Tissue Culture Echinacea Best Practices for Success

Echinacea from tissue culture are not difficult to establish if certain protocols are followed. The critical factor to understand is that these have been grown in a lab under high humidity, low light and warm temperatures. Lower humidity, higher light levels and temperature extremes should be avoided until the TC has been acclimated.









ARRIVAL & HANDLING PRIOR TO OPENING CONTAINERS

- Open box in a cool room or office. Do not open in a warm or sunny/lighted greenhouse.
- Be aware of the temperature inside the box upon arrival. If it is warm (i.e. above 75°F) it is beneficial to remove the containers and place in a cooler at 65°F to slowly cool before opening.
- TC should be planted immediately upon arrival. However, if the TC is not going to be planted immediately (i.e., same day), remove the vessels from the box and store the <u>unopened</u> containers in a lighted (<800 f.c.) room at 65-75°F. A germination or growth chamber with higher humidity is ideal. DO NOT PLACE IN A COOLER OR IN AN AREA WITH EXCESSIVELY LOW HUMIDITY (<50%).

PROPAGATION HOUSE PREPARATION

- Benches and surrounding area should be sanitized with a quaternary ammonia or hydrogen peroxide product.
- 'Tents' should be set up on the benches. The purpose of the tents is to maintain very high (95%) humidity and reduce the light intensity to 200-300 f.c.
 - Domes can be constructed using lightweight tubing, or rectangular frames can be easily put together using PVC tubing and connectors.
 - Enclose the tents with plastic sheeting. White greenhouse poly is ideal. Woven propagation cloth may also be used.
 - Shade cloth should then be placed over the tents or hung overhead.

TRANSPLANTING

- Open petri dishes or vials to be planted in a controlled environment. Ideal environment would be 65-75°F and very humid, on the verge of foggy.
- Remove plantlets and rinse the nutrient agar from the roots (this makes it easier for planting).
- Grade the plantlets for larger with longer roots and smaller with shorter roots. Plant accordingly for more uniform trays. *There may be a small percentage with no roots. These normally do not root and the number should be documented. These may be planted or discarded.
 - Plant into trays filled with a well-drained media, a pH of 5.8-6.2, and an EC<1.0. Controlled release fertilizers or high starter charge is not recommended. *Heavier media may benefit from loosening up with additional perlite.
 - 1. Water trays to a moisture level 2-2 ½ (slightly moistened, tray is still light and media is not settled).

2. Using a small plastic spatula or similar tool, hold plants by the leaves and gently press the roots into the media, being careful not to damage. (Lift media slightly first if media is too firm.) *Be sure the crown is above the media surface and free of any particles.

3. Water plantlets in as soon as possible to a moisture level 4. Avoid saturation as this can slow root development.









PLACE TRAYS IN PROPAGATION HOUSE

***Photoperiod**: Echinacea require long days to become established and actively grow. However, long days also triggers flowering. Darwin Perennials recommends that Sombrero and Double Scoop Echinacea be provided with a 14 hour photoperiod. (A longer photoperiod may be used, but not shorter than 14 hours.) This is enough to provide growth and bulking, and short enough to slow flower bud development. It is recommended any flower buds that do develop in the liner stage be removed.

- Transport planted trays to propagation area.
 - Avoid direct sunlight.
 - Avoid strong airflow and low humidity.
 - Place trays inside tents and lower the sides to contain humidity.
 - Ideal media temperature is 72-78°F
 - Ideal light level is 40-60 umol.
 - Ideal humidity is 95%
 - Some air exchange is beneficial, such as through the benchtop, but not air flow.
- Mist as necessary to maintain high humidity. Monitor media moisture to maintain a constant level 4 to encourage root development.

ACCLIMATION

After 5-7 days Echinacea begin to acclimate and the leaves will stand up. Media moisture can be allowed to dry to a 3 before watering to a 4.

- Gradually reduce humidity and increase light levels by opening up the sides of the tents.
- By day 10 plants will be rooting in. Media moisture and humidity can be further reduced. Light levels can be increased to 80-100 umol. DLI of 4 mol. *Photoperiod should be extended to 14 hours when natural days are shorter.
- Liquid feed program can begin at 50ppm-75ppm N by day 14.
- Increase DLI to 8-10 mol by day 21.

** The use of Configure PGR has been found to be effective for bulking Echinacea from tissue culture. One spray application of 300ppm can be used around week 8, once liners are well established.

** To control stretch in the liner, Daminozide (B-9), or Daminozide/Chlormequat (Cycocel) may be used. Always trial first!

MOISTURE SCALE

5=Saturated. Media is wet and drains freely. Water is easily displaced with a light touch. **4=Medium Wet.** Media is glistening but water is not draining freely. Water is displaced slightly with a squeeze.

3=Medium Dry. Media is not dark black and glistening. On the verge to changing to light brown. No water is easily displaced when squeezed but moisture can be felt.

2=Dry. Media has changed to light brown. No moisture can be felt.

1=Very Dry. Media is light brown and may be pulling away from the sides of the container. Plant is wilting.