CHEMICAL ANALYSES OF DEER BLADDER URINE AND URINE COLLECTED FROM SNOW

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Urinalysis has provided a new avenue for identification and study of nutritional indices in white-tailed deer (Odocoileus virginianus). Initial studies examined urea nitrogen (Warren et al. 1981, 1982; Waid and Warren 1984; DelGiudice et al. 1987a), electrolytes, and hydroxyproline; all showed potential as indicators of nutritional condition (DelGiudice et al. 1987a, 1988). During early undernutrition and starvation, deer conserve urea nitrogen, sodium, and potassium by renal tubular reabsorption; and urinary excretion of these characteristics decreases progressively (DelGiudice et al. 1987a, 1988; DelGiudice and Seal 1988). Similarly, decreases in urinary hydroxyproline have been associated with declining nutrition (Whitehead 1965, McCullagh 1969). As nutritional deprivation is prolonged, catabolism of endogenous protein, including bone collagen, increases, thereby dramatically increasing urinary urea nitrogen and hydroxyproline, as well as sodium, potassium, and calcium (Del-Giudice et al. 1987a, DelGiudice and Seal 1988).

The practicality of collecting such physiological data for research and management purposes is often limited. It is expensive, time-consuming, and often difficult to capture and immobilize free-ranging animals to acquire data on physiological condition (DelGiudice et al. 1987b).

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We recently demonstrated that urine deposited in snow (snow-urine) by wolves (Canis lupus) can be analyzed biochemically to reflect nutritional status (DelGiudice et al. 1987b, Mech et al. 1987). Our objective here is to document the feasibility and accuracy of collecting and chemically analyzing snow-urine of white-tailed deer for characteristics that have shown potential as nutritional indices.

METHODS

We collected a total of 8 urine samples from 6 captive, adult white-tailed deer (5 F, 1 M) by catheterization or cystocentesis (Kreeger et al. 1986) from 10 January to 21 February 1985 (2 deer were sampled twice). We aliquoted samples into 5-cc falcon tubes, and stored them at -20 C. Samples were assayed within 3 months for urea nitrogen (U), creatinine (C), sodium (Na), and potassium (K) as described by DelGiudice et al. (1987a), and the remainder of each sample was refrozen.

On 12 December 1986, each urine sample was thawed and poured into the snow. An hour later, the snow-urines were collected by using a plastic bag as a glove to prevent contamination of the sample by skin contact. We collected the most concentrated (i.e., yellow) portion of the snow-urine sample from a depth of about 8 cm. Bags were sealed, and samples were thawed at room temperature, swirled thoroughly to assure a homogeneous mixture, and then aliquoted into 5-cc plastic tubes. Aliquots were stored frozen and assayed within 2 weeks as described above. We collected 3 samples of urine-free snow randomly and analyzed them as controls.

We tested reproducibility and potential effects of time on chemistries (nutritional indices) after urine is deposited in snow with 6 additional urine samples from 6 different adult female deer. These deer were on various diets and were not chosen to reflect a particular nutritional plane. Samples were aliquoted into 5 12-cc falcon tubes. On 3 December 1987, we deposited 4 aliquots of each urine sample individually into the snow

Table 1. Ratios of urinary chemistries to creatinine in white-tailed deer bladder urine stored frozen and analyzed within 3 months of collection versus snow-urine from the same samples after 22 months of storage at -20 C.

Urine chemistry ratios ^a	Bladder (n =		Snow- (n =		
	ī	SE	Ī	SE	t-test
U:C	3.4	0.6	3.7	0.7	P > 0.10
$Na:C \times 100$	27.4	6.8	25.8	7.6	P > 0.10
$K:C \times 100$	45.9	7.7	44.3	7.8	P > 0.10

 $^{^{\}perp}$ U:C = urea nitrogen: creatinine, Na:C = sodium: creatinine, and K:C = potassium: creatinine.

and marked the location of each sample. The fifth aliquot of each urine sample was kept frozen at -20 C. Subsequently, 1 snow-urine sample from each of the 6 original bladder urines was collected at 24, 48, 72, and 120 hours after deposition and assayed for U, C, Na, and K.

Metabolite data from individual urine samples were compared as ratios to creatinine, which is a nonproteinous, nitrogenous compound. The urinary excretion of creatinine is constant over 24 hours in individual animals (Vestergard and Leverett 1958, Coles 1980: 246). Creatinine ratios correct for extraneous variability, estimate 24-hour excretion, and correct for dilution by snow.

RESULTS AND DISCUSSION

There were no differences between mean U:C, Na:C, and K:C ratios in the bladder urine samples compared to those in the snow-urine samples (Table 1). Bladder urine samples were

assayed and stored frozen 22 months before being used to make the snow-urine samples. The lack of any difference between the U:C, Na:C, and K:C ratios of the urine and snow-urine samples strongly supports the validity of deep-freeze storage for long-term sample preservation. Urine-free snow, analyzed as a control, contained no U or C and negligible amounts of Na ($\bar{x} = 0.039 \, \text{mEq/L}$, SE = 0.004) and K ($\bar{x} = 0.001 \, \text{mEq/L}$, SE = 0.001).

There were no significant differences among mean U:C, Na:C, and K:C ratios of bladder urine samples and snow-urine samples collected between 24 and 120 hours after deposition of urine into snow (Table 2). Maximum ambient temperatures were ≤0 C for the first 2 days after urine deposition, but averaged 1.4 C on days 3-5. Concentrations of U, Na, K, and C remained stable for the 24- to 72-hour collections; however, notable decreases in chemistry concentrations occurred by the 120hour collection (Table 2). Average dilutions of all chemistries between the 24- and 120-hour collections were similar and ranged from 70.9 to 75.2%. However, ratios of these chemistries to creatinine remained unchanged for all 4 collections (Table 2).

These results demonstrated the feasibility of analyzing snow-urine of deer as a means of collecting physiological data that may be used

Table 2. Characteristics of white-tailed deer bladder urine and snow-urine collected 24, 48, 72, and 120 hours after deposition.

Urine characteristics ^a			Snow-urine Hours after deposition								
	Bladder urine		24		48		72		120		
	Ī	SE	ž	SE	Ī	SE	ī	SE	ī	SE	
U (mg/dl)	493.0 ^b	57.6	103.0	12.0	95.6	8.8	95.2	7.6	30.0	2.7	
Na (mEq/L)	52.0	8.8	10.9	1.7	10.1	1.4	10.5	1.4	2.7	0.3	
K (mEq/L)	78.1	6.6	16.6	2.0	15.3	1.0	15.8	1.2	4.6	0.5	
C (mg/dl)	106.0	11.9	22.1	3.2	20.3	2.0	20.4	2.1	6.4	0.8	
U:C°	4.7	0.4	4.8	0.5	4.8	0.4	4.8	0.4	4.8	0.4	
$Na:C \times 100^{c}$	50.0	6.9	51.8	6.9	51.5	7.3	54.0	8.4	44.4	5.8	
$K:C \times 100^{\circ}$	75.8 -	5.1	77.5	4.2	77.4	4.7	79.4	5.6	73.2	4.4	

Definitions given in Table 1.

n = 6 for each collection.

 $^{^{\}circ}$ No differences (P < 0.10) among means

for nutritional assessment. As with free-ranging wolves, this method circumvents many constraints associated with traditional techniques of physiological data collection (DelGiudice et al. 1987c).

MANAGEMENT IMPLICATIONS

Collecting fresh samples after a recent snowfall permits a more accurate reference of deer metabolic data and the derived nutritional assessment to a specific time. Our data showed that as long as ambient temperatures remain below freezing, urine chemistry ratios in snow may remain unaltered for days after urine is deposited. However, post-deposition time should be minimized once temperatures rise above freezing. Although we found that ratios remained constant over several days, despite daily maximum temperatures above freezing, more prolonged snowmelt conditions may dilute chemistries to concentrations below the measuring capabilities of laboratory instruments. The effects of snowmelt on this technique probably will vary with snow depth and depth of the most concentrated part of the snow-urine sample. Snow-urine analysis should permit more economical, continuous, and sensitive monitoring of the condition of deer in winter yards.

LITERATURE CITED

Coles, E. H. 1980. Veterinary clinical pathology. W. B. Saunders Co., Philadelphia, Pa. 562pp.

DELGIUDICE, G. D., L. D. MECH, U. S. SEAL, AND P. D. KARNS. 1987a. Winter fasting and refeeding effects on urine characteristics in white-tailed deer. J. Wildl. Manage. 51:860–864.

——, AND U. S. SEAL. 1988. Classifying winter undernutrition in deer via serum and urinary urea nitrogen. Wildl. Soc. Bull. 16:27–32.

indices of wild mammals: a new direction for wild-life and habitat management systems. *In* Proc. XVIIIth Congr. Int. Union Game Biol., Jagiellonian Univ., Krakow, Poland. In Press.

ing and fasting on wolf blood and urine charac-

teristics. J. Wildl. Manage. 51:1-10.

nary hydroxyproline to dietary protein and fasting in white-tailed deer. J. Wildl. Dis. 24:75–79.

Kreeger, T. J., G. D. DelGiudice, U. S. Seal, and P. D. Karns. 1986. Methods of urine collection for male white-tailed deer. J. Wildl. Dis. 22:442–445.

McCullagh, K. 1969. The growth and nutrition of the African elephant. I. Seasonal variations in the rate of growth and the urinary excretion of hydroxyproline. East Afr. Wildl. J. 7:85–90.

MECH, L. D., U. S. SEAL, AND G. D. DELGIUDICE. 1987. Use of urine in snow to indicate condition of wolves.

J. Wildl. Manage. 51:10-16.

Vestergard, P., and R. Leverett. 1958. Constancy of urinary creatinine excretion. J. Lab. and Clin. Med. 51:211–218.

Waid, W. D., and R. J. Warren. 1984. Seasonal variations in physiological indices of adult female white-tailed deer in Texas. J. Wildl. Manage. 20:212–219.

Warren, R. J., R. L. Kirkpatrick, A. Oelschlaeger, P. F. Scanlon, and F. C. Gwazdauskas. 1981. Dietary and seasonal influences on nutritional indices of adult male white-tailed deer. J. Wildl. Manage. 45:926–936.

B. Whelan. 1982. Energy, protein, and seasonal influences on white-tailed deer fawn nutritional indices. J. Wildl. Manage. 46:302–312.

WHITEHEAD, R. G. 1965. Hydroxyproline creatinine ratio as an index of nutritional status and rate of growth. Lancet ii:567-570.

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