

University of Al-Maarif

Medical Laboratory Technique Department.

2nd. Stage

Practical Molecular Biology

The Molecular Biology Laboratory Introduction

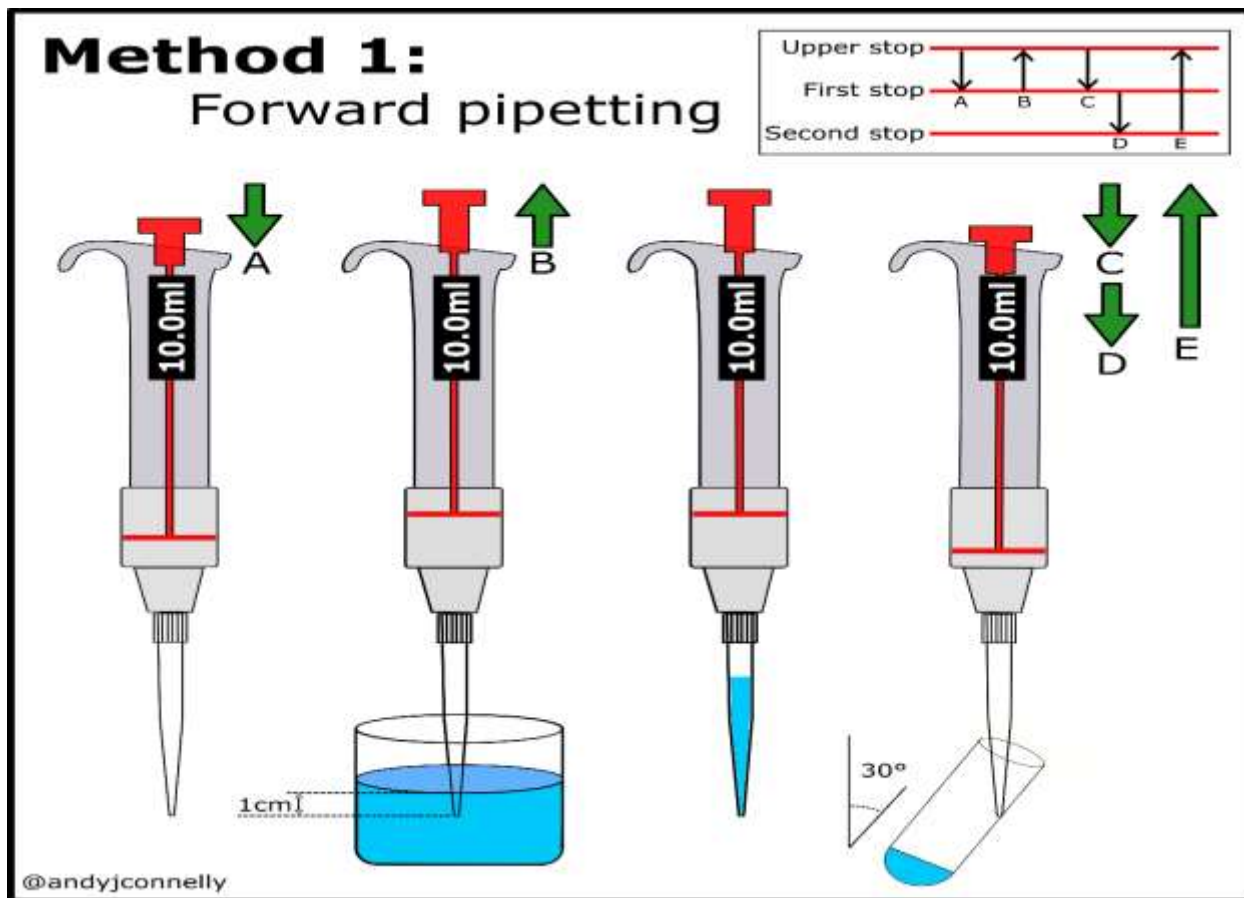
Analysis of the cell component DNA involves the use of specialty equipment, techniques, and protocols. Standard equipment used in the molecular biology laboratory includes pipettes, centrifuges. In this lab exercise, students will learn the basic techniques involved in **pipetting** and correct use of a **centrifuge** while learning to follow a simple protocol.

Pipettors:

Choose the most appropriate volume pipette (ex. If you want to pipette 300 μ l, use the 200-1,000 μ l pipette) and corresponding tip (push pipette into pipette tip without touching tip). Adjust the volume on the pipette by turning the knob to the desired volume (for the above example, the volume should read 030 NOT 300). Depress the pipette until you feel resistance (don't force it). Insert pipette tip into liquid and then release. Depress the pipette again to discharge the liquid being careful not to contaminate the tip during transfer. Release pipette tip into desired tube by pressing white button. never go beyond the volume capacity specified on the pipette.

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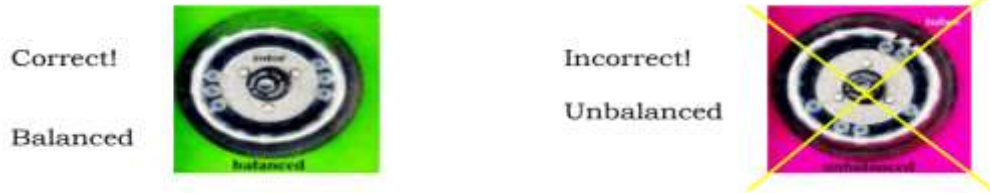
Note: To prevent **contamination** of your samples and materials, keep lids covering pipette tips and sample tubes closed as much as possible and avoid contact with the pipette tip when transferring liquid. It is helpful to have tubes and mixtures ready before you prepare pipette tip. Also, wear gloves when doing any work in the lab.



Place centrifuge tubes directly across from each other in the centrifuge. For example, if the centrifuge has 18 slots and you have 6 tubes, place three tubes in slots 1-3 and three tubes in slots 10 – 12. It is **CRITICAL** that the centrifuge is balanced (see below). Make sure that the tubes have similar volumes of sample and an even number of tubes. You may have to add an extra tube with water to balance

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the centrifuge. Alternatively, if you can coordinate with another group to combine tubes in order to balance, this would save time. Replace spray guard (if present), close lid, and set desired rpms and time (this is usually specified in protocols).



Protocol definition

A **protocol** is a detailed set of instructions used to perform a specific task. Before starting any procedure, it is highly recommended that you read the protocol in entirety first to make sure you have all necessary materials and understand the procedure. Also, reading the protocol first will help you determine the most efficient and time saving methods for completing the task.

Safety Guidelines

There are certain chemicals we use throughout these laboratory protocols that are dangerous upon exposure. Acquiring knowledge about using them properly is quite important, to remain healthy.

Phenol: is a strong corrosive chemical and quickly cause serious chemical burns. It also causes cellular damage by precipitating and denaturing proteins [changing shape of proteins]. In addition, phenol is a respiratory irritant and readily penetrates human skin.

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Chloroform: is a strong carcinogen [cancer causing agent], it can cause tissue and organ damage. It can also be inhaled or absorbed through the skin. Its effects are cumulative.

Ethidium bromide: is a strong mutagen [causing mutation], by virtue of its insertion between the bases of DNA. Gloves should always be worn when handling gels or buffers containing this chemical. One can use disposable tubes to mix the gel with Ethidium Bromide then use it. This would prevent contamination of the bench work as well as the materials being used.

UV light: used to illuminate the DNA, stained with Ethidium Bromide, is dangerous. Protect your eyes and face and hands by wearing a UV blocking face shield as well as wearing gloves.