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Drought stress application for smaller plants based on soil water holding capacity

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ABSTRACT

The method of applying drought stress in the small soil-filled containers for relatively small plants. The described method is working well for Arabidopsis, tomato and cowpea, but does require quite high time commitment in the manual setting, as it relies on daily monitoring of the pot weight and adjustment of the target weight by manual watering. Alternatively - the method can be also used for imposing the salt stress treatment - by growing the plants at 50% Water Holding Capacity, and subsequently immersing the pots in 200 mM NaCl / 0 mM NaCl for final treatment of 100 mM NaCl / Control treatment respectively.

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drought, water holding capacity, soil experiment, small plants, Arabidopsis , tomato, cowpea

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Material preparation

1 Prepare pots with the soil of choice for the experiment. Make sure that each pot is filled with approximately the same amount of soil and that the soil is pushed with approximately the same strength. Make sure to mark the pots with a unique ID for each pot.

2 Leave the filled pots overnight to air-dry

3 Measure the weight of each pot - this is going to be your DRY WEIGHT. We usually use a scale with 0.01 g precision level for this purpose

4 Soak the pots by placing them in a water-filled tray as well as spraying them with the spray bottle from the top. Leave the pots for soaking between 3-12 hours and fill the water in the tray if necessary. We usually leave the pots to soak up the water overnight.

5 Let the wet pots drain for 30 minutes on an absorbent surface - such as a paper towel - to get rid of excess water

6 Measure the weight of each pot - this is going to be your WET WEIGHT or pots at 100% Water Holding Capacity. We usually use a scale with 0.01 g precision level for this purpose

Seed germination

7 Treat the seeds of the species you intend to grow for this experiment as you require. For Arabidopsis - we usually put them at 4C for 24h prior to putting them into the soil.

8 Put the seeds into 100% Water Holding Capacity pots for germination

9 Let the plants germinate and grow until the desired target stage prior to imposing the drought stress. We usually germinate the plants for 2-3 weeks for Arabidopsis, 3 weeks for tomato and 2-3 weeks for cowpea prior to the imposition of drought stress.

Please note - depending on the species and their transpiration rate - it might take thevarious amount of time to reach desired water holding capacity.

Calculate target water holding capacity

10 Using the DRY and WET weight, measured in steps 3 and 6 respectively, we can now calculate the target weight of each pot for a desired Water Holding Capacity (WHC). Usually - we use 60% WHC for "Control treatment" and between 40% - 10% WHC for drought stress depending on the species, where Arabidopsis will show reduced growth rate already at 40% WHC, while more resilient species like cowpea will only show differences at 20% WHC, and wild resilient species such as wild tomato (Solanum pimpinellifolium) will show differences at 10% WHC.

Also - take into account that some species will require to adjust WHC for control conditions as well.

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11 Calculate desired WHC for each pot by using the following formula:

TARGET WEIGHT = (((WET WEIGHT - DRY WEIGHT) / 100) * TARGET_WHC) + DRY WEIGHT

e.g. calculation for 60% WHC, with 145.45 g DRY WEIGHT and 279.31 g WET WEIGHT will be:

TARGET WEIGHT = (((279.31 - 145.45) / 100) * 60) + 145.45 = 225.766 g

Make sure to calculate TARGET WEIGHT for each individual pot separately - as the differences in DRY and WET WEIGHT will result in slightly different target weights for desired water holding capacity

Start drought treatment

12 Once your plants reached a desired developmental stage, withhold the watering until the desired target weight is reached. For Arabidopsis, it usually takes 1-2 days to reach the desired target weight, while cowpea and tomato can reach their target weight within 1 day.

13 Maintain the drought/control treatment by weighing the pots every day and adding the water to reach the TARGET weight.

If you are interested in evapotranspiration between your treatments/genotypes - you might also want to record the pot weight before and after watering - and track the plant water use and evaporation by calculating the difference between pot weight after watering and pot weight before watering on the next day.

Please note - as your plants get bigger, the plant weight is becoming a factor in the above calculations that can no longer be ignored. Therefore the above method is only applicable for relatively young seedlings of tomato / cowpea (up to 4-5 weeks old), and longer period of time for Arabidopsis (up to 8 weeks old).

14 Throughout the drought exposure, you can monitor the plant growth, photosynthetic efficiency and other physiological parameters.

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