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The Effect of Herbage Availability and Season of Year on the Rate of Liveweight Loss during Weighing of Fasting Ewe Lambs

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Abstract: Sheep (*Ovis aries*) liveweight and liveweight change can contain errors when collection procedures are not standardized, or when there are varying time delays between removal from grazing and weighing. A two-stage study was conducted to determine the effect of herbage availability and season of year on the rate of liveweight loss during fasting and to develop and validate correction equations applied to sets of delayed liveweights collected under commercial conditions. Results showed that ewe lambs offered the Low herbage availability lost up to 1.7 kg and those offered the Medium or High herbage availability lost 2.4 kg during 8 h of delayed weighing without access to feed or drinking water. The rate of liveweight loss varied by season, herbage availability and farm ($p < 0.05$). Applying correction equations on matching liveweight data collected under similar conditions, provided more accurate estimates (33–55%) of without delay liveweight than using the delayed liveweight. In conclusion, a short-term delay prior to weighing commonly associated with practical handling operations significantly reduced the liveweight recorded for individual sheep. Using delayed liveweights on commercial farms and in research can have significant consequences for management practices and research results globally, therefore, liveweight data should be collected without delay. However, when this is not feasible delayed liveweights should be corrected, and in the absence of locally formulated correction equations, the ones presented in this paper could be used.

Keywords: accuracy; correction equations; gut-fill; liveweight; liveweight loss

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1. Introduction

Liveweight (LW) is an indicator of the physical state of an animal, and change in LW is a useful tool to assess how an animal is responding to its current environment [1,2]. Liveweight is a measure of total body mass and includes muscle, fat, bone, organ, body fluids, and gut fill [2]. Advances in technology have led to commercially available automated weighing systems which combine electronic scales and use of radio frequency identification (RFID). These automated systems make it easier to regularly collect and utilize liveweight data of individuals over time [3].

Liveweight is relatively stable over a short period of time (a few minutes), but alters over longer time periods in response to environmental and physiological conditions [2,4]. Liveweight measurements can be affected by a number of factors including gut-fill (digesta and urine), growth, nutrition, health, stress, physiological state, and genotype [3,5]. The contents of the rumen (fluid and feed) can account for between 10% and 23% of total liveweight in ruminants [6,7]. Liveweight fluctuations due to gut-fill in ruminants can be affected by factors influencing feed intake such as age and size of the animal, time of day relative to sunrise, ambient temperature, and differences in grazing behavior, and time since last meal [2,4,6,8–11].

In the southern hemisphere sheep production is mainly extensive in nature and pastoral based. In New Zealand, the flock sizes on average are greater than 2500 sheep [12].

Automatic weighing systems can record up 400 weights per hour without interruptions (https://www.livestock.tru-test.com (accessed on 10 December 2020)), thus, requiring 6–7 h to weigh an average flock. Further, mustering and routine on-farm sheep handling in addition to weighing can increase the length of time sheep are restricted from accessing feed and water supplies while waiting to be weighed. Delays in weighing can lead to weight loss due to a reduction in gut-fill and body fluids [8–10]. In ewe lambs, varying levels of weight loss have been reported within flocks waiting to be weighed. Previously we have reported losses of 1.8 (4.7% of initial weight) to 2.9 (6.7% initial weight) kg after 8 h [13]. Hughes [6] reported losses of 0.5–1.2 kg (1.8–3.8% of initial liveweight) after 6 h and 1–1.7 kg (3.7–5.3% of initial liveweight) after 12 h. Burnham, Morel [9] and Wishart, Morgan-Davies [2] reported liveweight losses after 6 h of 4.2 kg (9.8% of initial liveweight), 4.8 kg (7.8% of initial liveweight), and 2.9 kg (5.6% of initial liveweight), respectively. These levels of liveweight loss can interfere with the accuracy of comparison of liveweights, and changes in liveweight over time.

Several strategies can be used to reduce variability in liveweight including removal of feed and water for fixed periods of time prior to weighing, standardizing weighing procedures, taking multiple liveweights readings per individual per day over successive days, weighing at a specific time relative to sunrise, standardizing the feed offered prior to weighing and/or increasing the number of animals and repetitions of a study [2,4]. Such methodologies to reduce variation are time consuming and, therefore, not practical for on-farm commercial use. Thus, there is a need for a new approach to determine and adjust for variations in liveweight among animals across time. The on-going improvements in weighing equipment, software, and data management [3,10] may offer a solution, as there is capacity for the time stamping of individual animal weights. Liveweight is used as a measure of an animal's productivity providing a basis for decision making regarding sheep management. Inaccurate liveweights can lead to wrong conclusions where individual animal growth performance or a comparison of liveweights is required. It is, thus, imperative that accurate liveweights are determined and used in sheep management.

Prefasting gut-fill has been found to be important in determining the rate of sheep liveweight loss during fasting [14–16]. The degree of gut-fill, retention time of particles in the gastrointestinal tract, and passage rate can be affected by the quality and quantity of dry matter intake in ruminants [17–20]. Therefore, it is likely that differences in the type and amount of herbage availability offered to sheep can result in variation in liveweight loss during fasting.

To date, the effect of herbage availability, season, and their interaction on the liveweight loss of young sheep during fasting has not been reported. The aim of this study, therefore, was to firstly, investigate the effect of herbage availability (Low, Medium, and High) and season on the rate of liveweight loss in ewe lambs when removed from herbage. Secondly, to generate and validate ewe lamb liveweight loss correcting equations. If such equations could be developed, they could then be incorporated into modern weighing systems to allow for more accurate liveweight data recording. It was hypothesized that differences in herbage availability and season would affect the rate of liveweight loss when ewe lambs were fasted.

2. Materials and Methods

2.1. Overall Study Design

This was a two-stage study. Stage one (calibration stage) profiled the liveweight and liveweight loss of ewe lambs offered three feeding herbage availability levels (Low, Medium, and High) over two seasons (autumn and early winter). Stage two (validation stage) evaluated liveweight loss correction equations developed from stage one, on different ewe lambs.

2.2. Stage 1: Calibration

2.2.1. Location

The experimental site was at Massey University's Keeble farm, 5 km south of Palmerston North (40°24′ S and 175°36′ E), New Zealand. The experiment was conducted from 22 March 2019 to 4 April 2019 (autumn) and repeated from 18 June 2019 to 1 July 2019 (winter). Weather data for both seasons is presented in Figure 1.

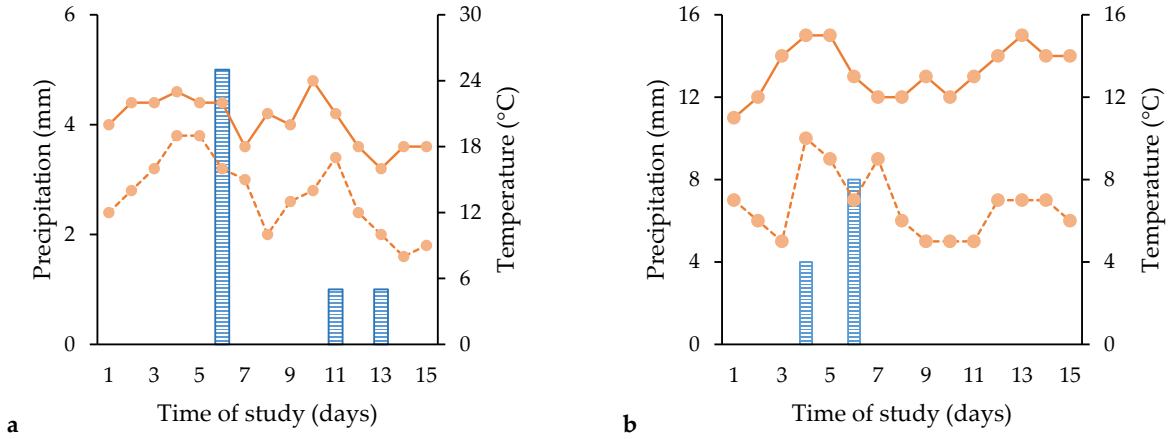

Figure 1. Daily precipitation (stripped bars) and temperature (solid line: maximum, dashed: minimum) during the study time in autumn (**a**) and winter (**b**). Data from: https://cliflo.niwa.co.nz (accessed on 6 August 2019).

2.2.2. Study Animal Conditions, Experimental Design, and Feed Management

A total of 180 Romney ewe lambs were used in this study. In autumn (from 30 March to 16 April 2019), 90 ewe lambs (6–7 months of age) were selected for the study. In winter (27 May to 13 June 2019) a different group of 90 ewe lambs (8–9 months of age) were selected. The lambs were obtained from Massey University's Keeble farm and all had electronic identification ear tags (EID) and were weighed individually. The ewe lambs were randomly assigned on day one, to one of three ryegrass based herbage availability levels; 700–900 kg DM/ha (Low herbage availability target range, $n = 30$), 1100–1300 (Medium, $n = 30$), and ≥1400 (High, $n = 30$) (Figure 2). These three herbage availability levels were selected as they represented the range of potential masses these ewe lambs might be offered in normal farm practice in New Zealand. Previous studies have shown that herbage masses of 800–1000 kg DM/ha, 1200–1400 kg DM/ha have been associated with maintenance and daily liveweight gains of 120–160 g/d, respectively [21,22]. The herbage areas were 1.9 ha (Low), 2.1 ha (Medium), and 2.0 ha (High). The study had the approval of Massey University ethics committee (protocol number: MUAEC 18/98).

Figure 2. Herbage availability (Low herbage availability target range: 700–900 kg DM/ha; Medium: 1100–1300 kg DM/ha; High: >1400 kg DM/ha) offered to ewe lambs during the study time.

2.2.3. Liveweight Measurement

Ewe lambs were weighed using Tru-Test^{TM} MP600 load bars and XR5000 weigh head (Tru-Test Group, Auckland, New Zealand). The weighing system collected liveweights at a resolution of 0.1 kg for liveweights between 0 and 50 kg and 0.2 for weights between 50 and 100 kg. At day seven, lambs were weighed immediately after arriving at the weighing facility from their paddock (within ten minutes of removal from herbage), and then again at hourly intervals for the following 8 h, in their respective herbage availability levels which were weighed in the same group sequence. During their stay at the weighing facility, ewe lambs did not have access to feed or water. After 8 h, the ewe lambs were returned to their paddocks. This procedure occurred on two more occasions within each season, while the lambs grazed their respective herbage availability levels (autumn: days 7, 11, and 14; winter: days 7, 12, and 14).

2.2.4. Herbage Sampling, Mass and Quality

To determine the grazing herbage dry matter (DM) mass and ensure that the herbage availability levels were maintained within the desired ranges over the study period, rising plate meter heights were recorded at least two days before weighing of the ewe lambs and on the day of weighing. Masses were estimated using a rising herbage plate meter (plate diameter of 355 mm; Jenquip, Fielding, New Zealand) calculated from 200 readings (R) per herbage availability level/paddock. Sward heights were calculated using plate meter readings using the equation below.

$$\text{Sward height (cm)} = \left(\frac{R_1 - R_2}{200}\right) \quad (1)$$

where R_2 is the final meter reading and R_1 is the rising plate meter reading before the first measurement of the plate.

Sward height data collected within each paddock were converted to herbage mass according to an equation developed by [23].

$$\text{Herbage mass (kg DM/ha)} = 200 + 158 \times \text{sward height (cm)} \quad (2)$$

Herbage grab samples to represent what the lambs were consuming were collected at random for nutritional quality analysis across herbage availability levels and pooled within herbage availability level and day of collection, at days 7, 11, and 14 in autumn (n = 9) and 7, 12, and 14 in winter (n = 9). Samples were collected between 9.00 AM and 12.00 PM at each sampling time. Samples were divided into two and either freeze-dried and stored for further chemical analysis or used for dry matter determination (percentage dry matter and the ratio of live/green to dead matter).

Samples for each herbage availability level collection on each day, were mixed and a subsample of approximately 50 g fresh weight was recorded. The subsamples were then oven dried at 70 °C to a constant weight. The oven-dried herbage was then ground to pass through a 1-mm sieve and analyzed for crude protein (CP), crude fiber (ADF), neutral detergent fiber (NDF), and digestible organic matter (OMD) using near-infrared reflectance spectroscopy (NIRS; Model: FOSS NIRSystems 5000, Silver Spring, Maryland, USA) [24–26] calibrated for high water soluble carbohydrate (WSC) grasses (FeedTech, AgResearch Grasslands, Palmerston North). Additionally, a prediction of the metabolizable energy (ME) of the feed was determined using organic matter digestibility (OMD*0.16 MJ/kg) [27,28]. The NIRS system estimates forage composition by comparing the spectral scan with a database of spectral and analytical information (predetermined from wet chemistry) to give an estimate of chemical composition [26].

The fresh herbage sample was weighed before being oven dried at 70 °C for 48 h, and then reweighed to determine its dry matter content (DM) % using the formula below.

$$\text{DM \%} = \frac{(\text{Fresh weight} - \text{Dry weight})}{\text{Fresh weight}} \times 100 \quad (3)$$

Further, a subsample (approximately 20 g) of fresh herbage was sorted into live and dead matter and then oven dried for dry matter estimation. The dry samples (live and dead) were then weighed separately to determine their dry weights. The proportion of live (green) matter to dead was calculated per herbage availability level as follows.

$$\text{Live matter } \% = 100 \times \left(\frac{\text{Dry weight of green herbage}}{\text{Total dry weight (green + dead)}}\right) \quad (4)$$

2.2.5. Statistical Analyses

All analyses were conducted using the R program version [29]. During the analysis, residuals (error term) were visually explored using residual plots (i.e., for potential outliers based on Cook's distances [30], for normality using qqplots and heteroscedasticity using residual vs. fitted plots). Additional tests undertaken included, Shapiro–Wilk test [31,32] for normality and the Breush–Pagan test for heteroscedasticity [33]. Extreme outliers were excluded from the final analysis basing on their influence (outliers with Cook's distances >4/sample size, were then considered influential points) in the final model [34]. The final model residuals met the assumption of normality, linearity, and homoscedasticity. There was, however, temporal autocorrelation in the residuals based on visual inspection of the autocorrelation plots.

Prior to analysis, data were partitioned into two while maintaining the class balance for different herbage availability level and physiological state of groups as follows: 70% of the measurements were used to train the model (training set: data from which the equations were derived), and the remaining 30% were used to cross-validate the model (test dataset). The ewe lamb liveweight loss training dataset was expanded using a 1000-fold cross validation resampling technique and using different splits each time. To predict ewe lamb weight loss, a linear mixed effects model with polynomial time effect and a first order correlation structure was fitted using R program [29] with the nlme package extension [35]. All effects in the model were compared based on Sidak's multiple-comparisons tests as in [13]. Herbage availability (H) together with season (S) were fitted as fixed variables, holding time (T: first and second order polynomial) as covariate while an individual sheep effect was fitted as a random effect. Initially, all variables were fitted including their two-way ($S \times H$, $S \times T$, $S \times T^2$, $H \times T$, $H \times T^2$) and three-way ($S \times H \times T$ and $S \times H \times T^2$) interactions and then the nonsignificant ones eliminated through backward selection. The model with the least Bayesian information criterion (BIC) and Akaike's information criterion (AIC) values (minimal model) was retained.

Herbage mass was estimated using a general linear model fitted using the generalized least squares method (gls) in nlme package with herbage availability level, season, and sample days as fixed effect. Two-way herbage availability $\times$ season interactions nested within sample days, or three-way herbage availability $\times$ season $\times$ sample days were tested. The model with nesting structure having had the least AIC and BIC values was selected as most fitting for further analysis. All model effects were compared using the minimal model.

2.3. Stage 2: Validation

2.3.1. Location

Data collection for the validation of the equations was conducted on two different Massey University farms (Tuapaka and Riverside), New Zealand. Tuapaka farm located 15 km north-east of Palmerston North City (40°20′ S, 175°43′ E) and Riverside farm was located 11 km north to north-west of Masterton (40°50′ S, 175°37′ E). The weather details for the validation sites are presented in Figure 3 below.

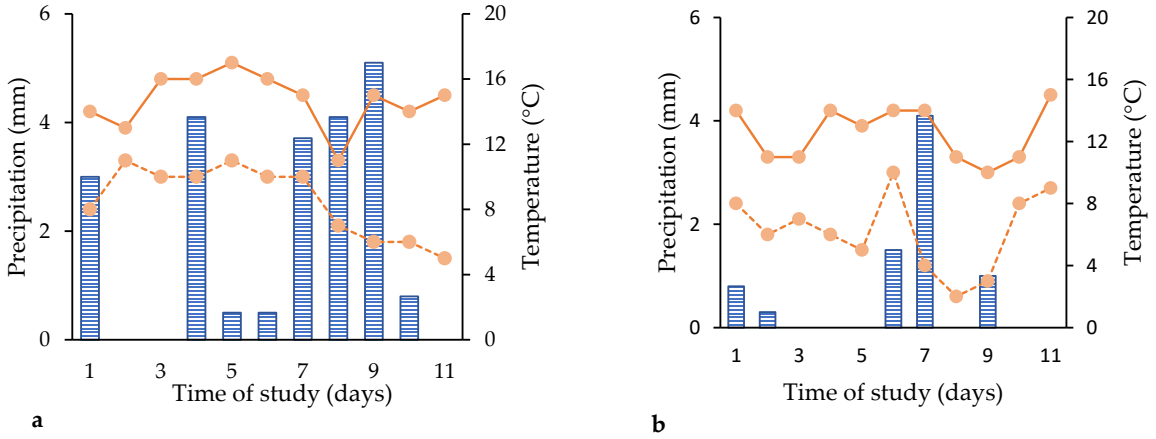

Figure 3. Daily temperature (solid line: maximum, dashed: minimum) and precipitation (stripped bars) during the validation period for Tuapaka farm (**a**) and Riverside farm (**b**). Data from: https://cliflo.niwa.co.nz (accessed on 1 September 2020).

2.3.2. Study Animals, Experimental Design, and Feed Management

Validation was conducted using eight-month-old Romney ewe lambs (n = 90) at Tuapaka farm from the 30th July to 8th June and at Riverside farm from the 7th to 16th July (n = 90) in the winter of 2020. On day one, ewe lambs (n = 30) were randomly allocated on to one of three herbage availability levels (Low, Medium, and High as per the calibration stage). The herbage was a ryegrass and white clover-based sward mix. The herbage availability areas were 1.5 ha (Low), 3.7 ha (Medium), and 2.0 ha (high) for Tuapaka, and 3.0 ha (Low), 4.1 (Medium), and 3.1 ha (High) for Riverside.

2.3.3. Liveweight Measurement

Ewe lambs were placed on their respective herbage availability levels/paddocks (only one paddock per herbage availability level) for three days (days −3 to day 0) prior to start of the study. The ewes were weighed on days 4 and 6 at Tuapaka farm and days 4 and 7 at Riverside farm. Ewe lambs were weighed in their respective herbage availability groups immediately after arriving at the weighing facility from their paddock (within 10 min of removal from herbage to get the without delay weight), and then hourly in the same group sequence for the following 6 h. During their stay at the weighing facility, ewe lambs did not have access to feed and water. After 6 h, the ewe lambs were returned to their paddocks. The study had the approval of Massey University ethics committee (protocol number: MUAEC 19/53).

2.3.4. Herbage Sampling, Availability Determination, and Herbage Quality

Herbage availability was recorded on the first day (day one), first weighing (within 2–4 days) and last weighing day (within 5–7 days) of the study only. Herbage samples were collected on each day of weighing and analyzed for quality parameters and for dry matter percentage and proportion of live to dead matter as per the calibration stage of study.

2.3.5. Statistical Analyses

The validation datasets generated in stage two were collected using different groups of ewes. Two datasets, each containing 630 records of liveweights (seven weights taken in 6 h including the "without delay") from 90 ewe lambs were collected at each study farm. The 6-h fasting period was considered a more practical period of delay that may occur during routine handling and weighing of a flock of sheep [2].

To determine if the rate of liveweight loss was consistent across farms and study stages, data from the winter season in stage one, from Keeble farm (n = 1730), using up to 6 h of fasting, was pooled with the two validation datasets from Tuapaka farm (n = 1078) and Riverside farm (n = 1257). A mixed effects model with a first-order correlation structure was

fitted using R program [29] with the nlme package extension [35]. All effects in the model were compared as in stage one. Herbage availability (H) and farm (F) were fitted as fixed variables, holding time (T: first and second order polynomial) as covariate, and individual ewe lamb was fitted as a random effect. All variables were fitted including their two-way ($F \times H$, $F \times T$, $F \times T^2$, $H \times T$, $H \times T^2$) and three-way ($F \times H \times T$ and $F \times H \times T^2$) interactions. Additionally, models were refitted with herbage availability and time effects nested within farm. The model without nesting structure having had the smallest AIC and BIC values (loglikelihood ratio, $p < 0.001$) was selected as most appropriate for further analysis. The nonsignificant model terms were eliminated, and the minimal model subsequently selected as in the calibration stage. Season was not considered as validation data were only collected in winter.

For herbage mass prediction, a general linear model was fitted as in calibration stage with herbage availability level, farm and sample days as fixed effect. Further, two-way farm $\times$ herbage availability interactions nested within sample days, and three-way herbage availability $\times$ farm $\times$ sample days were tested. The model with nesting structure was selected as most fitting for further analysis. All model effects were compared using the minimal model.

Following the linear mixed effects regression model analysis in stage one, six separate correction equations were generated at stage one, representing each herbage availability offered (Low, Medium, and High) and season (autumn, winter). This resulted in six liveweight loss equations. The formula for computing the corrected liveweight (cW_0) is given below.

$$cW_0 = dWt + Wlt \quad (5)$$

where, dW_t was the delayed or observed weight measurement at time (t) and Wl_t was ewe weight loss after time (t) off feed (t = 0, . . . , 6 h) computed using the separate or consolidated weight loss equations generated in stage one.

Even though up to 6 h of fasting would be the quintessential delayed time during on-farm weighing, 8 h were preferred for developing the liveweight correction equations and the subsequent without LW predictions. This is because the 8-h-based correction equations covered more data (time points) and thus, more accurately modeled the liveweight loss trend within and after the 6-h fasting period.

Liveweight correction equations were deployed to predict the without delay liveweight on validation datasets collected during winter from two farms (Tuapaka and Riverside). Validation data could not be collected on ewes in autumn due to the COVID-19 lockdown imposed in New Zealand from March to June. Several metrics (Table 1) were used to assess the quality of models, including the coefficient of determination (R^2), Lin's concordance correlation coefficient (CCC), bias, root mean squared error (RMSE), residual prediction deviation (RPD), and the ratio of performance to interquartile distance (RPIQ) [36–39]. The success of the predictions for individual samples was determined using the relative percent error (RPE). The best model would have the highest R^2, CCC, RPD, and RPIQ, and the lowest RMSE and RPE. In addition, RPD has been classified [37,40] into three different categories, weak prediction (RPD < 1.4), reasonable (1.4 < RPD < 2.0), and excellent (RPD > 2.0). In a similar manner, RPIQ has been divided into four categories, very poor prediction (RPIQ < 1.4), fair (1.4 < RPIQ < 1.7), good (1.7 < RPIQ < 2.0), very good (2.0 < RPIQ < 2.5), and excellent (RPIQ > 2.5) [41].

Each validation was conducted using 1000-fold cross validation (bootstrap) with three repeats. In theory as the number of times a bootstrap is conducted increases (large number of folds), the bootstrap standard deviation approximates sample standard error [42]. Consequently, 1000 bootstraps were conducted to estimate the descriptive statistics on accuracy and error metrics (mean, standard deviation, interquartile range).

Table 1. Goodness of fit and accuracy measures of the calibration (stage one) equations applied to the validation (stage two) data sets.

Metric	Equation
Bias	$\text{Bias} = \frac{1}{n}\sum_{j=1}^{n}\left(x_j - y_j\right)$
Root mean square error	$\text{RMSE} = \sqrt{\frac{\sum_{j=1}^{n}\left(x_j - y_j\right)^2}{n}}$
Relative prediction error	$\text{RPE} = \frac{\text{RMSE}}{\bar{x}} * 100$
Residual prediction deviation	$\text{RPD} = \frac{SD_{y_j}}{\text{RMSE}}$
Ratio of performance to interquartile distance	$\text{RPIQ} = \frac{(Q_3 - Q_1)}{\text{RMSE}}$
Coefficient of determination	$R^2 = \frac{\sum_{j=1}^{n}\left(x_j y_j - \overline{xy}\right)^2}{(\sum_{j=1}^{n} x_j^2 - \bar{x}^2)(\sum_{j=1}^{n} y_j^2 - \bar{y}^2)}$
Lin's concordance correlation coefficient	$\text{CCC} = \frac{2\rho SD_x SD_y}{SD_x^2 + SD_y^2 + (\bar{x} - \bar{y})^2}$

where n is sample size, x_j and y_j are the actual and predicted values, respectively, and $\bar{x}$ and $\bar{y}$ are their respective means. ρ is the Pearson's correlation coefficient between the observed and predicted values. SD is standard deviation, Q1 and Q3 are the 25th and 75th quartiles, respectively.

3. Results

3.1. Calibration Stage

3.1.1. Herbage Mass and Chemical Composition

The mass of available herbage (kg DM/ha), differed among herbage availability target levels ($F_{2,12}$ = 153.7, $p < 0.001$) and between seasons ($F_{1,12}$ = 4.60, $p < 0.05$) but not ($F_{2,12}$ = 0.06, $p = 0.941$) period of study (time from day 0 to day 14) (Table 2). Further, the interaction between herbage mass and season was not significant ($F_{2,12}$ = 0.50, $p = 0.613$). The proportion of herbage that was considered live (green) and thus edible differed by season ($F_{1,12}$ = 9.4, $p < 0.001$) and increased with herbage availability ($F_{1,12} = p < 0.05$).

Table 2. Estimated post feeding herbage mass (least squares means) and proportion of live dry matter (%) of Low, Medium, and High herbage availability levels (kg DM/ha) offered to ewe by season (autumn, winter) during calibration.

Item	Autumn	Winter
Herbage mass (kg DM/ha)		
Low	854.6 ^{a}	924.46 ^{a}
Medium	1216.06 ^{b}	1219.16 ^{b}
High	1911.2 ^{c}	1870.4 ^{c}
Proportion of live dry matter (%)		
Low	19.5 ^{a}	56.4 ^{c}
Medium	30.7 ^{b}	74.5 ^{d}
High	36.6 ^{b}	83.0 ^{d}

^{abcd}: different superscripts denote significant difference at $p < 0.05$ across rows and columns within measurement type. Herbage availability: Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300 kg DM/ha, High: ≥1400 kg DM/ha). All tests and comparisons were based on Sidak's multiple comparison methods. Standard error of mean difference (herbage mass: availability level: 33.1 kg DM/ha, season: 23.2 kg DM/ha; proportion of dry matter: 2.15%. Single SEM value for live dry matter comparison across rows and within columns indicates a significant herbage availability × season interaction).

The herbage chemical composition varied ($p < 0.01$) by season of year but not ($p > 0.05$) herbage availability (Table 3). Dry matter, NDF, and ADF were greater ($p < 0.05$) in autumn, while, DM, CP, and ME were greater ($p < 0.01$, Table 3) in winter. Within herbage availability, there were seasonal differences ($p < 0.05$) for all components. Within season, however, the herbage availability levels did not differ ($p > 0.05$) in all components.

Table 3. Herbage quality parameters for grab samples of the Low, Medium, and High herbage availability treatments offered to ewe lambs during autumn and winter (least square means). Analysis conducted using near-infrared reflectance spectroscopy (NIRS) method.

	Autumn			Winter		
Parameter	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
DM %	26.4 ^{b}	26.1 ^{b}	26.7 ^{b}	19.1 ^{a}	18.7 ^{a}	19.5 ^{a}
CP %	16.8 ^{a}	18.3 ^{ab}	18.4 ^{ab}	21.6 ^{abc}	25.8 ^{bc}	27.3 ^{c}
NDF %	52.9 ^{c}	52.0 ^{c}	50.5 ^{bc}	43.2 ^{ab}	42.2 ^{ab}	39.1 ^{a}
ADF %	30.1 ^{c}	30.2 ^{c}	28.3 ^{bc}	23.5 ^{ab}	23.1 ^{ab}	21.8 ^{a}
ME (MJ/kg)	9.5 ^{a}	9.5 ^{a}	9.8 ^{a}	11.5 ^{b}	11.4 ^{b}	11.4 ^{b}

DM: dry matter; CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber (ADF); ME: metabolizable energy. Herbage availability: Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300 kg DM/ha, High: ≥1400 kg DM/ha. ^{abc}: different superscripts within a row denote significant difference at $p < 0.05$. All tests and comparisons were based on Sidak's multiple comparison methods. Standard error of mean difference (Herbage mass: availability level: 1.34, season: 1.09; proportion of dry matter: 2.15, a single value for both live and dead matter represents mean comparisons across rows). Standard error of mean difference % (DM: 0.58, 1.62; CP: 1.09, 1.34; NDF: 1.09, 1.33; ADF: 0.76, 0.93; ME: 0.18, 0.23 for effect of season and herbage availability, respectively).

3.1.2. Effect of Herbage Availability and Season on Liveweight Loss

Overall, the liveweight loss data were highly variable as indicated by the coefficient of variation (CV = 31–48%) (Table 4). The overall liveweight loss of lambs did not vary by herbage availability ($F_{2,\ 4173} = 0.53$, $p = 0.589$) or season ($F_{1,\ 4173} = 0.13$, $p = 0.722$) over the 8-h period. However, this loss in liveweight varied linearly ($F_{1,4173} = 114.6$, $p < 0.001$) but not nonlinearly ($F_{1,4173} = 0.34$, $p = 0.558$) with fasting time. All two-way and three-way interactions were nonsignificant ($p > 0.05$) except for herbage availability × time (first order polynomial) ($F_{2,4173} = 4.35$, $p = 0.01$). The rate of liveweight loss stayed uniform (straight line) over time in autumn for each herbage availability level and for ewe lambs offered the Low herbage mass during winter (Figure 4). However, for ewe lambs offered the Medium and High herbage masses in winter, liveweight loss was greater in the first 4 h of fasting ($p < 0.05$) compared to the last 4 h in winter.

Table 4. Mean initial (without delay) and final weight and prediction parameters with standard errors in parentheses, for ewe lamb liveweight loss (kg) based on herbage availability (L: Low, M: Medium, H: High) offered to ewes by season (autumn, winter) and fasting time (1–8 h). CV is the coefficient of variation and adjusted R^2 is a measure of goodness of fitness of the model. All models were significant at $p < 0.05$.

Herbage Availability	Liveweight (kg)		Predictor			Coefficient of Variation (CV)	Adjusted R^2
	Initial	**Final**	**Intercept**	**Time**	**Time ^{2}**		
				Autumn			
Low	36.8 (0.42)	35.0 (0.39)	0.01 (0.08)	0.20 ^{a} (0.02) **	ns	0.31	0.69
Medium	37.6 (0.43)	35.9 (0.41)	0.11 (0.08)	0.23 ^{a} (0.02) **	ns	0.41	0.69
High	37.6 (0.40)	35.6 (0.38)	0.05 (0.08)	0.27 ^{ab} (0.02) **	−0.020 ^{ab} (0.003) *	0.45	0.71
				Winter			
Low	47.3 (0.33)	45.4 (0.32)	0.10 (0.08)	0.22 ^{a} (0.02) **	ns	0.48	0.6
Medium	48.1 (0.32)	46.0 (0.32)	0.13 (0.09)	0.35 ^{bc} (0.02) **	−0.012 ^{a} (0.002) **	0.39	0.71
High	48.5 (0.34)	46.3 (0.32)	0.02 (0.08)	0.42 ^{c} (0.02) **	−0.020 ^{ab} (0.002) **	0.42	0.67

Initial liveweight: liveweight without delay. Final liveweight: liveweight after 8 h of fasting. ^{abc} superscripts within the predictor columns (Time and Time ^{2}) per category (autumn, winter, overall), denote significant difference at $p < 0.05$. ns denotes not significant at $p > 0.05$. *, ** denote significance at $p < 0.05$ and $p < 0.01$, respectively. Model goodness of fit: the higher R^2 the better. All contrasts based on Sidak's multiple-comparisons tests.

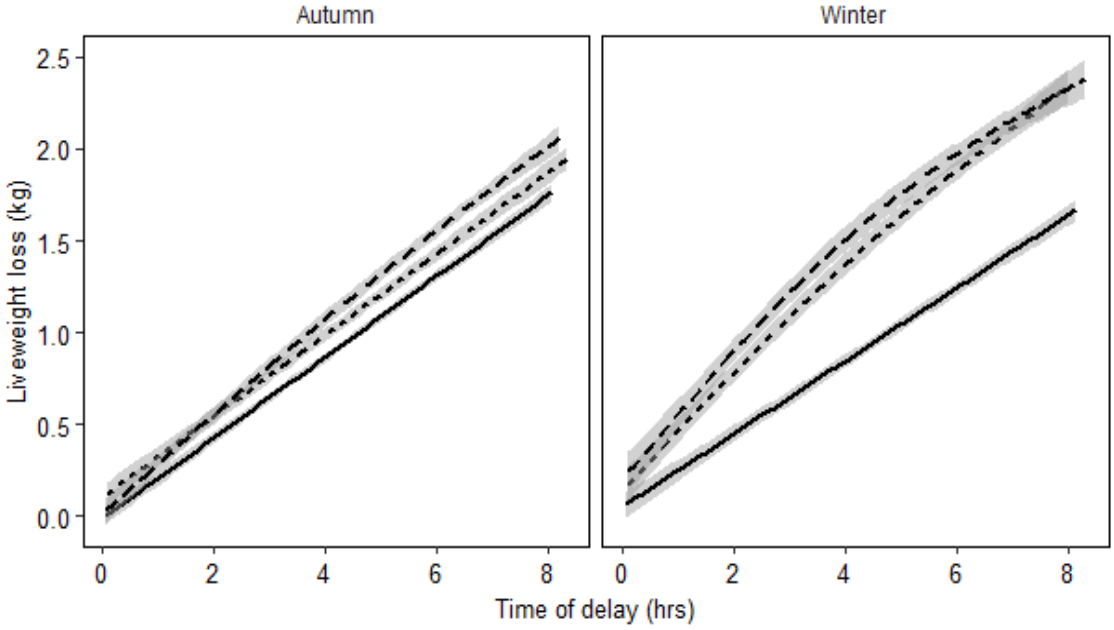

Figure 4. Change in liveweight (with 95% CI, grey shade) for herbage availability (Low: solid line, Medium: dashed line, High: long dashed line) in autumn and winter over 8 h of fasting.

Although overall ewe lamb liveweight loss was comparable ($p > 0.05$) among herbage mass availability levels and seasons, the rates of weight loss varied by herbage availability and season ($p < 0.01$) (Table 4). Generally, the rate of liveweight loss was greater ($p < 0.01$) for winter than autumn. In autumn the rate of liveweight loss was greater ($p < 0.05$) among lambs offered the high herbage mass than either Medium or Low herbage mass. In winter, the rate of liveweight loss was greater ($p < 0.01$) for lambs offered either High or than Low herbage availability level (Table 5).

Table 5. Estimated post-feeding herbage mass (least squares means) and proportion of live matter (%) of Low, Medium, and High herbage availability target levels (kg DM/ha) offered to ewe lambs during the period of study on Tuapaka farm and Riverside farm during validation.

Item	Tuapaka Farm	Riverside Farm
Herbage mass (kg DM/ha)		
Low	940 ^{ab}	882.3 ^{a}
Medium	1284.3 ^{b}	1232.5 ^{b}
High	1910.8 ^{d}	1530.3 ^{bc}
Proportion of live dry matter (%)		
Low	61.7 ^{a}	85.3 ^{bc}
Medium	93.4 ^{bc}	71.2 ^{ab}
High	94.5 ^{c}	86.2 ^{bc}

^{abcd}: different superscripts denote significant difference at $p < 0.05$ across rows and in columns for herbage availability and proportion of live matter. Herbage availability (Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300, High: $\geq$ 1400). Standard error of mean difference (110 kg DM/ha; 6.20% for herbage mass and proportion of dry matter, respectively. Single SEM value for herbage availability and live matter comparisons across rows and within columns indicates a significant herbage availability level $\times$ farm interaction).

After 8 h of fasting in autumn ewe lambs lost 1.54 kg (4.2% of initial weight), 1.60 kg (4.3% of initial weight), and 2.0 kg (5.3% of initial weight) for Low, Medium, and High herbage availability levels, respectively. In winter, ewe lambs lost 1.50 kg (3.2% of initial weight), 2.60 kg (4.8% of initial weight), and 2.62 kg (5.4% of initial weight) for Low, Medium, and High herbage availability levels, respectively (Figure 5).

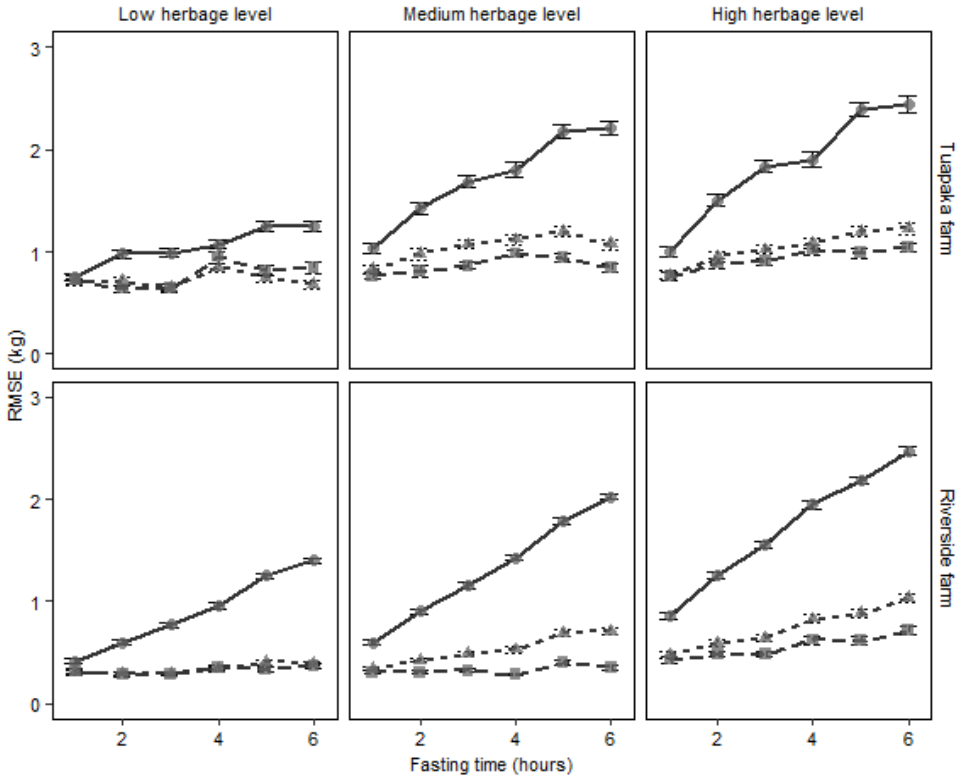

Figure 5. Change in root mean square error (RMSE) with associated standard deviation for the prediction of true ewe liveweight over time of fasting when individual respective correction equations (solid line with circular points: no model, dotted line with triangular points: autumn model, dashed line with square points: winter model) for each target herbage availability (Low, Medium, and High) and season generated in stage one were applied to the data collected in the winter season of 2020 to predict the without delay liveweight by farm (Tuapaka or Riverside). Herbage availability (Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300, High: ≥1400).

3.2. Validation Stage

3.2.1. Herbage Mass and Chemical Composition

Herbage mass differed among herbage availability target levels and farms ($p < 0.01$) but not period between sample days ($F_{1,12} = 0.90$, $p = 0.361$) (Table 5). With the exception of herbage availability × farm ($F_{2,12} = 4.48$, $p = 0.035$), all interactions between herbage availability, farm, and sample days ($p > 0.05$). The variability (range) in herbage mass was greatest in the High availability target level. Although the aim was to maintain the herbage availability within the preset target ranges (i.e., 700–900, 1100–1300, and ≥1400 kg DM/ha), at Tuapaka farm the availability levels were slightly out of range due to unpredictable pasture growth. Consequently, Tuapaka farm had greater herbage availability levels offered to ewe lambs in both the Medium and High groups than on Riverside farm. The proportion of herbage that was considered live (green) and thus edible differed by season and increased with herbage availability ($p < 0.05$). Further, Tuapaka farm had greater live matter proportions than Riverside farm.

All herbage chemical components varied by herbage availability ($p < 0.01$). Herbage NDF and ADF were different ($p < 0.05$), while, DM ($F_{1,8} = 0.11$, $p = 0.849$) and ME ($F_{1,8} = 1.05$, $p = 0.395$) did not vary between farms (Table 6). There were no significant herbage availability × farm interactions ($p > 0.05$) for all chemical components. Metabolizable energy was greater at Tuapaka farm than Riverside farm ($p < 0.05$). Dry matter was lower for Medium and High herbage availability levels but was comparable ($p > 0.05$) for all herbage availability levels at Riverside farm. Crude protein and NDF increased with herbage mass while ADF decreased with increasing in herbage availability across farm ($p < 0.05$).

Table 6. Herbage quality parameters for grab samples of the Low, Medium, and High herbage availability levels offered to lambs during Winter season on Tuapaka farm and Riverside farm.

	Tuapaka Farm			Riverside Farm		
Parameter	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
DM %	17.9 ^{b}	16.5 ^{a}	16.7 ^{a}	17.8 ^{b}	18.6 ^{b}	17.9 ^{b}
CP %	22.8 ^{a}	26.9 ^{b}	26.9 ^{b}	24.6 ^{a}	27.2 ^{b}	27.4 ^{b}
NDF %	39.1 ^{a}	42.2 ^{b}	43.2 ^{b}	39.1 ^{a}	42.9 ^{b}	43.3 ^{b}
ADF %	21.8 ^{a}	23.1 ^{b}	23.5 ^{b}	22.3 ^{a}	19.9 ^{a}	19.7 ^{a}
ME (MJ/kg)	11.5 ^{b}	11.4 ^{b}	11.4 ^{b}	10.3 ^{a}	10.8 ^{a}	10.5 ^{a}

^{ab}: different superscripts denote significant difference at $p < 0.05$ in a row, across herbage availability and farm. DM: dry matter; CP: crude protein, NDF: neutral detergent fiber NDF; ADF: acid detergent fiber; ME: metabolizable energy. Herbage availability (Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300, High: ≥1400). SEM values represent all comparisons across rows. All tests and comparisons were based on Sidak's multiple comparison methods. Standard error of mean difference % (DM: 0.40, 0.41; CP: 0.58, 0.46; NDF: 0.66, 0.52; ADF: 0.81, 0.73; ME: 0.08, 0.06 for effect of herbage availability and farm effect, respectively).3.2.2. Variability in liveweight loss trends at calibration and validation stage.

This section presents a comparison in the ewe liveweight loss trends during a 6-h fasting period between the calibration dataset (from Keeble farm) and two validation datasets (from Tuapaka farm and Riverside farm) (Table 7). The overall mean liveweight loss did not vary ($F_{2,261} = 0.54$, $p = 0.581$) between farms and among herbage availability ($F_{2,261} = 0.78$, $p = 0.460$) over the 6-h fasting period. However, the overall liveweight loss varied linearly ($F_{1,3778} = 21.43$, $p < 0.001$) but not nonlinearly ($F_{1,3778} = 0.21$, $p = 0.650$) with fasting time. All two-way time-based interactions were significant ($p < 0.001$). However, the herbage × farm interaction was not significant ($F_{1,261} = 0.45$, $p = 0.769$) and all three-way interactions were not significant ($p > 0.05$). The rate of liveweight loss varied among farms ($F_{2,3778} = 11.9$, $p < 0.001$) and among herbage availability levels ($F_{2,3778} = 46.7$, $p < 0.001$). The proportion of variance explained by each model (adjusted R^2) was greatest for Riverside farm and least for Keeble farm. Further, the variability in data was highest at Tuapaka farm (CV = 44–55%) and was lowest on Riverside farm (CV = 23–31%).

Table 7. Mean initial (without delay) and final weight and prediction parameters with standard errors in parentheses, for ewe lamb liveweight loss (kg) during a 6-h fasting period in winter, by herbage availability (Low, Medium, High) and farm (* Keeble, Tuapaka, and Riverside). CV is the coefficient of variation and adjusted R^2 is a measure of goodness of fitness of the model. All models were significant at $p < 0.05$.

		Liveweight (kg)		Predictor			Coefficient of Variation (CV)	Adjusted R^2
Farm	**Herbage Availability**	**Initial**	**Final**	**Intercept**	**Time**	**Time^2**		
* Keeble	Low	47.3 (0.33)	45.4 (0.32)	−0.28 (0.107) ^{e}	0.23 (0.033) ^{a}	0.01 (0.005) ^{e}	0.48	0.48
	Medium	48.1 (0.32)	46.0 (0.32)	0.35 (0.117) ^{cd}	0.45 (0.036) ^{c}	−0.02 (0.006) ^{cd}	0.39	0.62
	High	48.5 (0.34)	46.3 (0.32)	−0.62 (0.117) ^{bcd}	0.55 (0.036) ^{cd}	−0.03 (0.005) ^{bc}	0.42	0.58
† Tuapaka	Low	38.1 (0.26)	37.1 (0.24)	−0.62 (0.146) ^{abcd}	0.40 (0.046) ^{bc}	−0.03 (0.007) ^{bcd}	0.55	0.50
	Medium	41.5 (0.39)	39.5 (0.33)	−1.24 (0.150) ^{a}	0.80 (0.046) ^{e}	−0.06 (0.007) ^{a}	0.43	0.65
	High	42.6 (0.40)	40.3 (0.36)	−1.13 (0.153) ^{ab}	0.79 (0.047) ^{e}	−0.06 (0.007) ^{a}	0.44	0.65
† Riverside	Low	40.6 (0.45)	39.3 (0.44)	−0.13 (0.13) ^{de}	0.27 (0.041) ^{ab}	−0.01 (0.006) ^{de}	0.31	0.75
	Medium	43.8 (0.46)	41.8 (0.44)	−0.39 (0.137) ^{cd}	0.46 (0.043) ^{c}	−0.02 (0.006) ^{cd}	0.24	0.84
	High	43.9 (0.49)	41.4 (0.47)	−0.75 (0.144) ^{abc}	0.68 (0.045) ^{de}	−0.04 (0.007) ^{ab}	0.23	0.85

Initial liveweight: liveweight without delay. Final liveweight: liveweight after 8 h of fasting. Asterisks *,† attached to farm name indicate the dataset used for the analysis (*: Calibration dataset, †: Validation dataset). ^{abcde}: different superscripts within the predictor columns (Time and Time^2) per herbage availability and season denote significant difference at $p < 0.05$. Herbage availability (Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300, High: ≥1400). Model goodness of fit: the higher R^2 the better. All contrasts based on Sidak's multiple-comparisons tests.

3.2.2. Using Separate Correction Equations on Validation Datasets to Predict without Delay Liveweight

The regression equations derived in the calibration phase (8 h of fasting) were validated against two independent datasets (6 h of fasting) collected on lambs from two different farms (Tuapaka and Riverside) using the correction equations (equation 4.1). The validated results showed that the ewe lamb liveweight correction equations for all feeding levels by season developed in stage one of the present study, predicted liveweight with substantial accuracy as shown by their low RPE (0.75–2.93%) and high R^2 (87.9–99.3%) and RPIQ (3.33–16.8) values as compared to not using any correction method (Table 8, Figure 5).

Table 8. Initial, final, and predicted liveweights, measures of goodness of fit and accuracy (Bias, RMSE, RPE, RPD, RPIQ, r^2, CCC) for liveweight correction models based on 8 h of fasting (from autumn and winter, 2019) applied onto the independent datasets (validation datasets) collected from Tuapaka farm and Riverside farm in Winter (2020) during a 6-h fasting period after the lambs were offered the Low, Medium, and High herbage availability.

Farm	* Correction Equation (Model)	Herbage Availability	Liveweight (kg)			Bias	RMSE	RPE %	RPD	RPIQ	r^2 %	CCC %
			Actual Initial	Actual Final	Predicted Final							
Tuapaka	None	Low	38.2 (0.26)	37.1 (0.24)		−0.75	1.06	2.78	1.92	2.45	87.9	80.8
		Medium	41.5 (0.38)	39.5 (0.33)		−1.49	1.77	4.26	1.67	2.34	80.6	75.6
		High	42.6 (0.40)	40.3 (0.36)		−1.62	1.91	4.49	1.63	2.26	81.9	73.2
	From autumn	Low	38.2 (0.26)	37.1 (0.24)	38.5 (0.24)	−0.06	0.75	1.96	2.72	3.95	87.9	92.4
		Medium	41.5 (0.38)	39.5 (0.33)	41.2 (0.33)	−0.62	1.05	2.53	2.82	3.95	92.6	94.8
		High	42.6 (0.40)	40.3 (0.36)	41.9 (0.36)	−0.58	1.06	2.49	2.93	4.08	92.6	91.8
	From winter	Low	38.2 (0.26)	37.1 (0.24)	38.6 (0.24)	0.25	0.78	2.04	2.62	3.33	87.9	91.6
		Medium	41.5 (0.38)	39.5 (0.33)	41.0 (0.33)	−0.24	0.87	2.1	3.4	4.77	92.6	94
		High	42.6 (0.40)	40.3 (0.36)	42.7 (0.36)	−0.35	0.94	2.21	3.31	4.6	92.6	95.5
Riverside	None	Low	40.6 (0.45)	39.3 (0.44)		−0.84	0.96	3.55	2.42	3	87	92.9
		Medium	43.8 (0.46)	41.8 (0.44)		−1.27	1.41	3.9	2.18	3.25	84.9	85.5
		High	43.9 (0.49)	41.4 (0.47)		−1.67	1.8	4.1	2.1	3.07	89.4	80.9
	From autumn	Low	40.6 (0.45)	39.3 (0.44)	40.6 (0.44)	−0.16	0.34	0.84	10.26	12.12	99.3	99.5
		Medium	43.8 (0.46)	41.8 (0.44)	43.5 (0.44)	−0.42	0.55	1.26	6.46	10.1	99.2	99.2
		High	43.9 (0.49)	41.4 (0.47)	43.0 (0.47)	−0.64	0.77	1.76	4.91	7.18	99.1	96.6
	From winter	Low	40.6 (0.45)	39.3 (0.44)	40.7 (0.44)	0.14	0.33	0.81	10.57	13.11	99.3	99.5
		Medium	43.8 (0.46)	41.8 (0.44)	43.4 (0.44)	−0.04	0.33	0.75	10.77	16.82	99.2	98.9
		High	43.9 (0.49)	41.4 (0.47)	43.8 (0.47)	−0.42	0.57	1.3	6.62	9.69	99.1	99.4

Herbage availability (Low herbage availability target range: 700–900 kg DM/ha, Medium: 1100–1300, High: ≥1400). Interpretation of measures: The best model has the highest RPD (residual prediction deviation), RPIQ (ratio of performance to interquartile distance), r^2 (coefficient of determination), CCC (Lin's concordance correlation coefficient), and the lowest root mean square error and RPE (relative prediction error). RPD (<1.4: weak, 1.4 < RPD < 2.0: reasonable, >2.0: excellent). RPIQ (<1.4: very poor, 1.4 < RPIQ < 1.7: fair, 1.7 < RPIQ < 2.0: good, 2.0 < RPIQ < 2.5: very good, >2.5: excellent). * Correction equation (None indicates delayed liveweight considered).

Prediction error varied ($p < 0.05$) with time of fasting, herbage availability, and farm. The prediction error (RMSE) increased with ewe lamb liveweight loss over time (Figure 5). Prediction error was highest in the High herbage availability and lowest in the Low herbage availability. The prediction error was also greater ($p < 0.05$) for Tuapaka farm than Riverside farm in all herbage availability levels. Further, prediction error varied by season from which the prediction model was developed ($p < 0.05$). Liveweight correcting models developed in winter were more accurate in predicting the without delay liveweight (i.e., directly off herbage) than those from autumn for Medium and High herbage availability levels but not for the Low herbage availability. Low herbage availability weight correcting equations had comparable accuracy or prediction error regardless of model season. Using the herbage availability and season specific correcting models to predict the without delay liveweight when lambs were offered the High herbage availability prior to fasting increased the prediction accuracy of the without delay liveweight estimates by 50.5% and 58.8% for models developed in autumn and winter, respectively, compared to using the delayed weights (not immediately off herbage). The correcting equations increased the accuracy of the without delay liveweight estimates in lambs offered Medium herbage availability by 48.1% and 58.8% using models developed in autumn and winter, respectively, compared to the delayed weights. The correcting equations increased the accuracy of the without delay

liveweight estimates in lambs offered Low herbage availability by 44.1% and 41.2% using models developed in autumn and winter, respectively, compared to the delayed weights.

4. Discussion

The current study was conducted in two stages aimed at, firstly, calibration by determining the effect of herbage availability and season on the rate of liveweight loss of ewe lambs during fasting and, secondly, validation by determining if without delay liveweight of ewe lambs could be accurately predicted from delayed liveweights.

4.1. Calibration Stage

The findings indicated that ewe lambs lost a significant amount of liveweight (autumn: between 4.2% and 5.3% of initial weight, winter: 3.2–5.4% of initial weight) between each weighing throughout the fasting period. The magnitude of this change is likely to influence the reliability of liveweight measures which will likely have implications for management decisions both on-farm and for research trials, unless it can be corrected for. The findings support our previous study which profiled liveweight losses of Romney ewe lambs offered either grass or herb-clover based swards [13].

The current study indicated that the rate of liveweight loss was affected by both herbage availability and season of year, suggesting that different equations may be required to accurately correct for liveweight if animals are off pasture for periods of greater than 60 min. The variation in ewe lamb liveweight loss rate by herbage availability was likely due to differences in gut-fill volume and differences in the chemical composition of the pasture [43]. The DM content of the herbage was consistently lower in the High and Medium herbage availability levels than Low herbage availability level. It is, therefore, possible that the ewe lambs were consuming more water from the Medium and High herbage availability levels than the Low herbage, with this excess water being excreted faster through urine, than would herbage via fecal defecation. The seasonal differences in the chemical composition of the feeds may also have been responsible for the differential lamb liveweight loss. The lower proportions of CP and ME, but with correspondingly higher fiber (DM, NDF, and ADF) may have been responsible for the lower rate of lamb liveweight loss in autumn compared to winter. Greater structural carbohydrate and higher levels of fiber are known to increase water holding capacity in the sheep gut and thus reduce the rate of ruminal flow [7]. In drier seasons, the proportion of fermentable carbohydrates and pectin content decrease while the structural carbohydrates (NDF and ADF) increase, however, in wetter seasons the reverse is true [44–49]. Therefore, it is not surprising that autumn (dry season) herbage had the highest DM and fiber and thus, the lowest rate of lamb liveweight loss compared to winter. The seasonal differences in liveweight loss could also be attributed to the higher ambient temperature experienced in autumn compared to winter during the study period. Exposure to colder temperatures can increase reticulo-rumen motility, increase the passage rate of gut particles, and reduce the gut-fill retention time [50].

4.2. Validation Stage

The significant polynomial regression obtained between liveweight loss and time off feed, and the subsequent linear association between delayed and without delay liveweight, suggests there is a relationship between weight loss and without delay liveweight. This is predicated on the hypothesis that the amount of weight lost per unit time varies depending on herbage availability and season. It was observed that the weight prediction equations tended to be more linear rather than curvilinear when animals were offered low herbage availability, or high DM%, but were curvilinear when herbage availability or when time off herbage was increased.

A comparison of liveweight loss trends using calibration and validation datasets demonstrated significant differences in overall liveweight loss and liveweight loss rates between farms. A significant farm × time interaction indicates differences in overall

liveweight loss rates among farms. Further, the results indicated a greater CV% associated with this liveweight loss, which was highest at Tuapaka farm and lowest at Riverside farm. The herbage availability target ranges varied in availability levels and dry matter content (Tables 2, 3, 5 and 6) which might explain the differential weight losses on different farms. Additionally, at both Keeble farm and Tuapaka farm, liveweights were recorded manually by the operator whereas at Riverside farm, weights were automatically recorded. Comparison weighing was done using two 20 kg loads at the start of each weighing. However, it is possible that some error was introduced while the operator forgot to readjust the scale reading to zero each time a "shy" ewe rapidly and violently rammed into the crate gates shifting the position of the crate. An automated weighing system regularly readjusts the scale to zero, thereby reducing the error introduced due to shifts in the position of the crate.

Ideally, weighing without any delay (immediately off pasture) should provide ewe lamb liveweight measurements with the least error. However, if this is not achievable, the validating process has demonstrated that correction equations can be used to supply corrected liveweights (cW_0) that are more accurate estimates of the without delay liveweight (aW_0) than a delayed liveweight (dW_t). This provides a major step towards achieving improved (precise) liveweight measurement in sheep production.

All correction equations were based on the 8-h fasting period. This provided more data and, therefore, explained more variability in ewe liveweight loss. The precision of the correction equations was significantly impacted by herbage availability, season, the period of delay in recording, animal weight, and farm. This is in partial agreement with Wishart, Morgan-Davies [2] who reported a significant impact of grazing location on the precision of liveweight correction equations, but not time of delay before the weighing of ewes. Their study showed that the precision of the correction equations was affected by the factors associated with fluctuations in gut-fill [2,4].

The correction equations were substantially more stable when predicting without delay liveweight in ewe lambs offered the Low herbage availability than the Medium or High herbage availability. The consistently stable precision associated with the Low herbage availability was likely due to the higher DM% which might have caused greater water retention in the gut than for ewe lambs feeding on a lower DM% (Medium and High). In addition, the lower quantity (kg DM/ha) of herbage within the Low availability could have restricted the gut-fill thereby eliciting a response to reduce ruminal emptying. Lambs offered the High herbage availability had access to wider herbage availability ranges (1400–2200 kg DM/ha) than those offered the Low availability (700–900 kg DM/ha) which likely explains their greater error rates. However, it has been previously reported with mature ewes that intakes do not increase above a herbage availability of approximately 1400 kg DM [51].

Riverside farm had more accurate liveweight estimates regarding the calibration dataset than Tuapaka farm and these differences in prediction accuracy could be explained by the variations in herbage availability levels offered to ewe lambs especially in the Medium and High herbage availability. The herbage availability estimates (Medium and High) offered to ewe lambs on the Tuapaka farm were slightly greater than those on Riverside farm. Further, the differences in prediction accuracy could also be attributed to variation in herbage dry matter percentage DM % between farms at the time of the study. This was not unexpected as Tuapaka farm is located in an area which receives more rainfall compared to Riverside farm. Overall, the results appear to suggest that increased DM % resulted in a more accurate estimate of without delay liveweight.

In the current study, all predictions were executed on a dataset collected over one season (winter). We however, validated all the liveweight correcting equations developed from the two seasons (autumn and winter). It was not surprising that the correction equations developed for winter gave more accurate estimates than those for autumn, given the timing of the validations. However, results suggest that applying an equation from a

different season to predict the without delay liveweight from delayed liveweight is a better option than using the delayed weights themselves.

The validations were conducted using a range of herbage availability levels and liveweights which should cover most situations for an extensive sheep system grazing a ryegrass-based pasture. The use of simple and multiple linear regression equations based on time stamps to predict liveweight loss and to predict without delay liveweight in sheep has been previously reported [2]. They predicted the without delay liveweight based on time off pasture with no reference to nutritional differences and did not provide an indication of how accurate their models were compared to not using the equation. The current study corroborates the suggestions made by Wishart, Morgan-Davies [2] that the differences in quality and quantity of herbage as well as environmental factors [6,7] which impact liveweight variation, contribute to the differences between sheep from different across farms and feeding levels.

The results of the present study demonstrated that it is possible to obtain substantially accurate estimates of without delay liveweight of lambs offered varying availability levels of herbage prior to weighing and in different seasons of the year. It is important to correct for liveweight losses associated with handling and delayed weighing of sheep. The developed equations utilized time recorded by the weighing systems to compute the time period from pasture to weighing and adjust for weight. To use these equations if incorporated into modern weighing systems, the time when ewe lambs are removed off pasture, would need to be manually entered.

5. Conclusions

The present study showed that ewe lambs lose a significant amount of liveweight while feed and drinking water are restricted. The study demonstrated that the rate of ewe lamb liveweight loss can be predictable over a period and is dependent on herbage availability offered and season. Further, the study demonstrated that these liveweight losses can be substantially accounted for using sets of correcting equations. These equations could be incorporated into weighing systems to quickly supply farmers accurate, without delay, liveweight measurements. Future studies should explore how to understand location-related variability in liveweight loss observed in the current study. Further, the extent to which the liveweight correcting equations can be generalized to ewe lambs from different locations and breed is warranted.

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References

1. Brown, D.J.; Ball, A.J.; Huisman, A.E. The influence of ewe weight at mating on lamb performance and reproduction of the ewe. *Proc. Assoc. Adv. Anim. Breed. Genet.* **2005**, *16*, 306–309.
2. Wishart, H.; Morgan-Davies, C.; Stott, A.; Wilson, R.; Waterhouse, T. Liveweight loss associated with handling and weighing of grazing sheep. *Small Rumin. Res.* **2017**, *153*, 163–170. [CrossRef]

3. Brown, D.J.; Savage, D.B.; Hinch, G.N.; Hatcher, S. Monitoring liveweight in sheep is a valuable management strategy: A review of available technologies. *Anim. Prod. Sci.* **2015**, *55*, 427–436. [CrossRef]
4. Coates, D.B.; Penning, P. Measuring animal performance. In *Field and Laboratory Methods for Grassland and Animal Production Research*; Mannetje, L., Jones, R.M., Eds.; CABI Publishing: Wallingford, UK, 2000; pp. 353–402.
5. Kenyon, P.R.; Maloney, S.K.; Blache, D. Review of sheep body condition score in relation to production characteristics. *N. Z. J. Agric. Res.* **2014**, *57*, 38–64. [CrossRef]
6. Hughes, J.G. Short-term variation in animal live weight and reduction of its effect on weighing. *Anim. Breed. Abstr.* **1976**, *44*, 1976.
7. Moyo, M.; Nsahlai, I. Rate of Passage of Digesta in Ruminants; Are Goats Different? In *Goat Science*; Kukovics, S., Ed.; IntechOpen: London, UK, 2018; pp. 39–74. [CrossRef]
8. Cranston, L.; Ridler, A. Pasture measurement. In *Livestock Production in New Zealand*; Stafford, K.J., Ed.; Massey University Press: Auckland, New Zealand, 2017; pp. 88–96.
9. Burnham, D.L.; Morel, P.C.H.; Kenyon, P.R.; Morris, S.T.; Stafford, K.J. The effect of fasting on live weight in hogget wethers and pregnant mature ewes: Brief Communication. *Proc. N. Z. Soc. Anim. Prod.* **2009**, *69*, 112–114.
10. Wilson, R.F.; Hardwick-Smith, J.A.; Logan, C.M.; Corner-Thomas, R.A.; Bywater, A.C.; Greer, A.W. Brief communication: Optimizing a fasting protocol to assess live weight of sheep. *Proc. N. Z. Soc. Anim. Prod.* **2015**, *75*, 223–224.
11. Hogan, J.P.; Petherick, J.C.; Phillips, C.J. The physiological and metabolic impacts on sheep and cattle of feed and water deprivation before and during transport. *Nutr. Res. Rev.* **2007**, *20*, 17–28. [CrossRef] [PubMed]
12. Semakula, J.; Corner-Thomas, R.; Morris, S.; Blair, H.; Kenyon, P. The effect of herbage type prior to fasting on the rate of liveweight loss during fasting in ewe lambs. *Proc. N. Z. Soc. Anim. Prod.* **2019**, *79*, 131–134.
13. Kirton, A.H.; Quartermain, A.R.; Uljee, A.E.; Carter, W.A.; Pickering, F.S. Effect of 1- and 2-days' ante-mortem fasting on live weight and carcass losses in lambs. *N. Z. J. Agric. Res.* **1968**, *11*, 891–902. [CrossRef]
14. Thompson, J.; O'Halloran, W.; McNeill, D.; Jackson-Hope, N.; May, T. The effect of fasting on liveweight and carcass characteristics in lambs. *Meat Sci.* **1987**, *20*, 293–309. [CrossRef]
15. Kirton, A.; Moss, R.; Taylor, A.G. Weight losses from milk and weaned lambs in mid Canterbury resulting from different lengths of starvation before slaughter. *N. Z. J. Agric. Res.* **1971**, *14*, 149–160. [CrossRef]
16. Haaland, G.L.; Tyrrell, H.F. Effects of Limestone and Sodium Bicarbonate Buffers on Rumen Measurements and Rate of Passage in Cattle. *J. Anim. Sci.* **1982**, *55*, 935–942. [CrossRef]
17. Varga, G.A.; Prigge, E.C. Influence of forage Species and Level of Intake on Ruminal Turnover Rates. *J. Anim. Sci.* **1982**, *55*, 1498–1504. [CrossRef]
18. Alwash, A.H.; Thomas, P.C. The effect of the physical form of the diet and the level of feeding on the digestion of dried grass by sheep. *J. Sci. Food Agric.* **1971**, *22*, 611–615. [CrossRef] [PubMed]
19. Kaske, M.; Groth, A. Changes in factors affecting the rate of digesta passage during pregnancy and lactation in sheep fed on hay. *Reprod. Nutr. Dev.* **1997**, *37*, 573–588. [CrossRef] [PubMed]
20. Nicol, A.; Brookes, I. The metabolisable energy requirements of grazing livestock. In *Pasture and Supplements for Grazing Animals*; Rattray, P.V., Brookes, I.M., Nicol, A.M., Eds.; New Zealand Society of Animal Production: Hamilton, New Zealand, 2007; pp. 151–172.
21. Penning, P.; Hooper, G. An evaluation of the use of short-term weight changes in grazing sheep for estimating herbage intake. *Grass Forage Sci.* **1985**, *40*, 79–84. [CrossRef]
22. Hodgson, J.; Matthews, P.N.P.; Matthews, C.; Lucas, R.J. Pasture measurement. In *New Zealand Pasture and Crop Science*; White, J., Hodgson, J., Eds.; Oxford University Press: Oxford, UK, 1999; pp. 56–65.
23. Khodabux, K.; L'Omelette, M.S.S.; Jhaumeer-Laulloo, S.; Ramasami, P.; Rondeau, P. Chemical and near-infrared determination of moisture, fat and protein in tuna fishes. *Food Chem.* **2007**, *102*, 669–675. [CrossRef]
24. Haese, E.; Krieg, J.; Grubješić, G.; Feyder, A.; Rodehutscord, M. Determination of in situ ruminal degradation of phytate phosphorus from single and compound feeds in dairy cows using chemical analysis and near-infrared spectroscopy. *Animal* **2020**, *14*, 1461–1471. [CrossRef] [PubMed]
25. Corson, D.; Waghorn, G.; Ulyatt, M.; Lee, J. NIRS: Forage analysis and livestock feeding. *Proc. N. Z. Grassl. Assoc.* **1999**, *61*, 127–132. [CrossRef]
26. Dowman, M.G.; Collins, F.C. The use of enzymes to predict the digestibility of animal feeds. *J. Sci. Food Agric.* **1982**, *33*, 689–696. [CrossRef]
27. Roughan, P.G.; Holland, R. Predicting in-vivo digestibilities of herbages by exhaustive enzymic hydrolysis of cell walls. *J. Sci. Food Agric.* **1977**, *28*, 1057–1064. [CrossRef]
28. R Core Team. *R: A Language and Environment for Statistical Computing*; R Version 3.4.4 (2018-03-15); R Foundation for Statistical Computing: Vienna, Austria, 2016.
29. Dhakal, C.P. Multiple Regression Model Fitted for Rice Production Forecasting in Nepal: A Case of Time Series Data. *Nepal. J. Stat.* **2018**, *2*, 89–98. [CrossRef]
30. Peat, J.; Barton, B. *Medical Statistics: A Guide to Data Analysis and Critical Appraisal*; John Wiley & Sons: Hoboken, NJ, USA, 2008.
31. Shapiro, S.S.; Wilk, M.B. An analysis of variance test for normality (complete samples). *Biometrika* **1965**, *52*, 591–611. [CrossRef]
32. Breusch, T.S.; Pagan, A.R. A Simple Test for Heteroscedasticity and Random Coefficient Variation. *Econometrica* **1979**, *47*, 1287–1294. [CrossRef]

33. Cook, R.D. Detection of Influential Observation in Linear Regression. *Technometrics* **1977**, *19*, 15–18. [CrossRef]
34. Pinheiro, J.; Bates, D.; DebRoy, S.; Sarkar, D.; R Core Team. *nlme: Linear and Nonlinear Mixed Effects Models*; R package Version 31-137; R Foundation for Statistical Computing: Vienna, Austria, 2018. Available online: https://cran.r-project.org/web/packages/nlme (accessed on 20 June 2020).
35. McDowell, M.L.; Bruland, G.L.; Deenik, J.L.; Grunwald, S. Effects of subsetting by carbon content, soil order, and spectral classification on prediction of soil total carbon with diffuse reflectance spectroscopy. *Appl. Environ. Soil Sci.* **2012**, *2012*, 294121. [CrossRef]
36. Bellon-Maurel, V.; Fernandez-Ahumada, E.; Palagos, B.; Roger, J.-M.; McBratney, A. Critical review of chemometric indicators commonly used for assessing the quality of the prediction of soil attributes by NIR spectroscopy. TrAC. *Trends Anal. Chem.* **2010**, *29*, 1073–1081. [CrossRef]
37. Moriasi, D.N.; Arnold, J.G.; Van Liew, M.W.; Bingner, R.L.; Harmel, R.D.; Veith, T.L. Model evaluation guidelines for systematic quantification of accuracy in watershed simulations. *Trans. ASABE* **2007**, *50*, 885–900. [CrossRef]
38. Botchkarev, A. Performance Metrics (Error Measures) in Machine Learning Regression, Forecasting and Prognostics: Properties and Typology. *arXiv* **2019**, arXiv:1809.03006.
39. Kodaira, M.; Shibusawa, S. Using a mobile real-time soil visible-near infrared sensor for high resolution soil property mapping. *Geoderma* **2013**, *199*, 64–79. [CrossRef]
40. Nawar, S.; Mouazen, A.M. Predictive performance of mobile vis-near infrared spectroscopy for key soil properties at different geographical scales by using spiking and data mining techniques. *Catena* **2017**, *151*, 118–129. [CrossRef]
41. Efron, B. Estimation and Accuracy after Model Selection. *J. Am. Stat. Assoc.* **2014**, *109*, 991–1007. [CrossRef]
42. Pino, F.; Mitchell, L.K.; Jones, C.M.; Heinrichs, A.J. Comparison of diet digestibility, rumen fermentation, rumen rate of passage, and feed efficiency in dairy heifers fed ad-libitum versus precision diets with low- and high-quality forages. *J. Appl. Anim. Res.* **2018**, *46*, 1296–1306. [CrossRef]
43. Crampton, E.W.; Jackson, I.R.C. Seasonal Variation in Chemical Composition of Pasture Herbage and the Relation to Its Digestibility by Steers and Sheep. (Pasture Studies—XXVI). *J. Anim. Sci.* **1944**, *3*, 333–339. [CrossRef]
44. Särkijärvi, S.; Niemeläinen, O.; Sormunen-Cristian, R.; Saastamoinen, M. Changes in chemical composition of different grass species and -mixtures in equine pasture during grazing season. In *Forages and Grazing in Horse Nutrition*; Saastamoinen, M., Fradinho, M.J., Santos, A.S., Miraglia, N., Eds.; Wageningen Academic Publishers: Wageningen, The Netherlands, 2012; pp. 45–48.
45. Warly, L.; Fariani, A.; Ichinohe, T.; Fujihara, T. Seasonal changes in nutritive value of some grass species in West Sumatra, Indonesia. *Asian Australas. J. Anim. Sci.* **2004**, *17*, 1663–1668. [CrossRef]
46. Mir, S.; Ahmed, H. Effect of Season on the Chemical Composition and Nutritive Value of Shrub Foliage (*Cotoneaster* spp.) of Sub-Alpine Pasture of Kashmir Valley. *Int. J. Livest. Res.* **2017**, *7*, 289–294. [CrossRef]
47. Litherland, A.J.; Woodward, S.J.R.; Stevens, D.R.; McDougal, D.B.; Boom, C.J.; Knight, T.L.; Lambert, M.G. Seasonal variations in pasture quality in New Zealand sheep and beef farms. *Proc. N. Z. Soc. Anim. Prod.* **2002**, *62*, 138–142.
48. Ekanayake, W.; Corner-Thomas, R.; Cranston, L.; Hickson, R.; Kenyon, P.; Morris, S. Characterisation of the nutritional composition of plant components of a herb-clover mix during November to May in New Zealand. *Proc. N. Z. Soc. Anim. Prod.* **2019**, *79*, 162–167.
49. Bernabucci, U.; Bani, P.; Ronchi, B.; Lacetera, N.; Nardone, A. Influence of Short- and Long-Term Exposure to a Hot Environment on Rumen Passage Rate and Diet Digestibility by Friesian Heifers. *J. Dairy Sci.* **1999**, *82*, 967–973. [CrossRef]
50. Kennedy, P.M. Influences of cold exposure on digestion of organic matter, rates of passage of digesta in the gastrointestinal tract, and feeding and rumination behaviour in sheep given four forage diets in the chopped, or ground and pelleted form. *Br. J. Nutr.* **1985**, *53*, 159–173. [CrossRef] [PubMed]
51. Morris, S.; Kenyon, P. The effect of litter size and sward height on ewe and lamb performance. *N. Z. J. Agric. Res.* **2004**, *47*, 275–286. [CrossRef]