

Ion Reporter™ Software 5.18 Release Notes

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Revision History

Revision	Date	Description
D.0	June 06, 2022	<p>Revised release notes for Ion Reporter™ Software 5.18.4, which includes software updates for Analysis workflow variant calling and annotation improvements and feature enhancements for assays and panels and Support for OncoPrint™ Myeloid MRD Assay.</p> <p>The following issues were fixed in Ion Reporter™ Software 5.18.4: IR-48307, IR-48225, IR-48035, IR-47891, and IR-47890/IR-47397, IR-47762.</p> <p>The following Known Issues are added to the release notes: IR-48456, IR-48321, IR-48431, IR-48306, IR-47724, IR-48221, IR-48071, IR-47914, IR-47889, IR-47881, IR-47872, IR-47902, IR-47743, IR-47762, IR-47743, IR-45509, IR-47672, IR-47870, IR-47869/IR-47903, IR-47896/IR-47610, IR-47908, IR-48185.</p>
C.0	31 Aug 2021	<p>Updated release notes for Ion Reporter™ Software 5.18.2, which includes software updates for new analysis workflows, updates for MAF and ExAC filters and updates to OncoPrint™ Extended 5.18v2 and TMB (Non-germline Mutations) 5.18v2 filter chains.</p>
B.0	02 July 2021	<p>Updated release notes for the following versions: Released on thermofisher.com:</p> <ul style="list-style-type: none"> • Ion Reporter™ Software 5.18.0.0 on Connect • Ion Reporter™ Software 5.18.0.1 on Connect • Ion Reporter™ Software 5.18.0.2 on Connect • Ion Reporter™ Software 5.18.1.0 on Connect <p>Ion Reporter™ Server release:</p> <ul style="list-style-type: none"> • Ion Reporter™ Software 5.18.1.0
A.0	30 April 2021	<p>New release notes for Ion Reporter™ Software 5.18.</p>

New features and enhancements in Ion Reporter™ Software 5.18.4

Analysis workflow variant calling and annotation improvements and feature enhancements for assays and panels

OncoPrint™ Comprehensive Assay Plus provides comprehensive genomic profiling with analysis of low-frequency somatic variants (SNPs, Indels, CNVs) from over 500 genes, tumor mutation burden (TMB), Loss-Of-Heterozygosity, and microsatellite instability (MSI). The assay includes the following improvements:

- CNV: Updated Gene-level, BRCA exon CNV parameters to enhance performance. Disabled CNV calling from genes with less than 7 amplicons.
- Variant calling: Sequence variant baseline improvements (suppressed recurrent variant in KMT2B)
- MSI: Optimized algorithm for more robust performance across different environmental temperatures
- Fusions: Updated exon tiling imbalance baseline to enhance performance

Support for the OncoPrint™ Myeloid MRD Assay

Ion Reporter™ Software 5.18 supports detection of ultra-low allele-frequency variants and fusions with the new OncoPrint™ Myeloid MRD Assay. The software supports 8 new analysis workflows that feature detection of hotspot and de novo somatic variants (SNPs, indels), gene fusions in targeted RNA libraries, integrated analysis of FLT3 gene internal tandem duplications (FLT3-ITDs), and time series visualizations to track variants for the same sample over time. Visualization graphs can be added to customizable reports for reporting longitudinal MRD test results.

Security enhancements

Ion Reporter™ Software 5.18.4 includes updated versions of two components: Spring Framework v5.3.18 and Apache HTTP Web Server v2.4.53. These updated component versions eliminate two recently identified security vulnerabilities.

Updates for Ion Reporter™ Software 5.18.2

Updates to OncoPrint™ Extended 5.18v2 and TMB (Non-germline Mutations) 5.18v2 filter chains

Ion Reporter™ Software 5.18.2 changes the behavior of the MAF and ExAC filters which are part of the OncoPrint™ Extended 5.18v2 filter chain and the TMB (Non-germline Mutations) 5.18v2 filter chain, the default filter chains included with the new OncoPrint™ Comprehensive Plus Assay analysis workflows w2.3. We recommend that the new OncoPrint™ Comprehensive Plus Assay analysis workflows w2.3, which includes the new filter chains, be used.

To use the updated TMB (Non-germline Mutations) 5.18v2 filter chain, you must copy and edit the Tumor Mutational Burden-enabled analysis workflow (for example, OncoPrint™ Tumor Mutation Load – w3.3 – DNA – Single Sample). In the Parameters step, under Annotation, select the Tumor Mutational Burden tab. Find the Tumor Mutational Burden Filter Chain parameter, then change the value to TMB (Non-germline Mutations) 5.18v2. This change allows calculation of Tumor Mutation Burden (TMB) scores for the OncoPrint™ Tumor Mutation Load Assay that is more similar to Ion Reporter™ Software

5.16 results.

Updates to MAF and ExAC filters

Ion Reporter™ Software 5.18.2 changes the behavior of the MAF and ExAC filters, which are used to create custom filter chains. The MAF component is adjusted in Ion Reporter™ Software 5.18.2 to correct allele normalization. The EXAC component has been adjusted for correct allele normalization and been switched from Allele matching to Locus matching.

New Features in Ion Reporter™ Software 5.18

gnomAD annotations for hg19 analysis workflows

gnomAD (The Genome Aggregation Database), which is the successor of the ExAC database, is now included in Ion Reporter™ Software as an annotation source. The version of gnomAD added to the software is hg19 gnomAD exome data v2.

Non-targeted fusion detection improvements

In the non-targeted fusion detection method, the panel primers are used to detect fusions between novel combinations of known driver and partner genes. The sequencing reads are mapped to a broader reference, such as the whole-exome. Mapping the reads to a broader reference allows for the detection of multiple configurations of driver and partner genes as well as detection of novel breakpoints between the known partner and driver genes.

In Ion Reporter™ Software 5.18, low confidence non-targeted fusions are now identified in VCF output files with key value pairs. The types of non-targeted fusions identified are as follows: out-of-frame sequences, partner-partner fusions, and secondary isoform fusions.

Updates to OncoPrint™ Extended Filter Chain

The non-targeted fusion condition has been removed from the filter chain. High-confidence non-targeted fusions are now annotated with OncoPrint Gene Class and OncoPrint Variant Class by the OncoPrint™ Variant Annotator plugin, and are now filtered in based on this annotation.

Customizable logo in report footers

In previous releases, the footer of reports and report templates created in Ion Reporter™ Software include the Thermo Fisher Scientific logo. You can now remove this logo and replace the image with a custom logo.

Improvements to BRCA analysis workflows

- BRCA Expanded and Gynecological analysis workflows, those that contain both BRCA1 and BRCA2, have Whole Gene Deletion and Exon Deletion/Duplication CNV calling. CNV baselines for these two panels are enhanced for exon deletion/duplication calling.
- All analysis workflows with any BRCA gene detection have BRCA-specific enhancements to TVC parameters and hotspot files, including position-specific parameter overrides and blacklist positions from the OncoPrint™ BRCA Research Assay workflows.

Change to MAF filter in Ion Reporter™ Software

In Ion Reporter™ Software 5.14 and earlier, the MAF filter searches MAFs of all alleles that are associated with a locus. Alleles at the locus found by the filter are returned as filtered results if they fall within the specified MAF range (except `rma=1`). In Ion Reporter™ Software 5.16, the MAF filter searches only the allele(s) that are specified by the genotype (excluding `rma=1`). The allele-based MAF filter results can be downloaded as Filtered Variants.

However, the Analysis Results screen in Ion Reporter™ Software 5.16 continues to show the locus-based MAF values when no MAF filter is applied. As a result, users may observe some variants which have displayed MAF values within the filtered in range are not returned by the new MAF filter on the Analysis Result screen. To avoid the discrepancy, use allele view, which is not expected to have this issue because there is only one variant allele shown on each line.

End of life for Ion Reporter™ Software 5.6 analysis workflows

Ion Reporter™ Software 5.6 analysis workflows are now retired from the Ion Reporter™ Software 5.18. Version 5.6 analysis workflows will also be unavailable after servers are upgraded Ion Reporter™ Software 5.18. Analyses that are launched through analysis workflows from Ion Reporter™ Software 5.6 and earlier will continue to be available to open and create reports for in the latest version of Ion Reporter™ Software. Each new version of Ion Reporter™ Software will continue to retire at least one of the earliest versions of the currently released Ion Reporter™ Software analysis workflows.

Java version required for the Integrative Genomics Viewer (IGV)

Java 8 is required to launch IGV from Ion Reporter™ Software. Java 8 is included with the software. If you are running Java 9 or later, you must have Java 8 also for JNLP to work correctly.

Ion AmpliSeq™ Designer access code is required to import panel designs

An AmpliSeq Designer access code has been added to enhance security for the import of panel designs from ampliseq.com. You must authenticate your account by providing ampliseq.com username and the access code to import panel designs for Ion AmpliSeq™ and Ion AmpliSeq™ HD Made-to-Order, On-Demand, or Ready-to-Use research panels from ampliseq.com into Ion Reporter™ Software. The imported files are used when you create an analysis workflow for use with the Ion AmpliSeq™ Designer panels.

You are required to generate an access code for your ampliseq.com account just one time, if you save the access code when it is generated. If you do not save the access code, you have the option to regenerate a new one when you sign in again on ampliseq.com. You will be required to set a new access code when the code expires.

1. Go to ampliseq.com and sign into your account.
2. Click **My Account > Manage Access Code**.
3. Generate an access code.
 - a. In the Manage Access Code dialog box, select an Access Code Expiration in the dropdown menu.
 - b. Click **Generate**.
 - c. Click **Copy to Clipboard**.

The access code is available for use on the clipboard.

- d. (Optional) Click **Save** to save the access code for use when you return to ampliseq.com.
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You can now use the access code to import panel files or planned runs from ampliseq.com.
For more information, see the Ion Reporter™ Software help system.

Software assessment service

Assessment to help optimize Ion Reporter™ Software analysis workflows for new and established assays on a new version of Ion Reporter™ Software. Contact your bioinformatics support team to learn more.

Issues fixed in Ion Reporter™ Software 5.18

Issue number	Description
IR-48225	A security vulnerability was detected in third party libraries, that is, Spring Framework versions 5.3.17 and below. Ion Reporter™ Software included these third-party libraries and used versions that were impacted by these vulnerabilities. The issue is fixed as part of Ion Reporter™ Software 5.18.4 patch release, which includes an upgrade to Spring Framework version 5.3.18.
IR-48035	A security vulnerability affects Apache HTTP web server version 2.4.46. Ion Reporter™ Software used these third-party libraries and used versions were impacted by these vulnerabilities. The issue is fixed as part of Ion Reporter™ Software 5.18.4 patch release, which includes an upgrade to Apache HTTP server version 2.4.53.
IR-48307	For Ion Reporter™ Servers that were upgraded from the 5.16 image, the Oncomine BCR IGH-LR (5.16 and 5.18) analysis workflows yielded 0 reads. This issue is fixed in Ion Reporter™ Software 5.18.4.
IR-47891	Two BRCA1 Exon 8 amplicons were removed from the OCA Plus Assay baseline. With only one amplicon covering that exon, a potential issue arises if that amplicon is flagged as an outlier by the VCIB algorithm. This causes ‘nan’ values to be calculated for the copy number distribution due to divide by zero errors, which caused the BRCA exon deletion calling pipeline and the overall analysis to fail. The ‘nan’ values are now covered in the VCF record by the BRCA exon deletion algorithm. This issue is fixed in Ion Reporter™ Software 5.18.4.
IR-47890/IR-47397	Memory issues in Ion Reporter™ Software 5.18.0 caused runs to fail occasionally when Oncomine™ Comprehensive Plus - w2.3 (DNA and Fusions – Single Sample, DNA – Single Sample, Fusions – Single Sample, and Annotate Variants – Single Sample) analysis workflows are used. This issue is fixed in Ion Reporter™ Software 5.18.4.
IR-47730/ FST-16768	<p data-bbox="378 1041 1404 1129">Ion Reporter™ Software 5.18.2 includes adjustments to the TMB (Non-germline Mutations) 5.18v2 filter chain and a new Oncomine™ Extended 5.18v2 filter chain for customers with licenses for the Oncomine™ analysis workflows.</p> <p data-bbox="378 1136 1404 1325">To use the updated TMB (Non-germline Mutations) 5.18v2 filter chain, you must make a custom analysis workflow. Copy and edit a Tumor Mutational Burden-enabled analysis workflow (for example, Oncomine Tumor Mutation Load – w3.3 – DNA – Single Sample). In the Parameters step, under Annotation, select the Tumor Mutational Burden tab. Find the Tumor Mutational Burden Filter Chain parameter, then change the value to TMB (Non-germline Mutations) 5.18v2.</p> <p data-bbox="378 1331 1404 1486">In Ion Reporter™ Software 5.18.1.0 and earlier, the preinstalled TMB (Non-germline Mutations) filter chain and the Oncomine™ Extended filter chain allele normalization for long INDELS did not occur properly for MAF and ExAC components. Allele normalization for long INDELS has been fixed and also the ExAC component has been switched from allele matching to locus matching in the updated filter chains.</p>
IR-47762	Eight parameters for fusion detection in AmpliSeq HD analysis workflows for Liquid Biopsy released with Ion Reporter™ Software 5.16 do not match what is used in previous versions of AmpliSeq HD analysis workflows. This change in the workflow files was unintentional. The values are corrected for AmpliSeq HD analysis workflows for Liquid Biopsy that contain fusion detection in Ion Reporter™ Software 5.18. To avoid any potential issues, we recommend that you use the Ion Reporter™ Software 5.18 version of workflows, or Ion Reporter™ Software 5.14 version of workflows or earlier.
IR-47730	In Ion Reporter™ Software 5.18.1.0 and earlier, allele normalization for long INDELS did not occur properly in custom filter chains that included the MAF and EXAC filters. The MAF and EXAC components are adjusted in Ion Reporter™ Software 5.18.2 to correct allele normalization. The fix applies to custom filter chains that use these components.

Issue number	Description
IR-47682	Ion Reporter™ Servers that run on Dell T430 computers displayed total storage incorrectly in the Dashboard screen. The total space was shown as 20 TB, although the server has 15 TB of total usable storage. This issue is fixed in Ion Reporter™ Software 5.18.
IR-47654	The list of filtered out variants in the Analysis Results screen was empty when a custom filter chain that included Drugbank = Filtered In was applied to an analysis. This issue is fixed in Ion Reporter™ Software 5.18.0.2.
IR-47618	The generation of visualization reports for BRCA analyses resulted in corrupt PDF reports if a user started to generate the visualization report without first selecting the BRCA tab in the analysis results screen. The BRCA section was also blank during the report configuration steps. The issue is fixed in Ion Reporter™ Software 5.18.0.2. Users can now generate a visualization report and view the correct images in the BRCA section of the report configuration screen without first opening the BRCA tab.
IR-47676	Ion Reporter™ Servers displayed available storage incorrectly in the Dashboard screen. This information was not updated each time the storage space changed. This issue is fixed in Ion Reporter™ Software 5.18.1.0.
IR-47658	Filtered out Variants tab displayed all the Variants of the analysis instead of the results as in Ion Reporter™ Software 5.18. This issue is fixed in Ion Reporter™ Software 5.18.0.2.
IR-46733	When fusion variants were detected as NOCALL, the No Call Reason column in the Analysis Results table does not represent the reason for no call. The No Call Reason information for fusion variants remained available in the non-Filtered VCF file that is included in the ZIP file for the All Variants download includes all variants for an analysis. This issue did not affect No Call Reason results for other types of variants. This issue is fixed in Reporter™ Software 5.18.
IR-46683	If you launched an analysis workflow that included multiple barcodes and different analysis workflows selected for each barcode, the following error message appeared: "IonReporter:ERROR:Row2:Selected workflow name does not match a previous sample with the same SetID, with workflow name in row 1." The issue did not affect immune repertoire analyses, as launches of multiple immune repertoire analyses at the same time was supported in Ion Reporter™ Software 5.16. This issue is fixed in Reporter™ Software 5.18.
IR-46824	Restoration of archived analyses that originally contained sample(s) that are deleted from the system at the time of restoration was not allowed. An error message indicates that VCF or BAM sample files are missing. This issue is fixed in Reporter™ Software 5.18.
IR-46830	In Immune Repertoire Result, the date and time shown in the Sequenced On column was not correctly displayed. The correct sequencing date and time remained available in the Torrent Suite™ Software and in sequencing run information in the QC Report for the sample that is uploaded. This issue is fixed in Reporter™ Software 5.18.
IR-47662	Changes made to the Tumor Mutational Burden Filter Chain parameter in Tumor Mutational Load analysis workflows did not work correctly after upgrades to Ion Reporter™ Software 5.16. This issue is fixed with the release of software for Ion Reporter™ Server 5.18.0.2.
IR-47659	Users whose emails exceeded 32 characters were unable to complete archive processes and received the following error message. "Sorry, we were unable to complete your requested action. If the problem continues to happen, contact technical support." This issue is fixed in Ion Reporter™ Software 5.18.1.0.

IR-47629	In Ion Reporter™ Software 5.16, when the command "ir_sysinfo" was run, the internal checks to track "Mac address", "IP Address" will not reflect actual values due to the Ubuntu™ operating system upgrade. Another check with for "Network Connection" also does not reflect actual values when the network interface has not been assigned to eth0. This issue is fixed in Ion Reporter™ Software™ 5.18.1.0. Network interface checks are now extended to include em1 and eno1 in addition to eth0.
IR-47664	The Integrative Genomics Viewer (IGV) became unresponsive when zoomed to level 9 and showed an error message. This issue is fixed in Ion Reporter™ Software™ 5.18.1.0.
IR-47713	In Ion Reporter™ Software 5.18.0.0, 5.18.0.1, and 5.18.0.2 there are rare instances when two alleles are adjacent to one another, one a real SNP and the other an INDEL based on a sequencing error. The Sequence Variant Baseline and software code incorrectly used a footprint – the part of reference sequence that is thought to have been changed by an allele – for the variant candidate that was larger than appropriate. This is a rare case and happens only when a complex allele is formed with both a real variant and a sequencing error proximal to one another, and only when the footprint may affect the grouping of alleles. This issue is fixed in Ion Reporter™ Software 5.18.1.0.
IR-47652	Archival and restoration of analyses and samples were not allowed if any of the following processes are in progress: Analyses running or pending, uploads of data through the IonReporterUploader plugin, uploads of BAM or VCF files, transfers of samples, analyses sharing. This issue is fixed in Ion Reporter™ Software 5.18.0.2.
IR-47691	If an analysis was deleted, the samples used in the analysis could not be archived or restored. This issue is fixed in Ion Reporter™ Server 5.18. Samples can be archived and restored if the analysis that uses the samples is deleted.
IR-44642	In Chrome™ browser version 80 and later, an analysis or sample could not be edited by other users in the organization if one user started an edit process, then closed the browser and exited Ion Reporter™ Software before an edit action was complete. The issue occurs when these actions are done in the following screens. Analysis Results screen – If a user clicks a link to open an analysis, clicks Edit, or clicks Actions > Edit in the Analysis screen, then closes the Analysis Results screen before they save edits Generate Report screen – If a user clicks Generate Report from the Analysis Results screen, then closes the Generate Report wizard before the final report generation is cancelled or published. Define Samples screen – In the Samples tab, if a user clicks a sample link (or clicks Define Sample > Manual), to open the Define Samples page, then closes the screen before the Review step is complete and edits are saved This issue was fixed in Ion Reporter™ Software 5.16.
IR-47692	transProbabGapDecay is an unexposed parameter that scales with the Transition Penalty for analyses that use ReproSeq and Low-Pass Whole-Genome Aneuploidy workflows. The parameter was introduced in Ion Reporter™ Software 5.6 with an incorrect constant value for the calculation. 6.02E-5 was used instead of the intended value of 6.02E-7. This issue was fixed in Ion Reporter™ Software 5.14.

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-48456	When customizing a report template with an additional Image section, if the image section is placed below the footer of a page, that image section will not be displayed on the PDF preview or final PDF. Workaround: When you add the Image section, use the section positioning arrows to place the additional Image section above the footer of the page.
IR-47724	When the hostname for an Ion Reporter™ Server is changed, additional “@IonReporterT430” queues are present because the host cleanup script does not remove the default hostname. This issue has no impact on analyses.
IR-48306	Exon Tile baseline is failing with error "Segmentation fault (core dumped)" in rare cases. In tests, this failure occurs once in 150 baseline creation attempts. Workaround: Relaunch the baseline creation will likely resolve the failure since this issue rarely occurs.
IR-48431	The hg19_clinvar_20200329.vcf annotation file contains special characters for some lines. For such cases, the Clinvar track in IGV does not show variant information. This is due to an issue in the IGV application.
IR-48321	The error message "30012 IOException Occurred" occurs when viewing Alpha Diversity plots in metagenomics analysis results for analyses that use only custom reference files and are launched with workflows that are copied and customized, or are newly created.
IR-48221	In Ion Reporter™ Software 5.18, homozygous losses (LOH) data is shown in the Homologous Recombination Repair (HRR) summary table for the genes with homologous copy loss. The correct software behavior is for the HRR summary to show only the CNV values for homozygous deleted genes.
IR-48071	In the OncoPrint™ Comprehensive Plus - w2.3 (DNA and Fusions- Single Sample, DNA – Single Sample, Fusions – Single Sample, and Annotate Variants – Single Sample) analysis workflows in Ion Reporter™ Software 5.18.2, the current stringency level for the gene-level copy loss threshold, maximum fold difference, also known as FD, is causing reporting of likely false-positive calls. This threshold has been updated from 0.85 to 0.70 in Ion Reporter™ Software 5.18.4. Although 0.70 threshold is more stringent, BAP1 copy loss calls will still be called because the reported FD is 0.5.
IR-47914	Multiple BAM files that are used to define a sample in Ion Reporter™ Software must have the exact same Sample Name as it is defined in the RG (Read Group) tag in all the BAM files. If BAM files having different values for SM (sample name) key in @RG tag are used in a single fusion sample the analysis will fail. Workaround: Update the values for SM keys in the BAM files and rerun the analysis.
IR-47889	A RUNX1 insertion, 597dupG, results in a P200A frame shift mutation that is known to be present in a CAP sample has been shown to sometimes have a false negative call if there is a signal discrepancy between the two strands. Workaround: Contact your Field Bioinformatics Scientist (FBS) for further assistance. A fix for this issue is planned for a future version.
IR-47881	Occasionally on sequencing systems under a high computational load, mapping fails on some blocks of data. These blocks do not get merged into the final results and do not get picked by further processing, which leads to gaps in information. This can cause the BAM file to be missing information or otherwise not correctly generated. When the failure to properly generate the BAM files is not correctly identified as an error, the analysis can complete without properly failing, leading to potential missing data. Workaround: In most cases, the issue can be resolved if the analysis is launched again. In rare cases when the problem recurs, downsampling the data set may avoid the issue. Contact your Field Support Representative for assistance with this approach.

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-47872	In the OncoPrint™ Comprehensive Plus - w2.4 (DNA and Fusions – Single Sample, DNA – Single Sample, Fusions – Single Sample, and Annotate Variants – Single Sample) analysis workflows, a stringent SVB BSTRAND threshold is causing an INDEL variant call to be suppressed in one of the system-installed controls. Workaround: Update the SVB BSTRAND threshold to accommodate the expected variant call and test the new threshold on a larger cohort to ensure that this change does not result in new false-positive calls.
IR-47902	The visualization graph for OncoPrint™ BRCA analysis workflows in Ion Reporter™ Software 5.10 is not shown in the software. Workaround: The graph is available in the ZIP file when analysis results files are downloaded.
IR-47743	Exons with GAIN or LOSS values sometimes get incorrectly converted to NOCALL when using the OncoPrint™ Comprehensive Plus analysis workflows. The value is shown in the Filter column in the analysis results screen. This occurs because input VCF records sometimes have Filter values that are not equal to 'PASS' or 'NOCALL'. The software assumes these are the only two values, and converts any non-PASS record to NOCALL. Exons 22-27 are then reported as NOCALL instead of merged into the GAIN variant.
IR-44509	You can use the visualizations in the software to confirm deletions and duplications. Whole gene deletion or duplication results can be confirmed in the pre-corrected view. Workaround: Use the post-corrected visualization to confirm exon deletions and duplications. For example, if a BRCA1 deletion (BRCA1DEL) or a BRCA2 deletion (BRCA2DEL) for a GeneCNV subtype is shown in the Call Details of the CNV results table, review the visualization to verify the deletion of the gene. Click Pre-corrected and Compare the SampleID (sid) amplicons to the BRCA1 genes.
IR-47672	Three amplicons in the OncoPrint™ BRCA Panel have been identified as having lower than typical coverage on one strand. Analysis results that use the OncoPrint™ BRCA Research Somatic - 530 - w3.5 and w3.6 analysis workflows where the threshold for “cov in each strand” is 100 can lead to false negative calls due to inadequate coverage on the requirement for both strands. Workaround: The BED file can be modified to apply reduced min_cov_both_strands value (20 instead of 100) for those three amplicons only.
IR-47870	Batch archival process can sometimes stop running or fail. Contact your Support representative if the issue persists.
IR-47869/ IR-47903	Some NAS storage devices might experience issues when using the device for archiving data. Workaround: If possible, try to use another NAS device. If the issue persists contact your Support representative.
IR-47896/ IR-47610	If large numbers of VCR or BAM files are uploaded with the IonReporterUploader command-line utility on a Microsoft™ Windows computer, uploads may stop. Workaround: Add a maximum of 5 files each time that files are uploaded.
IR-47908	Failed analysis results can occur when the Ion Reporter™ Software analysis workflow OncoPrint™ BRCA Research Somatic - 530 - w3.6 - DNA - Single Sample is used. Workaround: Use a down-sampled BAM file to re-launch the analysis. If you are using Ion Reporter™ Software on Connect and have analyses that fail with the use of OncoPrint™ BRCA Research Somatic - 530 - w3.6 - DNA - Single Sample, contact your Support representative.
IR-48185	In rare cases when barcoded and non-barcoded samples are combined in the same sequencing run, a NullPointerException error is received when downloading variant files in Ion Reporter™ Software. Workarounds: 1) Delete coverageAnalysis plugin output in Torrent Suite™ Software, then re-upload samples using the IRU plugin and complete the analysis on Ion Reporter™ Software. 2) Upload the BAM file using IRUCLI command-line utility by downloading it from Torrent Suite™ Software, then complete the analysis in Ion Reporter™ Software.

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-47734	In Ion Reporter™ Software 5.14, changes were made to genotype parsing that impacted some valid VCF files with a genotype that includes a single allele such as those on the Y chromosome, male nonpseudautosomal regions of X, or mitochondrion. The issue impacts only VCF files that are not generated by Torrent Suite™ Software, Ion Reporter™ Software, or Genexus™ Software. This issue does not impact VCFs generated by these Thermo Fisher Scientific software products.
IR-47689	DRA (Disease Research Area) annotations include links to the external main MESH database main web page. When a user clicks on a specific annotation on the DRA column in Ion Reporter™ Software, a MESH main page opens, instead of the direct link to the page with details for an annotation. Workaround: Use these steps to open the direct link for the annotation: From the main web page that is opened, for example: https://meshb.nlm.nih.gov/search#%2Frecord%2Fui%3Fui=D008577 , copy and paste the ID "D008577" into the search box on the page, then select the MESH Unique ID option and click "exact match" to start a search. This will open the direct link for the annotation: https://meshb.nlm.nih.gov/record/ui?ui=D008577 .
IR-47612	The preview for visualization reports and final reports preview are not available if you use version 14 of the Safari® browser on macOS® Big Sur. Workaround: Use Google Chrome™ browser or Mozilla Firefox™ browser to view the report previews.
IR-47648	The preview for final reports is not available when a custom report template from Ion Reporter™ Software 5.16 or earlier is used for an analysis that is launched in Ion Reporter™ Software 5.18. The issue occurs because a new report footer section that allows users to customize a footer logo was added to Ion Reporter™ Software 5.18. An error occurs with the report preview because the configurable footer section is not present in earlier versions of the software. Workaround: If you want to change the default Thermo Fisher Scientific logo in a custom report template, you must use Ion Reporter™ Software 5.18.
IR-47706	For Fusion isoforms such as MTAP-CDKN2B_AS1_004.M7C5, some fusion panels use a "Do Not Report" flag internally to suppress false calls caused by background coverage. The final VCF file correctly excludes the output for the call. However, the "Fusion Overall Call" in the top of the Analysis Results screen reports the call. Workaround: We recommend that the call in the "Fusion Overall Call" and intermediate files be ignored, unless the call is present in the both Analysis Results fusion data and the final VCF export.
IR-46409	Messaging and information on RAID status on Ion Reporter™ Server might not initially include the correct warning symbol. Workaround: To view the correct RAID status icon, in the Admin tab, click System Services, then click RAID Status and click the icon for the individual drives.
IR-47647	When using Microsoft™ Internet Explorer browser version 20H2, the status of all accounts on the IonReporterUploader plugin configuration screen are red or 'waiting', even though some of the accounts are available to select in the screen. Workaround: Use Google Chrome™ browser or Mozilla Firefox™ browser to view the correct statuses for accounts when configuring the IonReporterUploader plugin.
IR-47510	Upon launching the AmpliSeq Microbiome Health analysis workflows for the same sample, the abundances calculated and reported on the 16S rRNA Gene tab in Ion Reporter™ Software 5.18 (Family Relative Abundance% and Gene Relative Abundance% columns in the visualization) might show some minor variation from one run to another. This occurs when there are reads that have multiple best hits upon mapping to the reference; the mapping method used by the software picks any one target at random, which leads to different estimations. 16S regions are very similar to each other, so it is very likely that a given read is equidistant from several reference sequences. As a result, such results are expected. These variations in analysis results do not impact the overall quality of the results.

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-44638	<p>In Ion Reporter™ Software 5.14 and later, a software update to the Minor Allele Frequency filter which allows users to select the Include Unannotated Variants option causes a discrepancy between the total number of variant rows shown in the table and the counts given in the software for the total number of Filtered In and Filtered Out variants.</p> <ul style="list-style-type: none"> - In Ion Reporter™ Software 5.12 and earlier, the Include Unannotated Variants checkbox for the filter is disabled, so you cannot not select this option, and the Total Variants count does not add up to 100 percent of the variants because unannotated variants are not included in the count of Total Variants. Instead, the Filtered Out and Filtered In variants includes only variants that meet the criteria entered for Filtered In variants, such as a Range of 0.0 to 0.5 plus the Filtered Out variants, but does not include the unannotated variants. <p>In Ion Reporter™ Software 5.14 and later, if the Include Unannotated Variants checkbox is selected for the filter, the count of Total Variants (both Filtered Out and Filtered In) adds up to 100 percent of the variants, as expected because the unannotated variants are included in the Total Variants count. If the Include Unannotated Variants checkbox is not selected, the count of total variants might not add up to 100 percent.</p>
IR-47610	<p>While uploading a large number of multiple VCF files with the IonReporterUploader command-line utility on a windows computer, uploads may stop. Workaround: Add a maximum of 5 VCF files for each file upload.</p>
IR-45389	<p>If exon-levels CNVs are detected in a panel, not all of the CNVs may be represented for that gene in CNV heatmaps. This occurs due to how the software identifies CNVs.</p> <p>In the CNV heatmap for a set of CNVs, for a specific genomic segment, ploidy for variants is analyzed in the ascending order of the variant positions based on the following rules:</p> <ul style="list-style-type: none"> • If a single variant with the highest ploidy is identified, the software analysis will 1) begin with one variant before the segment, and 2) include all of the variants after the segment that have the highest ploidy, and 3) include the single variant that was identified as having the highest ploidy variant. • If there are multiple variants identified as having the highest ploidy, the following variants are included in the analysis: 1) the last identified variant that has the highest ploidy, and 2) one variant before the multiple variants and, 3) all variants after the multiple variants. <p>NOCALL variants are not counted.</p>
IR-31124	<p>When visualizing REFERENCE calls for hotspot alleles in the Liquid Biopsy tab of Analysis Results, some records might have empty values in Mol Counts, Mol Freq, Detection Limits. This happens when consecutive REF calls occur. The empty values are equivalent to values reported in the first record of the group of consecutive REF calls that are found in the genome, and shown in the BED file, or as sorted by the software in consecutive rows of the Analysis Results table.</p>
IR-44334	<p>In audit records, all actions performed by the user are categorized as Add or Modification. Audit Records that include records of a deletion are captured as a Modification. Workaround: Users can view the status of the deleted record as part of the audit record details. To find the record of a deletion, look at the Status field in the audit details. If a record has been deleted, the status will be Deleted.</p>
IR-41058	<p>When using the option to download variants as a current results TSV file from the Analysis Results screen, the column order and variants that are exported in the TSV file are the same as the variants shown in the table of analysis results in Ion Reporter™ Software. However, due to the way data is stored and then sorted in the software, the sort order between the screen and the TSV file might be different. The variants in the TSV file are always sorted by the Locus column.</p>

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-42886	When a CNV record from MyVariants is exported, the export might contain comma-separated ploidy values. When importing the record back into Ion Reporter™ Software, after making edits to the file, the Copy Number field is not imported if the CNV record includes multiple copy number values. Workaround: For a successful import, edit the file to either 1) remove all ploidy values or 2) keep a single ploidy value in the column.
IR-45381	The analysis name is incorporated into Immune Repertoire output files. Long sample names, and/or analyses names which are based on long sample names, can prevent some PDF output files from being generated by the Immune Repertoire analysis workflows. The issue is due to limitations on the number of characters that can be used in file names. To resolve this issue, reduce the length of the analysis name, then re-launch the analysis, or reduce the length of the sample name in Ion Reporter™ Software, then reanalyze the sample.
IR-41021	The list of filters for some filter chains that appear in the Filter Options section of the Analysis Results screen are not ordered consistently. However, the results of the filtering for the variants will appear the same each time that a filter chain is applied. This does not affect how filter chains function.
IR-36949	When you download Ion Reporter™ Software logs on a Microsoft Windows™ operating system, the Microsoft™ Windows 7-Zip File Manager or WinRAR software are the recommended tools to extract the downloaded ZIP files.
IR-44743	The BAM file named "Demo AmpliSeq Exome CNV case" that is included with Ion Reporter™ Server is corrupted and will fail when used with workflows. Workaround: Select another Exome BAM file to demonstrate workflows until the file is updated in a later version.
IR-33625	For OncoPrint™ BRCA analysis workflows, the default canonical transcript has been changed from NM_007300.3 to NM_007294.3. If a custom transcript set is selected when the preferred BRCA1 transcript is NM_007300.3, the exon numbering for BigDup and BigDel CNV variants are still based on the numbering that is in the NM_007294.3 transcript. As a result, Exon 13 in transcript NM_007300.3 is not used in Exon Deletion/Duplication variant calling.
IR-41252	While viewing a specific chromosomal region, if you zoom in on the VCF track in Ion Reporter Genomic Viewer (IRGV), the drawing that represents the CNV can disappear from the screen. This is due to a calculation in the software of the offset for drawing representations of SNVs, CNVs, MNVs, and so on. The CNV is visible again when you zoom in to a point where the retrieved data block of data from the index file is different from the previous block of data.
IR-33725	Column sorting for the "Mol Counts" and "Mol Freq" columns is not correct for ascending sort in the LOD view for liquid biopsy analyses. This is due to the way the data is stored in the internal database.
IR-40286	Quality control for CNV calling of BAM files can fail in several ways. When CNV calling fails, analyses will complete but all CNV hotspots will be NOCALLs and no de-novo CNV calling will occur. One or more of the following can cause the CNV calling to fail: MAPD value is greater than "MAPD threshold," aligned reads less are than "min-read-count," percentage of amplicons with no reads assigned greater than "min-non-zero-amplicons-percent," percentage of reads assigned to amplicons less than "min-aligned-read-percent", or the median read count across amplicons is less than "-min-median-reads-per-amplicon."
IR-41823	In Ion Reporter™ Software, p-values in Analysis Results in the Summary view in the column named "p-value" are rounded to 5 decimal places (between 0.00001-0.99999) when displayed on the screen. Very small p-values that are less than 0.00001 are rounded to 0.00001 by default when displayed on screen. Very large p-values that are greater than 0.99999 are rounded to 0.99999 by default when displayed on screen.

Known issues and limitations in Ion Reporter™ Software 5.18

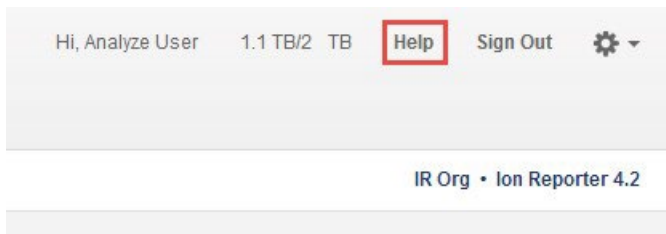
Issue number	Description
IR-39523	Differences in the predefined filter chains and similar custom filter chains can occur if both gene models (refGene and ensGene) are not considered. The gene model used in the analysis must match the gene model that is used by the filter chain.
IR-34781	The non-targeted fusion detection is not fully supported for Ion AmpliSeq™ HD and TagSeq fusions analysis workflows. The molecular family counts for any non-targeted fusion candidates that are observed in the sample are not computed. The molecular family count is reported as 0, and the Detection call is made based on only the read counts, such as is the case with Ion AmpliSeq™ fusions analysis workflows.
IR-35322	<p>The variantCaller plugin for Torrent Suite™ Software 5.10 and later includes the use_fd_param=1 for default Ion AmpliSeq™ HD somatic and germline settings. The parameter, when set to true, uses the new FD parameters. Ion Reporter™ Software 5.10 and later has partial support for FD parameters with the following limitations:</p> <ul style="list-style-type: none"> • The use_fd_param parameter is not visible in the Ion Reporter™ Software, but it can be enabled with the import of a variant calling JSON file that includes use_fd_param=1. • The related min_ratio_for_fd parameter (default 0.1) is not available in Ion Reporter™ Software, and it cannot be set or changed in Ion Reporter™ Software 5.10 and later analysis workflows. <p>Contact your Field Bioinformatics Scientist (FBS) for further assistance if you would like to change these parameters in Ion Reporter™ Software.</p>
IR-33433	In Ion Reporter™ Software, both the Allele View VCF file and the Locus View VCF file includes values in the FDP and DP fields that are the same; however, each allele is displayed as a single line of record in the Allele View VCF file.
IR-34146	VCF files that contain information for more than one sample are currently not supported using Allele (Proper) view. For example, the VCF output files for tumor-normal analysis workflows contain genotype information for both normal and tumor samples. As a result, the analysis is expected to fail, if the Allele view option is enabled in a custom Ion Reporter™ Software analysis workflow.
IR-34768	<p>If user-created Custom sample attributes share the same name as predefined Ion Reporter™ Software sample attributes, the import of samples might use either the custom or predefined attribute, depending on which attribute type is encountered first by the software. This issue is present in Ion Reporter™ Software 5.8 and later.</p> <p>Workarounds: The workaround for this issue is to, ideally, not create custom sample attributes for which an Ion predefined attribute is available. Alternatively, if using custom attributes with the same name as an Ion predefined attribute, you can confirm that the attribute values for the samples use the correct attribute after upload, or you can choose to add the values for those attributes after the samples are successfully uploaded.</p>
IR-34789	When files or reports are downloaded from the Ion Reporter™ software, there will be a mismatch in time stamp data between what is shown on the screens in the software and in the downloaded file. This is due to the difference in user and server time zones. The data shown on the software screens will match the client's time zone and the data shown on the downloaded files will match the server's time zone. One exception to this difference is the visualization report where the downloaded PDF will show client time stamp under the 'launched on' field.
IR-30957	In the results for TagSeq Liquid Biopsy analyses, CNVs contain only the start coordinate position of the variant in the 5.6 version of software.
IR-29790	If you use Table Preferences to add sample attribute columns in the Samples tab that were created in Ion Reporter™ Software 5.4 or earlier, in order to access those preferences in Ion Reporter™ Software 5.6 or later, you must delete the entire Table Preference, then recreate it.

Known issues and limitations in Ion Reporter™ Software 5.18

Issue number	Description
IR-29159	When creating a new annotation-only analysis workflow, OncoPrint™ customers with analysis workflows available to their Ion Reporter™ Software organization will see the availability of the OVAT plugin. Functionality for the OVAT plugin is highly dependent upon the panel used for the assay, so only data from supported panels should be run through analysis workflows containing the OVAT plugin.
IR-24941	When editing a filter chain, canceling during the edit should bring the user back to the filter chain, as it was before the edit with no changes. In Ion Reporter™ 5.4 Software and later, Some edits might persist if a user cancels changes made in filter-chain dialog box. To get back to the same state before edits were made, the page needs to be reloaded to clear all edits made before the cancel.
IR-29709	In some older versions of the Firefox and Safari browsers, the choice of a filter chain will successfully filter the data and list the correct number of variants, but a statement "No Filters Selected" will be present where the filters in the filter chain and their individual filtering results should be listed. A workaround is to use the Chrome browser.
IR-23462	Currently For some older analyses when the BAI file is missing from the input BAM folder, then BAM and Reads Coverage tracks do not load in IGV or Ion Reporter™ Genomic Viewer screens. Workaround: Re-run the analyses, which will generate the BAI file and cause successful loading of both the BAM and Reads Coverage tracks. Ion Reporter™ Software 5.2 does not require you to specify the sample types for RNA/Fusions samples. However, if you do not specify RNA samples as RNA type, you do not get correct results.
IR-23379	In DNA and Fusions analysis workflows, cellularity is not required for NTC RNA samples, but is still required for NTC DNA samples.
IR-25252	When you create an hg19 annotation set, do not use the annotation source DrugBank version 20150107. Use instead annotation source DrugBank version 1 or DrugBank version 20161212. If you use the DrugBank version 20150107 in an hg19 annotation set, you will not be able to create filter chain of DrugBank for any analysis.
IR-31487	A known bug that affected the variant calling module in Ion Reporter™ Software 5.4 caused the results for variants with a very long (>1024 bytes) variant_ID to not be recorded in the VCF file. The variant_ID is a unique key for a variant that, due to improvements to ensure uniqueness for any variant in a sample, was constructed using additional variant data fields in Ion Reporter™ Software 5.2 and 5.4 compared to variant IDs from earlier software versions, which used fewer fields to construct a variant's variant_ID. Rare variants with extremely long (>1024 bytes) variant_IDs were excluded due to exceeding the byte length limit of the variant ID field. The bug was fixed in Ion Reporter™ Software 5.6. Due to this fix, if there is variant in the analysis with a variant_ID length >1024 bytes, the number of variants will be the same for the different analysis workflow versions.

Documentation

You can access product documentation through the help link at the top right of Ion Reporter™ Software. This link opens the help in a new tab (depending on your browser settings).




Software documentation is also available at

<https://www.thermofisher.com/us/en/home/technical-resources/technical-referencelibrary/next-generation-sequencing-support-center/ngs-software-support.html>.

Compatibility with Torrent Suite™ Software

In Ion Reporter™ Software on Connect requires an access code is required to configure user accounts for the IonReporterUploader plugin and to use Ion Reporter™ Software command-line utility (IRUCLI).

To create an access code for use with the IonReporterUploader plugin setup and to use Ion Reporter™ Software command-line utility (IRUCLI):

1. Sign into Ion Reporter™ Software.
2. Click  **(Settings) > Manage Tokens**.
3. Click **Set New Access Code**, then enter an access code in the New Access code field.

The access code must contain at least 6 characters. The maximum length of the access code is 50 characters.

4. Select an expiration time in the Access code Age dropdown menu, then click Save and Generate.

Note: The IRU token is for use with the IonReporterUploader command-line utility and is not required for this procedure.

The access code that you must use to set up the IonReporterUploader is shown in the **Manage Tokens** dialog box and is available on the clipboard. Save this access code for use in future account setups. Alternatively, you can reset the access code as needed.

The IonReporterUploader plugin versions are compatible with the following Torrent Suite™ Software and Ion Reporter™ Software versions.

Version Compatibility Matrix		
Ion Reporter™ Uploader plugin	Compatibility with Torrent Suite™ Software	Compatibility with Ion Reporter™ Software
Ion Reporter™ Uploader plugin 5.18.0.21 or later	Torrent Suite™ Software 5.16 and earlier	Ion Reporter™ Software versions 5.10, 5.12, 5.14, 5.16, and 5.18
Ion Reporter™ Uploader plugin 5.16.0.27 or later	Torrent Suite™ Software 5.16 and earlier	Ion Reporter™ Software versions 5.6, 5.10, 5.12, 5.14, and 5.16


IonReporterUploader plugin

A new version of the IonReporterUploader plugin is available: Ion Reporter™ Uploader plugin 5.18.0.21 or later. This version of the plugin is already installed when Torrent Suite™ Software 5.16 is installed or upgraded.

New plugin versions might become available between released of Torrent Suite™ Software or Ion Reporter™ Software. If you need to upgrade IonReporterUploader plugin on a Torrent Server of version 5.8 or later that is connected to the Internet, you can use the off-cycle upgrade procedures described in the Torrent Suite™ Software User Guide.

If your Torrent Server is not connected to the internet, a debian (.deb) package is available from <http://iru.ionreporter.thermofisher.com>. Use of the debian (.deb) package is the only option to update IonReporterUploader plugin in Torrent Suite™ Software 5.8 and later.

Ion Reporter™ Software command-line utility (IRUCLI)

A ZIP file of the Ion Reporter™ Software command-line utility (IRUCLI). This allows you to upload files to Ion Reporter™ Software from a computer other than the Torrent Server. You can also reach <http://iru.ionreporter.thermofisher.com> through the **Download Ion Reporter™ Uploader** link that is in Ion Reporter™ Software when you click  **Settings > Ion Reporter Uploader**.

The most current IonReporterUploader plugin and Ion Reporter™ Software command-line utility (IRUCLI) are always available for download from <http://iru.ionreporter.thermofisher.com>.

Further information

The plugin version format is “plugin_name p.q.r.s”, where “p.q” represents the major/minor version number of the Torrent Suite™/Ion Reporter™ Software with which the plugin is used. The “r” represents the patch number and “s” represents the number of builds done on this patch.

IRUCLI 5.6 and later requires a Java version of 1.7 or higher to function. Therefore, IRUCLI installed on an older Torrent Server running Java 1.6 does not work anymore unless you perform an additional setup.

IonReporterUploader plugin configuration

Use the following IonReporterUploader plugin configuration setting for Ion Reporter™ Software accounts to transfer data to Ion Reporter™ Software on Connect:

Server: 40.dataloader.ionreporter.thermofisher.com

This is the default shown when configuring an Ion Reporter™ Software on Connect account in the Torrent Suite™ Software.

Note: The previous server address, 40.dataloader.ionreporter.lifetechnologies.com, will continue to work, but please use the new address going forward.

Ion Reporter™ Server: Update Operating System

Ion Reporter™ Software 5.18 is designed to work with the Ubuntu™ operating system version 18.04 and is not compatible with earlier Ubuntu™ operating system versions. Messages can periodically appear asking if you want to update your Ubuntu™ software. Do NOT update your Ubuntu™ operating system, if prompted to do so. Doing so without help from support or the Ion Reporter™ Server-specific instructions causes the Ion Reporter™ Software on the server to stop working correctly due to changes between Ubuntu™ versions.

To upgrade the Ubuntu™ Operating System software to 18.04 on your Ion Reporter™ Server, contact your Field Bioinformatics Specialist (FBS), Field Support Engineer (FSE), or Field Application Scientist (FAS) for assistance.

Contact your local Ion Torrent™ Field Bioinformatics Specialist (FBS) for specific instructions and help to upgrade the Ubuntu™ operating system of your Ion Reporter™ Server. Do NOT update the Ubuntu™ operating system of your Ion Reporter™ Server without the guidance and assistance of an Ion Torrent™ bioinformatics representative. For other options and assistance with updating an Ion Reporter™ Local Server, contact your local FBS.

Update Ion Reporter™ Server Software to 5.18

To upgrade Ion Reporter™ Software to 5.18, you must have an up-to-date software license. Upgrades of the Ubuntu™ operating system is part of a separate server support contract.

The first year of software license and hardware support comes with the purchase of the Ion Reporter™ Server (4487118). In subsequent years, a combined Ion Reporter™ Software license and Hardware support contract (ZG10SCIONSERVER) is required to update the Ion Reporter™ Software through the online command line process, or as part of an on-site visit from a support representative to upgrade the software. This yearly license allows upgrading as many times as desired to as many Ion Reporter™ Software versions as are released during that year. Contact your local FBS, FSE or other service representative for online instructions to update Ion Reporter™ Software or to schedule an onsite visit.

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