



Analyzing the Effect of Pipette Tip Geometries on Fluid Velocity and Shear Strain Rate: Biomek Wide Bore vs. Standard Pipette Tips

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Introduction

Throughout the scientific literature, sample preparation protocols specify the need for “wide bore” pipette tips and cannulas to minimize the inadvertent harm, which can be applied to native biological sample matrices during sample preparation. Loss of biological sample integrity can occur when pipetting fragile cell suspensions, hybridomas, embryoid bodies, algal coagulates, protein aggregates, and genomic materials due to the pipette tips’ small internal orifice diameter and the speed of aspiration and dispense during pipetting. This has led researchers to report, “cutting 2 to 3 mm from the end of the tips” to increase the internal diameter, or bore, of standard pipette tips. This practice achieves the intended result albeit in an imprecise manner and with a shortening of the tip, which reduces the pipette tip volume and limits labware access.¹

Biomek Automated Workstations, and their associated software, provide greater control and reproducibility over pipetting techniques to minimize the disruption of sample integrity vs. manual pipetting, but the internal diameter of the pipette tip orifice may demand a slowing of pipetting speeds to compensate for the turbulence [and shear] caused within the narrow diameter pipette tip. Biomek Wide Bore Pipette Tips have been designed to address such sample preparation challenges. Through the use of Computational Fluid Dynamics (CFD) software, physical forces and flow characteristics within a pipette tip during pipetting have been simulated. In this virtual CFD world, comparisons of pipette tip orifice diameters can be analyzed for such parameters as fluid (sample) velocity, wall shear, and shear strain rate. Less wall shear effects and shear strain rates are desirable in preserving the biological integrity of native samples during sample preparation. Wall shear occurs within a pipette tip at the plane of contact between the sample fluid and the inside wall of the pipette tip. Shear strain rate indicates the rate of change in displacement of the faces of a sheared layer of sample fluid.



Figure 1. Biomek Span-8 pod with P250 (i-Series* 230 µL) wide bore pipette tips.

Simulation Design

Biomek P250 (i-Series 230 µL) standard and wide bore pipette tips were analyzed at a constant volumetric flow rate with both deionized (DI) water and 80% glycerol as comparative sample fluids representing typical biological sample matrices. The simulations were designed using laminar flow conditions of uninterrupted, unidirectional sample flow along the inside wall of the pipette tip. Biomek software default technique files for aspirate and dispense speeds for the respective sample fluids were used in the models. (See Table 1 below.)

The primary computational fluid dynamics software used for this analysis was ANSYS CFD Software. Simulations were run on a 180 GB, 160-processor, 64-bit computer.

Sample Fluid	Viscosity(cP) @ 20°C	Flow Rates	Standard Orifice ID	Wide Bore Orifice ID
Water, DI	1.0	90 µL s ⁻¹	0.6 mm	1.4 mm
Glycerol, 80%	60.1	30 µL s ⁻¹		

Table 1. Simulation design parameters.

Results and Discussion

The CFD software simulation exposed significant differences in fluid (sample) velocities and shear strain rates on the inside of the pipette tips for both viscosities of sample fluids tested; e.g., DI water and 80% glycerol. As presented in Table II, a reduction in maximum fluid velocities of 3.95 times and 4.75 times is measured when the pipette tip orifice (or bore) is increased from 0.6 mm to 1.4 mm for DI water and 80% glycerol, respectively. Maximum shear strain rates are also reduced with the larger orifice diameter by factors of 13 times for DI water and 10 times for 80% glycerol.

Sample Fluid	Analysis	Standard Orifice Pipette Tip	Wide Bore Orifice Pipette Tip	x Reduction (Standard/Wide Bore)
Water, DI	Velocity, maximum	0.415 m s ⁻¹	0.105 m s ⁻¹	3.95x
	Shear Strain Rate, maximum	3982 s ⁻¹	313 s ⁻¹	13x
Glycerol, 80%	Velocity, maximum	0.172 m s ⁻¹	0.036 m s ⁻¹	4.75x
	Shear Strain Rate, maximum	1005 s ⁻¹	99 s ⁻¹	10x

Table 2. Analysis results for velocity and shear strain of standard vs. wide bore orifice pipette tips with both DI water and 80% glycerol sample fluids using normalized radii values.

Effect on Fluid Velocity

Visualization of the reduction in sample velocities for DI water and 80% glycerol are shown in Figures 2 and 3, respectively. The color scale and associated color images of the pipette tips are defined by the progressively warmer colors representing increasing fluid velocities; e.g., green to yellow to orange to red. The progressively cooler colors represent decreasing fluid velocities; e.g., green to blue to indigo to violet. Figure 2 shows the ellipsoidal velocity contours for a standard orifice pipette tip (left) and a wide bore pipette tip (right). The standard orifice pipette tip produces a comparatively higher fluid velocity at the tip and sample interface in relation to a wide bore orifice; indicated by the “hot spot” of red and surrounding warm colors on either side of the tip and sample interface vs. cooler blue to violet colors for that of the wide bore tip.

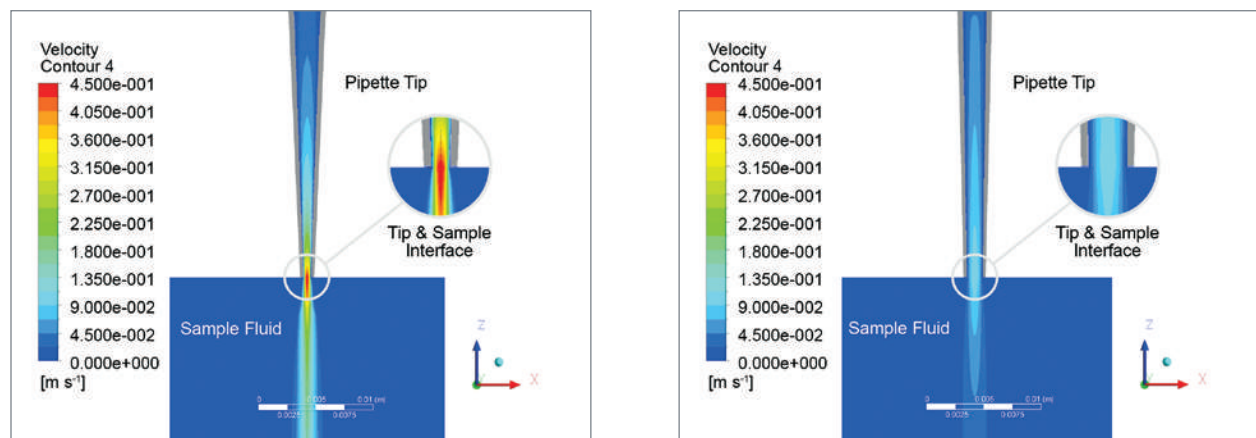


Figure 2. Fluid (sample) velocity through the pipette tip's orifice for a standard orifice pipette tip (left), and a wide bore orifice pipette tip (right) using DI water as the model sample fluid.

Figure 3, representing the velocity contour of 80% glycerol, shows similar comparative results to DI water (Figure 2) for a standard orifice pipette tip (left) and a wide bore pipette tip (right). The standard orifice pipette tip produces a comparatively higher fluid velocity at the tip and sample interface in relation to a wide bore orifice; indicated by the “hot spot” of red and progressive warm colors starting at the tip and sample interface vs. cooler blue to violet colors for that of the wide bore tip. The more ellipsoidal contours apparent on either side of the tip to sample interface with DI water vs. 80% glycerol are due to the higher velocities or faster [default software] flow rates used for water with Biomek Liquid Handling Workstations and modeled accordingly for this study.

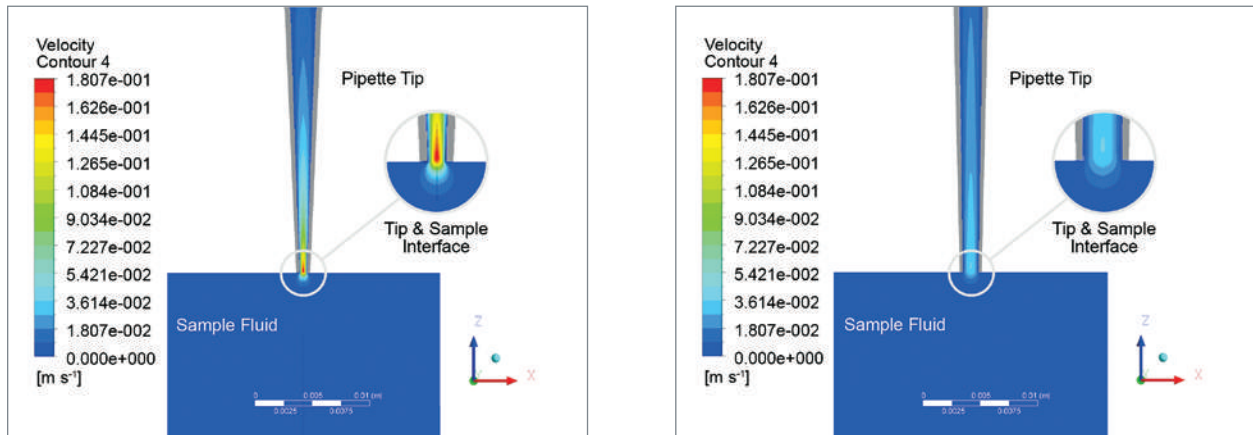


Figure 3. Fluid (sample) velocity through the pipette tip’s orifice for a standard orifice pipette tip (left), and a wide bore orifice pipette tip (right) using 80% glycerol as the model sample fluid.

Effect on Shear Strain Rate

Visualizations of the reduction in shear strain rates for DI water and 80% glycerol are shown in Figures 4 and 5, respectively. The color scale and associated color images of the pipette tips are defined by the progressively warmer colors representing increasing shear strain rates; e.g., green to yellow to orange to red. The progressively cooler colors represent decreasing shear strain rates; e.g., green to blue to indigo to violet.

Figure 4 shows the shear strain rate contours on the inner wall for a standard orifice pipette tip (left) and a wide bore pipette tip (right). The standard orifice pipette tip produces a comparatively higher shear strain rate at the tip and sample interface in relation to a wide bore orifice; indicated by the “hot spot” of red and migrating warm colors as the fluid is aspirated into the tip vs. cooler blue to violet colors for that of the wide bore tip.

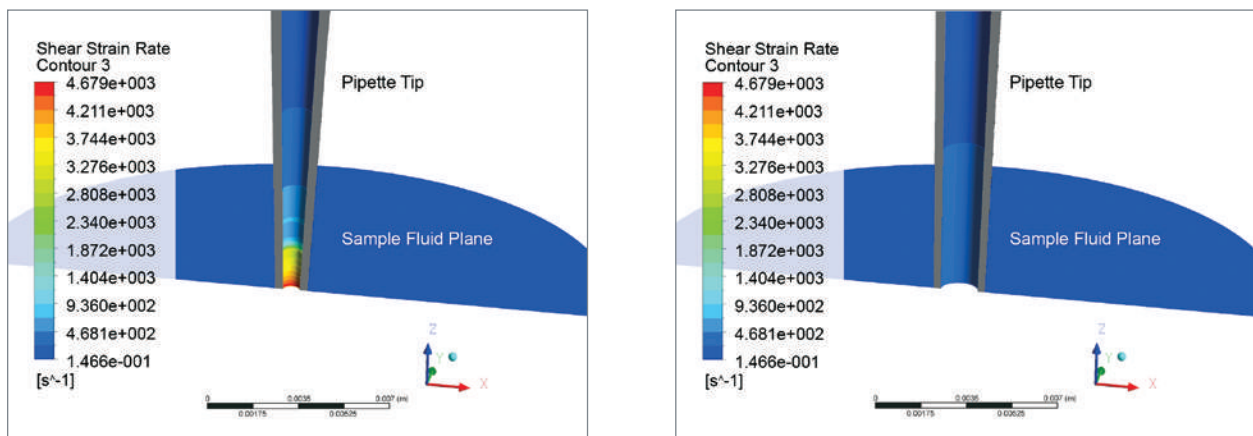


Figure 4. Shear strain rate at the pipette tip’s inner wall for a standard orifice pipette tip (left), and a wide bore orifice pipette tip (right) using DI water as the model sample fluid.

Figure 5, representing the shear strain rate contours of 80% glycerol, shows similar comparative results to DI water (Figure 4) for a standard orifice pipette tip (left) and a wide bore pipette tip (right). The standard orifice pipette tip produces a comparatively higher shear strain rate at the tip and sample interface in relation to a wide bore orifice; indicated by the “warmer” yellowish-green color at the tip and sample interface vs. cooler indigo to violet colors for that of the wide bore tip.

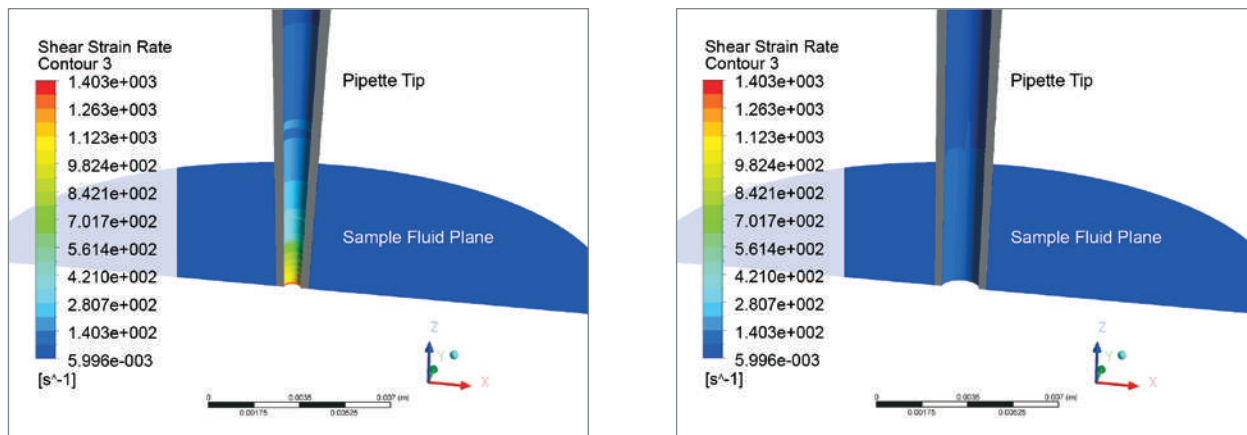


Figure 5. Shear strain rate at the pipette tip's inner wall for a standard orifice pipette tip (left) and a wide bore orifice tip (right) using 80% glycerol as the model sample fluid.

Conclusions

Computational Fluid Dynamics (CFD) software is a powerful tool for simulating physical forces and flow characteristics within a pipette tip during pipetting. The product development tool not only is invaluable for predicting the impact of fluid behavior within a given design but also provides high-quality visualizations and enables quantitative analysis to be conducted on the simulation models.

The results of this study demonstrate significant differences in fluid (sample) velocities and shear strain rates on the inside of the pipette tips for both DI water and 80% glycerol. The larger orifice, wide bore pipette tips reduced the velocities and shear strain rates for both fluid types. These reduced flow characteristics can be applied to the pipetting of biological sample matrices where the loss of biological sample integrity can occur as reported in the scientific literature at large.

In addition to minimizing disruption of sample integrity, Biomek Wide Bore Pipette Tips reduce sample preparation processing times by allowing faster pipetting speeds. At twice the internal diameter of a standard orifice tip, Biomek Wide Bore Tips can transfer viscous reagent solutions, such as glycerol, from reservoir to destination labware much more efficiently and expediently. The larger diameter bore also minimizes tip clogging when pipetting particulate-laden or fibrous sample matrices as found in biomass studies.

¹As common as the term “wide bore” is in the literature, there is no industry standard, which defines what dimensional size(s) this represents. Life Technologies' Applied Biosystems defines wide bore pipette tips on their website as “> 1 mm.” Further reference to the cutting of pipette tips to achieve a wide bore tip feature occurs in Ambion's “Genomic DNA Preparation from RNAlater Preserved Tissues” and Gibco BRL's DNAzol Reagent product insert. In comparison to hypodermic needles, the industry-recognized reference series *Current Protocols in Molecular Biology* (1994) 2.3.1-2.3.7, John Wiley & Sons, Inc., specifies the use of a “large-bore (15-G) collecting needle” for the purification of genomic DNA from plant tissue. A 15-G (or 15 gauge) needle has an internal diameter of 1.37 mm.