

SEASONAL RELATIONSHIPS OF THYROID, SEXUAL AND ADRENOCORTICAL HORMONES TO NUTRITIONAL PARAMETERS AND CLIMATIC FACTORS IN WHITE-TAILED DEER (*ODOCOILEUS VIRGINIANUS*) OF SOUTH TEXAS

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(Received 6 April 1983)

Abstract—1. Annual cycles of serum testosterone (T), estradiol 17- β (E), thyroxine (T₄), triiodothyronine (T₃) and cortisol (C) and their relationship with dry matter intake (DMI), ambient temperature and daylength (DL) were examined in four male and four female white-tailed deer.

2. Serum T in bucks correlated ($P < 0.05$) with DMI during the rut. In does, E correlated ($P < 0.05$) with DMI and body wt. Both serum T and E were DL and temperature dependent.

3. Serum T₄ in bucks correlated ($P < 0.05$) with body wt and increasing temperature while T₃ correlated ($P < 0.05$) with DMI and body wt. In does, T₄ was significantly ($P < 0.05$) correlated with DMI and body wt, while T₃ was not.

4. Serum C levels were not correlated ($P > 0.05$) with either DMI or body wt.

5. It appears that serum T₄ and T₃ in bucks and T₄ in does offer the best year-round indicators of nutritional stress in deer.

6. Male DMI was temperature and DL dependent. In does, only a short-term effect on DMI was found.

7. After the breeding season in bucks, and throughout the year in does, DL and temperature may control intake via the thyroid hormones.

INTRODUCTION

White-tailed deer (*Odocoileus virginianus*) in North America display a seasonal fluctuation in feed consumption and body wt (French *et al.*, 1960; Holter *et al.*, 1977; Moen, 1978). Both male and female deer voluntarily decrease their intake in the winter. It is believed that this hypophagia assists white-tails in reproduction and environmental adaptation (McMillin *et al.*, 1980). In boreal regions, feed consumption generally increases in the summer and is suppressed in adult males during the fall rut or breeding season. There is a short compensatory intake period after the rut, followed by restricted intake the remainder of the winter. In southern environments, there is no winter compensatory period, but there is an additional mid-summer intake suppression, probably due to high temperature and humidity (Short *et al.*, 1969; Wheaton and Brown, 1983). In addition to these geographical differences in feed intake, the latitude also affects the timing of the breeding season (Robinson *et al.*, 1963).

In south Texas, because of its more temperate climate, the deer have a more complex environment to which to adapt (i.e. mild winter cold stress but severe summer heat stress). Meyer (1982) reported that summer was more critical to these deer than winter in terms of the nutritional adequacy of the diet. Marked reductions in feed consumption by sheep and cattle

can be caused by high environmental temperatures or rations with a high calorogenic effect. Thus, thermodynamic mechanisms may dominate regulation of feed intake independently of other factors (Conrad, 1966).

The annual cycles of testosterone (T), estradiol 17- β (E), thyroxine (T₄), triiodothyronine (T₃), and cortisol (C) have been reported in deer (McMillin *et al.*, 1974; Bubenik *et al.*, 1975a; Bubenik *et al.*, 1977; Mirarchi *et al.*, 1981). Testosterone levels in bucks are highly correlated with the rut (McMillin *et al.*, 1974; Bubenik *et al.*, 1977; Mirarchi *et al.*, 1977) while E is the principal ovarian steroid regulating mating activity in does (Wade, 1972). In deer, T₄ and T₃ are metabolic hormones correlated with feed intake (Seal *et al.*, 1978). They also have been used as indicators of nutritional stress (Bahanak *et al.*, 1981). Cortisol is a main metabolic steroid in ruminants regulating protein and carbohydrate metabolism and also has been used as an indicator of general stress in deer (Bubenik *et al.*, 1977).

While McMillin *et al.* (1980) reported some hormonal relationships with hypophagia in northern white-tailed deer, no one has studied this phenomenon in southern white-tails. This project was designed to investigate the annual cycles of hormonal changes and their interrelationship with hypophagia and ambient climate in south Texas white-tailed deer.

MATERIALS AND METHODS

Four male and four female 1½-year-old white-tailed deer were housed individually in 5 × 5 m covered pens and fed a complete pelleted diet† (18.93% C.P. and 2.45 Kcal DE/g DM) *ad libitum* from 1 October 1979 to 30 September 1980. Average daily dry matter intake (DMI) was recorded weekly

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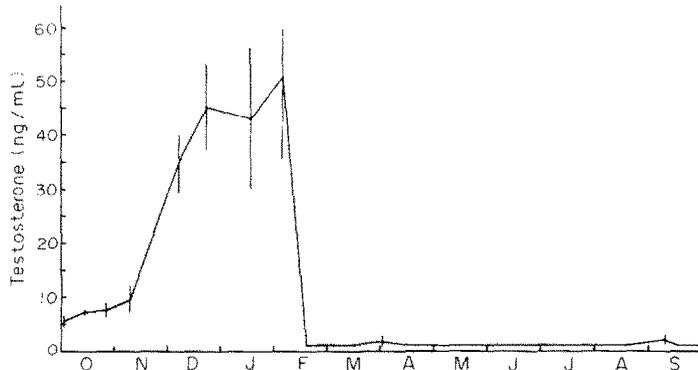


Fig. 1. Annual cycle of serum testosterone, mean (\pm SD) of four male yearling white-tailed deer.

while body wt was measured and blood samples were collected bi-weekly. For blood collection, the deer were tranquilized with xylazine hydrochloride (Rompun), and samples were taken from the jugular vein by venepuncture. After centrifugation, serum samples were stored at -20°C for further analysis.

Temperature data were taken from the climatological substation at Texas A & I University, Kingsville. Daylength (DL), rainfall and relative humidity (RH) data were provided by the U.S. Naval Air Station in Kingsville.

Testosterone, E, T_4 and T_3 were measured on duplicate samples using radioimmunoassays kits from Wien Labs, Inc.* while C was measured by an assay from Corning Medical Scientific Co.† The T and E assays utilized anti-hormone-protein complex antibodies and ^3H as a tracer. The T_4 and T_3 assays utilized anti- T_4 or T_3 protein complex antibodies and ^{125}I as a tracer. The C assay utilized anti-cortisol-protein complex antibodies and ^{125}I as a tracer.

Due to the sample size, bi-weekly hormonal levels and weekly intake data were pooled into six periods of 2-month duration and statistically analyzed by analysis of variance with a SPSS computer program. Pearson correlation coefficients were analyzed by deer and by sex to determine correlations between hormones, intake and climate data. The statistical significance level was set at $P < 0.05$.

Three climatological periods in the study were: Period 1, from October through January, as temperature and DL decreased; Period 2, from February through May, as temperature and DL increased; and Period 3, from June through September, as temperatures were generally greater than 27°C and DL decreased. In determining these periods, biological events were also considered, since the first period includes the rut or breeding season, the second period is the period of sexual quiescence, and the third period corresponds to the antler growth period in bucks. Since attempting to correlate blood values with contemporary environmental data might encounter substantial variance due to prior events (Seal *et al.*, 1972), lagged climatic data were also correlated with bi-weekly blood parameters in addition to divided periods. Temperature and DL from the previous 1–7 weeks was lagged to examine short-term (1–2 week) and long-term (7 week) effects on hormonal changes and DMI (i.e. T1 was average temperature data from current week, T2 was averaged temperature data from current and previous week, etc.).

RESULTS

Annual cycles in serum hormone levels

Serum T in bucks (Fig. 1) increased slightly from mid

to late autumn, then increased dramatically and peaked (50.6 ng/ml) during late winter. In February, it dropped abruptly to a baseline (1.0 ng/ml) until autumn. The changes of T levels confirm the study of testicular changes in Texas by Robinson *et al.* (1963) in which changes lagged 2 months behind those of Northern white-tails (Mirarchi *et al.*, 1977; McMillin *et al.*, 1980).

Serum E levels in does (Fig. 2) increased gradually from October until early winter, then dropped abruptly to the nadir in late winter (2.0 pg/ml) and early spring (4.0 pg/ml). The level then increased gradually until autumn except for a short interruption during early summer.

Serum T_4 levels (Fig. 3) in bucks decreased in late autumn and remained low through the winter. An increase occurred in April, and then the levels dropped again in July and were low through September. The T_4 levels in does displayed a more distinct pattern. The level decreased steadily from mid-autumn to the nadir (4.72 $\mu\text{g}/\text{dl}$) in early spring. It then peaked abruptly (8.51 $\mu\text{g}/\text{dl}$) in mid-spring. The level dropped gradually again until mid-summer (4.66 $\mu\text{g}/\text{dl}$) and remained low until September.

When six periods of equal duration were divided, serum T_4 levels were significantly different among periods. In bucks, the highest levels were found in October–November and the lowest in August–September. In does, the differences were more distinct, with the highest levels in both October–November and April–May and the lowest in July–August. However, there was no significant difference between sexes.

Serum T_3 levels (Fig. 4) in bucks increased from mid to late autumn, then decreased abruptly in early winter. A gradual increase through the winter was followed by a decrease in the spring. The levels increased and peaked (196 pg/dl) in mid-summer, then dropped gradually to the nadir (92 pg/dl) in early autumn. In does, the levels fluctuated throughout the year with the peaks in early winter and mid-summer (225 and 209 pg/dl, respectively), with the lowest levels occurring in late spring and late summer (116 and 121 pg/dl, respectively). T_3 levels were significantly lower during December–January in the bucks. Due to large variations, no significant differences were found in other 2-month durations and between sexes.

Serum C levels (Fig. 5) in both bucks and does fluctuated greatly throughout the year. The nadir in bucks (12.5 ng/dl) and in does (24.2 ng/dl) occurred in

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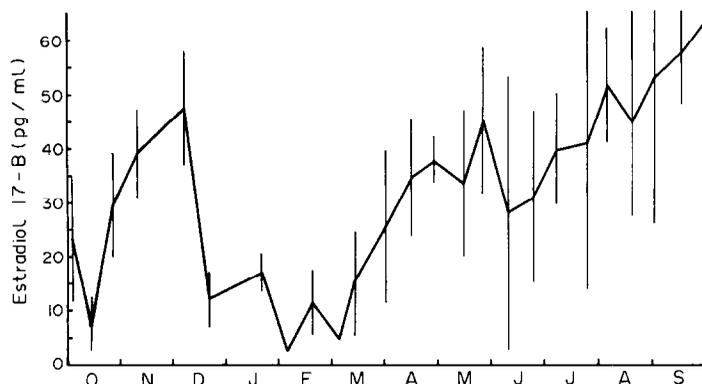


Fig. 2. Annual cycle of serum estradiol 17- β mean (\pm SD) of four female yearling white-tailed deer.

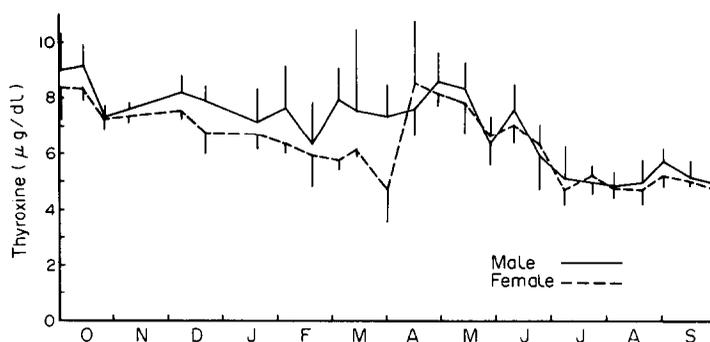


Fig. 3. Annual cycle of serum thyroxine (T_4), mean (\pm SD) of four male and four female yearling white-tailed deer.

late winter. Due to large variations, there were no seasonal and sex differences in serum C levels. However, in most 2-week periods, overall higher C level were found in the female deer (Fig. 5).

Annual cycles in feed intake and body wt

Dry matter intake (g/deer/day) in bucks declined significantly in early and late winter (Fig. 6). An overall

decrease of 40% through the winter was followed by a gradual increase through the spring and summer. A slight decrease in late spring was followed by a short-term, abrupt increase (40%) in mid-summer. In does, the pattern of DMI was similar to that found in the bucks with the significantly lowest intake in early and late winter and an overall decrease of 50%. A gradual increase in early spring was followed by another drop

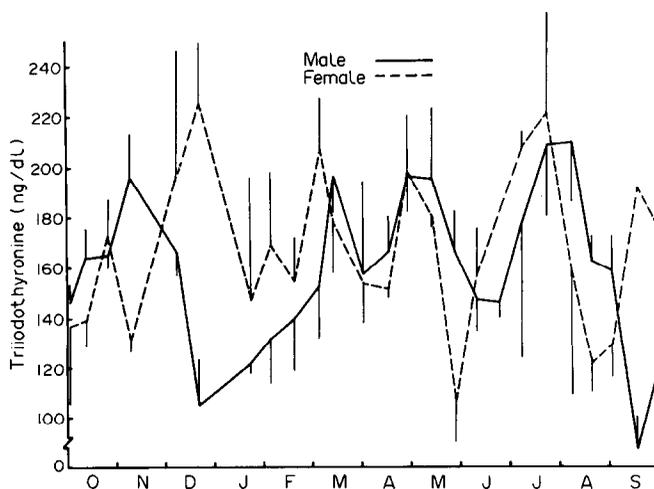


Fig. 4. Annual cycle of serum triiodothyronine (T_3), mean (\pm SD) of four male and four female yearling white-tailed deer.

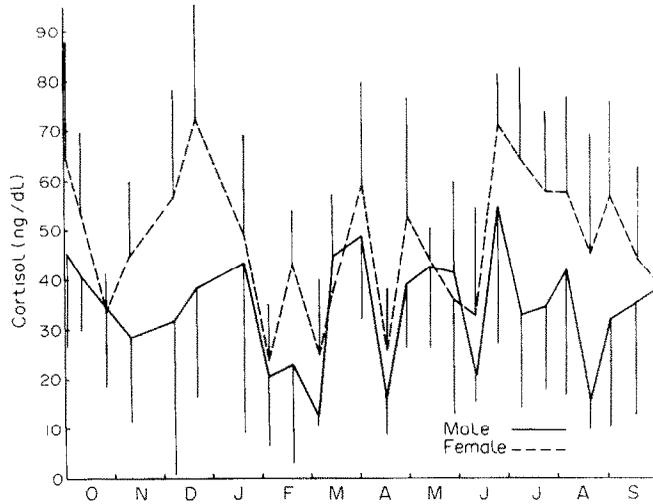


Fig. 5. Annual cycle of serum cortisol, mean (\pm SD) of four male and four female yearling white-tailed deer.

(30%) in late spring. DMI in December–January and February–March was significantly lower than in other 2-month durations in both sexes.

Body wt in bucks (Fig. 7) decreased slightly to the lowest (39.5 kg) in late winter with a total loss of 14%. A slight increase through the spring was followed by

another loss (5%) in May. A gradual increase in the summer was followed by a third drop (8%) in August. In does, body wt increased slightly through the autumn and winter. In late winter an abrupt drop (20%) occurred. Body wt in does decreased slightly to the nadir (34.4 kg) in late spring for a total loss of 11%.

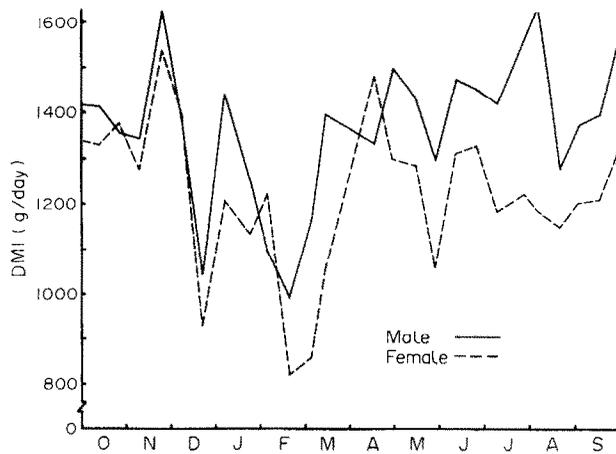


Fig. 6. Dry matter intake of four male and four female yearling white-tailed deer fed a pelleted diet.

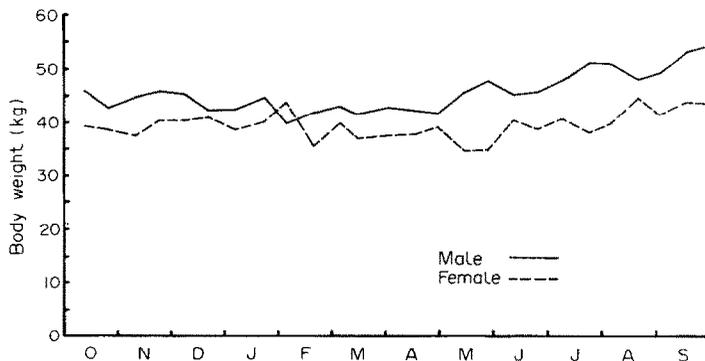


Fig. 7. Body wts of four male and four female white-tailed deer.

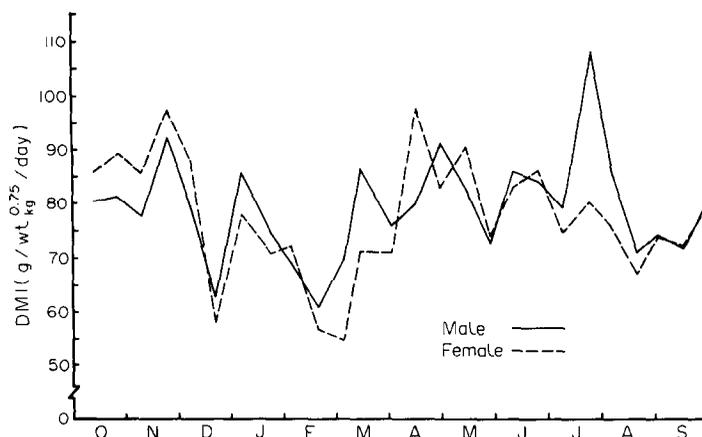


Fig. 8. Dry matter intake (DMI) per unit metabolic body size (body wt^{0.75}) in four male and four female yearling white-tailed deer fed a pelleted diet.

A gradual increase in the summer was followed by another drop (8%) in late summer. There were four declines in DMI/body wt^{0.75} throughout the year in both bucks and does (Fig. 8). Two declines occurred in early winter (December) and late winter (February) with a drop of 30 and 20% respectively in the bucks and 36 and 29% respectively in the does. The other two declines occurred in late spring and mid-summer (17 and 34% in the bucks and 18 and 16% in the does, respectively). An abrupt increase of DMI in mid-summer was followed by the greatest loss (34%) in the bucks.

Correlations between hormones, DMI or body wt and climate data

In bucks, serum T levels were significantly correlated with DMI, DL and temperature ($r = -0.42$, -0.69 and -0.67 , respectively). Serum T₄ levels were significantly correlated with body wt or temperature ($r = -0.68$ and -0.44 , respectively) while T₃ was significantly correlated with DMI and DL ($r = 0.45$ and 0.40 , respectively). DMI was significantly correlated with both DL and temperature ($r = 0.51$ and 0.60 , respectively), while body wt was correlated with temperature ($r = 0.57$) only. However, in Period 1 during the rut, T levels were significantly correlated with DMI, DL and temperature ($r = -0.82$, -0.73 and -0.76 , respectively). In Period 2 after the winter, T₄ and T₃ were significantly correlated with DMI ($r = 0.84$ and 0.86 , respectively). During summer heat stress in Period 3, T₄ was significantly correlated with body wt ($r = -0.68$).

In does, serum E levels were significantly correlated with DL and temperature ($r = 0.40$ and 0.54 , respectively), while only T₄ was significantly correlated with DMI and body wt ($r = 0.43$ and -0.46 , respectively). During the breeding season in Period 1, T₄ correlated significantly with DL and temperature ($r = 0.71$ and 0.86 , respectively). However, in Period 2 after the winter, E was significantly correlated with DL and temperature ($r = 0.90$ and 0.79 , respectively). T₄ was correlated with DMI and temperature ($r = 0.86$ and 0.76 , respectively), while T₃ was correlated with temperature ($r = 0.76$) only. During the summer heat in Period 3, E was significantly correlated with DL and

temperature ($r = -0.91$ and -0.77 , respectively), while C levels were significantly correlated with body wt ($r = -0.84$).

DISCUSSION

Annual cycles in hormone levels

Serum T levels, which were high during the rut, followed the same pattern as that found in northern states (McMillin *et al.*, 1974; Bubenik *et al.*, 1977), except that they occurred two months later. Serum E levels in the does peaked in early winter and also corresponded with the breeding season. The short-term interruption in the increase of E level during early summer, however, might have been due to initial high ambient temperature and humidity (Thatcher, 1974). The increased levels in the last few months of the study might have been due to increased age and the maturation of the reproductive tract in the does.

The lack of sex differences in T₄ levels was consistent with the study by Bubenik and Bubenik (1978). Seal *et al.* (1972) found that there were no significant differences in T₄ levels throughout the year in northern does with good diets. Bubenik and Bubenik (1978) found no significant differences over the year in mature deer, but they did find significantly higher concentrations in male juveniles. This explains our findings of higher T₄ levels in the initial months and lower levels in the final months of the study as the result of increased age. However, they also may have been due to increasing temperature (Hefco *et al.*, 1975). The T₄ concentrations, ranging from 4.5 to 11.7 µg/dl, were far lower than those found in Canada by Bubenik and Bubenik (1978) (13–24 µg/dl). This could be due to latitude, since in warmer weather rat T₄ concentrations tend to be lower than in colder weather (Hefco *et al.*, 1975).

Winter cold coupled with the breeding season seemed to have a more critical effect on does than bucks, since there was a 20% weight loss in the does and only 14% in the bucks. Hoffman and Robinson's (1966) histological study of thyroid glands in wild white-tailed deer in Maryland showed evidence for the possibility of lowered metabolic activity during winter. During winter, a lowered metabolic rate, as revealed

by lowered T_4 levels, might facilitate not only compensatory rest after the breeding season, but also adaptation to severe winter cold (Bubenik and Bubenik, 1978). The decreased energy intake resulting from winter voluntary hypophagia would result in decreased thyroid activity (Seal *et al.*, 1972). Thus, a primary alteration in appetite, with resulting secondary changes in thyroidal hormone economy, could explain the metabolic adaptations seen in the deer in the northern winter (McMillin *et al.*, 1980). The significant and negative correlation between T_4 and temperature found in our study indicated the existence of an inverse relationship (Hefco *et al.*, 1975). This result contrasts with those from Hoffman and Robinson (1966) and Bubenik and Bubenik (1978) in northern environments. In south Texas, the deer face a less severe winter cold and a more severe summer heat stress (Meyer, 1982). This climate resulted in different responses of deer to the environment in both hormonal changes and hypophagia (Wheaton and Brown, 1983).

In the bucks, T_4 levels were significantly correlated with temperature in Periods 1 and 2, but not in Period 3. This might be due to small sample size, but also it is possible that high temperature in Period 3 interfered with T_4 changes. From a biological viewpoint, it is possible that increased T_4 levels, coupled with increased temperature and DL somewhat in Period 2, provide a better environment for the bucks in initiating antler growth (Bubenik, 1982; Brown *et al.*, 1983). Increased T_4 levels might be a result of, rather than a cause of, increased energy intake (McMillin *et al.*, 1980). The significant correlation between T_4 levels and DMI during this period in males and females ($r = 0.84$ and 0.95 , respectively) supports this concept. Results from either lagged temperature or DL provide evidence that serum T_4 levels are both temperature and DL dependent in the bucks. In the does, the lack of a significant correlation of T_4 with either DL or temperature indicates that T_4 changes may be independent of both temperature and DL. However, in late May as temperature increased to 27°C , the significant drop of T_4 levels in both male and female deer confirms the study of Hefco *et al.* (1975) that the fall of T_4 production in the heat is the direct consequence of an elevated temperature. It seems that a short-term (1-week) effect of temperature on serum T_4 levels exists in deer.

Serum T_3 levels are directly dependent on energy intake (Seal *et al.*, 1978). This was true in our study, especially during late winter and late spring. This might be due to the effect of increased DMI (McMillin *et al.*, 1980). Increased DL and temperature may be the mover for increased DMI and in turn resulted in increased T_3 levels during Period 2. Results from lagged DL showed that it played a more important role in serum T_3 changes in bucks. The lack of significant correlations between serum T_3 and any of the climatic parameters indicated that serum T_3 levels in does might be independent of DL and temperature. It is also possible that individual variations mask a significant correlation between serum T_3 and DL or temperature. However, Bahanak *et al.* (1981) suggested that although total serum T_3 and T_4 concentrations in does did not always correlate well with the degree of thyroid gland activity, they were still good

indicators of nutritional stress. In our findings, serum T_4 and T_3 in the bucks and T_4 in the does were good indicators for studying hypophagia if coupled with gonadal hormone changes.

The lack of sex and seasonal differences in C levels agrees with the findings of Seal *et al.* (1972) and Bubenik *et al.* (1975a). The large variations within groups agree with the finding of Bubenik *et al.* (1975a) of large variations in C levels between individual deer.

Annual cycles of feed intake and body wt

Significantly lowered DMI in December and February indicated that winter, coupled with rutting season, had a significant effect on DMI in South Texas white-tails (Wheaton and Brown 1983). Cowan (1971) attributed the short-term intake suppression during mid-summer to the association with hormonal changes triggered by the shortened days after the summer solstice. Wheaton and Brown (1983) and Holter *et al.* (1977) attributed this to warm and humid weather. In August–September all deer had heavier body wts than in February–March or April–May. The increased body wt during the final months could be the result of increased age, but the significantly lowered body wt during late winter could be the result of winter cold coupled with rutting activities. Early summer weight loss (17%) in bucks and late spring weight loss (10%) in does, which agree with findings by Short *et al.* (1968), may have been due to the effects of initial short-term high ambient temperatures.

Interrelationships among hormone levels, DMI, body wt and climate

The negative correlation between serum T level and DMI, especially during the rut (December–January) confirms the hypothesis offered by McMillin *et al.* (1980) that fall hypophagia is directly under the control of the annual T concentrations in bucks, and that high concentrations of T trigger a behavioral response of appetite suppression in the hypothalamus. This is consistent also with Mazur's (1974) findings that there was an inverse relationship between intake and circulating plasma T concentrations in male deer during the rut and winter, but that the relationship was less clear in the spring and summer.

Daylength has its effects primarily by changing the sensitivity of the hypothalamus to circulating steroids in red deer stags (*Cervus elaphus*) (Lincoln and Kay, 1979). Also Mirarchi *et al.* (1977) pointed out that decreasing DL was associated with increased pituitary LH secretion, causing a rise in circulating LH levels which in turn stimulated the interstitial cells of Leydig to produce T. Studies by McMillin *et al.* (1974) suggested that the T cycle was an inherent circannual rhythm which could be synchronized to the photoperiod. They suggested, however, that under constant, unequal daily periods of light and dark, the approximately yearly T cycles were independent of nutrition and temperature changes. It seems that in our study T levels are dependent on both DL and ambient temperature changes.

The lack of a significant correlation between serum E levels and DMI indicated that the breeding season in does seemed to have a less distinct effect on hypophagia than T levels did in bucks. According to Wade's

(1972) study in rats, E is the principal ovarian steroid regulating body wt in females and acts on separate neural loci to inhibit DMI. There was no such relationship found in the deer in our study. Serum E levels increased along with increased DL and temperature, especially in Period 2. In Period 3, as temperatures were usually greater than 27°C, the negative correlations between E levels and both DL and temperature may have been due to the suppression of E from high ambient temperatures and humidity or due to the effects of DL changing from increasing to decreasing. Correlations of lagged temperature and DL up to 7 weeks with E indicated that both had overall long-term effects on serum E levels.

The difficulty in studying blood C levels was reported by Bubenik *et al.* (1977), in which an elevation of blood C concentrations as much as 350% in white-tailed deer was found in just 15 min. However, in attempting to avoid the sudden increase of C levels in our study, Rompun was used as tranquilizer to facilitate bleeding without causing a significant stress. The large variation in this study was not due to sudden seasonal fluctuations, but due to individual differences (i.e. some animals had overall high C concentrations while others had lower concentrations throughout the year). It is possible that large individual variations mask a clear seasonal trend and any correlation with other parameters (Bubenik *et al.*, 1977).

During and after the rut, significant correlation of DMI with DL supported the studies by French *et al.* (1960), Cowan (1971) and Simpson and Suttie (1983). Initial high temperature (27°C), however, had a significant short-term effect on DMI in bucks and does, i.e. in early summer. According to the study by McDowell *et al.* (1969) in cattle, during the first week of elevated temperatures, the decreased DMI might be the result of the effect on the appetite center and the tendency to let a high respiration rate interfere with feeding. Conrad (1966) also pointed out that thermostatic mechanisms may dominate regulation of feed intake independently of other factors. Thompson *et al.* (1963) found that adverse effects of the hot environment became conspicuous soon after the initial heat exposure in dairy heifers. Our study in deer showed the same result; the suppression of DMI by initial high ambient temperature was apparent.

In bucks, body wt was significantly correlated with temperature. Results from lagged temperature also provided evidence that temperature had both short- and long-term effects on body wt in bucks. In does, the climatic effects on body wt were less clear than those in bucks. However, winter cold caused a 20% loss in body wt in a short period, and the initiation of high temperature in late spring caused a 12% body wt loss. It seems that short-term climatic effects were clear in the does.

In conclusion, the hypophagia found in winter in both male and female South Texas white-tailed deer is no doubt an endogenous rhythm synchronized by environmental factors and the breeding season. Winter cold coupled with the breeding season and of initial and prolonged heat in late spring and summer result in a unique form of hypophagia somewhat different from that found in the Northern states. Due to the number of animals in this study, the results cannot be considered conclusive. Nonetheless it is evident that the

thyroid hormones along with the sex steroids can function as indicators of nutritional condition, and may play a role as the link between environmental cues and feed intake in deer.

Acknowledgement—The authors wish to thank C. A. Wheaton for collection of the feed intake and body weight data, and R. Bingham for his assistance in the statistical analysis of the data. This work was supported by the Caesar Kleberg Foundation for Wildlife Conservation and N.I.H. grant No. 506-RR-08017-09.

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