

We germinate achenes using a pregermination and germination protocol that uniformly breaks dormancy, synchronizes emergence, and germinates almost every viable seed (Feghahati and Reese 1994). We follow the published methodology except we use no fungicide and use different containers.

### **Pregermination treatment (14 days)**

Put seeds on blotters<sup>1</sup> in petri dishes<sup>2</sup>. Dampen blotter thoroughly with Florel solution<sup>3</sup>.

Put in refrigerator (4 degrees C) with 24 h/day low levels of light ( $4.2 \times 10^{-6} \text{ mol m}^{-2} \text{ s}^{-1}$  PAR) from fluorescent tubes. In our lab, this level of light occurs in the glass door fridge when the inside and overhead lights are on. Maintain moisture levels with Florel solution<sup>3</sup>.

### **Germination treatment (14 days or more)**

Water dishes and put in incubator (25 degrees C) with 16h/day high light ( $1.65 \times 10^{-4} \text{ mol m}^{-2} \text{ s}^{-1}$  PAR). Maintain moisture level with water (or florel solution).

Most sprouts emerge in about 24h. After daily germination tapers off (~5 days) we moisten with the Florel solution instead of water. We get fungus on the seed coats, it doesn't seem to hamper germination (fungicide does though).

### **Planting Sprouts**

We check for sprouts daily and plant a sprout in a plug tray<sup>4</sup> when its radicle emerges 1 mm.

-----

1 Anchor blotters: Anchor Steel Blue Seed Germination Blotter (SDB2)  
<http://www.seedpaper.com/seedpapers.htm>

2 Falcon petri dishes, 60 x 15 mm, disposable polystyrene, Falcon Brand with Falcon stock #1007.

3 Florel solution: 365  $\mu\text{l}$  Florel / 100 ml H<sub>2</sub>O

4 288 plug trays square deep (Landmark plastic, Akron, OH # P-288SQD)  
[http://www.landmarkplastic.com/product\\_details.asp?kar=&id=61](http://www.landmarkplastic.com/product_details.asp?kar=&id=61)

Feghahati, S. M. J., and R. N. Reese. 1994. Ethylene-, light-, and prechill-enhanced germination of *Echinacea angustifolia* seeds. *Journal of the American Society of Horticultural Science* **119**:853-858.