

1.0 Procedure title

Use of the plate reader for absorbance endpoint

2.0 Related Procedures

Creating a protocol file for the SpectraMax Pate Reader for Absorbance Endpoint

AUTO001a

3.0 Procedure impacts and concerns

- **3.1** Safety N/A
- 3.2 Quality N/A
- 3.3 Delivery N/A
- 3.4 Environmental N/A
- **3.5** Cost N/A
- 3.6 Compliance N/A

4.0 Responsibilities

Document Owner Manage content and distribution
Process Owner Responsible for content and process validation
Plant Manager Responsible for implementation and conformance

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5.0 Process

5.1 Process description

The plate reader is to be used for measuring absorbance at required wavelength(s) in 96-well plate assays. This procedure was based on a Molecular Devices Spectra Max 190 (or M2) controlled with Softmax Pro software. The version of your software will depend on the specific make and model of your plate reader; however, the general definitions of settings are presented here.

If there is no protocol file for your assay, then create one. Then, you may reuse a protocol file to maintain your assay setting on the plate reader. There will be adjustments in settings, template, and display. Finally, the protocol will be saved.

5.2 Process diagram



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5.3 Process steps

5.3.1 Turn on the plate reader.

- If the plate reader is not turned, then switch on the main power located on the rear of the instrument.
- ii. Allow the instrument to remain powered on for at least 1 hour before use.
- iii. It is common practice to leave the instrument on indefinitely when the reader is used often by lab members.

5.3.2 Open the SpectraMax Softmax Pro Software.

i. Double click the SoftMax Pro icon on the Windows desktop.

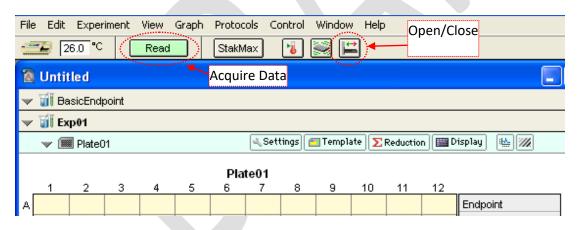


5.3.3 Create a protocol file, if one does not exist for your assay.

i. Follow procedure AUTO001a for creating a protocol file.

5.3.4 Place the plate in the reader drawer.

- i. Open the plate reader drawer by pressing the Open/Close Drawer icon, or the button on the reader front panel.
- ii. Place the color developed plate into the drawer, and ensure proper orientation. Well A1 must be in the upper left corner of the drawer.



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Best practice is to gently move the plate to the upper right corner to ensure the same orientation every time; however, the readers are often designed to align the plates automatically.

5.3.5 Acquire the absorbance data.

- i. Click the green read button.
- ii. If data from a previous read was still on-screen, then you will be prompted to replace the data. Hit OK to replace the data.

The drawer will automatically close, and acquisition will begin.

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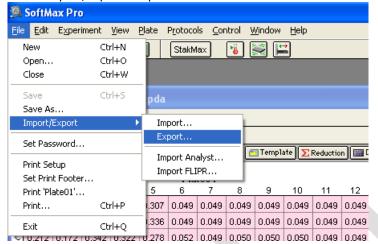
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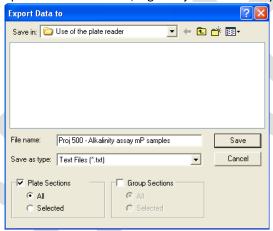
5.3.6 Export the data.

i. Select File > Import/Export > Export



Export the data to ASCII (text) format, when the reader has finished acquiring data for the plate read. Note: The timestamp of the exported file serves as time-point reference for kinetic studies.

ii. Type in a descriptive name for the file, e.g. "Proj 500 - Alkalinity mP samples".



- iii. Click SAVE.
- iv. The data are stored in text file format, and can be easily imported into, or opened with, Excel.
- 6.0 Required documents
- 6.1 Input documents
 Assay protocol.
- 6.2 Output documents

Absorbance data file in text format with timestamp of the read associated with the file.

TBD

- 7.0 Document control
- 7.1 Revision history

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Procedure document Use of the plate reader for absorbance endpoint

Procedure number AUTO001b

R0 – Initial Release – <Editor name>

R1 - <Editor name>

<Date>

7.2 Document approval

<Name>

<Approval date>

7.3 Document reviewers

<Name> <Name>

date> <Last reviewed date>

<Last reviewed

8.0 Risk analysis

TBD

<Owner> <RPN>

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