Zinc Toxicity and Its Effects on *Arabidopsis thaliana* and Its Mutants *zip-2* and *nca1-1*

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Experimental design

1. Background info:
   - Zinc is a micronutrient in the soil that plays a critical part in the growth of plants.
   - Zinc toxicity-an excessive amount of zinc in the soil can cause catastrophic effects to the plant (as more than 200 ppm will kill it)
   - The effects of zinc toxicity-expressed in discoloration, stunting of growth, underdevelopment of growth, and even deterioration of root cells (Rout 2009).
   - These destructive effects on plants had become the focus of the experiment and the test subject was Arabidopsis *thaliana*. Not only was this research based on the Colombian Wild Type, but also two mutants by the names of *nca1* and *zip2*.
   - The function of the *nca1* mutant is to have slowed down catalase activity, which causes the plant to grow more gradually (Li 2015) as well as having the ability to have facilitated diffusion specifically with metal ions (see **Figure 1**)
   - The *zip2* mutant's function is silencing genes for a more rapid growth, which causes it to grow not as healthily as the Wild Type (Milner) as well as having an extra metal diffuser to aid in the bonding and transferring of metal (see **Figure 1**)

Question:

- How does an increased amount of zinc impact the stem length and biomass of the Arabidopsis thaliana Wild Type, as well as its mutants *zip-2* and *nca1-1*?

1. Setups
   - Lights above four blue bins that held our plants (see **Figure 2**)
   - Inside these blue bins held green trays with our plants made separate by their type and treatment and grew from pods (see **Figure 3**)
   - There was also also a gray bin for watering our plants with a zinc solution (20.0g of zinc sulfate per l liter of water, 16.15g per l liter of water was used earlier in the experiment), and a bin for watering plants with tap water (see **Figures 9 and 10**)
2. Variables (Dependant/Independent)
   - Independent variable: 20.0g of zinc sulfate per l liter of water, 16.15g per l liter of water (Depending on the date of the experiment)
   - Dependent variables: stem height and biomass (weight of dry mass)
   - Controlled variables: light, air quality, size of pods, water, heat, and levels of other micronutrients

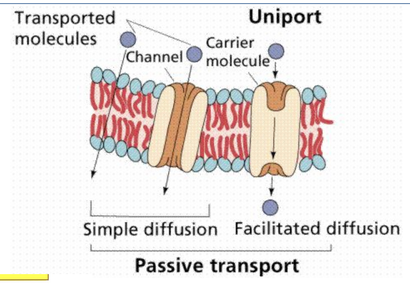

Figure 1 *the left side describes simple diffusion and this is how* zip2 *is able to transfer zinc through each cell membrane. The right side illustrates facilitated diffusion, and this is how* nca1-1 *mutant transfers and bonds with zinc*

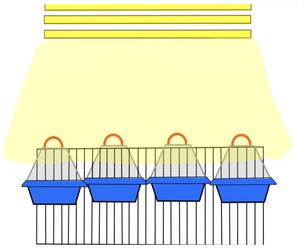

Figure 2 *light and covers on our bins*

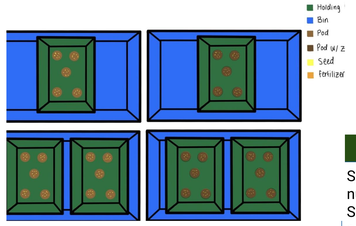

Figure 3 *plant arrangement*

Results

Stem Height:

Wild Type (0.1% statistical chance these conclusions are inaccurate)-Wild Type with zinc solution grew taller quicker than the control

nca1-1 (90% statistical similarity between their results)-The stem height of the *nca1-1* controlled plants and plants with added zinc had similar measurements throughout the experiment, with the control doing slightly better in the beginning, then later the *nca1-1* with zinc solution surpassing its growth (as seen in the stem height graph)

zip-2-The *zip-2* plants with the zinc solution grew significantly slower than the controlled *zip-2* plants

Biomass:

Wild Type-Wild Type control plants had a significantly larger biomass than Wild Type plants with zinc solution.

nca1-1-The nca1 control also had a larger biomass than the treatment

zip-2-*zip-2* control plants had a significantly larger biomass than the *zip-2* with zinc solution (as shown by our measurements seen in the graph above).

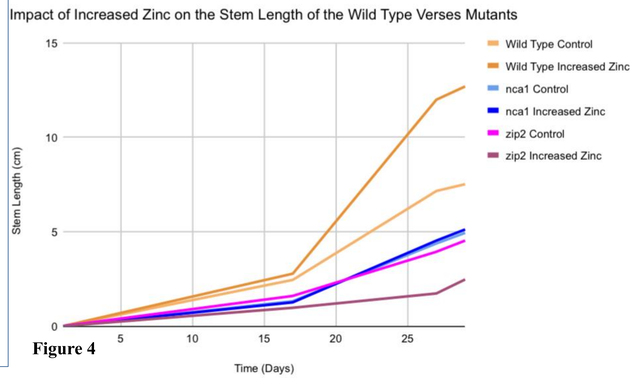

Figure 4

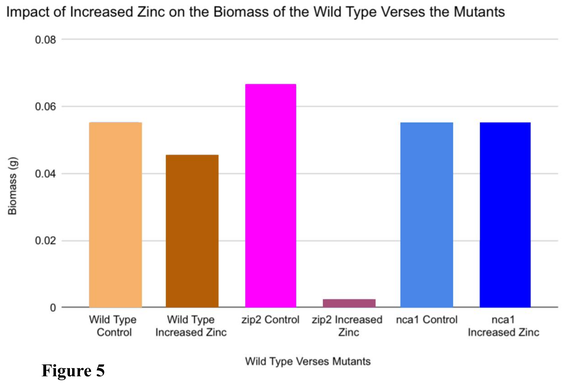

Figure 5

Figure 6 *(Wild Type control vs increased zinc)*

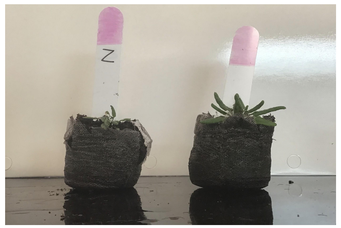

Figure 7 *(zip2 control vs increased zinc)*

Figure 8 *(nca1 control vs increased zinc)*

Conclusions

Our Independent Variable does cause our dependent variable because:

- The Colombian Wild Type control and increased zinc proved its divergence through statistical evidence
- Our Wild Type with added zinc grew taller than the control (see **Figure 4**), but had a smaller biomass to depict that the increased growth was a response to stress (see **Figure 5**)
- Premature flowering was also displayed as a result of stress (see **Figure 6**)
- The *zip2* results exemplified a critical stunting of growth in the increased zinc, as it had a large difference in both its biomass (see **Figure 5**) and stem height (see **Figure 4**)
- The lacking size and strength of the leaves in the *zip2* plant compared to the control also illustrated this difference (**Figure 7**)

However:

- The *nca1* results indicated a strong resemblance between the results of both the stem height (see **Figure 4**) and biomass (see **Figure 5**) displaying that zinc toxicity does not strongly impact its growth
- Discoloration and premature flowering was also exhibited in the *nca1* plants with increased zinc (see **Figure 8**)

Errors in our experiment that could have led to inaccurate results

- Not taking the covers off the top of our green trays: It was not anticipated that the elongated time of the covers on top of our plants would cause the growth of fatal fungus (see **Figure 2**)
- Taking the covers off the top of our green trays: When the tops were removed to stop the fungus, the plants became dehydrated more quickly (see **Figure 3**)
- Leaving the plants unattended: After we returned from a three day absence, our subjects were critically dehydrated and we needed to water our plants with an extra 200 mL of water (see **Figures 9 and 10**)
- A watering day without added zinc: Water was added to our plants over a long weekend but zinc was not added to the water as we had done in the past

Figure 9

Figure 10

Acknowledgements

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