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Low Retention Tips

Application Note

Performance comparison of low retention pipette tips from various manufacturers

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Abstract
Pipetting accuracy and precision are the key factors for successful experiments in the laboratory. The best pipetting performance is achieved, when the pipette and the tip match perfectly with each other, and the tip being used is optimal for the liquid being transferred.

When pipetting liquids with low surface tension (e.g. detergents), they tend to leave a film of liquid on the inner surface of standard polypropylene pipette tips, leading to inconsistency and inaccuracy in pipetting as well as loss in volume of valuable samples.

Various technologies are being used to modify the standard pipette tips to mitigate this issue. This application note describes the comparison of various manufacturers' low retention tips, when handling liquids that contain detergents. The results show that Sartorius Low Retention Tips ensure near complete recovery of the sample, when pipetting liquids of low surface tension. When comparing to other low retention tips from various manufacturers, Sartorius tips secured the best sample recovery and the best chemical resistance.

Introduction
In many molecular biology applications the increased sensitivity of the detection methods call for extreme reliability and reproducibility in pipetting. In DNA and protein analysis methods the reagents and/or the samples often contain detergents. Pipetting liquids that contain detergents can be problematic, when using standard pipette tips. Often residue of liquid remains in the tip due to differences in surface energies between the sample and the plastic pipette tip. This, sometimes invisible, film on the tip's inner surface causes imprecision in pipetting, and a loss of valuable samples or expensive reagents.
Sartorius has utilized an advanced technology to create an extremely hydrophobic yet durable surface on their Low Retention Tips. This feature helps users to clearly reduce the amount of residue in the tip, when handling detergents or other liquids that have a low surface tension. Better reproducibility in pipetting is especially beneficial in sensitive applications like PCR, or real-time PCR.

In this application note we:
1. compare various pipette tips with low retention feature, when pipetting common detergent solutions used in the molecular biology laboratories.
2. show the results of a test, where the chemical resistances of various manufacturers' low retention tips have been compared.

Materials and methods
Materials
- Sartorius Optift and SafetySpace™ standard and Low Retention Tips: 120 µl, 200 µl, 350 µl, 1000 µl
- Sartorius Picus electronic pipettes: 5-120 µl, 10-300 µl, 50-1000 µl, aspirating at speed 4, dispensing at slowest speed 1
- Sartorius mLINE® mechanical pipettes: 20-200 µl, 100-1000 µl
- Detergent solutions: 0.1% Triton X-100, 10% Tween 20, 10% SDS, Colored 10X PCR buffer (contains detergents, density reagent and tracking dyes)
- Other chemicals: Isopropanol, Acetonitrile, DMF (dimethylformamide)
- Green food dye
- Low retention pipette tips from five other manufacturers
- Sartorius MC5 Microbalance, Sartorius Analytical Balance BP211D
- Spectrophotometer for 96-well microplates (Biotek)

Gravimetric method
The liquid residue remaining in the pipette tip, after dispensing, was measured using the gravimetric method. A small glass container filled with a test solution was placed on the balance and the weight was set to zero. A desired volume of the test solution was then aspirated from the container and dispensed back. The balance reading, indicating the amount of liquid that remained in the tip, was then recorded. This was repeated for each detergent solution listed above.

Absorbance test
In the absorbance test a colored test solution (green food dye dissolved in distilled water) was used to determine the residual liquid in the pipette tips after dispensing. The maximum nominal volume of the tested tip was used to aspirate the green test solution. The liquid was then dispensed directly back into its container. The tip was then rinsed five times with distilled water using the maximum volume of the tip. The absorbance of this solution was then measured using a spectrophotometer (405 nm), and the results were compared to the reference solution. The absorbance of the tip rinsing solution directly correlates to the amount of residue in the tip.

Test for chemical resistance

1000 µl of solvents: Isopropanol, Acetonitrile and Dimethylformamide, were aspirated and dispensed 20 times each with a 1000 µl pipette tip. Afterwards, the tips were rinsed three times with distilled water. The effect of this treatment to the performance of the selected low retention tips was analyzed with the absorbance test using colored liquid as a test solution. The test was repeated with 6 tips for every solvent. The results of the chemically treated tips were compared to non-treated standard tips and non-treated low retention tips.

Results

Comparison of residual liquid volumes

The performance of 5 other manufacturers' low retention tips were compared with Sartorius' standard and Low Retention Tips, when pipetting commonly used detergent solutions. With all the liquids tested, Sartorius Low Retention Tips retained the least amount of residue (Figure 1a). Some of the competitors' low retention tips performed worse than even Sartorius' standard tips, suggesting that there are significant differences in performance among low retention tips available on the market.

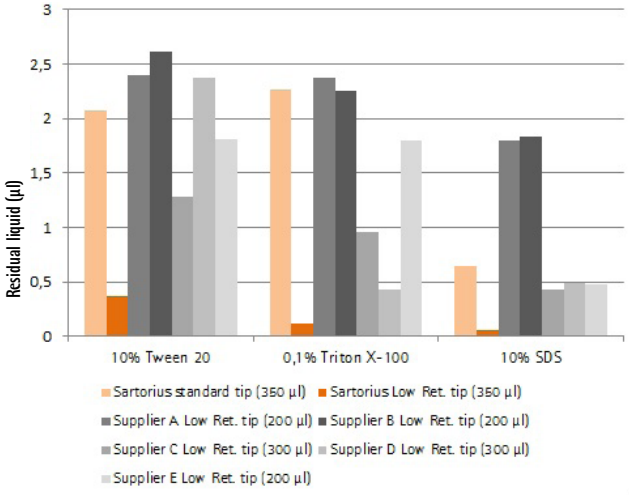

Figure 1a. Comparison of residual liquid amounts.

Low retention tips from five manufacturers were compared with Sartorius standard and Low Retention Tips, when pipetting 200 µl of the following detergents: 10% Tween 20, 0,1% Triton X-100 and 10% SDS. Pipette tips of volumes 200 µl, 300 µl and 350 µl (size depending on the manufacturers' offering and compatibility) were used with Picus electronic pipette 10-300 µl. The remaining liquid amount in the tip was measured using the gravimetric analysis method described in the Materials and methods. The test was repeated for 10 tips of each manufacturer.

In another experiment a colored detergent-containing PCR buffer was used as a test solution to compare pipetting precision of various low retention tips and to see how the pipetting volume affects liquid retention in the tip. As shown in Figure 1b, Sartorius Low Retention Tips retained the least amount of residue with both volumes. Even Sartorius standard tips retained similar levels of residue as some of the competitors' low retention tips. The use of Sartorius Low Retention Tips resulted in best pipetting precision, supporting the benefits of these tips e.g. in PCR setup. The data also shows that the differences in residual liquid amount between the standard and the low retention tips grow with the increase in pipetted volumes. This is due to the liquid adhering to a larger surface area inside the tip.

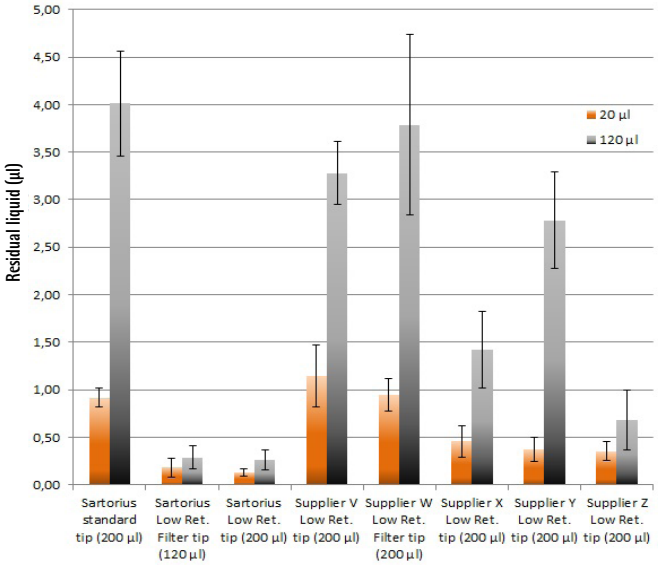

Figure 1b. Comparison of residual liquid amounts and pipetting precision. A coloured PCR buffer was used in 2 volumes.

Low retention tips from five other manufacturers were compared with Sartorius standard and Low Retention Tips (filtered and non-filtered). Pipette tips of volume 120 µl, 200 µl and 300 µl (size depending on each manufacturers' offering and compatibility) were used with Sartorius mLINE® 20-200 µl to aspirate and dispense both 20 µl and 120 µl of a colored PCR buffer. The remaining liquid amount in the tip was measured using the gravimetric analysis method described in Materials and methods. The test was repeated for 10 tips of each manufacturer. The error bars show the standard deviations.

Effect of surface tension

The effect of liquid surface tension on the amount of residue left in the tip, after dispensing, was tested by using various concentrations of Isopropanol, which has a low surface tension (23 mN/m) (Figure 2). The stronger the Isopropanol concentration and the lower the surface tension, the more beneficial it was to use highly hydrophobic low retention tips rather than standard pipette tips to minimize the reagent loss and pipetting imprecision. Similar benefits could not be seen with water or other aqueous solutions, due to the larger differences in surface energies between the highly hydrophobic low retention tip and water (72 mN/m).

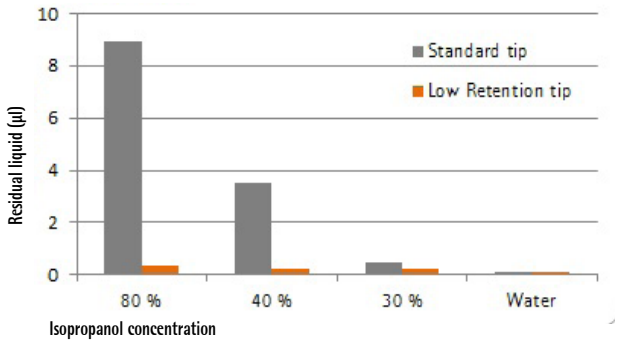

Figure 2. Effect of liquid surface tension to the amount of residue in the tip.

1000 µl of various concentrations of Isopropanol (30%, 40%, 80%) and distilled water were aspirated and dispensed with both Sartorius standard and Low Retention Optifit Tips (1000 µl) using Sartorius Picus electronic pipette (1000 µl). The residual liquid in the tips was measured using the gravimetric analysis method described in the Materials and methods. The test was repeated for 10 tips of each manufacturer.

Chemical resistance of low retention pipette tips

Several technologies are being used to create low retention surfaces to pipette tips. The most stable methods produce tips, which have complete coverage in terms of hydrophobicity, and are non-leaching. As shown in the Figure 3, there is a significant variance in the tested low retention tips in terms of chemical resistance. With some of the competitor tips the low retention feature was significantly reduced after treatment with the selected solvents. The performance of Sartorius Low Retention Tips after chemical testing was at the same level as without any chemical treatment, suggesting that these tips are inert and chemically resistant. Autoclaving the Sartorius Low Retention Tip also had no effect on the performance of the tips (data not shown).

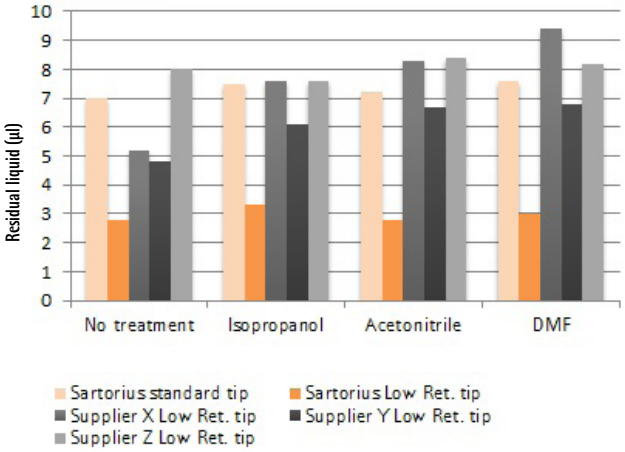

Figure 3. Comparing the chemical resistance of low retention pipette tips.

Low retention tips from three manufacturers were compared with Sartorius standard and Low Retention Tips. The test for chemical resistance was performed as described in the Materials and methods using 1000 µl pipette tips with Sartorius Picus electronic pipette (1000 µl). The test was repeated for six tips of each manufacturer.

Discussion

The test results show that Sartorius Low Retention Tips clearly reduce the liquid residue in the tip, when handling detergents, or other liquids with low surface tension. The data also suggests that the low retention tips available on the market can differ quite significantly in performance and chemical tolerance. Out of the various tips tested, Sartorius Low Retention Tips secured the best sample recoveries, best precision and the best chemical resistance.

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