

Fusion Software Release Notes

Software Version: 2.2.0

This file provides information about Phenolmager Fusion and PhenoCycler Fusion software, version 2.2.0. Please read this information carefully as it supersedes all printed material or online help.

Overview:

Fusion software provides the interface to running the Phenolmager Fusion and PhenoCycler Fusion Imaging Systems.

System Requirements:

- U.S. English Version of Microsoft® Windows 10 64-bit
- .NET 4.7 and .NET 4.8
- Minimum monitor resolution: 1600x1200 at 96 DPI
- Minimum RAM: 32 GB

Software Installation:

To install on Windows 10:

1. Log in as an administrator
2. Close all open software programs
3. Double click on the Setup.exe
4. Follow the installer's instructions

Note: Earlier versions of Fusion software are not compatible with version 2.2.0. Please uninstall all prior versions of Fusion software before installing 2.2.0.

Fusion Software Version 2.2.0

Phenolmager Fusion System

New Features and Enhancements:

- **16-bit QTIFF file format:** Users can now select a 16-bit final QTIFF file format for whole-slide Fusion workflows. This feature includes support for exposure settings and snapshots.
- **Focus band selection:** Option to choose focus bands other than DAPI for whole-slide Fusion scans.
- **Saturation protection update:** Updated saturation protection mechanisms. Note: Older protocols may require updates.

Bug Fixes:

- **Microscope command retry:** Added a retry when the microscope is unable to complete a command.
- **Camera timeout recovery:** Implemented a recovery mechanism for specific camera timeout errors.
- **Protocols lacking DAPI filter now enabled:** Permits protocols that don't use the DAPI filter.
- **Instrument serialization fix:** Corrected instrument serialization to be culture-independent and to ensure proper decimal and group separator numbers.
- **Height-Sensing calculation update:** Introduced a fix to apply the proper offset to the height-sensing calculation.

PhenoCycler-Fusion System (1-slide setup)

New Features and Enhancements:

- **16-bit QTIFF file format:** Users can now select a 16-bit final QTIFF file format for PhenoCycler workflows.
- **Saturation protection update:** Updated saturation protection option.
- **Post-Processing time improvement:** Improved post-processing times for PhenoCycler workflows.
- **Pre-Flight Check instructions:** Improved instructions in the pre-flight check of PhenoCycler workflows.
- **Simplified error messaging:** Improved clarity in error messages when two experiments cannot be run together at the same time.

Bug Fixes:

- **Waste calculation update:** Adjusted to handle scale variations and prevent negative values.
- **Height-Sensing calculation update:** Introduced a fix to apply the proper offset to the height-sensing calculation.

PhenoCycler-Fusion System (2-slide setup)

New Features and Enhancements:

- **16-bit QTIFF file format:** Users can now select a 16-bit final QTIFF file format for PhenoCycler workflows.
- **Saturation protection update:** Updated saturation protection option.
- **Post-Processing time improvement:** Improved post-processing times for PhenoCycler workflows.
- **Pre-Flight Check instructions:** Improved instructions in the pre-flight check of PhenoCycler workflows.
- **Simplified error messaging:** Improved clarity in error messages when two experiments cannot be run together at the same time.

Bug Fixes:

- **Fluidics improvement:** Changes made to prevent potential signal carryover issues.
- **Waste calculation update:** Adjusted to handle scale variations and prevent negative values.
- **Height-Sensing calculation update:** Introduced a fix to apply the proper offset to the height-sensing calculation.

Known Issues (All systems):

- **Snapshot file naming:** The slide ID seen in PhenoChart is a unique set of alphanumeric characters which don't correspond to the original file name assigned by the user.

Fusion Software Version 2.1.0

Phenolmager Fusion System

New Features and Enhancements:

- Live View Feature Updates:
 - Introduced the ability to set exposure times within Live View

- Introduced capabilities like AutoExpose and AutoFocus
- Snapshot Feature for Fluorescent Slides:
 - Added a snapshot feature for 4-slide fluorescent acquisition
- Estimated Times for Reference Acquisition:
 - Users can now view estimated times for reference acquisition

PhenoCycler-Fusion System (1-slide setup)

New Features and Enhancements:

- Live View Feature Updates:
 - Live View now also available with PhenoCycler filters
- Whole-Slide Scanning with PhenoCycler filters:
 - Users can now create and automate whole-slide scanning of up to 4 slides with standard coverslip and using PhenoCycler filters
- Improved Sample Validation:
 - There are improvements to the Sample Validation feature with the PhenoCycler Fusion run, allowing users to validate sample masks with greater confidence

PhenoCycler-Fusion System (2-slide setup)

New Features and Enhancements:

- Live View Feature Updates:
 - Live View now also available with PhenoCycler filters
- Whole-Slide Scanning with PhenoCycler filters:
 - Users can now create and automate whole-slide scanning of up to 4 slides with standard coverslip and using PhenoCycler filters
- Improved Sample Validation:
 - There are improvements to the Sample Validation feature with the PhenoCycler Fusion run, allowing users to validate sample masks with greater confidence
- Leak Detection Feature Enabled:
 - Leak Detection feature enabled as default within the PhenoCycler-Fusion 2.0 workflow
 - Includes a tool within the UI that allows testing the dryness of the leak sensor

Known Issues (All systems):

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan
- The file name for snapshots is not used for the slide ID in the metadata

Fusion Software Version 2.0.0

Phenolmager Fusion System

- No updates to the Phenolmager workflows

PhenoCycler-Fusion System (1-slide setup)

New Features and Enhancements:

- Improved workflow during preflight routine
- New tissue finding algorithm with user review prior to scanning

PhenoCycler-Fusion System (2-slide setup)

New Features and Enhancements:

- Parallel automation of 2-slide PhenoCycler assays
- Faster fluidics
- Automated in situ RNA detection (available as beta functionality)
- Leak detection
- Improved workflow during preflight routine
- New tissue finding algorithm with user review prior to scanning

Known Issues (All systems):

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan

Fusion Version 1.0.8

New Features and Enhancements:

- Improved run estimation times for PhenoCycler Fusion runs

Bug Fixes:

- Fix for edge artifacts in the final processing of PhenoCycler Fusion qptiffs; this issue was identified in 1.0.7

Known Issues:

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan

Fusion Version 1.0.7

New Features and Enhancements:

- Allows choosing the tissue detection algorithm prior to starting the PhenoCycler Fusion run
- Improved background subtraction to reduce the appearance of artifacts near areas of high autofluorescence; this scheme may affect the pixel signal intensity of marker signals located near areas of high autofluorescence. This update will increase post-processing time; the extent of the increase depends on the size of the experiment and the sample size

Bug Fixes:

- Updated water requirement for the PhenoCycler preflight check
- Updated error message for excessively large registration errors in post-processing
- Updated error handling for significant robot errors

Known Issues:

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan
- Run estimation for PhenoCycler Fusion scans is often lower than the actual needed time

Fusion Version 1.0.6

New Features and Enhancements:

- Live Preview

Bug Fixes:

- Improved memory management for PhenoCycler Fusion scans and post-processing
- Fix for disk estimation based on largest possible sample size

Known Issues:

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan.
- The preflight check does not accurately request the amount of water needed
- Run estimation for PhenoCycler Fusion scans is often lower than the actual needed time

Fusion Version 1.0.5

New Features and Enhancements:

- Reference Viewing
- Update the reference site search to use the stages preferred direction
- Improve the signal to background ratio in the FITC filter

Bug Fixes:

- Focus fix: update the threshold for discarding bad focus points
- Focus fix: handle the case where only 3-4 good focus points are found
- Focus fix: handle dim samples
- Tissue Finding fix: improve the signal to noise ratio using longer exposures and slower stage motion
- Tissue Finding fix: improve the ability to find multiple sections on the same slide
- Fix to not start the scan when filters or reference imagery is missing
- Keep the fork engaged after acquiring label imagery
- Installer fix to retain the filter kit and objectives folder on uninstall
- Handle & characters in experiment design QPTIFF serialization
- Fix UI typos

Known Issues:

- Fluorescence slide scanning protocols without a DAPI filter will fail to scan.

Fusion Version 1.0.4

Bug Fixes:

- Optimize the z stage parameters
- Update the LED brightness

Known Issues:

- QPTIFF serialization does not handle & characters in an experiment design
- Focus failures occur on dim PhenoCycler samples
- Focus failures occur when only a few focus points are found

Fusion Version 1.0.3

New Features and Enhancements:

- New ability to cancel PhenoCycler scans
- Improved status reporting for PhenoCycler scans
- Improve how focus is calculated
- Center dialogs

Bug Fixes:

- Fix for “noise” on the right hand side of scans
- Handle background subtraction better for all exposure cases
- Fix to handle a deleted study folder
- Fix for Z acquisition illumination
- Check for lost Z robot motor steps
- Change the icon in the preflight check dialog
- Handle a powered off PhenoCycler cleanly
- Fix to make the software prompt when the protocol needs saving

Fusion Version 1.0.2

New Features and Enhancements:

- Disk space availability display
- Increased performance when writing data

- Updated tamper evidence algorithm
- Updated wording and information
- Additional manufacturing support.

Bug Fixes:

- Confirm the brightfield filter is present before launching

Fusion Version 1.0.1

New Features and Enhancements:

- New saturation protection algorithm: DAPI-only
- Improved fluidics for image quality
- Improved focus algorithm for BF & Opal slide scanning
- Updated scanning resolutions
- Updated wording and information
- Additional manufacturing support.

Bug Fixes:

- Update for stage issue when taking FL overview

Fusion Version 1.0.0

New Features:

- Whole slide scanning of Opal and PhenoCycler stained samples
- New PhenoCycler Experiment Designer application

Documentation:

- Complete documentation is accessible by selecting Help within the Fusion software

Support:

Contact Akoya Biosciences Customer Care by phone or online.

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