the 4 wk, all cows had received all treatments once.

On measurement days, cows were milked in the parlor at approximately 0700 h and moved to the research facility without having had access to grain in the dairy during milking. They were then individually restrained in stalls using a neck yoke. Treatment forages were weighed and placed near each stall before cows arrived, but the feed was not placed in bins until the preset start time for measurements. Cows were offered their allocated forage ad libitum during the measurement period, which depending on the forage ranged between

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1.1 and 3.2 kg DM/cow for a 20-min eating session including refusals.

The FC and RP were both in the vegetative stage when cut from pure stands using electric hand shears (Handypiece, Fielding, New Zealand) at an estimated grazing height of ~5 cm above ground level. In both cases, fresh forage was cut each morning and offered to the cows as cut. Alfalfa hay was purchased in Victoria, Australia, and loosened slightly from the bale before being presented to the cows. The RS was taken from wrapped bales that had been ensiling for at least 3 wk. The ryegrass used for silage was in the vegetative stage at the time of cutting. The RS was presented to the cows without any additional processing.

Bolus Collection and Sampling

To collect the swallowed bolus, each cow’s ruminal fistula was opened, and ruminal contents were partially evacuated (approximately one-third), exposing the cardia (Minnee et al., 2019). The solid fraction of the ruminal contents was extracted by hand, and the liquid fraction was removed using a plastic cup. Digesta was stored in a clean bucket covered with a tarpaulin to reduce heat loss as much as possible.

Four cows were measured simultaneously on each sampling day. Five minutes was allocated to partially evacuate the rumen, then feed was made available to the cows and a 20-min sampling period commenced. Swallowed boli were intercepted by samplers placing a hand at the opening of the cardia as soon as feed was offered and collecting one bolus at a time (Minnee et al., 2019). Collected swallowed boli were placed individually in labeled aluminum containers during the 20-min eating session. A 20-min eating session was chosen to avoid the removed ruminal contents becoming cold. The time of each swallow was recorded. On an average of 2.2 occasions per 20-min eating session, the samplers could feel that they had missed or dropped a bolus. Missed or dropped boli were recorded for swallow rate calculations and calculated swallow mass.

Immediately after the 20-min eating session, ruminal contents were returned, the fistula was cleaned, and the fistula closed. The temperature of the removed digesta was monitored using a digital thermometer (Genia, Hilaire de Chaleons, France) and the temperature did not drop below 37°C. The fresh weight of each bolus was weighed and recorded, and then boli were oven-dried at 105°C for 48 h. Finally, the dry weight of each bolus was recorded.

Approximately 3 kg of DM of a barley and maize (50:50) grain mixture was offered after the conclusion of the sampling period. Cows had ad libitum access to water in the stalls. After finishing their grain, cows were moved to a nearby paddock where they received ad libitum AH. After the evening milking, cows were returned to the main herd until they were required for sampling the following week.

Sampling, Analyses, and Calculations

Dry matter concentration of each forage was determined daily by collecting 10 subsamples of approximately 30 g each from each forage. All samples were taken immediately after cutting the fresh forages or collecting the silage from the bales, before filling the cows’ feeders. Daily samples for each forage type were pooled and then dried in an oven at 105°C for 24 h. Required quantities of forage offered to individual cows were weighed into individual feed bins, with each forage weight then calculated on a DM basis. Refusals were collected and weighed after each feed to facilitate the calculation of DMI for each cow (DM offered minus DM refused). The DM recovery rate was calculated as the sum of DM obtained from the captured boli divided by the DMI. The swallowed bolus mass (g DM/bolus) was calculated from DMI divided by the total number of swallows; hereafter called calculated swallowed bolus mass. The observed swallowed bolus mass was the weight of the dried intercepted boli.

Representative samples for nutritive analysis of forages offered were collected each day before being offered to the cows, by subsampling from different locations in the feed bin. Daily forage samples were composited on a weekly basis for each forage type. These composited samples were subsequently frozen, freeze-dried, ground to pass through a 0.5-mm sieve, thoroughly mixed and subsampled. Samples were sent to a commercial laboratory (Dairy One Forage Laboratory, Ithaca, NY) for analysis of concentrations of CP (AOAC International, 2000; Method 990.03), ADF (ANKOM Technology Method 14), NDF (ANKOM Technology Method 15), lignin (ANKOM Technology Method 9), NFC (calculated by difference as 100% − [CP% + NDF% + Fat% + Ash%]; NASEM, 2001), starch (as per Xylem YSI Life Sciences Application Note Number 222LS-02), and ash (AOAC International, 2000; Method 942.05).

Concentrations of estimated ME were calculated using the following formula (NASEM, 2001):

\[ \text{ME 1x (Mcal/kg)} = 1.01 \times \text{digestible energy 1x} - 0.45. \]

For descriptive purposes, mean nutritive characteristics of each of the 4 forages are presented in Table 1.
Indirect Swallow Counting

The video recording method relied on observing swallows. For this purpose, cameras were set in 2 locations near the cows in the stalls: one camera on the top of the feed bin in front of the cow and another on a pole near the left-hand side of the cow. Furthermore, video recording of swallows was improved with supplementary light using an LED Work Light (Arlec, WL0007, Arlec Australia Pty Ltd., Blackburn North, Victoria) adjacent to the stalls. Video recordings from the cameras were played on a VLC media player (VLC project, VideoLAN organization, Paris, France), and swallows were counted by visual observation of the swallowing process at the pharyngeal phase. If the pharyngeal movement was not clearly visible, a pause taken after mastication chews followed by another prehension bite was assumed to indicate a swallow. Swallows were sometimes difficult to see on video recordings due to the movement of cows. Thus, the number of swallows was counted from the footage of 2 cameras with synchronized timestamps.

The audio recording method used a microphone positioned on the forehead of the cow and was adapted from the methods of Laca and WallisDeVries (2000) and Navon et al. (2013). Cows were fitted with microphones (Transducer PT-01, Perytek, Warragul, Victoria, Australia) to allow acoustic counting of swallows. The digital recorder of the microphone (Tascam DR-05X, 1910417, TEAC Corporation) was fastened to a halter just behind the cow’s head, and the microphone was placed firmly against the forehead of the cow using a Velcro elastic band (Laca and WallisDeVries, 2000; Navon et al., 2013). Audio data were recorded at 44.1 kHz and 16-bit resolution sampling frequencies. The recorded data were saved in an secure digital memory card in Waveform Audio file format (Clapham et al., 2011). Audio data collected using microphones were processed with the Wavepad Sound Editor software (NHC software 6.93). Using this software allowed for identifying peak and trough patterns in the audio data and entire sound traces from each cow were examined. From a preliminary analysis, we observed that trough patterns in the audio data corresponded to the timing of physically captured swallows. Thus, each trough in the audio data were identified as a swallow.

Feeding Behavior

Jaw movement recorders (RumiWatch, Itin+Hoch GmbH, Liestal, Switzerland) were fitted onto cows as described by Norbu et al. (2021). These recorded the number of prehension bites, mastication chews, and total jaw movements. Recorded behaviors were translated into 5-min summaries using RumiWatch converter software 0.7.3.36. Bite rate was calculated as the number of prehension bites per 20 min. Bite mass (g DM/bite) was then calculated for each cow from the DMI divided by the number of prehension bites.

Statistical Analyses

Data (n = 12) on observed swallowed bolus mass, bite mass, bite rate, and swallowing rate was averaged within four 5-min intervals of each 20-min measurement period for each cow before statistical analysis and analyzed using REML mixed models in GenStat 20 (VSN International Ltd., Hemel Hempstead, UK). The statistical model comprised factorial fixed effects of forage type by time interval, and random effects of the cow by day split for the time interval, plus sampler. The trends of observed swallowed bolus mass, bite mass, bite rate, and swallowing rate against the duration since the start of the eating were presented graphically, including 5% least significant intervals. Dry matter intake, DM recovery rate, calculated swallowed bolus mass,
fresh swallowed bolus mass, total swallows, and eating behaviors were averaged within the day for each cow before analysis and analyzed with ANOVA. For this, the statistical model excluded all terms involving time interval but was otherwise the same as described above. The rationale for using 2 different analyses was to see the trend of observed swallowed bolus mass, bite mass, bite rate, and swallowing rate over time spent eating, where the REML analysis avoids some of the problems of biased variance estimates. Differences of $P < 0.05$ were considered significant. Residuals were checked graphically for normality, and constant variance and data transformation were applied, if necessary, to meet these distributional assumptions.

A generalized linear model with binomial model error distribution and logit link function was used to analyze the number of swallows detected by video recording and audio recording compared against the actual number of boli captured physically. The linear predictor in the model was as follows:

$$\text{logit}(p) = u + C + D + T,$$

where $u$ is a constant, $C$ is the cow effect, $D$ is the day effect, $T$ is the treatment effect, and $p$ is the proportion of swallows detected.

Further, Lin’s concordance correlation coefficient (LCCC) analysis (Lin, 1989) was calculated to evaluate video recording and audio recording of the number of swallows against the number of physically captured bolus. The concordance correlation coefficient (LCCC = $r \times$ bias correction factor [Cb]) measures both precision (Pearson correlation coefficient $r$) and accuracy (Cb). The $r$ measures how far each observation deviates from the best-fit line and the Cb measures how far the line of best fit is from a 45-degree angle through the origin. The r, as well as the LCCC and the Cb, were interpreted as negligible (0.00–0.30), low (0.30–0.50), moderate (0.50–0.70), high (0.70–0.90), and substantial (0.90–1.00; Hinkle et al., 2003).

## RESULTS

### Means and Variation of DMI, Swallowed Bolus Mass, Swallow Number and Rate, and Eating Behaviors

Table 2. Overall means, SD, ranges, and CV (irrespective of treatment) for DMI, swallowed bolus mass, swallow number and rate, DMI recovery, and eating behaviors of cows for the 20-min measurement

<table>
<thead>
<tr>
<th>Item</th>
<th>Mean</th>
<th>SD</th>
<th>Minimum</th>
<th>Maximum</th>
<th>CV %</th>
</tr>
</thead>
<tbody>
<tr>
<td>DMI (kg/cow)</td>
<td>1.2</td>
<td>0.45</td>
<td>0.2</td>
<td>2.2</td>
<td>37.5</td>
</tr>
<tr>
<td>Observed bolus mass (g DM/bolus)</td>
<td>17.4</td>
<td>6.9</td>
<td>6.3</td>
<td>32.6</td>
<td>39.4</td>
</tr>
<tr>
<td>Calculated bolus mass (g DM/bolus)</td>
<td>18.6</td>
<td>6.96</td>
<td>6.3</td>
<td>33.9</td>
<td>37.4</td>
</tr>
<tr>
<td>Fresh bolus mass (g/bolus)</td>
<td>150</td>
<td>25.9</td>
<td>102</td>
<td>207</td>
<td>17.3</td>
</tr>
<tr>
<td>DM recovery (%)</td>
<td>93.8</td>
<td>9.1</td>
<td>50.9</td>
<td>100</td>
<td>9.7</td>
</tr>
<tr>
<td>Swallows per 20 min</td>
<td>66.3</td>
<td>16.5</td>
<td>31</td>
<td>117</td>
<td>24.8</td>
</tr>
<tr>
<td>Bite mass (g DM/bite)</td>
<td>2.9</td>
<td>1.74</td>
<td>0.9</td>
<td>8.7</td>
<td>60.0</td>
</tr>
<tr>
<td>Prehension bites per 20 min</td>
<td>485</td>
<td>204</td>
<td>105</td>
<td>985</td>
<td>42.1</td>
</tr>
<tr>
<td>Prehension bites (no./bolus)</td>
<td>7.3</td>
<td>3.52</td>
<td>2.1</td>
<td>12.3</td>
<td>48.2</td>
</tr>
<tr>
<td>Mastication chews per 20 min</td>
<td>1,050</td>
<td>251.30</td>
<td>299</td>
<td>1,579</td>
<td>23.9</td>
</tr>
<tr>
<td>Mastication chews (no./bolus)</td>
<td>16.9</td>
<td>6.42</td>
<td>7.6</td>
<td>36.8</td>
<td>38.0</td>
</tr>
<tr>
<td>Total jaw movements per 20 min</td>
<td>1,535</td>
<td>217.50</td>
<td>548</td>
<td>1,814</td>
<td>14.2</td>
</tr>
<tr>
<td>Total jaw movements (no./bolus)</td>
<td>24.2</td>
<td>5.46</td>
<td>13.4</td>
<td>39.6</td>
<td>22.6</td>
</tr>
</tbody>
</table>

1Calculated swallowed bolus mass is the swallowed bolus mass derived from DMI divided by total swallow counts.

2Dry matter recovery percentage is the amount of DM recovered in the boli (including an adjustment for known missed boli) as a percentage of measured DMI.

Means, SD, and ranges of the variables including coefficient of variation (CV) for swallowed bolus mass, swallows, and eating behaviors of cows are shown in Table 2. Mean-observed bolus mass was 17.6 g DM/bolus, and mean-calculated bolus mass was 18.6 g DM/bolus. The mean swallow rate was 66.3 swallows/20 min, and mean bite mass was 2.9 g DM/bite. The CV was greater than 30% for observed swallowed bolus mass, calculated swallowed bolus mass, bite mass, bite rate, prehension bites, swallows, and mastication chews. Moreover, the CV was 9.7% for the DMI recovery rate, 17.3% for the fresh swallowed bolus mass, and 14.2% for total jaw movements.

### Swallowed Bolus Mass, Swallow Rate, and Bite Mass

Observed swallowed bolus mass, calculated swallowed bolus mass, and fresh swallowed bolus mass were affected by forage type (Table 3). Observed swallowed bolus mass was lower in cows offered FC or...
RP than in cows offered AH or RS with the lightest swallowed bolus mass in cows offered FC. Similarly, the calculated swallowed bolus mass was lower in cows offered FC or RP than in cows offered AH or RS. However, the observed swallowed bolus mass in cows offered FC was lower than RP. Fresh swallowed bolus mass was lower in cows offered AH than in the other forages. The DM recovery rate was not affected by forage type.

Swallowed bolus mass and bite mass over the 20-min eating bout is presented in Figure 1 (a and b, respectively). Swallowed bolus mass was not affected by time spent feeding, but at every 5-min interval, swallowed bolus mass was greater in cows offered the AH or the RS than cows offered the FC or RP. Bite mass was lower in cows offered FC or RP than in cows offered AH or RS (Table 4). Bite mass over the 20-min eating bout is presented in Figure 1(b). Bite mass was not affected by time spent feeding in cows offered FC or RP, whereas bite mass in cows offered AH or RS decreased as the eating bout progressed. Bite mass was lower in cows offered FC or RS at every 5-min interval than cows offered AH or RS, with the greatest bite mass in cows offered RS at all intervals.

Bite rate was greatest in cows offered FC and lowest in cows offered RS, with cows offered RS and AH being intermediate (Table 4). Bite rate over the 20-min eating session is presented in Figure 1(c). Bite rate was not affected by time spent feeding in cows offered FC, RP, and RS. However, bite rate in cows offered AH sharply increased from the first 5-min interval to the third 5-min interval during the eating session and then decreased after the third 5-min interval.

Swallowing rate was greater in cows offered FC, than in cows offered AH, RS, and RP (Table 3). Swallowing rate over the 20-min eating bout is presented in Figure 1 (d). Swallowing rate in cows did not change for any of the forages over the duration of the 20-min eating bouts. However, at every 5-min interval, swallowing rate was greater in cows offered FC than in cows offered other forages.

### Bites and Chews Per Bolus

The number of prehension bites, the number of mastication chews, the number of total jaw movements and their contribution to each bolus in cows offered different forages are presented in Table 4. The number of prehension bites was affected by forage type but not the number of mastication chews or total jaw movements.

Prehension bites per bolus were affected by forage type, being lowest in cows offered RS and highest in cows offered RP and AH. Mastication chews per bolus was affected by forage type, being lowest for cows offered FC and highest in cows offered AH and RS. The number of mastication chews per 20 min was not affected by forage type. Total jaw movements per 20 min was not affected by forage type but total jaw movements per bolus was lowest in cows offered FC and highest in cows offered AH and RS.

### Evaluation of Methods for Indirect Swallow Counting

The percentage of swallows detected by video recording and audio recording compared against the physical...
For every forage type, the number of swallows counted with the video recording showed a lower detection percentage than the audio recording when compared with the physical capturing of the bolus. The video recording detected between 68% and 77% of the captured boli and this capturing of boli are shown in Table 5. For every forage type, the number of swallows counted with the video recording showed a lower detection percentage than the audio recording when compared with the physical capturing of the bolus. The video recording detected between 68% and 77% of the captured boli and this

Table 4. Effect of forage type on eating behaviors of cows.

<table>
<thead>
<tr>
<th>Item</th>
<th>FC</th>
<th>RP</th>
<th>AH</th>
<th>RS</th>
<th>SED</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bite mass (g DM/bite)</td>
<td>1.6c</td>
<td>2.2ab</td>
<td>3.2b</td>
<td>4.8a</td>
<td>0.52</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Bite rate (bites per 20 min)</td>
<td>553a</td>
<td>503ab</td>
<td>371b</td>
<td>81.00</td>
<td>0.005</td>
<td>0.005</td>
</tr>
<tr>
<td>Prehension bites (no./bolus)</td>
<td>7.3ab</td>
<td>8.0a</td>
<td>8.4*</td>
<td>5.5b</td>
<td>0.95</td>
<td>&lt;0.018</td>
</tr>
<tr>
<td>Mastication chews per 20 min</td>
<td>1,030</td>
<td>1,005</td>
<td>1,076</td>
<td>1,089</td>
<td>68.90</td>
<td>0.839</td>
</tr>
<tr>
<td>Mastication chews (no./bolus)</td>
<td>14.6 (2.6)b</td>
<td>16.8 (2.8)ab</td>
<td>19.3 (2.9)a</td>
<td>19.8 (2.9)a</td>
<td>1.07 (0.07)</td>
<td>0.001</td>
</tr>
<tr>
<td>Total jaw movements per 20 min</td>
<td>1,582</td>
<td>1,508</td>
<td>1,460</td>
<td>88.80</td>
<td>0.499</td>
<td></td>
</tr>
<tr>
<td>Total jaw movements (no./bolus)</td>
<td>20.9b</td>
<td>24.2ab</td>
<td>27.3a</td>
<td>24.2ab</td>
<td>2.10</td>
<td>&lt;0.042</td>
</tr>
</tbody>
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*a-c Group means with different superscripts are significantly different (P < 0.05).

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<tr>
<td>Mastication chews per 20 min</td>
<td>1,030</td>
<td>1,005</td>
<td>1,076</td>
<td>1,089</td>
<td>68.90</td>
<td>0.839</td>
</tr>
<tr>
<td>Mastication chews (no./bolus)</td>
<td>14.6 (2.6)b</td>
<td>16.8 (2.8)ab</td>
<td>19.3 (2.9)a</td>
<td>19.8 (2.9)a</td>
<td>1.07 (0.07)</td>
<td>0.001</td>
</tr>
<tr>
<td>Total jaw movements per 20 min</td>
<td>1,582</td>
<td>1,508</td>
<td>1,460</td>
<td>88.80</td>
<td>0.499</td>
<td></td>
</tr>
<tr>
<td>Total jaw movements (no./bolus)</td>
<td>20.9b</td>
<td>24.2ab</td>
<td>27.3a</td>
<td>24.2ab</td>
<td>2.10</td>
<td>&lt;0.042</td>
</tr>
</tbody>
</table>

*a-c Group means with different superscripts are significantly different (P < 0.05).

Numbers inside parenthesis are arithmetic mean and within the parenthesis are on log scale. FC = fresh chicory; RP = fresh perennial ryegrass pasture; AH = alfalfa hay; RS = perennial ryegrass silage; SED = standard error of the difference.
Forage type offered to cows affected swallowed bolus mass, with a greater swallowed bolus mass observed in cows offered the 2 conserved forage treatments (AH and RS) than the 2 freshly cut forage treatments (FC and RP). The greater swallowed bolus mass observed in the cows when they were offered the 2 conserved forages may be attributed to the greater moisture concentration and bulk density of the fresh forages (Soder et al., 2009). It is possible that the greater moisture and bulk density of feeds causes a physical restriction to the amount of feed DM ingested in each swallow. There may be other factors including particle size and structural plant characteristics such as plant fibrousness, tensile and shear strength, and plant canopy structure that are known to affect the feeding behavior of herbivores (Laca et al., 2001), and that may be affecting swallowed bolus mass. However, these were not measured in this current research and may be a limitation of this research.

The mean swallowed bolus mass in cows offered FC or RP in the current study is similar to the values reported by Minnee et al. (2019), who observed a mean swallowed bolus mass of 9 g for cows fed chicory and 13 g for cows fed ryegrass. The mean swallowed bolus mass of cows offered AH in our experiment was also similar to that of cows offered timothy (Phleum pratense L.) hay (20 g; Gill et al., 1966). To our knowledge, this is the first report of swallowed bolus mass of cows offered RS. The time spent eating did not affect swallowed bolus mass of cows. If the swallowed bolus mass is consistent throughout the day, then it may be possible to estimate the DMI of a cow as the product of the predicted bolus mass and the number of swallows a cow takes. However, we acknowledge it is uncertain whether the 20 min measurement period used in the current experiment with stall fed cows is sufficient to draw conclusions about the variation in swallowed bolus mass over 24 h in grazing cows. Contrary to our findings, previous research has reported that swallowed bolus mass was lower at the beginning of a 30-min eating session than later in the eating session (Gill et al., 1966; Minnee et al., 2019). The reason for these inconsistent effects of time spent eating on swallowed bolus mass is not known. We suggest that future research evaluates the variation in swallowed bolus mass over 24 h to better understand bolus mass and the factors affecting it, with the aim to use predicted swallowed bolus mass in the calculation of DMI.

**DISCUSSION**

Forage type offered to cows affected swallowed bolus mass, with a greater swallowed bolus mass observed in cows offered the 2 conserved forage treatments (AH and RS) than the 2 freshly cut forage treatments (FC and RP). The greater swallowed bolus mass observed...
In cows offered FC or RP, bite mass was greater and bite rate was lower than in cows offered AH or RS. This indicates that feeds that resulted in greater swallowed bolus mass also resulted in greater bite mass and lower bite rate. This is presumably related to the lower DM content of the fresh forages compared with conserved forages as, generally, bite mass increases as cattle graze forages with greater sward bulk density (Casey et al., 2004). The range of bite mass observed in cows in the current study offered FC or RP was within the range of 0.2 to 4.0 g DM/bite reported in the meta-analysis by Boval and Sauvant (2019). Similar to the calculation of DMI using the swallowed bolus mass, DMI can be calculated as the product of bite rate and bite mass (Hodgson, 1982).

To calculate DMI based on the predicted bolus mass and the number of swallows a cow takes requires counting the number of swallows. In our evaluation of the 2 indirect methods for counting swallows, audio recording detected 92% to 96% of the physically captured bolii, indicating a miss rate of about 5% of swallows, while video recording only detected 68% to 78% of the physically captured bolii, indicating a miss rate of about 25% of swallows. There was substantial concordance between the audio recording and the physical capturing of the bolus for counting swallows. This finding indicates that video recording was not an accurate method for counting swallows, but the accuracy of audio recording was good. Our experience of events being missed in the video recording is similar to a previous report where prehension bites were missed, mainly due to quick bites or an obscured view (Li et al., 2021). We used 2 video cameras with different views to minimize the risk of views being obscured, but it was still often difficult to see the swallowing process due to the movement and position of the cow’s head. The difficulty in ensuring a clear view of the cow’s neck, and the resulting low swallow recording rate in our experiment indicate that visual observation or video recording is unlikely to be suitable for counting swallows as part of a method to estimate the DMI of dairy cows. Moreover, the video recording method is unpractical to use in grazing cattle, whereas the audio recording method based on a microphone attached to the cow’s forehead and a data recorder placed behind the cow’s head on a halter is more practical to use in grazing cattle. Acoustic monitoring of animal behaviors has been suggested previously as a method to accurately identify eating behaviors, as animals make different sounds for different jaw movement events, and these sounds can be recorded with a microphone (Laca and WallisDeVries, 2000). Previous studies have evaluated the use of microphones to identify jaw movements of cattle (Navon et al., 2013; Galli et al., 2019), as well as chewing and intake estimates (Galli et al., 2006; Chełotti et al., 2018). The audio recording method may be used in future research as an indirect method to count swallows in cattle and generate a further understanding of the relationship between intake and swallows.

CONCLUSIONS

The the mass of the swallowed boli is greater in cows consuming FC or fresh perennial ryegrass than AH or perennial RS. Using a microphone attached to the cow’s forehead was shown to provide an accurate indirect measurement of the number of swallows a cow takes. However, a method involving video recording of the swallowing process at the pharyngeal phase is less accurate. Together this information could be used to develop a future method for estimating DMI in cattle.

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ORCIDS

N. Norbu: https://orcid.org/0000-0001-8135-6989
P. S. Alvarez-Hess: https://orcid.org/0000-0003-3681-8165
B. J. Leury: https://orcid.org/0000-0001-9173-2730
M. L. Douglas: https://orcid.org/0000-0002-4709-4572
S. R. O. Williams: https://orcid.org/0000-0003-1321-6487
V. M. Russo: https://orcid.org/0000-0002-5361-6015
M. C. Hannah: https://orcid.org/0000-0001-6708-5393
M. J. Auldist: https://orcid.org/0000-0002-5363-0319