384-Channel 30 µL Tip Accuracy and Precision with Multiple Tip Attachments

Technical Overview

Summary

Precision and accuracy of the 30 µL tips are stable/constant over 100 cycles of tip attachment and detachment. Accuracy and precision are nearly as good on the 100th liquid transfer as they are on the first.

Introduction

Many laboratory automation systems run cycles of liquid handling processes. In any liquid handling process, there is a decision between fixed and disposable tips. Disposable tips have advantages in certain cases, such as eliminating carryover between plate to plate dispensing by allowing tip changes, and therefore increasing throughput time by removing the necessity to wash tips. On the other hand, disposable tip usage requires increased built-in tip capacity and potentially larger consumable and disposal costs.

For certain applications, it may be advantageous to re-use a set of disposable tips. A buffer addition or a backfill step that occurs in all plates of an automation process could potentially use a single set of tips, as there is less likelihood of cross contamination if those tips are constantly exposed to only one solvent or buffer type.

When re-using tips, it is important to consider if the process involves repeated cycles of tip attachment/detachment. This could affect the integrity of the liquid transfer. The plastic tip can become warped over repeated cycles of tip attachment and detachment, which could impact accuracy and precision.

In this study, the precision and accuracy of the Agilent 30 µL disposable tip after multiple tip attachment was explored on the Agilent Bravo Automated Liquid Handling Platform. The results demonstrate that the tip maintains very good accuracy and precision after repeated cycles of tip attachment.

Materials

- Bravo with a 384-channel ST Disposable Tip Head
- Agilent 384ST 30 µL disposable tips (product no.11484-202)
- Agilent 384-well manual fill reservoir (product no. 08104-001)
- 384-well polystyrene, clear flat bottom plates (Greiner 781101)
- Tartrazine solution, dissolved in dimethylsulfoxide [DMSO]
- UV/Vis Spectrophotometer with a 405 nm filter (Thermo Multiskan Ascent)

Method

60 mL of tartrazine solution is poured into a manual fill reservoir. The tartrazine reservoir is placed on position 2 of the Bravo. A rack of 30 µL disposable tips is placed on position 3. A 384-well polystyrene plate is placed on position 5. Agilent Works software liquid classes for 0.5-1 µL and 30 µL dispense are utilized. A VWorks protocol is created and run in the following manner:

1. Tips are pressed onto the head.
2. 1 µL (for low dispense) or 30 µL (for high dispense) tartrazine solution is aspirated from the manual fill reservoir. Aspirate parameters are 6 mm from the bottom of the plate, and preceded by 0.5 µL (for 1 µL aspirate) or 2 µL (for 30 µL aspirate) pre-aspirate volumes.
3. 1 µL (for low dispense) or 30 µL (for high dispense) tartrazine solution is dispensed into a 384-well plate (every 10th cycle) or back into the reservoir (every other cycle). Dispense parameters are 0.05 mm (for 1 µL dispense) or 0 mm (for 30 µL dispense) from the bottom of the plate, and followed by dispensing a 0.5 µL (for 1 µL dispense) or 2 µL (for 30 µL dispense) blowout volume.
4. Tips are unloaded into the tip rack.
5. Dispensed volumes are diluted to 50 µL with the addition of water. Plates are centrifuged at 1800 rpm for 60 seconds to ensure full mixing and consistent well menisci. Absorbance is read at 405 nm.

Accuracy is calculated based on an equation derived from a tartrazine/DMSO calibration curve consisting of data points at 0.3, 0.5, 1, 2, 3, 4, 5, and 6 µL (for 1 µL dispense) or 5, 10, 20, 30, 40 and 50 µL (for 30 µL dispense), compared to the actual absorbance value in each well. Absorbance values in each well are used to determine the precision of the dispense. Coefficient of Variance (CV) calculations were made by dividing the standard deviation by the mean.

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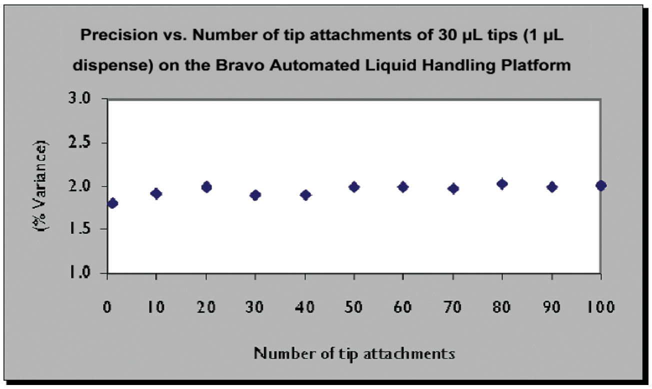
Results

Precision remains virtually unchanged (CVs of 2% and below for low volume transfers, and 1% for high volume transfers), suggesting that the integrity of the tip is maintained, and also suggesting that there is not an excess build-up of liquid in the tips between transfer cycles. Accuracy drops slightly (<3% at low volumes and <1% at higher volumes) over 100 cycles. This may be caused by the adsorption of water into the tips and reservoir, which dilutes the tartrazine concentration and causes the dispense to appear less accurate. Given that precision remains consistent over 100 cycles, the accuracy shift would appear not to be the result of changes to the tip and its seal.



Tip Attachment #	CV (%)	Accuracy (± %)
1	1.8	0.8
10	1.9	-0.3
20	2.0	-0.7
30	1.9	-1.1
40	1.9	-1.1
50	2.0	-2.3
60	2.0	-2.0
70	2.0	-2.2
80	2.0	-2.7
90	2.0	-2.9
100	2.0	-3.2

Figure 1 & Table 1. Accuracy and Precision results for 1 µL dispensing with multiple cycles of tip attachment (1-100 cycles).

Tip Attachment #	CV (%)	Accuracy (± %)
1	1.0	1.3
10	1.0	0.9
20	1.0	0.9
30	1.0	0.6
40	1.0	0.6
50	1.0	0.7
60	1.0	0.7
70	1.1	0.6
80	1.0	0.6
90	1.0	0.6
100	1.0	0.4

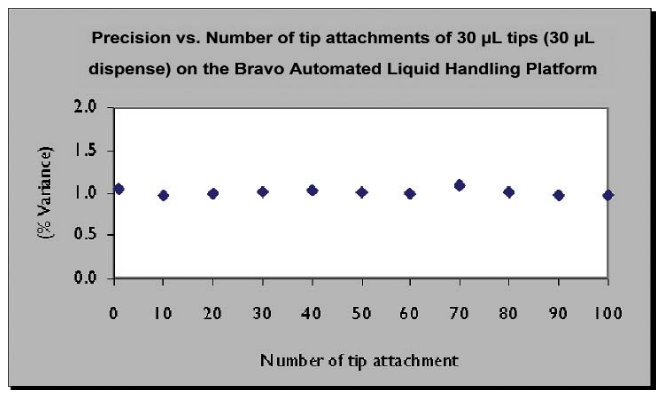


Table 2 & Figure 2. Accuracy and Precision results for 30 µL dispensing with multiple cycles of tip attachment (1-100 cycles).

Conclusion

In summary, Agilent's 30 µL disposable tips are capable in providing precise pipette transfer for both low and high dispense even after multiple tip attachments. Accuracy for 30 µL disposable tips may show a slight change for both low and high dispense after a number of tip attachments, but it still is repeatable and the fall-off is not significant.

Please contact your sales representative or Agilent Applications Support if you have particular questions regarding your specific application. Supplemental information (protocol files and data analysis spreadsheets) are also available upon request.

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