

Original Article

## Carbon and Nitrogen Stable Isotope Values for Plants and Mammals in a Semi-Desert Region of Mongolia

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### Abstract

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Little information exists on the isotopic signatures of plants and animals in Mongolia, limiting the application of stable isotope analysis to wildlife biology studies. Here we present plant and mammal carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) isotope values from a desert-steppe region of southeastern Mongolia. We analyzed 11 samples from 11 plant species and 93 samples from 24 mammal species across Ikh Nart Nature Reserve, and compared these numbers to isotope values reported from other areas of Mongolia. Our plant and mammal  $^{13}\text{C}$  and  $^{15}\text{N}$  values were similar to those from a similar arid steppe region and more enriched than those from less arid habitats. Habitat variation within and between study sites has an important influence on  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  variation. Our results supplement current knowledge of isotopic variation in Mongolia and provide a reference for future stable isotope research in Mongolia and similar Asian steppe ecosystems.

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### Introduction

Stable isotope analysis (SIA) is a rapidly developing method in wildlife studies that is efficient, cost-effective and minimally invasive (Ben-David & Flaherty, 2012). Since its potential applications were first explored in the late 1970s and early 1980s (DeNiro & Epstein, 1978, 1981), SIA has been used to address a variety of topics for many different ecosystems and organisms (Dawson *et al.*, 2002). In the wildlife field, it has primarily been used to investigate diet and

foraging behavior, movement patterns (including migration), and resource use (Gannes *et al.*, 1998; Ben-David & Flaherty, 2012).

Stable isotopes are isotopes (element forms with differing numbers of neutrons in the nucleus) that do not decay over time (Fry, 2006). SIA measures the relative amounts of target isotopes (reflecting the conditions of the environment) in individuals. For example, areas of low water availability are associated with a higher rate of

assimilation of heavier nitrogen isotopes by plants and animals (Murphy & Bowman, 2006). The ratio of heavy to light nitrogen isotopes in plant and animal tissue can therefore be used to assess water availability in their environment.

Isotopes commonly used in wildlife studies include two forms of carbon ( $^{12}\text{C}$  and  $^{13}\text{C}$ ) and nitrogen ( $^{14}\text{N}$  and  $^{15}\text{N}$ ). Carbon isotope ratios ( $\delta^{13}\text{C}$ ) of plants and animals vary depending on the photosynthetic pathway of primary producers in the community of interest (Chen *et al.*, 2007; Koch, 2007). Nitrogen isotope ratios ( $\delta^{15}\text{N}$ ) vary based on water availability, the concentration of animal and human waste in soil and water, and the trophic level and age of the organism (Overman & Parrish, 2001; Murphy & Bowman, 2006; Koch, 2007, Hyodo *et al.*, 2012). Target isotope ratios are obtained by analyzing samples of the species of interest: commonly stem, leaf, and seed for plants, and hair, feathers, muscle, bones or blood for animals, depending on the nature of the study (Koch, 2007; Ben-David & Flaherty, 2012).

Isotopic signatures in samples can be used to describe trends in the movement of elements

through individuals and through ecosystems (West *et al.*, 2006). For example,  $\text{C}_4$  plants have an alternative photosynthetic pathway to more common  $\text{C}_3$  plants (Bender, 1968; Chen *et al.*, 2007) and are significantly more enriched in  $^{13}\text{C}$  than  $\text{C}_3$  plants. The resulting dichotomy of  $\delta^{13}\text{C}$  values (Post, 2002) can be traced up a food web from herbivores consuming the plants, to the  $\delta^{13}\text{C}$  signature of the carnivores consuming the herbivores. Isotope models can be used to estimate the diet of a species of interest based the isotope signature of the focal species and of potential food sources (Koch, 2007). They can also be used to predict the origin of an individual or population based on the similarity of sample isotopic values to those from potential locations of origin (Ben-David & Flaherty, 2012).

Effective application of SIA requires a basic understanding of the general distribution of different isotope forms in the system of interest. Ideally, a predictive isotope study will have access to a series of isotopic measurements that capture the variation between individuals and between species at the study site, between different habitat

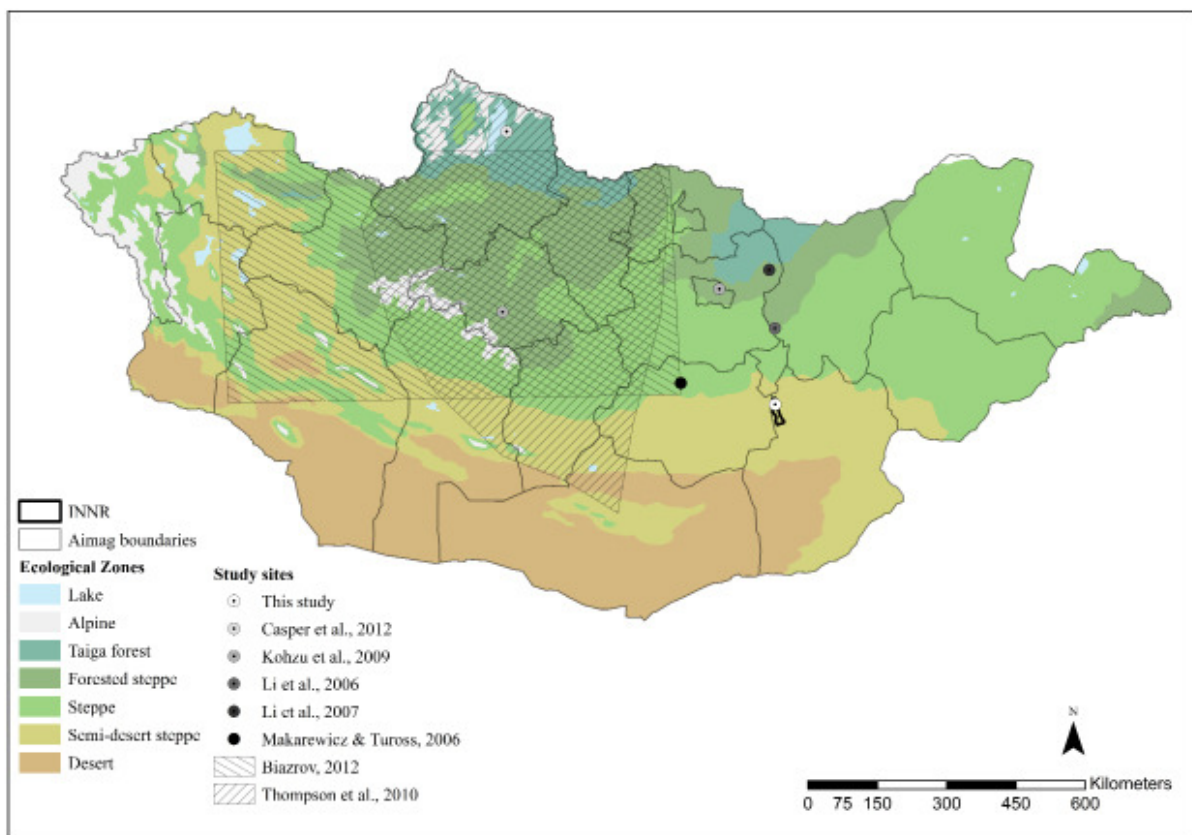


Figure 1. Locations of previous plant and mammal isotope studies in Mongolia, including Ikh Nart Nature Reserve (INNR). Locations presented as points for studies that occurred at small scales, and larger polygons for studies that covered broader areas.

Table 1. Plant and mammal stable isotope studies previously conducted in Mongolia.

Study	Location	Aimag	Latitude	Longitude	Samples collected
Li <i>et al.</i> , 2006	Kherelenbayan-Ulaan	Khenti	47.2 N	108.7 E	plants
Makarewicz & Tuross, 2006	Baga Gazaryn Chuluu	Dundgovi	46.2 N	106.0 E	plants, mammals
Li <i>et al.</i> , 2007	Mungunmorit	Tuv	48.4 N	108.7 E	plants, soil
Kohzu <i>et al.</i> , 2009	Gachuurt	Ulaanbaatar city	48.0 N	107.2 E	plants, mammals, arthropods
Kohzu <i>et al.</i> , 2009	Khangai	Arkhangai	47.5 N	100.9 E	plants, mammals, arthropods
Thompson <i>et al.</i> , 2010	Multiple locations through central Mongolia	Arkhangai, Bulgan, Orkhon, Ulaanbaatar city, Umnugovi, Uvs	43.5 to 51.2 N	93.5 to 107.0 E	mammals
Biazrov, 2012	Khangai plateau	Arkhangai, Bayankhongor, Bulgan, Govi-Altai, Khovd, Khuvsgul, Uvurhangai, Selenge, Tuv, Uvs, Zavkhan	46.0 to 50.0 N	92.0 to 106.0 E	plants
Casper <i>et al.</i> , 2012	Lake Khuvsgul	Khuvsgul	51.0 N	100.8 E	plants

types, and across broader areas such as ecozones (Bond & Diamond, 2011; Newsome *et al.*, 2012). Once this inherent variation is known and controlled for, it is possible to isolate variation specifically due to the factors of interest (e.g., isotope enrichment from food to consumer or variation in values due to water availability) and use this variation to answer questions about diet, movement, and resource use.

Here we present carbon and nitrogen isotope values of several mammal and plant species from a semi-desert steppe region of Dornogobi *aimag* (province), Mongolia, and compare them to the values reported by other SIA research projects in Mongolia. Although our samples do not represent a comprehensive species list, to date there have been few stable isotope studies conducted in Mongolia, and even fewer exploring variation in plant and mammal values (Fig. 1, Table 1). Therefore our study, although not comprehensive, serves as a supplement to current knowledge of the Mongolian isoscape and as a reference for future isotope research in Mongolia and similar arid steppe regions.

## Materials and Methods

### Study Area

We collected samples of major forage plants, and hair samples from domestic and wild ungulates, small mammals, and a carnivore species in Ikh Nart Nature Reserve (hereafter

Ikh Nart), Dornogobi *aimag* (province), Mongolia (45°43'N, 108°39'E). Ikh Nart, a 666 km<sup>2</sup> protected area with a unique landscape (Myagmarsuren, 2000), was established in 1996 to conserve a population of globally threatened argali sheep (*Ovis ammon*) (Reading *et al.*, 2011). Ikh Nart lies at the margin of steppe and semi-desert ecozones (Mallon, 1985) and includes areas of rugged rocky outcrops, open plains, and steep drainages (Reading *et al.*, 2011). Shrubs, grasses, and forbs are the primary vegetation of the plains. Rocky areas and drainages often include trees (e.g. *Ulmus pumila* and *Salix sp.*) (Jackson *et al.*, 2006). Climate is arid (<200 mm of annual precipitation) and annual temperatures typically range from -40°C to +40°C.

### Sample Collection

We collected samples from plant species identified as major components of wild ungulate and livestock diet within the reserve (Wingard *et al.*, 2011). Major component plant species, defined as those with a >5% occurrence in livestock and argali diet (Wingard *et al.*, 2011), included the C<sub>3</sub> species *Bassia dasyphylla*, *Allium sp.*, *Ajania achilleoides*, *Artemisia frigida*, *A. ruthifolia*, *Convolvulus spp.*, *Oxytropis spp.*, *Dracocephalum foetidum*, *Agropyron cristatum*, *Stipa sp.*, and *Haplophyllum dauricum*, and one C<sub>4</sub> plant, *Cleistogenes squarrosa*.

We collected livestock samples from carcasses and from live animals at herder campsites. Over

one hundred nomadic pastoralist households occur in and around the reserve (Davie *et al.*, 2014). Households typically raise up to five species of livestock: goats (*Capra aegragus*), sheep (*Ovis aries*), horses (*Equus ferus caballus*), cattle (*Bos taurus*) and camels (*Camelus bactrianus*). Only two species of wild ungulates are year-round residents: argali sheep and Siberian ibex (*Capra sibirica*). We opportunistically collected argali and ibex samples from carcasses and observed bedding sights.

Many species of small mammals are found in Ikh Nart, including hedgehogs (Erinacidae), hares (Leporidae), pika (Ochotonidae), voles (Arvicolidae), hamsters (Cricetidae), jerboa (Dipodidae), gerbils (Gerbilidae) and marmots (Sciuridae) (Murdoch *et al.*, 2006). We analyzed samples from species in each family. Hedgehog spine samples were collected from hedgehogs captured as part of an ongoing radio-tracking study (see Batdorj, 2012). We collected tolai hare (*Lepus tolai*) samples opportunistically from carcasses, including those located in raptor nests (primarily Golden Eagle [*Aquila chrysaetos*] and Cinereous Vulture [*Aegypius monachus*]). Siberian marmot (*Marmota sibirica*) samples were from shed hairs gathered from the resting areas of burrow entrances. All other small mammal samples (rodent and pika) were collected from individuals captured in small mammal box traps (XLK folding box trap, 3 x 3.75 x 12", H.B. Sherman Traps, Tallahassee, Florida, USA). Box trap grids are established at five sites in the study area as part of an ongoing survey by another project (G. Wingard, pers. comm.). We surveyed each grid once per month during June, July, and August. Traps were opened from 18:00 to 07:00 the following day, and checked daily. We collected hair samples by gently pulling 30-50 hairs from the flank of captured animals.

We obtained wolf hair samples from the pelts of hunted wolves. We used these samples to represent the carnivore community. Other carnivores occur in Ikh Nart, such as red fox (*Vulpes vulpes*), corsac fox (*Vulpes corsac*), badger (*Meles leucurus*) and Pallas' cat (*Otocolobus manul*) (Murdoch *et al.*, 2006), but we were unable to analyze samples from these species during our study.

### Stable Isotope Analysis

We estimated  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values by measuring the ratio of isotopes in the gases of

combusted samples using a stable isotope ratio mass spectrometer (IRMS). We determined the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures for each individual livestock, wild ungulate, and wolf sample. We combined individual samples from small mammals and plants into one bulked sample for each species. For example, we collected 14 tolai hare (*Lepus tolai*) samples across the reserve, and combined them all into one sample for analysis. We did this to provide a data point for each species representing the average isotopic value for the species in the reserve.

### Sample Pre-treatment

We dried plant samples (stems and leaves) in an oven from 24 to 72 hours at 50°C, and then ground them to a powder using a Wig-L-Bug (Crescent Dental, Model 3110B).

We sonicated hair samples in glass beakers of 50 ml deionized H<sub>2</sub>O for two three-minute intervals using a tabletop ultrasonic cleaner (Fisher Scientific FS6) to remove coarse debris. After sonication, we rinsed samples under a ventilation hood in a 2:1 chloroform-methanol solution to remove oils and fine debris (Merkle *et al.*, 2011). We allowed hairs to air dry in aluminum weighing boats under a ventilation hood for 15 minutes, and then transferred them to an oven (VWR 1320) to dry for 24 to 72 hours at 50°C.

### Sample Analysis

We combusted samples for stable carbon and nitrogen isotopes offline and then processed them separately on a dual inlet V.G. SIRA II isotope ratio mass spectrometer (IRMS). The results for carbon and nitrogen were reported in per mil (‰) units using the delta ( $\delta$ ) notation:

$$\delta \text{HX} = [(R_{\text{sample}}/R_{\text{standard}}) - 1] * 1000,$$

where,  $\delta \text{HX}$  represents the measurement of difference relative to a standard (V-PDB for C and atmospheric N<sub>2</sub> for N), H is the heavy isotope mass of element X, and R is the ratio  $^{13}\text{C}/^{12}\text{C}$  or  $^{15}\text{N}/^{14}\text{N}$  (Peterson & Fry, 1987). Precision of sample measurements was  $\pm 0.05\text{‰}$  for C and  $\pm 0.1\text{‰}$  for N, based on replicate standards (USGS-22 and IAEA-N1) and internal-lab standards. Mean values for each species or group were reported as mean  $\delta^{13}\text{C}$  or  $\delta^{15}\text{N} \pm 1 \text{ SD}$ . We tested for significant differences between ungulate groups using a Kruskal-Wallis nonparametric Analysis of Variance and considered differences significant when  $P < 0.05$ .

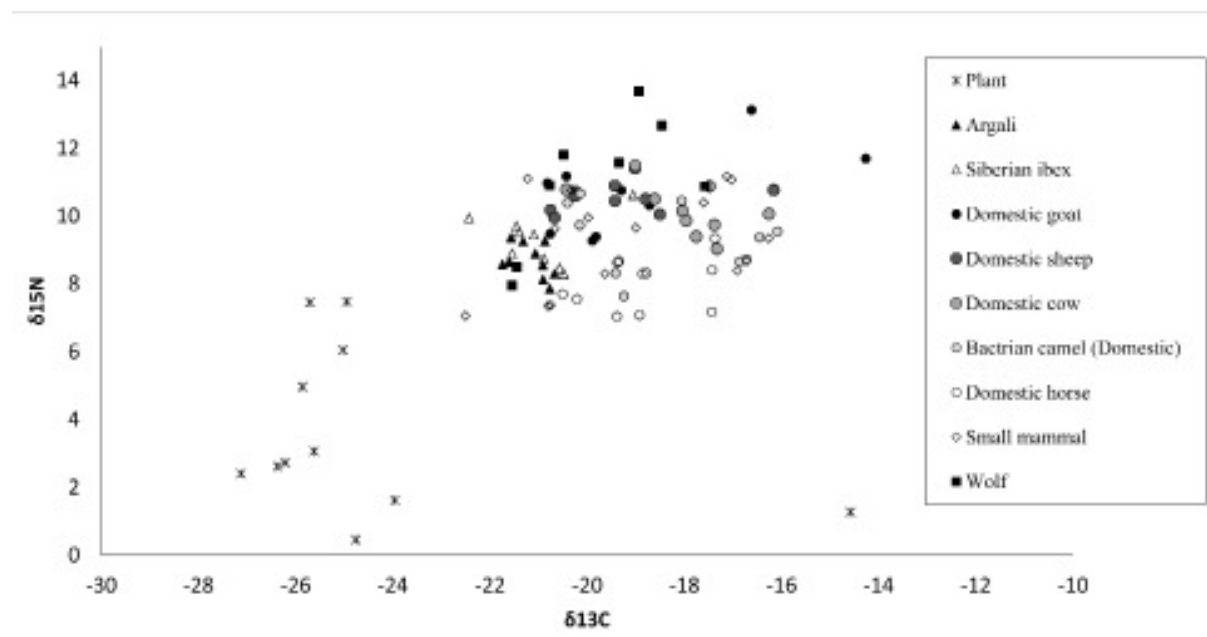


Figure 2. Distribution of plant, wild and domestic ungulate, small mammal, and wolf  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values from Ikh Nart Nature Reserve, Mongolia. Plant and small mammal values represent  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values for one bulked sample ( $N = 3$  to 10 individuals) for each species.

Table 2.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values for bulked samples of plant species in Ikh Nart Nature Reserve, Mongolia.  $N =$  number of individual plants included in a bulked sample.

Group/Family	Species	N	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$
<b>C<sub>3</sub> Plants</b>				
Amaranthaceae	<i>Bassia dasyphylla</i>	3	-25.7	7.5
Amaryllidaceae	<i>Allium sp.</i>	8	-25.0	6.0
Asteraceae	<i>Ajania achilleoides</i>	3	-27.1	2.4
	<i>Artemisa sp.</i>	6	-25.6	3.1
Convolvulaceae	<i>Convolvulus sp.</i>	4	-25.0	7.5
Fabaceae	<i>Oxytropis sp.</i>	3	-26.2	2.7
Lamiaceae	<i>Dracocephalum foetidum</i>	3	-25.9	5.0
Poaceae	<i>Agropyron cristatum</i>	3	-26.4	2.6
	<i>Stipa sp.</i>	5	-24.0	1.6
Rutaceae	<i>Haplophyllum dauricum</i>	3	-24.8	0.5
<b>C<sub>4</sub> Plants</b>				
Poaceae	<i>Cleistogenes squarrosa</i>	3	-14.6	1.3

## Results

We collected 44 samples from 11 plant species and 173 samples from 24 mammal species (Tables 2, 3 and 4). We therefore analyzed 11 plant samples (1 bulked sample per species, Table 2), 16 small mammal samples (1 bulked sample per species, Table 4), 28 wild ungulate and wolf samples (Table 3), and 50 livestock samples

(Table 3). Plant species had lower  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures than the mammals sampled with the exception of *C. squarrosa* (Fig. 2; Table 2). *C. squarrosa* was the most enriched in  $^{13}\text{C}$  at -14.6 ‰, while *A. achilleoides* was the most depleted at -27.1 ‰ (Table 2). The mean  $\delta^{13}\text{C}$  value for plants was  $-24.6 \pm 3.4$  ‰. *Convolvulus sp.* was the most enriched in  $^{15}\text{N}$ , at 7.5 ‰, and *H. dauricum* was the most depleted in  $^{15}\text{N}$ , at 0.5 ‰. The mean

Table 3. Mean  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values for wild ungulates, livestock, and wolves in Ikh Nart Nature Reserve, Mongolia. N = the number of samples analyzed for each species.

Group/Family	Species	Common name	N	$\delta^{13}\text{C}$		$\delta^{15}\text{N}$	
				Mean	SD	Mean	SD
Wildlife							
Bovidae	<i>Capra sibirica</i>	Siberian ibex	10	-20.9	0.9	9.3	0.8
	<i>Ovis ammon</i>	Argali sheep	10	-21.1	0.4	8.7	0.5
Canidae	<i>Canis lupus</i>	Gray wolf	8	-19.8	1.5	11.0	1.9
Livestock							
Bovidae	<i>Bos tarus</i>	Domestic cow	10	-18.0	1.1	10.2	0.7
	<i>Capra aegragus</i>	Domestic goat	10	-18.9	2.2	10.6	1.2
	<i>Ovis aries</i>	Domestic sheep	10	-19.3	1.4	10.6	0.4
Equidae	<i>Equus caballus</i>	Domestic horse	10	-18.8	1.1	8.1	0.7
Camelidae	<i>Camelus bactrianus</i>	Bactrian camel (domestic)	10	-18.3	1.7	9.3	1.0

Table 4.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values for bulked samples of small mammal species in Ikh Nart Nature Reserve. N = number of individuals included in a bulked sample.

Family	Species	Common name	N	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$
Erinaceidae	<i>Hemiechinus auritus</i>	Long-eared hedgehog	4	-20.3	10.8
	<i>Mesechinus dauuricus</i>	Daurian hedgehog	3	-20.0	10.0
Leporidae	<i>Lepus tolai</i>	Tolai hare	14	-20.8	7.4
Ochotonidae	<i>Ochotona pallasii</i>	Pallas's pika	11	-22.5	7.1
Arvicolidae	<i>Alticola semicanus</i>	Mongolian silver vole	5	-17.6	10.4
Cricetidae	<i>Allocricetulus curtatus</i>	Mongolian hamster	8	-20.7	9.6
	<i>Cricetulus barabensis</i>	Striped dwarf hamster	2	-19.6	8.3
	<i>Eolagurus luteus</i>	Yellow steppe lemming <sup>1</sup>	2	-21.7	
	<i>Phodopus roborovskii</i>	Desert hamster	4	-21.2	11.1
	<i>Phodopus campbelli</i>	Campbell's hamster	1	-19.0	9.7
Dipodidae	<i>Allactaga sibirica</i>	Siberian jerboa	7	-16.7	8.7
	<i>Dipus sagitta</i>	Northern three-toed jerboa	4	-16.3	9.4
Gerbillidae	<i>Meriones meridianus</i>	Mid-day gerbil	8	-16.9	8.4
	<i>Meriones unguiculatus</i>	Mongolian gerbil	11	-17.1	11.2
Muridae	<i>Mus musculus</i>	House mouse	2	-17.0	11.1
Sciuridae	<i>Marmota sibirica</i>	Siberian marmot	9	-20.8	7.4

<sup>1</sup> Only a carbon value is presented as the sample was too small to obtain a nitrogen value.

$\delta^{15}\text{N}$  value for plants was  $3.6 \pm 2.5$  ‰.

The isotope signatures of wild ungulates and livestock formed three distinct clusters. Mean argali and ibex values were significantly depleted in  $^{13}\text{C}$  ( $H = 34.62$ ,  $DF = 1$ ,  $P < 0.001$ ) and  $^{15}\text{N}$  ( $H = 7.70$ ,  $DF = 1$ ,  $P = 0.006$ ) relative to the livestock values. Argali values ( $\delta^{13}\text{C} = -21.1 \pm 0.4$  ‰,  $\delta^{15}\text{N} = 8.7 \pm 0.5$  ‰) were more depleted and showed less variation than ibex ( $\delta^{13}\text{C} = -20.9 \pm 0.9$  ‰,  $\delta^{15}\text{N} = 9.3 \pm 0.8$  ‰) (Table 3).

$^{15}\text{N} = 9.3 \pm 0.8$  ‰) (Table 3).

Mean domestic goat ( $\delta^{13}\text{C} = -18.9 \pm 2.2$  ‰,  $\delta^{15}\text{N} = 10.6 \pm 1.2$  ‰) and sheep ( $\delta^{13}\text{C} = -19.3 \pm 1.4$  ‰,  $\delta^{15}\text{N} = 10.6 \pm 0.4$  ‰) values were very similar, but goat values showed much more variation than sheep (Table 3). Mean domestic cow ( $\delta^{13}\text{C} = -18.0 \pm 1.1$  ‰,  $\delta^{15}\text{N} = 10.2 \pm 0.7$  ‰) and camel ( $\delta^{13}\text{C} = -18.3 \pm 1.7$  ‰,  $\delta^{15}\text{N} = 9.3 \pm 1.0$  ‰) numbers were slightly more enriched in  $^{13}\text{C}$  than sheep and

goats (Table 3). On average, horses were the most depleted in  $^{15}\text{N}$  of all ungulate species ( $\delta^{15}\text{N} = 8.1 \pm 0.7 \text{‰}$ ), although their  $\delta^{13}\text{C}$  values ( $\delta^{13}\text{C} = -18.8 \pm 1.1 \text{‰}$ ) were similar to those of goats and sheep (Table 3).

The values of the small mammal species showed a high degree of variation (Fig. 2; Table 4). The northern three-toed jerboa (*Dipus sagitta*) was the most enriched in  $^{13}\text{C}$  of all mammals sampled (Table 2), while Pallas's pika (*Ochotona pallasii*) was the most depleted in  $^{13}\text{C}$  of all mammals. The Mongolian gerbil (*Meriones unguiculatus*) was the most enriched and Pallas's pika was the most depleted in  $^{15}\text{N}$  of all mammals sampled (Fig. 2; Table 4).

On average, wolves were more enriched in  $^{15}\text{N}$  than any of the ungulate species ( $\delta^{15}\text{N} = 11.0 \pm 1.9 \text{‰}$ ), but values were not as high as would be predicted given the trophic enrichment reported in other wolf diet studies (Szepanski *et al.*, 1999; Khozu *et al.*, 2009; Urton & Hobson, 2005; Derbridge *et al.*, 2012). Average wolf  $\delta^{13}\text{C}$  values ( $\delta^{13}\text{C} = -19.8 \pm 1.5 \text{‰}$ ) were, on average, higher than livestock and lower than wild ungulates (Fig. 2; Table 3).

### Discussion

Stable isotope analysis can provide insight into the movement of elements across trophic levels and thus serve as a tool for understanding wildlife behavior and ecology (Gannes *et al.*, 1998; Dawson *et al.*, 2002; Fry, 2006; Ben-David & Flaherty, 2012). However, inferences about the movement of isotopes are limited by information on the spatial distribution of isotopes in a landscape. Our study obtained isotope signatures from several trophic levels (autotrophs, herbivores, carnivores) from Ikh Nart. These values may be combined with those from other isotope studies in Mongolia to reveal broader trends in isotope distribution, helpful in improving the accuracy and precision of wildlife-focused SIA studies in the future.

Previous plant and mammal isotope studies have been limited to areas in north/central Mongolia. Plant studies have explored variation of plant isotopic ratios due to elevation (Khozu *et al.*, 2009; Biazrov, 2012; Casper *et al.*, 2012), difference in water sources (Li *et al.*, 2007), and desertification from vehicle travel (Li *et al.*, 2006). Mammal studies have focused on the relationship between plant and mammal values within food

webs. Khozu *et al.* (2009) described the variation in isotope signatures across trophic levels at two study sites (Gachuurt, Ulaanbaatar and Khangai, Arkhangai *aimag*). Makarcwicz & Tuross (2006) also explored the difference in livestock and wild ungulate isotopes due to diet. Thompson *et al.* (2010) focused on the variation in human isotope values across Asia, including central Mongolia.

Overall, Ikh Nart plants were more enriched in  $^{13}\text{C}$  and  $^{15}\text{N}$  than the mean values reported from study areas at Gachuurt ( $\delta^{13}\text{C} \approx -26.5 \text{‰}$ ,  $\delta^{15}\text{N} = 0.5 \text{‰}$ ) and Khangai ( $\delta^{13}\text{C} \approx -26.5 \text{‰}$ ,  $\delta^{15}\text{N} = 0.0 \text{‰}$ ) (Khozu *et al.*, 2009). Ikh Nart signatures were similar to those at Baga Gazaryn Chuluu, Dundgovi *aimag* (Makarcwicz & Tuross, 2006), although the range of Ikh Nart nitrogen signatures was smaller than those reported at Baga Gazaryn Chuluu ( $\delta^{15}\text{N} = -0.8$  to  $17.5 \text{‰}$ ,  $\delta^{13}\text{C} = -28.0$  to  $-11.2 \text{‰}$ ) (Makarcwicz & Tuross, 2006). Variation in plant values for carbon and nitrogen isotopes are generally related to the abundance of  $\text{C}_4$  plants present in the ecosystem, water availability, and variation in water use efficiency within and between species in different soil moisture conditions (Chen *et al.*, 2007). The similarity in values between Ikh Nart and Baga Gazaryn Chuluu is probably due to the similarity of the habitat type. The lower values reported by Khozu *et al.* (2009) were from less arid habitats.

Our ungulate isotope numbers formed three distinct clusters consisting of wild ungulates (ibex and argali), non-equid livestock, and horses. A similar trend was reported in isotope values in the dentin collagen of argali, ibex, and domestic sheep and goats at Baga Gazaryn Chuluu, with significant differences in both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  between wild ungulates and domestic sheep and goats (Makarcwicz & Tuross, 2006). Mean argali and ibex values were  $-19.2\text{‰}$  and  $-19.5\text{‰}$  and  $8.8\text{‰}$  and  $8.1\text{‰}$  for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively, while mean domestic goat and sheep values were  $-18.2\text{‰}$  and  $-17.5\text{‰}$  and  $9.8\text{‰}$  and  $11.2\text{‰}$  for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively (Makarcwicz & Tuross, 2006). Makarcwicz and Tuross (2006) attributed the differences in wild ungulate and domestic sheep and goat values to human manipulation of livestock diet, particularly the provisioning of supplemental hay to domestic sheep and goats during the winter. Supplemental hay sampled included a high proportion of  $\text{C}_4$  plants (annuals not available to wild ungulates during winter months) resulting in  $^{13}\text{C}$  enriched

numbers for domestic livestock (Makarowicz & Tuross, 2006). Previous work at Ikh Nart, using fecal analysis to assess ungulate diet, found a high degree of overlap between domestic sheep and goats and argali in both summer (72%) and winter (95%) (Wingard *et al.*, 2011). Livestock at Ikh Nart are provided supplemental feed during periods of severe weather or increased energy requirements (e.g. spring lambing and kidding). Further research will be necessary to fully explore the influence of food and water sources on the differences in wild ungulate and livestock  $\delta^{13}\text{C}$  values.

Previous studies have found that increased human and livestock density can lead to  $^{15}\text{N}$  enrichment, through the increased presence of nitrogenous wastes in water and soil (Hyodo *et al.*, 2012). Sheep and goats in Ikh Nart live in large herds (mean herd size =  $519 \pm 278$  individuals; Davie *et al.*, 2014) that are generally penned during the winter, and frequently graze within a one-kilometer radius of herder *ger* camp sites year round. Both penning and frequent grazing near human habitation could contribute to enriched  $^{15}\text{N}$  in sheep and goats relative to wild ungulates. Domestic horses and camels tend to travel and graze at a greater distance from human habitation, which could explain the more depleted  $^{15}\text{N}$  values of the camels and horses in our study.

We are unaware of any reported isotope values for small mammals in Mongolia, making it difficult to interpret our results. However, it is possible to consider our values relative to existing information on the diets of Ikh Nart species and related species in other parts of the world. Long-eared hedgehogs (*Hemiechinus auritus*) and Daurian hedgehogs (*Mesechinus dauuricus*) primarily feed on insects, but will also consume lizards, birds (and eggs), and small mammals (Murdoch *et al.*, 2006; Batdorj, 2012). A carnivorous diet would be expected to place hedgehogs at a higher trophic level than small mammals feeding on seeds and vegetative plant parts, due to enriched nitrogen values. Our hedgehog values were enriched in  $^{15}\text{N}$ , but not as much as some herbivorous species (desert hamster and Mongolian gerbil) (Fig. 2; Table 4).

Hamsters, gerbils, and voles feed on seeds and vegetative plant parts (Randall, 1994; Liu *et al.*, 2013). Mice and jerboas have similar diets, but may feed on insects as well (Randall, 1994; Shiels *et al.*, 2013). Our hamster, jerboa, gerbil, and mouse values were all enriched in  $^{13}\text{C}$  and  $^{15}\text{N}$

relative to hares, pika, and marmot. This may be due to a greater amount of  $\text{C}_4$  plants in their diet, frequent foraging around human *ger* camps (staple human foods are often based on  $\text{C}_4$  plants and therefore enriched in  $^{13}\text{C}$ ), or extensive feeding on the millet seeds (a  $\text{C}_4$  plant) used to bait the box traps during capture. The home ranges of the smaller mammals would be more limited than that of livestock, which could result in reduced access to water (associated with enriched  $^{15}\text{N}$ ) (Murphy & Bowman, 2006; Newsome *et al.*, 2010).

Hares and marmots feed on grasses and forbs, although studies in North America have found strong preferences for forbs over grass in both species (Daniel *et al.*, 1993; Nagy, 1994; Stallman & Holmes, 2002), possibly due to the higher water content of forbs (Stallman & Holmes, 2002). Studies of pika diet have found strong preference for  $\text{C}_3$  plants (Ge *et al.*, 2013), which is consistent with the depleted  $^{13}\text{C}$  of our pika sample. Pika in India and in North America preferred forage rich in nitrogen and with a high moisture content (Dearing, 1996; Bhaltacharyya *et al.*, 2013). The low  $\delta^{15}\text{N}$  of hares, marmots, and pika may reflect preferential foraging on plants with high moisture content and low  $\delta^{15}\text{N}$  values, or the foraging location, as plant  $\delta^{15}\text{N}$  values vary depending on the nitrogen content of the soil on which they grow (Gannes *et al.*, 1998; Murphy & Bowman, 2006).

Wolves from Ikh Nart were enriched in both carbon and nitrogen relative to mean wolf values reported by Kohzu *et al.* (2009) for Mongolia (Table 3;  $\delta^{13}\text{C} \approx -21 \text{‰}$ ,  $\delta^{15}\text{N} \approx 8.0 \text{‰}$ ). This is consistent with our more enriched ungulate and plant samples. The trophic enrichment of nitrogen from wild ungulates and horses relative to wolves was consistent with the enrichment values described for earlier studies of 2 to 5‰ (DeNiro & Epstein, 1981; Urton & Hobson, 2005). The nitrogen values of sheep and goats relative to wolves, and the carbon values of domestic prey relative to wolves, did not show the enrichments generally predicted in other wolf diet studies (Roth & Hobson, 2000; Urton & Hobson, 2005; Kohzu *et al.*, 2009). Further research on the isotope signatures of differing trophic levels would help clarify this discrepancy.

Our study represents an initial sketch of the isotopic landscape of Ikh Nart and contributes to a broader understanding of isotopic variability



across trophic levels in Mongolia. Variation in ecozone and habitat type both appear to influence general trends in carbon and nitrogen values. Further studies, with larger samples sizes and more varied sampling points, will be necessary to more accurately describe trends in carbon and nitrogen isotopes values in Ikh Nart, and across Mongolia. However, stable isotope analysis remains a powerful method that can provide valuable information on the structure, flow, and movement of resources among wildlife populations.

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