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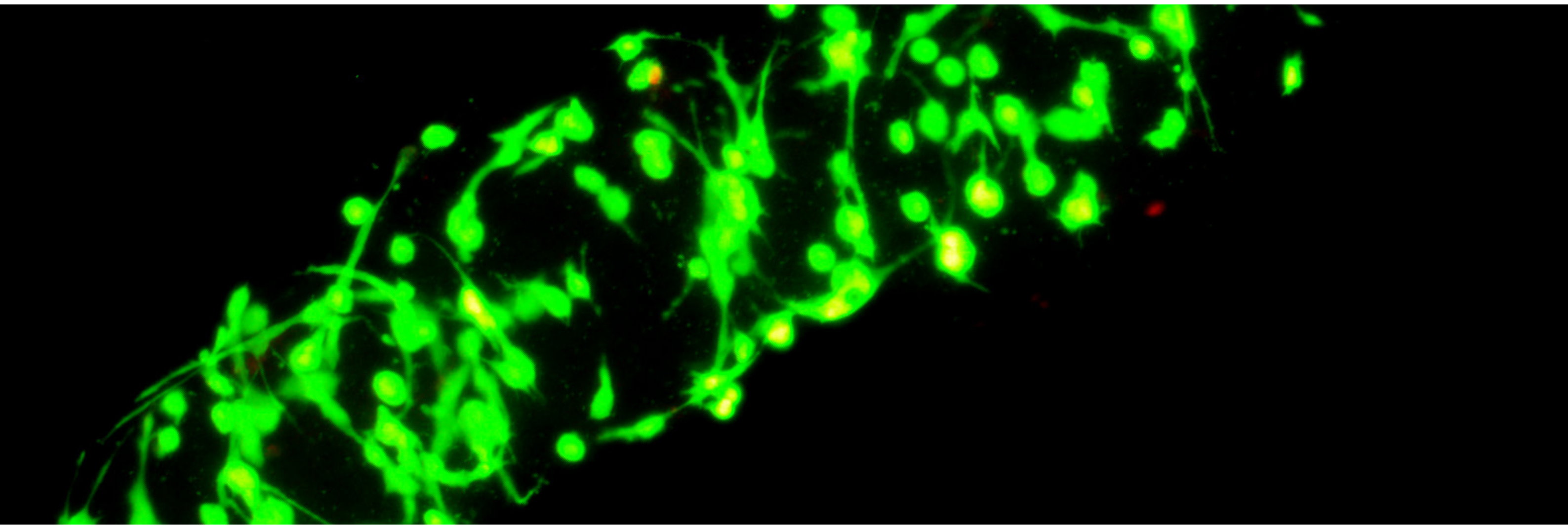
I.DOT One & the Artel MVS®

Determining Accuracy and Precision of the I-DOT One noncontact low volume dispensing platform using the Artel Multichannel Verification System (MVS)



Abstract

The Immediate Drop on Demand Technology (I.DOT) is a new approach for non-contact low volume dispensing, developed at the Fraunhofer Institute for Manufacturing Engineering and Automation IPA, Stuttgart, Germany. The Artel Multichannel Verification System (MVS) was used during development of the I.DOT and is now utilized at Dispendix GmbH, Stuttgart, for quality control of the series systems. In this Technical Note we provide typical data for inaccuracy and imprecision for the I.DOT One dispensing platform utilizing the Artel MVS. The results for single channel and multi channel operations were all within specification of the I.DOT system. These results demonstrate that the I.DOT One can produce both accurate and precise non-contact liquid transfers while providing a highly flexible platform in terms of throughput, volumes and liquids.

Abstract

Recent improvements in liquid handling technology outline a paradigm shift towards automated and noncontact systems and also a trend towards smaller sample volumes, commonly called low volume dispensing [1], [2]. There are several technology approaches for low volume dispensing, such as acoustic droplet ejection, piezo based systems, syringe pumps, positive displacement micropipettes or solenoid valves.

It is commonly understood that users demand a high degree of accuracy and precision of liquid handling platforms regardless of the application, volume range and / or liquid types. The "Immediate Drop on Demand Technology" (I.DOT) is a new approach for nano- to microliter liquid handling tasks. The general principle is simply based on a hole in the bottom of a micr otiter plate well. Capillary forces keep the sample liquid in the cavity (Figure 1). By applying a well-defined pressure pulse on top of the well a droplet is formed and a high precise and accurate nanoliter droplet is released into or onto (nearly) any destination. Larger volumes are achieved by applying 100 pulses per second.

The current format of the I.DOT Dispensing Plate is a 96 well MTP. Each of the dispensing wells can be addressed individually, i.e. each well can dispense a different volume, different liquid types can be used in a single run, position on the target plate. The I.DOT One system is basically designed and built around the I.DOT plate. It accommodates one I.DOT plate as a source plate and one SBS compatible plate (96, 384, 1536 MTP) as destination, flat objects can be used too. 8 pressurized channels are sealing 8 dispensing wells at a time. Depending on the plate layout the I.DOT system can be either operate in a parallel dispensing mode or wells are applied with the pressure pulse one by another. This flexibility enables extremely fast combinatorically dispensing tasks.

In this Tech Note the performance verification and subsequent optimization of the I.DOT One (Figure 2) noncontact low volume dispensing platform is shown. Data for inaccuracy and imprecision was generated using the Artel MVS (Multichannel Verification System, Figure 3).

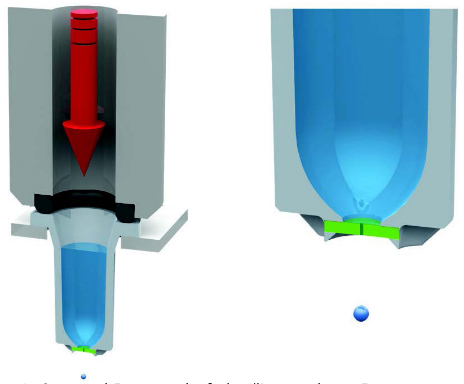


Figure 1. *General Principal of the "Immediate Drop on Demand Technology" (I.DOT).*

Figure 2. *I.DOT*

Figure 3. *Artel MVS (Multichannel Verification System).*

Materials and Methods

An MVS (Artel, Inc., Westbrook, Maine), Multi Channel Verification System, was used to measure the performance of the I.DOT One. The analysis performed is based upon Artel's dual-dye ratiometric photometry approach and provides measurements traceable to International System of Units (SI) through reference standards developed and maintained by the National Institute of Standards and Technology (NIST, USA) and National Physical Laboratory (NPL, UK), with system specifications of inaccuracy < 3% and imprecision < 1.5 % CV when measuring in 384well Artel verification plates. The MVS is a complete system consisting of the following components: a microtiter plate reader, a bar code reader, a microtiter plate shaker, a calibrator plate, sample and diluent solutions, dimensionally characterized microtiter plates (Artel verification plates), and Data Manager software which automatically calculates individual volume values, inaccuracy and imprecision and summary statistics by well, row, and column. [3]

The MVS Sample Solutions Range C (Volume Range: 500 nL – 2.490 nL) and Range E (Volume Range: 30 nL – 290 nL) were used.

The I.DOT Silica Plate (60 µm orifice, droplet volume between 2 nl and 12 nl for water) was used for uniformity and linearity test. 8 Silica Wells were filled with 50 µl MVS Sample Solution C each and 8 Silica Wells were filled with 10 µl MVS Sample Solution E (see Figure 6).

Dispensing protocols were set up using the "I.DOT Assay Studio Software".

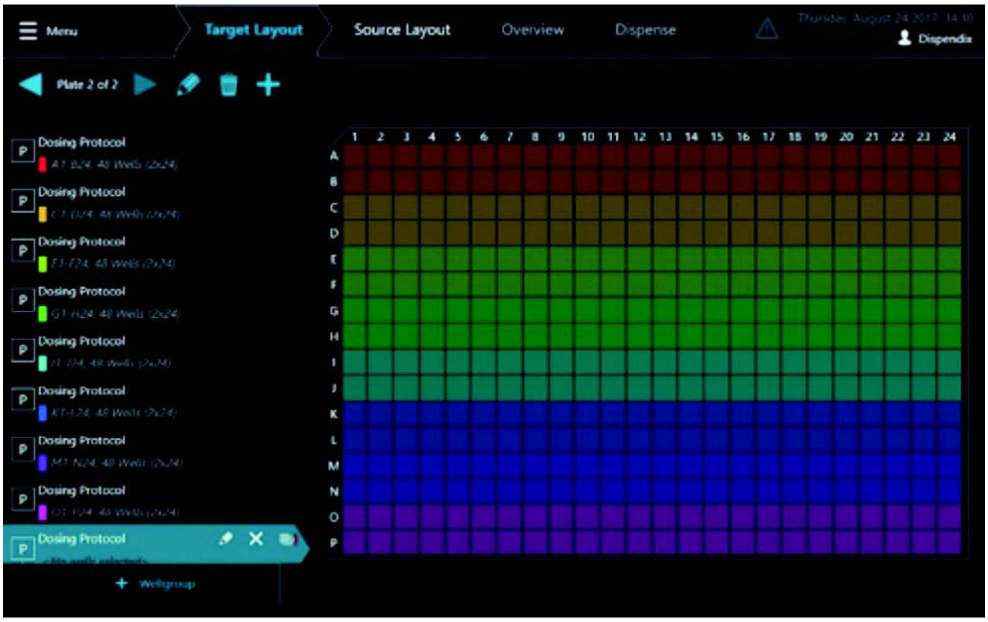


Figure 4. *Target Plate Layout for Uniformity test.*

For the uniformity test, 500 nl of Reagent C was dispensed into 48 wells (48 replicates per dispensing channel and well) of a 384-Well Verification Plate (see Figure 4).

For the linearity test, a range between 10 nL and 1.500 nL was dispensed into a 384-Well Verification Plate (see Figure 5).

Reagent E was used for the range between 10 nL and 250 nL (10 nL, 50 nL, 100 nL and 250 nL).

Reagent C was used between 500 nL and 1.500 nL (500 nL, 750 nL, 1.000 nL and 1.500 nL).

3 replicates per dispensing channel and volume were dispensed.

Further steps were carried out according to Artel´s protocol, except the shaking time of the plates. Shaking time was 300 seconds instead of 120 seconds as outlined in the protocol.

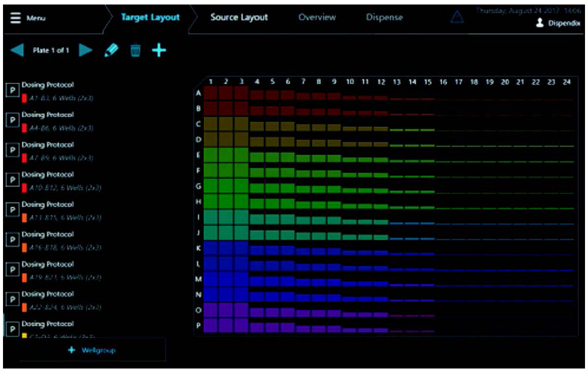


Figure 5. *Target Plate Layout for Linearity test.*

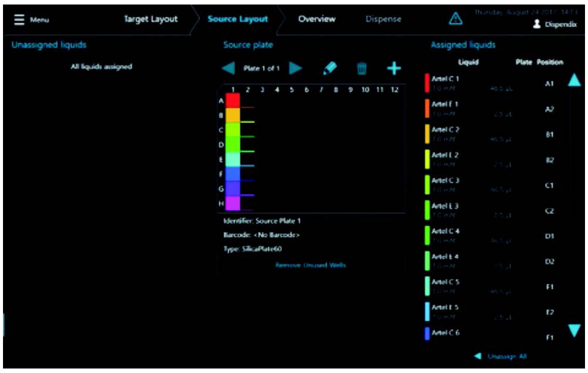


Figure 6. *I.DOT Source Plate Layout. Row A: Artel Solution C, Row B: Artel Solution C.*

Results

Uniformity Test

All eight pressurized channels at the I-DOT were used. Per dispensing channel 500 nl were dispensed into 48 wells of a 384 well plate. The initial dispensing results showed that the dispensing channel #8 delivered too low volumes, which were out of spec (see Table 1).

Table 1. *TMVC results initial run.*

Group 1 Channel Statistics

Channel	Mean Volume	Inaccuracy	Standard Deviation	CV	Channel
1	0.50372	0.74%	0.00525	1.04%	Passed
2	0.48143	-3.71%	0.00763	1.58%	Passed
3	0.49700	-0.60%	0.01076	2.16%	Passed
4	0.49885	-0.23%	0.00297	0.60%	Passed
5	0.50942	1.88%	0.00574	1.13%	Passed
6	0.49734	-0.53%	0.01585	3.19%	Passed
7	0.48747	-2.51%	0.00304	0.62%	Passed
8	0.37786	-24.43%	0.00542	1.43%	Failed

The channel was adjusted and another run was performed using channel #8 only. The channel setting could be brought back into specification (see Table 1).

Table 2. *Result for Dispensing Channel 8 after adjustment.*

Group 1 Channel Statistics

Channel	Mean Volume	Inaccuracy	Standard Deviation	CV	Channel
1	0.52167	4.33%	0.00340	0.65%	Passed

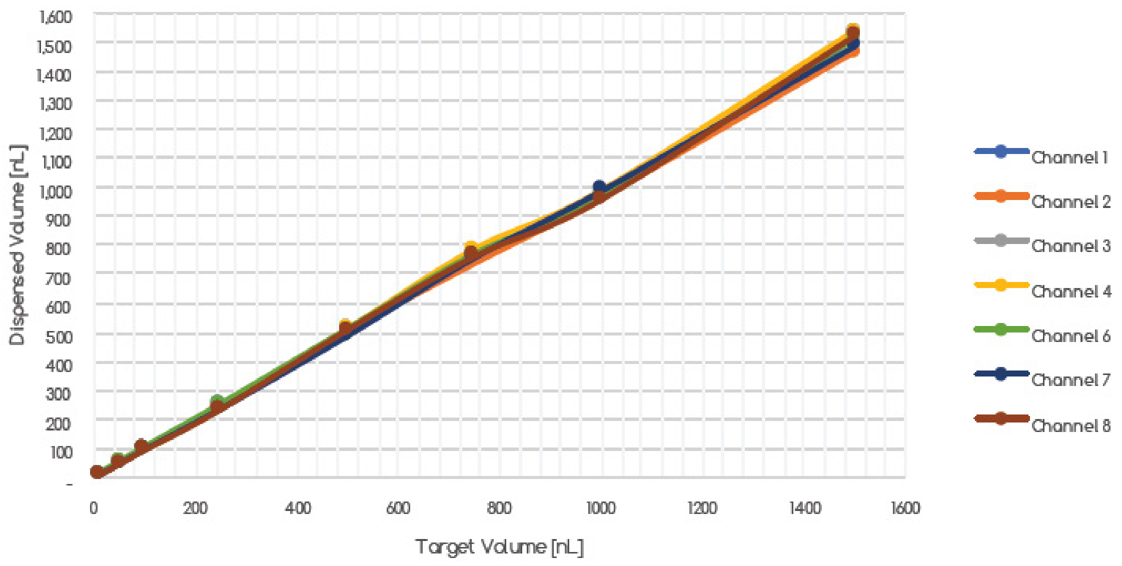
Linearity Test

All eight pressurized channels at the I.DOT were used. Reagent E was dispensed in a range from 10 nL to 250 nL and Reagent C in a range between 500 nL and 1.500 nL. CV´s of all channels were in the same range of the uniformity test. 3 replicates per dispensing channel and volume were dispensed. Both Reagents were dispensed into the same 384 well plate and analyzed together on the MVS System.

The coefficient of regression "r" was calculated for all individual channels and over all channels (see Table 3 and Graph 1). The coefficient of regression can be used as a quality criteria for the accuracy.

Table 3. *Coefficient of regression for individual channels and over all channels.*

Channel	R	R^2
1	1,0110	1,0221
2	0,9707	0,9423
3	1,0106	1,0213
4	1,0192	1,0388
5	0,9783	0,9571
6	0,9982	0,9964
7	0,9940	0,9880
8	1,0046	1,0092
All Channels	0,9983	0,9967



Graph 1. *Linearity: Target volume vs. despened volume.*

Conclusion

In this study, Artel´s MVS has been utilized to provide data for imprecision (% coefficient of variation) and relative inaccuracy (% systematic error) for the I.DOT One non-contact liquid handling system. The data presented in this Tech Note are typical for the I.DOT and can be expected from optimized protocols.

The MVS system is an exceptional helpful system for developers and supplier of liquid handling systems to optimize the dispensing performance already during development, production and final testing.

In summary, the results generated by the MVS clearly show that the I.DOT One system meets or exceeds all of Dispendix's specifications for both relative inaccuracy and coefficient of variation.

References:

1. Frost & Sullivan, "Western European Liquid Handling Market - Increasing Demand for High Throughput Data Drives Adoption of Automated Liquid Handling Instruments," 2014.
2. HiStec Low Volume Dispensing Survey 2016, March 2016
3. Multichannel Verification System (MVS): a dual dye ratiometric photometry system for performance-verification of multichannel liquid delivery devices, JALA (10)1: 35-42, 2005all of Dispendix's specifications for both relative inaccuracy and coefficient of variation.

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