



Feb 07, 2020

Collecting Duckweed Samples For Microbial Community Profiling

PLOS One

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dx.doi.org/10.17504/protocols.io.98zh9x6



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EXTERNAL LINK

https://doi.org/10.1371/journal.pone.0228560

THIS PROTOCOL ACCOMPANIES THE FOLLOWING PUBLICATION

Acosta K, Xu J, Gilbert S, Denison E, Brinkman T, Lebeis S, Lam E (2020) Duckweed hosts a taxonomically similar bacterial assemblage as the terrestrial leaf microbiome. PLoS ONE 15(2): e0228560. doi: 10.1371/journal.pone.0228560

ATTACHMENTS

S1_Protocol.pdf

MATERIALS

| NAME ~ | CATALOG # | VENDOR ~ |
|---|-----------|-----------------|
| Miracloth | 475855 | Merck Millipore |
| Nalgene™ Rapid-Flow™ Sterile Disposable Filter Units with CN Membrane, 1000mL, 0.2μm pore | 127-0020 | Thermo Fisher |
| Sklar Sterile Disposable Scalpels | 06-3111 | |

Processing Water Samples

- Get 150 mL 0.2 micron filter unit. Lay sterilized miracloth on top of filter unit.
- Collect 150-300 mL of water sample and pour through sterilized Miracloth into filter unit.
- Filter water sample through 0.2 micron filter unit by turning vaccum on. 3
- Cut 0.2 micron filter from unit using sterile scalpel. 4
- Place into 5 mL centrifuge tube.
- Flash freeze and store at -80C until processing.

Processing Rinse Solution And Duckweed Tissue Samples

- Collect 1-3 grams of fresh weight Duckweed from environment sample. (See "Separating Duckweed From Other Solids" to obtain pure Duckweed tissue from environment.)
- Place into 50 mL falcon tube.

- 9 Wash tissue 2x's with sterile water.
- 10 Rinse tissue 3-5x's with 30-40 mL rinse solution. Filter rinse solution through 0.2 micron filter each time.
- 11 Filter a total of 100-150 mL of rinse solution through 0.2 micron filter.
- 12 Cut filter using sterile scalpel, transfer to 5 mL centrifuge tube, flash freeze, and store for 16S rRNA amplicon sequencing.
- 13 Wash rinsed Duckweed tissue 2x's with sterile water.
- 14 Transfer tissue to 5 mL centrifuge tube.
- 15 Flash freeze and store for 16S rRNA amplicon sequencing.

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