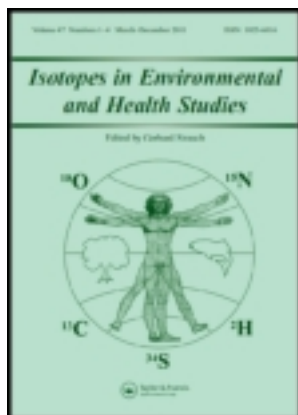


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### Food webs in Mongolian grasslands: The analysis of <sup>13</sup>C and <sup>15</sup>N natural abundances

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## Food webs in Mongolian grasslands: The analysis of $^{13}\text{C}$ and $^{15}\text{N}$ natural abundances

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Overgrazing often lowers species richness and productivity of grassland communities. For Mongolian grassland ecosystems, a lack of detailed information about food-web structures makes it difficult to predict the effects of overgrazing on species diversity and community composition. We analysed the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures of herbaceous plants, arthropods (grouped by feeding habit), wild and domestic mammals, and humans in central Mongolia to understand the predominant food-web pathways in this grassland ecosystem. The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of mammals showed little variation within species, but varied considerably with slope position for arthropods. The apparent isotopic discrimination between body tissue and hair of mammals was estimated as 2.0‰ for  $\delta^{13}\text{C}$  and 2.1‰ for  $\delta^{15}\text{N}$ , which was large enough to cause overestimation of the trophic level of mammals if not taken into account when using hair samples to measure isotopic enrichment.

**Keywords:** carbon-13; food selectivity; grazing food chain; herbaceous plants; isotope ecology; nitrogen-15; spatial scale; trophic enrichment

### 1. Introduction

Grasslands occupy more than 30% of the land surface of the Earth and are the representative landscape of the semi-arid region of the Eurasian continent [1]. The primary productivity of grassland communities is mainly provided by perennial herbaceous plants, which support organisms at higher trophic levels, including arthropods, wild and domestic mammals, and humans. On the Mongolian steppes, grassland ecosystems have increasingly been degraded by human activities such as agriculture and nomadism as well as by global warming [2,3]. Although traditional nomadic grazing on the Mongolian steppes has continued for centuries, government-driven land privatisation has led to enhanced grazing pressure from livestock around large cities such as

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Ulaanbaatar [4,5]. Overgrazing often lowers the species richness and productivity of grassland communities, thereby allowing grazing-tolerant (less edible or unpalatable) plant species to increase in abundance and distribution [6]. Such changes in grassland community composition might decrease the efficiency of energy transfer between trophic levels, and this might propagate through the food chain. However, the lack of detailed information about grassland food-web structures makes it difficult to predict the effects of overgrazing on species diversity and community composition of Mongolian grassland ecosystems.

Stable carbon and nitrogen isotope signatures ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) of the organisms provide useful information about the trophic relationships of ecosystems [7–9]. Enrichment in  $\delta^{13}\text{C}$  of 0–1‰ and enrichment in  $\delta^{15}\text{N}$  of  $\sim 3.3$ ‰ have been widely reported in association with trophic transfer of organic carbon and nitrogen [10,11]. The base of food webs and trophic positions of organisms both in terrestrial and aquatic ecosystems can be identified using these empirical relationships of trophic fractionation in  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values [11,12]. Moreover, if spatial variation in isotopic values of basal resources (primary producers and detritus) exists, the spatial extent of an energy channel from a trophic base to consumers can also be deduced [13,14]. However, few studies have examined the trophic fractionation of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values in diverse invertebrates and vertebrates in terrestrial grassland ecosystems [11]. Furthermore, although stable isotope analyses have been applied to grassland communities [15], most of the previous studies have focused only on a specific trophic group or species to analyse their diets [16,17]. To our knowledge, there are few empirical data that describe the comprehensive food webs in grassland ecosystems [18]. We analysed the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures of herbaceous plants, arthropods with various feeding habits, wild and domestic mammals, and humans in central Mongolia to understand the predominant food-web pathways in this grassland ecosystem.

## 2. Materials and methods

### 2.1. Site description

Field sampling was conducted during summer in 1999 and 2000 at two field sites in central Mongolia: the Gachuurt site (48° 01'N, 107° 11'E; 30 km northeast from Ulaanbaatar) and the Khangai site (47° 31'N, 100° 56'E; 430 km southwest from Ulaanbaatar) (Figure 1a). The Gachuurt site is located on the southwestern edge of the Khentii mountains where the mean annual temperature is about 0 °C (range = –25 °C (January) to 15 °C (July)) and the annual precipitation is very low (200–300 mm). Most precipitation falls during spring and autumn. Grasslands cover the southern slope of the Gachuurt site, whereas the northern slope is covered with natural larch forests (Figure 1b). Sampling was conducted on a southeastern slope that was covered with herbaceous plant species (mean slope = 29 %). The Khangai site is located on the northeastern edge of the Khangai mountains where the mean annual temperature is about 0 °C (range = –25 °C (January) to 13 °C (July)). The annual precipitation is 300–400 mm, and most precipitation falls during spring and autumn. Grassland is the dominant land cover around the Khangai site, except in riparian areas where well-developed willow forests prevailed. Sampling was conducted on a southeastern slope of the Khentii mountains (mean slope = 5.3 %).

### 2.2. Sample collection and preparation

At each of the Gachuurt and Khangai sites, we established five 5 m × 5 m quadrats along an elevational gradient on the selected slope (from the top of the slope to the valley bottom) (Figures 1a and b). Adjacent sampling quadrats were separated by 60–220 m at the Gachuurt

site and 320–1050 m at the Khangai site (Figure 1a). The flora within the sampling quadrats showed high species diversity (>20 species) and often differed between the quadrats (Table 1, online supplementary material). Therefore, one shoot of every herbaceous plant species was collected for each quadrat. Arthropods (mainly insects and spiders) were collected within 20 m of each quadrat (Table 2, online supplementary material). So we collected herbaceous plants and arthropods from five locations. The organism samples were dried at 45 °C for 72 h and were ground to a fine powder for stable isotope measurements.

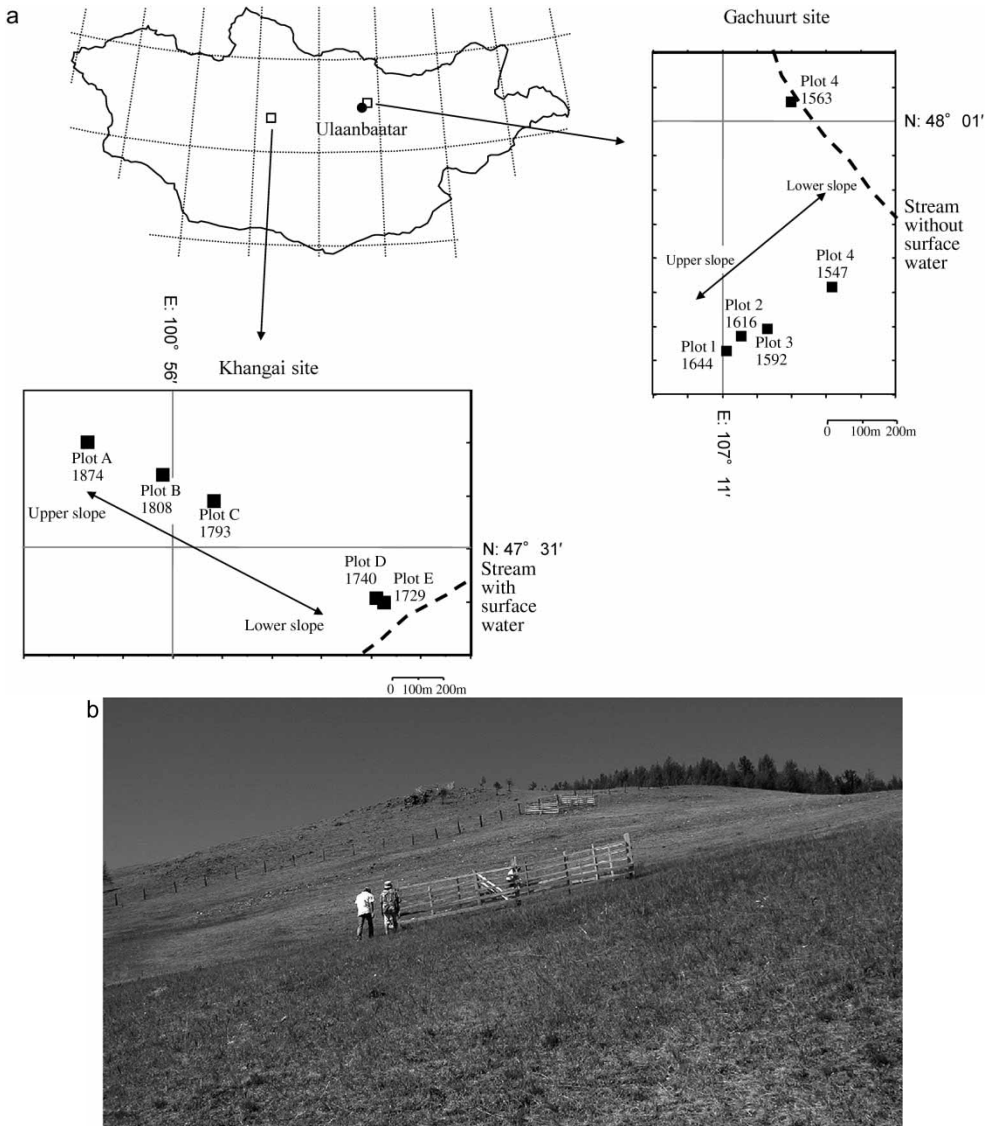


Figure 1. (a) Location of the two study sites in Mongolia. Herbaceous plants and arthropods were sampled within and around five quadrats arranged from the top of the slope to the valley bottom at each site in the following order: plot 1, 2, 3, 4, and 5 at the Gachuurt site, and plot A, B, C, D, and E at the Khangai site. Values associated with the plot names indicate elevation. (b) View looking uphill from the top of the slope at the Gachuurt site. The upper three quadrats (5 m × 5 m) corresponding to plots 1, 2, and 3 are visible on the slope. Grasslands cover the southern slope of the mountain range and the northern slope is covered with larch forest.

Samples of herbivorous mammals and dogs were collected from the hair of domestic animals (i.e. sheep, goats, cows, yaks, horses, and dogs) kept by local nomads living in Arkhangai and Tov region, which includes the Gachuurt and Khangai sites. Samples of carnivorous mammals were collected from the fur of foxes and wolves hunted around the Khangai Mountains during 1995–1998. Human hair samples were collected from local nomads around the Khangai site and from people in Ulanbator. The fur and hair samples were rinsed with 70 % ethanol and cut into smaller pieces with scissors (<1 mm length) for stable isotope measurement.

### 2.3. Stable isotope measurement

The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of the samples were measured using a continuous flow isotope ratio mass spectrometer (CF/IRMS: EA1108, Fisons; DeltaS, Finnigan). Stable isotope ratios are expressed in  $\delta$  notation as the difference in parts per thousand (‰) from the standard:

$$\delta X = (R_{\text{sample}}/R_{\text{standard}} - 1) \times 1000,$$

where  $X$  is  $\delta^{13}\text{C}$  or  $\delta^{15}\text{N}$ ,  $R$  is  $^{13}\text{C}/^{12}\text{C}$  or  $^{15}\text{N}/^{14}\text{N}$ , and appropriate standards were Vienna Pee Dee Belemnite (VPDB) and atmospheric  $\text{N}_2$  for carbon and nitrogen, respectively. Analytical precision was better than  $\pm 0.2$  ‰ for both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values.

### 2.4. Statistical analysis of the data

The average  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of the herbaceous plants, arthropods, and mammals were statistically compared by various factors such as plot (Figure 2), food habit (Figure 3), and trophic level (Figure 4). Multiple comparison of values was conducted by the Tukey-Kramer HSD test, and different letters indicate significantly ( $p < 0.05$ ) different means in the figures.

### 2.5. Classification of arthropods by feeding habit

The arthropods collected were grouped into four trophic guilds based on their feeding habit: carnivores, omnivores (feed on more than one trophic level), detritivores (including coprophagous and soil-feeding animals), and herbivores (feed on foliar or non-foliar parts of live plants). Arthropods often exhibit life-history omnivory; that is, they feed on different trophic levels at different stages of their life cycle. Although most of our arthropod samples were adults, their values might have been affected by feeding habits during their larval stage that differed (in trophic level) from the feeding habit of their adult stage. This is because the stable isotope ratio of a consumer represents the average value of its diet over a certain period of time prior to the time of sampling; the length of the period is determined by the consumer's tissue turnover rate [13]. Therefore, arthropods that exhibit life-history omnivory were also classified as omnivores. The family and feeding guild of both larval and adult stages of analysed specimens are summarised in Table 2 (online supplementary material).

## 3. Results

### 3.1. Herbaceous plants

The  $\delta^{15}\text{N}$  values of herbaceous plants decreased from the valley bottom towards the top of the slope (Figure 2). Significant negative correlations were found between the  $\delta^{15}\text{N}$  values of herbaceous

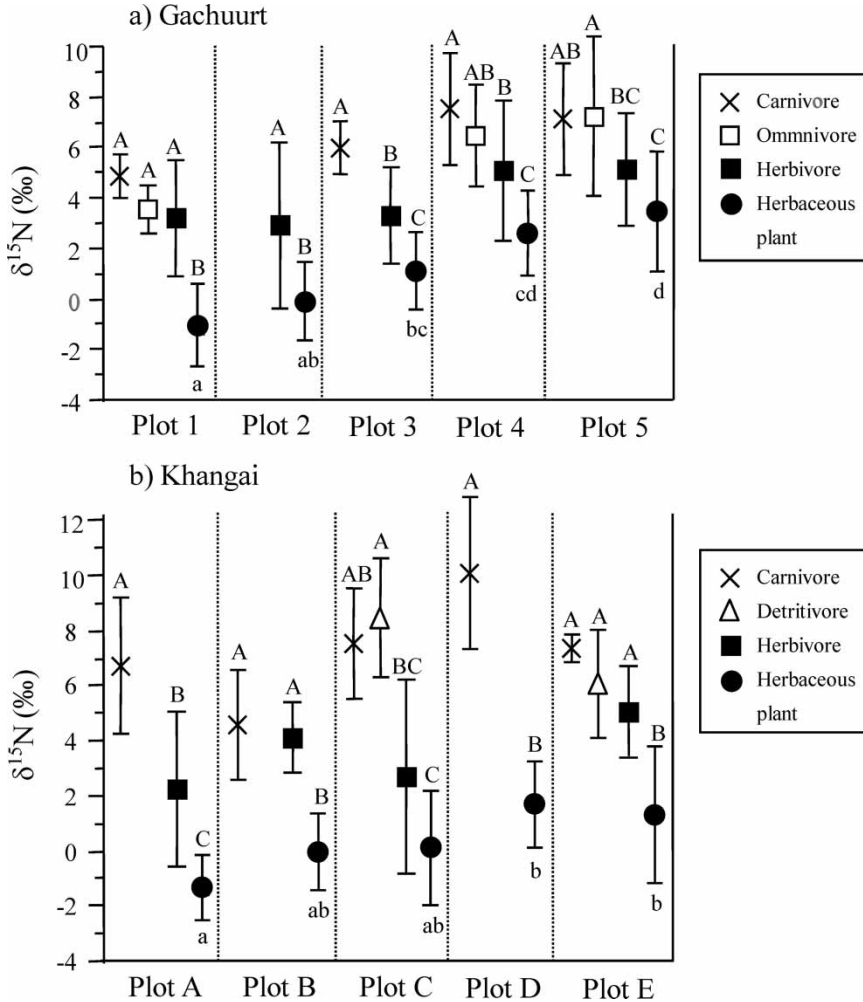


Figure 2. Mean  $\delta^{15}\text{N}$  values ( $\pm 1$  SD) of herbaceous plants and arthropods collected around the five sampling quadrats on the slope at the Gachuurt (a) and Khangai (b) sites. Mean  $\delta^{15}\text{N}$  values were compared among the three different feeding habit groups and herbaceous plants. Statistically significant differences between groups within each plot are expressed by different capital letters on the upper SD bar. Differences between plot means of  $\delta^{15}\text{N}$  of herbaceous plants are expressed by different small letters under the lower SD bar.

plants and elevation at both the Gachuurt ( $r = -0.65$ ,  $P < 0.0001$ ,  $n = 108$ ) and Khangai ( $r = -0.57$ ,  $P < 0.0001$ ,  $n = 59$ ) sites. If data from the Fabaceae family (atmospheric  $\text{N}^2$  fixers) are excluded,  $\delta^{15}\text{N}$  values of herbaceous plants correlated more strongly with elevation at both the Gachuurt ( $r = -0.71$ ,  $p < 0.0001$ ,  $n = 98$ ) and Khangai ( $r = -0.75$ ,  $P < 0.0001$ ,  $n = 53$ ) sites. The  $\delta^{15}\text{N}$  values in the uppermost quadrat and the lowest quadrat on the slope differed by 4.4 and 3.5 ‰ (on average) for the Gachuurt and Khangai sites, respectively (Figure 2). Plant species composition differed among plots (Table 1, online supplementary material). However, this trend was a genuine function of slope position rather than species composition for several reasons.  $\text{N}^2$ -fixing plants that would be depleted in  $\delta^{15}\text{N}$  were distributed throughout the slope. For example, the family representative of  $\text{N}^2$ -fixing plants, Fabaceae, was present in all plots on the slope at a frequency that was not significantly different among plots (i.e. 2/26, 2/28, 4/25, 1/13, and 1/15 in plots 1, 2, 3, 4, and 5, respectively) (Table 1, online supplementary material). In addition, for

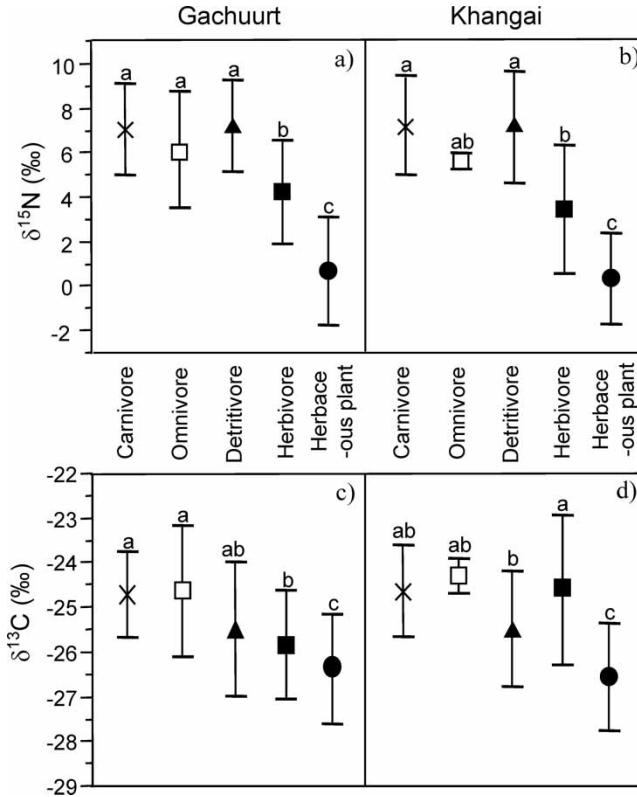


Figure 3. Mean  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values ( $\pm 1$  SD) of herbaceous plants and arthropods (herbivores, detritivores, omnivores and carnivores) at the Gachuurt (a and c) and Khangai (b and d) sites. All individuals collected from the five sampling quadrats were combined for each trophic group. There is no significant difference ( $P \geq 0.05$ ) between groups with the same letter.

several species that were present in all plots,  $\delta^{15}\text{N}$  values also increased from the top of the slope to the lower slope. For example,  $\delta^{15}\text{N}$  values of *Leontopodium leontopodioides* (Asteraceae) at the Gachuurt site increased from the top of the slope to the bottom of the slope as follows:  $-2.9$  (plot 1),  $-1.1$  (plot 2),  $+2.7$  (plot 3), and  $+3.2$  (plot 4) (Table 1, online supplementary material). Similarly, for *Arenalia capillaries* (Caryophyllaceae) at the Khangai site,  $\delta^{15}\text{N}$  increased from the top of the slope (plot A) to the bottom of the slope (plot D) as follows:  $-0.7$ ,  $-0.5$ ,  $+1.1$ , and  $+3.2$  (Table 1, online supplementary material). The relationship between  $\delta^{15}\text{N}$  difference and elevation was therefore not an artefact of non-uniform distribution of particular plant species with inherently high or low  $\delta^{15}\text{N}$  values.

In contrast to the  $\delta^{15}\text{N}$  values, the spatial gradient of  $\delta^{13}\text{C}$  values of herbaceous plants along the slope was not as clear. Although a significant positive correlation between  $\delta^{13}\text{C}$  and elevation was found at both the Gachuurt site ( $r = 0.21$ ,  $P = 0.03$ ,  $n = 108$ ) and the Khangai site ( $r = 0.37$ ,  $P = 0.006$ ,  $n = 53$ ), the mean  $\delta^{13}\text{C}$  values of herbaceous plants in the uppermost quadrat (plot 1 or A) and the lowest quadrat (plot 5 or E) differed only by  $1.5$  ‰ at the Gachuurt site and  $1.1$  ‰ at the Khangai site ( $P < 0.05$  by the  $t$ -test in both cases).

### 3.2. Arthropods

As observed in the isotopic signatures of plants,  $\delta^{15}\text{N}$  values of herbivorous arthropods showed a significant negative correlation with elevation at the Gachuurt site ( $r = -0.33$ ,  $P = 0.002$ ,  $n = 88$ ; Figure 2a). Significant negative correlations of  $\delta^{15}\text{N}$  values with elevation were also



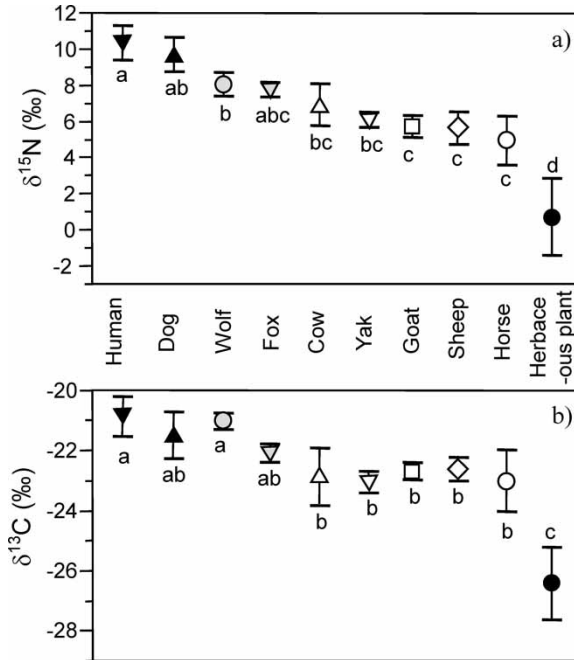


Figure 4. Mean  $\delta^{15}\text{N}$  (a) and  $\delta^{13}\text{C}$  (b) values ( $\pm 1$  SD) of mammal hairs collected from central Mongolia. There is no significant difference ( $P \geq 0.05$ ) between species with the same letter. Mean  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values of herbaceous plants collected at the Gachuurt and Khangai sites are also shown.

found for carnivorous ( $r = -0.34$ ,  $P = 0.030$ ,  $n = 42$ ) and omnivorous arthropods ( $r = -0.49$ ,  $P = 0.020$ ,  $n = 22$ ) at the Gachuurt site (Figure 2a). At the Khangai site, however, no significant relationships were found between  $\delta^{15}\text{N}$  values and elevation for any of the trophic groups (herbivores,  $r = -0.32$ ,  $n = 32$ ; detritivores,  $r = 0.19$ ,  $n = 14$ ; carnivores,  $r = -0.31$ ,  $n = 15$ ; omnivores,  $r = -0.36$ ,  $n = 5$ ;  $P > 0.05$  for all).

Arthropod  $\delta^{15}\text{N}$  values were enriched relative to  $\delta^{15}\text{N}$  values of herbaceous plants (Figures 2a and b); the differences in  $\delta^{15}\text{N}$  values between herbaceous plants and arthropod consumers (herbivores, omnivores, detritivores, and carnivores) were all significant at both the Gachuurt and Khangai sites (Figures 3a and b;  $P < 0.0001$ ). The mean trophic enrichment of  $\delta^{15}\text{N}$  between herbaceous plants and herbivores was 3.5 and 3.1 ‰ at the Gachuurt and Khangai sites, respectively, when all the samples collected from the five quadrats were combined.  $\delta^{15}\text{N}$  values of carnivores and detritivores were significantly higher than those of herbivores (2.8–3.7 ‰ on average) at both study sites ( $P < 0.0001$ ). Omnivore  $\delta^{15}\text{N}$  was also significantly enriched relative to herbivore  $\delta^{15}\text{N}$  values at the Gachuurt site ( $P < 0.001$ ), but not at the Khangai site.

As with  $\delta^{15}\text{N}$  values, arthropod  $\delta^{13}\text{C}$  signatures were enriched relative to  $\delta^{13}\text{C}$  values of herbaceous plants. The differences in  $\delta^{13}\text{C}$  values between herbaceous plants and arthropod consumers were all significant at both the Gachuurt and Khangai sites (Figures 3c and d;  $P < 0.01$ ). The mean  $\delta^{13}\text{C}$  enrichment from herbaceous plants to herbivores was 0.6 and 2.0 ‰ at the Gachuurt and Khangai sites, respectively. The differences in  $\delta^{13}\text{C}$  enrichments between carnivores, detritivores, omnivores, and herbivores were less clear (Figures 3c and d).

### 3.3. Mammals

The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of mammal hair were enriched relative to those of herbaceous plants at the Gachuurt and Khangai sites (Figure 4). The mammals that showed the most enriched  $\delta^{15}\text{N}$

and  $\delta^{13}\text{C}$  values were Mongolian people and dogs, followed by carnivorous mammals (wolves and foxes) and domestic herbivorous mammals (Figure 4). The average  $\delta^{15}\text{N}$  values of domestic herbivorous mammals (i.e. sheep, goats, cows, yaks, and horses) were depleted by 3.1 ‰ relative to those of Mongolian carnivores (i.e. people, dogs, wolves and foxes) and were enriched by 5.2 ‰ relative to those of herbaceous plants. The average  $\delta^{13}\text{C}$  values of domestic herbivorous mammals were depleted by 1.5 ‰ relative to those of carnivores and were enriched by 3.5 ‰ relative to the average  $\delta^{13}\text{C}$  signatures of herbaceous plants (Figure 4).

The within-species variations of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures of carnivorous and herbivorous mammals were much smaller than the corresponding variations in herbaceous plants and arthropod trophic groups (Figures 3 and 4), despite the fact that mammal hair samples were collected from a wide area that included the two study sites.

## 4. Discussion

### 4.1. $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ trophic enrichment in the Mongolian grassland food webs

Information on  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  enrichments associated with trophic transfers of carbon and nitrogen is increasingly being accumulated [10,11]. However, empirical data on isotopic enrichments in grassland food webs are relatively scarce. The present study revealed trophic enrichments in arthropods ranging from 0.0 to 1.1 ‰ for  $\delta^{13}\text{C}$  and from 2.8 to 3.7 ‰ for  $\delta^{15}\text{N}$  in Mongolian grasslands (Figure 3). These trophic fractionations accord well with the general patterns reported from various biomes or ecosystems. For example, Post [11] reviewed isotopic studies of both aquatic and terrestrial food webs, reporting mean enrichments per trophic level of  $0.4 \pm 1.3$  ‰ for  $\delta^{13}\text{C}$  and  $3.4 \pm 1.0$  ‰ for  $\delta^{15}\text{N}$ . Thus, the generally accepted fractionation values of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  are also applicable to the trophic analysis of terrestrial arthropods in Mongolia. However, it should be noted that, in grassland food webs, where the major source of carbon and nitrogen are herbaceous plants, there may be a different isotopic baseline. For example, detritivores (including soil feeders and coprophagous arthropods) showed enriched  $\delta^{15}\text{N}$  relative to herbivores, and their  $\delta^{15}\text{N}$  values were comparable to carnivore  $\delta^{15}\text{N}$  values at both study sites (Figures 2 and 3a and c). The enrichment of  $\delta^{15}\text{N}$  in detritivores probably resulted from the fact that soil organic matter and dung are usually enriched in  $\delta^{15}\text{N}$  owing to selective loss of  $^{14}\text{N}$  during biogeochemical transformations such as ammonia evaporation and denitrification [19,20]. If this is the case, the analysis of trophic position by using consumer  $\delta^{15}\text{N}$  may require the identification of multiple end members that eventually support higher trophic level organisms in this ecosystem [12].

In addition to the general trophic enrichment of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , mammal samples showed the possibility of tissue-specific isotope fractionation. Isotopic discrimination between herbivorous mammals and herbaceous plants (+3.5 ‰ for  $\delta^{13}\text{C}$  and +5.2 ‰ for  $\delta^{15}\text{N}$ ) was relatively large (Figures 4 and 5). We consider that these unexpected  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  enrichments could be due to the relatively large fractionation of isotopes into mammal hair during hair formation. Although we measured mammal hairs to represent the values of individuals, mammal hair is composed mainly of keratin and therefore differs from most other body tissues in its turnover rate and protein, lipid and carbohydrate composition [21,22]. As a result, hair isotopic signatures often show distinct values. In fact, relatively large isotopic discriminations between mammal hairs and food sources have been reported for seals, sea lions [23], monkeys [24], and humans [25]. In contrast, the discrimination between hair samples of herbivores and carnivores in our study (+1.5 ‰ for  $\delta^{13}\text{C}$  and +3.1 ‰ for  $\delta^{15}\text{N}$ ) was close to, or in agreement with, the generally accepted stepwise enrichments of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  along food chains (Figures 4 and 5) [10,11]. This suggests that isotopic differences between body tissue and hair are similar at different trophic levels (Figure 5). Assuming that the enrichment factors (+1.5 ‰ for  $\delta^{13}\text{C}$  and +3.1 ‰ for  $\delta^{15}\text{N}$ ) between herbivorous and carnivorous

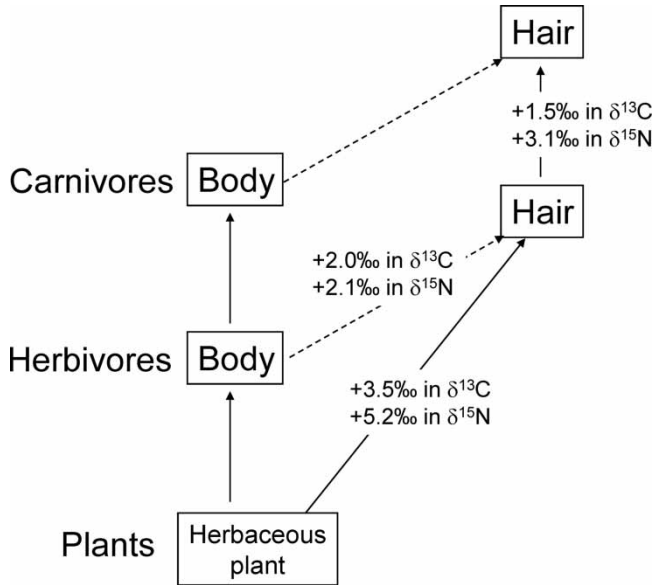


Figure 5. Schematic diagram of isotope trophic enrichment (solid arrows) and tissue-specific isotope fractionation (dashed arrows) as the determinants of  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values of mammal hairs. The mean values of isotope enrichment and fractionation are shown on the corresponding arrows.

mammals are applicable to enrichment between herbaceous plants and body tissues of herbivorous mammals, the apparent isotopic discrimination between body tissue and hair can be estimated as 2.0 ‰ ( $=3.5-1.5$ ) for  $\delta^{13}\text{C}$  and 2.1 ‰ ( $=5.2-3.1$ ) for  $\delta^{15}\text{N}$  (Figure 5). These isotopic shifts from body tissues to hair seem to be large when analysing the food source or trophic position of mammals, especially in food webs with multiple resource bases. Thus, when delineating food webs by using stable isotopes, we should take into account isotopic discrimination between the tissue types analysed [22,26]. We infer from the present study that about +2 ‰ enrichment in both  $\delta^{13}\text{C}$  and in  $\delta^{15}\text{N}$  can occur between the body tissue and hair of mammals.

#### 4.2. Food-web characteristics deduced from $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ patterns

The stable carbon and nitrogen isotope ratios of consumers represent the average values of their food sources and trophic positions. In arthropod food webs, the general  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  enrichments per trophic level reviewed by Post [11] were applicable when the primary diets of herbivorous and carnivorous arthropods were herbaceous plants and herbivores, respectively. In addition, at the Gachuurt site,  $\delta^{15}\text{N}$  values of carnivores and herbivores increased down the slope, which could only be explained by the herbaceous plants that were the basal N sources for the arthropods having a similar  $\delta^{15}\text{N}$  increase pattern down the slope. These facts confirmed that, at both the Gachuurt and Khangai sites, herbaceous plants generally formed the primary producers of arthropod food webs. However,  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of higher trophic groups may also vary due to food selectivity, the spatial extent of food acquisition, and the metabolic rate of the consumers when the isotopic values of their food resources vary spatially and temporally [22,27]. For example, we observed variability of  $\delta^{13}\text{C}$  in arthropods within trophic groups. At the Gachuurt site,  $\delta^{13}\text{C}$  enrichment in Hymenoptera species that feed on non-foliar parts (mean  $\delta^{13}\text{C} = -24.0 \pm 0.7$  ‰) was significantly ( $p < 0.05$  by t-test) greater than that in other groups except Diptera (see Table 2, online supplementary material). The difference in  $\delta^{13}\text{C}$  between the two herbivore groups appears to reflect the greater enrichment of  $\delta^{13}\text{C}$  within non-foliar parts (e.g. stems, roots, and seeds) than in foliage [28,29]. This suggests

that the variability of  $\delta^{13}\text{C}$  within trophic groups may be partly attributed to species-specific differences in the diets of the consumers (Table 2, online supplementary material).

In addition, the spatial scale of energy channels from primary producers may have also influenced the variability of consumer isotope values. At both the Gachuurt and Khangai sites there was a negative correlation between  $\delta^{15}\text{N}$  values of herbaceous plants and elevation. Although the underlying mechanism for this correlation was unclear, it might reflect different activities in N mineralisation, denitrification, and  $\text{N}^2$ -fixation along the soil moisture gradient of the slope, with resultant differences in isotope fractionation at the different slope positions [30,31]. At the Gachuurt site, the negative correlation between  $\delta^{15}\text{N}$  values and elevation was also seen in herbivorous arthropods and even omnivores and carnivores (Figure 2a). This fact suggests that arthropod consumers gained food resources from a relatively restricted area of the slope, implying no apparent upward or downward movements of carbon and nitrogen via trophic transfer in the food web. On the slope of the Khangai site, however, no negative correlation between  $\delta^{15}\text{N}$  values and elevation was found for any of the arthropod trophic groups (Figure 2b). Considering that the faunal assemblies were similar between the two sites (see Table 2, online supplementary material), the difference in elevational gradient of consumer  $\delta^{15}\text{N}$  values might be a result of the spatial scale of arthropod food webs caused by differences in the landform. At the Khangai site, where the slope gradient (5.3 %) is much gentler than the Gachuurt site (mean slope = 29 %), it is likely that there is more movement of arthropods between the valley bottom and the top of the slope than there is at the Gachuurt site, resulting in less clear trends in isotopic values with elevation.

The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of mammals exhibited incremental enrichment along food chains. The  $\delta^{15}\text{N}$  values of humans and domestic dogs indicate that they occupy the highest trophic position among mammals. The mean  $\delta^{15}\text{N}$  values of wolves ( $8.1 \pm 0.6$  ‰) and foxes ( $7.7 \pm 0.4$  ‰) were both significantly lower than those of Mongolian people ( $10.3 \pm 0.9$  ‰) (Figure 4a). The reason for this difference is unclear. The  $\delta^{15}\text{N}$  values of cattle were higher than those of other domestic herbivorous mammals except for yaks (Figure 4a). This is because our samples included many calves that exhibited relatively high  $\delta^{15}\text{N}$  values (7.5–9.1 ‰), presumably resulting from them drinking milk with high  $\delta^{15}\text{N}$  values [32].

One of the salient features of our results was that the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of mammals displayed little variation within species. This low variability for mammals suggested non-selective feeding habits, broader spatial scales of food acquisition, and/or slower metabolic rates compared with arthropod consumers. Cabana and Rasmussen [33] also showed that large organisms tend to have lower variability in  $\delta^{15}\text{N}$  values than smaller organisms in aquatic ecosystems. These results suggest that grazing effects on grassland vegetation might be different with respect to duration and locality, depending on whether the overgrazing is caused by mammals or arthropods. Although the effects might be specific to certain landscape features (e.g. steep slopes), it is possible that arthropod consumers have a direct grazing effect on local scale biodiversity and biomass of plants of grassland communities, whereas mammals have effects at much larger spatial and temporal scales. Furthermore, overgrazing by livestock mammals can alter the abundance and species composition of arthropod communities that depend on local primary productivity [2]. The present study suggests that in addition to delineating the food-web pathways, the spatial and temporal variation of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures between organisms and landscapes can provide useful information on the structure and dynamics of grassland ecosystems.

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