TruSight Oncology 500 Local App Software

Release Notes

V2.2.1

For TruSight Oncology 500 Assay

October 23, 2024

Introduction

These Release Notes detail the key features and known limitations to software components for the Local TSO500 App v2.2.1.

This software is intended for use with the TruSight Oncology 500 Assay.

The software package includes:

- trusight-oncology-500-ruo-2.2.1.8.zip
- md5sum.txt
- resources
- trusight-oncology-500-ruo-dockerimage-ruo-2.2.1.8.tar
- trusight-oncology-500-ruo.img
- TruSight_Oncology_500_RUO.sh

NEW FEATURES:

- The TSO 500 Docker image OS is updated to Oracle Linux 8
- The TSO 500 blocklist is updated to remove PMS2CL pseudogene
- Run folders from NovaSeq 6000Dx (RUO mode) are supported

DEFECT REPAIRS:

- Illumina Connected Annotations (formerly, Illumina Annotation Engine, Nirvana) in updated to v3.2.7. This includes the following enhancements and bug fixes:
- RefSeq content version 105.2020102
- Reduced the HGVS c. error rate by 54% and HGVS p. error rate by 20%. Currently the HGVS c. error rate is 0.00527% and the HGVS p. error rate is 0.00737%.
- Fixed issues related to incorrect CDS coordinates in some edge cases
- Improved detection of frameshifts when variants partially overlap the coding sequence

KNOWN ISSUES:

- Local app may use additional available hardware/compute resources, recommendation is to not run multiple local app instances on a single node.
- When using NovaSeq XP workflow, sample sheets require lane information and may provide unclear error messages when lane information is excluded.
- An error will occur when trying to run more than 96 samples on a single node.

- Rare instances of low multiplexed runs (3 DNA samples in a run) cause delayed results when performing read collapsing.

PRODUCT LIMITATIONS:

- Performance not verified using read lengths other than 2 x 101.
- FASTQ Generation is not supported with read lengths other than 2 x 101 or 2 x 151.
- Unmapped long insertions are not likely to occur on shorter indels because there is sufficient reference-matching sequence in the reads. Product claims only indels up to 25 base pairs.
- Complex variants are specifically output only for a specific region of the EGFR gene, component and phased variants would both be contained in the output.
- Incorrect calculation of variant allele frequency can occur in variants near the start and end of genomic reads, but variation in read start and end positions in an enrichment assay is sufficient to make incorrect variant allele frequency in output variants a low-probability situation.
- Germline estimation using high tumor purity (>70%) can impact estimation, due to somatic and germline variants appearing with similar variant allele frequency.
- Germline estimation uses latest publicly available population data and estimated to be representative of targeted population, the impact of rare germline mutations is expected to be limited.
- Poor quality wild type reads may align as chimeric and be miscalled during RNA analysis.
- When filtering fusion variant candidates, strand consideration is not used in annotation, which allows contiguous sequences to be intragenic, when they are on different strands.
- Manta will not call fusions where both breakpoints map to the same gene transcript: FIP1L1-PDGFRA (when both breakpoints overlap Ensembl transcript ENST00000507166) and GOPC-ROS1 (when both breakpoints overlap Ensembl transcript ENST00000467125).
- Manta may not always call fusions in the following situations, which have been exclusively observed in synthetic commercial controls:
    - Multiple fusion breakpoints from a single fusion gene pair with breakpoints within approximately 150 base pairs of each other (observed with ETV6-ABL1 and ETV6-NTRK3 fusions).
    - Multiple fusions from two different gene pairs with breakpoints within approximately 150 base pairs of each other (observed with IRF2BP2-NTRK1, TFG-NTRK1, SQSTM1-NTRK1 fusions).
    - Breakpoint(s) are located in region(s) with high homology (observed with fusions with breakpoints on SEPT14 exon 10).
- Gathering output requires all samples scattered to be gathered at once, gathering a subset of results is not supported.
- The Illumina Connected Annotations (formerly, Illumina Annotation Engine, 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%.

Release History

Revision	Release Reference	Originator	Description of Change
00	CN 1114469	Svetlana Bureeva	Initial Release