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We use this protocol and it's working

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Transplanting Arabidopsis

Forked from Transplanting Arabidopsis

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ABSTRACT

Transplanting Arabidopsis grown from seed on agar plates to soil trays.

IMAGE ATTRIBUTION

Meghan Burns

GUIDELINES

Growth Conditions for Arabidopsis

Rivero L, Scholl R, Holomuzki N, Crist D, Grotewold E, Brkljacic J. Handling Arabidopsis plants: growth, preservation of seeds, transformation, and genetic crosses. Methods Mol Biol. 2014;1062:3-25. doi: 10.1007/978-1-62703-580-4_1. PMID: 24057358.

Maturity Stages

Plants produce their first flowers within 4–5 weeks, and seeds can be harvested 8–10 weeks after planting.

Lighting

Optimum light intensity is 120– 150 µmol/m 2 s. Higher intensities may result in death of some seedlings, but are tolerated by older plants; purpling of leaves is the first symptom of high-light stress. Very low light intensities may result in weak and chlorotic plants. Arabidopsis is a facultative Luz Rivero et al. 21 long-day plant. Plants flower rapidly under continuous light or long-day (>12 h) photoperiods, while under short days (<12 h), flowering is delayed, favoring vegetative growth. Plants grow well under a cycle of 16-h light/8-h dark or under continuous light.

Temperature

Optimum temperature is 22–23 °C. Night temperatures should be maintained 2–4 °C lower than the day temperature. The temperature range for Arabidopsis growth is 16–25 °C. Lower temperatures are permissible, but higher temperatures are not recommended, especially for germination through early rosette development. Temperatures above 28 ºC are better tolerated by more mature plants (past early

rosette stage). In general, high temperatures result in a reduced number of leaves, flowers, and seeds. At lower temperatures, growth is slow, favoring the vegetative phase, and flowering is delayed.

Humidity
Plants tolerate low (20–30 %) relative humidity well, but depletion of soil moisture may occur in these conditions. Plant sterility may result from very high (>90 %) relative humidity. Mild humidity (50–60 %) is considered optimal for plant growth; however, low humidity (<50 %) is recommended for silique maturation.

Watering
Water from the bottom up once or twice a week until soil is saturated. Pour off any remaining standing water.
Plants should not be overwatered to avoid development of algae, fungi, fungus gnat larvae, and other pests who thrive on overly wet soil. Algae can be manually scraped off and the soil allowed to dry.

Fertilizer
Osmocote ® 14-14-14 (14 % nitrogen, 14 % phosphate, 14 % potassium) is an extended time-release fertilizer, feeding up to 3 months from planting. Apply in amounts according to the label.

Vernalization (Species Dependent, A. thaliana flowers successfully without vernalization)
Some winter-annual natural accessions require a period of cold to initiate flowering, a process known as vernalization (e.g., Galdo-1, Monte-1, Cit-0, Dog-4, Istisu-1, Valsi-1, Mir0, Tamm-2). Young rosettes (2–4 weeks old) of late flowering accessions should be placed at 4 °C for 4–7 weeks to accelerate flowering.

MATERIALS

- Tweezers/Forceps (Fisher Scientific 10-270)
- Small dish filled with distilled water
- Soil, Berger BM6 HP Myco Organic Fertilized Peat Moss Mix (Hummert International 10121000)
- Tray insert 3.5" square, 18 cell (True Leaf Market TRAY INSERT - 18 CELL)
- Propogation tray, 20"L x 10"W, no holes (Better Grow Hydro 5630)
- Plastic plant label stakes (A.M. Leonard VP-PS6)
- Sharpie (Fisher Scientific 19-166-600) or laser printable labels for identifying pots (Order information varies based on printer)

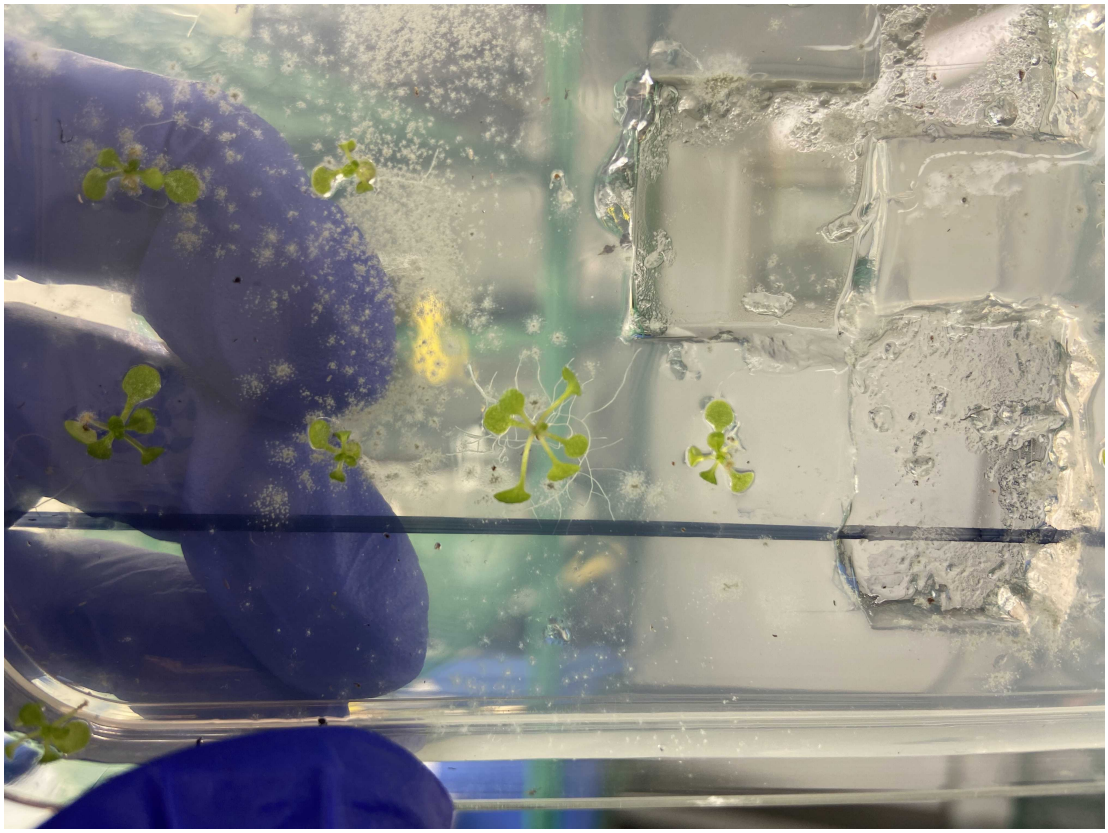
BEFORE START INSTRUCTIONS

Seedlings will be ready to transplant in approximately 10 to 14 days after planting when they've formed one to two true leaves. Do not wait too long to transplant. The root system begins to develop rapidly after the first true leaves. Damaging roots during transplant will drastically affect development and result in non-uniform plants.

If planting transgenic plants, review all USDA-APHIS requirements for the RIPE APHIS permit for handling transgenic materials before beginning any experiment.

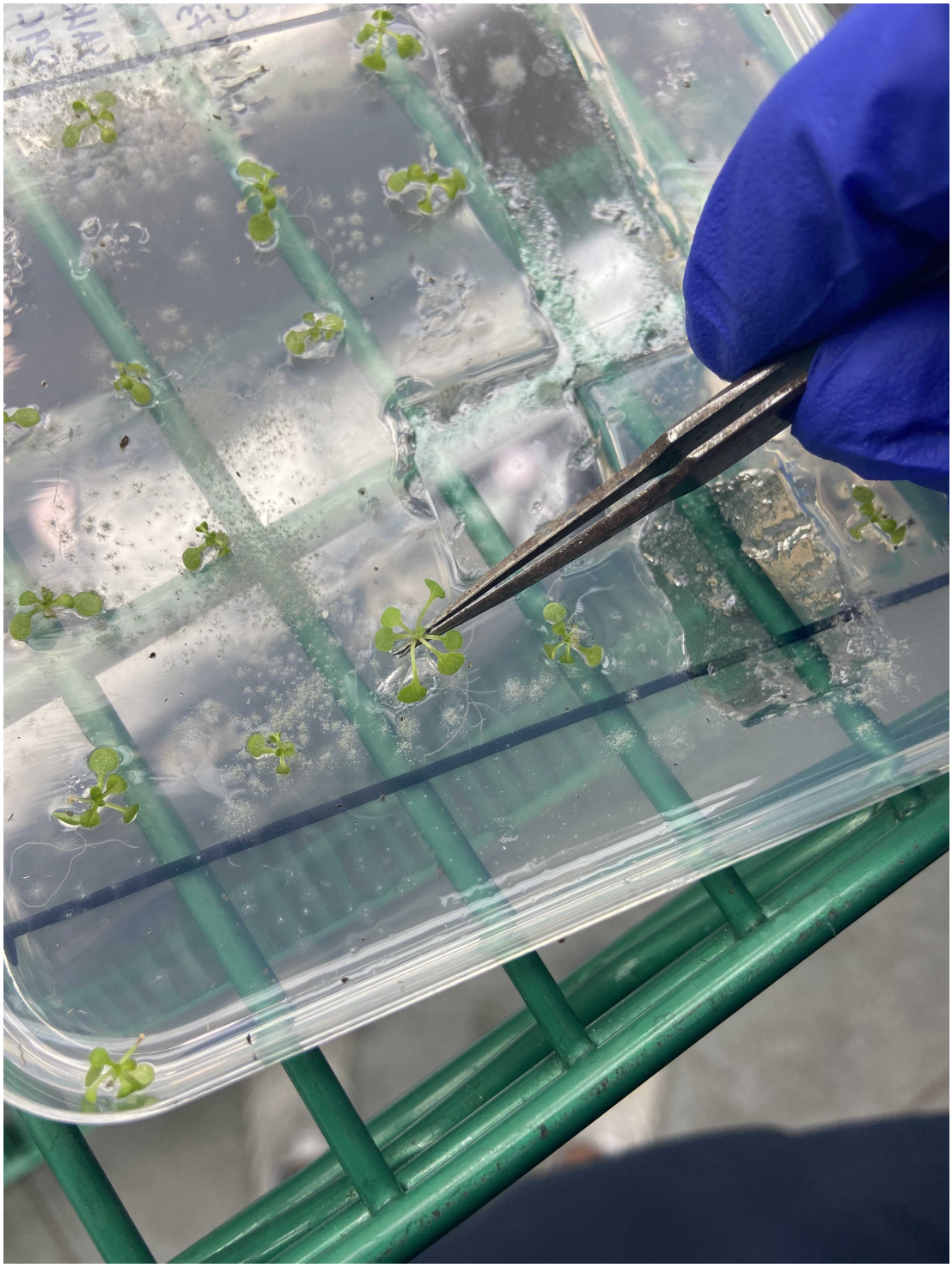
1 Take soil and place it into the trays. Fill about 3/4 of the way, press down gently, and then add water to the top. Fill completely with soil and add more water to the top. When complete it should look like the image below.

2 Look at the Arabidopsis on the agar plate and determine which one looks the best. Typically this means it is the plant with the longest roots and greenest leaves.

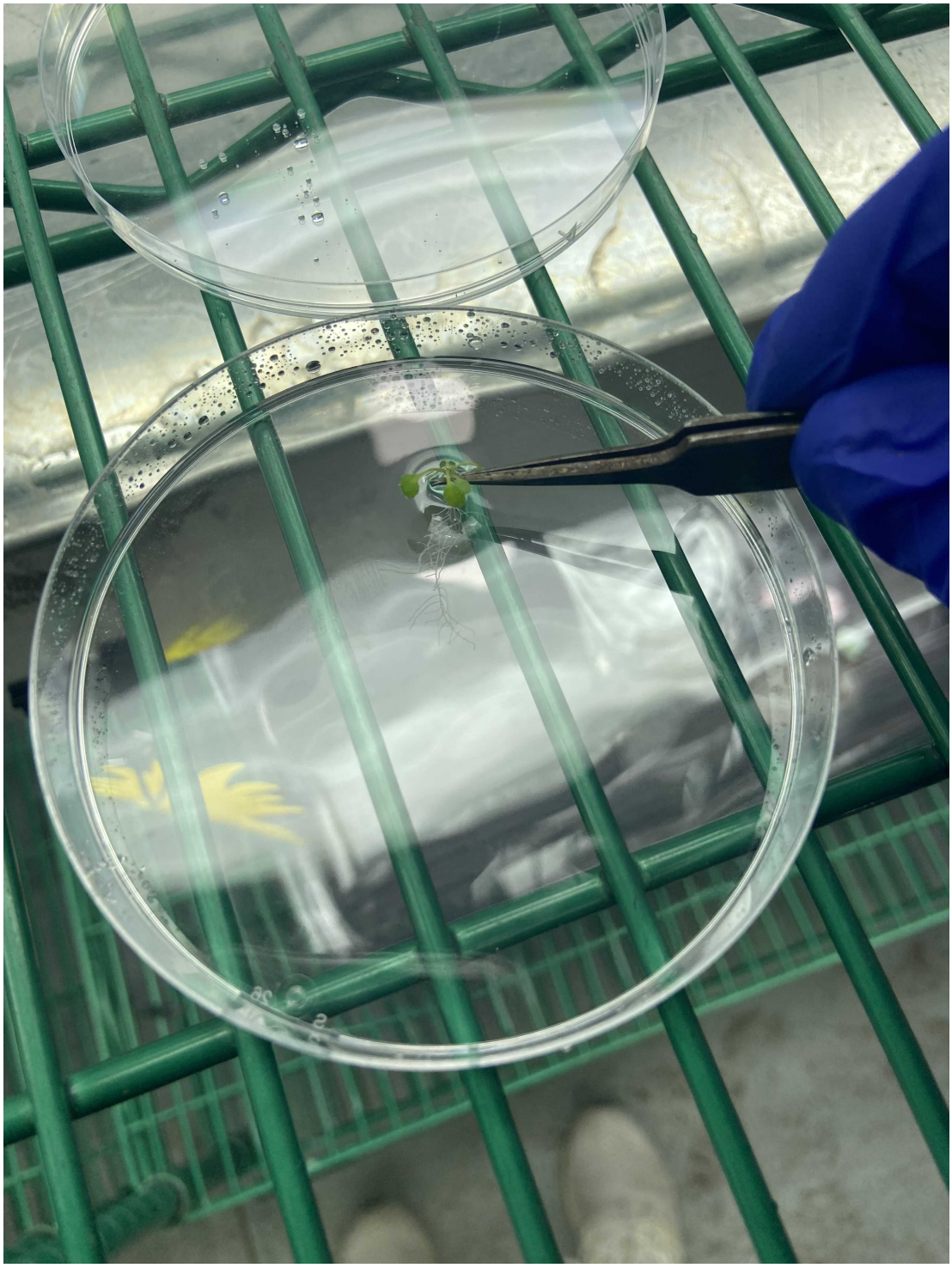


Ideal plant is the plant in the middle.

3 Take a pair of tweezers and place them at the base of the plant. Pull up very slowly, ensuring that all of the roots come out of the agar in one piece.



4 Once the plant is out of the agar, rinse it in a Petrie dish filled with distilled water to get all of the



agar off.

5 Take the pair of tweezers and grab the plant by the base of the root and stick the roots down into the dirt of the new tray. Make sure the roots are all the way down and the leaves are above the

dirt.

6 Pat down the dirt around the leaves and water the tray from the bottom up.

7 Label the tag with the plant and plate number and the genotype.


CS 737
Plot 3 Plot 4