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Climate Change: The Effect of Increasing Temperature on Caenorhabditis Elegans

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ABSTRACT

The effect of temperature change on organisms continues to be an area of interest as global temperatures rise. The *Caenorhabditis elegans* (*C. elegans*) is a small, transparent nematode that is commonly used as a model organism in biology research due to its short life cycle with distinct phases of life. The *C. elegans* wild-type N2 strain*, Strain A*, and *Strain B* were observed at 22ºC, 22.5ºC, and 23ºC. The progeny numbers for all three strains at the indicated temperatures were quantified and the embryonic lethality of each was calculated. An increase in temperature was correlated with an increase in progeny number for *Strain A* and *Strain B,* while no statistical difference waws found in wild-type N2. A decrease in embryonic lethality from the first day of exposure to the new surrounding temperature to the second day was observed for all three strains. These results suggest. That temperature change can have a significant effect on the progeny number and the adaptability of *C. elegans* to their surrounding s and genetic variation can affect an organism's physiological response.

Introduction

Caenorhabditis elegans (*C. elegans*) are small, transparent, soil-dwelling nematode worms that are commonly used as a model organism in the fields of biology and genetics. It is about 1 millimeter in length and can be found in a variety of environments, including composts, decaying fruits and vegetables. *C. elegans* are widely studied because it reproduces rapidly and has a simple and well-defined nervous system, a short lifespan, and a transparent body that allows for easy observation of its internal structures. *C. elegans* are highly adaptable to differing circumstances, and the nematode has been used to study a wide range of biological processes, including development, genetics, neurobiology, and aging.

The rate of living theory states that an organism's metabolism, or the rate at which it uses energy, is positively correlated with its rate of aging and therefore lifespan. According to this theory, organisms with a higher metabolic rate age faster and have shorter lifespans, while those with a lower metabolic rate age slower and have longer lifespans. This is because a higher metabolism requires more energy to be used, which in turn generates more harmful byproducts such as reactive oxygen species (ROS) leading to a faster accumulation of cellular damage. The rate of living theory has been traditionally used to explain the relationship between temperature and lifespan (Loeb, J., & Northrop, J. H., 1916). However, recent studies have shown that genes also play a critical role in regulating lifespan in *C. elegans* in response to temperature changes.

Temperature changes trigger a variety of complex behaviors in *C. elegans*. Due to the simplicity of the nervous system in *C. elegans*, the mechanisms of physiological behaviors in *C. elegans* have been extensively studied at the circuit and molecular levels. A temperature of 20°C is the canonical condition for maintaining *C. elegans* in the lab with *E. coli* OP50 bacteria as the worm's diet (Brenner S., 1974). However, 15°C–25°C temperature maintenance with *C. elegans* is considered physiological. Temperatures outside of this range are considered harmful as the extreme temperatures threaten the organism's survival and the physiology and development of *C. elegans* (http://www.wormbook.org). Exposure to different temperatures has been shown to

have major effects on the physiological behavior of *C. elegans*, such as longevity or developmental time (Gómez-Orte et al., 2017). For example, the mean lifespan of *C. elegans* maintained at 20°C increases 60% when kept at 16ºC or decreases 50% when kept at 24°C (Klass, 1977). While much is known about temperature regulation and aging of *C. elegans*, it remains largely unclear how temperature affects progeny number/fertility and embryonic lethality of various strains of *C. elegans.*

One area of research that has garnered significant attention is the effect of temperature on *C. elegans* development and behavior. This research paper will explore the various ways in which temperature impacts *C. elegans*, including its progeny number, embryonic lethality, and metabolism. The Wild-type N2 strain, *Strain A*, and *Strain B* will be observed. Understanding the temperature sensitivity of *C. elegans* can provide insight into the broader effects of temperature on biological systems and potentially inform the development of temperature-based therapies.

Materials and Methods

The *C. elegans* strains tested were the wild type (N2), *Strain A,* and *Strain B*. All *C. elegans* strains were maintained on MYOB plates seeded with *Escherichia coli* (*E. coli*) OP50 at 20ºC. The hermaphrodites were raised at 20°C until the L4 stage was reached. *Strain A* and *Strain B* were derived from the wild-type Bristol N2 strain and vary in genetic makeup. Individual L4 hermaphrodites were transferred to new plates and allowed to produce progeny for 0-24 hours in temperature-controlled incubators at 22ºC, 22.5ºC, and 23ºC for genetic analysis. After 24–48 hours, the young adult hermaphrodite was moved to a new plate and placed at the same temperatures. All plates were seeded with *E. coli.* Progenies on each plate were allowed to develop for 22-24 hours before quantifying the number of dead eggs and larvae. To reduce internal incubator differences, plates were always placed on the same shelf and in the same location.

Results

Wild-Type N2 Strain

In the Wild-type N2 strain, no pattern between an increase in surrounding temperatures and progeny number was observed (see Figure 1). Average progeny numbers of 25.67, 64.67, and 49.67 were calculated for surrounding temperatures of 22ºC, 22.5ºC, and 23ºC respectively. A t-test was performed to measure the correlation between temperature and progeny number in the Wild-type N2 strain. A p-value of 0.081 and a linear regression line of y = -493.333 + 24x were calculated. No relationship between the increase in temperature and progeny number in the Wild-type N2 strain was observed, $t(7) = 1.565$, $p = .081$. The null hypothesis $H_0 : \beta = 0$ is accepted as there was no statistically significant evidence that $\beta > 0$ in 95% confidence intervals.

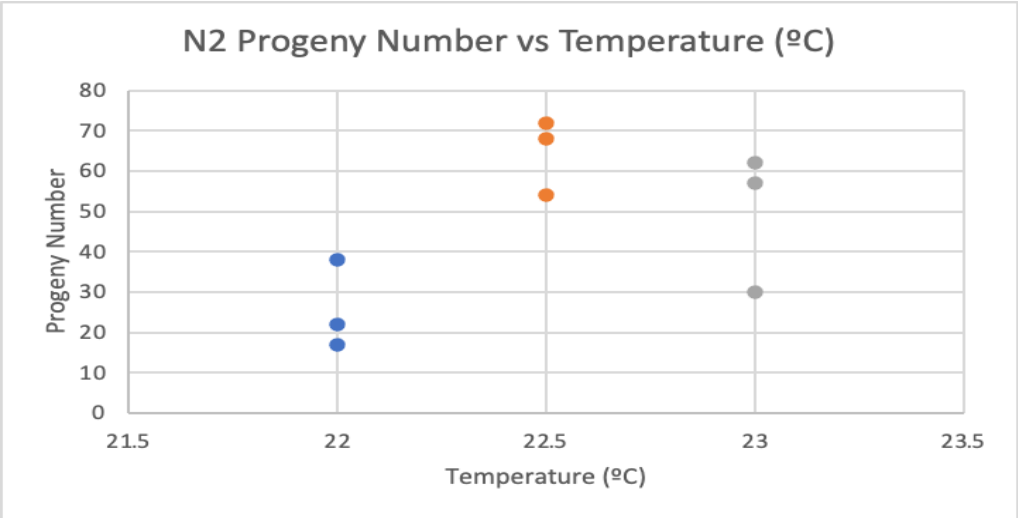

Figure 1. Wild-type N2 Progeny Number vs Temperature (ºC), 0-24 hours. The progeny numbers at the three temperatures are depicted. Each data point represents a plate of the N2 *C. elegans*.

Strain A

In *Strain A*, an increase in progeny number was observed with an increasing surrounding temperature (see Figure 2). Average progeny numbers of 46.21, 61.93, and 72.36 were calculated for surrounding temperatures of 22ºC, 22.5ºC, and 23ºC respectively. The correlation between temperature and progeny number in *Strain A* was $t(69) = 4.052$, $p = 6.550 \times 10^{-5}$. A linear regression line of $y = -518.278 + 25.782x$ was calculated. The null hypothesis $H_0 : \beta = 0$ is rejected as there was statistically significant evidence that $\beta > 0$ in 95% confidence intervals.

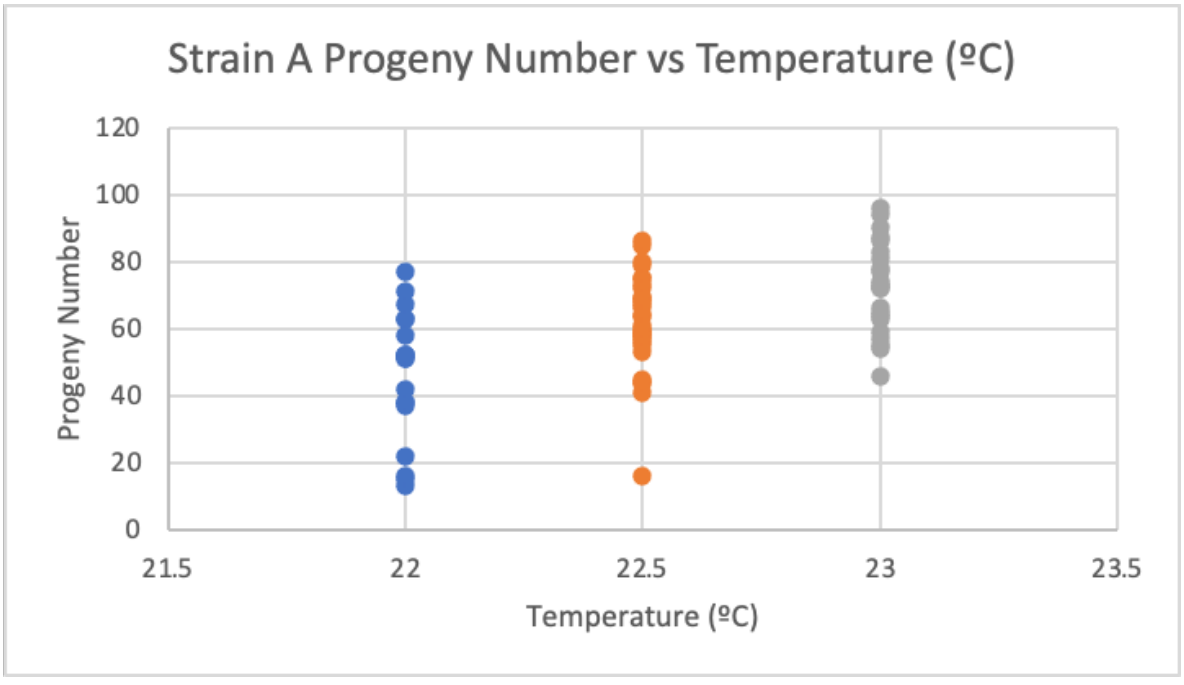

Figure 2. Strain A Progeny Number vs Temperature (ºC), 0-24 hours. The progeny numbers at the three temperatures are depicted. Each data point represents a plate of the *Strain A C. elegans*.

Strain B

In *Strain B*, an increase in progeny number was observed with an increasing surrounding temperature (see Figure 3 3). Average progeny numbers of 50.63, 69.41, and 71.70 were calculated for surrounding temperatures of 22ºC, 22.5ºC, and 23ºC respectively. A linear regression line of y = -393.840 + 20.382x was calculated, and the correlation between temperature and progeny number in *Strain B* was t(72) = 3.893, p = 1.095 x 10^{-4}. Thus, the null hypothesis H_0 : β = 0 is rejected as there was statistically significant evidence that β > 0 in 95% confidence intervals.

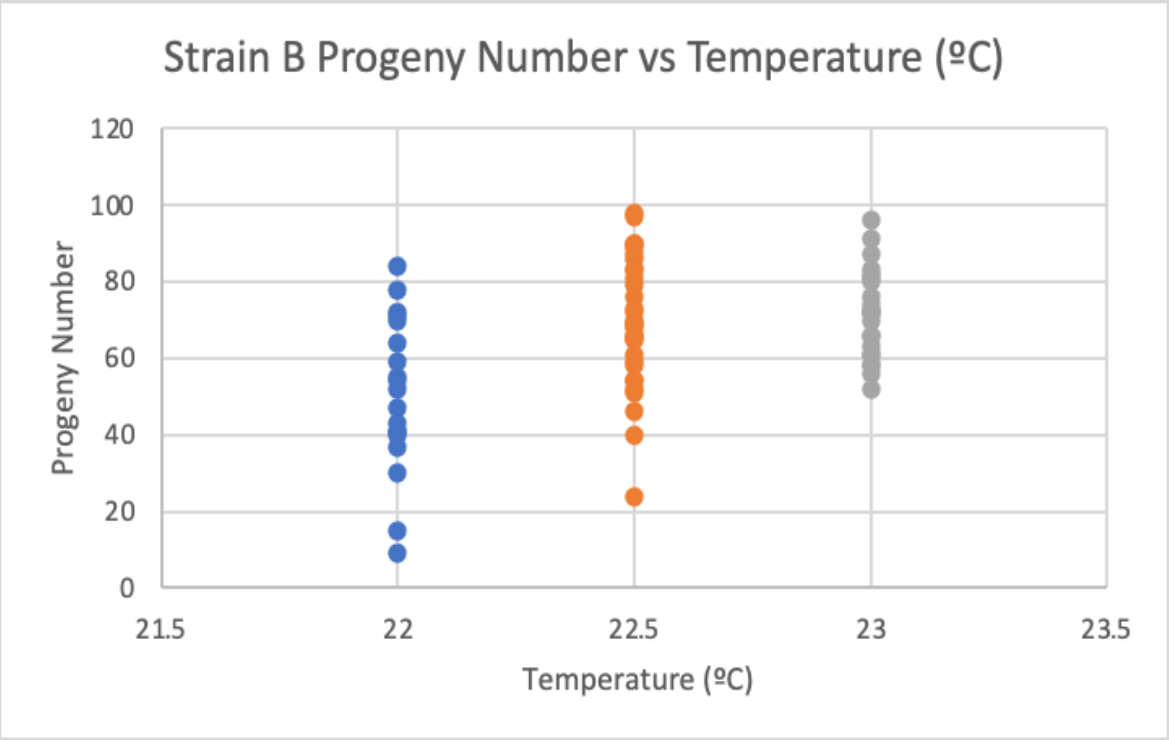

Figure 3. Strain B Progeny Number vs Temperature (ºC), 0-24 hours. The progeny numbers at the three temperatures are depicted. Each data point represents a plate of the *Strain B C. elegans*.

Conclusion

There was no correlation between an increase in temperature and progeny number found for the Wild-Type N2 strain of *C. elegans*. Following a t-test, there was no statistical evidence to disprove the null hypothesis, indicating that increasing growth temperatures did not affect the total progeny number of Wild-type N2 strain within the 0-24 hour time frame.

However, genetic differences between strains of *C. elegans* have been shown to impact the nematode's behavior in different temperature settings. In Strain A and Strain B, a relationship between progeny number and temperature was found. As the surrounding temperature increased, an increase in the number of eggs and larvae was counted. The t-tests performed were in agreement that there was a correlation between the two variables. The results suggest that increasing surrounding temperatures in the physiological range (15ºC-25ºC) leads to

an increase in metabolism in certain strains of *C. elegans*. The increased metabolism may result in an increase in reproductive activity and more progeny laid during the 0-24 hour period. This relationship was only observed for two of the three strains tested. This suggests that the response of the nematode to the change in surrounding temperature may differ by strain and genetic makeup.

In summary, a correlation between increased reproductive activity and increasing surrounding temperatures was observed for *Strain A* and *Strain B* at temperatures of 22ºC, 22.5ºC, and 23ºC. This correlation was not observed for the Wild-type N2 strain. The data show that differences in genetic make up of *C. elegans* affects the nematode's physiological response to different surrounding temperatures. The relationship between progeny number and temperature suggests that an increase in surrounding temperature may boost metabolism of *C. elegans* and increase reproductive activity. The results demonstrate the significant effect temperature has on the behavior of *C. elegans*. Observing and analyzing the effects of temperature on *C. elegans* may contribute to the development of new temperature-based therapies.

Applications

This experiment demonstrates that within one species, genetic makeup can drastically change an organism's physiological response to outer stimulus, such as changes in environment temperature. The three strains observed within this study demonstrated different optimal temperature ranges for survival. As global temperatures continue to fluctuate, it is important to be aware of this difference in reaction even within one species, and to treat differences within one group with sensitivity.

Limitations

This study observed the *N2, Strain A* and *Strain B* types of *C. elegans* at 22ºC, 22.5ºC, and 23ºC. The behavior of other strains of *C. elegans* at temperatures varying from those tested within this study are not shown in the results.

Future Research

Future research with different temperatures for the same strains may be tested. The same temperatures with different *C. elegan* strains may also be tested to further understand different strains and their reactions to various surrounding temperatures.

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