

**INTERNATIONAL BACCALAURETE BIOLOGY
EXTENDED ESSAY**

The effect of number of riped *Citrus sinensis* (Orange) (as a source of ethylen) on the deterioration of *Malus domestica* (Apple)

How does the number of riped *Citrus sinensis* (Orange) (as a source of ethylen) effect on the deterioration of *Malus domestica* (Apple) ?

Word count:3.717

TABLE OF CONTENTS

Introduction.....	2
Research Question.....	4
Hypothesis.....	4
Method Development and Planning.....	5
Variables.....	6
Material list.....	7
Procedure.....	7
Safety.....	9
Results and Analysis.....	10
Discussion.....	12
Statistical Test.....	13
Evaluation.....	15
Conclusion.....	17
Referances.....	19
Appendix.....	21

Introduction

Ethylene is a plant hormone secreted by fruits and vegetables. It is produced in the plant by deteriorating tissues and stem nodes. While doing a research on the internet about ethylene and oranges I found that Dartmouth plant biologist Eric Schaller once said *“Ethylene is an important natural plant hormone used in agriculture to force the ripening of fruit, but it can also be a problem for growers who are storing fruits or vegetables. Ethylene is used not because it induces ripening, because it doesn't affect ripening of citrus fruit, but it does affect the skin color of the citrus fruit. It produces a more uniform 'orange-colored' skin for the fruit,” he says, using cosmetics to improve agricultural marketing.*¹ Ethylene, which is a hydrocarbon, is a molecule that exists in the form of gas. Why it is important? Because seed germination, plant root growth, flowering and deterioration also have a lot of relationships and effects on the growth and development of the plant².

Nutrients, fiber, and antioxidants exist in fresh fruits and vegetables. They are living organisms that will go to waste if not consumed within a reasonable amount of time, and their nutrient benefits will deteriorate. Microorganisms such as bacteria and mold, cause most fruits and vegetables to deteriorate. Microorganisms enhance the deterioration process. Bacteria and molds, expediting the degradation process. As ripening occurs, some fruits will emit ethylene. Ethylene is responsible for texture changes, softening, color changes, and other

¹Blumberg, J., 2013. A Biologist Considers Apples and Oranges, Rice and Rubber | Dartmouth. [online] Home.dartmouth.edu. Available at: <<https://home.dartmouth.edu/news/2013/04/biologist-considers-apples-and-oranges-rice-and-rubber>> [Accessed 29 September 2013].

² (2015). Byju's: <https://byjus.com/> received from Bakırcı, M. Ç., Erdem, B., & Budak, G. (2010). Evrim Ağacı: <https://evrimagaci.org/>

ripening processes³. Since ethylene is in a gaseous state, it affects other fruits around the ethylene produced in the fruit. The image I saw on the fruit bowl on the kitchen counter helped me do my thesis on the deterioration of fruits. An apple and an orange were standing side by side in the bowl. I've seen rot in oranges and apples. I wonder if the orange had any effect on the deterioration of the apple? If I give another example, there were a few products in the fruit and vegetable section of the fridge at home along with oranges. I noticed that these products deteriorate in a short time, but the fruits and vegetables that are on the top shelf, without having any contact with an orange, do not deteriorate. In my research, I found that it really causes the fruits and vegetables next to it to deteriorate in a short time thanks to the ethylene secreted by the orange into the external environment⁴. After these observations, I decided my research question which is **“How does the number of riped *Citrus sinensis* (Orange) (as a source of ethylene) effect on the deterioration of *Malus domestica* (Apple) ?”**

Every fruit and vegetable has a certain period of deterioration. Except for the factors that accelerate and slow down this deterioration period. From the point of view of marketing, it is desirable that the shelf life of semi-processed vegetables or fruits in terms of microbiological, and nutritional should be at least 4-7 days, but preferably up to 21 days⁵. Seeing this state of the fruits at home, I wanted to discuss the topic of the experiment through oranges and apples. I can easily access the fruits and vegetables I want in the country I live in. The apple I used in the experiment is referred to as the “Amasya Apple” (*Malus domestica*). The orange is

³ Healthy Options. 2022. *Why Do Fruits & Vegetables Go Bad? - Healthy options, Philippines : News Digest*. [online] Available at: <<https://www.healthyoptions.com.ph/newsdigest/boost-your-immunity/why-do-fruits-vegetables-go-bad>> [Accessed 14 February 2022].

⁴ Sezer, E., & Ayhan, Z. (2017). Meyve ve Sebzelerde Etilen Tutucu İçeren Aktif Ambalajlama Sistemlerinin Uygulanması ve Raf Ömrüne Etkisi. *Akademik Gıda*, 15(2), 182-191.

⁵ OLUK, C. A. (2018). Yarı İşlenmiş Meyve ve Sebzelerin Muhafaza Yöntemleri. *Uluslararası Doğu Akdeniz Tarımsal Araştırma Enstitüsü Dergisi*.

referred to as the “Washington Orange” (*Citrus sinensis*). The Washington orange is a huge, pot-bellied, thick-fruited orange with a peel that peels readily. The orange slices are also prominent and easily distinguished from one another. It's also great for orange juice because it doesn't leave a bitter aftertaste and squeezes out easily⁶. For the Amasya apple, the fruit production of a tree that yields fruit one year declines the following year, which is a characteristic of the marble apple. It usually has a greenish tint, with one side being red and the other yellow. It has a thin shell, a lovely aroma, and a pleasing flavor. It's tough and long-lasting⁷.

Research Question

How does the number of riped *Citrus sinensis* (Orange) (as a source of ethylen) effect on the deterioration of *Malus domestica* (Apple) ?

Hypothesis

If the number of oranges increases (as a source of ethylen) the deterioration zone diameter in apple increases.

Alternative hypothesis: As the number of oranges increases, the deterioration zone of apple increases.

Null hypothesis: There is no relationship between the number of oranges and the deterioration zone in apples.

⁶ Bağışıklık.com. 2022. Portakal Hakkında Her Şey | Bağışıklık. [online] Available at: <<https://bagisiklik.com/wellness-onerileri/portakal-hakkinda-her-sey/>> [Accessed 19 January 2021].

⁷ Tr.wikipedia.org. 2022. Amasya elması - Vikipedi. [online] Available at: <https://tr.wikipedia.org/wiki/Amasya_elmas%C4%B1> [Accessed 14 November 2021].

Method Development and Planning

The purpose of the experiment is to examine the diameter of the deterioration zone on the apple. I first made my experiment using orange peel. Because I thought that it would be more advantageous both in terms of costs and in terms of materials to use only the peel instead of using more fruit than I would have tried five times. Before starting the experiment, I washed the fruits separately in vinegar water . The protective property of vinegar is derived from the acetic acid component . The main purpose of adding vinegar is to remove the possible harmful biological factors that may be present on the food⁸. Vinegar is a very important element for fruits and vegetables ⁹.Vinegar neutralizes microorganisms¹⁰.I cut the orange peels so that they are 14 cm, 28 cm, 42 cm and 56 cm, respectively. I took five plastic containers and washed them all so that the germs or dirt left in them would be cleaned. There was only be one apple in the first container. In the second container, 14 cm of orange peel and an apple. In the third container, 28 cm of orange peel and an apple. In the fourth container, 42 cm of orange peel and an apple. On the last container, there were a 56 cm orange peel and an apple. I was going to look at how the length of the orange peel affects the size of the deterioration of the apple in exactly 30 days. But when I got to the end of the thirty days, I didn't get the results I wanted. That's why I chose to use a whole orange instead of an orange

⁸ Harvard T:H: Chan. (1946). *Harvard T:H: Chan School Of Public Health*.
<https://www.hsph.harvard.edu/> received from

⁹ *Sirkenin Besin Degerleri-ve Faydalari*. (2021, 07 25). <https://www.cnnturk.com/>:
<https://www.cnnturk.com/saglik/sirkenin-besin-degerleri-ve-faydalari> received from

¹⁰ *Elma Sirkesinin Faydalari*. (2020, 06 22). Medical Park:
<https://www.medicalpark.com.tr/elma-sirkesinin-faydalari/hg-2301> received from

peel. The reason I look at the experiment in a 30-day process is that fruits and vegetables generally have a deteriorate time of 3 or 4 weeks¹¹.

Variables

Independent variables	The number of orange (0, 1,2,3,4,5)
Dependent variable	Diameter of deterioration zone after 30 days
Controlled variables	100 gr of one apple (±1.00 gr) The width of one apple is 18 cm (±0.05 cm) 150 gr of one oranges (±1.00 gr) The width of one orange is 25 cm (±0.05 cm) Softness and hardness of the fruits Room temperature 20°C (±0.06 °C) Containers (20 cmx25cm)
Uncontrolled variables	Chemical substance found on the fruit Microorganizms on the fruits Contamination Surface area of the fruits Genetics of the fruits.

¹¹ Singh, Z. ve Khan, A. (2010). Stewart Postharvest Review

Material list

- 75 oranges
- 30 apples
- 0.5 x 5 liter of vinegar
- 1x 5liter of water for apples
- 1x 5 liter of water for oranges
- 20x25 cm of 6 plastic containers
- 1 precision kitchen scales
- Ruler

Procedure

A) Preperation of apples and oranges for the experiment

- 1) Wash apples and oranges in 1 liter of water but in seperaely
- 2) And wash oranges and apples separately in 1 liter of water with 0.5 liter of vinegar.
- 3) After washing the fruits , wipe the fruits with a dry napkin

B) Prepation of containers for the experiment

- 1) Clean the containers with water to get rid of the germs inside of the container
- 2) Wipe all of the container with a dry napkin.

C)Placing apples and oranges in containers

- 1) Take one of the plastic containers and put only one apple.
- 2) Second container must have 1 orange and one apple
- 3) Third container must have 2 orange and one apple
- 4) Fourth container must have 3 orange and one apple
- 5) Fifth container must have 4 orange and one apple
- 6) Sixth container must have 5 oranges and one apple.
- 7) After closing all of the containers , put them in front of a window where there will be enough natural light for the experiment.
- 8) Analyze the experiement without opening the containers.

D) Measuring the diameter of deterioration zone on surface of apple after 30 days

- 1) Measure the deterioration zone by using ruler
- 2) Take note of all the results.

Repeat all of the previous steps (A B, C, D) five times

Safety

I didn't do anything against the ethical rules during the experiment process. Having vinegar in the materials I use has not caused any harm to my experiment or my environment. But because I was looking at the deterioration zone, it started to smell in my room towards the end of the thirty day. When fruits expires and begins to smell, it is usually due to the growth of deterioration germs such bacteria and mold. Smells can come from two places: chemicals released by the microorganisms as they destroy the fruits, or chemicals created directly by the microorganisms. Those toxic scents could be the result of bacterial invasion. Scented chemicals linked to fruit deterioration have been found to have a critical function in microbial interactions¹².

¹² E. Wolfe, B., 2022. *Why does rotting food smell bad?.* [online] Tufts Now. Available at: <<https://now.tufts.edu/articles/why-does-rotting-food-smell-bad>> [Accessed 15 December 2014].

Results and Analysis

Raw Data Table

Number of oranges	Trials	The diameter of the deterioration in the apple after 30 days (± 0.01 cm)	The size of orange (± 0.01 cm)	The size of apple (± 0.01 cm)	Room temperature (C°)
None	1	0, 00	25	18	20
	2	0, 00	25	18	20
	3	0, 00	25	18	20
	4	0, 00	25	18	20
	5	0, 00	25	18	20
1	1	0, 00	25	18	20
	2	0, 00	25	18	20
	3	0, 00	25	18	20
	4	0, 00	25	18	20
	5	1, 05	25	18	20
2	1	0, 00	25	18	20
	2	0, 00	25	18	20
	3	0, 00	25	18	20
	4	1, 40	25	18	20
	5	1, 65	25	18	20
3	1	0, 00	25	18	20
	2	0, 00	25	18	20
	3	1, 88	25	18	20
	4	2, 25	25	18	20
	5	2, 35	25	18	20
4	1	0, 00	25	18	20
	2	2, 35	25	18	20
	3	2, 50	25	18	20
	4	2, 65	25	18	20
	5	3, 00	25	18	20
5	1	3, 44	25	18	20
	2	3, 68	25	18	20
	3	3, 79	25	18	20
	4	3, 95	25	18	20
	5	4, 05	25	18	20

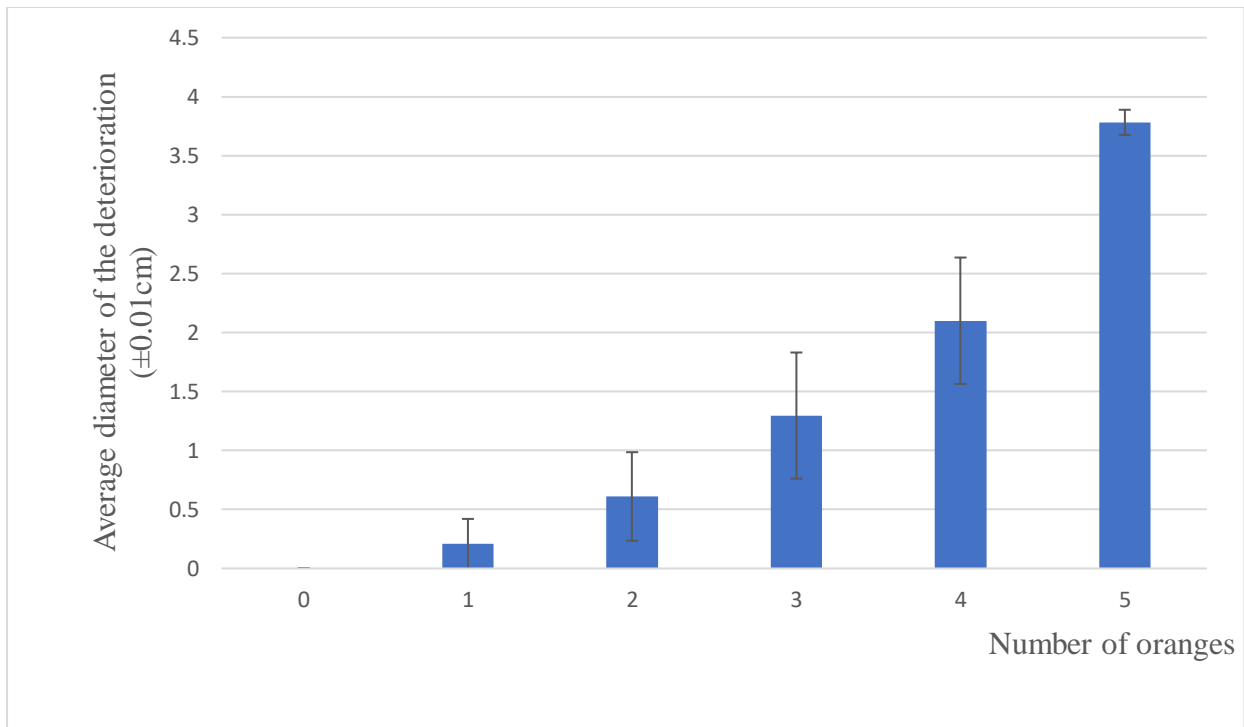
Table 1: Deterioration zone diameters of apples in containers containing different numbers of orange (0,1,2,3,4,5) after 30 days.

Processed Data Table

Number of oranges	Average diameter of the deterioration (± 0.01 cm)	Standart deviation	Standart error
None	0	0	0
1	0.21	0.47	0.21
2	0.61	0.84	0.38
3	1.29	1.20	0.53
4	2.10	1.20	0.54
5	3.78	0.24	0.11

Table 2: Standart deviation, standart error , the average deterioration zone diameters of apples in containers containing different numbers of oranges (0 , 1,2,3, 4, 5) after 30 days.

Graph



Graph:1 Average diameter of the deterioration zone of apple in containers containing different numbers of oranges (0,1,2,3,4,5) after 30 days. Error bars represent standard deviation.

Discussion

When I looked at the process data table which is table 2 , I saw that there is not much decency between the values. In the container where there is no orange, there was no rotting or softening of the apple. In the container where there is one orange, we see that the deterioration begins with the softening of the apple wich has 0.42 cm deterioration zone. As the number of oranges increases, the size of the deterioration zone numbers goes like 0 , 0.42, 0.61, 1.29, 2.10, 3.78 . In the container with five oranges, the apple has the largest deterioration size. If we draw a judgment from these results, the orange did not do much effect on the apple, on deterioration. On the other hand, there is an increase in the graph, although it is not very regular. As the number of oranges increases day by day, the deterioration zone on the apple increases .

Statistical Test

The analysis of variance (ANOVA) is a statistical method that divides a data set's observed aggregate variability into two parts: systematic variables and variables. Random factors have no statistical impact on the supplied data set, whereas systematic variables do. In a regression research, analysts utilize the ANOVA test to examine the impact of independent factors on the dependent variable¹³.

H_1 : As the number of oranges increases, the deterioration zone of apple increases.

H_0 : There is no relationship between the number of oranges and the deterioration zone in apples.

SUMMARY

	<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>	<i>stdev</i>	<i>std error</i>
0		5	0	0	0	0	0
1		5	1,05	0,21	0,2205	0,469574	0,21
2		5	3,05	0,61	0,7055	0,83994	0,375633
3		5	6,48	1,296	1,43033	1,195964	0,534851
4		5	10,5	2,1	1,43625	1,198436	0,535957
5		5	18,91	3,782	0,05687	0,238474	0,106649

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	50,74003	5	10,14801	15,81733	6,4E-07	2,620654
Within Groups	15,3978	24	0,641575			
Total	66,13783	29				

¹³ KENTON, W., 2022. *How Analysis of Variance (ANOVA) Works*. [online] Investopedia. Available at: <<https://www.investopedia.com/terms/a/anova.asp>> [Accessed 6 October 2021].

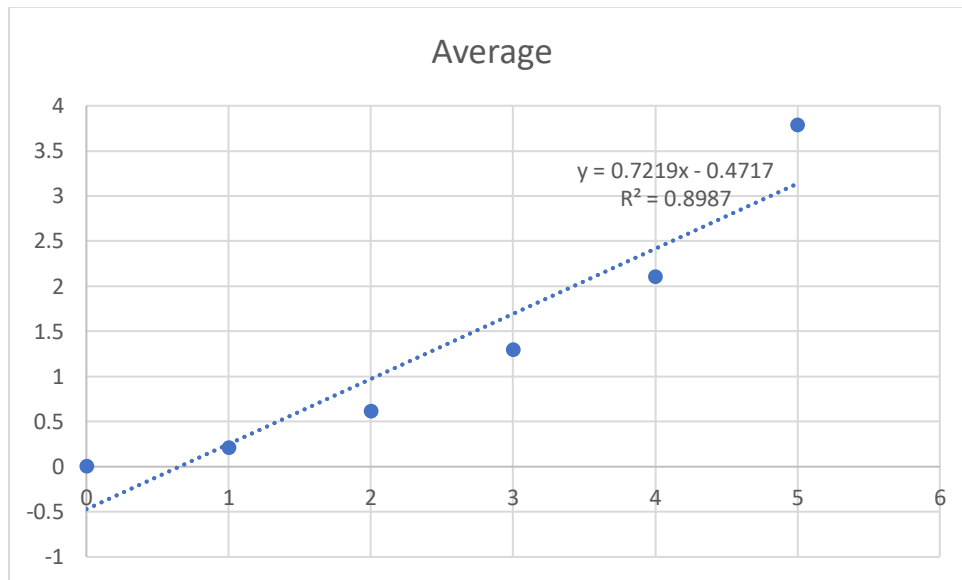
By doing this statistical test, I looked at the connection of experiment with the hypothesis and whether the significant relationship was significant or not. And the value for this is $P < 6.4 \times 10^{-7}$ and because it is closer to plus one, I can say that my experiment has a positive progress. With this value which can be considered as $P < 0.05$, I rejected my H_0 hypothesis which is: "There is no relationship between the number of oranges and the deterioration zone in apples.". As a result of the test, the alternative hypothesis is more significant for this experiment. I also looked for the Pearson correlation coefficient which is $-1 < 0,947975502 < +1$. The test statistic Pearson's correlation coefficient assesses the statistical link, or connection, between two variables¹⁴. Because it is based on the expected and actual results, it is known as the best method for analyzing the relationship between variables of interest. The result of my ANOVA test is $P = 6,4E, \times 10^{-07}$. As I mention in my hypothesis that increase in the number of oranges, the deterioration zone of apple increases. As a result of the fact that the value is close to plus one, the correlation also increases the more. According to my Pearson correlation result, there is a strong relationship between the increase in the number of oranges and the deterioration zone of the apple.

I also looked at regression for this test. Regression analysis is a mathematical method of determining which of those factors has an effect¹⁵. But the slope in the regression goes up to a certain number of oranges (*Graph 2.*). Finally, it continues at a constant rate because the number of oranges increases and the number of apples will remain constant. The apple must also deteriorate for a certain period of time, and as soon as this period wears out, the apple will shrink. The oranges are in contact with apple so I could not comfortably examine the apple for the deterioration zone. I can only experiment up to a certain number of oranges. As a

¹⁴ (Pearson's Correlation Coefficient)

¹⁵ Gallo, A., 2022. *A Refresher on Regression Analysis*. [online] Harvard Business Review. Available at: <<https://hbr.org/2015/11/a-refresher-on-regression-analysis>> [Accessed 4 November 2015].

result, the experiment turned out to be consistent with my hypothesis . There is a direct proportion between the deterioration diameter of the apple and the increase in the number of oranges. But if I had increased the number of oranges even more, I wouldn't have been able to measure the deterioration diameter of apples.



Graph 2: The regression analyses results for the average diameter of the deterioration zone in apple.

Evaluation

In the experiment, first the orange rotted, and then the apple began to soften and rot. The experiment was supposed to be in a closed environment. When the fruits are left in a closed container, the temperature becomes trapped in the container and the humid environment is exposed. Humidity is one of the factors affecting the deterioration of plants in the environment. Since apples and oranges belong to the class of closed seeds, their rotting is contagious¹⁶. Apples began to deteriorate in the last weeks of the thirty-day. An apple usually begins to deteriorate after 3 weeks, if it is in an environment with room temperature. A fatty

¹⁶ NKFUCOM. 2018. *Açık Tohumlu ve Kapalı Tohumlu Bitkiler Arasındaki Farklar*. [online] Available at: <<https://www.nkfu.com/acik-tohumlu-ve-kapali-tohumlu-bitkiler-arasindaki-farklar/>> [Accessed 3 November 2018].

material with limited water and air permeability covers the surface of the cells in the outermost layer of the fruits. The cells secrete substance to keep them from coming into contact with the outside air. The 'cuticle' is the outermost layer. Cutin and epicuticular wax are two components of the cuticle layer. In apples, the thickness of this above-shell structure is about 3 microns. However, depending on the variety, the environment in which it grows, and the climatic conditions, the structure and thickness of this layer can vary¹⁷. This natural wax protects the apples from moisture loss, improves their firmness, and slows down their natural decomposition. The natural wax on an apple's fruit is made up of roughly fifty specific elements that belong to at least a full different chemical groups¹⁸. Ursolic acid, the primary cyclic substance of apple fruit wax, is highly water-repellent. Ursolic acid has been proven to prevent cancer cells and can be used as a raw material for the development of more effective bioactive components such as anticancer medicines. Vinegar is the one that can assist you get rid of this wax. Vinegar will dissolve the waxy surface and, if left in touch for long enough, will completely remove it. This waxy layer protects the fruit from evaporation and acts as a natural wall, preventing some pathogens from entering. The amount of waxy coating varies according on the species, harvest ripeness, and storage and disposal¹⁹. Perhaps it affects the shelf life, since the top of the apples is covered with wax. After being harvested, apples are washed to minimize pesticide remains before being purchased in the shop. The wax is also cleansed during this procedure. Since this waxy coat prevents humidity from evaporating from the apple, its removal reduces the fruit's storage duration. As a result, producers coat the

¹⁷ Ekolojikpazarlar.org. 2022. Her parlak elma parafini mi işaret ediyor? | 100% Ekolojik Pazarlar. [online] Available at: <<http://ekolojikpazarlar.org/?p=2681>> [Accessed 16 February 2022].

¹⁸ Schwarcz, J., 2022. *Why do they spray wax on apples?*. [online] McGill. Available at: <<https://www.mcgill.ca/oss/article/you-asked/why-do-they-spray-wax-apples->>> [Accessed 20 March 2017].

¹⁹ KEMBLE, J., 2022. Is There Wax on Apples? | BestFoodFacts.org. [online] Best Food Facts. Available at: <<https://www.bestfoodfacts.org/wax-on-apples/>> [Accessed 19 August 2013].

fruit with a small layer of wax to prevent water evaporation while also improving the appearance of the apple ²⁰. I also let the fruits stand for 5 minutes in vinegar water separately before using them. Perhaps because vinegar does not completely dissolve the preservative on the fruits, the oranges began to rot early. In fact, in order to get better results from the experiment, I could have kept the fruits in vinegar water longer so that I could be sure that they were free of germs on them. The result of the experiment was to look at the size of the deterioration, but because there was softening in the apple, I should have continue the experiment directly through softening. I was able to collect very less data, precisely because the deterioration was not in the apple. But I don't have a technological device in my house that can measure the softness of fruits.

Conclusion

Obviously, the experiment didn't work out the way I wanted. While waiting for more deterioration in the apple, it turned out in the orange. Based on these data and experiment, I came to the conclusion that the amount of ethylene secreted by the apple is greater than the amount secreted by the orange. If I think about another conclusion, we should not put apples next to other fruits. Because of the closed environment, the hormones they secrete, microorganisms that vinegar cannot destroy one hundred percent spread around. It probably splashes on the tops of the fruits and promote the onset of deterioration. There are too many factors that trigger deterioration. I have done only one of them, that is, my experiment and research on the hormone ethylene. In the process, I chose a regular experiment by trying a lot and different ways. As a result of my research, I have new knowledge about fruits and their

²⁰*Why Do Fruits & Vegetables Go Bad?* (1995, October 27). Retrieved from Healthy Options.

deterioration. I have learned about which fruits and vegetables I should store in the refrigerator, that room temperature is an important factor, and the chemical substances on the fruits.

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Appendix



Figure 2. The material that I had use in my experiment. My reason for using the scaler was just to see how many grams of apples and oranges ,I had bought from the supermarket.