# EFFECTS OF RELOCATION AND ENVIRONMENTAL FACTORS ON LOGGERHEAD SEA TURTLE (*Caretta caretta*) NESTS ON CAPE ISLAND

A thesis submitted in partial fulfillment of the requirements for the degree

# **MASTER OF SCIENCE**

in

# **ENVIRONMENTAL STUDIES**

by

# MELISSA KENNEDY BIMBI OCTOBER 2009

at

# THE GRADUATE SCHOOL OF THE COLLEGE OF CHARLESTON

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#### ABSTRACT

# EFFECTS OF RELOCATION AND ENVIRONMENTAL FACTORS ON LOGGERHEAD SEA TURTLE (*Caretta caretta*) NESTS ON CAPE ISLAND

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MASTER OF SCIENCE in ENVIRONMENTAL STUDIES by MELISSA KENNEDY BIMBI OCTOBER 2009 at

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Cape Island is the highest-density nesting beach of the northern nesting assemblage of the Northwest Atlantic population of loggerhead sea turtles (*Caretta caretta*). In order to determine the effect of nest relocation, *in situ*, hatchery and individually relocated nests were monitored throughout the peak of the 2007 nesting season and the entire 2008 nesting season. MicroDAQ<sup>TM</sup> LogTag temperature data loggers ( $\pm 0.1^{\circ}$ C error) were placed in the approximate center of nests during the entire incubation duration. Environmental factors such as sand characteristics, vegetation, inundation, and elevation were also examined. Hatchery nests incubated at cooler temperatures than *in situ* nests and had longer incubation durations. Individually relocated nests incubated at similar temperatures as *in situ* nests and had similar incubation durations. Inundation was significantly higher in *in situ* nests, and elevation was significantly lower in inundated nests. Hatch and emergence success were similar between all nest types. This research suggests that nest relocation, when used correctly, remains an important management tool for sea turtle conservation and the need for it may increase with rising sea levels.

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#### **CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW**

The loggerhead sea turtle (*Caretta caretta*) is listed as endangered on the International Union for the Conservation of Nature (IUCN) Red List and threatened in the U.S. under the Endangered Species Act (USFWS and NMFS 1978, MTSG 1996). Loggerheads inhabit the continental shelves and estuarine environments along the margins of the Atlantic, Pacific, and Indian Oceans. In the continental U.S., loggerheads nest from Louisiana to Virginia and major nesting concentrations occur on the coastal islands of North Carolina, South Carolina, and Georgia, and on the Atlantic and Gulf coasts of Florida (NMFS and USFWS 2008).

All species of sea turtle embryos and hatchlings face many threats on their natal beaches worldwide including poaching, native and non-native predators, storm and tidal inundation, beach erosion, and coastal development and artificial lighting (Witherington 1999). Nest protection projects have been implemented globally in order to aid in the recovery of the all sea turtle species. Nest relocation is a management tool often used when nests are oviposited below the spring high tide line or on highly erosive beaches. This management practice has effectively increased nest productivity (Hopkins and Murphy 1983, Stancyk *et al.* 1980, Eckert and Eckert 1990). However, in the Southeastern U.S., concerns have recently arisen about the high percentage of relocated nests and the possible negative effects of this practice on sex ratios, hatch success, hatchling emergence, and hatchling fitness (Morreale *et al.* 1982, Godfrey *et al.* 1997, Pintus *et al.* 2009, Moody 2000, Pilcher and Enderby 2001, Booth *et al.* 2004, Adam *et al.* 2007).

Loggerheads like all other sea turtles have an environmental sex determination (ESD) mechanism in the form of temperature dependent sex determination (TSD) (Standora and Spotila 1985, Janzen and Paukstis 1991), which occurs during the middle third of incubation (Yntema and Mrosovsky 1980, Vogt and Bull 1982, Mrosovsky and Pieau 1991). The range of incubation temperatures in which both males and females are produced is called the transitional range of temperature (TRT) (Godfrey et al. 1997). The TRT is centered around the pivotal temperature, which is the temperature that produces a 1:1 sex ratio. For loggerheads, the pivotal temperature is 29–30°C and the TRT extends 2 to 3°C above and below the pivotal temperature (Mrosovsky 1994, Wibbels 2005). Incubation temperatures above the pivotal temperature produce female biased sex ratios, and incubation temperatures below produce male biased sex ratios (Wibbels 2005). Incubation temperatures outside the TRT result in 100% males or 100% females (Godfrey et al. 1997). Species with TSD are vulnerable to environmental factors that influence temperature; therefore, any nest manipulation of the incubation environment, even for conservation purposes, has the potential to alter and skew sex ratios of all life stages of sea turtle populations (Godfrey et al. 1997).

Studies have found that relocated nests can incubate at different temperatures than *in situ* nests (Hoekert *et al.* 1998, Başkale and Kaska 2005, Mrosovosky and Yntema 1980, Tuttle 2007, and Pintus *et al.* 2009) causing skewed sex ratios (Morreale *et al.* 1982, Godfrey *et al.* 1997). Hoekert *et al.* (1998) and Başkale and Kaska (2005) both reported that relocated nests incubated at warmer temperatures than *in situ* nests. Mrosovosky and Yntema (1980) found the incubation of eggs in styrofoam boxes above ground produced mostly males. However, García *et al.* (2000), Tuttle (2007),

and Pintus *et al.* (2009) found no difference between relocated and *in situ* nest temperatures.

Studies have also found that relocated nests can have higher or lower hatch success rates than *in situ* nests (Wyneken *et al.* 1988, Hoekert *et al.* 1998, Moody 2000, Kornaraki *et al.* 2006, Pintus *et al.* 2009). Wyneken *et al.* (1988), Hoekert *et al.* (1998), Kornaraki *et al.* (2006), and Tuttle (2007) reported higher hatch success rates in relocated nests than *in situ* nests. Moody (2000) and Pintus *et al.* (2009) reported lower hatch success rates in relocated nests than *in situ* nests than *in situ* nests. However, García *et al.* (2000) reported no difference between the two. This suggests that local differences in relocation practices and environmental conditions play an important role in determining incubation temperature and nest success.

The purpose of this study was to determine the effects of relocation and environmental factors on loggerhead nest incubation temperature, incubation duration, hatch success, and emergence success on Cape Island. Chapter 2 examines the differences between *in situ* and hatchery nests during the 2007 and 2008 nesting seasons. Chapter 3 examines the differences between *in situ* and individually relocated nests during the 2008 nesting season. Chapter 4 evaluates both methods of relocation and examines the current management practices on Cape Island.

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# CHAPTER 2: HATCHERY USE AT THE HIGHEST DENSITY NESTING SITE FOR THE NORTHERN NESTING ASSEMBLAGE OF THE NORTHWEST ATLANTIC LOGGERHEADS

#### To be submitted to the Journal of Wildlife Mangement

Abstract: Cape Island is the highest-density nesting beach of the northern nesting assemblage of the Northwest Atlantic population of loggerhead sea turtles (*Caretta caretta*). In order to determine the effect of nest relocation, *in situ*, and hatchery nests were monitored throughout the peak of the 2007 and 2008 nesting season. MicroDAQ<sup>TM</sup> LogTag temperature data loggers ( $\pm 0.1$ °C error) were placed in the approximate center of nests during the entire incubation duration. Differences in environmental factors such as sand characteristics and elevation were also examined between *in situ* and hatchery nests. Hatchery nests incubated at cooler temperatures than *in situ* nests and had longer incubation durations. Hatch and emergence success were similar between both nest types. This research suggests that hatcheries, when situated in habitat similar to *in situ* nests, can be an important management tool for sea turtle conservation on beaches with high predation rates and severe erosion.

**Keywords:** *Caretta caretta*, loggerhead sea turtle, nest relocation, incubation temperature, hatchery, management

# Introduction

Nests relocated into hatcheries can skew sex ratios, reduce hatch success, and delay hatchling release potentially causing a reduction in fitness (Mrosovosky and Yntema 1980, Morreale *et al.* 1982, Godfrey *et al.* 1997, Hoekert *et al.* 1998, Mortimer 1999, Moody 2000, Pilcher and Enderby 2001, Başkale and Kaska 2005, Tuttle 2007, and Pintus *et al.* 2009). In order to control raccoon predation, hatcheries have been used on Cape Island since 1979. Cape Island, the largest island in the

Cape Romain National Wildlife Refuge (CRNWR), is the highest-density nesting beach of the northern nesting assemblage of the Northwest Atlantic population of loggerhead sea turtles (Caretta caretta) (NMFS and USFWS 2008). The U.S. Fish and Wildlife Service (USFWS) manages CRNWR and implements predator management programs, conducts nesting surveys, and carries out nest protection measures for the recovery of loggerheads. Land-based predators on Cape Island include raccoons (Procyon lotor), ghost crabs (Ocpode quadrata), rats (Ratus norvegicus), mink (Mustela vison), and sea gulls (Larus sp.) (USFWS 2007, 2008). In response to predation and severe erosion, the USFWS relocates nests into hatcheries during the peak of nesting season when more than ten nests are oviposited per day. Hatcheries are self-releasing and constructed with a PVC pipe frame with hardware cloth covering the sides, and a metal frame lid with chicken wire attached to the frame. The hatcheries have a capacity of either 50 or 100 nests and are located either seaward or landward of the primary dune depending on habitat availability and suitability. Hatch success for nests relocated into hatcheries on Cape Island ranges from 61.7% to 90.2% with a median of 80.1% (USFWS 2008).

Given the concerns about the effect of hatchery use on incubation temperature and nest success, we examined several questions regarding nest relocation into hatcheries on Cape Island: (1) Are hatchery nest incubation temperatures during the sex determining period warmer than *in situ* nest incubation temperatures? (2) Are there differences between *in situ* and hatchery hatch and emergence success?

# Methods

# Study Site

Cape Island (32°59' N, 79°20' W) is a 10 km undeveloped barrier island located off the coast of South Carolina, USA (Figure 1). The island is the northeast portion of

South Carolina's only cuspate foreland (cape), and has one of the most erosional coastlines in the state (Hayes and Michel 2008, Hopkins-Murphy and Murphy 1983), which loses approximately 7.6 m of beach/year (USFWS 2008). The majority of the beach is backed by salt marsh (*Spartina sp.*). Sea Oats (*Uniola paniculata*) and Sea Rocket (*Cakile harperi*) are the dominant vegetation on the remaining sand dunes. The intertidal zone slopes steeply down from the base of the dunes. The tidal range is 0–3 m, but it can be higher depending on spring tides and wind direction. At the lowest tide, the width of the beach ranges from 10–30 m seaward of the sand dunes, and at the highest tide the width ranges from 0–3 m.

# Nest Relocation

We identified nests by walking the fresh incoming turtle crawls to the body pit and used a 1 cm diameter blunt steel probe to confirm the presence and location of the nest by probing along the centerline where thrown or disturbed sand covered the incoming crawl. Once we located the egg chamber, we either left the nest in place (*in situ*) or relocated it into a hatchery if it was located below the spring high tide line or in a highly erosional area. In 2008, we also monitored nests that were not placed into hatcheries, but were individually relocated (see Chapter 3). For *in situ* nests, we removed sixty eggs from each nest,  $\frac{1}{2}$  of the average clutch size for Cape Island (USFWS unpublished data), and inserted a data logger upright in the center of the nest with sand, and covered the nest with a 5x10 cm welded 14 gauge wire cage with 61x61x41 cm dimensions over the nest to protect it from raccoons (*Procyon lotor*). We marked the nest with a numbered orange tipped PVC pole standing approximately 1 m behind the center of the nest.

All hatchery nest egg chambers were dug to a depth of 55 cm in order to match the mean nest depth on Cape Island (Sarah Dawsey, USFWS, personal communication). We placed half of the clutch in the new egg chamber, and inserted a data logger upright in the center of the nest with the sensor facing north, and placed the remaining eggs around and above the data logger. We placed data loggers in a subset of nests in each hatchery and marked these nests with numbered survey flags.

#### Incubation Temperature

We placed MicroDAQ LogTag temperature data loggers with a ±0.1°C accuracy in the approximate center of *in situ* and hatchery nests during the entire incubation duration of the 2007 and 2008 nesting season. We labeled the loggers, sealed them in food saver plastic, and set them to record every ½ hour before the field season started. We also placed the loggers in an incubator at 30°C before and after deployment in order to determine accuracy and consistency.

# Environmental Characteristics

We measured environmental characteristics during the 2008 season in order to determine if the variability could be related to environmental conditions or nest relocation. We collected sediment samples 1 m from each *in situ* and each hatchery by excavating 25 cm of sand with a garden trowel, then driving in a plastic corer (3.65 cm diameter, 55 cm long) with a rubber mallet until it was level with the surface of the sand. We emptied the contents of the core into a pre-weighed, labeled quart-size plastic freezer bag and stored each sample in a cooler. The push core represented an integrated average of sediment characteristics within the egg chamber. We froze all samples for later analysis of sand grain phi size, silt/clay fractions, moisture content, calcium carbonate (CaCO<sub>3</sub> content), and organic matter (see Chapter 3 for methods).

#### Elevation

We determined the exact location and elevation of each *in situ* nest and hatchery monitored in this study using a Trimble R-8 GNSS (Global Navigation Satellite System). This unit has a horizontal accuracy of  $\pm 5 \text{ mm} + 0.5 \text{ ppm}$  (parts-permillion) Remote Monitoring System (RMS) and a vertical accuracy of  $\pm 5 \text{ mm} + 1$ ppm RMS. For *in situ* nests, we placed the unit on the sand surface next to the cage. For each hatchery, we placed the unit next to each of the four corners and averaged the four values to get an average elevation.

# Nest Success

Three days after the first emergence of hatchlings, we inventoried all nests with data loggers in order to determine the hatch success and emergence success. We excavated all nest contents and separated eggs or hatchlings into the following categories: empty shells (50% or more intact), unhatched eggs, pipped eggs (eggs with openings that contained a dead hatchling), dead hatchlings, and live hatchlings. We calculated hatch success and emergence success for each nest in accordance with the methods in Miller (1999). We defined hatch success as the number of hatchlings that hatch out of their shell and calculated it as follows:

# total # hatched x 100 total eggs

We defined emergence success as the total number of hatchlings out of the total clutch that emerged from the nest and calculated it as follows:

# <u>total # hatched – total # live and dead hatchlings</u> x 100 total eggs

We defined and calculated an additional term, hatched emergence success, as the number of hatchlings out of the hatched eggs that emerged from the nest and calculated it as follows:

# total # hatched – total # live and dead hatchlings x 100 total hatched eggs

The purpose of this additional calculation was to detect if nest relocation reduced emergence ability. We defined incubation duration or incubation-to-emergence period as the number of days beginning with the date of the morning after the nest deposition through the date of the first emergence (Miller *et al.* 2003).

#### Data Analysis

We downloaded temperature data into LogTag Analyzer and exported the data into Microsoft Excel. To calculate incubation temperature, we averaged temperatures during the middle third of incubation, which is the sex-determining period (Yntema and Mrosovsky 1980, Vogt and Bull 1982, Mrosovsky and Pieau 1991). We used nest inventory data to calculate incubation duration, hatch success, emergence success, and hatched emergence success. All data were exported into Minitab 15 for analysis and were transformed to meet the assumptions of parametric statistics. We used an analysis of variance (ANOVA) to determine differences between *in situ* and hatchery nest incubation temperatures and incubation durations ( $\alpha = 0.05$ ). Since hatch and emergence success data were not normally distributed, we used a Kruskal-Wallis test to determine differences between *in situ* and hatchery nest hatch and emergence success.

#### Results

Hatchery nest incubation temperatures were significantly cooler than *in situ* nests  $(F_{1,223}=123.99, p < 0.000)$  (Figure 2). Hatchery nest incubation temperatures were significantly warmer in 2007 than in 2008  $(F_{1,223}=237.89, p < 0.000)$  (Figure 2). Hatchery nest incubation durations were significantly longer than *in situ* nests both years  $(F_{1,123}=100.22, p < 0.000)$  (Figure 3) and hatchery nest incubation durations were significantly shorter in 2007 than in 2008  $(F_{1,123}=152.30, p < 0.000)$  (Figure 3).

Hatch success and emergence success were not significantly different between hatchery and *in situ* nests in 2007 (n = 125, Z = ±0.91, p = 0.363; Z = ±1.0, p = 0.319, respectively) (Figure 4a and 4b) or in 2008 (n = 122, Z = ±0.39, p = 0.697; Z = ±0.02, p = 0.988, respectively) (Figure 4a and 4b). However, they were significantly different between the three hatcheries in 2008 (n = 60, Z = -1.10, -1.90, 2.66, p = 0.020) because hatchery 2 had a lower hatch and emergence success than the other hatcheries (Figure 4a and 4b). Conversely, hatched emergence success was significantly higher in hatchery nests as compared to *in situ* nests in 2007 (n = 125, Z = ± 9.33, p < 0.000) (Figure 4c) and 2008 (n = 152, Z = ± 4.74, p < 0.000) (Figure 4c). However, it was not significantly different among hatcheries in 2007 (n = 53, Z = ±0.03, p = 0.976) (Figure 4c) or 2008 (n = 60, Z = -0.27, 0.11, 0.18, p = 0.472) (Figure 4c). Elevation and all sediment characteristics except silt/clay content were higher for 2007 hatcheries than 2008 hatcheries (Table 1).

#### Discussion

Hatchery nests incubated at cooler temperatures and had longer incubation durations during the 2007 and 2008 nesting season, but there was a lot of variability between seasons. Hatcheries in 2007 and 2008 did alter the nest incubation environment and possibly altered sex ratios compared to *in situ* nests because incubation temperatures were cooler. Cooler temperatures also extended the incubation duration. The differences between hatchery and *in situ* nest incubation durations in 2007 and in 2008 were 1.6 days and 6.3 days, respectively. Davenport (1997) reported that cooler incubation temperatures slowed development. However, we did not determine how cooler incubation temperatures influenced the development of embryos in our study. It is possible that the location of the hatchery influenced incubation temperatures (Table 1). There was little variability in temperature between

nests in the hatcheries; therefore, poor site selection could impact all nests in the hatchery. The hatcheries were located in areas with a higher calcium carbonate content and a smaller phi size, which translates to larger sand particle size, than *in situ* nests (Table 1). In a study by Speakman et al. (1998), large shell fragments and large particle size correlated with reduced conductivity of heat because larger shell fragments and particle sizes trap more air and act as an insulator; therefore, heat transfers less efficiently. This may explain why hatchery nests were incubating at cooler temperatures than *in situ* nests. It is also possible that the higher moisture content found in hatchery sand samples also contributed to the cooling effect on hatchery nests (Table 1). Hatcheries were also located at lower elevations, which also could have had a cooling effect on incubation temperatures (Table 1). Foley (1998) reported cooler incubation temperatures in areas closer to the water in a study in the Ten Thousand Islands of Florida. In that study, 46.1% of nests experienced some degree of groundwater inundation and groundwater was present at a depth of about 0.5 to 0.75 meters. The influence of groundwater on Cape Island has not been determined.

Nest relocation into hatcheries on Cape Island did not decrease overall hatch or emergence success as compared to *in situ* nests. Analysis of preliminary South Carolina Department of Natural Resources (SCDNR) data for the 21 nest protection projects in South Carolina from 2000 through 2006 shows similar results. Median hatch success for *in situ* nests and relocated nests were 82.1% and 83.7%, respectively. Median emergence success for *in situ* nests and relocated nests was 77.9% and 79.7%, respectively (SCDNR unpublished data). Hatched emergence success, however, was significantly higher in hatchery nests than *in situ* nests also indicating that nest relocation on Cape Island does not decrease the overall emergence

success of the hatched eggs. It is unclear why *in situ* nests had a lower hatched emergence success, but it is possible that roots in the egg chamber from vegetation around *in situ* nests and compaction and sand deposition from overwash events were influencing factors. Hatcheries were typically located away from vegetation and areas subject to inundation. It is also unclear why hatch success and emergence success were significantly different among hatcheries in 2008, but not in 2007. However, it does illustrate the importance of hatchery site selection and the consideration of environmental characteristics influential on nest incubation. García *et al.* (2003) reported no significant differences between hatchery and *in situ* nest incubation temperatures during the sex-determining period or hatch success. Morreale *et al.* (1982) suggested that beach hatcheries can maintain natural sex ratios if care is taken to mimic the *in situ* environment.

In 1999, the Marine Turtle Specialist Group recommended that hatcheries only be used on beaches where *in situ* nest protection is impossible because depredation rates are high (Mortimer 1999). In the past, raccoon predation rates on Cape Island have been high, which is why hatcheries were started. However, in 2007 and in 2008 predation rates were 0.0% and 0.2%, respectively, due to intensive trapping efforts from 2004–2008 (USFWS 2007, USFWS 2008). Even with raccoon predation rates currently low, the USFWS continues to relocate nests into hatcheries during the peak of nesting season because suitable habitat for relocation is limited and hatcheries can hold more nests than individually caged nests in the same amount of space. The National Marine Fisheries Service (NMFS) and the USFWS Recovery Plan deems hatchery use on Cape Island appropriate at this time since the conservation benefits outweigh the conservation risks (NMFS and USFWS 2008). Given the accelerated

loss of suitable habitat on Cape Island, hatchery site selection will become increasingly more challenging.

#### **Summary and Conclusions**

Hatcheries in 2007 and in 2008 did alter the nest incubation environment and possibly altered sex ratios as compared to *in situ* nests because incubation temperatures were cooler. However, relocation into hatcheries did not decrease overall hatch or emergence success as compared to *in situ* nests. Since there was little variability in temperature between nests in the hatcheries, poor site selection could impact all nests in the hatchery illustrating the importance of hatchery site selection.

Cape Island continues to experience accelerated erosion from both natural and man-made sources (USFWS 2008). Increases in sea-level will bring higher tides as well as the increased potential for stronger storms (Karl *et al.* 2009) compounding existing conditions and increasing the need for nest relocation. Hatcheries will need to continue to be used on Cape Island because suitable habitat for relocation is limited and hatcheries can hold more nests than individually caged nests in the same amount of space. Since CRNWR is the largest rookery for the northern nesting assemblage of Northwest Atlantic loggerheads, conservation efforts carried out here will go further towards the recovery of this assemblage and the population. If this assemblage is lost, it will not be replaced by recruits from other nesting assemblages due to high nesting site fidelity (Schroeder *et al.* 2003). Although hatcheries limit the incubation environment variability compared to natural nesting areas, careful site selection and consideration of environmental characteristics present on the natural nesting beach, make hatcheries a feasible management strategy on Cape Island in order to aid in the recovery of this species.

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Figure 1. Location of Cape Island.



□ In Situ 
□ Average Hatchery 
□ Hatchery 1 □ Hatchery 2 □ Hatchery 3

**Figure 2.** Incubation temperature comparison between *in situ* and hatchery nests in 2007 and 2008. Data are means ( $\pm$ SE). \*Sex predictions based on Wibbels 2005.



Figure 3. Incubation duration comparison between *in situ* and hatchery nests in 2007 and 2008. Data are means ( $\pm$ SE).







Figure 4. Comparison of 2007 and 2008 *in situ* and hatchery hatch, emergence, and hatched emergence success. Data are means ( $\pm$ SE).

а

				Sediment Characteristics					
				Moisture			Organic	Sand	
				Content	Silt/Clay	CaCO <sub>3</sub>	Matter	Phi Size	Elevation
Year	Nest Type	Size	Location	(%)	(%)	(%)	(%)	( <b>φ</b> )	(m)
2007*	Hatchery 1, 2	50, 100	Landward of dune, north end	9.05	0.00	88.63	1.12	1.2	2.4
2008	Hatchery 1	100	Seaward of dune, north end	8.63	0.00	44.30	0.80	0.9	1.9
2008	Hatchery 2	50	Seaward of dune, south end	7.56	0.00	23.73	0.70	0.7	2.1
2008	Hatchery 3	100	Landward of dune, north end	7.60	0.00	25.06	0.29	0.7	1.9
2008	In situ		Above SHTL** on island	3.24	0.12	3.03	0.19	1.3	2.5

**Table 1.** Comparison of hatchery and *in situ* locations and sediment characteristics. Sediment characteristics were determined from one sample collected at each hatchery and *in situ* nest. \*Data collected in 2008. \*\*Spring high tide line.

# CHAPTER 3: NEST RELOCATION: THE GOOD, THE BAD AND THE REALITY

#### To be submitted to Conservation Biology

**Abstract:** Cape Island is the highest-density nesting beach of the northern nesting assemblage of the Northwest Atlantic population of loggerhead sea turtles (*Caretta caretta*). In order to determine the effect of nest relocation, *in situ* and individually relocated nests were monitored throughout the peak of the 2008 nesting season. MicroDAQ<sup>TM</sup> LogTag temperature data loggers with  $\pm 0.1$ °C accuracy were placed in the approximate center of nests during the entire incubation duration. Environmental factors such as sediment characteristics, vegetation, inundation, and elevation were also examined. Individually relocated nests incubated at similar temperatures and had similar incubation durations as *in situ* nests, but both nest types varied with the time of season. Inundation was significantly higher in *in situ* nests, and elevation was significantly lower in inundated nests. Hatch and emergence success were similar between relocated and *in situ* nests, but both measures were significantly lower in inundated nests. This research suggests that nest relocation, when used correctly, remains an important management tool for sea turtle conservation and the need for it may increase with rising sea levels.

**Keywords:** *Caretta caretta, in situ*, nest relocation, temperature, management, conservation

# Introduction

Loggerheads like all other sea turtles have an environmental sex determination (ESD) mechanism in the form of temperature dependent sex determination (TSD) (Standora and Spotila 1985, Janzen and Paukstis 1991), which occurs during the

middle third of incubation (Yntema and Mrosovsky 1980, Vogt and Bull 1982, Mrosovsky and Pieau 1991). The range of incubation temperatures in which both males and females are produced is called the transitional range of temperature (TRT) (Godfrey *et al.* 1997). The TRT is centered around the pivotal temperature, which is the temperature that produces a 1:1 sex ratio. For loggerheads, the pivotal temperature is 29–30°C and the TRT extends 2 to 3°C above and below the pivotal temperature (Mrosovsky 1994, Wibbels 2005). Incubation temperatures above the pivotal temperature produce female biased sex ratios, and incubation temperatures below produce male biased sex ratios (Wibbels 2005). Incubation temperatures outside the TRT result in 100% males or 100% females (Godfrey *et al.* 1997). Species with TSD are vulnerable to environmental factors that influence temperature; therefore, any nest manipulation of the incubation environment, even for conservation purposes, has the potential to alter and skew sex ratios of all life stages of sea turtle populations (Godfrey *et al.* 1997).

Studies have found that relocated nests can incubate at different temperatures than *in situ* nests (Hoekert *et al.* 1998, Başkale and Kaska 2005, Mrosovosky and Yntema 1980, Tuttle 2007, and Pintus *et al.* 2009) causing skewed sex ratios (Morreale *et al.* 1982, Godfrey *et al.* 1997). Hoekert *et al.* (1998) and Başkale and Kaska (2005) both reported that relocated nests incubated at warmer temperatures than *in situ* nests. Mrosovosky and Yntema (1980) found the incubation of eggs in styrofoam boxes above ground produced mostly males. However, García *et al.* (2000), Tuttle (2007), and Pintus *et al.* (2009) found no difference between relocated and *in situ* nest temperatures.

Studies have also found that relocated nests can have higher or lower hatch success rates than *in situ* nests (Wyneken *et al.* 1988, Hoekert *et al.* 1998, Moody 2000,

Kornaraki *et al.* 2006, Pintus *et al.* 2009). Wyneken *et al.* (1988), Hoekert *et al.* (1998), Kornaraki *et al.* (2006), and Tuttle (2007) reported higher hatch success rates in relocated nests than *in situ* nests. Moody (2000) and Pintus *et al.* (2009) reported lower hatch success rates in relocated nests than *in situ* nests. However, García *et al.* (2000) reported no difference between the two. This suggests that local differences in relocation practices and environmental conditions play an important role in determining incubation temperature and nest success. Nest relocation is a management tool often used when nests are oviposited below the spring high tide line or on highly erosive beaches. This management practice has effectively increased nest productivity (Hopkins and Murphy 1983, Stancyk *et al.* 1980, Eckert and Eckert 1990). Concerns have recently arisen about the high percentage of relocated nests and the possible negative effects of this practice on sex ratios, hatch success, and hatchling emergence (Morreale *et al.* 1982, Godfrey *et al.* 1997, Moody 2000, Booth *et al.* 2004, Adam *et al.* 2007, Pintus *et al.* 2009).

The Cape Romain National Wildlife Refuge (CRNWR) comprises 25–32% of the nests laid in South Carolina, USA (Hopkins-Murphy and Murphy 1983). Cape Island, the largest island in CRNWR, is the highest-density nesting beach of the northern nesting assemblage of the Northwest Atlantic population of loggerhead sea turtles (*Caretta caretta*) (NMFS and USFWS 2008). The U.S. Fish and Wildlife Service (USFWS) manages CRNWR and implements predator management programs, conducts nesting surveys, and carries out nest protection measures for the recovery of loggerheads. In response to a 3 m tidal amplitude and severe erosion, the USFWS relocates nests deemed vulnerable to tidal inundation. Since approximately half of the nests on Cape Island are relocated, we wanted to determine the effect of relocation on nest success so we addressed the following questions: (1) Are relocated nest

incubation temperatures warmer than *in situ* nest incubation temperatures? (2) Are there differences between *in situ* and relocated nest hatch and emergence success? (3) Are there differences between *in situ* and relocated nest incubation environments? (4) What effect does tidal inundation have on nest hatch and emergence success?

#### Methods

#### Study Site

Cape Island (32°59' N, 79°20' W) is a 10 km undeveloped barrier island located off the coast of South Carolina, USA (Figure 1). The island is the northeast portion of South Carolina's only cuspate foreland (cape), and has one of the most erosional coastlines in the state (Hayes and Michel 2008, Hopkins-Murphy and Murphy 1983), which loses approximately 7.6 m of beach/year (USFWS 2008). The majority of the beach is backed by salt marsh (*Spartina sp.*). Sea Oats (*Uniola paniculata*) and Sea Rocket (*Cakile harperi*) are the dominant vegetation on the remaining sand dunes. The intertidal zone slopes steeply down from the base of the dunes. The tidal range is 0–3 m, but it can be higher depending on spring tides and wind direction. At the lowest tide, the width of the beach ranges from 10–30 m seaward of the sand dunes, and at the highest tide the width ranges from 0–3 m.

# Nest Relocation

We identified nests by walking the fresh incoming turtle crawls to the body pit and used a 1 cm diameter blunt steel probe to confirm the presence and location of the nest by probing along the centerline where thrown or disturbed sand covered the incoming crawl. Once we located the egg chamber, we either left the nest in place (*in situ*) or relocated it if it was located below the spring high tide line or in a highly erosional area. For *in situ* nests, we removed sixty eggs from each nest, <sup>1</sup>/<sub>2</sub> of the average clutch size for Cape Island (USFWS unpublished data), and inserted a data logger upright in the center of the nest with the sensor facing north (Figure 2). We placed the eggs back in the egg chamber, packed it with sand, and covered the nest with a 5x10 cm welded 14 gauge wire cage with 61x61x41 cm dimensions over the nest to protect it from raccoons (*Procyon lotor*). We marked the nest with a numbered orange tipped PVC pole standing approximately 1 m behind the center of the nest.

Relocated nests were moved to similar areas as *in situ* nests laid above the spring high tide line. We counted the eggs, placed them in a bucket, and covered them with a hand towel and damp sand from the egg chamber. We measured the original egg chamber depth in centimeters for all relocated nests. We used post hole diggers to dig a hole approximately 50–55 cm deep (average depth of *in situ* nest chambers on Cape Island, S. Dawsey, personal communication, 2007) and bowled out the bottom of the hole to create the new egg chamber and measured the depth of the new nest cavity. We placed half of the clutch in the new egg chamber, and inserted a data logger upright in the center of the nest with the sensor facing north. We placed the remaining eggs in the new chamber and packed it with sand. These nests were caged and marked as described above (Figure 2). Nests were also relocated into hatcheries during the 2008 nesting season, which were discussed in Chapter 2.

# Incubation Temperature

We placed MicroDAQ LogTag temperature data loggers with a  $\pm 0.1$  °C accuracy in the approximate center of *in situ* and relocated nests during the entire incubation duration of the 2008 nesting season. We labeled the loggers, sealed them in food saver plastic, and set them to record every ½ hour before the field season started. We also placed the loggers in an incubator at 30°C before and after deployment in order to determine accuracy and consistency. Since data loggers were the limited resource

for this study, we determined the sample size needed by using a power analysis of temperature data collected in 2007. We calculated power based on the standard deviation of *in situ* nest incubation temperatures during the middle third of incubation because *in situ* nests exhibited more variability, which gave a more conservative estimate of power. In order to detect a  $\frac{1}{2}$  °C difference with 95% power (actual power = 0.953533), we needed a minimum sample size of 48 for each treatment during the nesting season. Based on this analysis, we inserted data loggers in 12 *in situ* and 12 relocated nests during each of four time periods: 16–24 May, 9–14 June, 3–8 July, and 29 July–4 August, 2008.

#### Environmental Characteristics

We included environmental characteristics in order to determine if the variability detected was due to environmental conditions or nest relocation. We collected the samples on July 8, 2008, for each nest laid in May and June and on August 19, 2008, for each nest laid in July and August since nests were laid throughout the season. We collected sediment samples 1 m from each *in situ* and relocated nest by excavating 25 cm of sand with a garden trowel, then driving in a plastic corer (3.65 cm diameter, 55 cm long) with a rubber mallet until it was level with the surface of the sand. We emptied the contents of the core into a pre-weighed, labeled quart-size plastic freezer bag and stored each sample in a cooler. The push core represented an integrated average of sediment characteristics within the egg chamber. We froze all samples for later analysis of sand grain phi size, silt/clay fractions, moisture content, calcium carbonate (CaCO<sub>3</sub> content), and organic matter.

# Moisture Content

We thawed all samples, wiped off any condensation from the bag, and weighed the sample in the bag. We emptied the contents of the bag into to a clean pre-weighed

600 mL beaker, and recorded the weight. We placed each sample in a drying oven (60°C) until dry, transfered it to a desiccator to cool, and then weighed the sample. We weighed the empty bag after it air dried in order to determine how much of the sample remained in the bag, so we could calculate the error. We calculated percent moisture as follows:

# wet sample weight – dry sample weight x100 wet sample weight

#### Silt/Clay Fractions

We processed each sample for silt/clay content using methods described in Plumb (1981). We took a 20g sub-sample from each core sample after processing it for moisture content. We added twenty mL of dispersant (6.2g/L sodium hexametaphosphate) to each sample, placed a 1000 mL graduated cylinder under a funnel and 63 µm sieve, and rinsed the sample through the sieve with distilled water until the water was clear. We then added distilled water to the graduated cylinder up to the 1000 mL mark, placed a stop top on the graduated cylinder, and shook it to suspend any particles. After 20 seconds, we pipetted 20 mL from 20 cm below the surface. We extracted the fluid into a clean pre-weighed 100 mL beaker and rinsed the pipette with distilled water. We dried the samples in a drying oven, placed them in a desiccator to cool, and weighed them to get the mass of the silt/clay fraction. We calculated the percent silt/clay as follows:

# mass of silt/clay\_x 100 20g

We rinsed the remaining sand fraction from the sieve into a clean pre-weighed 100 mL beaker for further processing.

*CaCO*<sub>3</sub> *Determination* 

We dried and weighed the sand fraction from the above analysis and then acidified (leached) it with 10% hydrochloric acid (HCl). After the sample stopped bubbling, we rinsed it over a 63  $\mu$ m sieve and placed it in a clean pre-weighed crucible. We dried the sample in a drying oven and placed it in the desiccator to cool. We weighed the sample and then combusted it at 700°C in a muffle furnace for 2 hours. We placed the sample back in the desiccator to cool and then weighed it again. We calculated CaCO<sub>3</sub> content as follows:

# (mass of sample before acidification – mass of sample after acidification) x 100 total mass

#### Sand Particle Phi Size

We poured the sample from the above analysis through a stack of sieves (2 mm, 1.4 mm, 1 mm, 710  $\mu$ m, 500  $\mu$ m, 355  $\mu$ m, 250  $\mu$ m, 180  $\mu$ m, 125  $\mu$ m, 90  $\mu$ m, and 63  $\mu$ m). We placed the stack of sieves on a shaker for 15 minutes. We brushed the contents of each sieve into a square plastic container and brushed them into a weigh boat and weighed the contents. We calculated mean phi size based on methods described in Folk (1980) as follows:

$$\sum_{i=-1.0}^{4.0} (i * \text{mass}_i)$$

sand total mass

*i*=phi size

# Organic Matter

We took a 3g sub-sample from each core sample after it was processed for moisture content and placed it in a labeled, pre-weighed crucible. We weighed the sample and combusted it in a muffle furnace at 550°C for 2 hours. We cooled the sample in a desiccator and weighed it. We calculated organic matter as follows:

# <u>3g – mass of combusted sample</u> x 100 3g

#### Vegetation

We determined plant stem densites by placing a 52 cm diameter and a 100 cm diameter plastic hoop around each nest and counting the number of stems within each hoop. We identified vegetation to the lowest practical taxonomic level. We also noted presence or absence of vegetation within 1 meter of the nest. The dominant vegetation was comprised of Sea Oats (*Uniola paniculata*) and Sea Rocket (*Cakile harperi*).

#### Elevation

We determined the exact location and elevation of each *in situ* and relocated nest monitored in this study using a Trimble R-8 GNSS (Global Navigation Satellite System) placed on the sand surface next to the cage. This unit has a horizontal accuracy of  $\pm 5 \text{ mm} + 0.5 \text{ ppm}$  (parts-per-million) Remote Monitoring System (RMS) and a vertical accuracy of  $\pm 5 \text{ mm} + 1 \text{ ppm}$  RMS.

## Tidal Inundation

We recorded the elevation of tidally inundated nests on the day of the first overwash event, which occurred on August 19, 2008. This was an opportunistic component of the study and some of these nests were not necessarily the same nests monitored with data loggers. Tidal inundation was defined as nests that were washed over during the previous high tide. We also recorded inundations on a daily basis in order to compare hatch and emergence success rates with nests not subjected to inundation. It should be noted that only the frequency of inundation was recorded; the duration and the degree of inundation were not recorded.

Nest Success

Three days after the first emergence of hatchlings, we inventoried all nests with data loggers in order to determine the hatch success and emergence success. We excavated all nest contents and separated eggs or hatchlings into the following categories: empty shells (50% or more intact), unhatched eggs, pipped eggs (eggs with openings that contained a dead hatchling), dead hatchlings, and live hatchlings. We calculated hatch success and emergence success for each nest in accordance with the methods in Miller (1999). We defined hatch success as the number of hatchlings that hatch out of their shell and calculated it as follows:

# total # hatched x 100 total eggs

We defined emergence success as the total number of hatchlings out of the total clutch that emerged from the nest and calculated it as follows:

# total # hatched – total # live and dead hatchlings x 100 total eggs

We defined and calculated an additional term, hatched emergence success, as the number of hatchlings out of the hatched eggs that emerged from the nest and calculated it as follows:

# total # hatched – total # live and dead hatchlings x 100 total hatched eggs

The purpose of this additional calculation was to detect if nest relocation reduced emergence ability. We defined incubation duration or incubation-to-emergence period as the number of days beginning with the date of the morning after the nest deposition through the date of the first emergence (Miller *et al.* 2003).

#### Data Analysis

We downloaded temperature data into LogTag Analyzer and exported the data into Microsoft Excel. To calculate the incubation temperature, we averaged temperatures during the middle third of the incubation, which is the sex-determining period (Yntema and Mrosovsky 1980, Vogt and Bull 1982, Mrosovsky and Pieau 1991). We only used middle third temperatures in our analysis. We used nest inventory data to calculate incubation duration, hatch success, emergence success, and hatched emergence success. All data were exported into Minitab 15 for analysis and were transformed to meet the assumptions of parametric statistics. We used an analysis of covariance (ANCOVA), analysis of variance (ANOVA), and two sample T-tests to determine differences between *in situ* and relocated nests ( $\alpha = 0.05$ ). For our ANCOVA analysis, we included nest type, temperature, incubation duration, and nest success as responses and sediment characteristics, presence of vegetation, and elevation as covariates to determine the difference between *in situ* and relocated nest incubation environments. We used ANOVA to examine the effect of tidal inundation and elevation on hatch and emergence success. Since hatch and emergence success data were not normally distributed even following transformation, we used a Kruskal-Wallis test to determine differences between in situ and relocated nest hatch and emergence success. We used a Principal Components Analysis (PCA) to determine the influence of environmental characteristics on hatch and emergence success in *in situ* and relocated nests.

# Results

#### *Temperature*

Incubation temperature during the middle third of incubation, which is the sexdetermining period, was significantly different between time periods throughout the

season ( $F_{3,84} = 16.86$ , p < 0.000) but not between *in situ* and relocated nests ( $F_{1,86} = 0.70$ , p = 0.405). All mean temperatures were above the pivotal temperature although the last *in situ* time period was close to pivotal (Figure 3).

#### Incubation Duration

Incubation duration was significantly longer for nests laid in May ( $F_{1,89} = 0.96$ , p < 0.000), but was not significantly different between *in situ* and relocated nests ( $F_{3,87} = 20.57$ , p = 0.138) (Figure 4).

# Physical Environmental Characteristics

The first principle component (PC1) derived from all environmental characteristics accounted for 24.2% of the variability and the second principle component (PC2) accounted for 17.6% of the variability (cumulatively 41.8%) (Table 1). PC1 was mostly driven by elevation, moisture content, and the presence of vegetation and PC2 was mostly driven by phi size, organic matter, calcium carbonate, and the presence of vegetation (Table 1). All environmental characteristics were positive (increased from left to right) in PC1 except for organic matter and moisture content. All environmental characteristics were positive (increased from bottom to top) in PC2 except for CaCO<sub>3</sub> and presence of vegetation (Figure 5). Hatch and emergence success as well as incubation duration were not significantly correlated with environmental characteristics (Table 2). Nest depth was not significantly different between *in situ* nests prior to relocation and relocated nests (2 Sample T test, T = -0.39, p = 0.697). The incubation environment was not significantly different between *in situ* and relocated nests (Table 3).

# Hatch and Emergence Success

No significant differences were detected between *in situ* and relocated nest hatch success (n = 136, Z =  $\pm 0.49$ , p = 0.620) and emergence success (n = 136, Z =  $\pm 0.78$ , p

= 0.435; Figure 6). Hatched emergence success, however, was significantly higher in relocated nests (n = 136, Z =  $\pm 2.14$ , p = 0.032; Figure 6).

#### Tidal Inundation

Tidal inundation occurred at least once in 64.6% (62 of 96) of *in situ* nests and 15.2% (7 of 46) of relocated nests. Elevation was significantly lower in inundated *in situ* nests ( $F_{1,139} = 4.50$ , p = 0.036) and was significantly lower in nests with multiple inundations as compared to non-inundated nests ( $F_{5,135} = 4.43$ , p = 0.001; Figure 7). Hatch and emergence success were significantly lower in inundated nests ( $F_{1,131} = 117.73$ , p < 0.000,  $F_{1,131} = 47.71$ , p < 0.000; Figure 8). Both hatch and emergence success dropped to less than 50% after one inundation event (Figure 8). Forty percent of inundated nests produced zero hatchlings while less than five percent of non-inundated *in situ* and relocated nests produced zero hatchlings.

# Discussion

Nest relocation is an effective conservation method when sites are chosen carefully (Wyneken *et al.* 1988). Our results suggest that nest relocation on Cape Island did not significantly alter the incubation temperatures during the sex-determining period of incubation relative to similarly positioned *in situ* nests. Other studies have found that relocated nests can incubate at different temperatures than *in situ* nests (Hoekert *et al.* 1998, Başkale and Kaska 2005, Mrosovosky and Yntema 1980, Tuttle 2007, and Pintus *et al.* 2009) causing skewed sex ratios (Morreale *et al.* 1982, Godfrey *et al.* 1997). Hoekert *et al.* (1998) and Başkale and Kaska (2005) both reported that relocated nests incubated at warmer temperatures than *in situ* nests. Mrosovosky and Yntema (1980) found the incubation of eggs in styrofoam boxes above ground produced mostly males. However, García *et al.* (2000), Tuttle (2007) and Pintus *et al.* (2009) found no difference between relocated and *in situ* nest temperatures.

Incubation duration was not significantly different between *in situ* and relocated nests, but was significantly different between time periods. Both *in situ* and relocated nest incubation durations for nests oviposited in May 2008, were significantly longer possibly due to cooler air temperatures coinciding with the beginning of the incubation period. Incubation duration was not significantly different between *in situ* and relocated nests, which is consistent with incubation temperature not being significantly different between the two. Analysis of preliminary South Carolina Department of Natural Resource (SCDNR) data for the 21 nest protection projects from 2000 through 2006 show similar results. Mean incubation durations for all documented *in situ* and relocated nests in South Carolina between 2000 and 2006 were 57.0 and 56.3 days, respectively (SCDNR unpublished data). Egg chamber dimensions were not measured and duplicated for each individual relocated nest; however, nest depths of original egg chambers and new egg chambers were not significantly different indicating that this was unlikely to have a significant effect on incubation temperature or incubation duration.

The incubation environment was not significantly different between *in situ* and relocated nests. This was likely because relocated nests were purposely placed in areas where *in situ* nests, deemed to be safe from inundation, were incubating. Caldwell (1959) reported a range of 1.3% to 4.2% moisture content for Cape Island in 1939. In our study, moisture content ranged from 2.9% to 3.2% between *in situ* and relocated nest samples collected twice during the season. However, we only collected moisture content samples once for each nest on two separate days during the season, so moisture content is not accurately represented in this study. However, since *in situ* nests were more likely to be inundated than relocated nests, moisture content would likely be higher in *in situ* nests because they were typically closer to the water.

In 2008, nest relocation on Cape Island did not decrease overall hatch or emergence success relative to *in situ* nests. McElroy (2009) found no difference between *in situ* and relocated nest hatch and emergence success in a study on Sapelo Island, Georgia, which is consistent with our results. Analysis of preliminary SCDNR data for the 21 nest protection projects in South Carolina from 2000 through 2006 also shows similar results. Median hatch success for *in situ* nests and relocated nests were 82.1% and 83.7%, respectively, and median emergence success for *in situ* nests and relocated nests was 77.9% and 79.7%, respectively (SCDNR unpublished data). Hatched emergence success, however, was significantly higher in relocated nests than *in situ* nests indicating that nest relocation on Cape Island does not decrease the emergence ability of hatchlings. We do not know why *in situ* nests had a lower hatched emergence success, but it is possible that roots in the egg chamber from vegetation around *in situ* nests and compaction and sand deposition from overwash events were influencing factors. Relocated nests are typically located away from vegetation and areas subject to inundation.

Tidal inundation mostly occurred in *in situ* nests (64.6%) although it also occurred in relocated nests (15.2%). Data for the degree and duration of tidal inundation were not collected, but both were presumably greater in nests located closer to the water. Elevation was significantly lower in inundated *in situ* nests, which explains why they were experiencing multiple inundations throughout the incubation period. Tidal inundation from spring tides began in late August near the end of the nesting season and near the end of the incubation period (median = 50 days, n = 44) for nests laid earlier in the season. The timing of inundation likely resulted in late developmental mortality. Hatch and emergence success were significantly lower in inundated nests, and both measures dropped to less than 50% after one inundation event. This

suggests that the duration of these events were long enough to significantly limit gas exchange (Ackerman 1980) and cause asphyxiation of embryos (Foley et al. 2006). Salinity may also have played a role in the reduction of hatch and emergence success in inundated nests. Foley (1998) reported a negative correlation between both moisture content and salinity relative to nest success. In a previous study conducted on Cape Island between 1991 and 2004, once every two weeks, all nests regardless of location were left *in situ* in order to determine the fate of the nest. Forty-eight percent (272/571) of these nests were inundated. Inundated nests had a mean hatch success of 0% and non-inundated nests had a mean hatch success of 66.72% (USFWS unpublished data). Foley et al. (2006) documented 31% egg mortality in their study in south Florida despite frequent inundation. We found 24% total egg mortality in our study, with inundated nests having 38% egg mortality and non-inundated nests having 19% egg mortality. We also found 100% egg mortality in 40% of inundated nests. Aside from storm events or spring high tides that cause occasional inundation, nests on Cape Island are typically inundated due to low elevations and severe beach erosion. Foley et al. (2006) also suggested that nests laid lower on the beach in south Florida may produce males, the rarer sex, and should be left *in situ* since these nests produce some hatchlings. We did not measure incubation temperatures in in situ locations below the spring high tide line on Cape Island, so we cannot estimate the predicted sex ratios of nests located below the spring high tide line. However, even if the ratio of males is greater in nests located lower on the beach, the hatch and emergence success would be greatly reduced based on the study conducted on the island between 1991 and 2004.

The location of a nest affects the fitness of both parents through the survival of their offspring (Wood and Bjorndal 2000). By relocating nests that are not likely to

produce hatchlings because of lethal tidal inundation, we are possibly increasing the fitness of both parents. It has been proposed that relocating nests selects for "poor nesters" and artificially increases their fitness (Mrosovosky 2006). However, no evidence currently exists to indicate that nest site selection is a heritable trait (Pike 2008). In fact, a study by Pfaller *et al.* (2008) found that loggerheads scatter their reproductive effort across multiple breeding seasons and no one individual out of 295 individually-identified turtles in that study had more than one season of unsuccessful nest sites. They also found that a large number of unsuccessful nest sites relates to unsuccessful nest site selection by a large percentage of the population, not from a small number of individuals. Therefore, their study suggests that nest relocation does not distort the gene pool, but does enable conservation.

#### **Conservation Implications**

Our study suggests that nest relocation continues to be a sound conservation practice when carried out properly. A large component of successful relocation is careful site selection as well as consideration of environmental factors for a particular nesting beach. Since CRNWR is the largest rookery for the northern nesting assemblage of Northwest Atlantic loggerheads, conservation efforts carried out here will go further towards the recovery of this assemblage and the Northwest Atlantic population. If this assemblage is lost, it will not be replaced by recruits from other nesting assemblages due to high nesting site fidelity (Schroeder *et al.* 2003).

Also, global climate change is an increasing threat to sea turtles. It has the potential to exacerbate existing threats, such as decreasing suitable nesting habitat and increasing inundation risk (Fish *et al.* 2005, Mazaris *et al.* 2009), as well as introduce new ones. Studies have documented earlier nesting seasons and warmer nest incubation temperatures (Hawkes *et al.* 2007, Glen and Mrosovsky 2004, Hays *et al.* 

2003, Mazaris *et al.* 2008, Pike *et al.* 2006, Weishampel *et al.* 2004). Although sex was not confirmed, it is likely that the majority of hatchlings produced in 2008 were female biased because the mean incubation temperatures during the sex-determining period were above the pivotal temperature (Mrosovsky 1994, Davenport 1997, Wibbels 2005).

#### **Summary and Conclusions**

Individually relocated nests incubated at similar temperatures and had similar incubation durations as *in situ* nests since relocated nests were purposely placed in areas where *in situ* nests were incubating. Inundation was significantly higher in *in situ* nests since *in situ* nests are more likely to become inundated because relocated nests are typically located away from areas subject to inundation. Hatch and emergence success were similar between relocated and *in situ* nests, but both measures were significantly lower in inundated nests. This research suggests that nest relocation, when used correctly, remains an important management tool for sea turtle conservation and the need for it may increase with rising sea levels.

Loggerheads as well as other sea turtles continue to face threats throughout their entire life cycle. In order to conserve sea turtles, global conservation efforts need to continue for all life stages and nest relocation will continue to enable conservation on beaches subject to severe erosion.

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Figure 1. Location of Cape Island.



Figure 2. Data logger placement in *Caretta caretta* nests.



**Figure 3.** Mean *in situ* and relocated nest temperatures recorded during the middle third, which is the sex-determining period, of incubation during four different time periods throughout the 2008 nesting season. Data are means ( $\pm$ SE). \*Sex predictions based on Wibbels 2005.



**Figure 4.** Mean *in situ* and relocated nest incubation durations during four different time periods throughout the 2008 nesting season. Data are means ( $\pm$ SE).



**Figure 5.** Plot of environmental characteristics on the first two principal components derived from PCA.

☑ Hatch Success ☑ Emergence Success ☑ Hatched Emergence Success



**Figure 6.** Comparison of hatch success, emergence success, and hatched emergence success percentages between *in situ* and relocated nests. Data are means (±SE).



Figure 7. Differences in elevation between inundated *in situ* and relocated nests. Data are means  $(\pm SE)$ .



**Figure 8.** Mean hatch and emergence success and elevation plotted against tidal inundation. Data are means±standard errors.

Variable	PC1	PC2
Organic Matter	-0.168	0.461
Calcium Carbonate	0.020	-0.459
Silt/Clay	0.307	0.032
<b>Vegetation Present</b>	0.457	-0.404
Phi Size	0.387	0.531
Elevation	0.551	0.286
Moisture Content	-0.464	0.219

**Table 1.** Coefficients of environmental variables for the first two principal components.

Parameter	PC1	PC2
Hatch Success	-0.148	0.130
Emergence Success	-0.131	0.134
Incubation Duration	0.008	-0.156
p>0.10 for all coefficients		

**Table 2.** Correlations between PCA components and hatch and emergence success and incubation duration.

Characteristic	In Situ	Relocated	p-value
Moisture Content (%)	3.24±0.0015	0.0286±0.0011	0.626
Silt/Clay (%)	$0.12 \pm 0.0010$	$0.0004 \pm 0.0002$	0.464
$CaCO_3$ (%)	$3.03 \pm 0.0086$	$0.0650 \pm 0.0201$	0.272
Organic Matter (%)	$0.19 \pm 0.0003$	0.0189±0.0096	0.569
Sand Phi Size ( $\phi$ )	$1.3 \pm 0.0556$	1.2958±0.0479	0.925
Elevation (m)	$2.5 \pm 0.0689$	2.3573±0.0654	0.126

**Table 3.** Environmental characteristic comparison between relocated and *in situ* nests. Data are means ( $\pm$ SE).

#### **CHAPTER 4: SYNTHESIS**

Nest relocation is an effective conservation method when sites are chosen carefully (Wyneken et al. 1988). This study clearly indicates that nest relocation continues to be a sound conservation practice when carried out properly. However, it also illustrates the importance of hatchery location and the careful consideration of environmental characteristics prior to site selection. Individually relocated nests in 2008 mimicked in situ nest incubation environments since they were relocated next to similarly positioned in situ nests. Hatcheries in 2007 and 2008, however, did alter the nest incubation environment and possibly altered sex ratios compared to in situ nests because incubation temperatures were cooler. Cooler temperatures also extended the incubation duration. The differences between hatchery and *in situ* nests in 2007 and in 2008 were 1.6 days and 6.3 days, respectively. Davenport (1997) reported that cooler incubation temperatures slowed development. However, we did not determine how cooler incubation temperatures influenced the development of embryos in our study. Hatcheries will likely need to continue to be used on Cape Island because suitable habitat for relocation is limited and hatcheries can hold more nests than individually caged nests in the same amount of space.

Besides skewed sex ratios, reduction in hatch success, and delayed hatchling release (Mrosovosky and Yntema 1980, Morreale *et al.* 1982, Godfrey *et al.* 1997, Hoekert *et al.* 1998, Mortimer 1999, Moody 2000, Başkale and Kaska 2005, and Pintus *et al.* 2009), other concerns about hatchery use include the increase in predator concentration on land and in the water due to the increase of hatchling density. In the water, Stewart and Wyneken (2004) reported a higher hatchling survival rate (95%)

on a natural nesting beach on the Atlantic coast of Florida than Wyneken and Salmon (1997) reported for a hatchery site (72%) on the Atlantic coast of Florida. Depredation rates ranged from 4.6% to 34% in seven in-water hatchling predation studies with the greatest depredation rate occurring at a hatchery site (Whelan and Wyneken 2007). Relative to land predators, Ratnaswamy and Warren (1998) and Stancyk et al. (1980) suggested that intensive removal of raccoons from barrier beaches may increase ghost crab abundance. It is possible that ghost crabs have increased as a result of the raccoon trapping program. However, studies suggest that an increase in the ghost crab population does not necessarily increase predation rates. Caut et al. (2006) reported a 48.9% ghost crab predation rate of nests and a mean predation rate of 4.1 eggs per nest on Awala Yalimapo beach in French Guiana. Barton and Roth (2008) found no correlation between ghost crab density and the proportion of eggs consumed by ghost crabs. However, predation rates for hatchlings on their way to the water were not addressed in either study. In 2007, all hatcheries were landward of the primary dune and in 2008, one hatchery was landward of the primary dune and the other two were seaward of the primary dune. Placement of nests farther inland increases the likelihood of hatchling desiccation, misorientation, and predation (Wood and Bjorndal, 2000, Mrosovosky 2006). Further research is needed to determine if there is a significant difference between *in situ* and hatchery nest land-based depredation since hatchlings from hatcheries on Cape Island may have to traverse longer distances to the ocean depending on the hatchery location. Although suitable habitat is limited and hatcheries can hold more nests than individually caged nests in the same amount of space, hatcheries should be used as a last resort and should be placed seaward of the primary dune whenever possible in order to reduce predation risk (Figure 1).

CRNWR continues to experience severe erosion (USFWS 2007, 2008). Increases in sea-level resulting in higher tides as well as the increased potential for stronger storms (Karl *et al.* 2009) will compound existing conditions increasing the need for nest relocation. Since CRNWR is the largest rookery for the northern nesting assemblage of Northwest Atlantic loggerheads, conservation efforts carried out here will go further towards the recovery of this assemblage and the population. If this assemblage is lost, it will not be replaced by recruits from other nesting assemblages due to high nesting site fidelity (Schroeder *et al.* 2003).

Loggerheads as well as other sea turtles continue to face threats throughout their entire life cycle and global climate change is an emerging threat. It has the potential to exacerbate exisiting threats such as decreased suitable nesting habitat and increased inundation risk (Fish *et al.* 2005, Mazaris *et al.* 2009) as well as introduce new threats. In order to conserve sea turtles, global conservation efforts need to continue for all life stages and nest relocation should on beaches subject to severe erosion in order to enable conservation.

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Figure 1. Nest relocation decision tree.