



Field sampling of root-associated microbes for DNA/RNA extraction

Version 2

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SoWa Anaerobic and Molecular Microbiology



ABSTRACT

This protocol describes a procedure for sampling plant roots in the field for future DNA and RNA extraction for microbiome analysis. The protocol is deliberately designed to be simple and requires no electronic equipment. Root samples are preserved in LifeGuard Soil Preservation Solution for protecting against nucleic acid degradation.

TAGS

dna

plant

Show tags

PROTOCOL STATUS

Working

We use this protocol in our group and it is working

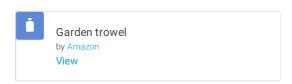
MATERIALS

NAME ~	CATALOG #	VENDOR \vee
Micro-spatula set	AT16.1	Carl Roth
LifeGuard Soil Preservation Solution	12868-100	Qiagen
Scissors	HCT7.1	Carl Roth
Technical-grade ethanol (70%)	T913.1	Carl Roth
Paper towels	Y03.1	Carl Roth
Microcentrifuge tubes 2 ml	CK06.1	Carl Roth
Garden trowel	View	Amazon
Disposable pasteur pipettes	EA61.1	Carl Roth
Tweezers set	PX40.1	Carl Roth
Cooling box	AA46.1	Carl Roth
Cooling packs	E447.1	Carl Roth
STEPS MATERIALS		
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BEFORE STARTING

- Clean spatulas using 70% ethanol
- Sample a triplicate of plant individuals spaced out a few metres apart from each otherMake sure you are reall sampling individual plants and not offshoots of the same plant.
- 9 Using a garden trowel, carefully dig out the plants while keeping the root system intact (as much as possible, of course).



3 While holding the plant by the shoot, shake the root system hard enough so that all loose soil is removed from it. Take care to damage the plant as little as possibleYou can use a spatula to remove large soil aggregates that are attached to the roots.

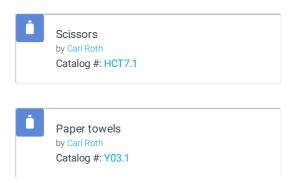


From the remaining root system (plus soil particles plus attached to the roots), trim a 'representative sample' of roots using scissors or scalpel. It is usually best to trim the roots onto a piece of paper towel.





5 Cut the trimmed out roots a little so that they fit into a 2.0 ml tube.



6 Place about 2-3 g of that cut out sample into a 2.0 ml tube.







NOTE

 $The \ root\ tissue\ should\ make\ up\ at\ least\ half\ or\ more\ of\ the\ mass,\ while\ the\ remaining\ attached\ soil\ should\ make\ up\ the\ rest$

7 Press the sample a little into the bottom of the tube to decrease its volume.

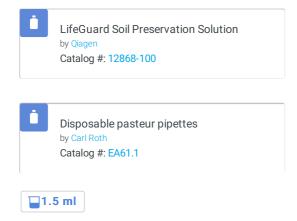


NOTE

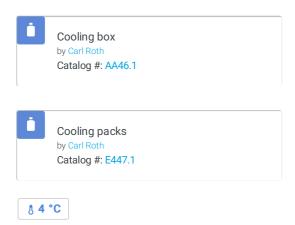
Make sure the roots do not take up more than 1/2-2/3 of the volume

It is of course possible to split each sample into several separate tubes, depending on the specific type of roots, and submerge each with LifeGuard solution

8 Add as much LifeGuard solution so that the sample is submerged in about twice of its volume (about 1.0 – 1.5 ml).
Best is to use a disposable Pasteur pipette for dispensing the solution.



9 Place the tubes in cooling (around 4 °C) and keep them cooled until you reach the lab. The solution will protect nucleic acids even at room temperature for several days, but cooling is preferred.



1) In the lab, store the samples in a freezer (-20 - -80 °C).

₹ -20 °C or -80 °C

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