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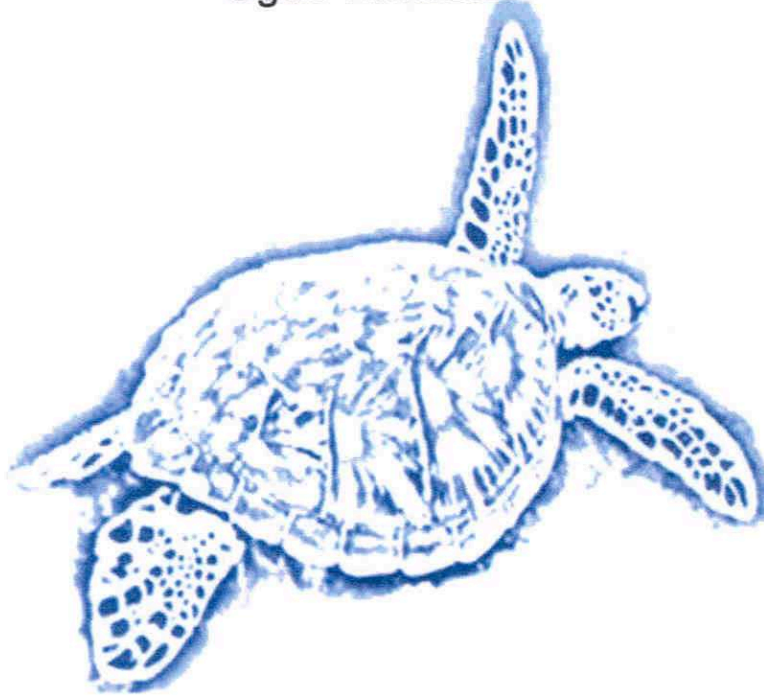Pamukkale University
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NEST TEMPERATURES AND SEX RATIO VARIATIONS AMONG THE HATCHLINGS AND EMBRYOS OF LOGGERHEAD TURTLES ON DALAMAN BEACH, TURKEY

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INTRODUCTION

Many reptiles have no sex chromosomes. Sexual differentiation of sea turtle hatchlings is determined by egg incubation temperature, usually during the middle third of development (Yntema and Mrosovsky 1980, Janzen and Paukstis 1991, Mrosovsky 1994, Kaska et al. 1998). Temperature-dependent sex determination has been the subject of a number of studies (review in Mrosovsky 1994). When eggs are incubated at constant temperatures, there is a narrow range of temperatures around which ca. 50 % of each sex will be produced; wider ranges above this temperature produce females and below this threshold produce males (Bull 1980). The temperature at which an equal sex ratio is produced has been termed the pivotal temperature (Yntema and Mrosovsky 1980, Mrosovsky and Pieau 1991). The hatchling sex ratio depends on the proportion of embryonic development that occurs above and below the pivotal temperature during TSP. The sex ratio estimations on sea turtles were reviewed recently by Freedberg and Wade (2001).

The population survival of sea turtles depends on the sufficient production of both sexes. Estimates of the sex ratio have been obtained by combining the nesting distribution with the sexing of sampled hatchlings from different times during the season, from pivotal incubation durations and nest temperature of a nest during the middle third of the incubation period (Standora and Spotila 1985, Mrosovsky 1994, Marcovaldi et al. 1997, Kaska et al. 1998, Godley et al. 2001a).

It is known that sex determination in sea turtles is temperature dependant and this phenomenon is called temperature depend sex determination. Sex ratios of loggerhead turtles in the Mediterranean have been studied especially during the last few years. Kaska et al. (1998) found a mean sex ratio of 81.6 % females in loggerhead clutches laid over the 1995 and 1996 nesting seasons. Godley et al. (2001a, b) reported very short incubation durations for loggerhead turtles in Cyprus, implying warm, feminising conditions (89-99 % females based on incubation durations and mean incubation temperatures). The pivotal temperatures in studies of loggerhead turtles all cluster within one degree of 29 °C (Mrosovsky 1994, Marcovaldi et al. 1997). Mrosovsky et al. (2002) reported recently the pivotal temperature for loggerhead turtles in the Mediterranean (by using two clutches from Greece) as 29.3 $^{\circ}$C and the pivotal incubation duration as 52.6 days. For the eastern Mediterranean, Kaska et al. (1998) used mean temperatures in the middle third of incubation to indicate a pivotal temperature just below 29 °C and the pivotal incubation duration later calculated as 59.9 d, close to the values of 59.3 and 61.7 d for Brazil and the USA, respectively. These data indicate that the physiology of Mediterranean loggerhead turtles is quite similar to that of conspecifics in the Americas with respect to thermal influences on sexual differentiation. Mrosovsky et al. (2002) reported also that hatchling sex ratio on some Mediterranean beaches is female biased but probably varies within this region. We, therefore, aimed to investigate the sex ratio of hatchlings on Dalaman beach, where there was no similar study done before.

MATERIALS AND METHODS

Temperature was measured using "Tiny talk" temperature recorders (Orion Components (Chichester) Ltd., UK). The device fits within a 35 mm film case. The accuracy of the device was tested under laboratory conditions against a standard mercury thermometer, and they were found to have a mean resolution of 0.35^oC (min. 0.3^oC, max. 0.4^oC) for temperatures between 4^oC and 50^oC. They were launched by computer for a recording period of 60 days with readings taken at 90 min. intervals. This gave 16 readings per day. They were placed at one (either top or bottom) or two (any two levels) of the nest, during the oviposition or after excavating the nest in the morning of laying (approximately 10 hours after oviposition).Temperature data were offloaded to a computer and the gonads of the sacrificed 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 µm from the middle of the gonad, and stained with the Periodic Acid Schiff reaction (PAS) and Harris' haematoxylin. Sex designation was based on the development of the cortical and medullary regions and the presence or absence of seminiferous tubules (Yntema and Mrosovsky 1980). The middle third of the incubation period was calculated from the total incubation period, from the night of laying to the day of first hatching. The temperature data were analysed as Mean temp. 2/3 period = 0.0716 percent female + 25.114 as the formula obtained from Kaska et al. (1998).

RESULTS AND DISCUSSION

A total of 34 nest temperatures were recorded (8-12-14 for the years 2002-4 respectively). By analyzing the nest temperatures during the middle third of the incubation period, the mean temperatures during this period ranged from 28.4 to 31.9^oC. The sex ratios were estimated between 46% and 95% with a mean of 76% females by using the temperature data.

The mean sex ratios were obtained as 85% females (n=190) by histological examination of the gonads of dead hatchlings and embryos. The majority of the embryonic mortalities were found at early (6-7) and late (>26) stages. When these embryonic mortalities were compared in terms of depths, the highest percentages (45%) were found at middle levels and bottoms (35%) of the nests and less (20 %) mortalities at top levels. When the sex ratio of dead hatchlings and embryos was compared between the different levels, a 94% female sex ratio was obtained at the top level but only 64 % at the mid and bottom levels. The temporal and spatial sex ratio variations were also studied.

The nesting season started in mid May and continued until August. The hatching season started in July and continued until the first few weeks of the October, but there were no samples collected in October. Although our sample sizes were not high, we divided these hatchlings for every two weeks (from the begging of the July to the end of September) periods for the hatching season, there were statistical differences between the percentages of the both sexes produced during these periods (x^2=11.39; df=5; P<0.05), there were slight increases in the male percentages at the beginning and the end of hatching seasons compared to the middle hatching season.

The temperatures of nests close to the sea may be cooler, and therefore may be potentially producing more males, nests further inland may be exposed to warmer temperature conditions, and therefore producing more females. The relocation of nests to a safer area (or hatchery) may increase the hatching success, but the nature sex ratio and the sex ratio after the relocation

might be different. The investigations of sex ratios are becoming important due to global warming and the relocation of nests. Since nearly all the natural sex ratios are highly female dominated, production of both sexes may be necessary in the future, since an endangered species can only be conserved by the presence of both sexes in nature. If we have more unfertile eggs in the future or if we are getting 100% female hatchlings from the beach, we may have to think of producing male hatchlings.

Sex ratio estimations and their biological and ecological implications are clearly a complex issue. There is an intricate interplay between nest location, nest depth, nest temperature, duration of hatching, selective predation and other mortalities within and outside the nest, along with changing conditions from year to year and from beach to beach.

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