B7 Pipettes

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What are the goals of this module?

Pipettes are omnipresent in Molecular Biology labs. They are an integral part of most experiments we will be performing.

We use pipettes to:

- **measure** a volume of liquid precisely and accurately
- **transfer** a known volume of liquid from one container to another

After working through this module, you will understand and demonstrate proper pipetting technique. You will be able to:

- **select** the appropriate pipette for a given task
- **use** the selected pipette in the proper manner
- **dispose** of any waste that results in the proper manner

Image source: commons.wikimedia.org

What do you notice about these pipettes?

Take a minute to notice some of the **similarities** & **differences**:

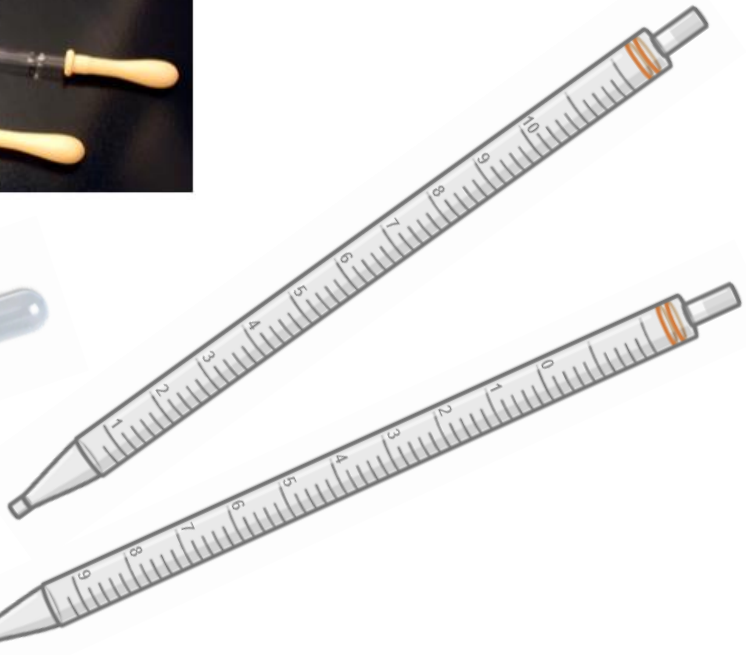
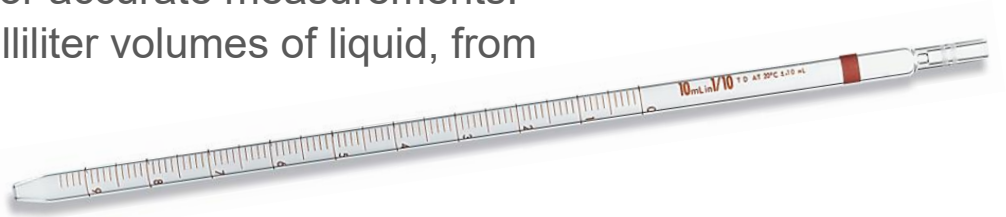


Image sources:
us.vwr.com
commons.wikimedia.org

Types of pipettes

Serological Pipettes are the clear choice for accurate measurements. They are frequently used for transferring milliliter volumes of liquid, from less than 1 mL to up to fifty 50 mL.



- **Blow-out:**
  - A blow-out pipette has a continuous etched ring or two small, close continuous rings located near the top of the pipette.
  - This means you need to get the last drop of liquid out of the pipette

- **Self-draining:**
  - Without these markings, a pipette is self-draining, and the last drop should **not** be forced out.

Image source: commons.wikimedia.org

Types of pipettes

Pasteur Pipettes are used to transfer uncalibrated volumes up to about 2.5 mL

The commonly recognized form is a glass tube tapered to a narrow point (a pipette) and fitted with a rubber bulb at the top, although many styles of both plastic and glass droppers exist.

The Pasteur pipette name is from the French scientist Louis Pasteur, who used a variant of them extensively during his research.

How to pipette correctly

This approximately 3-minute video demonstrates correct pipetting procedure:

https://www.youtube.com/watch?v=WGLivRvsh5w

Check your understanding # 1: How to keep pipettes sterile

Which end should you open?

Answers to “Check your understanding” questions are on the last page of the module

Check your understanding # 2: How to correctly read measurements

Which images show that 2 mL of liquid have been drawn into the pipette?

A

B

C

D

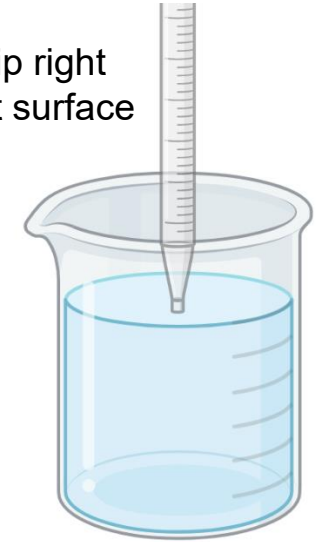
E

F

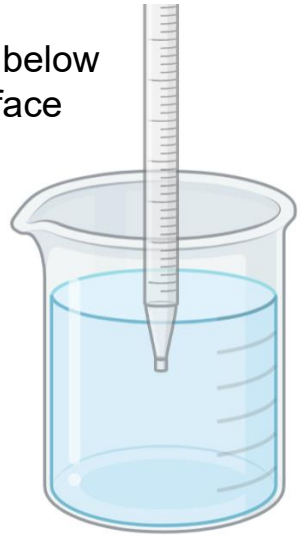
Check your understanding # 3: How to properly immerse the pipette tip

Which image shows the appropriate pipette position when drawing up liquid?

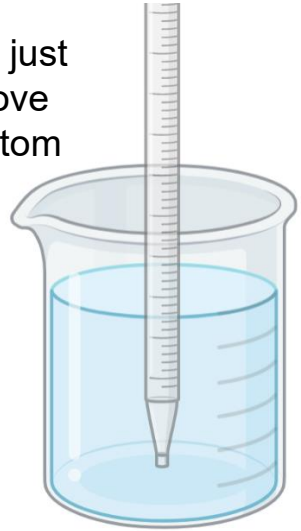
A
Tip right at surface



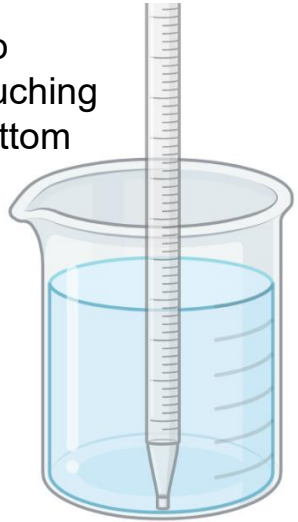
B
Tip below surface



C
Tip just above bottom



D
Tip touching bottom



Pipette fillers

Biology 140 labs will use pipette pump style pipette fillers. They are typically color coded to indicate their maximum volume:

- Blue: up to 2 ml
- Green: up to 10 ml
- Red: up to 25 ml (not used in B140)

Use your thumb to draw up liquid:

or to dispense liquid:

Image source: Forsyth Tech CTL https://www.youtube.com/watch?v=97SSzo5PGQs

Check your understanding # 4: Correct pipetting process

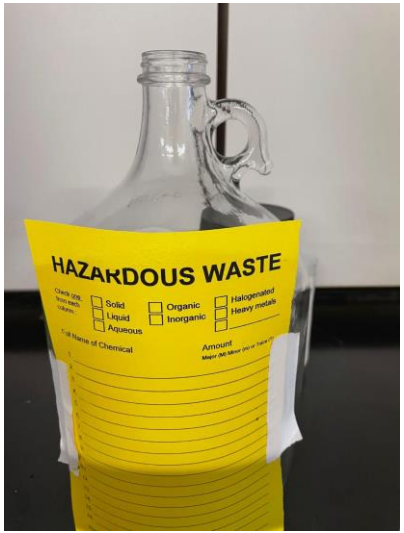
To transfer **7 mL** of buffer:

1. Choose the appropriate pipette
    a. 2 mL
    b. 50 mL
    c. 10 mL
    d. 25 mL
    e. 5 mL
    f. 1 mL
    g. Pasteur
2. Choose the appropriate pipette pump
    a. Blue
    b. Green
3. For the pipette you chose, should you
    a. push out the last drop of solution, or
    b. just let it drain without forcing out the last drop?

Waste Disposal

All waste must be disposed of properly:

- **Liquid waste** must be transferred to the appropriately labeled glass jug



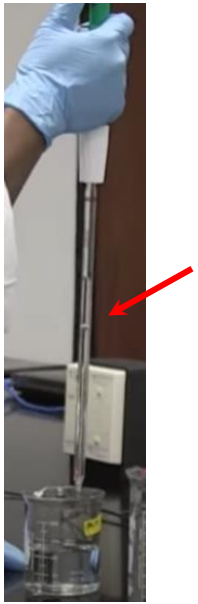
- **Used pipettes** that are not contaminated with any hazardous chemicals or bacteria should be placed in the labeled bin

If you have any questions, please consult with your lab instructor.

Troubleshooting

What should you do if…

you get bubbles in the solution you are drawing in to the pipette?



What should you do if…

the tip touches something (your hand, the counter, etc.) and becomes contaminated?

Troubleshooting answers

What should you do if…

 you get bubbles in the solution you are drawing in to the pipette?

Dispense liquid back into the original container and try again

What should you do if…

 the tip touches something (your hand, the counter, etc.) and becomes contaminated?

Dispose of pipette properly and try again

If you have any questions, please consult with your lab instructor.

Conclusion

If you feel you have mastered the objectives of this module, please schedule an appointment to complete a posttest with the Science Learning Center assistants. Please let us know if you have any questions!

Answers to “Check Your Understanding” questions

1. A
2. A and F

   Note: There are two different scales, one counting up and one counting down. The volume is read at the bottom of the meniscus.

1. B

   Note: The tip should be submerged far enough that it remains in solution after the desired volume of liquid has entered the pipette. If it is submerged too deeply, droplets on the outside of the pipette can lead to volume errors.

1. I: **C** 10 mL pipette
   II: **B** Green pipette pump
   III: **A** “Blow out” the last drop
   (note the two close orange rings on the 10 mL pipette)