DRAGEN TSO500 ctDNA Analysis Software on ICA

Release Notes

V2.6.1

For TruSight Oncology 500 ctDNA Assay

October 11, 2024

Introduction

These Release Notes detail the key features and known limitations of software components for the DRAGEN TSO500 ctDNA v2.6.1 Analysis Software on ICA. Below is a summary of the changes included in DRAGEN TSO 500 ctDNA v2.6.1 Software on ICA. For full details, please consult the DRAGEN TSO 500 ctDNA v2.6.1 on ICA Software User Guide available on the support website.

This software is intended for use with the TruSight Oncology 500 ctDNA and TruSight Oncology 500 ctDNA v2 assays.

- Software Version: 2.6.1
- DRAGEN software version 3.10.18

NEW FEATURES:

- Replaced DRAGEN BCL Convert with BCL Convert (non-DRAGEN) on ICA to improve processing time

FIXED ISSUES:

- Fixed an issue related to fusion calling sensitivity for samples with high number of candidate Structural Variants (SVs). SV calling involves 3 steps: 1) genome-wide graph generation of coarse resolution SV candidates, merging and filtering 2) refinement of SV candidates and 3) DNA Fusion Filtering (DNAFF). Samples that generate high number of candidate SVs, including but not limited to samples that have high number of chimeric reads or high library prep conversion efficiencies, may have had missed calls in previous software versions (v2.1.1 to v2.6.0) due to filtering of low-support SV candidates. The updated SV caller enhances the graph merging and filtering processes to recover previously missed SVs, reducing false negatives.

KNOWN ISSUES:

- There are minor differences in outputs of exon_cov_report and gene_cov_report from DRAGEN TSO 500 Analysis Software on ICA and DRAGEN server/NovaSeq 6000Dx Analysis Application
- NTC sample (No-Template Control samples with 0 reads) is marked as 'False' (under the header 'Completed_All_steps') in the metrics output file instead of being marked 'TRUE'.
- The sample sheet should not have blank rows between samples in the [Data] section, this may cause a run failure.
- Performance not verified using reads other than 2 x 151, paired end, dual index.
- The software does not notify the user when InterOp files for RunQC are missing or corrupted.
- ICA does not provide sufficient debugging information, when the user auto-launches the

TSO 500 ctDNA v2.5 pipeline and the input run folder contains additional, unexpected data that reduces available space for analysis.

- Analysis fails when starting from V1 sample sheets due to missing adapter sequences in V1 sample sheet template. Users are recommended to start with V2 sample sheet template or add adapter sequences manually.
- Pipeline does not exit early and continues to the next DragenCaller step due to TSO500 ctDNA FASTQ validation failure if Fastq_list.csv is missing.
- When storage is not selected as "XLarge" for NovaSeqX 25B flow cells and at least "Large" for every other flow cells the run will fail a disk space check.
- High chimeric read count results in incorrect TMB calculation.
- In the V2 CNV cutoff bed file, gene "MYCL" should be listed instead of "MCYL1".
- Variant consequences are not assigned consistently for co-occurring variants when stop_gained is introduced.

PRODUCT LIMITATIONS:

- The sample sheet must be configured as described in the provided templates, the User Guide or by using BaseSpace Run Planning tool.
- Sample sheets generated for auto-launch on ICA are not compatible and cannot be reused without changes for DRAGEN TSO500 ctDNA Analysis Software on a Local DRAGEN server, and vice versa.
- ICA run time depends on ICA instance availability, it will be affected by region and traffic
- Added validation for the storage size selection generates an error if "Small" or "Medium" values are selected ("Large" is required as a minimum) but the error message appears with a delay.
- The values in the Run Metrics section will be listed as 'NA' if the analysis was started from FASTQs or if the analysis was started from BCLs but the InterOp files are missing or corrupted.
- Germline estimation uses the latest publicly available population data and is estimated to be representative of targeted population, the impact of rare germline mutations is expected to be limited.
- The Illumina Annotation Engine (aka Nirvana) may report incorrect HGVS c. and HGVS p. notation for small variants occurring in RefSeq transcripts that exhibit transcript sequences differing from the genomic reference (i.e., RNA-edits). Currently the HGVS c. error rate is 0.00527% and the HGVS p. error rate is 0.00737%.
- The CNV caller has slightly higher noise for sample types that are not included in the baseline used for normalization (eg., cell lines). The baseline samples consist of mostly healthy donor clinical samples and SeraCare-contrived samples.
- MSAF output has had limited testing and needs to be used with caution. Updates to the small variant calling have led to an increased MSAF in samples with higher DNA input.

Release History

Version	Workflow#	Author	Description of Change
00	CN 1114085	Svetlana Bureeva	Initial Release