**Protocol 1: The microbial flora of cheese**

**Introduction**

Microorganisms play an important role in the food industry. It is possible to observe certain elements of the microbial flora of certain foods when they are present in large numbers, for example in yogurt or cheese. In some cheeses, we can find more than 250 different species of microorganisms.

In this laboratory, you will be able to observe species grouped in two kingdoms of life: fungi and bacteria. To observe the latter, you must use the immersion method of observation.

**Objective**: Identify the morphological characteristics of fungi and bacteria.

**Preparatory questions**

1. Why is it important to clean the blade with an alcohol solution?

2. What is the advantage of using methylene blue instead of water?

3. What are the characteristics of microorganisms that make them particularly effective in growing on cheese?

**Safety**

* Never touch your face when working with microorganisms.
* Dispose of the sample and cover slip as directed by the teacher
* Wash the slide and the microscope stage thoroughly according to the teacher’s instructions
* Wash your hands thoroughly after the lab.

**Material**

* Microscope
* Slide and cover slip
* Adhesive paper or cotton swabs
* Alcohol
* Pliers
* Pipette
* Water or methylene blue (optional)
* Immersion oil
* Lens paper
* Lens cleaner
* 2 different types of bloomy rind cheese
* Image of yeast, mould, hull, streptococcus, staphylococcus, bacillus *(see presentation for example)*

**Method**

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| **Dry method — tape** | **Wet method — tape** |
| 1. Clean the slide with an alcohol-based solution
2. Let dry.
3. Cut a 1.5 to 2 cm piece of tape, avoiding touching the adhesive side.
4. Using the pliers, apply the adhesive side to the bloomed surface of the cheese.
5. Place the tape (sticky side towards the slide) on the slide.
6. Observe at highest magnification.
7. Repeat the steps for the second type of cheese.
 | 1. Clean the blade with an alcohol-based solution.
2. Let dry.
3. Place a drop of dye or water on the slide.
4. Cut a 1.5 to 2 cm piece of tape, avoiding touching the adhesive side.
5. Using the pliers, apply the adhesive side to the bloomed surface of the cheese.
6. Place the tape (sticky side towards the slide) on the slide.
7. Let dry for 2 minutes.
8. Observe at highest magnification.
9. Repeat the steps for the second type of cheese.
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| **Dry method — smear** | **Wet method — smear** |
| 1. Clean the slide with an alcohol-based solution
2. Let dry.
3. Rub the cheese with a cotton swab.
4. Spread the sample on the slide.
5. Cover with a coverslip.
6. Observe at highest magnification.
7. Repeat the steps for the second type of cheese.
 | 1. Clean the slide with an alcohol-based solution
2. Let dry.
3. Rub the cheese with a cotton swab.
4. Spread the sample on the slide.
5. Add a drop of food coloring or water.
6. Cover with a coverslip.
7. Observe at highest magnification.
8. Repeat the steps for the second type of cheese.
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**Observation**

1. Make a biological drawing of each of the organisms you observe.
2. Identify the cellular structures of the organisms observed.
3. Estimate the number of each of these microorganisms.

**Analysis**

1. Using the illustrations of the organisms used in the cheese industry, identify the organisms observed as accurately as possible. Justify your findings based on the specific characteristics of each taxon.
2. Why is it important to understand and control the cheese ecosystem? Give at least two specific examples.
3. What are the growth conditions that allow these types of microorganisms to develop on cheese?
4. In order to ensure consumer safety, the cheesemaker must have a good understanding of the ecosystem of each of the different types of cheese they make. What are the procedures, techniques or principles to be carried out in the manufacture of cheese that ensure the safety of consumers? Give two different procedures and explain the role and importance of each of them in establishing the specific cheese ecosystem.