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Characteristics of a Green Turtle (*Chelonia mydas*) Assemblage in Northwestern Florida Determined During a Hypothermic Stunning Event

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A hypothermic stunning event (i.e., cold-stunning event) during late Dec. 2000 and early Jan. 2001 involving an unprecedented number of sea turtles provided an opportunity to characterize the green turtle (*Chelonia mydas*) assemblage in St. Joseph Bay (Gulf County, Florida). In addition to 388 green turtles, the 401 cold-stunned turtles comprised 10 Kemp's ridleys (*Lepidochelys kempii*) and three loggerheads (*Caretta caretta*). Most (337/401) of the turtles survived and were eventually released. To place this event in perspective, we categorize sea turtle cold-stunning events in the eastern United States as either acute or chronic. Acute cold-stunning events, like the one in St. Joseph Bay, occur only during unusually cold winters in shallow-water areas (< 2 m), where sea turtles are year-round residents. These are short-lived (< 2 wk) events with low mortality rates (< 30%) that affect principally green turtles. Chronic cold-stunning events occur every winter in areas where sea turtles are seasonal residents. These are long-lived (1–3 mo) events with high mortality rates (> 60%) that affect primarily Kemp's ridleys. All of the green turtles from St. Joseph Bay were neritic-phase juveniles, and the mean straight-line carapace length of this group was 36.6 cm (range = 25.0–75.3 cm, SD = 8.9). This assemblage of juvenile green turtles is the first documented along the northern Gulf of Mexico. Sequencing of mtDNA (mitochondrial DNA) from tissue samples taken from 255 of the green turtles revealed that about 81% were from the nesting populations in the United States (Florida) and Mexico (Yucatan). This assemblage is unusual in the United States because it does not have a substantial representation from the nesting population in Costa Rica (Tortuguero), the Atlantic's largest green turtle nesting population. Based on necropsies of 51 of the green turtles, the sex ratio of this assemblage was female-biased (3.25 females:1 male), which may be a result of warm incubation temperatures on the nesting beaches in Florida. The majority of the material found in the gastrointestinal tracts of the green turtles that died was turtle grass (*Thalassia testudinum*). This was the first time turtle grass has been identified as the primary diet of juvenile green turtles anywhere in the continental United States. Green turtles in St. Joseph Bay appear to have few direct threats, but the seagrass upon which these turtles primarily forage has suffered extensive damage from boat propellers.

Green turtles (*Chelonia mydas*) inhabit tropical and warm-temperate oceans worldwide. After completing an initial oceanic life stage (Carr et al., 1978; Carr and Meylan, 1980), green turtles become residents of neritic zones, where they feed on seagrasses or macroalgae (Bjorndal, 1980; Hirth, 1997). Green turtle assemblages consisting of only juvenile animals are found in many nearshore areas (Carr and Caldwell, 1956; Mendonça and Ehrhart, 1982; Coyne, 1994; Ehrhart and Redfoot, 1995; Shaver, 2000). As these turtles approach maturity, they leave these developmental habitats and move to adult resident habitats (Carr et al., 1978; Musick and Limpus, 1997).

In 1978, the breeding populations of green turtles in Florida and along the Pacific coast of Mexico were listed as endangered under the U.S. Endangered Species Act. Green turtles from other breeding populations were listed as threatened. Some of the primary tasks of the associated recovery plan are to determine the distribution, abundance, and population characteristics of green turtles and to identify threats to their survival throughout their range (National Marine Fisheries Service and U. S. Fish and Wildlife Service, 1991). In the continental United States, green turtles are found primarily from Massachusetts through Texas. Assemblages of juvenile green turtles within this range have been studied in North Carolina (Epperly et al., 1995), along the central and southern Atlantic coast of Florida (Mendonça and Ehrhart, 1982; Wershoven and Wershoven, 1992a; Ehrhart and Redfoot, 1995; Bresette et al., 1998; Bresette et al., 2000), in the Florida Keys (Schroeder et al., 1998), along the central and southern Gulf coast of Florida (Carr and Caldwell, 1956; Schmid, 1998; Witzell and

Schmid, 2004), and along the southern coast of Texas (Coyne, 1994; Shaver, 2000). An assemblage of adult green turtles likely occurs southwest of the Florida Keys (Schroeder et al., 1996).

Data on the distribution of green turtles are also collected opportunistically when dead or debilitated animals wash ashore (i.e., strand). However, sea turtle strandings may provide only relatively small, infrequent samples of animals. Furthermore, because carcasses and moribund turtles can drift long distances before washing ashore, it may be difficult to determine from what specific area they originated. An exception to this generalization occurs when a hypothermic stunning event causes a large number of sea turtles from a specific area to strand during a short period of time.

Hypothermia in cheloniid sea turtles disrupts metabolic pathways, leading to imbalances in blood chemistry (Lutz et al., 1989; Carminati et al., 1994.). Turtles become lethargic and float at the surface when water temperatures drop below 10°C (Schwartz, 1978). Because they are disabled, these cold-stunned sea turtles can be easily retrieved from the water or after they wash ashore (Witherington and Ehrhart, 1989; Morreale et al., 1992). The environmental conditions necessary to cause the cold-stunning of large numbers of sea turtles restrict these events to relatively small, well-defined areas (Witherington and Ehrhart, 1989; Shaver, 1990; Morreale et al., 1992). Therefore, the areas the turtles inhabited just before stranding can be easily determined.

In the United States, cold-stunning events involving large numbers of sea turtles have been documented in Long Island Sound, NY (Morreale et al., 1992; Still et al., 2002); in the Indian River Lagoon system along the central-east coast of Florida (Witherington and Ehrhart, 1989); and in Laguna Madre in southern Texas (Shaver, 1990). Green turtles, Kemp's ridleys (*Lepidochelys kempii*), and loggerheads (*Caretta caretta*) have all been known to become debilitated by cold-stunning (Witherington and Ehrhart, 1989; Morreale et al., 1992; Shaver, 1990).

Here we report on a record sea turtle cold-stunning event that involved principally green turtles. This event occurred in northwest Florida (St. Joseph Bay, Gulf County) and provided the opportunity to determine the characteristics of and the threats to this assemblage.

Materials and Methods

Cold-stunned sea turtles were found in the southwestern portion of St. Joseph Bay (Fig. 1) beginning on 31 Dec. 2000. The turtles were initially found and collected by the Gulf Coast Turtle Patrol (GCTP), a nonprofit conservation organization permitted by the Florida Fish and Wildlife Conservation Commission (FWC) to conduct sea turtle stranding and salvage activities in Florida. The GCTP was joined in the effort to search for and collect cold-stunned turtles by staff from the Apalachicola National Estuarine Research Reserve and the FWC. Most of the workers searched by foot along the shoreline of the bay, but one small boat was also used to locate torpid turtles that had not yet drifted ashore. Cold-stunned sea turtles were collected from St. Joseph Bay through 12 Jan. 2001 and were transported each day by vehicle to Gulf World, a permitted sea turtle rehabilitation facility in Panama City Beach, FL (about a 90-km drive). Most of the turtles became active during transport and were placed into a large, heated, outdoor tank at Gulf World. The turtles that did not quickly resume normal activity or those that were in need of continuing medical attention were put into smaller, heated, indoor tanks.

On 7 Jan. 2001, we began the process of measuring, examining, tagging, and taking blood or skin samples from the turtles that were being held at Gulf World. A graduated caliper was used to take Straight measurements of Carapace Length from the nuchal notch to the posterior marginal tip (SCL). During this time, the turtles were also examined for any external anomalies such as evidence of trauma or disease. Turtles with parallel gashes across the head, carapace, or plastron were thought to have been struck by a propeller. Shark bites were indicated by crescent-shaped wounds that were usually most distinctive on the carapace or plastron. Numerous cuts forming a crescent-shaped "dotted line" also indicated a shark bite. Evidence of disease included the presence of one or more tumors or a noticeable degree of emaciation. A turtle was determined to have fibropapilloma-like tumors when at least one verrucose tumor was present (see Herbst, 1994, for an overview of this disease). Emaciation was determined by appraising the overall body condition of the turtle. Turtles with a distinctly concave plastron and a prominent supraoccipital were determined to be emaciated. Each turtle was also checked for external flipper tags and scanned for the presence of internal passive integrated transponders (PIT tags) by using Pocket Readers from Destron Technologies.

The turtles were tagged following standard methodologies and considerations (Balazs, 1999). Inconel metal tags (size 681, National Band and Tag Company) were applied to the

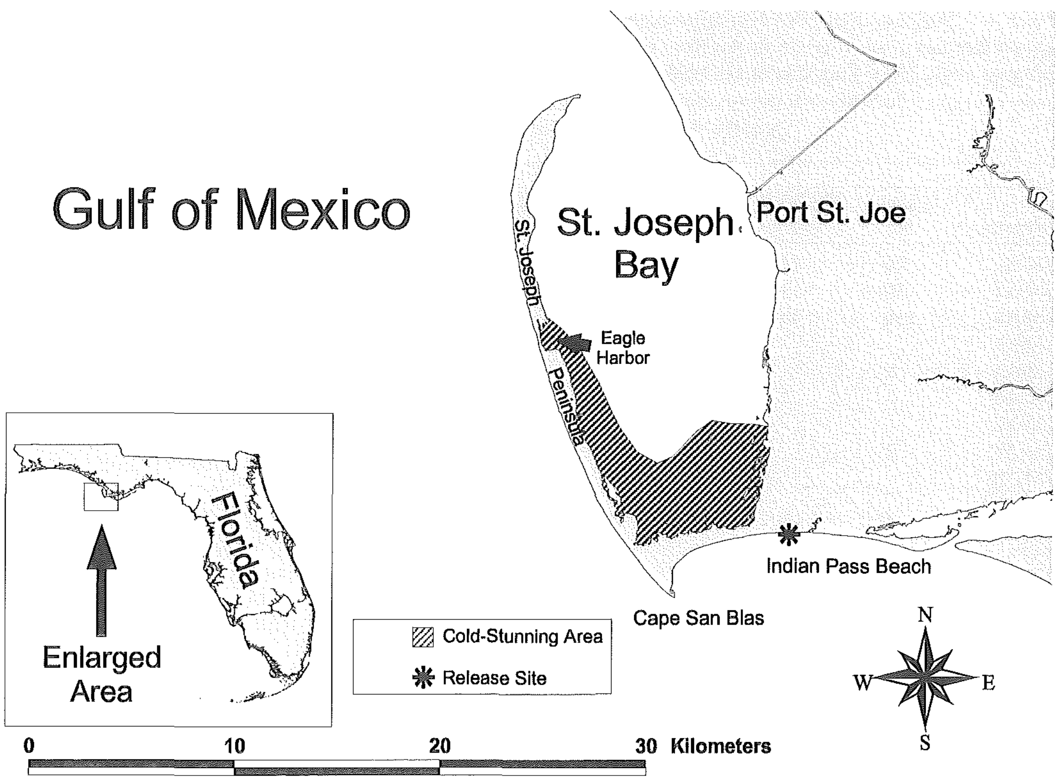

Fig. 1. Area in St. Joseph Bay where 401 cold-stunned sea turtles (388 green turtles, 10 Kemp's ridleys, and three loggerheads) were found washed ashore or floating and the site where most of the same turtles were eventually released after recovering at a nearby rehabilitation facility.

trailing edge of both front flippers of each turtle (typically in the first proximal scute). The PIT tags were placed along the dorsal surface of one of the front flippers in the area between the radius and ulna and the trailing edge.

To determine the genetic identity of the live turtles, either a 3-ml blood sample was taken from the dorsal cervical sinus with a vacutainer system using a 22-gauge needle or a semicircular skin sample was taken from the trailing edge of one rear flipper using a 6-mm-diameter punch. The samples were obtained and preserved as described by Dutton (1996).

Gross necropsies were conducted on turtles that were found dead and on those that later died at a rehabilitation facility. During necropsy, the overall body condition was assessed by noting the robustness of the pectoralis muscles and the extent and color of fat overlying the peritoneum. Atrophied pectoralis muscles and darkened fat (from condensation of melanocytes due to mobilization of fat stores) indicated poor nutritional condition. The gonads of each turtle were also examined and the gender determined. The ovary was identified by the presence of follicles or, when follicles were not clearly visible, by its grainy, heterogeneous surface and by its attachment to the peritoneal wall along only one lateral edge. The testis was identified by its smooth, homogenous surface and by its attachment to the peritoneal wall by its entire dorsal surface. The gastrointestinal (GI) tract was removed from each necropsied turtle and frozen for later examination. A 1-cm^3 pectoralis muscle sample to determine genetic identity was collected from each dead turtle and placed into a 3-ml tube with a 20% DMSO (dimethyl sulfoxide) solution saturated with sodium chloride (Dutton, 1996).

The frozen GI tracts were later thawed at room temperature and cut along their full length to examine gut contents. All material was removed and placed into separate dissecting dishes, which indicated where in the GI tract the material was found. Material from the esophagus, stomach, and each 50-cm section of the intestine was kept separate. The material in each dissecting dish was processed thoroughly with probes and forceps under a dissecting microscope (or a compound microscope when necessary). The majority of material was turtle grass (*Thalassia testudinum*), so care was taken to look for any fragments of any other seagrasses or any macro-

TABLE 1. Mitochondrial DNA haplotype composition and frequency of green turtles found cold-stunned in St. Joseph Bay and those of green turtles from rookeries around the Atlantic and adjacent waters (Allard et al., 1994; Lahanas et al., 1994; Encalada et al., 1996; and Lahanas et al., 1998). The asterisk denotes haplotypes for which rookery-origin information is not yet available. Haplotype designations follow those given by Bjorndal and Bolten (unpubl. data) at http://accstr.ufl.edu/cmmtdna.html.

		Green turtle nesting stocks							
Haplotype	St. Joseph Bay	United States (Florida)	Mexico (Yucatan)	Costa Rica (Tortuguero)	Aves/ Suriname	Brazil	Ascension Island	Guinea Bissau	Cyprus
CM-A1	70	11	7						
CM-A2	10	1							
CM-A3	116	12	5	395	3				
CM-A4				1					
CM-A5	9		1	32	40				
CM-A6					1				
CM-A7					1				
CM-A8	1					8	16	19	
CM-A9						5	1		
CM-A10							3		
CM-A11						1			
CM-A12						2			
CM-A13									8
CM-A14									1
CM-A15			1						
CM-A16	16		1						
CM-A17	3		2						
CM-A18	9		3						
CM-A20				2					
CM-A21				3					
CM-A22*	3								
CM-A26*	8								
CM-A27*	5								
CM-A28*	3								
CM-A29*	2								
TOTAL	255	24	20	433	45	16	20	19	9

algae. All plant material was separated by species, dried at 55°C for 4 d, and then weighed.

DNA was isolated from packed red and white blood cells, skin samples, or muscle samples by using either standard phenol/chloroform extraction techniques (Sambrook et al., 1989) or the Fast Prep DNA isolation kit (Bio101®). Amplification of mtDNA was performed with polymerase chain reaction (PCR) methodology (Innis et al., 1990), using the primers HDCM2 and LTCM2 designed to target 481 bp (base pair) at the 5′ end of the control region of the mitochondrial genome (Encalada et al., 1996). Cycle sequencing reactions with fluorescently labeled dideoxynucleotides were performed, and sequencing products from both light and heavy strands were analyzed with an Applied Biosystems model 3100 genetic analyzer. Sequences were aligned against reference data from the ca. 480-bp segment of the mtDNA control region corresponding to the region reported by Encalada et al. (1996). Haplotype nomenclature follows that reported by Lahanas et al. (1994, 1998), Encalada et al. (1996), and Bjorndal et al. (2005) as summarized on the Marine Turtle DNA Sequence website maintained by the Archie Carr Center for Sea Turtle Research at the University of Florida (http://accstr.ufl.edu/cmmtdna.html).

Baysesian mixed-stock analysis was performed with haplotype frequency data using the program BAYES (Pella and Masuda, 2001). Estimates of contributions by different nesting stocks to the St. Joseph Bay aggregation were based on Bayesian analysis using Markov Chain Monte Carlo estimation from 10,000 resamplings of four stock mixtures composed of green turtles from seven major nesting stocks. Published haplotype frequency data were used as potential source stock baselines (see Table 1). The Mediterranean stock was not included in the analyses, because no Mediterranean haplotypes were detected among the cold-stunned green turtles from St. Joseph Bay.

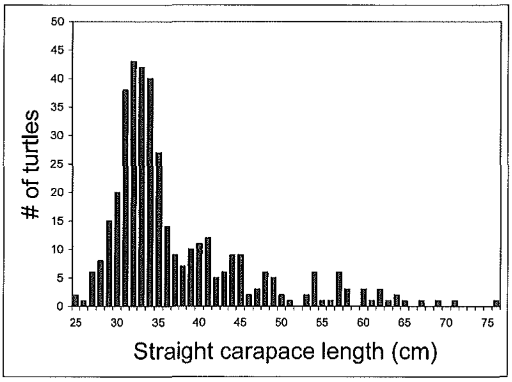

Fig. 2. Size–frequency distribution of 387 green turtles found cold-stunned in St. Joseph Bay (Gulf County, FL) during late Dec. 2000 and early Jan. 2001. The mean was 36.6 cm (SD = 8.9). The carapace length of one of the green turtles could not be determined because of carapace damage.

Results

The 401 cold-stunned turtles comprised 388 green turtles, 10 Kemp's ridleys, and three loggerheads. Overall, 341 turtles were found alive (333 green turtles, six Kemp's ridleys, and two loggerheads), and 60 turtles were found dead (55 green turtles, four Kemp's ridleys, and one loggerhead). All of the cold-stunned turtles were found either washed ashore or floating in the southern and southwestern portions of St. Joseph Bay (Fig. 1). Turtles were found from 31 Dec. 2000 through 12 Jan. 2001, but most (N = 361; 90%) were found between 2 Jan. and 5 Jan. Water temperatures in the south end of St. Joseph Bay during the first few days of January were measured as low as 6°C (T. Summers, pers. obs.). The size-frequency of the green turtles is given in Figure 2. The mean SCL of the 10 Kemp's ridleys was 33.4 cm (range of 26.5–46.0 cm). The SCLs of the three loggerheads were 38.1, 48.9, and 55.4 cm.

Of the 341 live turtles that were taken to Gulf World, one green turtle died shortly after arrival. Most of the turtles (291 green turtles, three Kemp's ridleys, and two loggerheads) quickly resumed normal activity and were released at a site along the Gulf of Mexico a short distance southeast of St. Joseph Bay (at Indian Pass Beach, east of Cape San Blas; Fig. 1) on 7, 8, and 9 Jan. The water temperature at the release site was 14°C (A. Redlow, pers. obs.). Nine more green turtles were released at this same location on 16 Jan. Two of the remaining green turtles had fibropapilloma-like tumors. One of these turtles had a single tumor (smaller than 1 cm in diameter) on its neck. The other turtle had numerous tumors on most areas of soft tissue (except for the eyes and mouth), and some of the tumors were up to 4 cm in diameter. Both turtles were strong and active. Because fibropapillomatosis (FP) is likely a contagious disease (Herbst, 1994) that had not been previously documented in northwest Florida (Foley et al., 2005), we released these turtles on 25 Jan. in central-west Florida (Pinellas County, about 300 km southeast of St. Joseph Bay), where FP is commonly found on green turtles (Foley et al., 2005). The final 33 turtles (30 green turtles and three Kemp's ridleys) needed longer-term rehabilitation and were divided among six permitted sea turtle rehabilitation facilities in Florida (Clearwater Marine Aquarium, EPCOT's Living Seas, Florida Aquarium, Gulfarium, Mote Marine Laboratory, and Sea World). The most common ailments were the inability to dive, eye infection, and cloacal infection. One of the green turtles had ingested fishing line, and one end of the line had come out of the cloaca. Three of the green turtles that were taken for longer-term rehabilitation subsequently died, but the 30 other turtles recovered and were eventually released back into St. Joseph Bay along the west-central shore of St. Joseph Peninsula in Eagle Harbor (27 turtles) or at Indian Pass Beach (three turtles) (see Fig. 1).

Three of the turtles found during the cold-stunning event had been previously tagged. One of the green turtles that were found alive and then released on 9 Jan. had living tags (from switching a plug of plastral tissue with a plug of carapacial tissue) on the posteriormost left costal scute of the carapace and on the left pectoral scute of the plastron. This turtle was from a nest that hatched on Xcacel Beach (Quintana Roo, Yucutan, Mexico) in 1997. It was either one of the many thousands of green turtle hatchlings that were living-tagged after hatching and then released 2–3 d later at the same beach or one of the 200 green turtle hatchlings that were raised in captivity for 1 yr and then released on Xcacel Beach in 1998 (A. Negrete, pers. comm.). The SCL of this turtle upon recapture was 38.7 cm. One of the green turtles found dead had spent the previous winter at the J.L. Scott Aquarium in Biloxi, MS. This turtle was confiscated late in 1999 from an individual who likely captured it incidental to commercial fishing. The aquarium released this turtle just offshore of Deer Island, MS, on 20 April 2000. One of the Kemp's ridleys found dead had a single flipper tag and a PIT tag. This 1998 year-class turtle was captive-reared at the National Marine Fisheries Service's (NMFS) Galveston Laboratory and released offshore of Galveston, TX, in June 1999. The SCL of this turtle upon recapture was 36.7 cm.

Some of the green turtles found during the cold-stunning event had old, healed wounds. The most commonly observed signs of previous trauma were notches (generally a "U" shape) along the margin of the carapace (noted for 26 turtles) or along the trailing edge of one or more flippers (noted for 14 turtles). Fifteen of the green turtles were missing all or part (at least 25%) of one rear flipper; one was missing about 75% of the left front flipper; and two were missing a part of two flippers. Most of these turtles (16/18) had only rear-flipper injuries, and most of these turtles (13/18) had lost all or part of a flipper (front or rear) only on the left side. Although any (or all) of these flipper injuries could have come as a result of a shark bite, none of the injuries had definitive shark-bite characteristics. In addition to the green turtle that had ingested fishing line, two other green turtles had old, healed injuries along the jaw or face that may have resulted from being previously caught on hook and line. One green turtle had old, healed propeller wounds on its carapace, and three green turtles had unusual humps in some part of the carapace that may have resulted from a previous trauma or a congenital abnormality. None of the live turtles were determined to be emaciated.

Necropsies were conducted on 51 of the green turtles. Eight of the green turtles found dead were not necropsied because their viscera had been completely or mostly eaten by vultures (black vultures, *Coragyps atratus*, and turkey vultures, *Cathartes aura*). Four of the necropsied green turtles had also been fed upon by vultures, and although much of their viscera remained, the GI tracts had been damaged and were not collected for diet analysis. Clotted blood indicated that some of the turtles were still alive when attacked by the vultures. The three green turtles that later died at a rehabilitation facility were necropsied, but their GI tracts were not collected for diet analysis.

Only one of the necropsied green turtles appeared to be emaciated. Six of the green turtles had moderate to high numbers of white to tan nodules (0.02 cm diameter) distributed multifocally on the external surfaces of the GI tract and the liver and within the mesentery. These nodules were similar in appearance to the granulomas with intralesional larval cestodes that are regularly seen in necropsies of sea turtles from Florida (A. Foley, pers. obs.). Direct examination of the gonads revealed that 39 of the green turtles were female and 12 were male (3.25 females:1 male). The sex ratio of this group was female-biased ($P < 0.001$, Goodness of Fit for a 1:1 sex ratio, $X^2 = 26.93$).

Of the 44 green turtle GI tracts examined, all contained recently ingested submerged aquatic vegetation. Turtle grass was present in the upper GI tract (esophagus, stomach, and small intestine) of all of the green turtles. Shoal grass (*Halodule wrightii*) was present in the upper GI tract of 36 (82%) of the turtles. However, in all but five of the green turtles that had ingested both turtle grass and shoal grass, the dry weight of the shoal grass was less than 1% of the dry weight of the ingested turtle grass. Only one green turtle ingested more (by dry weight) shoal grass than turtle grass. Manatee grass (*Syringodium filiforme*) was found in only one GI tract. The amount of manatee grass that was ingested in this one GI tract was about the same (in dry weight) as the amount of turtle grass that was ingested (but they were not mixed, and turtle grass had been eaten more recently). Macroalgae was found in seven (16%) of the GI tracts. Genera of macroalgae identified in the samples were *Laurencia* and *Enteromorpha*. However, in all but one of the green turtles that had ingested macroalgae, the dry weight of the macroalgae was less than 1% of the dry weight of the ingested turtle grass. The green turtle that had ingested the greatest amount of macroalgae still ingested about six times more (by dry weight) turtle grass than macroalgae. One green turtle had also ingested two small pieces (one about 1 cm^2 and the other about 4 cm^2) of cellophane-like plastic.

Thirteen haplotypes were identified from mtDNA control region sequences obtained from 255 samples (Table 1). The most common haplotype was CM-A3 (45%), which has been reported at several nesting beaches, including Florida, Mexico, Costa Rica, Aves Island (in the center of the Caribbean Sea), and Surinam. The second most common haplotype was CM-A1 (27%), which has been found in Florida and Mexico. The other haplotypes were relatively rare and included five haplotypes that have not been reported to date from any nesting population. Bayesian mixed-stock analysis determined that the green turtles from St. Joseph Bay comprised a mixed stock of individuals principally (81%) from United States (Florida) and Mexico (Yucatan) nesting populations, but with an estimated 15% (1–30% CV) from the nesting population in Costa Rica (Tortuguero) (Table 2).

Of the 327 green turtles found cold-stunned in St. Joseph Bay during this event and subsequently released, 46 have been recaptured back in the southern half of St. Joseph Bay after 3–39 mo at large. Most of these turtles (42/46) had been

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TABLE 2. Mean estimated stock mixtures of green turtles found cold-stunned in St. Joseph Bay based on mixed-stock analysis using BAYES [10,000 resamplings (20,000 MCMC samples) of four stock mixtures composed of green turtles from seven major Atlantic nesting stocks]. Median and 95% confidence limits (2.5% and 97.5% quantiles) are shown.

Stock	Mean (standard deviation)	Median (upper quantile, lower quantile)
United States (Florida)	0.4889 (0.1205)	0.4900 (0.2544, 0.7112)
Mexico (Yucatan)	0.3231 (0.0928)	0.3130 (0.1704, 0.5299)
Costa Rica (Tortuguero)	0.1507 (0.0917)	0.1444 (0.0086, 0.3381)
Suriname/Aves	0.0210 (0.0149)	0.0187 (0.0009, 0.0553)
Brazil	0.0051 (0.0050)	0.0036 (0.0001, 0.0183)
Ascension Island	0.0054 (0.0052)	0.0039 (0.0002, 0.00192)
Guinea Biseau	0.0058 (0.0054)	0.0041 (0.0002, 0.0199)

released at Indian Pass Beach (outside of St. Joseph Bay, see Fig. 1) after cold-stunning. Many of the recaptures (36/49) were made in the south end of St. Joseph Bay by sea turtle researchers from the University of Florida (E. McMichael, pers. comm.). All of those turtles appeared healthy and were released. Eleven of the recaptures occurred during a subsequent cold-stunning event in southern St. Joseph Bay on 26 Jan. 2003 (this included two turtles also recaptured by the University of Florida researchers) (E. McMichael, pers. comm.). The other recapture within St. Joseph Bay was of a green turtle released at Indian Pass Beach and then found dead in the southern end of the bay 3 mo later. Only three of the 327 green turtles released after the current cold-stunning event were recaptured outside of St. Joseph Bay. One green turtle was found dead in Pensacola Bay, FL (about 200 km west-northwest of St. Joseph Bay) 1.5 mo after its release in Eagle Harbor. Another green turtle was found dead on the beach of St. George Island, FL (about 50 km east of St. Joseph Bay), 50 mo after release. The third green turtle was recaptured (alive and apparently healthy) by an Army Corps of Engineers relocation trawler just offshore of the south end of South Padre Island near Brownsville, TX (T. Bargo, pers. comm.) after 60 mo at large.

DISCUSSION

We classified previous sea turtle cold-stunning events (i.e., where at least dozens of turtles are cold-stunned) in the continental United States as being either acute or chronic (Table 3). According to our criteria for classifying these events, St. Joseph Bay is the third area in the United States where an acute event has been documented. To our knowledge, the only sea turtle cold-stunning event outside the United States occurred in the southern Adriatic Sea and involved only immature loggerheads (Bentivegna et al., 2002). As reported, the cold-stunning event in the Adriatic Sea appeared to be similar to the acute cold-stunning events documented in the United States.

The cold-stunning event in St. Joseph Bay involved more turtles than any other previous cold-stunning event. The next largest acute cold-stunning event occurred in Dec. 1989, when 256 sea turtles were found cold-stunned in the Mosquito Lagoon and adjacent waters (Schroeder et al., 1990). The record chronic cold-stunning event occurred during Nov. and Dec. 1999 and Jan. 2000, when 277 sea turtles were found cold-stunned along the shore of Cape Cod Bay (Still et al., 2002).

All the bodies of water in the United States where sea turtle cold-stunning events have occurred (Long Island Sound, Cape Cod Bay, Mosquito Lagoon, St. Joseph Bay, and Laguna Madre) share certain characteristics. All are enclosed to a large extent and are either closed at the southern end or are open there only to a limited extent. If there is a large opening into these water bodies, it occurs only at the northern end. Perhaps in an attempt to travel south when water temperatures drop near tolerable limits, sea turtles become effectively trapped and fail to escape these bodies of water in time to avoid cold-stunning.

Cold-stunned green turtles have been previously documented in St. Joseph Bay but only in small numbers. The Sea Turtle Stranding and Salvage Network (STSSN, coordinated nationally by NMFS and in Florida by FWC) documented a few cold-stunned green turtles over the course of several days during Dec. 1983 and Jan. 1984, again during Jan. 1989, and then again during Jan. 1994. One cold-stunned green turtle was documented in Jan. 1996 and another in Dec. 1997 (Foley, unpubl. data). All of these turtles were found in Eagle Harbor (see Fig. 1), where there is a small marina that is part of the St. Joseph Peninsula State Park.

TABLE 3. A comparison of acute and chronic sea turtle cold-stunning events in the United States.

Type of cold-stunning event	Sea turtle residency in area	Water depth of area	Event occurrence periodicity	Duration of event	Mortality	Species primarily affected	Areas of occurrence
Acute	Year-round (winter water temperatures usually warm enough for sea turtles to occur year-round)	Extensive shallow areas (< 2 m) that can cool quickly	Irregular (only during unusually cold winters)	Short (< 2 wk)	Low (< 30%)	green turtle, loggerhead	Mosquito Lagoon, FL (Witherington and Ehrhart, 1989; Schroeder et al., 1990), Laguna Madre, TX (Shaver, 1990); St. Joseph Bay, FL (present study)
Chronic	Seasonal (winter water temperatures usually drop below that tolerated by sea turtles)	Can include areas of deeper water (> 5 m)	Regular (every winter)	Long (1–3 mo)	High (> 60%)	Kemp's ridley, loggerhead	Long Island Sound, NY (Meylan and Sadove, 1986; Morreale et al., 1992; Gerle et al., 2000), Cape Cod Bay, MA (Still et al., 2002; Still et al., 2003)

Because all but three of the 49 recaptured green turtles from the current cold-stunning were found back in the southern end of St. Joseph Bay (some after more than 3 yr at large), we assume the current event provided an opportunity to study green turtles that were typically residing for 1 or more years in St. Joseph Bay. Large aggregations of green turtles (at least several hundred animals) have been documented as far north as Levy County, FL, in the eastern Gulf of Mexico (Carr and Caldwell, 1956) and as far north as Matagorda Bay, TX, in the western Gulf of Mexico (Hildebrand, 1982). The green turtle aggregation in St. Joseph Bay is farther north than any other documented in the Gulf of Mexico and the only one known to occur along the northern shore.

All of the green turtles that were found in St. Joseph Bay were likely neritic-phase immatures. Green turtles begin life in the water with an epipelagic, oceanic phase that can last for several years (Carr et al., 1978; Hirth, 1997). The smallest green turtles from St. Joseph Bay were close to the minimum size at which green turtles are known to begin their benthic, neritic phase (Hirth, 1997). The largest green turtles from Joseph Bay were too small to be within the range of sizes typical of adult green turtles (Hirth, 1997) and were likely to have been prepubescent based on the size of pubescent green turtles in other studies (Limpus et al., 1994; Musick and Limpus, 1997). As with almost all other near-shore areas in the continental United States where green turtles are known to occur, St. Joseph Bay is exclusively a developmental habitat for juvenile green turtles. The only area in the continental United States where there have been indications that resident adult green turtles may occur is in southern Florida, southwest of the Florida Keys (Schroeder et al., 1996).

In some areas, neritic-phase juvenile green turtles may pass through two successive developmental habitats. Juvenile green turtles found oceanside of Florida's east-central coast tend to be smaller than the juvenile green turtles found in adjacent lagoons and other estuaries (Henwood and Ogren, 1987; Guseman and Ehrhart, 1990). In southern Texas, juvenile green turtles found along the rocky shoreline of a pass opening to the Gulf of Mexico tend to be smaller than the juvenile green turtles found farther inshore in the adjacent bays and other estuaries (Coyne, 1994). Green turtles shifting from an oceanic phase to a neritic phase may first reside along nearshore areas directly adjacent to offshore waters (the first neritic-phase developmental habitat), where they feed predominantly on macroalgae. Next, the turtles move farther inshore into bays and other estuaries (the second neritic-phase developmental habitat), where they feed primarily on seagrasses.

It appears unlikely that green turtles in St. Joseph Bay spend an earlier portion of their neritic phase in another nearshore developmental habitat because of the large number of relatively small individuals. Most (61%) of the green turtles from St. Joseph Bay were less than 35 cm SCL, and only 15% were greater than 45 cm SCL. The mean SCL of green turtles from St. Joseph Bay tended to be between that of turtles in the first (Guseman and Ehrhart, 1990; Coyne, 1994) and second (Ehrhart, 1983; Coyne, 1994) neritic-phase developmental habitats of other areas. This may indicate that St. Joseph Bay has both the smaller and larger juvenile green turtles that could be segregated in other areas where juvenile green turtles occur.

Results of the mixed-stock analysis indicate that about 81% of the green turtles found in St. Joseph Bay belong to the nearby nesting populations in the United States (Florida) and Mexico (Yucatan). The more distant green turtle nesting population in Costa Rica (Tortuguero) is the largest in the Atlantic (Troëng and Rankin, 2005) and an order of magnitude greater than either the nesting population in Florida or in Mexico (FWC, unpubl. data; J. Zurita, pers. comm.), yet is represented by only about 15% of the green turtles in St. Joseph Bay.

The fact that five haplotypes found among the green turtles from St. Joseph Bay have not yet been reported from any nesting population indicates that further sampling is needed for more accurate mixed-stock analyses (see Bjorndal et al., 2005). In addition to several smaller nesting populations in the Caribbean and Gulf of Mexico that have not been sampled, the large population that nests in Florida also remains undersampled, and it is likely that the unidentified haplotypes are present there at low frequencies. Given the wide confidence limits of the estimated stock-contribution proportions, the point estimates from the current analysis should be interpreted with caution.

However, it is clear that the representative stock for the green turtle assemblage in St. Joseph Bay is different from that of nearby green turtle assemblages offshore of Hutchinson Island (along the Atlantic coast of Florida) and in the Bahamas. There is a distinct gradient between these three sites in the representation of turtles from the nesting populations in the United States and Mexico and the nesting population in Costa Rica (Tortuguero) (Table 4). Individuals from Costa Rica are relatively rare in St. Joseph Bay, more common offshore of Hutchinson Island, and most common in the Bahamas. Individuals from either the United States or Mexico are most common in St. Joseph Bay, less common offshore of Hutchinson Island, and relatively rare in the Bahamas.

TABLE 4. Representation of green turtle rookeries in the assemblages of juvenile green turtles from two different areas in the North Atlantic and one in the Gulf of Mexico. The mixed-stock analysis of green turtles from Hutchinson Island (Atlantic coast of Florida) was conducted by Bass and Witzell (2000) and that in the Bahamas was conducted by Lahanas et al. (1998). The representation of the rookeries in each assemblage is different ($P < 0.001$, $X^2 = 162.7$).

Location of green turtle assemblage	Representation by rookery		
	Costa Rica	United States/ Mexico	Others
St. Joseph Bay, FL	15%	81%	4%
Hutchinson Island, FL	53%	42%	5%
Bahamas	80%	5%	15%

The sex ratio of the juvenile green turtles from St. Joseph Bay was biased toward females. A prevalence of females has typically been the case when the sex ratios of sea turtle assemblages have been determined (Wibbels, 2003). A group of cold-stunned juvenile green turtles found in the Indian River lagoon system had a female-biased sex ratio (1.75 females:1.0 male) (Schroeder and Owens, 1994). Nevertheless, several assemblages of juvenile green turtles have been found to have an equal number of males and females (Bolten et al., 1992; Meylan et al., 1992; Wibbels et al., 1993; Limpus et al., 1994; Limpus and Reed, 1985; Koga and Balazs, 1996). Because the green turtle assemblage in St. Joseph Bay has a high proportion of turtles from the Florida nesting population, the female-producing incubation temperatures often documented in Florida (Mrosovsky and Provancha, 1992) could be a contributing factor to the female-biased sex ratio of green turtles in St. Joseph Bay.

Turtle grass is recognized as the most common forage of green turtles in the Caribbean (Bjorndal, 1997), but to our knowledge, the present study is the only one to document turtle grass as a major component of the diet of juvenile green turtles anywhere in the continental United States. Other studies in Florida have found that juvenile green turtles feed principally on manatee grass (Mendonça, 1983), shoal grass (Mendonça, 1983; Wershoven and Wershoven, 1992b), or macroalgae (Wershoven and Wershoven, 1992b; Redfoot, 2000). In southern Texas, juvenile green turtles living in bays were found to feed mainly on shoal grass (Coyne, 1994). In a study of the diets of juvenile green turtles in Long Island Sound, almost all had fed on macroalgae, and about half had also fed on eelgrass (*Zostera marina*) (Burke et al., 1992).

Juvenile green turtles that live in seagrass meadows may feed principally on seagrasses, even when a large amount of macroalgae is present (Mendonça, 1983). They have been known to feed primarily on the most abundant species of seagrass (Mendonca, 1983) or to selectively feed on the least abundant species of seagrass (Coyne, 1994). Turtle grass is the most prevalent species of seagrass in St. Joseph Bay (Florida Department of Environmental Protection, 1997) and was found to be the primary dietary item in the juvenile green turtles recovered from St. Joseph Bay.

Sea turtles living in nearshore areas are susceptible to a variety of mortality factors associated with human activities (National Research Council, Committee on Sea Turtle Conservation, 1990). In Florida, the most commonly documented human-related mortality factors are injuries from collisions with boats and entanglement in or ingestion of anthropogenic debris (Singel et al., 2003). Among the 410 sea turtles recovered from St. Joseph Bay during the current cold-stunning event, these anomalies were rare. One turtle had propeller wounds, two turtles had ingested monofilament fishing line, one turtle had ingested a small piece of cellophane-like plastic, and one turtle was entangled in monofilament fishing line. In addition, none of the 69 stranded sea turtles found in St. Joseph Bay during 1980–2000 and documented by the STSSN were noted as having propeller wounds or as being entangled (Foley, unpubl. data).

The incidental take of sea turtles in commercial and recreational fisheries can also be a source of significant mortality for sea turtles living in areas such as St. Joseph Bay (National Research Council, Committee on Sea Turtle Conservation, 1990). In Florida, nearshore gill-net fisheries have been specifically implicated in the lethal take of immature green turtles (Ehrhart et al., 1990). A statute banning gill nets in nearshore waters took effect in Florida on 1 July 1995. Still, illegal gill-netting continues to some extent in St. Joseph Bay. On 18 Oct. 2000, the STSSN documented four juvenile green turtles that were found dead in a 50-m piece of abandoned gill net in the southwestern corner of St. Joseph Bay (A. Foley, unpubl. data). The STSSN has not reported any hook-and-line captures of sea turtles from St. Joseph Bay (A. Foley, unpubl. data), but two of the green turtles found during the current cold-stunning event had old injuries to the jaw or to the side of the face that could have been caused by hook-and-line capture.

Perhaps the most worrisome discovery regarding potential mortality factors of sea turtles from St. Joseph Bay was the occurrence of FP in two of the green turtles from the current cold-stunning event. Fibropapillomatosis is a debilitating, often fatal, disease characterized by single to multiple tumors that may reach up to 30 cm in diameter (Herbst, 1994). Rates of occurrence in some parts of the world have reached levels as high as 92% (Herbst, 1994). Although FP is commonly documented on green turtles from the southern half of Florida, it had not been previously reported anywhere in the northern half of Florida (Foley et al., 2005). St. Joseph Bay and much of the surrounding area is currently afforded some environmental protection. Most of St. Joseph Bay is designated as an aquatic preserve by the State of Florida and as a Gulf Ecological Management Site by the U.S. Environmental Protection Agency. Much of the land surrounding the bay is either a state park, national wildlife refuge, or a state buffer preserve. However, despite the protected status of the area, seagrass meadows in St. Joseph Bay have been extensively damaged by boat propellers (Sargent et al., 1995).

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