### HiSeq X Instrument Software Release Notes

HiSeq Control Software (HCS) v3.3.76.1

Real-Time Analysis (RTA) v2.7.6

**Recipe Fragments v3.3.9** 

Microsoft .Net Framework 4.5.1

Sequencing Analysis Viewer (SAV) v1.10.2

**BaseSpace Broker v2.9.0.2** 

Run Copy Service v1.0.19.0

For HiSeq X<sup>®</sup> Systems

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### Introduction

These Release Notes detail the key changes to instrument software components for the HiSeq X since the release of the previous HiSeq X Control Software (HCS) package. The previous release contained HCS v3.3.39, RTA v2.7.1, Run Copy Service 1.0.16.0, Recipe Fragments v3.2.4, and BaseSpace Broker v2.5.2.28.

If you are upgrading from an earlier version than HiSeq X Control Software v3.3.39, review the release notes for HCS v3.3.39. These release notes contain a list of features and bug fixes introduced in that version.

The software included in this release is customer-installable and does not require additional support from trained Illumina service personnel.

The Recipe Fragments installation overwrites any custom recipe files. Therefore, recreate any custom recipes after receiving this update.

The BaseSpace Send Instrument Health data option is activated during this software installation. After receiving this update, update your BaseSpace Send Instrument Health settings.

The software package includes:

- HCS v3.3.76.1
- RTA v2.7.6
- Recipe Fragments v3.3.9
- Microsoft .Net Framework 4.5.1 no change
- SAV v1.10.2 no change
- BaseSpace Broker v2.9.0.2
- Run Copy Service v1.0.19.0

### I. HiSeq X Control Software v3.3.76.1

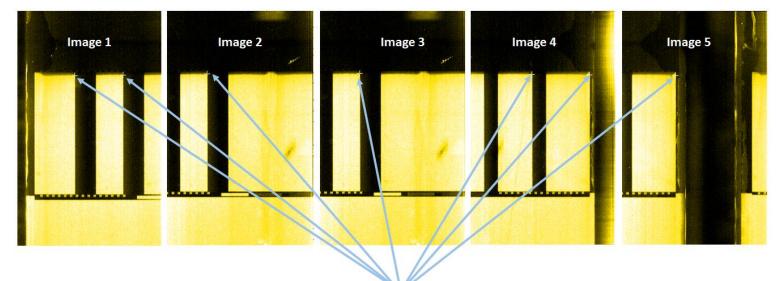
**New Features:** 

- Addition of BaseSpace domain configurability and BaseSpace integration with Illumina SeqLab.
- Simplified system health log files generated in run folders.
- Added Performance Monitor to HCS Menu | Tools to record computer hardware performances.
  - This feature defaults to "off" on install.
  - Feature is intended for Illumina Field Staff to troubleshoot hardware issues on instrument control PC (feature not intended for customer use).

**IMPROVEMENTS:** 

- Updated the binary file for flow cells ending in barcode "ALXX" to ensure proper autocentering.
- Improvement to the auto-center algorithm and retry attempts.

• In previous HCS versions, only 1 finger within a lane was used for alignment. Auto-center now uses multiple fingers within a lane for alignment. As a result, the auto-center crosshair alignment is assigned to multiple fingers within a lane.



Accurate Crosshairs

- When auto-center is unsuccessful for a particular lane, HCS retries alignment in the next lane until all 8 lanes have been attempted (total of 16 retries per flow cell, 8 retries per flow cell surface).
- Expanded the list of acceptable flow cell barcode configurations.
  - The remaining unique barcodes with the current flow cell barcode configuration is depleting. To ensure positive tracking of flow cells, the list of acceptable barcode configurations for HiSeq X flow cells has been expanded.
- Updated the disk space check to occur before reagent priming.

#### **DEFECT REPAIRS:**

- The Pause feature is enabled.
  - HCS users can now pause a run to check run components or replenish reagents. After a paused run is resumed, RTA automatically restarts.
  - The recommended procedure for staggering runs on Flow Cell A and Flow Cell B have been updated. For instructions, see the *HiSeq X System Guide* (*document # 15050091*).

#### KNOWN ISSUES:

- Selecting the Abort button to stop a run during imaging puts the camera in an unknown state and stops the adjacent run.
  - To recover from this issue, restart HCS.
  - The sequencing run for either flow cell cannot be resumed.

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### II. RTA v2.7.6

**IMPROVEMENTS:** 

• New Q-table calibration trained on a broad range of genomic samples has been updated to provide accurate Q-scores for human and non-human whole-genome sequencing.

**DEFECT REPAIRS:** 

- Improved phasing and prephasing estimation.
- Improved the run time of intensity correction algorithms.
- When RTA cannot write files on the S (SSD) drive, instead of crashing, the software logs an error and subsequent sequencing data for that tile is corrupt.

KNOWN ISSUES:

• PhiX error rate quantized at 0.2% at lowest allowed spike-in of 0.1%. Illumina recommends using a minimum spike-in of 0.5% to recover.

#### **III.** Recipe Fragments v3.3.9

**IMPROVEMENTS:** 

- Updated the following recipe fragments:
  - Reagent and flow cell barcode masks
  - Paired-end (PE) reagent names

**DEFECT REPAIR:** 

None

KNOWN ISSUES:

• None

#### **IV.** Sequencing Analysis Viewer v1.10.2

• No changes in this release.

#### V. BaseSpace Broker v2.9.0.2

#### **New Features:**

• Addition of BaseSpace Sequence Hub domain configurability and integration with Illumina SeqLab.

**IMPROVEMENTS:** 

- Updated how frequently system health logs are uploaded to BaseSpace Sequence Hub:
  - During a sequencing run: after every cycle.
  - Between sequencing runs: once every 24 hours or after HCS closes.

**DEFECT REPAIR:** 

• None

KNOWN ISSUES:

None

**EXTERNAL CONSIDERATIONS:** 

• For customers with network firewall limitations, the addition of basespace-dataeast.s3-external-1.amazonaws.com to whitelist addresses is required to enable BaseSpace Sequence Hub functionality.

#### VI. Run Copy Service v1.0.19.0

**New Features:** 

- New file copy user interface indicates the following:
  - $\circ$  File copy in progress but the network connection is slow.
  - $\circ$  RCS is not running.
  - RCS is running but not copying.
- Configurable value that defines the limit for the number of files that have not been copied.