Application Guide

INTEGRA

Performance of Low Retention GripTips from INTEGRA

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Abstract

Accuracy and precision are of key importance in many pipetting applications. It has been demonstrated on several occasions: best results are obtained by following the recommended pipetting techniques and working with an optimized pipetting system in which tips and pipettes have been designed to perfectly fit together.
In this context, the nature of the pipetted liquid and its intrinsic physicochemical properties should not be neglected. If standard tips made out of polypropylene provide ideal accuracy, precision and liquid recovery when pipetting water, these results may be significantly different when handling other substances.
Indeed, 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 on the inner wall of the tips and ensure optimal liquid recovery and best pipetting results in combination with the full range of INTEGRA's pipettes.
This Application Guide illustrates the advantages of using Low Retention GripTips when pipetting low surface tension liquids.

Why using Low Retention Tips instead of standard Tips

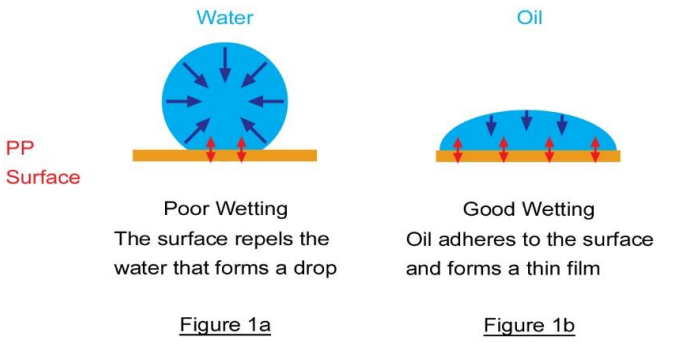
Polypropylene (PP) is the material of choice to produce pipette tips because of its hydrophobic properties and low surface energy. Indeed, a liquid having a high surface tension, as water does, will have a low affinity for polypropylene, because the interaction forces inside the liquid (blue arrows, Figure 1a) are stronger than the interaction 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).

On the contrary, if a liquid has a low surface tension - or low interaction forces inside the liquid - like 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).

Pipetting water with standard polypropylene tips is therefore the optimal solution 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 pipette tip inner wall (Figure 1c). This effect leads to pipetting inaccuracy and inconsistency as well as loss of precious reagents.

This specific case can occur in several application domains, 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.



Blue arrows: interaction forces inside the liquid (cohesion).
Red arrows: interaction forces between the liquid and the surface (adhesion).

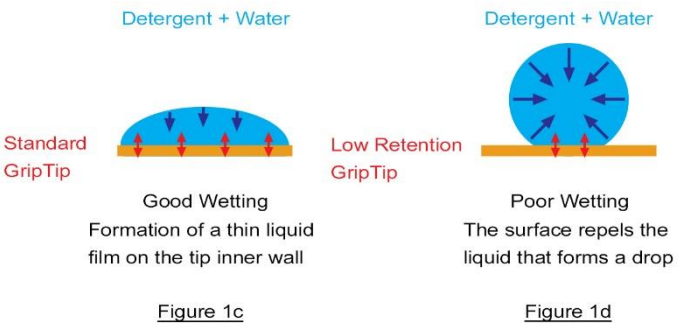


Figure 1: *Illustration of the interaction forces inside a liquid and between a liquid and a surface in four different configurations (1a, 1b, 1c and 1d).*

A 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. Known as Low Retention Tips, these tips prevent low surface tension samples from spreading out and "wetting" the inner wall of the tips, allowing them to bead-up for a maximum liquid recovery (Figure 1d).

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

However, 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 opted to use a unique polypropylene blend to mold Low Retention GripTips with heightened hydrophobic properties.

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. GripTips never fall off and are always perfectly aligned, resulting in superior accuracy and precision results.

To cover the need in handling viscous and low surface tension liquids with the same accuracy and reliability than the ones offered by INTEGRA's pipettes and standard GripTips with standard water-based liquids, INTEGRA has introduced new Low Retention GripTips.

A simple visual check using a 300 µl 12-channel VIAFLO II electronic pipette to pipet a concentrated green dye already exhibits the performance of the Low Retention GripTips. Picture 1 shows standard GripTips on the left side and Low Retention GripTips on the right.

Picture 1: *Pipetting a concentrated green dye with a 300 µl 12-channel VIAFLO II electronic pipette featured with 6 standard GripTips on the left side and 6 Low Retention GripTips on the right side.*

Comparing residual liquid amounts

To demonstrate the efficiency of Low Retention GripTips in comparison with standard GripTips, multiple tests were performed in the controlled environment of a calibration lab. Based on a gravimetric method, we compared the residual liquid amounts in both types of pipette tips after dispensing a full volume of a low surface tension liquid.

In a first series of tests, we evaluated the performance of 300 µl sterile Low Retention GripTips, in comparison 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 II 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 result consistency.

Figure 2 presents the results for the three tested solutions, highlighting the average residual liquid amounts in each type of tips together with the corresponding error bars.

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

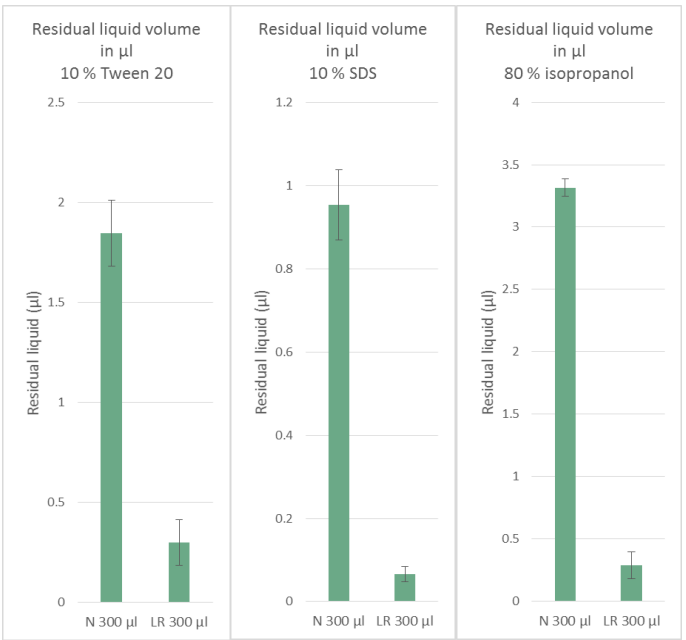


Figure 2: *Averages and error bars of residual liquid amounts (in µl) after dispensing a 10 % Tween 20 (left side), 10 % SDS (middle) and 80 % isopropanol solution (right side) with 300 µl standard GripTips (N) and sterile Low Retention GripTips (LR).*

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 remaining quantity of liquid 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 (in µl) given in percentage of the full volume that has been pipetted with 300 µl standard GripTips (N) and sterile 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 on the full volume range of Low Retention GripTips: 12.5 µl, 125 µl, 300 µl and 1250 µl.

The residual amounts of liquid in 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 II 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 GripTip volume in order to ensure the repeatability and accuracy of the results.

Figure 3 below shows the averages of residual liquid amounts given in µl and corresponding deviations for the four GripTip volumes in their standard and Low Retention sterile 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 that has been pipetted. For each size of GripTips, we observed a significant decrease of the remaining solution quantity in tips when using Low Retention GripTips.

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

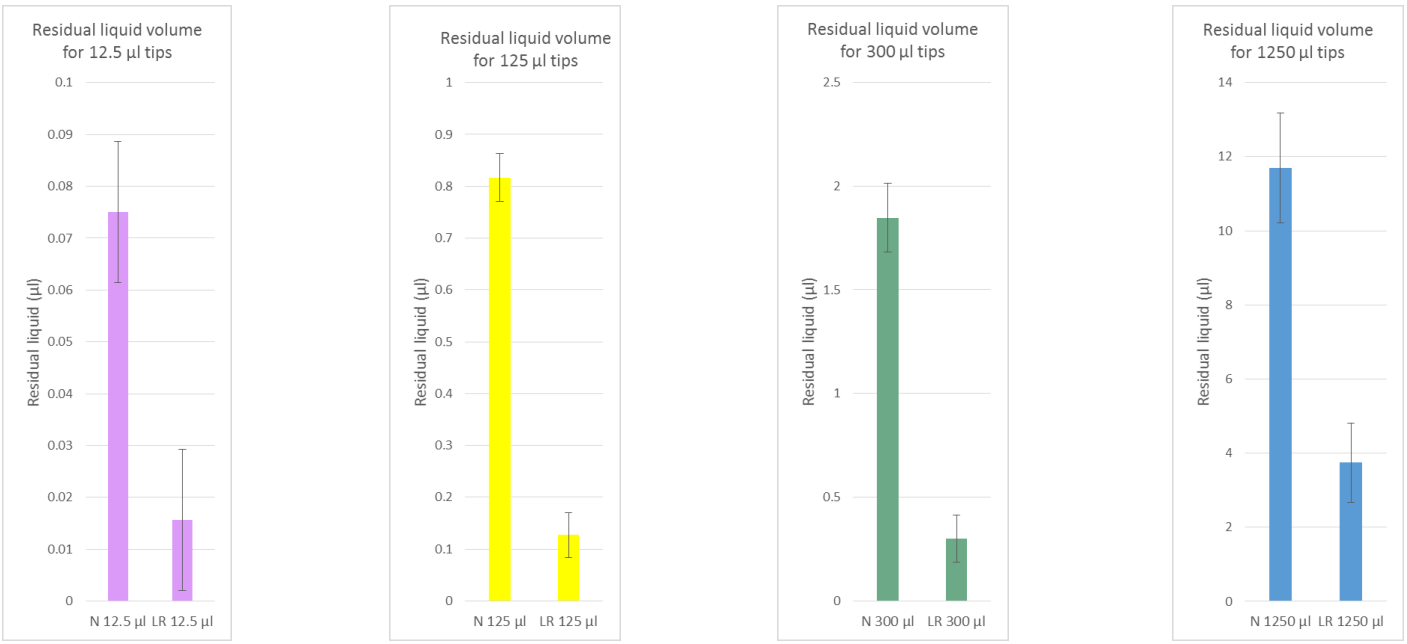


Figure 3: *Comparison of the residual liquid amounts in standard GripTips (N) and sterile Low Retention GripTips (LR) of all available volume range after pipetting a solution of 10 % Tween 20 in distilled water: average of liquid amount in µl and corresponding error bars (number of tested tips: 12 tips of each type).*

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

	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 %

Effects of liquid surface tension

Effect of surface tension is usually evaluated by testing various concentrations of isopropanol (iPrOH) in water. Indeed, 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). By modifying the volume ratio of isopropanol and water, test solutions having different surface tension values can easily be prepared.

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

To this end, we used 300 µl standard GripTips and sterile Low Retention GripTips together with a single channel VIAFLO II electronic pipette and the previously described custom program. Solutions of 0, 20, 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 ten times to ensure the quality and consistency of our data.

Results of this test series are presented in Figure 4. 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 20 % 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 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.

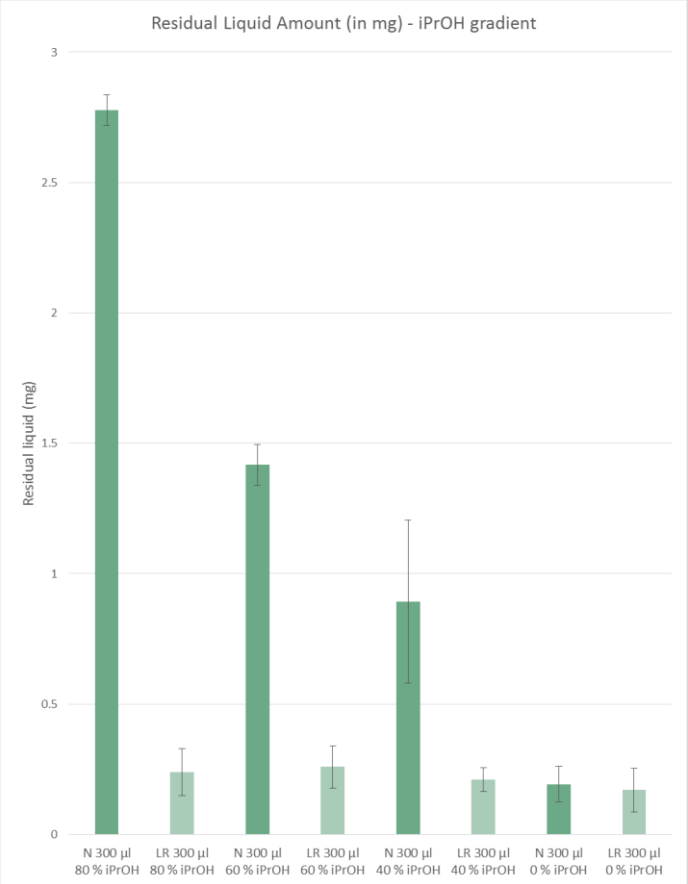


Figure 4: *Comparison of the residual liquid amounts in 300 µl standard GripTips (N, in dark green) and sterile Low Retention GripTips (LR, in light green) when pipetting different concentrations of isopropanol in water. The calculation of the error bars is based on 10 measurements for each experiment.*

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 experiments in Life Science.
Best pipetting practices and optimized pipetting systems, in which tips and pipettes perfectly fit together, have been developed to overcome these issues and to provide scientists with highly effective pipetting solutions.
However, standard tips in polypropylene were initially designed to pipette water-based solutions, and they reach their limits when handling low surface tension liquids, especially in terms of maximum liquid recovery.
To provide an overall solution for the full range of its manual and electronic pipettes, INTEGRA has introduced Low Retention GripTips.
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 residual liquid amount.
Together with INTEGRA's pipettes, Low Retention GripTips therefore offer optimal liquid recovery and best pipetting results for this domain of applications.

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