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USE OF URINE IN SNOW TO INDICATE CONDITION OF WOLVES

L. DAVID MECH,[1] U.S. Fish and Wildlife Service, Patuxent Wildlife Research Center, Laurel, MD 20708
ULYSSES S. SEAL, Veterans Administration Medical Center, Research Service, Minneapolis, MN and Department of Biochemistry and Department of Fisheries and Wildlife, University of Minnesota, St. Paul, MN 55108
GLENN D. DELGIUDICE, Department of Fisheries and Wildlife, University of Minnesota, St. Paul, MN 55108

Abstract: Urine deposited in snow by wild gray wolves (*Canis lupus*) and by fed and fasted captive wolves was analyzed for urea nitrogen, calcium, sodium, potassium, and creatinine. Ratios of the elements with creatinine were considerably higher for fed than for fasted animals, and ratios for fed wolves compared favorably with ratios from wolf urine in snow along trails leading from kills. Thus, wolf urine in the snow can indicate whether wolves have fed recently, and a series of such urine collections from any given pack can indicate relative nutritional state.

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Urinalysis theoretically could indicate the approximate interval between when a carnivore last fed and when it urinated (DelGiudice et al. 1987). Urine contains metabolic byproducts, including urea, that result from the digestion of meat and exhibit definite patterns of excretion related to protein intake. Furthermore, urinary ratios of sodium : creatinine, potassium : creatinine, and calcium : creatinine also can be used to distinguish recently fed from fasted wolves (DelGiudice et al. 1987). Because most of the wolf range is snow-covered for long periods, frozen urine can be collected from along wolf trails in the snow (Peters and Mech 1975, Rothman and Mech 1979). Analyses of frozen urine samples from the same individual, pack, or population collected over a period could then indicate the relative welfare of those respective animals. We report here an attempt to test this technique and describe the methods by which such assessments can be made.

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[1] Mailing address: North Central Forest Experiment Station, 1992 Folwell Avenue, St. Paul, MN 55108.

Table 1. Effects of feeding and fasting on selected chemical components ($\bar{x} \pm$ SE) in urine deposited in snow by captive and free-ranging wolves in Minnesota, 1980. (No. of urine samples in parentheses.)

Components	Captive: Fed (11)	Captive: Fasted (10)	Free ranging: Around deer kills[a] (234)	Free ranging: Along trails (9)
Urea nitrogen (U) (mg/dl)	968 ± 168	744 ± 73	817 ± 24	1,076 ± 95
Creatinine (C) (mg/dl)	21.7 ± 3.3	154.3 ± 14.8	15.1 ± 1.0	29.7 ± 4.4
U:C[b]	44.4 ± 3.5	4.9 ± 0.2	80.8 ± 2.6	43.7 ± 6.9
Calcium (Ca) (mg/dl)	0.45 ± 0.08	0.45 ± 0.08		1.12 ± 0.67
Ca:C × 1,000[c]	21.6 ± 3.07	3.4 ± 0.79		30.7 ± 12.80
Sodium (Na) (mEq/liter)	16.4 ± 6.1	2.4 ± 0.8		18.8 ± 2.03
Na:C × 100[d]	82.6 ± 28.1	2.1 ± 1.1		78.3 ± 14.1
Potassium (K) (mEq/liter)	31.2 ± 3.2	20.7 ± 3.5		46.7 ± 7.2
K:C × 100[b]	173.2 ± 20.6	15.7 ± 3.5		160.1 ± 18.1

[a] Within 100 m of 15 wolf-killed deer.
[b] Fed vs. fasted wolves and fasted wolf samples vs. trail samples, $P < 0.0001$.
[c] Fed vs. fasted wolves, $P < 0.05$.
[d] Fed vs. fasted wolves and fasted wolf samples vs. trail samples, $P < 0.01$.

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STUDY AREA AND METHODS

Our approach involved studying: (1) captive wolves under controlled conditions and (2) collecting frozen urine around depredated carcasses and along the trails of free-ranging, radio-collared wolves from a companion study.

The captivity study compared urine from fasted wolves with that of wolves fed white-tailed deer (*Odocoileus virginianus*) meat ad libitum in Minnesota during January 1980. Urine was collected by catheterization and the differences in composition between the 2 treatment groups were reported by DelGiudice et al. (1987). Urine from frozen deposits in the snow along the edges of the wolf pens also was compared with the urine obtained via catheterization from the same wolves.

Secondly, urine was collected from deposits made by wild wolves in the snow around 15 deer kills in the Superior National Forest of northeastern Minnesota in winter 1980 and from along trails in snow made by radio-tagged wolf packs (Mech 1979) after they left kills. Urine was collected from snow with clean plastic scoops and placed in sterile, plastic tubes to minimize bacterial contamination.

Snow-urine samples were stored frozen (−20 C) and subsequently assayed for urea nitrogen (U), creatinine (C), calcium (Ca), sodium (Na), and potassium (K), as described by DelGiudice et al. (1987). Snow-urine samples were diluted 1:50 and 1:10 for U and C determinations, respectively, and 1:200 for Na and K. Samples were not diluted for Ca analyses. Snow-urine samples from around deer kills were not assayed for Na, K, or Ca.

Urinary constituents were analyzed as ratios to C for reasons explained by DelGiudice et al. (1987) and to correct for dilution by snow. Data are presented as means and standard errors of the means. Multiple comparisons of data from the fed and fasted wolf groups and from samples collected along trails were made by 1-way analysis of variance (ANOVA); significant differences between groups were determined by orthogonal contrasts (Hintze 1982). The minimal level of significance accepted was $P = 0.05$.

RESULTS AND DISCUSSION

In the captive wolves fasting and feeding U:C, Ca:C, Na:C, and K:C ratios showed the same patterns in the urine samples collected from snow (Table 1) as in the ones collected by catheterization (cf. Table 1 and DelGiudice et al. [1987]). Thus, it appears that mixing of the urine with snow does not substantially distort these ratios, lending confidence to the values obtained from wild wolf urine voided into snow. Significantly greater values for all of these ratios were found in fed wolves.

Mean U:C ratios of urine sample groups collected from snow near individual deer kills ranged from 35.0 ± 5.9 to 102.3 ± 10.3 and averaged 80.8 ± 2.6 (Table 1). Ratios from individual urine deposits collected along wolf trails leading away from kills varied from 14.6 to

82.8 and averaged 43.7 ± 6.9 (Table 1). The values from along trails were comparable to those from well-fed captive wolves (DelGiudice et al. 1987), but the values from around kills were higher.

There are probably 2 reasons for the higher mean U:C values from around deer kills. First, longer fasting, greater energy expenditure between feedings, and greater consumption by free-ranging wolves probably resulted in increased urea synthesis and excretion. Changing inductions of urea cycle enzymes with varying protein intake (Walser 1973), and rapidly increasing enzyme concentration with increasing dietary protein intake (Saheki et al. 1978), act as "short-term regulatory mechanisms . . . modulating the rate in which urea loads are excreted" (Bovee 1984:621). Furthermore, the canine kidney possesses great functional reserve for excretion of excess protein metabolites, which may be an evolutionary adaptation to infrequent eating (Bovee 1984).

The 2nd reason why free-ranging wolves have higher U:C ratios in urine near kills may be because in winter wolves probably derive most of their excretory water from food. (We have never seen them eat snow.) That water is most available for electrolyte and nitrogen excretion while food is being absorbed and metabolized (Pitts 1944). Since increased protein intake increases renal blood flow and filtration rate in the canine, this probably leads to a greater and more immediate excretion of urinary urea in free-ranging wolves. Between feedings, glomerular function is reduced to conserve water. The captive wolves had free water available at all times so probably were more hydrated. Captive wolves also consumed food daily, thus more evenly distributing urea excretion.

Mean Na:C, K:C, and Ca:C ratios in snow-urine samples collected along wolf trails leading away from kills closely paralleled those values from snow-urine samples in captive, fed wolves (Table 1). These values from free-ranging and captive wolves were somewhat greater than those from urine samples obtained via catheterization from captive, fed wolves (DelGiudice et al. 1987), probably related to slight differences (i.e., hours) in the periods between eating and urine deposition or sampling. The importance of Na, K, and Ca and mechanisms responsible for the observed patterns of their urinary excretion in fed and fasted wolves have been discussed elsewhere (DelGiudice et al. 1987).

These preliminary results from both the captive study and from wild wolves indicate that the technique of collecting wolf urine from the snow and examining the U:C, Na:C, K:C, and Ca:C ratios can be a useful method of determining whether wolves have recently fed or not. The cost of analyzing large quantities of urine samples for urea and creatinine is estimated at approximately $0.25/sample for reagents, and the analyses can be conducted with equipment and technicians available at most hospitals and commercial analytical laboratories.

Obviously, consideration must be given to the proper sampling scheme in order to use this technique to draw valid conclusions about the relative condition of wolves from different packs, areas, years, etc. For example, if individual packs are being compared, one must be certain that urine attributed to a given pack was really left by that pack. The use of radio-collared wolves would help in this respect. In addition, each pack must be sampled frequently enough throughout a winter to obtain a valid representation of their actual mean time since feeding. It would then be reasonable to conclude that a pack that has a significantly longer time since feeding is in poorer condition than the same size pack that feeds more frequently.

Because this technique depends on metabolic relationships that are basic among a wide variety of mammals, we propose that it should be usable with any mammal inhabiting regions that have snow for a significant part of the year.

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