# Investigating The Relationship Between Ethylene Exposure And Ripeness Of Bananas

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## Abstract

This study investigated the relationship between ethylene exposure and the ripening of bananas. We predicted that the proximity of an unripe (experimental) banana to a ripe (ethylene source) banana will increase the rate of ripening because a higher concentration of ethylene surrounds the ripe banana, thereby inducing accelerated ripening of the experimental banana. To test this, we placed unripe bananas 0 cm, 15 cm, and 30 cm away from a ripe banana. The appearance of dark spots on the surface of the experimental banana was quantified after one week using Adobe Photoshop. Analysis performed on our data using a one-way ANOVA test indicated no statistical significance in the difference of our treatments ($p = 0.76895$, $p > 0.05$ on a 95% confidence interval). These findings reveal that banana ripening is not affected by ethylene exposure at different distances, failing to reject the null hypothesis. Thus, the ripening rate of unripe bananas is not increased at closer proximity to ripe bananas.

## Introduction

*Ethylene* is a gaseous plant hormone that induces fruit ripening (Burg & Burg 1965). Specifically, ethylene increases cellular respiration and signals enzymes that convert starch into sugars, degrade cell walls and break down chlorophyll (Maduwanthi & Marapana, 2019; Borkar et al., 2008; Nishiyama et al., 2007; Burg & Burg, 1965). As a result, when fruits mature, the increase in ethylene causes them to sweeten, soften, produce unique aromas, and turn from green to yellow or brown (University of Maryland Extension, 2021; Maduwanthi & Marapana, 2019). While ethylene has been used to induce ripening and make fruit readily available for consumption artificially, high ethylene concentrations can result in spoilage and increase the fruit's susceptibility to pathogen attacks (University of Maryland Extension, 2021, Maduwanthi & Marapana, 2019). Therefore, managing ethylene exposure is essential to increase fruit shelf-life, produce marketable fruits, and minimize loss of profit (University of Maryland Extension, 2021).

Generally, fruits are categorized into two groups based on their ripening mechanisms: climacteric and non-climacteric (University of Maryland Extension, 2021; Maduwanthi & Marapana, 2019). Non-climacteric fruits such as cherries, grapes, and strawberries will not ripen after harvest and do not require ethylene (University of Maryland Extension, 2021). In contrast, climacteric fruits such as bananas, avocados, and apples continue to ripen after harvest are sensitive to ethylene concentrations (University of Maryland Extension, 2021). Notably, climacteric fruits are autocatalytic, meaning an initial exposure to ethylene promotes further ethylene production resulting in accelerated ripening (University of Maryland Extension, 2021). Respectively, ripe fruits secrete higher ethylene concentrations than unripe fruits (Burg & Burg 1965).

This study investigates the relationship between ethylene exposure and the ripening of fruits, particularly bananas (*Musa acuminata*). Previous research concluded that exogenous ethylene treatment could increase respiration rate and endogenous ethylene levels, thus inducing fruit ripening (Dominguez & Vendrell, 1994; Burg & Burg 1965). Moreover, due to the substantial secretion of ethylene from ripe fruits, an increase in ethylene concentrations in the surrounding environment is expected, which might accelerate the ripening of nearby fruit (Dominguez & Vendrell, 1994; Burg & Burg 1965). Hence, we hypothesize that the proximity of an unripe banana to a ripe banana will determine the ripening rate of the unripe banana. We predict that the ripe banana will serve as a source of exogenous ethylene; correspondingly, as the distance between the ripe and unripe banana increases, a decreased rate of ripening in the unripe banana should be observed.

## Methods

To investigate the effect of proximity on banana ripeness, we placed two bananas some distance apart from each other then we determined their degree of ripeness as a percentage of dark spots after one week.

First, we placed an unripe, green-colored banana (experimental banana) X distance away from another ripe, yellow-colored banana (ethylene source). The four treatments were: placing the experimental bananas either 0cm, 15cm, or 30cm away from an ethylene source - and a control treatment. The control treatment is where we placed the experimental banana without an ethylene source. The distance between the bananas was measured using a measuring tape and a right-angled triangle ruler.

**Figure 1.** Bookshelf with the first replicates of the four conditions (from top to bottom): 30cm, 15cm, 0cm, and control.

As demonstrated by Figure 1, each treatment was isolated on a different shelf in a tall bookshelf and left for one week. We placed the bookshelf 2m away from an east-facing window to induce natural light conditions. We used a thermostat and a thermometer to maintain the room temperature between 20 and 23 degrees Celsius throughout the experiment. Each treatment was replicated three times for reliability.


**Figure 2.** A shelf with the first replicate of the 15cm treatment. The distance between the two bananas was measured from the centers of the cis and trans faces.

Figure 2 shows how we measured the distance between the two bananas from the centers of the cis and trans faces of the bananas. We placed the measuring tape against the edge of the bookshelf and used the right-angled triangle ruler to ensure that the centers of the bananas were perpendicular and equidistant to the edge of the bookshelf.

All bananas used in this experiment are from the Dole brand and grown in Costa Rica. The bananas used in each replicate are from the same respective bunch. For example, we chose all experimental bananas in the second replicate from the same banana bunch and all ethylene sources in the third replicate from the same bunch.

After one week, we placed the experimental bananas in a dark room where we took photographs of all their four sides against a white-paper background. We used an iPhone 11 camera to take the photographs; we turned the flash function on to supply artificial light for consistency in the photographs.

**Figure 3.** A screenshot of the front side of the 0cm experimental banana (1st replicate) with the dark-spots represented in white using Adobe Photoshop.

Using Adobe Photoshop (2022 version), the photos of all four sides of each banana were normalized to size then merged to produce a 3-dimensional visualization of each banana. Using the quick-selection tool and threshold tool, we selected the areas of dark spots on the bananas, as illustrated in Figure 3. Then, we used the measurement tool to find the total number of pixels of dark spots and the number of pixels making up the total surface area of the banana. Equation (1) shows how we calculated the percentage of dark spots on the skin of each banana after one week. For consistency, we did not measure the surface area around the stem of the bananas.

$$\% \, of \, Dark \, Spots = \frac{Surface \, Area \, of \, Dark \, Spots \, (pixels)}{Total \, Surface \, Area \, of \, Banana \, (pixels)} \times 100\% \qquad (1)$$

We compiled the data onto Microsoft Excel, where we calculated the mean percentage of dark spots for each treatment across the three replicates. We performed a one-way ANOVA test for the four independent treatments to determine if a significant difference between the percentage of dark spots at a 0.05 significance level exists. We used a bar chart to illustrate the results.

## Results

Within the first 3 days for replicates one and two, green areas of the bananas turned yellow, and by the fourth day, there were little to no green areas left, and dark spots started to appear. By the seventh day, the bananas looked similar, all a deep-yellow color with many dark spots covering at least 50% of the banana. On the other hand, for replicate three, the banana color was slightly yellow for days one to four and still contained dark-green areas on day 5. On day 6, not all bananas had dark spots, but those that did had very few. By day seven, all bananas had dark spots covering 5 - 20% of the banana, but much less than the previous replicates. Overall, there is a trend present in all replicates; as the distance between the bananas increased, the percent coverage of dark spots also increased.

Figure 4 shows the mean percentage of dark spots on the ripening experimental banana when the two bananas are separated for the four treatments: control (17.5%), 0 cm (24.2%), 15 cm (27.8%), and 30 cm (34.1%). Over three replicates, the control treatment yielded the lowest surface area percentage of dark spots, while the 30 cm treatment yielded the greatest.  A significance level of 0.05 was used, and the ANOVA test produced a $p$-value of 0.76895, indicating that the differences between the treatment groups are not statistically significant.

Visually, the figure shows a positive correlation between distance and the percentage of dark spots.

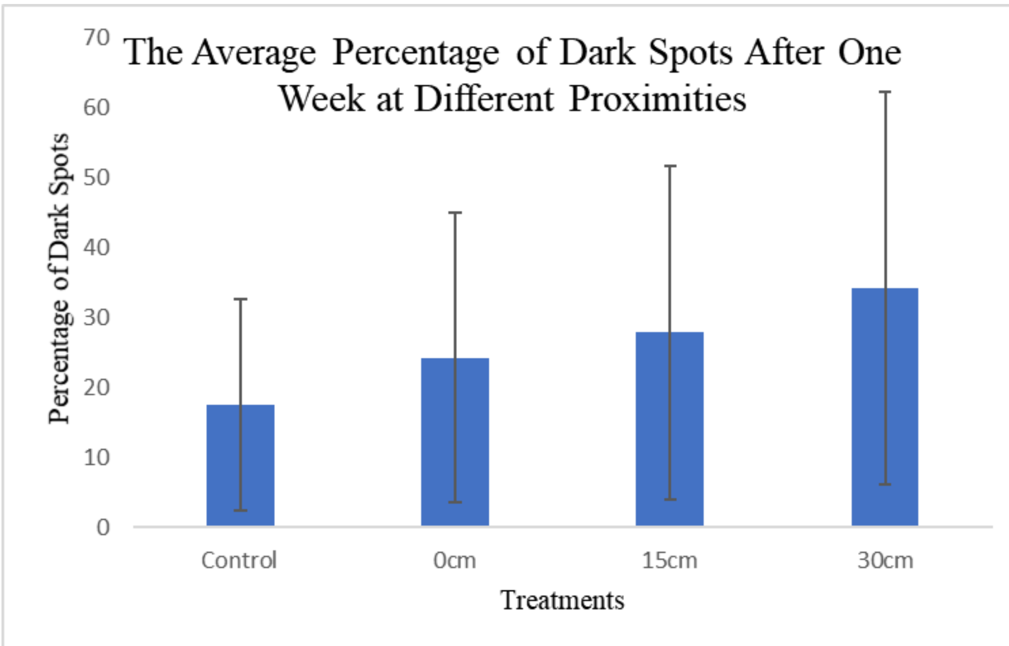


**Figure 4**. Mean percentage of dark spots for each treatment after one week of ripening. The bars represent surface area percentage of dark spots of the experimental banana for control (n = 3), 0 cm (n = 3), 15 cm (n = 3), 30 cm (n = 3). Errors bars illustrate ∓95% confidence intervals. A $p$-value of 0.76895 was obtained on the ANOVA test.

## Discussion

We observed more dark spots on the surfaces of experimental bananas when placed further away from the ethylene source. Respectively, fewer dark spots were observed on unripe bananas when closer to the ripe bananas. This observation suggests that the ripening rate increases further from the ethylene source as measured by the percentage of dark spots covering the banana surface over one week. However, since our findings were statistically insignificant, we cannot support this claim.

A one-way ANOVA test determined no statistical significance between exposure of an unripe banana to ethylene at different distances. Particularly, the test determined a p-value of 0.76895 ($p > 0.05$), which is much larger than the significance level with a 95% confidence interval. Furthermore, the error bars in Figure 4 indicate a large uncertainty among the treatments. Therefore, we can conclude that the mean percentage of the four conditions is insignificant. Moreover, these results show that the null hypothesis cannot be rejected; our results do not support our hypothesis that exposure to an ethylene source at a shorter distance will increase the ripening rate.

This experiment was conducted to determine the durability of unripe climacteric fruits, such as bananas, when stored with ripe climacteric fruits. Our main concern was to determine how bananas at different ripeness levels influence the ripening rate of unripe bananas when ethylene gas is released in a commercial environment. Although our hypothesis was not supported by our results, our findings pave the way towards determining and experimenting on variables that can potentially influence the ripening process of climacteric fruits.

Ethylene is a gaseous plant hormone that plays a significant role in the biological aging (senescence) and ripening of climacteric fruits (Maduwanthi & Marapana, 2019). As ethylene concentrations increase, senescence occurs and the fruit becomes sweeter and softer in texture. Eventually, the fruit spoils and turns brown, developing a mushy texture, and a sweet yet pungent smell and taste (University of Maryland, 2021; Maduwanthi & Marapana, 2019). Thus, when it comes to commercial uses of climacteric fruits, ethylene can promote delicious sweet fruit that is highly profitable or spoil the fruit, decreasing the quality of the product (Borkar et al., 2008). The shelf-life of climacteric fruits is important to provide the best quality fruits to consumers and generate revenue. Furthermore, understanding the ripening and spoilage of fruits is important to decrease wastage and prevent the ripening of neighbouring fruits. Borkar et al. (2008) studied the effectiveness of using non-perforated ethylene absorbent storage packages containing banana bundles to delay the ripening process. Their results showed using an absorbent to trap ethylene gas was significantly effective in increasing the shelf-life of bananas. Another study performed by Maduwanthi & Marapana (2019) observed the ripening of bananas through artificially inducing ripening with ethylene gas. The ethylene vapours used to induce ripening came from ethylene gas previously passed through ripe bananas. We incorporated some elements from Maduwanthi & Marapana's (2019) study to obtain results that could help us determine appropriate storage of bananas to increase shelf-life as achieved by Borkar et al. (2008).

Since the experiment results do not support the hypothesis, the results should be interpreted cautiously. Some factors that could explain the results achieved are differences in temperatures throughout the day and night, accumulation of ethylene gas from previous replicates that could influence later replicates, and altitude and pressure difference of bananas (perhaps negligible since altitude difference is small). Additionally, using Adobe Photoshop (2022 version) to build observations can lead to inaccurate results, depending on how the editing software is used. In this case, when developing the image, some regions of the images may have overlapped and cropped out, causing an underestimation of the percentage of dark spots covering the bananas. Lastly, the final replicates presumably used bananas of a different brand. These particular bananas ripened at a significantly lower rate and produced only a few dark spots, accounting for the large error bars in Figure 4. Further experimentation is required using proper methods to control extraneous variables, and different measures of ripeness, such as measuring the concentration of ethylene, observing changes in texture, taste, or scent, or a combination of various characteristics associated with ripening.

## Conclusion

This experiment aimed to determine whether the rate of ripening of a banana is affected by how far it is from an ethylene-abundant ripe banana. Overall, our study concludes that there is no significant relationship between the rate of ripening of unripe fruits and their placement at different proximities from ethylene sources. The results failed to support our hypothesis that there is an increase in the rate of ripening at closer proximity to the ethylene source. However, we now understand that the durability of bananas might be influenced by factors other than distance. Future experiments with an improved setup or a different experimental approach could find better ways to extend the shelf-life of climacteric fruits.

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