

Plasma Corticosterone Concentrations Associated with Acute Captivity Stress in Wild Loggerhead Sea Turtles (*Caretta caretta*)

Lisa F. Gregory,^{*,1} Timothy S. Gross^{†,‡} Alan B. Bolten,^{‡,§} Karen A. Bjorndal,^{*,§} and Louis J. Guillette, Jr.^{*}

^{*}Department of Zoology, [†]Biotechnologies for the Ecological, Evolutionary and Conservation Sciences (BEECS) Program, [§]Archie Carr Center for Sea Turtle Research, and [‡]Department of Wildlife Ecology and Conservation, University of Florida, Gainesville, Florida 32611

Accepted July 3, 1996

Plasma corticosterone concentrations were measured in wild loggerhead sea turtles (*Caretta caretta*) in response to acute captivity (capture, serial bleeding, and restraint up to 6 hr). In general, concentrations of corticosterone dramatically increased 1 hr after capture, peaked at 3 hr, and decreased by 6 hr. Initial corticosterone concentrations were significantly lower in animals captured by tangle net than in those captured by trawl and were thought to more closely represent baseline levels. Significant effects of season and size class on corticosterone concentrations were found for turtles captured by trawl. Corticosterone concentrations of small turtles captured in summer were higher than those of large turtles captured in the same season and of all turtles captured during winter. In winter, corticosterone concentrations for small turtles were higher than those for large turtles at 3 hr after capture. Large turtles captured during winter experienced the slowest rate of increase in plasma corticosterone and a decline at 3 hr after capture. Although cloacal temperatures were significantly higher in summer samples, corticosterone concentrations of large turtles did not differ between seasons until 1 hr after capture. In addition, several large turtles during summer did not experience an increase in corticosterone concentrations 1 hr after capture. It is possible that the

lower corticosterone response of large turtles captured during summer may be associated with reproductive condition. © 1996 Academic Press, Inc.

It is widely accepted that exposure to stressors increases secretion of glucocorticoids, cortisol, and corticosterone in a wide variety of gnathostome vertebrates (Stephens, 1980; Greenberg and Wingfield, 1987). These hormones are believed to have a role in adapting the body to stressors that present an immediate threat to homeostasis (Axelrod and Reisine, 1984). Although most generalizations about stress are derived from studies of domesticated endotherms, recent efforts have demonstrated stress-induced elevations of corticosterone in a variety of reptiles (Elsey *et al.*, 1990; Mahapatra *et al.*, 1991; Moore *et al.*, 1991; Grassman and Hess, 1992; Lance, 1994). Several reptilian studies have demonstrated situations where corticosterone concentrations do not increase in response to stimuli normally considered stressful (Whittier *et al.*, 1987; Valverde *et al.*, 1993).

General conclusions about endocrine responses to stress are difficult to make among and within species across studies (Moore *et al.*, 1991). Plasma corticosterone levels are often influenced by exposure to handling or captivity stress before the actual experiment begins. Many studies utilize animals that have been

¹ To whom all correspondence should be addressed.

bred or raised in captivity and may respond differently from wild, free-ranging species. Conducting a well-controlled experiment utilizing wild animals is difficult because of the extreme diversity of factors that can affect glucocorticoid secretion (e.g., time of day, reproductive condition, nutrition, social status). A particular problem faced by investigators of stress responses in wild ectotherms is the difficulty of separating the effects of ambient temperature from other physiological parameters. Finally, variations in radioimmunoassay techniques must be taken into account before meaningful comparisons of hormone values can be made among studies.

All species of sea turtles occurring in United States waters are listed as threatened or endangered under the Endangered Species Act of 1973 and subsequent amendments. Research and conservation efforts have produced a large body of literature on sea turtle biology and conservation theory (Bjorndal, 1995). However, little published data dealing with stress-induced elevations of corticosterone exist. One study of green sea turtles (*Chelonia mydas*) demonstrated that plasma corticosterone concentrations increased in response to capture and serial bleeding and were higher in turtles with fibropapillomas (Aguirre *et al.*, 1995).

The purpose of the present study was to describe plasma corticosterone concentrations in a wild, free-ranging, ectothermic vertebrate. This study investigated alterations in plasma corticosterone concentrations associated with acute captivity stress, defined here as the stress associated with capture, serial bleeding, and restraint up to 6 hr, in the loggerhead sea turtle (*Caretta caretta*). In addition, the effect of (1) capture method, (2) season, and (3) size class on stress-induced alterations of plasma corticosterone concentrations were examined. To our knowledge, no study has examined the effects of size class on plasma corticosterone concentrations in reptiles. Such data will provide new insight into the endocrinology of the stress response in these unique marine reptiles.

MATERIALS AND METHODS

Capture Methods

Trawl. A shrimp trawler equipped with two 18.3-m mongoose style nets (20.3-cm mesh stretch) was used

to capture turtles in the Port Canaveral Ship Channel, Cape Canaveral, on the Atlantic coast of Florida. Trawling was conducted monthly over 3 consecutive days from June through August 1992 (summer) and January through March 1993 (winter). "Tow time" is defined as the interval between the deployment of the net (i.e., when the net reaches the bottom of the channel) and the start of retrieval. Eight tows (two time, 25–30 min) were utilized on each of the 3 days between the hours of 0700 and 1500. Turtles were removed from the trawl within 15 min after the start of retrieval. Additional tows using a shorter trawling period (tow time, 10 min) were conducted intermittently after the eighth tow on the 3rd sampling day. Animals captured in short tows ($n = 6$) were examined to hopefully establish a more reliable baseline for plasma corticosterone.

Tangle net. Turtles were collected by tangle net at Corrigan Reef, east of the Cedar Keys, on the Gulf coast of Florida. A large mesh tangle net (50 m in length, 50.8-cm stretch mesh, and 20 meshes deep) held in place at each end by a 10-kg anchor was used to capture turtles from May through November 1992. Netting was conducted biweekly for 1–3 consecutive days, although tide and weather conditions greatly influenced the netting schedule. The net was continuously monitored for turtle entanglement and checked hourly to ensure that no turtle was caught in the lead line and to prevent net fouling by stingrays. It was observed that turtles immediately surface when they first encounter the net. Turtles were removed from the net within 15 min of first sighting (i.e., when the turtle's head first broke the water's surface).

Sampling Procedure

All blood samples were obtained from the dorsal cervical sinus using 20-gauge needles and sterile, sodium-heparinized vacuum tubes. Immediately after collection, samples were centrifuged (1200g) for 5 min. Plasma was transferred into cryovials, frozen in liquid nitrogen within 15 min of sampling, and then stored at -70° until assayed. An initial blood sample (7 ml), designated "Time zero" (T-0), was taken from all turtles immediately after removal from the nets to determine plasma corticosterone, testosterone, and estradiol concentrations (sex steroid analyses in Bolten *et al.*, unpublished). The time between the start of the

tow or the first sighting to the T-0 sample was recorded in minutes and designated "Start-T-0." Additional serial blood samples (3 ml) were taken to determine subsequent plasma corticosterone concentrations over time. After the initial blood sample and between serial samples, turtles were held on their dorsal side to minimize activity and prevent injuries. Animals were covered with wet, white sheets on the trawler to prevent overheating. Animals captured by tangle net were protected from the sun by a canopy. Cloacal temperatures were monitored and turtles reaching a temperature of 32° were released immediately ($n = 1$; captured by trawl during summer).

Initial and serial blood sampling times (in minutes following T-0) for turtles captured by trawl were T-0 ($n = 107$), T-60 ($n = 91$), and T-180 ($n = 59$). Six-hour samples (T-360; $n = 17$) were taken opportunistically during the winter months to further characterize the corticosterone response induced by acute captivity. Initial and serial blood sampling times for turtles captured by tangle net were T-0 ($n = 11$), T-30 ($n = 9$), T-60 ($n = 8$), T-180 ($n = 8$), T-360 ($n = 3$). Sample sizes decrease over time due to logistical problems such as limited trawl availability and inclement weather. However, to control for possible effects of hemodilution, no intermediate blood samples were skipped (e.g., all turtles captured by trawl and sampled at T-360 were also sampled at T-0, T-60, and T-180).

Measurements

Minimum straight-line carapace length (MSCL; Pritchard *et al.*, 1983) was measured with metal calipers. Total tail length (posterior-most margin of the anal scutes to the tip of tail) was measured with a fiberglass tape measure. Cloacal temperatures were taken with an Atkins T Thermocouple thermometer (Model No. 39641-T).

Animals with MSCL < 80.0 cm were classified as small and those with MSCL \geq 80.0 cm as large, based on estimates for subadult and adult *C. caretta* from Henwood (1987). We prefer not to use the terms subadult and adult (or immature and mature) because the reproductive status of turtles in the present study was not determined. All turtles sampled at Corrigan Reef were small. The sex of small turtles could not be determined based on plasma sex steroid concentrations (Bolten *et al.*, unpublished). The sex of large

turtles was determined primarily on sexual dimorphism of tail morphology (Pritchard *et al.*, 1983). For the present study, large turtles with total tail lengths > 35.0 cm were classified as males; large turtles with total tail lengths < 28.0 cm were classified as females. A bias against large males may exist since short-tailed, immature males are known to reach carapace lengths over 80 cm (Limpus, personal communication). Effects of sex on plasma corticosterone concentrations in large turtles were not determined due to small sample size within season.

Radioimmunoassay

Plasma samples from *C. caretta* were analyzed for corticosterone using a standard radioimmunoassay (RIA). Antiserum (#07-120016, lot #3R3-PB) and tritium-labeled corticosterone were purchased from ICN Biomedicals (Costa Mesa, CA). Corticosterone titers in nanograms per milliliter were calculated using the standard curve generated in the assay. Values were determined by a 4-parameter logistic model using a program supplied by Beckman Inc. (EIARIA Curve Fit Program).

The extraction efficiency was determined by adding tritium-labeled corticosterone (10,000 cpm) to 10 samples ($50 \mu\text{l} \times 4$) from turtles of various sizes and collected at various times and seasons. Samples were allowed to equilibrate at 4° for 24 hr. A single extraction with 5 ml of anhydrous ether produced an extraction efficiency of $70 \pm 10\%$. A second extraction substantially increased the extraction efficiency to $94 \pm 3.2\%$. Consequently, all samples prior to RIA analysis were extracted twice with 5 ml of anhydrous ether and afterward corrected for the extraction efficiency.

Standard curves were prepared in buffer with known amounts of radioinert corticosterone (0, 0.78, 1.56, 3.125, 6.25, 12.5, 25, 50, and 100 ng/ml) purchased from Amersham Corp. (Arlington Heights, IL). The minimum concentration per tube that was distinguishable from zero was 0.176 ng/ml. Cross-reactivities of the corticosterone antiserum with other steroids were 6.1% for deoxycorticosterone; <1% for progesterone and cortisol; <0.1% for aldosterone, 20-alpha dihydroprogesterone, testosterone, and 11-deoxycortisol; and <0.01% for all other steroids examined.

A pooled *C. caretta* sample (from animals of various sizes and collected at various times and seasons) was

TABLE 1
Initial Mean Plasma Corticosterone Concentrations, Cloacal Temperatures, and Start-T-0 for Small Loggerhead Turtles (*Caretta caretta*) Captured by Tangle Net and Trawl

Capture method	Initial plasma corticosterone (ng/ml)	Initial cloacal temperature (°)	Start-T-0 (min)
Tangle net (<i>n</i> = 11)	0.55 ± 0.15	27.21 ± 0.89	8.27 ± 0.84
Trawl (<i>n</i> = 51)	2.07 ± 0.35	20.77 ± 0.50	37.77 ± 0.89

assayed serially in 0-, 10-, 20-, 30-, 40-, and 50- μ l volumes (final volume of 50 μ l with charcoal-stripped plasma). This inhibition curve was parallel to the standard curve, with the test for homogeneity of regression indicating that the curves did not differ. Further characterization of the assay involved measurement of known amounts of radioinert corticosterone (0, 0.78, 1.56, 3.125, 6.25, 12.5, 25, 50, and 100 μ g/ml) in 50 μ l charcoal-stripped plasma [$Y = 11.5 + 0.973X$; Y , amount of corticosterone measured (μ g/ml); X , amount of corticosterone added (μ g/ml); $R^2 = 0.9341$]. Inter-assay and intra-assay coefficients of variation were 15.0 and 5.7%, respectively.

Statistics

Statistical analyses were performed on raw and log-transformed data using the SuperANOVA general linear modeling program (Abacus Concepts, 1991). Corticosterone concentrations (ng/ml) were log transformed to obtain homogeneity of variance. All reported probability values were obtained from log-transformed data. Data from initial samples were subjected to analyses of variance (ANOVA) and analyses of covariance. Start-T-0 and cloacal temperature were used as covariates. Data from serial samples were

analyzed by repeated measures ANOVA. Due to uneven sample sizes, a conservative approach (Bonferroni) was used to interpret mean comparison contrasts. The P value for each contrast was multiplied by the number of comparisons made for each model (Jandel Scientific, 1994). All statistical significance is accepted at $P < 0.05$. All graphs and mean values reported for corticosterone (mean \pm SE) were obtained from raw data.

Effects of capture method were determined for small turtles as no large turtles were captured with the tangle net. Thus, effects of size class could only be determined for turtles captured by trawl. In addition, a seasonal effect could not be determined for animals captured by tangle net as no turtles were captured when water temperatures fell below 20°C.

RESULTS

Initial Samples

Mean initial corticosterone concentrations of small turtles captured by trawl were 3.7 times higher ($P = 0.0001$; Table 1) than levels from animals captured in a tangle net. Mean Start-T-0 was significantly greater ($P = 0.0001$; Table 1) for animals captured by trawl and accounted for the variation due to capture method.

A significant interaction ($P = 0.0005$; Table 2) between season and size class was observed for turtles captured by trawl. Mean initial corticosterone concentrations were significantly higher ($P = 0.0001$; Table 2) for small turtles and turtles captured in summer. Small turtles captured in summer had a mean corticosterone concentration over 3.5-fold higher ($P = 0.0004$; Table 2)

TABLE 2
Initial Mean Plasma Corticosterone Concentrations and Cloacal Temperatures for Small and Large Loggerhead Turtles (*Caretta caretta*) Captured by Trawl during Summer and Winter

Size class	Initial plasma corticosterone (ng/ml)		Initial cloacal temperature (°)	
	Summer	Winter	Summer	Winter
Small	4.71 ± 0.96 (<i>n</i> = 12)	1.26 ± 0.24 (<i>n</i> = 39)	27.23 ± 0.25	19.07 ± 0.16
Large	1.19 ± 0.08 (<i>n</i> = 46)	1.04 ± 0.22 (<i>n</i> = 10)	28.13 ± 0.17	18.14 ± 0.13

than small turtles captured in winter and large turtles captured during either season. No significant difference in initial corticosterone concentrations occurred between size classes during winter or for large turtles between seasons. As expected, cloacal temperatures were significantly higher ($P = 0.0001$; Table 2) in turtles captured during summer and accounted for the variation due to season.

No effect of time of day was observed on initial corticosterone concentrations. In addition, no significant difference in initial corticosterone concentrations was found between animals captured in 10-min tows and those in all other tows.

Serial Samples

Corticosterone concentrations changed significantly ($P = 0.0001$; Fig. 1) over time in response to acute captivity stress in small turtles captured by both trawl and tangle net. No significant variation in corticosterone concentrations between capture method was detected for T-60, T-180, and T-360 samples.

A significant ($P = 0.0005$) three-way interaction among time, season, and size class was detected. Serial corticosterone concentrations for small turtles captured during summer were markedly higher ($P \leq 0.0189$; Fig. 2) than those for large turtles captured during the same season and those for small turtles

captured in winter. Turtles captured during winter did not exhibit a difference in serial corticosterone concentrations between size class except at T-180 ($P = 0.0008$; Fig. 2). Large turtles exhibited a significant difference in corticosterone concentrations between seasons at T-60 and T-180 ($P \leq 0.0104$; Fig. 2).

DISCUSSION

Acute Captivity Stress

This study demonstrates that capture, restraint, and repeated bleeding of wild *C. caretta* are generally associated with increased plasma corticosterone concentrations, with highest concentrations occurring 3 hr after capture. Corticosterone concentrations of small turtles captured by trawl in winter and by tangle net declined by the 6th hr of capture. Large turtles captured by trawl during winter were the only animals that did not experience any significant alterations of plasma corticosterone in response to acute captivity.

Initial corticosterone concentrations of small turtles captured by trawl were similar to those from four small *C. caretta* captured by trawl in the Port Canaveral Ship Channel on 29 May 1982 and exposed to acute captivity stress (Wibbels *et al.*, 1987). However, mean

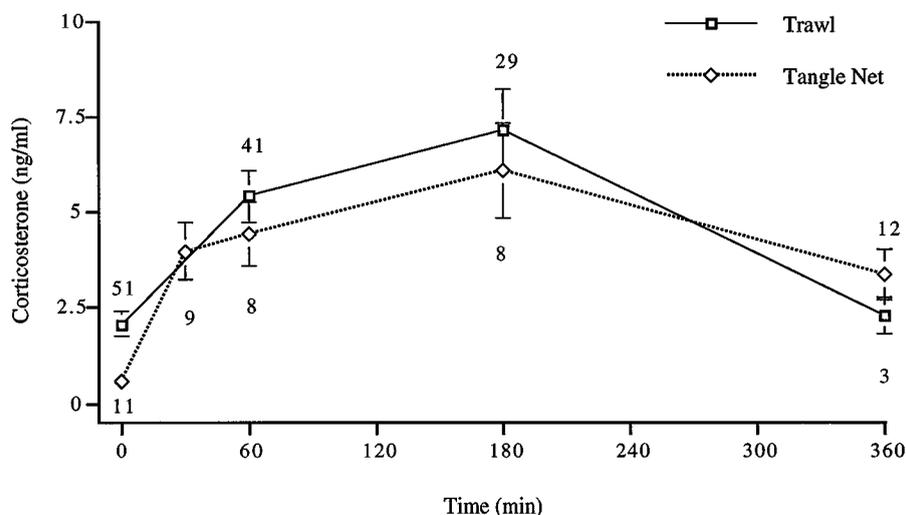


FIG. 1. Mean plasma corticosterone concentrations over time for small loggerhead turtles (*Caretta caretta*) captured by trawl or tangle net. Numbers by means (\pm SE) indicate sample sizes.

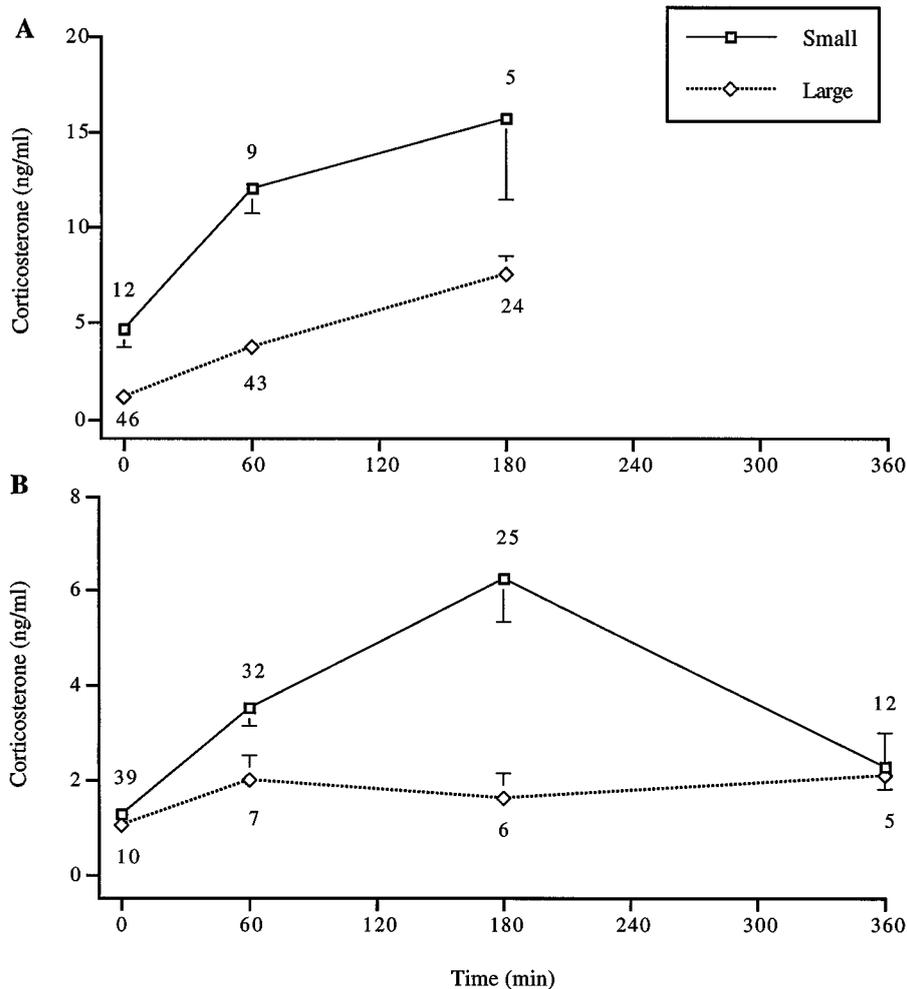


FIG. 2. Mean plasma corticosterone concentrations over time between size class for loggerhead turtles (*Caretta caretta*) captured by trawl during summer (A) and winter (B). Numbers by means (\pm SE) indicate sample sizes.

corticosterone concentrations after 3 hr of capture in the latter study were almost twice as high as T-180 samples from small turtles captured during summer. This variation in mean corticosterone concentrations over time may not be a true intraspecific difference. In Wibbels *et al.* (1987), five to six blood samples (total volume not published) were obtained per individual turtle, whereas in the present study, only two blood samples (total volume = 10 ml) were taken by 3 hr after capture. Repeated handling during blood sampling and hemodilution (Callard, 1975) may alter corticosterone concentrations in *C. caretta* and should be evaluated before accurate comparisons can be made among studies.

Relatively fast rates of corticosterone secretion in

response to acute stress can occur in reptiles. In the tree lizard, *Urosaurus ornatus*, wild caught animals subjected to only 10 min of handling stress had corticosterone concentrations 6.6 times higher than animals bled immediately after capture (Moore *et al.*, 1991). The fact that initial plasma corticosterone concentrations in small *C. caretta* captured in a tangle net displayed a 7.2-fold increase after 30 min of restraint suggests that significant increases in corticosterone may occur at even shorter time intervals.

Corticosterone concentrations within a particular treatment group can be highly variable (Gist and Kaplan, 1976; Morris, 1982; Dauphin-Villemant and Xavier, 1987; Summers and Norman, 1988). For example, in the present study, three apparently healthy

small *C. caretta* were captured on 13 August 1992 during the same tow and in the same net. Initial corticosterone concentrations were 2.3, 10.3, and 2.0 ng/ml and by the 3rd hr of captivity, concentrations had risen to 12.8, 21.6, and 7.4 ng/ml, respectively. Initial variability could be due to the time each turtle spent in the trawl, genetic variability, or a precapture experience (e.g., a recent encounter with a shark). Nonetheless, the two animals that initially had similar corticosterone concentrations after removal from the trawl demonstrated strikingly different hormone levels in response to the same stressor. This individual variability in plasma hormone concentrations is not due to season, size class, diurnal rhythm, or reproductive condition, but to some other factor not accounted for in the present study (e.g., genetic variability, nutritional condition). The animal with the highest initial corticosterone concentration (11.85 ng/ml) was a small *C. caretta* recaptured by trawl in June with a severely damaged, necrotic right flipper and two deep propeller wounds on its carapace. Hence, individual differences in corticosterone concentrations may provide clues to underlying physiological conditions and thus may lead to a better understanding of the role of stress-induced elevations of corticosterone.

Capture Method

Initial corticosterone concentrations of small turtles captured by trawl were significantly higher than turtles captured by tangle net and were attributed to higher Start-T-0 times. Significant differences in corticosterone concentrations were not detected at subsequent sampling times. This might suggest that the stress of holding and sampling the animal on deck induces a similar adrenocortical response regardless of capture method. However, a seasonal effect on corticosterone concentrations was observed for turtles captured by trawl and may account for the lack of significance between capture method for serial samples.

Plasma corticosterone concentrations ranged from nondetectable levels (<0.05 ng/ml; T-0; tangle net) to 25.15 ng/ml (T-180; trawl). Mean initial corticosterone concentrations of turtles captured by tangle net are among the lowest values reported for reptiles (see Lance and Elsey, 1986; Schwantes, 1986; Elsey *et al.*, 1991; Moore *et al.*, 1991 for comparable studies utilizing wild reptiles) and are similar to initial levels

experienced by healthy, wild green turtles captured by hand at Kaneohe Bay, Oahu, Hawaii (Aguirre *et al.*, 1995). These T-0 samples represent a unique data set for free swimming *C. caretta* and may be indicative of baseline levels of corticosterone in this species. However, to assume that the higher initial corticosterone concentrations of *C. caretta* captured by trawl are due only to higher Start-T-0 times may be premature. The loggerhead population at Port Canaveral may have higher baseline levels of corticosterone than the population at Corrigan Reef. Thus, a location effect (e.g., human disturbances in the Port Canaveral Channel) may have been an additive influence not controlled for in this study.

Season

Initial and serial corticosterone concentrations of small *C. caretta* captured by trawl during summer were significantly higher than those in *C. caretta* captured during winter. Nearly all the variation in corticosterone concentrations of small turtles between seasons was due to temperature. Magnuson *et al.* (1990) reviewed evidence for increased mortality in turtles captured by trawl in summer compared to those towed under similar conditions in winter. The differential mortality was assumed to be caused by a higher respiratory demand due to increased metabolism at higher water temperatures. A higher metabolic rate in small turtles due to higher water temperatures may be the basis for the variability of corticosterone concentrations between seasons. However, *in vitro* incubated adrenal glands undergo seasonal secretory changes regardless of temperature, indicating that water temperature may not be the only factor driving seasonal effects (Tam *et al.*, 1972; Bradshaw, 1986).

No significant difference in corticosterone concentrations was observed between seasons for large *C. caretta* until 1 hr after capture. The lack of a between-season variation in initial corticosterone concentrations of large *C. caretta* is of particular interest since cloacal temperatures of large turtles were 10° higher in the summer. Ambient temperature may not affect initial concentrations of corticosterone in large turtles or some other factor may alter the adrenal response to stress between seasons.

Size Class

Movement patterns relative to size class are well established for loggerhead turtles on the east coast of Florida (Henwood, 1987; Schmid, 1995). Small turtles are least abundant between May and August. Large females utilize the Port Canaveral Channel during the nesting season and large males are prevalent several months prior to the nesting season. In the present study, all but two large turtles captured during summer were female and all but one large turtle captured during winter were male.

Corticosterone concentrations for small turtles were markedly higher than those for large turtles at all sampling times during summer and at T-180 during winter. At least three factors, age, metabolism, and reproductive condition, may influence this variability. The notion that older animals have experienced more stress than younger animals and become desensitized to environmental stressors is not supported in mammalian literature (Selye, 1976). Adult agamid lizards (*Amphibolurus nuchalis*) often die under harsh environmental conditions (e.g., intense heat, drought) and have high, chronic plasma corticosterone concentrations comparable to juveniles that survive and grow under the same conditions (Bradshaw, 1986). Slower metabolic rates may influence plasma corticosterone concentrations in large turtles. However, the present data suggest otherwise, as we observed similar rates of increase in plasma corticosterone concentrations for small and large turtles between T-60 and T-180 during summer.

Several avian species of the Sonoran Desert demonstrated a suppression of the adrenocortical response when exposed to acute captivity stress during the breeding season (Wingfield *et al.*, 1992). Initial corticosterone concentrations of nesting and adult female *C. caretta* captured by trawl during the nesting season were low and exhibited no significant difference (Schwantes, 1986). While nesting, some olive ridley sea turtles (*Lepidochelys olivacea*) did not exhibit an adrenocortical response when turned over and sampled repeatedly (Valverde *et al.*, 1993). In the present study, several large females captured in summer did not exhibit any change in plasma corticosterone after 1 hr of acute captivity stress. The mean MSCL for large female *C. caretta* was 91.81 ± 0.78 cm, well within the range reported by Bjorndal *et al.* (1983) for females

nesting in June and July on the east coast of Florida. Although we did not verify reproductive status, it is reasonable to assume that large females in the present study were reproductively active. These findings may support a hypothesis that species breeding under severe conditions or where disruption of breeding is great show resistance to acute stressors (Wingfield, 1988). The high energy demands of nesting, both in terms of reproductive output (over 400 eggs during a nesting season; Frazer, 1984) and muscular activity, may depress the stress response in this species. Studies that compare stress-induced alterations of corticosterone concentrations between reproductive and nonreproductive adults are needed to further clarify these data.

ACKNOWLEDGMENTS

The authors thank Peter Eliazar for his assistance with field work at Port Canaveral and Jeffrey Schmid for his expertise in netting turtles at Corrigan Reef. We are grateful to Captain Eddie Chadwick and the crew of the *Mickey Anne* for their hard work on all trawling excursions. We thank Brian Bowen and Dennis Haney for their critical review of the manuscript. Support for the research was provided by the Marine Entanglement Research Program of the National Marine Fisheries Service (to K.A.B., A.B.B., and L.J.G.), the U.S. Army Corp of Engineers (to K.A.B. and A.B.B.), and National Marine Fisheries Service Southeast Fisheries Science Center (to K.A.B., A.B.B., and Jeffrey Schmid). Additional financial support for this study was provided by the following programs located at the University of Florida: Biotechnologies for the Ecological, Evolutionary, and Conservation Sciences—Reproductive Analysis Laboratory; The Archie Carr Center for Sea Turtle Research; and the Department of Zoology.

REFERENCES

- Abacus Concepts (1991). "Superanova: Accessible General Linear Modeling," 4th ed. Abacus Concepts, Inc., Berkeley, CA.
- Axelrod, J., and Reisine, T. D. (1984). Stress hormones: Their interaction and regulation. *Science* **224**, 452–459.
- Bjorndal, K. A., Ed. (1995). "Biology and Conservation of Sea Turtles." Revised ed. Smithsonian Institution Press, Washington DC.
- Bjorndal, K. A., Meylan, A. B., and Turner, B. J. (1983). Sea turtles nesting at Melbourne Beach, Florida. I. Size, growth and reproductive biology. *Biol. Conserv.* **26**, 65–77.

- Bradshaw, S. D. (1986). "Ecophysiology of Desert Reptiles." Academic Press, New York.
- Callard, G. V. (1975). Control of the interrenal gland of the freshwater turtle *Chrysemys picta* *in vivo* and *in vitro*. *Gen. Comp. Endocrinol.* **19**, 69–79.
- Dauphin-Villemant, C., and Xavier, F. (1987). Nychthemeral variations of plasma corticosteroids in captive female *Lacerta vivipara jacquin*: Influence of stress and reproductive state. *Gen. Comp. Endocrinol.* **67**, 292–302.
- Else, R. M., Joanen, T., McNease, L., and Lance, V. (1990). Stress and plasma corticosterone levels in the american alligator—Relationships with stocking density and nesting success. *Comp. Biochem. Physiol.* **95A**, 55–63.
- Else, R. M., Lance, V. A., Joanen, T., and McNease, L. (1991). Acute stress suppresses plasma estradiol levels in female alligators (*Alligator mississippiensis*). *Comp. Biochem. Physiol.* **3**, 649–651.
- Frazer, N. B. (1984). A model for assessing mean age-specific fecundity in sea turtle populations. *Herpetologica* **40**, 281–291.
- Gist, D. H., and Kaplan, M. L. (1976). Effects of stress and ACTH on plasma corticosterone levels in the caiman *Caiman crocodilus*. *Gen. Comp. Endocrinol.* **28**, 413–419.
- Grassman, M., and Hess, D. L. (1992). Sex differences in adrenal function in the lizard *Cnemidophorus sexlineatus*. II. Responses to acute stress in the laboratory. *J. Exp. Zool.* **264**, 183–188.
- Greenberg, N., and Wingfield, J. C. (1987). Stress and reproduction: Reciprocal relationships. In "Hormones and Reproduction in Fishes, Amphibians and Reptiles" (D. O. Norris and R. E. Jones, Eds.), pp. 461–503. Plenum, New York.
- Henwood, T. A. (1987). Movements and seasonal changes in loggerhead turtle *Caretta caretta* aggregations in the vicinity of Cape Canaveral, Florida (1978–1984). *Biol. Conserv.* **40**, 191–202.
- Jandel Scientific (1992–1994). "SigmaStat Statistical Software for Windows User's Manual," pp. 7–61. Jandel Scientific, San Rafael, CA.
- Lance, V. A. (1994). Life in the slow lane: Hormones, stress, and the immune system in reptiles. In "Perspectives in Comparative Endocrinology" (K. G. Davey, R. E. Peter, and S. S. Pobe, Eds.), pp. 529–534. National Research Council of Canada, Ottawa, Ontario.
- Lance, V., and Else, R. M. (1986). Stress-induced suppression of testosterone secretion in male alligators. *J. Exp. Zool.* **239**, 241–246.
- Magnuson, J. J., Bjorndal, K. A., DuPaul, W. D., Graham, G. L., Owens, D. W., Peterson, C. H., Pritchard, P. C., Richardson, J. I., Saul, G. E., and West, C. W. (1990). "Decline of the Sea Turtles: Causes and Prevention." National Academy Press, Washington DC.
- Mahapatra, M. S., Mahata, S. K., and Maiti, B. (1991). Effect of stress on serotonin, norepinephrine, epinephrine and corticosterone contents in the soft-shelled turtle. *Clin. Exp. Pharmacol. Physiol.* **18**, 719–724.
- Moore, M. C., Thompson, C. W., and Marler, C. A. (1991). Reciprocal changes in corticosterone and testosterone levels following acute and chronic handling stress in the tree lizard *Urosaurus ornatus*. *Gen. Comp. Endocrinol.* **81**, 217–226.
- Morris, Y. A. (1982). "Steroid Dynamics in Immature Sea Turtles." Master's thesis, Texas A&M University, College Station, Texas.
- Pritchard, P., Bacon, P., Berry, F., Carr, A., Fletemeyer, J., Gallagher, R., Hopkins, S., Lankford, R., Marquez, R., Ogren, L., Pringle, W., Jr., Reichart, H., and Witham, R. (1983). "Manual of Sea Turtle Research and Conservation Techniques" (K. A. Bjorndal and G. H. Balazs, Eds.). Center for Environmental Education, Washington DC.
- Schmid, J. R. (1995). Marine turtle populations on the east-central coast of Florida: Results of tagging studies at Cape Canaveral, Florida, 1986–1991. *Fish. Bull.* **93**, 139–151.
- Schwantes, N. L. (1986). "Aspects of Circulating Corticosterone in Sea Turtles." Master's thesis, Texas A&M University, College Station, Texas.
- Selye, H. (1976). "The Stress of Life." McGraw-Hill, New York.
- Stephens, D. B. (1980). Stress and its measurement in domestic animals: A review of behavioral and physiological studies under field and laboratory conditions. *Adv. Vet. Sci. Comp. Med.* **24**, 179–210.
- Summers, C. H., and Norman, M. F. (1988). Chronic low humidity-stress in the lizard *Anolis carolinensis*: Changes in diurnal corticosterone rhythms. *J. Exp. Zool.* **247**, 271–278.
- Tam, W. H., Phillips, J. G., and Lofts, B. (1972). Seasonal changes in the secretory activity of the adrenal gland of the cobra (*Naja naja*). *Gen. Comp. Endocrinol.* **19**, 218–224.
- Valverde, R. A., MacKenzie, D. S., Owens, D. W., and Amoss, M. S. (1993). Corticosterone release in response to ACTH and physical disturbance in nesting olive ridley sea turtles, *Lepidochelys olivacea*. *Proc. XII Int. Congress Comp. Endocrinol.*
- Wibbels, T., Owens, D. W., Morris, Y. A., and Amoss, M. S. (1987). Sexing techniques and sex ratios for immature loggerhead sea turtles captured along the Atlantic coast of the United States. In "Ecology of East Florida Sea Turtles" (W. W. Witzell, convener and Ed.), pp. 65–73. NOAA Tech. Rep. NMFS-53.
- Wingfield, J. C. (1988). Changes in reproductive function of free-living birds in direct response to environmental perturbation. In "Processing of Environmental Information in Vertebrates (M. H. Stotson, Ed.), pp. 121–148. Springer-Verlag, New York.
- Wingfield, J. C., Vleck, C. M., and Moore, M. C. (1992). Seasonal changes of the adrenocortical response to stress in birds of the Sonoran desert. *J. Exp. Zool.* **264**, 419–428.