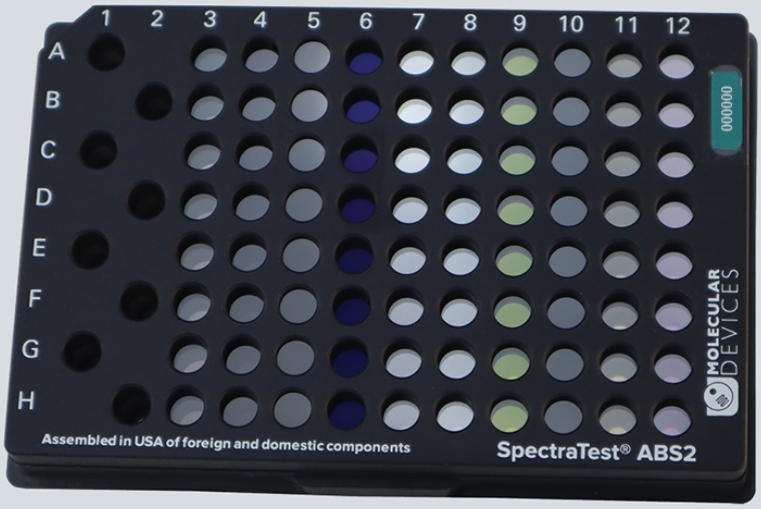

SpectraTest ABS2 Absorbance Validation Plate

User Guide

MOLECULAR DEVICES

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Product manufactured by Molecular Devices, LLC.
3860 N. First Street, San Jose, California, 95134, United States of America.
Molecular Devices, LLC is ISO 9001 registered.

Contents

Chapter 1: SpectraTest ABS2 Validation Plate Overview

Molecular Devices® microplate readers are designed to provide consistent performance for many years. The SpectraTest® ABS2 Absorbance Validation Plate from Molecular Devices enables you to validate the optical performance of the following instruments:

- FlexStation® 3 Multi-Mode Microplate Reader
- SpectraMax® ABS Microplate Spectrophotometer
- SpectraMax® ABS Plus Microplate Spectrophotometer
- SpectraMax® i3 Multi-Mode Platform
- SpectraMax® i3x Multi-Mode Detection Platform
- SpectraMax® iD3 Multi-Mode Microplate Reader
- SpectraMax® iD5 Multi-Mode Microplate Reader
- SpectraMax® M2 Multi-Mode Microplate Reader
- SpectraMax® M2e Multi-Mode Microplate Reader
- SpectraMax® M3 Multi-Mode Microplate Reader
- SpectraMax® M4 Multi-Mode Microplate Reader
- SpectraMax® M5 Multi-Mode Microplate Reader
- SpectraMax® M5e Multi-Mode Microplate Reader
- SpectraMax® Plus 384 Microplate Spectrophotometer

The SpectraTest ABS2 Absorbance Validation Plate is a comprehensive optical validation package. The SoftMax® Pro Data Acquisition and Analysis Software Protocol Library includes instrument specific protocols that automatically read the validation plate, perform the required test measurements, and make the required calculations. The software also enables you to customize the test report format.

The validation plate is compatible with the StakMax® Microplate Handling System for automated plate transfer to reduce time spent validating the instrument daily. See the *StakMax Microplate Handling System User Guide*.

Before you run the validation protocol, confirm that the time and date settings on the computer are correct. The SoftMax Pro Software uses the computer system settings for the time and date stamps.

ISO-17025

SpectraTest ABS2 Absorbance Validation Plate is a tool of metrology. The American Association for Laboratory Accreditation (A2LA) has granted accreditation to the Laboratory Quality System under ISO/IEC 17025 for validation plate calibration and re-calibration processes.

Package Contents

The SpectraTest ABS2 Absorbance Validation Plate package contains the following items:

- Protective case
- Validation plate with protective plastic sleeve
- Signed paper Certificate of Calibration (not in picture below)
- Protocol installation instructions (not in picture below)
- USB drive that contains:
  - SpectraTest ABS2 Absorbance Validation Plate User Guide with instructions to use the plate and the protocols.
  - (SN_Date_Cert#)_EZinCert.pdf that includes the information to use the EZinCert process to insert and verify the certification values in the protocol.
  - Protocols to use with the SpectraTest ABS2 Absorbance Validation Plate.
  - Installation files to install the protocols into the SoftMax Pro Software.
  - Certification file with a .pdf extension.

Validation Packages Part Numbers

Part Number	Item Name	Compatible Instruments
0200-6117	SpectraTest ABS1 Absorbance Validation Plate	FlexStation 3, SpectraMax 190, SpectraMax 340PC384, SpectraMax ABS, SpectraMax ABS Plus, SpectraMax i3, SpectraMax i3x, SpectraMax iD3, SpectraMax iD5, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e, SpectraMax Plus 384, VersaMax
0200-6191	SpectraTest ABS2 Absorbance Validation Plate	FlexStation 3, SpectraMax ABS, SpectraMax ABS Plus, SpectraMax i3, SpectraMax i3x, SpectraMax iD3, SpectraMax iD5, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e, SpectraMax Plus 384
0200-5060	SpectraTest FL1 Fluorescence Validation Plate	FlexStation 3, Gemini EM, Gemini XPS, SpectraMax i3, SpectraMax i3x, SpectraMax iD3, SpectraMax iD5, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e
0200-6186	SpectraTest LM1 Luminescence Validation Plate	FlexStation 3, SpectraMax i3, SpectraMax i3x, SpectraMax iD3, SpectraMax iD5, SpectraMax L, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e
0200-2420	Cuvette Absorbance Validation Set	SpectraMax ABS Plus, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e, SpectraMax Plus 384
0200-7200	Multi-Mode Validation Plate	FilterMax F3, FilterMax F5, SpectraMax i3*, SpectraMax i3x*, SpectraMax iD5*, SpectraMax Paradigm * Specific read modes or cartridges.

Care and Handling

Treat the optical standards with care to retain their validity. The SpectraTest ABS2 Absorbance Validation Plate is vulnerable to ambient contamination. Do not expose to extreme temperatures and do not expose to direct sunlight for an extended period of time. When the plate is subjected to significant temperature changes, leave the plate in the storage sleeve until it reaches the ambient temperature to avoid condensation issues.

When not in use, keep the plate in the plastic storage sleeve in the storage case to protect the optical surfaces from dust, scratches, and corrosion. Do not touch the wells with your fingertips. Do not store the plate in the case without first putting the plate in the storage sleeve.

Inspect the plate before all plate runs to look for dust and dirt. If you observe dust on the plate, blow moisture-free, clean canned air across both sides of the plate. Do not use air from “house” air lines and do not blow on the plate with your mouth to clean it. See Maintenance and Troubleshooting on page 25.

Certificate of Calibration

Each validation plate comes with a Certificate of Calibration that contains information specific to the individual validation plate for which it is created. In addition to details that are relevant for ISO 17025 compliance, the following information is included:

- Serial Number
- Certificate Number
- Certification Date

Factory certification of the validation plate's secondary standards is done using a reference instrument that is reserved exclusively for SpectraTest ABS2 Absorbance Validation Plate calibration and is checked for accuracy at fixed intervals of time. Molecular Devices recommends that you have the SpectraTest ABS2 Absorbance Validation Plate recertified yearly. See Recertification on page 26.

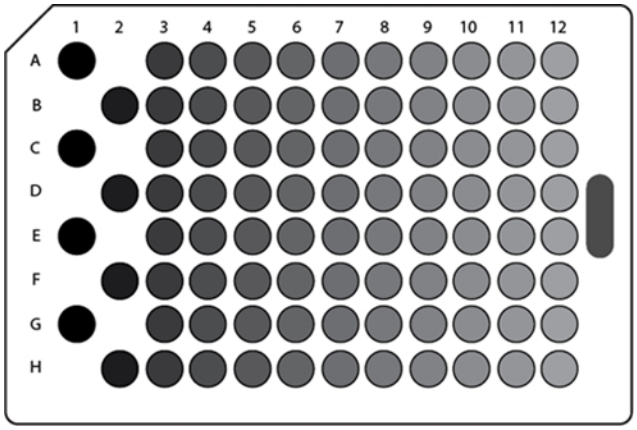
Available Tests

The SpectraTest ABS2 Absorbance Validation Plate enables you to qualify the performance of the system by testing optical specifications that are critical to achieve quality results. See Acceptability Criteria on page 21.

Each ABS2 validation protocol is specific to the instrument you test and includes the following tests:

- **Baseline Noise Endpoint and Kinetic** - Measures the stability of the optical detection system (100% Transmission).
- **Photometric Accuracy (Linearity)** - Measures the accuracy or linearity of the optical density measurement.
- **Photometric Precision (Reproducibility)** - Measures the precision or reproducibility of the optical density measurement.
- **Wavelength Accuracy** - Measures the accuracy of the assigned wavelength.
- **Wavelength Precision** - Measures the precision or reproducibility of the wavelength selection.
- **Ultimate Dark** - Measures the signal when the light beam is completely obstructed (0% Transmission).
- **Optical Alignment** - Determines whether the carriage is aligned and the light beam passes through the center of the well.
- **Stray Light** - Measures the purity of the monochromatic light that passes through the sample.

The following indicates the columns related to the available tests.



SpectraTest ABS2 Absorbance Validation Plate Configuration

Test	Columns	Configuration
Baseline Noise Tests	No plate in drawer	100% Transmittance
Photometric Accuracy and Photometric Precision	3 through 6	3: NG11, light gray glass, 2.0 mm thick (0.25 A) 4: NG4, dark gray glass, 2.0 mm thick (1.00 A) 5: NG4, dark gray glass, 3.8 mm thick (2.00 A) 6: Non-reflective filter, 1.5 mm thick (3.00 A)
Wavelength Accuracy and Precision	11 and 12	11: Light pink glass, 2.0 mm thick (Holmium oxide) 12: Purple glass, 2.0 mm thick (Didymium)
Ultimate Dark and Optical Alignment	1 and 2	Checkerboard strip alternating 100% T and 0% T. Ultimate Dark uses the wells with 0% T. Optical Alignment uses the wells with 100% T.
Stray Light	7 through 10	7: Clear glass, 2.0 mm thick (250 nm) 8: Light green glass, 2.0 mm thick (340 nm) 9: Yellow glass, 3.0 mm thick (405 nm) 10: Dark red glass, 3.0 mm thick (650 nm)

Chapter 2: Validation Protocols

The SpectraTest ABS2 Absorbance Validation Plate ships with a USB drive. If your version of the SoftMax Pro Software does not contain the protocols for the SpectraTest ABS2 Absorbance Validation Plate, you can run a software application on the USB drive to copy these protocols from the USB drive to the SoftMax Pro Software Protocol Library.

When you use the default installation, the Protocol Library is in the following path:

C:/Program Data/Molecular Devices/SMP<n.n>/Protocol Library

The protocol installation software application creates a folder named Reader Validation-Plate ABS2 and places the ABS2 validation plate protocols into this folder.

Download Validation Protocols

If needed, you can obtain the latest version of the validation protocols by contacting Molecular Devices support via the web site https://www.moleculardevices.com/support.html

To manually copy the protocol files from the download site:

1. Create a new folder (sub-directory) on the hard drive to contain the protocol file and give it a name of your choice.

   Note: To be consistent with the current Protocol Library naming convention, the location and name would be the following C:/Program Data/Molecular Devices/SMP<n.n>/Protocol Library/Reader Validation-Plate ABS2.

2. Locate the protocol file to download. The protocol file name includes the instruments for which it is intended, such as FlexStation 3 ABS2. Select the protocol that is for the instrument you plan to validate.
3. Save the protocol file in the folder you create.

Note: When you download protocol files or the protocol installation application, after the download, you may need to right-click the file and open the Properties dialog to select the Unblock check box.

Protocol File Sections

The experiments and sections in each SpectraTest ABS2 Absorbance Validation Plate protocol file contain settings that are for a specific instrument. You should read all Note sections in each experiment. For additional information and instructions, see the *SoftMax Pro Data Acquisition and Analysis Software User Guide* or the application help.

The protocols contain the following sections:

 The **ABS2 <instrument>** experiment contains the following Note sections:

-  **Introduction**: Contains protocol information.
-  **Instructions**: Contains information to get you started.
-  **EZinCert**: Contains a field that enables you to use the EZinCert method to enter the Certificate of Calibration information into the protocol. See Entering Certificate Information on page 15.
-  **Results**: Displays a summary of the result of the validation protocol after you run the protocol.
-  **Detailed Results**: Displays detailed results of the validation protocol after you run the protocol.

 The **NoPlate** experiment contains the Baseline Noise tests that you run with the instrument plate drawer empty (no validation plate in the drawer):

-  **Instructions**: Contains additional information for some instruments.
-  **Endpoint** (three plates): Contains plate settings for Endpoint read type data acquisition.
-  **Kinetic** (three plates): Contains plate settings for Kinetic read type data acquisition.
-  Group sections perform calculations.

 The **ABS2Plate** experiment contains the tests you run with the validation plate inserted in the instrument plate drawer:

-  **PhotoAccPrec** (multiple wavelength-specific Plate sections): Plate settings for data acquisition.
-  **WaveAccPrec** (multiple wavelength-specific Plate sections): Plate settings for data acquisition.
-  **OptAlignandUltDark**: Plate settings for data acquisition (two separate plates for the ABS2 SpectraMax iDx protocol and the ABS2 SpectraMax i3x protocol).
-  **Stray Light**: Plate settings for data acquisition.
-  Group sections perform calculations.

The **Appendix** experiment contains the following Note sections (do not change the content of these sections):

- **Recertification**: Contains contact information for recertification.
- **Instrument Test Criteria**: Contains criteria that are specific to the instrument type.
- **Instrument Tolerances**: Contains the source of the test criteria values.
- **CertInfo**: Contains fields that are related to the information you enter into the EZinCert Note section.
- **CertData**: Contains fields that are related to the information you enter into the EZinCert Note section.
- **Dictionary**: Contains a definition for each acronym in the protocol.
- **DataPoint Diagnostics**: Contains a summary of missing data points, if any.
- **Revision**: Contains a description of the changes made to the protocol.

Chapter 3: Entering Certificate Information

Before you run a validation protocol you must enter information from the Certificate of Calibration that accompanies the validation plate into the SoftMax Pro Software. Enter the information one time before the initial use of the protocol and then again each time Molecular Devices recertifies the validation plate and sends you a new Certificate of Calibration.

To enter certification information:

1. Insert the USB drive that shipped with the validation plate into a USB slot on the computer.
2. Locate and open the (SN_Date_Cert#)_EZinCert.pdf file using Adobe, Internet Explorer, or Microsoft Edge.

   Note: Do not open with Chrome or Notepad as the data does not copy correctly.

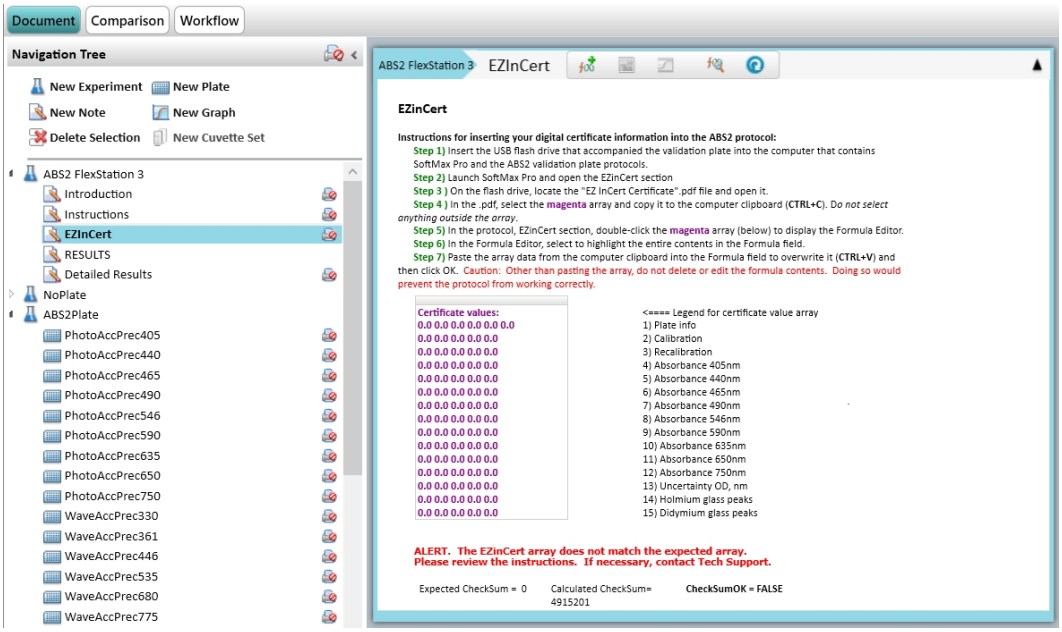
3. This file contains a group of values that have a shaded background. Select the values with the shaded background and copy this information to the computer clipboard (Ctrl+C).

```
(0~ 0~ 00001~ 0~ 0~ 561839442)&
(0~ 8~ 15~ 2019~ 2)&
(0~ 8~ 15~ 2020~ 3)&
(405~0.312~ 1.193~ 2.239~ 2.929)&
(440~0.314~ 1.138~ 2.141~ 2.943)&
(465~0.28~ 1.057~ 1.981~ 2.954)&
(490~0.289~ 1.097~ 2.067~ 2.958)&
(546~0.285~ 1.061~ 2.004~ 2.948)&
(590~0.302~ 1.129~ 2.145~ 2.941)&
(635~0.302~ 1.089~ 2.074~ 2.935)&
(650~0.306~ 1.087~ 2.065~ 2.934)&
(750~0.309~ 0.839~ 1.551~ 2.902)&
(0~0.0077~ 1.2~ 0~ 0)&
(0~360.4~ 445.6~ 536.1~ 0)&
(0~329.1~ 681~ 773.3~ 0)
```

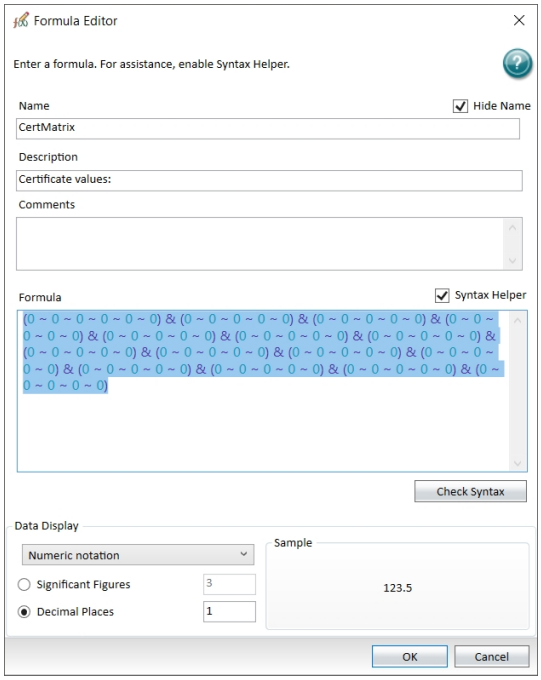
   Note: This image is a representation of the values in the file. Your section values may appear different.

4. In the SoftMax Pro Software, select the Home tab and click **Protocol Manager** > **Protocol Library** > **Reader Validation-Plate Abs2** > **<instrument>** to open the instrument-specific validation protocol.
5. In the Navigation Tree, expand the **ABS2<instrument>** experiment and select the **EZinCert** Note section.

6. Double-click the magenta **Certificate Values** field to display the Formula Editor dialog.



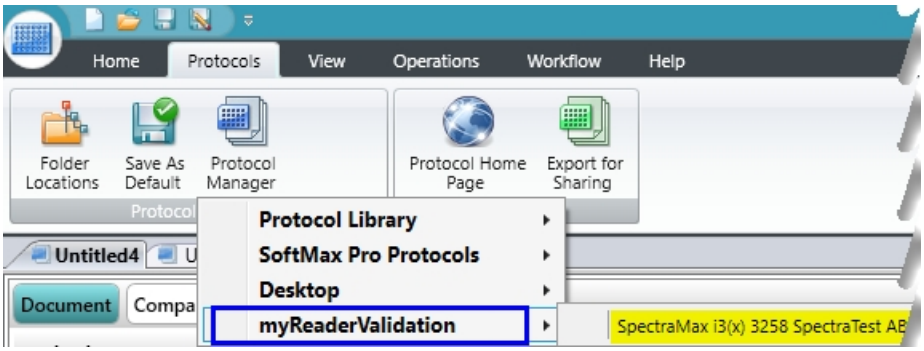
7. Wait until the content of the Formula field loads and displays colors. Then, starting at the bottom of the **Formula** field, drag the cursor upward to highlight the contents of the Formula field.



8. Paste data from the EZinCert certification information file (Step 3) over the highlighted formula content (Ctrl+V).
9. Wait until the array parses and the Formula displays highlighted in colors. Then click **Check Syntax** to verify that the certificate information formula syntax is valid. If the syntax is not valid, copy and paste the data from the worksheet into the Formula Editor until the syntax is valid.

10. Click **OK** to close the Formula Editor dialog.
11. Click **Save As** to save the file with a new name in order to save the certificate information and to prevent over writing the original protocol.
12. In the Save As dialog, click the **Save As Type** drop-down and select **Protocol Files** to save the file as a protocol.

> Tip: Name the file with the validation expiration date and instrument type, for example ABS2 2020-Jan-22 SpectraMax i3x and save the new protocol file. You can use the Folder Locations feature to save the file to the folder of your choice.



13. In the Navigation Tree, expand the **ABS2 <instrument>** experiment and select the **Results** Note section. The **Summary Result** should display "Incomplete" because the protocol has yet to be run. If not, repeat the steps in this section.

The new protocol is now ready for use with the validation plate.

Chapter 4: Running Validation Protocols

Now that you have used the EZinCert process to enter the data from the Certificate of Calibration and renamed the protocol, you are ready to run the validation protocol.

To run the validation protocol:

1. Power on the instrument and wait for the instrument to complete the start-up routine.
2. Start the SoftMax Pro Software.
3. Confirm that the instrument and the software are connected and communicating properly. If not, a red X displays over the instrument icon in the upper-left corner of the software window.

   Note: Do not insert the validation plate in the drawer. The first experiment section is done with an empty plate drawer.

4. Open the protocol file that contains the certification data you entered.
5. Click **Save As** to save the file as a data file with a name of your choice.
6. Expand the **NoPlate** experiment and select the **Endpoint1** Plate section.
7. Click **Read**. The instrument reads all Plate sections in the experiment.

   Note: If a message displays regarding no plate being in the instrument, click **OK** to have the read proceed with no plate.

8. Place the validation plate in the instrument drawer with well A1 in the A1 drawer position.

   Note: For the FlexStation 3SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, and SpectraMax M5e you must first insert an adapter plate.

9. Expand the **ABS2Plate** experiment and select the **PhotoAccPrec405** Plate section.
10. Click **Read**. The instrument reads all Plate sections in the experiment.
11. When all Plate sections are read, remove the validation plate from the drawer, return it to its protective plastic sleeve, and place the validation plate and sleeve in the storage case.

    CAUTION! To protect the optical surfaces from dust, scratches, and corrosion, do not store the plate in the case without its protective sleeve.

12. Click **Save** to save the data file.
13. Expand the **ABS2 <instrument>** experiment and select the **Results** Note section.
    - If the results are within acceptable limits and the Overview of Test Results area displays "Acceptable". Save the data file and print the report.
    - If the data is Out of Specification, see Troubleshooting on page 25.

Chapter 5: Interpreting Test Results

The Acceptable/Out of Specification limits for the tests are based on instrument specifications plus other applicable tolerances. Where a filter is used, the tolerance is determined from the tolerances quoted by the NIST or NMI on the primary standard they supply, plus a different tolerance for the production of the secondary standard. All OD filters are NIST or NMI traceable to Starna SRM1930 and RM-H6 standard set in the range of 0.5 to 3.0OD for all 9 wavelengths, as well as Holmium and Didymium filters.

Acceptability Criteria

The acceptability criteria for each test shown in the following tables are derived from a combination of the error of the instrument (or published specification for the instrument), the uncertainty of the measurement, and the uncertainty of the standard.

Table 5-1: Acceptability Criteria: SpectraMax iD3 and SpectraMax iD5 Tests

Test	Validation Plate Columns	Acceptable/Out of Specification Criteria
Endpoint Baseline Noise	No plate	Maximum deviation ≤ 0.007 OD
Kinetic Baseline Noise	No plate	Maximum deviation ≤ 0.007 OD | Drift | ≤ 0.5 mOD/min
Optical Alignment	1 and 2	Minimum values ≤ 0.015
Ultimate Dark	1 and 2	Minimum OD ≥ 3.3
Photometric Accuracy	3 through 6	| Average OD – Cert. Val. | ≤ (0.01*Cert. Val. + 0.010)
Photometric Precision	3 through 6	Stdev (Values of Replicates) ≤ (0.01*Cert. Val. + 0.003)
Stray Light	8 through 9	Minimum OD ≥ 2.3
Stray Light	7 and 10	Minimum OD ≥ 2.8
Wavelength Precision	11 and 12	Stdev (Peak Values of Replicates) ≤ 1.0 nm
Wavelength Accuracy	11 and 12	| Average Peak Value – Cert. Val. | ≤ 3.0 nm

Table 5-2: Acceptability Criteria: SpectraMax i3 and SpectraMax i3x Tests

Test	Validation Plate Columns	Acceptable/Out of Specification Criteria
Endpoint Baseline Noise	No plate	Maximum deviation ≤ 0.005 OD
Kinetic Baseline Noise	No plate	Maximum deviation ≤ 0.005 OD | Drift | ≤ 0.5 mOD/min
Optical Alignment	1 and 2	Minimum values ≤ 0.015
Ultimate Dark	1 and 2	SpectraMax i3: Minimum OD ≥ 3.3 when wavelength is between 400 nm and 650 nm SpectraMax i3: Minimum OD ≥ 2.5 when wavelength is < 400 nm or > 650 nm SpectraMax i3x: Minimum OD ≥ 3.3
Photometric Accuracy	3 through 6	| Average OD – Cert. Val. | ≤ (0.01*CertVal. + 0.010)
Photometric Precision	3 through 6	Stdev (Values of Replicates) ≤ (0.01*Cert. Val. + 0.003)
Stray Light	7 through 10	Minimum OD ≥ 2.2
Wavelength Precision	11 and 12	Stdev (Peak Values of Replicates) ≤ 1.0 nm
Wavelength Accuracy	11 and 12	| Average Peak Value – Cert. Val. | ≤ 3.0 nm

Table 5-3: Acceptability Criteria: FlexStation 3, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, and SpectraMax M5e Tests

Test	Validation Plate Columns	Acceptable/Out of Specification Criteria
Endpoint Baseline Noise	No plate	–0.003 ≤ all values ≤ 0.003
Kinetic Baseline Noise	No plate	–0.003 ≤ all values ≤ 0.003 | Drift | ≤ 0.2 mOD/min
Optical Alignment	1 and 2	–0.003 ≤ all values ≤ 0.015
Ultimate Dark	1 and 2	Minimum OD ≥ 3.3
Photometric Accuracy	3 through 6	| Average OD – Cert. Val. | ≤ (.01*Cert.Val. + 0.006)]
Photometric Precision	3 through 6	Stdev (Values of Replicates) ≤ (0.01*Cert. Val. + 0.003)
Stray Light	7 through 10	Minimum OD ≥ 2.2
Wavelength Precision	9	Stdev (Peak Values of Replicates) ≤ 0.2 nm
Wavelength Accuracy	11 and 12	| Average Peak Value – Cert. Val. | ≤ 3.0 nm

Table 5-4: Acceptability Criteria: SpectraMax ABS, SpectraMax ABS Plus, and SpectraMax Plus 384 Tests

Test	Validation Plate Columns	Acceptable/Out of Specification Criteria
Endpoint Baseline Noise	No plate	–0.003 ≤ all values ≤ 0.003
Kinetic Baseline Noise	No plate	–0.003 ≤ all values ≤ 0.003 | Drift | ≤ 0.2 mOD/min
Optical Alignment	1 and 2	–0.003 ≤ all values ≤ 0.015
Ultimate Dark	1 and 2	Minimum OD ≥ 3.3
Photometric Accuracy	3 through 6	| Average OD – Cert. Val. | ≤ (.01*Cert.Val. + 0.006)]
Photometric Precision	3 through 6	Stdev (Values of Replicates) ≤ (0.01*Cert. Val. + 0.003)
Stray Light	7 through 10	Minimum OD ≥ 3.15
Wavelength Precision	9	Stdev (Peak Values of Replicates) ≤ 0.2 nm
Wavelength Accuracy	11 and 12	| Average Peak Value – Cert. Val. | ≤ 2.0 nm

Chapter 6: Maintenance and Troubleshooting

At the time of delivery, all validation plates meet the specifications defined by Molecular Devices. You are responsible for maintaining the plates in a clean, dry, and covered environment. Validation plate maintenance requires the same care that you would give to all optical components.

- Store the plate in the plastic sleeve in the storage case when not in use.
- Inspect the plate before all plate reads. Look for dust and dirt.
- If you observe dust on the plate, you can blow moisture-free, clean canned air across both sides of the plate to clean it.

  CAUTION! Do not use air from “house” air lines on the plate, and do not blow on it with your mouth.

- If a well needs more cleaning, you can use a high-purity ethanol or methanol, such as HPLC-grade reagent alcohol, and a tightly woven cotton swab. Loosely woven cotton swabs can leave behind fiber residues. The alcohol solution can contain methanol or isopropanol, but must not contain more aggressive hydrocarbon solvents such as ethyl acetate or ketones.

  CAUTION! Do not touch the inside of the plate wells with cleaning tools other than a clean, tightly woven swab. Do not use acetone or other nonpolar solvents to clean the plate.

Troubleshooting

In the absence of data, the Results should display "No Result." If the EZinCert information is missing or invalid, the Summary Result will indicate as such. The Summary Result is a status indicator.

If one or more test results are Out of Specification, perform the following troubleshooting procedures:

- Confirm that the certificate values you enter match the values on the most recent Certificate of Calibration.
- Make sure you ran the tests with well A1 of the validation plate in the A1 drawer position and the plate was positioned such that the serial number and logo were visible. If the plate was positioned incorrectly, reposition it and repeat the test.
- Check that you used the correct plate adapter, if applicable. The FlexStation 3, SpectraMax M2, SpectraMax M2e, SpectraMax M3, SpectraMax M4, SpectraMax M5, SpectraMax M5e require the use of an adapter plate when you run the SpectraTest ABS2 Absorbance Validation Plate.
- Check the plate for dirt, dust, or other defects. Dust is not always visible. You can blow moisture-free, clean canned air across both sides of the plate to clean it and then repeat the test. Do not use air from “house” air lines on the plate, and do not blow on it with your mouth. If the results are still unacceptable, you can try cleaning with alcohol. See Maintenance and Troubleshooting on page 25.
- Check the Data Point Diagnostic or Data Diagnostics section. The summaries in this section check for errors that can be generated by missing data points. Data might be missing for one or more of the following reasons:

- The data points are outside of the reduction limits set in this protocol for a specific test.
- There is an instrument problem.
- There is a problem with the transmission of information between the instrument and the software.
- There is a problem with the computer.

  For all of these conditions, except the first one, the software displays the message: “data points are missing, please check your data.” If there is a section with missing data, you should re-run the Plate sections indicated in the Data Diagnostics report to check for an intermittent data transmission or computer problem.

If the results are still unacceptable, contact Molecular Devices Technical Support.

Recertification

Molecular Devices recommends annual recertification of your validation plates in order to ensure that they meet specifications and to ensure data accuracy of your plate reader.

Factory certification of the validation plate’s secondary standards is done using a reference instrument that is reserved exclusively for SpectraTest ABS2 Absorbance Validation Plate calibration and is checked for accuracy at fixed intervals of time.

You must return the validation plate to Molecular Devices to have it recertified. Only Molecular Devices has the necessary knowledge and equipment to recertify SpectraTest validation plates.

Note: Do not send the USB drive with the validation plate. If you send the USB drive, you will be charged for additional shipping and handling.

Each validation plate you return to Molecular Devices for recertification is measured as found.

- If the validation plate is found to be in tolerance, it is disassembled, cleaned, reassembled, and then returned with a new Certificate of Calibration.
- If the validation plate is found to be out-of-tolerance (OOT), you will be contacted to recommend and authorize the next steps.

The suggested recertification date (Next Calibration Date) is on the Certificate of Calibration. After you reserve a place in the recertification program, you will be notified when to return the validation plate (without the USB drive) to Molecular Devices.

Contact us to schedule your recertification.

- North America: Customer.Relations@moldev.com
- Europe : Service.EU@moldev.com
- China: Support.China@moldev.com
- Rest of World: Please contact your local sales representative. See Obtaining Support on page 27.

Note: Please contact Molecular Devices well before the recertification date to reserve a place in the recertification program and for pricing information. A minimum of one month is recommended.

Obtaining Support

Molecular Devices is a leading worldwide manufacturer and distributor of analytical instrumentation, software, and reagents. We are committed to the quality of our products and to fully supporting our customers with the highest level of technical service.

Our Support website, www.moleculardevices.com/service-support, has a link to the Knowledge Base, which contains technical notes, software upgrades, safety data sheets, and other resources. If you still need assistance after consulting the Knowledge Base, you can submit a request to Molecular Devices Technical Support.

You can contact your local representative or Molecular Devices Technical Support at 800-635-5577 X 1815 (North America only) or +1 408-747-1700. In Europe call +44 (0) 118 944 8000.

To find regional support contact information, visit www.moleculardevices.com/contact.

Please have your instrument serial number, validation plate serial number or Work Order number, and your software version number available when you call.

WARNING! BIOHAZARD. It is your responsibility to decontaminate components of the instrument before you request service by a service engineer or you return parts to Molecular Devices for repair. Molecular Devices does not accept items that have not been decontaminated where it is applicable to do so. If parts are returned, they must be enclosed in a sealed plastic bag stating that the contents are safe to handle and are not contaminated.

Appendix A: ABS1 Validation Plate vs. ABS2 Validation Plate

The ABS2 validation plate is the next generation of the ABS1 validation plate. The data generated with the ABS1 validation plate and protocols is not comparable with the ABS2 validation plate because the underlying test algorithms may be different. The results are comparable because the ABS2 validation plate includes expanded test parameters that do not exist for the ABS1 validation plate.

The tables on the following pages list how the ABS2 validation plate compares with the ABS1 validation plate for each reader.

Table A-1: Test Updates From ABS1 to ABS2

Test	SpectraMax Plus 384	SpectraMax ABS Plus	SpectraMax ABS	M-Series FlexStation 3	SpectraMax i3(x)	SpectraMax iDx
Endpoint Baseline Noise	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No change.
Kinetic Baseline Noise	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No statistically significant difference.	No change.
Photometric Accuracy	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range and more wavelength coverage.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.
Photometric Precision	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.	Expanded dynamic linear range and more wavelength coverage.	Expanded dynamic linear range, more wavelength coverage, and improved statistical significance.
Wavelength Accuracy	Expanded wavelength range. New software algorithm improves resolution to subnanometer. Normal Read instead of Speed Read.	Expanded wavelength range. New software algorithm improves resolution to subnanometer. Normal Read instead of Speed Read.	Expanded wavelength range. New software algorithm improves resolution to subnanometer. Normal Read instead of Speed Read.	Expanded wavelength range. New software algorithm improves resolution to subnanometer.	Expanded wavelength range. New software algorithm improves resolution.	Expanded wavelength range. New software algorithm improves resolution.
Wavelength Precision	More wavelength coverage and improved statistical significance.	More wavelength coverage and improved statistical significance.	More wavelength coverage and improved statistical significance.	More wavelength coverage and improved statistical significance.	More wavelength coverage.	More wavelength coverage and improved statistical significance.
Optical Alignment	No change.	No change.	No change.	No change.	Superior test method (made equal to iDx).	No change.
Ultimate Dark	No change.	No change.	No change.	No change.	Improved test method and limits	No change.
Stray Light	No change.	No change.	No change.	No change.	No change.	No change.

Table A-2: ABS1 vs. ABS2 Details

	SpectraMax Plus 384		SpectraMax ABS Plus		SpectraMax ABS		SpectraMax M-Series FlexStation 3		SpectraMax i3(x)		SpectraMax iDx	
	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2
Endpoint Baseline												
# Wavelengths (λ)	3	3	3	3	3	3	3	3	3	3	3	3
Reads/ Well	6	3	6	3	6	3	4	3	4	3	3	3
# Wells/ λ	96	96	96	96	96	96	96	96	48	24	24	24
Reads/ Plate/ λ	576	288	576	288	576	288	384	288	192	72	72	72
Kinetic Baseline												
# Wavelengths	3	3	3	3	3	3	3	3	3	3	3	3
Reads/ Well	21	10	21	10	21	10	13	10	10	10	10	10
# Wells/ λ	96	96	96	96	96	96	96	96	48	12	12	12
Reads/ Plate/ λ	2016	960	2016	960	2016	960	1248	120	480	120	120	120
Photometric Accuracy												
# Wavelengths (λ)	5	9	5	9	5	9	4	9	3	9	3	9
Reads/ Well/ λ	1	11	1	11	1	11	1	10	7	7	3	7
# Wells/ Column	8	8	8	8	8	8	8	8	8	8	8	8
Reads/ Column/ λ	8	88	8	88	8	88	8	80	56	56	24	56
Column==> Nominal OD												
Columns Used (Nom ODs)	4	3	4	4	4	4	4	3	4	3	4	4
Max OD Tested	1.6	2	1.6	3	1.6	3	1.6	2	1.6	2	1.6	3
Photometric Precision												
# Wavelengths (λ)	1	9	1	9	1	9	1	9	3	9	3	9
Reads/ Well	6	11	6	11	6	11	4	10	7	7	3	7

Table A-2: ABS1 vs. ABS2 Details (continued)

	SpectraMax Plus 384		SpectraMax ABS Plus		SpectraMax ABS		SpectraMax M-Series FlexStation 3		SpectraMax i3(x)		SpectraMax iDx	
	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2
Photometric Precision (Continued)												
# Wells/ Column	8	8	8	8	8	8	8	8	8	8	8	8
Reads/ Column/ λ	48	88	48	88	48	88	32	80	56	56	24	56
Column==> Nominal OD												
Columns Used (Nom ODs)	4	3	4	4	4	4	4	3	4	3	4	4
Wavelength Accuracy												
# Peaks	6	6	6	6	5	5	4	6	4	6	5	6
# Wells/ Peak	8	8	8	8	8	8	8	8	8	8	3	8
Wavelength Precision												
# Peaks	*	6	*	6	*	6	*	6	4	6	5	6
# Replicates (Wells)		8		8		8		8	8	8	3	8
*ABS1 test method for these readers is indirect. It is based on repeated OD measurements on the spectral slope of a long-pass filter. OD repeatability is correlated with wavelength repeatability. The updated ABS2 test method harmonizes to the SpectraMax i3(x) and iDx test method of repeated scans and calculating the standard deviation of peak positions.												
Optical Alignment												
# Wavelengths	3	3	3	3	3	3	3	3	3	3	3	3
Reads/ Well	1	1	1	1	1	1	1	1	1	**	**	**
# Wells/ Wavelength	8	8	8	8	8	8	8	8	8	**	**	**
Reads/ Plate/ Wavelength	8	8	8	8	8	8	8	8	8	**	**	**
** The ABS1 test method for most readers involve single endpoint reads through open holes. The ABS2 test method for the i3(x) and iDx has been updated to a well scan of a single open hole using reader features specific to the i3(x) and iDx.												

Table A-2: ABS1 vs. ABS2 Details (continued)

	SpectraMax Plus 384		SpectraMax ABS Plus		SpectraMax ABS		SpectraMax M-Series FlexStation 3		SpectraMax i3(x)		SpectraMax iDx	
	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2	ABS1	ABS2
Ultimate Dark												
# Wavelengths	3	3	3	3	3	3	3	3	3	3	3	3
Reads/ Well	1	1	1	1	1	1	1	1	1	1	1	1
# Wells/ Wavelength	8	8	8	8	8	8	8	8	8	8	8	8
Reads/ Plate/ Wavelength	8	8	8	8	8	8	8	8	8	8	8	8
Stray Light												
#Wavelengths	4	4	4	4	3	3	4	4	4	4	4	4
Reads/ Well	1	1	1	1	1	1	1	1	1	1	1	1
# Wells/ Wavelength	8	8	8	8	8	8	8	8	8	8	8	8
Reads/ Plate/ Wavelength	8	8	8	8	8	8	8	8	8	8	8	8

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