DRAGEN TSO500 ctDNA Analysis Software

Customer Release Notes

V2.6.0

For TruSight Oncology 500 ctDNA Assay

June 21, 2024

Introduction

These Release Notes detail the key changes to software components for the DRAGEN TSO500 ctDNA v2.6.0 Analysis Software on DRAGEN server. For full details, please consult the DRAGEN TSO 500 ctDNA v2.6.0 Analysis 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: 2.6.0
- DRAGEN software version 3.10.17

The software package includes:

- dragen_tso500_ctdna_2.6.0.tar – a tar file of the DRAGEN TSO 500 ctDNA v2.6.0 Analysis Software docker image
- dragen_tso500_ctdna_2.6.0.sif – singularity image format file of the DRAGEN TSO 500 ctDNA v2.6.0 Analysis Software
- uninstall_DRAGEN_TSO500_CTDNA-2.6.0.sh - bash script used to uninstall DRAGEN TSO500 ctDNA
- resources/ - workflow resource bundle for DRAGEN TSO500 ctDNA v2.6.0 Analysis Software
- DRAGEN installer:
  - dragen-3.10.17-8.el8.x86_64.run - DRAGEN installer for Oracle Linux 8
  - dragen-3.10.17-8.el7.x86_64.run - DRAGEN installer for CentOS 7
- check_DRAGEN_TSO500_CTDNA-2.6.0.sh - bash script used to validate DRAGEN TSO500 ctDNA installation is successful
- install_DRAGEN_TSO500_CTDNA-2.6.0.run - script used to install TSO500 ctDNA
- DRAGEN_TSO500_CTDNA-2.6.0.sh - bash script used to launch DRAGEN TSO 500 ctDNA

NEW FEATURES:

- DRAGEN TSO 500 ctDNA runs now supports NovaSeq X Plus data with an offboard DRAGEN server.
- Per sample gene and exon level coverage metric reports were added to results folder.
- FastQ QC Metrics have been added: Percentage of bases above Q30 (PCT_Q30_BASES) has been added to SARJ and Metrics Output.
- The metrics output file has been updated to report the percentage of soft clipped bases (PCT_SOFT_CLIPPED_BASES) as a new metric.
- DRAGEN version has been updated to v3.10.17
- Nirvana version has been updated to v3.2.7
- Custom, user-provided samplesheet file names can now include spaces.
- DRAGEN TSO 500 ctDNA Software can now be installed without Docker for launching analyses

with Apptainer.

FIXED ISSUES:

- Fixed an issue where FASTQ validation errors were being ignored and the software continued to the next analysis step.
- Fixed an issue in the V2 CNV cutoff bed file: gene "MYCL" should be listed instead of "MCYL1".
- Fixed an issue where an MNV was being incorrectly classified as TMB eligible.
- Fixed an issue where FastqGeneration step fails when using S4 flow cells and the run folder size is up to 1.2 TB, and FASTQ files up to 4.2 TB.

KNOWN ISSUES:

- Moving or modifying files during the analysis may cause the analysis to fail or provide incorrect results.
- Using control-c during a running analysis may cause an FPGA error. To recover from an FPGA error, shut down and restart the server.
- 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.
- Some contrived samples such as SeraCare Complete Mutation Mix, which have multiple structural variants (SVs) and high library conversion efficiencies, could generate a high number of chimeric reads and high number of candidate SVs. Occasionally, the SV caller may filter some of the reads and lead to occasionally missing fusions. In such cases downsampling the FASTQs can help recover those fusion calls. Contact your local support team for additional details and a workaround.
- 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.
- FastqGeneration step fails when using S4 flow cells and the run folder size is up to 1.2 TB, and FASTQ files up to 4.2 TB. This is due to FASTQ files being duplicated in both the Nextflow works folder as well as the Logs_Intermediates/FastqGeneration folder causing the disk space to run out before the FastqGeneration step could be completed.
- ctDNA pipeline fails for an NTC sample (No-Template Control samples with 0 reads) due to absence of Evidence BAM File.
- In the V2 CNV cutoff bed file, gene "MYCL" should be listed instead of "MCYL1".
- ctDNA pipeline fails for an NTC sample (No-Template Control samples with 0 reads) due to absence of Evidence BAM File.

- 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.
- Positive control samples can miss fusions due to SV caller step issue.
- High chimeric read count results in incorrect TMB calculation.

PRODUCT LIMITATIONS:

- The sample sheet must be configured as described in the provided templates, 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.
- 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

Revision	Release Reference	Originator	Description of Change
00	1109299	Manavi Abrol	Initial Release