



CrowdScience

Science Box

MICROBIOLOGY

Instruction

Table of contents

Introduction	2
List of box components.....	4
Safety precautions.....	5
Day 1 - Soil sample collection.....	6
Day 2 - Bacteria isolation from soil sample.....	7
Day 3 - Bacteria selection and transfer.....	10
Day 4 - Antibiotic producer search test.....	13
Day 5 - Results analysis.....	15

Day 1

Soil sample collection



1. Choose a location for sample collection.

Choosing the right location will increase your chances of finding unique producer bacteria. A forest edge, forest, meadow, cropland, or wooded area far from city center are the best options.

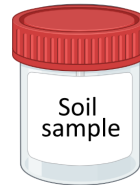
2. Put down the discription of the selected location:

- a) geographic location (GPS coordinates)
- b) relief type (mountains, plains and e.g.)
- c) relief form (ravine, hill, valley, slope and e.g.)
- d) history of the location (pasture, fire place, lake and e.g.)
- e) type of vegetation around (grassy, woody, desert and e.g.)

3. Put on a pair of gloves and take a a sterile container labeled "Soil Sample".

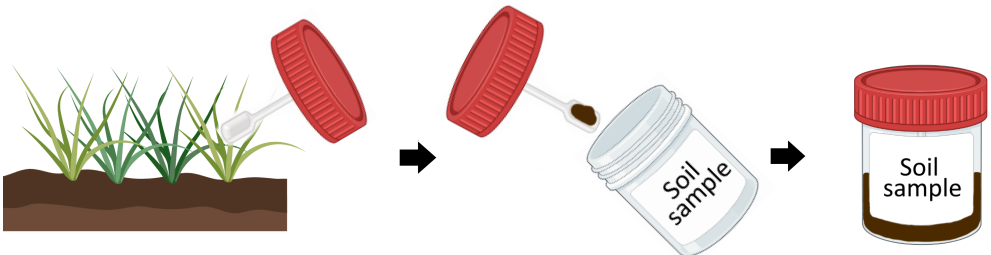


Pair of gloves



"Soil sample" sterile container

4. Collect soil to the container using a scoop. Make sure to fill the container up to the middle.



5. Go straight to Day 2 or store your sample in the refrigerator at +4C° before continuing the experiment.

Day 2

Bacteria isolation from soil sample

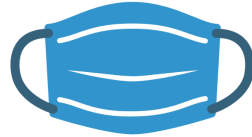
1. Put on a robe, mask and a pair of gloves.



A robe

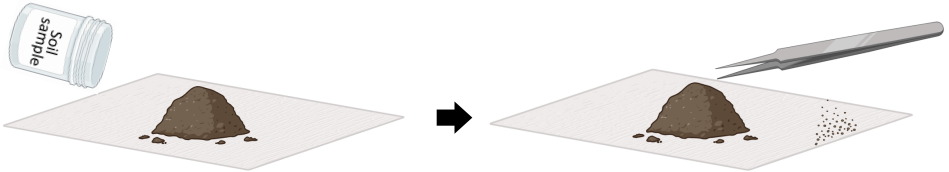


Pair of gloves



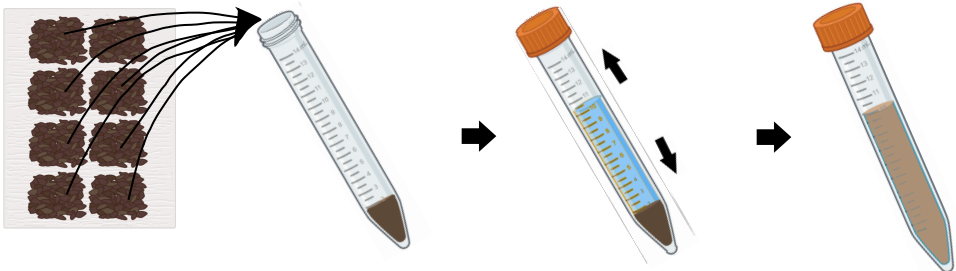
A mask

2. Take a clean sheet of paper or foil and pour the contents of the "Soil sample" container onto the prepared sheet.



3. Use tweezers to carefully remove foreign inclusions (roots, stones, twigs, broken glass, etc.) until your soil sample looks homogeneous.

4. Evenly distribute the soil sample on the surface of the sheet, divide it into 8 sections, take a pinch of the sample from each section and transfer them to the 15 ml test tube, signed as "Soil Suspension". Fill the test tube with soil up to the 2 ml mark.



5. Pour filtered tap water into the same test tube to the 10 ml mark. Tightly close the tube with the lid and shake it thoroughly, making sure the soil sample is evenly distributed throughout the water volume.

6. Pour the entire contents of the test tube into the container labeled "Dilution 1". Pour 10 ml of water into the "Soil suspension" test tube and repeat the procedure from **step 5** so that all soil residues are transferred into the container.

7. Carefully wipe one of the gloves you are wearing with an alcohol wipe.

8. Gently use your finger to further homogenize the suspension in "Dilution 1" container by rubbing the clumps of soil against the container walls.

! This helps to detach the bacteria from the
■ substrate and transfer them to the solution.

9. Pour filtered tap water into the "Dilution 1" container up to the 100 ml mark and mix it thoroughly.

10. Prepare a series of 10-fold dilutions:

a) in a 15 ml tube signed as "Dilution 2", pour 9 ml of water and add 1 ml of solution from the "Dilution 1" container up to the 10 ml mark;

b) screw the tube tightly and mix the contents thoroughly by turning the tube up and down 15 times;

c) similarly prepare dilution 3 from dilution 2, dilution 4 from dilution 3, and dilution 5 from dilution 4.