BLOOD UREA NITROGEN LEVELS OF WHITE-TAILED DEER AS AN INDEX OF CONDITION AND NUTRITIONAL INTAKE

by

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Thesis submitted to the Graduate Faculty of the

Virginia Polytechnic Institute and State University

in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

in

Wildlife Management

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July, 1974
Blacksburg, Virginia 24061

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ACKNOWLEDGEMENTS

I would like to express sincere appreciation to all members of my graduate committee for their guidance and helpful suggestions during the course of my study. Dr. Roy L. Kirkpatrick, committee chairman, provided invaluable advice, supervision, and encouragement throughout the study. Dr. Henry S. Mosby is thanked for his willingness to advise and make recommendations regardless of his always heavy workload. Thanks are extended to Dr. Patrick F. Scanlon for his interest shown and his advice given. Recognition is also due Dr. Harold E. Burkhart who, although not a committee member, provided statistical counseling when needed.

I am grateful to the Southeastern Cooperative Wildlife Disease Study group for providing serum samples from wild deer. I would also like to thank the Virginia Commission of Game and Inland Fisheries and the Radford Army Ammunition Plant personnel for their help in obtaining fawns.

Special thanks are offered to Wesley Abler who used the same fawns in his project and who devoted many hours of his time in rearing and sampling fawns. Thanks are also extended to the following graduate students who helped in collecting blood samples and caring for fawns; Edward Reed, Ralph Mirarchi, Bob Zepp, Mack Walls, Joe Lint, John Estep, Tom Jones, and several others.

Appreciation is extended to Wanda Morehead for her technical assistance in analysis of blood samples.

I am grateful to the Division of Forestry and Wildlife for providing financial assistance during my graduate study and the Virginia Cooperative Wildlife Research Unit for incurring many of the incidental costs created by the captive fawn project.

Finally, I would like to thank my fiancee, Nancy St. Clair for her typing the many drafts of my thesis, and for her patience, understanding, and encouragement during the latter part of my graduate career.

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INTRODUCTION

The classical objective of wildlife managers is to provide the public with the maximum number of hours of outdoor recreation by means of the wildlife resource without impairing that resource for future use. A biologist is continually concerned with the deterioration of wildlife populations and habitats. However, to evaluate populations and habitats from quantitative view is not sufficient; the quality or condition must also be evaluated if managers are to achieve their long-run objective. Any technique that would assist biologists in both quantitative and qualitative evaluations could further elucidate ecological-nutritional relationships and could help assure that neither wildlife populations nor habitat would be seriously impaired for future use.

Any reproducible structural or biochemical measurement that can be taken from an animal so that the results of such measurements can be related to the relative condition of the animal, the surrounding environment, or preferably both may be called a "physiological index." Most of the indices thus far developed have had the disadvantage of either being too subjective or requiring dead or dying animals. There is great need of such indices for the white-tailed deer (Odocoileus virginianus). The white-tailed deer has become one of the most popular game species in the United States, particularly in the Southeast. Populations

are increasing, interactions between humans and deer are intensifying, hunter pressure is increasing, and the demand for quantity and quality of animals is increasing.

An ideal index should be one that could be measured without injury to the animal. Blood parameters would seem to be logical measurements since deer could be immobilized (usually by remote injection of drugs) or live-trapped and released unharmed after drawing a blood sample. However, the problem with many blood parameters is their extreme variability and inconsistency. Blood urea nitrogen (BUN) levels seem to be a reasonably stable parameter for use as a physiological index.

The specific objectives of the present study were (1) to determine if BUN levels in white-tailed deer collected from several areas of the Southeast could be related to body weight, season, sex, age, subjective measurements of condition, and proximate analysis of rumen contents, and (2) to determine the influence of protein and energy levels on BUN values of captive deer.

LITERATURE REVIEW

Physiological Indices

Bailey (1969) compiled information on the use of rumino-reticular contents and blood constituents as measures of nutritional condition in deer. Twenty-one characteristics from 23 studies were examined. Deer in these studies were arbitrarily classified by Bailey (ibid.) into condition categories of either good, normal, or poor. Most of the measurements, when compared between studies, had extreme variation and lacked the sensitivity to differentiate between animals in the condition classes. This seemed especially true of such characteristics as erythocyte counts, packed cell volume, hemoglobin, blood glucose, serum proteins, and other serum biochemical and hematological values which are prone to change because of excitement, muscular activity, endocrine responses, or other reactions caused by environmental stressors.

Klein (1962) used Sitka deer (Odocoileus hemious sitkensis) rumen content analyses to test known differences in the range quality (determined by the forage analyses) between two islands in Alaska. Nitrogen percentage of rumen contents was significantly different between the two islands in three of four age groups tested and crude fiber percentages were significantly different in two of two age groups tested. It was concluded that rumen content analysis could be employed for evaluation of forage and range quality of wild ruminants. Brüggemann et al. (1968) also

concluded that rumen content analyses could be used to provide information on food selection and digestive processes.

Porterfield (1970) obtained serum from captive deer and wild deer and performed serum lipid and protein analyses, but failed to find sufficient evidence to support assessment of nutritional status of the deer from serum lipid and protein levels. Data from wild deer were analyzed by use of a stepwise multiple regression technique. Body weight was chosen as the dependent variable and independent variables were lipid fractions, protein fractions, albumin/globulin ratio, sex, age, condition index, and season. Data from captive deer were similarly analyzed except that mean weekly food consumption was the dependent variable. All independent variables remained the same except lipid fractions were excluded.

An index for assessment of nutritional status of the Columbian black-tailed deer (Odocoileus hemious columbianus) was developed by Bandy et al. (1956). Body weight was predicted first from heart girth and then body weight was predicted from hind foot length and a comparison of ratios of the two body weight estimates was made for individual deer. This index was based on the contention that skeletal growth and hind foot length, once attained, are not influenced as much as heart girth is by nutritional levels. Bandy et al. (ibid.) noted that even if the animal was on a low plane of nutrition, the heart girth would be influenced to a greater relative extent than would skeletal measurements. An

index value of 1 supposedly indicated a "normal" animal. A value less than 1 indicated poor condition, and a value greater than 1 indicated excellent condition.

Klein (1964) measured body weights, and femur and hind foot lengths from Sitka deer from two islands in Alaska, one of which had been shown previously by Klein (1962) to be higher quality habitat. Significant differences between islands in femur/hind foot ratios were found in three of four age groups. The author felt that femur/hind foot ratios are not as influenced by genetics as body weights and therefore are more reliable for evaluating nutritional condition than body weights.

A very detailed paper by Riney (1955) described different techniques for determining condition (total fat reserves) of red deer (Cervus elaphus). The measurements used were kidney fat index, back fat, abdominal fat, fat content and appearance of bone marrow, girth measurement, body weight, and antler characteristics. Data from 500 red deer showed kidney fat index to be the most satisfactory for detecting deer of different condition and from different environments.

Ransom (1965) plotted percent femur fat against a kidney fat index for 34 female white-tailed deer of different ages. The results showed that the kidney fat index could be used to assess conditions when the kidney fat index value was 30 or above, but where the index fell below 30, best evaluation could be achieved using percent femur fat. The conclusion was that kidney fat

reserves are not completely exhausted in animals before the initiation of femur fat mobilization and that kidney fat indices should be used for fat deer and femur fat indices for deer in poor condition. The fat content of tissue underlying the grinding teeth of the mandible was examined by Baker and Lueth (1966) as a possible index of condition. Moderate correlation coefficients (0.396, 0.478, and 0.507) were found between mandibular cavity tissue (MCT) fat content and femur fat, general condition class, and weight, respectively. Means of MCT fat content segregated deer with regard to poor and fair condition classes, but lacked the sensitivity to differentiate well between fair and good condition classes. The authors recommended further research to investigate the deposition and usage of MCT fat, to elucidate the mechanisms involved, and to improve the techniques and assays required, before the index could be implemented as a management tool.

Dauphine (1971) analyzed data from 411 barren-ground caribou (Rangifer tarandus groenlandicus) to determine changes in their physical condition. Condition was again used to mean total fat reserves. The data collected included body weights, back fat, abdominal fat, kidney fat, and fat content of femur marrow. The author concluded that kidney fat and abdominal fat were effective indices over the entire range of condition, but a better evaluation of condition could be achieved if all four indices were examined and evaluated collectively.

Two of the less scientific methods that have been used to estimate condition of deer have been visual observations and antler size. Riney (1960) described a subjective system for assessing condition of ungulates. The technique was based on symptoms of inanition exhibited by the animals and animals were classified by visual observation into good, medium, or poor condition classes. Antler size has long been used by sportsmen to reflect the "quality" of deer, and in most cases antler size and growth of antlers are indicators of nutritional intake (French et al, 1956; Long et al. 1959). Neither of these measurements are considered to be reliable indices of the nutritional status of deer because of individual animal variation and the subjectiveness involved.

Blood Urea Nitrogen

No literature reviewed dealt directly with the use of blood urea nitrogen (BUN) levels as an index of condition for deer and deer habitat, but several authors have provided supporting evidence that BUN levels could be used as a physiological index.

Lewis (1957) in his experiments with sheep, concluded that BUN levels were dependent on the diet of the animal and that BUN levels paralleled rumen ammonia levels after a delay of 4-8 hours. Preston et al. (1965) conducted experiments to determine if BUN levels could be related to protein intake in finishing lambs. In all experiments, BUN was significantly increased by increasing protein levels fed to lambs. Preston et al. (1961) in an earlier experiment showed similar results for steer calves.

Four Jersey steers were used in a nitrogen depletion-repletion experiment by Biddle and Evans (1973). The steers were placed on three consecutive 4 week periods to standarize, to deplete, and to replete body nitrogen. The protein level for the periods was 15, 5, and 14 percent, respectively. Plasma urea nitrogen levels were significantly lower in the depletion period than in either the standardization or repletion periods.

Ullrey et al. (1968) found BUN values to differ between white-tailed deer fed balsam fir and those fed white-cedar. BUN values for deer on the balsam fir diet decreased from pretreatment values while BUN values for deer on the cedar diet increased. Cedar has more crude protein than does balsam fir. Teeri (1958) sampled fawns on a 16 percent crude protein diet and showed that all fawns exhibited similar BUN values when on this diet. Seal and Erickson (1969) tested differences in BUN values of white-tailed deer of different sex and ages (adult male vs. adult female vs. juvenile male vs. juvenile female) but found no significant differences. Seal et al. (1972b) showed some seasonal differences in BUN values, but the deer on the high diet consistently exhibited higher BUN levels than did animals on the moderate diet. The high diet consisted of white cedar browse, ad libitum, supplemented with pelleted feed and the moderate diet consisted of white cedar browse only.

Franzmann (1972) working with bighorn sheep (Ovis canadensis) found the BUN values reflected protein intake. He also determined

that BUN values, unlike numerous other blood parameters, are not significantly affected by excitement. Seal et al. (1972a) also determined that BUN levels were significantly unaffected by restraint or certain immobilizing drugs in the white-tailed deer.

The literature has not been limited entirely to ruminants as work has been done also with monogastric animals. Fonnecbeck and Symons (1969) tested the effects of diet on BUN levels in horses and concluded that horses fed high protein diets had higher BUN levels than those fed low protein diets. Eggum (1970) utilized 17 feeding experiments with rats to conclude that a direct correlation existed between the protein content in the diet and BUN levels. He also showed that, in pigs, BUN levels increased for the first 3-4 hours after feeding and then reached a plateau. Dror and Bondi (1969) calculated a cubic prediction equation for BUN levels in rams based on protein as a percentage of the theoretical requirement for maintenance. Preston et al. (1965) calculated a prediction equation between BUN levels and protein intake for lambs. The correlation coefficient (r) had a value of 0.99. Eggum (op. cit.) calculated an equation for the same two variables in his study and found an r value of 0.95.

All of the above literature seems to substantiate that BUN levels are somewhat predictive and could feasibly be used as an index of animal or habitat condition for white-tailed deer.

Metabolic Pathways Leading to Urea Synthesis

A knowledge of protein metabolism, ammonia formation, and urea formation, excretion and recycling is fundamental to the understanding of BUN levels and pathways. The explanation below was taken from information presented by Hungate (1966), Allen (1970), Phillipson (1970), and Fontenot (1971).

Ruminants

Digestion breaks proteins into their basic building blocks, amino acids. Digestion is accomplished by protease enzymes cleaving peptide bonds of proteins to produce short-chained amino acids. The fate of these amino acids is twofold: (1) either they are absorbed through the rumen or lower gastro-intestinal (GI) tract (2) or they are subjected to the action of microbes present in the rumen. The amino acids in the former case are transported to deficient tissues and organs throughout the body via the systemic circulation. Amino acids in the latter case may be used by the microbial population for their own body protein or for energy purposes. End products of their activities include carbon dioxide, ammonia, and short-chained fatty acids. Ammonia is the principal product when considering urea synthesis. Ammonia is absorbed through the rumen wall where it enters the portal blood. Most of the absorbed ammonia is transported to the liver and converted to urea by the Krebs-Henseleit cycle. Urea levels may be measured in the systemic circulation. Rumen microbes are not able to synthesize and assimilate large amounts of dietary

protein into microbial protein, except when large amounts of energy are also available. As a result, large amounts of ammonia are produced when high levels of protein are digested. These high levels of ammonia result in high levels of ammonia in the systemic circulation. High energy levels would lower the levels of BUN since the high level of energy would allow the microbes to be more efficient in their utilization of dietary protein and less ammonia would be produced and thus less urea formed. An additional capability of ruminants is the ability to recycle urea and to use it as a nitrogen source during periods of low dietary protein intake. The urea that escapes urinary excretion is either absorbed directly through the rumen wall or is introduced into the rumen via saliva. Houpt (1959) showed that urea was recycled and that energy sources had an influence on recycling. Supplemental carbohydrates increased the amount of urea recycled in sheep on a low protein diet when compared to sheep on a low protein diet with no supplemental energy sources. Systemic blood levels of urea would be lowered in animals which were on low protein diets, not only because of low ammonia production, but also possibly because of the amount of urea removed by the recycling mechanism.

Nonruminants

Protein digestion in nonruminants is basically the same process as for ruminants except that all absorption of free amino acids occurs through the small intestine and lower in the

GI tract. Amino acids in excess of the body requirements are catabolized for energy by the liver with an end product of ammonia. The resulting ammonia is converted to urea in the liver by the Krebs-Henseleit cycle and excreted in the urine by the kidney. BUN levels are also detectable in nonruminants.

MATERIALS AND METHODS

Captive Deer

Fawn Procurement

Twelve female fawns were captured at the Radford Army Ammunition Plant near Radford, Virginia during the spring of 1973 using the method described by Downing and McGinnes (1969). Twelve additional female fawns were obtained from the Virginia Commission of Game and Inland Fisheries and were from various regions of the state. Five additional female fawns were born to captive does in the V.P.I. & S.U. deer pen facilities. All fawns were successfully reared to 3½ months of age using techniques described by Buckland et al. (1974).

Experimental Design

The design used in this study was a 2x2 factorial with energy and protein as the factors. High and low levels of both energy and protein produced four treatments: high energy-high protein (HEHP), high energy low protein (HELP), low energy-high protein (LEHP), and low energy-low protein (LELP). Ullrey et al. (1970) reported daily digestible energy maintenance requirements for deer to be 158 kcal per kg body weight.$^{0.75}$ Our objective was to formulate high energy rations to contain twice the amount of daily energy required to maintain a 45.4 kg deer or approximately 5597 kcal. The basis for this was to assure that the fawns were provided enough energy for both maintenance and growth. Daily

food consumption per deer was estimated to be 1.82 kg. After considering food consumption, high energy rations were formulated to contain 3080 kcal/kg. It was decided to try to achieve a 30 percent restriction in energy, i.e. provide 2156 kcal/kg for low energy rations. The basis for this was to restrict the animals enough to show differences in treatments, but not to restrict the animals to such an extent to retard their growth for future studies. The goal for high protein rations was in the range of 15-20 percent and the goal for low protein rations 7-11 percent. Ullrey et al. (1967) reported that female fawns were capable of maintenance and growth on a 7.8 percent crude protein diet, although the growth was less than for fawns on higher protein diets.

Formulation of acceptable rations proved to be more difficult than was originally expected. The first formulation of rations (Table 1) was fed beginning on 15 September, but was not suitable. The deer, especially the ones on low energy rations, did not eat and began deteriorating in condition. It was thought the amount of straw in the rations was undesirable and not palatable to the deer. On 26 September, low energy rations were reformulated to contain more orchardgrass hay and no straw. After this change, low energy animals continued to decrease food consumption and to decline in condition. Until this time, the rations had been ground, but not pelleted. It was decided to pellet the rations to increase palatability and to eliminate any selective feeding

Table 1. Formulation of original rations fed to captive fawns.

Constituent	High energy High protein (percent)	High energy Low protein (percent)	Low energy High protein (percent)	Low energy Low protein (percent)
Alfalfa meal	36.0	0.0	0.0	19.8
Soybean oil meal	14.0	0.0	31.8	0.0
Orchardgrass	4.8	30.0	0.0	30.0
Straw	0.0	2.8	56.0	30.0
Corn	33.0	55.0	0.0	8.0
Molasses	9.0	9.0	9.0	9.0
PO_4	0.7	0.7	0.7	0.7
Mineralized Salt	1.5	1.5	1.5	1.5
Vitamin premix	1.0	1.0	1.0	1.0

that may have occurred earlier. On 11 October, unused amounts of the ground high energy rations were taken to the mill and pelleted. Unused amounts of the low energy diets were discarded. New low energy ration formulations were also pelleted. These new low energy rations contained neither straw nor orchardgrass, but corncobs were substituted as roughage and filler. Both low energy diets increased in percent crude protein and digestible energy as a result of these changes, but the rations proved to be more palatable. The final change in rations was made on 25 October when all but the low energy-high protein rations were reformulated. This final formulation (Table 2) was an attempt to make percent crude protein and digestible energy more equitable between protein treatments and energy treatments. Proximate analyses and digestible energy values of the initial formulation and the final formulation are presented in Table 3 and Table 4, respectively.

Actual restriction in energy was only 18 percent as opposed to the desired 30 percent. All digestible energy values for the constituents are based on sheep digestive values taken from tables in Crampton and Harris (1969).

All animals were weighed on 14 September 1973. The eight heaviest animals were assigned to Group I and started on the experimental feed regimes the following day. Within the group, animals were assigned randomly to one of four treatments with treatments assigned randomly to pens. The remaining fawns were weighed on 28 September, the eight heaviest animals assigned to

Table 2. Formulation of final rations fed to captive fawns.

Constituent	High energy High protein (percent)	High energy Low protein (percent)	Low energy High protein (percent)	Low energy Low protein (percent)
Alfalfa meal	30.8	15.0	46.8	32.8
Soybean oil meal	16.0	0.0	16.0	0.0
Corn	35.0	52.8	0.0	15.0
Corn cobs	6.0	20.0	25.0	40.0
Molasses	9.0	9.0	9.0	9.0
PO_4	0.7	0.7	0.7	0.7
Mineral salt	1.5	1.5	1.5	1.5
Vitamin premix	1.0	1.0	1.0	1.0

Table 3. Proximate analysis and calculated energy values for original rations.

Component	High energy High protein (percent)	High energy Low protein (percent)	Low energy High protein (percent)	Low energy Low protein (percent)
Dry matter	88.61	57.04	87.66	87.82
Crude protein	17.00	9.25	18.19	9.44
Ether extract	2.36	2.93	1.18	1.96
Crude fiber	16.62	15.44	27.02	28.47
Ash	8.63	6.80	9.10	9.26
Nitrogen free extract	55.39	65.58	44.51	50.87
Calculated digestible energy values (kcal/kg)	3025	3052	2180	2182

Table 4. Proximate analysis and calculated energy values for final rations.

	High energy High protein (percent)	High energy Low protein (percent)	Low energy High protein (percent)	Low energy Low protein (percent)
Dry matter	87.95	86.46	87.83	86.90
Crude protein	18.19	9.19	18.19	10.00
Ether extract	1.66	1.99	2.18	1.28
Crude fiber	15.15	12.92	21.41	23.73
Ash	7.83	4.90	9.41	7.56
Nitrogen free extract	57.17	71.00	48.81	57.43
Calculated digestible energy values (kcal/kg)	3052	3075	2507	2489

Group II, randomized as above, and begun on experiment 29 September. Group III, comprised of the next eight heaviest animals, was randomized and started on experimental rations 14 October.

All animals within the same group, assigned to the same treatment, were housed two per pen. Groups also represented age differences; Group I contained the oldest fawns, Group III contained the youngest fawns, and Group II was intermediate. Grouping was necessary to reduce the possible variation in treatment effects due to different initial body weights and/or ages.

Food Consumption

Daily food consumption was recorded for each pair of animals within treatments. These values were averaged on a monthly basis to give mean average daily consumption per month. Since some animals within treatments died during the experiment, some values represented consumption data for one fawn only.

Sampling and Weighing

All experimental animals were bled and weighed one day prior to being placed on experimental rations, and every 4 weeks thereafter for a period of 24 weeks. The data included 1 set of pre-experimental data, 5 sets of experimental data, and 1 set of post-experimental data for each animal.

Each fawn was physically restrained and blood sampling was accomplished by jugular phlebotomy using Vacutainers (Becton-Dickinson). Fawns were always bled prior to weighing and during

the morning hours to minimize the effects of excitement and diurnal variation. Body weights were obtained by use of a modified platform scale.

BUN Analysis

Serum samples were analyzed for BUN by a diacetyl monomixe procedure (Sigma Chemical Company, 1971). Concentrations of BUN were determined by use of a Bausch and Lomb Spectronic 20 colorimeter. Results from preliminary samples that were run in duplicate indicated that the procedure was accurate and reproducible and therefore future samples were not run in duplicate. A standard curve was established over the range of BUN values expected. A blank solution and at least three standard urea solutions were run with each set of serum samples to assure the validity of the assay. If the standards were not consistently in agreement with corresponding values on the curve, a new set of standards was prepared and a new standard curve established. Consistency in pipetting was accomplished by using transfer pipets for the reagents and Eppendorf microliter pipets for making dilutions.

Data Analysis

Initially, the sample size of the experiment was 24 animals, but through the course of the experiment six animals were lost due to illnesses and accidents. Of these six animals, one was euthanized just prior to the last sampling period. A final experimental BUN value was calculated for this animal by the method of missing data calculation described by Snedecor (1956).

One degree of freedom was subtracted when the test mean square was computed in order to compensate for the calculated value. No missing data values were calculated for the weight gains analysis, since it was thought that weights could not be justifiably predicted because of the lack of consistency in weight gains between animals in the same treatment. Values from the other animals lost were incomplete and were omitted from the BUN and weight gains analyses.

Serum BUN values were analyzed by analysis of variance using a least squares regression procedure from the Statistical Analysis System (SAS Barr and Goodnight, 1971). Sources of variation were partitioned into main effects (group, energy, and protein) and their interactions and monthly values and their interactions. A different mean square term was used to test main effects than was used to test monthly effects. The former test term was an estimate of the variation in BUN values between animals on the same treatment and was an estimate of the variation in BUN values between animals on the same treatment in the same pen during the same month and was designated by month x animal (pen). The tests were based on expected mean square values which were determined by Burkhart (pers. comm.) for the model of analysis, which was considered to be purely random.

Monthly weight gains were also analyzed by analysis of variance using the SAS package. Group, energy, protein and their interactions were the sources of variation.

Wild Deer

Serum samples from wild deer were provided by the Southeastern Cooperative Wildlife Disease Study (SCWDS) group stationed at the University of Georgia, Athens, Georgia. Deer were collected as part of a disease research project. All animals were shot with high powered rifles without regard to sex, age, or condition. Blood was taken, processed for sera, and then frozen for future use. Five deer were killed during each season in six areas of the southeastern United States. During the first year (1967-68) of the study, collections were made from one area each week for six successive weeks during each season in the following order: (1) A. P. Hill Military Reservation in Virginia, (2) Forks Game Management Area in South Carolina, (3) Chocolocco Game Management Area in Alabama, (4) Daniel Boone Game Management in North Carolina, (5) Eglin Air Force Base in Florida, and (6) Fort Stewart Military Reservation in Georgia. During the last two years (1968-69, 1969-70) of the study, deer were again collected seasonally, but the order of sampling was random. Daniel Boone Game Management Area and Eglin Air Force Base were not sampled during the third year. In all years the spring collections were made between mid-April and late May, the summer collections between mid-July and late August, the fall collections between mid-October and late November, and the winter collections between mid-January and late February.

A visual estimate of physical condition was made for each deer at the time of necropsy. The condition was classified as excellent,

good, fair, or poor and were later coded for statistical purposes as 4, 3, 2 and 1, respectively.

BUN Analysis

Analysis of serum for BUN was identical to the procedure used for captive deer.

Data Analysis

BUN values were analyzed by analysis of variance using the SAS package previously described. Three areas had one missing sample, however; the BUN values were estimated by substituting the mean of the respective subgroup for the missing value. One degree of freedom was subtracted from the appropriate error mean square for each missing sample. Two areas were not sampled during the third year of the study, and the data were analyzed by area to prevent a confusing of the results. An analysis of variance table was produced for each area with year and season as main effects.

Partial correlations, by season, were also determined between BUN values and other variables that were available on the individual deer. This was accomplished by using an option of the SAS package. Kirkpatrick et al. (1969) had previously obtained rumen samples from the SCWDS for the first year deer and had conducted proximate analyses and food habits analyses. Analyses were later performed on deer collected during the second and third year (unpubl.). Age, weight, and physical condition data were also available for correlation.

RESULTS

Captive Deer

Food Consumption

Food consumption data are presented in Table 5. No statistical analysis was performed. After the first month of the study, all animals, regardless of group or treatment, decreased their food consumption. Animals on low energy diets tended to eat slightly more than animals on high energy diets, but overall mean food consumption seemed to be equitable between groups and between treatments.

Weight Gains

Mean weight gains are presented in Table 6. The mean squares from the analysis of variance of these data are presented in Table 7. Mean weight gains for animals on high protein rations (2.21 kg) were significantly higher ($P < .05$) than those of animals on low protein rations (1.55 kg). There were no significant group or energy effects and no significant interactions. All sources were tested with the estimate of variance between animals on the same treatment in the same pen which was denoted animal (pen).

BUN Values

The overall mean BUN value for fawns before the experiment began was 17.4 mg/100 ml. Pretreatment group BUN means (mg/100 ml) were as follows: Group I, 13.7; Group II, 21.2; and Group III, 17.4 Post-treatment group BUN values (mg/100 ml) in the same above order were 18.4, 23.1, and 19.6. The overall mean was 20.4 mg/100 ml.

Table 5. Average food consumption (kg/deer/day) of captive fawns on experimental rations.

Treatment	Month of experiment 1	2	3	4	5
Group I					
HEHP[b]	--	1.61	1.38[a]	1.07[a]	0.57[a]
HELP[c]	--	1.77[a]	1.43[a]	0.87[a]	0.72[a]
LEHP[d]	--	1.75[a]	1.66[a]	1.28[a]	0.94[a]
LELP[e]	--	1.70	1.69	1.38	1.03
Group 2					
HEHP	1.36	1.57	1.30	1.07	0.93
HELP	1.30	1.33	1.10	1.06	0.64[a]
LEHP	1.26	1.42	0.90	1.34[a]	0.84[a]
LELP	1.28	1.60	1.38	1.13	0.93
Group 3					
HEHP	1.27	1.27	1.04	0.80	0.92
HELP	2.03	0.95	0.84	0.60[a]	0.85[a]
LEHP	1.13	1.20	1.08	0.95[a]	1.15[a]
LELP	1.13[a]	1.36	1.14	0.86	1.14
Overall monthly means					
HEHP	1.31	1.48	1.24	0.98	0.81
HELP	1.67	1.35	1.12	0.84	0.74
LEHP	1.20	1.46	1.21	1.19	0.98
LELP	1.20	1.60	1.40	1.12	1.03

Overall treatment means

HEHP	1.16
HELP	1.14
LEHP	1.21
LELP	1.27

a Values are for one fawn only; other values are average values for two fawns.
b High energy-high protein.
c High energy-low protein.
d Low energy-high protein.
e Low energy-low protein.

Table 6. Mean weight gains (kg) of captive fawns on experimental rations for a 20 week period.[a]

Treatment	Group 1	Group 2	Group 3	Overall mean
HEHP[c]	(1)[b] 2.04	(2) 2.81	(2) 1.70	(5) 2.21
HELP[d]	(1) 2.32	(2) 2.10	(2) 1.15	(5) 1.76
LEHP[e]	(1) 2.46	(1) 2.06	(1) 2.12	(5) 2.21
LELP[f]	(2) 1.68	(2) 1.51	(1) 0.28	(5) 1.33

a Animals were weighed every 4 weeks.
b Animal sample size.
c High energy-high protein.
d High energy-low protein.
e Low energy-high protein.
f Low energy-low protein.

Table 7. Mean square from the analysis of variance of weight gains for captive deer.

Source	Degrees of freedom	Mean square
Group	2	5.88
Energy	1	2.24
Protein	1	9.57*
Group x energy	2	0.62
Group x protein	2	1.42
Energy x protein	1	2.66
Group x energy x protein	2	1.12
Animal (pen)	6	1.32

* ($P < .05$).

These latter values were obtained after placing all fawns on a standard ration for one month after feeding of experimental rations was complete.

Mean BUN values are presented in Table 8. The mean squares from the analysis of variance of these data are presented in Table 9. BUN values were significantly lower ($P < .01$) for animals in high energy groups (16.4 mg/100 ml) than in low energy groups (17.5 mg/100 ml). Protein had an opposite effect. Animals on high protein rations (23.0 mg/100 ml) exhibited significantly higher ($P < .001$) BUN values than did animals on low protein, rations (11.3 mg/100 ml). There was no significant group effects or main effect interactions when testing with animal (pen) as the error term.

The remaining sources of variation were tested with the appropriate term of month x animal (pen). The only source that was significant was the interaction between month and protein ($P < .001$).

Wild Deer

Seasonal means of BUN values, by area, are presented in Table 10. Results from overall analyses (not considering areas separately) included significant three-way interactions which could not be easily explained. Analysis by area removed one component of the interaction and made interpretation less difficult.

The results from the analysis of variance, by area, of BUN values for deer collected by the SCWDS are presented in Table 11.

Table 8. BUN means (mg/100 ml) of 19 captive fawns on experimental rations for a 20 week period.

Treatment	Sample	Months of Sampling 1	2	3	4	5
Group 1						
HEHP[a]	(2)	22.0	26.0	20.5	19.5	20.5
HELP[b]	(1)	5.3	6.5	8.0	10.0	7.5
LEHP[c]	(1)	32.3	28.5	25.0	24.5	24.5
LELP[d]	(2)	9.5	10.8	11.5	15.0	10.8
Group 2						
HEHP	(2)	30.5	20.5	17.3	18.8	19.2
HELP	(2)	6.5	7.0	10.0	11.2	17.5
LEHP	(1)	30.5	31.0	24.0	24.5	23.0
LELP	(2)	12.8	9.8	18.5	17.0	15.8
Group 3						
HEHP	(2)	21.8	20.8	21.5	25.2	25.8
HELP	(2)	5.8	11.8	10.8	13.2	8.5
LEHP	(1)	27.5	20.0	24.0	20.0	18.5
LELP	(1)	13.5	10.0	13.0	13.5	10.5
Overall monthly means						
HEHP	(6)	24.7	22.4	19.8	21.2	21.8
HELP	(5)	6.0	8.8	9.9	11.8	11.9
LEHP	(3)	30.1	26.5	24.3	23.0	22.0
LELP	(5)	11.6	10.2	14.6	15.5	12.7

Overall treatment means

HEHP	(30)	22.0
HELP	(25)	9.7
LEHP	(15)	25.2
LELP	(25)	12.9

a High energy-high protein.
b High energy-low protein.
c Low energy-high protein.
d Low energy-low protein.

Table 9. Mean square from the analysis of variance of BUN values for captive deer.

Source	Degree of freedom	Mean square
Group	2	20.35
Energy	1	236.47**
Protein	1	3330.92***
Group x energy	2	40.39
Group x protein	2	27.59
Energy x protein	1	0.40
Group x energy x protein	2	10.25
Animal (pen)	7	17.21
Month	4	5.82
Month x protein	4	72.86***
Month x energy	4	16.02
Month x group	8	6.34
Month x energy x protein	4	2.64
Month x group x protein	8	18.61
Month x group x energy	8	10.63
Month x group x energy x protein	8	9.31
Month x animal (pen)	27[a]	8.30
Total	93	

** ($P < .01$).
*** ($P < .001$).
a One degree of freedom subtracted for missing data calculation.

Table 10. Seasonal means of BUN values (mg/100 ml) for deer collected from selected areas in the Southeast, 1967-1970.

Year	Season			
	Spring	Summer	Fall	Winter
	A. P. Hill Military Reservation, Virginia			
1[a]	13.2	5.7	6.7	12.5
2[b]	14.1	7.4	9.1	8.5
3[c]	12.1	11.5	20.7	12.1
	Forks Game Management Area, South Carolina			
1	10.6	11.1	8.5	14.2
2	9.1	10.0	16.3	13.8
3	14.2	2.8	6.8	9.2
	Chocolocco Game Management Area, Alabama			
1	6.9	12.0	4.9	13.6
2	12.4	3.9	7.0	14.2
3	4.1	9.4	7.5	7.8
	Daniel Boone Game Management Area, North Carolina			
1	14.2	13.2	4.7	3.7
2	7.9	13.2	4.9	4.8
	Eglin Air Force Base, Florida			
1	8.1	7.7	3.4	11.6
2	10.6	8.3	6.3	8.1
	Fort Stewart Military Reservation, Georgia			
1	6.3	7.1	9.6	20.9
2	6.8	5.5	9.9	8.1
3	9.2	8.2	5.9	6.7

a 1967-68.
b 1968-69.
c 1969-70.

Table 11. Mean squares of BUN values for deer collected from selected areas of the Southeast 1967-70.

Area	Source of Variation			
	Year	Season	Year x Season	Error
A. P. Hill Military Reservation, Virginia	(2)[a] 132.33***	(3) 68.40**	(6) 73.80***	(47) 12.92
Forks Game Management Area, South Carolina	(2) 86.55*	(3) 53.29	(6) 72.11**	(48) 22.11
Chocolocco Game Management Area, Alabama	(2) 31.18	(3) 79.42***	(6) 71.82***	(48) 71.45
Daniel Boone Game Management Area, North Carolina	(1) 15.62	(3)200.74***	(3) 28.91	(32) 10.71
Elgin Air Force Base, Florida	(1) 3.91	(3) 50.56*	(3) 21.42	(32) 11.64
Fort Stewart Military Reservation, Georgia	(2) 79.26	(3) 75.48	(6) 91.34*	(46) 37.42

a Degrees of freedom are in parentheses to the left of each mean square.
*** ($P < .001$)
** ($P < .01$)
* ($P < .05$)

There were significant differences in BUN values between years in A. P. Hill Military Reservation ($P < .001$), and Forks Game Management Area ($P < .05$). Significant differences between seasons were found for A. P. Hill Military Reservation ($P < .01$), Chocolocco Game Management Area ($P < .001$), Daniel Boone Game Management Area ($P < .001$), and Eglin Air Force Base ($P < .05$). However, there were significant year x season interactions in four of the six areas.

Partial correlation coefficients, by season, between BUN values and other variables are presented in Table 12. BUN values for deer collected in the spring were significantly correlated with age (r=0.32), and weight (r=0.28). During summer significant correlations were found between BUN and crude protein (r=0.33), ash (r=0.26), unidentifiable finely ground matter (r= -0.31), and fungi (r=0.35). During fall, BUN values were significantly correlated with weight (r=0.36), crude protein (4=0.44), ether extract (r= -0.28), crude fiber (r= -0.27), fungi (r=0.25), and soft fruit (r= -0.27). BUN values in winter were correlated with ash (r= -0.23), nitrogen free extract (r= -0.29), and unidentifiable finely ground matter (r=0.26).

Table 12. Partial correlation coefficients, by season, between BUN values and several other variables of deer collected from selected areas of the Southeast, 1967-70.

Variable	Season			
	Spring	Summer	Fall	Winter
Characteristics				
Age	0.32**	-0.16	0.11	0.09
Weight	0.28*	0.00	0.36**	0.07
Condition	-0.18	-0.06	-0.09	0.17
Proximate analysis				
Dry matter[a]	0.19	-0.05	-0.04	0.10
Crude protein	-0.11	0.33**	0.44***	0.14
Ether extract	0.02	-0.04	-0.28*	-0.18
Crude fiber	0.05	-0.18	-0.27*	-0.23***
Ash	-0.08	0.26*	0.14	0.61
Nitrogen	0.09	-0.27	-0.02	-0.29*
Food habits				
Ufgm[b]	-0.09	-0.31*	-0.09	0.26*
Leaves of woody plants	0.09	-0.11	-0.20	0.02
Herbaceous stems and leaves	-0.05	0.16	-0.05	0.04
Acorns	0.08	0.08	0.06	-0.11
Fungi	-0.22	0.35**	0.25*	-0.12
Grasses and sedges	0.16	-0.20	0.12	0.15
Soft fruit	-0.12	-0.19	-0.27*	-0.12
Woody stems and buds	-0.11	-0.03	-0.08	-0.01
Hard fruit	0.02	-0.09	0.13	-0.04

a This variable and all remaining variables are expressed as percentages.
b Unindentifiable finely ground matter.
* ($P < .05$).
** ($P < .01$).
*** ($P < .001$).

DISCUSSION AND CONCLUSIONS

The decline of food consumption throughout the study may have been a result of voluntary food restriction by the animals. Voluntary food restriction in deer has been documented by Long et al. (1965). Another explanation is that animals on low energy rations may have eaten more to make up for the deficit in energy, but is not likely since rations did not differ as much in energy as was originally planned. Although no statistical analysis was performed, it appears that the quantity of food consumed was not a significant source of variation for the BUN data.

The effect of protein on weight gains of fawns was similar to results reported by Ullrey et al. (1968). In his study, fawns that consumed high protein diets weighed more than fawns on low protein diets.

Protein is important to the growth of young animals and this was verified by the weight gains analysis. Energy probably also has an effect, but the effect may have been masked, since the experimental design had only an 18 percent restriction in energy intake, whereas there was about a 50 percent restriction in protein intake.

BUN values for fawns on experimental rations compared favorably with values reported by Tumbleson et al. (1968) and Seal and Erickson (1969). Some of the results from the analysis of variance of BUN values were as expected and agreed with the

results of Preston et al. (1961) who used a similar experimental design with steer calves. Their rations consisted of different levels of energy and different levels of protein. Steers on high energy rations, when compared to steers on low energy rations, exhibited lower BUN values. Animals on high protein rations, when compared to animals on low protein rations, exhibited higher BUN values. High energy intake allows the microbial population of ruminants to be more efficient in their utilization of protein and not as much ammonia is produced and converted to urea as would be produced with low energy diets. If energy intake is constant, animals on high protein diets would have greater BUN values. High protein intake results in a greater production of ammonia and larger amounts of urea which create a higher BUN concentration. If protein is constant, animals on low energy diets would have greater BUN values.

The significant month x protein interaction for captive fawn data could not be explained other than to say that the effect of protein intake on BUN values was not the same from month to month, and no pattern could be discerned.

Significant differences in BUN values of SCWDS deer between years in two areas may be attributed to differences in seasonal climatic conditions that occurred during the years. Differences in precipitation, temperature, and other climatic conditions could cause differences in the quantity and quality of vegetation available to deer and the nutritive intake would be reflected by the BUN

level. Annual fluctuations in climate and vegetation may have been fairly consistent in the four other areas, since no annual differences were shown in BUN levels.

Three of the four areas that had significant seasonal differences in BUN values exhibited conflicting patterns. Deer from A. P. Hill Military Reservation had lowest values during spring for all three years sampled. This agreed with the results of Skeen (1974) who determined BUN values to be higher for deer collected during spring than for deer collected during other seasons in Western Virginia. Deer from Daniel Boone Game Management Area had lowest values during winter and next to lowest during fall for both years sampled. Deer from Eglin Air Force Base exhibited lowest values during fall for both years sampled. No pattern could be determined for the remaining three areas. The significant year x season interactions in four of the six areas meant that seasonal differences in BUN levels, in addition to varying between areas, also varied within the same area between collection years. Some of these seasonal differences may be attributed to the seasonal occurrence of certain food items. Two examples follow. Succulent vegetation, which is relatively high in protein, is most prevalent in spring. Acorns, which are low in protein, but relatively high in energy, are present in greatest abundance in fall. Differing amounts of these and other foodstuffs which occur seasonally could logically alter protein and energy intake and the differing intakes would be reflected in

BUN levels. If BUN values are to be assumed a measure of nutritional regime, then it must be concluded that nutritional regime varied widely between and within the areas of the Southeast that were sampled.

Correlation coefficients and their significance may be misleading unless carefully examined. With a large enough sample size, significant correlations may be determined between almost any two variables. One must consider the coefficient of determination (r^2) of correlations to determine if biological significance was equal to statistical significance. The definition of an r^2 value is the amount of variation of the dependent variable (BUN value) that could be explained by its relationship with the independent variable. The basis for biological significance is left to individual researchers, but most would probably be confident in a correlation with an r^2 value of 0.50 (50 percent). This would mean a correlation coefficient (r) would have to have a value of 0.70 or higher before it was considered biologically significant. No correlations in this study were found to be of that magnitude. Some of the significant correlations were thought to be coincidental and were not explainable. The correlation of BUN value and age was considered a coincidental correlation since it was only significant in one season. The reason for this was unknown since other seasonal r values were very low and Seal and Erickson (1969) failed to fine age differences in BUN levels in their study. Body weight

was significantly correlated with BUN values during the spring and fall. The other two seasons had exceptionally low r values. Condition was not correlated with BUN values, but the results may have been misleading. Condition, a discreet variable (excellent, good, fair, or poor), was coded and entered into a correlation matrix. The results may be invalid since the other measures are continuous variables. Weight and condition are related and it follows that condition and BUN values should be related, especially where weight was found to be correlated with BUN values. It is thought by the author that a measure such as kidney fat index (a continuous measure of condition) would be significantly correlated with BUN values. It was expected that the correlation between BUN and crude protein of rumen contents, would be higher and more consistent than was found since BUN values had been shown in the former part of this study to be significantly affected by protein intake. The reason for low r values in the winter and spring was not known. Ether extract, which is a measure of fat, and crude fiber, which is an indirect measure of digestibility followed similar trends and both exhibited negative correlations with BUN level only during the fall. No explanation could be given. The correlation of BUN with ash content which was significant in summer and winter could not be explained. Nitrogen free extract, a measure of energy, had negative correlations with BUN values during three of the four seasons and the correlation was significant only in winter. The negative correlations of BUN and energy

follow the results of the captive deer results. No reasonable explanations or assumptions could be made for any of the statistically significant food habits correlations. A different type of analysis of these data may render further explanations. A multiple regression analysis, by season, would show what variables explain the largest amount of variation in BUN values. It should again be emphasized that correlations do not imply cause and effect relationships.

The opposite effects that protein and energy had on BUN values may make the use of BUN values undesirable as a physiological index, but is still believed that BUN values could be useful as a management tool to evaluate trends on a particular deer range from year to year and to compare and rank similar physiographic areas. Baseline data for an area should be collected over a range of conditions and subsequent BUN values could be compared with what was considered to be a normal or average year. One problem that could be encountered is the misinterpretation of a high BUN value. This could be a result of either a low energy-high protein diet or an extremely poor diet, which resulted in protein catabolism. It is not known at what level of dietary protein intake that deer begin catabolizing body protein, but the result is a high BUN level. Franzmann (1972) determined this level to be around five percent for bighorn sheep. He has suggested (pers. comm.) that experimental deer be placed on varying levels of protein intake to determine at what level catabolism occurs. Another variable, such as serum glutamic-

oxalacetic transaminase (SGOT), that could be measured and used in conjunction with BUN may help determine when catabolism occurs. Amino acids, in excess of what are needed for protein synthesis, cannot be stored and are degraded and the resulting α-keto acid is used for energy if the animal has need for energy. SGOT is responsible for catalyzing the transfer of an amino acid group of glutonic acid to form the resulting α-ketoglutaric acid (White et al. 1968). A high BUN value implies either a high protein intake and an excess of amino acids, some of which had been converted to urea or a high level as a result of protein catabolism. A high SGOT level would indicate that amino acids were in excess of normal intake and that some had been converted into energy producing compounds. A high level of both BUN and SGOT would verify that a higher than average BUN value was attributable to a high dietary protein intake and not to body protein catabolism. Tumbelson et al. (1968) reported a significant ($P < .01$) correlation (0.49) between the level of SGOT activity and serum BUN levels.

The age of the serum samples from the SCWDS may have influenced the BUN results. Samples were frozen for 4-7 years before analysis was begun. During this time if the samples were thawed and exposed to bacterial contamination, ureas may have been formed and some urea hydrolyzed to ammonia, thus decreasing the urea content. It is not known whether or not samples were exposed to freezing and thawing or contamination, but it is proposed that more field studies similar to the SCWDS

project be done to study the feasibility of using BUN levels as a management technique, and that the sera be assayed promptly to prevent possible deterioration.

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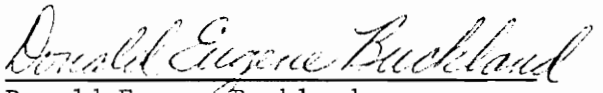

Donald Eugene Buckland

BLOOD UREA NITROGEN LEVELS OF WHITE-TAILED DEER AS AN INDEX OF CONDITION AND NUTRITIONAL INTAKE

by

Donald Eugene Buckland

(ABSTRACT)

Serum samples from 95 captive fawns on experimental rations and 317 samples from wild deer collected from six areas of the southeastern United States were analyzed for blood urea nitrogen (BUN) content by use of a diacetyl monoxime procedure.

Captive fawns were fed one of the following rations for a 20 week period: high energy-high protein (HEHP); high energy-low protein (HELP); low energy-high protein (LEHP); or low energy-low protein (LELP). Percent crude protein and calculated digestible energy values (kcal/kg) for the four rations were HEHP 18.2, 3052; HELP 9.2, 3075; LEHP 18.2; 2507; and LELP 10.0, 2489. Overall means for BUN values (mg/100 ml) for the four treatment groups were 22.0, 9.3, 25.2, and 12.8, respectively. Analysis of variance showed that high protein levels significantly increased BUN values ($P < .001$), while high energy levels significantly decreased them ($P < 0.01$). Analysis of variance for wild deer BUN values showed seasonal effects on BUN levels for four of the six areas, but no consistent seasonal trends were determined either between or within areas. Partial correlation

coefficients, by season, resulted in several significant correlations between BUN values and age, weight, physical condition, proximate analysis of rumen contents, and food habits. Although significant, many of the correlations were not explained and were considered to be coincidental.

The extreme variation in BUN values for wild deer was undesirable and the opposite effects that protein and energy had on BUN values for captive deer makes interpretation of nutritional intake difficult. For these reasons, BUN values may have limited use as a physiological index.