TECHNICAL BULLETIN

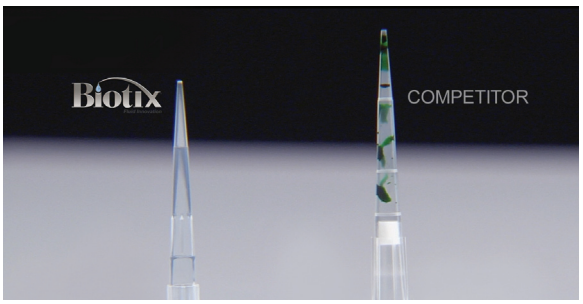
X-Resin™

Setting a New Standard in Sample Delivery

INTRODUCTION

The high cost of rare biological samples, coupled with the increasing sensitivity of today's molecular methods, demands high precision pipetting by laboratory technicians[1]. Even minor variations in sample delivery can impact data quality for expression analysis[2] and next generation sequencing[3] technologies. For a number of years, pipette tip manufacturers have tried to market the efficiency of their tips for sample delivery through gravimetric studies using water, viscous dye comparisons, and even high resolution photos showing "smoothness" of the inner tip wall. These methods, however, fail to provide quantitative assessment of tip efficiency for delivery of molecules in a given substrate. In essence, are sample molecules left behind in tips due to inherent retention of plastic?

In an extensive comparison against the leading pipette tip brands on the market, Biotix tips with X-Resin technology proved to be the most effective in preventing sample loss during pipetting.



This technical bulletin provides an overview of data generated from a comprehensive study conducted by MRIGlobal*, a leading national research institute. The study design consisted of evaluating sample loss following dispense of three different sample types: fluorescently labeled DNA; fluorescently labeled protein; and nanoparticles utilizing nanoceria with detection by ICP-MS. An electronic pipettor was utilized to assure equivalent handling of liquids for each brand of tip, thereby limiting user introduced variance. Samples were processed in triplicate and then analyzed in duplicate.

Table 1: Pipette Tips Used in Study

Manufacturer	Part Number	Lot Number	Description
Biotix	M-0100-9FC	0309 65464	100 µl Filter Pipette Tip, Sterile
Competitor A	TF-100-L-R-S	302-28-151	100 µl MAXYmum Recovery Filter Tip, Sterile
Competitor M	2065	09440809	ART™ 100 µl Filtered Pipette Tip, Sterile

*MRIGlobal is an independent, not-for-profit research organization. More information at www.MRIGlobal.org

Biotix™ Fluid Innovation

DNA Challenge

Methods

Human DNA diluted to 20 µg/ml was used in the DNA challenge. The DNA solution was labeled with fluorescent dye as per Invitrogen's Qubit dsDNA HS Assay Kit (Cat. #Q32851) protocol. One hundred microliters of the fluorescent DNA solution was drawn up and down with the pipettor 3 full times, with a final dispense back to the original tube. One hundred microliters of molecular grade dH_2O was drawn up and down 3 times in the tip and dispensed into a fresh 0.5 ml tube. This procedure was repeated in triplicate for each of the three brand pipette tips. DNA solutions were analyzed on the Qubit 2.0 Fluorometer for residual fluorescent signal associated with retention of DNA solutions on the pipette tip. A reading of "Too Low" indicates less than 0.010 µg/ml dsDNA detected by the fluorometer.

Results

There was a distinctive and measureable difference in sample loss due to residual DNA solution left in the tips following dispensing of the sample (Table 2 and Figure 1). Loss of sample due to residual DNA left was 0.25%, 0.71% and 1.75% for Biotix, Competitor A and Competitor M tips, respectively. Biotix tips demonstrated the best consistency and efficiency of DNA sample delivery among the three brands.

Figure 1:
Graph of Residual DNA Carryover on Tips

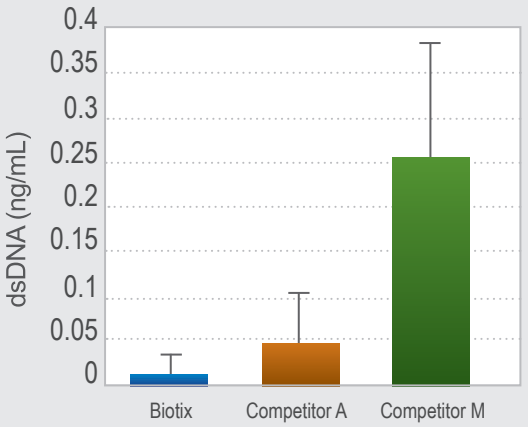


Table 2: Residual DNA Carryover

Sample	5 µl	10 µl	Mean (µg/ml)	SD
Negative Control	Too Low	Too Low	Too Low	Too Low
DNA 20 µg/ml	22.000	11.000	16.500	7.778
Biotix – 1	0.036	0.047	0.042	0.008
Biotix – 2	Too Low	Too Low	Too Low	Too Low
Biotix – 3	Too Low	Too Low	Too Low	Too Low
Competitor A – 1	0.119	0.115	0.117	0.003
Competitor A – 2	Too Low	Too Low	Too Low	Too Low
Competitor A – 3	0.028	0.426	0.035	0.010
Competitor M – 1	0.130	0.448	0.289	0.225
Competitor M – 2	0.367	0.203	0.285	0.116
Competitor M – 3	0.205	0.185	0.195	0.014

Protein Challenge

Methods

Bovine Serum Albumin (BSA) at 5 mg/ml was used in the protein challenge. The BSA solution was labeled with fluorescent dye as per Invitrogen's Qubit Protein Assay Kit (Catalog # Q332111) protocol. Fluorescent BSA was pipette up and down three full times, with final dispense back to the original tube. Next, one hundred microliters of molecular grade dH_2O was pipetted up and down three times in the tip, and then dispensed into a fresh 0.5 ml tube. The procedure was repeated in triplicate for each of the three brand pipette tips. The Qubit 2.0 Fluorometer was used to measure any residual fluorescent signal associated with retention of the protein solutions on the pipette tips. In the data generated, "Too High" represented samples with greater than 5 mg/ml, and "Too Low" represented samples with less than 12.5 µg/ml.

Results

As observed in the DNA analysis, there was measurable sample loss due to residual protein solution left in tips following dispensing of the sample (Table 3 and Figure 2). The greatest average measured loss of sample due to residual solution left on the tip was 0.32%, 1.47% and 1.87% for Biotix, Competitor A and Competitor M tips, respectively. Biotix demonstrated the least amount of protein sample loss in this analysis.

Figure 2:
Graph of Residual Protein Carryover

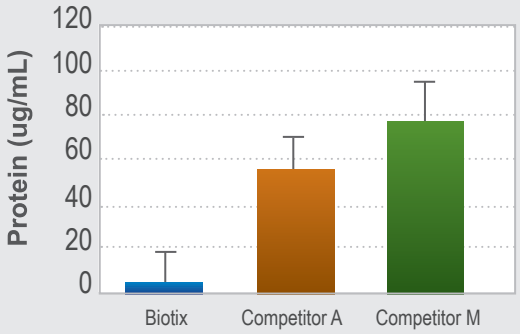


Table 3: Residual Protein Carryover

Sample	5 µl	10 µl	Mean (µg/ml)	SD
Negative Control	Too Low	Too Low	Too Low	Too Low
BSA 5mg/ml	Too High	Too High	Too High	Too High
Biotix – 1	Too Low	Too Low	Too Low	Too Low
Biotix – 2	Too Low	31.9	Too Low	Too Low
Biotix – 3	Too Low	Too Low	Too Low	Too Low
Competitor A – 1	52.5	39.5	46	9.192
Competitor A – 2	50.8	48.2	49.5	1.838
Competitor A – 3	76.7	69.6	73.15	5.02
Competitor M – 1	83.5	83.4	83.45	0.071
Competitor M – 2	58	52.2	55.1	4.101
Competitor M – 3	97.3	89.7	93.5	5.374

Nanoparticle Challenge

Methods

Cerium oxide nanoparticles (30 nm) at 82,000 ng/ml was used in the nanoparticle challenge. The CeO_3 solution was pipetted up and down three full times, with final dispense back to the original tube. Then, using the same tip, one hundred microliters of molecular grade dH_2O was pipetted up and down three times and dispensed into a fresh 0.5 ml tube. This procedure was repeated in triplicate for each of the three brands of tips. The wash solutions were analyzed on an Inductively Coupled Plasma Mass Spectrometer (ICP-MS) for residual CeO_3 associated with retention of the solution on the pipette tip.

Results

There was a measurable difference in sample loss due to residual cerium nanoparticles in solution left on the tips following dispensing of the samples (Table 4 and Figure 3). The greatest average measured loss of sample due to residual solution left in the tips was 0.005%, 0.02% and 0.06% for Biotix, Competitor A and Competitor M tips respectively. Biotix tips demonstrated the least amount of nanoparticle sample loss in this challenge.

Figure 3: Graph of CeO_3 Carryover

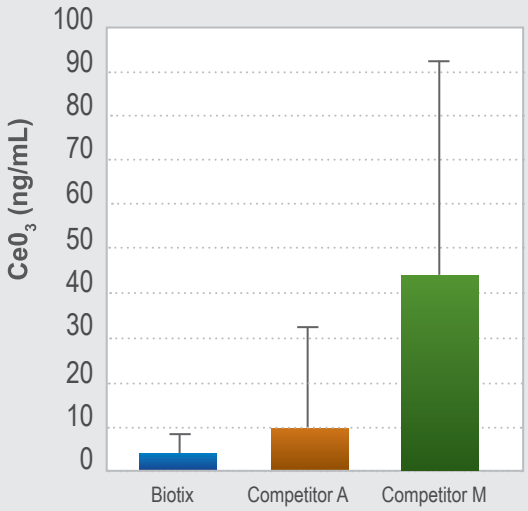


Table 4: Residual CeO_3 Carryover

Sample	Replicates	Mean (ng/ml)	SD	Range
Biotix	3	4.328	4.019	0.433 – 8.49
Competitor A	3	17.287	14.556	1.36 – 29.9
Competitor M	3	44.107	48.700	9.52 – 99.8
Negative Control	2	>0.019	N/A	N/A
Positive Control	2	73545.5	8703.777	67391 – 79700

Testing performed by independent research laboratory, MRIGlobal, showed Biotix brand pipette tips to have superior performance in sample delivery as tested across three unique sample types: fluorescently labeled DNA; fluorescently labeled protein; and nanoparticles. Compared with competitive tips in this study, Biotix tips consistently had the least amount of sample retention in all three challenges. When working with precious samples or using advanced technologies that are sensitive to minor variations in pipetting, Biotix should be the tip of choice to ensure assay performance and data accuracy.

For more information about Biotix pipette tips featuring X-Resin, visit us at our website or contact us at the address below.

References

(1) Curry, D., MHale, C., and Smith, M., "Factors Influencing Real-Time RT-PCR Results: Application of Real-Time RT-PCR for the Detection of Leukemia Translocations", Molecular Biology Today, (2002) 3: 81.

(2) Applied Biosystems, "User Bulletin #2", ABI Prism™ 7700 Sequence Detection System, Oct. 1, 2001, pp.3.

(3) Illumina, Inc., "Liquid Handling", TruSeq™ Sample Preparation Best Practices and Troubleshooting Guide, June 2011, pp. 4

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