

Protocol for Painting Gametophyte Biomass onto Spools

1. Ensure your work area is clean and sterilize any surfaces that will be used with a generous amount of ethanol.
2. Gather supplies:
 - ☐ Gloves
 - ☐ Ethanol spray bottle
 - ☐ Paper towels or kimwipes
 - ☐ Pasteurized seawater
 - ☐ Small beaker
 - ☐ Large artists' paint brush (about 1 inch wide)
 - ☐ Wound seed spool (wound with Langman seedstring)
 - ☐ Tray
 - ☐ Pre ground and weighed gametophyte mix
 - ☐ Sharpie
 - ☐ Timer
3. Note: The gametophytes you are using for seeding should have been ground 24-48 hours before painting. Seed at a density of 0.1g per 100ft.
4. Ensure all supplies are sterile.
 - Spray ethanol on the surface of the tray, wipe with a paper towel, and allow it to air dry.
 - Soak paint brushes in ethanol for 2 minutes, shake them off, rinse with seawater, and then swirl and soak them in seawater.
 - Sterilize the small beaker and rinse with seawater.
5. Set up your space by getting a tray, a wound spool, and a paintbrush.
 - The tray will collect excess water while you are painting so it should be cleaned prior to using.
6. Place the spool on the tray.
7. Pour your pre-weighed biomass into the small beaker.
 - GreenWave uses 50mL falcon tubes to separate out the biomass for each spool the day before painting.
8. Top off the beaker with sterile seawater to 80mL total.
9. Swirl the beaker to suspend the biomass.
 - Do this occasionally throughout the painting process to help ensure even coverage.
10. Dip the clean brush into the mixture and begin painting the spool downward from the top.
 - You should paint all the way around the spool in ~4 inch sections moving downward. This way, if you run out of biomass for some reason the top section of the spool will still be usable.
11. Use the brush to paint all of the biomass onto the spool in an even coat.
 - The string on the spool should absorb the majority of the 80mL of water.
 - If you notice the string is consistently not absorbing all of the water, adjust the volume. The volume absorbed will depend on how tightly your spools were wound and may vary.
12. Once all of the gametophyte mixture is on your spool, look over the spool to make sure they are evenly distributed. Use the brush to move the biomass around if needed.
13. Place the spool in a humid and chilled place, like your nursery, for 15 minutes.
14. After 15 minutes, gently place your spool into your grow tank.

15. Keep the spools in the dark with low water flow for 24 hours.
 - The tank should have as low water flow as possible during this time. If you are using chillers, try to limit the water flow as much as possible.
16. Treat the spools according to the Environmental Conditions Chart (page X).
17. Dump the water from the tray.
18. Clean the brush and beaker between gametophyte mixes.
 - You can use the same brush and beaker to paint multiple spools with the same gametophyte mix.

