



## Impact of Temperature Elevation on Seawater Snail *Planaxis sulcatus*

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### ARTICLE INFO

#### Article History:

Received: June 10, 2021

Accepted: July 27, 2021

Online: Aug. 9, 2021

#### Keywords:

*Planaxis sulcatus*;  
Elevated temperature;  
Global warming;  
Climate change;  
Red sea.

### ABSTRACT

To assess the effect of temperature on wet weight, shell length, and shell aperture of seawater snail *Planaxis sulcatus*, an experimental trial was conducted to examine 72 samples of this species. The specimens were collected from the area of Al-Rayis coast on the western side of Saudi Arabia and were divided into 3 groups; A, B, and C (24 snails/group). Group A was the control group. Group B was treated by increasing water temperature about 5°C above the control group, and about 10°C above the control group for group C. There were no significant differences in weekly wet weight measurements with respect to the intragroup comparison and the intergroup comparison of the three groups. However, regarding shield length, it increased at week 1 with a pairwise comparison of weekly records showing only a significant difference between the 1<sup>st</sup> week and that of the onset of the study in group A. While, in group B, results showed only a significant difference between week 3 and week 6, and no significant difference was detected in group C. In addition, no significant differences were recorded when the 3 groups' shield lengths were compared. Hence, it is recommended to conduct complex experiments that depend on exposing snails to several climate change factors, such as increased acidity and salinity in addition to high temperature.

### INTRODUCTION

Since the year 1950, the climate has witnessed an obvious change, including temperature change, hurricanes and high levels of saline seawater precipitation amounts (IPCC, 2014), and this has remarkably been due to the natural global effect or to human activities (Gravili *et al.*, 2017). Global warming is one of these climate changes spotlighted by the Intergovernmental Panel on Climate Change (IPCC) which predicts that global mean sea water temperature will increase from 2–4.5°C by 2100 (scenario RCP 8.5, (Intergovernmental Panel on Climate Change, 2013). Inc. global warming of 1.5°C - 2.5°C is predicted to cause extinction of about third of biological species (Dow & Downing, 2011).

*Planaxis sulcatus* belongs to the Anemalia kingdom, Mollusca phylum, Gastropoda class, Neotaenioglossa snails order, family of Planaxis, species: *Sulcatus* (Born, 1978). Gastropods are the largest class of mollusks including about 30,000 marine

species. The Furrowed Clusterwink (*Planaxis sulcatus*) is found commonly in the rocky intertidal environments. The conical shell, speckled with patterns of white spots on a greenish-brown background of *P. sulcatus*, grows up to a length of 35 mm in adult individuals. The *P. sulcatus* are herbivorous, feeding primarily on microalgae growing on the substrates in the habitats (Houbrick, 1987). Females are usually larger than males (OHGAKI, 1997).

The *P. sulcatus* withdraws into its shell behind the operculum attaching itself to the substratum during low tides (Ruppert, 2004). Therefore, it can be seen on rocks and stones in aggregates or taking shelter in rock pools, crevices and under large rocks during the low tides (Houbrick, 1987). Snails are of medical importance as they act as intermediate hosts for many parasites during their life cycles (Dore, 1991). Most of the *P. sulcatus* act as hosts for many trematode species and are usually heavily infected by one or more species of trematodes (Rohde 1981).

The daily and seasonal variation in temperature affects the metabolism of the *P. sulcatus*, their activity level and energy balance. Furthermore, mostly the rates of physiological and biochemical activities are affected by temperature (Precht & Herbert, 1973). Notably, the increase in behavioral activity is attributed to the increase of sea water temperature; the outcome of oxygen consumption increase. This, in return, leads to a decrease in the energy storage in the body of marine organisms causing a decline in physical growth rate (Levinton, 2009). Consequently, the global warming is expected to decrease body size of those organisms (Mazurkiewicz *et al.*, 2020). Therefore, warming decreases their biomass through affecting interaction with food web due to increasing energy demand and metabolic processes (Bruno *et al.*, 2015). Marigómez *et al.* (2017) reported a gradual reduction in weight of the sea cucumber, *Apostichopus japonicus*, due to the gradual rise in the environmental temperature where it lives. The previous authors added that, an additional increase in current temperatures is feared to exacerbate global warming negative effects, as these organisms are likely to be affected by sea warming. Thus, it is important to predict effects of climate changes on these organisms. However, these physiological and biochemical activities do not depend mostly on environmental temperature as these activities can compensate for variations (Precht & Herbert, 1973). This compensation is called acclimation if it is induced by a single environmental factor (such as temperature) in the laboratory. If this compensatory change is due to the temperature changes, it is called thermal acclimation (Yanzeel, 1998).

Kim *et al.* (2004) examined the effects of the harmful red tide on the dinoflagellate, *Cochlodinium polykrikoides*, and recorded optimum growth rates at temperatures 21 to 26°C and salinities from 30 to 36°C. They added that, when temperature was lower than 10°C, no growth occurred. Moreover, they noted that growth occurred at temperature from 15 to 30°C and salinities from 20 to 36°C. Therefore, this study aimed to assess the effect of temperature on wet weight, shell length and shell aperture of seawater snail *Planaxis sulcatus*.

## MATERIALS AND METHODS

### Materials:

- 72 *Planaxis sulcatus* snails
- Aquaria (10 L to each one) to place snails
- Cage to keep snails in the bottom of the aquarium
- Digital thermometer (Aqua Medic with an accuracy of  $\pm 1$  ° C) to measure water temperatures
- Heaters (50W) to provide suitable water temperatures
- Measuring ruler cm to measure the snail aperture
- Balance to measure the weight of the snail (KERN Precision Balance PCB scale)
- Plastic tie to tie the cage
- Distilled water to balance salinity (when necessary)
- Caliper cm to measure the length of the snail
- Digital salinity (Aqua Medic with an accuracy of  $\pm 2$  ppt)
- pH monitor (Aqua Medic with a resolution of  $\pm 0.01$  pH)

### Collection of samples

The samples were collected from the area of Al-Rayis coast in the western side of Saudi Arabia (Red sea) where the surface water temperature was 23°C (measured 30 cm below the surface of the seawater). Snails were then transferred to the lab in plastic containers with some rocks containing algae as a temporary food for snails.

*Planaxis sulcatus* snail was chosen due to its availability in the region and the surrounding areas and it is not one of the endangered species, in order to preserve the biodiversity and abundance of aquatic life in the area.

### Adaptation

Samples in the lab were left for adaptation for five days, then transferred to the Aquaria. Temperature was gradually raised by 0.5°C per day for group B, and 1°C per day for group C to reach the target temperature of each aquarium.

### Grouping

Seventy-two samples were divided into 3 groups; A, B and C. Each group contained 24 snails. Group A was the control group. Group B was treated by increasing water temperature about 5°C above the control group, while group C was treated by increasing water temperature about 10°C above the control group.

### Measurements

Growth measurements for all samples (wet weight, shield length and shield aperture) were evaluated after 10 days adaptation period qualified to reach the required temperature. Measurements were evaluated once per week.

### Statistical analysis

The collected data were coded, processed and analyzed using Statistical Package of Social Science (SPSS) program for windows (version 16) (Chicago, IL, USA). Quantitative data were presented in mean and standard deviation (SD).

Repeated measures ANOVA test was performed to test significant difference throughout the study period for each group. Bonferroni post hoc test was performed to detect pairwise significance throughout study period. To test the significance between different groups, one way ANOVA test was used. A  $p < 0.05$  was considered statistically significant, and all analyses were two-sided. Line graphs were established to aid with the demonstration of results.

## RESULTS

During the study period, the seawater measurements of studied groups revealed that in group A, the mean temperature, salinity and acidity were  $25.3 \pm 1.2^{\circ}\text{C}$ ,  $39.08 \pm 0.73$  ppm and  $8.10 \pm 0.09$ , respectively. Seawater measurements of temperature, salinity and the pH increased gradually in group B ( $29.6 \pm 1.2^{\circ}\text{C}$ ,  $39.55 \pm 0.95$  ppm, and  $8.06 \pm 0.10$ ) and group C ( $35.4 \pm 0.6^{\circ}\text{C}$ ,  $39.76 \pm 1.57$  ppm, and  $8.02 \pm 0.13$ ), respectively.

Changes in the wet weight of studied snails were considered. It was noticed that, in group A, the wet weight started at  $1.00 \pm 0.27$  g in the beginning of the study to reach  $0.94 \pm 0.25$  g by the end of 6<sup>th</sup> week without any significant differences between weekly measures of the wet weight (Fig. 1). Regarding group B, the wet weight started at  $0.98 \pm 0.26$  g and reached  $1.06 \pm 0.23$  g by the end of 6<sup>th</sup> week, without any significant differences between weekly measures of the wet weight as seen in Fig. (1). In group C, the wet weight started at  $1.02 \pm 0.19$  g in the beginning of the study to reach  $0.99 \pm 0.14$  g by the end of 6<sup>th</sup> week without any significant differences between weekly measures of the wet weight (Fig. 1).

Changes in shield length were regarded for the studied species of snails. It was noticed that in group A, the shield length started at  $1.32 \pm 0.13$  g in the beginning of the study to reach  $1.27 \pm 0.12$  by the end of 6<sup>th</sup> week. Pairwise comparison of weekly shield length records showed only a significant difference between the 1<sup>st</sup> week with reading ( $1.29 \pm 0.17$ ) and that of the onset of study ( $1.32 \pm 0.13$ ) (Fig. 2). Regarding group B, the shield length started at  $1.30 \pm 0.07$  in the beginning of the study to reach  $1.32 \pm 0.11$  by the end of the 6<sup>th</sup> week. Pairwise comparison of weekly shield length records shows only a significant difference between reading during week 3 ( $1.25 \pm 0.1$ ) and that of the end of study ( $1.32 \pm 0.11$ ) (Fig. 2). For group C, the shield length started at  $1.30 \pm 0.10$  in the beginning of the study to reach  $1.30 \pm 0.06$  by the end of the 6<sup>th</sup> week without any significant differences between weekly measures of shield length (Fig. 2).

Changes in in shield aperture of the snails were addressed throughout this study. It was noticed that in group A, the shield length started at  $0.99 \pm 0.14$  g in the beginning of the study to reach  $1.00 \pm 0.11$  by the end of the 6<sup>th</sup> week; without any significant

differences between weekly measures of shield opening (Fig. 3). Regarding group B, the shield length started at  $0.98 \pm 0.08$  in the beginning of the study to reach  $1.00 \pm 0.06$  by the end of the 6<sup>th</sup> week; without any significant differences between weekly measures of shield opening (Fig. 3). Regarding group C the shield length started at  $0.99 \pm 0.06$  in the beginning of the study to reach  $0.98 \pm 0.04$  by the end of the 6<sup>th</sup> week without any significant differences between weekly measures of shield opening (Fig. 3).

Comparison between the three studied groups regarding the weekly assessed wet weight of snails is presented in Table (1). It revealed no significant difference in the wet weight at any station of the study. Regarding the shield length, comparison of the weekly assessment revealed no significant difference in the shield length at any station of the study (Table 2). In addition, the comparison between the three studied groups regarding their weekly assessed shield aperture of snails showed no significant difference in the shield aperture at any station of the study (Table 3).

**Table 1.** Comparison between the studied groups regarding the wet weight (g) of the *Planaxis sulcatus* throughout study period (n = 24)

| Study period | Wet weight in (g)        |                               |   |
|--------------|--------------------------|-------------------------------|---|
|              | Group A<br>Mean $\pm$ SD | Group B<br>Mean $\pm$ SD      | Group C<br>Mean $\pm$ SD                    |
| Week 0       | 1.00 $\pm$ 0.27          | 0.98 $\pm$ 0.26<br>P1 = 0.868 | 1.02 $\pm$ 0.19<br>P2 = 0.893<br>P3 = 0.738 |
| Week 1       | 1.06 $\pm$ 0.34          | 1.05 $\pm$ 0.26<br>P1 = 0.943 | 1.02 $\pm$ 0.16<br>P2 = 0.801<br>P3 = 0.833 |
| Week 2       | 1.04 $\pm$ 0.34          | 1.08 $\pm$ 0.22<br>P1 = 0.761 | 1.00 $\pm$ 0.14<br>P2 = 0.773<br>P3 = 0.384 |
| Week 3       | 0.97 $\pm$ 0.27          | 1.07 $\pm$ 0.23<br>P1 = 0.481 | 0.99 $\pm$ 0.14<br>P2 = 0.853<br>P3 = 0.436 |
| Week 4       | 0.95 $\pm$ 0.26          | 1.06 $\pm$ 0.23<br>P1 = 0.383 | 0.99 $\pm$ 0.14<br>P2 = 0.652<br>P3 = 0.478 |
| Week 5       | 0.94 $\pm$ 0.25          | 1.06 $\pm$ 0.24<br>P1 = 0.392 | 0.99 $\pm$ 0.13<br>P2 = 0.629<br>P3 = 0.514 |
| Week 6       | 0.94 $\pm$ 0.25          | 1.06 $\pm$ 0.23<br>P1 = 0.382 | 0.99 $\pm$ 0.14<br>P2 = 0.637<br>P3 = 0.495 |

One way ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.

P1 compare group B vs. group A

P2 compare group C vs. group A

P3 compare group B vs. group C

**Table 2.** Comparison between the studied groups regarding the length (cm) of *Planaxis sulcatus* throughout study period, (n = 24)

| Study period | Shield length in (cm)    |                               |   |
|--------------|--------------------------|-------------------------------|---|
|              | Group A<br>Mean $\pm$ SD | Group B<br>Mean $\pm$ SD      | Group C<br>Mean $\pm$ SD                    |
| Week 0       | 1.32 $\pm$ 0.13          | 1.30 $\pm$ 0.07<br>P1 = 0.702 | 1.30 $\pm$ 0.10<br>P2 = 0.724<br>P3 = 1.00  |
| Week 1       | 1.29 $\pm$ 0.17          | 1.34 $\pm$ 0.13<br>P1 = 0.485 | 1.28 $\pm$ 0.06<br>P2 = 0.898<br>P3 = 0.231 |
| Week 2       | 1.29 $\pm$ 0.18          | 1.29 $\pm$ 0.12<br>P1 = 0.769 | 1.29 $\pm$ 0.09<br>P2 = 0.861<br>P3 = 0.841 |
| Week 3       | 1.26 $\pm$ 0.15          | 1.25 $\pm$ 0.10<br>P1 = 0.600 | 1.27 $\pm$ 0.09<br>P2 = 0.591<br>P3 = 0.992 |
| Week 4       | 1.27 $\pm$ 0.12          | 1.29 $\pm$ 0.12<br>P1 = 0.303 | 1.29 $\pm$ 0.08<br>P2 = 0.340<br>P3 = 0.751 |
| Week 5       | 1.27 $\pm$ 0.12          | 1.32 $\pm$ 0.11<br>P1 = 0.170 | 1.32 $\pm$ 0.07<br>P2 = 0.169<br>P3 = 0.735 |
| Week 6       | 1.27 $\pm$ 0.12          | 1.32 $\pm$ 0.11<br>P1 = 0.172 | 1.30 $\pm$ 0.06<br>P2 = 0.306<br>P3 = 0.442 |

One way ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.

P1 compare group B vs. group A

P2 compare group C vs. group A

P3 compare group B vs. group C

**Table 3.** Comparison between the studied groups regarding the aperture (cm) of *Planaxis sulcatus* throughout study period, (n = 24)

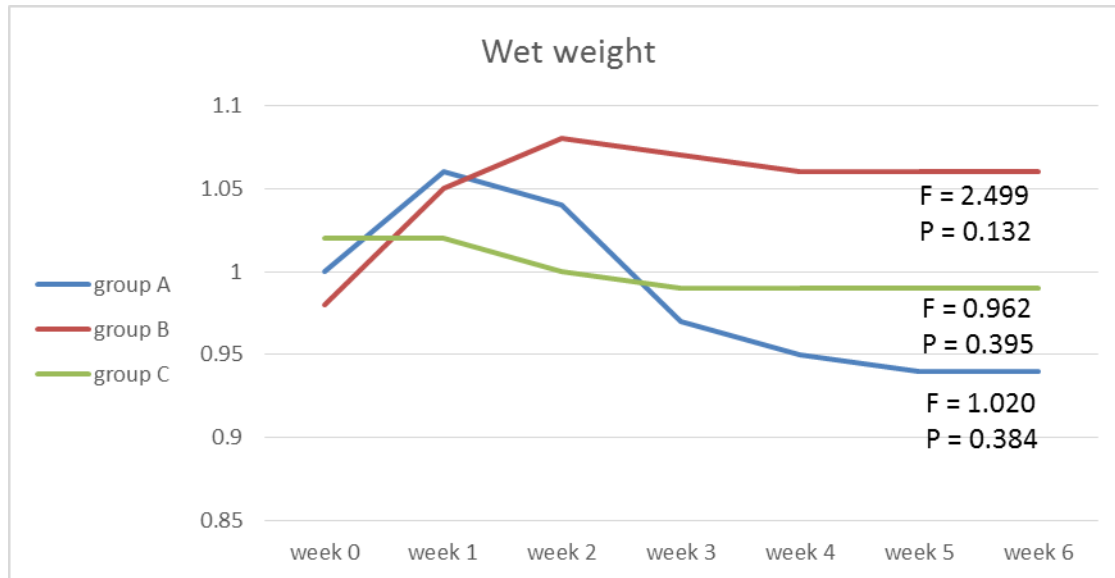
| Study period | Shield aperture (cm)     |                               |   |
|--------------|--------------------------|-------------------------------|---|
|              | Group A<br>Mean $\pm$ SD | Group B<br>Mean $\pm$ SD      | Group C<br>Mean $\pm$ SD                    |
| Week 0       | 0.99 $\pm$ 0.14          | 0.98 $\pm$ 0.08<br>P1 = 0.768 | 0.99 $\pm$ 0.06<br>P2 = 0.937<br>P3 = 0.726 |
| Week 1       | 0.99 $\pm$ 0.14          | 0.98 $\pm$ 0.08<br>P1 = 0.874 | 0.99 $\pm$ 0.06<br>P2 = 0.874<br>P3 = 1.00  |
| Week 2       | 0.99 $\pm$ 0.13          | 1.00 $\pm$ 0.08<br>P1 = 0.834 | 1.00 $\pm$ 0.05<br>P2 = 0.873<br>P3 = 0.905 |
| Week 3       | 1.00 $\pm$ 0.10          | 0.99 $\pm$ 0.07<br>P1 = 0.923 | 1.00 $\pm$ 0.05<br>P2 = 1.00<br>P3 = 0.882  |
| Week 4       | 0.99 $\pm$ 0.10          | 1.00 $\pm$ 0.05<br>P1 = 0.913 | 0.99 $\pm$ 0.04<br>P2 = 0.988<br>P3 = 0.869 |
| Week 5       | 0.99 $\pm$ 0.11          | 1.00 $\pm$ 0.07<br>P1 = 0.850 | 0.99 $\pm$ 0.05<br>P2 = 0.936<br>P3 = 0.674 |
| Week 6       | 1.00 $\pm$ 0.11          | 1.00 $\pm$ 0.06<br>P1 = 1.00  | 0.98 $\pm$ 0.04<br>P2 = 0.687<br>P3 = 0.527 |

One way ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.

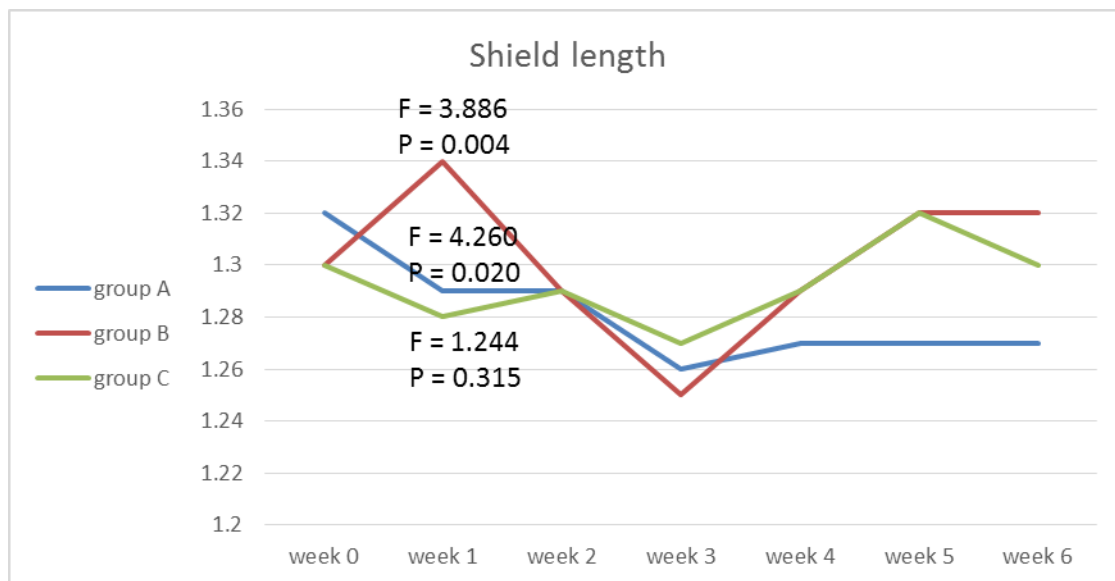
P1 compare group B vs. group A

P2 compare group C vs. group A

P3 compare group B vs. group C

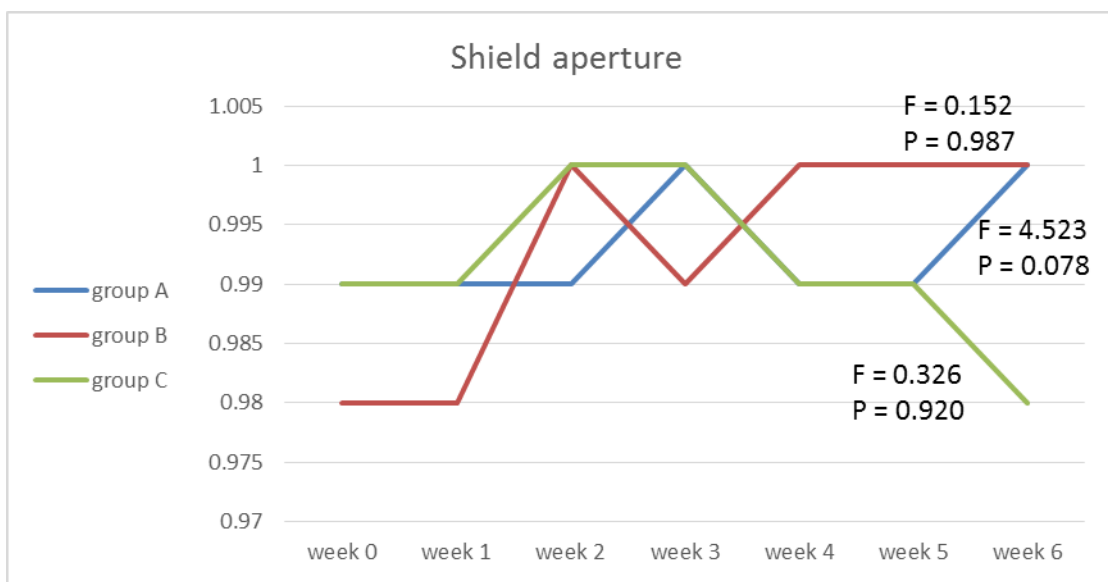


**Fig. 1.** Mean wet weight (g) of *Planaxis sulcatus* in the studied groups C throughout the study period (n = 24) using repeated measures. ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.



**Fig. 2.** Mean shield length (cm) of *Planaxis sulcatus* in the studied groups C throughout the study period (n = 24) using repeated measures. ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.





**Fig. 3.** Mean shield aperture (cm) of *Planaxis sulcatus* in the studied groups C throughout the study period (n = 24) using repeated measures. ANOVA test was used with Bonferroni post hoc test. A p-value less than 0.05 was considered statistically significant.

## DISCUSSION

There is a high biodiversity in the Red Sea which is located in one of the warmest regions in the world (Dreano *et al.*, 2016). Although species found are small sized, it is still an aquatic ecosystem that has not been studied sufficiently when compared to other water ecosystems such as the Caribbean Sea and the Great Barrier Reef (Bruckner *et al.*, 2013; Berumen *et al.*, 2013). Few studies have examined the impact of temperature elevation on sea snail in the Red Sea, especially at the coast of the kingdom of Saudi Arabia on which almost no studies was conducted.

This study is an experimental animal study including 72 *Planaxis sulcatus* snails collected from Al-Rais region in the kingdom of Saudi Arabia. This work was organized to study the physiological impact of near future temperature elevation due to climate changes' effect on sea snail growth at the coast of kingdom of Saudi Arabia on the Red Sea. The current findings revealed that, the seawater measurements of temperature, salinity and the pH increased gradually in group B and group C. This resulted in no significant differences between weekly measures of the wet weight in intragroup comparison of the three groups A, B and C. In addition, when comparing the 3 groups, wet weight of studied snails also showed no significant differences. However, regarding shield length, it increased at week 1 with pairwise comparison of weekly shield length records that showed only a significant difference between the 1st week reading and that of the onset of the study in group A; the control group. In group B, the shield length increased at week 6 with pairwise comparison of weekly shield length records showing only a significant difference between week 3 and that of the 6<sup>th</sup> week. While, group C

showed no significant differences between weekly measures of shield length. In addition, when comparing the 3 groups, the shield length of the studied snails also showed no significant differences.

The shield aperture of the studied snails showed no significant differences between weekly measures in group A, B and C throughout the study period. Furthermore, when comparing the 3 groups, no significant differences was detected. The present results showing no differences between the control group and others with temperature elevation may be due to acclimation. These results are in accordance with a previous study which investigated acclimation temperature on thermal resistant at upper and lower lethal temperature. Snails acclimated to 10, 20 and 30C showed reasonable heat and cold acclimation (**Al-Khateeb, 2006**).

This adaptation helps gastropods to survive in environments such as the intertidal zone where low tide temperature exceeds 52°C on coasts which are tropical and rocky (**Marshall et al., 2013**). Organisms living in the low tide zone can tolerate higher temperature (**Knox, 2001**). The reason for their survival in high temperature may be due to their cone shaped and high-spired shells (**Vermeij, 1973**).

A previous study concluded that the pyramid-building behavior in the *P. sulcatus* is a thermoregulatory adaptation. Therefore, temperature can be predicted to influence the pyramid-building behavior in the *P. sulcatus* as the previous authors did not observe pyramid-building behavior among the Gulf periwinkles during winter in eastern Bahrain (**Kaminski & Garrison, 2020**). But, this behavior was not addressed in the current study.

Eminently, similar changes in depth range can be expected, as species shift down in the water column to escape warm surface waters. While these migrations seem like viable adaptations, it is unclear how successful species can be when moving across these distances and depths. Life history characteristics that rely on other environmental cues, such as day length and tidal cycle, may not adapt fast enough for continued success (**Galland et al., 2009**).

It is worthy to mention that water warming has many consequences; one of which is elevation of sea level. Water expands and water surface rise when its temperature is elevated. By now, this is limited to the surface layer which is only a few hundred meters deep (**Domingues et al., 2008**). This heat diffuses overtime to more depth, leading to more expansion and higher sea level due to melting of inland glaciers and continental ice sheets as those resting on Antarctica. Previous studies estimated that sea level would rise by 0.5-0.8 m over 1990 levels by 2100, and that this rise might be more than one meter. Recent studies concluded that the mean sea-level rise of 0.5-0.8 m over 1990 levels by 2100 is likely, and that a rise of more than one meter in thae predicted time is possible (**Richardson et al., 2009**). These significant changes cause more dangerous storm surges and flooding that can occur regularly (**McMullen, 2009**). Changes in the ocean

temperature and climate change have significant impact on marine ecosystems and human beings depending on them (Sommerkorn & Hassol, 2009).

## CONCLUSION

In conclusion, most results were statistically non-significant, which meant no significance difference was detected between the three groups regarding wet weight, shield length and shield aperture. Hence, no relationship was found between temperature elevation and those parameters. Therefore, it is recommended to conduct complex experiments that depend on exposing snails to several climate change factors, such as increased acidity and salinity in addition to high temperature.

## ACKNOWLEDGMENTS

The author would like to thank Hossam Alharbi, Turki ALMuzaini, Eyad Alharbi, Abdulrahman Almughadoi on their effort to collect part of the information in this work.

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