

## SHORTER CONTRIBUTIONS

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### Invasion of New Aquatic Habitats by Male Freshwater Turtles

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Long-term field research often reveals how organisms respond to stochastic environmental events such as droughts (Gibbons et al., 1983) or changes in population structure or species composition over time (Tinkle, 1979; Parker, 1984; Pechmann et al., 1991). Studies on aquatic turtle populations have been conducted for more than 25 years on the US Department of Energy's Savannah River Site (SRS) near Aiken in the Upper Coastal Plain of west central South Carolina (Gibbons et al., 1982; Gibbons, 1990a; Frazer et al., 1991). One conclusion from these studies is that adult males residing in small, isolated wetlands are more likely than females to move overland and to move greater distances (Morreale et al., 1984).

Factors reported to stimulate overland movements by freshwater turtles include travel to and from hibernacula (Bennett, 1972; Gibbons, 1986), pond drying/filling (Cagle, 1944; Sexton, 1959; Gibbons et al., 1983), nesting activity of females (Ernst et al., 1994), and mate-searching by males (Parker, 1984; Gibbons, 1986). Although male and female conspecifics probably exhibit similar terrestrial activity in some situations, differences in reproductive strategies should produce distinct terrestrial activity patterns. Previous studies have suggested that females sometimes travel long distances on land to nest but that males may also travel long distances overland seeking aquatic habitats containing females to inseminate (Morreale et al., 1984; Gibbons, 1986; Brown and Brooks, 1993). If these predictions are correct, males should be more likely than females to encounter new aquatic habitats, including those without conspecific populations. Therefore, colonizing or invading nonresident species should have male-biased sex ratios relative to established resident species. Here we report on captures of 10 species of aquatic turtles and compare sex ratios of six established resident species to those of *Kinosternon baurii* and *Chrysemys picta*.

*Materials and methods.*—Observations described here are based on data collected from 1967–1993 at Ellenton Bay, a freshwater wetland lo-

cated on the SRS. Ellenton Bay is a 10-ha Carolina bay with a maximum water depth of approximately 2 m. Although the bay contains water year-round during most years, surface area and water depth vary seasonally and annually (Gibbons and Semlitsch, 1991).

Turtles were captured using a variety of techniques including aquatic traps, pitfall traps along drift fences, and hand captures. All captured turtles were brought to a laboratory facility, where they were identified to species, measured, sexed, and given individual marks (Gibbons, 1990b). Each turtle was then released at the point of capture. Depending on the species, sexual maturity in males was determined by the presence of enlarged tails or elongated foreclaws. The identification of individuals not readily distinguishable as *K. baurii* or *K. subrubrum* was confirmed by performing the calculations presented by Lamb (1983).

*Results.*—Ten species of aquatic turtles were captured at the Ellenton Bay drift fence between 1967 and 1993: *Trachemys scripta*, *K. subrubrum*, *Pseudemys floridana*, *Sternotherus odoratus*, *Chelydra serpentina*, *Deirochelys reticularia*, *Pseudemys concinna*, *Clemmys guttata*, *Chrysemys picta*, and *K. baurii*. The first six species are known to nest at Ellenton Bay, based on the capture of hatchlings, and have been captured in most years since 1967. These species are considered resident populations. Only mature individuals have been verified for *K. baurii* (n = 30), *C. picta* (n = 5), *C. guttata* (n = 5), and *P. concinna* (n = 1). Hence, these species are considered nonresidents.

Individuals of *C. guttata*, *C. picta*, and *K. baurii* were first captured at Ellenton Bay in 1980, 1980, and 1987, respectively. Prior to 1980, 1663 adults of the six resident turtle species had been captured. An additional 1045 adults of these species were captured at Ellenton Bay after 1980. Of the 2708 mature animals of these six species captured from 1967–1993, 45–64% were males, depending on the species (Table 1).

All *K. baurii* and *C. picta* captured through 1993 at Ellenton Bay have been males (Table 1). One of five *C. guttata* and the one *P. concinna* were females. The residency times for individuals ranged from 14–32 days for *C. guttata*, 21–89 days for *C. picta* and 10–577 days for *K. baurii*. The single *P. concinna* has apparently remained in Ellenton Bay since 1980 based on recaptures over several years.

TABLE 1. NUMBERS OF ADULTS OF EACH SPECIES OF AQUATIC TURTLE CAPTURED AT ELLENTON BAY BETWEEN 1967 AND 1993 AND THE PERCENTAGE THAT WERE MALES.

Species	N	% males
Resident species		
<i>Deirochelys reticularia</i>	363	64
<i>Chelydra serpentina</i>	41	61
<i>Sternotherus odoratus</i>	214	55
<i>Kinosternon subrubrum</i>	860	54
<i>Trachemys scripta</i>	1098	53
<i>Pseudemys floridana</i>	132	45
Non-resident species		
<i>Kinosternon baurii</i>	30	100
<i>Chrysemys picta</i>	5	100
<i>Clemmys guttata</i>	5	80
<i>Pseudemys concinna</i>	1	0

*Discussion.*—The six resident species at Ellenton Bay occur abundantly at several nearby aquatic habitats. The Savannah River, human-made borrow pits, a small stream system, and several ephemeral wetlands are all within 2 km of Ellenton Bay, and overland movement by aquatic turtles between Ellenton Bay and several of these wetlands has been documented through mark-recapture methods (Morreale et al., 1984; Burke et al., 1995). The closest known breeding population of *C. picta* is 18.8 km away, near Jackson, South Carolina. No female *C. picta* has ever been caught in any habitat on the SRS. *Clemmys guttata* has been captured in the vicinity of a marsh-like habitat 600 m away, which is presumably the source of those found at Ellenton Bay. The marshlike area appears more characteristic of the typical habitat of the species (Ernst et al., 1994) than does a Carolina bay.

The nearest habitat where female *K. baurii* have been captured is Four Mile Swamp, 3.4 km away. The nesting behavior of *K. baurii* is not known on the site, but that of *K. subrubrum* has been studied extensively. The average distance from Ellenton Bay of 25 *K. subrubrum* nests was 49.3 m, with a range of 17.3–90.0 m (Burke et al., 1994). Although *K. baurii* nest in autumn (pers. obs.), we assume that the two species have similar nesting patterns spatially. If true, females of *K. baurii* residing in Four Mile Swamp would be unlikely to travel as far as Ellenton Bay during nesting forays.

Longer, more frequent overland travel by males relative to females has been observed in several species of terrestrial and freshwater turtles, including *T. scripta* (Morreale et al., 1984;

Gibbons, 1986) and *Gopherus berlandieri* (Rose and Judd, 1975). Morreale et al. (1984) found that male *T. scripta* were three times more likely than females to move between aquatic habitats. Consequently, males are more likely than females to immigrate into new aquatic habitats, including those unoccupied by females. One possible explanation for this phenomenon is that frequent or long-distance travel by males, especially to new aquatic habitats, will increase encounter rates, and perhaps mating events, with females. We conclude, therefore, that the occurrence of *K. baurii*, as well as *C. picta*, at Ellenton Bay is a result of relatively recent invasion by adult males that have made long-range excursions not typically undertaken by females.

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### Phylogenetic Relationships of Western Atlantic Snappers (Family Lutjanidae) Based on Mitochondrial DNA Sequences

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Despite the importance of lutjanids, their taxonomy remains problematic. Most of the tax-

onomic uncertainty involves the large subfamily Lutjaninae. Much of the uncertainty stems from morphological similarity within the group and the evident ability of several species to hybridize (Domeier and Clarke, 1992; Loftus, 1992). The subfamily Lutjaninae has three genera represented in the western Atlantic: *Lutjanus*, *Ocyurus*, and *Rhomboplites*. Separation of monotypic *Ocyurus* and *Rhomboplites* from *Lutjanus* is based mainly on cranial differences and on several other morphological characteristics (Evermann and Marsh, 1900). Three other genera are represented in the western Atlantic (*Etelis*, *Pristipomoides*, and *Apsilus*) with a total of 15 species.

Recent advances in DNA technology and molecular genetics have provided new methods for examining evolutionary relationships of closely related taxa. Mitochondrial DNA (mtDNA) has proved especially useful in resolving phylogenetic relationships among species that differ little in morphology (Bucklin et al., 1992; Bowers et al., 1994). Development of the polymerase chain reaction (PCR) (Saiki et al., 1988) and the discovery of highly conserved regions of the genome that can be targeted to make “universal” primers for PCR (Kocher et al., 1989) now make acquisition of DNA sequence available for a wide range of taxa. In this study, we compare DNA sequences from portions of two mitochondrial genes, 12S rRNA (12S) and cytochrome *b* (*cyt b*), to examine phylogenetic relationships of 14 species of snappers occurring in the western Atlantic Ocean.

*Materials and methods.*—Specimens were collected in 1992 and 1993 by hook and line near Miami and the Florida Keys. All species of snappers found in the western Atlantic were collected except for black snapper (*Apsilus dentatus*), which is not usually found near Miami or the Florida Keys. Fish were held on wet ice until taken to Miami for storage at  $-70^{\circ}\text{C}$ . Total genomic DNA was extracted from frozen gonad or muscle by using standard phenol/chloroform extraction (Hillis et al., 1990). Double-stranded DNA was amplified in 50  $\mu\text{L}$  reactions containing 50 mM KCl, 10 mM Tris-HCl, 1.5 mM  $\text{MgCl}_2$ , 0.1% Triton X-100, 200  $\mu\text{M}$  of each dNTP, 1  $\mu\text{M}$  of each oligonucleotide primer, 0.5 unit of *Taq* polymerase, and 1  $\mu\text{L}$  of DNA sample. Two gene regions of the mitochondrial genome were amplified: a 359 bp region of the *cyt b* gene and a 450 bp region of the 12S gene. Primers used to amplify these regions were shortened versions of those described in Kocher et al. (1989): *cyt b*, L14841, GCTTCCAT-CCAACATCTCAGCATGATG; and H15149, GCAGCCCTCAGAATGATATTTGTCC-