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# Fungal Extraction from Beetle Galleries

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1 Works for me

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Bark Beetle Mycobiome Research Coordination Network

ABSTRACT

This protocol describes how to extract fungi from beetle galleries.

This protocol is part of the Bark Beetle Mycobiome (BBM) Research Coordination Network. For more information on the BBM international network: Hulcr J, Barnes I, De Beer ZW, Duong TA, Gazis R, Johnson AJ, Jusino MA, Kasson MT, Li Y, Lynch S, Mayers C, Musvuugwa T, Roets F, Seltmann KC, Six D, Vanderpool D, & Villari C. 2020. Bark beetle mycobiome: collaboratively defined research priorities on a widespread insect-fungus symbiosis. Symbiosis 81: 101–113 <a href="https://doi.org/10.1007/s13199-020-00686-9">https://doi.org/10.1007/s13199-020-00686-9</a>.

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# **Materials**

### (all autoclaved)

- eppendorf vials
- Pestles or some other grinding tool
- scalpel
- Reserve hood time

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- strong forceps
- vials
- ethanol in petri dish for dipping the forceps & scalpel into
- vial rack
- (chisel)
- (pruner)
- pipette
- 1000 uL pipette tips
- EtOH bath

For field sampling of beetle associated fungi:

- ethanol
- squirt bottle (or separate vials with ethanol)
- hard forceps
- scalpel
- scalpel blades
- lighter (for forceps & scalpel sterilization)
- ziploc bags
- permanent markers (alcohol-resistant)

#### **Procedure**

Scrape piece of 1 gallery in the hood, scrape into

- a. 1 ml of 1X PBS AUTOCLAVED phosphate buffer saline solution in tube
- 3. mortar & pestle
- 4. plate directly, refrigeration decreases growing success
- 5. Plate three different dilutions, refer to the dilution protocol.