

## Sex ratio estimations of *Chelonia mydas* hatchlings at Samandağ Beach, Turkey

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Received: 12.01.2015 • Accepted/Published Online: 22.02.2016 • Final Version: 09.06.2016

**Abstract:** Samandağ Beach, an important nesting location for endangered green turtles (*Chelonia mydas*), is located at the most eastern part of the Mediterranean in Turkey. Hatchling sex ratio can be one of the key factors for green turtle protection, which is under the threat of global climate change. In this study, sex ratios of hatchlings were estimated by histological examination of dead hatchlings ( $n = 190$ ) and by nest temperatures ( $n = 14$ ) at Samandağ Beach between the 2003 and 2007 nesting seasons. The histological examination of dead *C. mydas* hatchlings and temperature-recorded nests indicated a female-biased sex ratio on Samandağ Beach. By histological examination, top and bottom levels of nests were recorded as 94% and 64% of females. Sex ratios during the middle third of the incubation period ranged from 39% to 97% for females with a mean of 74%. Distance from the sea was correlated with the middle third of incubation temperature ( $r = -0.604$ ,  $P = 0.022$ ) and female ratio ( $r = -0.573$ ,  $P = 0.032$ ). While nest temperatures and nest parameters such as hatching success, clutch size, nest distance from the sea and from vegetation, nest depth, and the day of emergence were not found significantly different among years ( $P > 0.05$ ), incubation duration was found significantly different among years ( $P < 0.05$ ). The results showed a female-dominated sex ratio by both histology and nest temperature. The knowledge of hatchling sex ratios provides information on demographic parameters, and coupled with appropriate conservation measures this can make important contributions to studies of climate change effects on green sea turtle populations.

**Key words:** Sex ratio, sand temperature, *Chelonia mydas*, green turtle, Samandağ

### 1. Introduction

The sex ratio of sea turtle hatchlings is a demographic variable that is particularly vulnerable to environmental conditions, because the sex of sea turtles is determined by their incubation temperature (temperature-dependent sex determination). The relationship between sex and incubation temperature is characterized by a pivotal temperature, where female and male individuals are in equal proportion (Bull, 1980). In most sea turtles, embryos that incubate at high temperatures ( $>29.0$  °C) produce a larger proportion of females, while cooler nests ( $<29.0$  °C) produce a greater proportion of males (Mrosovsky, 1994). Therefore, the intranest temperatures during incubation periods can be used for two of the factors for estimation of overall sex ratio of sea turtles (Yntema and Mrosovsky, 1980; Kaska et al., 1998; Broderick et al., 2000). Incubation duration is related to nest temperature; while a long duration implies low temperatures and more males, a short duration implies high temperature and more females (Mrosovsky et al., 1999). Nest temperature, which varies spatially and/or temporally (Hays et al.,

1999), is determined by natural environmental conditions, and anthropological factors may also be effective factors on determining the nest temperature (Mrosovsky, 1988; Kaska et al., 1998). Therefore, local climatic conditions influence the overall sex ratio of sea turtles and both existing and future populations may be affected by global climate change. Knowing the sex ratio of nesting beaches is important for the future of the species and effective conservation planning, because long-term survival depends on both female and male production (Janzen, 1994a).

Very few studies have estimated hatchling sex ratios of *Chelonia mydas* in Turkey. Kaska et al. (1998) recorded only five *C. mydas* nest temperatures and obtained a female-biased sex ratio in Turkey. A female-biased sex ratio was also estimated by using only sand temperatures on Akyatan Beach, Turkey (Casale et al., 2000). Broderick et al. (2000) also studied the sex ratio of *C. mydas* on Alagadi Beach, Cyprus, and estimated that there was a female-biased sex ratio of hatchlings. They stated that at least 86% to 96% of hatchlings produced from 1993 to

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1998 at this site were females. Kılıç and Candan (2014) also studied green turtles from Turkey, and the estimated sex ratio of hatchlings and dead embryos based on histological examination determined a female bias, as well.

Nesting of green turtles in the Mediterranean is confined to the eastern part of the basin and Samandağ Beach is one of the most important nesting beaches for *C. mydas* in Turkey (Yalçın-Özdilek, 2007), having 20% of the nesting on the Turkish coasts (Türkozan and Kaska, 2010). The number of *C. mydas* nests per season on Samandağ Beach was reported to be between 16 and 621 per year (mean = 241 nests) during the last two decades (Yalçın-Özdilek, 2007; Yalçın-Özdilek and Sönmez, 2011). Since Samandağ Beach is also one of the most important nesting beaches in the Mediterranean, studying sex ratios of hatchlings on these beaches could be important for the estimation of adult populations' sex ratios and ultimate population growth. Despite several studies on the nesting environment of *C. mydas* on Samandağ beach, (Özdilek et al., 2006; Yalçın-Özdilek and Sönmez, 2006; Yalçın-Özdilek et al., 2006, 2007), the sex ratio of *C. mydas* hatchlings is missing from the literature.

The goal of our study was to estimate the sex ratio of hatchlings by gonad histology of dead hatchlings and record temperatures of nests laid by green turtles on Samandağ Beach, Turkey. We tested the correlations between nest temperature, and hence the female ratio, and hatching success, clutch size, nest depth, distance from the sea and vegetation, incubation duration, and hatchling emergence during the 2003–2007 nesting seasons. Based on these results, information is provided that can be used as a basis to study climate change effects on green sea turtle populations.

## 2. Materials and methods

Data were collected from Samandağ Beach (36°07'N, 35°55'E), located in the eastern Mediterranean region of Turkey. Samandağ Beach is about 14 km in length, extending from Çevlik Harbor to the north to Sabca Promontory to the south. The study area corresponded to a 9-km stretch of beach between Çevlik Harbor to the north and the mouth of the Asi River to the south (Figure 1), because this section has the highest nesting activity (Yalçın-Özdilek and Sönmez, 2011). The studied section of the beach was subdivided into 1000-m sectors starting at Çevlik Harbor throughout the south in order to allow easier control of spatial distribution and to indicate the location of the nests. This was to check for the exact positioning of turtle activity and egg chambers by measuring to the nearest post (Öz et al., 2004)

Nest temperature was recorded using Tiny Talk temperature data loggers (Orion Components, Chichester, UK) that were housed in 35-mm film cases. The accuracy

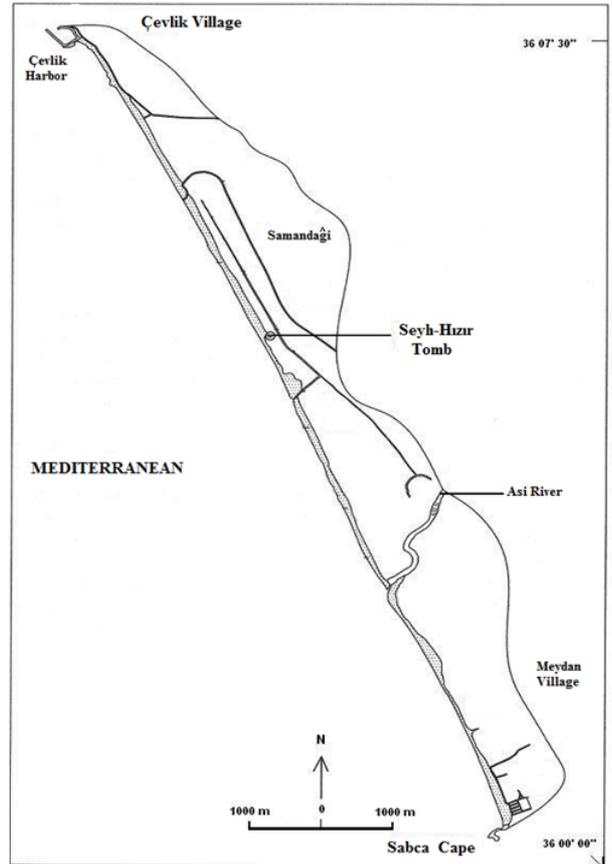


Figure 1. Map showing study area of Samandağ Beach.

of the device was tested under laboratory conditions against a standard mercury thermometer and had a mean resolution of 0.35 °C (minimum 0.3 °C, maximum 0.4 °C) for temperatures between 4 and 50 °C. Data loggers were programmed to record temperatures at 90-min intervals (16 readings per day) for a period of 90 days. Recorders were placed on the top of the eggs (Kaska et al., 1998; Öz et al., 2004) during oviposition or after excavating the nest in the morning of laying (about 10 h after oviposition). In the 2003 and 2006 nesting seasons, one nest was selected and two data loggers were placed in a nest; one of the data loggers was placed on the top of eggs and another was placed on the bottom eggs (Kaska et al., 1998; Öz et al., 2004) in order to compare temperatures between the levels. Temperature data were downloaded from data loggers to a computer and analyzed.

In order to analyze sex determination, dead late-term embryos and dead hatchlings were collected during excavation. All nests were checked every day to control for potential depredation, and nests under risk from land predators were screened off with a metal grid, with a 9-cm mesh placed above the nest at a depth of 20 cm

from the surface. Biological data such as distance of nests from the sea and vegetation, hatching success, incubation duration, clutch size, nest depth, and duration of hatchling emergence were collected during daily monitoring of sea turtles on Samandağ Beach between the 2003 and 2007 nesting seasons. Distance from sea (DFS) and distance from vegetation (DFV) for each nest were measured with a flexible tape ( $\pm 0.1$  m) from nest egg chamber and in a straight line. Hatching success (HS) was calculated as a percentage of hatchlings in a clutch (Yalçın-Özdilek, 2007). Incubation duration (ID) of each nest was defined as the number of days between the night when the clutch was laid and the night when the first group of hatchlings emerged (Broderick et al., 2001). Eight to 10 days after the last observed emergence of hatchlings, nests were excavated by hand (Yalçın-Özdilek, 2007). During this excavation, clutch size (CS) of each nest was determined by counting the number of unhatched eggs and hatched shell fragments one by one (Başkale and Kaska, 2005). Unhatched eggs were classified as containing no yolk, dead embryos, and no embryo visible to the naked eye. Nests were carefully excavated to avoid possible differences in the number of eggs originally laid. Concurrently, nest depth (ND) of each nest was measured with a flexible tape to the nearest centimeter as the straight vertical distance from the sand surface to the deepest point of the nest. The duration of hatchling emergence (DHE) was calculated as the number of days between the first and the last group of hatchlings that emerged from the nest.

Two types of nest temperatures were calculated in this study. One was the nest temperature recorded during the entire incubation period and as the representative of total incubation duration temperature. The other was the temperature taken during the middle third of the incubation period to estimate sex ratio. Sex ratios of hatchlings were estimated using two methods. The first method used the mean temperature during the middle third of the incubation period (the middle third of the incubation temperature =  $0.0714 \times \text{female ratio} \% + 25.125$ ) (Kaska et al., 1998). The second method used to estimate the sex ratios consisted in sexing all dead hatchlings and late-stage dead embryos (>stage 25) (Kaska and Downie, 1999) found in suitable conditions by histological analysis of the gonads (Yntema and Mrosovsky, 1980). Gonads of hatchlings were dissected and preserved in Bouin's solution for sex determination. The gonads were cut in half transversely and one half was embedded in paraffin wax, sectioned at 8–10  $\mu\text{m}$  from the middle of the gonad, and stained with periodic acid–Schiff reaction (PAS) stain and Harris hematoxylin (Yntema and Mrosovsky, 1980; Kaska et al., 1998). Sex designation was based on development of the cortical and medullary regions and the presence or absence of seminiferous tubules (Yntema

and Mrosovsky, 1980). Based on these histological criteria, only those individuals clearly identified as female or male were chosen. Sex was determined independently by two researchers.

In order to compare the mean nest temperatures among years and locations, nests built within each 10–14 days apart were chosen since it was assumed that they belonged to different females based on an interesting period for *C. mydas* ranging from 0 to 14 days (Broderick et al., 2002).

Normality and homogeneity of tests were determined before any comparison of data groups. The Levene statistic and Kolmogorov–Smirnov tests were used to test for homogeneity of variances. The two different methods used for placement of data loggers and sex ratio results of gonad histology and temperature recorder were compared by independent sample t-test and one-sample t-test. The Kruskal–Wallis test was used when normality and homogeneity of data were not found ( $P < 0.05$ ). Correlation between variables was tested with the Spearman rho. Results were considered significant at the 0.05 level. Analyses were conducted with SPSS 22.0. All data are reported as means  $\pm$  SD.

### 3. Results

A total of 14 nests were examined for nest temperature and the descriptive statistics are shown in Table 1. In addition to these nests, two nests were examined for top and bottom level nest temperatures, and the descriptive statistics of their features are shown in Table 2. The two different methods used for placement of data loggers were assessed together. The two different methods showed no significant differences for mean nest temperature ( $P > 0.05$ ) and middle third period of the nest temperatures ( $P > 0.05$ ). As seen in Table 3, the highest values of DFS and DFV of nests were 42 and 48.5 m in 2004 and 2007, respectively. Mean incubation duration was 54.2 days over the entire study period (Table 3). In 2006, nests had highest mean values for ND (77.3 cm) and HS (91.7%) (Table 3). In 2003, CS had the smallest value (91.6), whereas DHE had the largest value (1.3) (Table 3). Mean temperatures recorded during the middle third of the incubation duration ranged from 27.9 to 32.1  $^{\circ}\text{C}$  over the entire study period (Table 3). Mean nests temperatures were 30.5  $^{\circ}\text{C}$  for all nests and years (Table 3). Female ratios based on the middle third period of the nest temperatures were obtained between 39% and 97% over the years with a mean value of 74% (Table 3).

In the 2003 nesting season, a total of 190 dead late-term embryos and dead hatchlings were collected to analyze sex determination. Among them, 38 specimens were collected from the nests that had data loggers. The histological examination of dead *C. mydas* hatchlings ( $n = 38$ ) collected from four temperature-recorded nests indicated a female-dominant sex ratio (79%). Based on

**Table 1.** The descriptive statistics of temperature-recorded nests in Samandağ Beach, Turkey (distance from sea (DFS), nest depth (ND), clutch size (CS), mean nest temperature (NT), middle third temperature (MTT)).

Nest no.	Laying date	DFS (m)	ND (cm)	CS	NT (°C) (min-max)	MTT (°C)
N 71	10.07.2003	19	62	90	30.9 (29.8-31.8)	30.6
N 74	12.07.2003	19	74	115	32.2 (30.6-33.5)	31.6
N 80	15.07.2003	49	61	70	31.0 (29.4-31.8)	30.9
N 119	04.07.2004	75	76	194	28.3 (26.9-29.8)	27.9
N 124	04.07.2004	35	69	113	30.0 (28.3-31.8)	29.6
N 130	05.07.2004	22	75	95	30.2 (25.8-32.2)	30.3
N 132	05.07.2004	36	70	153	30.8 (28.3-34.8)	30.7
N 19	18.07.2005	22	75	79	29.6 (26.1-31.0)	29.3
N 20	18.07.2005	47	64	178	31.2 (30.2-33.0)	30.9
N 140	26.06.2006	35	74	138	29.8 (27.9-32.2)	30.7
N 148	29.06.2006	21	73	115	32.0 (30.2-33.5)	31.4
N 165	29.06.2006	18	85	127	32.0 (30.2-33.5)	32.1
N 19	25.06.2007	20	73	130	30.6 (29.1-31.8)	30.3
N 21	27.06.2007	29	68	72	31.4 (29.1-32.6)	31.0

**Table 2.** Descriptive statistics of temperature-recorded nests for level comparisons in the nest chamber. DFS = Distance from sea; ND = nest depth; CS = clutch size; NT = mean nest temperature; MTT = middle third period temperature; FR = female ratio.

Nest no.	Level	Laying date	DFS (m)	ND (cm)	CS	NT (°C) (min-max)	MTT (°C)	FR (%)
N 96	Top	29.07.2003	25	84	56	29.9 (28.7-31.4)	29.9	76
	Bottom					28.4 (27.6-29.4)	28.4	46
N 367	Top	03.08.2006	23	66	154	33.4 (28.7-34.8)	31.4	100
	Bottom					31.9 (26.9-33.0)	32.6	88.4

**Table 3.** Annual summary of mean characteristics of 14 nests with standard deviation and min-max for nest parameters. DFS = Distance from sea; ND = nest depth; CS = clutch size; NT = mean nest temperature; MTT = middle third period temperature.

Year	DFS (m)	DFV (m)	ND (cm)	CS	HS (%)	DHE (day)	ID (day)	NT (°C)	MTT (°C)	FR (%)
2003	29.0 ± 17.3	7.0 ± 12.1	65.6 ± 7.2	91.6 ± 22.5	72.9 ± 1.3	1.3 ± 0.6	49.3 ± 2.3	31.4 ± 0.7	31.0 ± 0.5	83.0 ± 7.2
	(19-49)	(0-21)	(61-74)	(70-115)	(71.4-73.9)	(1-2)	(48-52)	(30.9-32.2)	(30.6-31.6)	(77-91)
2004	42.0 ± 22.9	22.8 ± 27.8	72.5 ± 3.5	138.8 ± 44.1	69.3 ± 37.4	1.25 ± 0.5	57.8 ± 2.6	29.8 ± 1.1	29.6 ± 1.2	62.9 ± 16.8
	(22-75)	(0-58)	(69-76)	(95-194)	(15.8-99.1)	(1-2)	(55-60)	(28.3-30.8)	(27.9-30.7)	(39.3-77.6)
2005	34.5 ± 17.7	11.5 ± 3.5	64 ± 0.0	178 ± 0.0	44.9 ± 63.6	1 ± 0.0	48.0 ± 0.0	30.4 ± 1.1	30.1 ± 1.1	69.6 ± 16.3
	(22-47)	(9-14)	(64-64)	(178-178)	(0-89.9)	(1-1)	(48-48)	(29.6-31.2)	(29.3-30.9)	(58-81.1)
2006	24.7 ± 9.1	36.7 ± 18.2	77.3 ± 6.6	126.6 ± 11.5	91.7 ± 10.2	1 ± 0.0	57.7 ± 4.0	31.3 ± 1.3	31.4 ± 0.7	79.3 ± 23.4
	(18.0-35.0)	(17-53)	(73-85)	(115-138)	(80.3-100)	(1-1)	(53-60)	(29.8-32.0)	(30.7-32.1)	(52.8-97)
2007	24.5 ± 6.4	48.5 ± 4.9	70.5 ± 3.5	101 ± 41	15.1 ± 13.4	1.5 ± 0.7	55.5 ± 2.1	31.0 ± 0.6	30.7 ± 0.5	76.8 ± 6.8
	(20-29)	(45-52)	(68-73)	(72-130)	(5.6-24.6)	(1-2)	(54-57)	(30.6-31.4)	(30.3-31.0)	(72-81.6)
Total	31.9 ± 16.2	24.4 ± 21.8	71.1 ± 6.5	122.3 ± 37.3	63.7 ± 35.9	1.2 ± 0.4	54.2 ± 4.9	30.7 ± 1.1	30.5 ± 1.1	73.7 ± 15.8
	(18-75)	(0-58)	(61-85)	(70-194)	(0-100)	(1-2)	(48-60)	(28.3-32.2)	(27.9-32.1)	(39.3-97)

temperature data from the same year, the estimated female sex ratio was 83%. When comparing results for the same nests for sex ratio from histological examination and from middle third nest temperature data for sex ratio, sex ratios were similar ( $P > 0.05$ ). Overall histological examination of specimens indicated an 85% female-biased sex ratio.

Histological examination of hatchling sex ratios resulted in 94% females at the top and 64% at the bottom. These results were found to be significantly different in terms of the chi-square test ( $\chi^2 = 4.5$ ;  $df = 1$ ;  $P < 0.03$ ) with top levels of nests producing higher rates of female hatchlings while higher rates of males were found at the bottom levels of nests. In the 2003 nesting season, the estimated female ratios from nest temperature at the bottom and upper levels of nest chambers were 46% and 67%, respectively (Table 2). Records for the middle third period of the incubation temperature of the bottom and top levels of two nests (nest number 96 in 2003 and nest number 367 in 2006) were compared and the temperatures at the bottom levels were slightly cooler than the levels above, recorded as 28.5 °C and 29.9 °C in nest 96 in 2003 and 31.4 °C and 32.6 °C in nest 367 in 2006 at the bottom and the top, respectively ( $Z_{N96-2003} = 10.38$ ;  $n = 566$ ;  $P < 0.001$ ;  $Z_{N367-2006} = 8.04$   $n = 324$ ;  $P < 0.001$ ).

When the nest features were compared according to years with the Kruskal–Wallis test, no significant difference among years was found for mean nest temperatures (NT), mean middle third of temperatures (MTT), sex ratios (FR), hatching success (HS), clutch size (CS), distance from sea (DFS) and vegetation (DFV), nest depth (ND), and duration of hatchling emergence (DHE) (Table 4). However, incubation duration was significantly different among years (Table 4). The incubation durations of the 2005 nesting season were lower than those in the 2004 and 2006 nesting seasons. Incubation durations ranged from 48 to 60 days with a mean of  $54.2 \pm 4.9$  days (Figure 2).

Maximum increase in nest temperature was recorded as 6.5 °C during the 2004 nesting season (Figure 3). Similarly, the maximum decrease in nest temperature was recorded as 4.1 °C during the 2005 nesting season. The cause of this decrease was nest flooding at the 19th day of the incubation period (Figure 3). This clutch did not produce any hatchlings.

The perpendicular distances from the nests to the sea and to the vegetation varied between 18 and 75 m and 0 and 58 m, respectively (Table 2). The location of the nests in terms of distance from the sea was correlated with NT ( $r = -0.594$ ,  $P = 0.025$ ), MTT ( $r = -0.604$ ,  $P = 0.022$ ), and FR ( $r = -0.573$ ,  $P = 0.032$ ), whereas distance from vegetation was not correlated with those variables ( $P > 0.05$ ).

#### 4. Discussion

Gonad histology and nest temperature are both commonly used for estimating the sex ratio of sea turtles (Mrosovsky, 1994; Kaska et al., 1998; Broderick et al., 2001). In this study, we observed a female-biased sex ratio in green turtle hatchlings on Samandağ Beach. Most studies around the world and also in the Mediterranean showed a female-biased sex ratio in *C. mydas* populations (Mrosovsky, 1994; Kaska et al., 1998; Broderick et al., 2000; Kılıç and Candan, 2014). Samandağ shows a similar pattern to that of Akyatan, Northern Cyprus, and Sugözü in terms of sex ratio (Broderick et al., 2000; Casale et al., 2000; Kılıç and Candan, 2014). The range of mean nest temperatures in this study (between 28.3 and 32.2 °C;  $n = 14$ ) is similar to that reported by Kaska et al. (1998) (29.5 to 31.3 °C;  $n = 5$ ,  $P > 0.05$ ), Broderick et al. (2000) (29.8 to 32.5 °C;  $n = 18$ ,  $P > 0.05$ ), and Kılıç and Candan (2014) (28.1 to 32.6 °C  $n = 8$ ,  $P > 0.05$ ). The differences or similarities might result from local temperature variations at Samandağ Beach. Many environmental factors related to the nest location, such as distance from the sea, nearness of vegetation, being under sand dunes, sand albedo, climatic conditions like heavy rain and storms, and other factors related to both the climate and geomorphology of the beach, such as inundation and erosion, affected nest temperature and therefore sex ratio of hatchlings (i.e. Janzen and Paukstis, 1991; Mrosovsky, 1994; Ackerman, 1997; Godfrey et al., 1997; Hays et al., 2001).

Previous studies have indicated that a female-biased population, i.e. insufficiency of males, might cause unsuccessful fertilization of available females (Broderick et al., 2000). Broderick et al. (2000) estimated that the pivotal incubation period for green turtle populations in the Mediterranean was  $\geq 56$  days. According to their research, with the conservative assumption that 100%

**Table 4.** Comparison of nest features among years. DFS = Distance from sea; ND = nest depth, CS = clutch size; NT = mean nest temperature; MTT = middle third period temperature.

	DFS	DFV	ND	CS	HS	DHE	ID	NT	MTT	FR
Chi-square	3.679	5.520	5.251	5.063	5.995	2.033	9.812	4.749	6.188	3.667
df	4	4	4	4	4	4	4	4	4	4
Asymp. sig.	0.451	0.238	0.262	0.281	0.200	0.730	0.044	0.314	0.186	0.453

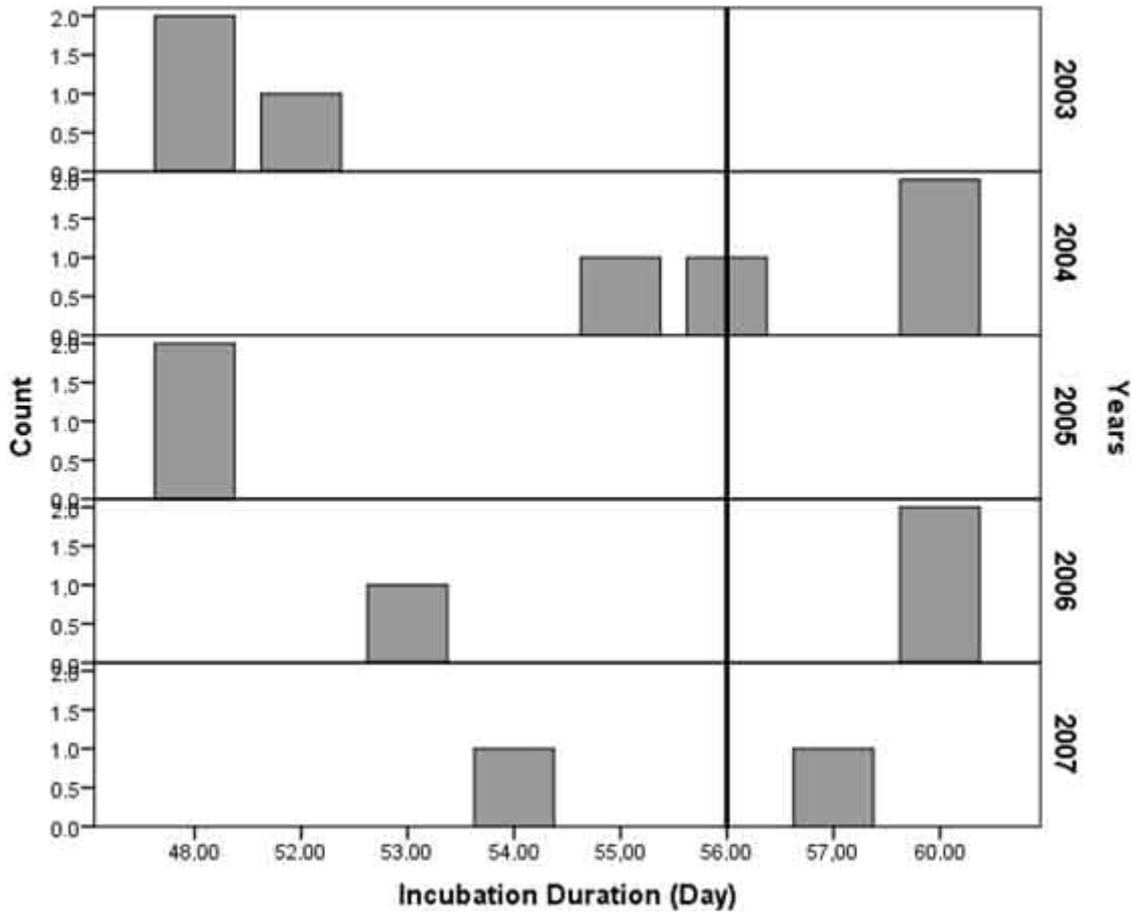


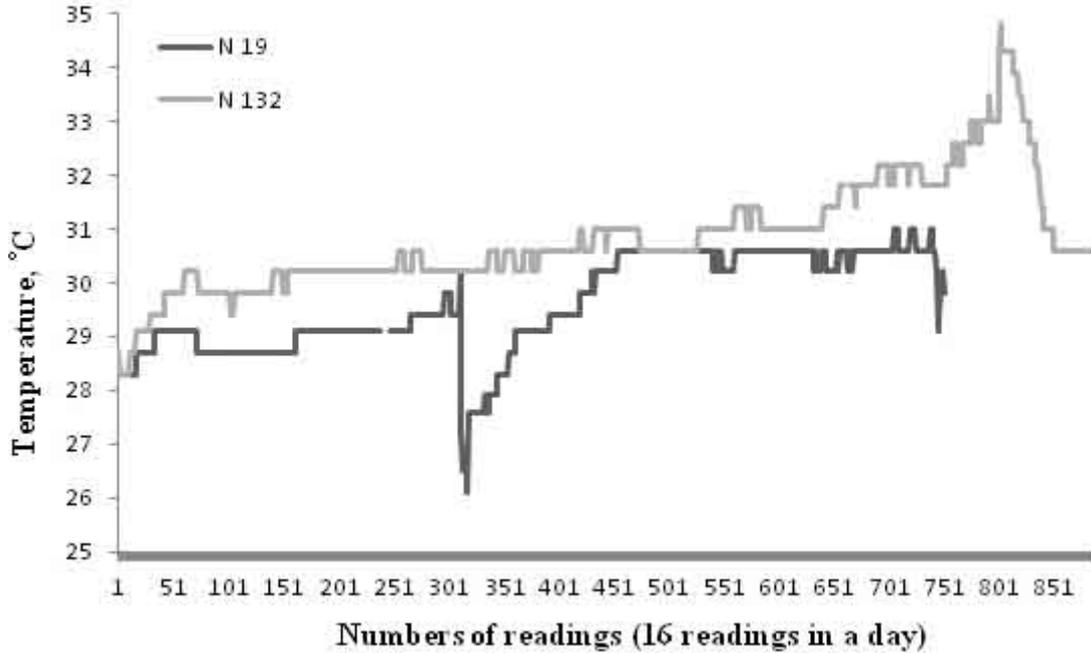
Figure 2. Frequency of incubation durations in examined nests at Samandağ Beach.

males are produced by nests with incubation periods of  $\geq 56$  days, Samandağ Beach, having a few nests incubating for 56 days or longer, could also be expected to have a higher production of male hatchlings. Although distance from vegetation showed no correlation with female ratio, one of total examined nests in the 2004 nesting season was not female-dominant (FR = 39.3%). This nest was covered by vegetation and vegetation on the beach could be an important role in producing male hatchlings. Spotila et al. (1987) stated that *C. mydas* nests in areas shaded by vegetation produced significantly more male hatchlings than nests in open areas at Tortuguero, Costa Rica. Similarly, in nonmarine turtles, Janzen (1994b) stated that vegetation cover was the most consistent predictor of the sex ratio for painted turtle (*Chrysemys picta bellii*) in Illinois. Moreover, female ratio and nest temperatures showed negative correlations with distance from sea. In other words, the female ratio decreased in nests closer to the vegetation. More data are needed to support this hypothesis for Samandağ Beach. As a result of PCA analysis, Türkozan et al. (2003) stated that distance from sea was inversely related to nest temperature and distance

from vegetation for loggerhead turtle nests at Fethiye, Turkey. However, Kılıç and Candan (2014) found a positive correlation between nest temperature and distance from sea on Sugözü Beach, Turkey. Although Samandağ and Sugözü are both located on the eastern Mediterranean, local differences such as rain and sand albedo (Hays et al., 2001) could vary between them.

Thermal variations within a nest could cause asynchronous patterns of emergences in loggerhead turtles (Houghton and Hays, 2001). Emergences of green turtles from the nests did not show an asynchronous pattern at Samandağ Beach (Table 3). The emergence of all *C. mydas* hatchlings in a nest is completed generally within a day; it rarely takes 2 days at Samandağ Beach. Therefore, results from our study do not support the relationship between intranest temperature differentiation and asynchronization emergence behavior in *C. mydas* hatchlings.

Hatching success is important for the estimation of the future generation of an endangered species. Global climate change, as predicted, would increase nest temperature, resulting in lower hatching success (Hawkes et al., 2007). However, there was no significant correlation between



**Figure 3.** The maximum increase and decrease in two nests' temperatures throughout the incubation duration.

hatching success and nest temperature at Samandağ Beach. Previous studies of Samandağ Beach indicated that hatching success was more related to nest moisture, and particularly inundation, than to sand temperature (Yalçın-Özdilek et al., 2007). Similarly, Özdemir and Türkozan (2006) stated that moisture has been associated with hatching success on Northern Cyprus. However, Türkozan et al. (2003) found that there was no correlation between hatching success and moisture and nest temperature for loggerhead turtle nests on Fethiye Beach, Turkey.

One of the possible effects of climate change on sea turtles populations is the critical control on sex ratios (Hays et al., 2003). However, Mazaris et al. (2015) stated that predictions based only on climate can be insufficient for protection of sea turtle populations, but it could be used for building the resilience of existing and potential nesting areas. They also warned that spatial climatic trends at a global scale cannot exactly estimate the distribution of sea turtle nesting sites. Hays et al. (2003) recorded an increase of about 0.13 °C in nest temperature for the last 100 years on Ascension Island. It is difficult to say that this was a result of global warming based only on results presented in this study for green turtle sex ratios on Samandağ Beach. Maximum increases and decreases in nest temperature can have an effect on hatchling survival (present study) and sex ratio (Kaska et al., 1998). Mrosovsky and Yntema (1980) stated that a change of 1–2 °C can make a considerable difference in the sex ratios of hatchlings. If the temperature decreases by 1 °C, the duration of incubation will increase by 5 days (Mrosovsky and Yntema, 1980). In this study,

we found 6.5 °C of maximum temperature increase during the incubation period. This nest with the maximum temperature increase had a longer incubation duration (55 days) than the mean incubation duration (54.2 days) of the entire study. This value was lower than the maximum temperature increase of 9.6 °C found for green turtles on Cyprus by Kaska et al. (1998). However, there is no significant differences between them ( $P > 0.05$ ). These differences among beaches could have resulted from local temperature variations, climatic conditions like heavy rain and storms, and other factors related to both climate and geomorphology at each beach (Janzen and Paukstis, 1991; Mrosovsky, 1994; Godfrey et al., 1997). Metabolic heating during incubation should not be ignored as a cause for the increasing nest temperatures found during incubation (Godfrey et al., 1997). This study showed that at least 4 °C less in temperature will result in mortality increase for green turtles on Samandağ Beach. The nest temperatures might be affected by environmental changes, such as inundation by high tides and storms (Kaska et al., 1998). In addition to rapid temperature decline, the osmotic conditions in that nest chamber could have resulted in embryonic mortalities (Ackerman, 1997).

Relocation (or hatchery) of nests to safer areas may increase hatching success (Wyneken et al., 1988; Başkale and Kaska, 2005; Sönmez and Yalçın Özdilek, 2013), but the natural sex ratio (Godfrey and Mrosovsky, 1999) and hatchling morphology (i.e. scute deviation and production of smaller hatchlings) and survival of hatchlings (Türkozan and Yılmaz, 2007; Sönmez et al., 2011) after relocation

can be different. Nest relocation over the long term may also distort gene pools (Mrosovsky, 2006). Sönmez and Yalçın Özdilek (2013) suggested that nests within ~20 m of perpendicular distance from the sea were under risk of erosion and if nests in that section were not relocated then all would most probably be flooded and disappear. From this point of view, we suggest that nests are under risk of erosion and the temperature characteristics of the beach sections should be well known and taken into consideration when translocating nest on Samandağ Beach. The conservation aspects of the sex ratio of hatchlings, such as collecting data with temperature recorders in beach sections and nests, and research on metabolic heating also need to be continuously monitored over different years and throughout the season.

In conclusion, Samandağ Beach has a female-biased sex ratio based on histological examination and nest temperatures for green turtle hatchlings. The sex ratio was negatively correlated with the distance of nests to the sea, but it was not correlated with distance of nests to the

vegetation. However, vegetation on the beach could be an important role in producing male hatchlings. At least 4 °C less in temperature can affect the hatchlings' survival during incubation. Given the importance of climate change effects on sea turtle populations, this research could be useful for effective conservation planning for both existing and future populations.

#### Acknowledgments

We particularly want to thank the Scientific and Technological Research Council of Turkey (TÜBİTAK) since this study was partially supported by the projects coded YDABAG-103Y058 and YDABAG-104Y055. We must also express gratitude to the Hatay Environmental and Forestry Directorate. Many thanks also to the hard-working MKÜ Biology Department volunteer students, to Dr Hasan Göksel Özdilek for his help in field research, and to lecturer Graham H Lee for proof-reading the text. We thank Dr Graeme Hays and Dr John Rowe for reviewing and giving valuable advice for this manuscript.

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