

**Sand temperature profiles at turtle nesting sites in the Red Sea:  
implications for hatchling sex ratios**

Thesis by  
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## **EXAMINATION COMMITTEE PAGE**

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**ABSTRACT**

Sand temperature profiles at turtle nesting sites in the Red Sea: implications for hatchling sex ratios

Lyndsey Tanabe

Climate change poses a serious threat to species that demonstrate temperature dependent sex determination (TDS), including marine turtles. Increased temperatures can result in highly female skewed sex ratios and decreased hatchling success. *In situ* sand temperature data was collected from the nesting depth of hawksbill and green turtles at five study sites along the coast of the Red Sea. The sand temperature profile at four of the sites exceeded the pivotal temperature of 29.2°C (commonly cited in literature) throughout the study duration, which suggests feminization of turtles could be occurring, but further studies need to identify the pivotal temperature in this region. The percentage of days exceeding the commonly cited maximum thermal threshold (33 and 35°C) was calculated for each site at 30 and 50 cm. Sand temperature recordings were as high as 36.0°C at 30 cm depth, and 35.3°C at 50 cm. This suggests that the turtle hatchlings in some areas of the Red Sea could already have high mortality rates due to high temperatures, unless they are locally adapted to these high temperatures. The Red Sea is home to five out of the seven extant species of marine turtles in the world, but not much is known about these populations. The Red Sea is an understudied region of the world, but it has the potential to provide insight on how species might adapt to future climate change due to its high and variable water temperatures (range of 20°C to 35°C) and high salinity (40 PSU). Sites with lower sand temperatures (and lower risk of feminization) may represent priority areas for conservation efforts, particularly in regions facing imminent coastal development.

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## **LIST OF ABBREVIATIONS**

BSR – breeding sex ratio

CITES - Convention on the International Trade of Endangered Species

IPCC- International Panel on Climate Change

IUCN - International Union on the Conservation of Nature

KAUST – King Abdullah University of Science and Technology

PT- pivotal temperature

SST - sea surface temperature

TDS - temperature-dependent sex determination

TRT - transitional range of temperature

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## 1. Introduction

### 1.1 Turtles and threats

Marine turtles first appeared during the late Triassic period 220 million years ago (Li et al., 2017) and have since survived and adapted to climatic changes throughout their evolutionary history (Harris, 1993). Despite survival through past climatic shifts, present day rates of change are very rapid, leading to a rising concern of how modern populations will respond to anthropogenic climate change (Root & Schneider, 2002). There are currently seven extant species of marine turtles found worldwide, all sharing similar life history traits. In general, turtle hatchlings emerge from their nest at night, and spend their early life stages in the open ocean feeding on planktonic food (Carr, 1986). After this pelagic stage, juveniles undergo a behavioral shift and migrate to coastal waters to forage (Carr & Caldwell, 1956). Upon maturity, estimated between 20-50 years of age (Goshe et al., 2010), they will migrate back to their natal beaches to reproduce and nest. The exact mechanism on natal homing is unknown, but a common hypothesis is that turtles imprint on the unique geomagnetic signature of their birth place, and then they use this information to return and reproduce (Lohmann et al., 2008a). During the mating season, a single female can mate with several males, known as polyandry (Lee & Hays, 2004). A few weeks after mating, adult female turtles crawl onto their natal beach at night and take at least two hours to dig their nest pit. The number of eggs in a nest, called a clutch, varies by species and ranges from 50-200 eggs. An individual female lays between 2-8 clutches per nesting season, and she will nest every 2-4 years (Esteban et al., 2017). Thus, turtle populations demonstrate many life

history traits that make them vulnerable to environmental change and anthropogenic impacts. These characteristics include a long pelagic maturation period, late sexual maturity, large clutch sizes with high juvenile mortality, and migrations between nesting and feeding locations (Van Buskirk & Crowder, 1994). Thus, nearly all species of marine turtles are included in the International Union on the Conservation of Nature's Red List of Endangered Species (IUCN, 2018) and are protected by Appendix I of the Convention on the International Trade of Endangered Species (i.e., no international trade is permitted) (CITES, 2018). Despite this protection, illegal turtle fisheries continue in many countries.

Turtles have been over-exploited for centuries for their meat, oil content, and intricate shells. The IUCN (2018) reported that in every major ocean basin, there has been an 84-87% decline of hawksbill females nesting annually over the past three generations (Mortimer & Donnelly, 2007). This is likely attributed to the degradation of nesting beaches, harvest of juveniles and adults for food, incidental by-catch, and overall marine habitat degradation (Meylan & Redlow, 2006). The turtle shell trade has severe impacts on sea turtle populations. It is estimated that over the last century, millions of hawksbill turtles have been killed for the markets of Europe, the United States, and Asia (Meylan & Redlow, 2006). Turtle shell trade has existed in the Red Sea region for at least 2,000 years (Hirth & Latif, 1980). Moreover, egg collection and exploitation of marine turtles continue in many parts of the world, despite protection (Meylan & Redlow, 2006; Seminoff, 2004). All of the countries surrounding the Red Sea legally protect marine turtles through national laws and international agreements,

although enforcement at sea and around nesting sites is significantly lacking (Mancini et al., 2015).

## 1.2 Temperature-dependent sex determination

Marine turtles demonstrate temperature-dependent sex determination (TSD), which is a common trait of many reptiles. While the sex of mammals is determined by sex chromosomes at the time of fertilization, the sex of marine turtles is determined by the nest temperature during the middle third of incubation (Mrosovsky, 1980). Thus, there are significant implications to these populations in the face of climate change because small changes in temperature can cause dramatic changes in sex ratio (Bull, 1979).

For marine turtles, eggs incubated at  $\sim 29^{\circ}\text{C}$  produce a 50:50 sex ratio, called the pivotal temperature (PT), which differs slightly between species and geographic location (Mrosovsky & Pieau, 1991; Godfrey & Mrosovsky, 2006; King et al., 2013).

Temperatures above the PT produce predominantly females and vice versa (Mrosovsky, 1994). The transitional range of temperature (TRT) is the range of incubation temperatures in which both female and males are produced. This value is generally not more than  $2\text{-}3^{\circ}\text{C}$ , and is centered around the pivotal temperature (Mrosovsky, 1994).

Temperatures above or below the TRT result in 100% males or females. For all species of marine turtles, there is a thermal range of  $25\text{ to }33^{\circ}\text{C}$  in which egg incubation is successful (Miller, 1985; Spotila & Standora, 1985; Howard et al., 2014), and temperatures towards the higher threshold (prolonged exposure above  $33^{\circ}\text{C}$ ) can result

in morphological abnormalities and hatchling mortality (Hays et al., 2003; Hawkes et al., 2007; Laloë et al., 2017; Packard et al., 1977). The frequently cited maximum thermal thresholds of 33°C (Miller, 1997) and 35°C (Ackerman, 1997) were based on studies conducted in the field and through constant incubation laboratory experiments, respectively. There are no Red Sea specific values of thermal maximum thresholds, so the commonly cited temperatures of 33 and 35°C were used in this study to assess the percentage of days exceeding these thermal thresholds. The effects of constant incubation at high temperature versus fluctuating high temperatures have substantial differences on hatchling survival (Bowden et al., 2014).

### 1.3 The Red Sea

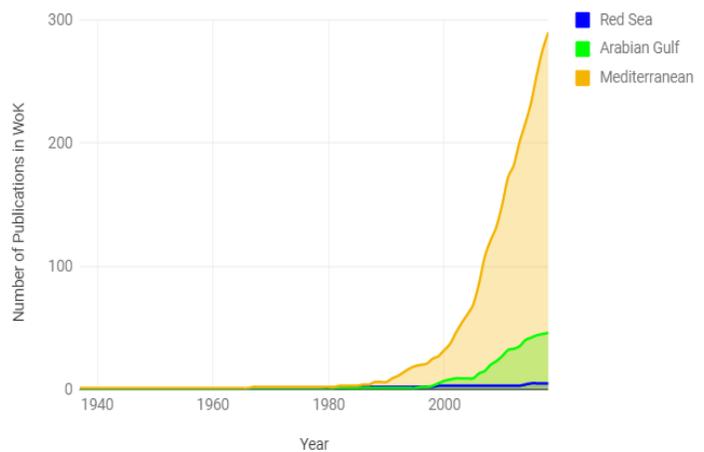
The Red Sea is a semi-enclosed body of water with high biodiversity (Stehli & Wells, 1971) and endemism (DiBattista et al., 2016), supporting a variety of coastal and marine habitats. Some species inhabiting the Red Sea are vulnerable to oceanic warming from anthropogenic climate change (Sawall et al., 2015), with current mean temperatures already exceeding those of other tropical regions (Cantin et al., 2010). Despite high temperatures, the Red Sea hosts one of the largest reef ecosystems in the world, but some organisms are already close to their thermal limits (Osman et al., 2017).

Five of the seven extant species of marine turtles have been observed within the Red Sea: the green turtle (*Chelonia mydas*), hawksbill (*Eretmochelys imbricata*), loggerhead (*Caretta caretta*), olive-ridley (*Lepidochelys olivacea*), and leatherback (*Dermochelys coriacea*) (Frazier & Salas, 1984). The hawksbill and green sea turtle are

the most common species and are known to nest along the coastline and on sandy islands throughout the Red Sea (Attum et al., 2014; Hanafy, 2012; Pilcher & Al-Merghani, 2000). The nesting season in Egypt is thought to extend from May to September, with a peak in late July and early August (Hanafy & Sallam, 2003). In Saudi Arabia, less is known about nesting season duration. Pilcher & Al-Merghani (2000) suggested that in Ras Baridi, nesting occurred from July to November, with a peak in September, and an average incubation duration of 57 days (Pilcher, 1999). In the Red Sea, it is estimated that there are about 1,500 females nesting per year (Hanafy, 2012). The Red Sea is an understudied region of the world (Berumen et al., 2013). Decades of war and political unrest have prevented surveys in Somalia, Eritrea, Sudan and Yemen, and therefore the status of sea

turtles in these areas are unknown (Mortimer & Donnelly, 2007). There have been very few publications about marine turtles in the Red Sea compared to other water bodies in the region such as the Mediterranean or Arabian

Gulf (Fig. 1). A Web of Knowledge search indicates there are currently only five publications with the topic of turtles in the Red Sea, whereas the Arabian Gulf has 46 and the Mediterranean has 290. This demonstrates a major knowledge gap in the Red Sea, despite having foraging and nesting habitats throughout the latitudinal gradient.



**Figure 1.** Cumulative number of Web of Knowledge marine turtle publications through time from three regions: the Mediterranean (yellow), Arabian Gulf (green), and the Red

#### 1.4 Turtles and climate change

Climate change is affecting species and ecosystems worldwide (Root et al., 2003; Walther et al., 2002) and the impact is particularly adverse for populations that exhibit temperature-dependent sex determination, including marine turtles. Climate change is expected to negatively alter the viability of sea turtle populations in areas that experience rising temperatures (Butt et al., 2016; Fuentes et al., 2011). In the past century alone, the global mean air and sea temperatures have risen 0.8°C and this is predicted to increase by 2.6°C by 2100 (IPCC, 2014). The impact of these changes on marine turtle nesting and foraging grounds is a growing field of study (Laloë et al., 2017; Poloczanska et al., 2009; Saba et al., 2012). In addition to sex determination and hatchling success, incubation temperature influences growth rate (Gillooley et al., 2001), incubation duration (Mrosovsky & Yntema, 1980), morphology of hatchling (Du & Ji, 2003), and long-term physiology and behavior traits (Sibly & Atkinson, 1994). Given the severe consequences that climate change poses to marine turtles, it is important to identify behavioral changes that turtles could make in order to adapt to changes in temperature.

#### 1.6 Objectives

The main objective of this study was to evaluate the current sand temperatures profiles at nesting sites in the Red Sea at the average depths of hawksbill (30 cm) and green turtle (50 cm) nests. These temperature profiles were compared between five different sites to assess if there are any site-specific trends. We also used *in situ*

temperature data to assess the seasonal changes and depth comparison of temperature at each study site to identify the possibility for turtles to alter their behavior in order to nest in cooler sands. Fine-scale differences between temperature logger station locations within an individual site were measured to assess the importance of nest-site selection. Daily temperature fluctuations were calculated to identify the amount of environmental change that occurs at nesting depth, to assess if parameters measured in constant incubation experiments (pivotal temperature, transitional range of temperature, thermal threshold, etc.) are relevant to the natural nest conditions found in the Red Sea. *In situ* measurements recorded at nesting depths of hawksbill and green turtles were used to predict the hatchling sex ratio by using a maximum likelihood mathematical model. In addition, the percentage of days exceeding the possible thermal thresholds of 33 (Miller, 1997) and 35°C (Ackerman, 1997) was determined.

## **2. Methods**

### **2.1 Study sites**

The Red Sea environmental conditions are relatively extreme due to the high temperatures throughout the region. This marine ecosystem is one of the most saline bodies of water, reaching up to ~40.0 PSU in the north, located in one of the hottest regions of the world (Edwards, 1987). The temperature is persistently high throughout the year (up to 35°C in the summer, and about 24°C in the spring), with very little precipitation, high evaporation rates, and no significant fresh water input. This study

included five sites within the Red Sea distributed across  $\sim 8^\circ$  latitude ( $27\text{-}19^\circ$  N). Each study site was located in an area with evidence of turtle nesting activity.

There is a gradient of environmental conditions along the latitude of the Red Sea. Salinity increases from south to north as temperature decreases. The five sites in this study were selected to cover these strong temperature and environmental gradients (Fig. 2). The northernmost site in this study is Small Gobal Island (N  $27^\circ 40' 49.31''$  E  $33^\circ 48' 13.92''$ ) located in Egypt near the Gulf of Suez. This island has recorded nesting activities of hawksbill turtles (Hanafy, 2012). Ten hawksbill nests were recorded during the 2018 nesting season beach (I. Elsadek, personal communication, October 2018). Small Gobal Island is comprised of sandy beaches and uplifted marine limestone cliffs. There is sparse low vegetation which was highly dispersed. This island is located approximately 50 km away from the port in Hurghada, so it has few visitors, consisting mainly of scuba divers on liveaboards who rarely visit the beach.

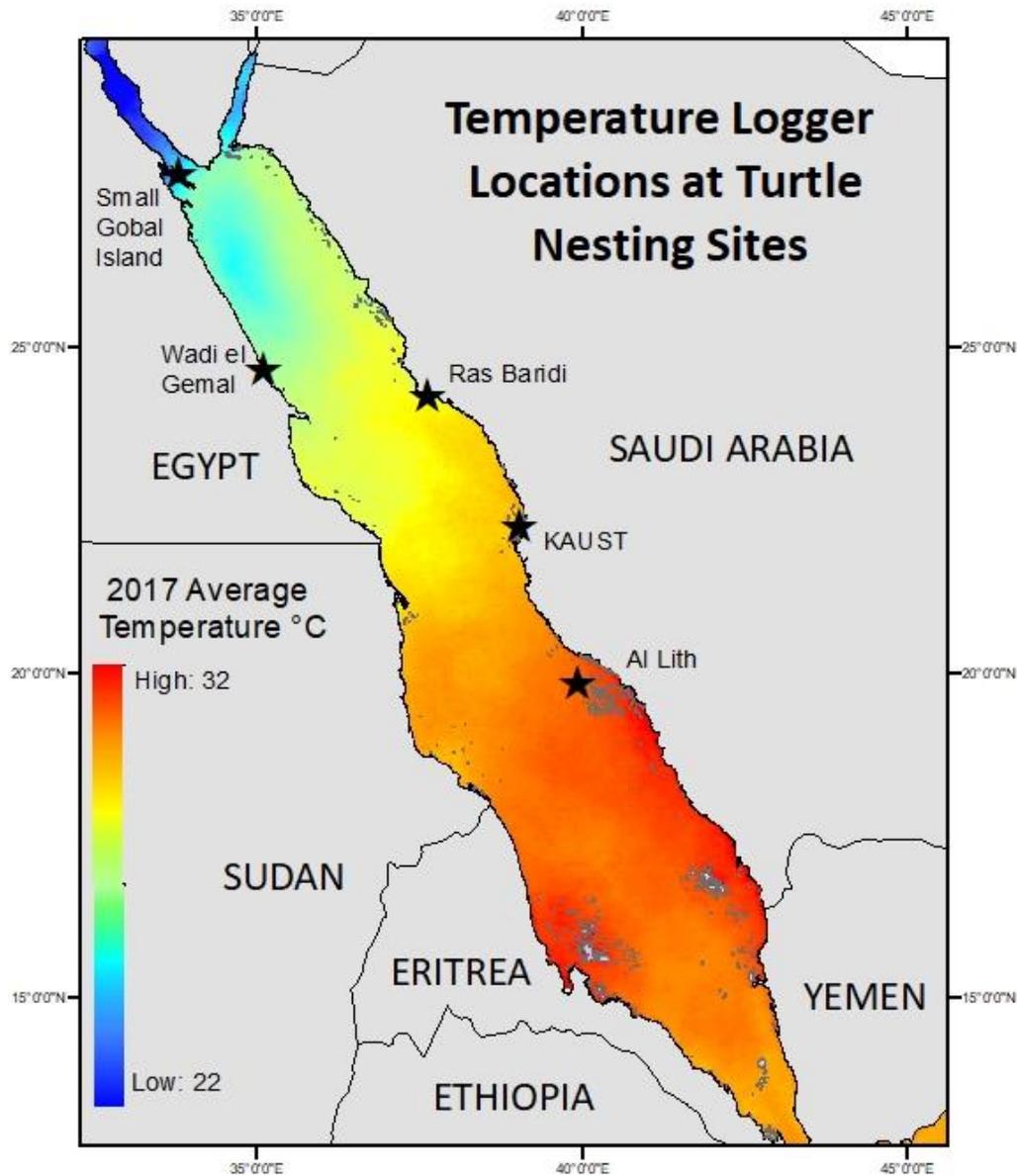
Wadi el Gemal (N  $24^\circ 39' 51.59''$  E  $35^\circ 06' 33.09''$ ) is an Egyptian National Park located south of Masrsa Alam. This site is sometimes referred to as Ras Bagdadi in some publications, and has evidence of green turtle nesting (Hanafy, 2012). This beach has limited access due to the protection afforded by the National Park, but the fringing reef is a commonly-used dive and snorkel boat site. Fresh tracks and old nests were observed during temperature logger deployment. Twenty green turtle nests were counted during the 2018 nesting season beach (I. Elsadek, personal communication, October 2018). There was some low-lying vegetation around these nests.

The northernmost study site in Saudi Arabia is a stretch of shoreline known as Ras Baridi, north of Yanbu (N 24°16'24.33" E 37°37'35.3"). Ras Baridi is located near the Yanbu Cement Company, which uses limestone from a nearby fossil reef in its cement production. Research indicates that cement dust from the factory results in decreased hatchling emergence from the hardened dome made by the accumulating dust over the clutch during the initial stages of the filling-in process by the adult female (Pilcher, 1999). In 1983, a study suggested that due to the high environmental temperatures in this region and the design of the factory, the kiln dust that is produced in the firing process is not reused, and it is estimated that over 120 tonnes of this dust is released per day (MEPA, 1983). Despite proximity to the cement factory, there is some consensus that this is the main green sea turtle nesting area in Saudi Arabia (Pilcher, 1999; Pilcher & Al-Merghani, 2000). The rookery is composed of small sandy coves along an exposed fossil reef that extends 2-3 meters above sea level. The beach surface has small halophytic plants above the dune line.

The Thuwal study site is located off of King Abdullah University of Science and Technology (KAUST) in the central Red Sea. The Thuwal study site was established on a sandy unnamed island (N 22°15'9.3" E 39°01'40.2"). No record of existing turtle nesting in this area was found in literature. Evidence of sea turtle egg shells were found and local fishermen have witnessed emergence of turtle hatchlings in the early spring. The sandy island is small (~100 meters in length) with little vegetation.

Al Lith is the southernmost site in this study, located about 50 km offshore within the Farasan Banks. This study site was on Marmar Island (N 19°50'17.8" E

39°55'38.9"). The island is about 800 meters long and 150 meters wide. This was the site with the greatest evidence of nesting (turtle egg shells, tracks, and a hatchling corpse). There was both hawksbill and green turtles in the area, but only evidence for green turtle nesting was found during this study.



**Figure 2.** Study locations at turtle nesting sites in the Red Sea. From North to South, these sites include Small Gobal Island, Wadi el Gemal, Ras Baridi, KAUST, and Al Lith.

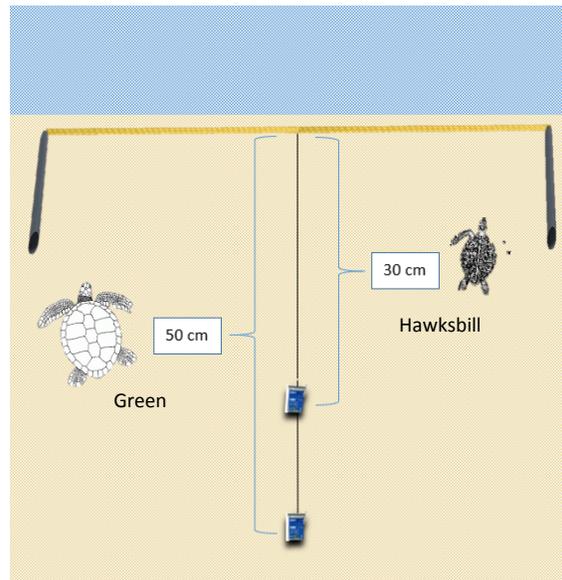
## 2.2 Measuring sand temperature

HOBO Pendant® Temperature Data Loggers (henceforth referred to as “loggers”), UA-001-64, Onset® Computers, were used to record sand and water temperature. The accuracy reported by the manufacturer is  $\pm 0.53^{\circ}\text{C}$  and the resolution is  $0.14^{\circ}\text{C}$ .

Temperature measurements were recorded every 15 minutes. Loggers

were buried at 30 and 50 cm, the average depths of hawksbill and green turtle nests, respectively. Loggers were deployed from May-September 2018 (Table 1), because turtles are recorded to nest mainly in the spring and summer in this region (Hanafy, 2012; Pilcher & Al-Merghani, 2000). To facilitate the location and retrieval of the loggers, they were attached together on a string, which was secured to a rope buried just under the sand (Fig. 3). The rope was tied to small pieces of PVC pipe to ensure that it stayed in place throughout deployment. GPS coordinates and photos were taken at each site.

At each study site, there were a total of 12 temperature loggers deployed, with variable numbers of loggers retrieved (Table 1). Each site had four stations of sand temperature measurement, at both 30 cm and 50 cm depths. In addition, there were four in-water loggers deployed at approximately five meters depth. For the islands (Al



**Figure 3.** Methods of deploying sand temperature loggers at depth of hawksbill and green turtle nests.

Lith and Thuwal), loggers were placed on the north, east, south, and west of the island and surrounding reef. For the beach study sites, loggers were spread between 50 and 500 meters apart, depending on the length of the sandy portion of the beach.

**Table 1.** Temperature logger retrieval information for each site (listed from North to South). Start and end dates are in the year 2018, and the number of loggers retrieved include those deployed at 30 and 50 cm in the sand, and 5 m in the ocean.

Site	GPS	Start date	End date	# days	# loggers retrieved
<b>Small Gobal Island</b>	N 27°40'49.31" E 33°48'13.92"	May 28	October 4	130	3
<b>Wadi el Gemal</b>	N 24°39'51.59" E 35°06'33.09"	May 29	September 21	116	7
<b>Ras Baridi</b>	N 24°16'24.33" E 37°37'35.3"	June 2	September 19	110	10
<b>Thuwal</b>	N 22°15'9.3" E 39°01'40.2"	May 10	September 15	129	10
<b>Al Lith</b>	N 19°50'17.8" E 39°55'38.9"	May 3	September 20	141	4

## 2.3 Sand temperature comparison between study sites

### 2.3.1 Comparison of mean sand temperatures

To compare the mean sand temperature at 30 and 50 cm depth at each study site, HOBOware was used to assess if the data was suitable for analysis. If the logger was dislodged from its deployed depth, the data showed extreme ranges in temperature so the measurements after the dislodgement were omitted from analysis. HOBOware was also used to find the average daily temperature for each logger. At each of the five study sites, the mean daily temperature measurement was calculated for 30

and 50 cm depth. These mean values were plotted with the theoretical pivotal temperature (29.2°C) over time from May 30<sup>th</sup>-September 20<sup>th</sup>, 2018. This time period was selected because the majority of the five study sites had loggers deployed for this duration. In addition to reporting daily means, the maximum temperatures recorded at each site were also determined.

### 2.3.2 Comparison of monthly mean temperatures

Monthly averages were used to compare temperatures between study sites to assess how many months exceeded the theoretical pivotal temperature (PT) of 29.2°C. The average and standard variation were calculated for the loggers deployed at the same depth at each study site. This comparison was used to assess how the sex ratios could differ month-to-month at the five turtle nesting sites.

### 2.3.3 Comparison of water temperature

The mean daily water temperatures were calculated at each site, to determine if they follow a similar pattern to the sand loggers. Water temperature can be used as a proxy for sand temperature (Laloë et al., 2014). Finding a proxy for sand temperature is of particular interest in the Red Sea region, because many of the turtle nesting sites are not easily accessible for direct measurements. In Egypt, some the major nesting sites such as Zabarged Island and Large Gobal Island in Egypt (Hanafy, 2012) are strictly military-controlled. Other regions, such as the Farasan Islands in southern Saudi Arabia (Hagan, 2006) and the Dahlac Islands in Eritrea (Teclमाराम et al., 2009) have

important nesting sites, but are difficult for researchers to access due to political conditions in the region.

Daily mean temperatures were plotted from May 30<sup>th</sup>-September 20<sup>th</sup>, 2018. Unfortunately, no temperature loggers were retrieved from Small Gobar Island, so we had to omit this site from the comparison.

#### 2.3.4 Daily temperature fluctuation

Many publications use constant temperature incubation experiments to identify important parameters such as pivotal temperature, transitional range of incubation, and thermal thresholds. Assessing the temperature fluctuations can identify if constant incubation experiments are applicable to natural nests in the Red Sea region. Mean daily temperature fluctuations over time were calculated by subtracting the minimum recorded temperature from the maximum for each sand logger for each day. The daily fluctuation was plotted for each study site at each depth (30 and 50 cm) from May 30<sup>th</sup> to September 20<sup>th</sup>. Mean temperature fluctuation from the entire study duration was also calculated.

#### 2.3.5 Daily temperature profiles

After analyzing daily temperature fluctuations over time, the sand temperature profile over the course of a single day was evaluated. Temperature measurements from 0:00 (midnight) until 23:45 at each depth and site were extracted from June 1<sup>st</sup>, July 1<sup>st</sup>, and August 1<sup>st</sup>, except for Ras Baridi which the 3<sup>rd</sup> of each month were chosen because loggers were not yet deployed on June 1<sup>st</sup>. These three months were chosen because

June was generally the coolest month, and then the subsequent months displayed an increase in temperature. The thermal profiles of each site and depth were plotted to identify the warmest and coolest time of the day.

#### 2.3.6 Temperature increase from June to September

The temperature increase from June and September was assessed to determine how a shift of nesting season could impact the temperature profile of the nest. The temperature increase between June and September was calculated by compiling logger data and finding the mean value for each depth at each site. This mean value from June was then subtracted from the mean temperature in September.

#### 2.3.7 Sand temperature exceeding 33 and 35°C

The percentage of days with recorded temperatures greater than 33 and 35°C was calculated for each depth. These two temperature values are commonly used as the maximum thermal range of successful incubation. To calculate this percentage, the number of days that recorded temperatures that exceeded these values were counted for each site by depth, and then this value was divided by the total number of days (105). This analysis was done between June 3<sup>rd</sup> and September 16<sup>th</sup>, which are the dates that obtain data from each site.

#### 2.4 Fine Scale patterns in temperature

The fine scale patterns of temperature were assessed to identify temperature profile differences within a single study site. This information could provide insight on

the importance of nest location selection for female turtles at a single nesting site. Data was selected for dates corresponding to the maximum duration of loggers which were deployed and successfully collecting data. At each site and depth, the daily mean temperature from each logger was plotted over time. Next, the temperature profile of each site was analyzed to assess if they differed significantly from each other, with comparisons made between loggers at the same depth. The data did not meet the ANOVA assumption of normality of residuals and were non-parametric, thus, Mann-Whitney U tests were used to assess significance between two loggers at different stations, and Kruskal-Wallis tests were used to compare data between three or more locations.

To identify the relationship between sand and water temperature, correlation and regression analyses were conducted. Mean daily temperature and daily minimum and maximum values over the dates of deployment were plotted, separated by depth. If water temperature was available, it was plotted to compare the temperature profiles between sand and water.

## 2.5 Hatchling sex ratio prediction

To predict the sex ratios, a maximum likelihood mathematical model developed by Girondot (1999) was used, referred to as the Hill equation. This equation describes the relationship between pivotal temperature ( $P$ ), the value that produces equal

number of each sex, the incubation temperature measured in the sand ( $t$ ), and hatchling sex ratios ( $sr$ ):

$$sr(t) = \frac{1}{1 + e^{\frac{1}{S}(\ln(P+K) - \ln(t+K))}}$$

where  $sr(t)$  is the sex ratio of females, at a specific temperature,  $t$ .  $S=-0.0336281$ ,  $P=29.2$ , and  $K=0.1$ . Mean weekly temperature was used at the  $t$  value. The  $S$  parameter defines the shape of transition from masculinizing to feminizing temperature. Since there have not been any turtle incubation experiments in the Red Sea region,  $S$  and  $K$  values were based on theoretical values recorded in laboratory studies (Laloë et al., 2014). This equation has been used to estimate sex ratios at nesting sites in the Cape Verde Islands (Laloë et al., 2014), Georgia, USA (LeBlanc et al., 2012a), Texas, USA (LeBlanc et al., 2012b) and Ascension Island (Godley et al., 2002). The pivotal temperature value of 29.2°C was used because it appears to be illustrative for all species of sea turtles (Ackerman, 1997). Thus, these sex ratios are rough estimates, but notably are not based on data from Red Sea turtle populations.

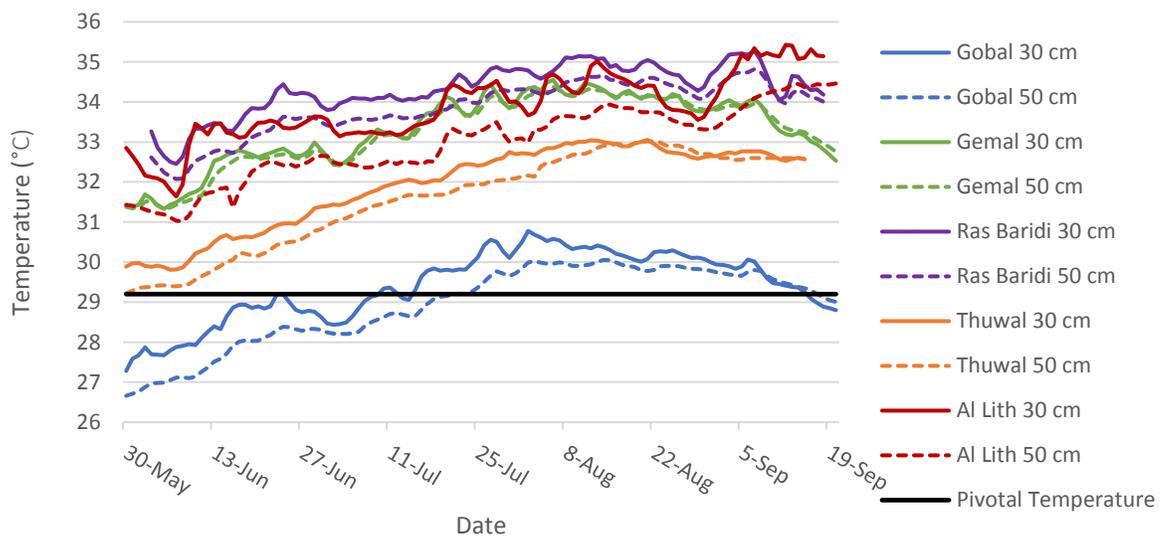
### 3. Results

#### 3.1 Sand temperature comparison between study sites

##### 3.1.1 Comparison of mean sand temperatures

A comparison between the five study sites from May 30<sup>th</sup>-September 19<sup>th</sup>, 2018 was conducted to assess the sand temperature patterns in the Red Sea (Fig. 4). Overall, temperatures recorded at 30 cm were warmer compared to the loggers buried at 50 cm in the same location. All study sites except Small Gobal Island had sand temperatures well above the PT (29.2°C), which suggests that there could have been a high ratio of

female sea turtles produced during the 2018 nesting season. Ras Baridi demonstrated the highest sand temperatures, with a maximum measurement of 36.0°C at 30 cm depth, and 35.3°C at 50 cm. This was followed by Marmar Island in Al Lith, the southernmost study site, which had a maximum temperature of 35.9°C at 30 cm, and 34.5°C at 50 cm depth. Wadi el Gemal had similar temperatures to Al Lith, with a maximum temperature of 35.0°C at 30 cm and 34.1°C at 50 cm depth. Furthermore, the Thuwal study site had sand temperatures consistently cooler than Ras Baridi, Wadi el Gemal, and Al Lith, with a maximum temperature of 33.5 °C at 30 cm depth, and 33.0 °C at 50 cm depth. Finally, the lowest sand temperatures were recorded in the northernmost study site, Small Gobal Island, which had a maximum of 31.0°C at 30 cm, and 30.5 °C at 50 cm depth.



**Figure 4.** Average temperature between May 30<sup>th</sup> and September 19<sup>th</sup>, 2018 at the five different study sites. Measurements at 30 cm depth (solid line) and 50 cm depth (dashed line) are compared. Pivotal temperature (29.2°C) is depicted as the black line. At this temperature, it is presumed that 50% of hatchlings will be born male and female.

### 3.1.2 Comparison of monthly mean temperatures

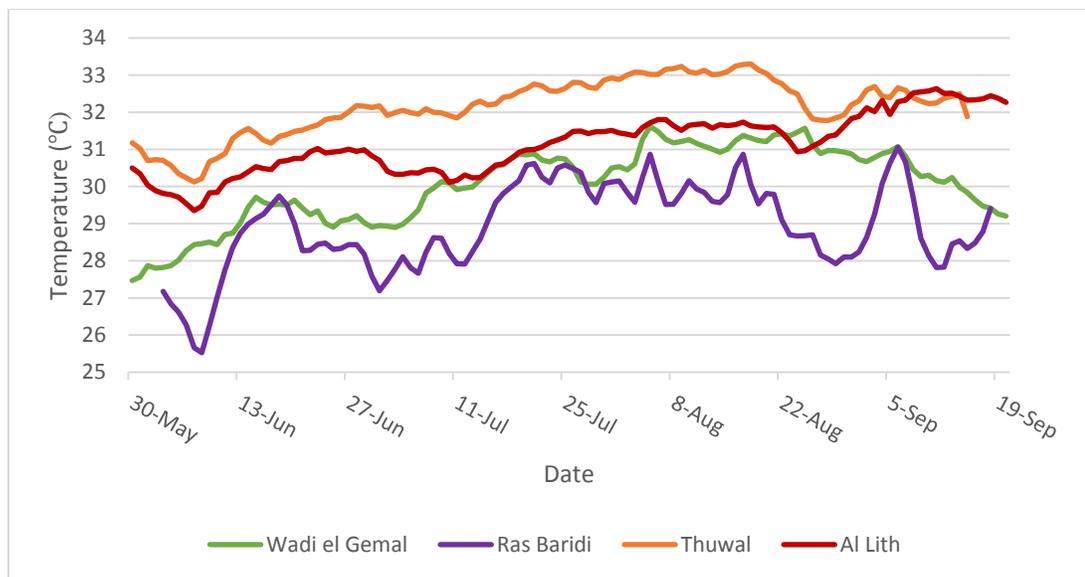
Monthly mean temperatures indicated that August was consistently the hottest month for sand temperature at all five study sites, except Marmar Island in Al Lith, which had a temperature increase in September at both 30 and 50 cm depth (Table 2). Wadi el Gemal, Ras Baridi, and Al Lith had monthly mean values that far exceeded the PT throughout the duration of the study. Small Gobal Island was the only study site that had evidence of hawksbill turtle nesting during logger deployment dates. Hawksbill turtles have shallower nests compared to green sea turtles, at about 30 cm depth. At this depth, the mean temperature in June was below PT, whereas July and August had warmer means, and September had an average temperature almost exactly at PT. Sandy Island in Thuwal had temperatures below PT in May, but then exceeded PT in June through September.

**Table 2.** Mean monthly sand temperatures between study sites at 30 and 50 cm depth. In general, temperatures start off cooler and then get warmer until August, and then decrease in September. Mean temperatures exceeding the pivotal temperature (PT) are in red, temperatures below PT are blue, and temperatures  $\pm 0.3^\circ\text{C}$  of the PT are green.

	May		June		July		August		September	
	30 cm	50 cm	30 cm	50 cm	30 cm	50 cm	30 cm	50 cm	30 cm	50 cm
<b>Small Gobal Island</b> N 27°40'49.31" E 33°48'13.92"	-	-	28.4 $\pm$ 0.5	27.7 $\pm$ 0.6	29.5 $\pm$ 0.7	28.9 $\pm$ 0.6	30.3 $\pm$ 0.3	29.9 $\pm$ 0.4	29.2 $\pm$ 0.5	29.3 $\pm$ 0.5
<b>Wadi el Gemal</b> N 24°39'51.59" E 35°06'33.09"	-	-	32.3 $\pm$ 0.6	31.7 $\pm$ 0.5	33.5 $\pm$ 0.6	32.8 $\pm$ 0.5	34.2 $\pm$ 0.3	33.6 $\pm$ 0.2	33.4 $\pm$ 0.6 (Sept 1-21)	33.1 $\pm$ 0.4 (Sept 1-21)
<b>Ras Baridi</b> N 24°16'24.33" E 37°37'35.3"	-	-	33.6 $\pm$ 0.7 (June 2-30)	32.9 $\pm$ 0.7 (June 2-30)	34.3 $\pm$ 0.5	33.8 $\pm$ 0.5	34.8 $\pm$ 0.5	34.4 $\pm$ 0.4	34.7 $\pm$ 0.6 (Sept 1-19)	34.4 $\pm$ 0.5 (Sept 1-19)
<b>Thuwal</b> N 22°15'9.3" E 39°01'40.2"	29.0 $\pm$ 0.7 (May 5-31)	28.5 $\pm$ 0.6 (May 5-31)	30.4 $\pm$ 0.6	29.9 $\pm$ 0.6	32.0 $\pm$ 0.4	29.9 $\pm$ 0.6	32.8 $\pm$ 0.4	32.3 $\pm$ 0.4	32.7 $\pm$ 0.4 (Sept 1-17)	32.3 $\pm$ 0.4 (Sept 1-17)
<b>Al Lith</b> N 19°50'17.8" E 39°55'38.9"	32.8 $\pm$ 0.7 (May 3-31)	31.1 $\pm$ 0.6 (May 3-31)	33.0 $\pm$ 0.7	31.8 $\pm$ 0.8	33.7 $\pm$ 0.5	33.1 $\pm$ 0.3	34.2 $\pm$ 0.5	33.6 $\pm$ 0.2	35.1 $\pm$ 0.4 (Sept 1-21)	34.1 $\pm$ 0.4 (Sept 1-21)

### 3.1.3 Comparison of water temperature

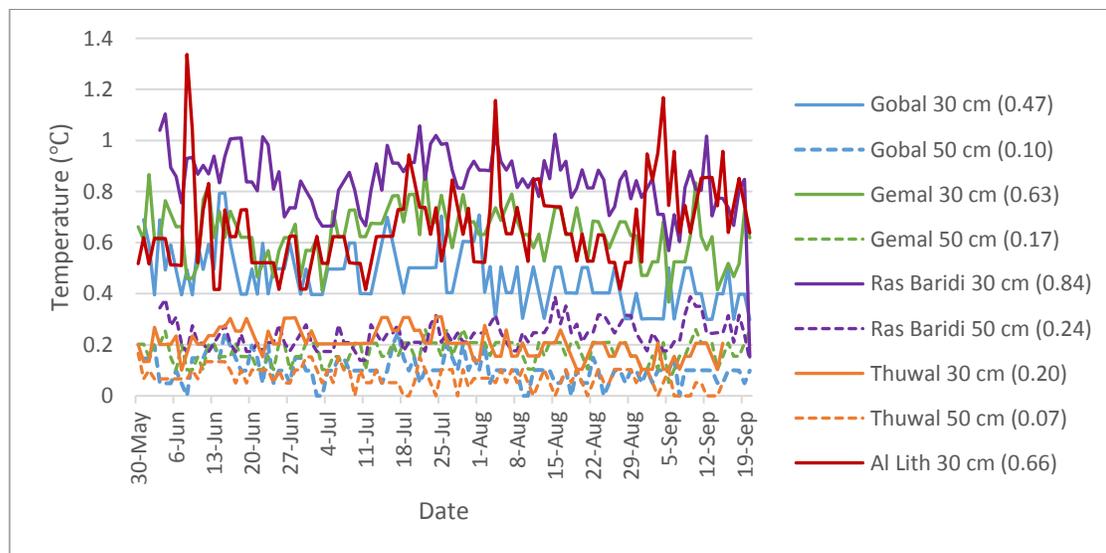
Mean water temperatures recorded at 5 meters depth at each site were plotted over time (Fig. 5). Daily means consisted of data from three loggers, four loggers, four loggers, and one logger from Wadi el Gemal, Ras Baridi, Thuwal, and Al Lith, respectively. No ocean loggers could be retrieved from Small Gobal Island, so this study site was omitted from this comparison. The results showed that the lowest sea temperature was recorded at Ras Baridi, which had the highest sand temperature (Fig. 4). Inversely, Thuwal demonstrated the warmest water temperature (Fig. 5) while having the second coldest sand temperatures (Fig. 4). Thus, the sites with the warmest sand temperature did not also have the warmest water temperature, and vice versa. In addition, the water temperatures also did not show a latitudinal trend as expected.



**Figure 5.** Average water temperature recorded at 5 meters depth from May 30<sup>th</sup>-September 20<sup>th</sup>, 2018. Wadi el Gemal contains data from three loggers, Ras Baridi and Thuwal contain data from four loggers, and Al Lith contains data from one logger.

### 3.1.4 Daily temperature fluctuation

The average temperature fluctuation varied daily and at different amplitudes between the different study sites (Fig. 6). Al Lith temperature fluctuations at 50 cm were omitted from the data due to extreme outliers. At all sites, the daily fluctuations were larger at 30 cm compared to 50 cm. Ras Baridi, the site with the highest sand temperatures, also exhibited the highest mean daily fluctuations at both 30 cm depth and 50 cm. The mean temperature fluctuations at 30 cm depth over the study duration exhibited higher fluctuations at the sites with higher sand temperature. The exception was the coldest site, Small Gobal Island, which had a larger daily fluctuation than Thuwal. It is also important to note that Al Lith demonstrated spontaneous spikes in temperature fluctuation (red line) over the studied time period. The daily fluctuations (as high as 1.6°C) suggest that parameters identified with constant incubation experiments may not be applicable to natural turtle nests in this region.

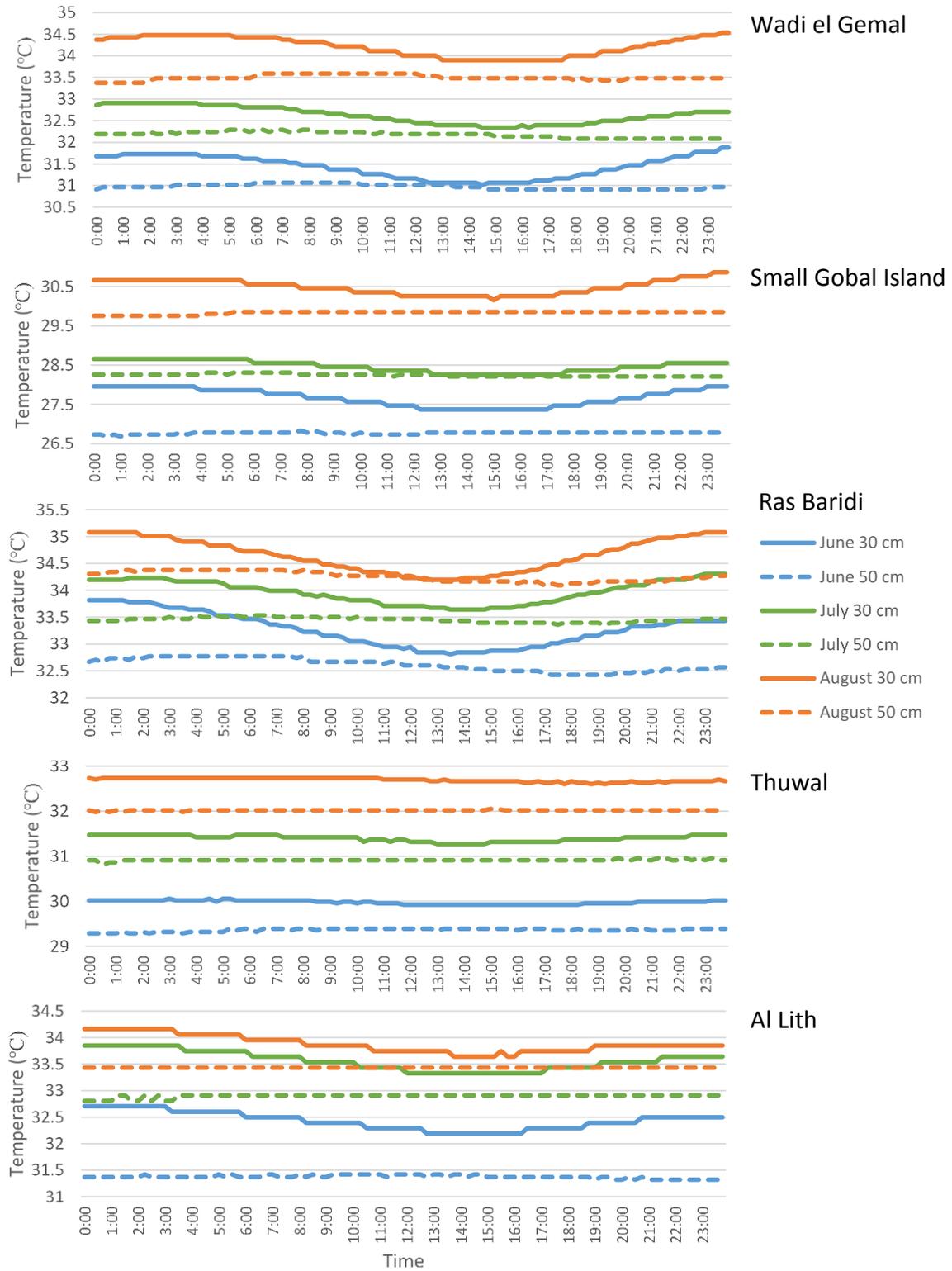


**Figure 6.** Average daily temperature fluctuations between May 30<sup>th</sup> and September 19<sup>th</sup>, 2018 at the five different study sites. Measurements at 30 cm depth (solid line) and 50 cm depth (dashed line) were compared. The mean daily temperature fluctuation over the study duration noted in parenthesis in the legend.

### 3.1.5 Daily temperature profiles

To assess the daily temperature profile at each site, 24-hour thermal profiles on June 1<sup>st</sup>, July 1<sup>st</sup>, and August 1<sup>st</sup> were compared (Fig. 7). When comparing the temperature profiles between the different dates at the same depth, the temperature fluctuations follow a similar trend, despite the temperature increasing by month. The temperature profiles were slightly different at each site. For example, at Ras Baridi, the warmest site, the profile at 30 cm depth showed that the temperature was lowest at around 13:00, and then warmed until 21:00, when it stabilized and stayed high until 02:00. Afterwards, from 02:00-13:00, temperatures gradually decreased. Alternatively, at Small Gobal Island, the coolest site, the temperature profile at 30 cm depth had a smaller temperature fluctuation throughout the day. At this site, the temperature was lowest from 12:00 until 17:00, and then increased until about 02:00, when it steadied off until 05:00 and began to drop in a stepwise pattern. As the results of the mean daily fluctuations suggested, Thuwal exhibited the smallest change of temperature within a single day. The temperature at this site stayed relatively constant throughout the day.

All sites exhibited a maximum temperature at night/early morning, and a minimum temperature in the afternoon. This shows that the sand temperatures displayed a lag of heating and cooling. This study did not focus specifically on understanding the thermodynamics of sand, and further studies should be conducted to examine these temperature patterns.

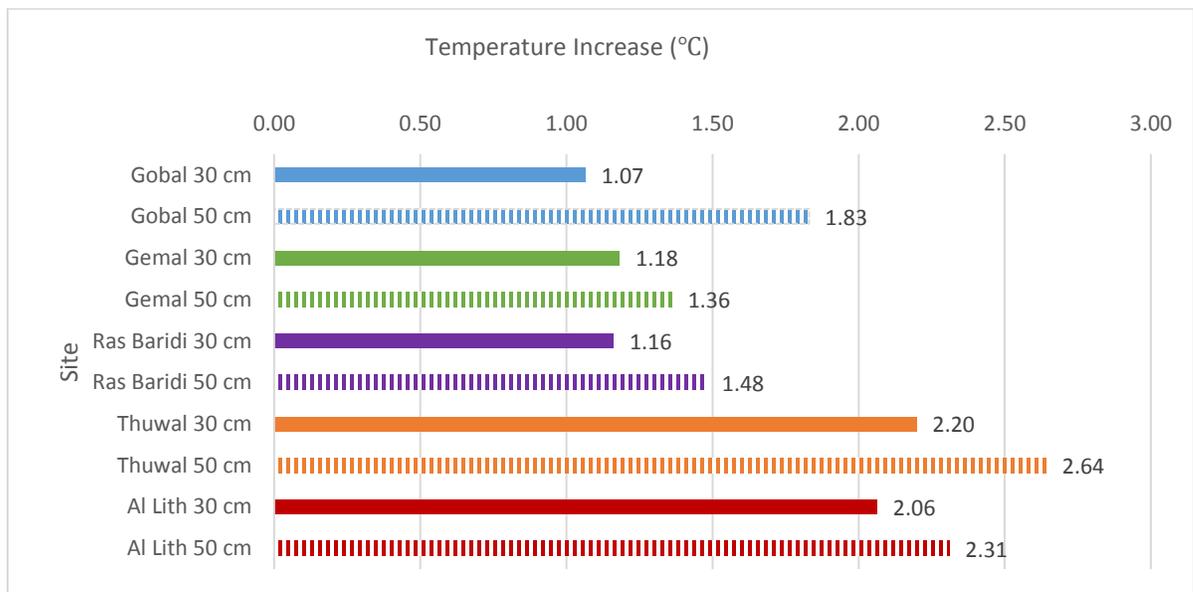


**Figure 7.** Daily thermal profiles from Small Gobal Island, Wadi el Gemal, Ras Baridi, Thuwal, and Al Lith during a 24-hour period. Temperatures between 30 cm (solid line) and 50 cm (dashed line) are compared between June (blue), July (green), and August (orange).

### 3.1.6 Temperature increase from June to September

The temperature increase from June to September was assessed at all sites for both 30 and 50 cm depth. Temperature increase was greater at 50 cm depth when compared to 30 cm depths at all sites (Fig. 8). This trend was most evident at Small Gobal Island, which demonstrated a 1.1°C increase at 30 cm, and a 1.8°C increase at 50 cm depth.

Despite Ras Baridi having the highest sand temperature of all the sites, it had a relatively small increase when it was compared to the coolest month (June), with an increase of 1.2°C at 30 cm, and 1.5°C at 50 cm depth. In comparison, Thuwal had the second coolest sand temperature overall during the months of deployment but exhibited the largest increase between June and September of 2.2°C at 30 cm depth, and 2.6°C at 50 cm depth.

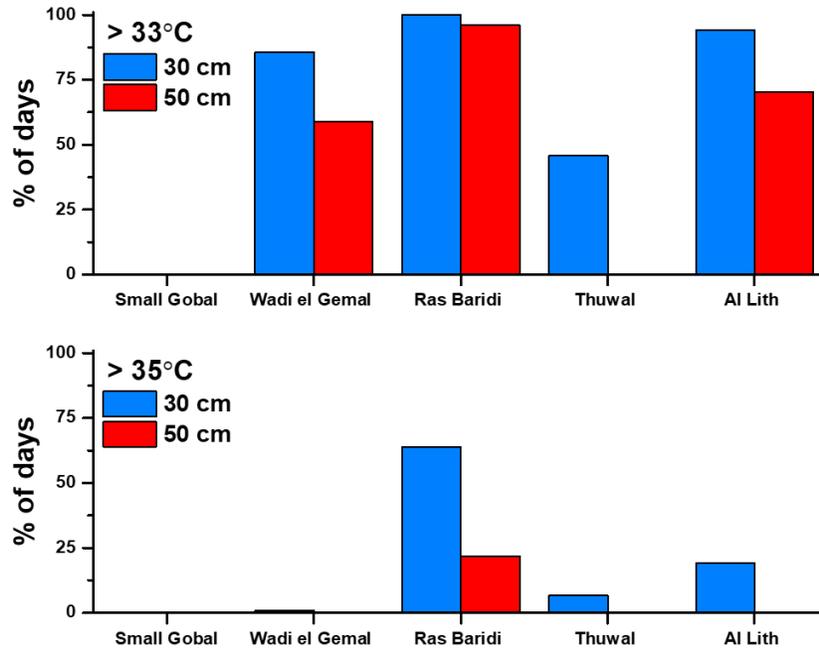


**Figure 8.** Temperature increase between mean June and September measurements at all sites, comparing values from 30 and 50 cm depth.

### 3.1.7 Sand temperature exceeding 33 and 35°C

The next analysis calculated the percentage of days between June 3<sup>rd</sup> and September 15<sup>th</sup> that had temperature recordings exceeded 33 and 35°C (Fig. 9). These two values are commonly cited in literature as the maximum thermal tolerance for successful incubation (Miller, 1997; Ackerman, 1977), while the minimum thermal threshold has been identified as 25°C (Howard et al., 2014). Importantly, Small Gobal Island had zero days crossing the thermal threshold limits for the time period studied, whereas Ras Baridi had 100% and 96% of the days where temperatures exceeded 33°C at 30 and 50 cm, respectively (Fig. 9).

The results illustrated in Fig. 9 show that the thermal threshold of 33°C is largely surpassed by the majority of sites at both 30 and 50 cm depth for at least 50 percent of the days studied (except for Small Gobal Island and Thuwal at 50 cm depth). However, if 35°C is considered as the thermal threshold for successful incubation, then only Ras Baridi faces a high threat with nearly 60% and 25% of the days exceeding 35°C at 30 and 50 cm depth, respectively. Furthermore, both Al Lith and Thuwal exhibit a small percentage of days with temperature exceeding 35°C at 30 cm depth (11% and 19%, respectively). These results illustrate a clear threat to both hawksbill and green sea turtles at our study sites, suggesting that green turtles at Ras Baridi are already facing threats of extreme high temperatures. In addition, turtles nesting at Wadi el Gemal, Thuwal, and Al Lith might also exhibit mortality from the high sand temperatures.



**Figure 9.** Percentage of days between June 3<sup>rd</sup> and September 15<sup>th</sup>, 2018 that recorded temperatures exceeding 33°C (top) and 35°C (bottom), these two temperatures are often considered the maximum thermal threshold of successful incubation for sea turtles. Comparison was made between temperatures recorded at 30 cm (blue) and 50 cm (red).

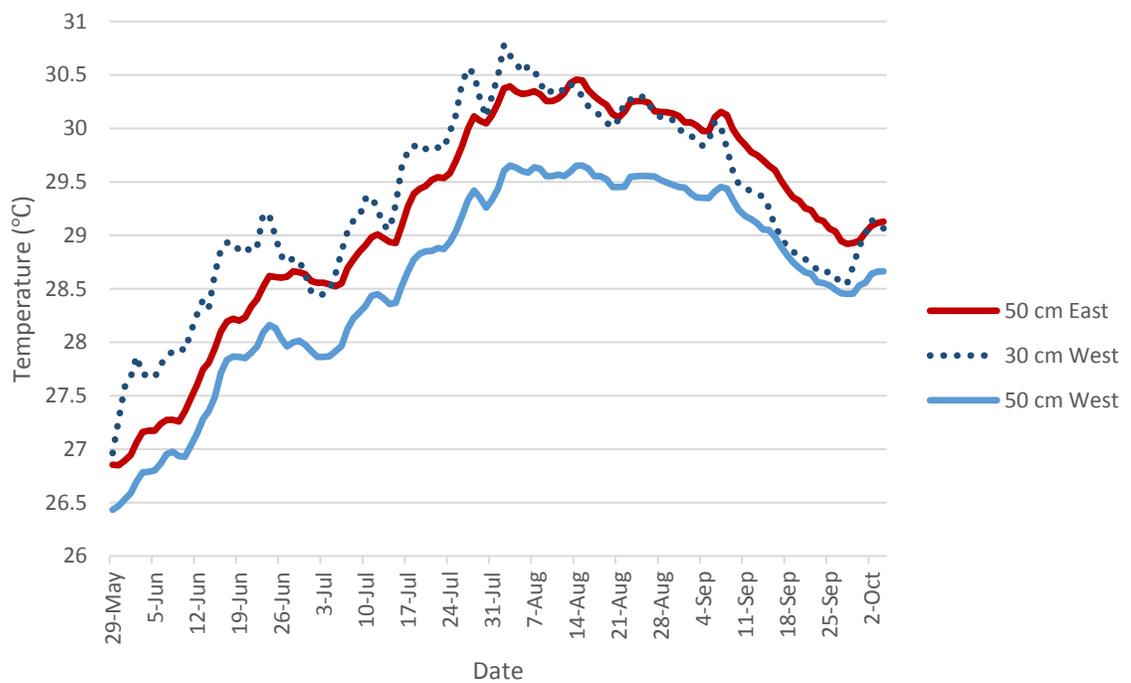
### 3.2 Fine-scale patterns in temperature

#### 3.2.1 Small Gobal Island



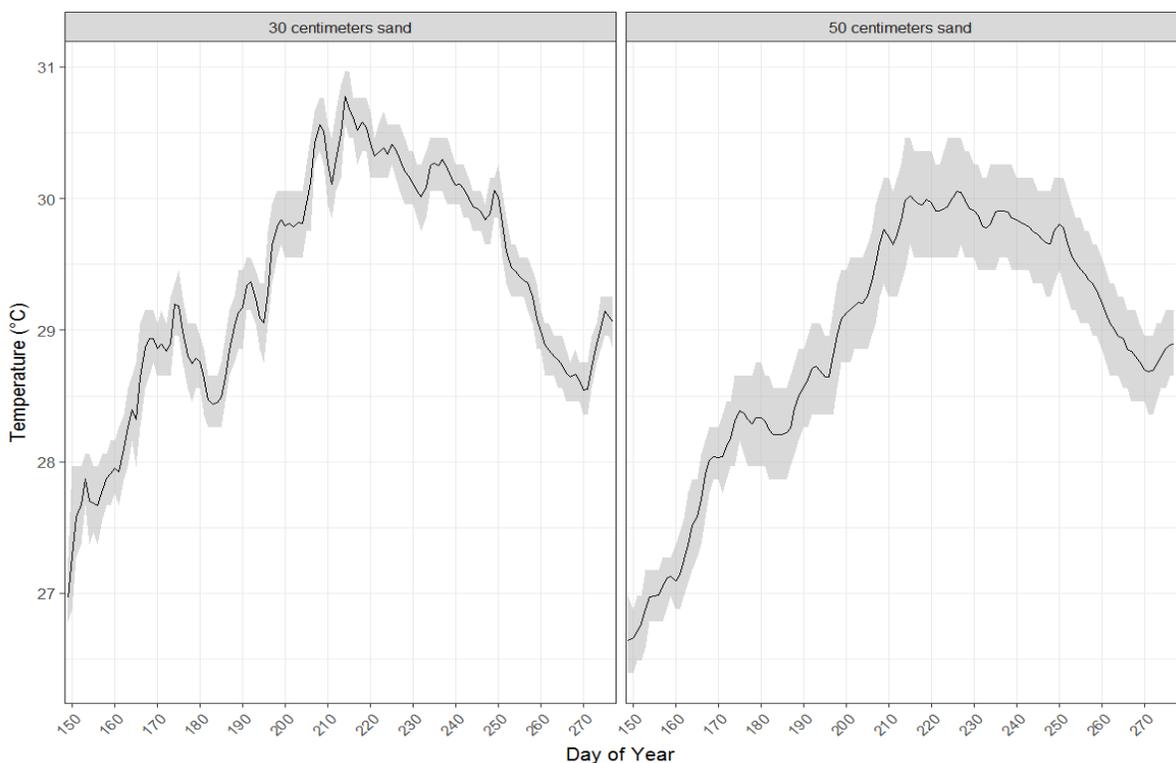
**Figure 10.** Two locations of sand temperature loggers at the Small Gobal Island study site. Source: "Small Gobal Island, Egypt". 27°40'45.19" N and 33°48'09.39" E. **Google Earth Pro V 7.3.2.5491.** June 23, 2016. October 14, 2018.

Temperature loggers on Small Gobal Island were deployed from May 29<sup>th</sup> until October 5<sup>th</sup>, 2018. Three out of eight sand temperature loggers were retrieved from two stations (East and West) (Fig. 10). During the 2018 nesting season, there were ten recorded hawksbill turtle *Eretmochelys imbricata* nests on this beach (I. Elsadek, personal communication, October 2018). At 50 cm depth, the East station temperatures were significantly higher than the West station ( $U=5313$ ,  $p<0.001$ ) (Fig. 11). The East station was as much as 0.8°C warmer compared to the West station at 50 cm, which occurred in August. All throughout the duration of the study, the East site was warmer than temperature at the West site at 50 cm depth.



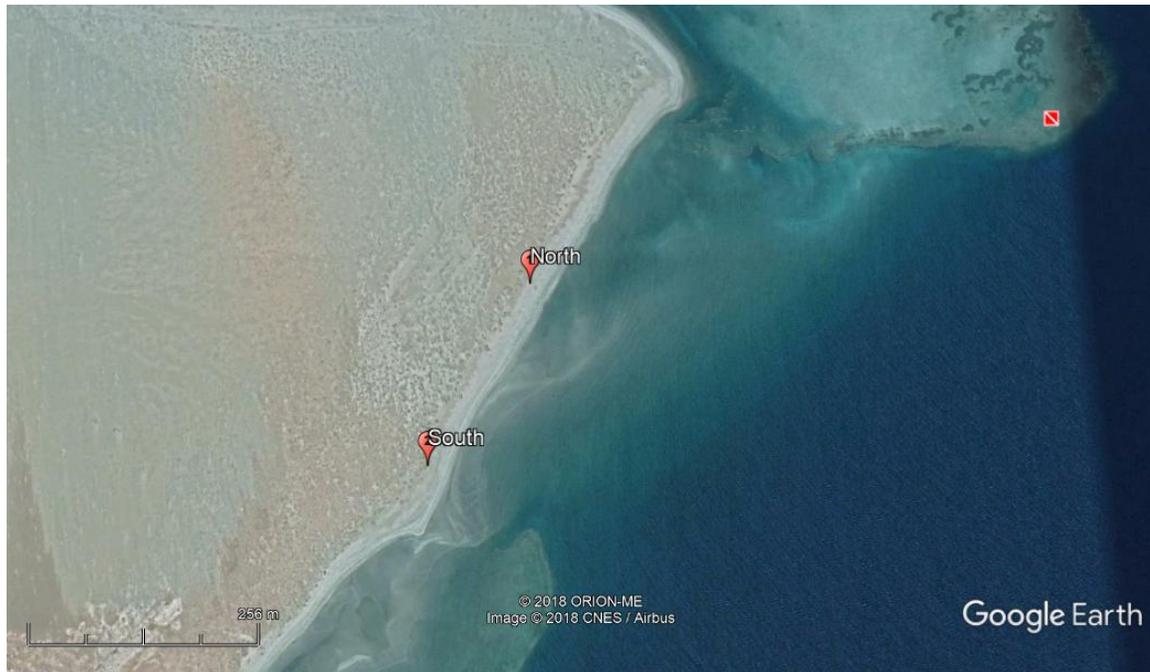
**Figure 11.** Temperature measurements between May 29<sup>th</sup>-October 4<sup>th</sup>, 2018 at the East and West site on Small Gobal Island, Egypt. The median temperatures are 29.35°C, 29.25°C, and 28.66°C at 50 cm East, 30 cm West, and 50 cm West, respectively.

At 30 and 50 cm depth, temperature measurements were the lowest when loggers were first deployed in May (day 150) with temperatures at 27°C and 26.5°C, respectively, and then increased until they peaked at ~30.5°C and 30°C in the end of July (day 210). The mean sand temperature then decreased to under 29°C and 28.5°C in late September (day 270) (Fig. 12). The range of maximum and minimum is larger at 50 cm, which could be because the data is from two loggers, whereas the 30 cm plot only includes data from one logger. Unfortunately, none of the in-water temperature loggers were retrieved. The loggers might have been removed from the reef due to the high density of scuba divers in this area.



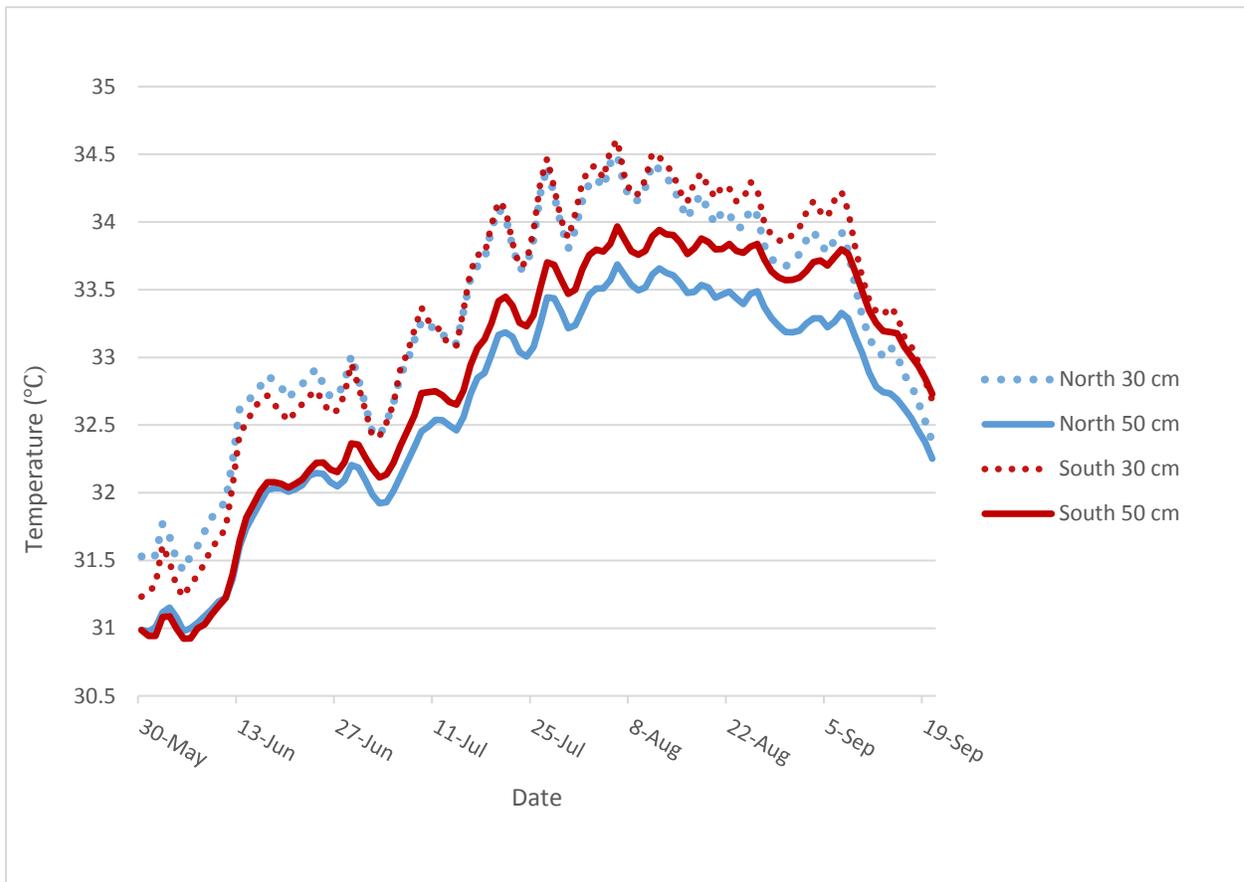
**Figure 12.** The mean hawksbill sea turtle *Eretmochelys imbricata* nesting beach sand temperature (black line) at 30 cm (left) and 50 cm (right) between May 29<sup>th</sup> and October 4<sup>th</sup> on Small Gobal Island, Egypt. The turtles are expected to bury their eggs at about 30 cm on Small Gobal Island. The shaded grey areas represent the range of minimum and maximum temperatures. The 30 cm graph contains data from one logger, whereas the 50 cm graph contains data from two loggers.

### 3.2.2. Wadi el Gemal



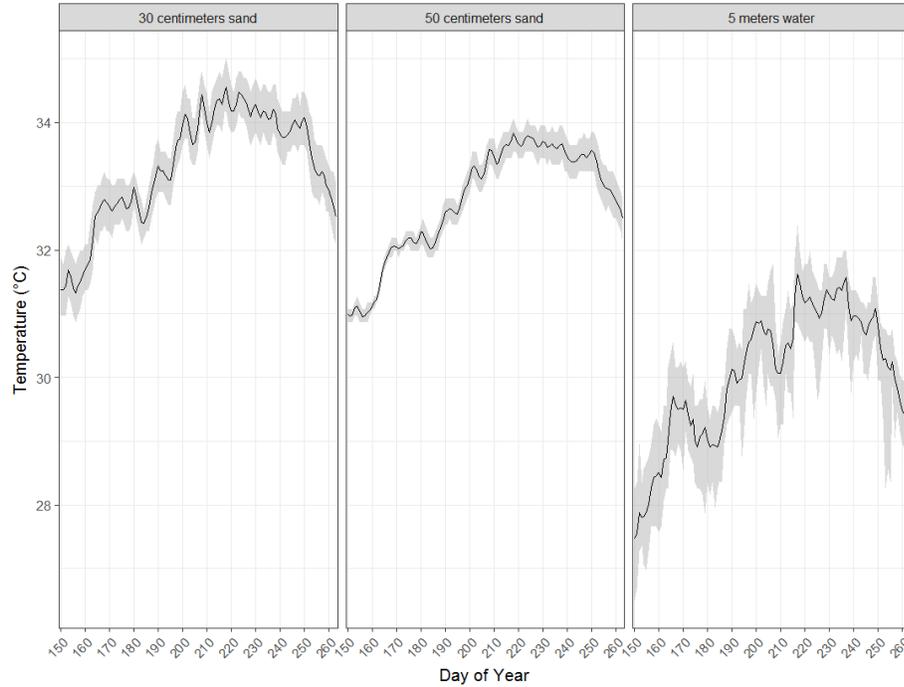
**Figure 13.** Two locations of sand temperature loggers at the Wadi el Gemal study site. Source: "Wadi el Gemal, Egypt". 24°15'35.87" N and 37°36'32.24" E. **Google Earth Pro V 7.3.2.5491**. July 10, 2018. October 14, 2018.

At the Wadi el Gemal study site, temperature loggers were deployed from May 30<sup>th</sup> to September 20<sup>th</sup>, 2018. Four out of eight sand loggers were retrieved from two stations (North and South). Temperature recorded at 30 cm are not significantly different between the North and South logger stations ( $U=6096$ ,  $p=0.420$ ) (Fig. 14). Conversely, daily temperatures at 50 cm are significantly higher at the South station compared to the North station ( $U=4283$ ,  $p=0.002$ ). The South station was as much as 0.5°C warmer compared to the North station at 50 cm, which occurred in September. At 30 cm, the temperature was as much as 0.3°C warmer at the South station compared to the North station.



**Figure 14.** Temperature measurements between May 30th-September 20th, 2018 at the North and South logger stations at Wadi el Gemal, Egypt. The median temperatures are 33.43°C, 32.81°C, 33.54°C, and 33.22°C at 30 and 50 cm North, and 30 cm and 50 cm South, respectively.

The water temperature at 5 m depth followed similar heating and cooling patterns to the sand temperature profiles, although the water temperature was on average  $3.3^{\circ}\text{C} \pm 0.1$  cooler than from 30 cm sand depth sand, and  $2.7^{\circ}\text{C} \pm 0.4$  cooler than 50 cm sand depth. Correlation and regression analyses were conducted to examine the relationship between sea and sand temperature. Mean sea temperature is positively correlated with mean sand temperature at both 30 and 50 cm depth. The regression relationship was stronger at 30 cm depth ( $R^2=0.906$ ,  $F(1, 112)=554.34$ ,  $p < 0.001$ ) than at 50 cm depth ( $R^2=0.891$ ,  $F(1,112)=914.37$ ,  $p < 0.001$ ).



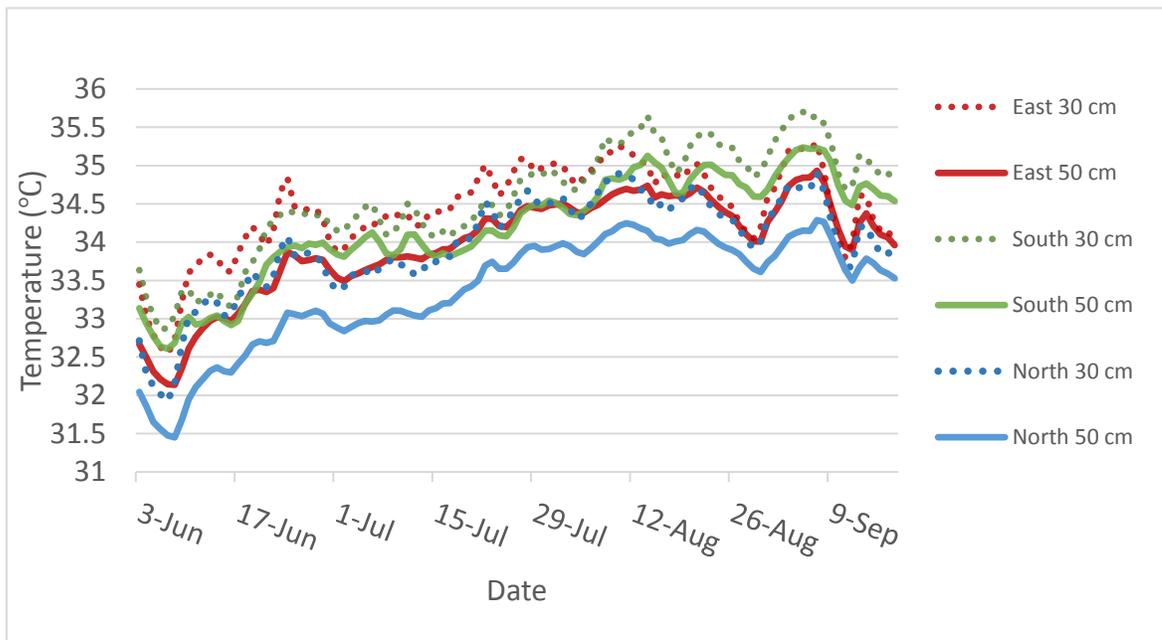
**Figure 15.** The mean green sea turtle *Chelonia mydas* nesting beach sand temperature (black line) at 30 cm (left) and 50 cm depth (middle), and mean water temperature recorded from 5 m depth (right) between May 30<sup>th</sup> and September 20<sup>th</sup> from Wadi el Gemal, Egypt. The turtles are expected to bury their eggs at about 50 cm at Wadi el Gemal. The shaded grey areas represent the range of minimum and maximum temperatures. The 30 and 50 cm graphs contain data from two loggers, and the 5 m water graph contains data from four loggers.

### 3.2.3. Ras Baridi



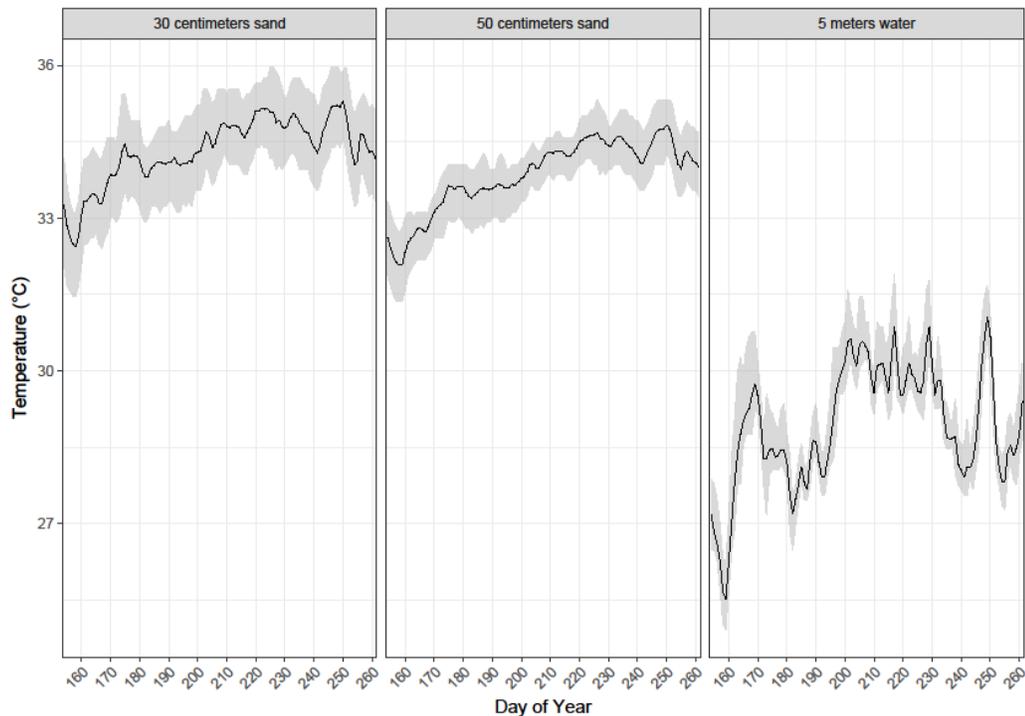
**Figure 16.** Three locations of sand temperature loggers at the Ras Baridi study site. The Yanbu Cement Company can be seen in the northwest section of the map. Source: "Ras Baridi, Saudi Arabia". 24°15'35.87" N and 37°36'32.24" E. Google Earth Pro V 7.3.2.5491. October 21, 2016. October 8, 2018.

Temperature loggers were deployed from June 3<sup>rd</sup>-September 18<sup>th</sup>, 2018 at the Ras Baridi study site. Six out of eight sand temperature loggers were retrieved, at three study stations (North, East, and South) (Fig. 16). The Kruskal-Wallis H test showed that there were significantly different temperatures at 30 cm depth between the three logger locations,  $\chi^2(2)=43.13$ ,  $p<0.001$ , with a mean rank score of 177.5 for the East station, 115.2 for the North station, and 194.8 for the South station (Fig. 17). This test also showed that there were significantly different temperatures at 50 cm depth between the three logger locations,  $\chi^2(2)=65.58$ ,  $p<0.001$ , with a mean rank score of 176.38 for the East station, 105.37 for the North station, and 205.76 for the South station. At 30 cm, the South logger station was as much as 1.1°C warmer than the North location. At 50 cm, the South location was as much as 1.3°C warmer than the North location.



**Figure 17.** Temperature measurements between June 3<sup>rd</sup>-September 18<sup>th</sup>, 2018 at the East, North, and South site of Ras Baridi. The median temperatures are 34.48°C, 34.06°C, 34.06°C, 33.64 °C, 34.59 °C, and 34.27 °C at 30 and 50 cm East, 30 and 50 cm North, and 30 and 50 cm South site, respectively.

The water temperature at 5 m depth followed a similar heating and cooling trend as the sand temperature, although the sea temperatures were markedly cooler. Water temperature recorded at 5 m depth was on average  $5.4^{\circ}\text{C} \pm 0.8$  cooler than from 30 cm sand depth sand, and  $4.9^{\circ}\text{C} \pm 0.9$  cooler than 50 cm sand depth. Correlation and regression analyses were conducted to examine the relationship between sea and sand temperature. Mean sea temperature is positively correlated with mean sand temperature at both 30 and 50 cm depth. The regression relationship was stronger at 30 cm depth ( $R^2=0.516$ ,  $F(1, 106)=113.22$ ,  $p < 0.001$ ) than at 50 cm depth ( $R^2=0.428$ ,  $F(1,106)=79.24$ ,  $p < 0.001$ ). The R value is relatively low at both sand depths, which could be due to the cooler water temperatures compared to sand at this study site.



**Figure 18.** The mean green sea turtle *Chelonia mydas* nesting beach sand temperature (black line) at 30 cm (left) and 50 cm depth (middle), and mean water temperature recorded from 5 m depth (right) between June 3<sup>rd</sup> and September 18<sup>th</sup> at Ras Baridi. The turtles are expected to bury their eggs at about 50 cm at Ras Baridi. The shaded grey areas represent the range of minimum and maximum temperatures. The 30 and 50 cm graphs contain data from three loggers each, and the 5 m water graph contains data from four loggers.

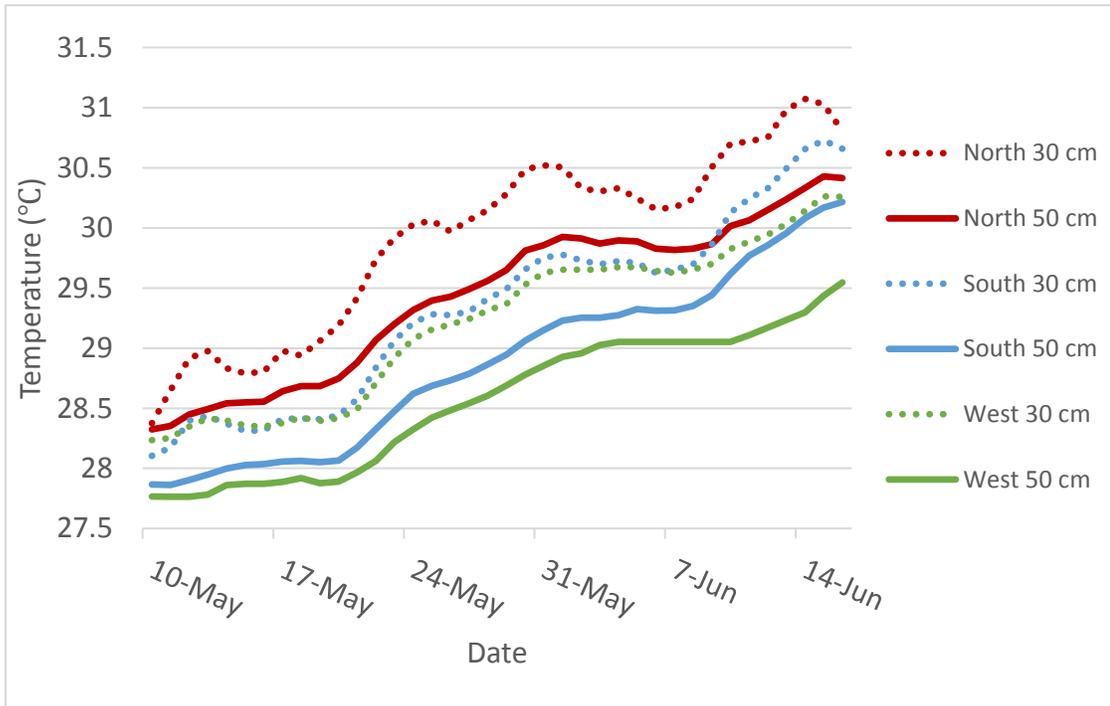
### 3.2.4 Sandy Island, Thuwal



**Figure 19.** Four sand logger locations on Sandy Island, Thuwal. Source: "Sandy Island, Saudi Arabia". 22°15'09.56" N and 39°01'40.77" E. **Google Earth Pro V 7.3.2.5491.** August 14, 2018. October 10, 2018.

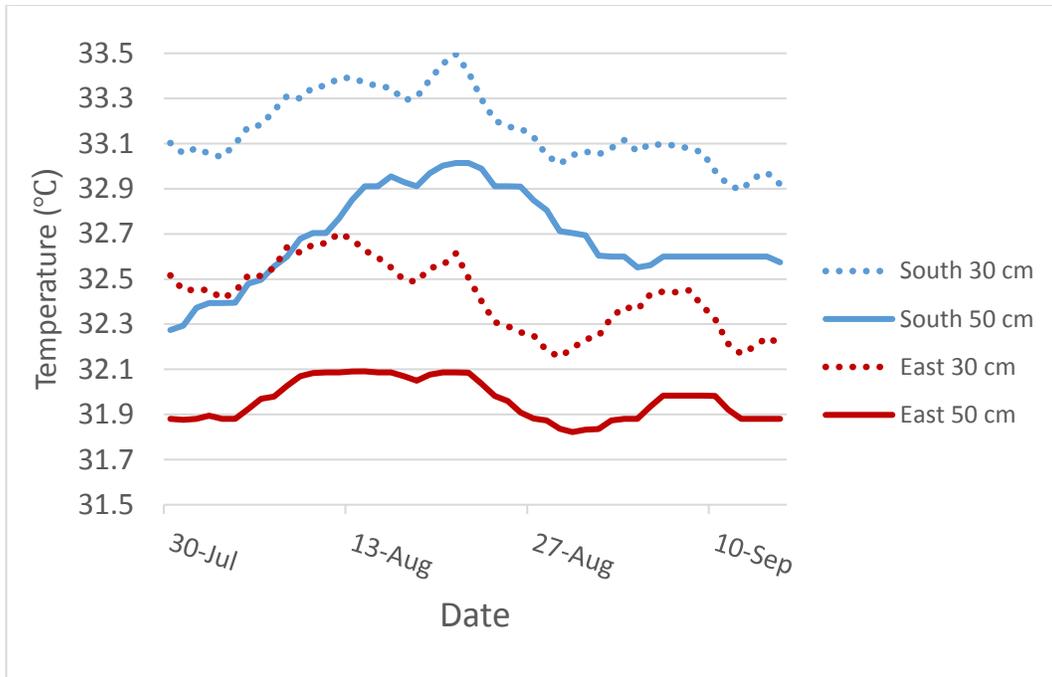
At the Sandy Island, Thuwal site, loggers were deployed from May 9<sup>th</sup> to September 16<sup>th</sup>, 2018 at four different stations (North, East, South, and West) (Fig. 19). Temperature data was collected at 30 and 50 cm depths from the North station (May 9<sup>th</sup>-August 3<sup>rd</sup>), South station (May 9<sup>th</sup>-September 15<sup>th</sup>), West station (May 9<sup>th</sup>-June 16<sup>th</sup>) and the East station (July 30<sup>th</sup>-September 15<sup>th</sup>). Due to the varying dates, two comparisons were conducted. From May 9<sup>th</sup>-June 16<sup>th</sup>, the Kruskal-Wallis H test showed that there were significantly different temperatures at 30 cm depth between the three logger locations,  $\chi^2=18.38$ ,  $p<0.001$ , with a mean rank score of 75.93 at the North site, 51.34 at the South site, and 45.22 at the West site (Fig. 20). Similarly, the Kruskal-Wallis H test also showed that temperatures differed significantly between sites at 50 cm depth,  $\chi^2=25.48$ ,  $p<0.001$ , with a mean rank score of 77.74 at the North site, 55.07 at the South site, and 39.70 at the West site. At 30 cm, the temperature at the North logger

station was as much as 1.0°C warmer than at the West logger station. At 50 cm, the temperature at the North logger station also was as much as 1.0°C warmer than at the West logger station.



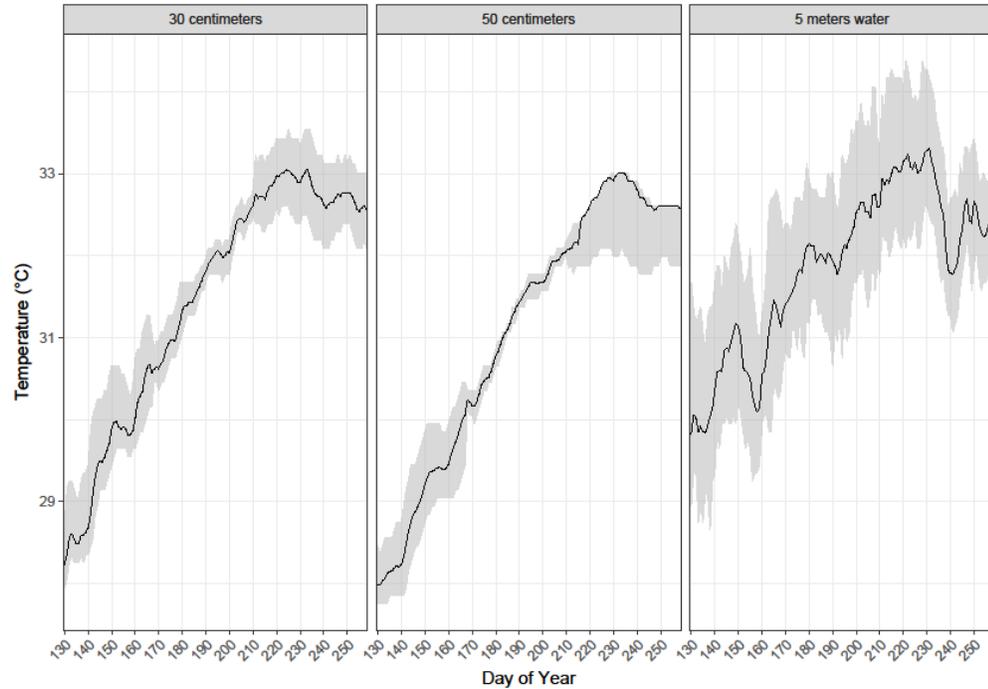
**Figure 20.** Temperature measurements between May 9<sup>th</sup> and June 16<sup>th</sup>, 2018 at the North, South, and West site of Sandy Island, Thuwal. The median temperatures are 30.15°C, 29.56°C, 29.4°C, 28.87 °C, 29.31 °C, and 28.80 °C at 30 and 50 cm depth at the North, South, and West site, respectively.

From July 30<sup>th</sup>-September 15<sup>th</sup>, 2018, temperatures recorded at 30 cm are significantly higher at the South logger location compared to the East ( $U=0$ ,  $p<0.001$ ) (Fig. 21). Similarly, daily temperatures at 50 cm are significantly higher at the South logger location compared to the East ( $U=0$ ,  $p<0.001$ ). At 30 cm, the temperature at the South station was as much as 0.9°C warmer than at the East station. At 50 cm, the temperature at the South station was as much as 1.0°C warmer than at the East logger station.



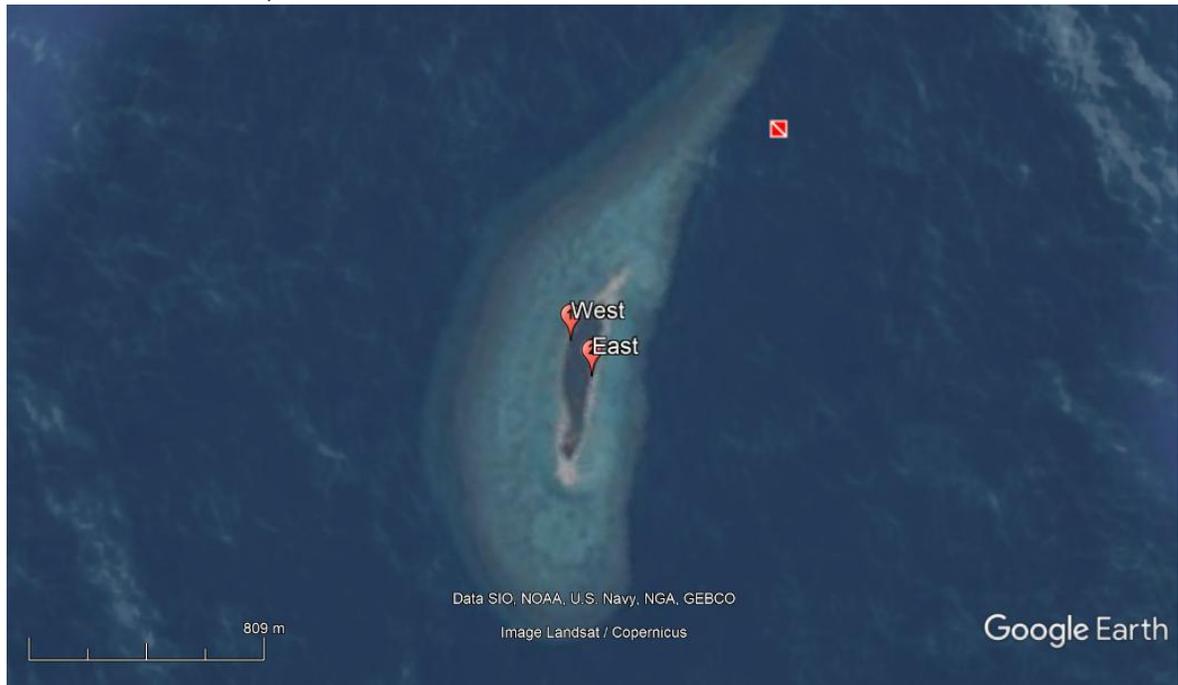
**Figure 21.** Temperature measurements between July 30<sup>th</sup> and September 15<sup>th</sup>, 2018 at the East and South site of Sandy Island, Thuwal. The median temperatures are 32.45°C, 31.96°C, 33.11°C, 32.60°C 30 and 50 cm depth at the East and South site, respectively.

Mean daily temperature measurements from 30 and 50 cm depth in the sand and 5 meters depth in the water were compared between May 9<sup>th</sup> and September 15<sup>th</sup>. Four ocean loggers were retrieved from 5 m depth, which followed similar heating and cooling patterns to the sand temperatures, although the sea temperature had a larger range (Fig. 22). Furthermore, the average water temperature was  $0.4^{\circ}\text{C} \pm 0.6$  warmer than the 30 cm sand depth, and  $0.8^{\circ}\text{C} \pm 0.8$  warmer than 50 cm sand depth. Mean sea temperature is positively correlated with mean sand temperature at both 30 and 50 cm depth, but the relationship is stronger at 30 cm depth ( $R^2=0.894$ ,  $F(1, 127)= 1,068.49$ ,  $p < 0.001$ .) compared to at 50 cm depth ( $R^2=0.841$ ,  $F(1, 127)=673.08$ ,  $p < 0.001$ ).



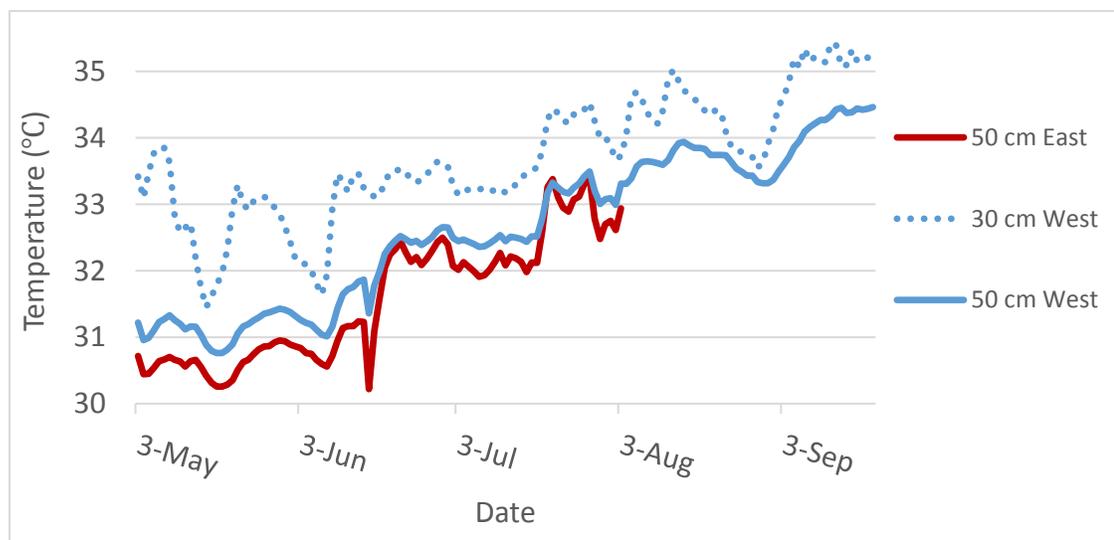
**Figure 22.** The mean sand temperature (black line) at 30 cm (left) and 50 cm depth (middle), and mean water temperature recorded from 5 m depth (right) between May 9<sup>th</sup> and September 16<sup>th</sup> from Sandy Island, Thuwal. The shaded grey areas represent the range of minimum and maximum temperatures. The 30 and 50 cm graphs contain data from 2-3 loggers, and the 5 m water graph contains data from four loggers.

### 3.2.5 Marmar Island, Al Lith



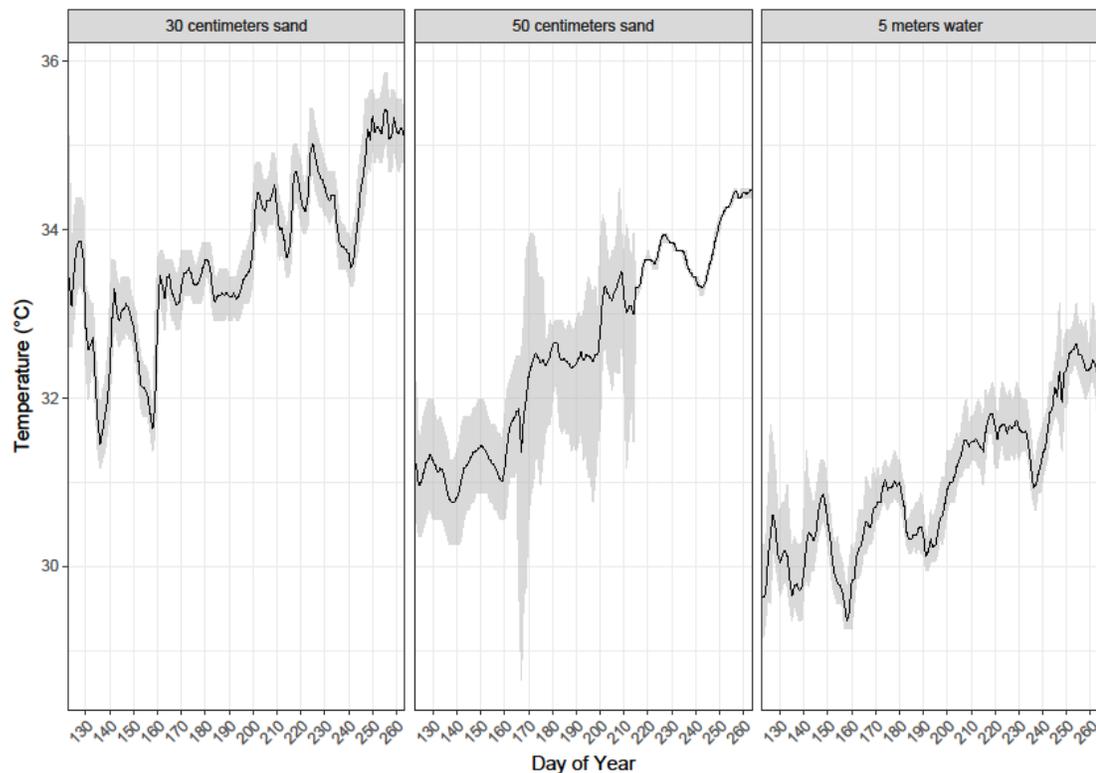
**Figure 23.** Two sand logger locations on Marmar Island, offshore of Al Lith. Source: "Marmar Island, Saudi Arabia". 19°50'17.8" N 39°55'38.9" E. **Google Earth Pro V 7.3.2.5491**. December 15, 2015. October 11, 2018.

Al Lith loggers were deployed on Marmar Island from May 3<sup>rd</sup>-September 20<sup>th</sup>, 2018 and collected data from two stations (East and West) (Fig. 23). There was nesting activity and adult tracks seen all around the circumference of the island, with less in areas with exposed rocks that prevented turtles from reaching the sand. The nests and tracks were primarily located on the northern and southern sandy areas of the island. The loggers from the North station could not be retrieved, which could be due to the high nesting activity. In addition, two loggers were recovered from the South station, but the loggers were displaced from their buried depth, so these data were omitted. Therefore, from May 3<sup>rd</sup>-June 13<sup>th</sup>, three out of eight loggers collected temperature measurements from the East and West stations on Marmar Island (Fig. 24). The temperature at 50 cm was significantly warmer at the West station compared to the East, ( $U=127$ ,  $p < 0.001$ ). At 50 cm depth, the temperature at the West logger station was as much as 1.1°C warmer than at the East logger station.



**Figure 24.** Temperature measurements between May 3<sup>rd</sup> and June 13<sup>th</sup>, 2018 at the East and West site of Marmar. The median temperatures are 30.66°C, 32.90°C, 31.68°C, 30 cm depth at the East site, and 30 and 50 cm depth at the West site, respectively.

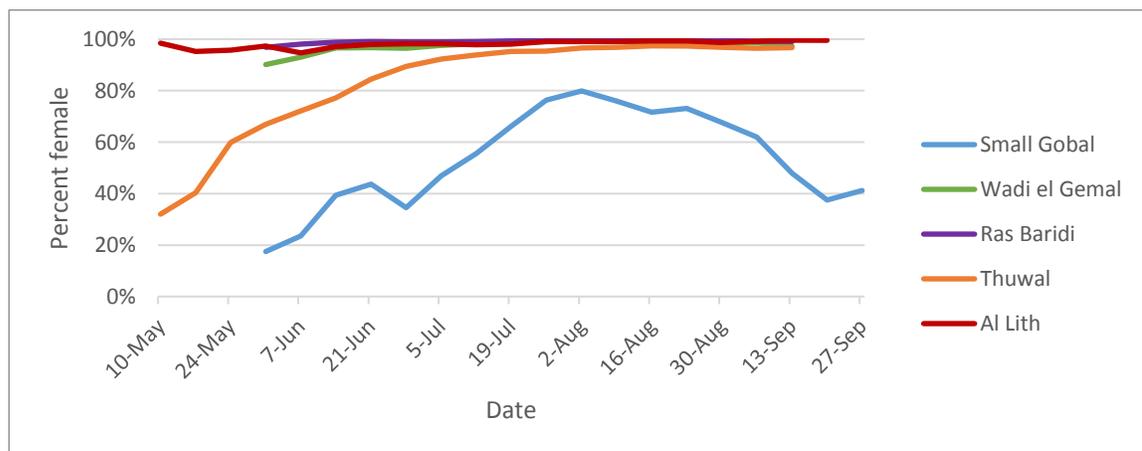
Only one water temperature logger was retrieved Al Lith. From May 9<sup>th</sup> to September 16<sup>th</sup>, the mean water temperature was  $31.0^{\circ}\text{C} \pm 0.8$ . The water temperature profile followed similar trends as the sand temperature but were on average  $2.7^{\circ}\text{C} \pm 0.4$  cooler than sand at 30 cm, and  $1.7^{\circ}\text{C} \pm 0.5$  cooler than sand at 50 cm. Correlation and regression analyses were conducted to examine the relationship between sea and sand temperature. Mean water temperature was positively correlated with mean sand temperature at both 30 and 50 cm depth. The regression relationship was stronger at 30 cm depth ( $R^2=0.848$ ,  $F(1, 139)=774.11$ ,  $p<0.000$ ) than at 50 cm depth ( $R^2=0.844$ ,  $F(1,139)=754.22$ ,  $p<0.000$ ).



**Figure 25.** The mean sand temperature (black line) at 30 cm (left) and 50 cm depth (middle), and mean water temperature recorded from 5 m depth (right) between May 9<sup>th</sup> and September 16<sup>th</sup> on Marmar Island. The shaded grey areas represent the range of minimum and maximum temperatures. The 30 cm graph has data from one logger, the 50 cm graph has data from two loggers until day 210, when one logger was moved, and the 5 m water graph contains data from one logger.

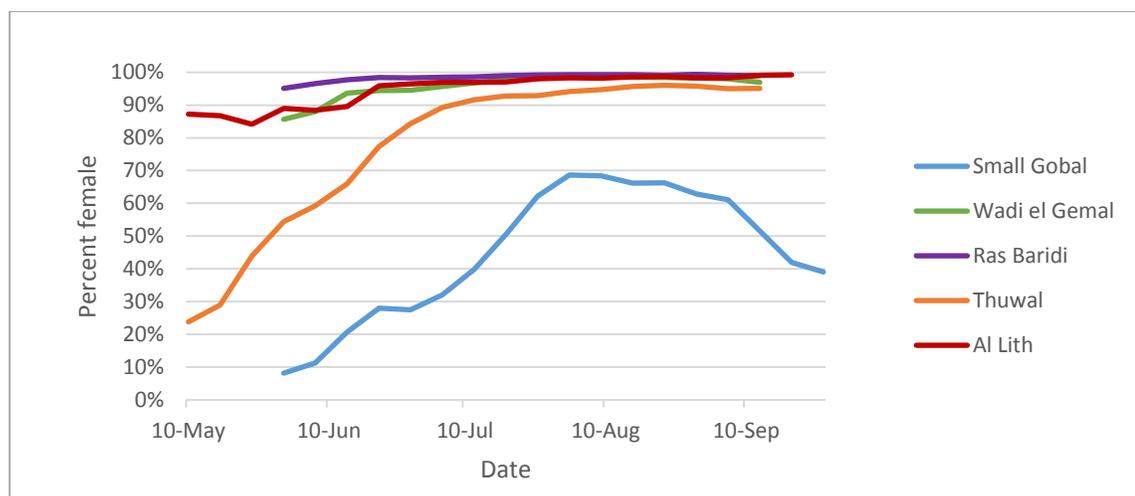
### 3.3. Hatchling sex ratio prediction

The maximum likelihood model resulted in highly female-skewed sex ratios at all sites except for on Small Gobal Island, the northernmost study site, and Thuwal during May. It is important to note that the results from the daily fluctuation assessment showed the temperature at nesting depth fluctuates as much as 1.6°C in a single day, which suggests that the pivotal temperature measured in a constant incubation experiment might not be applicable to natural nests. Nonetheless, assuming that the pivotal temperature is applicable to the Red Sea nesting sites, at 30 cm depth it is estimated that Small Gobal would have had the lowest female sex ratio of 17.5% in the end of May (Fig. 26). This percentage of females increased with time until the maximum of 80% in early August. After August, the percent of predicted females decreased to about 50% in late September. The Thuwal study site had the next lowest predicted female sex ratios, with as little as 32% in May, which increased until to over 90% from July until September. Ras Baridi, Wadi el Gemal, and Al Lith had extremely high predicted female sex ratios above 97% for the duration for deployment.



**Figure 26.** Predicted sex ratio (percent female) at five study sites in the Red Sea from May to September 2018<sup>th</sup>. Temperatures were recorded at a depth of 30 cm, which is the estimated mean temperature depth for hawksbill turtles (*Eretmochelys imbricata*). The maximum likelihood model used the pivotal temperature of 29.2°C.

The trends from 30 cm are very similar to the 50 cm, but the percentage of predicted females are slightly lower due to the cooler temperatures (Fig. 27). For example, on Small Gobar Island the predicted percent female was as low as 8% in late May, with a maximum of 69% in August. The Thuwal study site had a predicted percent female as low as 24% in May, which increased to a maximum of 96% in August. Wadi el Gemal, Ras Baridi, and Al Lith had predicted percent female between 85% and 99%.



**Figure 27.** Predicted sex ratio (percent female) at five study sites in the Red Sea from May to September 2018<sup>th</sup>. Temperatures were recorded at a depth of 50 cm, which is the estimated mean temperature depth for green sea turtles (*Chelonia mydas*) in the region. The maximum likelihood model used the pivotal temperature of 29.2°C.

#### 4. Discussion

Sand temperature varied greatly between and within study sites. Due to the large latitudinal gradient of the study, patterns of temperature from north to south were expected. However, latitude did not seem to significantly influence the thermal profile recorded at each study site. Similar results have been observed in the Great Barrier Reef, which compared nesting sites over four degrees of latitude and found no significant influence (Fuentes et al., 2010). In addition to the large-scale sand temperature differences between sites, significant differences between temperature

profiles within a single site were also found. This shows the complexity and variability of sand temperature at small spatial scales.

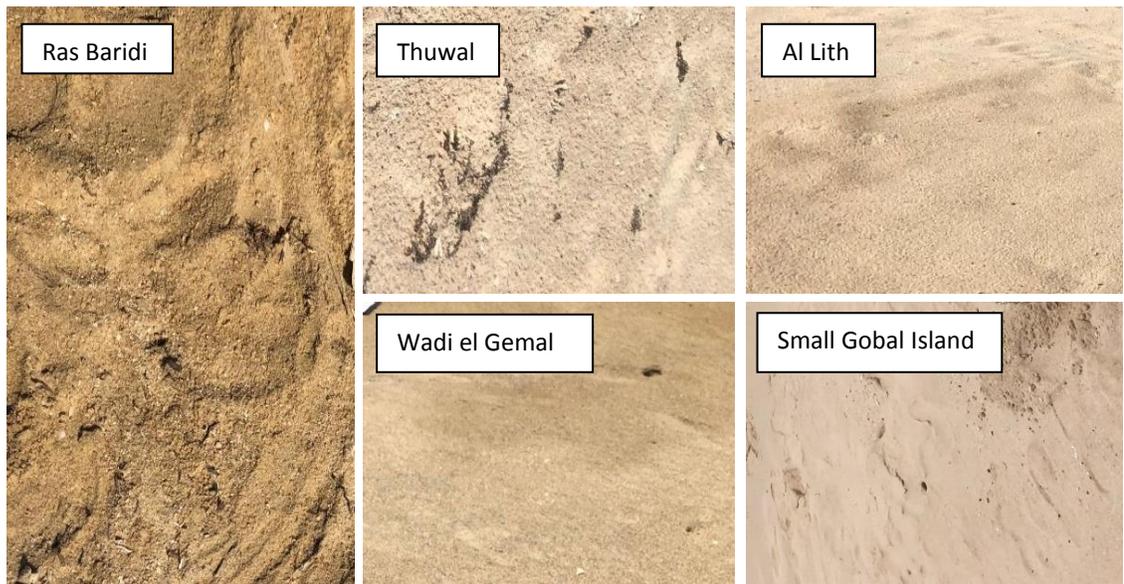
Many parameters attribute to the thermal profile of sand, including seasonal temperature changes (Mrosovsky et al., 1984), shading by vegetation (Morreale et al., 1982), grain size (Hays et al., 1995), color of sand (albedo) (Hays et al., 1995), rainfall (Laloë et al., 2016), and extreme high tides (Laloë et al., 2016). While these factors have been identified as causing thermal variations between nests, not much is known about how exactly these aspects dictate nest temperatures. In addition, studies generally focus on assessing how a single factor influences sand temperature. For example, in Costa Rica, a study found that green turtle nest temperatures were significantly affected by vegetation shading (Morreale et al., 1982). Alternatively, on the eastern coast of the United States, seasonality was found to be a significant factor in determining sex ratios of loggerhead turtles due to shifts in temperature, with predominantly males produced in the beginning and end of the nesting season and females during the middle (Mrosovsky et al., 1984).

It is important to note that many of the study sites (i.e. Small Gobar Island, Wadi el Gemal, Ras Baridi, and Thuwal) have little to no vegetation, so shading is not a cooling factor. Furthermore, the turtle nesting season coincides with the summer in Saudi Arabia, which is the hottest time of year, only receiving an average of about 2.50 mm of rain per month between June and September. Additionally, the Red Sea does not demonstrate dramatic tidal ranges, which is an important factor of cooling in other parts of the world (Laloë et al., 2016). Thus, the turtle nesting sites in the Red Sea do not

demonstrate cooling mechanisms that are crucial for regulating temperatures as reported in other areas of the world (Laloë et al., 2014; Houghton et al., 2007).

Sand albedo, a parameter known to significantly influence sand temperature (Hays et al., 1995), may explain the large-scale differences in sand temperature. Sand albedo is characterized by the percentage of the incident solar radiation reflected from the land surface (Hays et al., 1995). Studies conducted on Ascension Island and Cyprus have shown that sand temperature was higher at beaches with darker sand due to greater absorption of incident radiation (Hays et al., 2001). Temperature loggers buried at nest depth revealed seasonal changes in both light and dark beaches, but the lowest temperatures recorded on the dark beaches rarely dropped below the highest temperatures recorded at the light beaches (Hays et al., 2001).

Ras Baridi exhibited warmer temperatures than all the other sites, despite being located at a similar latitude as Wadi el Gemal. One observation from the field was that the sand at Ras Baridi was darker and coarser than the other sites, although this was not explicitly quantified. Thus, it is possible that the dark sand coloration of Ras Baridi could explain why it demonstrated warmer temperatures than the southernmost sites (Fig. 28). In order to fully assess this, further studies should quantify the sand albedo at each of these study sites.



**Figure 28.** Differences in sand color between the five study sites. Sand color (albedo) can significantly influence the temperature of sand, with darker colors generally with higher heating. These are just examples of sand colors, and there was some variation of sand color at each site.

By nesting at sites with a high range of thermal profiles, turtle populations can produce both male and female hatchlings. Unfortunately, these findings suggest that many of the nesting sites exceed the pivotal temperature for most of nesting season, and feminization could already be occurring if the assumed pivotal temperature is applicable to this region. To confirm this, further investigation should focus on measuring the sex ratio of turtles produced at the sites with highest nesting activity, such as Ras Baridi and the offshore islands of Al Lith, as well as the coolest nesting sites, such as Small Gobar Island or Thuwal.

Not only did the mean sand temperatures from each site fail to exhibit a latitudinal gradient, but water temperatures also did not show a latitudinal trend. In

general, the mean sea surface temperatures are warmer in the south and cooler in the north due to solar radiation differences between the regions (Chaidez et al., 2017; Raitzos et al., 2013; Karnauskas & Jones, 2018). Thus, it was expected that Al Lith would exhibit the warmest water temperature, followed by Thuwal, Ras Baridi, Wadi el Gemal, and Small Gobal Island. Surprisingly, the site with the warmest water temperature was Thuwal, followed by Al Lith, Wadi el Gemal, and Ras Baridi. Although the loggers were deployed at the similar depths (~5m), the difference could be attributed to the bathymetry of each reef where the loggers were deployed. Studies in the Red Sea have showed that the daily temperature range can vary drastically across the reef (Davis et al., 2011). This also highlights the role of fine scale variance, in this case due to localized hydrodynamics at the reef scape scale.

Results from this study indicated that there are some behavioral changes that turtles could make in order to change the incubation temperature of their nests. For example, temperature recorded at 30 cm was generally warmer compared to 50 cm at a given logger station. This means that it might be possible for female turtles to adjust their nest depth to reach cooler temperatures. In addition, there were seasonal differences up to 2.6°C between June and August, so females potentially could shift their nesting earlier in the season to adjust the incubation temperature of their clutch of eggs. Additionally, the sites demonstrated strikingly different sand temperatures. Small Gobal Island had a monthly mean up to 5.1°C cooler than Ras Baridi. Thus, it might be possible for female turtles to shift their nesting distribution to cope with rising temperatures. Although female turtles generally return to their natal beaches, there is

evidence that navigational error can result in redistribution (Lohmann et al., 2008b). Furthermore, results from my study indicated that sand temperature varied among logger stations within a single site, with a maximum of 1.3°C difference between logger stations at the same study site. This highlights the importance of nest location selection by nesting females. Within a single nesting beach, the thermal profile can differ. Some parameters involved in determining sand temperature that can vary on a single beach include vegetation, distance from high tide line, sand albedo, sand density, sand grain size, and the physical characteristics of the landscape. These small-scale variations make it difficult to use air temperature, sea surface temperature (SST), or land surface temperature as a proxy. The most accurate way to assess the nest temperature is visiting the site and deploying temperature loggers, which can be costly, time consuming, and difficult due to access restrictions in many locations.

Several sites had temperature recordings in August and September (the end of nesting season) which exceeded 35°C. Most sea turtle populations, including in the Red Sea, tend to nest during the hottest part of the year, which suggests that female bias might be an adaptive trait maximizing the reproductive potential (Laloë et al., 2014; Mrosovsky & Provancha, 1991). Nonetheless, the Red Sea sand temperatures are not only exceeding the presumed pivotal temperatures but are reaching the commonly cited maximum thermal tolerance. Thermal tolerance of developing embryos is not fully understood, but reviews on the topic cite a maximum thermal tolerance of either 33 or 35°C depending on the nesting beach (Miller, 1997; Ackerman, 1977). However, there is *in situ* evidence that turtle nests with fluctuating thermal regimes exceeding 35°C still

have successfully produced hatchlings (Hewaviseenthi & Parmenter, 2002; Booth & Evens, 2011; Wood et al., 2014). A review of thermal tolerances at different nesting sites indicated that there is a range of maximum values that is dependent on the location (Howard et al., 2014). There is currently no information on the maximum temperature threshold of turtles in Saudi Arabia or anywhere in the Middle East, so future studies should assess the thermal maximum on incubating eggs from this region of the world, which experiences extreme high temperatures compared to other turtle nesting sites.

In addition to mortality, high temperatures can cause reduced oxygen levels, which negatively affects muscle coordination, inhibiting the ability for turtles to crawl out of the nest (Matsuzawa et al., 2002). High nest temperatures can cause smaller hatchlings, which have decreased ability to emerge from the nest, and an increased probability of predation (Booth & Evans, 2011; Wood et al. 2014). Hence, further studies should identify the thermal maximum of sea turtle embryos at nesting sites in the Red Sea to assess the impacts that these high temperatures have on the populations.

Overall mean temperatures at the majority of our study sites were high compared to other turtle nesting sites around the world (Table 3). It is also important to note that the temperature surrounding natural nests have increased temperatures due to metabolic heating of embryos (Godfrey et al., 1996). This temperature increase can vary greatly depending on the number of eggs laid, and the number of successful hatchlings (Broderick et al., 2001). In Turkey, metabolic heating was computed at an important green turtle nesting site, which had a mean heating of 0.6°C during the middle third of the incubation period, when it is thought that sex is determined (Önder

& Candan, 2016). The temperature loggers from my study were not deployed inside a sea turtle nest, so the actual mean nest temperature could be slightly higher than reported.

**Table 3.** Mean nest temperature (°C) at different turtle nesting sites around the world. The species, geographic location, nesting season, and citation are also included.

SPECIES	GEOGRAPHIC LOCATION	NESTING SEASON	MEAN TEMPERATURE °C	CITATION
<i>Caretta caretta</i>	North Carolina, USA (Atlantic)	May-August	29.0-31.0	DeGregorio & Southwood Willard, 2011
<i>Chelonia mydas</i>	Ascension Island (Central Atlantic)	January-April	29.5-32.2	Godley et al., 2002
<i>Eretmochelys imbricata</i>	Barbados (Caribbean)	May-October	30.3-32.4	Horrocks & Scott, 1991
<i>Caretta caretta</i>	Greece (Mediterranean)	May-August	29.5-33.2	Godley et al., 2001
<i>Caretta caretta</i>	Turkey (Mediterranean)	May-August	26.7-32.1	Kaska et al., 2006
<i>Chelonia mydas</i>	St. Eustatius (Caribbean)	April-October	29.1-33.3	Laloë et al., 2016

The results of the sex ratio prediction showed an extremely high percentage of female hatchlings at all sites except Wadi el Gemal, which was the only site with evidence of hawksbill nesting. A challenge of using the Girondot (1994) statistical model on sex ratio is that the input ( $t$ ) assumes constant incubation temperature. This is because many pivotal temperature and transitional range of temperature studies are conducted in a laboratory setting, with one temperature throughout incubation. For *in situ* sand temperature, there is daily variation, especially at shallow depths. The results from this study showed daily temperature variation up to 1.6°C. This variation can

complicate simple predictions of sex ratio from daily mean temperature values (Bull, 1985). In order to validate the results from the sex ratio prediction, studies on the sex ratio of hatchlings at nesting sites through the Red Sea could be conducted.

Unfortunately, for immature turtles, there is a lack of external sex characteristics which make distinguishing sexes difficult (Moreno-Mendoza et al., 1999). Thus, direct assessment of sex requires sacrificing hatchlings for histological examination of the gonad (Mrosovsky & Benabib., 1990). Any work requiring the sacrifice of juvenile turtles must take careful consideration of the endangered status of the species. Indirect methods attempt to predict sex ratios involve using pivotal temperature, transitional range of temperature, sand temperature, or incubation duration, but these approaches only provide estimates of sex ratios. It is important to note that obtaining pivotal temperature and transitional range of temperature at a specific beach requires sacrificing large numbers (~200) of hatchlings (Wibbels, 1999; Godfrey & Mrosovsky, 2006), and these parameters are often defined by conducting constant incubation experiments.

Despite the complexity and variation of the natural environment, most studies on the effects of temperature on reptiles have been conducted under constant incubation temperature. These experiments have revealed that many phenotypic characteristics such as body size (Deeming, 2004), locomotion (Janzen, 1993), immunity response (Freedberg et al., 2008), behavior (Delmas et al., 2007), and sex (Bull & Vogt, 1979) are influenced by the incubation temperature. However, because these parameters were studied at a constant temperature, the applicability to natural nests is unknown. In order to understand how natural conditions effect phenotypic variation,

there are two options of study methods that could address the issue of environmental fluctuation. The first option is to conduct more studies in the field. The advantage of this option is that incubating eggs would experience a full range of environmental fluctuations. An alternative method would be to mimic temperature fluctuations from the field in the laboratory (Bowden et al., 2014). The advantage of this approach is that it would be possible to isolate the effects of temperature to a greater extent without confounding factors of variation (Bowden et al., 2014). Temperatures recorded in natural fluctuations, like from this study, could be used to set a temperature profile for a laboratory experiment. Designing a complementary field and laboratory experiment would be beneficial to get a better understanding of the effects of temperature on hatchling development (Bowden et al., 2014).

The results from the maximum likelihood model resulted in a high proportion of predicted female hatchlings. This finding is similar to sex ratios found in some other regions of the world. For example, a study conducted in the northern Great Barrier Reef in Australia found that 99.1% of juveniles were female (Jensen et al., 2018). Similarly, studies in Florida (Mrosovsky & Provancha, 1989) and Turkey (Özdilek et al., 2015) found 93% and 64-94% female hatchlings, respectively.

Due to the rising concern of highly female-skewed turtle hatchlings, studies have focused on quantifying sex ratios. The use of gonadal histology is considered the most accurate method for sexing individual hatchlings (Mrosovsky & Yntema, 1980). Due to the large number of hatchlings required to be sacrificed for this method, an alternative method is to study the sex ratios in adult turtle populations, but this can also have many

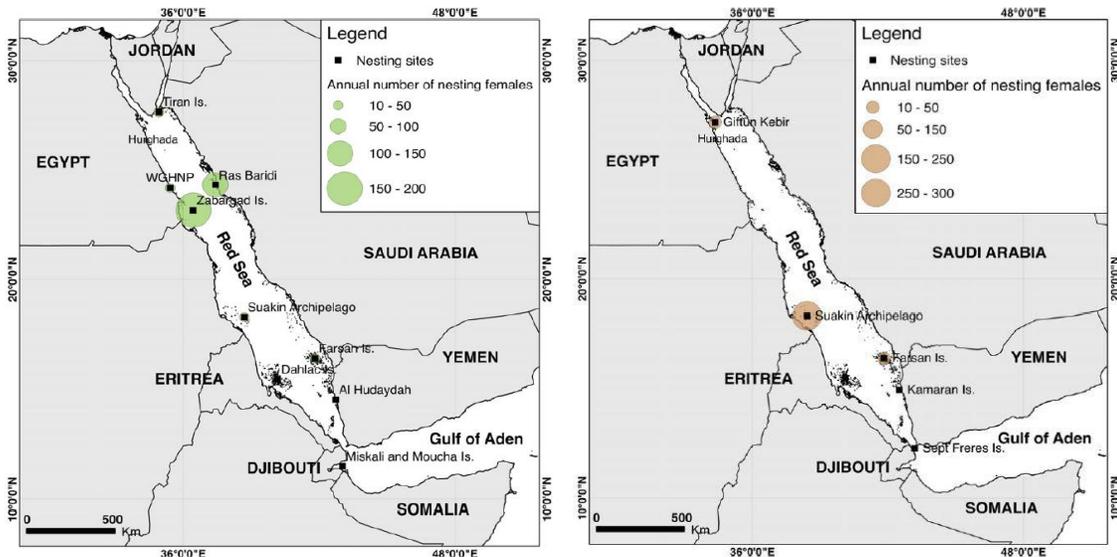
difficulties due to their highly dispersed distribution. Therefore, a functional alternative known as the breeding sex ratio (BSR) can be measured to infer adult sex ratios (Lasala et al., 2018). The BSR is the proportion of males and females that are successfully mating at one time (Stewart & Dutton, 2014). This can be assessed with parentage analyses, which can help quantify the number of males that contribute to nests. Sea turtles have been documented to have a promiscuous mating system, demonstrating both polyandry and polygyny (Crim et al., 2002). The sperm from more than one male can fertilize a single clutch (Fitzsimmons, 1998; Wright et al., 2012; Gonzalez-Garza et al., 2015; Chassin-Noria et al., 2017). To quantify the number of males fathering a single clutch of eggs, a parentage analysis can be conducted by using DNA microsatellites (Chassin-Noria et al., 2017). It has been suggested that female turtles can increase their chance of acquiring favorable genes by mating with multiple males (Alfaro-Núñez et al., 2015). However, there are risks involved due to the increased chance of predation, energy expenditure, and disease transmission (Alfaro-Núñez et al., 2015). In order to quantify the current adult sex ratio of Red Sea turtles, BSR should be assessed.

Furthermore, studies on the connectivity of marine turtles in the Red Sea should be conducted to evaluate if turtles from different latitudes are breeding. If there are turtle nesting sites with few males produced due to high temperatures, it is important to assess if males from other nesting areas are breeding with these females. Sea turtles are a highly migratory species and have been recorded crossing entire ocean basins (Lohmann et al., 2008b). Adults often migrate from their foraging ground to specific breeding and nesting sites (Hays et al., 2002). Breeding male turtles have demonstrated

tendencies to habitually return to the breeding ground located in the vicinity of their natal beaches (FitzSimmons et al., 1997). Inter-population breeding most likely occurs when individuals from different populations mix at foraging grounds or along migration routes. There has been one study published in the Red Sea that has assessed post-nesting migration patterns of green turtles in Egypt (Attum et al., 2014). These turtles were tagged on Zabargad Island, a key nesting site in the Red Sea, with an estimated 438 to 1,527 nests annually from June to August (Hanafy, 2012). One turtle migrated to Wadi el Gemal, where it resided for about 100 days before crossing the Red Sea towards Saudi Arabia. Another female turtle migrated 940 km south, passing through the territorial waters of Egypt, Sudan, and Eritrea. A third turtle traveled north to the feeding grounds nearby the Gamsha archipelago, the area of the study site Small Gobal Island (Attum et al., 2014). These results show the importance of international conservation for this highly migratory species. In addition, more tagging studies should be conducted to assess post-nesting migrations at other key nesting sites in the Red Sea, such as Ras Baridi or Al Lith.

The presence of turtle nests on the offshore islands of Al Lith in the Farasan Banks region has not been previously reported in publications (Fig. 29). A high density of fresh tracks and several carcasses of large nesting green sea turtles (presumably that had come ashore to nest) were encountered on the islands of MarMar, Dohra, Jadir, and Mulathu (Appendix 1). This demonstrates the need for more nest surveys in the Saudi Arabian region in order to document exactly where turtles are nesting. Further exploration should be conducted within the Farasan Banks because there is likely to be

high nesting activity on the sandy islands in this region. Little is known about the number of turtles nesting at each site, the species, or the nesting seasons. Thus, there is a clear need for baseline information on turtles in the Red Sea in order to evaluate how they are responding to climactic changes and anthropogenic factors.



**Figure 29.** Turtle nesting sites of green turtles (left) and hawksbill turtles (right) from a review compiling information on nesting turtles in the Red Sea by Mancini et al., 2015. Farasan Banks, located north of the Farasan Islands and east of the Suakin Archipelago, is not noticed as a turtle nesting site.

Turtles have survived millions of years on our planet through a dynamic climate, but the current rates of climate change are unprecedented (Brohan et al., 2006; IPCC, 2014), so it is unclear how turtles might adapt. There is evidence to suggest that sea turtles, and other reptiles with TSD, might not be capable of evolving fast enough to counter the negative impacts of climate change (Janzen, 1994). Conversely, other studies suggest that there are mechanisms that sea turtles could use to adapt to climate change which include changing the distribution of nesting grounds and digging deeper nests (Hays et al., 2001). It is also possible that turtles could shift their nesting season to a cooler time of year, which is a mechanism that has already been observed in some

parts of the world including Florida (Hays et al., 2003; Pike, 2014). It is even predicted that they might be able to adapt their pivotal temperature and thermal threshold (Davenport, 1989). Sea turtles have survived on this planet for at least 220 million years, meaning that they have persisted through dramatic temperature and sea level changes by developing new migratory routes and changing their nesting locations (Hamann et al., 2010). Nonetheless, the strong genetic structure seen in mitochondrial DNA suggests that sea turtle populations have deep demographic isolation due to strict natal homing (Dethmers et al., 2006). Populations must be self-sustaining in this case, thus the local-scale dynamics (such as hatchling success and sex ratio) are very important for the persistence of the local population. In addition, some studies suggest that it is unlikely that new colonization events contribute significantly to turtle populations (Gomulkiewicz & Holt, 2016). Furthermore, there are several management strategies that could be used to artificially lower the incubation temperature of turtle nests if it was determined that an intervention was needed. Some of these tested approaches include relocating turtle eggs (Tuttle & Rostal, 2010), shading (Wood et al., 2014), and cooling with water (Hill et al., 2015).

## **5. Conclusion**

The results from this study indicate that sand temperatures at nesting beaches do not follow the expected latitudinal gradient in the Red Sea. Sand temperature recorded differed among the study sites, throughout the nesting season, and between depths of 30 and 50 cm. This means that changing nesting location, earlier nesting season, or deeper nests could alter the sex ratio of turtle hatchlings. Furthermore, sand

temperature at a single site varied up to 1.3°C at comparable depths, highlighting the importance of nesting site selection for female turtles. Additionally, the commonly cited pivotal temperature of 29.2°C is exceeded at most sites from June to September. The maximum likelihood mathematical model also showed that the sex ratio may be highly female skewed (~99%) based on the pivotal temperature of 29.2°C. However, this study also demonstrated that temperature fluctuations can be as high as 1.6°C in a single day, which reveals that parameters determined in constant incubation experiments may not be applicable to natural nesting conditions. In addition, the presumed thermal maximum threshold of 33°C is exceeded as high as 100% of days from June 3<sup>rd</sup> to September 15<sup>th</sup>. Temperatures recorded at most of the study sites were higher compared to current published literature from other nesting sites around the world. However, there is a possibility that Red Sea turtle populations could be adapted to these higher changes and therefore have a higher pivotal and maximum thermal threshold compared to other populations.

### 5.1 Future Directions

To expand on the results from this study, a temperature projection should be modelled to assess how sand temperature might change in the future under various IPCC climate change scenarios. Other studies have used air temperature and sea surface temperature (SST) as a proxy for sand temperature. Remotely-derived land temperature data could also be used to create the projection in the Red Sea. In addition, for future studies I would suggest deploying air temperature loggers during the

duration of the study, and all Hobo temperature loggers should be calibrated prior to the study.

In general, research on how temperature influences the phenotype of turtles should consider thermal fluctuations. These studies should be done in the field, or in the laboratory but use parameters that mimic temperature fluctuations from the field. Understanding the mechanism of TSD and how it can adapt over time would provide key insight into how thermally-sensitive species will respond to the impacts of climate change. In order to conserve and protect sea turtles in the Red Sea, much more information is needed on the local populations. This includes movement patterns, sex ratios, pivotal temperature, transitional range, thermal thresholds, etc. Tagging studies can provide information on nesting sites, key foraging areas, and migratory routes, all of which is crucial to understand in order to implement policies to protect turtles in this region. Furthermore, in order to understand if nesting seasonality is shifting at the sites in the Red Sea, monitoring programs need to be implemented to better understand the current nesting season. In addition, aerial surveys should be conducted to assess turtle nesting densities.

High priority should be given to study the important nesting and foraging habitats of the northern Red Sea, which could potentially be a refuge from the effects of climate change in the future. The cool temperatures and sex ratio prediction mean that this could be an important nesting region for the survival of male turtles. In addition, this area will undergo a massive transformation due to the NEOM project on the northern Saudi Arabian Red Sea coast. This mega-city concept was developed under

Saudi Arabian's Vision 2030, with the aims of diversifying the economy. NEOM is proposed to span 26,000 sq. km across parts of Egypt, Jordan, and Saudi Arabia (Public Investment Fund, 2017). In addition, the Red Sea Project plans to develop 50 islands between the city of Umluj and Al Wajh, which could be another important nesting and foraging area for sea turtles. This construction will likely increase shipping traffic which can cause injury and mortality in many marine species, including turtles. All of these projects present potential challenges for the existing turtle populations, but the projects all also aim to be eco-tourism destinations. As the projects are all at very early stages of planning and development, there is an opportunity for Saudi to set a good example and proceed with these developments while preserving environmental conditions (including the critical nesting habitats for turtles). Careful planning and interaction with scientists will be necessary to achieve sustainability in the near future.

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**APPENDIX 1.**

The following photos provide evidence of the turtle nesting occurring on the offshore islands near Al Lith, in the Farasan Banks. Marmar Island (N 19°50'3.57" E 39°55'37.54") was visited on April 27<sup>th</sup>, and a recently deceased green sea turtle hatchling (Fig. 30) was observed on the southern end of the island. This shows that nesting begins in April, which is earlier than previously believed. Marmar is slightly unusual among islands in the Farasan Banks in that it has substantial vegetation, which means that there could be shading occurring at some of the nests. However, the areas with the highest density of tracks were the large sandy areas of the north and south of the island, where there was less vegetation, which we observed during our visits both in May and September (Fig. 31). The east and west sides of Marmar had exposed rocks along the shoreline that could be an obstacle to many turtles, which could explain why they mostly nest in the north and the south (Fig. 32). Dohra (N 19°49'28.65" E 39°53'58.13") is a sandy island about 700 meters long and 350 meters wide is approximately 2.7 km west of Marmar. When we visited in May, we observed several (approximately seven) turtle skeletons which were likely there for a substantial amount of time based on their advanced stage of decay (Fig. 33). However, when we returned in September we found a turtle carcass with flesh intact. We do not think this carcass was present during our first visit in May, suggesting that it was from this nesting season (Fig. 34). Dohra, Jadir (N 19°47'17.07" E 39°57'15.44"), and Malathu (N 19°45'0.53" E 39°54'36.69") all were sandy islands covered in turtle tracks and nests. It was unclear

how many of the nests were from this season.



**Figure 30.** A deceased green sea turtle hatchling (*Chelonia mydas*) found on the northern side of Marmar Island on May 2<sup>nd</sup>, 2018.



**Figure 31.** Several turtle tracks were observed on the northern side of Marmar island on September 21<sup>st</sup>, 2018.



**Figure 32.** There were less tracks observed on the east and west sides of Marmar, most likely due to the exposed rocks which can be seen on the right side of the photos.



**Figure 33.** Several adult sea turtle skeletons were found in the center of Dohra island on May 2<sup>nd</sup>, 2018



**Figure 34.** A deceased female green turtle (*Chelonia mydas*) observed on Dohra on September 21<sup>st</sup>, 2018.