

Performance of Low Retention GripTips from INTEGRA

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Abstract

Accuracy and precision are of key importance in pipetting applications. It has been demonstrated on several occasions that the best results are obtained by following the recommended pipetting techniques, and working with an optimized system where tips and pipettes have been designed to perfectly fit together. In this context, the nature of the liquid to be pipetted and its intrinsic physicochemical properties should not be neglected. Standard polypropylene (PP) tips that provide ideal accuracy, precision and liquid recovery when pipetting water may produce significantly different results when other substances are handled. Liquids containing detergents

or other low surface tension solutions form a thin film on the inner surface of the pipette tips, leading to pipetting inaccuracy, non-repeatability and, even worse, the loss of expensive or very precious reagents and samples. To overcome this issue, INTEGRA has introduced Low Retention GripTips, which prevent low surface tension samples from spreading out across the inner wall of the tips. Used in combination with INTEGRA's range of pipettes, this ensures optimal liquid recovery and best pipetting results. This application guide illustrates the advantages of using Low Retention GripTips when pipetting low surface tension liquids.

Why use low retention tips instead of standard tips?

Polypropylene is the material of choice to produce pipette tips because of its hydrophobic properties and low surface energy. A liquid that has a high surface tension – such as water – will have a low affinity for polypropylene, because the cohesive forces inside the liquid (blue arrows, Figure 1a) are stronger than the adhesive forces between the liquid and the polypropylene surface. As a result, the liquid will form a drop that will be repelled by the surface (**Figure 1a**).

In contrast, if a liquid has a low surface tension or low interactive forces inside the liquid, for instance olive oil, it will have a stronger affinity for polypropylene. As a result, the liquid will spread out and wet the polypropylene surface (**Figure 1b**).

Standard polypropylene tips are therefore the optimal solution for pipetting water, since water simply pearls off on the tip surface during dispensing.

However, some life science applications involve the use of viscous samples, detergents and other low surface tension liquids that tend to interact with standard pipette tips and form a thin liquid film on the on the inner wall of the pipette tip (**Figure 1c**). This effect leads to pipetting inaccuracy and inconsistency as well as loss of precious reagents.

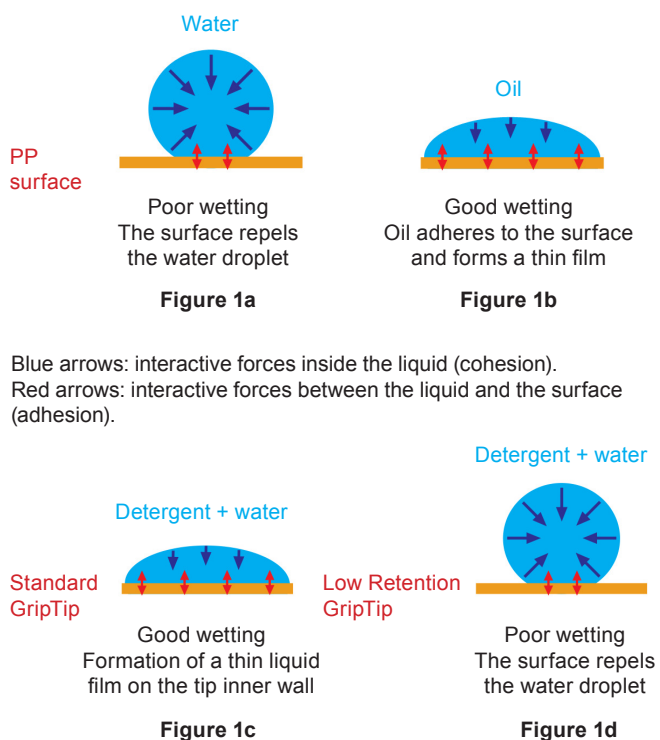


Figure 1: Illustration of the interactive forces inside a liquid and between a liquid and a surface in four different configurations.

This specific scenario can occur in several applications, for instance in cell culture, PCR and qPCR, DNA sequencing, cloning, protein analysis or PAGE. It can also be observed in all other pipetting steps involving the use of viscous or low surface tension liquids such as TRIS, Tween®, isopropanol, Master Mix, Triton X-100, SDS, whole blood, plasma and much more.

One way to overcome this issue is to modify the tip material in order to reduce its surface energy and thus the affinity of low surface tension liquids with the tip surface. Low retention tips prevent low surface tension samples from spreading out and 'wetting' the inner wall of the tips, allowing them to beadup for maximum liquid recovery (Figure 1d).

Performance of Low Retention GripTips versus standard GripTips

GripTip pipette tips have been designed in combination with INTEGRA's manual and electronic pipettes to ensure a perfect pipetting system. GripTips snap into place with minimal tip loading effort, providing a secure connection. They never fall off and are always perfectly aligned, resulting in superior accuracy and precision results.

INTEGRA has introduced new Low Retention GripTips to meet the need for tips that can handle viscous and low surface tension liquids with the same accuracy and reliability as that obtained when using INTEGRA pipettes and standard GripTips with standard water-based liquids.

A simple visual check using a 300 µl 12 channel VIAFLO electronic pipette to pipette a concentrated green dye demonstrates the performance of the Low Retention GripTip (Figure 2).

Comparing residual liquid amounts

Multiple tests were performed in the controlled environment of a calibration lab to demonstrate the efficiency of Low Retention GripTips compared with standard GripTips. Using on a gravimetric method, the residual liquid amounts in both types of pipette tip were compared after dispensing a full volume of a low surface tension liquid.

In the first series of tests, we evaluated the performance of 300 µl Low Retention GripTips, compared with standard GripTips of the same volume when pipetting three common types of low surface tension liquids: 10 % Tween 20 in distilled water, 10 % SDS in distilled water and 80 % isopropanol in distilled water. For this purpose, we used a 300 µl single channel VIAFLO electronic pipette with the following custom program: aspirate full volume of the pipette at speed 5 and dispense the full volume at speed 2. Results were obtained by differential weighing of tested tips. Each measurement was repeated 10 times per test solution and GripTip type to ensure the consistency of the results.

Low retention tip manufacturers usually choose between two manufacturing processes, involving either a silicone coating technique or the use of a polypropylene blend.

A blended polypropylene process is preferred over common silicone coating techniques, which can wash out or leach out with the sample and negatively impact results.

Therefore, INTEGRA has molded Low Retention GripTips from a unique polypropylene blend with heightened hydrophobic properties.



Figure 2: Pipetting a concentrated green dye with a 300 µl 12 channel VIAFLO electronic pipette featured with six standard GripTips (left) and six Low Retention GripTips (right).

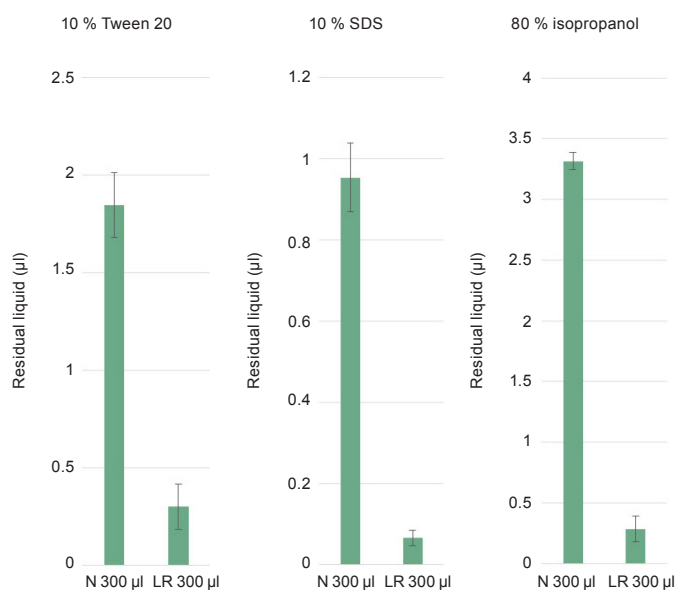


Figure 3: Averages and error bars of residual liquid amounts (µl) after dispensing solutions of 10 % Tween 20 (left), 10 % SDS (middle) and 80 % isopropanol solution (right) with 300 µl standard GripTips (N) and Low Retention GripTips (LR).

Figure 3 presents the results for the three solutions tested, highlighting the average residual liquid amounts in each type of tip together with the corresponding error bars.

We also calculated the residual liquid volumes as a percentage of the total volume that aspirated and dispensed, for each solution and GripTip configuration. This allowed us to get an understanding of the tested Low Retention GripTips in comparison with their standard version from a different perspective.

The values obtained are presented in **Table 1** and highlight the improvements in terms of maximum liquid recovery when using Low Retention GripTips with different types of low surface tension mixtures.

As a conclusion, this first series of experiments demonstrates the advantages of using Low Retention GripTips when pipetting standard low surface tension solutions. The quantity of liquid remaining in the tips is significantly and reproducibly reduced, providing an enhanced solution for users looking for accurate pipetting with a maximum liquid recovery.

	N 300 µl	LR 300 µl
10 % Tween 20	0.62 %	0.10 %
10 % SDS	0.32 %	0.02 %
80 % isopropanol	1.11 %	0.10 %

Table 1: Residual liquid volumes (µl) as a percentage of the full volume pipetted with 300 µl standard GripTips (N) and Low Retention GripTips (LR). Tested solutions: 10 % Tween 20, 10 % SDS and 80 % isopropanol in water.

Efficiency of Low Retention GripTips of each volume range

A second series of tests was performed to extend the results across the full volume range of Low Retention GripTips: 12.5, 125, 300 and 1250 µl.

The residual amounts of liquid in the standard and Low Retention GripTips were compared after dispensing a full volume of a 10 % solution of Tween 20 in distilled water. A single channel VIAFLO electronic pipette of each volume was used with the same custom program as in the first series of tests. Measurements were repeated 12 times for each tip volume in order to ensure the repeatability and accuracy of the results.

Figure 4 shows the average residual liquid amounts given in µl and the corresponding deviations for the four GripTip volumes in their standard and Low Retention versions.

The different charts indicate the efficiency of Low Retention GripTips in comparison with standard tips throughout the entire product range.

In **Table 2**, the residual liquid volumes were calculated as a percentage of the total volume pipetted. For each tip size, we observed a significant decrease in the volume of liquid remaining in tips when using Low Retention GripTips.

We can therefore confirm the efficiency of Low Retention GripTips independently of their size and shape. They provide scientists with an optimal solution to handle low surface energy liquids with accuracy and precision, with maximum liquid recovery.

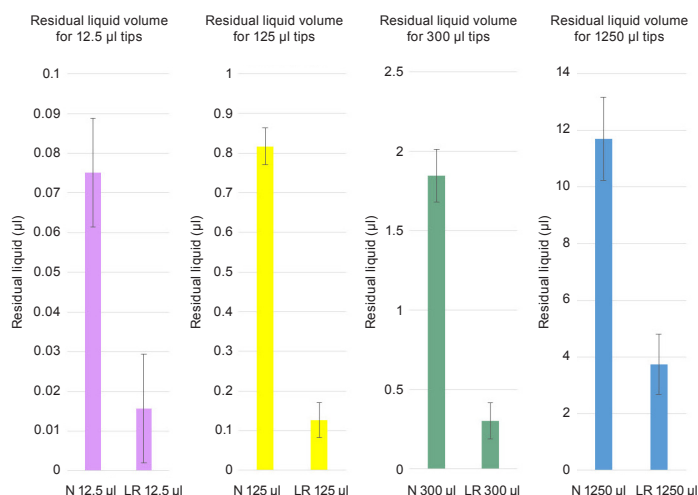


Figure 4: Comparison of the residual liquid amounts in standard GripTips (N) and sterile Low Retention GripTips (LR) of all available volume ranges after pipetting a solution of 10 % Tween 20 in distilled water. The graphs show the average liquid amount in µl and corresponding error bars. 12 tips of each type were tested.

	N	LR
12.5 µl	0.60 %	0.13 %
125 µl	0.65 %	0.10 %
300 µl	0.62 %	0.10 %
1250 µl	0.94 %	0.30 %

Table 2: Residual liquid volumes (µl) given in percentage of the full volume that has been pipetted with standard GripTips (N) and Low Retention GripTips (LR). Tested volumes: 12.5, 125, 300 and 1250 µl. Tested solution: 10 % Tween 20 in distilled water

Effects of surface tension

The effect of surface tension is usually evaluated by pipetting various concentrations of isopropanol in water. This alcohol has a low surface tension (23.00 mN/m at 20 °C) in comparison with water (72.80 mN/m at 20 °C). Test solutions of different surface tensions can easily be prepared by modifying the volume ratio of isopropanol and water.

This method was used to observe the influence of varying liquid surface tension on Low Retention GripTips and to compare it with standard GripTips.

We used 300 µl standard GripTips and Low Retention GripTips together with a single channel VIAFLO electronic pipette and the previously described custom program. Solutions of 40, 60 and 80 % isopropanol in distilled water were prepared and residual liquid amounts in tips were measured gravimetrically. For each configuration, the experiment was repeated 10 times to ensure the quality and consistency of our data.

Results of this test series are presented in **Figure 5**. When using standard GripTips, we observe a strong influence of the liquid surface tension on the quantity of liquid remaining in the tips, with a maximum of 2.78 mg of residual liquid after having pipetted a solution of 80 % isopropanol in water. In the case of Low Retention GripTips, the maximum remaining liquid amount in tips was 0.26 mg. The advantage of choosing Low Retention GripTips over standard GripTips is clearly highlighted by these measurements when pipetting low surface tension solutions.

Between 0 and 40 % isopropanol in water, the retention effects of standard GripTips lessen. The results are then close to the values obtained when using Low Retention GripTips. This observation can easily be explained by the fact that the energy difference between the liquid and polypropylene is high enough

Conclusion

Liquid handling precision at a microliter level is strongly influenced by several factors, which have to be identified and controlled to ensure the expected accuracy and repeatability of many life science experiments. Best pipetting practices and optimized pipetting systems, where tips and pipettes perfectly fit together, have been developed to overcome these issues and to provide scientists with highly effective liquid handling solutions. However, standard polypropylene tips were initially designed to pipette water-based solutions, and they reach their limits when handling low surface tension liquids,

to repel the liquid on the inner wall of the pipette tip leading to a higher liquid recovery. The advantage of using Low Retention GripTips over standard GripTips is therefore reduced when pipetting high surface tension liquids.

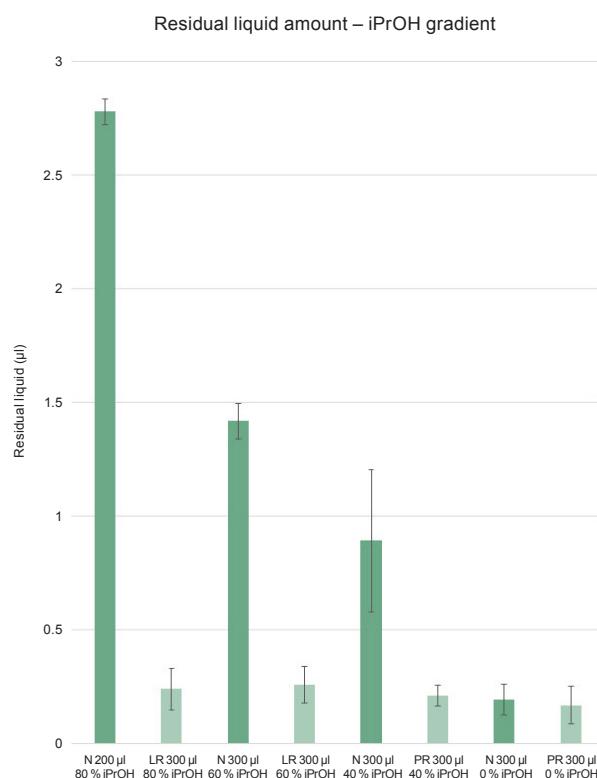


Figure 5: Comparison of the residual liquid amounts in 300 µl standard GripTips (N, dark green) and Low Retention GripTips (LR, light green) when pipetting different concentrations of isopropanol in water. The calculation of the error bars is based on 10 measurements for each experiment.

especially in terms of maximum liquid recovery. INTEGRA has introduced Low Retention GripTips to provide an overall solution for its full range of manual and electronic pipettes. The results presented in this application guide clearly show the benefits of using Low Retention GripTips over standard GripTips when pipetting low surface tension liquids, especially in terms of low amount of residual liquid. Together with INTEGRA's pipettes, Low Retention GripTips therefore offer optimal liquid recovery and best pipetting results for life sciences applications.