

Preparation of packaged medium from the Orchid Seedbank

Unless otherwise labeled, all packets are for a final volume of 1 liter of medium. Media are tested and prepared here with distilled water; we do not guarantee gelling if used with tap water.

To prepare medium:

- 1) Pour ~800 mL of distilled water into a container that will hold at least 1 liter at final volume. 1-liter Erlenmeyer flasks are ideal for this. Other containers will work as well.
- 2) Add the contents of the packet. Media which contain charcoal will be dark; this material is hard to clean out of clothes and porous surfaces. Although not toxic, try to avoid making unnecessary amounts of dust when pouring.
- 3) Stir until components are dissolved. Break up any large chunks. Not all ingredients are soluble; agar dissolves only in hot water. You may heat the water if desired, but it is safer to pour cold medium.
- 4) Add distilled water to a final volume of 1 liter. If you add too much water, the medium may not gel well. Not enough water will result in a medium that is more concentrated, and not all plants will do well with this.
- 5) Dispense medium into containers. Stir or swirl container between pouring to ensure agar is evenly distributed. If your finished medium has some containers in which the medium is too hard and some that are too soft (or liquid) when cool, you are not distributing the agar evenly.
- 6) Cap your containers. Place containers in pressure cooker or autoclave.
- 7) Autoclave at 15 psig (121-123 C) for the desired period of time. We use 22 minutes.
- 8) Allow autoclave to cool to where it is cool enough to touch. Remove containers while warm; containers should cool in a reasonably clean area. As containers cool, they pull in air, which may increase the risk of contamination. Swirl containers before they gel to ensure distribution of charcoal.

Once containers have cooled, they can rest for several days (to check for contamination, ensuring the autoclaving was performed correctly), or be used immediately if you are certain your techniques are correct.