

TECHNICAL NOTE

Stability of the Precision of Different CyBi®-Well vario Heads after Multiple Tip Washing Routines

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Key words

CyBi®-Well vario, tip washing, stability of precision after multiple washing routines, fluorescence, buffer, DMSO

Summary

We used different CyBi®-Well vario heads with different tip types to determine the stability of the precision after 1, 3, 10 and 20 tip washing routines. Every tip washing routine consisted of 3 tip washing cycles with our active shallow well Tip Wash Station 96 (see Fig. 1). Different Fluorescein concentrations solved in buffer and DMSO were used as dye solutions to measure the precision of different test volume ranges after repeated tip washing routines. At the described experimental conditions even after 20 tip wash routines the precision remained stable with handling buffer and DMSO solutions, respectively.

Introduction

The possibility of automated and effective washing of disposable tips seriously reduces the costs for many liquid handling applications in drug research and life science. The proper function of a tip wash station corresponding to the tip number and tip length is a condition for the multiple use of tips without risk of cross contamination. The high efficiency of different CyBio Tip Wash Stations was already demonstrated (1, 2).

This Technical Note was prepared to determine the stability of the precision after multiple washing cycles. The experiments were done with different CyBi®-Well vario heads and different tip types. The precision was measured after repeated tip washing routines with the active shallow well Tip Wash Station 96, the results were discussed.



Fig.1: Tip wash trough of the active shallow well Tip Wash Station 96 from CyBio

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Materials and Reagents

- CyBi®-Well vario with 96/250 µL head (CyBio # OL 3381-27-830), 96/25 µL head (CyBio # OL 3381-27-730), and 96/60 µL head (CyBio # OL 3381-24-780)
- Active shallow well Tip Wash Station 96 (CyBio # OL 3397-25-160)
- 250 µL deep well tips (CyBio # OL 3800-25-559N)
- 25 µL tips (CyBio # OL 3800-25-533N)
- 60 µL tips (CyBio # OL 3800-25-535N)
- Black flat bottom 96 well plates PS (Greiner bio-one # 655 076)
- Fluorescein-Sodium (Standard Fluka # 46960)
- Fluorescein, free acid (Standard Fluka # 46955)
- PBS (Sigma # P3813)
- DMSO (SeccoSolv Merck Darmstadt # 1.02931.1000)
- Adhesive foil (Nunc # 236269)
- Omni Trays (Nunc # 140156) as disposable reservoirs

- PolarStar (BMG Labtechnologies) with filter set 485 nm (ex) and 520 nm (em)

Methods

Fluorescence measurement

All fluorescence measurements were performed in black 96 well plates (final volume 200 µL per well, final Fluorescein concentration = 300 nM). To obtain a test solution with low surface tension and higher viscosity Fluorescein was dissolved in DMSO, for a test solution with high surface tension and lower viscosity Fluorescein-Sodium was dissolved in PBS buffer. For further experimental details see Tab. 1 and (3) and (4).

Experimental conditions

All sets of experiments were started with the preparation of three start precision measurement plates with fresh tips. Then one washing routine consisting of 3 washing cycles was performed with the active shallow well Tip Wash Station 96 and again 3 precision measurement plates were prepared with the same set of tips. Deionized water was used as washing liquid. This procedure was repeated. After 3, 10 and 20 tip washing routines, respectively, again 3 precision measurement plates were prepared and measured. The precision results in Tab. 1, Fig. 2 and Fig. 3 show the mean of three measurements each.

Results and Discussion

In Fig. 2 the complete data set is shown for the precision stability experiment with the CyBi®-Well vario 96/250 µL head and 250 µL deep well tips after pipetting of 5 µL 12 µM Fluorescein solution in buffer and DMSO, respectively. The precision results after the different numbers of washing routines vary in a small range (less than 1%) and even after 20 washing routines the precision error is still about 2% for pipetting Fluorescein solution in buffer and less than 1.3% for pipetting Fluorescein solution in DMSO thus meeting the specification.

Similar results were obtained with the 96/25 µL head and 25 µL tips as well as with the 96/60 µL head and 60 µL tips. In Fig. 3 the complete data set is shown for the precision stability experiment with the CyBi®-Well vario 96/25 µL head and 25 µL tips after pipetting of 2 µL 60 µM Fluorescein solution in buffer and DMSO, respectively. Also in this example precision stability experiment the precision results after the different numbers of washing routines vary in a small range only (less than 0.5%) and even after 20 washing routines the precision error is still in the specification range of less than 2% for pipetting both Fluorescein solutions.

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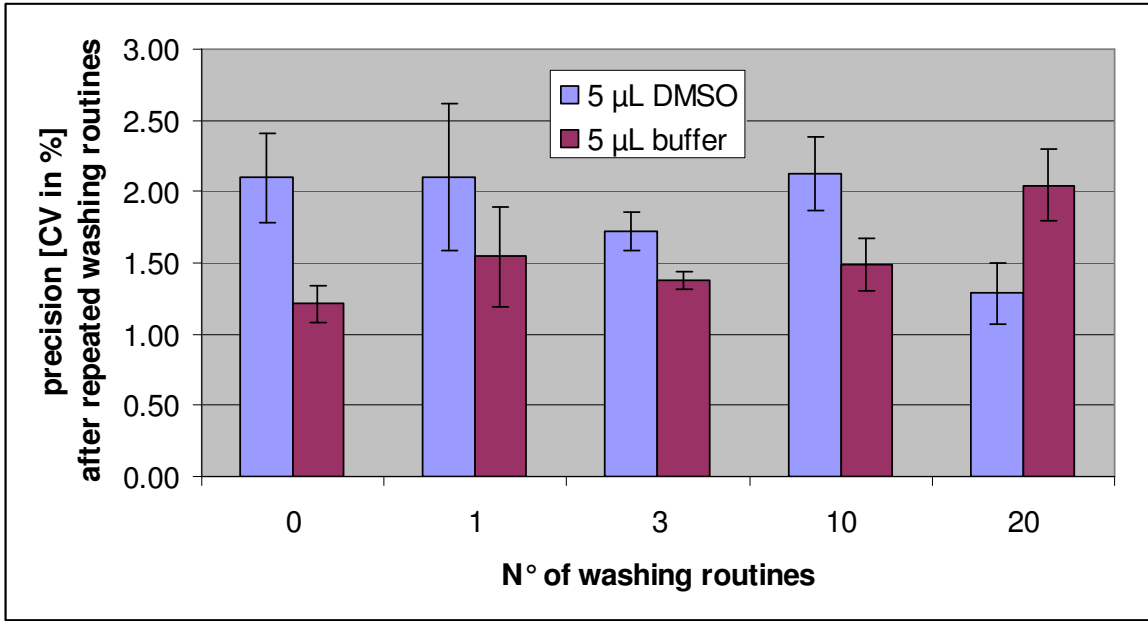


Fig. 2: Example precision data obtained with the CyBi®-Well vario 96/250 µL head and 250 µL deep well tips after pipetting of 5 µL Fluorescein solution in buffer and DMSO, respectively, after repeated washing routines, n=3.

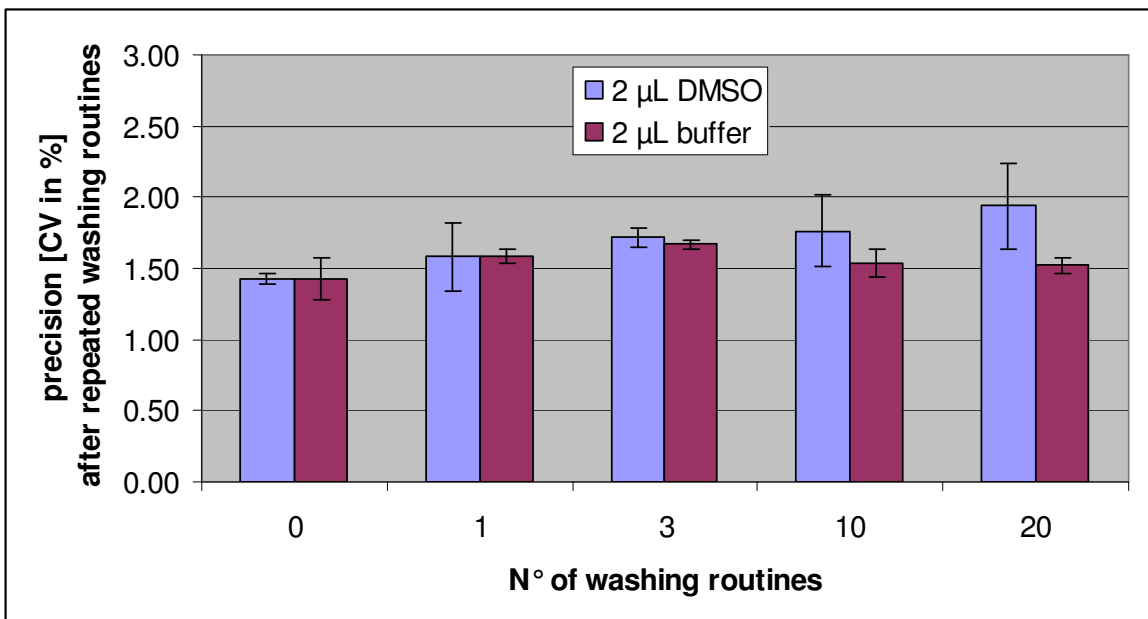


Fig. 3: Example precision data obtained with the CyBi®-Well vario 96/25 µL head and 25 µL tips after pipetting of 2 µL Fluorescein solution in buffer and DMSO, respectively, after repeated washing routines, n=3.

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In Tab. 1 the mean start precision values and the mean precision error after 20 washing routines are summarized for all precision stability experiments.

test volume [μL]	forward volume [μL]	Fluo conc. [μM]	solvent	Mean start precision [CV in %]	Mean precision after 20 wash routines [CV in %]
96/250 μL Head, 250 μL long tips					
100	100	0.6	buffer	1.24	1.84
5	195	12	buffer	1.21	2.04
100	100	0.6	DMSO	1.31	1.28
5	195	12	DMSO	1.99	1.28
96/25 μL Head, 25 μL tips					
10	190	6	buffer	1.27	1.50
2	198	30	buffer	1.43	1.52
10	190	6	DMSO	1.60	1.65
2	198	30	DMSO	1.42	1.94
96/60 μL Head, 60 μL tips					
50	150	1.2	buffer	1.50	1.41
5	195	12	buffer	1.54	1.40
50	150	1.2	DMSO	1.58	1.14
5	195	12	DMSO	1.60	1.98

Tab.1: Experimental conditions, mean start precisions and mean precision after 20 wash routines for different CyBi®-Well vario heads with different tip types and different volumes of Fluorescein test solutions in buffer and DMSO, respectively, $n=3$.

This data demonstrate the high stability of the precision of the CyBi®-Well vario with different pipetting heads and tip types and the high quality of CyBio's tip washing technology. Even after 20 washing routines the precision errors have been in the range of 2% and lower for different liquids and for high and low volumes, respectively.

References:

1. Prüfer, H. and Busch, M. (2007); "Performance Data demonstrating the Efficiency of the Tip Wash Station of the CyBi®-Well Family", TechNote CyBio AG, www.cybio-ag.com
2. Undisz, K. (2008); "Start-up Routine and Tip Washing Efficiency Data of the Active Deep Well Tip Wash Station 384", Application Note CyBio AG, www.cybio-ag.com
3. Undisz, K., Prüfer H. and Hermann, H. (2007); "Precision of the CyBi-Well vario 96/25 μL Head, Example Data of Different Liquids, Pipetting Modes and Disposable Tips with Fluorescence Readout", TechNote CyBio AG, www.cybio-ag.com
4. Undisz, K., Prüfer H. and Hermann, H. (2007); "Precision of the CyBi-Well vario 96/250 μL Head, Example Data of Different Liquids, Pipetting Modes and Disposable Tips with Fluorescence Readout", TechNote CyBio AG, www.cybio-ag.com