Isolation and growing of microorganisms on pure culture

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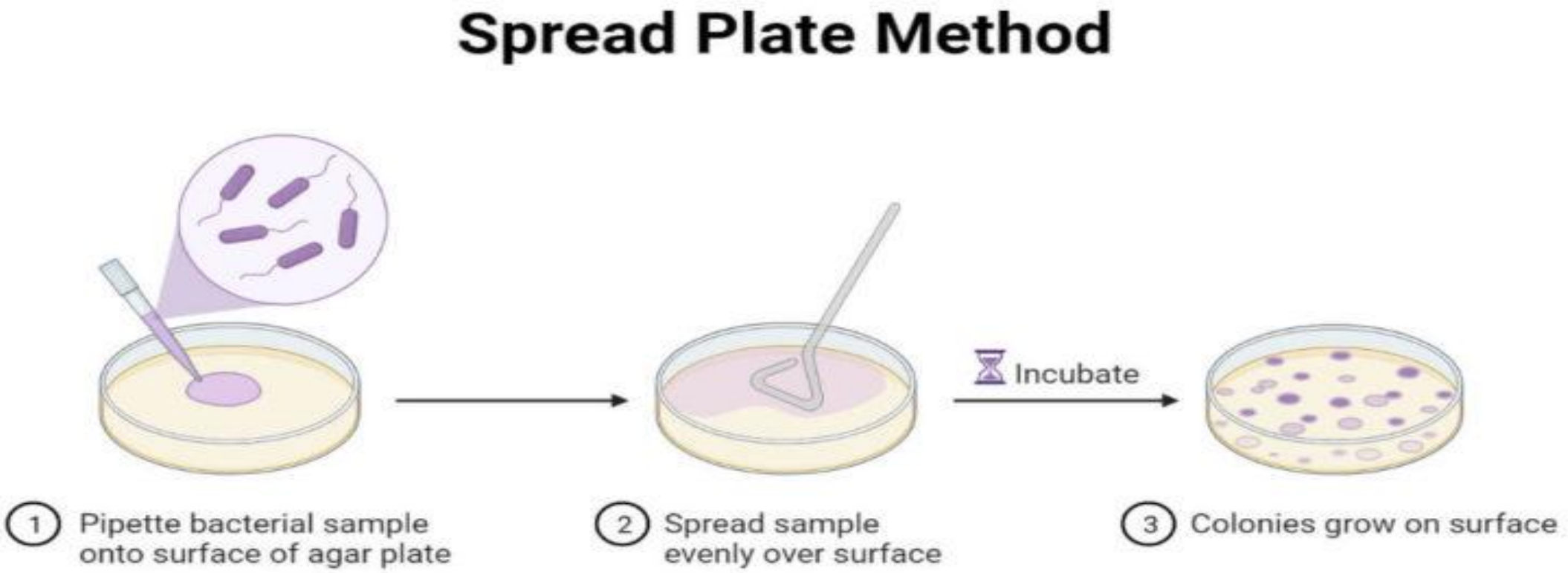
- When bacteria or any other microorganism is grown on a food medium in the laboratory, it is then called a microbial culture which is defined as bacteria or microorganisms growing on a specific culture medium.

- Different types of bacteria growing on the same food medium may appear completely different, so it is very important to know the external appearance or characteristics of the cultures to distinguish the different types, which then helps in diagnosing the type. Therefore, a pure culture must be obtained in order to accurately determine its characteristics before studying and determining the characteristics of the culture.

- Pure culture When it consists of a group of cells all derived from a single cell, the culture is considered purer one.

- There are many methods used to isolate and grow microorganisms purely on culture media in the laboratory:-

1) spread plate method



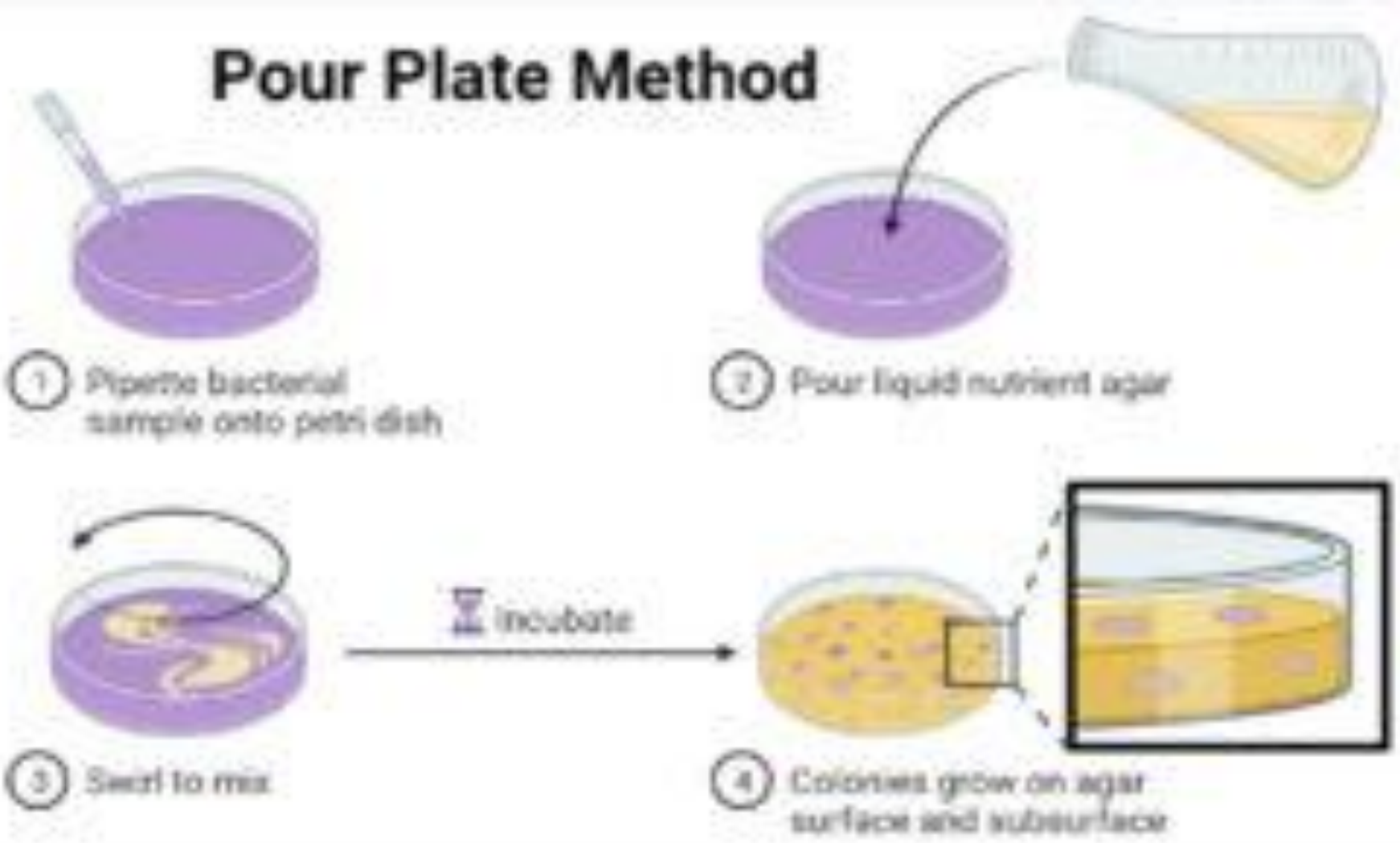
Advantages of Spread Plate Technique

- It is a simple, easy, and quick method of culturing microorganisms.
- A very low microbial load can be detected.
- The colony morphology of a microorganism can be studied by this method. The size of colonies produced when cultured by this method will be larger than those produced by the sample species using the pour plate method.
- It is a qualitative as well as quantitative isolation method which facilitates isolation as well as an enumeration (i.e. calculation of CFU/mL) of microorganisms.
- It is the most appropriate method for culturing the aerobic microorganism.
- There is very least chance of contamination if one used sterile glass beads as a spreader. This is because all the action will be done by closing the lid of the culture plate.

Disadvantages of Spread Plate Technique

- It requires extra tools like a spreader.
- It needs the sample to be in liquid or suspension form and needs to be serially diluted, hence; it is a little complex process.
- Solid or semisolid samples must be suspended prior to inoculation. It is very difficult if the sample is not easily soluble.
- It doesn’t support sufficient growth of Microaerophiles and anaerobes.
- It is unsuitable if the microbial load in the sample is too high. So, the sample must be serially diluted to reduce the microbial load at 20 – 300 CFU/mL. To get this dilution range, we may even need to do pilot testing.
- There is a chance of gouging the media during spreading, especially while using glass rod by one with not enough skill, and if the media is not properly solidified.

What is Pour Plate Method?



Advantages And Disadvantages

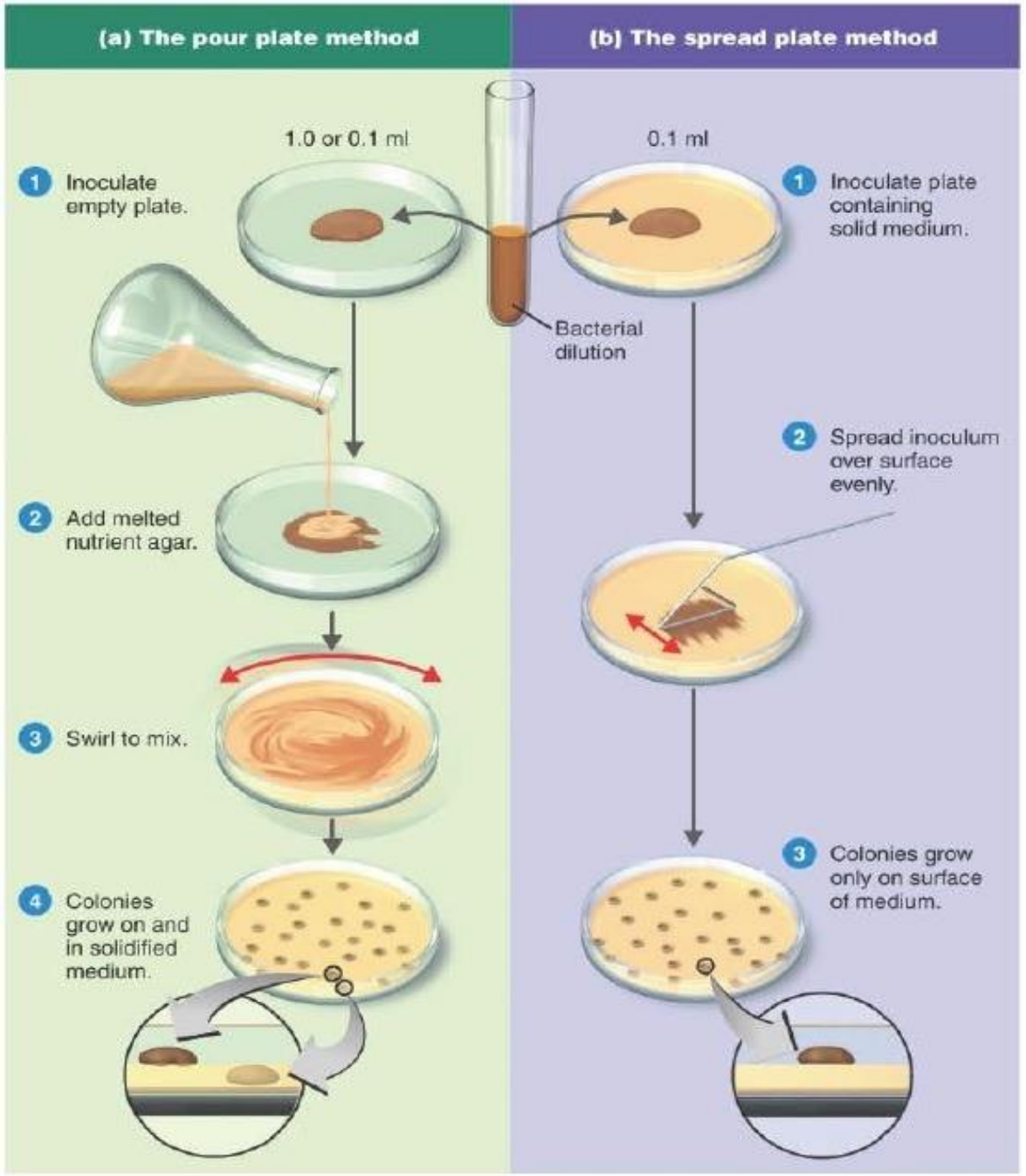
Advantages of Pour Plate Technique

- It is easy to perform and doesn't require extra tools or materials for inoculation.
- Don't require previously solidified agar medium.
- Don't have a risk of gouging during inoculation as like in streaking and spreading.
- Even a very low microbial load can be detected.
- Along with the isolation of microorganisms, their isolated colonies can be obtained. The number of CFU/mL can be calculated as well.
- It can use any type of specimen like clinical or environmental samples, liquid or solid (it can be dissolved).

Disadvantages of Pour Plate Technique

- There is a need for serial dilution of the sample otherwise too numerous colonies will be formed that can't be counted or identified as discrete.
- Heat-sensitive organisms can be affected by molten media at 40–45°C.
- It requires some time for the growth of organisms and the formation of colonies. There will be less supply of oxygen to microbial cells at the bottom of the solidified media, hence growing slowly.
- Colonies may be smaller than in streaking or spreading which increases the chance of overlooking them.

- Comparison between the Spread Plate Technique and Pour Plate Method



streaking method

Apparatus required:

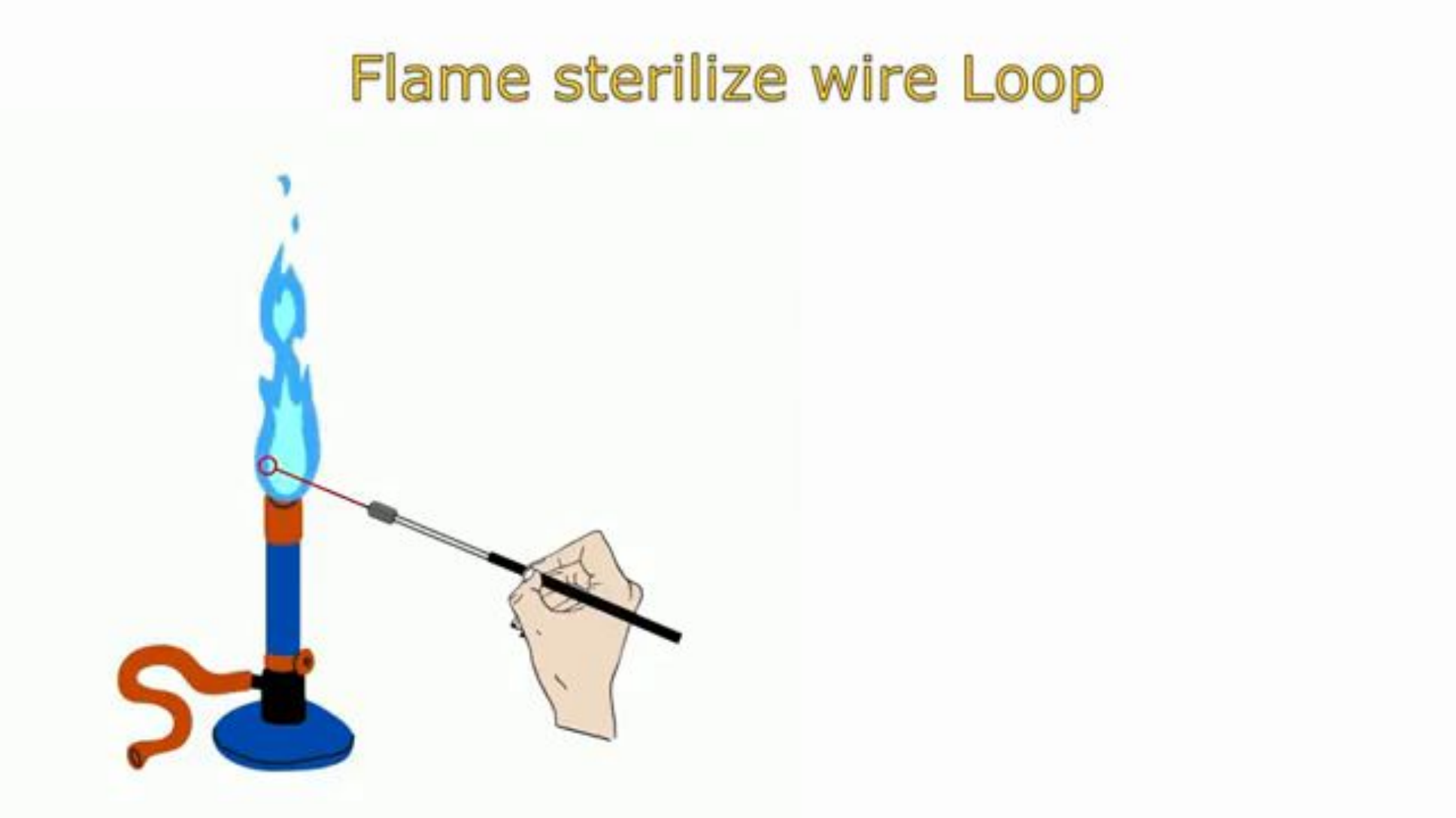
T Streak

Nutrient Agar Plate

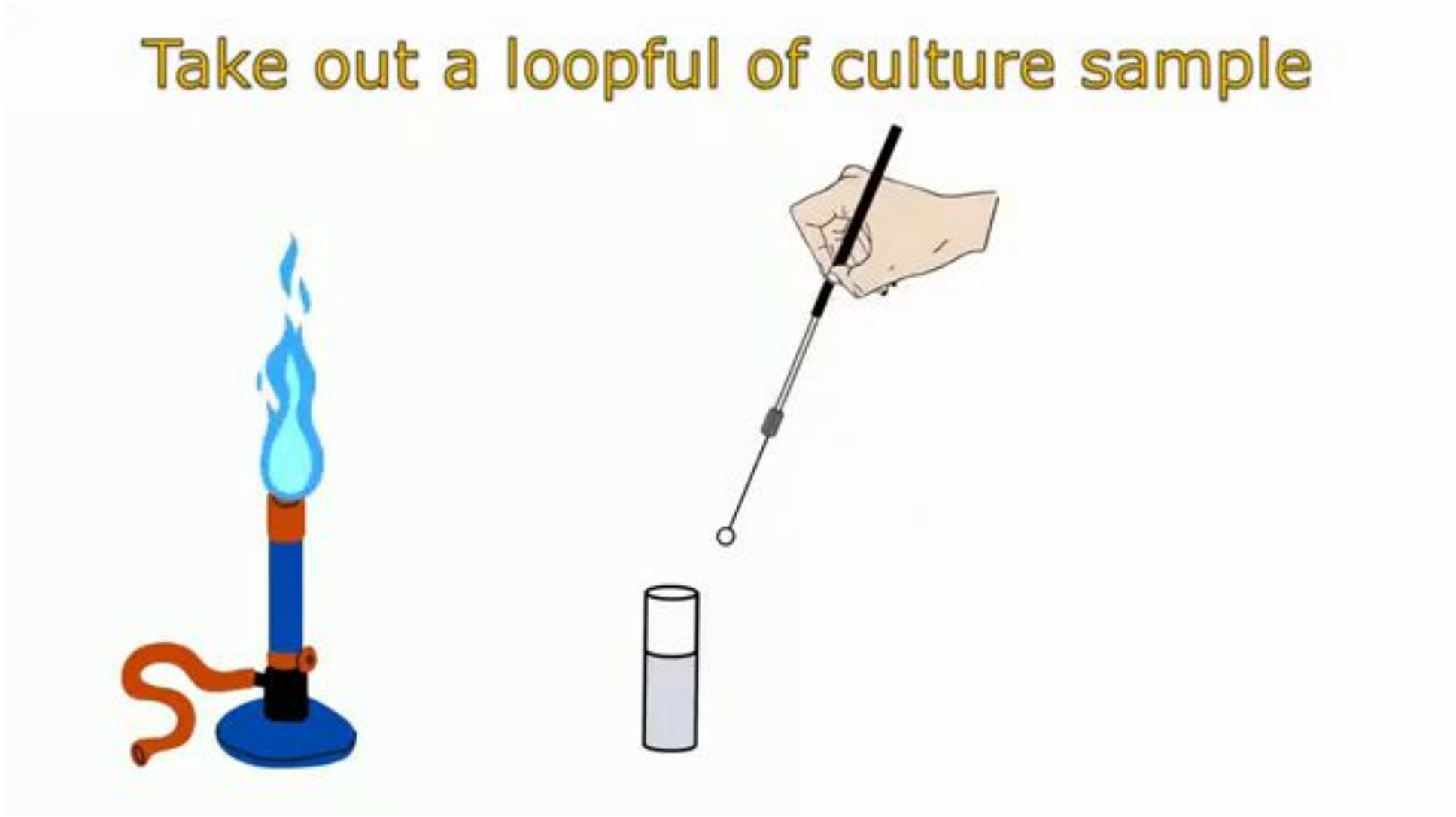
Quadrant streak

Radiant Streak

Zig-zag streak



Continuous streak



H.W

What are the advantages and disadvantages of streaking method ?