

# Nest site selection of loggerhead sea turtles: The case of the island of Zakynthos, W Greece

Antonios D. Mazaris<sup>a,\*</sup>, Yiannis G. Matsinos<sup>a</sup>, Dimitris Margaritoulis<sup>b</sup>

<sup>a</sup> Biodiversity Conservation Laboratory, Department of Environmental Studies, University of the Aegean, GR-81100 Mitilene, Greece

<sup>b</sup> ARCHELON, The Sea Turtle Protection Society of Greece, Solomou 57, GR-10432 Athens, Greece

Received 21 June 2005; received in revised form 13 April 2006; accepted 21 April 2006

## Abstract

The sandy beaches of Zakynthos Island support the largest single nesting aggregation in the Mediterranean Region of the endangered loggerhead turtle *Caretta caretta*. The present study attempts to determine possible correlations between a series of habitat variables and nest site selection. Nesting activities, including total and nesting emergences were examined in response to the recorded biotic and abiotic variables. The results of the analysis indicate that beach width is the most critical habitat variable affecting nest site selection. Further analysis of nesting performance implies that sea turtles use multiple environmental cues for nest site selection during the different steps of the nesting processes such as emergence from the surf and nesting. Nevertheless, we caution that a detailed study needs to be conducted over a more extensive period of time to verify these suggestions.

© 2006 Elsevier B.V. All rights reserved.

**Keywords:** CART; Mediterranean; Nest site selection; Nesting activities; Nesting success; Sea turtles; Zakynthos Island

## 1. Introduction

A better understanding of the nesting biology of sea turtles is linked to our ability to evaluate the effect of various human disturbances upon reproductive success, thus improving the efficiency of our conservation plans (Antworth et al., 2006). The position of the nest has a significant effect on the reproduction success of sea turtles. Physical and chemical characteristics of the nest influence hatching success, hatchling emergence success, sex ratio but also embryonic development, fitness and robustness of hatchlings (Packard and Packard, 1988; Horrocks and Scott, 1991; Janzen, 1994;

Resetarits, 1996; Matsuzawa et al., 2002; Zbinden et al., 2006). On the other hand, the position of the nest affects inundation risk of the nest, seaward orientation of hatchlings and the risk of predation for eggs and hatchlings (Flower, 1979; Janzen and Paukstis, 1991; Godfrey and Barreto, 1995; Shine, 1999).

Identification of the possible cues driving nest site selection has received considerable attention (Miller, 1997). Results from a series of studies have identified several physical and chemical parameters associated with nest site location (e.g. width, slope and vegetation cover of the beach, salinity, particle size of the sand, pH, organic content, conductivity, water content, sand temperature) (for a review see Mortimer, 1995; Garmestani et al., 2000). Sea turtles are likely to use multiple environmental factors in nest site selection either by integrating environmental information or by

\* Corresponding author. Tel.: +30 22510 36298; fax: +30 22510 36298.

E-mail address: amzar@enz.aegean.gr (A.D. Mazaris).

using critical thresholds that must be reached for each one of the environmental factors (Wood and Bjorndal, 2000). However, despite the considerable amount of work, our understating of the environmental cues controlling nest site selection is still limited (Miller et al., 2003). The understanding of the role of different cues on nest site selection could be further improved by extending our research, which has hitherto been intensively focused on successful emergences, to include a combination of nesting and non nesting emergences (Miller et al., 2003).

In this study, we examine the importance of a series of biotic and abiotic parameters on nest site selection of loggerhead sea turtles in Zakynthos Island Greece, the largest known population of the species in the Mediterranean. To investigate the effect of the studied parameters on overall nesting process, we analyse the relation of abandoned and successful nesting but also on nesting success, defined as the proportion of emergences resulting in the construction of a nest, for a series of sites.

## 2. Methods

### 2.1. Study area

The Island of Zakynthos is located in Western Greece. At the south-eastern side of the Island, in the Bay of Laganas, six nesting beaches of the loggerheads support the largest documented nesting population in the Mediterranean (Margaritoulis et al., 2003). The location of the nesting beaches within the Bay of Laganas are shown in Fig. 1. Nesting of loggerheads occurs for mid May until the beginning of August.

### 2.2. Data collection

#### 2.2.1. Beach sections

We divided nesting beaches into a total of 91 sections. The upper limit of each section was designed with respect to the natural (i.e. dense vegetation, steep slopes) or human-induced (i.e. wall, hut) structures that bounded the sandy shores. The length of each section was determined based on a preliminary analysis of sand characteristics and a visual examination of the topography and structure of the beach.

#### 2.2.2. Nesting activity

In 2001, from May to August, all emergences of adult females were monitored at each section. Those emergences that have resulted in the construction of a nest were determined. Morning and night surveys were conducted on a daily basis with the collaboration of the

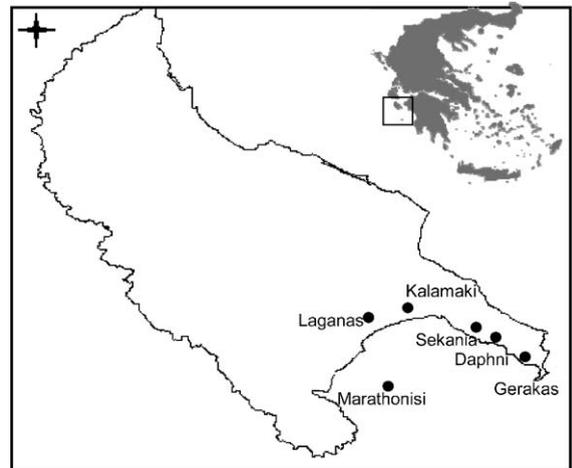


Fig. 1. Map of Zakynthos Island featuring study sites (ordered from west to east: Laganas (~600 m), Gerakas (~600 m) and a beach located at the island of Marathonisi (~360 m) within the gulf of Laganas).

personnel of ARCHELON, the Sea Turtle Protection Society of Greece. The distance of each nest from the sea was determined by using a tape measure. Based on the recorded data, we calculated nesting success at each section as the proportion of emergences resulting to a nest.

#### 2.2.3. Habitat variables

A total of 16 variables were assessed in each section (Table 1). Length (i) was measured as the horizontal distance between the edges of each section. Section width (ii) was calculated as the mean value of the distances between the upper higher water marks (taken by five points) and the objects that backed the beach. Beach slope (iii) was measured with an inclinometer positioned at the higher water mark. Soil samples were collected by various locations within each section by the use of an extemporary soil tube. During sample collection the upper soil zone was removed and samples collected down to a depth of 5–10 cm. Samples collected from the same section were pooled and taken for laboratory analyses. All samples were air dried before performing further analyses as outlined in Allen (1989). Soil texture (iv) was classified by particle size analyses performed by using a net of sieves. Organic content (v) was determined by combusting soil samples in a furnace at 500 °C for 5 h. The loss of weight after combustion was used to determine the organic content of the samples. A pH analysis (vi) was conducted by taking a 20 g oven-dried subsample and mixing it with de-ionized water in a 1:5 mixture. This mixture was stirred for 30 min and left for the same duration in

Table 1  
Variables included in the analysis

Habitat variables
(i) Section length
(ii) Section width
(iii) Slope
(iv) Soil texture
(v) Organic content
(vi) pH
(vii) Sand color
(viii) Clay cover
(ix) Substratum type
(x) Percentage covers of grass/herbaceous
(xi) Percentage covers of shrub
(xii) Percentage covers of tree
(xiii) Percentage of bare area
(xiv) Silhouette of the area backing the beach
(xv) Presence of roads – stream edges
(xvi) Artificial construction/objects

calmness before a pH reading was taken. Sand colour (vii) of air-dried samples was quantified using the [Hurst \(1977\)](#) rating system, which is calculated based on [Munsell colour notation system \(1954\)](#). A white background was used during colour determination.

The existence of thick zones of clay (viii) at the surface of the sand was coded based on 5 categories representing low to high percentages of cover (<5%, 6–10%, 10–30%, 31–50%, <50%). We describe substratum type (ix) of each section as area with sand, mostly sand, half sand/half rock or mostly rock. We used the Braun-Blanquet scale ([Braun-Blanquet, 1965](#)) to describe the vegetation cover at the area that backed the sections; we applied the method four times for describing grass/herbaceous cover (x), shrub (xi) and tree (xii) percent cover or bare area (xiii).

Height of the objects (xiv) backing the beach (e.g. human buildings, trees) was also evaluated by measuring the elevation from the lower to the top of the highest object backing a section by using a inclinometer; measurements were taken while standing in the middle width of each section. The presence of stream edges and roads was also recorded and included in the analysis as dummy variables (xv).

An additional variable was included in the analysis to describe human structures, thus potential disturbances. Buildings and other human objects (xvi) positioned immediately behind the beach (i.e. representing the upper bound of the section) and high structures located within a 10-m zone from the end of the polygons edge were noted by 1, higher constructions of fixtures (such as houses) located within 10–20 m from the upper part of the beach were indexed as 2, lower structures within this zone were grouped as category 3, while distinctive

structures (houses, hotels, etc.) located in longer distances were coded as 4.

Atmospheric climatic variables such as air humidity and temperature were not considered because the studied sites are closely located. Sand temperature and humidity were also not included, because the experimental area was large enough to properly monitor these parameters at each section or even provide some representative values. We should also mention that the varying widths of the sections, but also their topological heterogeneity due to natural material and human placed objects, hindered stratified sampling to account for the above variables (for such an experiment see [Stoneburner and Richardson, 1981](#); [Foote and Sprinkel, 1994](#)).

Data collection was designed to be obtained at three discrete intervals, reflecting initiation, meantime and end of nesting season, at the end of May, mid July and beginning of August, respectively; for each one of these three discrete intervals, a continuous sampling was obtained in an attempt to accomplish sampling of all nesting beaches within limited time difference.

#### 2.2.4. Data analysis

Classification and Regression Trees (CART) analysis ([Breiman et al., 1984](#); [Urban, 2002](#)) was performed at a section level ( $n=91$ ) to investigate whether the recorded habitat variables as predictors account for differences among nesting and total nesting emergences. Each CART was a three-class model, where the three grouping variables were entered as relative abundance indicating low, medium and high values, respectively. Nesting abundance was characterized as low ( $n_L$ ), medium ( $n_M$ ) and high ( $n_H$ ) when the observed nests per section were 0–5, 5–20 and >20, respectively. Similarly, total emergences and nesting success were classified into groups of relative low, medium and high values (for emergences  $e_L$ : 0–20,  $e_M$ : 20–50,  $e_H$ : >50, and for nesting success data  $ns_L$ : 0–0.2,  $ns_M$ : 0.2–0.4,  $ns_H$ : >0.4, respectively). According to our sampling protocol, data were collected at three discrete periods; however, no significant differences were obtained (soil texture, pH, sand colour, organic content), therefore mean values produced by using data from the three sampling intervals were included in the analysis. CART analysis was performed using S-Plus (version 6.2).

### 3. Results

The classification tree for the three classes of nesting data ( $n_L$ ,  $n_M$  and  $n_H$ ) had 14 terminal nodes ([Fig. 2a](#)). Width, organic content, beach slope, length, sand texture, pH, bare ground and grass cover were identified

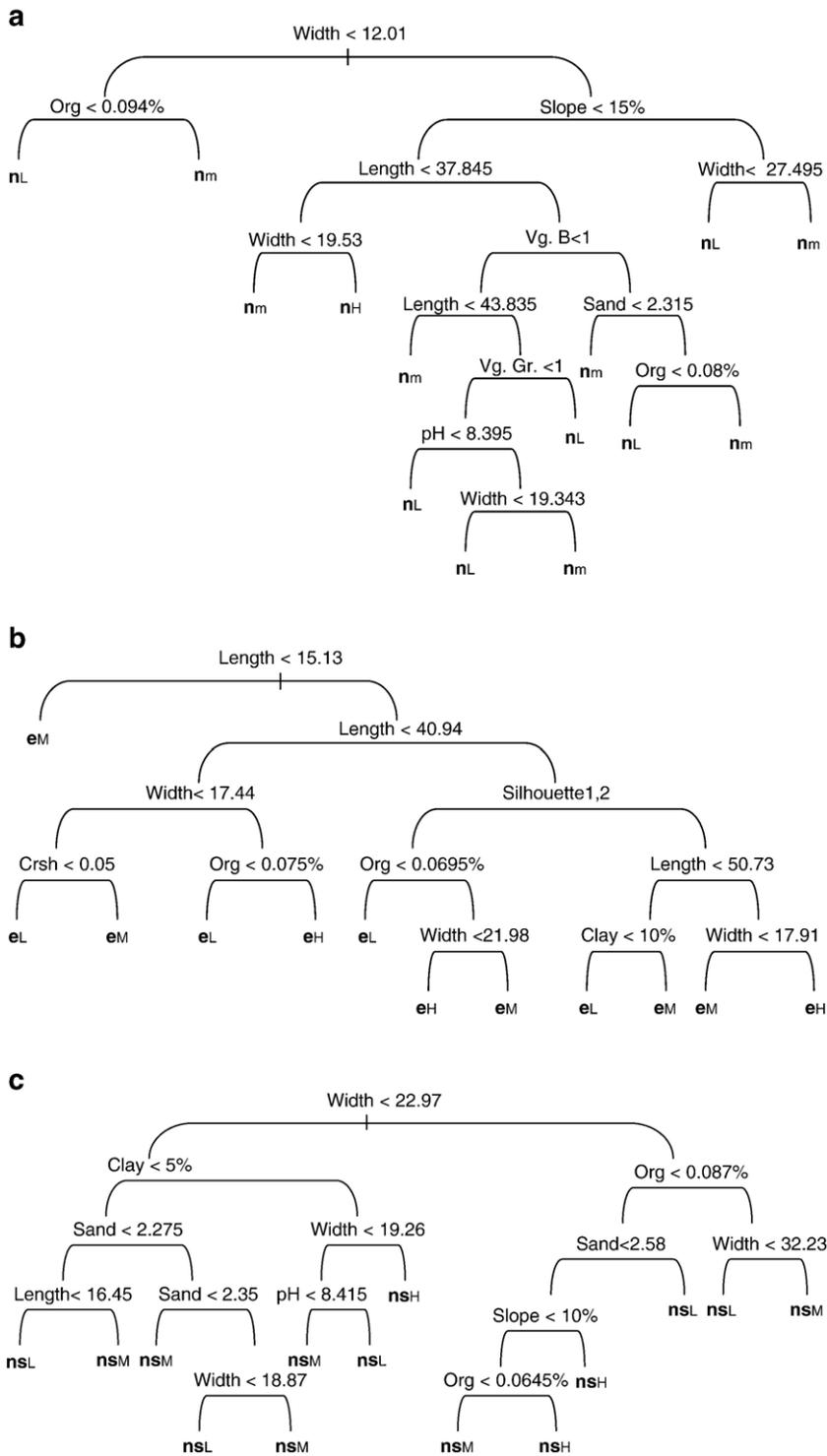


Fig. 2. (a) Classification tree for the three relative groups of nesting emergences indicating low ( $n_L$ ), medium ( $n_M$ ) and high ( $n_H$ ) abundance of nests. (b) Classification tree for the three relative groups of total emergences indicating low ( $e_L$ ), medium ( $e_M$ ) and high ( $e_H$ ) observation of emergences. (c) Classification tree for the three relative groups of nesting success data indicating low ( $e_L$ ), medium ( $e_M$ ) and high ( $e_H$ ) nesting success.

as important variables for the partition of nesting emergences data (misclassification error rate: 0.209). The most important split occurred at the width of 12.01 m. For sectors with lower width (less than 12.01 m) the classification tree showed that low nesting occurred when organic content was lower than 0.094%; under the same conditions medium nesting occurred when organic content was higher than 0.094%. Several nodes were identified for wider sectors (width >12.01 m), with the next significant split occurring on variable slopes (15%). Under these conditions higher nesting abundance occurred at sectors with a length of less than 38 m and width of at least 19.53 m. Other nodes, indicating significant splits, thus identifying explanatory variables, were partitioned according to the percentage of bare ground. Moreover, sand texture and length were recognized to be of moderate importance for data classification.

The classification tree for emergence data ( $e_L$ ,  $e_M$ ,  $e_H$ ) had 12 terminal nodes (Fig. 2b) (misclassification error rate: 0.219). Beach length was recognized as an important explanatory variable partitioning the observations into two groups, with medium abundance of emergences occurring at sectors with a length lower than 15.13 m. For longer sites (length >15.13 m) another division based on length was performed. Under these conditions, the silhouette of the objects that backed the beach and beach width were also important leading to significant splits. High abundance of emergences was then observed at sectors with relatively lower lengths (length <40.94) which were characterized by a width of less than 17.44 m and organic content higher than 0.75. In the same manner, terminal nodes identifying higher emergences also occurred regarding beach width.

The first split in the CART model developed to describe classes of nesting success, occurred at widths of 22.97 m (misclassification error rate: 0.2308) (Fig. 2c). The classification tree identified clay cover and organic content as the next important variables. Sand texture and beach width were further recognized as critical predictor variables. Following the divisions of the tree, beach width, slope and organic contents were recognized as variables distinguishing higher nesting success.

#### 4. Discussion

The results of the analysis performed for nesting emergences and nesting success data, identified beach width as the most important variable. Variables that are associated with the quality of a site on micro scale such as organic content, sand texture and clay layers were found to be important in determining nesting success.

The CART analysis identified the high importance of length into classifying total emergences. Among the other parameters used as predictors for total emergences, width of the beach and silhouette of the objects that backed the beach were found to have a high importance.

The choice of a wider beach provides nesting females with more potential nesting sites in terms of favourable micro-environmental characteristics (Miller et al., 2003). The fact that we identified section length as a variable of high importance in classifying non-nesting emergences could simply reflect an effect due to our sampling protocol (i.e. comparison of sections with different lengths). It has been shown, however, that objects that backed the beach are factors significantly correlated with nesting behavior, which is similar to what has been observed in other loggerhead studies (Salmon et al., 1995). The interpretation of this finding could be that before emergence or at the beginning of the crawl, sea turtles probably use the height of the objects at the upper part of the site as an indicator to evaluate its width. This behaviour could prevent the expenditure of energy resulting from a false crawl.

In conclusion, we demonstrate that environmental variables used to classify each category of nesting activity data (i.e. actual nesting emergences, emergences and nesting success) share some general characteristics that allow their grouping. Habitat variables that were found to better differentiate nesting success appear to represent micro environmental conditions. On the other hand, the habitat variables that are closely associated with emergences could be considered among those that could be detected by an animal even before it begins its nesting process. In a similar way, nesting emergences were better classified by variables associated with the ability of an animal to avoid a false crawl. These results are consistent with the suggestion made by Wood and Bjørndal (2000) that sea turtles use multiple cues for the selection of the nest site. Although we cannot state whether sea turtles use integrated patterns of associations of environmental factors or examine each factor separately, it is clear that they use different environmental cues during different steps of the nesting processes (i.e. emerging from the surf, ascending the beach, nest placement). The use of multiple cues at the different stages of the nesting process links the energetic cost of searching for a nest site to the benefits of selecting a favourable site. Hence, it is possible that nest site selection is a stepwise procedure in terms of the selection of the suitable site; at each step, starting before the emergence from the sea, the females may use the available environmental information to proceed and orient their crawl.

Overall, our results suggest that width of the nesting beach is a strong factor that controls nest site selection. Therefore, this parameter is of high concern for beach management conservation. However, the dynamics of complex systems, such as sandy beaches, should be studied over a more extensive period of time. In this manner, protection measures should be reviewed and studies on the repeatability of the nest site selection could further improve our knowledge and thus our conservation efficiency. On the other hand, there is evidence to support the use of multiple environmental cues at different stages of the nesting process. It will be worthwhile to examine biotic and abiotic factors of the beach to which nesting females are responding during the distinctive stages of the nesting process such as emergence from the surf, ascending of the beach, digging of the egg chamber, and to investigate the patterns of their associations.

### Acknowledgments

We wish to thank all volunteers that have participated in the 2002 Zakynthos Monitoring Project of ARCHELON, the Sea Turtle Protection Society of Greece. The work of A.D.M was partially supported through the HERAKLEITOS research program of the Greek Ministry of Education and Religious Affairs and the European Union. The manuscript was improved by insightful comments of Prof. Chronis Tzedakis and four anonymous referees. [RH]

### References

- Antworth, R.L., Pike, D.A., Stiner, J.C., 2006. Nesting ecology, current status, and conservation of sea turtles on an uninhabited beach in Florida, USA. *Biological Conservation* 130, 10–15.
- Allen, S.E., 1989. *Chemical Analysis of Ecological Materials*, 2nd edn. Blackwell, Oxford, p. 543.
- Braun-Blanquet, J., 1965. *Plant Sociology: The Study of Plant Communities*. Hafner, London, p. 439. Translation by Fuller, G.D., Conard, H.S.
- Breiman, L., Friedman, H.J., Olshen, A.R., Stone, J.C., 1984. *Classification and Regression Trees*. Chapman and Hall, New York.
- Flower, L.E., 1979. Hatchling success and nest predation in the green sea turtle, *Chelonia mydas*, at Tortuguero, Costa Rica. *Ecology* 60, 946–955.
- Foote, J., Sprinkel, J., 1994. Beach compactness as a factor affecting nesting on the west coast of Florida. In: Bjorndal, K.A., Bolten, A.B., Johnson, D.A., Eliazar, P.J. (Eds.), *Proceedings of the Fourteenth Annual Workshop on Sea Turtle Biology and Conservation*. NAOA Technical Memorandum NMFS-SEFCS-351, pp. 217–220. Comps.
- Garmestani, A.S., Percival, H.F., Portier, M.K., Rice, A.G., 2000. Nest-site selection by loggerhead sea turtle in Florida's Ten Thousand Islands. *Journal of Herpetology* 34, 504–510.
- Godfrey, M.H., Barreto, R., 1995. Beach vegetation and seafinding orientation of turtle hatchlings. *Biological Conservation* 74, 29–32.
- Horrocks, J.A., Scott, N.M., 1991. Nest site location and nest success in the hawksbill turtle *Eretmochelys imbricata* in Barbados, West Indies. *Marine Ecology. Progress Series* 69, 1–8.
- Hurst, V.J., 1977. Visual estimation of iron saprolite. *Geological Society of America Bulletin* 88, 174–176.
- Janzen, F.J., 1994. Vegetational cover predicts the sex ratios of hatchling turtles in natural nests. *Ecology* 75, 1593–1599.
- Janzen, F.J., Paukstis, G.L., 1991. Environmental sex determination in reptiles: ecology, evolution, and experimental design. *The Quarterly Review of Biology* 66, 149–179.
- Margaritoulis, D., Argano, R., Baran, I., Bentivegna, F., Bradai, M.N., Caminas, J.A., Casale, P., De Metrio, G., Demetropoulos, A., Gerosa, G., Godley, B.J., Haddoud, D.A., Houghton, J., Laurent, L., Lazar, B., 2003. Loggerhead turtles in the Mediterranean: present knowledge and conservation perspectives. In: Bolten, A., Witherington, B. (Eds.), *Loggerhead Sea Turtle*. Smithsonian Books, Washington D.C., pp. 175–198.
- Matsuzawa, Y., Sato, K., Sakamoto, W., Bjorndal, K.A., 2002. Seasonal fluctuations in sand temperature: effects on the incubation period and mortality of loggerhead sea turtle (*Caretta caretta*) pre-emergent hatchlings in Minabe, Japan. *Marine Biology* 140, 639–646.
- Miller, J.D., 1997. Reproduction in sea turtles. In: Lutz, P.L., Musick, J.A. (Eds.), *The Biology of Sea Turtles*. CRC Press, Boca Raton, pp. 51–81.
- Miller, J.D., Limpus, C.L., Godfrey, M.H., 2003. Nest site selection through hatchling emergence. In: Bolten, A., Witherington, B. (Eds.), *Loggerhead Sea Turtle*. Smithsonian Books, Washington D.C., pp. 125–143.
- Mortimer, J.A., 1995. Factors influencing beach selection by nesting sea turtles. In: Bjorndal, K.A. (Ed.), *Biology and Conservation of Sea Turtles*. Revised edition. Smithsonian Institution Press, Washington, D.C., pp. 45–52.
- Munsell, A.H., 1954. *Munsell Soil Color Charts*. Munsell Color Co., Baltimore, Md.
- Packard, G., Packard, M., 1988. Physiological ecology of reptilian eggs and embryos. In: Gans, C., Huey, R.B. (Eds.), *Biology of the Reptilia. Ecology B. Defense and Life History*, vol. 16. Alan R. Liss, Inc., New York, pp. 523–605.
- Resetarits Jr., W.J., 1996. Oviposition site choice and life history evolution. *American Zoologist* 36, 205–215.
- Salmon, M., Reiners, R., Lavin, C., Wynneken, J., 1995. Behavior of loggerhead sea turtles on an urban beach: I. Correlates of nest placement. *Journal of Herpetology* 29, 560–567.
- Shine, R., 1999. Why is sex determination by nest temperature in many reptiles? *Trends in Ecology and Evolution* 14, 186–189.
- Stoneburner, D.L., Richardson, J.I., 1981. Observations on the role of temperature in loggerhead turtle nest site selection. *Copeia* 238–241.
- Urban, L.D., 2002. Classification and regression trees. In: McCune, B., Grace, B.J. (Eds.), *Analysis of Ecological Communities*. MjM Software Design, Gleneden Beach, Oregon, pp. 222–232.
- Wood, D.W., Bjorndal, K.A., 2000. Relation of temperature, moisture, salinity, and slope to nest site selection in loggerhead sea turtles. *Copeia*, pp. 119–128.
- Zbinden, J.A., Margaritoulis, D., Arlettaz, R., 2006. Metabolic heating in Mediterranean Loggerhead sea turtle clutches. *Journal of Experimental Marine Biology and Ecology* 334, 151–157.