Pipetting Tips and Tricks

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Improve your results

“Even the warmth of your hand can influence pipetting results.”

Want to know why?

METTLER TOLEDO recently released a series of tips and tricks emails for life scientists. These tips and tricks covered the areas of weighing, pipetting and pH measurement. The emails were so well received, we compiled them to make them available to anyone who is interested.

By describing common mistakes and how to improve technique, this booklet about pipetting tips and tricks will help you to achieve more accurate results.

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METTLER TOLEDO

Summary

Pipette results are dependent on their reliable performance. Are you sure your pipette is dispensing the correct amount? Although regular pipette checkups can ensure functioning and accurate pipettes, did you know that how you handle your pipette greatly influences its performance?

Pipetting technique directly affects the success and repeatability of your experiments. This booklet is about tips and tricks for proper pipetting. It will help you to achieve more accurate results, by describing common mistakes and describing how to improve your technique.

1. Aspirating at the Correct Angle

Aspirating at the correct angle

The immersion angle of your pipette tip should be as near to 90 degrees as possible and not deviate more than 20 degrees from vertical. If the angle is more than this, the liquid level will be lower than when the pipette was calibrated. This can cause too much liquid to be drawn into the tip, resulting in inaccurate aspirations. For example, at an angle of 30 degrees from vertical, up to 0.7% too much liquid can be aspirated.

- Read more on aspirating at the correct angle in the Academia World of Wonders eBook.

2. Maintaining Consistency

Consistent pipetting rhythm

Use a consistent pipetting rhythm from sample to sample. Instead of rushing or rapid operation, get into a rhythm for each step in the pipetting cycle.

Large volume pipettes

For larger volumes – typically 1 ml or greater – pause about 1 second or more after sample pickup with the tip still in the liquid. This will allow the sample to be fully aspirated. Withdraw the tip only when aspiration is complete.

Smooth plunger action

Maintain consistent speed and smoothness when pressing and releasing the plunger. Uncontrolled aspiration can cause bubbles, splashing, aerosols and contamination of the pipette shaft and piston.

Electronic pipettes

Electronic pipettes deliver the greatest pipetting consistency by virtually eliminating inconsistencies due to variability in user technique.

- Electronic Singlechannel Pipettes
- Electronic Multichannel Pipettes
- Liquidator96 pipetting system

3. Pre-rinsing Tips

Pre-rinse tips at least twice

Dispensing liquid from a pipette leaves a coating of the liquid on the tip, making the expelled volume slightly less than it should be. Whenever a new tip is used, pre-rinse at least twice with the liquid being dispensed before pipetting, to compensate for this liquid film. Pre-rinsing also helps neutralize capillary effects in micro-volume pipettes and, for macro-volume pipettes, equalizes the air temperature inside the pipette with the sample temperature.

Exceptions to pre-rinsing

Pre-rinsing can negatively affect results when pipetting very warm or cold solutions. Pre-rinsing is not recommended for pipetting very cold solutions, such as those from an ice bath, or for solutions above 37°C, as it may result in errors of up to 5%.

- BioClean Tips

4. Avoiding Temperature Variation

Maintain a constant room temperature

Large or sudden temperature changes should be avoided, since air inside the pipette cannot adjust quickly, which can affect the volume of sample dispensed. Pipetting at a constant temperature can improve accuracy by up to 5%.

Allow time for equilibration

Pipettes are susceptible to variation in the temperature of the samples dispensed. Cold liquids tend to deliver in excess, while warm liquids may deliver smaller volumes than expected. Unless otherwise specified, allow sufficient time for the temperature of your pipettes and liquids to equilibrate before use.

Hand warming effects

When pipetting for long periods, heat from your hand can warm the air inside the pipette, causing it to expand and leading to inaccurate results. Use pipettes made from high grade PVDF polymers, avoid continually holding the pipette in your hand between pipetting cycles and always put the pipette back on its stand after pipetting.

- Read more in the Academia World of Wonders eBook.

5. Ensuring Complete Dispensing

Ensure proper touch-off

Greatest accuracy and sample-to-sample reproducibility are achieved by ensuring that the last remaining droplet dispenses fully and does not adhere to the tip end. This is particularly important for micro-volume pipetting, due to the small sample volumes involved. For most applications, dispensing with the end of the tip resting against the vessel wall reduces or eliminates the amount of sample remaining in the tip. Remove the pipette by sliding the tip end along the side wall to release any remaining droplet at the tip orifice. This technique can increase accuracy by 1% or more. For applications where this is not appropriate, use thin wall tips – which allow the droplet to fully release – to dispense the sample either above the liquid surface or directly into the liquid.

- Electronic Singlechannel Pipettes
- Electronic Multichannel Pipettes

6. Routine Balance Testing

Correct tip filling
The best results are achieved by aspirating at 50% of the maximum tip volume. Pipetting volumes of between 35 and 100% of nominal volume is recommended. This puts less demand on operator technique and improves results by 1%. Pipetting volumes less than 10% of the nominal volume is problematic.

Immersion depth
The correct tip immersion depth can improve accuracy by up to 5%, and is particularly important for micro-volume pipettes. The tip should be immersed between 1-2 mm for micro-volume pipettes and up to 6-10 mm for large-volume pipettes, depending on the tip size. If the tip is immersed too far, the volume of gas in the tip is compressed and can cause too much liquid to be aspirated. Liquid retained on the tip surface can also distort results. Not immersing the tip far enough can draw air in, causing air bubbles and inaccurate volumes.
• Liquidator96 pipetting system

7. Posters

Good Pipetting Techniques - Get a Free Poster!
Pipetting technique directly affects the success and repeatability of your experiments. Luckily there are simple measures to ensure that your pipetting results are always accurate.

Refresh your knowledge!
Click here to request a free copy of the Good Pipetting Techniques Poster!