Technical Note

Measuring the Precision and Accuracy of the CyBi®-Well vario 384/25 µl Head using the ARTEL Multichannel Verification System (MVS®)

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Introduction

In laboratory automation precise and accurate liquid handling over a large volume range is essential to meet the challenges of modern drug discovery and life science applications. The accuracy and precision demands of liquid handling tasks are growing with increasing throughput and decreasing volumes.

In this study the sophisticated and well accepted ARTEL Multichannel Verification System (MVS®) was used to evaluate the precision as well as the accuracy of the 384/25µl head of the CyBi®-Well vario (Fig.1), one of the most frequently used pipetting heads of CyBio´s flexible simultaneous pipetting platform.

***Figure 1**: The CyBi®-Well vario with 384/25 µl head, 5-position linear carriage, tip wash station and 2 stackers*

Materials

Devices

- CyBi®-Well vario with 384/25 µl head (for addition of MVS® sample solution)
- 384/60 µl head (for addition of MVS® diluent and MVS® baseline solution)
- CyBi®-Composer version 2.30.00.01
- MVS® components (MVS® Microplate Shaker, MVS® Microplate Reader, MVS® Calibrator Plate, MVS®Barcode Reader, MVS® Data Manager Software)

Consumables

- 60 µl tips (CyBio # OL 3899-25-515-N)
- 25 µl tips (CyBio # OL 3800-25-513-N)
- OmniTrays (Nunc # 140156) as disposable reservoirs
- Microplate Lids (Corning # 3898)
- MVS® 384 well Verification Plates (MVS-245)

Reagents

- MVS Solutions Start-up Pack (MVS-208)
- MVS Range D Sample Solution (MVS-206)
- MVS Range E Sample Solution (MVS 207)
- DMSO (Merck Darmstadt # 1.02931.1000)

Methods

General Liquid Handling Rules

All liquid handling methods were set up using CyBio Composer Software which allows the flexible setting of all liquid handling parameters. The "10 Tips To Improve Your Pipetting Technology" from the ARTEL homepage (www.artel-usa.com) were considered. The detailed experimental conditions are:

1. All solutions were used at room temperature
2. Prewet the pipette tips always 3x
3. Reduced piston speed (4 µl/s)
4. Reduced stage speed (50 rpm)
5. Final volume always 55 µl per well
6. Break of 1s following every aspiration and dispensing step
7. Aspiration always with overstroke (reverse pipetting mode)
8. Pipetting of the destination volume as a part of the total volume into the test plate
9. Always wet pipetting with tips immersed about 1-2 mm into the provided diluent solution
10. Ejecting the residual volume with maximum blow out volume back into the reservoir
11. Perform the ARTEL MVS Analysis immediately as described in (1).

Pipetting of MVS® Baseline Solution and MVS® Diluent

MVS® Baseline Solution and MVS® Diluent were pipetted into the MVS® 384 well Verification Plates with the CyBi®-Well vario and a 384/60 µl head according to the general liquid handling rules. The volumes were adjusted to have always a final volume of 55 µl per well. The prepared plates were lidded and immediately used for the MVS® liquid handling performance tests.

Pipetting of MVS® Sample Solution

The MVS® Sample Solution corresponding to the destination volume was pipetted with the CyBi®-Well vario and a 384/25 µl head according to the general liquid handling rules, all volumes were pipetted in the reverse pipetting mode. 10 µl were additionally aspirated for destination volumes < 25 µl and in the first dispensing step the destination volume was dispensed back into the reservoir. Then the destination volume was dispensed into the destination plate and the residual volume was ejected with maximum blow-out volume back into the reservoir. For volumes < 1 µl the first dispensing step back into the reservoir was decreased to 5 µl. Liquid handling in this volume range often is used for compound handling. Thus volumes less than 1 µl also were analysed using the ARTEL MVS® Stock Sample Solution in DMSO. The pipetted plates were analysed immediately with the ARTEL MVS® System.

ARTEL MVS® Analysis

The ARTEL MVS® Analysis was performed as described in (1) immediately following the final pipetting step into the destination plate. For each volume 3 plates were measured and mean errors of accuracy and precision were calculated automatically using the ARTEL Data Manager software.

Results and Discussion

The experimental data concerning relative inaccuracy and precision error as determined by the MVS® are shown in Table 1.
Over the whole volume range the results meet CyBio´s specification claims for the 384/25µl head of the CyBi®-Well vario which are defined to have a precision error of CV ≤ 1% in the volume range between > 5 µl and 25 µl and CV ≤ 2% in the volume range between 2 µl and 5 µl. Also in the low volume range < 2 µl the data show the excellent pipetting performance of the 384/25µl head of the CyBi®-Well vario.

Table 1: *MVS® data for the evaluation of the liquid handling performance of the CyBi®-Well vario with 384/25 µl head.*

Target Volume [µl]	MVS® Stock Sample Solution	Relative Inaccuracy ± [%]	Precision error [CV in %]
25	Range A Standard	2.20	0.78
10	Range A Standard	2.40	0.78
5	Range B Standard	1.82	0.67
2.5	Range B Standard	3.04	0.85
1	Range C Standard	2.10	1.37
0.5	Range C Standard	-0.60	4.43
0.25	Range D Standard	-1.16	8.26
0.5	Range C DMSO	5.40	2.09
0.25	Range D DMSO	14.80	4.98

The CyBi®-Well vario with the 384/25µl head was shown to have a high quality pipetting performance even at sub-microliter volumes and is able to precisely span 2 logs of volumes with one pipetting tool. The ease of use of the ARTEL MVS® make it a fast and reliable system to quickly ascertain optimal method parameter settings and instrument compliance.
The CyBi®-Well vario with the 384/25µl head is used very often for compound handling applications especially in the sub-microliter range. Thus we tested the liquid handling performance in this low volume range additionally with the corresponding MVS® Stock Sample Solutions in DMSO at the same experimental conditions. The results in Table 1 show that in this volume range using DMSO solution results in an increased precision while the accuracy was decreasing. This demonstrates the high complexity of liquid handling processes and points out the importance of physiochemical properties of the liquids for the pipetting result. This influence is growing with decreasing volumes. The ARTEL MVS® also can be used to analyse such complex interrelationships.

References

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