INTERNATIONAL BACCALAUREATE BIOLOGY EXTENDED ESSAY

Investigating The Effect Of Different Nutrients (Flour As A Control, Cucumber, Potatoes, Carrots, Lettuce) On *Tenebrio molitor* (Mealworms) Over A Six-Week Period

Research Question

How Does Different Nutrients (Flour As A Control , Cucumber, Potatoes, Carrots, Lettuce) Effect The Growth Of Mealworms (*Tenebrio molitor)* In A Six-Week Period?

Word Count: 4000

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INTRODUCTION

Background Information

Mealworms are the larvae of mealworm beetles. They can be found in many parts of the world. Mealworms, which generally prefer warm, humid and dark areas, show their living activities in places where there is rotten wood, leaves, flour and food. Burrowing and eating are a top priority when it comes to being a mealworm, and they will eat just about anything. They will eat grains, vegetables, any organic material, fresh or decaying.[1]

In addition, these creatures are very important for the ecosystem. Mealworms, which are constantly feeding activities, decompose and clean unwanted materials. It is an indispensable food source for many animals. Spiders, birds, rodents, reptiles, and even humans in the future can feed on mealworms. In the world where there are important problems such as climate change and global warming, mealworms are a good option for the continuity of human life, considering the eating conditions that may change in the future as mealworms are also used for the consumption of domestic animals such as fish and turtles. In addition, mealworms have historically been a part of Asian culture. Consumption of insects, recently marketed as healthy snacks, also dates back centuries in some cultures.[2]

In May 2017, mealworms were approved as food in Switzerland. In June 2021, dried mealworms were allowed as new food in the European Union after the European Food Safety Authority deemed the larvae safe for human consumption.[3]

The main reason for me to choose this experiment is that I have learned from the books I have read before that mealworms are a good solution against possible future food shortages or damage to the ecology in the world, and I want to take advantage of such an opportunity and conduct such an experiment. Especially since my family is very knowledgeable and interested

in nature, I researched such a topic and took care to organize the experiment in a way that would be beneficial to nature and living things.

Life Cycle

Like every holometabolic insect, there are four life stages: Egg, larva, pupa, and adult (can be seen from the *Figure 1*) When female bettles find a dark and safe environment, they lay about 300 white, bean-shaped eggs. The areas where they spawn are usually dishes or flour bowls.

Larvae called mealworms hatch from the eggs. Mealworms, which have a milky white color at first, become yellow and brownish in time. While they are larvae, they consume the food in their environment and change their skin as they feed. Mealworms transform from larva to pupa if the temperature drops and hibernate. Pupae are C-shaped, thick and white-colored structures. Their color gets darker as they progress.[4]

When the temperature rises again, adult insects emerge from the pupa. Insects that initially have white and orange colors turn black over time. Although they are slow-moving insects, they have wings so they can dominate areas. Adults measure between 1.25 and 1.8 centimeters and can live between 3 and 12 months.[5]

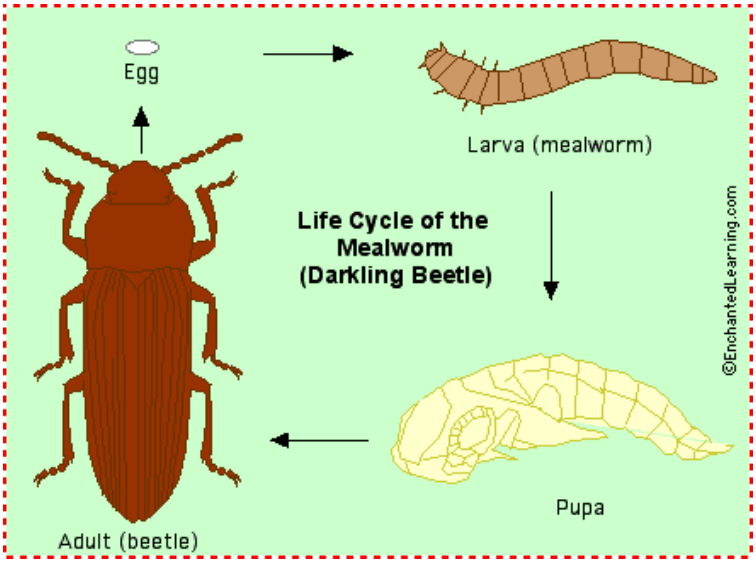

Figure 1: Life Cycle of the Meal Worm

Since I didn't have a great camera that can greatly zoom the lifecycle of a mealworm, I used an image from the website www.enchantedlearning.com that shows the life cycle and physical structure of mealworms well.[6]

Observation and Experiment

Although egg life is not easily noticed by an observer, mealworms that have reached larval size and have a yellow color are in a good condition for observation. The enlarged larvae live and appear prominently in grains, plastic containers, dead animals or invasive plants. In addition, the larvae sometimes live in the pantry of the houses, such as grain and flour, and occupy the food in the house. This situation, which is unfavorable for humans, can also cause large-scale destruction in grain production facilities. Adults are known to secrete a defensive chemical that both irritates and stains human skin, and this can make it difficult to contain infestations.[7]

In this investigation, five groups of mealworms will be placed in plastic containers at the egg stage and given four different nutrients as potato, carrot, cucumber and lettuce and a test group with no addittional nutrient for a period of siz weeks, the differences in their growth will be observed. The most important goal in this experiment is to find out which food will give the mealworms the most efficient growth.

Mealworms can be easily stored and cared for in the cupboard. All that needs to be done is to remove the boxes that are kept at certain intervals from the cabinet and give food. Since it will be easy to grow and reproduce in large numbers, the experiment is also likely to proceed correctly and unhindered.

As a result, considering the nutritional benefits of mealworms in ecology and in the future, it is predicted that the most beneficial nutrient in the breeding phase of mealworms will be determined as a result of this investigation and it will reach a conclusion.

In order to go deeper in the research and better understand the results of the experiment, it is necessary to have a good understanding and knowledge of the content of the four nutrients to be used in the experiment and their benefits for growth.

If the content and differences of these four nutrients are known, the mineral that is most beneficial to mealworms can be determined from the content of the food fed to the test group that showed the most growth as a result of the experiment.

The contents and information of the four nutrients used in the experiment are as follows:

Cucumber

100 grams of cucumber contains 15 calories, 4 grams of carb, 0.6 grams of protein, 0.7 grams of fiber. Eating 100 grams of cucumber also supplies approximately 20% of our recommended Vitamin K need. It is also involved in processes like temperature regulation and the transportation of waste products and nutrients in the body.[8]

Carrot

The nutrition facts for two small-to-medium raw carrots (100 grams) are:

41 Calories, 0.9 grams of protein, 9.6 grams of carbs, 4.7 grams of sugar and 2.8 grams of fiber. Carrots are also great resources to get Vitamin A which has an important role in growth, development and immune function. It also increases the eye health.[9]

Potato

Potatoes are great sources of many minerals. One medium baked potato (estimatedly 150 grams including the skin) provides 161 calories, 0.2 grams of fat, 4 grams of protein, 36.6 grams of carb and 3.8 grams of fiber. Potatoes also meets most of our daily need of potassium, vitamin B6 and vitamin C.[10]

Lettuce

Lettuce has a high nutrient content yet being very low in calories. Three cups of lettuce (nearly 90 grams) contain 11 calories, 1 gram of protein, 0.2 grams of fat and 1 gram of fiber. Lettuce is an excellent vegetable for vitamin K as three cups of it nearly provides 1.5 times more of our daily vitamin K need.[11]

Flour As Control Group

For the control group, no supplementary food was given. Namely, the flour in which mealworms live is sufficient for the mealworms to continue their vital activities. However, in this experiment, since the effect of different nutrients on the growth of the mealworms will be investigated by giving different foods to the mealworms, the control group was taken as a criterion and no food supplements were given to the control group.

Research Question: How does different nutrients (Flour as a control, cucumber, potatoes, carrots, lettuce) effect the growth of mealworms (*Tenebrio molitor*) measured by a six-week period?

Hypothesis

H_0: There is no difference in the development of animals fed different types of food.

H_1: There are differences in the development of animals fed with different types of food.

My hypothesis in this study is that different types of food can improve mealworms. Because, as I mentioned in the article, the types of nutrients to be given include minerals that support development. I also think that there will be differences in the growth of worms fed with different types of food. Because not every food has the same effect on the development of the body.

Variables

Type Of Variable	Name of Variable	Method of Management and/or Measurement
Independent Variable	Type of nutrient given (cucumber, potato, carrot, lettuce)	Different types of nutrients are given to mealworms in order to explore mealworms growth rate. 5 grams per week is given to each group.
Dependent Variable	The growth time of mealworms	The time taken for mealworms to grow is measured by controlled monitoring. No mealworms were harmed.
Controlled Variable	Temperature	The temperature in the place where the experiment was carried out was always kept constant and at a temperature where the mealworms could survive.
	Size and type of mealworms	All 100 mealworms (5x20) were taken from the same laboratory and used as the same species.
	Growth level of mealworms	All mealworms were at the same maturity level (larvae) when the experiment began.
	Size of boxes that mealworms are stored in	The boxes in which mealworms were kept had the same dimensions and volume in all data sets. (1500 mL)
	Whole wheat flour	The same whole wheat flour was used in the same quantity in all data groups. Flour was given for once and the same flour stayed till the end of the study.

Table 1: Variables

Material List

100 Mealworms (5 x 20)

5 Plastic Storage Boxes (5 x 1)

Weight (to measure the nutrients)

Knife (for cutting large food pieces)

Nutrients Apple, Potato, Carrot, Lettuce (5 grams every week)

1500 mL Whole Wheat Flour (300 x 5 mL)

Measuring Cylinder

Method

1. First of all, you need five storage boxes to prepare the experimental setups. Take these boxes and open their containers.
2. Then you will need to pour the flour into the boxes with which you will care for the mealworms. In order to provide equal levels of flour to all groups, first pour the whole wheat flour you have taken into the measuring cylinder and calculate it to be 300 milliliters.
3. After this, empty the flour stuck in the measuring cylinder into the box and do it for other four boxes. Notice that the flour is evenly distributed throughout the box. (You do not need to renew the sand during the experiment. Sand is quite sufficient for an experiment of this duration.)
4. You will need to place the mealworms in the boxes you have prepared. You can obtain mealworms from laboratories or live food vendors. After purchasing the mealworms, place them in each box, 20 mealworms each. You must have used a total of 100 mealworms. Your experimental setup should be like mine in *Figure 2*.

5. After preparing the mealworms, you need to move on to the experiment phase and measure and feed regularly. For the feeding phase, take one of the four foods to be used in the experiment and prepare a portion of 5 grams. Since foods will be larger than 5 grams, measure the food you consume by cutting it with a knife and weighing it on a scale. Do this step once a week for all foods and do not skip feeding. Otherwise, the development process of the mealworms will change and you will get different results in the experiment. For example, if you forget to feed, the development of mealworms will slow down.
6. During this process, check the mealworms daily and note the stage of their growth. If the larvae is transformed to pupae or insect on the day you check, make a note of the day it is. Pay attention especially if it is the first time of formation, because the first larva, pupa or insect formations help you determine the speed of trying different foods. I also checked it daily because this process in the development of mealworms can change daily and there should be no errors in the dates in order to strengthen the experiment.
7. Continue this entire process throughout the experiment, take measurements for six weeks. Taking it for six weeks is the ideal time to measure the progress of mealworms. When the experiment period is over, note one by one how much of each period is in each group. As a result of the experiment, it can be concluded that the food used in the most developing group is the most efficient food for mealworms.

Figure 2: My Experimental Setup

Risk Assesment

Safety

For safety, I took care to use gloves during the controls in the experiment. I have never touched any mealworms with my bare hands. Also, for my own health, I made sure to clean my hands before and after handling the mealworms.

For the safety of the mealworms, I kept all test groups in the same room under constant temperature, pressure and humidity. During the experiment, I did not physically harm any mealworms and I routinely checked them for possible problems.

Additionally, for the safety of the place where you conduct the experiment and the mealworms, you do not have to close the lids of the boxes you put the mealworms in. Make sure that the mealworms get enough air, do not worry about the mealworms escaping when you leave the lids of the vertical boxes open, because mealworms cannot physically climb vertical places.

Environment

During the experiment, I separated the residues of the four foods I used as organic waste and threw them away. I applied the same waste system to the flour remaining in the bowl after the experiment was over. The purpose of this practice is to avoid mixing organic waste with other waste.

Additionally, I did not take the mealworms out of their container until the experiment was over, in order to prevent anything that could potentially harm the contents of the experiment and the environment. In order to keep the mealworms' closed environment stable, I made sure that their environment was at room temperature and constant pressure. Also, when the experiment was over, I did not kill the mealworms or release them into the wild, I took them back to the seller I bought them from.

Ethics

Since the experiment will be carried out on living insects, it may be thoughtful to harm the living thing. However, in this experiment, there is nothing that will harm the mealworms. Mealworms will be cared for under healthy temperature and pressure, not starving and given adequate space, simply by changing their food. The food will also be the food the mealworms are already consuming. The purpose of giving different foods is to compare the proportions of minerals, vitamins and substances contained in the food and the growth rates of mealworms. Thus, the healthiest growth condition of mealworms will be determined and it will be aimed to get the highest yield.

All mealworms used in the experiment were taken from the same laboratory and returned to the laboratory as they were when the experiment was completed without any death. In this regard, every ethical consideration has been taken in conducting an experiment.

It can also be thought that the mealworm group, which was not given any supplementary food among the test groups, was harmed. Not giving mealworms supplementary food may slow their development, but it will not kill them. Because mealworms can survive by consuming the nutrients in the flour they are in. In short, no mealworms were killed in this experiment. It was aimed to measure the development time in their natural growing environments by giving supplementary nutrition to only four of the five groups.

DATA

Time/ Nutrient Given	Control Group	Cucumber	Lettuce	Potato	Carrot
Week 6 (End of the experiment)	20 Larvae 0 Pupa 0 Adult	11 Larvae 4Pupa 5 Adult	10 Larvae 6 Pupa 4 Adult	16 Larvae 3 Pupa 1 Adult	5 Larvae 8 Pupa 7 Adult

Table 2: The Number Of Larvae, Pupa And Adult (Insect) At The End Of 6-Week Period

Table above shows the development of every group at the end of the 6-week period. The table shows how many of the 20 mealworms turned into pupae or insects, or stayed in the larvae stage from each group.

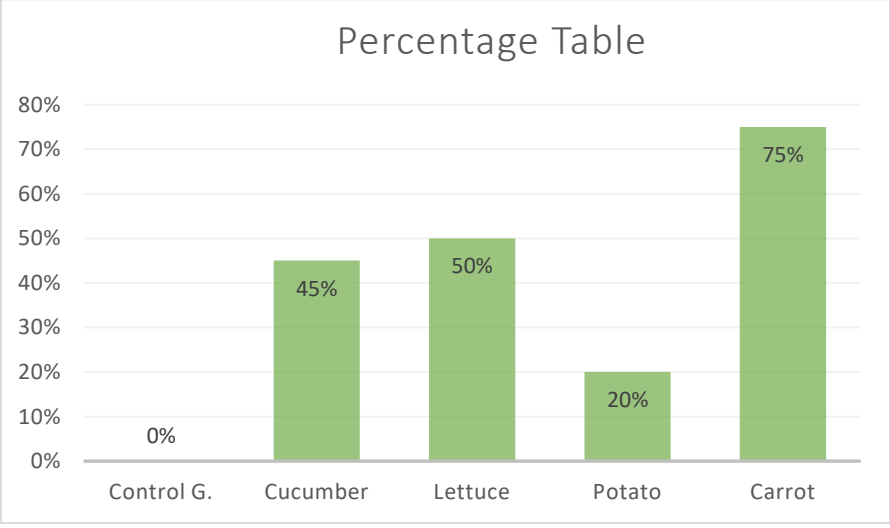

Table 3: Percentage Graph Showing What Percentage Of Mealworms Transition To The Post-Larval Stage In Each Group

The graph above shows percentage of how many of 20 mealworms from each group turned into pupae or insects at the end of the experiment.

As seen in the graph, it is understood that the most pupa and adult formation is in carrots, and the least pupa and adult formation is in potatoes. In the control group, which was not given extra food, no trace of the mealworms transitioning to the post-larval stage was found during the experiment.

First Pupa Formation	Carrot
First Insect Formation	Carrot
Most Pupa and Insect Formation	Carrot
Least Pupa and Insect Formation	Control Group

Table 4: Detailed Information On Fastest, Slowest Formations Of Pupa And İnsects From The Experiment Groups.

Table above gives a detailed information on fastest, slowest formations of pupa and insects from the experiment groups.

Chi-Square Test

The chi-square test is a hypothesis that is used when you want to determine if there is a relationship between two categorical variables. Now, for this investigation, we will be making a chi-square test in order to find if there is relationship with the nutrient given to the mealworms and their growth speed. The reason why we are doing chi-square test and not any other statistical application is the fact that we have two categories with the possibility of relationship just like in any other chi-square test. So, my hypothesis on this test is there is a relationship between different types of nutrients given to the mealworms and the growth speed of mealworms.

For the statistical calculation, www.socscistatistics.com website is used.[12]

Results						
	Cucumber	Lettuce	Potato	Carrot		***Row Totals***
Larvae	11 (10.50) [0.02]	10 (10.50) [0.02]	16 (10.50) [2.88]	5 (10.50) [2.88]		42
Pupa	4 (5.25) [0.30]	6 (5.25) [0.11]	3 (5.25) [0.96]	8 (5.25) [1.44]		21
Adult	5 (4.25) [0.13]	4 (4.25) [0.01]	1 (4.25) [2.49]	7 (4.25) [1.78]		17
Column Totals	20	20	20	20		**80 (Grand Total)**

The chi-square statistic is 13.0308. The *p*-value is .042549. The result is significant at $p < .05$.

Table 5: Chi-Square Test

Here is my Chi-square test screenshot made from the website www.socscistatistics.com. Also, the control group is not involved in this test because as there is no pupa or adult formation as seen from the table 2. So, from this calculation, it can be seen that there is a relationship between different types of nutrients given to the mealworms and the growth speed of mealworms. So, as I thought in my hypothesis, different types of nutrients have different effects on pupal and adult development. The result is significant at when p-value is lower than 0.05. This situation proves the investigation is significant and there is a relation.

Also, this result shows that there is a significant difference between the data groups. For example, feding mealworms with carrot and potato actually differ their growths. This shows that carrot is actually a different and a more valuable nutrient for mealworms.

CONCLUSION

As a result of the data obtained in the study and the chi-square test, the most productive and fastest growing mealworm group was the group fed with carrots. The group with the slowest growth was the group that was fed with potato. In the control group, the pupa and adult did not develop for 6 weeks, and it was determined that they continued to live as larvae during this period and did not die at this stage. Also, these results showed us that there is a significant difference between the nutrients given to the mealworms, the statistics showed the most efficient one is carrot.

As I predicted in my individual hypothesis, giving additional nutrients to the mealworms affected their growth. Compared to the control group every type of additional nutrient sped up the process of mealworms growth. Additionaly my chi-square test showed that there is a relationship between the nutrient given to the mealworms and their development, with supporting my hypothesis too.

The most efficient nutrient was carrot, which contains vitamin A, which is a great support for growth. As a result, it can be concluded that foods containing vitamin A, such as carrots, are the most effective foods in the development of mealworms. As can be seen in the data obtained in the experiment, 75% of the mealworms fed with carrots progressed to the post-worm period, and with 40%, there was a significant difference even with cucumber, which is the closest group to carrots.

Discussion And Evaluation

Discussion

As mentioned in the Introduction section, mealworms have many uses around the world. In addition to being consumed as food, they are used in many biological fields as decomposers and even as converters of polystyrene into organic matter. The result of this experiment has a very important place in increasing the productivity of mealworms. The use of foods containing vitamin A, which supports development, such as carrots and cucumber, in the nutrition of mealworms not only made the lives of mealworms healthier, but also indirectly showed that the productivity in the areas of use would increase.

In short, this experiment can help people and institutions doing research in the field of ecology to increase the productivity and development of mealworms. By making the development of mealworms, which have a fast life cycle, healthier and faster, the process progresses more efficiently in short-term experiments and production.

If we go deeper into the research, as predicted, carrot food, which contains vitamin A, transitioned 75% of the mealworms to the post-worm stage. Although not 100%, the effect of carrots increased noticeably throughout the experiment. It was observed that none of the mealworms in the control group could transition to the post-worm period during the experiment. After the control group, the group that showed the least improvement was potatoes. If we attribute this result to a reason, water consumption is the least in these two groups. Because wholemeal flour generally has a dry structure, the mealworms fed only with wholemeal flour in the control group were able to survive, but could not transition to the larval or pupal stage due to mineral deficiency.

If we compare this experiment with other research on the internet, it is seen in other articles that mealworms are fed with juicy and vitamin-rich foods such as carrots and apples. The use of

carrots in this experiment and their selection as the most efficient food is supported by other articles, and it is globally understood that carrots are a good food.

Talking about how this research can be carried forward, as mentioned throughout the essay, mealworms can be used in many areas, especially in the addition of organic matter. In cases such as necessary soil cleaning, releasing mealworms into the soil in the healthiest and most developed form can make the separation and cleaning of such areas faster and more efficient. In addition, the fact that mealworms are used as feed for many living creatures such as fish today and many studies predict that they will be consumed by humans in the future is supported by this research on how mealworms can be used as an efficient food.

Evaluation

As for the strengths of the experiment;

Conducting the experiment in humidity and at sufficient temperature is one of the most important and robust features of this experiment. This made the results of the experiment stronger because mealworms live a stress-free and healthy life in warm and humid environments and continue their development better.

The high number of mealworms used in the experiment minimized the number of possible errors in the experiment. By using approximately 100 mealworms, the research area in the experiment was increased and the margin of error was planned to be minimal.

It is very important to provide enough food for all mealworms in the experiment to get maximum efficiency from the experiment. Frankly, since the experiment is investigating how the mealworms will grow fastest depending on the nutrients, it is necessary to make sure that none of the mealworms are left without food.

Measurements in mealworms were in 6 weeks period, but the developments were checked daily and the obtained data were written down at the end of the week. Daily control was important throughout the experiment to control the instantaneous development of mealworms and to quickly intervene in case of a possible problem. Although this increased the effort spent on the experiment, it made it easier to reach definitive results in the experiment.

Now, here are the limitations in my experiment;

The fact that the mealworms used in the experiment were very small due to their biological structure made it difficult for a human to control the experiment. Because it was quite difficult to find and control the mealworms living in the whole wheat flour on a daily basis, but it was the most logical option I had.

In feeding the mealworms, I gave enough food for all the mealworms, but there is no guarantee that all the mealworms can access the food. The reason for this restriction was that it was pointless and impossible for me to follow every second of the experiment with the mealworms in wholemeal flour.

Further Investigation

For further investigation, it is worth mentioning that since the mealworms used in this experiment were taken from the same laboratory, their genetic diversity is low and their development may be affected similarly. In order to make this study more comprehensive, it can be said that this experiment can be performed on mealworms taken from different laboratories and even different amounts and types of nutrients can be given.

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