

## Bone marrow and kidney fat as indicators of condition in reindeer

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**Abstract:** Back-fat depth, kidney-fat index (KFI), fat and triglyceride contents in femur and metatarsal marrows were measured from 92 semi-domesticated reindeer (*Rangifer tarandus tarandus* L.) in northern Finland during 1980-84. The age of the reindeer varied from 5 months to 10 years and marrow fat content was estimated by five different methods: standard ether extraction, oven-drying (Neiland, 1970), reagent-dry assay (Verme and Holland, 1973), compression method (Greer, 1968) and visual estimation (Cheatum, 1949). The kidney-fat index (Riney, 1955) was significantly related ( $r=0.892$ ) to the percent marrow fat in femur and metatarsus. The oven-drying method gave high correlations with results obtained by ether extraction and reagent-dry methods. Oven-drying produced a lower mean dry-weight value than reagent-drying ( $1.31 \pm 0.36\%$ ). The amount of compression had a concave relationship with the ether-extractable fat content of femur and metatarsal marrows. The subjectiveness of the visual estimation technique limited its use. Femur marrow fat contents (dry-weight %) in reindeer which died from starvation varied between 2 - 8%. Metatarsal fat contents were slightly higher. Adult males had a mean triglyceride content of  $46.8 \mu\text{mol/g}$  in August in the femur. Adult females had slightly lower values. The amount of triglycerides was significantly related to the ether-extractable fat content of the femur ( $r=0.914$ ) and metatarsal ( $r=0.911$ ).

**Key words:** reindeer, fat, marrow, kidney, condition, Finland

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### Introduction

Good condition of reindeer (genus *Rangifer*) in autumn is very important to winter survival and optimal reproduction (e.g. Reimers, 1982; Larsen, 1985). Physical condition, physiological condition, and simply «condition» are, however, ambiguous concepts that frequently refer to the general state of health of animals, apparently inferred from their relative fatness (Anderson, 1981). According to LeResche *et al.* (1974) the nutritional status of individual wild animals can be determined grossly or finely and also acutely or chronically. All body tissues reflect nutritional status to a greater or lesser extent. Some indices are more sensitive than others, and some are also more easily obtained than others. No one

index can give all the answers, and therefore, several indices are generally used.

Storage of fat in the strategy of arctic survival is a general principle of vital importance to the Spitsbergen reindeer (*R. t. platyhyncus*) (Krog *et al.*, 1976; Reimers *et al.*, 1982). The body condition of animals has been defined usually as levels of fat deposited in different parts of the body. Fat animals are not necessarily in good health, but animals with relatively high levels of fat usually also have adequate levels of other resources within the body. Fat is deposited first in bone marrow, then around the kidney, and finally subcutaneously.

Harris (1945) and Rausch (1950) described the depletion of fat deposits in starving deer. As the

deer enter an energy deficient state during winter and spring, fat, protein and mineral reserves are gradually used. Fat reserves are generally utilized sequentially starting with the subcutaneous deposits, followed by the omental and mesenteric, kidney, heart and finally bone marrow fat (Random, 1965; Bear, 1971). As this happens the colour of the marrow changes from white to red and the marrow becomes gelatinous in consistency.

Fat content of bone marrow has long been related to the physiological condition of animals (Jackson, 1928), and several techniques have been used to determine the marrow fat levels in different *Cervidae*-species. Prior to 1970, procedures for determining marrow fat content usually consisted of either crude visual estimates based upon marrow colour and consistency, or extraction procedures which were relatively expensive and also time consuming. Neiland (1970) reported that percent fat in the marrow of barren-ground caribou (*R. t. granti*) was almost identical to percent oven-dry weight. The quick dry-weight method is today most commonly used to determine bone marrow fat content as an indication of the condition of ungulates.

Our purpose was to describe the quantitative relationship between the kidney-fat index and the amount of fat in femur and metatarsal marrows, and then to outline the methods for estimating energy reserves in reindeer. Marrow fat content was estimated by five different methods and comparisons were made.

## Material and methods

Femurs and metatarsal bones were collected from the hind legs of 92 dead or killed semi-domesticated reindeer (*R. t. tarandus* L.) in northern Finland during different seasons in 1980-84. Age, sex, date dead/killed, anatomical measurements (back length, tarsal length and chest width, see Nieminen and Helle, 1980), and general condition were recorded for each animal. Reindeer ages were estimated by tooth wear and the pattern of eruption (Miller, 1974). The condition of the reindeer was classified into three groups: good (group I), fair (group II) and poor (group III), according to measurements and organ fats (Suomus *et al.*, 1975).

Back-fat depth was measured and kidney and perirenal fat deposits were removed and weighed

to the nearest 0.1 g following Riney (1955). The index for each animal was calculated using the fat from both kidneys in order to minimize variation.

The femurs and metatarsal bones were removed and frozen ( $-20^{\circ}\text{C}$ ) in polythene bags until determined. Storage period ranged from 1 to 4 months. Samples of marrow for analysis were taken from the central portions of the bones after noting evidence of dehydration. Then samples were visually classified into four different groups (Cheatum, 1949).

A 60 mm section of marrow was removed from mid-way along each bone and marrow fat determinations were done by compression (Greer, 1968), oven-drying (Neiland, 1970), and reagent-drying (Verme and Holland, 1973) methods.

Bone marrow triglyceride content was determined fully enzymatically (Boehringer GmbH, Mannheim). About 5 - 10 g sample of fresh bone marrow was extracted with di-isopropylether and n-buthanol (6:4). The solvent was washed and evaporated with methanol. The fat residue was then soluted in 6% albumine and determined. All marrow and fat values were placed in age cohorts: calves (5 - 12 months), yearlings (13 - 24 months), and adults (>36 months).

## Results

Back-fat was the only deposit to disappear in most reindeer during winter. The trend in mean thickness of back-fat with age and season is shown in Fig. 1 and Table 1. During the first 2 years of life, males and females accumulated little back-fat except in late summer and early autumn. In males 3 years and older, however, substantial accumulations of back-fat were also present in

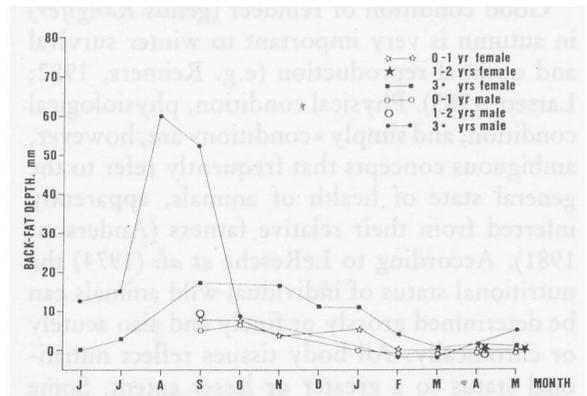


Fig. 1. Seasonal variation in mean back-fat depth in relation to age in male and female reindeer.

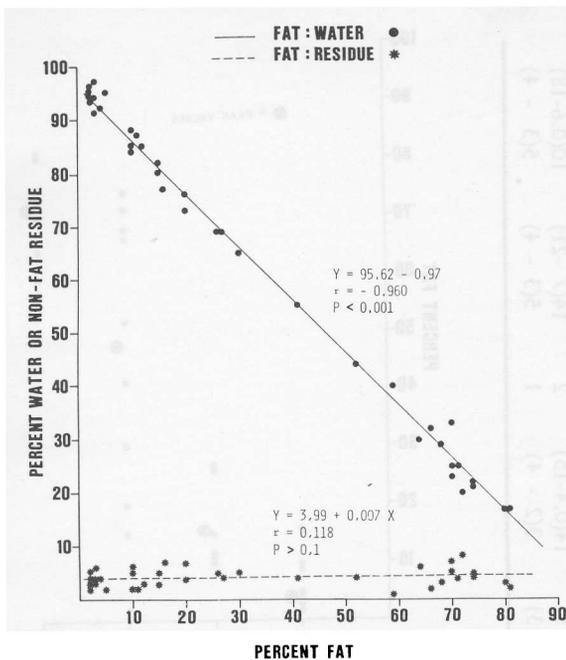


Fig. 2. The relationships between fat and water, and between fat and non-fat residue in reindeer femur marrows. Percent water determined by oven-drying; percent fat determined by ether extraction.

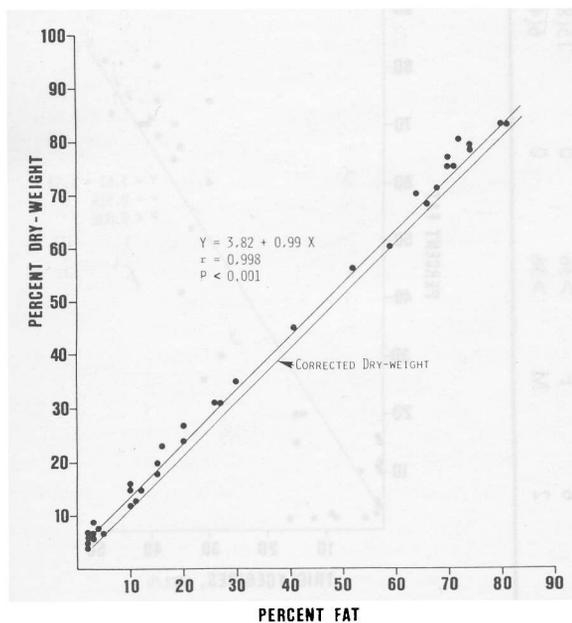


Fig. 3. Comparison of dry-weight, corrected dry-weight, and fat content of reindeer femur marrows. Dry-weight corrections determined from Fig. 2 and calculated according to method by Neiland (1970).

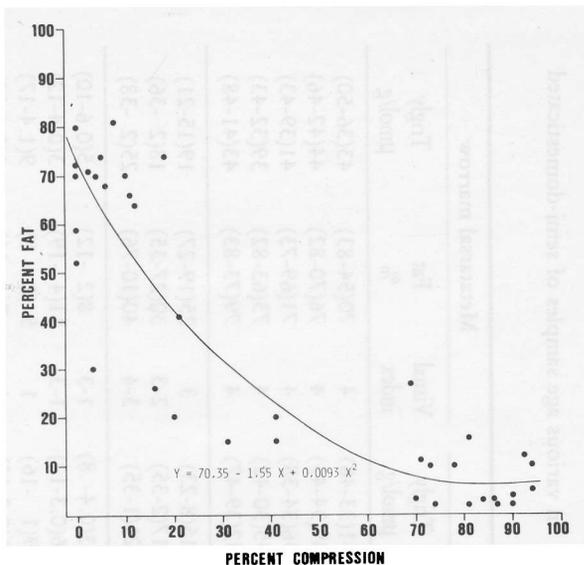


Fig. 4. The relationship between fat content and compression in reindeer femur marrows. Percent fat determined by ether extraction.

spring and summer. Adult males had a mean back-fat thickness of 56.0 mm (range 18.3 - 68.0 mm,  $n=5$ ) during autumn. In September - November the mean thickness of the back-fat depth in females (17.2 mm) was about one-third that of males. The greatest individual back-fat depth in a female (34.0 mm) was half the largest measurement recorded among adult males.

Kidney-fat was present in all reindeer. The amount of kidney-fat varied with the season in males and females of all ages. A significant increase in the mean kidney-fat index occurred during autumn in the calves and adult males. The female kidney-fat index peaked in late autumn and early winter and gradually declined during winter.

Percentage femur and metatarsal marrow fats had less variation with age and season than other fat deposits. Fat was present in the marrow, especially in the metatarsal marrow of all reindeer regardless of age and season, ranging from 2% to 83%. In adult females marrow fat content had almost the same seasonal variation as did kidney and subcutaneous fat deposits. Values were high in October - January, and gradually declined during late winter and spring.

Ether extraction following oven-drying enabled definition of inversely linear relationships of fat to water in reindeer femur ( $r=-0.960$ ) and metatarsal marrows ( $r=-0.999$ ) (Fig. 2). 8 marrows (4 femur, 4 metatarsal) frozen 4 months

Table 1. The condition, back-fat depth (mean and range), kidney-fat index and fat and triglyceride contents in various age samples of semi-domesticated reindeer, F=female, M=male.

Condition	Group	N	Sex	Age months	Back-fat depth mm	Kidney-fat index	Femur marrow			Metatarsal marrow			
							Visual index	Fat %	Trigly $\mu\text{mol/g}$	Visual index	Fat %	Trigly $\mu\text{mol/g}$	
Good	I	7	F	5-12	6.7(4 - 8)	49(46-52)	3-4	68(52-81)	41(33-48)	4	70(54-83)	43(36-50)	
		7	M	5-12	6.0(5 - 7)	54(43-64)	3-4	71(68-74)	39(34-43)	4	76(70-82)	44(42-46)	
		2	M	13-24	9.0(6 - 12)	68(66-70)	4	70(68-72)	36(34-38)	4	71(69-73)	41(39-43)	
		10	F	>36	14(11-18)	48(19-62)	3-4	67(59-74)	35(30-42)	4	73(63-82)	39(32-43)	
		10	M	>36	42(15-68)	66(60-71)	3-4	74(70-80)	42(39-47)	4	79(73-83)	43(41-48)	
	Fair	II	3	F	5-12	5(2-8)	15(8 - 21)	3	20(15-26)	15(8-23)	3	25(19-27)	19(15-21)
			5	F	>36	3(2-9)	31(20-50)	2-3	29(20-41)	17(2-35)	2-3	30(27-35)	18(2 - 36)
		4	M	>36	4(0-8)	29(15-56)	2-3	36(11-66)	21(1-35)	3-4	40(10-76)	25(2 - 38)	
		III	12	F	5-12	0	11(8 - 14)	1-2	5(2 - 10)	3(0.4 - 8)	1-3	8(2 - 12)	5(0.6-10)
			15	M	5-12	0	4(3 - 5)	1-3	10(2 - 16)	6(0.3-12)	1-3	11(4 - 19)	3(0.4-12)
4	F		13-24	0	17(10-24)	1	3(2 - 5)	9(1 - 16)	1	5(2 - 8)	9(1.4-17)		
Poor	5	M	13-24	0	4(3 - 5)	1	6(2 - 10)	6(0.3-12)	1	7(4 - 11)	6(0.4-12)		
	6	F	>36	0	15(8 - 24)	1-2	10(3 - 15)	14(0.4-15)	2	14(7 - 21)	10(0.6-18)		
	2	M	>36	0	6(4 - 9)	1	2(1 - 3)	3(2 - 4)	1	5(3 - 4)	5(3 - 4)		

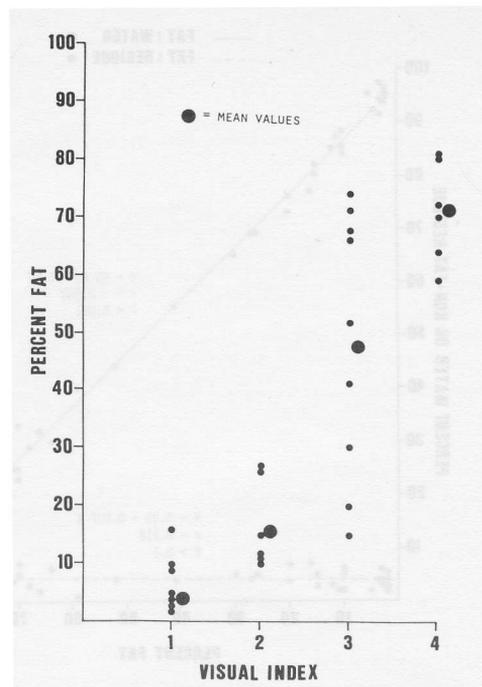


Fig. 5. Comparison of ether extraction and visual assessment method of reindeer femur fat estimation.

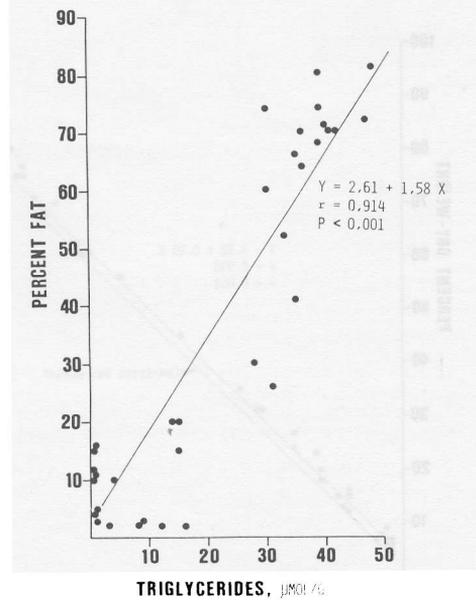


Fig. 6. The relationship between fat content and triglyceride content in reindeer femur marrow. Percent fat determined by ether extraction; triglycerides determined fully enzymatically.

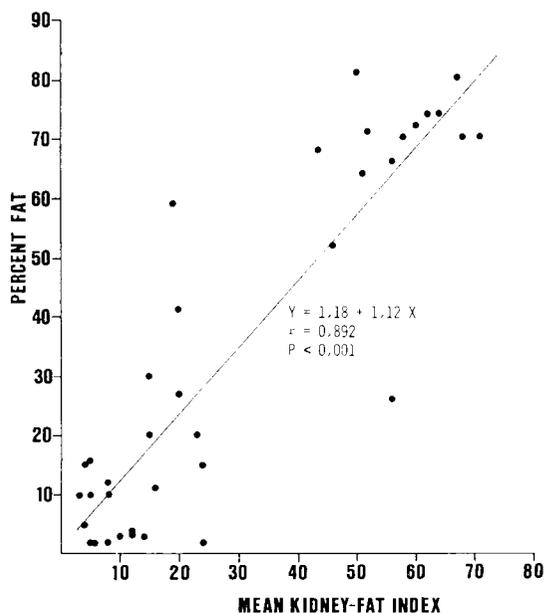


Fig. 7. The relationship between femoral fat content and the kidney-fat index in reindeer. Percent fat determined by ether extraction.

before chemical analysis gave  $3 \pm 2\%$  higher fat content than a fresh sample. However, the amount of non-fat residue in the marrows would be the prime source of error in dry-weight determinations of marrow fat content. Closer approximation of the fat value using the oven-dry method can be calculated by subtracting a correction factor for the non-fat residue (see Neiland, 1970). Fig. 3 shows the relationships of fat content to dry-weight and corrected dry-weight in the femur. Graphs of residue vs. dry-weight for the oven-dry and reagent-dry methods were very similar. Oven-drying produced, however, a lower mean dry-weight value than reagent-drying ( $1.3 \pm 0.36\%$ ). Dry-weight values for the femur and metatarsal marrow samples determined by oven-dry and reagent-dry methods were highly correlated with ether extractable content ( $r=0.998$ ;  $r=0.998$ ;  $r=0.989$ ;  $r=0.990$ , respectively).

The comparison of the compression and extraction methods of assessing marrow fats are shown in Fig. 4. The amount of compression had concave relationships with the ether-extractable fat content of femur and metatarsal marrows. The amount of overlap observed between fat values for samples allocated to different visual index grades was also significant. The subjecti-

veness of this visual estimation technique limited its use (Fig. 5).

The amount of marrow fat triglycerides varied with the season in males and females of all ages. A significant increase in the mean triglyceride values occurred during autumn. Adult males had a mean triglyceride content of  $46.8 \mu\text{mol/g}$  in August and  $40.8 \mu\text{mol/g}$  in September in femur marrow samples. Adult females had slightly lower values (mean  $30.4 \mu\text{mol/g}$  in September). Very low triglyceride values were found during winter and spring in femur and metatarsal marrows. The amount of triglycerides was significantly related to the ether-extractable fat content of the femur ( $r=0.914$ , Fig. 6) and metatarsus ( $r=0.911$ ).

Percentage marrow fat is plotted against kidney-fat index in Fig. 7. Percent marrow fat in femur and metatarsus was significantly correlated with the kidney-fat index ( $r=0.892$  and  $r=0.887$ , respectively).

## Discussion

Riney (1955) in reference to ruminants stated that «fat can be taken as a direct measure of the condition reflecting the metabolic level or goodness of physiologic adjustment of an animal to its environment». Measurements of fat deposits have been widely used as indices of energy reserves and general nutritional condition in cervids. Growing attention has been also focused on the importance of fat reserves for winter survival of many cervids (see Gasaway and Coady, 1974). Mautz (1978) refers to northern living cervids as semihibernators. According to Franzmann *et al.* (1978) Alaskan moose (*Alces alces*) lost 30% of their body weight from early winter to late spring. It is estimated that 75 - 80% of such a weight loss may be from fat stores, wherein each gram of fat contributes 38.9 kJ (Gasaway and Coady, 1974). The caloric content of body fat in white-tailed deer (*Odocoileus virginianus*) is estimated by Robbins *et al.* (1974) as 9.49 kilocalories (39.7 kJ) per gram.

The ability to store large amount of fat is a common feature among all arctic mammals, and according to Ringberg *et al.* (1980) the Svalbard reindeer are unique in this regard. The calves are born smaller than are other *Rangifer* subspecies. No subcutaneous or perivisceral white fat is found in newborn calves, but during autumn

dissectable body fat of Svalbard reindeer makes up 14 - 17% of the total body weight among calves and yearlings and 20 - 22% among the adults (Reimers, 1982). Maximum body fat concentration in the Svalbard reindeer (28.7 - 40.1%, Reimers *et al.*, 1982) is higher than in semi-domesticated reindeer and also higher than reported for other cervid species: less than 20% in white-tailed deer (Robbins *et al.*, 1974) and less than 11% in roe deer (*Capreolus capreolus*) (Weiner, 1973).

In Svalbard reindeer maximum average back-fat depths in males and females do not exceed 83 and 48 mm, respectively (Reimers *et al.*, 1982). The back-fat depths of calves and yearlings are 30.1 - 38.3 mm, which are 5 - 6 times greater than found in Finnish reindeer (Table 1) and also in Kaminuriak caribou (*R. t. groenlandicus*) of the same age (Dauphine, 1976). Upper extreme values on adult males range from 51 - 76 mm on Alaskan reindeer (Palmer, 1934), 75 mm on Swedish reindeer (Jacobi, 1933), 68 mm on Finnish reindeer (Table 1), 40 mm among Kaminuriak caribou (Dauphine, 1976), 71 mm on caribou on Coats Island, N.W.T. (Parker, 1975), 80 mm on wild reindeer in the U.S.S.R. (Shaposnikov, 1955) and 50 mm on wild reindeer in southern Norway (Reimers, 1982). Compared with semi-domestic reindeer wild forest reindeer (*R. t. fennicus*, Lönbn.) in Finland had very little body fat (Nieminen and Helle, 1980).

Fat content of the carcass varies considerably with sex and season in the genus *Rangifer*. Before the rut average back-fat depth in the Svalbard reindeer males is measured at 83 mm, which corresponds to a total dissectable fat weight of 24 kg or 22% of the total body weight (Reimers, 1982). McEwan (1968) reported accumulation of about 20 kg fat in a large male caribou at the end of the summer growth period. According to Spiess (1979), a 110 kg caribou stag killed in late September had about 22 kg (20% of the body weight) fat and 50 kg (55%) meat. This animal provided 198 000 kcal (828 828 kJ) or 66 man-days (3000 kcal/man-day) worth of fat-supplied calories. The femur, tibia, humerus, radius and metapodial bones of a 2-year-old, July-killed male caribou yielded about 520 g of fat from the marrow. An 80 kg female killed in December has a theoretic 10% fat content (8 kg) and 40 kg of meat. This female represents about 34 man-days of food, more than twice what a 90 kg stag killed at the same time represents.

Øritsland (1970) estimated the total body fat as being 15 kg (14.3% of the body weight) in a male Spitsbergen reindeer killed in August. According to Kleiber's (1961) formula  $m(\text{basal metabolism}) = 70W^{0.75}$ , this could last for 60 days at basal metabolic level and thermal neutrality. Counting seven months from October through April this would be able to supply about 30% of the basic demand for energy from fat storage during winter (Krog *et al.*, 1976). Compared to pre-rut weights in August adult male body weights were decreased, however, 24% by mid October, the most active rutting period (Reimers, 1982). Franzmann *et al.* (1978) also reported a loss of over 10% for Alaskan moose during the rut, whereas McMillan *et al.* (1980) showed 15 - 20% during the rut of white-tailed deer. By the end of winter; the fat reserves of young reindeer are almost totally depleted (see Table 1), while most surviving adults still have a few kilograms left. According to Krog *et al.* (1976) at the end of March the fat content may be at least 2 kg, of which about 500 g is located around the kidneys.

Bone marrow fat is the last reserve to be mobilized, and its level reflects condition only at the lower end of an overall animal condition (e.g. Riney, 1955; Dauphine, 1976). Thus, the use of marrow fat is dependent upon the initial level of other fat reserves and also the degree and duration of nutritional stress. Bischoff (1954) found that the percentage of femur marrow fat was not well correlated with other fat deposits or femur marrow colour but was strongly correlated with the visual estimate of marrow consistency. Femurs have, however, usually been used for fat content determination in ungulates.

According to Dauphine (1976) the mobilization of deposit fat in caribou of both sexes occur in the sequence back-fat, kidney-fat, abdominal fat, and marrow fat. The exhaustion of the back-fat, and the first use of the marrow fat, coincides with the cessation of the decline on body weight and with a 50% decline in the two visceral fat indices. Ransom (1965) reported that marrow fat in white-tailed deer was mobilized only when kidney-fat had declined to ca. 30% of kidney weight. Cheatum (1949) believed, however, that the health of adult white-tailed deer was not affected until the fat content of the marrow dropped below 25%.

Klein (1968) found no marrow fat in the medullae of long bones from starved reindeer and took that as evidence that all fat was absent from the marrow at the time of death. Femur marrow fat contents (dry-weight %) in reindeer dead from starvation in the present study varied between 2 - 8%. Metatarsal fat contents were slightly higher. Neiland (1970) reported levels as low as 5% in caribou shot in Alaska. The lowest femur marrow fat value recorded from suspected winter kills was 6.1% for Alaskan moose calves and 5.5% for adults (Franzmann and Arneson, 1976).

Cheatum (1949) reported that fat is mobilized from proximal leg bones in deer after other body fat deposits have been exhausted. Bischoff (1954) found, however, no correlation between fat levels in the femur and tibia in mule deer. (*O. hemionus*). In accordance with the present results Peterson *et al.* (1982) reported that fat mobilization was first evident in the limbs of moose in the femur and humerus, then the tibia and metatarsus, and finally the radius and metacarpus.

Many studies indicate that femur marrow could be fat-depleted and the animal dead from malnutrition with the distal bones still containing considerable fat. Cheatum (1949) suggested that fat mobilization might be more advanced in proximal bones because they are closer to the body core and warmer in winter. It seems also reasonable to expect reduced circulation within capillary networks and retarded metabolic processes in colder extremities.

Since its introduction by Riney (1955) the kidney-fat index (KFI) perhaps has been the most widely measure of cervid carcass fat. KFI correlate adequately with percentage of femur marrow fat, carcass density and weight, and depth of rump fat in mule deer (Anderson, 1981). In agreement with present results the measure of kidney-fat is, however, highly variable and consequently requires a large sample. In addition, the KFI — based on one kidney — is not very comparable to indexes using both kidneys. The required number of mature mule deer ranged by season from 88 (winter) to 221 (summer) for males and 114 (winter) to 532 (summer) for females (Anderson *et al.*, 1972). The required number of mature deer (to provide an estimate of depth of back-fat within 10% of the true mean at the 95% confidence level) ranged from 381 (summer) to 2595 (spring) for

males and 383 (winter) to 2192 (summer) for females.

The choice of a suitable index of relative fatness may be influenced much by sex, time and manpower available, physical facilities and constraints on collecting procedures. Thus, for female mule deer eviscerated carcass weight may be the best choice, but this may not necessarily be so for males (Anderson, 1981). Both the kidney-fat index and femur and metatarsal marrow fats may be useful in combination for reindeer. When using the bone marrow, caution must be used because in younger animals erythropoiesis may still be occurring in the femur, and the marrow would be red in colour. Present results indicate that the oven-dry method is nearly as convenient as visual and compression methods and as accurate as standard ether extraction.

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