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Winter is coming! Can you handle the freeze?

Featured scientist: Doug Schemske from Michigan State University

Research Background:

Doug is a biologist who studies plants from around the world. He often jokes that he chose to work with plants because he likes to take it easy. While animals rarely stay in the same place and are hard to catch, plants stay put and are always growing exactly where you planted them! Using plants allows Doug to do some pretty cool and challenging experiments. Doug and his research team carry out experiments with the plant species Mouse-ear Cress, or *Arabidopsis thaliana*. They like this species because it is easy to grow in both the lab and field. *Arabidopsis* is very small and lives for just one year. It grows across most of the globe across a wide range of latitudes and climates. *Arabidopsis* is also able to pollinate itself and produce many seeds, making it possible for researchers to grow many individuals to use in their experiments.

Doug wanted to study how *Arabidopsis* is able to survive in such a range of climates. Depending on where they live, each population faces its own challenges. For example, there are some populations of this species growing in very cold habitats, and some populations growing in very warm habitats. He thought that each of these populations would adapt to their local environments. An *Arabidopsis* population growing in cold temperatures for many generations may evolve traits that increase survival and reproduction in cold temperatures. However, a population that lives in warm temperatures would not normally be exposed to cold temperatures, so the plants from that population would not be able to adapt to cold temperatures. The idea that populations of the same species have evolved as a result of certain aspects of their environment is called **local adaptation**.





Doug, and two members of his team, setting up the reciprocal transplant experiment in Scandinavia.

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To test whether *Arabidopsis* is locally adapted to its environment, Doug established a **reciprocal transplant experiment**. In this type of experiment, scientists collect seeds from plants in two different locations and then plant them back into the same location (home) and the other location (away). For example, seeds from population A would be planted back into location A (home), but also planted into location B (away). Seeds from population B would be planted back into location B (home), but also planted at location A (away). If populations A and B are locally adapted, this means that A will survive better than B in location A, and B will survive better than A in location B. Because each population would be adapted to the conditions from their original location, they would outperform the plants from away when they are at home ("home team advantage").

In this experiment, Doug collected many seeds from warm Mediterranean locations at low latitudes, and cold Scandinavian locations at high latitudes. He used these seeds to grow thousands of seedlings. Once these young plants were big enough, they were planted into a reciprocal transplant experiment. Seedlings from the Mediterranean location were planted alongside Scandinavian seedlings in a field plot in Scandinavia. Similarly, seedlings from the Scandinavian locations were planted alongside Mediterranean seedlings in a field plot in the Mediterranean. By planting both Mediterranean and Scandinavian seedlings in each field plot, Doug can compare the relative survival of each population in each location. Doug made two local adaptation predictions:

- Scandinavian seedlings would survive better than Mediterranean seedlings at the Scandinavian field plot.
- 2. Mediterranean seedlings would survive better than Scandinavian seedlings planted at the Mediterranean field plot.



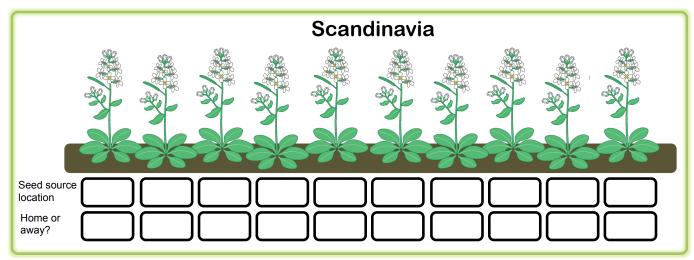
Mouse-ear Cress, or *Arabidopsis* thaliana, growing in the field.

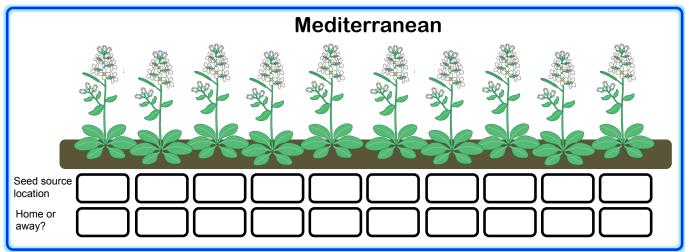
<u>Scientific Question 1</u>: Are populations of *Arabidopsis* locally adapted?

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<u>Draw your local adaptation predictions</u>: Below is a diagram of two field plots: one at a Scandinavian location and one at a Mediterranean location. Think about how seedlings originally from Mediterranean and Scandinavian locations would survive in a field plot at home versus away.

- 1. Start by looking at the diagram of the field plots. There are 10 seedlings in each plot- 5 from the Mediterranean population and 5 from the Scandinavian population. Fill in the first row of boxes with an **M** or **S** to represent the seed source location (where the seed was collected).
- 2. Next, think about whether each seedling is in its home or its original (away) location. Write "home" or "away" in the second row of boxes below each plant.
- 3. Now draw your survival predictions. Based on what you learned in the background information about local adaptation, which plants do you think will survive in each field plot? Put an X over the plants from each population that you predict will not survive. Think about whether the seedlings will have higher survival at home or away.





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Scientific Data 1:

Use the data from Table 1 to answer scientific question 1:

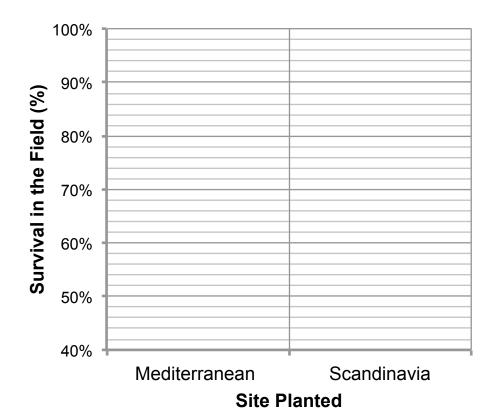
Source of Seeds	Site Planted	Home or Away?	Survival in the Field (%)
Mediterranean	Mediterranean		92%
Mediterranean	Scandinavia		48%
Scandinavia	Mediterranean		62%
Scandinavia	Scandinavia		73%

What data will you graph to answer the question?

Independent variable(s):

Dependent variable(s):

Draw your graph below:



Mediterranean Seeds

Scandinavian Seeds

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Interpret the data:
Make a claim that answers the scientific question.
What evidence was used to write your claim? Reference specific parts of table 1 or th graph.
Explain your reasoning and why the evidence supports your claim. Connect the data back to what you learned about local adaptation in populations of the same species.

<u>Doug's next steps:</u> The data from Doug's reciprocal transplant experiment show that the *Arabidopsis* populations are locally adapted to their home locations. Now that Doug confirmed that populations were locally adapted, he wanted to know how it happened. What is different about the two habitats? What traits of *Arabidopsis* are different between these two populations? Doug now wanted to figure out the mechanism causing the patterns he observed.

Doug originally chose *Arabidopsis* populations in Scandinavia and the Mediterranean for his research on local adaptation because those two locations have very different climates. The populations may have adapted to have the highest survival and reproduction based on the climate of their home location. To deal with sudden freezes and cold winters in Scandinavia, plants may have adaptations to help them cope. Some plants are able to protect themselves from freezing temperatures by producing chemicals that act like **antifreeze**. These chemicals accumulate in their tissues to keep the water from turning into ice and forming crystals. Doug thought that the Scandinavian population might have evolved traits that would allow the plants to survive the colder conditions. However, the plants from the Mediterranean aren't normally exposed to cold temperatures, so they wouldn't have necessarily evolved freeze tolerance traits.

To see whether freeze tolerance was driving local adaptation, he set up an experiment to identify which plants survived after freezing. Doug again collected seeds from several different populations across Scandinavia and across the Mediterranean. He chose locations that had different latitudes because latitude affects how cold an area gets over the year. High latitudes (closer to the poles) are generally colder and low latitudes









Setup of Doug's freeze tolerance experiment. Seeds germinated (1) and grew up into seedlings (2). Seedlings were placed in a freezer (3), and Doug measured the percent of seedlings that survived, and how many froze to death (4).

(closer to the equator) are generally warmer. Doug grew more seedlings for this experiment, and then, when they were a few days old, he put them in a freezer. Doug counted how many seedlings froze to death, and how many survived, and he used

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these numbers to calculate the percent survival for each population. To gain confidence in his results, he did this experiment with three replicate genotypes per population.

Doug predicted that if freeze tolerance was a trait driving local adaptation, the seedlings originally from colder latitudes (Scandinavia) would have increased survival after the freeze. Seedlings originally from lower latitudes would have decreased survival after the freeze because the populations would not have evolved tolerance to such cold temperatures.

Scientific Question 2: Are there differences in freeze tolerance between Scandinavian and Mediterranean populations of *Arabidopsis*?

<u>What is the hypothesis?</u> Find the hypothesis in the Research Background and underline it. A hypothesis is a proposed explanation for an observation, which can then be tested with experimentation or other types of studies.

Scientific Data 2:

Use the data from Table 2 to answer scientific question 2:

Source of				Survival after
Seeds	Trial #	Population	Latitude	freezing (%)
Scandinavia	1	RYGG	59.38	50
Scandinavia	2	RYGG	59.38	50
Scandinavia	3	RYGG	59.38	53
Scandinavia	1	HAMM	59.78	47
Scandinavia	2	HAMM	59.78	49
Scandinavia	3	HAMM	59.78	53
Scandinavia	1	INNF	62.5	96
Scandinavia	2	INNF	62.5	96
Scandinavia	3	INNF	62.5	98
Scandinavia	1	RODA	62.8	71
Scandinavia	2	RODA	62.8	77
Scandinavia	3	RODA	62.8	80
Mediterranean	1	SPE	41.76	3
Mediterranean	2	SPE	41.76	5
Mediterranean	3	SPE	41.76	5
Mediterranean	1	POB	41.35	7
Mediterranean	2	POB	41.35	16
Mediterranean	3	POB	41.35	17
Mediterranean	1	MUR	41.34	36
Mediterranean	2	MUR	41.34	37
Mediterranean	3	MUR	41.34	44
Mediterranean	1	VDM	42.03	40
Mediterranean	2	VDM	42.03	49
Mediterranean	3	VDM	42.03	53

	Average	SD	SE
Scandinavia - Survival After Freezing %		20.45	6.17
Mediterranean - Survival After Freezing %		18.95	5.71

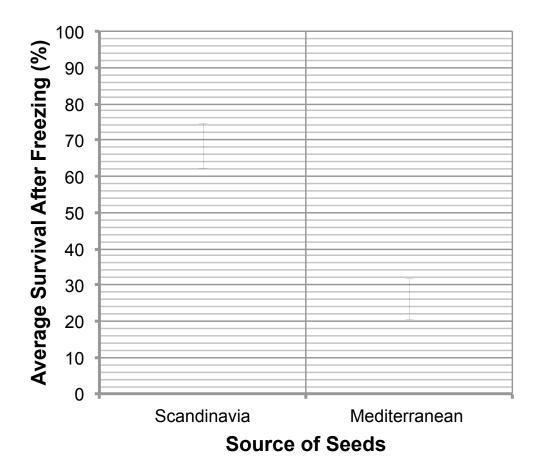
^{*}Standard deviation (SD) tells us about the amount of variation in the data. A large SD means there is a lot of variation around the mean, while a small SD means the data points all fall very close to the mean. Standard error (SE) tells us how confident we are in our estimate of the mean, and depends on the number of replicates in an experiment and the SD. A large SE means we are not very confident, while a small SE means we are more confident.

What data will you graph to answer the Scientific Question 2?

Independent variable(s):

Dependent variable(s):

Draw your graph below:



Name
Interpret the data:
Make a claim that answers the scientific question.
What evidence was used to write your claim? Reference specific parts of table 2 or the graph.
Explain your reasoning and why the evidence supports your claim. Connect the data back to what you learned about freeze tolerance and its effects on survival.
Your next steps as a scientist: Science is an ongoing process. What new question do you think should be investigated? What future data should be collected to answer your question?