The Effect of Differing the Environmental H+ Ion Concentration (pH) of a Pole Bean Plant on the Concentration of CO_2 Gas the Plant Produces during Respiration

Abstract: The effect that varying pH on the production of CO_2 during respiration was tested by exposing pole beans to 4 to 10 pH solutions; a respirometer was used to measure the CO_2 output by five of beans over a set time period of about 300 seconds. The results demonstrate that the pH that resulted in the greatest CO_2 production was 8. The data showed a downward parabolic shape with pH 8 equal to the global maxima. A multiple regression analysis indicated a p-value of 0.383 (less than alpha=0.05 and therefore significant), and the data had a very strong correlation with an R^2 value of 0.9036.

1. Introduction:

1.1. Background Information:

Plant life plays an extremely important role in today's society. They are primary producers without which life would not exist. Trees reduce pollution in the atmosphere and innovations in agriculture are responsible for the sustenance of the growing humans population on Earth. Through respiration, nutrients are converted into adenosine triphosphate (ATP). There are many factors that affect the rate of this respiration, one being pH of the plants environment. This is important due to the many enzymes involved in this process. Dehydrogenase catalyzes the formation of acetyl -CoA as a part of the Krebs Cycle. NAD+ serves as an oxidizing agent for glucose. These enzymes/coenzymes all have a pH at which they are most efficient in carrying out their functions. The closer the environmental pH is to this ideal H+ concentration, the more efficient respiration will be. Consequently, as the pH of the environment strays from the optimum pH, the enzymes protein folding structures change and respiration is slowed. The more efficient respiration is the more CO2—a byproduct of the cellular process—will be released; as a result, the change in CO2 concentration can be used to indicate the efficiency of respiration.

Pole beans –the main specimen being examined—are otherwise known as green pole beans and wax pole beans and are in the Fabacaea family. They are native to North America, and they prefer full sun and well-drained soil. Additionally, they require average fertility and a pH between 6.0 and 6.8.

1.2. Question and Hypothesis:

As a result, the question being asked is how does the concentration of H+ ions in a plant's environment affect its rate of respiration? At what pH is most effective? The hypothesis being tested: If plants are placed in environments of varying pHs, then the plants with pHs closer to a neutral pH of 7 will have a maximized rate of respiration because this is the pH closest to the pH of water—a substance fundamental to life—at room temperature.

2. Materials and Procedure:

The materials needed: Vernier CO_2 meter, corresponding plastic container, 35 pole beans, 10 pH and 4 pH buffer solutions, 7 beakers, Computer with Vernier software, pH sensor, and distilled water**.**

1. Completely submerge all pole beans in distilled water for 24 hours
2. Fill one beaker with 80 mL pH 4 buffer solution and one with 80 mL 10 pH buffer solution and one with 80 mL distilled water. Label them pH 4, pH 10, and pH 7 respectively
3. Use the pH sensor to mix 4 pH buffer solution and distilled water to form 80 mL of 5 pH solution and 80 mL of 6 pH solution.
4. Use the pH sensor to mix 10 pH buffer solution and distilled water to form 80 mL of 8 pH solution and 80 mL of 9 pH solution.
5. Label each solution with its corresponding pH level
6. Completely submerge 5 pole beans in each beaker of solution for 5 minutes
7. Place all beans from the 4 pH solution into the plastic container
8. Seal the container using the CO_2 meter
9. Using the computer and Vernier software, measure the change in CO_2 concentration in the container for 5 minutes (300 seconds).
    a. Find the difference in CO_2 concentration (to be represented after this point as: $[CO_2]$) by subtracting the initial CO_2 concentration from the CO_2 concentration after 300 seconds in the respirometer.
10. Plot the data on a graph with time on the x axis and concentration of CO_2 on the y axis

11. Repeat steps 7-9 using the remaining pH solutions

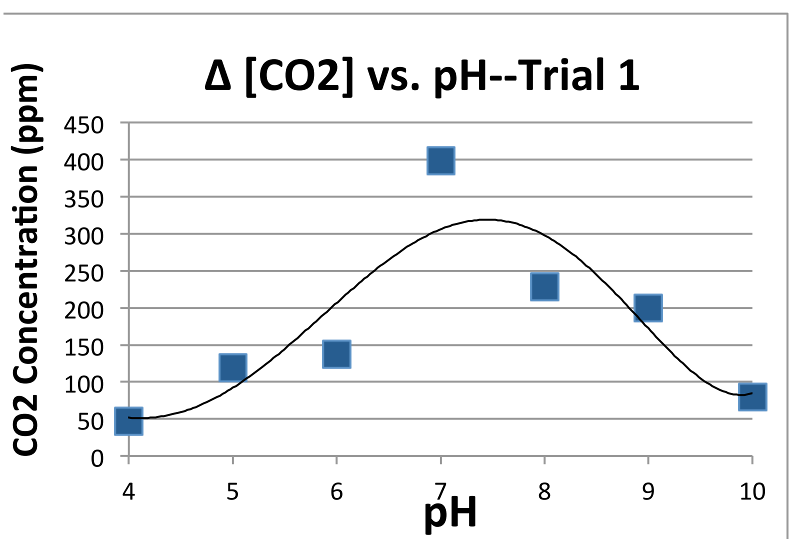
2.1 Observations:

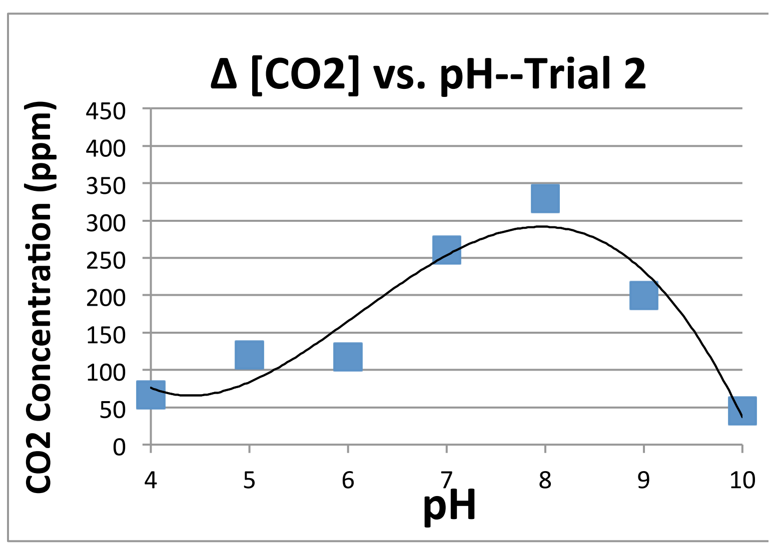
The pole bean is a plant that grows on a vine and has tuberous roots. The plant has red flowers and the beans grow in green pods. The beans that were used in this study had off white-cream color and were about 2 centimeters in length. After being soaked in water, they were moist to the touch. A thin film like layer covered the beans and some of the beans had visible embryos

3. Results:

	pH	$[CO_2]$ initial (ppm)	$[CO_2]$ final (ppm)	Difference (Δ) (ppm)
Trial 1	4	1178.67627	1225.662354	46.98608399
	5	1212.45	1332.1521	119.7020996
	6	1212.45	1350	137.55
	7	1411.437988	1809.692383	398.2543945
	8	1576.843262	1805.419922	228.5766602
	9	1503.43	1703.216553	199.7865527
	10	1292.266846	1372.2229	79.95605469

	pH	$[CO_2]$ initial (ppm)	$[CO_2]$ final (ppm)	Difference (Δ) (ppm)
Trial 2	4	1088.67627	1155.662354	66.98608399
	5	1142	1262.1521	120.1520996
	6	1244.812012	1362.1521	117.3400879
	7	1440	1700	260
	8	1460	1790	330
	9	1503.43	1703.216553	199.7865527
	10	1305	1350	45



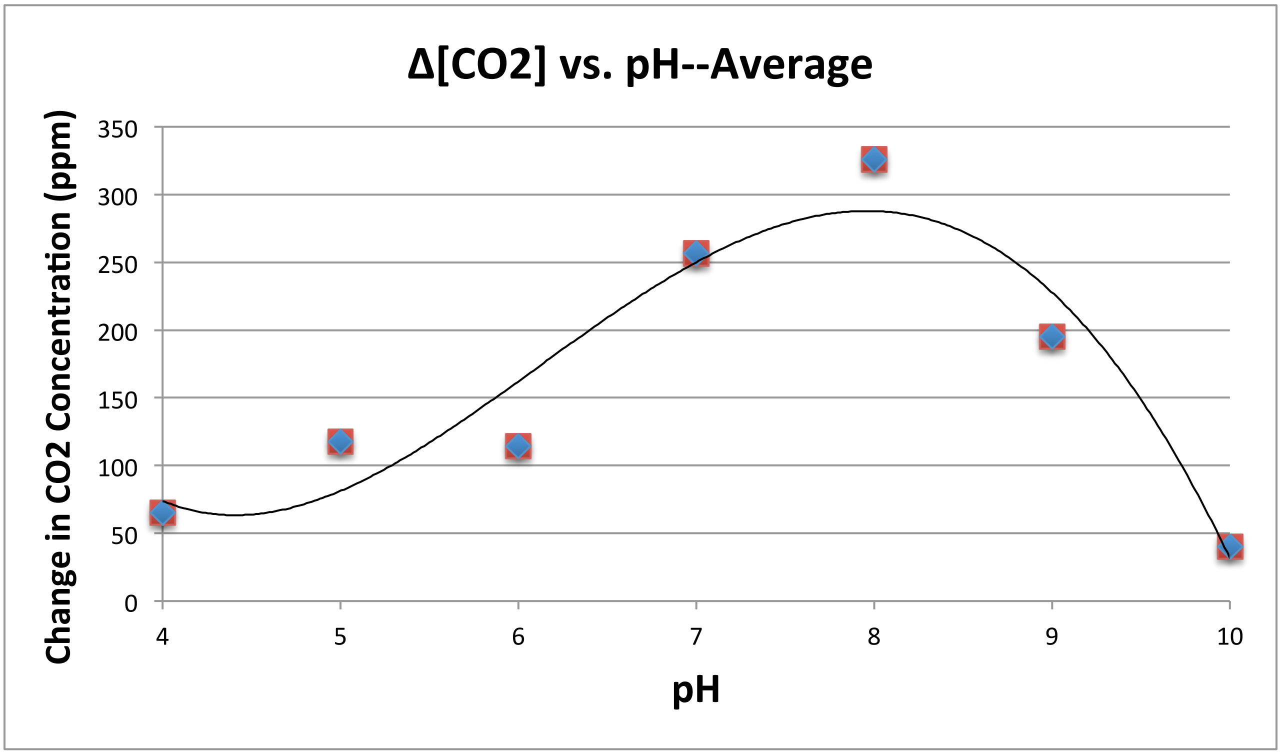


$[CO2]\text{-hat} = 0.699x^5 - 21.056x^4 + 235.07x^3 - 1200.6x^2 + 2807.7x - 2339.7$

$R^2 = 0.7635$

$[CO2]\text{ - hat} = 0.3929x^4 - 19.731x^3 + 273.39x^2 - 1394x + 2439.9$

$R^2 = 0.9039$



$[CO2]\text{-hat} = 0.3929x^4 - 19.731x^3 + 273.39x^2 - 1394.5x + 2439.9$

$R^2 = 0.90346$

4. Statistical Analysis:

Polynomial Regression: Saturday, January 3, 2014, 9:47:11 AM

Order 0
[CO2] = 7.000

Order 1
[CO2] = 6.322 + (0.00421 * [pH])

Order 2
[CO2] = 8.096 - (0.0234 * [pH]) + (0.0000754 * [pH]^2)

Order 3

[CO2] = 13.907 - (0.174 * [pH]) + (0.00107 * [pH]^2) - (0.00000184 * [pH]^3)

Order 4
[CO2] = 2439.9 – (1394.54 * [pH]) + (273.39 * [pH]^2) – (19.731 * [pH]^3) + (0.3929 * [pH]^4)

Regression Results:

Order	**MSres**	**MSincr**
0	4.667	4.667
1	5.364	-0.698
2	6.143	-0.779
3	5.286	0.858
4	4.342	0.313

Regression Results: Incremental

Order	**Rsqr**	**F**	**P**
0	0.000		
1	0.0421	0.220	0.659
2	0.0803	0.366	0.578
3	0.311	1.649	0.289
4	0.391	2.103	0.103

Regression Results: Overall

Order	**Rsqr**	**F**	**P**
0	0.000		
1	0.0421	0.220	0.659
2	0.122	0.279	0.625
3	0.434	0.766	0.446
4	0.9034	0.839	**0.383**

5. Analysis of Results:

5.1 Summary of Computer Output:

Under each order column is the degree of the equation (order of the equation) modeling the graph. The higher the order, the more accurate the equation in terms of following the graph, but each order provides its own details.

The regression equations are followed by three statistical regression tests. The first section shows an estimate of the variance in the data for each equation. The next two sections show the correlation of each regression equation with our data. Based on the p-value that this output returns for the fourth-order equation (0.383), p is less than a standard alpha value (α) of 0.05. As a result, it can be reasonably assumed that there is a correlation between these data and that they are statistically significant.

5.2. General Analyses:

The major trend in the data was the parabolic shape that the graph took on, with its vertex at pH 8 and its lowest points at pH 4 and pH 10. This relative maxima was at pH 8; the further the pH was from this optimal pH, the less CO_2 was produced, and therefore, the less efficient respiration was.

Another notable feature from the data is that pH's above 8 seem to decrease CO_2 production more than pHs below 8; four pH levels (4, 5, 6, 7) fall below the vertex on the lower end, while only two (9, 10) fall below the vertex on the higher end. In other words, the curve is not symmetrical; as a result, pHs above the optimal of 8 seem to detract from respiration efficiency than pH's below 8 do.

Ultimately, though, the data seems to follow a reasonable trend without much of an outlier. The cumulative graph of the difference in CO_2 production between the pHs has a strong correlation with an R^2 value of approximately .9036. From this, it is reasonable to assume that 90.36% of the variance in CO_2 concentration can be attributed to pH. It was also found that the graph of most closely fits a fourth-order polynomial equation defined by $[CO2] = 0.3929 * [pH]^4 - 19.731 * [pH]^3 + 273.39 * [pH]^2 - 1394.54 * [pH] + 2439.9$

6. Conclusion:

From the data, it can be concluded that the optimal pH for CO_2 production is 8. Evidence to support this conclusion is found within the data, which was found to be repeatable through multiple trials; pH 8 was found to be correlated to the largest change in CO2 concentration and therefore, be most efficient. The results were statistically significant because the p-value of 0.383, located in the overall regression results, is less than $\alpha = 0.05$. The reasoning behind this is also fairly simple; a pH too acidic or too basic (too close to 4 or 10) would have a negative effect on the cells, potentially killing them. The optimal level would have to be near something that is reasonably found in plants, which would be close to the level of water. Reactions that take place during respiration and photosynthesis would change the pH level of the solutions around them somewhat, though, leaving 8 as a reasonable choice.

The hypothesis was not completely refuted. Although pH 7 was not found to be the optimal pH, it was very close, and the statement of the optimal pH being closer to a neutral pH is still true. This pH is still within a range that will not harm the components that go into pH and is healthy for bodily functions.

7. Sources of Error:

1. Only previously used pH buffer solution was available, so the pH solutions may have been incorrectly created. Also, since these were used to calibrate the pH probe, all subsequent pH readings may have been skewed.

2. The pH probes seemed to be inaccurate; for example, a professionally standardized pH solution of 4 read 4.4 on the pH probes used. Even though each read was adjusted based on this standard, the results may have been easily corrupted by defective equipment.

3. For some of the pole beans in the trials, the pH of the solutions may have killed the plant and stopped all respiration activity. This would affect the CO_2 measured because fewer than five pole beans would be respirating.

4. In the background research done on pole beans, it was discovered that they prefer pH 6-6.8. This is in discordance with the optimal pH 8 that was discovered in this lab. A possible reason is that the pole beans used were old and as a result, are not representative of the entire pole bean population. Additionally, only two trials were done, further decreasing precision of these data.

8. Future Testing:

In the future, instead of using pH solutions that were prepared in the lab, professionally standardized pH solutions could be used. Also, various chemicals that plant life is frequently is contact with (such as acid rain and waste near a plant) could be used to further test the effects of pH on this plant life. Furthermore, the pole beans could be placed in the "full sun." This may result in differing results. Further trials with pole beans and with additional species of plants and beans could be done to confirm and improve data.

9. Incorporation of AP Statistics feedback:

After receiving the feedback, the experiment was largely modified. Originally, the procedure had many sources of error (as the students demonstrated); these sources of bias were considered. Still, the ultimate product was quite different than the initial research plan submitted to AP Stats. The procedure was simplified and more statistical analysis (such as multiple regression and R^2) were introduced.