**Experiment II: Group sampling**

**Lab ID: Name:**

**10.05** Field Trip

Weight (grams) GPS coordinates

Sample A

Describe the location and surrounding environment.

Images of the sampling site:

Weight (grams) GPS coordinates

Sample B

Describe the location and surrounding environment.

Images of the sampling site:

**15.05** Data analysis

1. **Data analysis of the focal point sample A**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  | **CFU** | **Dilution number** | **Dilution factor** | **CFU/ml** | **CFU/**  **g soil** | **Log (CFU)/ g soil** | **% ABr** |
| R2 |  |  |  |  |  |  |  |
| R2-Amp |  |  |  |  |  |  |  |
| R2-Chl |  |  |  |  |  |  |  |
| R2-Cip |  |  |  |  |  |  |  |
| R2-Kan |  |  |  |  |  |  |  |
| R2-Rif |  |  |  |  |  |  |  |
| R2-Van |  |  |  |  |  |  |  |

Note: Choose the highest dilution plate that yields approximately 30 – 300 colonies to count the number of colony-forming units (CFU) and to calculate CFU/ml.

For your own samples:

1. Calculate the bacterial CFU/ml by dividing CFU by the dilution factor.
2. Calculate CFU per gram of soil weight by multiplying CFU/ml by 20 and dividing the value by the weight of the soil sample A.
3. Calculate the log-transformed value of CFU for per gram of soil sample.
4. Make a figure (Figure 1) comparing the bacterial counts (Log of CFU/ g soil) on R2 agar plate without antibiotic and the resistant bacterial counts on R2 agar with each type of antibiotics. (Log value to one decimal point)
5. Calculate % ABr (antibiotic resistance) by dividing [CFU/g soil on R2-antibiotic] by [CFU/g soil on R2].
6. Make a figure (Figure 2) showing the % ABr for each kind of antibiotics tested in this experiment.
7. **Data analysis of the away site sample B**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  | **CFU** | **Dilution number** | **Dilution factor** | **CFU/ml** | **CFU/ g soil** | **Log of CFU/ g soil** | **% ABr** |
| R2 |  |  |  |  |  |  |  |
| R2-Amp |  |  |  |  |  |  |  |
| R2-Chl |  |  |  |  |  |  |  |
| R2-Cip |  |  |  |  |  |  |  |
| R2-Kan |  |  |  |  |  |  |  |
| R2-Rif |  |  |  |  |  |  |  |
| R2-Van |  |  |  |  |  |  |  |

Note: Choose the highest dilution plate that yields between 30 – 300 colonies to count the number of CFU and to calculate bacteria CFU/ml.

For your own samples:

1. Calculate the bacterial CFU/ml by dividing CFU by the dilution factor.
2. Calculate CFU for per gram of soil weight by multiplying CFU/ml by 20 and dividing the value by the weight of the soil sample B.
3. Calculate the log-transformed value of CFU for per gram of soil sample.
4. Make a figure (Figure 3) comparing the bacterial counts (Log of CFU/ g soil) on R2 agar plate without antibiotic and the resistant bacterial counts on R2 agar with each type of antibiotics. (Log value to one decimal point)
5. Calculate % ABr (antibiotic resistance) by dividing [CFU/g soil on R2-antibiotic] by [CFU/g soil on R2]
6. Make a figure (Figure 4) showing the % ABr for each kind of antibiotics tested in this experiment.
7. **Compare the results between the focal point (sample A) and the away location (sample B).**

For your own samples:

1. Make a figure (Figure 5) comparing the bacterial counts (log CFU/g soil) on R2 agar plate without antibiotic and the resistant bacterial counts on R2 agar with each type of antibiotic between the two locations A and B.
2. Make a figure (Figure 6) showing the % ABr for each kind of antibiotic between the two locations A and B.

Submit your Labenote II without the group results and step (IV)

For the group samples:

1. Combine the data from all group members to create a figure (Figure 7) comparing the bacterial counts (Log CFU/g soil) on R2 agar plate without antibiotic and the resistant bacterial counts on R2 agar with each type of antibiotics between the two locations (Error bar should be incorporated in the figure).
2. Combine the data from all group members to create a figure (Figure 8) showing the % ABr for each kind of antibiotic between the two locations (Error bar should be included). What results can be inferred from the group data?
3. **Nominate two most interesting bacteria found in your group’s Experiment II.**
4. Discuss among the group members and propose two candidate bacteria for “The most interesting bacteria in the IP-ABr2023”.
5. Take images of the two candidate bacterial colonies.

**16.05** Submit Labnote II by 6 PM

**17.05**

08: 15-12:00 Preparation for presentation.

12:00-13:15 Lunch break

13:15- 14:30 Presentation

14:45- 16:00 Overall results of Experiment I

Vote for the most interesting bug for IP-ABr2023

Feedback