Protocol to Grow Field Inoculum in the Greenhouse

Crust Bin/ General Greenhouse Setup

Materials

1020 trays with and without holes (greenhouse megastore is best place to purchase these)
Landscape fabric (weed cloth)
Sand (collected from the Moab dune)
Measuring cup
60% shade cloth

1. Cut the landscape fabric into rectangles large enough to line the bins with a little extra room. Fabric leftover from a previous growth cycle can be reused.
2. Sieve field collected sand through a 1 or 2 mm sieve to break up clumps and remove organics that can lead to contamination.
3. Set up the bins. From the bottom, the setup of the bins is: 1020 tray without holes, 1020 tray with holes, landscape fabric, 3 cups sieved sand. Inoculate the crusts (see inoculation protocol).
4. Use shade cloth to cover the crusts once the irrigation is set up and the bins are all inoculated.

Notes:

- Do not reuse sand from previous growth cycles because of possible contamination.

Irrigation Setup

Materials

Drip irrigation pressure regulator
½ inch PVC
½ inch irrigation line
¼ inch irrigation poly tubing
.5 gallon per hour pressure compensating emitters
Emitter hole punch
Goof plugs
Scissors
Zip ties
Silicone caulk
Various ½ inch irrigation and PVC fittings ie. elbows, couplings, Ts, end caps

1. Beginning at the valve, install the pressure regulator (along with a filter and backflow preventer, if needed in your system).
2. Install PVC next, until you reach a good starting point for your irrigation system.
3. Connect the ½ inch irrigation line to the PVC with an adapter and run it along the room. You will be running tubing from this main line to each bin, so arrange the line so that it is roughly equal distance from most bins. ½ inch irrigation fittings, such as ts and elbows, will allow you to split up the line and make this possible, as well as prevent kinks. Use end caps at the ends of your line.

Picture of the irrigation setup, showing 1/2 inch line with 1/4 inch tubing going into each bin.

4. Use the emitter hole punch to punch holes in the ½ inch line. There will be one hole for every bin. It's best that the hole is oriented towards the bin it corresponds with so that there is no tension on the emitter (tension = leaks).
5. Press the barbed side of the emitter into the hole. Repeat for all holes.
6. Connect the ¼ inch tubing to the emitter. Cut so that the tubing will reach the bin it corresponds with.
7. Insert the ¼ inch tubing between the two 1020 trays.
8. You may want to install a timer on the valve, although it helps to have someone present while watering in case a problem arises.

Notes:

- Use zip ties to help keep ½ inch line in place.
- You can move emitters if needed and plug the old emitter hole with a goof plug.
- Use silicone caulk to patch up any leaks.

Watering Protocol

1. Calculate how long you will need to run the irrigation to get the desired amount of water into your bins. We watered our crusts every 3 days, and wanted them to be completely dry by the next watering. This was achieved with around 600 mLs of water, or about 19 minutes of running the irrigation system (adjust according to flow rate of the emitter).
2. Turn the valve on and water for the calculated amount of time.
3. Turn the valve off.

Notes:

- If a bin did not get enough water for some reason, you can water manually using a plastic beaker. You can then either remove the entire top tray of the bin (carefully!!) and pour the water in the bottom tray, or just push the top tray of the bin to the side and pour the water in.

Inoculation Protocol

Materials

Inoculum
Water
Measuring spoon (1 tbs)
Plastic beaker

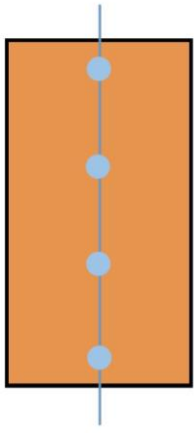
1. Sieve any inoculum to remove as much mineral soil as possible. Also remove large pieces of organic material.
2. Water the bins so the sand substrate is damp.
3. Measure out 4 tablespoons of sieved inoculum. Break up any chunks larger than pea size.
    a. We sued 4 tbs to achieve 20 – 35% cover of inoculum in our bins.
4. Add about 150 – 200 ml of water to the inoculum. Stir until you have a homogenous slurry.
5. Pour the slurry evenly across the surface of the bin. If there are globs of inoculum, you can gently spread them with the measuring spoon.

Chlorophyll A Collection Protocol

Materials

15 ml centrifuge tubes with ½ cm marked from opening
Coin envelopes

1. For each bin, you will take four samples along a transect and place them into the same envelope. Two samples should be taken near the edges of the bins and two should be in the middle.
2. To take soil samples, place centrifuge tube into the soil ½ cm. If the soil is damp, the sample should be easy to remove with the tube. If you are having trouble removing the sample, you can use a small piece of thin paperboard to scrape under the sample.
3. Deposit the sample from tube into a coin envelope.
    a. You may want to record level of development of the bin on the envelope.



Chlorophyll A sampling diagram. The blue line represents the transect and blue circles represent sampling areas.

4. Store samples in a cool, dark, and dry place until dry before chlorophyll a analysis.

Scraping Protocol

<u>Materials</u>

5 gallon buckets
Garden shovel

1. Let the bins dry completely before scraping.
2. To scrape, make an incision with the garden shovel across the length of the bin. Starting from this incision and moving towards the outside of the bin, scrape about ½ centimeter of soil from the top of the bin. Collect this in a 5 gallon bucket labeled as greenhouse inoculum. Repeat until you have removed all of the crust from the bin.
    a. If you have picked up some sand while scraping, you can hold the crust gently on the shovel and tilt, allowing the sand to flow out from under the crust. The goal here is to not collect any mineral soil in the inoculum bucket.
    b. Avoid areas in the bin where there doesn't seem to be any crust cover.
3. Dump any remaining sand into a 5 gallon bucket designated for waste.

Pictures of the crust scraping process.