Materials

Nylon membranes
- For fungal exclusion, use nylon membranes with a 0.45 μm pore size.
- Nylon membranes can be ordered through the institution's Fisher Scientific representative; they can help negotiate pricing. In 2014, the vendor that manufactured these membranes was Maine Manufacturing.
- The size of the roll can be custom ordered. Typically, we order 30 cm x 3 m rolls.
- These are custom ordered and take up to 5 weeks to be shipped.

Fabric glue
- This can be purchased in many different stores (ie. hardware stores, craft stores, Walmart, Amazon). The brand Tear Mender works well and dries almost instantly. It also holds up in hot, humid environments.

Duct tape
- Provides a final seal on the cages.

Metal tags, safety pins
- Used to label the cages. Tags can be ordered from Forestry suppliers and attached via a safety pin.

Chicken wire
- Used to protect cages in the field. This can be found at any hardware store.

Stakes
- Used to keep chicken wire and cages in place.

Methods

1. Cut nylon membranes to appropriate size. Typically, cages are 10 cm x 10 cm. Therefore, cages are cut to be 10 cm by 20 cm and folded in half. However, this may vary. Use gloves when handling the nylon membranes. Dirt or oil on your hands may affect the pores on these membranes.

2. Use fabric glue to close two edges. Leave one edge open so that litter or soil can be placed inside. Apply duct tape on these edges after the glue has dried.

3. If performing a litter decomposition experiment: Place appropriate amount of dried litter in the microbial cage.

4. Samples can now be sterilized via gamma irradiation. Sterilize bags and contents with at least 22 kGy gamma irradiation. Discuss with your facility how to appropriately package the cages, and if bags need to be rotated during this process. At UC Irvine, a maximum of 15 cages can be placed in one Ziplock bag to be gamma irradiated. There cannot be metal in the machine during gamma irradiation so do not put metal tags on the cages prior to this step.
5. A subset of cages and/or contents can be checked to ensure proper sterilization. But, at this point Ziplock bags should not be opened to keep cages sterile until they are ready to be used.

6. For litter: cages can now be inoculated and the last edge can be closed with glue and duct tape. For soil: soil can now be added to the cages and the last edge can be closed. For fungal inoculum: add 1 x 1 cm square of agar or X amount of liquid culture (make sure the inoculum is consistent across all samples, i.e. spores/ml) and the last edge can be closed.

7. Attach metal labels and record appropriately (see image below for final cage).

8. Depending on the downstream analyses the mass of each cage should be recorded after all contents are added.

9. In the field, protect cages in chicken wire to avoid destruction by insects and animals. Use stakes to fix wire and cages to the ground. When deploying cages, continue to use gloves to make sure the pores do not get blocked with dirt and oil.

10. After collection, be sure to use proper sterile technique when destructively sampling each cage.

Notes

Consider the ecosystem the microbial cages will be deployed in and how long they will be deployed for. This could affect the amount of litter placed inside and what steps you want to take to make sure the cages stay where they need to.
References

