PIPETTING TECHNIQUES





Reading and Adjusting the Volume

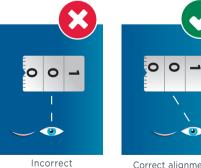
Hold the body of the micropipette in one hand and use the other hand to rotate the thumbwheel or the push button. With the push button, the volume can be easily adjusted with one hand. Push button volume adjustment is available on all MICROMAN pipettes and on PIPETMAN pipettes (except PIPETMAN L) manufactured after April 1995.



A Helpful Hint for Improving **Reproducibility and Accuracy**

Always finish setting clockwise for best reproducibility. This is how to obtain a clockwise volume setting:

- When decreasing the volume setting, slowly reach the required setting, making sure not to pass the setting.
- When increasing the volume setting, pass the required value by 1/3 of a turn and then slowly decrease to reach the volume, making sure not to pass the setting.



alignment: error

Correct alignment: accurate reading

To avoid parallax, hold the pipette in a horizontal position. Adjust the volume until the indicator is lined up with the desired volume.

NOTICE

To avoid internal damage to your pipette, never attempt to force the volume setting beyond the limits.



Air-Displacement / Forward Mode

The forward mode is the standard way of pipetting with an air-displacement pipette like PIPETMAN.



Preparation

Hold the instrument in a nearly vertical position. Depress the plunger smoothly to the first stop position.



Aspiration

Forward or Reverse Mode Pipetting

Immerse the pipette tip in the liquid*. Allow the plunger to move up smoothly to the rest position. Wait one second so that all the liquid has time to move up into the tip.



Dispense

Place the pipette tip at an angle (10° to 45°) against the inside wall of the receiving vessel. Depress the plunger smoothly to the first stop position.



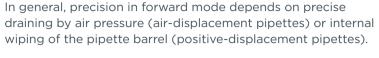
Purge

Wait one second, then depress the plunger to the second stop position. This purge stroke removes any remaining sample from the tip. Remove pipette tip end from sidewall by sliding it up the wall.



Home

Allow the plunger to move up to the rest position.



VOLUME	IMMERSION DEPTH (MM)
0.1 — 1 μL	11
1—100 μL	2-3
101—1000 μL	2-4
_1001 μL— 10 mL	3-6

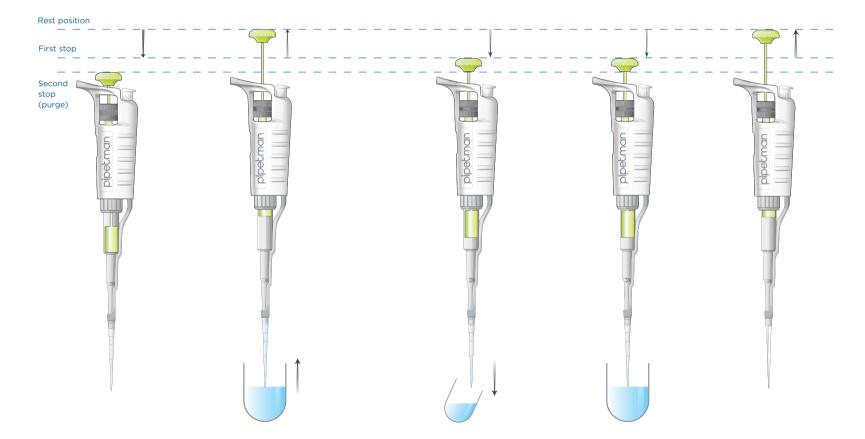
Pre-Rinsing

To obtain greater uniformity and precision of dispensing, it is better to provide identical contact surfaces for all aliquots. This is done by pre-rinsing with the same liquid as the one dispensed.

For pre-rinsing, aspirate with the tip, and then dispense back into the original reservoir or to waste.

Pre-rinse again when adjusting the volume.

* The immersion depth of your tip can have a significant effect on your results (for depth per model, see table above). If the tip is immersed too deeply, droplets will form on the outside of the tip and they will be deposited along with your sample. If the tip is not immersed deeply enough, vortexing will occur and your pipette will not aspirate the selected volume.



PIPETTING TECHNIQUES

Forward or Reverse Mode Pipetting

Air-Displacement / Reverse Mode

The reverse mode is only possible with air-displacement pipettes. It is sometimes used to pipette slightly viscous liquids.

In reverse mode pipetting, the purge stroke is used during preparation. During aspiration, an amount of liquid equal to the amount of purged air is added. This amount compensates for the liquid that remains inside the tip during dispensing.



Preparation

Hold the instrument in a nearly vertical position. Depress the plunger smoothly to the second stop position.



Aspiration

Immerse the pipette tip in the liquid. Allow the plunger to move up smoothly to the rest position. Wait one second so that all the liquid has time to move up into the tip.



Dispense

Place the pipette tip at an angle (10° to 45°) against the inside wall of the receiving vessel. Depress the plunger smoothly to the first stop position. Wait one second.



Re-Aspiration

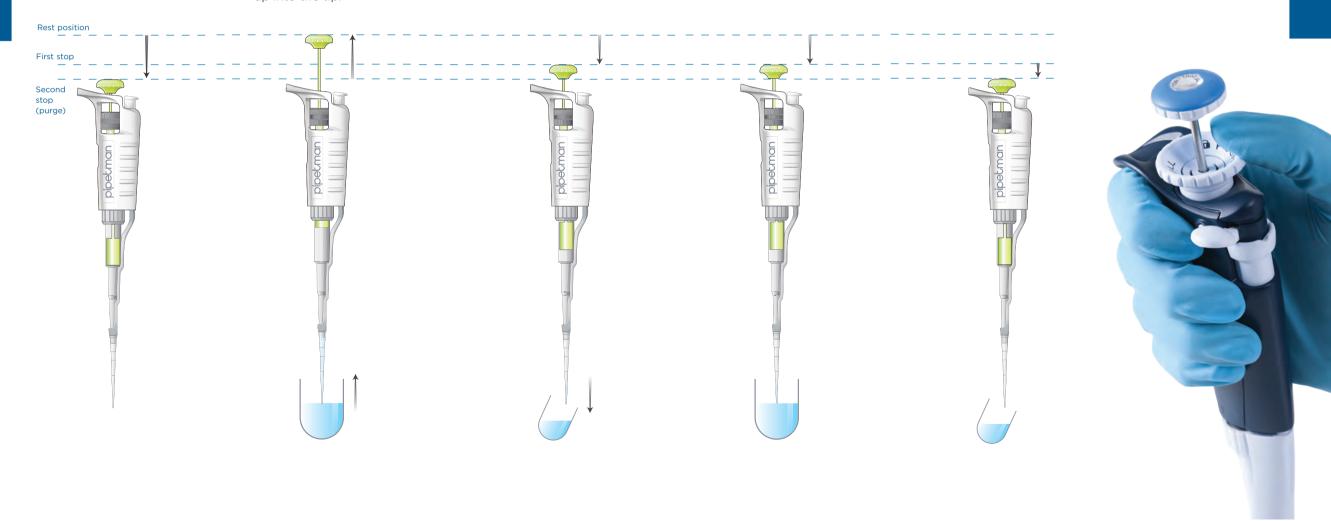
If the pipette tip is to be reused for the same sample, maintain the plunger in the intermediate position for subsequent immersion for the next pipetting cycle and restart operation 2.



or

Complete Purge

Wait one second and purge. If the pipette tip is not to be re-used, depress the plunger to purge position over an appropriate waste container and then eject the tip.



19 PIPETTING TECHNIQUES | GILSON GUIDE TO PIPETTING

PIPETTING TECHNIQUES

Forward or Reverse Mode Pipetting

Positive-Displacement / Forward Mode



Preparation

Press the plunger button to the first stop. The piston moves to the appropriate position.



Aspiration

the capillary.

Immerse the capillary/piston in the liquid*. Release the plunger, letting it move up to the home position. The piston moves up and the ambient pressure forces the desired volume of liquid through the orifice into



Dispense

Press the plunger button to the first stop. The piston moves down and expels the liquid out of the capillary.



Eiection

Press the plunger all the way down to the second and last stop. Capillary and piston are ejected without hand contact.



To avoid distorted results linked to the volume pipetted, ensure that no liquid is on the outside part of the tip. If necessary (viscous liquids, such as cream), wipe the outside of the tip or the capillary with a clean medical wipe. Do not touch the orifice. Choose a tissue that is resistant, lint-free, and inert to acids and solvents. Dispose of the tissue in a safe, hygienic manner.



When working with high risk specimens, do not wipe the disposable part. Make sure fluid depth penetration does not exceed the recommended immersion depth*.

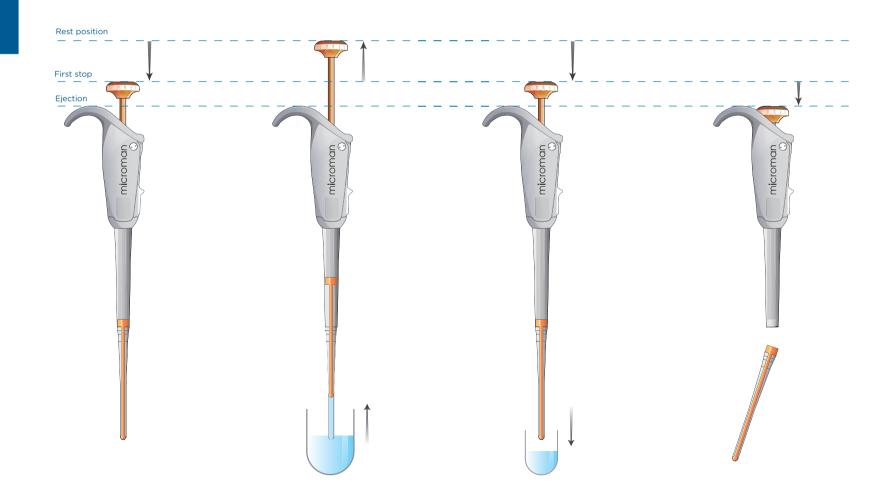


To pre-rinse, aspirate with the tip and then dispense back into the original reservoir or to waste.



When working with high risk specimens, do not wipe the disposable part. Make sure fluid depth penetration does not exceed the recommended immersion depth.

* The immersion depth of your tip can have a significant effect on your results (for depth per model, see table page 20). If the tip is immersed too deeply, droplets will form on the outside of the tip and they will be deposited along with your sample. If the tip is not immersed deeply enough, vortexing will occur and your pipette will not aspirate the selected volume.



21 PIPETTING TECHNIQUES | GILSON GUIDE TO PIPETTING

Tips for Mistake-Free Pipetting

How to Avoid Typical Pipetting Mistakes

MANY FACTORS MAY IMPACT PIPETTING ACCURACY

INFLUENCING PARAMETERS AND EFFECTS	CORRECTIVE MEASURES
Leaky/poorly seated pipette tips may affect accuracy by 0.5% to 50%	Using original or recommended pipette tips
Reuse of pipette tips may affect accuracy by up to 4%	Using pipette tips only once
The straightness of pipette tips may affect accuracy by up to 10%	Using appropriate tips only
The difference in vapor pressure of the liquid to be pipetted versus that of the water used for adjustment may affect accuracy by up to 2%	Sufficient pre-wetting of pipette tips
Failure to wipe pipette tip on the vessel wall can affect accuracy by up to 3%	Wiping of the pipette tip on the vessel wall (wiping distance 8 to 10 mm)*
Pipette tip immersion depth and handling angle during pipetting may affect accuracy by up to 1%	Holding pipette in a vertical position while pipetting
Irregular rhythm and timing during pipetting can affect accuracy by up to 1.5%	Applying a consistent pipetting technique
A leaky piston system can affect accuracy by 1% to 50%	Regularly checking the pipette and the volume aspirated
Uneven piston movement can affect accuracy by up to 0.5%	Smooth operation of piston

Information extracted from ISO 8655-2 - Annex B

Pipetting Ergonomics

Take a Few Minutes to Get **Organized and Ensure You Have:**

- 1. An appropriate posture
- 2. The right material Gilson offers various pipettes with forces adapted to user preferences. The forces of PIPETMAN L are some of the lowest.
- 3. The appropriate gestures
- 4. A good work organization

A good test is to see if you can rest your elbow comfortably on the work surface. If not, your receptacle may be too low or too high, find the right height.





Download the Gilson Ergonomics Poster for increased working comfort in your lab

www.gilson.com/resources/ergonomics.pdf

Take Time to Relax

- 1. If possible, try to switch periodically between different types of work.
- 2. Keep an appropriate, unrushed working speed. Let go of the pipette from time to time and give your fingers/hand a (micro) break.
- 3. Take frequent short breaks. Change your sitting position. Lean back and relax your shoulders and arms.

Special Attention Should be Paid to Smooth Pipetting

- 1. To facilitate uniform timing and motion, keep all necessary items within arm's reach.
- 2. Place the most frequently used objects in front of you. The more rarely used items can be placed a little further away from you.
- 3. The opening of the receptacle for used tips should be at the same height as the end of your pipette.

Use a Pipette Holder

Protect your pipette and always store it vertically on a pipette holder. Pipettes left on a workbench or stored in a drawer can easily come into contact with samples and become contaminated.



Figure 5 SINGLE® Pipette Holder



Figure 6 POWER CAROUSSEL® Stand

^{*} Gilson recommends touching the tip to the vessel wall at an angle of 10° to 45°.