

Standard Operating Procedure for the MVS® Multichannel Verification System

Company _____ Procedure #: _____

Procedure Title	Multichannel Pipette Calibration using the Artel MVS® System
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Prepared by	Effective date	Supersedes Procedure #

Review Date	Revision Date	Signature

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PRINCIPLE:

Accurate volumetric measurements and transfers are essential to all laboratories concerned with maintaining high levels of quality and productivity. The performance of pipettes varies substantially, and their accuracy cannot be taken for granted.

The MVS's unique, dual-dye photometric measurements are robust against environmental influences and traceable to SI units to enable comparison across operators, protocols, equipment, and locations. Using the Calibrator Plate, Verification Plates, and an Artel-certified Plate Reader, the MVS supports an unbroken chain of traceability to national and international standards.

The MVS complies with the Ratiometric Photometric Method according to ISO/IWA 15:2015 and can be used to fulfill test and calibration requirements of handheld or automated liquid handling systems according to CLSI QMS23:2019, ISO 17025:2017, cGMP, and cGLP. MVS System software can be used in laboratories compliant to 21 CFR Part 11.

Additionally, the MVS is effective for improving pipetting technique. Feedback is immediate and the system is used easily with minimal training. Pipette and operator performance can be verified when needed, where needed, and by personnel who use the pipettes, helping to ensure smooth workflow and optimum quality and efficiency in the laboratory.

METHOD:

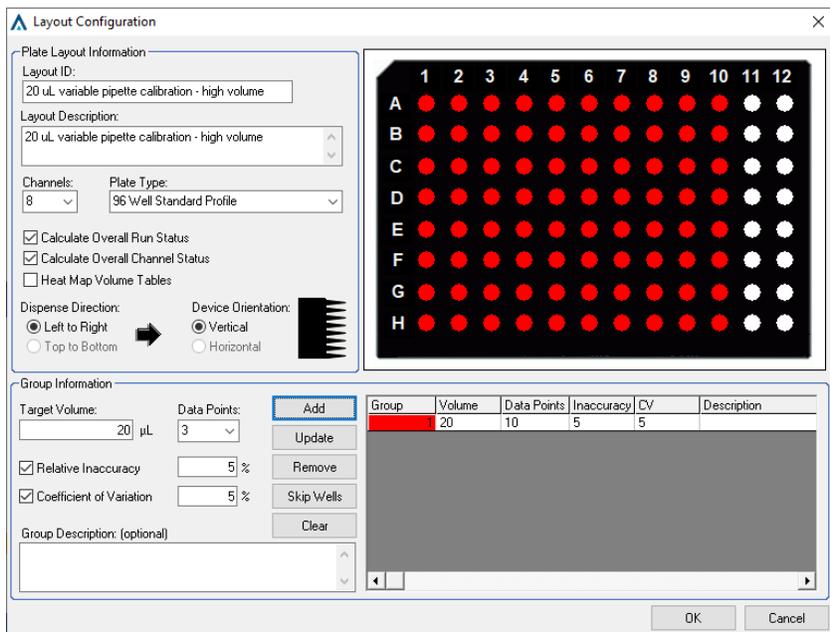
For pipettes already in service, calibrations are to be performed at least once every 12 months^{2, 3} but quarterly calibration is encouraged.¹ Adjustable volume pipettes should be tested at 3 volumes; nominal (the nominal value is the largest user-selectable volume setting; e.g., a 10-100 μ L pipette has a nominal volume of 100 μ L) volume, 50% of nominal volume and 10% of nominal volume of the minimum setting.^{1, 6, 7, 8, 9}

Each channel of a multichannel pipette needs to be tested like a single-channel pipette. Each channel of the multi-channel pipette must pass for the overall device to pass.^{7, 8, 9, 10, 11}

Calibrations require at least 10 replicate measurements per test volume.^{1, 5, 6, 7, 8, 9, 12}

NOTE: When performing pipette calibrations that contain 10 replicates, multiple verifications and multiple plates will need to be run.

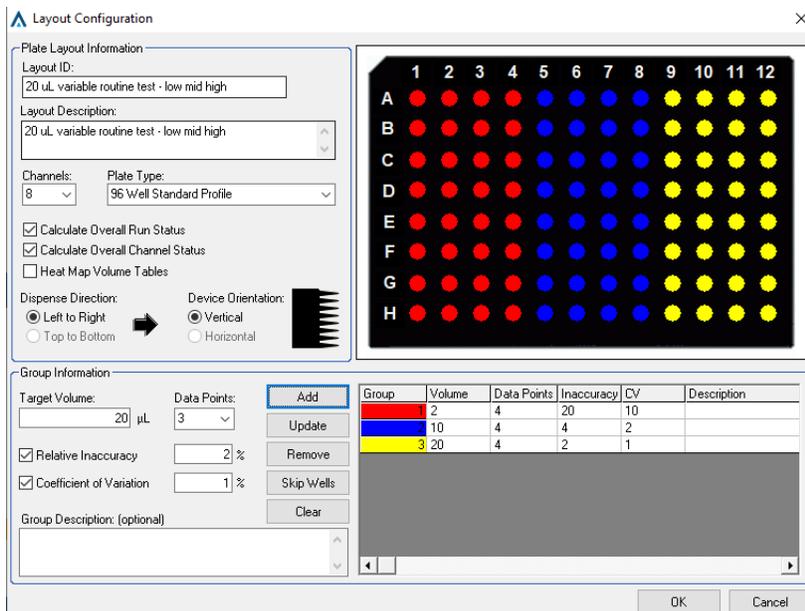
Example Pipette Calibration Plate Layout



Routine tests (quick checks) should be performed at least every 3 months.^{1, 4} Routine tests are intended to ensure the continuous fitness for purpose of the pipette between calibrations, and do not require an estimation of the expanded uncertainty of the measurement.

Routine tests require at least 4 replicates, which are needed for the calculation of the standard deviation (random error).^{5, 7, 12}

Example Routine Test Plate Layout



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Based on the risk factors of a pipette's use within an application, more frequent calibrations or routine tests should be performed. ^{1, 4, 5}

The performance requirements of the pipettes should be based on the required process/assay tolerances. See Artel Lab Report 5 for more details. ¹³

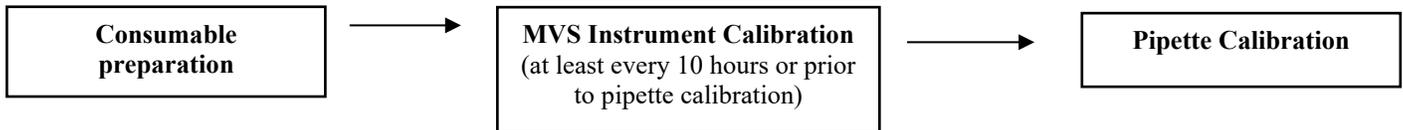
If the process tolerances are not known, single and multi-channel pipette should meet the following tolerances ¹:

- systematic error (trueness): max. 2% at nominal volume
- random error (precision): max. 1% at nominal volume

NOTE: The concept "accuracy" is related to errors – both, systematic and random errors contribute to the accuracy of a measurement.

PROCESS FLOW CHART:

Preparation → Instrument Calibration → Pipette Calibration



REQUIRED EQUIPMENT AND MATERIALS:

Equipment:

1. MVS[®] components (plate reader and power supply, shaker and power supply, barcode scanner, appropriate communication cables)
2. Computer with MVS System software installed
3. Pipettes to be calibrated
4. Appropriate pipette tips

Materials:

1. MVS[®] QualAssure Solutions (including Baseline, Diluent [if appropriate])
Declaration of traceability
2. MVS[®] Calibrator Plate
3. MVS[®] Verification Plates (96 well or 384 well)

Preparation:

The pipette to be calibrated should be clean and well-maintained. All reagents and pipettes should be allowed to equilibrate to the same ambient temperature before use.

Storage Requirements:

Instrument: When the MVS plate reader is turned on, the lamp is also on. It is recommended to turn it off when not in use.

QualAssure Solutions: MVS Aqueous QualAssure solutions should be stored room temperature (15°C to 30°C). For the highest degree of accuracy, Artel recommends that measurement occur as soon as possible after pouring QualAssure solutions from the capped amber bottles into open reservoirs and dispensing into microplates. To avoid contamination, do not return reagents to their original bottle after use.

MVS Calibrator Plate: The MVS Calibrator Plate should be stored in its carry case. Dye degradation is exacerbated by prolonged exposure to light. Store the Calibrator Plate at room temperature (15°C to 30°C). Prolonged exposure to extreme temperatures will degrade performance. Never allow the Calibrator Plate to freeze.

Clean dust from the glass surfaces according to the MVS Procedure Guide using the cleaning materials provided. Like any precision optical measurement tool, the Calibrator Plate must be maintained and cleaned.

If the MVS Calibrator Plate is shipped, it should be packed in an Artel approved shipping container with appropriate materials. The Calibrator Plate should always be shipped overnight. Contact Artel Technical Support for additional information regarding shipping instructions.

Use of the Calibrator Plate past the recertification date may negatively impact the overall MVS performance.

NOTE: MVS QualAssure solutions received frozen should be allowed to equilibrate at room temperature for several hours (or overnight), mixed by vigorously inverting the [Sample] bottle at least 20 times, and checking for precipitate. If no precipitate is visible, the solutions can be used without limitation for the specified shelf life. If precipitate remains, repeat mixing.

Instrument Preparation:

MVS Plate Reader: Turn on power to the MVS Plate Reader by toggling the power switch located on the side of the unit to the "I" position. The Plate Reader will run a self-test during which time plate carriage movement may be heard.

PLATE READER CALIBRATION:

MVS Calibrator Plate:

The MVS Calibrator Plate is used to normalize the absorbance measurements made by the MVS Plate Reader. Normalization is based upon an initial measurement made at the time of manufacture by a reference spectrophotometer at the Artel Laboratory, which provides results that are traceable to national or international standards. The Calibrator Plate is composed of a set of sealed cuvettes and pane of neutral density glass.

From the calibrator plate bar code, the MVS system software obtains critical information concerning the absorbance standards used in the calibrator plate. MVS software then compares and adjusts the plate reader accordingly.

The Plate Reader must be calibrated with the Calibrator Plate every 10 hours. The Calibrator Plate should also be re-read if the temperature changes by two degrees centigrade ($\pm 2^{\circ}\text{C}$) or more.

NOTE: MVS software will not prompt for re-calibration due to temperature fluctuations.

If the calibration values are not valid, MVS software will flag the operator. Refer to the troubleshooting section of the MVS Procedure Guide or contact Artel Technical Support.

PIPETTE CALIBRATION/VERIFICATION:

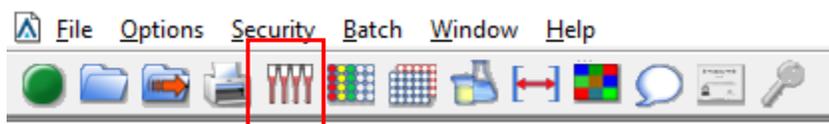
Preparation:

1. Prepare baseline plate (MVS software requires individual baseline readings for each plate type).
2. Prepare pipette calibration/verification plates as appropriate, dispensing both QualAssure solutions and/or Diluent as needed.

Procedure:

Adding a Pipette and a Plate Layout

1. Launch the MVS system software from the desktop icon and login (if appropriate).
2. If appropriate, add a pipette to the device list by selecting **File > Liquid Handler Setup** or click on the Liquid Handler Setup icon.

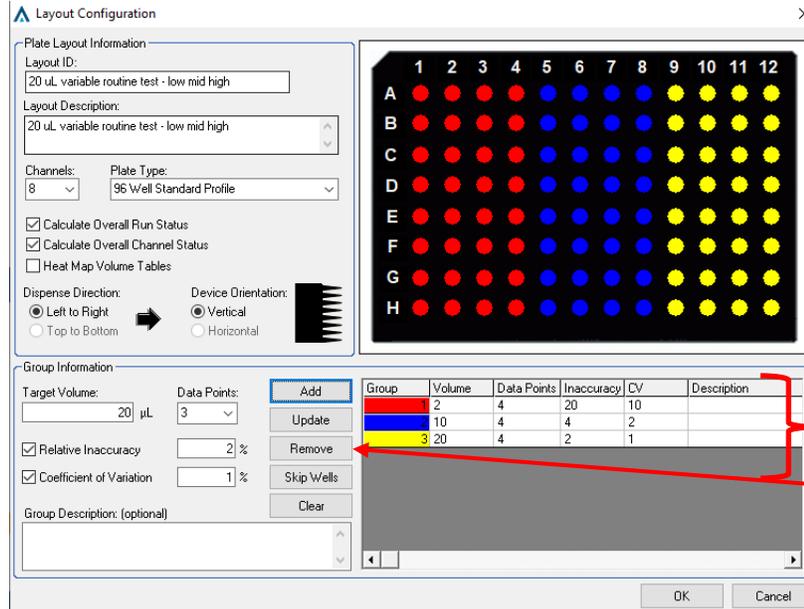


- a. Click **Add**.
 - b. Enter a unique **Device ID**, select the number of **Channels** and enter a **Description**.
 - c. The Manufacturer, Serial Number and Bar Code fields are optional.
 - d. Click **OK** until you return to the main menu.
3. If appropriate, add a plate layout by selecting **File > Plate Layout Setup** or click on the Plate Layout Setup icon.



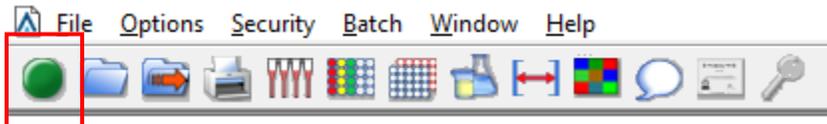
- a. Click **Add**, then select Volume Verification Layout
- b. Enter the **Layout ID**
- c. Enter the **Layout Description**
- d. Select the number of **Channels** and the **Plate Type**
- e. If appropriate, select the **Dispense Direction** and **Device Orientation**
- f. Enter the **Target Volume**
- g. Select the number of **Data Points**
- h. Confirm/check the boxes for **Relative Inaccuracy** and **Coefficient of Variation**, then enter the appropriate tolerances
- i. Click **Add**
- j. Repeat steps f-i as appropriate
- k. Click **OK** until you return to the main menu.

- l. To remove a layout group, click anywhere in the row of user-selected values for that layout group and click **Remove**.
- m. To clear all layout groups, click **Clear**.
- n. To add a row or column of empty cells, click **Skip Wells**.



Performing a Pipette Calibration/Verification

4. From the Home/main screen, select **File > Start New Verification** or, click on the green circle icon (if you hover on the icon, Start Verification will be displayed)



5. In the **Device List**, click to select the appropriate device. Click **OK**.
6. In the **Verification Setup** screen:
 - a. Click on the dropdown for the **Plate Layout** field and select the appropriate plate layout.
 - b. Click on the dropdown for the **Plate ID** field and select the appropriate plate type.
 - c. In the **Operator Name** field, enter the operator name (if appropriate)
 - d. In the **Verification Notes** field, enter notes as appropriate.
 - e. In the **Temperature, Pressure** and **Humidity** fields, enter information if appropriate.
7. Click **OK**.

8. Following the screen prompts, run MVS Calibrator Plate as needed. Before placing the MVS Calibrator Plate into the Plate Reader, check that bubble in each cuvette is in the window close to the bar code. If the bubble is visible in the lower part of the cuvette, hold the Calibrator Plate vertically and gently tap the side until all bubbles rise to the top. Ensure there are no dust particles, fibers or other interfering debris on the Calibrator Plate. If debris is present, remove only with the cleaning kit included with the Calibrator Plate.
9. Scan then insert plate into Plate Reader. Click **OK**.
10. Run baseline plate as needed. Scan baseline solution, then scan baseline plate (only MVS Verification Plates will require a scan of the plate barcode). Place baseline plate onto Plate Shaker and click **OK** (do not affix a plate cover on the plate). Once shaking is complete, insert baseline plate into Plate Reader and click **OK**.
 - a. The program will skip the baseline step if a valid baseline solution reading is already stored within the last 10 hours for the current Plate Reader.
11. Once the baseline plate has run, follow the MVS software prompts to scan QualAssure solution(s) and plate(s) as appropriate.
 - a. Only MVS Verification Plates will require a scan of the plate barcode.
 - b. Verification plates that are from a new lot code will require the user to scan the square bar code on the box of MVS Verification Plates.
12. When the Fill Plate screen is displayed, verify solutions have been dispensed correctly, then click **OK**.
13. Place plate in shaker and click **OK**. Do not affix a plate cover on the plate. Once shaking is complete, insert plate into Plate Reader and click **OK**.
14. Results will be displayed.
15. Right-click on the MVS report to display a menu with options to Change Limits, Add Comments, Validate E-Signature (if enabled), Toggle Heat Map, Export File or Close File.
 - a. Using the Heat Map feature can actively identify patterns or trends in data. See Special Conditions section for more information.

Special Conditions

Carryover Sample: residual in tip that carried over to the next well, i.e., the low reported volumes in column 3 may have been the result of droplets not transferred; and subsequently, residual droplets are transferred during the next replicate in column 4.

3	4
49.34	51.45
49.59	51.22
50.44	50.33
50.42	50.27
49.28	51.75
50.53	50.43
50.30	50.22
50.44	50.38

Pre-wetting of Tip Needed: the first dispense is lower than subsequent dispenses, indicating a need to pre-wet or to pre-wet more.

Group 1 Well Volumes (µL)

	1	2
A	49.87	50.42
B	50.24	50.22
C	49.99	50.44
D	49.97	50.28
E	50.05	50.51
F	49.95	50.37
G	49.85	50.39
H	49.99	50.31

Angled Aspirate: pipette was positioned at an angle during aspirate sequence, causing lower volumes on one half of the results and higher on the other half.



The first five tips are immersed too deep, causing more sample to flow

Group 1 Channel Statistics

Channel	5
1	51.47
2	51.43
3	51.55
4	51.30
5	51.47
6	51.24
7	51.09
8	51.02

The last three tips are immersed too shallow, causing less sample to flow

PROCEDURAL NOTES:

1. The MVS should be located in an area that is out of direct sunlight and away from local heat sources such as a heating vent. Exposure to excessive sunlight and/or heat can degrade the performance of the MVS QualAssure solutions and the Calibrator Plate.
2. Once an MVS Verification Plate read has been completed, the plate can be re-read within 60 minutes.
3. Be sure to close all solution bottles immediately after pouring an aliquot into a trough or receptacle.
4. Use MVS QualAssure solutions as quickly as possible.
5. Do not pour unused MVS QualAssure solutions back into their amber bottle.
6. Ensure that all MVS QualAssure solutions, MVS equipment, pipettes and tips are equilibrated at a stable, uniform temperature.
7. If MVS QualAssure solutions are frozen, warm slowly to room temperature. Gently invert bottle several times to mix the contents before use.
8. Always store the Calibrator Plate at room temperature inside its protective case to minimize exposure to dust and light.
9. The MVS Calibrator Plate should never be frozen, refrigerated or exposed to temperatures lower than 15°C or higher than 30°C, as irreversible damage may occur.
10. All components should be stored in the dark to the extent practical.
11. The MVS Plate Reader lamp is turned on when the reader is on. Estimated lamp life is approximately 6 months or 600 hours. The lamp assembly is a user replaceable part.
12. All MVS QualAssure solutions are non-hazardous. Refer to the SDS for more information on safety and disposal information.

LIMITATIONS OF THE PROCEDURE:

The systematic and random error of any pipette are affected by many factors.^{14, 15} The condition in which a pipette is maintained, technique used in aspirating/dispensing liquid, and type of pipette tips used are a few possible sources for discrepancy in results.

For meaningful results, it is recommended that standard laboratory pipetting techniques are used and the specific pipette manufacturer's instructions are followed. Each department performing pipette calibrations/verifications or pipette performance checks with the MVS should be responsible for determining the acceptable tolerance limits for systematic and random error with their pipettes. A calibration/verification or performance check should be repeated at least once for any failed pipette to rule out operator error.

REFERENCES:

1. [1] CLSI QMS23:2019
2. [2] ISO 8655-1:2002
3. [3] ISO/DIS 8655-1:2020
4. [4] ISO/DIS 8655-7:2020
5. [5] ASTM E1154-14
6. [6] ISO 8655-6:2002
7. [7] ISO/DIS 8655-6:2020
8. [8] ISO/DIS 8655-7:2020
9. [9] ISO/DIS 8655-8:2020
10. [10] ISO 8655-2:2002
11. [11] ISO/DIS 8655-2:2020
12. [12] ISO/DIS 23783-1:2021
13. [13] 19A3230_Artel Lab Report 5_Setting Tolerances for pipettes in the lab
14. [14] Curtis, R.H. Minimizing Liquid Delivery Risk: Pipettes as Sources of Error. *Am. Laboratory* 2007, 39 (7), 8-9.
15. [15] Carle, A.B.; Rumery, D.; Rodrigues, G. Best Practices for the Use of Micropipettes. *Am. Laboratory* June/July 2014
16. 7A6295_MVS Calibrator Plate Recertification Technical Bulletin
17. 15A5022_MVS Procedure Guide