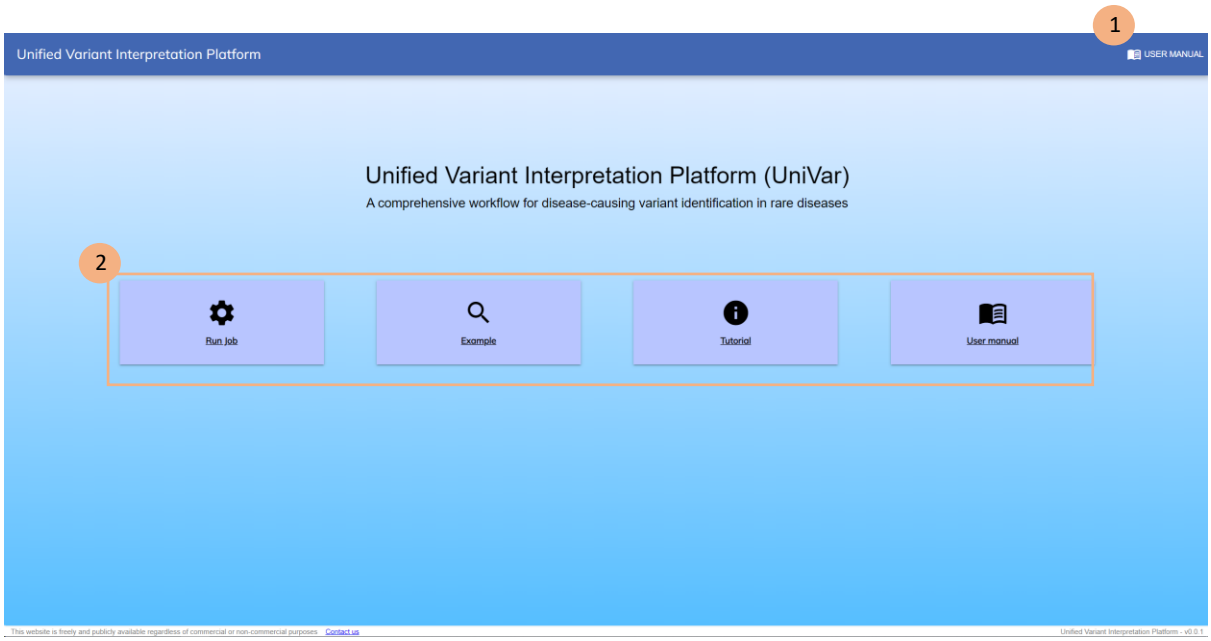
Unified Variant Interpretation Platform

User Manual

Version: 1.1

Table of Contents

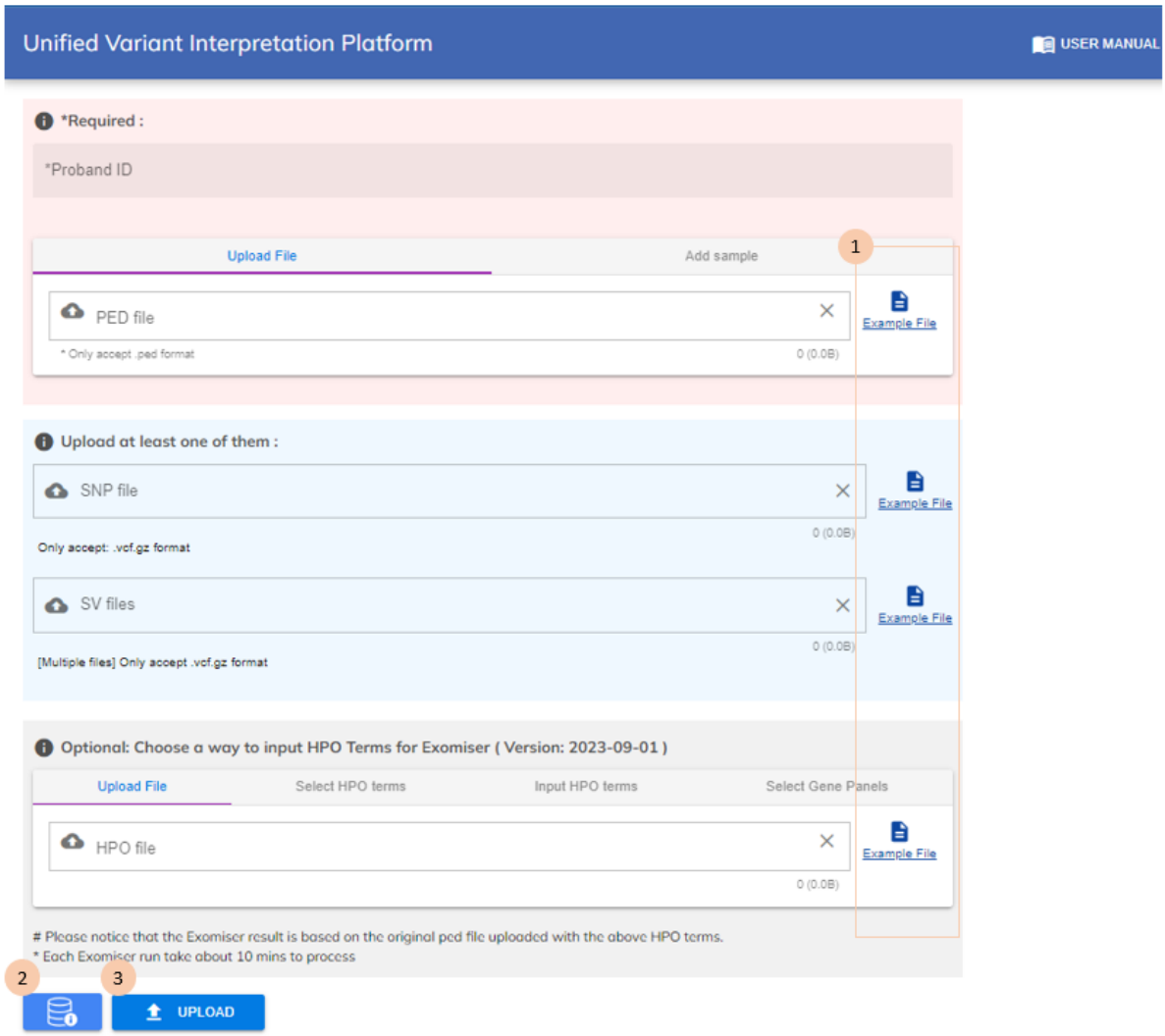
Landing Page



The landing page serves as the entry point for users and provides access to various pages and guides within the application.

1. Link to download user manual in PDF format (this document)
2. Menu buttons: Provide buttons for the following options: "Run Job," "Example Data," "Tutorial," and "User Manual"
    - "Run Job": Allows users to upload their own samples with ped, HPO terms, SNP VCF file, and SV VCF files. Refer to the Sample Upload Page section for more details
    - "Example": Case study demonstration to let first-time users explore the functionality of UniVar.
    - "Tutorial": Offers a downloadable PDF file with step-by-step instructions on how to use the application
    - "User Manual": Provides a download link to this document in PDF format, which helps users understand each page of the application

Sample Upload Page



The Sample Upload Page allows users to provide their own files for executing the UniVar's annotation pipeline and viewing the results in the Variant Tables Page.

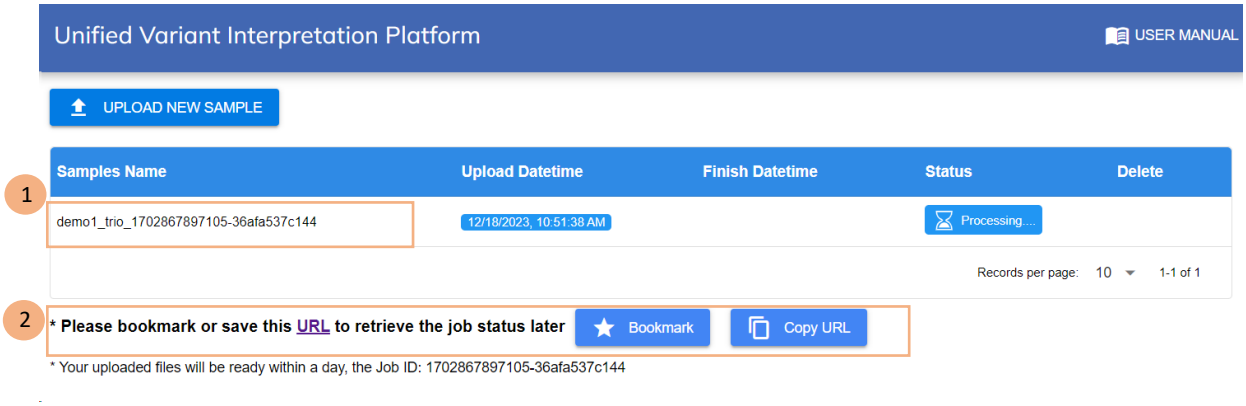
Users are required to provide the following items:

- Familial relationships among the samples (users can upload a PED file or input the details in the "Add sample" section)
- At least one VCF file in vcf.gz format:
    - Small variants file
    - Structural variants file (multiple files with different callers are accepted)
- HPO terms for prioritization (optional)
    - Users can upload an .hpo file
    - Select HPO terms in a drop-down list
    - Input HPO terms in string format, separated by commas, tabs or spaces
    - Select gene panel(s)

1. Example files of each type can be downloaded through the hyperlink provided next to each file selection box. These examples can help users understand the required file formats and serve as a source of example data in the Variant Tables Page.

2. Annotation pipeline information button: allows users to check the current details and version of the annotation database.
3. Upload button: Clicking this button initiates the file upload process, enabling users to submit their files for annotation.

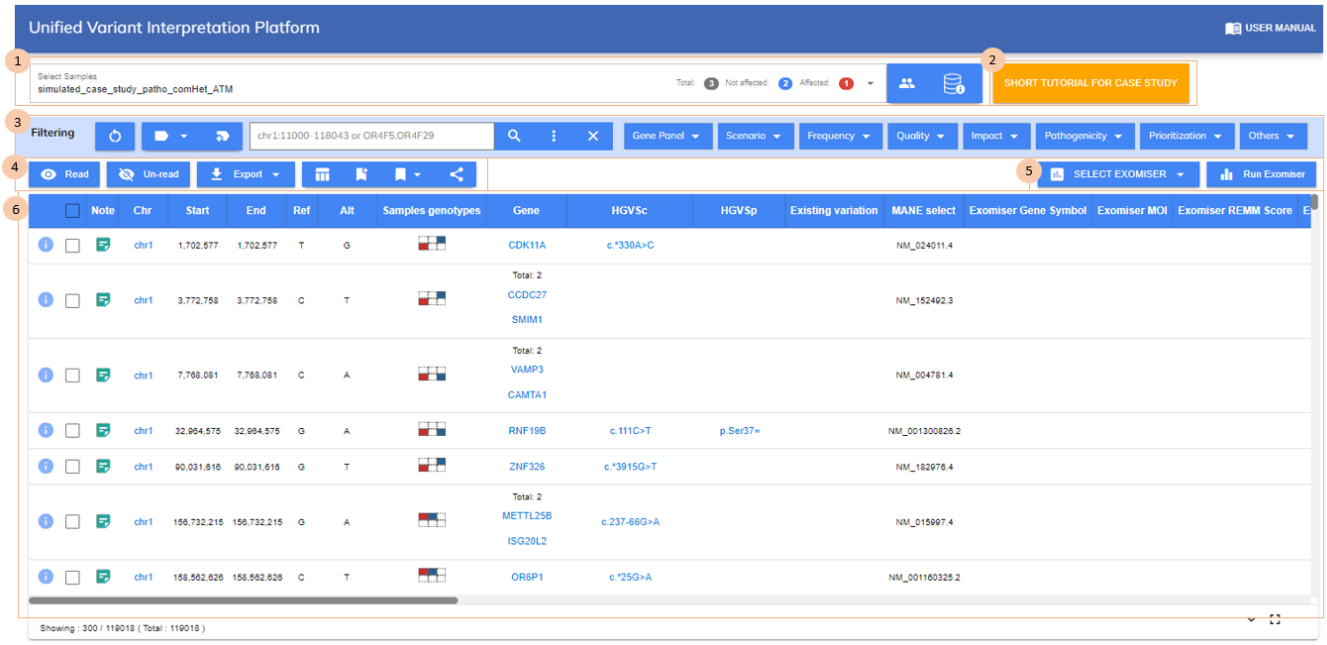
Job Status Page



The Job Status Page allows users to view the status of the files they have uploaded. Each time a user uploads files, a unique and private URL is generated. Users can save this URL to view the upload result later.

1. If the job is finished, users can click on the sample name to enter the Variant Table Page and browse the annotated results.
2. It is important for users to bookmark or save the provided URL. This allows them to retrieve the job status and access the results later.

Variant Tables Page

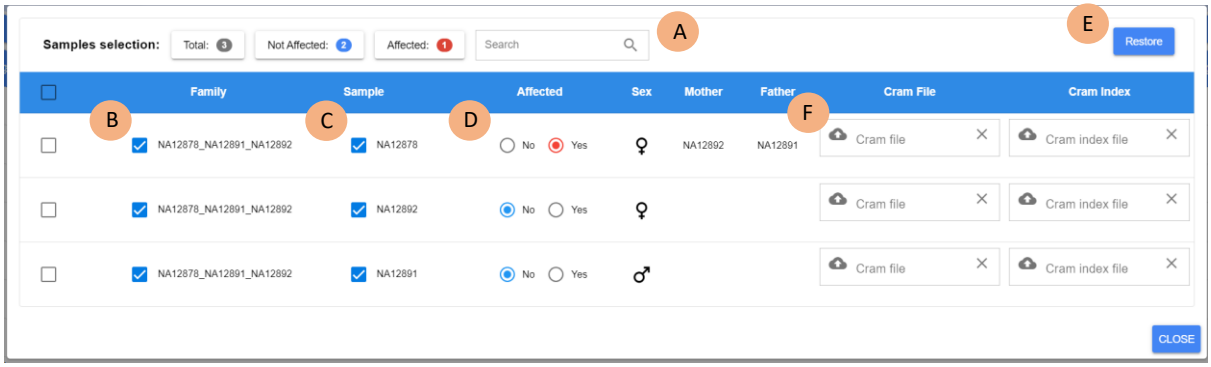


The Variant Tables Page provides detailed information about the annotated variants for further analysis and exploration.

1. Sample Panel: Sample selection, sample status and annotation pipeline information
2. Short tutorial for case study: the steps to identify the disease-causing variants in this case study presented in the paper
3. Filters Panel: Settings for filtering and prioritization
4. Control Panel: Miscellaneous settings for UI layout, data export and bookmarks
5. Exomiser Panel: Manage and submit Exomiser jobs for prioritization
6. Variants Panel: Display variants based on the selected filtering and prioritization options

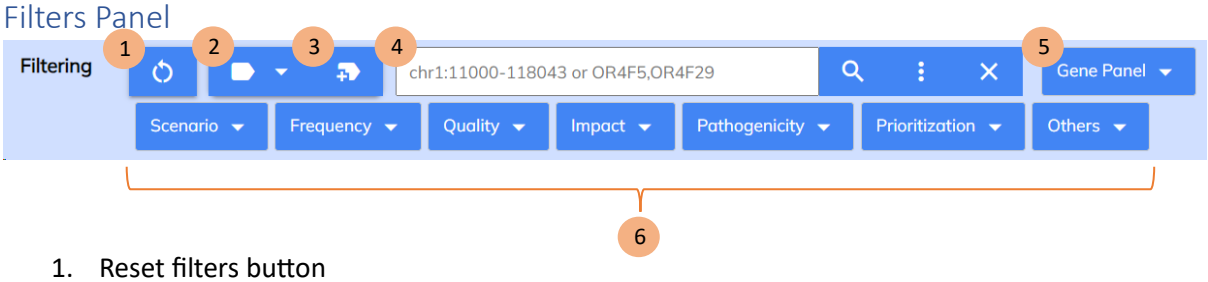
Sample Panel

1. Dropdown box for family selection
2. Sample status: View samples' family relationship, phenotype and gender



   A. Select all samples, only non-affected, or affected. User can use the search bar on the right to search samples based on sample name.
   B. Select/deselect family
   C. Select/deselect sample
   D. Sample phenotype based on the input pedigree file. User can modify the affected/unaffected status shown in the variant panel when necessary
   E. Restore the sample selection and the sample phenotype
   F. For the IGV feature
3. Annotation pipeline version details of the selected family in the dropdown box

Filters Panel

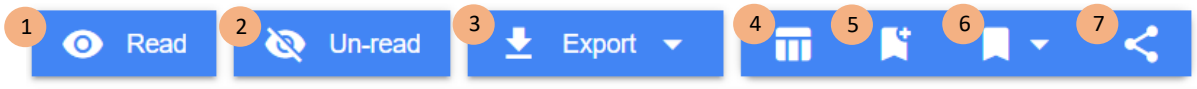


1. Reset filters button
2. Preset filters, including default presets provided by UniVar and presets saved by user. One of our preset is 'High risk (SNV/INDEL + SV)' filter, the parameter consists of (1) any scenarios that correspond to the mode of inheritance (MOI) in Mendelian disease (dominant, recessive, de novo, compound heterozygous and X-linked), (2) AF is 0.005 or less in any of the global population frequency database: gnomAD v2, gnomAD v3, 1KGP and inhouse, (3) genes that have a ClinGen HI score of 3 (sufficient evidence for HI) or 30 (gene associated with AR phenotype), (4) SV that are pLoF or SNV/INDEL that satisfy one of the following conditions: (4.1) high impact in the protein, predicted to cause protein truncation, loss of function or triggering nonsense mediated decay, (4.2) Polyphen score higher than 0.85, or SIFT score lower than 0.05, or CADD higher than 20, or REVEL higher than 0.5, and (4.3) reported as pathogenic or likely pathogenic in ClinVar
3. Save preset button. Save current applied filters as a preset.
4. Genomic location filter. Filter variants by chromosome coordinates or gene symbols. For filtering by gene symbols, multiple values (separated by commas) are supported.
5. Gene panel filters. Filter variants by gene panels from PanelApp UK, PanelApp AU and ClinGen Gene Curation Expert Panels. User can enter keywords to search gene panels. Multi-panel selection is supported.
6. Group of filters. Click to unfold.

Genomic Scenario Filters

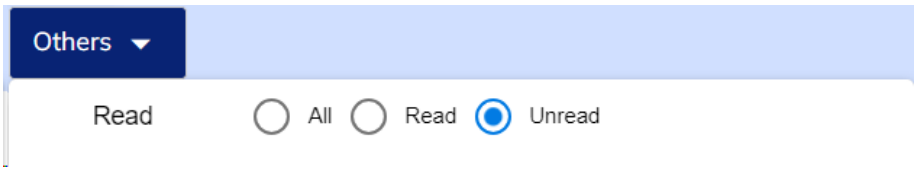
- None: Variants that are present in at least one selected sample
- Any scenario: Variants that match any scenario below (dominant/recessive/de novo/compound het/x linked)
- Dominant: Variants that are present in all affected individuals, and absent in the unaffected individuals
- Recessive: Variants that are homozygous in affected individuals, carried by the parents and not homozygous in unaffected individuals
- De novo: Variants that are present in all affected individuals, and not carried by the parents
- Compound het: pairs of variants affecting the same gene, one being carried by one parent, the second by the other parent, and both present in affected individuals.
- X linked: chromosome X recessive variants, carried by either affected sons and their mother, or by affected daughters (homozygous) and both their parents (and the father is affected). For dominant X-linked variants, use the “dominant” scenario while filtering on “chrX” in the Genomic location search box.

Control Panel



1. Mark selected variants as **Read**, which will showed with different background colour in the variants panel. In Filters Panel >> Others, there is a filtering option for selecting all variants marked as "Read".

2. Mark selected variants as **Unread**, which will showed with default background colour in the variants panel. In Filters Panel >> Others, there is a filtering option for selecting all variants marked as "Unread".



3. Export variants filtering results based on current applied filters as TSV or VCF
4. Column selection panel:

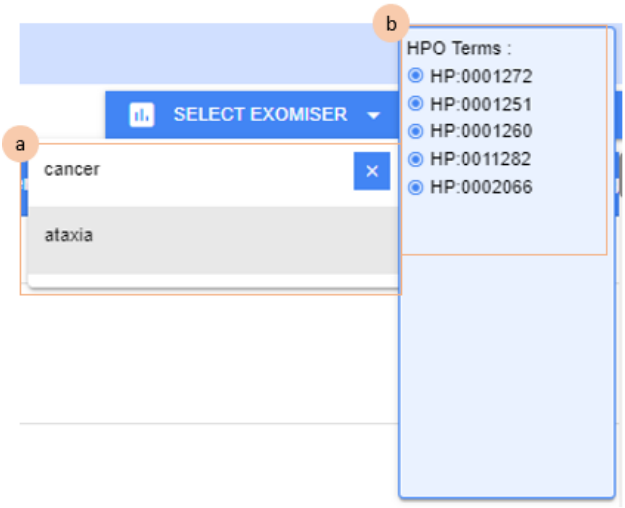
a. Columns for display in the combined view of both small variants and structural variants
b. Columns for display when viewing only small variants
c. Columns for display when viewing only structural variants
d. Freeze columns in the variants panel. Support maximum 5 frozen columns.

e. Current selected columns for display. To add a column, drag a column from the “Hidden Columns” panel and then drop in the “Display Columns”, and vice versa. Columns can be reordered using drag-and-drop.
f. Columns that are currently hidden
g. Dropdown list for filtering the hidden columns by category
h. Restore button. Reset the column selection settings to default.
i. Close button. Close the column selection panel

5. Save bookmark button. For saving the current analysis settings, like filtering, sorting, and column selection options.
6. Bookmark list button. For retrieving the saved analysis settings.
7. Sharing button. Copy a URL to clipboard for sharing the current analysis state with others who have same access to the selected family. Another user who opens this URL can see the same filtering and prioritization results in the variants panel.

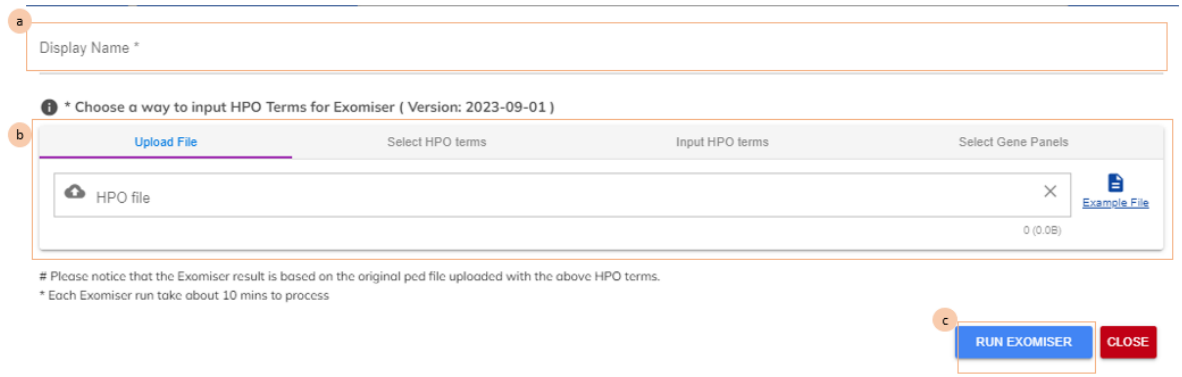
Exomiser Panel

1. Manage Exomiser jobs



a. Select a job from the drop-down list of Exomiser jobs to display the corresponding Exomiser results in the Variant Tables Page.
b. The selected HPO terms for each of the Exomiser jobs

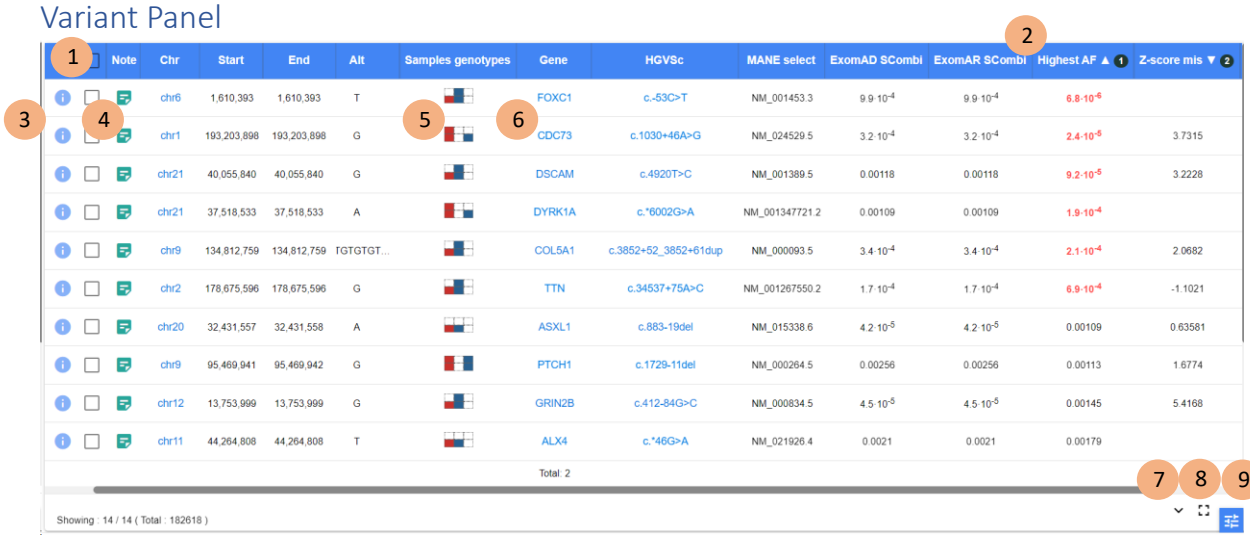
2. Submit Exomiser jobs



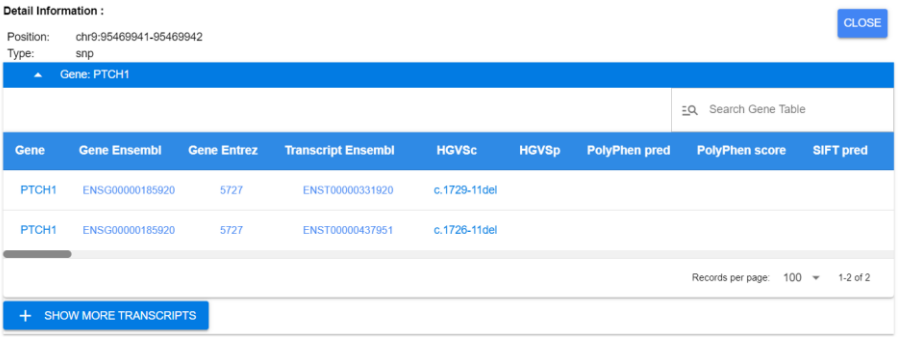
a. Enter a name to define this Exomiser job.
b. There are four ways to input HPO terms
- Upload an HPO file
- Select HPO terms from a drop-down list
- Input HPO terms in string format, separated by commas, tabs or spaces.
- Select gene panel(s)

c. Execute the current Exomiser job

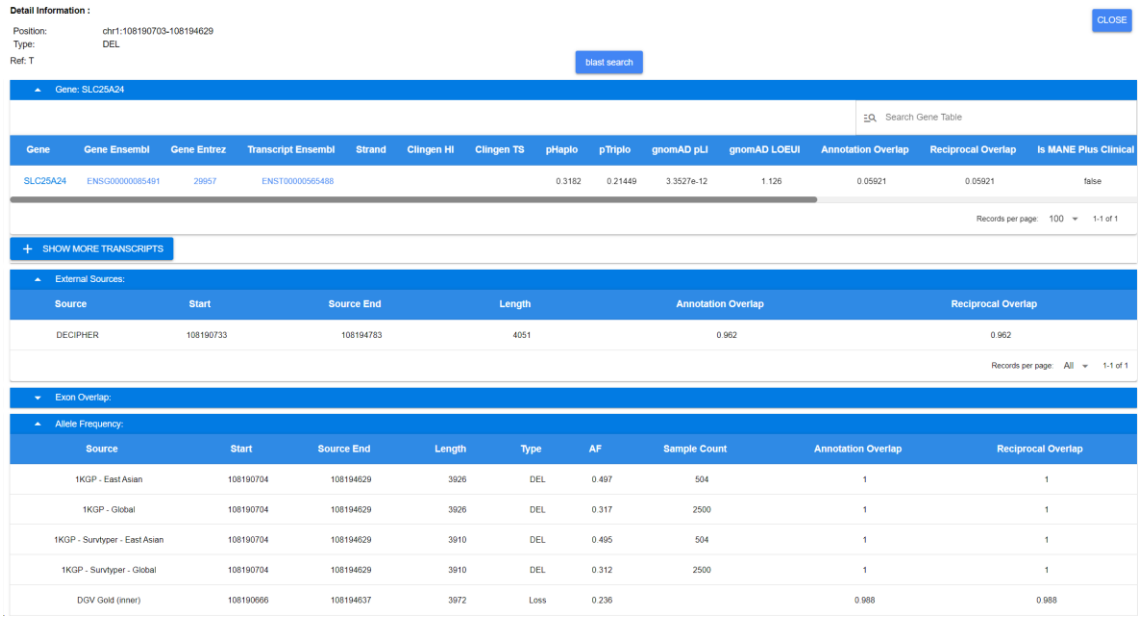
Variant Panel



1. Select variants to mark as “Read”/”Unread”
2. Click column names to sort by values. The arrow next to the column name indicates the sorting direction, which can be changed by clicking. Multi-column sorting is supported. The rightmost digit in the column header represents the sorting priority. To cancel column sorting, click the digit of the column header.
3. Detail information of the variant.
    - Small Variant: Show annotated transcripts information. Non-MANE transcripts are hidden by default. Click the “Show more transcripts” to view all transcripts.

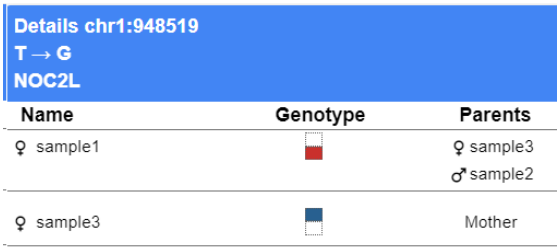


    - Structural Variant: Show annotated gene, related variants (overlapped external database records like Decipher), external sources, exon overlap, clinical interpretation, AF and Exomiser results.

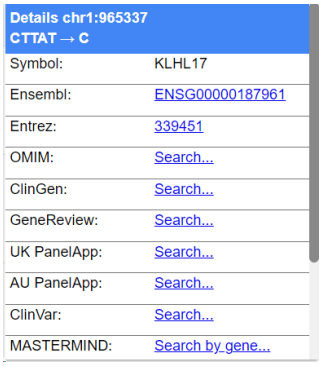


4. Note button. Click to save a note for a variant. In Filters Panel >> Others, there is a filtering option for selecting all variants with notes.

5. Genotypes. Each column represents one sample, and each square represents an allele. A square is filled if the allele is present. Affected sample is showed in colour red, and unaffected sample is showed in colour blue. User can click the value to view the details, like sample name and family relationship.

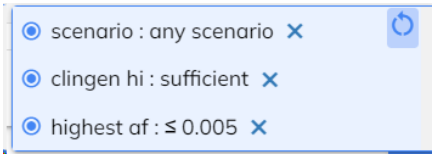


6. Annotated gene. Click the gene symbol can open a pop-up window for linking to external databases like Clingen, OMIM, GeneReview etc.



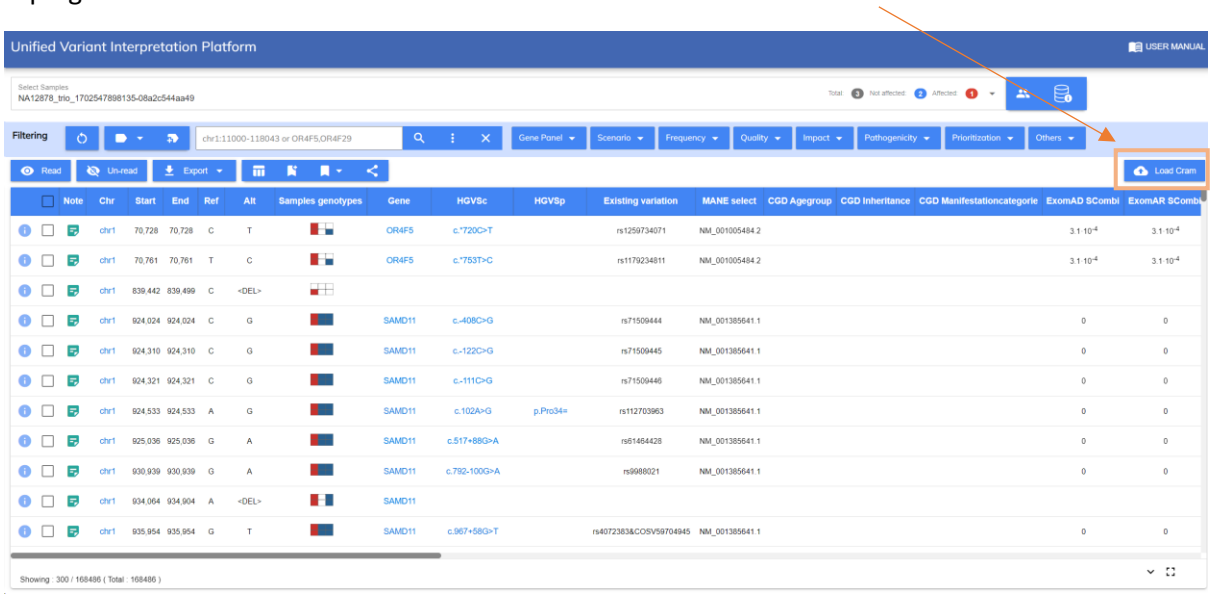
7. Expand button: load more variants into the variant list for scrolling.
8. Full screen button: show the variants panel in full screen mode.

9. Filter summary button: show all the applied filters. User can click the delete button to remove filter or the reset button to clear all filters when necessary.

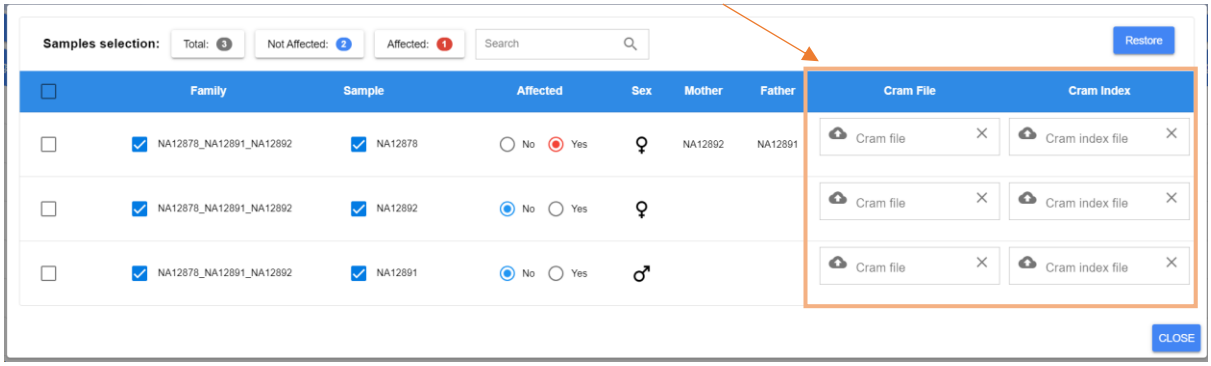


Reads alignment: Integrative Genomics Viewer (IGV)

If the local path of cram files is provided, user can view reads alignment of variants through Integrative Genomics Viewer (IGV). To use this feature, user first needs to click the "Load Cram" button on the top right corner of the variants table.



Then user can specify the local storage location of the cram files and the cram index files in the pop-up panel.



After that, user can view reads alignment of a variant by double clicking a row in the variants panel. An IGV will be opened with one track per sample, centred at the selected variant's position.