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Glycosylated Hemoglobin as a Stable Alternative to Serum Glucose in White-tailed Deer

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ABSTRACT: We compared serum glucose concentration and percent glycosylated hemoglobin (GH) in captive and wild white-tailed deer (*Odocoileus virginianus*) to determine stability of glucose relative to GH. Temporal changes in levels of serum glucose and GH were ascertained from serial blood samples collected from three captive deer over a 2-week period. State of glycemia also was determined for 17 wild deer that were collected from three populations in southeastern Oklahoma and southwestern Arkansas (USA). Concentration of serum glucose of captive deer decreased ($P = 0.04$) from 190.4 to 155.8 mg/dl over the 2 weeks; percent GH did not differ temporally ($P = 0.30$). Percent GH of wild deer did not differ ($P = 0.23$) when deer were separated into 2 groups (high and low state of glycemia) based on the median serum glucose concentration. We found a significant difference ($P = 0.04$) in percent GH among wild deer populations; serum glucose concentration did not differ ($P = 0.72$) among populations. Our results indicate that percent GH is more stable than serum glucose concentration and may be useful in population comparisons of nutritional condition.

Key words: Blood, glycosylated hemoglobin, *Odocoileus virginianus*, serum glucose, white-tailed deer, nutritional condition.

When assessing nutritional condition of white-tailed deer (*Odocoileus virginianus*), serum glucose is routinely included in metabolic profiles (Seal and Erickson, 1969; Seal et al., 1978; Morris and Bubenik, 1983; Wade and Warren, 1984). However, few studies have found differences in serum glucose concentrations that could be directly attributed to animal condition. Wesson et al. (1979) observed significant variation in serum glucose over a 90-min period in restrained deer and concluded that handling stress caused acute glycemia and reduced the utility of serum glucose as an index of carbohydrate metabolism.

Glycosylated hemoglobin (GH) (i.e., hemoglobin-bound glucose) has been used in human nutrition to assess serum glucose stability of diabetics (Gabbay et al., 1977; Jovanovic and Peterson, 1981). Blood glucose can readily diffuse into erythrocytes and binds primarily with terminal valine amino acids located in the beta chains of the hemoglobin molecule (Bunn et al., 1978). Binding of glucose to hemoglobin is irreversible (post Amadori rearrangement) and remains stable for the life of the hemoglobin molecule (ca. 120 days) (Peterson and Jovanovic, 1984). Correlations of GH with average serum glucose concentration indicate that GH can be useful as a long-term indicator of glucose status in humans (Koenig et al., 1976).

Use of GH as a long-term indicator of glucose status in wild and domestic ruminants has yielded inconclusive results. GH values for markhor (*Capra falconeri*), mouflon (*Ovis musimon*) and aoudad (*Ammotragus lervia*) were found to be low ($\leq 1\%$), possibly due to low erythrocyte permeability to glucose (Richter, 1986). However, a low GH concentration might be expected in some ruminant species because of their low blood glucose levels (i.e., 40–60 mg/dl; Fahey and Berger, 1988). On the other hand, Alayash et al. (1988) concluded that glycosylation of hemoglobin in domestic ruminants occurred similarly to that in humans and thus could be useful in assessing long-term glucose status. Our objective was to determine baseline values for GH in whole blood and

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evaluate stability of GH relative to serum glucose in captive and wild white-tailed deer.

Serum glucose concentration and percent GH were determined in serum and whole blood of wild deer that were collected in August 1988 from study areas in McCurtain County, Oklahoma (USA; 34°15′ to 34°25′N, 94°45′ to 94°50′W), and Howard and Pike counties, Arkansas (USA; 34°10′ to 34°20′N, 94°05′ to 94°15′W and 34°15′ to 34°20′N, 90°40′ to 93°50′W, respectively). Temporal changes in levels of serum glucose and GH also were determined in serial blood samples collected on 6, 14, and 21 March 1989 from three captive deer maintained on a high protein (16 to 18%) diet at the Caesar Kleberg Wildlife Research Institute (Kingsville, Texas 78363, USA). Blood samples from wild deer were collected via cardiac puncture immediately after animals were neck-shot; captive deer were immobilized with xylazine hydrochloride prior to blood collection and samples were collected from the jugular vein. Whole blood for GH analysis was stored in Vacutainer (Becton Dickinson, Rutherford, New Jersey 07070, USA) tubes containing EDTA(K_3).

Glucose concentration in serum was determined using the o-Toluidine method of Faulkner and Meites (1982). Levels of GH in whole blood were determined as a percentage of total hemoglobin using a Pierce GlycoTest Kit 42100 (Pierce, Rockford, Illinois 61105, USA) and procedures that are recommended by the manufacturer. All samples were determined in duplicate; if duplicates differed by >5%, samples were reanalyzed. We used ANOVA and *t*-tests to (1) examine the temporal stability of serum glucose concentration and percent GH in captive deer, (2) compare GH of wild deer relative to an arbitrary difference in glycemic status, and (3) compare levels of serum glucose and GH among the three populations of wild deer. Collections of wild deer were conducted on three areas that differed in cattle stocking rate, and we hypothesized that deer from the three areas differed in nutritional condition. A priori hypothesis testing was used to compare levels of serum glucose and GH of the three deer populations.

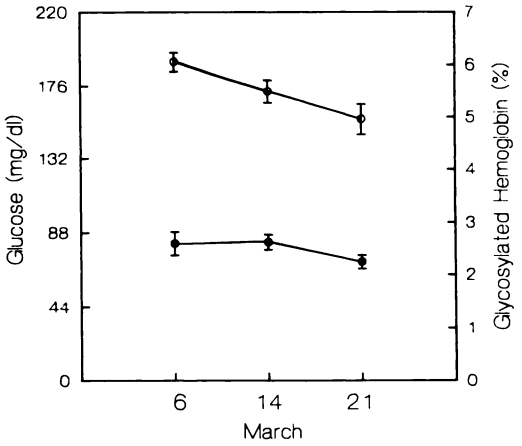


FIGURE 1. Percent glycosylated hemoglobin (solid circles) and serum glucose concentration (open circles) in blood samples collected on 6, 14, and 21 March 1989 from three captive white-tailed deer maintained at the Caesar Kleberg Wildlife Research Institute, Kingsville, Texas.

Serum glucose and GH levels of captive deer averaged 173.0 ± 6.2 mg/dl and 2.5 ± 0.1%, respectively. No significant correlation ($F = 2.87$, df = 1,7, $P = 0.13$) was found between serum glucose concentration and percent GH. Average serum glucose concentration of captive deer decreased from 190.4 to 155.8 mg/dl from 6 to 21 March ($F = 5.46$, df = 2,6, $P = 0.04$) (Fig. 1). However, percent GH remained stable ($F = 1.50$, df = 2,6, $P = 0.30$) over the 2-wk period. Because ration composition and dry matter intake of deer did not change, other than transient xylazine-induced anorexia (Warren et al., 1981), variation in serum glucose was unexpected and may have resulted from unknown stresses affecting deer during sample collection.

We collected 17 whole blood samples from deer on the three study areas and samples were transported to our laboratory for analysis. Serum glucose and GH levels of wild deer averaged 119.9 ± 8.4 mg/dl and 3.7 ± 0.4%, respectively. As with the captive deer, no significant correlation (*F*

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= 1.08, df = 1,15, P = 0.32) was found between serum glucose concentration and percent GH in whole blood. To evaluate stability of GH relative to serum glucose concentration of wild deer, we ranked glucose concentration and divided the 17 deer into 2 groups (high and low state of glycemia) based on the median glucose value for the sample. Deer with serum glucose concentrations >117.0 mg/dl were arbitrarily categorized as high glucose; deer with concentrations below this level were categorized as low glucose. This separation resulted in a significant difference (t = 4.58, df = 15, P = 0.0001) in serum glucose concentration. However, no difference (t = 1.25, df = 15, P = 0.23) was found in percent GH for the 2 groups of deer. We concluded that the concentration of serum glucose in wild deer was more variable than percent GH; this conclusion paralleled our captive study.

No difference (F = 0.128, df = 1,14, P = 0.72) was found in serum glucose concentration among the three deer populations (Fig. 2). However, percent GH was significantly higher (F = 5.33, df = 1,14, P = 0.04) in deer collected from Pike County, Arkansas, compared to deer from other study areas. The sample size for Pike County was small (n = 3) and thus, may not have been representative of that population. However, the difference in percent GH corroborated a difference in femur marrow fat (F = 12.14, df = 1,56, P = 0.001); Pike County deer had a higher level than deer from the other study areas (J. A. Jenks, unpubl. data).

Alayash et al. (1988) found that average percent GH in domestic sheep (3.2%) and goats (4.0%) was significantly lower than in humans (4.9%) but was above levels that have been reported for wild ruminants (Richter, 1986). In our study, mean percent GH of captive deer (2.5%) was below levels that have been observed in whole blood of sheep and goats (Alayash et al., 1988); however, GH in wild deer (3.7%) was similar to sheep and goats. The range of GH (1.6 to 5.4%) observed in our study

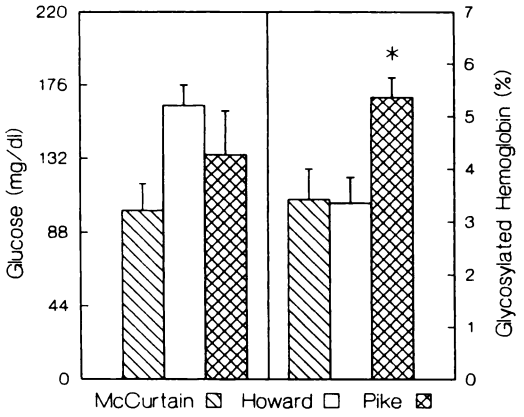


FIGURE 2. Percent glycosylated hemoglobin and serum glucose concentration in blood samples collected from wild white-tailed deer in McCurtain County, Oklahoma, and Howard and Pike counties, Arkansas in August 1988. Sample size was 5, 9, and 3 for McCurtain, Howard, and Pike counties, respectively. Asterisk denotes significant difference (P = 0.04).

was above average levels of GH for wild ruminants that were observed by Richter (1986).

Average serum glucose concentration has been related to percent GH (Koenig et al., 1976). Lack of significant correlations between serum glucose and GH in our study indicated that neither the single estimate of serum glucose concentration for wild deer nor the three estimates for the captive deer could be equated with average serum glucose. Glucose variability may have resulted from our blood collection methods or an unknown stressor affecting deer. Such instability in glucose concentrations renders these estimates unusable in assessing the glycemic status of deer. On the other hand, percent GH appeared to remain stable in both captive and wild deer and thus, provided a more accurate assay for assessing long-term glycemic status of deer.

Our study was not designed to compare levels of GH for captive and wild deer, but lower values of GH for captive (2.5%) relative to wild deer (3.7%) may have resulted from the difference in season in which blood samples were collected. Captive deer were sampled in March when some depression in metabolic rate may

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have resulted in a lowered GH level, despite consumption of a high protein diet. Conversely, wild deer were collected in August during a year with above normal fruit production, which may have resulted in maximized GH levels.

Future research on GH in deer should focus on the identification of factors (e.g., immobilization, seasonal variation) that can affect GH levels. Knowledge of these factors would enhance the diagnostic aspects of GH in assessing glycemic status of deer populations that are under differing nutritional constraints.

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