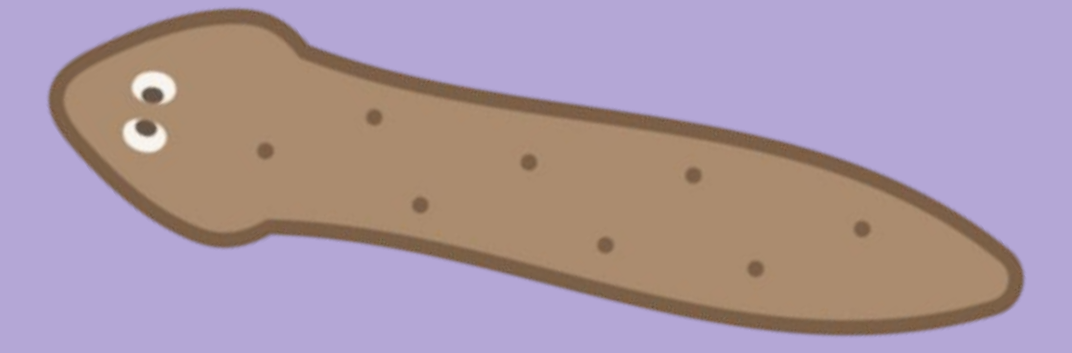
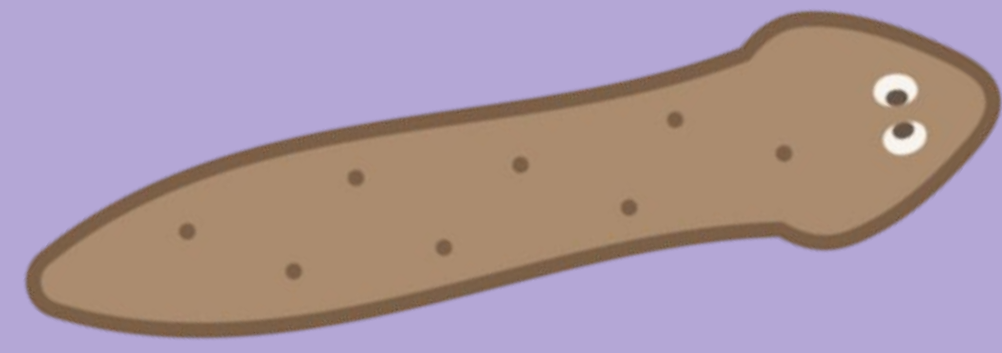


# The Effect of Calcium Concentration on Planarian Regeneration.

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## Abstract

Planarian regeneration occurs naturally after the organism is amputated. This process involves calcium signaling to start the growth of stem cells at the wound site to promote regeneration. To test the effect of additional calcium on the rate of regeneration, we put the planarian into solutions of 0, 0.1%, 0.5% and 1.0% solutions of CaCl and amputated them to observe the change in regeneration rate. The worms in the 0.5% and 1.0% exploded, likely due to an excess of calcium. The worms in 0.1% solution showed slightly improved regeneration, suggesting that calcium could have a positive effect on regeneration in low concentrations, but this observation was not supported by statistics.

## Introduction

Planarian regeneration generally takes place throughout a 14-day period, in which the planarian regenerate the part of their body that was removed. They first undergo wound closure, then their tissue is remodeled, then the eyespots reform if the head was cut off, and finally the pigment returns to the planaria. During regeneration,  $Ca^{2+}$  signaling plays a significant role in controlling stem cell behavior. While it is certain that calcium is needed for regeneration to occur, it is uncertain if an increased concentration of calcium ions in the water will affect the rate of regeneration. This research explores the testable question: **How does the concentration of calcium in the external environment affect the regeneration rate?** Based off our previous knowledge of planarian regeneration, we hypothesized that **if we increased concentrations of calcium it will speed up the process of regeneration.**

## Methodology

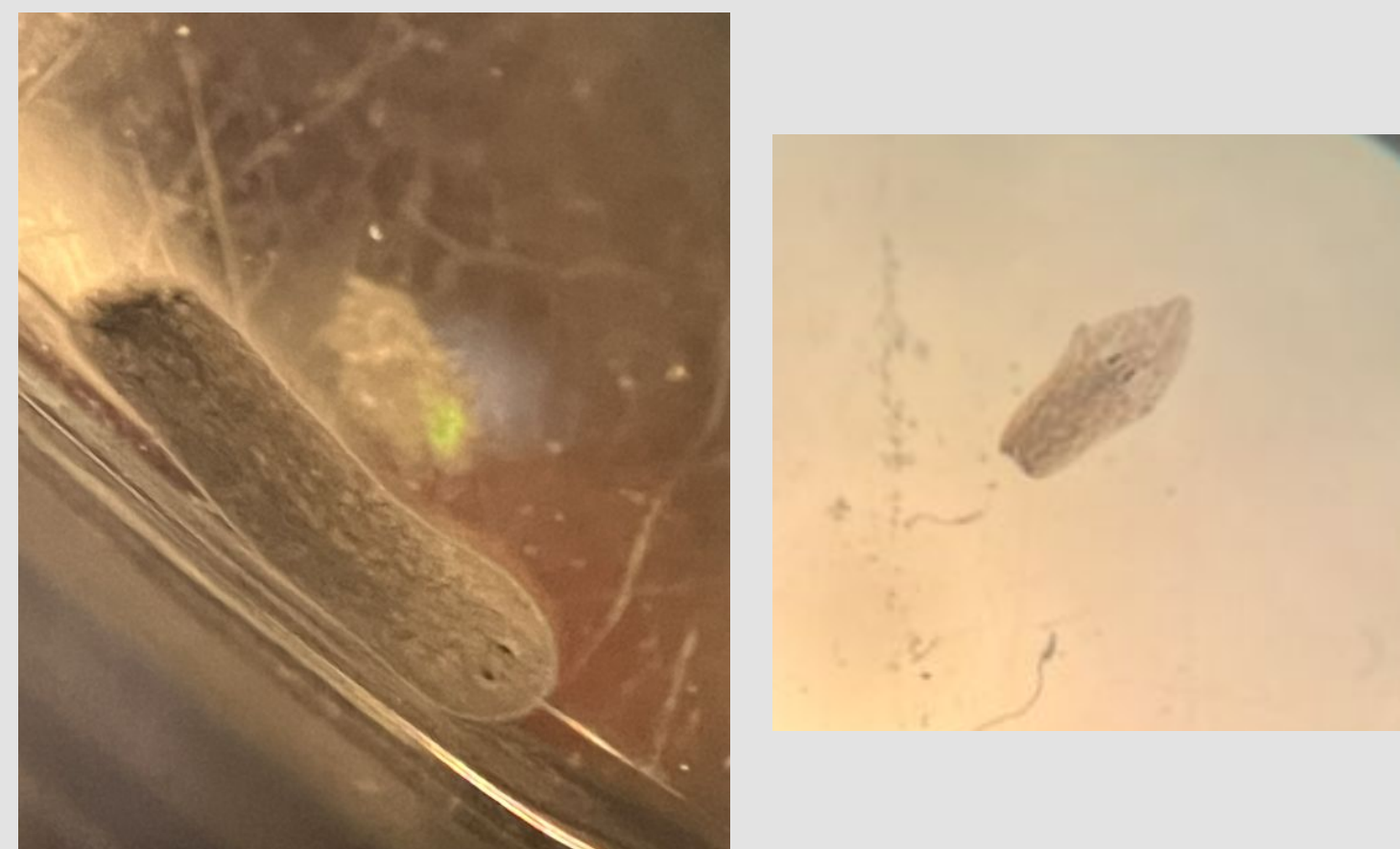
In order to conduct our experiment, we first experimented with planarians and observed their behavior and amputation. To set up our experiment we used our knowledge of planarian regeneration to formulate an experimental variable to improve the rate of regeneration. We landed on changing the concentration of calcium in the water, as calcium signaling was a crucial part of regeneration.

### Procedure:

**Experiment 1:** Increased the percentage of calcium to .1% (CaCl dissolved into water)

**Experiment 2:** Increased the percentage of calcium to .5% (CaCl dissolved into water)

**Experiment 3:** Increased the percentage of calcium to 1.0% (CaCl dissolved into water)



**Experiment 1:**

$$X^2 = 0.583$$

**Experiment 2:**

$$X^2 = 9$$

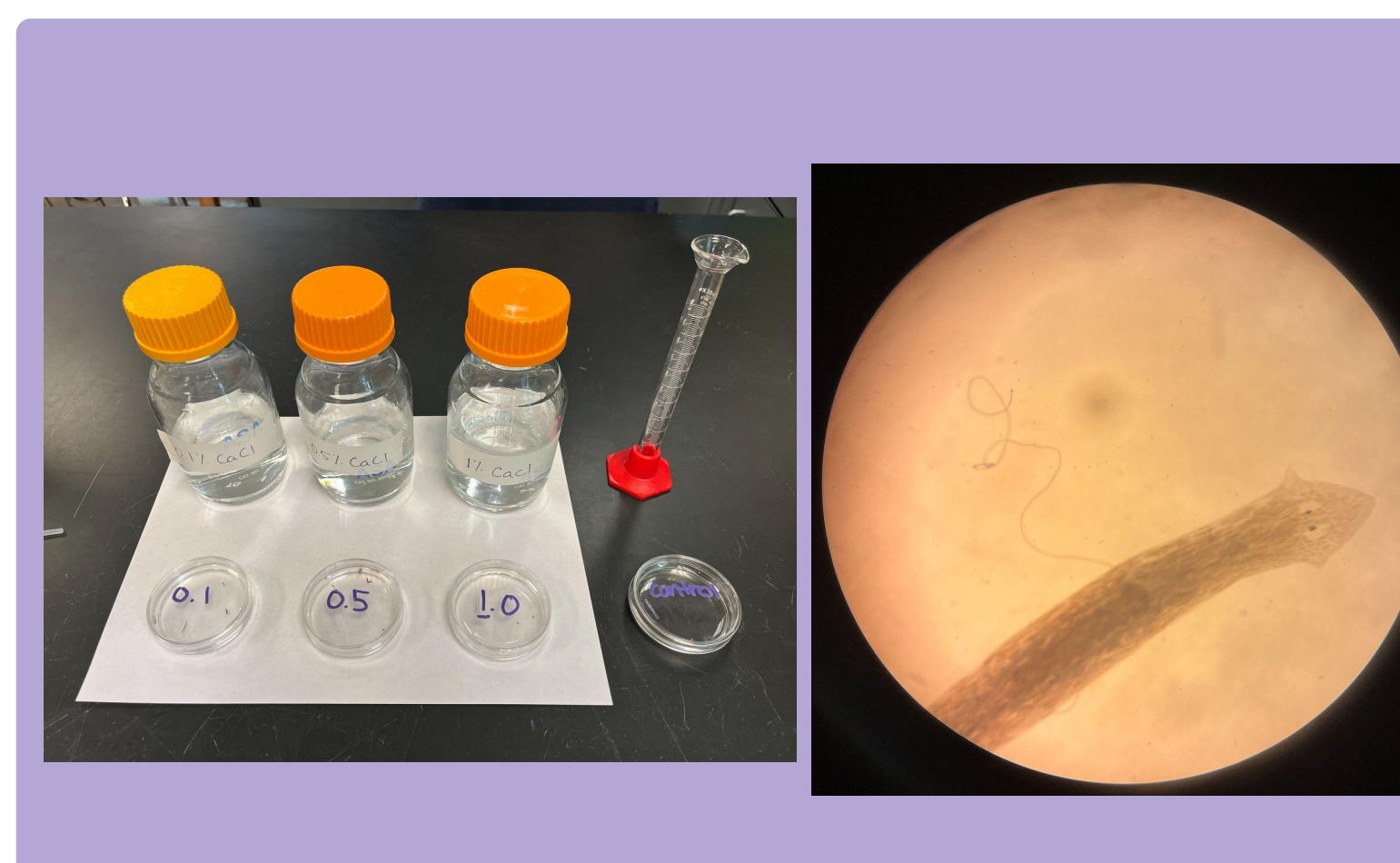
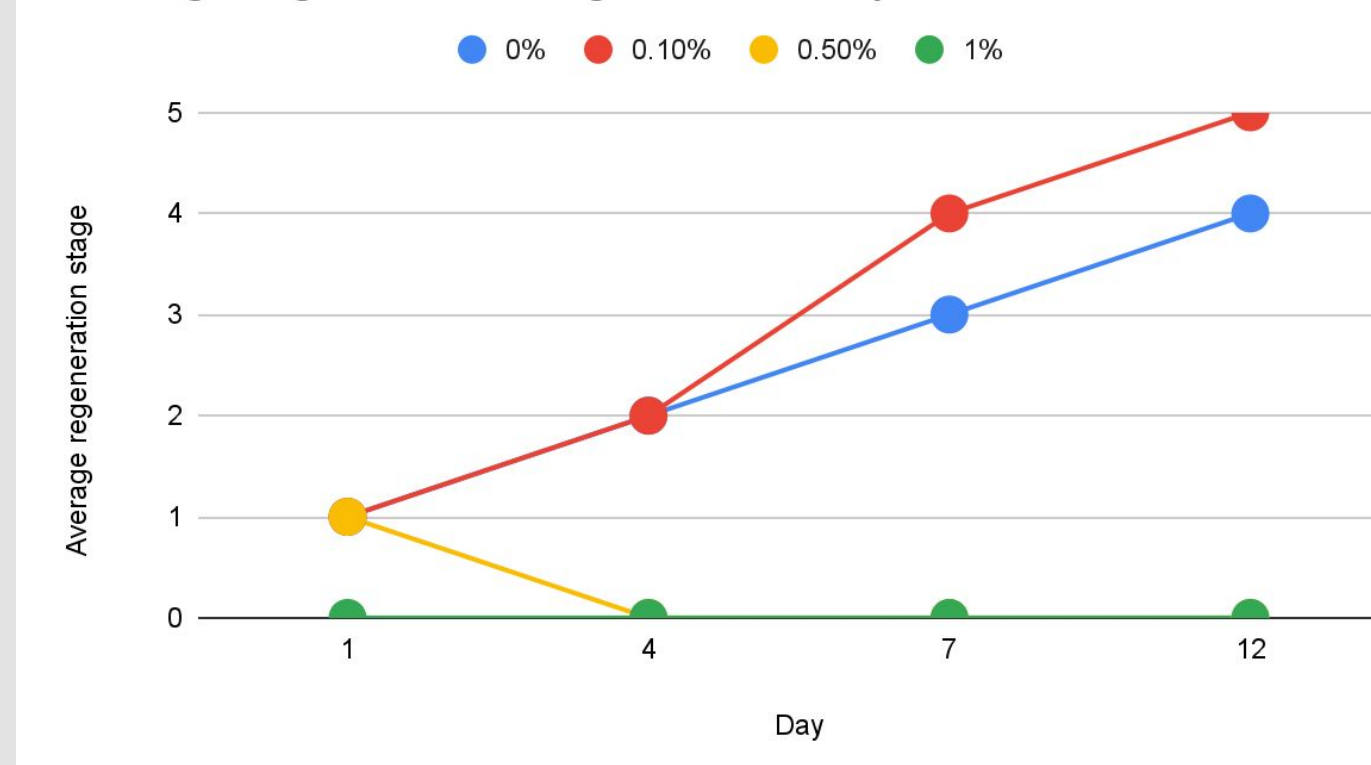
**Experiment 3:**

$$X^2 = 10$$

## Results

DAY	Average Regeneration Stage			
	1	4	7	12
Control group (0.0 %)	1	2	3	4
Experiment 1 (0.1%)	1	2	4	5
Experiment 2 (0.5%)	1	0 (died)	0 (died)	0 (died)
Experiment 3 (1.0%)	0 (died)	0 (died)	0 (died)	0 (died)

Average regeneration stage over 12 days



## Discussion

The data that we obtained showed that planarian do not regenerate faster when in an environment of increased calcium concentration. Even though calcium is needed to carry out regeneration, there is not enough data to confirm that calcium concentration affects the regeneration rate in a positive way. In fact, high percentages of calcium actually killed the planarian, as were the cases in the trials with 0.5% and 1.0% calcium. Therefore, our rate of regeneration actually stopped completely with increased calcium concentration because the worms did not survive. Although the planarian in the 0.1% calcium solution appeared to regenerate faster, there was not a great enough increase to statistically confirm this.

Our hypothesis was that if we increased concentrations of calcium, it would speed up the process of regeneration. Based on our data we found the opposite to be true for calcium concentrations of 0.5% and 1.0%. However, human error might have been the reason for some of these results. Due to many of the planarian being very small and thin, it was difficult to determine where to cut the worm so that the pharynx was not split in half. If the pharynx was cut, then the planarian would automatically die and not be able to regenerate the stem cells needed to regrow parts of their body. Using statistics, we accepted our null hypothesis for experiment 1 and rejected our null hypothesis for experiments 2 and 3 that increased calcium concentration has no effect on the rate of regeneration in planarian. We would conduct further studies by conducting additional experiments with calcium concentrations immediately above and below 0.1%.