

Determinants Influencing the Sex Ratio of Turtle Offspring

<https://github.com/christalzheng/HuangZheng>

Yuechen Huang; Shiqi Zheng

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Figure 1: A cute baby sea turtle

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1 Rationale and Research Questions

In the majority of species, sex is determined during fertilization; however, reptiles like turtles, alligators, and crocodiles exhibit a unique characteristic where sex determination occurs after fertilization. This process is known as temperature-dependent sex determination (TSD). In TSD, the sex of offspring is influenced by the temperatures experienced during egg incubation in the nest.

Our project is motivated by a keen interest in understanding TSD, particularly in turtles. While TSD was first described in a lizard, the order Testudines has played an important role in advancing scientists' understanding of TSD. Studies on species within Testudines brought TSD to the attention of the broader research community, and the term "temperature-dependent sex determination" also first appeared in a study of a community of North American turtles in 1979 (Yntema, 1976; Bull and Vogt, 1979).

We aim to test the TSD phenomenon by ourselves with a focus and thus choose to investigate a dataset containing plenty sex ratio information for the order Testudines. We will explore the following questions: 1. Does temperature affect the sex ratio of turtles? If so, how does temperature influence the sex ratio of turtles? 2. Is temperature the sole determinant of turtle sex, or are there other important factors?

2 Dataset Information

We use a dataset called Reptilian Offspring Sex and Incubation Environment (ROSIE). It is a dataset published in 2022 and contains over 7,000 individual measurements of offspring sex ratios in the order Testudines as well as SDM classifications for 149 species. This dataset obtained data by using the Web of Science to search for research published since the discovery of TSD (1966) until 31 December 2020.

We processed the original dataset by omitting all data that has been chemical treatment, selecting all TSD type data, all point contains male proportion data, converting date value to date object, and selecting all species information, mean temperature, latitude, longitude, captive, humidity, start date, end date, proportion male, and proportion female as the variables we will explore with.

Variables we used and wrangled for this project are shown in Table 1. More information about the ROSIE Dataset can be found at: <https://github.com/calebkrueger/ROSIE/tree/main>

Table 1: Summary of Dataset

Used Variables	Details
Species information	Order, Family, Genus, Species
Spatial information	Wild sampling location, or native range of species if captive or location not provided, Provided in average latitude and longitude
Captivity	Eggs from captive or/and wild individuals, 0 - wild and 1 - captive
Time	Time of turtle nesting/egg collection. Provided in start date and end date
Temperature	Mean of actual/recorded incubator temperature in degrees Celsius
Humidity	Relative humidity of incubation chamber between 0 and 1
Sex ratio	Proportion of male,

3 Exploratory Analysis

To have a better understanding of our data, we explore the distribution of different turtle species, variations in hatching temperatures across diverse turtle families, and distribution of data location.

3.1 Explore the species in the dataset

3.2 Explore hatching temperature among families

3.3 Explore data position

The location of wild sampling points, or the native range of species if captive, is plotted below as a map to show the spatial distribution of data. The world continent data is downloaded from ArcGIS database.

Distribution of data

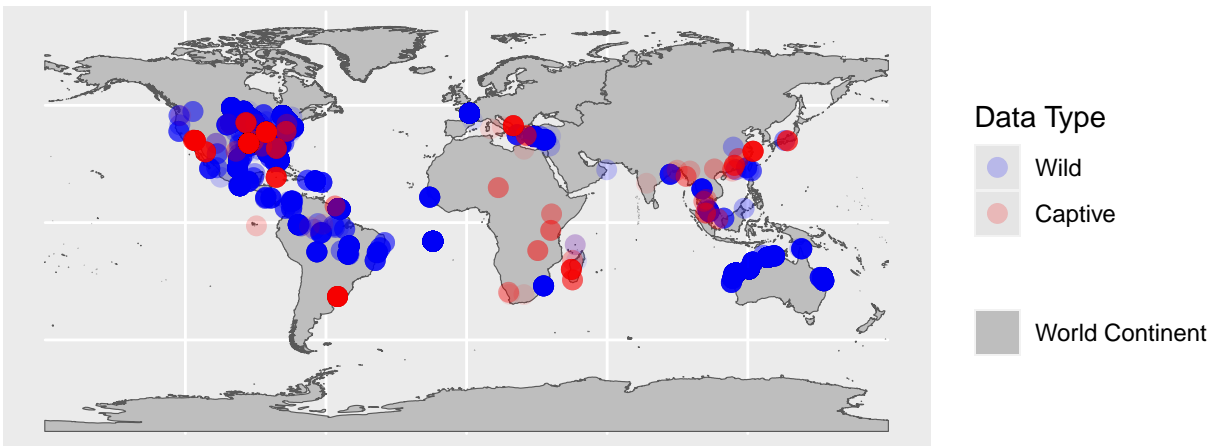


Figure 2: Spatial distribution of data

4 Analysis

4.1 Question 1: Does temperature affect the sex ratio of turtles?

4.1.1 how does temperature influence the sex ratio of turtles?

Table 2: Temperature-Sex Ratio Relationship

Temperature	Sex Ratio
> 32°C	Female
28-32 °C	Male:Female ~ 50:50
< 28°C	Male

4.2 Question 2: Are there other important factors influence sex ratio of turtles?

To answer this question, we explore the impacts of temporal and spatial variation on the sex ratio of turtles.

4.2.1 Temporal impacts

At Year Level Null Hypothesis: There is no effect of year on the sex ratio of turtles. Alternative Hypothesis: There is an effect of year on the sex ratio of turtles.

Based on the linear model, we reject the null hypothesis with a p value of 2.873e-16. This p-value also shows this model is statistically significant and a meaningful regression. But according to the R^2 value, only 1.495% of the total variance in sex ratio is explained by changes in year. The sex ratio is predicted to increase 0.0041539 with 1 year change. The results of model are clearly showed in Figure The blue line shows a overall slightly increasing trend between year and sex ratio, indicating that as time passed by there are more male turtles. This is contradicts with our intuition that time increases will cause temperature increases and thus will lead to more female turtles. But because temperature trends might not be strictly linear or may be influenced by other factors in long term scenario, and the very noisy scatter plot and low R^2 value both show that this pattern only explain limited part of data, the linear model results of year and sex ratio is still statistically significant and a meaningful regression.

Below is results of linear model and scatter plot of year and sex ratio.

```
##
## Call:
## lm(formula = Proportion_Male ~ Year, data = Cleaned_Data_omit)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -0.52675 -0.42290 -0.03557  0.48156  0.63525
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept) -7.8599014   1.0128760   -7.76 1.05e-14 ***
## Year         0.0041539   0.0005059    8.21 2.87e-16 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.412 on 4376 degrees of freedom
```

```
## Multiple R-squared:  0.01517,    Adjusted R-squared:  0.01495
## F-statistic: 67.41 on 1 and 4376 DF,  p-value: 2.873e-16

## 'geom_smooth()' using formula = 'y ~ x'
```

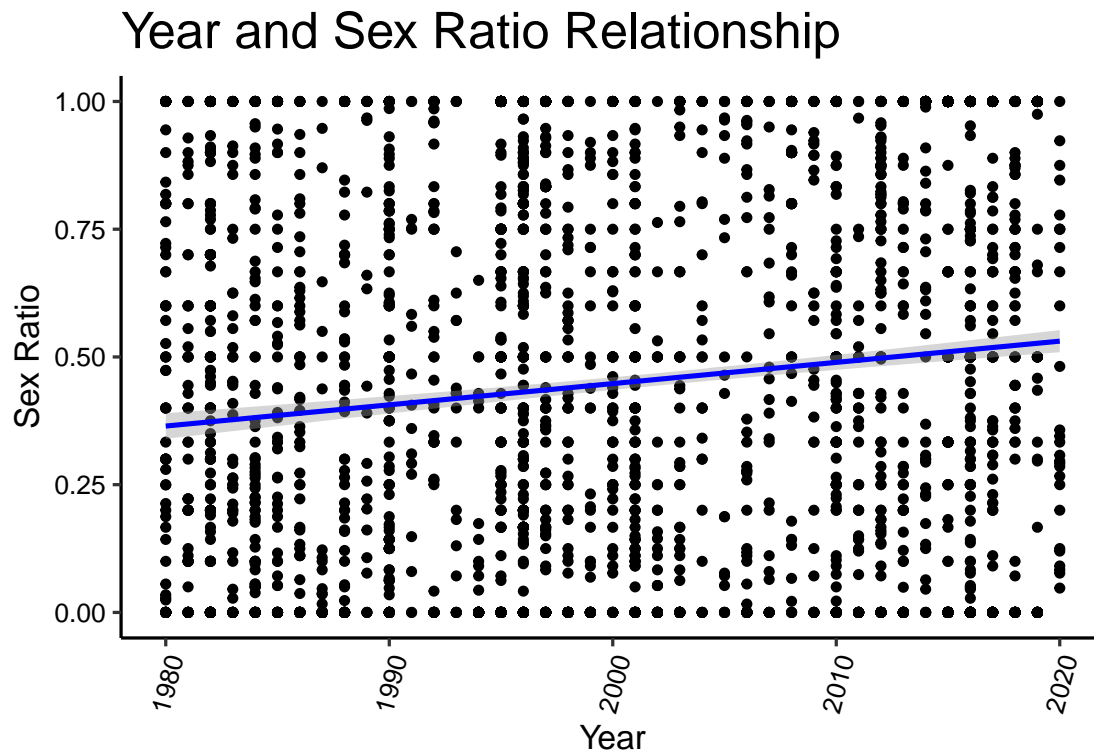


Figure 3: Year and sex ratio relationship

At Month Level Null Hypothesis: There is no effect of month on the sex ratio of turtles. Alternative Hypothesis: There is an effect of month on the sex ratio of turtles.

Based on the linear model, we reject the null hypothesis with a p value of $2.2e-16$. But according to the R^2 value, only 2.857% of the total variance in sex ratio is explained by changes in month. 7 of the 12 months had p-values of less than 0.05, with April, June, August, September, and December being the exception. The p-values of the statistically significant month ranged from $< 2e-16$ (January) to 0.01576 (February). The impacts of month may be influenced by spatial location and local climate. In order to explore monthly sex ratio distribution more clearly, we use box plot to show sex ratio by month for north hemisphere and south hemisphere respectively. The box plot for the north hemisphere clearly shows a trend of more male during colder season (November to April) and female during hotter season (May to October). This result is aligned with conclusion from Question 1 that higher temperature will cause more female turtles. The monthly distribution trend of sex ratio is not clearly for south hemisphere. This maybe because of lack of data in south hemisphere. But the highest proportion of male still appear in the colder season (July and August), and the rest of months all have less than 50% of male. Generally, the month impacts on sex ratio of turtles may also be influenced by the existence of breeding season.

Below is results of linear model and box plots of sex ratio by month for different hemispheres.

```
##
```

```

## Call:
## lm(formula = Proportion_Male ~ Month, data = Cleaned_Data_omit)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -0.6213 -0.4034 -0.0399  0.3882  0.6985
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)  0.47796    0.02046  23.363 < 2e-16 ***
## MonthFeb     -0.11185    0.04631  -2.415  0.01576 *
## MonthMar     -0.17643    0.06084  -2.900  0.00375 **
## MonthApr      0.02323    0.03586   0.648  0.51708
## MonthMay     -0.08198    0.03143  -2.608  0.00913 **
## MonthJun     -0.02139    0.02275  -0.940  0.34708
## MonthJul     -0.14990    0.02975  -5.039 4.86e-07 ***
## MonthAug     -0.00348    0.02879  -0.121  0.90380
## MonthSep     -0.07455    0.03845  -1.939  0.05256 .
## MonthOct     -0.16950    0.05546  -3.056  0.00225 **
## MonthNov      0.14339    0.02833   5.062 4.32e-07 ***
## MonthDec     -0.02247    0.03395  -0.662  0.50803
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.4092 on 4366 degrees of freedom
## Multiple R-squared:  0.03101,    Adjusted R-squared:  0.02857
## F-statistic: 12.7 on 11 and 4366 DF,  p-value: < 2.2e-16

```

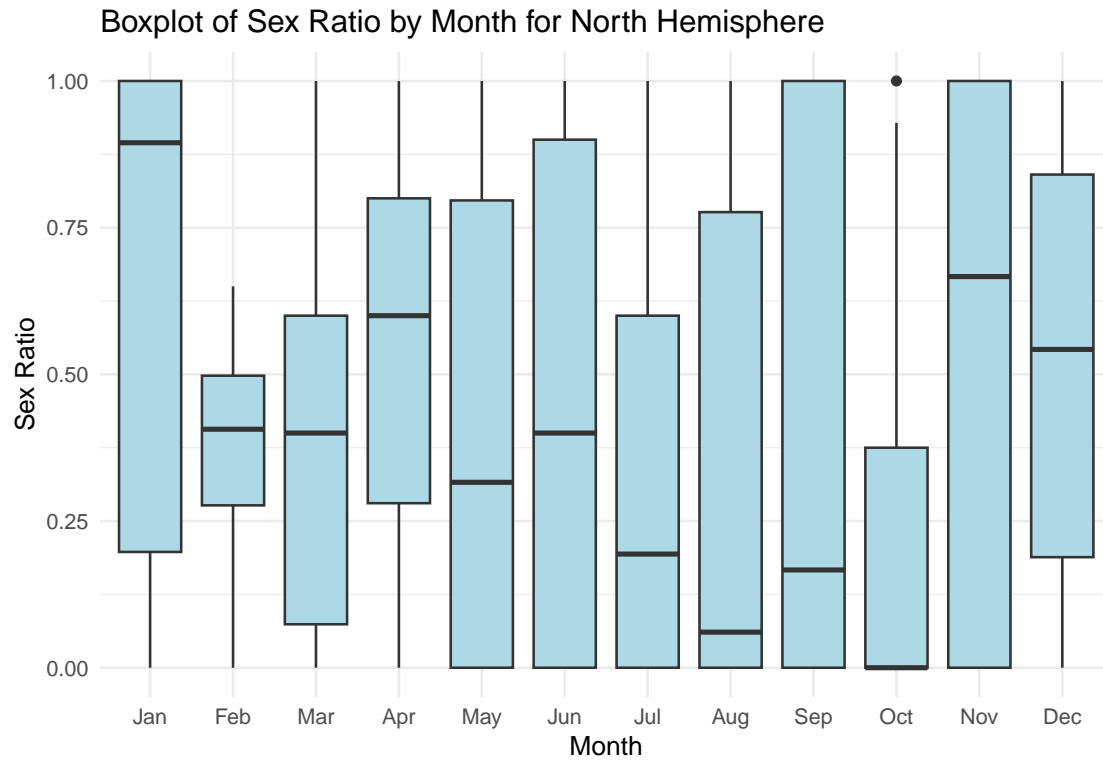


Figure 4: Sex ratio value distribution by month for north hemisphere

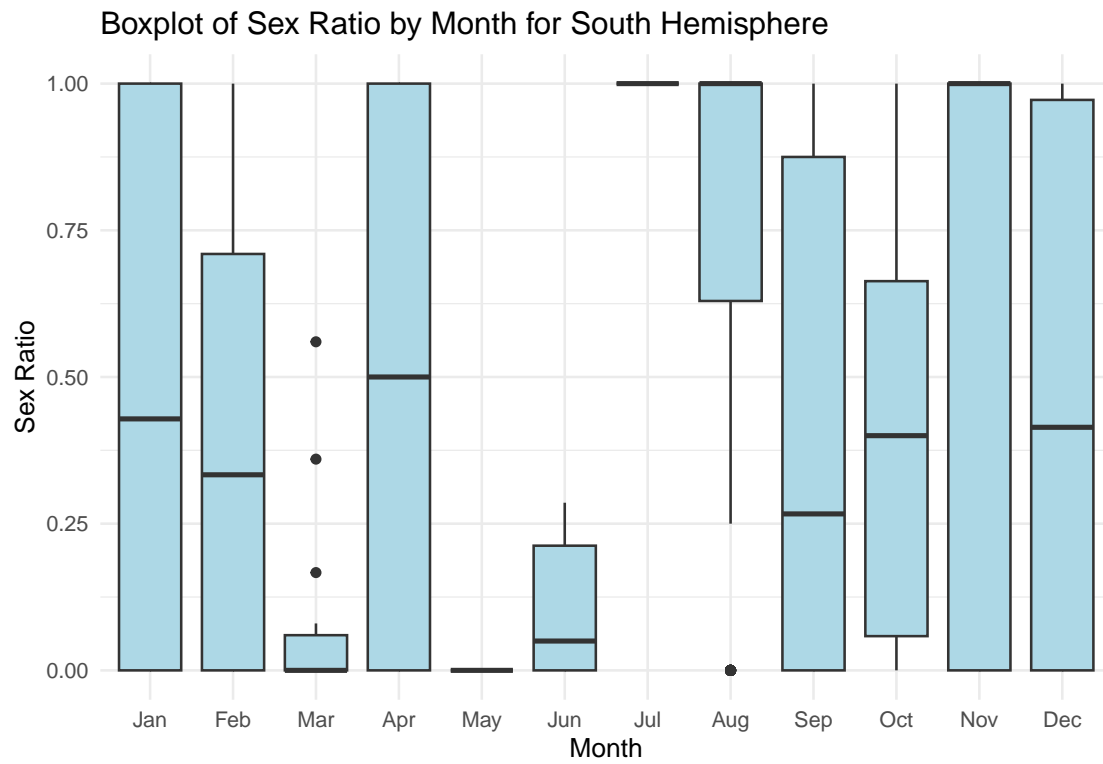


Figure 5: Sex ratio value distribution by month for south hemisphere

4.2.2 Spatial impacts

We created a map to visualize the spatial impact on the sex ratio of turtles. However, it was difficult to identify any distinctive spatial pattern of sex ratio distribution from the map alone. However, after combining the temperature data with the sex ratio distribution, we were able to observe that the distribution of sex ratio is influenced by the temperature at different positions. The results from the map are consistent with the findings from Question 1 that location with lower temperature will have higher male proportion.

Sex Ratio Distribution

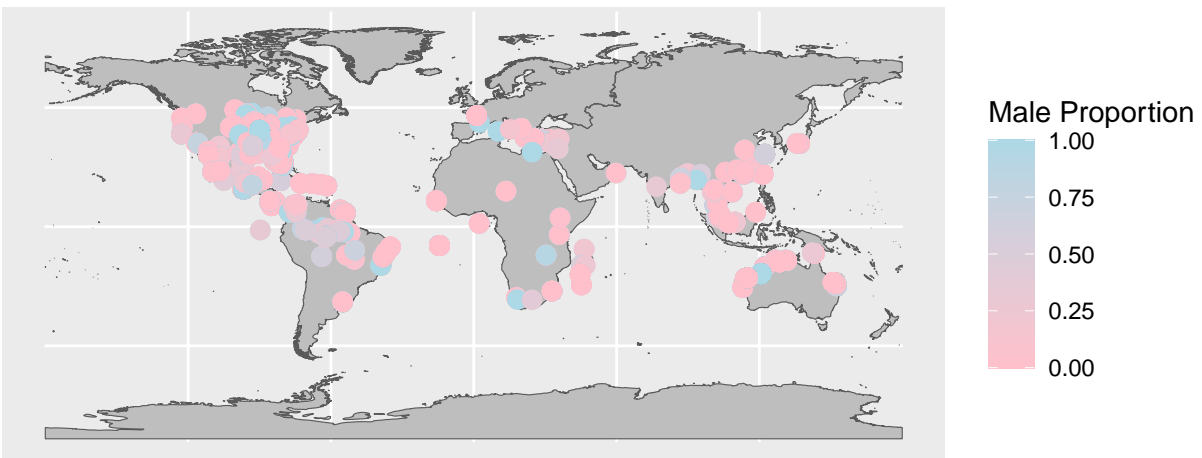
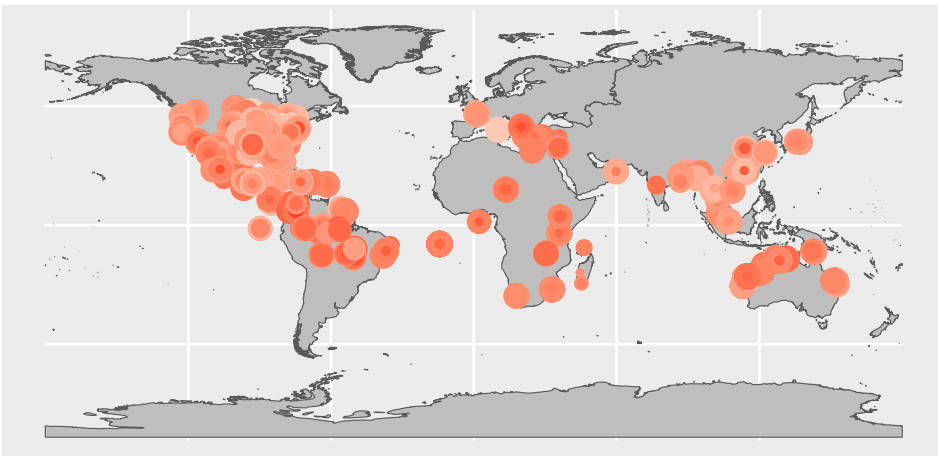
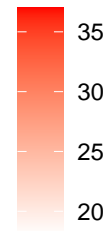


Figure 6: Sex ratio distribution on map

Sex Ratio Distribution



Temparatue



Male Proportion

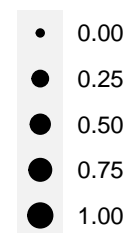


Figure 7: Sex ratio distribution with temperature on map

4.2.3 Best set of explanatory variables

Though month and year do have effects on the sex ratio of turtles, the trend between time and sex ratio may be confounded by the trend between time and temperature. We then want to explore what are best set of variables for sex ratio determinants. Thus, we run an AIC analysis to determine what set of explanatory variables is best suited to influence sex ratio. We use date data to provides the most detailed temporal information, including day, month, and year, which can capture fine-grained temporal patterns. The result form AIC shows that temperature alone is the best explanatory variables for the sex ratio of turtles. This result may be able to improve by having more valid data as there is only 63 observations due to limited amount of humidity data. Below is the detail of model results.

```
## Start: AIC=-131.4
## Proportion_Male ~ Mean_Temp + End_Date + Humidity + Captive
##
##           Df Sum of Sq    RSS    AIC
## - Humidity   1   0.00145 1.9449 -133.37
## - End_Date   1   0.01129 1.9548 -133.14
## - Captive    1   0.06095 2.0044 -132.01
## <none>                1.9435 -131.40
## - Mean_Temp  1   2.42999 4.3735  -96.90
##
## Step: AIC=-133.36
## Proportion_Male ~ Mean_Temp + End_Date + Captive
##
##           Df Sum of Sq    RSS    AIC
## - End_Date   1   0.08152 2.0265 -133.517
## <none>                1.9449 -133.365
## - Captive    1   0.09458 2.0395 -133.228
## - Mean_Temp  1   2.48308 4.4280  -98.342
##
## Step: AIC=-133.52
## Proportion_Male ~ Mean_Temp + Captive
##
##           Df Sum of Sq    RSS    AIC
## - Captive    1   0.05335 2.0798 -134.35
## <none>                2.0265 -133.52
## - Mean_Temp  1   2.58662 4.6131  -98.50
##
## Step: AIC=-134.35
## Proportion_Male ~ Mean_Temp
##
##           Df Sum of Sq    RSS    AIC
## <none>                2.0798 -134.347
## - Mean_Temp  1   2.7131 4.7929  -98.778
##
##
## Call:
## lm(formula = Proportion_Male ~ Mean_Temp, data = Cleaned_Data_remove)
##
## Coefficients:
## (Intercept)    Mean_Temp
##      3.4723      -0.1018
```

5 Summary and Conclusions

summary from question 1

The sex ratio of turtles can be influenced by temporal factors, although the impact is generally small. While the distribution of sex ratios across space does not exhibit any specific pattern, there is a noticeable trend where locations with lower temperatures tend to have a higher proportion of male turtles. Despite this, it appears that neither spatial nor temporal factors are significant determinants of the sex ratio of turtles. The analysis strongly suggests that temperature is the primary determinant.

// conclusion// below may be used as ref for conclusion “Researchers have also noted that the warmer the sand, the higher the ratio of female turtles. As the Earth experiences climate change, increased temperatures could result in skewed and even lethal incubation conditions, which would impact turtle species and other reptiles.” <https://oceanservice.noaa.gov/facts/temperature-dependent.html>



Figure 8: Smiling red-eared turtles

6 References

- Bull, J. J., & Vogt, R. C. (1979). Temperature-dependent sex determination in turtles. *Science*, 1186-1188.
- Krueger, C. J., & Janzen, F. J. (2022). ROSIE, a database of reptilian offspring sex ratios and sex-determining mechanisms, beginning with Testudines. *Scientific Data*, 9(1), 22.
- Yntema, C. L. (1976). Effects of incubation temperatures on sexual differentiation in the turtle, *Chelydra serpentina*. *Journal of Morphology*, 150(2), 453-461.
- Photo credit: <https://scitechdaily.com/why-does-temperature-determine-the-sex-of-turtles/>
<https://www.pinterest.jp/pin/61783826112891810/>