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Estimating winter energy balance and actual changes in nutrition of Mongolian grazing sheep

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ABSTRACT

Objective: We aimed to estimate the energy intake and expenditure of grazing Mongolian sheep during the winter months using indirect methods of bite mass, number of bites, ME content per bite, traveling distance, body condition, and blood parameters.

Materials and Methods: Energy intake was calculated as the bite mass × number of bites × ME per bite. Daily energy expenditure was calculated as the sum of the expenditures incurred for maintenance, grazing, and thermoregulating, as well as during pregnancy. We also investigated the sheep nutrition indicators (body condition and blood biochemical parameters) before and after winter.

Results and Discussion: Average daily bite number, bite weight, and daily feed ME of sheep were 12,094, 53 mg in March, and 1.16 Mcal, respectively, and thus, total daily ME of intake was 0.86 Mcal. The daily energy expenditure for maintenance, grazing, thermoregulation, and fetus were 1.58, 1.56, 2.27, and 0.41 Mcal, respectively, accounting for a total daily energy expenditure per sheep of 5.82 Mcal. The total protein (70.2 to 49.1 g/L), albumin (37.4 to 16.9 g/L), and leptin contents (37.1 to 10.9 ng/mL) in the blood decreased remarkably after winter.

Implications and Applications: The energy intake and expenditure results showed that the estimated total negative daily energy balance was 4.96 Mcal/d during the winter season. These results indicated that Mongolian sheep did not maintain a sufficient energy balance during winter and, thereby, required compensation for the negative energy balance using their body fat and protein stores.

Key words: number of bites, bite size, dryland

The authors declare no conflicts of interest.

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INTRODUCTION

Recently, the total number of livestock (sheep, goat, cattle, horse, camel) involved in Mongolian nomadism has reached over 60 million (National Statistical Office of Mongolia, 2003). Traditionally, Mongolian livestock grazes only on natural rangelands year-round and has been adapted to severe winter conditions. However, Mongolia has experienced significant declines in livestock numbers, with mortality rates reaching 23% in 2010 because of "dzud" (Nandintsetseg, et al., 2018). Dzud is a term used to describe extreme Mongolian winter conditions (i.e., snow and ice cover and lack of rangeland) leading to increased livestock mortality rates during the winter–spring season, mainly due to starvation. Physiological features of Mongolian livestock populations include the ability to overcome severe climatic conditions without any additional feeds and care. Additionally, high livestock mortality is caused by the combined effects of severe winters and subsequent feed restriction. To alleviate the mortality risk of livestock in dzud events, it is important to consider meteorological perspectives in predicting the occurrence of dzud and animal energy and nutrition. However, because of the difficulty in estimating the energy balance of free-grazing animals, such studies remain limited.

The mechanisms involving livestock mortality by dzud events are still unclear, but the energy intake of large grazing herbivores may decrease in winter due to other factors, such as (1) limited access to vegetation due to snow accumulation during winter, which forces animals to wade and paw through snow to consume underlying vegetation, thus increasing the amount of time required to encounter a food item (Robinson and Merrill, 2012), and (2) herbage being scarce and of lower quality in winter (Goodson et al., 1991; Liu et al., 2019). In addition, Mongolian plants may be completely covered with snow or ice in winter, compelling animals to move long distances to find edible forage, thus expending more energy (Dailey and Hobbs 1989; Yoshihara et al., 2009). This situation leads to a negative energy balance. If this negative energy balance is prolonged, animals may lose their entire body energy stores (e.g., fat and protein), leading to death. The effects of dzud on large herbivores may differ with regard to the intake and expenditure of energy. Therefore, it is important to evaluate these factors on grazing livestock to understand the mortality effects of the dzud events.

We aimed to estimate the dynamic changes in the energy intake and expenditure of Mongolian grazing sheep during winter. We also explored the sheep body condition and blood parameters before and after winter to validate the estimated energy balance model in winter.

MATERIALS AND METHODS

All animal procedures were approved by the Mie University Institutional Animal Care and Use Committee (No. 194), Japan. The study was conducted in Bayantsogt soum, Töv Province, Mongolia (48°07′N, 51°17′E), in the steppe ecological zone. The study site is located on a flat plateau approximately 1,000 m above sea level. Vegetation in the area is characterized by a mixture of grasses (e.g., *Stipa krylovii*) and forbs. The area has a long history (centuries) of grazing by domestic livestock under moderate grazing pressure. We studied the free-grazing sheep of a traditional Mongolian nomad family and the grassland in the vicinity of the soum.

We obtained winter climatic information from the nearest monitoring station (Meteorology and Hydrology of Mongolia; 32.7-km distance from the study site). Data from 2016 to 2017 from the Meteorology and Hydrology of Mongolia station reported that the mean temperature and precipitation were −14.5°C and 0.45 mm in November, −19.1°C and 0.03 mm in December, −24.0°C and 0.1 mm in January, and −16.0°C and 0.08 mm in February, respectively. Mean snow depth was 9.0 cm in November, 8.6 cm in December, 10.4 cm in January, and 11.3 cm in February.

More than 90% of the total Mongolian sheep are the "Mongol" sheep breed and are spread evenly across all regions. In 2016, 35 ewes in the first month of pregnancy, with a mean age of 4.46 yr ranging from 2 to 5 yr and BW of 44.6 kg ± 1.01 (SE), were randomly selected from the herd and used throughout this study. The ewes had unlimited access to natural rangeland from the morning (between 0700 to 0900 h) to evening (between 1600 to 1800 h) based on sunrise–sunset and were kept in a roofed shelter at night without any feed or supplement.

Estimating Energy Intake of Sheep

Estimated metabolizable intake was established by determining bite mass (mg of DM) × number of bites × ME content (Mcal/mg of DM). A neckband bite counter was attached to a collar on 3 middle-sized ewes during winter (from November 21 to the February 28) to count the jaw movements per day. The true number of bites (TNB) was predicted by using a correction equation (Kawamura et al., 2006):

$$\text{TNB} = 1.234 \times \text{JMT} + 2.524,$$

where JMT = jaw movement estimated by the bite counter.

The bite mass (**BM**) of grazing sheep was estimated by sward surface height (**SSH**) of forage plants (Edwards et al., 1995). We measured the SSH of the vegetation in the 14 randomly located 100-cm quadrates within the grazing area in winter (from the end of November to the beginning of March). The plants were clipped to the ground level in each species (Barthram et al., 2000). The clipped samples were dried in a forced-air oven at 70°C for 48 h to determine the dry weight. For the grasses, BM (g/bite) = −45.2 + 17.2 × SSH (cm), and for the forbs, BM = −33.4 + 17.2 × SSH. The average bite mass in this site was calculated by the estimated BM and the relative biomass weight of each plant species in the plots assuming that the sheep eat in proportion to the relative biomass of each species (Table 1).

Before the chemical analysis of plant samples, they were ground to pass through a 1-mm screen. The samples were analyzed for IVDMD by the pepsin–cellulase assay (Goto and Minson, 1977). The CP, neutral detergent insoluble CP (**NDICP**), and acid detergent insoluble CP (**ADICP**) contents were measured by determining N, using the combustion method (Elementar). A factor of 6.25 was used for the conversion of N into CP (AOAC, 1990). The ether extracts and fibers were determined using the diethyl ether and detergent methods. The ash was determined by incineration at 600°C for 3 h.

The content of the TDN was calculated using the equations following NASEM (2001):

$$\text{TDN} = \text{tdNFC} + \text{tdCP} + (\text{tdFA} \times 2.25) + \text{tdNDF} - 7,$$

$$\text{tdNFC} = 0.98\{100 - [(\text{NDF} - \text{NDICP}) + \text{CP} + \text{EE} + \text{Ash}]\},$$

$$\text{tdCP} = \text{CP} \times \exp[-1.2 \times (\text{ADICP}/\text{CP})],$$

$$\text{tdFA} = \text{EE} - 1.0,$$

$$\text{tdNDF} = 0.75[(\text{NDF} - \text{NDICP}) - \text{ADL}] \times \{1 - [\text{ADL}/(\text{NDF} - \text{NDICP})]^{0.667}\},$$

where tdNFC represents digestible nonfiber carbohydrates; EE represents ether extracts; tdCP represents truly digestible CP; tdFA represents truly digestible fatty acid; and tdNDF represents truly digestible NDF.

The content of daily DE and ME for each plant species found in the plots (*Stipa krylovii*, *Carex* spp., *Caragana leucophylla*, *Artemisia frigida*, *Artemisia adamsii*, *Poten-*

Table 1. Plant biomass, sward surface height (SSH), and estimated bite mass of the main species in winter

Item	*Stipa krylovii* (grass)	*Carex* spp. (grass)	*Caragana leucophylla* (shrub)	*Artemisia frigida* (forb)	*Artemisia adamsii* (forb)	*Potentilla acaulis* (forb)
Average biomass in the beginning and end of winter (g/m²)	10.10	1.64	1.29	0.50	1.36	0.57
Average SSH in the beginning of winter (cm)	7.16	6.18	5.64	5.68	6.77	2.33
Sampling size	(280)	(227)	(127)	(100)	(62)	(92)
Bite mass[1] in the beginning of winter (mg)	78.0	61.2	63.6	64.3	83.0	6.7
Average SSH in the end of winter (cm)	6.56	4.97	4.43	6.01	5.52	1.72
Sampling size	(100)	(98)	(21)	(21)	(63)	(9)
Bite mass in the end of winter (mg)	67.7	40.3	42.8	70.0	61.5	0.0

[1]Bite mass of grasses (g/bites) = −45.2 + 17.2 × SSH (cm). Bite mass of forbs = −33.4 + 17.2 × SSH.

tilla acaulis) were calculated using the following equations (NASEM, 1996):

$$\text{DE (Mcal/kg)} = 4.41 \times \text{TDN},$$

$$\text{ME (Mcal/kg)} = 0.82 \times \text{DE}.$$

Finally, we weighed and calculated the daily energy intake of sheep as Σ(Relative biomass of each species × daily intake of each species × ME of each species) according to their individual ME quality and biomass in the area.

In Mongolia, plant biomass is low, and most of the plants are completely covered with snow. Therefore, animals have been forced to eat edible forage nonselectively, based on our previous direct animal observation of foraging behavior (Yoshihara et al., 2009). Therefore, we calculated the relative intake of each plant species in proportion to their relative biomass.

Estimating Energy Expenditure of Sheep

Daily energy expenditure (E_E) of sheep was calculated as the sum of the expenditures incurred for maintenance (E_M), grazing (E_G), and thermoregulation (E_T), following (Tachiiri et al., 2017). The fetal energy requirement (E_F) was also included.

$$E_E = E_M + E_G + E_T + E_F$$

Metabolizable energy for maintenance was estimated using the DM digestibility (**DMD**), sheep monthly years (**T**), and average BW in November 2017 as follows:

$$E_M = [0.26 \times BW^{0.75}\exp(-0.03T/12)]/$$
$$\{[0.02(0.156 \times 100 \times DMD - 0.535)] + 0.5\}.$$

Based on previous studies (Lachica and Aguilera, 2005; Tachiiri et al., 2017), the daily energy for grazing (E_G) of sheep was estimated using the intake ME (E_I), DMD, traveling distance (D), slope angle of traveling path (θ), and BW as follows:

$$E_G = [0.05 \times E_I (0.9 - DMD) + (6.969 + 3.980 \times \theta)/$$
$$10^6 \times BW \times D]/[0.02 \times (0.156 \times 100$$
$$\times DMD - 0.535) + 0.5].$$

The traveling distance and slope angle were calculated by waypoints at 30-min intervals using Google Earth Pro (Google Inc.). We attached global positioning systems (VECTRONIC Aerospace) around the necks of the sheep for 20 d in winter (from November to December) to obtain waypoints. The energy required for thermoregulation and fetus were estimated using the Standing Committee on Agriculture (1990) guidelines. We then calculated the energy balance using the estimated intake and expenditure of energy during winter.

Sheep Nutrition Conditions

The chest circumference and BW of the 35 sheep were measured to check for body nutritional condition in November and the following March. We collected blood samples in evacuated EDTA tubes in the morning before the grazing. We measured blood albumin and total protein for the nutrition status in sheep (Ndlovu et al., 2007). We used leptin as a marker for the body fat condition because circulating leptin content is strongly related to body lipid content in sheep (Delavaud et al., 2007). Serum was separated from the blood samples, and the albumin, total protein, and leptin contents were measured with commercially available kits or an ELISA kit following the manufacturer's instructions (Cusabio Biotech Co.).

We compared the chest circumference, BW, total protein, albumin, and leptin contents in the blood of the sheep before and after winter using paired *t*-tests. All differences between comparisons with P-values < 0.05 were considered statistically significant.

Table 2. Nutritional composition of the main plant species collected in winter measured from our chemical experiment

Item[1]	*Stipa krylovii* (grass)	*Carex* spp. (grass)	*Caragana leucophylla* (shrub)	*Artemisia frigida* (forb)	*Artemisia adamsii* (forb)	*Potentilla acaulis* (forb)	Average
CP (%)	4.66	6.22	10.80	7.25	8.78	8.88	7.80
NDICP (%)	2.88	3.72	4.69	3.47	3.69	5.59	4.00
ADICP (%)	3.59	4.72	5.16	5.06	5.38	6.25	5.00
NDF (%)	69.8	64.0	64.0	54.8	44.2	38.0	55.8
ADL (%)	45.7	46.0	45.2	42.4	35.6	30.2	40.8
Ether extracts (%)	7.71	3.12	6.77	2.79	8.89	10.80	6.70
Ash (%)	5.90	3.95	3.10	3.30	1.40	1.70	3.20
tdNFC (%)	14.5	25.9	19.6	34.6	39.6	45.3	29.9
tdCP (%)	1.84	2.50	6.05	3.14	4.21	3.81	3.60
tdFA (%)	6.71	2.12	5.77	1.79	7.89	9.76	5.70
tdNDF (%)	3.58	1.77	1.75	0.80	0.31	0.08	1.40
TDN (%)	28.1	27.9	33.4	35.6	54.9	64.2	40.7
DE (Mcal/kg of DM)	1.24	1.23	1.47	1.57	2.42	2.83	1.80
ME (Mcal/kg of DM)	1.01	1.01	1.21	1.29	1.98	2.32	1.50

[1]NDICP = neutral detergent insoluble CP; ADICP = acid detergent insoluble CP; td = truly digestible; tdNFC = truly digestible nonfiber carbohydrates; tdFA = truly digestible fatty acid.

RESULTS AND DISCUSSION

The average SSH of each species in November and March were 5.6 and 4.9 cm, respectively (Table 1). The estimated average BM of each species decreased from 59.5 mg in November to 47 mg in March.

The average daily number of bites was 12,094 ± 4,422 SD among the 3 sheep and ranged from 4,296 to 23,802. The average daily number of bites increased from 10,653 in November and December to 13,718 in January and February ± 3,853 SD among the days.

The dominant grass type in Mongolian grasslands (*S. krylovii*) was found to be a low-quality source of nutrition in winter due to low contents of CP and TDN (Table 2). Forbs were of relatively high quality owing to greater CP and lower fibers and lignin. However, the forbs were less abundant, and thus, the relative species quantity-adjusted average plant ME was 1.16 Mcal/kg.

The DMD of the collected samples was 34.3%. The average daily traveling distance and slope angle of the traveling path for the sheep were 8,035 m (±6.52 SD) and 3.33° (±6.52 SD), respectively. The estimated daily energy expenditure for maintenance, grazing, thermoregulation, and fetus were 1.58, 1.56, 2.27, and 0.41 Mcal, respectively. The estimated total energy intake and expenditure are summarized in Table 3.

Chest circumference (82.2 to 82.2 cm, *P*-value = 0.998, t-statistic = 0.003, df = 34) and BW (44.6 to 43.0 kg, *P*-value = 0.349, t-statistic = 0.945, df = 34) of the sheep remained unchanged after winter (Figure 1). However, the total protein (70.2 to 49.1 g/L, t-statistic = 5.387), albumin (37.4 to 16.9 g/L, t-statistic = 9.573), and leptin contents (37.1 to 10.9 ng/mL, t-statistic = 6.743) in blood decreased during the study period (*P*-value <0.001, df = 34).

The estimated daily total energy of intake per sheep (0.86 Mcal) in the present study was less than the estimated energy of Mongolian ewes found by Tachiiri et al. (2017; 5.00 Mcal/d). This previous study estimated the intake energy using sheep BW and plant biomass; however, the intake amount of sheep and plant ME were not considered. This may account for the differences in results obtained in both studies. In addition, our estimated BM from the SSH is questionable because the pasture species used by Edwards et al. (1995) were ryegrass and white

Table 3. Summarized daily energy intake and energy expenditure of sheep

Item	Value
Energy intake	
Total bites per day (no.)	12,094
Bite size (mg)	61
Herbage metabolic energy (Mcal/kg)	1.16
Total energy intake (Mcal)	0.86
Energy expenditure (Mcal)	
Energy expenditure by maintenance	1.58
Energy expenditure by grazing	1.56
Energy required for thermoregulation	2.27
Energy required for fetus	0.41
Total energy expenditure	5.82

clover, which have different bulk densities, SSH, and morphologies than the species present in the Mongolian steppe rangelands. We estimated daily ME of expenditure (5.82 Mcal), which was comparable to the estimated metabolic energy (3.98–6.37 Mcal) by Tachiiri et al. (2017).

We calculated a daily negative balance of 4.96 Mcal from the energy intake and expenditure, which was lower than the estimated energy retention of grazing Tibetan yak (1.39 Mcal/d); however, the animal species and estimated methods were different (Ding et al., 2014). The negative energy balance of Mongolian sheep estimated here was attributed to the small number of biting times, BM, poor forage nutrition, and the increase in moving distance during winter. Robinson and Merrill (2012) reported that the bite rate of grazing ungulates is encounter limited and decreases when encounter rates are low due to plants being covered by snow. According to the nearest meteorological weather station to the study area, the snow depth during winter in the assessed year was 10 cm, such that snow accumulation would have completely covered most of the plants, constraining the foraging behavior of the sheep (Yoshihara et al., 2009). Indeed, the total number of bites of Mongolian sheep per day in winter was less than half of that in summer (Y. Yoshihara and B. Choijisuren, unpublished data). Moreover, the shortened Mongolian winter grass led to a smaller bite size of sheep because the plant height of *S. krylovii* in autumn was higher than 30 cm. In our previous study, Mongolian plant CP and NDF contents collected in summer were on average 16.3% and 37.7%, respectively (Yoshihara et al., 2019). The CP content in Mongolian winter plants was found to be less than half that of plants collected in summer, presumably due to winter withering. The mean traveling distance of sheep in winter was found to be approximately twice that in summer (Yoshihara et al., 2009).

The total protein and albumin content in the blood decreased by approximately 30 and 50% during winter, respectively, both of which were under the normal range (Plumb, 2018). Moreover, the Mongolian sheep lost ~70% leptin during winter, indicating that the nutrition condition of sheep deteriorated because they were required to compensate for the negative energy balance by using their fat and protein stores. Sheep fed approximately 32% restricted ME diet for 180 d showed a 10% decrease in the total protein content in the blood, indicating the severity of the Mongolian winter season for livestock (Song et al., 2018). The actual animal nutrient conditions were synchronized with our estimated energy balance, which further validated the estimated energy intake and expenditure model for grazing sheep in winter. However, we found

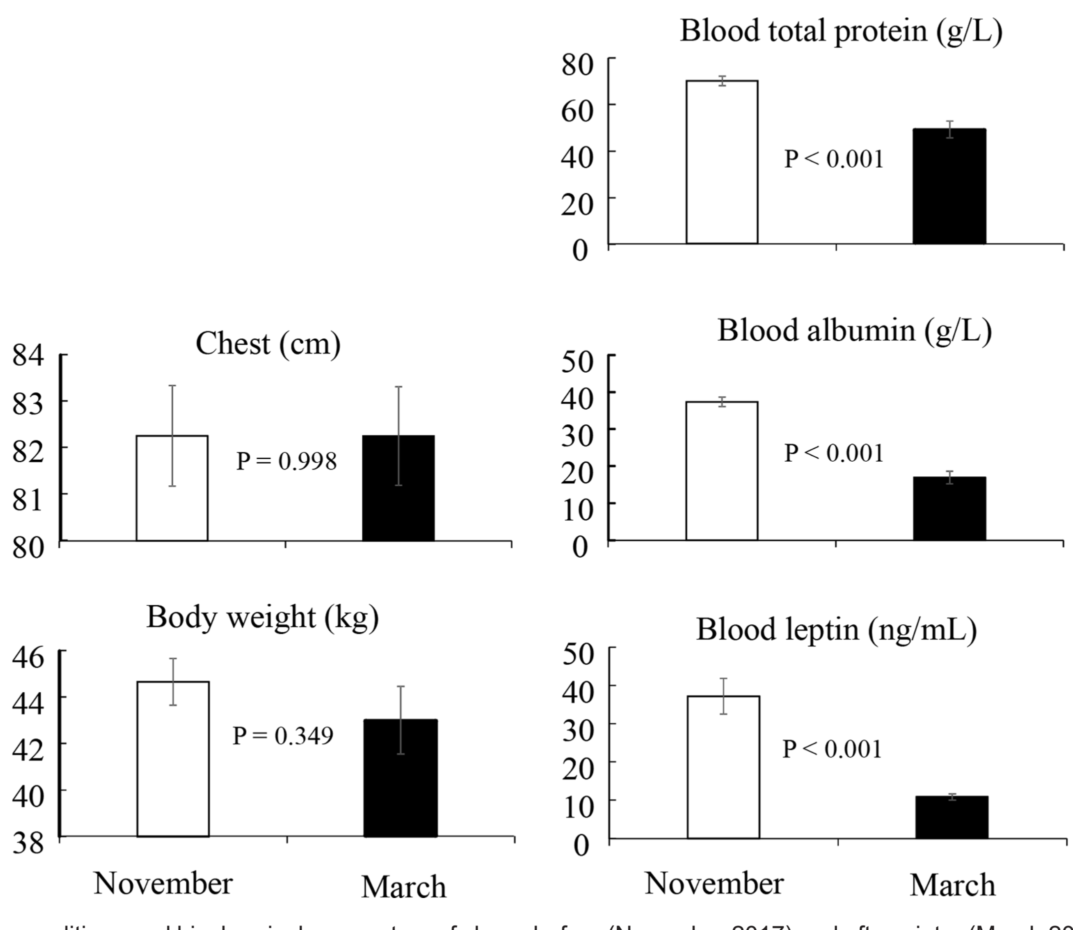

Figure 1. Body conditions and biochemical parameters of sheep before (November 2017) and after winter (March 2018). Error bars indicate SD.

no significant change in body size and weight during the study. One of the reasons for the better-than-expected body condition is that Mongolian sheep compensate for lost body size and weight with thicker fur to combat winter stress (Batsukh and Zagdsuren, 1991).

APPLICATIONS

According to our energy balance results, Mongolian nomads need to supply an additional 4.96 Mcal/d of forage to maintain the body condition of sheep during the winter season. These findings implied that if the nomads supply the feed with the same nutritional value as the plants, a daily supply of 4,276 g (4.96/1.16 Mcal) of field grass should be offered to each sheep. Although the specific value could be only applicable in this study as a starting point in calculating supplemental feeding strategies, the specific energy estimation values should be helpful to Mongolian nomads to alleviate the mortality risk of livestock, because the amount and timing of supplementary feed (mowed grass) is traditionally based only on experience and not on scientific findings.

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