Agilent Bravo Automated Liquid Handling Platform: 96-Channel 10 µL Tip Accuracy and Precision

Technical Overview

Summary

- Bravo, 96ST head, 10 µL tips
- 0.3 µL: 3.1% CV ± 1.5% accuracy
- 0.5 µL: 2.3% CV ± 0.8% accuracy
- 2.0 µL: 1.1% CV ± 0.8% accuracy
- >2 µL: equal or better than 2.0 µL CV and accuracy

Introduction

Automated liquid handling devices are used in many processes performed in laboratories today. Liquid handling automation increases sample processing and throughput(1) and improves accuracy (desired dispensed volume is equal to the actual dispensed volume) and precision (a narrow distribution of dispensed volume) when compared to a person operating a handheld liquid pipettor. Automated liquid handling devices can also significantly reduce the occurrence of errors in a process(2). To further the effort to automate routine laboratory processes, Agilent Automation Solutions has developed the Bravo Automated Liquid Handling Platform.

Bravo Features

The Bravo has nine deck positions which accommodate any 96-, 384-, or 1536-well SBS standard microplates. Deck positions can be configured with heating, cooling, and shaking stations, as well as maintaining open locations for tip boxes, sample microplates and reservoirs.

The Bravo can be used with 8-, 16-, 96-, or 384-channel fixed tip or disposable tip heads. Heads can be interchanged in a matter of seconds. The Bravo is designed to be used either standalone or as part of a robotic laboratory automation system. It is also designed to fit inside some models of laminar flow hoods and retain laminar flow, thereby opening up cell-based plating and cellular assays to an automated liquid handling platform.

Accuracy and Precision Testing

This technical overview describes a method to measure the accuracy and precision of the Bravo in conjunction with a 96 Channel ST Disposable Tip Head. The 10 µL disposable tips were tested at the lower end of the practical volume range (0.3-2 µL). Performance at volumes >2 µL meets or exceeds the 2 µL performance. Measurements are determined by dispensing a tartrazine/DMSO solution into dry microplates, filling the plates with water, and measuring tartrazine absorbance. The product's performance meets or exceeds CVs of 5% and accuracy of ±10% of the desired volume across the practical volume range.

Materials

- Bravo with a 96 Channel ST Disposable Tip Head
- Agilent 384ST 10 µL tips (product no. 10734-202)
- Agilent 96-well manual fill reservoir (product no. 08105-001)
- 384-well polystyrene, flat bottom plates (Greiner 781101)
- Tartrazine solution, 0.25% (w/v) 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 reservoir is placed on position 2 of the Bravo. A 10 µL tip box is placed on position 3. A 384-well polystyrene plate is placed on position 5. Agilent VWorks software liquid classes for 0.3-0.5 µL and

The Agilent Bravo has nine deck positions and can be configured with interchangeable 8-, 16-, 96-, or 384-fixed and disposable tip heads.

1-5 µL dispenses are utilized. A VWorks protocol is created and run in the following manner:

1. Tips are pressed onto the head.
2. 0.3, 0.5 or 2.0 µL tartrazine solution is aspirated from the manual fill reservoir. Aspirate parameters are 6 mm from the bottom of the plate, and preceded by a 0.4 µL pre-aspirate volume (1 µL for 2 µL dispense).
3. 0.3, 0.5 or 2.0 µL tartrazine solution is dispensed into one quadrant of a 384-well plate. Dispense parameters are 0 mm from the bottom of the plate, and followed by dispensing a 0.4 µL blowout volume (1 µL for 2 µL dispense).
4. The transfer is repeated for the other quadrants of the plate.

Dispensed volumes are diluted to 50 µL by the addition of water (in this case, with clean tips and a separate VWorks protocol). 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 0.1, 0.3, 0.5, 0.75, 1, 1.25, 1.5, 2, and 2.5 µL 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. The accuracy and precision results are typical, based on averages of three successive dispenses. Accuracy outliers are an average per plate. Results may vary, depending on individual experimental methods and liquid class optimization.

Results

Volume	0.3 µL	0.5 µL	2 µL
Precision (% CV)	3.1	2.3	1.1
% Accuracy (±)	1.5	0.8	0.8
Outliers (± 5%)	7	3	0
Outliers (± 10%)	1	1	0
Transfer time (seconds/plate)	24	24	27

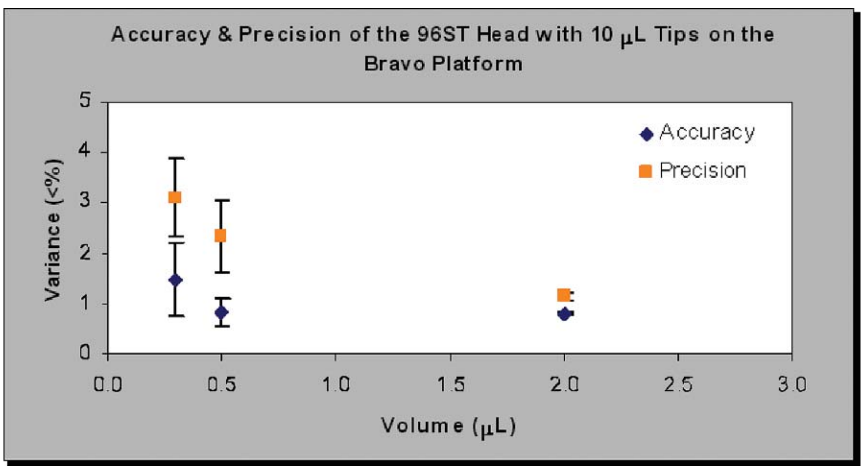

Plate Compatibility

Tip Type	96-Well Microplates	384-Well Microplates	1536-Well Microplates
96LT 200 µL	Yes	Yes	No
96LT 250 µL	Yes	Yes	No
384ST 10 µL	Yes	Yes	Yes
384ST 30 µL	Yes	Yes	Yes
384ST 70 µL	Yes	Yes	No

Conclusion

In summary, the liquid handling capabilities of the Agilent Bravo Automated Liquid Handling Platform in conjunction with a 96-Channel ST Disposable Tip Head provides precise, accurate and consistent liquid transfers. The 96-channel head, combined with Agilent's 10 µL disposable tips, can pipette across a low volume range (0.3-10 µL), making it ideal for applications requiring precise dispensing at low volumes. The highly configurable deck of the Bravo Automated Liquid Handling Platform along with its compact footprint makes it ideal for many applications in the laboratory including compound sample transfers, genomic applications, cell plating, and cell-based assays.

References:

1. Sofia MJ. Leveraging Compound Management Capabilities in Support of Drug Discovery: From Sample Archive to Sample Distribution — Driving Efficiency and Improving Productivity. Laboratory Robotics Interest Group Meeting Oral Presentation, Jan. 2005.
2. Holman JW, Mifflin TE, Felder RA, Demers LM. Evaluation of an automated preanalytical robotic workstation at two academic health centers. Clin Chem. 2002 Mar; 48(3):540-8.

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