

# Ion AmpliSeq™ SARS-CoV-2 Research Assay Parameter Sets for Genexus™ Software 6.2

## Release Notes

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### Contents

Revision History .....	1
Release Notes Overview .....	1
Ion AmpliSeq™ SARS-CoV-2 Research Assay in Genexus™ Software 6.2 Overview .....	2
Key Capabilities .....	2
Recommended number of samples per run .....	2
Sample input dependent differential process settings .....	2
SARS-CoV-2 Genexus™ Software plugins.....	2
Other plugins included in Genexus™ Software for data analysis.....	3
Known issues that affect the Ion AmpliSeq™ SARS-CoV-2 Research Assays in Genexus™ Software 6.2 .....	3
System Requirements .....	3
Installation procedure .....	4
Package and version information .....	4
Software packages versions.....	4

### Revision History

Revision	Date	Description
A.0	17 August 2020	New release notes for Ion AmpliSeq™ SARS-CoV-2 Research Assay Parameter Sets for Genexus™ Software 6.2

### Release Notes Overview

The Ion AmpliSeq™ SARS-CoV-2 Research Assays enable the processing and analysis of the Ion AmpliSeq™ SARS-CoV-2 Research Panel on the Ion Torrent™ Genexus™ Integrated Sequencer version 6.2. Install the parameter sets and the requisite SARS-CoV-2 Genexus™ Software plugins on the Genexus™ Software 6.2 to enable the assays. This document outlines the known issues to be fixed in subsequent releases.

# Ion AmpliSeq™ SARS-CoV-2 Research Assay in Genexus™

## Software 6.2 Overview

The Ion AmpliSeq™ SARS-CoV-2 Research Panel consists of two pools with amplicons ranging from 125 bp to 275 bp in length and covers >99% of the SARS-CoV-2 genome, including all serotypes. The assay is part of an automated, accurate, and targeted NGS workflow that enables coronavirus typing in under a day. This end-to-end solution includes the assay and plug-in suite that was developed in collaboration with researchers at the forefront of the SARS-CoV-2 crisis.

## Key Capabilities

### Recommended number of samples per run

Up to 16 samples occupying 2 lanes of a GX5™ chip per run are recommended to obtain sufficient depth of coverage for variant calling and sequence assembly.

### Sample input dependent differential process settings

Two assays are created in the software when the parameter sets are installed.

1. Ion AmpliSeq SARS-CoV-2 Research Assay (**Default assay**): to be used for > 200 viral copies, or virus not quantified.
2. Ion AmpliSeq SARS-CoV-2-LowTiter Research Assay (**Low Titer Assay**): to be used for ≤ 200 viral copies.

### SARS-CoV-2 Genexus™ Software plugins

- **COVID19AnnotateSnpEff**: Generates an annotated list of variants. You can use this plugin to identify and annotate variants with public or private databases.
- **IRMAreport**: Generates FASTA files of the consensus sequence for each barcoded sample. The Iterative Refinement Meta-Assembler (IRMA) identifies low frequency variants for highly variable RNA viruses.
- **AssemblerTrinity**: Generates a genome-guided or de novo viral sequence. The AssemblerTrinity plugin uses inchworm, chrysalis, and butterfly software modules sequentially, then builds de Bruijn graphs to resolve alternatively spliced isoforms and transcripts derived from paralogous genes to generate contig sequences.
- **AssemblyStats**: Provides QC metrics (% sequence identity of sample to reference sequence) corresponding to each of the IRMAreport and AssemblerTrinity assembled sequences
- **ControlStat**: Provides QC metrics for the 5 human expression control targets included in the Ion AmpliSeq™ SARS-CoV-2 Research Panel, and metrics for the relative expression of SARS-CoV-2 and human control targets in a sample.

## Other plugins included in Genexus™ Software for data analysis

- **coverageAnalysis:** Used to view statistics and graphs that describe the level of sequence coverage produced for targeted genomic regions. domains to enable detection of polymorphisms within the T-cell receptor variable gene. This assay reports the frequency and sequence features of clonotypes and performs secondary analyses of repertoire features. This assay uses the IMGT database of variable, diversity and joining genes as a reference
- **CustomerSupportArchive:** Generates a downloadable archive that a technical support representative can use to troubleshoot and diagnose problems with sequencing runs or with Genexus™ Software.

## Known issues that affect the Ion AmpliSeq™ SARS-CoV-2 Research Assays in Genexus™ Software 6.2

Issue number	Issue Summary	Impact and Mitigation
15753	Copying the Ion AmpliSeq™ SARS-CoV-2 Research Assay (with the <b>Manage Assays &gt; Copy</b> action) to modify settings does not proceed as expected.	This issue impacts both the default and the low titer assays. Contact your Field Bioinformatics Specialist (FSB) for assistance to modify any factory settings for the assays. This issue will be resolved in an upcoming software release.
15158	When making a copy assay of either of the two assays, if default plugins are upgraded, the upgraded plugins will not be auto-selected.	This issue impacts all assays that use Genexus™ plugins. As a workaround: <ul style="list-style-type: none"><li>• Manually select the upgraded plugins to run when copying a factory assay or modifying a draft assay (with the <b>Manage Assays &gt; Edit</b> action).</li><li>• Alternatively, run the plugins after the analysis is completed (with the <b>Sample Results &gt; ... (More Options) &gt; Run Plugins</b> menu). This issue will be resolved in an upcoming software release.</li></ul>
15773	Variant mis-calls are observed at reference genome positions 3037, 11083 and others.	Mis-calls are restricted to long homopolymer stretches. Variant calling optimization and position 3037 and 11083 mis-calls will be resolved in an upcoming software release.

## System Requirements

Ion AmpliSeq™ SARS-CoV-2 Research Assays are designed for use with Ion Torrent™ Genexus Software version 6.2 or later.

## Installation procedure

Ion AmpliSeq™ SARS-CoV-2 Research Assay parameter sets and the associated plugins are installed from **Thermo Fisher Connect** in Genexus software with the **Settings > Software Updates** menu or from a media storage device (USB) by qualified support personnel (FSE – Field Service Engineer).

## Package and version information

Release Date	15 August 2020
Release Version	2.0

### Software packages versions

The table below lists the package version included in respective zip files.

Product Name	Package	Version
Ion AmpliSeq™ SARS-CoV-2 Research Assay 2.0	Ts-Genexus-SARS-CoV-v2-GX5-w2.0-RNA_2.0.0_amd64	2.0
Ion AmpliSeq™ SARS-CoV-2-LowTiter Research Assay 2.0	Ts-Genexus-SARS-CoV-v2-GX5-w2.0-RNA_2.0.0_amd64	2.0

### SARS-CoV-2 Genexus™ Software plugins

Plugin	Version
COVID19AnnotateSnpEff	1.3.0.2
IRMAreport	1.3.0.2
AssemblerTrinity	1.3.0.2
AssemblyStats	1.0.0.0
ControlStat	1.0.1



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Products:  
Ion AmpliSeq™ SARS-CoV-2 Research Assay Parameter Sets  
SARS-CoV-2 Genexus™ Software plugins

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