Use and Maintenance of Micro-pipettes

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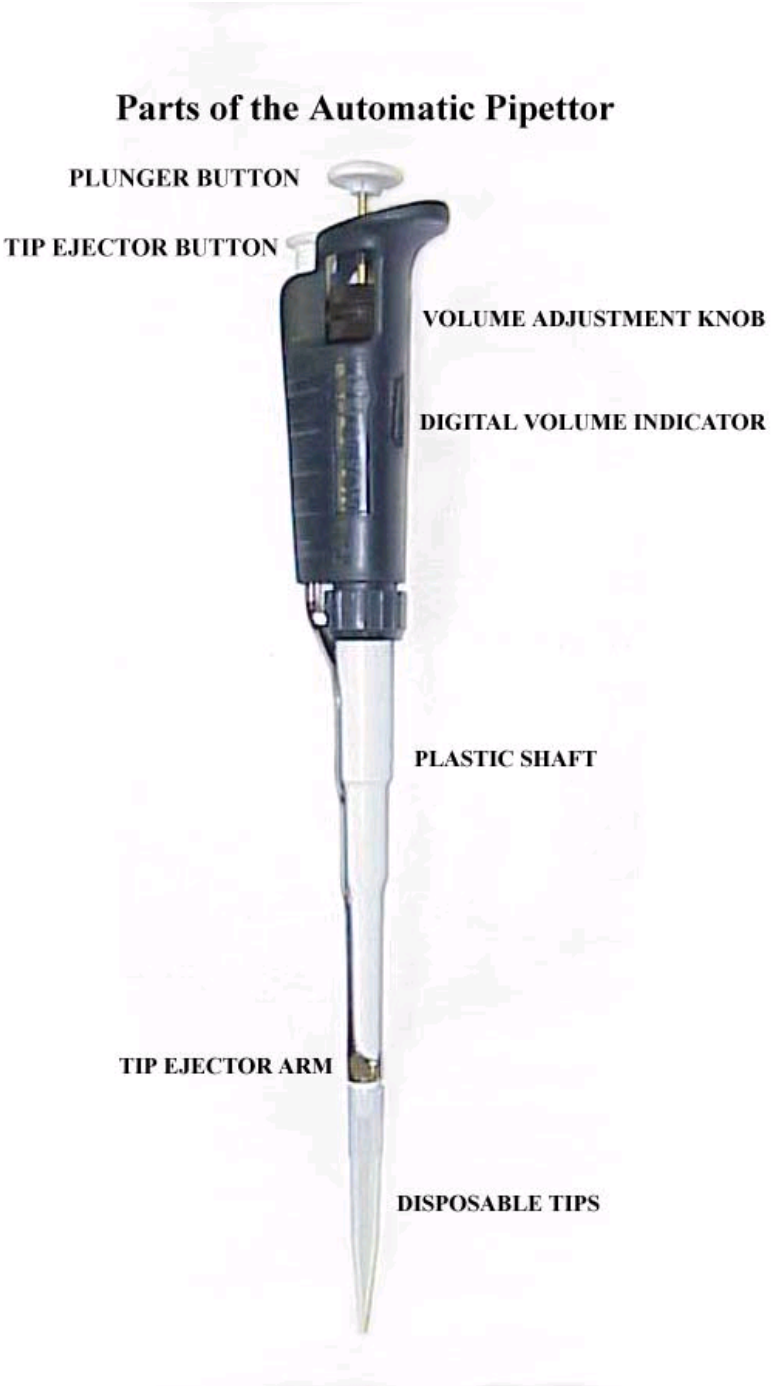
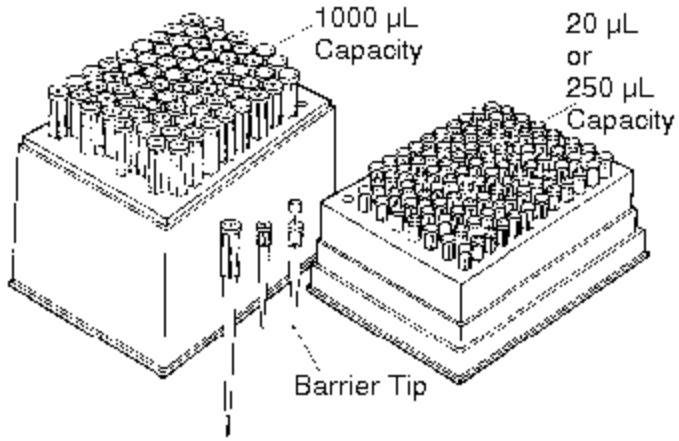
Introduction

- Automatic pipettes are used to accurately transfer small liquid volumes
- Glass pipettes are not highly accurate for volumes less than 1 milliliter (1 ml), but the automatic pipettes are both accurate and precise
- These are continuously adjustable digital pipettes
- Each pipette can be set to transfer any volume within its own volume range

Parts of the Pipette





Pipette tips

Operating the Micropipette

Step 1: Set the Volume

Pipettors – 3 Volumes:

Volume Adjustment Knob:

Digital Volume Indicator:

Operating the Micropipette

Step 1: (Continued) Read the Volume

How to Read the Volume Indicator:

		
(a): P-20 Model **6.86 μ l = 0.00686** **or 6.86 x 10^{-3} ml**	**(b): P-200 Model** **132.4 μ l = 0.1324** **or 1.324 x 10^{-1} ml**	**(c): P-1000 Model** **262 μ l= 0.262** **or 2.62 x 10^{-1} ml**

Operating the Micropipette

Step 2: Attach the Disposable Tip

Attaching the disposable tip

Example of tip sizes:

Operating the Micropipette

Step 3: Depress the Plunger to the First Stop

Step 4: Immerse Tip in Sample

Step 5: Draw up the sample

To aspirate the sample into the tip, allow the pushbutton to return slowly and smoothly to the fully extended UP POSITION. NEVER LET THE PLUNGER SNAP UP! This draws the exact calibrated volume into the tip if the tip remains below the liquid surface during withdrawal.

Step 6: Pause

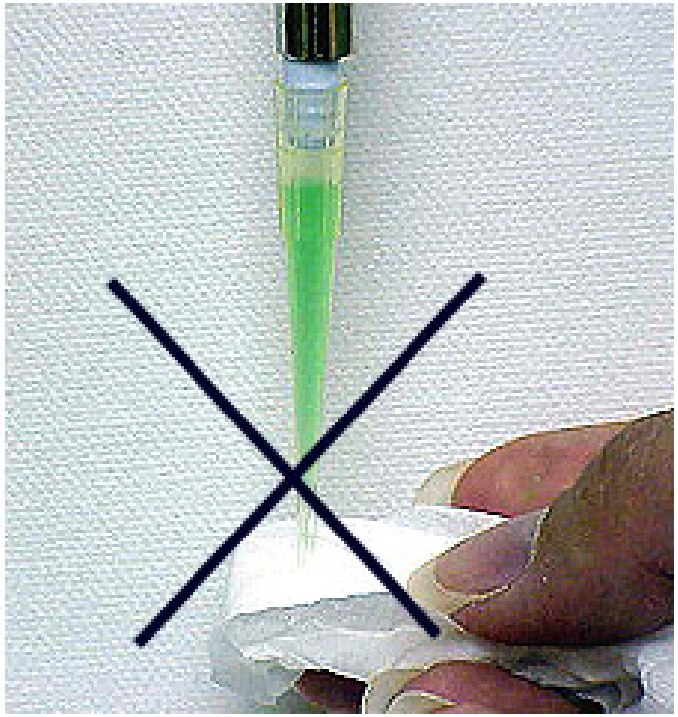
Wait a few seconds to ensure that the full volume of sample is drawn into the plastic tip. WAIT LONGER FOR LARGER VOLUMES. WAIT LONGER FOR MORE VISCOUS ("SYRUP-LIKE") SUBSTANCES.

Operating the Micropipette

Step 7: Withdraw the Tip

Remove the tip from the sample liquid. No liquid should remain on the OUTSIDE of the tip. Wipe away any droplets on the outside of the tip with a lint-free tissue, such as KIMWIPES, but only wipe droplets from the side of the tip. NEVER TOUCH THE TIP OPENING or you may absorb part of your sample.

Proper Droplet Removal



WRONG Droplet Removal

Operating the Micropipette

Step 8: Dispense the Sample

To dispense the sample from the pipette:
a) Touch the tip end to the side wall of the receiving vessel and
b) Depress the plunger to the FIRST STOP.
c) Pause for at least one second-- 1-2 seconds for P-1000, 2-3 seconds for P-5000, or longer for viscous liquids.
d) Press the plunger to the SECOND STOP (the second point, of greater resistance, at the bottom of the stroke) to expel any residual liquid in the tip (like "blowing out" a glass pipette).

<table>
<tr><td colspan="3">  </td></tr>
<tr><td>(a) Start Dispensing</td><td>(b) 1st Stop = Dispense</td><td>(c) 2nd Stop = Expel</td></tr>
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Operating the Micropipette

Step 9: Withdraw the Pipette

With the plunger fully depressed, withdraw the pipet from the receiving vessel carefully, sliding the tip along the wall of the vessel. Holding the tip against the side of vessel is especially important when transferring small volumes of liquid.

Step 10: Release the Plunger

Gently allow the plunger to return to the UP position. DO NOT allow it to SPRING BACK!

Step 11: Discard the Tip

Discard the tip by depressing the tip ejector button, as shown below. A fresh tip should be used for each sample to prevent sample carryover.

Press ejector button to discard tip.

Step-wise Operation of the Automatic Pipette

(1) Set the volume
(2) Attach disposable tip
(3) Depress the plunger to the first stop
(4) Immerse tip in sample
(5) Draw up the sample
(6) Pause
(7) Withdraw the tip
(8) Dispense the sample
(9) Withdraw the pipette
(10) Release plunger
(11) Discard the tip

Accuracy and Precision

- Accuracy means the closeness with which the dispensed volume approximates the volume set on the pipette
- Accuracy is specified as mean error, the average deviation of replicate measurements from the expected set volume
- Precision is the "scatter" or reproducibility of individual measurements of the same volume
- Precision can also be expressed as standard deviation

Accuracy and Precision (Continued)

- Relative accuracies are generally about 1% or less
- Precision is less than 0.5 % except when transferring the smallest recommended volume for a given pipette model
- Using the pipettes to transfer volumes which are below the recommended range will introduce larger errors

Pippetting Guidelines and Precautions

For optimal reproducibility, use the following pipetting procedures:

(1) Consistent SPEED and SMOOTHNESS when you press and release the PLUNGER

(2) Consistent pressure on the PLUNGER at the FIRST STOP

(3) Consistent and sufficient IMMERSION DEPTH

(4) Nearly VERTICAL POSITIONING of pipette

(5) AVOID ALL AIR BUBBLES: Since the plastic pipette shaft can be damaged if liquids are drawn beyond the tip into the shaft

(6) NEVER lay the pipette on its SIDE nor INVERT the pipette if liquid is in the tip

Thank You