# Pre-Lab Questions

1. What are two ways that some prokaryotes are able to survive in extreme environments?
2. Describe three ways that microorganisms in soil contribute to the ecosystem as a whole.
3. Give two examples of genera that produce endospores, and explain why they are important in human health.
4. What are coliforms? Why are they important in assuring water quality? Why is *E. coli* the indicator bacterium for fecal contamination?

**Experiment 1: Bacterial Isolation from Soil Samples**

## Post-Lab Questions

1. Observe the two sets of plates. Determine the CFU/mL of each solution (Normal and Heated), remembering to use a plate that has 30 – 300 colonies for statistical reliability. Is there a difference in the CFU/mL counts for the two treatment conditions? If so, what do you think accounts for this difference?
2. How are the colonies from the two treatment conditions different (e.g., size, color, shape)?
3. What effect does the heating and cooling procedure have on the microorganisms present in the soil samples?
4. What is the source of the colonies in the heated samples?

**Experiment 2: Characterizing Bacteria Isolated from Soil**

## Data Tables

Table 1: Gram Staining Observations

| Sample | Colony 1 | Colony 2 | Colony 3 |
| --- | --- | --- | --- |
| “Normal” |  |  |  |
| “Heated” |  |  |  |

Table 2: Experiment 2b Catalase Assay Results

| Colony | Catalase Assay Results (“Normal”) | Catalase Assay Results (“Heated”) |
| --- | --- | --- |
| 1 |  |  |
| 2 |  |  |
| 3 |  |  |

## Post-Lab Questions

1. In which soil type would be more likely to find a higher proportion of Gram-positive microorganisms? Why? (If necessary, use a textbook or reputable online source to inform your answer).
2. What effect should heat treatment have on the proportion of endospore producing microbes?
3. What effect does heat treatment have on the number of isolated colonies?
4. What conditions favor endospore formation?
5. Which treatment condition produced more catalase-positive bacteria? What can explain the difference in the number of catalase-positive bacteria?
6. Write out the reaction that is catalyzed by the enzyme catalase. Why is this enzyme important?
7. What would you conclude if all samples you tested were catalase-negative?
8. What types of controls could you include in the experiment to ensure that all components of the system are working properly?
9. Why did you perform multiple differential tests on the same three colonies for the Normal and Heat treatments? If you were able to observe your samples under a microscope, what conclusions can you make about the types of microorganisms that make up the colonies you observed?

**Experiment 3: Winogradsky Column**

## Data Tables

Sample Location:

Table 3: Experiment 3 Winogradsky Column Results

| Week # | Observations (colors, layers, etc.) |
| --- | --- |
| 1 |  |
| 2 |  |
| 3 |  |
| 4 |  |
| 5 |  |
| 6 |  |
| 7 |  |
| 8 |  |
| 9 |  |
| 10 |  |

## Post-Lab Questions

1. Where does carbon come from in a Winogradsky column?
2. Why is carbon important to living organisms?
3. What purpose does calcium sulfate serve in the Winogradsky column?
4. Why is a hydrogen sulfide gradient generated from bottom to top in a Winogradsky column?
5. How is energy synthesis different between cyanobacteria (growing at the top of the column) and green and purple sulfur bacteria (growing near the bottom 1/3 of the column)?