Quantifying the free living energy exchanges of Arctic ungulates with stable isotopes

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Abstract: When natural diets meet an animal's requirement for energy, other essential nutrients will usually be supplied in amounts at least sufficient for survival. Knowledge of the energy requirements of free ranging species under typical conditions are important in assessing both their nutritional needs and their ecological impact. The doubly labelled water (DLW) method is currently the most promising objective field methodology for estimating free living energy expenditure but expenditure is only equal to the energy requirement when an animal is in energy balance. Reproduction and seasonal cycles of fat deposition and utilization represent significant components of the energy budget of arctic ungulates but the information gained in the course of a typical DLW study may be used to estimate processes such as milk output and fat storage and mobilization in order to predict requirements from expenditure.

The DLW method has been exhaustively validated under highly controlled conditions and the introduction of innovations such as faecal sampling for the estimation of body water isotopic enrichment, the availability of appropriate correction factors and stoichiometries for known sources of error, and iterative calculation of unknown parameters, have produced a methodology suitable for use in truly free ranging species. The few studies carried out so far in arctic ungulates indicate that previous predictions have generally underestimated the true level of expenditure, that there is considerable between animal variation in the level of expenditure and that this is largely determined by physical activity.

The disadvantages of the DLW methodology are that it remains expensive and the isotope analysis is technically demanding. Furthermore, although DLW can provide an accurate value for free living energy expenditure, it is often important to have information on the individual components of expenditure, for example the relative contribution of physical activity and thermoregulatory thermogenesis, in order to interpret the values for overall expenditure. For these reasons the most valuable use of the DLW method in the field may be to validate factorial models and other approaches so that they may be used with confidence. Additional important information on the energy exchanges of free ranging animals may be obtained from other stable isotope methodologies. In addition to the use of the isotopes ^2H and ^{18}O in the DLW method, natural variations in the abundance of ^{13}C and ^{15}N in the arctic environment may be exploited to study diet selection in truly free living arctic ungulates.

Introduction

When natural diets meet an animal's requirement for energy, other essential nutrients such as protein, fats minerals and vitamins will usually be supplied in amounts at least sufficient for survival (Kay *et al.,* 1984). Consequently, energy is often the most important single factor in the nutrition of an animal and knowledge of energy expenditure under typical free-living conditions are important in assessing both energy requirements and the ecological impact of free-ranging animals. Arctic ungulates such as reindeer and caribou (*Rangifer tarandus*) may range over large areas therefore traditional methods of estimating energy expenditure such as chamber calorimetry are of limited use as they impose a significant degree of restriction on 'normal' activity which makes up a significant portion of the energy budget of free ranging species. Furthermore, the harshness of the arctic environment also influences expenditure in ways which are difficult to model; these include the effects of temperature and wind chill, and the additional energy expenditure which occurs as a consequence of foraging when the vegetation may be of poor quality or where access is made difficult by snow cover. In an effort to make more realistic estimates of energy expenditute, a number of workers have developed factorial models where values for the energy costs of metabolic

processes, activities, and the time spent in those activities are used to calculate the overall energy budget of the animal (e.g. Boertje, 1985; Tyler, 1987). Such models have been invaluable in studying the energy exchanges of truly free living animals in their natural habitat but the assumptions underlying these models have yet to be tested by comparison with objective measures of expenditure. The most promising method of estimating energy expenditure in free-living animals is the doubly labelled water (DLW) technique (Lifson & McClintock, 1966). This technique, which utilizes water labeled with stable isotopes, provides an integrated value for energy expenditure, typically over 1 to 3 weeks, and therefore is ideal to estimate requirements.

Energy expenditure is only equal to the enetgy requirement when an animal is in energy balance yet in arctic species fat deposition can make up a large part of the energy requirement in summer whilst mobilisation of body fat can supplement the energy requirement in winter. Similarly, productive processes such as pregnancy and lactation represent a significant part of the energy requirement of arctic ungulates. Since the DLW method only provides an estimate of energy expenditure, information on the magnitude of these other processes is required in order to estimate the more practically useful parameter of the energy requirement. This paper reviews new insights into the energy exchanges of arctic ungulates provided by the use of stable isotope methodologies and proposes novel ways in which stable isotopes may be used to study the various components of the energy budget of arctic ungulates (Fig. 1). The results for arctic species (Eurasian mountain reindeer; *R. t. tarandus,* Alaska caribou; *R. t. granti* and muskoxen; *Ovibos moschatus*) are compared with a non-arctic species (red deer; *Cervus elaphus*).

Stable isotopes

Isotopes are atoms with the same number of protons (atomic number) but different numbers of neutrons (mass number). They may be classified as either radioactive or stable and isotopes are available for many of the elements found in living systems (e.g. hydrogen, carbon, oxygen, nitrogen, sulphur and
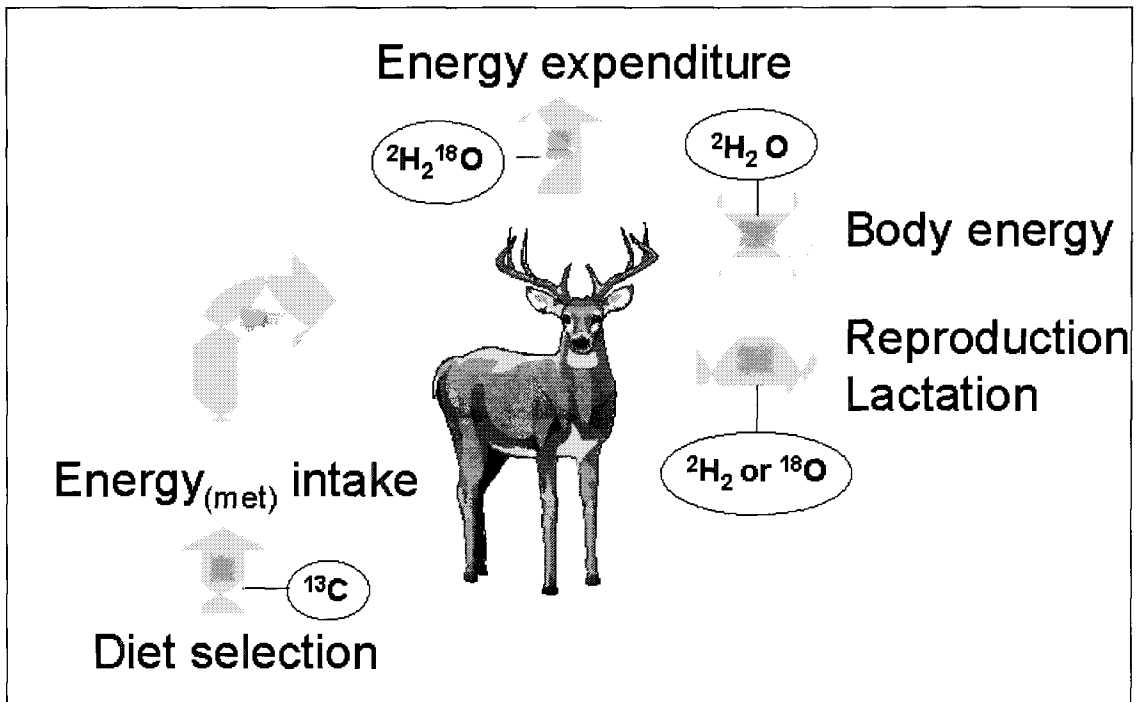

Fig. 1. The metabolisable (met) energy intake of an animal is equivalent to its requirement when this equals the energy expenditure plus the energy cost of reproduction/lactation, plus energy stored in the body or minus energy mobilised from body stores. All of these processes and the selection of diet can be measured in the field using a range of stable isotope methodologies.

many metals). The isotope concentration in biological samples is measured most precisely using gas isotope ratio mass spectrometry where the biological matrix is converted into a permanent gas (e.g. CO_2, H_2) prior to measurement. This conversion can be carried out 'off-line' or it can be automated and connected 'in-line' to standard laboratory techniques for isolation of individual molecules (e.g. gas chromatography-combustion-isotope ratio mass spectrometry). The isotopic results are presented as $\delta^{13}C_{PDB}$, the notation commonly used to report natural variations in ^{13}C abundance, or as parts per million (ppm) above background in tracer studies. Negative δ values indicate that the sample is depleted in ^{13}C relative to the standard (PDB: PeeDee Belemnite) and the numerical quantity expresses the depletion in parts per thousand or "per mil", designated by the symbol ‰.

In techniques such as the DLW method the only available tracer for oxygen is the stable isotope oxygen-18 (^{18}O) but the hydrogen of the body water could be labelled with either stable-isotopic deuterium (^2H) or radio-isotopic tritium (^3H). The latter has been used in the DLW validation studies of Fancy *et al.* (1986), presumably because ^3H is easier to measure using the widely available technique of scintillation counting. However, stable isotopes offer a number of advantages over radioactive isotopes: 1. Their use in free living animals studies does not result in the introduction of radioisotope contamination into the environment. 2. Stable isotopes can be measured more precisely than radioisotopes and in a comparison of water flux rate measurements in deer labelled with both ^2H_2O and ^3H_2O, the deuterium estimate of water flux was twice as precise as the tritium estimate (Haggarty *et al.*, 1998). Since the DLW method estimates CO_2 production from small differences between two isotopic flux rates which are dominated by water turnover, the improved precision offered by ^2H is an important consideration. 3. Stable isotopes are always present in the environment and natural variations in their concentration can be exploited to obtain information on the origin and inter-conversion of nutrients.

The doubly labelled water method

The DLW method is based on the observation that the hydrogen of body water is lost mainly as water whereas the oxygen of water is lost as water and CO_2. When the body water is labelled with ^2H and
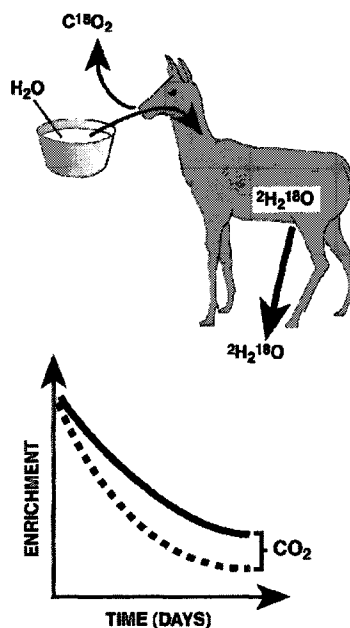

Fig. 2. Theoretical basis of estimating CO_2 production from the difference in washout rates of ^2H (—) and ^{18}O (····) from the body water.

^{18}O, the rate of dilution of ^2H gives a value for water flux and the rate of ^{18}O dilution gives a value for water flux plus CO_2 production and the production of CO_2 can be estimated from the difference between the two isotopic fluxes (Fig. 2).

This method (Lifson & McLintock, 1966) has now been successfully validated in ruminant (Fancy *et al.*, 1986; Midwood *et al.*, 1994) and monogastric (Haggarty *et al.*, 1994a) animals and may therefore be used with confidence in a wide variety of metabolic states if appropriate corrections are made for the processes which are known to introduce errors into the technique (Fancy *et al.*, 1986; Haggarty, 1991; Haggarty *et al.*, 1994a; Midwood *et al.*, 1989; 1993).

In order to calculate water flux and CO_2 production it is necessary to correct the isotope flux data for fractionated water loss (Haggarty *et al.*, 1988) and any change in pool size (Haggarty *et al.*, 1994a) during the course of the labelling period. There are, however, other processes which may introduce errors into the DLW estimate of CO_2 production; for example, loss of ^2H into products other than water will cause water flux to be overestimated and CO_2 production to be underestimated. This may occur during sequestration of ^2H into stable carbon-hydrogen bonds or by exchange of ^2H with labile positions on material which is subsequently export-
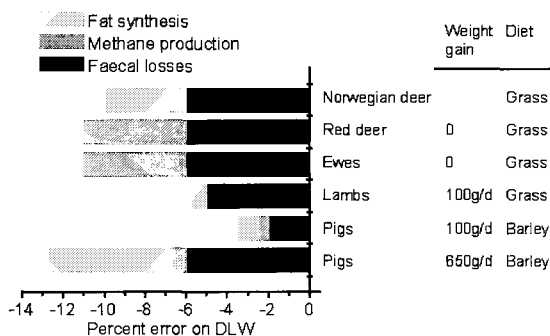
Fig. 3. The magnitude of errors which different seques-
tration/exchange processes introduce into the
DLW method vary between species and between
different physiological states.

ed from the body; faeces for example. The sequestra-
tion/exchange processes which have the potential to
introduce substantial errors into the DLW method
in production animals are fat synthesis, methane
production, export of faecal dry matter and lactation
(Haggarty, 1991; Midwood *et al.,* 1989; Haggarty *et
al.,* 1994a). The magnitude of these processes must
be estimated in order to make the necessary correc-
tions to the isotope flux data (Haggarty, 1991).
Some examples of actual effects (Fig. 3) illustrate
that there are wide variations in the relative impor-
tance of each error in different physiological states.
Also, the fact that all three processes result in an
underestimate of energy expenditure mean that the
errors are additive such that when they are com-
bined they can have a significant impact on the
accuracy of the DLW method. In addition, the
DLW method only provides an estimate for CO_2
production, therefore further information on oxygen
consumption, nitrogen loss and, in the case of rumi-
nants, methane production, is required to calculate
a value for energy expenditure. Under the highly
controlled conditions of a validation study it is pos-
sible to make direct measurements of faecal losses,
methane production and even fat synthesis, but the
need to carry out such measurements in a typical
DLW study would largely nullify the main advan-
tage of the DLW method which is that it can be
used in free-living animals in their natural environ-
ment. Although the DLW method was developed as
a method of estimating free living energy expendi-
ture, the necessity of correcting for these processes
and the typical DLW dosing and body water sam-
pling regime has severely restricted its use in truly
free-living animals. To overcome the first limita-

tion, an iterative method has been developed to cor-
rect for the above sources of error and provide esti-
mates of oxygen consumption, methane production
and nitrogen loss without the need to measure them
directly, using only the data which is typically
obtained in DLW studies in truly free-living ani-
mals (Haggarty *et al ,* 1998).

With respect to the second limitation, energy
expenditure may be calculated from DLW data
using the two-point or multi-point methods
(Haggarty, 1991) but the two-point approach has
the important advantage for studies in free-ranging
animals that the samples of body fluids (typically
blood) need only be taken on the first and last day of
the labelling period and not each day as with the
multi-point approach. Apart from the practical dis-
advantage of having to re-capture animals each day,
daily capture and sampling could, in itself, signifi-
cantly affect the energy expenditure of free-ranging
animals. In such animals, the two-point methodolo-
gy, where body fluids are sampled only at the begin-
ning and end of an experiment, typically lasting
between one and two weeks, would be a more feasi-
ble approach. A comparison of multi-point and
two-point calculations has shown that there is no
significant difference in the enetgy expenditure
derived by these approaches in ungulates (Haggarty
et al., 1998) therefore the two-point method may be
used with confidence. An ingenious way of further
reducing interference with the normal activity of
free ranging species during DLW measurements has
been proposed by Gotaas *et al.* (1997) who have
shown that faeces water can be used to estimate the
isotopic composition of the body watet. This
method offers the possibility of carrying out a DLW
study in free ranging ungulates requiring only one
capture in order to give the dose, the wash out of
isotope from the body can then be estimated by col-
lection of fresh faeces in the field and Gotaas *et al.*
(in press) have used this approach to estimate the
energy expenditure of free living reindeer.

Free living expenditure

Until the advent of objective methods such as DLW
the only way of estimating requirements under nor-
mal conditions was the factorial approach or, in a
compromise between truly free living and fully con-
fined, the measurement of food intake in penned
animals. It is therefore useful to put the few existing
DLW results in arctic ungulates in the context of
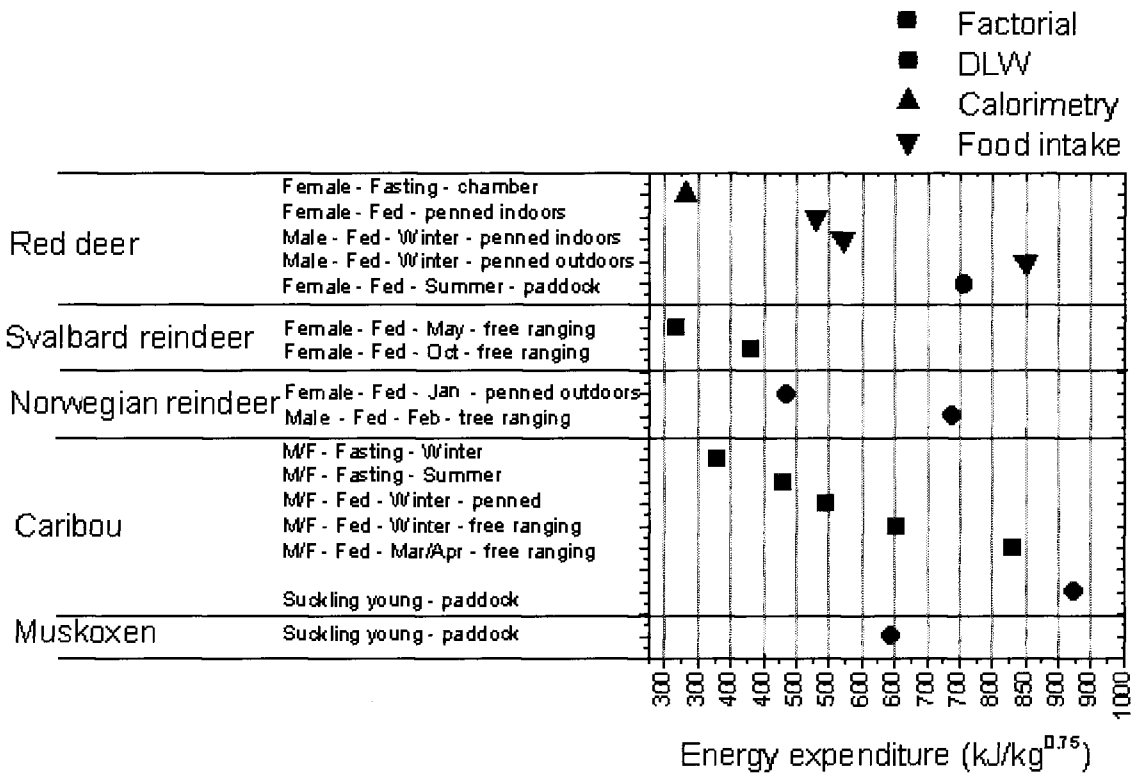data obtained using traditional methodologies.

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Fig. 4. Comparison of estimates of energy expenditure in three species of northern ungulates made using a range of methodologies: factorial, calorimetry, measured food intake and doubly labelled water (DLW).

To compare the metabolism of individuals within a species or to make between-species comparisons, the values for expenditure must be standardized in some way and the best method available is probably the Brody-Kleiber proportionality which suggests on empirical and theoretical grounds that expenditure should be expressed per $kg^{0.75}$ (Blaxter, 1989). When expressed in this way it can be seen from Fig. 4 that even within the factorial approach there is a very wide range in the estimated level of expenditure, both within and between species. Some of this variability will be due to differences between the studies in the sex of the animals, the season and the degree of restriction imposed but even after taking account of these effects the estimates are very variable. For example, factorial estimates of expenditure in free ranging Svalbard reindeer (*R. t. platyrhynchus*) in October (433 kJ/$kg^{0.75}$; Tyler, 1987) are considerably less than those for free ranging caribou in winter (652 kJ/$kg^{0.75}$; Boertje, 1985) and even studies within a species can be very variable with a second study in free living caribou in winter providing a value of (830 kJ/$kg^{0.75}$; Fancy, 1986).

Some factorial estimates have been very low with free ranging female Svalbard reindeer in May having an estimated expenditure (319 kJ/$kg^{0.75}$; Tyler, 1987) which is similar to the resting metabolic rate (standing) in captive animals (348 kJ/$kg^{0.75}$; Nilssen *et al.*, 1984). The relatively small number of DLW studies which have been carried out in large ungulates indicate that, whilst there are some exceptions, the DLW method generally provides higher values than have been derived from feeding or factorial studies. For example, the DLW derived expenditure of female red deer kept in a group in a field covering 3300 m² was 757 kJ/$kg^{0.75}$ during the summer (Haggarty *et al.*, 1998), a value that was higher than previous estimates of requirements with the only higher estimate coming from a feeding trial in stags penned outdoors in winter (850 kJ/$kg^{0.75}$; Fennessy *et al.*, 1980). A comparison of the outdoor feeding results with similar studies in animals indoors in the same season (570 kJ/$kg^{0.75}$; Kay *et al.*, 1984) suggest that fully one third of the outdoor requirements in winter in red deer are due to the effect of outdoor temperature. This finding is reinforced by
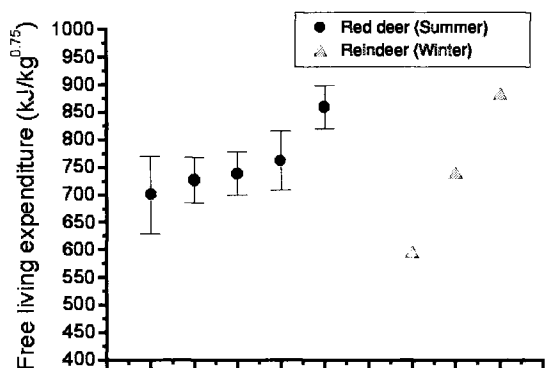
Fig. 5. Between animal variability in doubly labelled water (DLW) derived free-living energy expenditure for red deer (Haggarty *et al.*, 1998) and young male reindeer in winter (Gotaas *et al.*, in press).

the comparison of DLW derived free living energy expenditure in hinds in summer (757 kJ/kg$^{0.75}$) with the partially confined stags in winter (850 kJ/kg$^{0.75}$). Also included in Fig. 4 are some reports of DLW derived energy expenditure in caribou and muskoxen (Parker *et al.*, 1990) although the interpretation of these results is complicated by the fact that they were carried out during suckling.

Comparison of DLW results obtained in red deer with those from captive and free ranging reindeer are shown in Fig. 5. Given that the results for the red deer were obtained in summer where the thermal stress was minimal, whilst reindeer have a level of thermal insulation which protects them from the
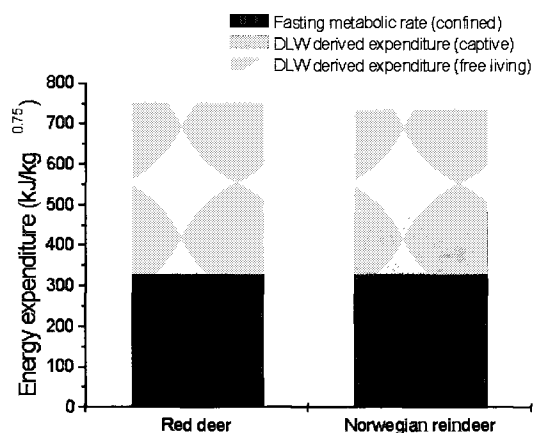

Fig. 6. Comparison of the components of free-living energy expenditure in ungulates from arctic (reindeer; Gotaas *et al.*, 1997; in press) and temperate (red deer; Haggarty *et al.*, 1998) regions.

effects of cold in winter (Tyler & Blix, 1990), the similarity in expenditure indicates that free living physical activity of both species is comparable since this will make up the largest proportion of heat production. This point is made clear in Fig. 6 where the DLW derived expenditure of free living reindeer (Gotaas *et al.*, in press) is compared with that of penned reindeer (Gotaas *et al.*, 1997). Whilst some of this difference may be explained by the fact that the free-living animals were male whilst the captive animals were female, it is striking that the non-fasting energy expenditure in the free living animals was up to 3 times that of the captive animals.

When a new method suggests that old assumptions may be in error it is natural to question the validity of the method. However, all the processes which could result in errors have been exhaustively described and, in most cases they result in an underestimate of expenditure (Haggarty, 1991; Haggarty *et al.*, 1994a; Midwood *et al.*, 1989; 1993; Gotaas *et al.*, 2000); the finding of generally higher than anticipated values for expenditure is therefore robust.

The DLW method may provide information in addition to the estimation of energy requirements of groups if a value for the precision of energy expenditure is available. For example, it can be seen that one of the red deer had a significantly higher level of expenditure compared to the rest of the group (Fig. 5). Since all the animals were exposed to the same climate and environment, this difference was presumably due to a higher physical activity, possibly reflecting the dominance of this hind within the social group. The equivalent values in free ranging reindeer shows that the mean level of expenditure was very similar to that in the red deer but that the range of values was much greater. No estimate of precision was available for these data but similar previous studies (Gotaas *et al.*, 1997) indicated a similar level of precision to that found in red deer, suggesting that in truly free ranging animals the energy expenditure difference between individuals is much larger than is found in farmed species. This is not only important with respect to the interactions between individuals within a herd but it also has implications for the number and type of individuals which have to be monitored (using DLW, the factorial method, or any other approach) in order to give a good estimate of the expenditure of the whole group. For example, Fancy & White (1985) have calculated that caribou breaking the trail at the head of the spring migration have an

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associated increment of minimal metabolism which is 2.5 times that in animals following the packed trail. Whilst the overall energetic effect of trail breaking would depend on the proportion of the day spent in this activity, such observations suggest that total energy expenditure measured by DLW could well be substantially different in these two groups of animals.

Cycles of fat deposition and mobilisation

Arctic ungulates such as Svalbard reindeer store very large amounts of fat during the short arctic summer and lose most of it during the arctic winter when there is no daylight and restricted access to forage (Tyler, 1987). Boertje (1985) has calculated that utilisation of body fat can provide up to 10% of the energy requirement of caribou in winter whilst Tyler (1987) suggested a figure of up to 25% in non-pregnant Svalbard reindeer. Conversely, Boertje (1985) has calculated that in summer the energy cost of fat deposition accounts for around 17% of the total requirement. Body composition has traditionally been estimated from a variety of techniques including dissection and chemical analysis (Ringberg et al., 1981; Adamczewski et al., 1987; 1995; Gerhart et al., 1995) but in order to derive values for energy requirements in the different seasons in truly free living animals it is necessary to evaluate the magnitude of these processes in the field, preferably without recourse to serial slaughter. This can be done isotopically by estimating the body water from isotope dilution where the body fat content is estimated using information on the hydration of non-fat tissues (e.g. Latsen & Blix, 1985; Parker et al., 1993). Measurement of the body water pool size is an integral part of the DLW method therefore a second measurement of body water using only a single isotope (most probably ^2H since this is much less expensive than ^{18}O) some weeks or months after the initial DLW measurement (the exact period over which the change is measured depends on the rate of fat loss) can provide an estimate of fat deposition/mobilisation (Haggarty et al., 1994).

The rate of deposition of fat can also be measured in the same way during the summer months but it should be kept in mind when combining such measurements with a DLW study that the sequestration of ^2H into body fat during fat synthesis can be a major source of error in the DLW method (Fig. 3) and one which has to be corrected for (Haggarty,

1990; Haggarty et al., 1990; 1994). Simple estimates of fat deposition cannot be used to estimate sequestration since ^2H incorporation into fat can occur during the normal process of fat turnover and fat synthesis has been detected in adult weight stable humans from the long term turnover of ^3H_2O (Snyder et al., 1968) and the incorporation of ^2H into circulating fatty acid (Leitch & Jones, 1993). Since the resulting error on the DLW method is specific to each species and physiological state, it may be necessary to sample the adipose stores where the maximum rate of deposition is occurring in order to measure ^2H incorporation directly. The long term sequestration of hydrogen isotopes into body fat is a factor that may also be relevant to the choice of hydrogen isotope (stable ^2H or radioactive ^3H) in DLW or body composition studies in free ranging species as the biological half life of the ^3H will be increased in the presence of significant sequestration (Snyder et al., 1968).

Lactation

Perhaps the biggest nutritional stress on female animals is reproduction and Boertje (1985) has calculated that the energy cost of lactation can account for up to 22% of the energy requirement of caribou. The best field method of measuring milk output is by isotopically labelling the body water of the mother and measuring the appearance of isotope in the body water of the suckling young (McFarlane et al., 1969). This has typically been done in ruminants using radiolabelled ^3H_2O and the technique has been used in reindeer and caribou calves (McEwan & Whitehead, 1970) and caribou and muskoxen (Parker et al., 1990; White et al., 1989). The results of these studies produced estimates of milk intake of around 1.5-1.6 l/day on day 20 of lactation with very little difference between species.

The disadvantages of long term radioisotope contamination of the environment are even more important when carrying out studies involving young which may retain the ^3H for a long time and there have been some attempts to replace ^3H with ^2H, at least in domestic species (Odwongo et al., 1985). However, as in the measurement of body composition, the normal use of the DLW method already provides much of the information required for this isotope transfer technique. Unfortunately, as with the estimation of body composition during fat deposition, the process of lactation can itself introduce significant errors into the DLW method. But

Select a subset of the herd
for detailed study using
existing field methodologies:
•*remote activity recording*
•*behavioral observations*
•*simulation modelling*
•*body composition*

Isotopic verification of field
methodologies in a subset of
the study group:
•*DLW*
•*milk transfer*
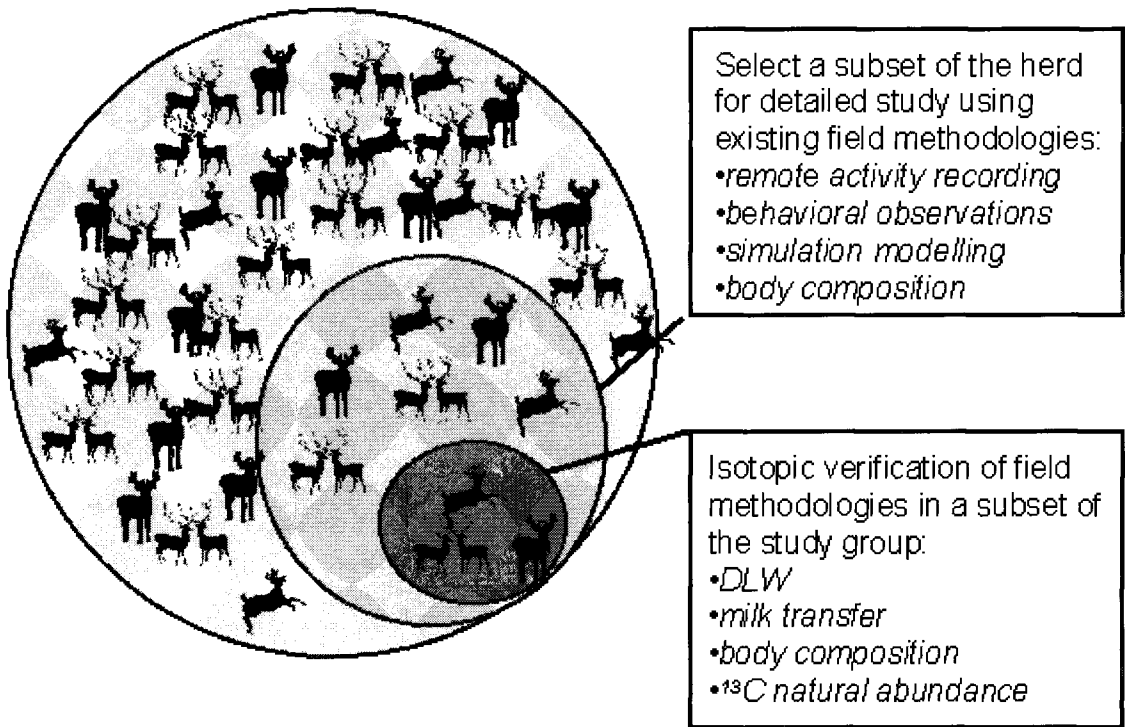•*body composition*
•*[13]C natural abundance*

Fig. 7. Isotopic methodologies are best used in combination with existing field techniques because of cost and because the field methodologies often provide additional information necessary to interpret the isotopic data.

the information collected in the process of estimating the transfer of nutrients in milk from the mother to the young is the same information needed to make the appropriate corrections to the DLW data (Haggarty *et al.,* 1990).

Diet selection and intake

The selection of forages which go to make up the intake of an animal can have a significant effect on expenditure since the quality of the forage will affect the energy cost of digestion and it will also have an effect via the energy cost of physical activity involved in the location and ingestion of the preferred grasses, shrubs, lichens and mosses. Some workers have attempted to estimate diet selection by observation and sampling (e.g. Boertje, 1990) but there is a stable isotope methodology which may also be useful in deriving this parameter. This approach is based on the fact that the ^{13}C concentration of biosynthetically derived organic compounds is determined by the ^{13}C abundance of the source material, the availability of the source material and kinetic isotopic fractionation effects during uptake,

assimilation and incorporation into living organisms. In the case of marine organisms the autotrophic fixation of inorganic carbon dissolved in seawater (Conway *et al.,* 1994) results in an average difference in ^{13}C abundance between marine and terrestrial biomass reported by different groups have been relatively constant, with marine sources being enriched by 13‰ (Craig, 1952; 1953), 14‰ (Wickman, 1952) or 12‰ (Degens, 1970) relative to terrestrial biomass. This difference is large enough in relation to the precision of isotopic measurements on modern isotope ratio mass spectrometers (±0.4‰) to measure the contribution of marine and terrestrial foods to total intake in the human food chain (Haggarty *et al.,* 1999) which is surely the most diverse of any species. Furthermore, the simultaneous measurement of natural variations in other isotopes such as ^{15}N would provide further detail on the dietary origin of nutrients.

In less complex diets, such as those found in arctic ungulates, the task of determining the dietary origin of nutrients is simpler. It has been shown that plants such as lichens, which lack a vascular system, have a characteristic ^{13}C composition which

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is generally lower than that of plants with a vascular system; the ^{13}C abundance of lichen has been reported as -18‰ with that of annual grass growing only a few feet away as -28‰ (Park & Epstein, 1960; Craig, 1953; Wickman, 1952). This kind of difference should be large enough to provide an estimate of the relative contribution of these forages to nutrient intake therefore the approach of exploiting natural ^{13}C abundance variations may be useful in studying the diet selection and intake of free living arctic ungulates even when consuming mixed diets.

Conclusions

With the introduction of innovations such as faecal sampling for the estimation of isotopic enrichment, the availability of appropriate correction factors and stoichiometries for known sources of error and iterative calculation of unknown parameters, the DLW technique can now be considered to be a truly free living methodology suitable for use in wild, free ranging species. Providing the appropriate corrections are made, this field methodology provides accurate values for free living energy expenditure in individual animals. The few studies carried out so far in arctic ungulates suggest that previous predictions have generally underestimated the true level of expenditure, that there is considerable between animal variation in the level of expenditure and that this is largely determined by physical activity.

Energy expenditure is only equal to the more useful parameter of the energy requirement when an animal is in energy balance. However, processes such fat deposition and utilisation and lactation represent a significant part of the energy budget of arctic ungulates. Much of the information gained in the course of a DLW study can be used to estimate milk output and the change in body fat and the additional information tequired to make these estimates is precisely the same information required to correct the DLW method for any errors caused by such processes. In addition to the use of the isotopes ^{2}H and ^{18}O in the DLW method, it is possible that natural ^{13}C abundance variations in the arctic environment may be exploited to study diet selection in truly free living arctic ungulates.

Some disadvantages of the DLW methodology are that it remains expensive, the stable isotope analysis is technically demanding and it requires access to sophisticated mass spectrometry facilities. Therefore, the use of this technique is likely to be limited to relatively small numbers within each study. Furthermore, although DLW can provide an accurate value for free living energy expenditure, it is often important to have information on the individual components of expenditure, e.g. the relative contribution of physical activity and thermoregulatory thermogenesis, in order to interpret the values for overall expenditure. For these reasons it may be that one of the most valuable uses of the DLW method is to validate existing field methodologies and simulation models (Fig. 7) so that they may be used with confidence in truly free living arctic ungulates.

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