

EVALUATING NUTRITIONAL CONDITION OF GRIZZLY BEARS VIA $\delta^{15}\text{N}$ SIGNATURES AND INSULIN-LIKE GROWTH FACTOR-1

ROBERT J. GAU, Wildlife and Fisheries Division, Department of Resources, Wildlife and Economic Development, Government of the Northwest Territories, #600 5102-50th Avenue, Yellowknife, NT X1A 3S8, Canada, email: rob_gau@gov.nt.ca
RAY CASE, Wildlife and Fisheries Division, Department of Resources, Wildlife, and Economic Development, Government of the Northwest Territories, #600 5102-50th Avenue, Yellowknife, NT X1A 3S8, Canada, email: ray_case@gov.nt.ca

Abstract: Changes in stable nitrogen isotope ($\delta^{15}\text{N}$) values in animal tissues may be an indicator of lean-tissue losses and thus reflect changes in nutritional condition. Adequate nutrient intake may be reflected by insulin-like growth factor (IGF-1) synthesis and secretion into the systemic circulation and thus also reflect changes in nutritional condition. However, the use of $\delta^{15}\text{N}$ and IGF-1 to estimate the nutritional condition of bears has yet to be validated with actual body compositions. We used bioelectrical impedance analysis (BIA) to estimate the body composition of a free-ranging population of grizzly bears (*Ursus arctos*) from the central Arctic of the Northwest Territories, Canada, 1995–96. We correlated serum IGF-1 concentrations to the percent of total body fat determined by BIA and their serum and erythrocyte $\delta^{15}\text{N}$ levels to lean body mass determined by BIA. Neither $\delta^{15}\text{N}$ nor IGF-1 had a significant relation with body composition. Ideas and problems for further research are presented and discussed.

Ursus 13:285–291(2002)

Key words: BIA, bioelectrical impedance analysis, body composition, body condition, grizzly bear, IGF-1, insulin-like growth factor, nutritional condition, stable nitrogen isotopes, *Ursus arctos*

Changes in the stable nitrogen isotope ($\delta^{15}\text{N}$) composition of animal tissues may indicate changes in the body condition of wild animals (Hobson et al. 1993, Gannes et al. 1997). Additionally, the level of insulin-like growth factor type-1 (IGF-1) hormone in serum has been used to estimate body condition in many domestic animals (Clemmons and Underwood 1991). Thus far, however, neither $\delta^{15}\text{N}$ nor IGF-1 has been reported in the literature as methods for determining body condition for bears. In fact, no indirect determination of body condition for bears has yet been validated via body composition (Cattet 1990, Farley and Robbins 1994, Gau and Case 1999).

Ratios of stable isotopes of nitrogen in animal tissues are indicative of the trophic level of an animal's diet (see reviews: DeNiro and Epstein 1981, Peterson and Fry 1987, Hobson 1999, Kelly 2000). However, increases of $\delta^{15}\text{N}$ levels in animal tissues of some species also result from nutritional or water stress and lean-tissue losses (Ambrose and DeNiro 1986, 1987; Hobson and Clark 1992; Hobson et al. 1993; Ponsard and Averbuch 1999). Recently, interest has increased in using $\delta^{15}\text{N}$ as a measure of body condition in free-ranging animals (Gannes et al. 1997, Ben-David et al. 1999, Kelly 2000).

Synthesis of the hormone IGF-1 and its secretion into the systemic circulation serves as an intermediate signaling mechanism that indicates to target cells that adequate nutrients have been ingested and are available (Clemmons and Underwood 1991). Subsequent research has successfully used IGF-1 levels to estimate the nutritional status of humans, rats, pigs, dogs, horses, and ruminants (Clemmons et al. 1981, Maes et al. 1983, Eigenmann et al. 1985, Breier et al. 1986, Hammond et al. 1990, McGuire et al. 1992, Webster et al. 1996, Rabkin 1997). However, the examination of IGF-1 levels in free-ranging wildlife has been limited (Adamczewski et al. 1992, 1998).

Bioelectrical impedance analysis (BIA) is a quick and noninvasive method in which an organism's resistance to conduction of a low-level alternating current is measured (Kushner 1992, Gales et al. 1994). Because the conductivity of body lipids is 4–5% that of lean tissue, body fluids, and bone, the body's electrical resistance is an indicator of total body water content (Farley and Robbins 1994). Knowing the body water content and mass of animals allows for the calculation of body lipid content due to the constancy of the composition of the fat-free mass (Johnson and Farrell 1988, Robbins 1993). Farley and Robbins (1994) developed the protocols and equations for use of snout-to-tail bioelectrical impedance analysis on bears. They concluded that BIA is a useful, reliable, and accurate method for estimating body composition. Recently, Hilderbrand et al. (1998) validated these conclusions in a single-blind study.

The objective of our study was to ascertain whether $\delta^{15}\text{N}$ and IGF-1 levels in blood reflect nutritionally stressful periods in a free-ranging population of grizzly bears. Thus, we determined the above parameters from a population of grizzly bears from the central Arctic of the Northwest Territories (NWT), Canada, and tested for correlations with accurate estimates of bear body composition.

STUDY AREA

Data were collected within an approximate 40,000 km² area of tundra in the central Arctic of the NWT. Research centered on the Government of the NWT Daring Lake Research Station (64°52'N, 111°37'W), located approximately 300 km northeast of Yellowknife, NWT, Canada. This region was a rocky upland area of the Canadian Precambrian Shield in which glaciation has resulted in patches of exposed bedrock and boulder fields, shallow rivers and

stream areas, numerous lakes, and glaciofluvial features like eskers (Mueller 1995). Vegetation included Arctic willow (*Salix* sp.), dwarf birch (*Betula glandulosa*), sedges (*Carex* sp.), and various grass and berry species. Most of the study area was treeless, although isolated stands of trees were found along its western boundary.

METHODS

In 1995 and 1996, we tranquilized adult male and lone female grizzly bears from a Bell 206B helicopter using a combination of zolazepam hydrochloride and telamine hydrochloride (Telazol®, Ayerst Laboratories, Montreal, Quebec, Canada) from a projected dart. Females with cubs were avoided to minimize the risk of cub abandonment. We determined body lipid content and lean body mass using a BIA Model 101A meter (RJL Systems, Detroit, Michigan, USA) and following the snout-to-tail protocol described by Farley and Robbins (1994) and Atkinson and Ramsay (1995). We placed immobilized bears on a plastic tarpaulin (to prevent loss of electrical current through direct ground contact) in a sternally recumbent position with the back legs fully extended, forelegs extended parallel to the length of the body, and the head straight and flush on the tarp between the forelegs. Lipids are a diverse class of compounds that include fats and oils; however, we assumed the total body lipid content to represent a bear's total body fat. Farley and Robbins (1994) reported the error in determining the total body fat by BIA for grizzly bears to be $\pm 2.2\%$. Also, even though $\delta^{15}\text{N}$ levels have a reported correlation with lean tissue loss, we used total lean mass (i.e. lean tissue plus the skeletal system) as an index of lean tissue. Total lean body mass is inherently correlated with lean tissue mass, and the former has been reported in other studies (Atkinson et al. 1996, Hilderbrand et al. 2000).

We collected blood from all captured bears from either the femoral artery or jugular vein. Samples were collected in 10 ml vacutainer serum separating tubes (Becton-Dickinson, Rutherford, New Jersey, USA). At the end of each day, samples were centrifuged, and serum was extracted and stored at -20°C .

Serum samples were assayed for IGF-1 by the Animal Biotechnology Centre, University of Saskatchewan (Saskatoon, Saskatchewan, Canada). Assays were performed following an acid-ethanol extraction (Breier et al. 1991) using a double-antibody radioimmunoassay developed for ovine and bovine serum by Kerr et al. (1990). Values for serial dilutions of a concentrated grizzly bear sample were parallel to the standard curve. All samples were analyzed in a single assay with an intra-assay CV of 4.0%.

The serum components of blood in bears have a protein

turnover rate over 4–10 days and in erythrocytes at least 40 days (Hilderbrand et al. 1996). Thus, the $\delta^{15}\text{N}$ levels in sera and the cellular fraction of blood may reflect the nutritional status of the animal for these 2 periods. Frozen serum and erythrocyte samples for $\delta^{15}\text{N}$ analysis were freeze-dried (FTS Systems Inc., Stone Ridge, New York, USA) for 60 hours, the time required to reach constant weight. Samples were then powdered with a mortar and pestle. A 1.0-mg portion of each powdered sample was weighed in a tin sampling capsule and assayed in a mass spectrometer (Europa Scientific 20/20, Crewe, United Kingdom) at the University of Saskatchewan Stable Isotope Facility in the Department of Soil Science. The natural abundance of the heavy-to-light stable nitrogen isotopes were reported in δ notation as parts per thousand (‰) deviations from a standard reference material according to the following equation:

$$\delta^{15}\text{N} = [(R_{\text{sample}} / R_{\text{standard}}) - 1] \times 1000$$

where R is the corresponding ratio $^{15}\text{N}/^{14}\text{N}$. The standard reference material used was atmospheric N_2 (air).

Statistical analysis followed Messier et al. (1987) and Gau and Case (1999), who previously correlated blood parameters to body compositions. Sex effects were assessed by 2-sample *t*-tests, and samples that were not significantly different were pooled. Relations between $\delta^{15}\text{N}$ and lean mass, and IGF-1 and the level of body fat, were evaluated via Spearman's rank correlation coefficients. Spearman's rank correlation is a robust test because no specific model (linear, curvilinear) of relation is assumed and variances can differ among values. Testing for relations between $\delta^{15}\text{N}$ and lean mass in bears was only considered when their total body fat levels were $\leq 10\%$. We assumed bears in extremely poor condition were more likely to catabolize lean tissue stores (i.e. bears with 10% total body fat may only have 5% usable fat in reserves assuming that cellular and structural components of adipose tissue account for 5% of total body fat levels). Statistical analyses were conducted with a statistical package for the social sciences (Norusis 1993). Means are presented with standard error and values of $P < 0.05$ were considered significant.

RESULTS

We captured 23 bears >3 years of age a total of 47 times between May and September 1995 and 1996. The only parameter to differ between sexes was serum IGF-1 ($t = 3.93$, 37 df, $P < 0.001$). The mean IGF-1 values for the male ($n = 19$) and female ($n = 20$) grizzly bears we sampled were 318.5 ng/ml (SE = 40.3) and 150.8 ng/ml (SE = 72.6), respectively. Sex effects on IGF-1 concentrations have

been previously observed (Davis et al. 1995). We found no significant difference in total lean body mass of males and females that had total body fat levels $\leq 10\%$ ($t = 1.3$, $n = 15$, $P = 0.22$). Thus, serum and erythrocyte isotope values were pooled across sexes for further analyses.

The mean serum and erythrocyte stable-nitrogen isotope values for the grizzly bears with $\leq 10\%$ total body fat we sampled were 7.7‰ (SE = 0.21; $n = 14$) and 6.6‰ (SE = 0.21; $n = 15$), respectively. Neither $\delta^{15}\text{N}$ (serum: $r_s = -0.03$, $n = 14$, $P = 0.91$; erythrocyte: $r_s = 0.19$, $n = 15$, $P = 0.50$), nor IGF-1 (males: $r_s = 0.29$, $n = 16$, $P = 0.27$; females: $r_s = -0.07$, $n = 20$, $P = 0.78$) were significantly correlated with body composition values (Figs. 1 and 2).

DISCUSSION

This study is among the first to compare quantitative body composition measurements of free-ranging grizzly bears to their $\delta^{15}\text{N}$ or IGF-1 levels. Although we did not find any significant correlations in the parameters we examined, there is inherent value our investigation. Our review has led to ideas for future research and illuminated some of the problems future researchers may need to overcome.

Insulin-like Growth Factor Type-1

Serum IGF-1 has been successfully used as an index of nutritional status for many species because higher concentrations indicate a better nutritional plane (Breier et al. 1986, Clemmons and Underwood 1991, McGuire et al. 1992). However, researchers have also acknowledged that a number of physiological or environmental factors can affect serum IGF-1 levels. Thus, despite the promise IGF-1 has in nutritional research for free-ranging animals, a number of concerns should be further explored in future research.

Synthesis of IGF-1 can be a response to injuries in cartilage, muscle, or arteries (Clemmons and Underwood 1991). Ambient temperature and photoperiod have also been suggested as factors influencing IGF-1 concentrations (Richards et al. 1995, Dahl et al. 1997, Webster et al. 1999). However, most researchers agree that the dietary intake of both protein and energy are among the most important factors in the stimulation of IGF-1 (Nap and Hazewinkel 1994, Thissen et al. 1994, Breier 1999).

Restrictive diet and refeeding studies have elicited IGF-1 fluctuations in various captive animals (Clemmons and Underwood 1991, Thissen et al. 1994). McGuire et al. (1995) demonstrated 50% reduction in IGF-1 levels after 2 days of feeding restrictions. Thus, the range of IGF-1 values we observed may have reflected recent feeding patterns and not overall body condition. Also, studies investigating IGF-1 levels often fail to acknowledge that

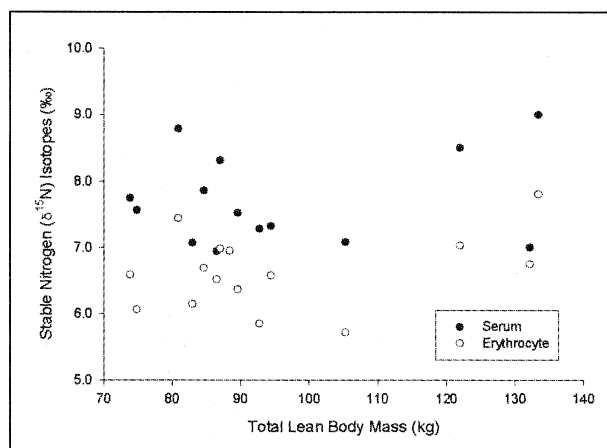


Fig. 1. Stable nitrogen isotope values for grizzly bear serum and erythrocytes versus total lean body mass for bears with $\leq 10\%$ total body fat as determined by bioelectrical impedance analysis for adult males and lone female grizzly bears from the central Arctic of the Northwest Territories, Canada, 1995–96.

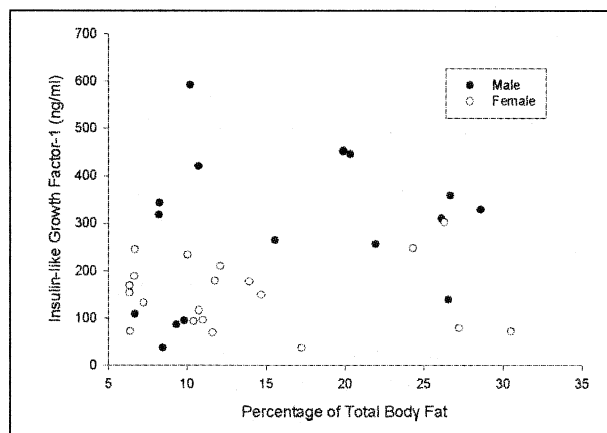


Fig. 2. Serum insulin-like growth factor-1 and the percent of total body fat as determined by bioelectrical impedance analysis for adult male and lone female grizzly bears from the central Arctic of the Northwest Territories, Canada, 1995–96.

IGF-1 concentrations generally have wide confidence limits in normal individuals, making it difficult to identify nutritional status (Thissen et al. 1994). Nutrient intake must often be decreased to a critical threshold before IGF-1 in serum declines. Above this threshold serum IGF-1 concentrations can remain normal over a broad intake of nutrients (Thissen et al. 1994).

Previous attempts to correlate IGF-1 to indices of body composition in humans have met with mixed results. Crist and Hill (1990) found a relation in females between the ratio of fat-free mass/fat mass and serum IGF-1 but no relation to the percent of body fat and serum IGF-1. Copeland et al. (1990) found an inverse relationship between serum IGF-1 and an index of body fat in obese male but not obese female subjects. Rudman et al. (1981)

found plasma somatomedin (IGF) levels tended to fall with an increasing index of adiposity for any given age in male and female subjects. However, the results for human subjects may not be comparable to our results. Attempts to correlate IGF-1 to indices of body composition in humans are usually done with obese subjects (Thissen et al. 1994). Obesity can be a chronic clinical condition for humans but is not prevalent in free-ranging grizzly bears for an extended period of time (Nelson et al. 1983).

There are discrepancies in the literature regarding how rapidly IGF-1 normalization occurs after malnourishment (Thissen et al. 1994). While some studies have reported depressed IGF-1 levels restored to normal on refeeding in 7–14 days (Clemmons and Underwood 1991, Tovar et al. 1999), IGF-1 recovery periods up to 29 days have also been reported (Thissen et al. 1994). For ursids, further research should be directed toward feeding trials on captive bears to determine baseline IGF-1 levels and suppression and normalization times to assess the viability of using IGF-1 to measure the nutritional status of free-ranging bears.

Stable Nitrogen Isotopes

Although interest has recently increased in exploring the viability of using $\delta^{15}\text{N}$ as a measure of body condition in free-ranging animals (Gannes et al. 1997, Kelly 2000), and promise has been shown for some species (Ambrose and DeNiro 1986, 1987; Hobson and Clark 1992; Hobson et al. 1993; Ponsard and Averbuch 1999), bears may present a problematic challenge.

The lack of relation between $\delta^{15}\text{N}$ and lean tissue levels we observed may suggest no protein catabolism had occurred in the animals we sampled. In fact, although some research on black bears (*U. americanus*) and polar bears (*U. maritimus*) has indicated bears may catabolize their body protein during a fast (Nelson et al. 1975, Atkinson et al. 1996), ursids in general may have the ability to adopt a protein-sparing state during food shortages (Nelson 1987, Atkinson et al. 1996). Thus as protein catabolism is conservative in bears, $\delta^{15}\text{N}$ may not be a reliable indicator of their nutritional status. Similar negative findings were also reported for Arctic ground squirrels (*Spermophilus parryii*; Ben-David et al. 1999).

Blood was chosen for analysis because it can be easily obtained from live animals and separated into serum and cellular components, each having its own particular protein turnover rate. Results can therefore be interpreted for 2 different periods from the same blood sample. We found no significant relation between serum $\delta^{15}\text{N}$ and the total lean body mass of grizzly bears. However, because the blood sera turnover rate is 4–10 days, $\delta^{15}\text{N}$ levels may be too sensitive to indicate changes in body condition for free-ranging bears. Although erythrocytes turnover after

40 days, they also did not indicate nutritional condition in the grizzly bears we sampled. Other body tissues have various turnover rates and further research may determine a tissue type more suitable for gauging the nutritional condition of free-ranging bears.

Stable isotope studies often investigate food webs with nitrogen signatures to differentiate gradients in marine, marine versus freshwater, or terrestrial versus marine ecosystems (Hobson 1999). However nitrogen isotope studies relying solely on the terrestrial ecosystem can have inherent problems because nitrogen isotope signatures vary geographically (Garten 1993, Kelly 2000) even over distances as small as a kilometer (Ponsard and Ardit 2000). This isotope variation becomes particularly confounding when interpreting nitrogen isotope results from northern ecosystems because the $\delta^{15}\text{N}$ signatures of primary producers (plants) can vary between and within species (Schulze et al. 1994, Nadelhoffer et al. 1996). In nutrient-deficient environments similar to our study area, intra- and inter-species differences for plants include variable root depths, various mycorrhizal root associations, moisture availabilities, rates of nitrification in the surrounding soil, and the utilization of different forms of nitrogen in the same soil horizon (Schulze et al. 1994, Chapin 1996, Michelsen et al. 1996, Nadelhoffer et al. 1996, Kielland et al. 1998, Michelsen et al. 1998). These factors can lead to significant differences in plant nutrition within the same ecosystem or habitat (Chapin et al. 1993, Kielland 1994). In our study area, for example, variability in primary producer $\delta^{15}\text{N}$ levels would thus lead to further $\delta^{15}\text{N}$ variability up the food web leading to barren-ground grizzly bears. Indeed, Gau (1998) found an 8.6‰ difference between various barren-ground grizzly bear plant foods sampled in the central Canadian Arctic. Griffith et al. (2000) also found similar results for some same sampled species in a study area contiguous to ours. Until future research can delineate the $\delta^{15}\text{N}$ deviations in the trophic levels below barren-ground grizzly bears, interpretations of their tissue's $\delta^{15}\text{N}$ levels will be confounded and spurious.

ACKNOWLEDGMENTS

Funding for our research was supplied by the Government of the NWT Department of Resources, Wildlife, and Economic Development; University of Saskatchewan; West Kitikmeot/Slave Society; Indian and Northern Affairs Canada; BHP Diamonds; Rescan Environmental Services; Diavik Diamond Project; Monopros Limited; Nunavut Wildlife Management Board; Northern Scientific Training Program; and the Polar Continental Shelf Project (no. 01298). Personal financial support to RJG was provided by the Science Institute of the Northwest Territories James M. Harrison Bursary and the Canadian

Wildlife Foundation Orville Erickson Memorial Scholarship. We gratefully acknowledge the field assistance and support provided by F. Messier, H.D. Cluff, P.D. McLoughlin, L. Buckland, and D. Olesen. Thank you to G. Perry, K. Hobson, and the late M. Ramsay for their assistance and guidance using stable isotopes and A. Lemke and G. I. Christison for their guidance using IGF-1. Earlier versions of this manuscript were improved by J. Roth and B. Laarveld.

LITERATURE CITED

- ADAMCZEWSKI, J.Z., P.J. FARGEY, B. LAARVELD, A. GUNN, AND P.F. FLOOD. 1998. The influence of fatness on the likelihood of early-winter pregnancy in muskoxen (*Ovibos moschatus*). *Theriogenology* 50:605–614.
- , A. GUNN, B. LAARVELD, AND P.F. FLOOD. 1992. Seasonal changes in weight, condition and nutrition of free-ranging and captive muskox females. *Rangifer* 12:179–183.
- AMBROSE, S.H., AND M.J. DENIRO. 1986. The isotopic ecology of East Africa mammals. *Oecologia* 69:395–406.
- , AND ———. 1987. Bone nitrogen isotope composition and climate. *Nature* 325:201.
- ATKINSON, S.N., R.A. NELSON, AND M.A. RAMSAY. 1996. Changes in the body composition of fasting polar bears (*Ursus maritimus*): the effect of relative fatness on protein conservation. *Physiological Zoology* 69:304–316.
- , AND M.A. RAMSAY. 1995. The effects of prolonged fasting on the body composition and reproductive success of female polar bears (*Ursus maritimus*). *Functional Ecology* 9:559–567.
- BEN-DAVID, M., C.J. MCCOLL, R. BOONSTRA, AND T.J. KARELS. 1999. ^{15}N signatures do not reflect body condition in Arctic ground squirrels. *Canadian Journal of Zoology* 77:1373–378.
- BREIER, B.H. 1999. Regulation of protein and energy metabolism by the somatotrophic axis. *Domestic Animal Endocrinology* 17:209–218.
- , J.J. BASS, J.H. BUTLER, AND P.D. GLUCKMAN. 1986. The somatotrophic axis in young steers: influence of nutritional status on pulsatile release of growth hormone and circulating concentrations of insulin-like growth factor 1. *Journal of Endocrinology* 111:209–215.
- , B.W. GALLAGHER, AND P.D. GLUCKMAN. 1991. Radioimmunoassay for insulin-like growth factor-1: solutions to some potential problems and pitfalls. *Journal of Endocrinology* 128:347–357.
- CATTET, M. 1990. Predicting nutritional condition in black bears and polar bears on the basis of morphological and physiological measurements. *Canadian Journal of Zoology* 68:32–39.
- CHAPIN, D.M. 1996. Nitrogen mineralization, nitrification, and denitrification in a high arctic lowland ecosystem, Devon Island, NWT, Canada. *Arctic and Alpine Research* 28:85–92.
- CHAPIN, F.S., III, L. MOILANEN, AND K. KIELLAND. 1993. Preferential use of organic nitrogen for growth by a nonmycorrhizal arctic sedge. *Nature* 361:150–153.
- CLEMMONS, D.R., A. KLIBANSKI, L.E. UNDERWOOD, J.W. MCARTHUR, E.C. RIDGEWAY, J.Z. BEITUS, AND J.J. VAN WYK. 1981. Reduction of plasma immunoreactive somatomedin C during fasting in humans. *Journal of Clinical Endocrinology and Metabolism* 53:1247–1250.
- , AND L.E. UNDERWOOD. 1991. Nutritional regulation of IGF-I and IGF binding proteins. *Annual Review of Nutrition* 11:393–412.
- COPELAND, K.C., R.B. COLLETTI, J.T. DEVLIN, AND T.L. MCAULIFFE. 1990. The relationship between insulin-like growth factor-I, adiposity, and aging. *Metabolism* 39:584–587.
- CRIST, D.M., AND J.M. HILL. 1990. Diet and insulinlike growth factor I in relation to body composition in women with exercise-induced hypothalamic amenorrhea. *Journal of the American College of Nutrition* 9:200–204.
- DAHL, G.E., T.H. ELSASSER, A.V. CAPUCO, R.A. ERDMAN, AND R.R. PETERS. 1997. Effects of a long daily photoperiod on milk yield and circulating concentrations of insulin-like growth factor-1. *Journal of Dairy Science* 80:2784–2789.
- DAVIS, M.E., M.D. BISHOP, N.H. PARK, AND R.C. SIMMEN. 1995. Divergent selection for blood serum insulin-like growth factor I concentration in beef cattle: I. Nongenetic effects. *Journal of Animal Science* 73:1927–1932.
- DENIRO, M.J., AND S. EPSTEIN. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochimica et Cosmochimica Acta* 45:341–351.
- EIGENMANN, J.E., J.J. DE BRUINE, AND E.R. FROESCH. 1985. Insulin-like growth factor I and growth hormone in canine starvation. *Acta Endocrinologica* 108:161–166.
- FARLEY, S.D., AND C.T. ROBBINS. 1994. Development of two methods to estimate body composition of bears. *Canadian Journal of Zoology* 72:220–226.
- GALES, R., D. RENOUF, AND G.A.J. WORTHY. 1994. Use of bioelectrical impedance analysis to assess body composition of seals. *Marine Mammal Science* 10:1–12.
- GANNES, L.Z., D.M. O'BRIEN, AND C. MARTÍNEZ DEL RIO. 1997. Stable isotopes in animal ecology: assumptions, caveats, and a call for more laboratory experiments. *Ecology* 78:1271–1276.
- GARTEN, C.T., JR. 1993. Variation in foliar ^{15}N abundance and the availability of soil nitrogen on Walker Branch watershed. *Ecology* 74:2098–2113.
- GAU, R.J. 1998. Food habits, body condition, and habitat of the barren-ground grizzly bear. Thesis, University of Saskatchewan, Saskatoon, Saskatchewan, Canada.
- , AND R. CASE. 1999. Evaluating nutritional condition of grizzly bears via select blood parameters. *Journal of Wildlife Management* 63:286–291.
- GRIFFITH, B., A. GUNN, D. RUSSELL, K. KIELLAND, AND S. WOLFE. 2000. Bathurst caribou calving ground studies: influence of nutrition and human activity on calving ground location. Final 1999 Annual Report to the West Kitikmeot Slave Study Society. University of Alaska-Fairbanks, Fairbanks, Alaska, USA.
- HAMMOND, A.C., T.H. ELSASSER, W.E. KUNKLE, T.S. RUMSEY, M.J. WILLIAMS, AND W.T. BUTTS. 1990. Effects of winter nutrition and summer pasture or a feedlot diet on plasma insulin-like growth factor I (IGF-I) and the relationship between

- circulating concentrations of IGF-I and thyroid hormones in steers. *Domestic Animal Endocrinology* 7:465–476.
- HILDERBRAND, G.V., S.D. FARLEY, AND C.T. ROBBINS. 1998. Predicting body condition of bears via two field methods. *Journal of Wildlife Management* 62:406–409.
- , ———, ———, T.A. HANLEY, K. TITUS, AND C. SERVHEEN. 1996. Use of stable isotopes to determine diets of living and extinct bears. *Canadian Journal of Zoology* 74:2080–2088.
- , C.C. SCHWARTZ, C.T. ROBBINS, AND T.A. HANLEY. 2000. Effect of hibernation and reproductive status on body mass and condition of coastal brown bears. *Journal of Wildlife Management* 64:178–183.
- HOBSON, K.A. 1999. Tracing origins and migration of wildlife using stable isotopes: a review. *Oecologia* 120:314–326.
- , R.T. ALISAUSKAS, AND R.G. CLARK. 1993. Stable-nitrogen isotope enrichment in avian tissues due to fasting and nutritional stress: implications for isotopic analysis of diet. *Condor* 95:388–394.
- , AND R.G. CLARK. 1992. Assessing avian diets using stable isotopes II: factors influencing diet-tissue fractionations. *Condor* 94:187–195.
- JOHNSON, R.J., AND F.D. FARRELL. 1988. The prediction of body composition in poultry by estimation in vivo of total body water with tritiated water and deuterium oxide. *British Journal of Nutrition* 59:109–124.
- KELLY, J.F. 2000. Stable isotopes of carbon and nitrogen in the study of avian and mammalian trophic ecology. *Canadian Journal of Zoology* 78:1–27.
- KERR, D.E., B. LAARVELD, AND J.G. MANNS. 1990. Effects of passive immunization of growing guinea-pigs with an insulin-like growth factor-1 monoclonal antibody. *Journal of Endocrinology* 124:403–415.
- KIELLAND, K. 1994. Amino acid absorption by arctic plants: implications for plant nutrition and nitrogen cycling. *Ecology* 75:2373–2383.
- , B. BARNETT, AND D. SCHELL. 1998. Intra-seasonal variation in the $\delta^{15}\text{N}$ signature of taiga trees and shrubs. *Canadian Journal of Forest Research* 28:485–488.
- KUSHNER, R.F. 1992. Bioelectrical impedance analysis: a review of principles and applications. *Journal of the American College of Nutrition* 11:199–209.
- MAES, M., L.E. UNDERWOOD, AND J.M. KETESLEGERS. 1983. Plasma somatomedin-C in fasted and refed rats: close relationship with changes in liver somatogenic but not lactogenic binding sites. *Journal of Endocrinology* 97:243–252.
- McGUIRE, M.A., D.E. BAUMAN, D.A. DWYER, AND W.S. COHICK. 1995. Nutritional modulation of the somatotropin insulin-like growth factor system: response to feed deprivation in lactating cows. *Journal of Nutrition* 125:493–502.
- , J.L. VICINI, D.E. BAUMAN, AND J.J. VEENHUIZEN. 1992. Insulin-like growth factors and binding proteins in ruminants and their nutritional regulation. *Journal of Animal Science* 70:2901–2910.
- MESSIER, F., J. HUOT, F. GOUDREAU, AND A.V. TREMBLAY. 1987. Reliability of blood parameters to assess the nutritional status of caribou. *Canadian Journal of Zoology* 65:2413–2416.
- MICHELSSEN, A., C. QUARMBY, D. SLEEP, AND S. JONASSON. 1998. Vascular plant ^{15}N natural abundance in heath and forest tundra ecosystems is closely correlated with presence and type of mycorrhizal fungi in roots. *Oecologia* 115:406–418.
- , I.K. SCHMIDT, S. JONASSON, C. QUARMBY, AND D. SLEEP. 1996. Leaf ^{15}N abundance of subarctic plants provides field evidence that ericoid, ectomycorrhizal and non- and arbuscular mycorrhizal species access different sources of soil nitrogen. *Oecologia* 105:53–63.
- MUELLER, F.P. 1995. Tundra esker systems and denning by grizzly bears, wolves, foxes, and ground squirrels in the central Arctic, Northwest Territories. File report no. 115. Renewable Resources, Government of the Northwest Territories, Yellowknife, Northwest Territories, Canada.
- NADELHOFFER, K.J., G. SHAVER, B. FRY, A. GIBLIN, L. JOHNSON, AND R. MCKANE. 1996. ^{15}N natural abundances and N use by tundra plants. *Oecologia* 107:386–394.
- NAP, R.C., AND H.A.W. HAZEWINKEL. 1994. Growth and skeletal development in the dog in relation to nutrition: a review. *Veterinary Quarterly* 16:50–59.
- NELSON, R.A. 1987. Black bears and polar bears—still metabolic marvels. *Mayo Clinic Proceedings* 62:850–853.
- , G.E. FOLK, E.W. PFEIFFER, J.J. CRAIGHEAD, C.J. JONKEL, AND D.L. STEIGER. 1983. Behavior, biochemistry, and hibernation in black, grizzly, and polar bears. *International Conference on Bear Research and Management* 5:284–290.
- , J.D. JONES, H.W. WAHNER, D.B. MCGILL, AND C.F. CODE. 1975. Nitrogen metabolism in summer starvation and in winter sleep and role of urinary bladder in water and nitrogen conservation. *Mayo Clinic Proceedings* 50:141–146.
- NORUSIS, M.J. 1993. SPSS for windows: base system user's guide, release 6.0. SPSS Inc., Chicago, Illinois, USA.
- PETERSON, B.J., AND B. FRY. 1987. Stable isotopes in ecosystem studies. *Annual Review of Ecology and Systematics* 18:293–320.
- PONSARD, S., AND R. ARDITI. 2000. What can stable isotopes ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) tell about the food web of soil macro-invertebrates? *Ecology* 81:852–864.
- , AND P. AVERBUCH. 1999. Should growing and adult animals fed on the same diet show different $\delta^{15}\text{N}$ values? *Rapid Communications in Mass Spectrometry* 13:1305–1310.
- RABKIN, R. 1997. Nutrient regulation of insulin-like growth factor-I. *Mineral and Electrolyte Metabolism* 23:157–160.
- RICHARDS, M.W., L.J. SPICER, AND R.P. WETTEMANN. 1995. Influence of diet and ambient temperature on bovine serum insulin-like growth factor-I and thyroxine: relationships with non-esterified fatty acids, glucose, insulin, luteinizing hormone and progesterone. *Animal Reproduction Science* 37:267–279.
- ROBBINS, C.T. 1993. *Wildlife feeding and nutrition*. Second edition. Academic Press, San Diego, California, USA.
- RUDMAN, D., M.H. KUTNER, C.M. ROGERS, M.F. LUBIN, G.A. FLEMING, AND R.P. BAIN. 1981. Impaired growth hormone secretion in the adult population: relation to age and adiposity. *Journal of Clinical Investigation* 67:1361–1369.
- SCHULZE, E.-D., F.S. CHAPIN, AND G. GEBAUER. 1994. Nitrogen nutrition and isotope differences among life forms at the northern treeline of Alaska. *Oecologia* 100:406–412.
- THISSEN, J.P., J.M. KETESLEGERS, AND L.E. UNDERWOOD. 1994. Nutritional regulation of the insulin-like growth factors.

Endocrine Reviews 15:80–101.

TOVAR, A.R., A. HALHALI, AND N. TORRES. 1999. Effect of nutritional rehabilitation of undernourished rats on serum insulin-like growth factor (IGF)-I and IGF-binding proteins. *Revista de Investigacion Clinica* 51:99–106.

WEBSTER, J.R., I.D. CORSON, R.P. LITTLEJOHN, S.K. STUART, AND J.M. SUTTIE. 1996. Effects of season and nutrition on growth hormone and insulin-like growth factor-I in male red deer. *Endocrinology* 137:698–704.

_____, _____, _____, _____, AND _____. 1999. Effects of photoperiod on the cessation of growth during autumn in male red deer and growth hormone and insulin-like growth factor-I secretion. *General and Comparative Endocrinology* 113:464–477.

Received: 24 February 2001.

Accepted: 25 February 2002.

Associate Editor: Hilderbrand.