

**Evaluation of Longevity in *Drosophila melanogaster*
Through 7-Keto Dehydroepiandrosterone**

Department of Science, AP Research

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Abstract

The objective of this study was to observe whether or not a natural metabolic supplement, claiming to increase metabolism, was effective in its claims. This study was performed with wild type *Drosophila melanogaster* as well as the natural metabolic-boosting supplement, 7-Keto Dehydroepiandrosterone, more commonly known as 7-Keto DHEA. With a lack of research backing the supplement's overall effectiveness, 7-Keto DHEA was expected to increase the metabolic rate of *D.melanogaster*, subsequently decreasing the organism's lifespan. Ultimately, it has been previously hypothesized that if the 7-Keto DHEA was effective, there would be a negative correlation regarding the lifespan of the *D. melanogaster*. Following the administration of the supplement, the lifespan of each *D. melanogaster* was recorded and an average was taken for each dosage level. In order to determine whether or not there was statistical significance, a Chi-Square test was utilized, concluding that there is a negative relationship between the administration of 7-Keto DHEA and *D. melanogaster*. Future research studies may determine whether or not these results are also applicable to humans, allowing consumers to be aware of the supplements that they are consuming.

Key Words: 7-Keto-DHEA, metabolism, lifespan, *Drosophila melanogaster*

Introduction

As defined by the Mayo Clinic, metabolism is, “the process by which the body changes food and drink into energy,” (Mayo Clinic 2022). As obesity rates have started to rise over time

	18 - 24	25 - 34	35 - 44	45 - 54	55 - 64	65 or older		18 - 24	25 - 34	35 - 44	45 - 54	55 - 64	65 or older
National							National						
Value	16.5	28.2	33.0	35.9	35.4	28.5	Value	19.5	30.9	35.5	38.1	36.3	29.3
95% CI	15.7 - 17.4	27.4 - 29.0	32.2 - 33.9	35.1 - 36.6	34.6 - 36.1	28.0 - 29.1	95% CI	18.5 - 20.6	30.0 - 31.9	34.5 - 36.5	37.1 - 39.0	35.4 - 37.2	28.6 - 29.9
Sample Size	23,516	41,784	46,647	62,722	88,110	145,669	Sample Size	22,611	38,280	44,948	52,842	69,210	125,950

Figure 1: This table comparison shows adult obesity rates from 2017 which is on the left to 2020 which is on the right (Centers for Disease Control and Prevention [CDC] 2020)

in the US,
the amount
of obese
adults has

climbed from 30.5% in 2017 to 41.9% in 2020 according to the Centers for Disease Control and Prevention (Centers for Disease Control and Prevention [CDC] 2022). The specific rates for obesity have also varied throughout the years 2017-2020 (**Fig. 1**). In order to combat this, many of these afflicted individuals have turned to easily-accessible weight loss drugs, which combat the need for a lot of the hard work previously associated with weight loss. Some of these recommended tactics according to Harvard School of Public Health, include, “limiting unhealthy foods and beverages, increasing physical activity, limiting television time, improving sleep, and reducing stress,” (Harvard 2022). However, there is also an increasing amount of online information, which includes the many “easy” ways to lose weight. Looking up “best supplements to use weight,” and clicking on the first source that pops up, WebMD states that, chitosan, chromium picolinate, conjugated linoleic acid, glucomannan, green tea extract, green coffee extract, guar gum, hoodia, 7-Keto DHEA, Ephedra, and bitter orange are the best supplements to take for weight loss (WebMD 2022).

In this source from WebMD, it states that, “[7-Keto DHEA] may help you lose pounds by boosting your metabolism so you burn more calories throughout the day,” however, later on, it

states, “...there still isn’t enough reliable evidence to rate how well it works,” (WebMD 2022).

Ultimately, there is high publicity that this is a beneficial supplement for weight loss, but it later



Fig 2: This image is the supplement that I will be using for my research, 7-Keto DHEA (Vitamin Shoppe 2022).

establishes that there is not enough evidence to fully evaluate whether or not it really is as effective as claimed. For my research, I will be using 7-Keto Dehydroepiandrosterone, or 7-Keto DHEA, as it is often recommended as a weight loss supplement (**Fig. 2**). 7-Keto DHEA is found naturally in the body, and functions to convert steroid hormones that include androgen and estrogen (WebMD 2022).

Overall, the effect of the weight loss supplement, 7-Keto Dehydroepiandristerone on the lifespan of *D. melanogaster* is relevant for potential future research. This ultimately led me to question, to what extent is 7-Keto DHEA an effective metabolite

as measured by lifespan on *Drosophila melanogaster*?

This study is being performed on *D. melanogaster*, meaning the study directly to humans cannot occur without further research. However, following the observation of the impact of 7-Keto DHEA on *D. melanogaster*, further research could be conducted in order to determine the impacts of weight loss supplements on the lifespan of humans. The current research and knowledge surrounding 7-Keto DHEA, demonstrate that it appears to be a promising weight loss supplement. However, if it has a negative correlation with the lifespan, the people that take this supplement should be informed, to make further decisions regarding the methods in which they would like to move forward, and the overall risks that they are willing to take.

Literature Review

Drosophila melanogaster

Drosophila melanogaster are an ideal model organism to use in my research because of their relatively short lifespan, averaging around 2-3 months. In addition, their genetic similarities to humans are roughly around 50% (Sun et. al 2015). While this does mean that this study does not directly apply to humans, there are still significant results that could potentially be obtained, sparking potential new research.

Throughout my research, I came across a study by Mateusz Molon that ultimately helped shape my academic research question. Within this academic paper, Molon observed the non-genetic factors that impact age, which includes temperature, level of stress, and diet. In addition, this study found that temperature may impact metabolism, which likely impacts the lifespan of the *D. melanogaster*. Throughout this study, it was observed that with an increased temperature, the amount of metabolic heat in many strains of wild type *D. melanogaster*, which represents a higher amount of heat flow through a metabolic process (**Fig. 3**) (Molon et. al 2020). An increased metabolic rate had also been observed to be directly proportional to a decreased life expectancy, essentially meaning that species that have a faster metabolism, have a shorter lifespan, and those with a slower metabolism live a longer life (Olshansky 2005). Therefore, through the observation of the increased metabolic heat produced at higher temperatures, the *D. melanogaster* are

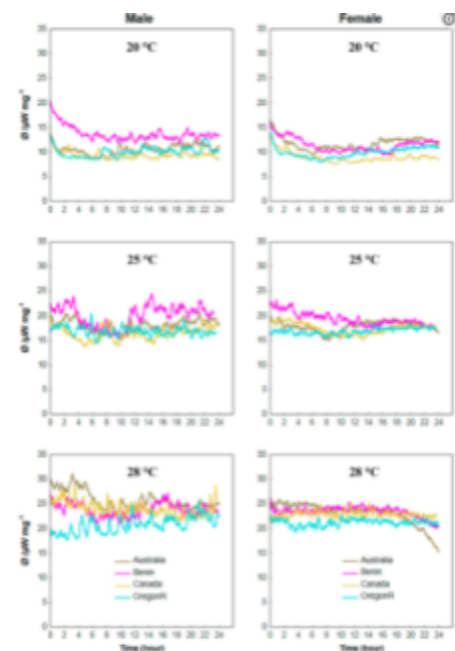


Figure 3: This figure shows the impact of higher temperatures on metabolic heat in *D. melanogaster* (Molon et. al 2020)

exhibiting a higher metabolism, and therefore, it is likely that it will decrease their lifespan. This ultimately connects to the rest of the study done by Molon, as when testing solely lifespan and temperature, the *D. melanogaster* had a decreasing lifespan as the temperature rose (Molon 2020).

Metabolism in Humans

Overall, metabolism occasionally shares a relatively similar process among many organisms, it can vary between the organisms based on the different sources of energy and carbon (LibreTexts 2022). Humans have a slow metabolism, however, a slow metabolism allows humans to live longer than some other

species (**Fig. 4**). Humans and other primates that are similar to humans burn 50% fewer calories per day than other mammals, ultimately meaning they have a slower metabolism.

Because of this, some of their life

processes are slowed down, and it

ultimately causes them to live longer (Ellis 2014). However, while humans and other primates similar to humans do have slower metabolisms, there are also differences between males and females to the rate of their respective metabolisms. Overall, women do tend to have a slower metabolism than men, however, this is more as a result of muscle mass, not gender. One of the reasons for this is that women have a greater proportion of slow twitch fibers, meaning that they use more fat during exercise than men, however after exercise, men use more fat than women. Consequently, men have more muscle mass, and they also have it at a more consistent rate,

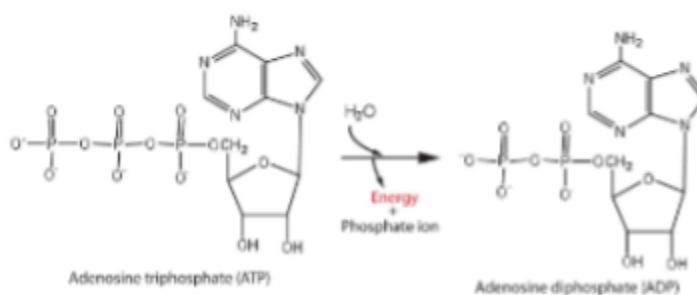


Figure 4: This figure depicts the energy produced through the hydrolysis of ATP to ADP (LibreTexts 2022).

explaining why their metabolism is often considered higher than women (Rogers 2022).

Although the differences between male and females are not exponential, they are still critical to consider when observing how metabolism is impacted with various supplements.

7-Keto DHEA

Learning about 7-Keto Dehydroepiandrosterone was vital in order to understand what exactly it does in the human body. It also explains why humans can potentially benefit from taking additional amounts of 7-Keto DHEA in this supplement form. In natural form, it has a role

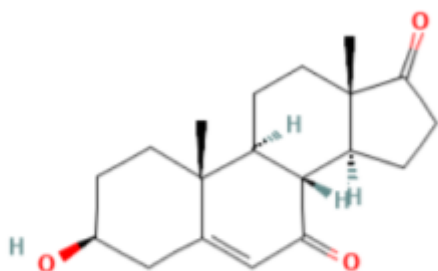


Figure 5: This figure depicts the chemical makeup of 7-Keto DHEA (PubChem 2022).

in the human blood as a serum metabolite, a prohormone, and a nutraceutical (PubChem 2022).

This essentially means that it regulates hormone expression, and helps with metabolic processes

(**Fig. 5**). In addition to this, because they are a nutraceutical, although they are monitored by the Food and Drug Administration, or the FDA, the regulation is much different as they provide a

health benefit beyond the traditional nutrients consumed in a typical day-to-day diet (News Medical 2022). These dietary supplements are monitored differently as a result of the Dietary Supplement Health and Education Act of 1994. This essentially stated that if specific items met the requirements to be considered a supplement, they would subsequently be monitored by the FDA in alternative ways (National Institutes of Health 1994). Because of this, there are more limitations that are placed on these supplements, but there are also gaps in how they are monitored since this act is so new. In the form of a supplement, 7-Keto DHEA is often taken for obesity, muscle strength, depression anxiety, etc. There is a possibility that 7-Keto DHEA is

successful in promoting weight loss in that it speeds up the metabolic processes of the body and converts energy consumed by food into heat, as opposed to storing it as fat (WebMD 2022).

The Established Gap

Regarding the current scholarly conversation there is a distinct correlation that establishes that increasing heat will consequently increase metabolism in *D. melanogaster* (Molon 2020). In addition, there is a correlation between increased metabolism leading to a decrease in lifespan (Olshansky 2005). The gap that I am exploring is in reference to whether or not 7-Keto DHEA is a successful weight loss supplement. There are claims that produce possible evidence that taking supplements of 7-Keto DHEA can increase metabolism, and lead to weight loss; however, there is still a debate as to whether or not it truly is successful in increasing metabolism. Although these results do not directly correlate to humans, by examining whether or not 7-Keto DHEA really is a successful weight loss supplement in *D. melanogaster*, which are a similar model organism to humans, it could potentially lead to further research in the future, directly involving humans in the process. As of now, the effectiveness of 7-Keto DHEA has not been fully established, allowing me to contribute to the scholarly conversation that is currently in place, and potentially sparking ideas for further research on this topic.

Methodology

Initial Fly Care

For my research, I had initially planned on purchasing the *D. melanogaster*; however, I was donated the flies by the Rock Canyon Biotechnology Department. This department has set up the initial vial, which contained the flies, 5 grams of Formula 4-24 Instant Drosophila Medium as well as 22 mL of tap water. Throughout the rest of my research, I will continue to use this same measurement for the media in my vials, only slightly differing for the vials that I add

the 7-Keto DHEA to. The control groups, however, will maintain the same exact measurements of the media in each vial. For the month of December, and slightly into January, I will allow the flies to breed and reproduce, in order to obtain a high amount of flies once I begin my research. Roughly twice a week I will split the flies into more vials in order to provide them more space to live, as well as to prevent overcrowding of any single vial. In addition, this will help to ensure that if one vial does contract a virus or mutation, it will not impact all of the other flies, just the flies in that vial, which will ultimately not be used in my research as it has the potential to skew the data.

Age Synchronization

A critical part in this study is to ensure that all of the *D. melanogaster* in each vial are the same age. Because I am looking at the impacts of metabolism on lifespan, it is vital that the flies in each of their respective vials are the same age, as my results will be based on how long they live. Through making sure that all of the flies in the vial are the same age, I will be able to collect accurate data, that will be representative of the impacts of 7-Keto DHEA, and allow me to determine whether or not there is a statistically significant impact in metabolism through taking this supplement, as monitored through the lifespan of the flies.

Collecting Virgins

Once it is time to start my trials, I need to ensure that all of my *D. melanogaster* are virgins. In order to successfully do this, I need to release all *D. melanogaster* that are adults, or in other words all of the *D. melanogaster* that can fly, or are hatched from their eggs. The reason that flies need to be released at such a specific time is because they are able to reproduce after 8-10 hours of life (Berg Lab 2022). Ultimately, I have to release the flies 8 hours before I am ready to sex them, which will ensure that they are virgins. In order to ensure that all of the flies

in the vial are the same age, all of the flies that I collect in a single day will be placed in the same vial based on their sex. Since it is unlikely that all of the 40-60 flies I need for each trial will hatch in the same time, this will occur over an extended period of time, in order to ensure that the ages remain constant.

Anesthetizing the *D. melanogaster*

As established above, temperature plays a vital role in the metabolism of the *D. melanogaster*. However, to reduce the higher chance of death that is associated with anesthetizing the flies using FlyNap, a gas anesthetic for flies, I will place the *D. melanogaster* into the freezer for roughly 5 minutes, where I will then select a few at a time to place under a microscope and determine the sex. Through this process, I will monitor whether or not they are still anesthetized, to ensure that I do not lose any of my flies during this process. I will place the flies under a microscope and begin to sex them, to ensure that the chances of reproduction among these flies are eliminated, allowing me to have consistent data, as well as keeping the age of the flies the same.

Sexing the *D. melanogaster*

In order to ensure that the *D. melanogaster* will not reproduce once I have determined

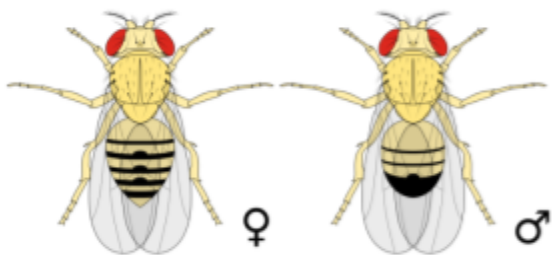


Figure 6: This figure shows the differences between male and female *Drosophila melanogaster* (Udayangani 2020).

them to be the same age, I need to separate the flies based on their respective sexes (**Fig. 6**). Following the anesthetization of my *D.*

melanogaster, I will place them under a microscope, using a petri dish.

Throughout this process, I will identify the sex of each of the flies through key details; in males I

will look for a shorter, and more rounded abdomen as well as sex combs, whereas in females I will look for a longer abdomen with a pointed end (Udayangani 2020). As the flies are still anesthetized, I will separate them into a male vial, and a female vial, ensuring that there is no chance of reproduction, and I will use these separated vials to ultimately begin collecting data for my study.

Making 7-Keto DHEA Media

My set up for my research will involve eight vials, at varying dosages. Because I have to separate out the males and females in order to achieve an accurate representation of the change in lifespan among the *D. melanogaster* there will be four dosages, with two vials per dosage, a male and a female vial. In research done by Kamal Patel, it is recommended to take anywhere from 200-400 mg per day in humans (Patel 2022). I then utilized this to determine the dosages for my various vials, creating a low dosage which was the equivalent of 200 mg, a medium dosage at the equivalent of 300 mg, and a high dosage which was the equivalent of 400 mg (**Fig. 7**). However,

	Low	Medium	High
Male	0.0018 g	0.0027 g	0.0036 g
Female	0.0021 g	0.0032 g	0.0043 g

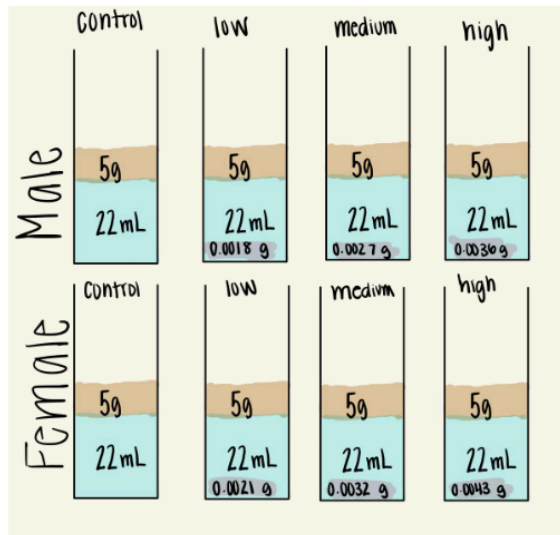
Figure 7: This table shows the dosages that were calculated at each dosage for the *D. melanogaster*, each of which were converted from the weight of a human, to the weight of the *D. melanogaster*.

in order to
establish a
negative
control,
one male

vial and one female vial will have the normal 5 grams of Formula 4-24 Instant Drosophila Medium, mixed with 22mL of tap water. In order to ensure that the *D. melanogaster* get the proper amount of nutrition, I will replace the media every two weeks, unless they have all died before this benchmark.

Finalized Experimental Design

In order to perform the research established in my research question, I ran one trial with



(Figure 8) This image shows the setup of my vials, each of which contained ten flies.

eight vials, two control vials, two low dosage vials, two medium dosage vials, and two high dosage vials (Fig. 8). Each of these sets will have one male vial and one female vial. Each vial will contain 10 flies, allowing for 80 data points. Each day, I checked in on my flies and observed whether or not any of them had died. I then waited for life to take its toll on the flies, or in other words, waited for them to die, recording how many flies

were alive each day until there were no flies left. I took these data points for each of the vials, and utilized these values to continue my research and draw conclusions based on further tests.

Results

Throughout the analysis of the lifespan of *D. melanogaster*, I was able to determine the significance of 7-Keto DHEA on their lifespan. Because I had determined that with an increased

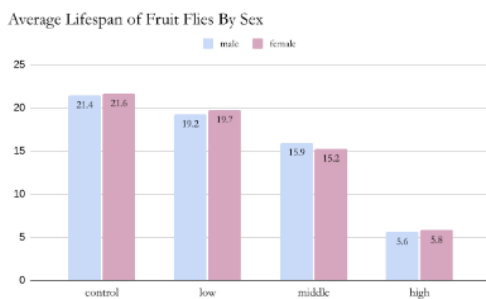


Figure 9: Comparison of metabolism after administering 7-Keto DHEA to the fruit flies as measured by their average lifespan in days

metabolism, there is a decreased lifespan, I was then able to use the correlation between this to measure the effectiveness of 7-Keto DHEA. After completing the overall analysis, I inputted the average data points for each sex, (Fig. 9). Although my initial question did not take the sex into account, it was important for me to put them side by side in order to determine that there were

no outstanding discrepancies which could serve as inconsistencies throughout my overall data analysis. Because there was no overall outstanding significance of either males or females consistently outliving the other, it was therefore appropriate for me to combine them into one average moving forward. After determining that it was appropriate to combine all of my data into one graph of the overall average, I compiled it into yet another bar graph, (Fig. 10). I was able to take this graph into consideration to observe that even just visually in this graph, any administration of 7-Keto DHEA did cause a decrease in lifespan, and therefore it increased their metabolism. As a result, I was able to determine that it did have an impact in their lifespan, and I just needed to determine whether or not this impact would be considered statistically significant.

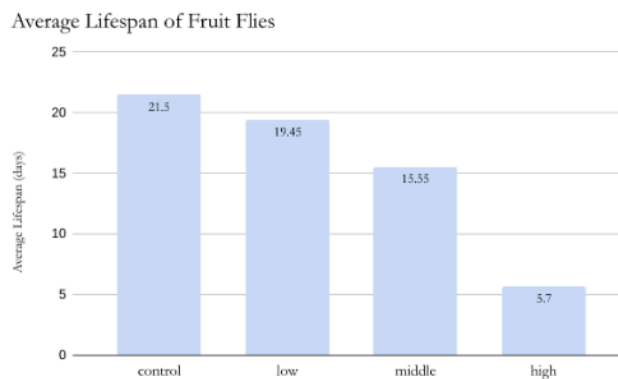


Figure 10: This graph depicts the overall average lifespan in days of the fruit flies at various dosages of 7-Keto DHEA

For my statistical test, I used a Chi square test which is used to understand the

$$\chi^2 = \sum \frac{(O-E)^2}{E}$$

- df = 3
- 95% confidence
- critical value = 7.815

Figure 11: This image depicts how to calculate the Chi Square test, the statistical test that I utilized in order to determine statistical significance in my study.

relationship between two or more categorical variables, (Fig. 11). For this specific study, I had four categorical variables, which were my dosages, the control, low, medium, and high. From this point forward, since I was using the overall average as opposed to the average by sex, there were four variables as opposed to eight. Based on this, I determined my degrees of freedom by subtracting one

from the four categorical variables in order to get my degrees of freedom of three. I wanted to perform this calculation at a 95% confidence interval, giving me a critical value of 7.815. Any value that is less than 7.815 is considered to be statistically insignificant, and any value greater is considered statistically significant. After determining the basics of these calculations, I needed to perform the calculation four times, (**Fig. 12**). For each

calculation, I needed to define what exactly my expected value was, and based on my specific study, this would be the average lifespan for the control vials.

When performing the initial calculation, this means that the value for the control is zero because the observed control value is equal to the expected value. I continued this calculation among all of the groups and for the low group I got a Chi Square value of 0.195, the medium group was 1.647, and the high group was 11.611. Through the Chi Square analysis, it was

determined that the dosage was statistically significant at the high dosage. Although there was a decrease in lifespan noted at all dosages, it was not considered to be statistically significant until the high dosages.

$$\frac{(21.5 - 21.5)^2}{21.5} = 0$$

$$\frac{(19.45 - 21.5)^2}{21.5} = 0.195$$

$$\frac{(15.55 - 21.5)^2}{21.5} = 1.647$$

$$\frac{(5.7 - 21.5)^2}{21.5} = 11.611$$

Figure 12: This figure depicts my Chi Square calculations to determine the statistical significance of 7-Keto DHEA on fruit flies.

Discussion

The data that I obtained did support my initial hypothesis in that the administration of 7-Keto DHEA would lead to a decreased lifespan and likely increased metabolism in *D. melanogaster*. I justified my hypothesis as a result of Dr. Mateusz Mołóń, who discovered the negative correlation between metabolism and longevity (Mołóń, 2020). Overall, this strengthens the claim

that by taking 7-Keto DHEA, there is likely an increase in metabolism. Not only this, it also examined the effectiveness of 7-Keto DHEA at various levels, and overall I found that there was decreased longevity in higher dosages, ultimately finding statistical significance at the high dosage. Because of the connections that I made in my research, it sparks new inquiries among other variations of these supplements as well as in other model organisms, until there is the potential for this to be applied to humans and since these supplements are not monitored by the FDA, this would allow for more accurate claims to be made by these supplement companies. In order to further determine whether or not there is a correlation, there is a need for further testing, in *D. melanogaster* as well as in other model organisms similar to humans, and potentially in humans as well.

Limitations

Although my data did show statistical significance in the high dosage, I was also held back by several limitations. As with any scientific study, there is always the risk for human error, such as in my measurements and recordings of death. I may have made small errors in my measurements which could alter the results, and I measured the deaths according to which days the flies died, not exactly what time they died. If I were to repeat this study, I could account for these in a more highly equipped lab which would be capable of more precise measurements, and ensure that I got the exact times of death of each fly to take a more accurate average. Another limitation that I faced was the fact that I mixed the supplement in, rather than sprinkling it on top, meaning that I

am unable to determine if each fly ate an equal amount, or if they ingested as much as they were intended to. In the future, I could try a mix of the methodologies and compare them among each other in order to determine if there really is a difference. In addition, there is really no way to combat the inconsistency in how much each individual fly ate, which means it would be a limitation regardless of the execution of my study. I also noted limitations in the lifespan of my control group of flies in that they did not live the average of 70 days, which may have arisen as a result of the way that I anesthetized the *D. melanogaster*. This is important to note moving forward, however, in my specific study, I was consistent in the way that I went about anesthetizing my *D. melanogaster*, and as a result, this limitation was likely consistent among all grounds. Overall, regardless of the limitations, my study still provides convincing evidence that there should be further research conducted on the impacts of 7-Keto DHEA on various model organisms and their metabolism as determined through their lifespan.

Implications and Further Steps

Overall, the fitness community plays a large role in society today, and more and more people are taking weight loss supplements or supplements to enhance their workout process. However, because these supplements do not have to be monitored or approved by the FDA, they consequently do not undergo the same precautions that other things endure in order to enter the shelves for people's consumption. As far as my research goes, it continues to add to the conversation regarding the research that needs to be done in order to determine whether or not the current advertisements are accurate among these dietary supplements, especially as they are not FDA monitored or approved. In my research, I determined that there is a plausible

correlation between increased metabolism and taking 7-Keto DHEA based on the decrease in longevity through the increased dosage of 7-Keto DHEA. Through further research, these results could eventually have the potential to be applicable to humans. Overall, advertisements play a large role in society and knowing whether or not the product that is being purchased is actually effective is critical in whether or not the product continues to be advertised in this way. Through further research, this could potentially be connected back to humans in order to determine the overall accuracy of these claims. Because of the growing obesity epidemic and the vast amount of money that is currently being spent on weight loss supplements in the United States alone in one year, there is an overall need to determine whether or not what people are consuming is actually effective or if false claims are being made. Although my research does not directly connect to humans, it is one step in the direction to determine the accuracy of advertisements and how they are being used and the overall consumption based on these advertisements. If it is able to determine the overall accuracy of these claims, although they are not monitored by the FDA, it would be one step in the right direction in order to determine the overall effectiveness of these supplements based on their advertisements. It is also important to address the fact that while yes, it may increase metabolism, as suggested by the decrease in longevity with higher dosages, there is also the possibility that there will be a decrease in lifespan, potentially in humans as well as in fruit flies.

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