

Hatching success of the Leatherback Sea Turtle,  
*Dermochelys coriacea*,  
in Natural and Relocated Nests  
on Gandoca Beach,  
Costa Rica



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## 0 Abstract

The hatching success of leatherback sea turtles (*Dermochelys coriacea*) of natural, relocated, and hatchery nests was determined in the nesting season of 2004 on Gandoca Beach, Costa Rica. Environmental factors such as nest temperatures, groundwater level, and the sand grain size distribution were examined for every nesting site and implicated on the hatching success. Natural nests as well as relocated nests show a hatching success of 45%, whereas hatchery nests have a success rate of 57%. Natural and relocated nests show fluctuations in temperature and are more likely to be affected by groundwater table, tidal inundation or beach erosion, while hatchery nests remain protected from environmental disturbance throughout the whole season. Sand grain composition and size was uniform within the hatchery nests. Relocated and hatchery nests show irregularities in grain size distribution. It is concluded that the interactions of environmental factors have an influence on the stability of the nesting environment providing successful incubation of the eggs. The hatchery provides a stable nesting environment and leads to adequate hatching success rates. The relocation strategy performed by the ANAI Sea Turtle Project contributes to successful development of nests, which are put in danger due to environmental disturbances.

## I Introduction

The leatherback sea turtle (*Dermochelys coriacea*) is considered a critically endangered species (Baillie, 1996) and populations are declining worldwide as a result of mainly anthropological impacts. Commercial use of leatherback products and incidental fishing resulting in juvenile and adult mortality cause major threats to this sea turtle species. Overlooking the dynamics of the world's population structure shows the extent of the threat. In 1982, Pritchard estimated that the global leatherback sea turtle population consists of 115'000 individuals. Until 1996 the global population declined to 35'400 individuals (Spotila et al., 1996). In less than one generation the population declined 70%. To prevent leatherback sea turtles from extinction a complete knowledge about their life cycle and nesting ecology is necessary. Conservation programs on nesting beaches provide an important effort to contribute to stable population dynamics.

Successful conservation on nesting beaches depends on various environmental factors as well as on conservation practices. The nesting beach is the incubator where embryonic development and successful incubation of the eggs depend on the presence of suitable conditions in the beach sand (Ackerman, 1997). Nest temperature, humidity and availability of respiratory gases are important factors that contribute to the development of the eggs. Interactions of beach material, their physical structure, the local climate, as well as the eggs in a clutch create a suitable microclimate for the development of the embryos.

The conditions on a beach are dynamic and vary among different beaches. Depending on local conditions, relocation of leatherback nests to a safe beach area or into artificial hatcheries is necessary. Relocation of nests is a common conservation strategy which embryonic and hatchling mortality (Swimmer, 1993). The decision on the safety of a relocation area largely depends on climatic circumstances and the dynamics of a beach. Founded knowledge about the danger of beach erosion or tidal inundation, the affection by the groundwater table, as well as the sand composition needs to be considered in order to successfully relocate leatherback sea turtle nests on a specific beach.

Gandoca Beach is one of the most important nesting beaches for the Caribbean leatherback population (Chacón et al., 2004). During the nesting season of 2004, the relocation conservation strategy conducted on Gandoca Beach, performed by the ANAI Sea Turtle Project, was examined in order to give recommendations on successful conservation strategies in the future.

We recorded hatching success of nests incubating under three different circumstances. Nests that were left *in situ* (natural nests), nests that were relocated to safer areas on the beach and nests relocated to an artificially constructed hatchery. Three environmental factors including nest temperatures, level of the groundwater, and sand grain size distribution were examined and their influence on the hatching success was recorded separately for each nesting site. The aim of this investigation was to examine the interactions of physical beach factors, local climatic conditions, and relocation strategies on Gandoca Beach. Furthermore, the influence

of these interactions on the hatching success was observed in order to improve relocation conservation strategies on Gandoca Beach.



## II General Information

### 1 Biology of Leatherback Sea Turtles

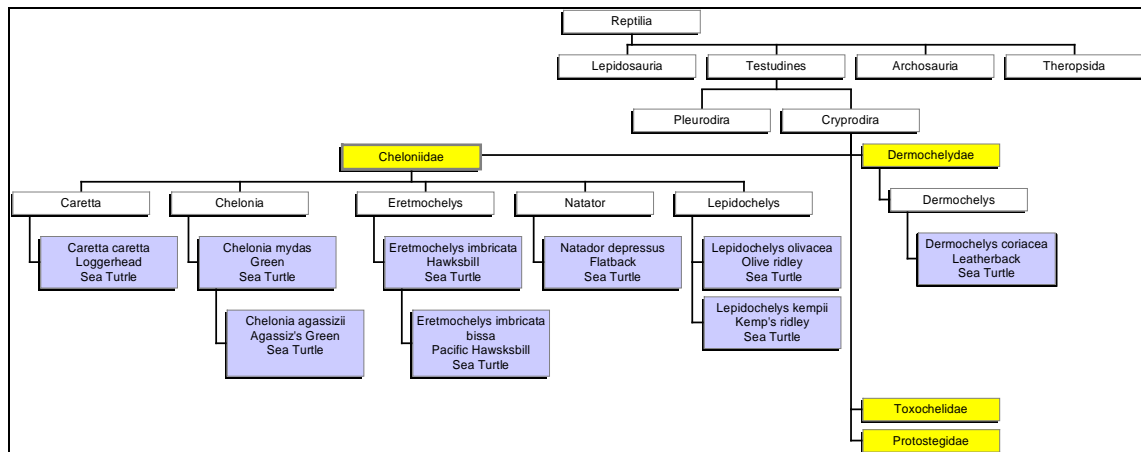
#### 1.1 Taxonomy

Together with terrestrial turtles, marine turtles belong to the order of Testudines, which is part of the Class of Reptilia. The evolution of the eggshell allowed the Reptilians as the first Vertebrates to colonize various terrestrial habitats independently of surrounding water (Storch et al., 1997). Reptilians show certain characteristics such as a vertebral column and an epidermal scale. They are all air breathing and exothermic (cold blooded; their external environment is primary source of body heat). Reptilians have an internal fertilization and the eggshell allows a water independent development of their eggs.

The Testudines (Turtles) are divided in three different families, Testudinoidea (recent amphibic and terrestrial turtles), Chelonoidea (marine turtles) and Trionychoidea (soft-shelled turtles). Numerous characteristics, genetic and morphological, distinguish marine turtles from other types of turtles. The shape of the marine turtle body shows hydrodynamic adaptations whereas the limbs are modified into flippers. The front limbs are modified into wing-like fore flippers and the hind flippers have become paddle-like or rudder-like (Wyneken, 1997).

The oldest sea turtle fossil (*Santanachelys*) is dating back to the middle Cretaceous Period (Gulko and Eckert, 2004). By the late Cretaceous (around 65 Million years ago) four distinct families of sea turtles have been described in which all the species are characterized by clear adaptations for marine life: Protostegidae, Toxochelidae, Cheloniidae and Dermochelidae.

Cheloniidae and the Dermochelidae are what is referred to “the living species of marine turtles”. The taxonomic family of the Cheloniidae includes five genera (*Caretta*, *Chelonia*, *Eretmochelys*, *Natator* and *Lepidochelys*) six species (*Caretta caretta*, *Chelonia mydas*, *Eretmochelys imbricata*, *Natator depressus*, *Lepidochelys olivacea* and *Lepidochelys kempii*) and two subspecies (*Chelonia agassizii* and *Eretmochelys imbricata bissa*). The Dermochelidae only includes one genera with one single species, *Dermochelys coriacea* (Tweksbury Institute of Herpetology, 2002, see Figure 1.1).



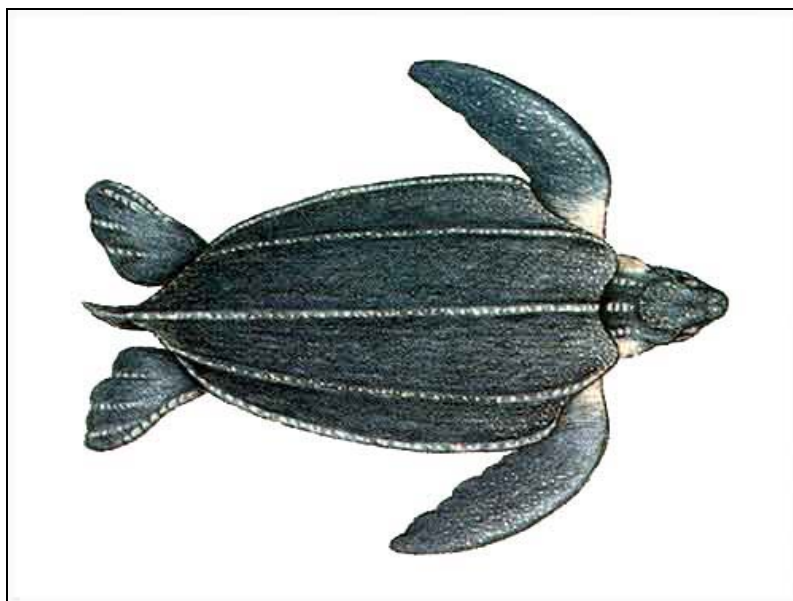
**Figure 1.1**  
Taxonomy of marine turtles with Cheloniidae and Dermochelyidae as the living species of marine turtles.

The generic name *Dermochelys* was introduced by Blainville (1816) whereas the specific name *coriacea* was first used by Vanelli (1761) and adopted by Linnaeus in 1766 (Rhodin and Smith, 1982). This name refers to the distinctive leathery, scaleless skin of the adult turtle. *Dermochelys coriacea* is known by a variety of common names like “leatherback” in English, “tortuga baula” or “tora” in Spanish and “tortue luthé” in French.

## 1.2 Morphological Characteristics

### 1.2.1 Description

The leatherback sea turtle shows striking morphological and physiological differences from other living sea turtles (Reina et al, 2002). Apart from its large size, the most obvious morphological difference is the shell, which is composed of thousands of small, polygonal bones covered by a black, flexible skin with a leathery appearance (Wood et al., 1996). The carapace of the leatherback is about 4cm thick and is made primarily of tough, oil-saturated connective tissue raised into seven longitudinal ridges. The front flippers are proportionally longer than in other sea turtles and may span 270 cm in an adult. The curved carapace length of adult females typically measures 130-175cm and their weight ranges between 250-500kg (Eckert, 2001). The largest leatherback on record (a male stranded on the coast of Wales in 1988) weighed 916 kg (Morgan 1989). The dorsal part of the body is black with scattered white and pink marks while the ventral part is mainly white. The spinal spot or “pink spot” on the top of the head of the turtle is an external characteristic, which differs by shape, color, size and pattern in every individual. Its function is uncertain but it may be a photosensitive organ.



**Figure 1.2**  
Adult leatherback sea turtle (Source: [www.ittiofauna.org](http://www.ittiofauna.org)).

### **1.2.2 Aquatic locomotion**

A marine turtles carapace is typically elongated along the anterior-posterior axis and appears streamlined, which associates with the emphasis of the pelagic life-style and well developed migratory behavior (Wyneken, 1997). The leatherback turtle is known for the longest vertical and horizontal migration and therefore their swimming patterns differ from those of the other sea turtle species. Functionally, sea turtles have exploited a mechanical system for propulsion that is not utilized by other turtle groups. With the elongated finger bones forming a major part of the limb, the flippers are relatively large in size and provide a powerful and fast locomotion. The flippers serve as wings (or propellers) as well as paddles. This morphological adaptation is reflected in distinctive behavioral and physiological characteristics, giving the marine turtles a remarkable ability to migrate over long distances through the oceans (Wyneken, 1997).

The flippers form a paddle that contributes to thrust production and as function a rudder in steering. With both fore-flippers simultaneously they carry out what is called a “power stroke” producing thrust to move vertically or horizontally. The flippers move upward and posteriorly, then downward and anteriorly (Wyneken, 1997). What differs compared to Cheloniidae is that the movements of a leatherback are rather upwards and downwards than anterior and posterior, which produces more thrust and may explain their ability to migrate over long distances without losing much energy.

## **1.3 Physiological Characteristics**

### **1.3.1 Thermal Biology**

According to Fair et al., 1972, leatherbacks are capable to maintain a corebody temperature as much as 18°C above the ambient water temperature. Due to a number of specific physiological adaptations, the leatherbacks are able to thermo-regulate in cold water. Their large size and a thick layer of subcutaneous tissue (6 to 7cm) favors heat retention from muscular activity and promotes a high volume to surface area ratio, which minimizes heat

loss. Different compositions of peripheral and central lipids and countercurrent vascular heat exchangers in the front and hind flippers allow thermoregulatory capabilities (James, 2001). Leatherbacks can use changes in blood flow to the skin and periphery to regulate body temperature, which allows them to maintain warm temperature in temperate oceans and avoid overheating in warm tropical waters. This allows them to migrate to temperate waters further than any other species of marine turtles. While nesting, it is typical to observe changes in color of their soft skin, appearing pale when they leave the water and changing to bright pink when they leave the nesting beach (Spotila et al., 1997). According to Spotila et al., the blood flow is more than ten times higher when skin appears pink compared to when the skin is pale. This increased blood flow helps to cool the turtle by bringing the heat to the skin surface where it can be transferred to the air.

### 1.3.2 Diving Physiology

*D. coriacea* belongs to the longest and deepest divers of the air-breathing vertebrates. They spend up to 97% of their time under water, where energetic and predatory costs are low, thus they can be considered truly subaquatic. The central feature of their diving ability involves an efficient oxygen transport system and extraordinary tolerance of hypoxia, which allows maximal use of limited oxygen stores (Lutcavage et al., 1996). Multi-chambered lungs full of elastic tissue provide large amounts of surface area for oxygen and carbon dioxide exchange and allow them to exchange oxygen at a faster rate than other reptiles. They are adapted to store the amount of oxygen needed for deep diving within their blood and other tissue and to make it available to the body cells. Even after lung collapse associated with an increased water pressure would normally cut off that oxygen supply, these storages maintain oxygen circuit to all vital organs (Gulko et al., 2004). In 1989 Eckert et al., recorded deepest dives from a leatherback sea turtle exceeding 1000m.



**Figure 1.3**  
Diving leatherback sea turtle (Source: Presentation  
Didiher Chacón)

### 1.3.3 Orientation, Navigation and Natal Beach Homing

Various theories about the orientation and navigation of all sea turtles have been suggested. The natal beach homing hypothesis proposes that adult sea turtles will return to lay their eggs on or near the same beaches they emerged from as hatchlings (Ackerman, 1997). Little is known about the underlying mechanism of long distance navigation in adult turtles and past

studies have been laboratory intensive undertakings, providing usually little information. The successful use of satellite tracking technology to reconstruct the paths of sea turtles allows interesting experiments about the navigational theories under natural conditions (Lohmann et al., 1996). There are two main theories about navigation and natal beach homing:

#### The chemical imprinting hypothesis

The chemical imprinting hypothesis suggests that sea turtle hatchlings imprint upon chemical cues unique to their natal beach and use this information years later to return to that same beach as adults for nesting (Lohmann et al., 1996). Experiments about chemical cues in natal beach recognition showed that during certain developmental periods turtles can acquire a preference for specific chemicals and retain it for at least two months. These experiments have been made under artificial conditions and it is not certain how such a process could occur under natural conditions. With the better understanding of oceanic currents and migratory paths of sea turtles, evidence against the use of chemical cues in long-distance navigation has begun to accumulate.

#### The magnetic map hypothesis

The observation that adults can swim directly to destinations from hundreds of kilometers away and return to their nesting sites, implies that turtles can determine their position relative to a goal (Lohmann, 1996). This so called “map sense” proposes that turtles determine their position using geomagnetic fields. The use of such a magnetic map allows precise journeying between nesting beaches and foraging sites.

### 1.4 Foraging

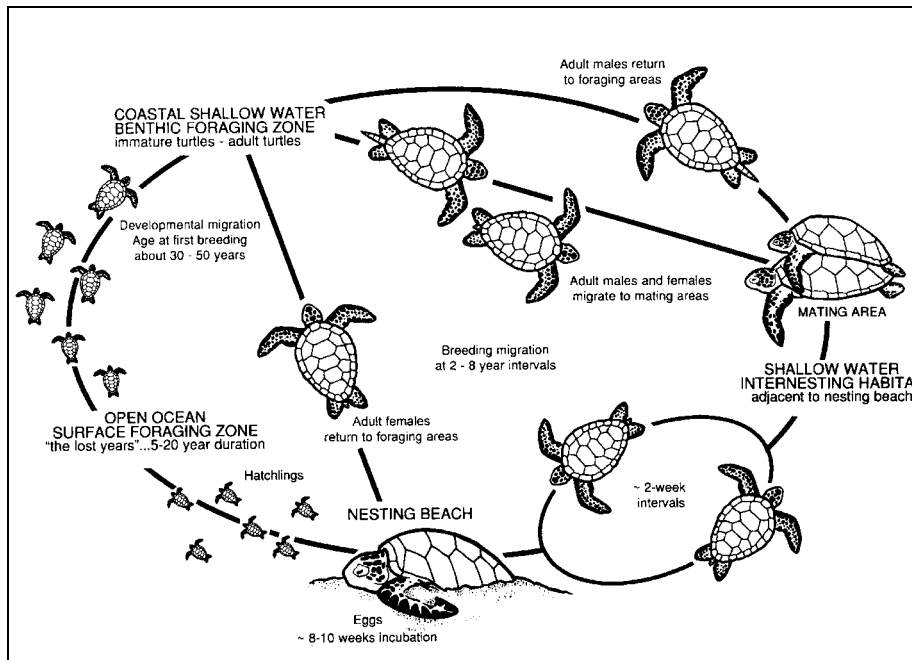
The leatherback is considered to be the most pelagic of all sea turtles, spending most of the time in the open ocean. They predominantly feed on jellyfish, salps and other gelatinous organisms, depending largely on the distribution of their prey (Bjørndal 1997). According to Plotkins (2003), *D. coriacea* does not appear to migrate to resident feeding grounds, but rather to swim continuously, possibly to areas of high food concentration. After Luschi et al., (2003), it is likely that current transportation allows leatherbacks to exploit macrop planktonic resources in the Indian Ocean over large areas with minimal effort. The distribution of jellyfish depends mainly on oceanographic factors, such as currents and predominantly occurs in colder waters, which explains their wanderings to most northern oceans of all living reptiles (up to Alaska). The leatherback post-nesting wanderings are therefore a consequence of their preference for oceanic current systems as foraging areas, and should not be considered migrations, a term usually referred to movements uninfluenced by the available resources. This has been shown previously in sharks and whales (Luschi et al., 2003). Characteristic diving thermal adaptations allow them to feed throughout the whole water column, from the surface to great depth (Bjørndal 1997). Possible morphological adaptations to the feeding are the sharply pointed upper and lower jaw cusps and the characteristic esophageal papillae. The shape of the jaw allows the leatherbacks to pierce jellyfish easily. The papillae are stiff projections with pointed tips, oriented posterior towards the stomach. By contracting the esophagus the saltwater is expelled while the papillae act to trap the food and facilitate the swallowing (Bjørndal, 1997).



**Figure 1.5**  
Esophageal papillae  
facilitating the swallowing of  
jellyfish (Source: Presentation  
Didiher Chacón)

### 1.5 Reproduction and Life cycle

The reproduction of the *D. coriacea* is similar to other species of sea turtles and occurs within the reproductive time where the animals move from foraging areas to mating areas, located in the tropics or subtropics. Usually the males then return to the foraging areas and the females move to nesting areas (Miller, 1996). Based on a skeletochronological analysis by Zug and Parham (1996), leatherback sea turtles reach sexual maturity after 13-14 years. Most experts agree that their life span is 30 years. The time between hatching and beginning of sexual maturity is called “the lost years”. Observations of hatchlings and juveniles during this period of their life cycle are missing. It is assumed that they spend the time drifting in ocean currents where they feed in driftlines (Gulko et al., 2004). Once they reached sexual maturity, nesting occurs, typically at intervals of two or three years. Females generally nest in 9-10 days intervals, depositing an average of 5-7 nests per nesting season on the selected beach (Eckert, 1999). They prefer to nest on beaches, which are easy accessible from the sea. The beach platform must be high enough in order not to be undulated by the spring tide or flooded by the water table below (Moritmer, 1995). The beach sand should provide optimal conditions for the development of the offspring (see chapter 2.4). Nesting typically occurs at night and approximately 70-80 fertile eggs are laid in each nest, along with a variable number of smaller infertile eggs (Eckert, 1999b).



**Figure 1.6**  
 Generalized life cycles of sea turtles, individual species vary in the duration phases. *Dermochelys coriacea* remain pelagic foragers throughout their lives. (Source: Miller, D. 1997. Reproduction in Sea Turtles. In: Lutz, The Biology of Sea Turtles. CRC Press, New York).

The nesting process follows a general pattern which contain the emergence from the surf, ascending to the beach, excavating the body bit, digging the egg chamber, oviposition, filling the egg chamber, filling in the body pit and returning to the sea (Miller, 1997).

Once the eggs are buried, they are left on their own and depend on optimal environmental conditions. Temperature, humidity and gas exchange in the nest must be within the embryonic tolerance and decide about their development (see chapter 2). After an incubation time of 55-75 days, the hatchlings usually emerge onto the surface of the beach during the early evening (Miller, 1997). Leatherback hatchlings are dorsally mostly black and covered with tiny polygonal or bead-like scales; the flippers are margined in white and rows of white scales appear as stripes along the length of the back. With an average carapace length of 60mm and an average weight of 50gr (Márquez, 1990), the hatchlings have to find the way to the sea immediately after hatching. The next time they return ashore is when they reached sexual maturity, after having lived approximately 15 years in the pelagic ocean.



**Figure 1.6**  
 Leatherback sea turtle hatchling (Photo by Didiher Chacón, 2005)

## 1.6 Distribution

*D. coriacea*, with the most extensive geographic range of any reptile, inhabits tropical and temperate waters of the Atlantic, Pacific, and Indian Oceans with the northern most latitude recorded at 70°15'N and the southern most at approximately 27°S (James, 2001). The nesting mainly occurs on tropical and subtropical beaches.

According to Spotila et al, (1996) major nesting beaches can be found in the following parts of the world:

**Western Atlantic:** French Guiana, Ya:loma:po and related Beaches, Suriname, Guayana, Brazil

**Caribbean:** Costa Rica, Panama, Trinidad, Colombia, Dominican Republic, St.Croix, USVI, Isla Culebra, Puerto Rico, Florida (Atlantic coast)

**Eastern Atlantic:** Babon, Pongar Pt., Ndiridi

**Indian Ocean:** South Africa, Sri Lanka, Andaman, Nicobar Islands

**West Pacific:** Irian Jaya, Papua New Guinea, Malaysia

**East Pacific:** Mexico, Costa Rica

The most northern known nesting location on the Atlantic coast is Blackbeard Island, Georgia. Very little is known about the distribution of post-hatchlings or juveniles. According to Eckert (1999), Leatherbacks with curved carapace lengths smaller than 100 cm seem to be limited to regions warmer than 26°C.

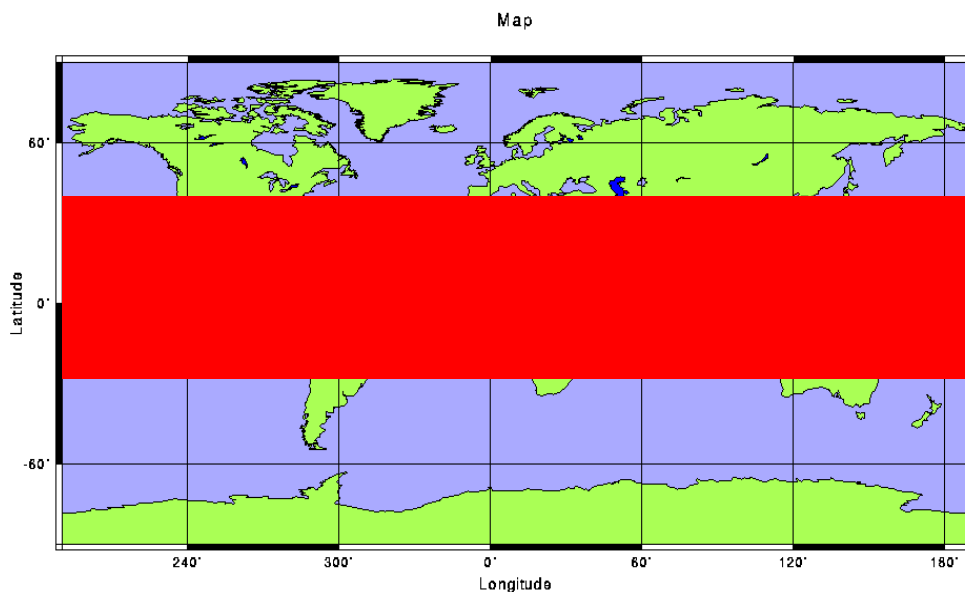


Figure.1.7

Worldwide distribution of leatherback sea turtles (Source: Chacón, 2004)

## 1.7 Population Trends

The survey of reproduction activity on nesting beaches is likely to be the most accurate way to get an overview of the global leatherback population. The first attempt to evaluate the global population was made by Ross (1979), who estimated that 29'000 to 45'000 adult leatherbacks existed in the world, not counting the rokeries of Eastern Pacific, which had not been discovered yet. In 1982 Pritchard estimated that the global population consists of 115'000 adult females, while the Mexican population supported up to 60% of the global total.



Spotila et al., made the most recent estimation in 1996. According to published data, unpublished information, and personal comments from 28 leatherback nesting sites, a estimation of 20'000 to 30'000 adult females represents the current leatherback sea turtle population. Taken together, this represents a reduction of the global population of 78% in 14 years, less than one generation (Sarti, 2000).

Based on the number of nestings known to date, it is feared that some of the most important populations have collapsed. Declining populations have been recorded in Malaysia and the East Pacific (Spotila *et al.* 2000). In the Mexican population for example, the number of nests has fallen from 5,080 to less than 100 annually in one of the main rookeries of the Pacific coast. In Costa Rica nests dropped from 1,646 to less than 500 nests in the main nesting beach of the Pacific coast. Leatherback turtle nesting in Caribbean Costa Rica has remained stable or experienced a slight decline over the past 15 years, a markedly different trend from the rapid nesting decline on the Pacific Beaches of Costa Rica (Spotila et al., 2000) According to Troeng et al., (2004), the leatherback rookery of Caribbean Central America represents one of the four largest remaining rookeries worldwide, together with French Guiana/Suriname, Gabon and Trinidad. For the coast of Africa, there are historical records of South Africa. Recent reports mention that West Africa has an important population with around 10,400 nests per season, but the total area occupied for the leatherbacks is not well known and there is no historical information available.

According to Spotilia et al, 1996, major colonies (1000 or more) still exist in French Guiana, in Suriname, in Gabon, and at Las Baulas Park (Playa Grande and Playa Langosta) in Costa Rica. Leatherbacks still nest in hundreds on Irian Jaya, Tortuguero and Gandoca/Manzanillo in Costa Rica, Trinidad, Dominican Republic, Columbia, Bocas del Torro in Panama, Guyana, Playa Tongaland in South Africa and the north coast of Papua New Guinea.

## 1.8 Threats

### 1.8.1 Natural Threats

The main natural threats are caused by a wide variety of natural predators who prey on eggs and hatchlings. On the nesting beach, predators like crabs, ants, lizards, seabirds, vultures, and raccoons harm the nests. They either acquire access to a nest by digging and feeding on the eggs or they prey on the hatchlings in the nest or when they are on the way to the ocean. Other serious predators are domestic animals like pigs and dogs. They usually discover the location of a nest by their sense of smell, borrow it up, and feed on the eggs or hatchlings. Once the hatchlings reach the sea, they are threatened from marine predators like seabirds, sharks, and other predatory fish, like catfish and jacks.

Environmental threats to sea turtle clutches can be caused by climatic changes, followed by decreasing or increasing sand temperatures or changes in groundwater level. Natural beach dynamics such as erosion, tidal inundation affect the nests and prevent them from successful incubation. Wooden debris washed ashore poses insurmountable obstacles to leatherback sea turtle hatchlings.

## 1.8.2 Human associated Threats

### Human presence on the beach

Vehicular traffic, foot traffic, and traffic of domestic animals such as cows walking on the beach can damage buried eggs and pre-emergent hatchlings (Lutcavage et al., 1996). People walking continuously upon developing nests cause compacting of the sand in the egg chamber. This influences the delicate microclimate of the clutch and possibly inhibits an optimal gas exchange. Inhibited gas exchange can have serious effects on the development of the eggs and the hatching success of the clutch. Furthermore, the compacting of the sand aggravates or inhibits the emerging of the hatchlings which can cause their death by suffocation or contusion. Human presence at night can cause turtle to abort nesting attempts, although controlled guided "turtle watch groups produce a minimal disturbance (Lutcavage et al., 1996). Organized turtle watching has an important conservation and education potential and became a common form of ecotourism. In Costa Rica often former subsistence turtle hunters or egg collectors have been trained as turtle guides and now derive income by leading nesting beach tours (Lutcavage et al., 1996).

### Commercial use of leatherback products

Leatherback sea turtles are mainly threatened because of their eggs, meat and oil. The eggs are harvested by local egg poachers and sold for assurance of their subsistence. People all over the world use sea turtle eggs for food. Some cultures believe that eating eggs promotes a long life, as turtles are amongst the longest living animals (Gulko, 2004). A common cultural reference is the belief in aphrodisiac qualities of eggs, particularly in Central America. Although there is no scientific prove of an aphrodisiac effect, they emphasize their nutritional and economic value to their families (Campbell, 2003). The oily meat is not widely favored but is typically prepared by sun-drying or stewing whereas the oil is used for medicinal purposes, generally in cases of respiratory congestion (Chacòn, 1999).

### Fisheries

According to the National Academy of Science (1990), a comprehensive review of sources of mortality in Sea turtles has found that incidental capture in shrimp trawls accounts for more mortality than all other sources of human activities combined. Trawling, logline fishing, driftnets, lobster and fish trap lines as well as gillnets, cause the greatest threat to juvenile and adult sea turtles worldwide. The result of these fishing activities is death by drowning due to forced submergence because of entanglement within the gear. Discharged or lost fishing gear also contributes to this problem through ingestion, entanglement, or blocking of access to feeding, nesting and basking areas (Gulko, 2004). According to Spotila et al., 1996, the incidental mortality in the Pacific Leatherback population is at least 1000 adults per year. Based on the population estimates, this is a 22% rate of mortality and simulations suggest that this would half the population in only 10 years even in the absence of egg poaching on nesting beaches. Leatherback populations in the Indian Ocean and Western Pacific Ocean cannot withstand even moderate levels of adult mortality and the current level of incidental mortality in commercial fisheries in these areas will cause the extinction of these populations if they continue. The laws in sea turtle fishery differ in every country and are hard to survey due to the fact that sea turtles are strongly migratory.

## Pollution

In the coastal zone, chronic pollution from industrial, agricultural waste, and urban runoff is a threat to sea turtles. To leatherback sea turtles plastic material and nonbiodegradable debris poses specific danger. As pelagic foragers, they prey on cnidarians and other gelatinous invertebrates, and it has been suggested that they may not be able to distinguish between gelatinous prey and transparent plastic, the type of debris most often found in their digestive track (Lutcavage et al., 1996). But also other types of debris originating from boats, ships, and from offshore oil, gas and chemical platforms cause serious danger to the leatherback sea turtles.

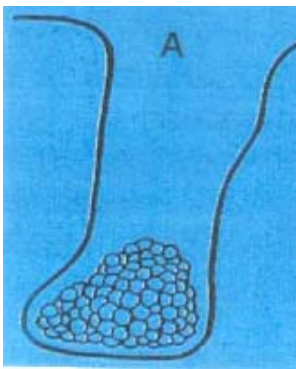
### 1.9 Conservation Status

The leatherback is classified as endangered by the World Conservation Union (Baillie and Groobridge, 1996). They are included in Annex II of the Protocol to the Cartagena Convention concerning Specially Protected Areas and Wildlife (SPA); Appendix I of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES); Appendices I and II of the Convention of Migratory Species (Bonn Convention); and Appendix II of the Convention on European Wildlife and Natural Habitats (Bern Convention) (Hykle, 1999). The species is also listed in the annexes of the Convention on Nature Protection and Wildlife Preservation in the western Hemisphere, a designation intended to convey that their protection is of “special urgency and importance” (IUCN, 2000). On the IUCN Red List of threatened species the leatherback sea turtle is classified as critically endangered since 2003.

## 2 Nesting Environment

### 2.1 Introduction

Once a leatherback female decided to nest, there are a few factors she needs to consider when choosing a good nest site. The beach area must be accessible from the sea, the angle of the beach must be high enough so that the tide can not routinely inundate the eggs and to prevent the underlying water table from reaching the nest. The beach sand substrate must be moist and porous enough to prevent collapse during construction of the egg chamber and to allow extensive gas diffusion (Gulko, et al., 2004). Leatherbacks alternately dig the egg chamber with their hind flippers and construct a nest in the shape of a “boot”, generally reaching 75cm in depth and 40cm in width.



**Figure 2.1**  
Nest shape of a *D. coriacea*  
Nest (Source: Chacón et al, 2004)

Besides the beach area, there are other important factors deciding on optimal embryonic development and high hatchling success of the clutch. Successful incubation depends on suitable conditions in the beach sand. Temperature, humidity, and gas exchange generate the microclimate for embryonic development of the sea turtle eggs. The composition of the surrounding sand and its compacting also contributes to suitable circumstances for development. The interaction of physical characteristics of the beach material, physical structures of the beach, local climate, and the eggs in the clutch, generate a specific microclimate and influence the incubation. The microclimate is dynamic and changes with the biological activity in the clutch and on the beach (Ackerman, 1997).

### 2.2 Temperature

The incubation temperatures in a sea turtle nest changes with diurnal beach temperatures and the exchanging of heat between the nest and the beach. Beach temperatures typically range between 26 to 35°C (Ackerman, 1997), but can increase or decrease rapidly with climatic changes like persistent solar radiation or rain. The diurnal temperature differences in the beach are lessened with the depth of a nest but still have a certain influence on the nest temperature. The thermal tolerance for development of sea turtle embryos incubated at constant temperatures ranges between 25 to 27°C and 33 to 35°C and is around 10°C wide (Ackerman, 1997). The incubation period typically increases with decreasing incubation temperatures.

Sea turtles have a temperature-dependent sex determination. The incubation temperature producing an equal number of both sexes (pivotal temperature) for leatherback sea turtles is 29.5°C and the sensitive period for sex determinations appears to be in the middle third of incubation (Mrosovsky, 1994). Below the pivotal temperature, male hatchlings will be produced and at warmer temperature, female hatchlings are being developed. The knowledge about temperature dependent sex determination plays an important role in conservation management. Eggs incubated under artificial conditions such as hatcheries or polystyrene incubators can influence their sex ratio and therefore propose a reaction to global population trends. Global climate changes (including global warming) might lead to the production of only female hatchlings and so to long term sex imbalances and may contribute to extinction of this species (Hays et al., 2003).

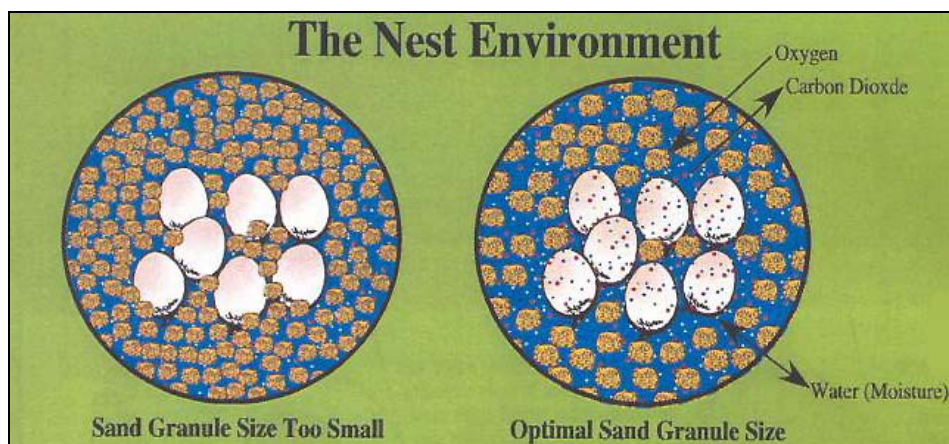
### 2.3 Water

The water exchange of sea turtle eggs occurs between a number of different compartments inside the egg, including the embryo and extra embryonic membranes such as the yolk sac, the chorioallantois, and the amnion (Ackermann, 1997). Very little is known about the water economy and the osmotic balance inside the sea turtle egg at any stage of embryonic development. Changing water sources of the beach like persistent rain or changes in groundwater influence the water balance and can generate a gravitational potential in the beach sand. The distribution of water in the beach is a result of the balance between water entering and water leaving a sand layer at any depth in the sand depth profile (Ackermann, 1997). If it is raining, water drains until it reaches equilibrium with the capillary forces caused by the interspaces between the sand grains. The water table also causes a gravitational potential in the beach and when surface drying occurs after a rainfall, water is lost from the sand to the atmosphere and the changing of the water balance causes a movement of water from the water table upwards. Depending on where the eggs in the clutch are located, the influence of the changing water potential can affect their development. Persistent rain or flooding of the nesting beach causes humid circumstances and mainly affect the eggs on the periphery of the clutch. Rainfall also causes decreasing temperatures, which aggravates the influence of the developing clutch and increases the incubation period. Depending on the beach the absence of rain can also affect the eggs. Therefore, according to Bilinsky et al., 2001, properly selected and monitored environmental conditions in artificial hatcheries in relocation programs is a very important purpose in effective sea turtle conservation management.

### 2.4 Gas exchange

Gas exchange in sea turtle clutches is necessary to support the metabolic activity of the developing embryo. Through the eggshell and associated eggshell membranes such as the chorioallantois, the embryo exchanges air and carbon dioxide. Sea turtle egg clutches develop in a medium, which allows gas exchange only through the gas filled fraction of the soil. O<sub>2</sub> and CO<sub>2</sub> move through convection or diffusion within the gas spaces of the surrounding sand grains or the interspaces between the eggs in the clutch. Convection in the beach can be induced by temperature variation, changes in atmospheric pressure, or displacement of soil air by water table movement (Ackerman, 1997). The composition of the sand, especially the size

of the grain, is an important factor for an optimal nesting environment. According to Gulko et al, 2004, small sand granulate is more likely to aggravate the exchange of O<sub>2</sub> and CO<sub>2</sub> and inhibits an optimal moisture level in the nest, which can be critical in maintaining the appropriate temperature, humidity, salinity and gas exchange necessary for embryonic development. The humidity of the sand is favored by the size of the pores of the grains and enables the substrate to retain water, which also decides over a successful construction of the nest. According to Mortimer, 1990, dry and coarse sand may be structurally unstable and therefore liable to collapse. Optimal particle size distribution varies from one locality to another, depending on patterns of precipitation, on mineral composition and shape of the particles composing the substrate.



## Figure 2.2

Sea turtle nests with sand grains of small and optimal size. Smaller grains provide less available open space for gas diffusion between the embryo and the nest (source:Gulko et al, 2004).

Sand compacted by heavy rain or human and domestic animal presence on the beach can also cause difficulties of essential gas exchange within the clutch and proposes difficulties to the emerging hatchlings on their way to the surface. Under the aspect of conservation, it has to be considered that relocated nests are located on areas with low human disturbance. Nests crowded together, for example occurring in a hatchery situation, will not allow an independent gas exchange. According to Ackerman, 1997, nests within a meter or less of each other are likely to “breathe” eachothers gases. Furthermore the selection of an optimal area for the construction of a hatchery is of importance. The sand in the selected area usually has to be sieved to avoid disturbing roots or other organic debris.

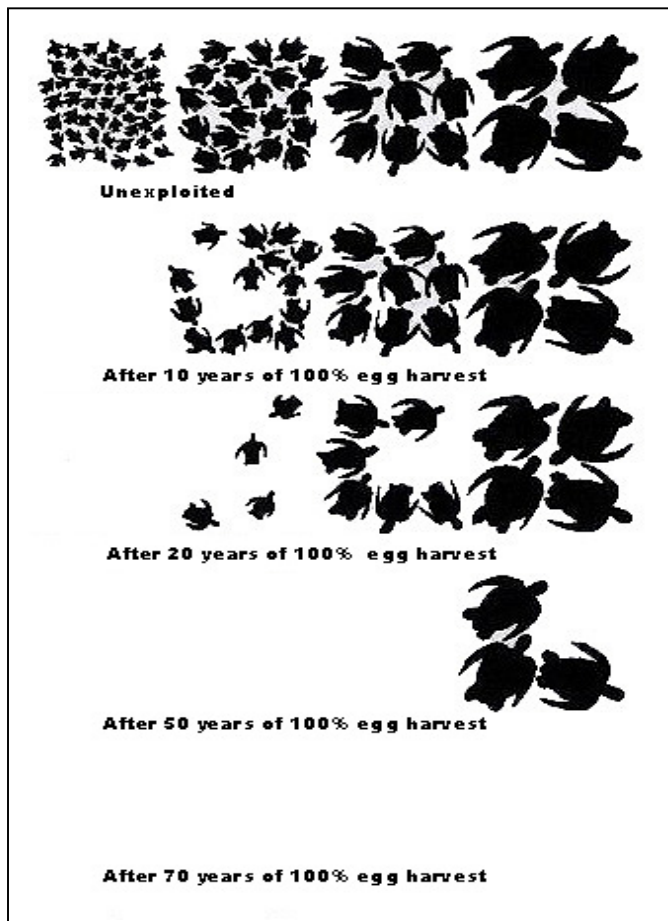
### 3 Sea Turtle Conservation

#### 3.1 Introduction

The World Conservation Strategy defined “Conservation” as “the management of human use of organisms or ecosystems to ensure such use is sustainable. Besides sustainable use, conservation includes protection, maintenance, rehabilitation, restoration, and enhancement of populations and ecosystems”. Due to various threats, worldwide populations of *Dermochelys coriacea* have been declining dramatically in the past 20 years and continue to do so. Without active intervention and management, global leatherback sea turtle populations are expected to continue to decline towards extinction (Eckert et al., 1999a). Conservation on a global level is necessary but has to follow certain guidelines in order to be successful. Effective conservation management in leatherback sea turtles includes continuative research and monitoring all aspects concerning their biology and habitat requirements. The interaction of their habitat use with the human use of habitat requires international laws on a political level in order to prevent habitat overexploitation. At the same time, the political, economic, and cultural conditions of people affected by conservation management actions need to be considered, and active participation of coastal people in marine turtle conservation needs to be promoted (Eckert et al., 1999a).

#### 3.2 Conservation management for long-lived species

Long-lived marine animals such as leatherback sea turtles generally grow slowly and mature at a late age and are therefore more vulnerable to excessive mortalities and rapid population collapse (Musick, 1999). In long-lived species the effect of overexploitation on a population, such as over harvesting of the eggs, is visible only after a certain time. Such a delayed effect is illustrated in Figure 3.1 after Moritmer, 1995b:



### Figure 3.1

This figure represents the destruction of a green turtle nesting population due to overharvesting of eggs. After 10 years 100% egg harvest, no hatchling turtles will remain in the population and the number of juvenile turtles will be reduced. The numbers of subadults and adults will be the same as in the unexploited population. After 20 years of 100% harvest, there will still be no hatchlings. Furthermore there will be fewer juvenile and subadults than in the population 10 years ago, but the number of breeding adults will have remained the same. After 50 years of 100% egg harvest, there will be no hatchlings, no juveniles, no subadults and the numbers of adults coming to the nesting beaches will have begun to decline. Only at this point the declining has become visible. After 70 years of 100% egg harvest the turtle population will be extinct.

Knowledge based on long-term based conservation is essential to predict the outcome of specific management actions. The biological constraints of sea turtles have to be taken into consideration in conservation efforts (Meylan et al., 2000). Research programs must address the root causes of turtle populations declines rather than seek short-term solutions that do not prevent long-term declines (Siegel et al., 2000).

### 3.3 Global Strategies for Conservation

Several global strategies for sea turtle conservation in general have been elaborated in the last century. They all focus on the same goal, namely to prevent existing population from declining further. For example the UICN Marine Turtle Specialist Group developed in 1995 a global strategy of sea turtle conservation which promotes (1) research and monitoring, (2) integrated management of marine turtles and their habitats, (3) capacity for conservation, research, and management, (4) public awareness, information sharing, and education, (5) community participation in conservation, (6) regional and international cooperation, and (7) evaluation of the status of marine turtles. This strategy is focusing predominantly on worldwide problems threatening marine turtles.

Regionally coordinated programs are important to focus on specific circumstances and promote adjusted interventions. Regional sea turtle programs are already on going in many areas, including the Mediterranean, western Indian Ocean, Northern Indian Ocean, South Pacific, and Caribbean. Cooperative tagging programs, conservation education campaigns, and exchange of technical information are common elements of these regional programs (Meylan et al., 2000). Programs on a regional level usually consist of different local conservation projects in the same region such as the participants of the Wider Caribbean Sea Turtle Conservation Network in the Caribbean region for example. The conception of conservation strategies on regional levels such as provided by the “Manual for Major Practices for Sea Turtle Conservation in Central America” (Chacon et al., 2000) for example are important requirements for successful sea turtle conservation on a global scale. Conservation efforts in the United States are largely guided by the recovery plans prepared for individual species of marine turtles by the national Marine Fisheries Service and the U.S. Fish and Wildlife Service.



International and regional political agreements such as reached by CITES (Convention on International Trade in Endangered Species) restrict the international trade in sea turtles and sea turtle products, whereas the Inter-American Convention for the Protection and Conservation of Sea Turtles addresses the intentional harvesting, accidental drowning of sea turtles in fisheries gear, and habitat protection (Meylan et al., 2000). The following insight into sea turtle conservation techniques will mainly focus on activities on nesting beaches, following the recommendations of the UICN Sea Turtles Specialist Group, 1999.

### 3.4 Conservation techniques on nesting beaches

#### 3.4.1 Beach survey

According to Schroeder et al., 1999, nesting beach surveys are the most widely implemented monitoring tools in use by the global sea turtle community. They are an important component of a comprehensive program to assess and monitor the status of sea turtle populations. It provides information on the number of nests deposited annually, the number of nesting females that are reproductively active annually, and annual nest productivity. Nesting beach surveys show the nesting activity within a nesting season. Precise information on geography of the survey area, available personnel and equipment, and nest density are imperative for successful beach surveys. To be successful over a long period of time, beach surveys need to be cost-effective, reproducible, quantitatively rigorous, and easily taught to others who will continue the surveys (Schroeder, et al., 1999). The nesting monitoring methodology usually is carried out by patrolling the beach on foot. Therefore, it is useful to divide the beach into equal zones (with beach markers located on the vegetation for example) usually looked after patrols consisting of one leader and some volunteers. The patrols are mainly carried out at night and are divided into different shifts. In some cases, especially when hatchlings are expected to emerge, the patrols may be set on the early morning as well. During each shift, the members of a patrol walk along the beach looking out for a turtle to come ashore or for eventual false crawls of turtles that did not nest.

#### 3.4.2 Data Collection

Once a female decided to come ashore, she is generally measured to relate body size to reproductive output, to determine minimum size at sexual maturity, and to monitor nesting female size for a particular rookery (Bolten, 1999). Straight or curved carapace length and carapace width is being measured with calipers or a flexible measuring tape. After the nest is built the shape of the nest, usually depth and width is measured. The numbers of fertile and infertile eggs laid in the clutch are reported during the egg laying process. At the same time, the existence of external metal tags or the evidence of previous external tags is checked and the tag identification numbers are recorded. In case that there are no tag evidences, the turtle receives new external tags and is recorded as a new female, nesting on the specific beach. Internal identification tags such as PIT tags (Passive Integrated Transponder tags) are small microchips (about the size of a grain of rice) that are injected into a turtle's shoulder muscle using a hand-held applicator gun. Researchers use hand-held scanners to detect PIT tags in turtles. The scanner reads the electromagnetic code of the tag and displays the identification number. Scanning turtles for PIT tags allows us to identify turtles that have previously been tagged on nesting beaches. Tagging a turtle is a very delicate issue and has to be dealt with

experienced personnel in order to prevent injuries or following infections. At the beginning of the nesting process, the final destination of the nest has to be decided. If the clutch is laid on a area of the beach where its successful incubation is not warranted, such as on zones of tidal inundation or danger of erosion, the eggs are usually relocated to an other area on the beach or into a hatchery. Eggs should be handled carefully and only relocated within a few hours of oviposition.

### 3.4.3 Relocations

Relocation of eggs to other parts of a beach is a common conservation practice used in order to reduce embryo and hatchling mortality due beach dynamic influences or human disturbance (Swimmer, 1993). Nonetheless, the relocation conservation strategy is controversial. The hatchling success might be negatively influenced by human interactions such as egg handling, choosing an appropriate relocation area, or by false reconstruction of the nest. Therefore some environmental factors need to be considered in order to ensure successful incubation of the nest. As explained in chapter 2.2, the new location for the eggs must provide adequate conditions of moisture, temperature, and gas exchange to support the developing embryos and be secure from predators and poachers (Miller, 1999). Leatherback sea turtles construct their nest in a specific shape, it is important to measure the depth and width of the natural nest and reconstruct it in the original shape.

The position of a relocated nest can be marked by using different techniques such as putting four wooden sticks with a colored tape around in the sand at the area of the clutch. On beaches where poachers are present, it is recommended to avoid any visual signs of the clutch location and use marking methods as the triangulation marking which will be explained in detail in Chapter 5.2.4. The location of a nest is also important to know to survey the areas in periods when the hatchlings are ready to hatch and gain information about their emergence.

### 3.4.4 Hatcheries

A hatchery is an artificially constructed and protected area on the beach (Chacon pers comm., 2004). It is usually delimited by a physical defense and monitored. The use of open air hatcheries has been criticized since it might influence the natural sex ratio of the hatchlings depending on where it is located (Marcovaldi et al., 1996). Shade areas produce more male and sunny areas more female hatchlings. A specific hatchery management is necessary to avoid such problems. Hatcheries are especially important in areas where nest predation or human traffic on the beach is the major source of mortality. In times of climatic changes such as storms or heavy rain, a relocation of eggs into the hatchery might be suggestive. A hatchery also has an impact on public awareness and can serve as an information center for sea turtle conservation.

### 3.4.5 Involving the public

Local communities associated with marine turtles and their habitats are often a strong force in the depletion of marine turtle populations and the destruction of their habitats (Eckert et al., 1999b). Conservation management projects need to include people in existing sea turtle projects and encourage them to participate in conservation activities instead of overharvesting

their resources. Reasonable conservation strategies can provide social and economic gains for local people. Although sustainable use is controversial in sea turtle conservation (Campbell, 2003), the use of sea turtle eggs under some conditions may be allowed such as shown in the example of Ostional in Costa Rica. Ostional is famous for its “arribadas” where olive ridley sea turtles nest in thousands at the same time. Following a specific conservation management, the local people are allowed to use the eggs to ensure their own existence but at the same time ensure the survival of the population (Campbell, 1998). The ANAI Sea Turtle Project on Gandoca Beach, the study area of my thesis, realizes an other example of community based conservation. The conservation strategy of ANAI will be explained in detail in chapter 4. Community based conservation also means to facilitate education programs in communities such as information events in local schools. Ecotourism is an other possibility to contribute to public awareness but remains controversial. Many examples of sea turtle ecotourism have been realized, but their effects on conservation, economies and sociology have not been monitored (Witherington et al., 2003). Turtle watching on nesting beaches or in the sea can lead to massive disturbance and harm rather than provide successful conservation.

### III Specific Information

#### 4 ANAI Sea Turtle Conservation on Gandoca Beach

##### 4.1 Introduction

Located on the southeast coast of Costa Rica Gandoca Beach is a major nesting beach for the leatherback Sea turtle in Caribbean Central America (Chacón, 1996). Gandoca Beach lies on the Caribbean coast of Costa Rica and belongs to the Talamanca region. The beach, where the Sea Turtle Project is located is part of the Gandoca/Manzanillo Wildlife Refuge, which is one of the most bio divers areas on the planet. This Refuge hosts over 2% of the worlds biodiversity and covers 4436 hectares of marine area and 5000 hectares of protected land (ANAI, 2004). At the same time when the Wildlife Refuge was established, in 1985, Asociación ANAI founded the ANAI Sea Turtle Conservation Project. The creation of the refuge and the marine turtle project lead the Asociación ANAI, the Costa Rican government, and local leaders of the gandocan community to start working together in a team to conserve biodiversity and at the same time considering the cultural and economic requirements of the community.

##### 4.2 Asociación ANAI

Asociación ANAI (Asociación de Nuevas Alquimistas Internacional) is a Costa Rican non-governmental organization, which was formed in 1978 to pursue what has since become known as sustainable development in Costa Rica's Talamanca region. The staff includes agronomists, foresters, biologists, economists, and educators. With the exception of the three founders, the staff and the Board of Directors are Costa Ricans. The mission of ANAI is to "help the people of Talamanca, design and implement a strategy linking socio-economic development, cultural strengthening and biodiversity conservation" (ANAI, 2004). ANAI's Talamanca initiative envisions a healthy mix of protected rainforests, wetlands and marine areas, managed forests, diverse agro-ecosystems, and human services like education, health care and ecotourism (ANAI, 2004). The Marine Turtle Program in Gandoca was one of the first initiatives within the Refuge and over the past 20 years it showed important development in socioeconomic aspects and conservation efforts.

##### 4.3 Community based Conservation in Gandoca

The village of Gandoca is accessible through a small road and can be considered as a fairly isolated community. Unlike other Caribbean coastal communities, which are typically populated by English-speaking Afro-Caribbean people, Gandoca is mainly hispanic. The people are mostly immigrants or the children of immigrants from other regions of Costa Rica, Nicaragua, Panama, Honduras and El Salvador. (Asociación ANAI, 2004). The main reason they colonized this region was because of working opportunities in a logging company in the 1940's. After the logging company left the area, people were forced to find other activities to ensure their survival. Many of them started working on the banana plantation or tried to settle in self-sustainable cultivation of corn and beans. They also began extracting natural resources including sea turtle derivates. Mainly leatherback sea turtle eggs but also green turtles and hawksbill turtles have been used for their meat and carapaces (Wagner, pers. Comm.). Until around 1980, the Sea Turtle nesting population at Gandoca remained largely unknown. Turtle

eggs were harvested by the small local population at levels that were probably sustainable. With the re-emergence of the Banana Companies and the improved road system in the area, poaching became a serious threat to the Sea Turtles. ANAI estimated that in the early 1980s, over 99% of the leatherback nests laid on Gandoca Beach were harvested (ANAI, 2004). With the establishment of the Gandoca/Manzanillo Wildlife Refuge and the founding of the Sea Turtle Conservation Program, the protection of the nesting colonies started. With the help of MINAI (Ministerio del Ambiente y Energía) and local people who are involved in conservation efforts by participating on nightly patrols, egg poachers have been successfully kept away. By 1990, poaching was fairly well controlled. ANAI established a Volunteer program, which involves volunteers from all over the world in protecting the nesting beach. The volunteers working for the Sea Turtle project are lodging with the local families and therefore provide an income for the local community which far exceeds the income for the eggs sold on the black market. Local people working as assistants, volunteer coordinator, or tour guides can ensure their income with conservation efforts and help to protect their resources instead of overexploit them. Guided tours or transport services as well as tourists visiting Gandoca Beach also generate an income and provide future economic potential for the community of Gandoca (ANAI, 2004).

#### 4.4 The Sea Turtle Conservation Program Management

Under the guiding of Didiher Chacón, a very experienced marine biologist and supervisor of my thesis, the Sea Turtle Project in Gandoca takes place every nesting season from the beginning of March to the end of July. The peak nesting period for the turtles is April/May with the first hatching turtles emerging from mid May to the end of the season (ANAI, 2004). As part of the conservation effort, ANAI recruits national and international volunteers to help with the research and conservation work. The staff usually consists of the coordinating biologist, local assistants and assistants from other countries. Besides this there are a lot of emergent assistants and about 500 volunteers during the whole nesting season (578 in the season of 2004). The work of the staff and the volunteers underlies the guidance of the biologist and follows the recommendations of the manual of nesting management of ANAI composed in 1990 and adjusted by the recommendations of the UICN/ SSC Marine Turtle Specialist Group (2000) and by Chacón (2001). A volunteer manual guides the volunteers through conservation activities but the importance to the individual volunteer and his interpretation remains a cultural and personal problem. Therefore the staff is very concerned to train all the volunteers in all conservation issues and give them a feeling that their help is of great importance for the project. The conservation efforts on the nesting beach are comparable to those described in Chapter 3.4. Following the recommendations of the Manual for Major Practices for Conservation of Sea Turtles in Central America (Chacón et al., 2000) nightly patrols measure and tag the nesting turtles and decide over a relocation of the eggs to other parts of the beach or to the hatchery. Hatching turtles are recorded, measured, weighted and released to the sea.

The relocation of nests also follows a certain strategy. According to Chacón (pers. Comm.), all conservation efforts concerning a leatherback sea turtle nest should first consider

conservation *in situ*, meaning the nest remains at the location where the female laid it. If this is not possible due to several environmental conditions (danger of inundation, beach erosion affection by the groundwater or human presence on the beach) the nests should be relocated in the same area as the natural nest but on the upper beach, strictly considering the recommendations of the ANAI Volunteer Manual. Relocation of a nest into the hatchery is the last step in conservation and is only recommended when human presence on the beach is intense and the intention of the visitors is not guaranteed. In Gandoca, this is usually the case during Eater Week (Semana Santa) when many tourists and locals come to the beach.

This current thesis is focusing on the relocation strategy conducted during the season of 2004 on Gandoca Beach and the following hatching success. It only shows a specific part of all conservational efforts made in Gandoca. The comprehensive conservational efforts are presented in detail in the Annual Nesting Report 2004 (Chacón, 2004).

## 5 Material and Methods

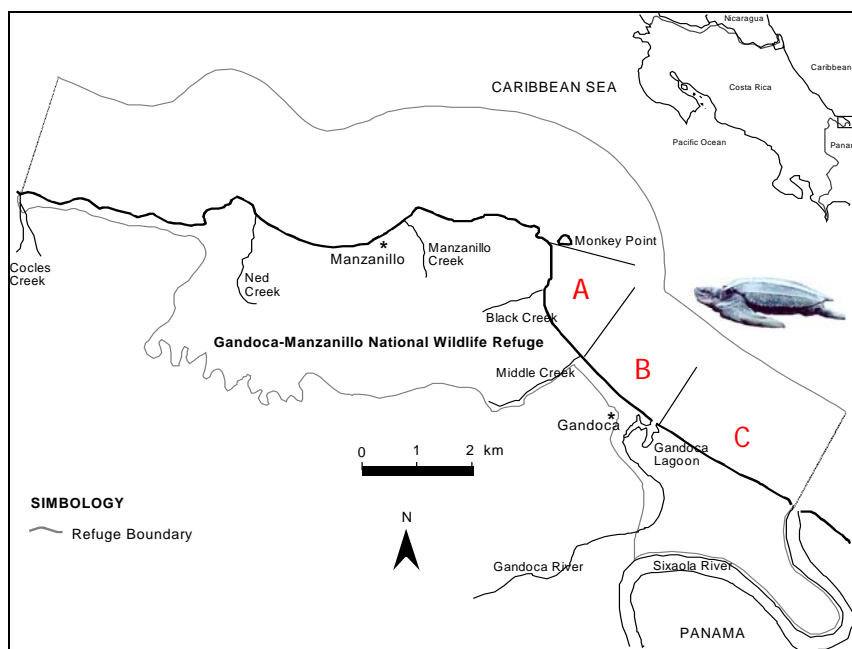
### 5.1 Experimental Design

#### 5.1.1 Study Area

##### Gandoca/Manzanillo Wildlife Refuge

The ANAI Sea Turtle Conservation Project and the studies for this thesis take place in the Gandoca/Manzanillo National Wildlife Refuge. The Refuge is located on the southeastern coast of Costa Rica and encompasses 4436 hectares of marine and 5000 hectares of terrestrial area; geographically it is found at 82°37'W, 9°37'.

Following the Holdridge life zone scheme, Gandoca/Manzanillo Wildlife Refuge shows characteristics of humid tropical forest within the whole Refuge. The climate of the refuge maintains the typical pattern of the Southern sector of the Costa Rican Caribbean coast. The annual average temperature ranges from 25°C to 27°C with a maximum of 31°C in the dry season (December to April) and a minimum of 20°C in the wet season (May to November). With a relative humidity between 86% and 88% this region of Costa Rica is considered as the most humid area of the whole country. The terrestrial environment of the refuge provides a very divers habitat with the dominance of wetlands mainly conformed by Yolillo palm trees (*Raphia taedigera*) and several species of mangroves.



**Figure 5.1**  
Gandoca/Manzanillo Wildlife Refuge on the southeastern coast of Costa Rica  
(Source: Marine Turtle Conservation Program, Asociación ANAI)

##### Gandoca Beach

Costa Rica's Gandoca Beach extending 8.85 km is limited north with Monkey Point (Punta Mona) and south with the river mouth of the Sixaola River forming the boarder to Panama (Rio Sixaola).



**Figure 5.2**  
Gandoca Beach extending 8.85km from Punta Mona to Río Sixaola (Source: Marine Turtle Conservation Program, Asociación ANAI)

Gandoca Beach is a black sand beach. It is considered as a typical high-energy beach with a medium to steep slope and a crenate to dentate shoreline. The width of the berm varies from 0 to 20m. The configuration of some parts of the berm and the height of the beach change over the course of the nesting and hatching process as a result of long shore currents, storm waves, and high spring tides. In general, this beach has a poorly developed berm during much of the year and is partly covered by assorted debris including logs, coconut husks, and a wider variety and amount of plastic articles most of which originate from the banana farms and are washed in the Sixaola river watershed. The high energy characteristics of this coast area are associated with narrow platforms and prevailing strong currents moving in a southerly direction. Although the shores are mostly sandy, there has been a recent input of organic waste and terrigenous sediment due to human induced changes along the Atlantic lowland, as a result of agricultural and touristic development. This sediment is assumed to be responsible for the degradation of some reefs and sea grass beds and may prevent access to the beaches (Chacón & McLarney, 1996)



**Figure 5.3**  
Gandoca Beach (Picture by Sue Furler, 2004).



The beach is continentally limited by the wetlands of Punta Mona and the Gandoca Lagoon where plant species such as the Yolillo palm trees (*Raphia taedigera*) and *Connosperma panamaensis* are dominant. The majority of mangrove species found in the Caribbean such as *Rhizophora mangle*, *Rhizophora racemosa*, *Avicennia germinans*, *Laguncularia racemosa* and *Conocarpus erecta* occur within the wetlands (Chaccon, 2004). Together with other species such as *Crassostrea rhizophorae*, *Megalops atlanticus*, *Bubulcus ibis*, *Casmerodius albus* and *Egretta caerulea* the wetlands form an ecosystem with an exceptional plant biodiversity. The coastal plain is characterized by humid tropical forest predominantly presented by the species *Priora copaifera*, *Pterocarpus officinale* and *Carapa guianensis*. The wetlands and the coastal plain provide an important habitat for a variety of threatened animal species such as the Collared Perycar ( *Tayassu tajacu*), the Spider Monkey (*Ateles geoffroyii*), the Ocelot (*Leopardus pardalis*), the Jaguarundi (*Herpailurus yaguarundi*), the Manatee (*Trichechus manatus*), the Neotropical Otter (*Lutra lonigcaudus*), the American Crocodile (*Crocodylus acutus*), the Cayman (*Caiman crocodilus*), the Red-lore Amazon Parrot (*Amazona autumnalis*) and the Great Curassow (*Crax rubra*).

Beside the Leatherback sea turtle (*Dermochelys coriacea*) other sea turtle species such as the Green turtle (*Chelonia mydas*) and the Hawksbill turtle (*Eretmochelys imbricata*) use Gandoca Beach as a nesting beach.

#### 5.1.2 Study requirements

As the studies for the current thesis take place within the ANAI Sea Turtle Program and their conservation efforts, (see chapter 4.4) there are a few environmental conditions set before the collection of the current data was possible.

##### Preparation of the beach

As recommended by Schroeder et al., 1999 (see chapter 2.3.4) Gandoca Beach was divided in equal zones in the beginning of the nesting season. Every 50 meters a wooden marker not less than 1.2 meters in height (ANAI Volunteer manual, 2004) were placed close to the vegetation line. The first marker was placed right after the rocky area of Punta Mona (Monkey Point). The southernmost marker was placed close to the mouth of Sixaola River. All wooden markers were placed on the border to the vegetation and the numbers painted on each marker were visible from the beach. The beach markers divide the beach into three different sectors. Sector A reaches from Monkey Point to Middle Creek, Sector B from Middle Creek to Gandoca Lagoon and sector C from Gandoca Lagoon to the Sixaola River (see Figure 5.1).

##### Construction of the hatchery

Following the recommendations of the Manual for Major Practices for Conservation of Sea Turtles in Central America (Chacon et al., 2000), two hatcheries were built during the nesting season of 2004 on Gandoca Beach. Hatchery A was built between the wooden markers 22 and 23 and Hatchery B between 65 and 66. The decision over the place of construction is based on the knowledge about the beach dynamics and experience of Didier Chacón and the local assistants. Before the construction was started, the sand was sieved through a 0.25cm sieve to

a depth of 90cm and thereby exempt from pieces of wood, roots and other debris, which could harm the incubating eggs.



Figure 5.4

**Local assistant and volunteers sieving the beach sand by constructing the hatchery  
(Photo by Arturo Herreda, 2004)**

After the sieving, the hatchery area was hedged by a physical defense and sand bags were put on the outside of the defense to avoid affection by the tide. Furthermore, a trench of 40cm depth was built around the hatchery area for protection from the sea. The inside area was divided into squares of 50 cm x 50 cm by a cord and the emanating rows and columns were provided with an identification number. The inside of the hatchery resembled a chessboard and the nests were placed in every other square of a column and shifted in every row. With this division there is an inter-square between each nest of a row and between the nests of a column providing enough space for independent incubation for each nest (see chapter 2.2.4 independent gas exchange). For protection of the nests from predators such as crabs feeding from the eggs and flies laying their larvae into the nests, baskets made of 0.5 cm metal mesh rings were prepared to cover each nest.



Figure 5.5 Hatchery nests relocated in rows with specific interspaces.

#### Hatchery

Half of the hatchery area was covered with a specific shade to compare the hatchling success of the nests in the sun and in the shade and to gain information about the sex determination of the specific nests.



**Figure 5.6**

The hatchery with protecting sand bags on the outside and nests covered with white basket in the inside. (Photo by Sue Furler, 2004)

Volunteers monitored the hatcheries 24 hours a few days before the first nest was expected until the last nest hatched. The monitoring time for Hatchery A was from the 1<sup>st</sup> of May to the 20<sup>th</sup> of August and for Hatchery B from the 18<sup>th</sup> of April to the 10<sup>th</sup> of August.

#### Nightly patrols

During the whole nesting season nightly patrols were set from 8:00 pm to 12:00 mn and from 12:00 mn to 4:00 am for each Sector (A, B and C). A patrol usually consists of one leader (local or international assistant) one emergent assistant and 5 to 7 volunteers. The patrols walk along the beach within their sector watching out for turtles coming ashore. Once a turtle is found specific data about the nesting turtle was collected following the conservation strategies of ANAI (see chapter 2.4.3). The position of the turtle was registered, it was measured by carapace length and width and it was tagged with external or internal tags if there were no previous tags from other nesting events. After the collection of the general information about turtle the decision over the final destination was made. Nests were either left natural, relocated on the beach or brought to the hatchery.

### 5.2 Data Collection

The data was collected from the 21<sup>st</sup> of March until the 2<sup>nd</sup> of August 2004. The data collection was divided in two different periods. In the first period the nest samples for the current study were selected and their final destination was set. In the second period, after the incubation, the hatchling success of the chosen study nests was determined.

#### 5.2.1 Sample size

Since the amount of nesting turtles on Gandoca Beach and total number of nests for the nesting season of 2004 was unknown the sample size was based on group selection. A representative sample of 30 nests per nesting site was chosen for inventory. The sample size for group selection provides a manageable data collection for one person and is possible over the available sampling time.

#### 5.2.2 Biometric data

For each nest regardless of the final destination a data sheet was created recoding the date of the nesting, the exact location of the nest, the depth and width of the nest (in cm) and the amount of fertile and infertile eggs. The depth of a nest was measured by putting a wooden stick to the bottom of the nest and remeasuring it with a flexible measuring tape. The width of the nest was measured by measuring the width of the right back flipper of the turtle. The amount of fertile and infertile eggs was registered for every nest but with differing counting methods for the different nesting sites.

### 5.2.3 Final destination of the nest

From the 21<sup>st</sup> of March on I attended one of nightly patrols. After the patrol collected the general data about the nesting turtle and made the decision about its final destination I took the nest as a sample nest for the specific category and proceeded as described below.

#### Natural nests

A nest was left natural when the distance from the nest to the sea was enough so that there was no danger of tidal inundation or erosion. The nest was not in danger to be affected by nearby creeks, which can flood the nests in periods of persistent rain. Furthermore, the risk of predation or the disturbance by humans was considered to be low, which was the case when the location of the nest was close to one of the two hatcheries because they were monitored 24 hours. The eggs in a natural nest develop under natural conditions without anthropological influence.

The amount of eggs was counted with the help of two tally counters, one for the fertile and one for the infertile eggs usually laid at the end of the egg laying process. Together with the data gained from the nesting event, the amount of eggs was registered on the data sheet for the specific turtle.

#### Relocated nests

If the nests need to be relocated because of environmental disturbance a plastic bag was put under the cloak of the turtle to collect the eggs. The bag was put to use right after the nest was constructed. At that point the female typically covers the hole with one of her flippers, which is the indication for applying the bag. After the infertile eggs begin to fall and the female moves one of her back flippers to start covering the eggs with sand, this is the exact moment to remove the bag. If the bag is not removed by then, the female will start to cover the nest with sand and the bag cannot be pulled out anymore without harming the eggs. In this case a flexible measuring tape or a long piece of cord should be put in the nest and held on the other end. After the female covered the nest and camouflaged the whole area the only way to find the exact location of the nest again is to dig along the cord to the center of the original clutch. After collection of the eggs the bag was closed immediately to avoid heat loss and guarded on a safe area until the female covered the nest and leaves the beach. The leader of the patrol decided where the nest was relocated following the recommendations of the ANAI Volunteer Manual considering that there is no driftwood around, the nest is not close to permanent or intermittent streams or rivers, there are no plant roots in the sand nearby that could interfere with the nest and that there are no paths, trails or houses nearby. For the reconstruction of the nest the exact measurements of the original nest measured during the nesting event were used.

The typical shape of a leatherback nest (see chapter 2.1) was considered and the reconstruction trained properly in the official nest reconstruction training implemented by the staff of ANAI. The eggs were handled very carefully and only with latex gloves. While relocating the eggs, they were counted and registered by type (infertile and fertile eggs) considering that the fertile eggs need to be put first into the nest and the infertile eggs on the top. After the relocation the area where the nest was located was camouflaged properly so that there was no evidence of the nest visible anymore. Especially footprints or handprints in the sand or spots of wet sand can indicate the nest location to poachers. Therefore the area was covered with dry sand and new footprints made leading to another area of the beach to distract from the original location of the clutch.



**Figure 5.7**  
Collection of leatherback sea turtle eggs during the egg laying process (Source: Marine Turtle Conservation Program, Asociación ANAI).

### Hatchery nests

A nest was only brought to the hatchery when human presence on the beach was intense and their intention was not foreseeable. This was especially the case during Easter week (Semana Santa, datum) when many tourists visit Gandoca Beach. Another important reason for relocating egg to the hatchery was periods of bad climatic conditions such as thunderstorms with persistent rain and rough sea and danger of beach inundation or erosion. For temporal reasons only the nests in Hatchery B were take as sample nests. The collection of the eggs and their handling was the same as for relocated nest with the difference that they were brought to the hatchery for relocation. The volunteers on hatchery duty usually did the reburying of the eggs and not by the leader of the patrol thus the information of the nest shape was written on a identification paper which was put into the bag together with the eggs. Following the instructions of the hatchery manual and the previous hatchery training obligatory for everybody attending a hatchery shift, the eggs were relocated according to the hatchery map. The information about the amount of fertile and infertile eggs and the position of the nest in the hatchery was registered on the data sheet together with the biometric data for that specific nest.



**Figure 5.8**

Hatchery inside with relocated sea turtle nests. The nests are relocated within the squares signaled by the white cord and protected by white baskets (photo by Arturo Herreda).

#### 5.2.4 Nest marking

The position of every nest was marked either directly after the nesting process or on the following day. In some nights the weather or light conditions did not allow to mark the nests right after nesting. In these cases, the leaders mentally memorized where they relocated the nests and showed me the exact location on the following day.

##### Marking with wooden sticks

If natural or relocated nests were located around the hatchery area, they were marked with four wooden sticks centering the clutch. A colored tape was put around the sticks and metal identification maker with the date of the nesting, the nesting category, the Sector, and the number of the nest was attached (Example: 21.03.04/**NB15**; Natural nest nr. **15** in sector **B**). This marking method was only used when the nests were under monitoring, the marking of nests with colored tapes on other parts of the beach would attracte poachers immediately.



**Figure 5.9**

Nest marked with wooden sticks and colored tape. The nest is equipped with a thermocouple (Photo by Sue Furler, 2004).

##### The triangle method

Most of the nests were marked with the triangle method as recommended by the Florida Fish and Wildlife Conservation Commission (2002). To mark the nest location, the exact distance from the precise or approximate clutch location to two selected marking points along the upper beach vegetation was measured. The marking points were beach markers if available or

special palm trees or other trees. First, the clutch was marked with a wooden stick and then exact distance from the first beach marker (A) to the clutch (B) was measured and registered. Then, the distance to the second beach marker (C) was measured continuously so that the registered measurement at beach marker C was the distance from A over B to C. Last, the distance between the two markers (A and C) was measured for reference and the measurement result in a triangle. The location of the clutch and the referring beach markers were drawn on a little map so it can be retrieved after the incubation. In cases where the specific beach marker trees were difficult to identify, they were marked with an adhesive tape and an identification number on it.

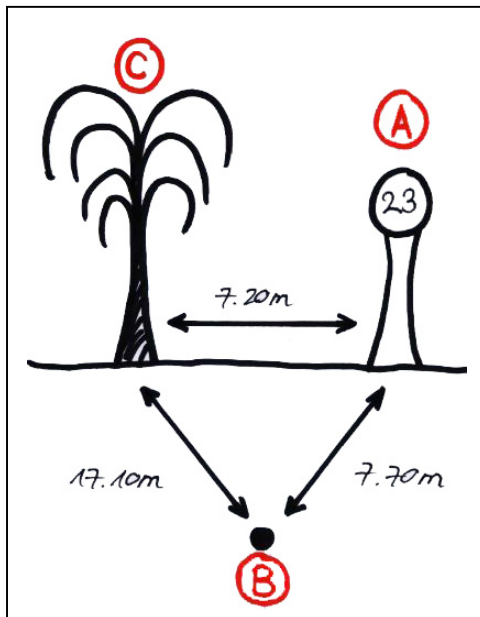


Figure 5.10

Map of a nest marked with the triangle method. The a beach marker (A) and the palm tree (C) are used as reference points for the measurements (source: Sue Furler, 2005)

The nests in the hatchery where not marked specifically but the identification number of the squares in which the nests were relocated (see chapter 5.1.2) was recorded (Example: **D17**; column **D**, row **17**).

### 5.2.5 Hatchling success

After an alert time of 50 days the incubating natural, relocated, and hatchery nests were monitored on a daily morning survey at 5am, using the maps for their exact location. Evidences of hatchling tracks were recorded, the tracks counted and the date registered. The nests where continuously monitored for further hatchling tracks and approximately seven days after the last hatchling track was registered the exhumation date was set. If no evidence for emerged hatchlings was found, the exhumation date was calculated using the average incubation time of nests located on similar areas and adding seven days to the estimated date. Hatchling emerging from the hatchery were counted, measured, weighed and released by the volunteers on hatchery duty. Hatchlings emerging from natural or relocated nests found the way to the sea by themselves.

To determine the hatchling success, nests ready for exhumation were dug up using latex gloves to prevent from egg parasites (larvae or ants) or infections caused by bacteria and fungus in the clutch. The whole content of the nest was taken out analyzed by separating them into different groups:

Infertile eggs

Living hatchlings

Dead hatchlings

Eggshells

Eggs without emerging evidence

The eggshells were counted representing the amount of live hatchlings emerged from the clutch. If more than 50% of the eggshell remained it was considered as the eggshell of one hatchling. Remaining eggshell pieces were puzzled together to the approximate size of an egg and registered. Pieces of eggshells with evidence of rotten or predated hatchlings or eggshells with a lot of larvae were separated and not considered.

The eggs without emerging evidence were opened and separated into eggs with embryos , eggs without development and eggs with fungus or bacterial infection. Appearing embryos were divided into different states. The states were identified considering the percentage of space the embryo occupied in comparison to the whole egg.

State I	0% to 25% of the egg is occupied by the embryo
State II	25% to 50% of the egg is occupied by the embryo
State III	50% to 75% of the egg is occupied by the embryo
State IV	75% to 100% of the egg is occupied by the embryo

The hatchling success from a nest was calculated using the following formula:

$$\text{Hatchling success} = \frac{(\text{Number of eggshells} - \text{Number of dead hatchlings}) \times 100}{\text{Total Number of fertile eggs}}$$

The total number of fertile eggs was the one counted at the relocation (for relocated and natural hatchery nests) respectively at the nesting event (natural nests). Calculating the hatching success of the hatchery nests the number of eggshells was replaced by the amount of emerged hatchlings counted for that specific nest at the hatchery shift.

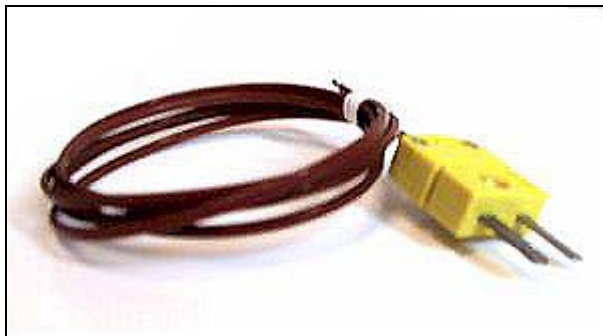
### 5.2.6 Nest Temperatures

On three natural nests, six relocated and six hatchery nests thermocouples (temperature measurement instruments) were installed and the nest temperature was measured during the whole incubation time. The measurements were done every six hours: 0600, 1200, 1800 and 2400. In hatchery nests three thermocouples were installed on nests incubating in the sun three on nests incubating in the artificial shade.

#### Thermocouples



The temperature was measured using 24-gauge Fe-Cn thermocouples (Type J) accurated to  $\pm 0.05^{\circ}\text{C}$ . A thermocouple of type J consists of a thermocouple plug with a ferric (Fe) and a constantan (Cn) contact. A plastic film canister with a cap protects the plug. A thermocouple wire is connected to the contacts of the plug ending in a temperature sensitive point on the other end of the wire, which measures the surrounding temperature.



**Figure 5.11**  
Yellow thermocouple plug with red wire (source: [www.onsetcomp.com](http://www.onsetcomp.com)).



**Figure 5.12**  
Thermocouple covered with a film Canister and fixed on a stick next to the nest (photo by Sue Furler, 2004)

#### Placing of the thermocouples

In natural nests the thermocouples were placed during the egg laying process, in relocated, and hatchery nests the placement was done during the relocation. After the female laid approximately 20 eggs or 20 eggs were put into the relocated nest, the temperature sensitive end of the thermocouple wire was gently put into the center of the clutch with its end pointing upward. While the female finished the egg laying process or the nest was filled with the remaining eggs, the wire was held along the one sidewall of the nest. Enough spare wire was left within the clutch so that a pulling out by the female while covering the nest with sand or by people bumping into them was impossible. The thermocouple plug protected by the film canister at the end of the wire was held until the female covered the nest and finished the nesting process or until the relocation was carried out. Thermocouple wires for natural nests need to be long enough to allow taking a distance to the female covering the nest. The spare wire was wound up on a wooden stick, which was put into the sand close to the nest. The thermocouple plug was fixated on the stick with a tape considering that the cap of the protecting film canister was pointing upwards to avoid saltwater coming into it and affecting the thermocouple contacts. Every thermocouple was marked with an identification tape stuck on the film canister

#### Temperature reading

The daily measurements were done by plugging the thermocouples into a temperature reader (Digi-sense<sup>®</sup>, Thermocouple thermometer, EW-91100-00 (Type J), Cole Parmer Instruments Co, Illinois, USA), which displayed the measurements in degree Celsius (Figure 5.12). The

temperatures were registered on a specific data sheet (see appendix 8.2). In case the display showed “error” the thermocouple plugs were opened with a screwdriver and the were wires cut and reconnected.



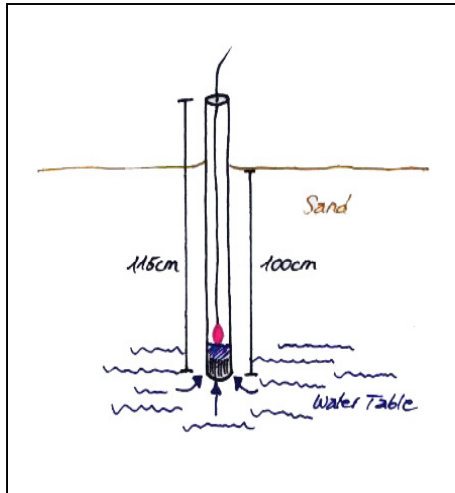
**Figure 5.13**  
Thermocouple reader  
(source: www.coleparmer.com).

#### 5.2.7 Rainfall measurements

On three different locations (ANAI Field station, Hatchery A and Hatchery B) the daily rainfall was recorded. The measurements were taken in  $\text{mm}^3$  per day using a plastic rain gage.

#### 5.2.8 Water table measurements

For measuring the water table three plastic tube of 115 cm length were put 100 cm into the sand thus 15 cm of the tube were visible on the beach (Figure 5.14/5.15). The end of the tube in the ground was closed but covered with little slots for the water to enter, the other end appearing on the beach surface was open. If the water table level in the ground increased because of rain or tidal inundation, the water reached the tube and entered through the slots into the tube. For the exact measurements of the water level in the tube a floating body (plastic egg) was attached to a cord and dropped along the tube until it floated on the water that entered the tube. The distance from the floating body to the end of the tube was measured on the cord. Since the tube is 115 cm long the measured distance was subtracted from 115 cm resulting in the value for level of the water in the tube. As the tube is only 100 cm in the ground the calculated value was subtracted from 100 cm resulting in the level or water table from 100 cm upwards. (Example: Measured distance from the floating body to the end of the tube: 106 cm,  $115 \text{ cm} - 106 \text{ cm} = 9 \text{ cm}$ ,  $100 \text{ cm} - 9 \text{ cm} = 91 \text{ cm}$  → the water table level comes up to 91 cm in the ground). Each sample site (natural, relocated, and hatchery) was equipped with one tube and the area was marked with a colored tape, the water table level was measured daily.



**Figure 5.14, Figure 5.15**

Water table measurement tube. Marked with sticks and colored tape (photo by Sue Furler, 2004).

### 5.2.9 Sand samples

Sand samples of 300g were taken on three natural, three relocated and three hatchery nests at a depth of 75cm. A grain size distribution analysis was made at the Institute of Geology, University of Basel, Switzerland. The sand samples were dehumidified for 24 hours in a drying chamber at a temperature of 105°C. Each sample was weighted and the initial weight registered. The sieving occurred in two steps using an automatic sieving machine. First each sample was sieved continuously during 10-15 minutes through piled sieves with the mesh sizes of 800µm, 630 µm, 500µm, 400µm, 315µm and 250µm, then the remaining sand in the each sieve was weighted. The sand grains smaller than 315µm were sieved again through sieves with mesh sizes of 200µm, 160µm, 125µm, 100µm, 80µm, and 63µm and the remaining sand weighted. For the grain size distribution the percentage weight proportion of the sand left in each sieve was calculated.

### 5.3 Statistical Analysis

The statistical analysis was conducted using the SPSS 12.0 statistical analysis program. One Way Analysis of Variance (ANOVA) was used for statistical comparison of the hatchling

success, the temperatures and sand grain size distribution of the different nests and nesting sites. Associations between nest temperatures and rainfall were tested using a Spearman rank-order correlation. A linear regression tested the relationships between hatchling success and nest depths. A One Way Analysis of Covariance (ANCOVA) was used for statistical comparison of the hatchling success and the nesting site considering the coefficient of variation in nest temperature.

## 6 Results

The results of this study are presented following the order of the data collection. After the presentation of biometric nest data the focus lies on the hatching success of the natural, relocated and hatchery nests, and trends for failing or successful incubation are demonstrated. The environmental conditions influencing the nests are demonstrated in further sections starting with the nest temperatures. The nest temperatures over the incubation are presented for the specific nesting sites separately and a comparison between the sites shows differences and similarities in incubation temperature under consideration of the influence on the hatching success. The influence of the water table level and the surrounding sand on a leatherback sea turtle nest are presented in the last sections of the current chapter.

### 6.1 Biometry of the nests

During the nesting season of 2004 there were a total of 269 nests of leatherback sea turtles recorded on Gandoca Beach. The staff of Asociación ANAI collected comprehensive data on the nesting and migration, the biometry, the nest management, and the nest monitoring (Chacón & Hancock, 2004, see Appendix, Table 8.1). Out of the nests recorded, 45 were left to incubate *in situ* (natural nests), 115 were relocated to other parts of the beach and a total of 99 nests were relocated to one of the two hatcheries (Table 6.1; after Chacón & Hancock, 2004). Out of this universal set specific biometric data was collected for the current study samples (21 natural nests, 30 relocated nests and 30 hatchery nests).

The following table illustrates their differences in shape (average depth and width) and the average number of fertile and infertile eggs for each nest. Nests samples incubating under natural conditions are in the average deeper (74.9 cm) than nest in the hatchery (72.2 cm) or relocated nests (71.3 cm) whereas relocated nests show the greatest average in width (43.3cm) compared to nest of the other nesting sites. All nests sampled for this study were exhumed in order to obtain information on hatching success, caused for reproductive failure and possible environmental factors affecting these parameters (see chapter 4.2).

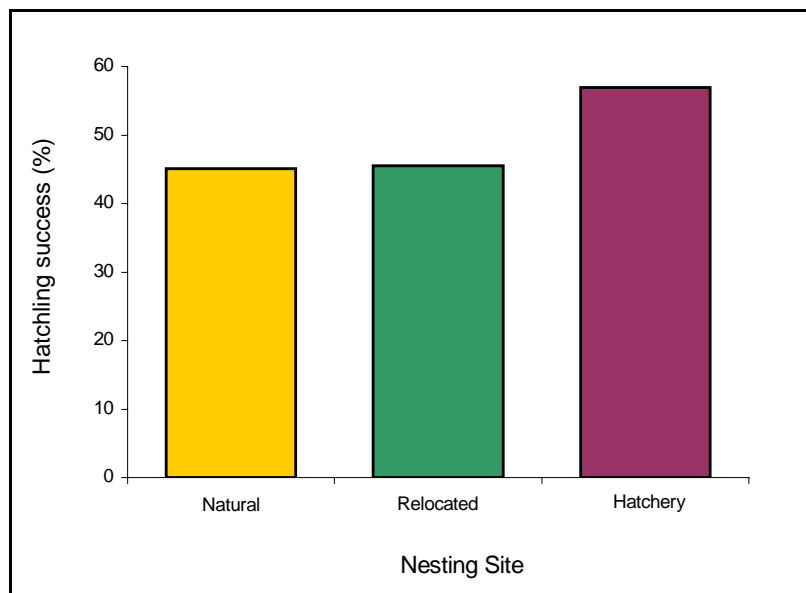
**Table 6.1**  
**Summary of data collection for natural, relocated and hatchery nests during the season of 2004.**

	Natural	Relocated	Hatchery	Total
Total number of nests 2004	45	115	99	269
Average incubation time (days)	64.4	64.3	68.8	65.8
Total exhumations 2004	21	53	99	173
Study samples	21	30	30	81
Average number of fertile eggs/nest	68	77	76	221
Average number of infertile eggs/nest	20	29	36	85
Average depth of nest (cm)	74.9	71.3	72.2	72.8
Average width of nest (cm)	42	43.3	40.6	41.9
Exhumations	21	30	30	81

## 6.2 Hatching success

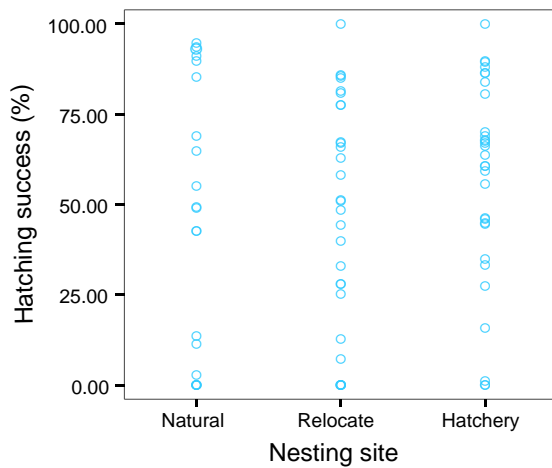
Since stable environmental conditions lead to high hatching success, the success rate of the observed nests gives important information on the environmental conditions on the different nesting site. Temperature, humidity, gas exchange generate the microclimate for successful embryonic development (see chapter 2.2.1). The hatching success was determined by analyzing the nest after the incubation on a specific exhumation. In hatchery nests, all hatchlings were counted bay direct observation.

The average hatching success for the study samples was 49.63% (S.D.= 32.3). The hatching success for sample nests left *in situ* was 45.13% (S.D.= 37.4). The hatching success recorded for relocated sample nests was 45.48% (S.D.= 32.4) and 56.95% (S.D.= 27.8) for hatchery nests sampled for this study (Fig. 5.2). It emanates that the success rate of hatchery nests is slightly higher than for natural and relocated nests.



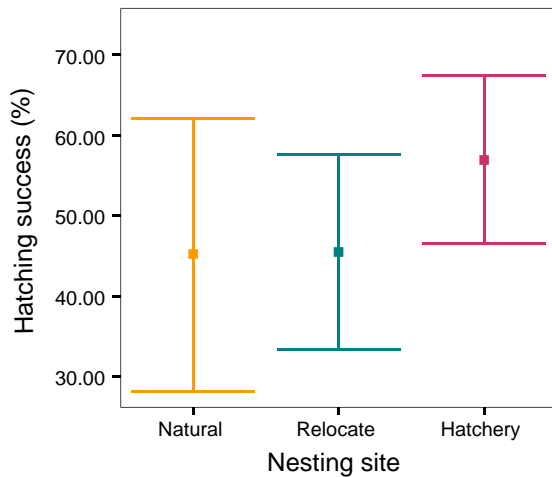
**Figure 6.2**  
Hatching success of natural, relocated and hatchery nests in %

Against the prospects no significant difference in hatching success was found between natural, relocated and hatchery nests (ANOVA,  $df= 2$ ,  $F= 1.225$ ,  $p= 0.299$ ). Natural nests show practically the same hatching success as relocated nests, whereas the success rate of hatchery nests is slightly higher. Focusing on the distribution of the success rates of the specific nests within a nesting site, the following scatter plot illustrates that the data is evenly distributed over all three nesting sites (Fig. 6.3). The hatching success of relocated and hatchery nests range from 0% to 100% whereas natural nests range from 0% to 96%.



**Figure 6.3**  
Scatter plots of the hatching success rates for natural, relocated and hatchery nesting sites.

The error bars in Figure 6.4 show the mean hatching success of each nesting site and the 95% confidence interval within which the success rates were observed. It emanates that the hatching success rates of relocated and hatchery nests lays within a narrower range than those of natural nests. This finding may be an indicator for slightly different environmental circumstances in the natural nesting site.



**Figure 6.4**  
Error bars showing a 95% confidence interval of the mean hatching success for the different nesting sites.

From some of the nests there were no emergences, thus a 100% mortality rate of the embryos. Beach erosion, tidal inundation, and bacterial infection as well failing embryonic development because of biological reasons can lead to failing incubation. Table 5.4 shows the amount of nests with a 0% hatching rate and the reason for failing incubation. These findings may

indicate that natural nests are more likely to be lost by erosion than relocated and hatchery nests, while relocated nests are more affected by tidal inundation than nests located on other parts of the beach. Mortality of the embryos caused by bacterial or fungal infection occurred only in hatchery nests.

**Table 6.5**

Nests with failing incubation due to biotic and abiotic factors				
	Natural	Relocated	Hatchery	Total
Study samples	21	30	30	81
Nests lost by erosion	3	2	0	5
Flooded nests	2	4	0	6
Nest destroyed by bacteria	0	0	2	2
Total of lost nests	5	6	2	13

### 6.3. Nest temperatures

The temperature inside the nests chamber is influenced by the surrounding climatic conditions. The climatic conditions on a nesting beach are dynamic. It is important to determine the incubation temperature and its changes, as sex in sea turtles is determined by these factors. For leatherback sea turtles, the pivotal temperature for sex determination is 29.5°C (see chapter xx), where below this temperature more males will be produced, and above will result in more females leatherback sea turtles. Besides this, strong oscillations in temperature, resulting from climatic variations may have a strong effect on the development of the sea turtle embryos, thus on the hatching success (see chapter 2.2).

In the first part of this chapter, the summary of the observed temperatures during the month of incubation will be presented. In the second part, the temperatures within the three different nesting sites are presented separately over the whole nesting season. In the third part, a comparison between the mean temperatures of the three sites and the influence of the rainfall will be demonstrated. In the last part, the influence of variation in temperatures on the hatching success of the different nesting sites will be explained.

#### 5.3.1 Nest temperatures during the month of incubation

The nest temperatures were observed in three natural nests, six relocated nests and six hatchery nests. The following table shows the incubation temperatures during the months when most of the nests were incubating (April, May, June and July) compared with the accumulated rainfall over the specific month. The lowest temperatures for all three nesting sites were recorded in May, coinciding with the highest rainfall, and thus indicating that heavy rainfall leads a decrease in nest temperatures. June is the month when the highest temperatures were recorded. Throughout the whole sampling period, the highest standard deviation of the mean incubation temperature at natural nests indicating less stability of incubation temperatures for these nests.



**Table 6.6**

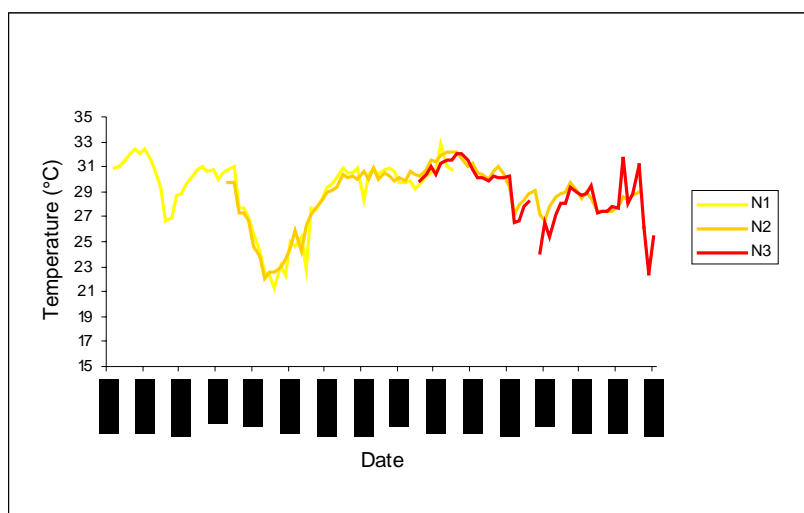
Incubation temperatures and rainfall over the month of April, May, June and July of Natural (N), relocated (R) and hatchery nests (H).

Incubation temperatures	April			May			June			July		
	N	R	H	N	R	H	N	R	H	N	R	H
Mean	30.3	29	28.7	27.3	26.7	25.8	30.2	30.7	30	27.7	30.5	28.6
Maximum	32.4	30.4	30.5	30.9	30.9	29.8	32.1	32.4	31.9	30.2	31.7	30.2
Minimum	26.7	25.4	25.1	21.9	20.6	20.4	26.9	28.3	29.1	22.5	27.9	26.6
S.D.	1.56	1.39	1.42	3.02	3.11	3.19	1.25	1.17	0.67	2.85	0.79	0.18
Accumulated rainfall (mm)	202.68			1203.95			543.7			477.1		
S.D. Rainfall	11.67			38.54			15.22			16.23		

### 6.3.2 Temperatures within the nesting sites

#### Natural nests

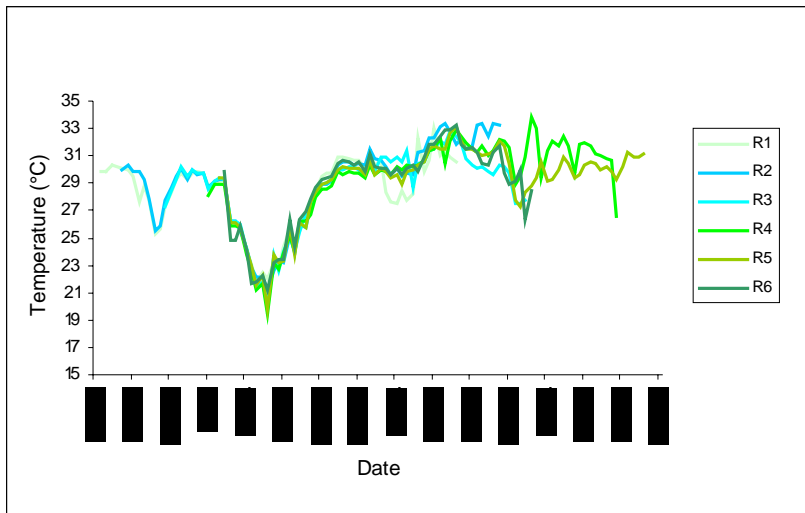
Because of practical restraints, thermocouples could only be placed in three natural nests. Figure 6.7 illustrates that the nests temperatures of all natural nests follow the same trend over the nesting season. This trend is characterized by a sharp decrease of incubation temperature in May, stable temperatures in June, and again irregularities in July were recorded for natural nests. Focusing on the average nest temperature over the incubation time a significant difference was observed between the different nests (ANOVA;  $df= 2$ ,  $F= 26.99$ ,  $p= 0.00$ ). The nest N2 had a significant lower average temperature than N1 and N3, which indicates that the natural nesting site may be more affected by environmental conditions. The nests N1 and N2 did not hatch because the sea flooded them several times during incubation. Nest N3 had a hatching success of 64.62% and was not affected by tidal inundations, as it was located on the upper beach.



**Figure 6.7**  
Daily mean nest temperatures of natural nests during incubation.

### Relocated nests

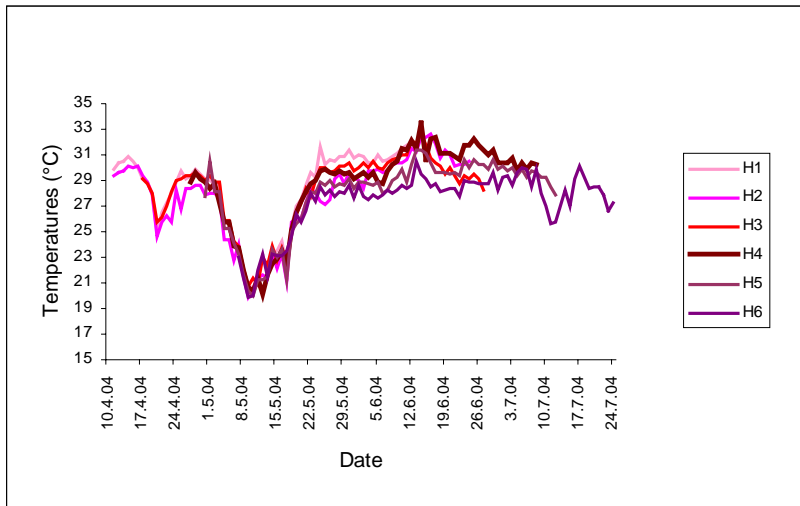
The daily mean temperatures of relocated nests follow the same distributions as natural nests (Fig. 6.8) but in the case of the relocated nests there was no significant difference found in their average temperatures over the incubation period (ANOVA;  $df= 5, F= 1.51, p= 0.185$ ). The hatching success of the observed relocated nests was 65.9% for R1, 48.3% for R2 and 81.6% for R3. Non or the remaining nests observed (R4, R5 and R6) hatched, since they were affected by inundation.



**Figure 6.8**  
Daily mean temperatures of relocated nests during incubation

### Hatchery nests

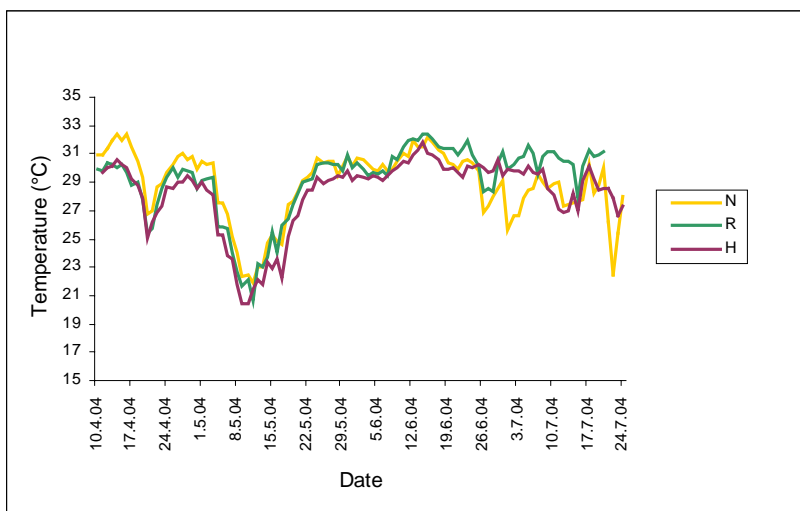
Figure 6.9 illustrates that the daily mean incubation temperatures of hatchery nests follow the same pattern as the natural and relocated nest with decreasing temperatures in April and May. In June and July the nest temperatures differ among each other but the temperature variation within the nest is less than in relocated and hatchery nest. In hatchery nests three thermocouples were installed in nests incubating in the sun and three in the artificial shade. No significant difference was found in the mean temperatures between the different hatchery nests (ANOVA;  $df= 5, F= 0.862, p= 0.506$ ). The hatching success of the observed nests ranges between 27.6% and 89.4%, none of the nests in the hatchery were flooded.



**Figure 6.9**  
Daily mean temperatures of hatchery nests during incubation.

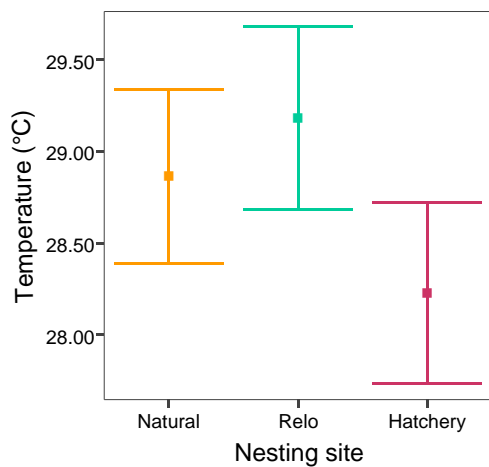
### 6.3.3 Comparison of the temperature over the three nesting sites

The following Figure illustrates the average of the daily mean temperatures of natural, relocated, and hatchery nests over the nesting season. The average temperatures of all nesting sites first follow the similar patterns with decreasing temperatures in the middle of April and a drastically temperature drop in middle May. In June and July the temperatures among the different sites are unstable. It emanates that natural nests underlie strong fluctuations in the second part of the season. Hatchery nests show lower temperatures throughout the whole season, middle temperatures were recorded for relocated nest in the first part of the season and increasing temperature rates in the second part compared to the other nesting sites.



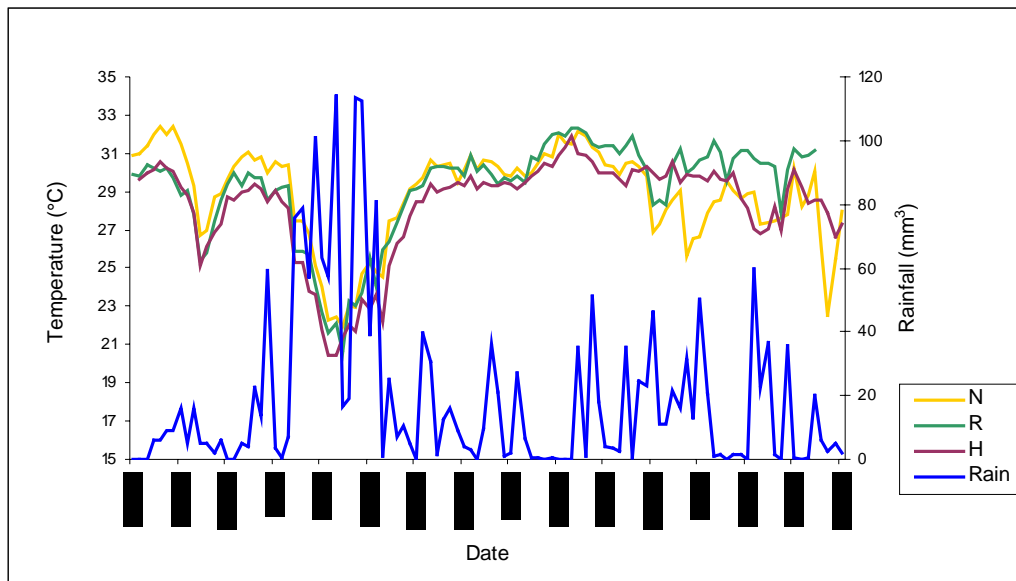
**Figure 6.10**  
Mean nest temperatures for natural (N), relocated (R) and hatchery nests (H) over the nesting season.

The diagram of the error bars clarifies the difference in the mean temperatures of the three nesting sites. Although the mean temperatures range between 28°C and 29.5°C, a One Way ANOVA indicated that there is a statistically significant difference between the three sites (df= 2, F= 3.84, p= 0.22). This shows that hatchery nests have a lower average temperature than natural and relocated nests. The variance of the temperatures appears to be similar over the nesting sites.



**Figure 6.11**  
Mean nest temperatures with 95% confidence interval for natural, relocated and hatchery nests.

Compared with the average daily rainfall over the nesting season (measured in mm<sup>3</sup> per day on three different locations) it states that there is a significant correlation with the average daily mean nest temperatures of the three different nesting sites and the rainfall (Pearson;  $r = -0.586$ ,  $p = 0.00$ ). Figure 5.11 illustrates the decreasing temperatures with increasing rainfall over the nesting season.

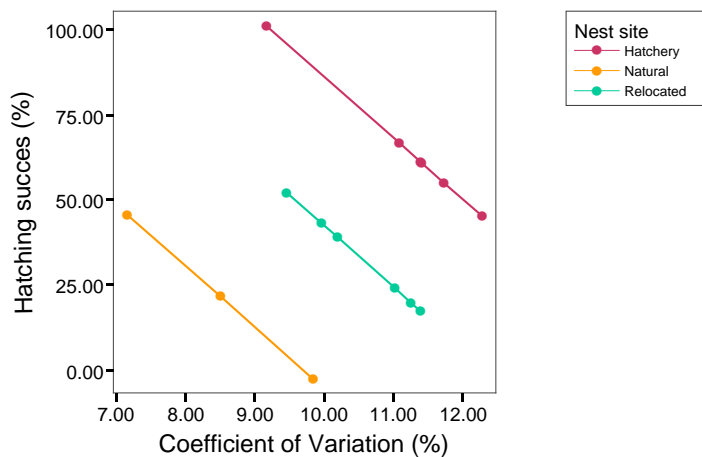


**Figure 6.12**

Mean nest temperatures for natural (N), relocated (R) and hatchery nests (H) compared with the daily rainfall over the nesting season.

#### 6.3.4 Effect of changing temperatures on the hatching success

Strong variations in temperature of a nest can influence the hatching success. The extent of temperature variations in a nest chamber can be estimated by calculating the coefficient of variation of the daily mean incubation temperature (Standard deviation of the temperature measurements in percentage of the mean temperature). The hatching success rates of the referring nests will indicate the extent to which variations in incubation temperature can influence development of sea turtle embryos and their survival. For all nests of the different nesting sites equipped with a thermocouple (three natural, six relocated and six hatchery nests) an ANCOVA indicated that there is statistically significant difference between the hatching success of the three nesting sites once the effect of the coefficient of variation in temperature is accounted for (ANCOVA;  $df= 2$ ,  $F= 5.12$ ,  $p= 0.027$ ). This adjusted model states that with higher variance of the mean incubation temperature the hatching success decreases in all of the three nesting sites and that hatchery nests have higher success rates than relocated and natural nests (Fig. 5.12).

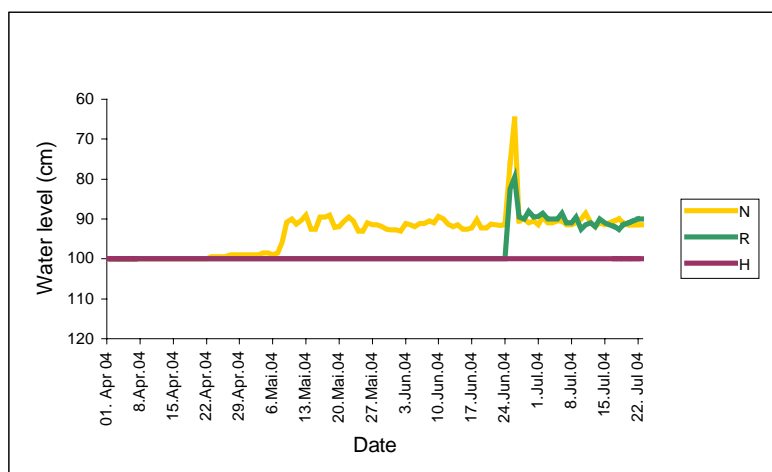


**Figure 6.13**  
Coefficient of variation of the nest temperature compared with the hatching success for natural, relocated and hatchery nests.

#### 6.4 Water table

The distance between the groundwater table and the nests has an important influence on the water exchange of the clutch. In times of persistent rain or tidal flooding, the water table level increases and can reach the nests. Humidity influences the embryonic development of the eggs and can lead to increasing hatchling mortality, if persistent.

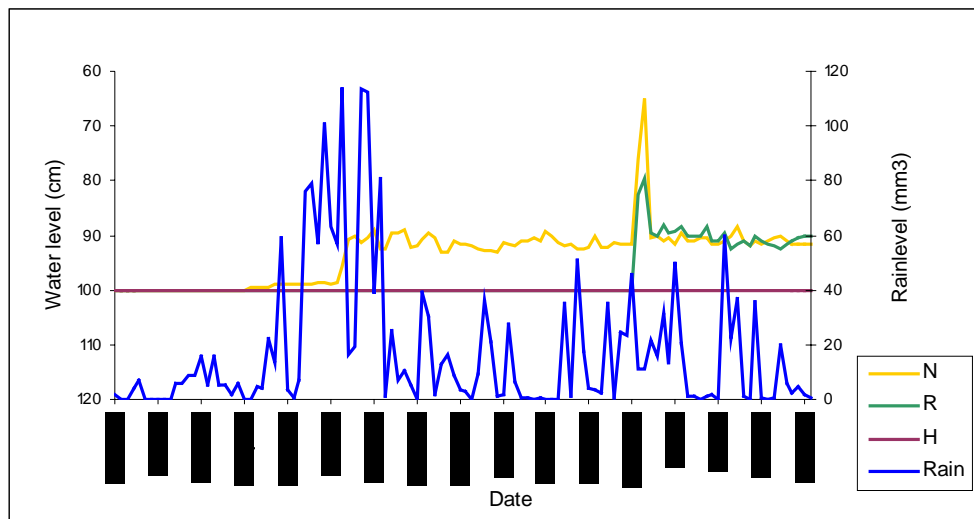
Figure 6.14 illustrates the water table level of the three nesting sites. The reference point is at a depth of 100cm. If the water table level exceeded 100cm in the ground, the specific lines for each nesting site signal it. Natural unmanipulated nests were affected by the groundwater almost during the whole season (Fig. 5.13, N). The water exceeded 100 cm from late April on and came up to 65 cm towards the surface in late June. Water table was recorded in relocated nests from late June until the end of the season with a peak depth of 79.5 cm (Fig.6.14, R). Over the whole incubation period, the groundwater table did not exceed 100 cm in the hatchery area (Fig. 6.14, H).



**Figure 6.14**

Water table level measured from 100cm up to the surface for natural area, relocated area and hatchery area.

Compared with the measurements of the rainfall measurements it appears that after a period of heavy rain in May the water table level in the natural nesting site constantly exceeded 100 cm (Fig. 6.15). The water table level on the relocated nesting site increased at the end of June but did not appear to be influenced by the rainfall. Hatchery nests were not affected by increasing water table level due to increasing rainfall.



**Figure 6.15**  
Water table level of natural, relocated, and hatchery nests compared with the rainfall over the nesting season.

Depending on the depth of a nest, the incubation conditions and subsequent development of the embryos may be affected by the water table level. The nest depth of observed natural nests ranges from 63 cm to 85 cm whereas relocated nests depths range from 45 cm to 89 cm and observed hatchery nests from 56 cm to 86 cm. There was no significant difference observed between the nest depths of the different nesting sites (ANOVA;  $df= 2$ ,  $F= 1.92$ ,  $p= 0.153$ ). Even if the hatching success of the different nesting sites is accounted for, the results obtained indicated that there is no significant relationship found between nest depths and the success rates at the three different nesting sites (ANCOVA;  $df= 2$ ,  $F= 1.22$ ,  $p= 0.29$ ).

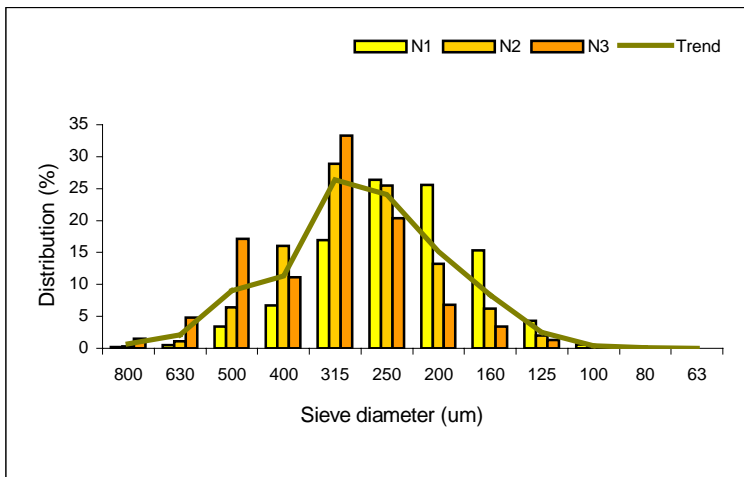
#### 4.5 Sand grain size distribution

The grain size of the sand is important for the successful incubation of the eggs and the development of the embryos. An optimal grain size allows optimal gas exchange within the clutch (see chapter 2.4) and contributes to stable environmental conditions.

Three samples of sand were taken at each nesting site at a depth of 75 cm and analyzed for mean grain size and distribution trends using sieves with different mesh sizes. The size of the sieve indicates the range of the sand grain size found within the sieve. The effectual grain size found in one sieve ranges between the next smaller and the next bigger sieve. (Example: Sand grains found in a sieve of 315  $\mu\text{m}$  diameter are bigger than 400  $\mu\text{m}$  but smaller than 250  $\mu\text{m}$ ).

### Natural Nests

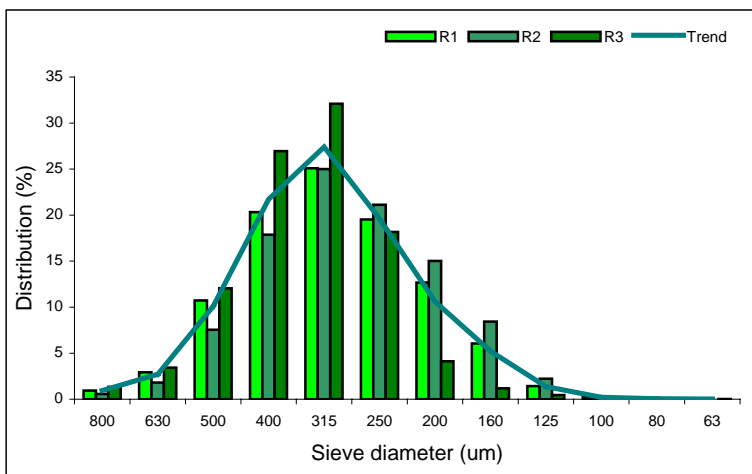
In two of three natural nests the main sand grains were found at a sieve size of 315  $\mu\text{m}$  (28.8% in N2 and 26.3% in N3). For one sample (N1), the main sand grains were observed at a sieve size of 250  $\mu\text{m}$  with 26.3%. The trend line shows the average grain size of all three samples peaking with 26.3% at 315  $\mu\text{m}$ . It emanates that there are differences in the grain size distribution within the three natural nests. N1 has more small grains than coarse grains and for N3 more coarse grains than small grains were recorded, whereas the grain sizes of N2 lies between the other two natural nests.



**Figure 6.16**  
Distribution of sand grain size for three samples of natural nests (N1, N2 and N3).

### Relocated Nests

The sand samples of the relocated nests show similar distribution as the natural nests. All samples have their main grain size at a sieve size of 315  $\mu\text{m}$  (25.0% in R1, 24.9% in R2 and 32.0% in R3). The average major grain size shown by the trend line also lays at 315  $\mu\text{m}$  with 27.3%. In relocated nests the distribution of the sand grains is more evenly, they all have similar ratios of coarse gain whereas R3 shows lower amounts of fine grains than the other relocated nests.



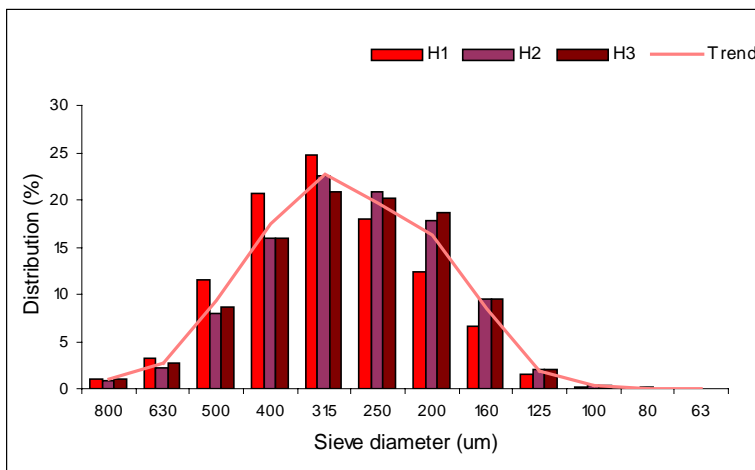


**Figure 6.17**

Distribution of sand grain sizes for three samples of relocated nests.

### Hatchery Nests

The sand grains of hatchery nests show a more even distribution in size within the three samples (Fig 6.18). The major grain size was found at the sieve size of 315  $\mu\text{m}$  (24.7% in H1, 22.5% in H2, and 20.7% in H3). In hatchery nests, the average major grain size is also at 315  $\mu\text{m}$  but with the lower percentage rate of 22.6%. For hatchery nests it states that the differences in grain size distributions over the three nests is are small. They all have similar amounts of coarse gains and fine grains within the nests.



**Figure 6.18**

Distribution of sand grain sizes for three samples of hatchery nests (H1, H2 and H3).

All three nesting sites show a normal distribution of the grain sizes. There is no significant difference in the average grain size distribution between the different nesting sites (ANOVA;  $df= 2, F= 0, p= 1$ ).

## IV Discussion

### 7 Hatching success

#### 7.1 Natural nests

In our study, the hatching success of nests incubating under natural conditions was 45.13%. These results are similar to previously recorded data by Eckert & Eckert (1990), who reported a hatching success for St. Croix leatherback nests of 40.2% in 1982, 48.9% in 1983, 55.5% in 1984 and 53.2% in 1985. Hirth & Orgen (1987) reported a success rate of 70.2% for natural leatherback nests in Jalova, Costa Rica. Leslie et al. (1996) recorded in Tortuguero a hatching success of 46.6% in 1990 and 56.7% in 1991. Alexander et al. (2002) reported on Sandy Point, St. Croix, U.S. Virgin Islands a hatching success of 57.5% for natural nests. For the nesting season of 2004 Chacón & Hancock, reported a hatching success of 54.9% for all excavated natural nests (not including nests lost by erosion with a 0% success rate). Three of our natural nests were lost by erosion and two of them had a 0% success rate because of tidal inundation. Leslie et al. (1996) reported the loss of five natural nests in 1990 and 20 nests in 1991 due to tidal inundation in Tortuguero. Alexander et al. (2002) lost 14 natural nests to erosion on U.S Virgin Island.

The hatching success for natural nests resulting from our study is largely comparable to the success recorded by Eckert & Eckert (1990) and Chacón & Hancock (2004). The hatching success for natural nests of our study samples appears diminished compared to some of the other studies that did not consider a 0% success rate for nests lost by tidal inundation or beach erosion. Unlike in our study, they did not include them into the sample set. The amount of natural nests on Gandoca Beach lost by erosion or inundation refers to what was found by Leslie et al. (1996) and Alexander et al. (2002).

Natural leatherback sea turtle nests are exposed to natural beach dynamics which differ at every beach and throughout the nesting season. Gandoca beach is a very dynamic beach, strongly changing due to long shore currents, storm waves, and high spring tides, depending on the climatic circumstances throughout the nesting season. Thus, most of the nests need to be relocated to safer beach areas or to the hatchery. A hatching success of 45.1% for natural nest is an adequate success rate for nests incubating under such unstable beach conditions, particularly if relatively low hatching success of this species compared to other sea turtle species is taken in account (Bell et al., 2003). The loss of five nests out of 21 nest samples (19%) provides an insight into the decision over the final destination of the nest as a conservation tool. For 19% of the natural nests, the decision about leaving them incubating *in situ* did not lead to a successful incubation. Taken into account that all translocated nests (30 relocated nests and 30 hatchery nests) would have had a failing incubation if they were left under natural conditions, it emanates that an estimation of over 80% of the total sample nests would have been effectively lost by beach erosion or tidal inundation. Furthermore, considering that the beach dynamics of Gandoca Beach are very difficult to pre-estimate, the present results indicate an effective conservation strategy to all intents and purposes but still leaves space for further conservation recommendations.

## 7.2 Relocated nests

The nests relocated to a safe beach area exhibit a hatching success of 45.4%. Whitmore & Dutton (1985) recorded a hatching success of 68.7% for relocated leatherback sea turtle nests in Suriname, whereas Alexander et al. observed a hatching success of 46.9% for relocated nests on Sandy Point, St. Croix, U.S. Virgin Islands in 2002. Chacón & Machado, recorded a 52.8% success rate of nests relocated on Gandoca Beach during the 2003 nesting season, whereas in 2004 (Chacón & Hancock) the observed hatching success for all relocated nests was 58.9% (excluding the nests lost by beach erosion). Out of 30 nests, two nests were lost to erosion and four nests were lost because of tidal inundation. Leslie et al. (2002) recorded between zero and three nests lost by erosion in the nesting seasons of 1982 through 2002 on the U.S. Virgin Islands.

The results of our study are similar to those found by Leslie et al. (2002) but it emanates that the success rate for our relocated study sample nests is lower compared to the other studies. Again it has to be taken into account that most of the present studies did not include nests lost by erosion with a 0% success rate. Still, the success of our relocated sample nests appears relatively low. Leslie et al. (2002) found similar amounts of nests lost by erosion. Bilinski et al. (2001) found that flooding of nests was certainly major cause of embryonic mortality in the years of 1997-1998 at Parque Nacional Marino Las Baulas, Costa Rica.

Explanations for the low hatching success for relocated sample nests on Gandoca Beach could be found in the relocation management, the environmental conditions or in biological circumstances. According to Wynecken et al. (1988) egg relocation is an effective conservation method, provided that sites are chosen carefully. The decision about the relocation area did not lead to successful incubation for 20% of the nests, which appears relatively high. Three of the nests lost by tidal inundation were relocated at the same beach area near the hatchery, which was considered to be a safe beach area. The rest of the relocated nests were distributed on different beach areas following the recommendations over where the safe beach areas are considering the specific climatic circumstances and beach conditions. Again it is very difficult to give recommendation over the Gandoca Beach dynamics and it needs a lot of experience and knowledge to make a decision over where the beach is considered to be safe in order to provide successful incubation of the eggs. A low success rate because of egg handling is not likely to be the reason for low hatching success of the relocated nests. Parameter (1980) recorded that eggs can be handled within a 12-hour period without reducing the hatching success induced by the detachment of embryonic membranes from the shell. Bell et al. (2003) recorded that handling of eggs in early stage of development was not likely the cause of death because the embryo has not yet attached to the eggshell membrane. If the handling of the eggs of the our study samples would have harmed them, a clear negative effect on the hatching success would have been recorded. Environmental conditions on beach areas where most of the nests were relocated to could have had a negative influence on their hatching success and will be further discussed in chapter 9. However, under assumption that the hatching success of the relocated nests would have been zero if they were not relocated, the relocation of these nests to other beach areas was an effective conservation strategy.

### 7.3 Hatchery nests

The hatching success for nests incubating in the hatchery was 56.9%. Alvarado et al. (1989) found a hatching success of 69.9% for hatchery nests in Mexico. Chacón & Machado (2003) recorded a success of 67.1% and 61.6% in 2004 (Chacón & Hancock) on Gandoca Beach. One of our hatchery sample nests was infected by bacteria and showed a 0% success rate. Wyneken et al. (1988) found that microbial pathogens can inflict egg failure

The results of our study show a slightly lower success rate as recorded for other nesting sites, but correspond to what was found in the nesting season of 2004 for all hatchery nests on Gandoca Beach.

The hatchery area is protected from tidal inundation and predation. On Gandoca Beach, the hatchery sand was sieved and exempt from wood, roots and other debris. This may have a positive influence on the incubation of the eggs. Nests were only brought to the hatchery in periods of high human presence on the beach or unstable climatic circumstances. Together this provides stable environmental conditions in the hatchery which should provide ideal conditions for successful embryonic development and hatching success. A success of 56.9% is an adequate hatching success for a hatchery. Considering that all these nests would have been lost if they were not relocated, the hatchery management of ANAI is a successful conservation practice. Relocation of marine turtle eggs to protected hatcheries is a common conservation practice to reduce embryo and hatchling mortality and increase hatchling recruitment (Marcovaldi et al., 1996). In areas where the beach conditions are unstable, which is the case on Gandoca Beach, a hatchery is a necessary and usually very successful conservation tool. Nonetheless, the use of open-air hatcheries has been criticized because of its possible influence on the sex ratio and their undisturbed development. The influence of the hatchery circumstances on the sex ratio of the current study samples will be discussed in chapter 9.1.3. Nests in crowded hatcheries are likely to influence the gas exchange among each other and this can lead to bacterial infection. In our study samples, just one of the 30 sample nests had failing incubation because of bacterial affection, this is not likely due to crowded hatchery circumstances. The ANAI hatchery management provides enough space between the nests so as to avoid negative interactions among them.

### 7.4 Comparison of the nesting sites

The hatching success recorded for natural nests was similar to the success rate of nests relocated to safe beach areas. Nests incubating in the hatchery showed a higher success rate compared to relocated and natural nests but there was no significant difference recorded between the three nesting sites. Whitmore & Dutton (1985) reported in Suriname a higher hatching success for relocated leatherback nests than for nests incubating under original conditions (natural nests). Alexander et al., (2002) reported on Sandy Point, St. Croix, U.S. Virgin Islands that the hatching success of relocated nests was significantly lower than for nests *in situ* (natural nests). Chacon & Machado reported in 2003 a higher hatching success for hatchery nests than for relocated and natural nests on Gandoca Beach representing the same trend over the past years.

The results of our study largely correspond to what was found by Chacón et al., in the past nesting seasons on Gandoca Beach. Compared to the hatching success for the total number of natural nests (52.91%), relocated nests (57.21%), and hatchery nests (61, 34%) recorded in the season of 2004 (Chacón & Hancock, 2004) it emanates that the success rate for our study samples follows similar proportions.

Relocated nests showed a similar hatching success as natural nests, indicating that the handling of the eggs, the reconstruction of the nests, and the new nest site selection did not affect their hatching success negatively. The fact that some of the relocated nests were still affected by tidal inundation and that their success rate was relatively low can be due to problems in nest site selection or differing environmental conditions on relocated nesting sites. A slightly higher hatching success for hatchery nest states for better environmental conditions providing successful incubation of the eggs. The decision over the final destination of the nests were favorable in most of the cases, thus the relocation conservation management provided by Acociación ANAI appears to be successful compared to nesting beaches with failing incubation in relocated or hatchery nests. The success rate ranges of natural nests was wider than for relocated and hatchery nests which may be an indicator for differing environmental conditions within the natural nests. Differing environmental circumstances on natural, relocated, or hatchery nesting sites provide different success rates and give a profound insight on reasons for successful or failing incubation of leatherback sea turtle eggs. The possible influence of the temperature, the level of water table, and the composition of the sand grains on the hatching success will be discussed in the following chapter.

## 8 Influence of environmental conditions

Successful incubation of leatherback sea turtle eggs largely depends on adequate environmental conditions. The observation of beach sand temperatures, the level of water table referring to the nests, and the composition of the surrounding sand may give an insight into these conditions and provide an explanation for low or high hatching success recorded for each nesting site.

### 8.1 Nest temperatures

#### 8.1.1 Nest temperatures over incubation

In May lowest temperatures were recorded for natural, relocated, and hatchery nests. The nest temperatures reached highest readings in June. Lowest precipitation was recorded in April, whereas highest rainfall occurred in May. In April, June, and July highest deviations from the mean nest temperatures were observed for natural nests.

Leslie et al. (1996) recorded a distinct decrease in sand temperatures in May due to rain in 1991 at Tortuguero, Costa Rica. Chacón & Machado (2003) recorded lowest and highest temperatures in May, which was also the month with highest precipitation.

Usually there are two periods of low temperatures and high precipitation at Gandoca Beach: February-March and May-July (Chacón et al., 1996). During the nesting season of 2004, precipitation in May was twice as high as in the nesting season of 2003. This provided drastically decreased temperatures in all of our study samples nests. Furthermore, relatively high amounts of rain in June and July compared to the nesting season of 2003 indicate that there were exceptionally wet conditions throughout the entire nesting season. The fluctuations observed in natural nests may be an indicator for unstable environmental conditions on areas where natural nests were incubating. Compared to nests in the protected hatchery, natural nests on Gandoca Beach are more likely to be affected by changing climatic conditions.

#### 8.1.2 Temperatures within the nesting sites

A significant difference in the average nest temperatures of natural nests was recorded, whereas relocated and hatchery nests showed similar nest temperatures within the site over the incubation period. Temperature fluctuations within natural and relocated nests were recorded in the second part of the season. There was a failing incubation observed in some of the natural and relocated nests; successful incubation was recorded for all hatchery nests equipped with a thermocouple.

It is difficult to find comparable data from other nesting beaches where the temperatures of three different nesting sites were observed. Leslie et al. 1996 found temperature fluctuations due to rainfall and cloud cover in natural nests of Tortuguero in 1990 and 1992.

Within natural nests high fluctuations in the nest temperatures were especially recorded in the second part of the season. The specific nest incubating in this period (N3) showed a success rate of 64.6%, thus it is not likely that these fluctuations affected the success rate negatively. Significantly lower temperatures and a failing incubation were recorded for N2 whereas N1 showed low variations in temperature but still a 0% hatching success. It appears that differences or fluctuations in temperature do not necessarily have a negative impact on the hatching success. In the case of our natural nests, failing incubation was more likely due to inundation of the nests. In relocated nests similar trends have been found. In the second part of the season clear fluctuations in temperatures were recorded. Although there were no significant differences in the temperatures, three of the nests failed in incubation. Again, this is a result of environmental disturbance such as tidal inundation or affection by the ground water table which will be discussed in detail in chapter 9.2. Hatchery nests did not show temperature fluctuations in the same extent as natural and relocated nests. All of the observed hatchery nests had a successful incubation (success rates between 27% and 89%), which clarifies the observation below.

#### 8.1.3 Comparison of the nesting sites

The temperatures of the different nesting sites all follow similar patterns beginning with a drop in the middle of April, followed by a distinct decrease in the middle of May. In June and July, clear differences were recorded between all the different sites. The mean incubation temperatures differed significantly between the three nesting sites. For hatchery nests lower

mean temperatures were recorded, than for relocated and natural nests. The temperatures strongly correlated with the rainfall recorded during the nesting season.

As aforementioned, the pattern of nest temperatures follows climatic conditions throughout a nesting season. Chacón & Machado (2003) recorded similar incubation temperatures in the nesting season of 2003 but with higher temperatures in April and May compared to the season of 2004. The rainfall in 2003 was less intensive throughout all the month of nesting. In 1990 and 1991 Leslie et al. (1996) recorded profound cooling effect on sand temperatures due to rainfall Tortuguero, Costa Rica.

On Gandoca beach periods, of low temperatures and high precipitation directly influence the incubation temperature and thus the sex ratio of the hatchlings (Chacón, 1999). For leatherback sea turtles the critical temperature is 29.5°C (Mrosovsky, 1994), where an equal number of males and females are produced. The mean nest temperature recorded for all our sample nests lies under 29.5°C, but the temperatures recorded for hatchery nests were significantly lower than for natural and relocated nests. Heavy rainfall lead to relatively low temperatures throughout the nesting season of 2004, which may have produced male hatchlings in most of the nests that were object of our study. Sex determination occurs in the second third of incubation and the temperatures reached during this critical period decide over the sex of the hatchlings. Especially nests that reached this important period in the month of May produced only male hatchlings. Mean incubation temperatures of 28°C recorded in the hatchery may have mainly lead to male leatherback hatchling. When using a hatchery as part of conservation the predominant production of one sex is not optimal. It has to be taken into account that the sex ratio of leatherback sea turtles varies with location, season, and year. It may be that the presumed long life span of this species allows the sex ratio to balance out over many years (Mrosovsky, 1994). In an artificial hatchery, metabolic heating can alter sex ratio of the hatchlings and influence the natural balance. It is not likely that the hatchery on Gandoca Beach promotes metabolic heating as the hatchery management provides enough space between the nests. As mentioned before, excessive rainfall can influence the sex ratio by lowering the ambient sand temperature which appears to be the reason for a predominant male hatchling production on Gandoca Beach.

#### 8.1.4 Effects of Temperature Variations

Increased variations in nest temperatures varying form the mean nest temperatures lead to a decrease in hatching success. There was a significant difference between the hatching success of the three nesting sites when considering variation in the temperature for each site. The tolerance for temperature fluctuations was higher in hatchery nests than in relocated and natural nests.

To my knowledge, no statistical analysis compared the coefficient of variations in temperature to the recorded success rate on three different nesting sites. According to Ackerman (1997) eggs in nature do not experience constant temperatures. Nest temperatures vary systematically over the course of incubation. However, the development of a sea turtle embryo may be inhibited by high fluctuations in temperature or hydric environment that may lead to deformed

or even undeveloped embryos (Bell et al., 2003). The results of our study show that within each nesting site the hatching success decreases with an increase in temperature variations. The hatching success appears to be less affected by variances in temperature within the hatchery nests than within relocated or natural nests. This may indicate that interactions with other environmental factors such as the moisture or the beach sand composition play an important role in creating the optimal microclimate for the incubating eggs. Considering that the hatching success of hatchery nests is significantly higher than for relocated and natural nests in this model, it is likely that the hatchery is an area with stable beach sand conditions tolerating variances in temperature. Although diurnal temperature changes naturally occur in sea turtle nests, high temperature variations in combination with moisture or unfavorable sand grain size appear to have a negative effect on the gas exchange within the clutch, and therefore on the successful development of the hatchlings. The extent of influence of the water table and the grain size composition of the beach sand on the hatching success will be discussed in the following chapter.

## 8.2 Influence of water table

The groundwater level was exceeding a depth of 100 cm in areas where natural and relocated nests were recorded. In the area of natural nests, the level increased from late April on and reached 65 cm towards the surface. Increasing water table measurements for relocated nests where recorded from late June until the end of the season with a peak of 79.5 cm on the same day as the peak was recorded for natural nests. The water table level never exceeded 100 cm in the ground in hatchery nests. Rainfall seemed to induce increasing water table level for natural nests in the beginning of May when heavy rainfalls were recorded on Gandoca Beach. According to the observed nests depths, some of the nests must have been affected by the groundwater, but there was no significant correlation with the nest depth and the hatching success found.

Leatherback sea turtle eggs are deposited in the sand, a substatum that limits respiratory exchange of gases such as O<sub>2</sub> and CO<sub>2</sub>. The gases are only free to move through the gas filled fractions of the soil (Ackerman, 1997). Respiratory gases are essential for the development of the embryo and they can only exchange through convection or diffusion. Convection on the beach can occur due to vertical movements of the water table caused by tidal pumping, which can induce displacement of the soil air and influence the gas exchange of the incubating eggs. The sea turtle embryo produces heat and carbon dioxide and consumes Oxygen. Thus, oxygen is a limiting factor for successful development of the embryo. Ackerman & Prange (1974) found little or no effect of water table movement on the gas exchange of *Chelonia mydas*, whereas Maloney et al. (1990) recorded that tidal pumping might be an important factor for displacement of soil air.

However, the observations made on Gandoca Beach show that on the natural nesting sites changes in the water table were recorded throughout the whole nesting season. This may have contributed to changing in availability of oxygen and influenced the success rate of natural nests negatively. In the natural nesting area, the water table level might even have flooded some of the nests entirely leading to embryo mortality. According to Ackerman et al. (1974)



the main reason for clutch failure during inundation appears to be impeded gas exchange when the eggs are in a moisture saturated environment. If the water table increases it is likely that chemical substances from decomposing processes deeper in the beach ground reach a nests and affect the eggs. The eggs are then in direct contact with water and such substances that promote putrefaction of the incubating eggs, starting with the decomposition of the eggshell.

Relocated nests were not as affected by a changing groundwater level in the same extent than natural nests. It emanates that in natural and relocated nests temperature fluctuations among the nests occur at the same time as the peak increase of the water level increased in the middle of June). It is more likely that in the second part of the season, the increasing water table contributed to temperature fluctuation in natural and relocated nests than the amount of rain measured during the observed time period. It is not clear in which extent the vertical movements of the water table are a result of tidal pumping or tidal inundation on Gandoca Beach. Nest incubating in the hatchery have neither been affected by tidal pumping nor tidal inundation, which may be again an indicator for stable circumstances in this area. Nonetheless, the composition of the sand in the hatchery may even contribute to a stable nesting environment.

### 8.3 Sand grain analysis

The sand grain size analysis of three nests out of every nesting site reflected that there are no significant differences in the average grain size distribution. They are all normally distributed with a main grain size of 315  $\mu\text{m}$ . Focusing on the grain size observations within a nesting site, it states that there is a small difference in their distribution. This may provide an important factor for the development of the eggs. The percentage of sand grains within a specific size varies along the different natural nests. Some of the natural nests show more coarse grains or more fine grains than others. A similar but alleviated trend was recorded for relocated nests. The grain size distribution of hatchery nests is evenly over along the different nests. Similar amounts of coarse or fine grains were recorded within all hatchery nests.

It is difficult to compare the grain size distribution of three nesting sites within a specific beach to other nesting beaches. Mortimer (1990) recorded that most of the world's major green turtle nesting beaches are characterized by moderately sorted sand with the mean particle diameters ranging from 0.2-1.0 mm (200-1000  $\mu\text{m}$ ). The grain size composition in a clutch is an important factor for gas and water exchange (Ackerman, 1980). Coarse textured sand provides larger interspaces for the respiratory gases and water exchange than fine textured sand. According to Ackerman (1981) a reduction of gas exchange increases incubation duration and decreases hatching success and embryonic growth rate in green turtles (*Chelonia mydas*). The effect of the sand grain sizes and their distribution on the hatching success is not well understood.

For further information on the availability of respiratory gases in the nests on Gandoca Beach, gas sampling devices would have lead to comprehensive results. With the analysis of the grain size distributions, first steps towards a better understanding have been made. In areas where

nests were left natural, the composition of the sand grains appears to be different, which could contribute to differences in oxygen availability and water content in the sand. In times of climatic disturbance (dry or wet conditions), the beach sand might not contribute in every way to balance these effects so that they do not affect the hatching success. Eggs in relocated nests underlie similar conditions but the difference is that there is a possibility to decide over where they are relocated. Thus, the areas with optimal beach sand could be estimated. Due to the fact that the hatchery sand has been sieved and cleaned from any sort of debris, the oxygen consumption and carbon dioxide production during decomposing of this organic material is reduced. This states for more availability of oxygen in hatchery nests providing an adequate environment for the incubating eggs. It is likely that the uniform composition of coarse and fine sand grains in the hatchery provides optimal interspaces between the sand grains. This could balance negative effects of climatic circumstances and the surrounding sand grains may function as a buffer for fluctuating temperatures or high moisture content in the beach sand.

## 9 Shortcomings and weaknesses

### 9.1 Sample size

Compared to previous nesting seasons, a relatively low number of female leatherback turtles nested on Gandoca Beach in the season of 2004 (268 recorded nests). According to Chacon & Hancock (2004), this is not an evidence for the declining of the population, but the result of natural fluctuations due to various environmental circumstances. In the nesting season of 2004, historic numbers of nests were recorded on Playa Chiriquí in Panama (100 km southeast of Gandoca), another important nesting beach for the Caribbean leatherback population. Abiotic factors such as the climatic conditions, beach dynamics and ocean currents influence the turtles choice of preferred nesting beaches. Because of the low nest numbers in Gandoca and the instable beach conditions, the proportion of natural nest was low and the foreseen 30 samples for natural nests were not existent. Thus, a comparison of the hatching success of only 21 (instead of 30) natural nests was necessary. This might have promoted a slight distortion of the results but does not affect the statistical analysis. The same problem occurred with the temperature measurements. Due to few nesting events with nest left natural, the installation of thermocouples was only possible for three nests (instead of the foreseen six). Furthermore, only nests close to the hatchery were equipped with thermocouples because this area was monitored 24 hours and the danger of their loss by curious beach visitors or nightly patrols was minimized. This fact constricted the possibility of thermocouple set even more.

### 9.2 Retrieving the nests

During the data collection, the marking of the nests right after the nesting or relocation was not always possible. Due to bad climatic conditions, bad visibility (new moon), or even due to the fact that I could not attend every single nesting event, some of the nests were marked on the next day. In some cases it was not possible to reconstruct the exact location of the nest, which lead to false marking and after incubation, the nest could not be found anymore. Thus, new nests of the referring nesting site replaced some of the samples, but in every case they were selected before their hatching success was calculated to avoid selection preferences.

### 9.3 Temperature measurements

Thermocouples installed to observe the temperature within the clutch are very sensitive to humidity and salt. Corrosion affected the wires and thereby the thermocouples were often broken and had to be repaired. They were maintained with every temperature reading, but still some of them had to be replaced because the readings were not authentic compared to the readings of other thermocouples. These irregularities might have affected the results of the temperature measurements.

### 9.4 Water table measurements

Because of monitoring reasons, the water table measuring tubes had to be placed close to the hatchery area. The hatchery area is a favorable area to leave nests natural and to relocate nests in danger of disturbances but does not represent the only predominant area for these nesting sites. Thus, it would have been informative to set more tubes on other parts of the beach where natural and relocated nest have been observed. However, water table tubes on beach areas without monitoring are likely to be lost to beach visitors or egg poachers.

## V Conclusions and Recommendations

### 10 Conclusions for sea turtle conservation on Gandoca Beach

In our study about the hatching success of natural, relocated and hatchery nests on Gandoca Beach it is concluded that the interaction of multiple environmental factors influence the incubation of leatherback sea turtle eggs. The surrounding sand provides the first important requirement for the incubating eggs, deciding over the availability of respiratory gases, the gas exchange within the clutch and the moisture conditions. If there are unbalanced conditions in the sand further negative climatic impacts as persistent rain causing changing temperatures, tidal inundation, beach erosion and changing water table levels provide increasing disturbance to the incubating eggs. These disturbance cumulate and their interaction lead to low success rates or failing development of the eggs.

Thus the hatching success largely depends on the local microclimate under which the eggs are incubating and it clearly states that most favorable conditions can be found in the hatchery. The sieved sand provides the basic requirement for balanced temperature and moisture conditions and the artificial protection from tidal inundation, erosion or affection by the groundwater creates a most favorable microclimate for the developing eggs.

Gandoca Beach is a very dynamic and rapid changing beach and the places where eggs are relocated are not guaranteed to be protected from climatic disturbance. The influence of these changing conditions on the hatching success emanates out of our study samples. Even if the beach sand conditions are favorable, the affection by the tide or increasing water table level harm the eggs and prevent their successful development.

Natural nests underlie the similar conditions as relocated nests, the microclimate the eggs are incubating in changes over the nesting site. This may lead to successful as well as failing incubation depending on how strong the interactions of beach sand composition and climatic affections emanate.

The ANAI Sea Turtle Project pursues an excellent conservation strategy only suggesting nests to be relocated when they are in serious danger of failing in incubation because of one of the described disturbances. This strategy allows the comparison of the relocation with *in situ* nests in multiple aspects, which is indispensable for a successful conservation of the nesting population. Due to the fact that Gandoca Beach is very dynamic and underlies rapidly changing climatic conditions the majority of the nests would be lost without human interference, which clearly states for the necessity of the relocation conservation strategy of ANAI.

Considering that the leatherback sea turtles are classified as critically endangered species (UICN Red list of threatened species, 1996) every effort must be made to protect them from extinction. The protection of the eggs during incubation and hatchlings during their first days of life could have a significant effect on overall stability of leatherback populations in the face

of increasing adult mortality (Spotila, et al., 1996). Thus the conservation management provided by ANAI is an effort of major importance to the survival of the caribbean leatherback population.

## 11 Recommendations over relocation strategies

Although the conservation strategies of ANAI lead to successful incubation for most of the nest, the current studies helped to establish detailed information about the nesting environment on Gandoca Beach. Further recommendations can improve the hatching success of the leatherback nests on the specific nesting sites and contribute to the survival of the population.

It is important to sensitize everybody working within the sea turtle project, especially the staff deciding over the relocation areas, for the interactions of the different factors influencing the incubation of the eggs. A specific repetitive training for the staff of ANAI about this particular subject is suggestive and can contribute to the awareness about the conditions of Gandoca Beach, leading to invigorated decision over relocations. The chosen area needs to be and stay safe from tidal inundation, beach erosion or affection by permanent or intermittent streams or rivers. Furthermore the areas with periodic changing of the groundwater level should be estimated and taken in account by deciding over an appropriate relocation area. It is recommended that the beach dynamics are properly observed throughout a nesting season and that any necessary adjustments concerning the relocations be guaranteed. The use of a geographical information system could help to establish a comprehensive map of the risk zones on Gandoca Beach. Zones with high water table levels due to wetlands or other geographical characteristics as well as zones with high risk of tidal inundation or erosion can be estimated and taken into consideration during the relocation process. Further investigations about the beach sand could contribute to improving hatching success on relocated or hatchery nests by including this information into the decision about an appropriate relocation area.

Considering the following guidelines the decision on an appropriate relocation area for leatherback sea turtle nests leads to successful incubation:

- Select a open area with a slope of over 45°
- Select a area without obstacles (wooden debris)
- Select an area which is not influenced by tidal sea water level changes (upper beach)
- Always select places away from permanent or intermittent streams or rivers
- Choose an area without human disturbance (houses, paths)
- Select a place without danger of beach erosion
- Use a map of the geographical characteristics of the beach and avoid places with wetlands or strong variations in water table level
- Select a area without organic material in the beach sand (plant roots, wooden debris)
- Choose an area with uniform sand grain sizes

Considering that the leatherback sea turtles nesting on Gandoca Beach belong to a population inhabiting one of the largest remaining rookeries worldwide, the Caribbean Central American rookery, the conservation efforts in other projects in the Caribbean should be included in a comprehensive conservation management. Further information about the nesting requirements and thus favorable relocation areas can contribute to improving hatching success on other beaches and sustain the stable population tendencies of the Caribbean leatherback population.

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## Appendix

Table 8.1

VARIABLE	VALOR
Período de registro y monitoreo en la playa	1 febrero-1 agosto
Nidos de <i>Dermochelys coriacea</i>	261
Nidos de <i>Eretmochelys imbricata</i>	6
Nidos de <i>Chelonia mydas</i>	1
<b><i>Dermochelys coriacea</i></b>	
<b>MARCAJE</b>	
Total de hembras marcadas con placas MONEL #49	98
Hembras marcadas con placa MONEL #49 durante 2004	53
Hembras marcadas con BIT durante 2004	30
Total de hembras con doble marcaje (placas y PIT)	47
<b>ANIDACIÓN Y REMIGRACIÓN</b>	
Hembras Neófitas	28
Hembras Remigrantes	70
Intervalo de reanidación	9-10 días
Número mínimo de reanidaciones	1
Número máximo de anidaciones	9
<b>BIOMETRÍA</b>	
Longitud Curva estándar del caparazón de las hembras adultas	151,3 cm
Ancho Curvo estándar del caparazón de las hembras adultas	110,2 cm
Longitud Recta estándar del caparazón de neonatos	61,9 mm
Ancho Recto estándar del caparazón de las hembras	42,4 mm
Peso promedio de neonatos	51,4 gr
<b>MANEJO DE NIDOS</b>	
Promedio de huevos normales / nido	79
Promedio de huevos vanos o infértiles / nido	35
Profundidad promedio de nido	71,7 cm
Ancho promedio de nido	41 cm
Proporción de nidos in-situ	19,1%
Proporción de nidos reubicados en la playa	46,1%
Proporción de nidos reubicados a viveros	34,7%
Proporción de nidos saqueados	1%
Proporción de nidos erosionados	4,6%
<b>RESULTADOS NIDOS</b>	
% Supervivencia neonatos en vivero	72,0%
% Supervivencia neonatos en nidos reubicados	58,9%
% Supervivencia neonatos en nidos in-situ	54,9%
Número de neonatos liberados en los viveros	4270
Estimación del total de neonatos nacidos	7246