KARYOMAP V2
KMAP SOFTWARE GUIDE

Vitrolife

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3. The Karyomapping products attempt to make a prediction of the genotype phase in test samples, however, the user is responsible for examining both the supporting and contrary evidence for this predicted phase before making a determination concerning the true phase. Results obtained from the use of KaryoMap v2 products should be confirmed using alternative methods. The KaryoMap v2 products may generate false positives, false negatives, or inconclusive results.
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    c. Consanguinity between relatives
    d. Sex chromosome abnormalities

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Revision history

Version	Date	Description of Change
1.0	28-Sep-2023	Initial release.
2.0	05-02-2025	• Multifactor Authentication (MFA) details added- Page 13 • Gene nomenclature changed from OMIM to HGNC- • Minor language updates • Page and table references updated

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1 INTRODUCTION

Karyomapping is a comprehensive method for genome-wide linkage-based analysis, to detect the inheritance of parental haplotypes. The KaryoMap v2 Solution utilises the Infinium BeadArray technology to generate genome-wide genotyping data for father, mother, reference (genomic DNA extracted from blood) and test samples (MDA-amplified DNA from 1-10 cells). The kMap software is a cloud-based software specially developed to support the KaryoMap v2 solution. The kMap analysis software (kMap) enables accessioning, laboratory planning, sample tracking, data analysis and reporting in a single software solution. This guide describes the features and functions of the kMap software. **Training is highly recommended before using the kMap software.** Refer to the ***"A Technical Guide to Karyomapping"*** for guidance on interpreting challenging data.

The kMap software user interface has been designed to guide the User through Accessioning, Laboratory Planning and Analysis, allowing faster assessment of results. A high-level overview of the kMap workflow to process samples through the KaryoMap v2 solution is shown in Figure 1.

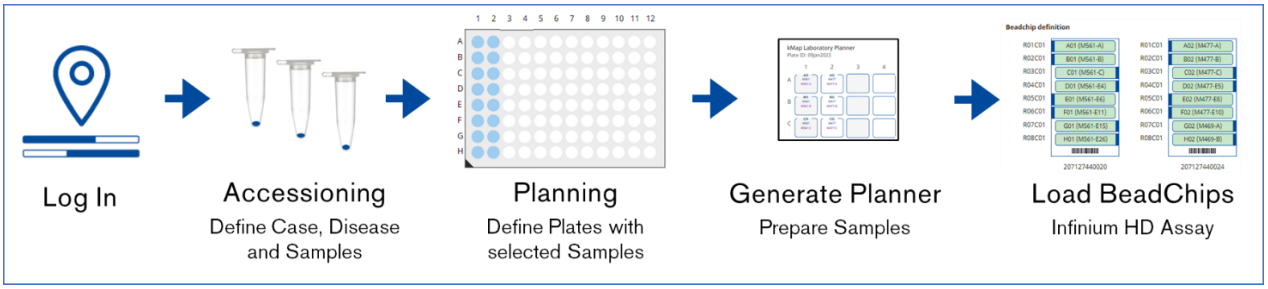


Figure 1: Run planning in the kMap software.

The genotyping data is generated in the form of GTC files after scanning the processed KaryoMap v2 BeadChips with the iScan or NextSeq 550. Scan data is then manually uploaded using the kMap-Uploader software to the User's kMap account and matched with the planned case definition and data is analysed automatically. Analysed data can be reviewed in drill-down levels, with each level providing more detailed information about the case and samples (Figure 2).

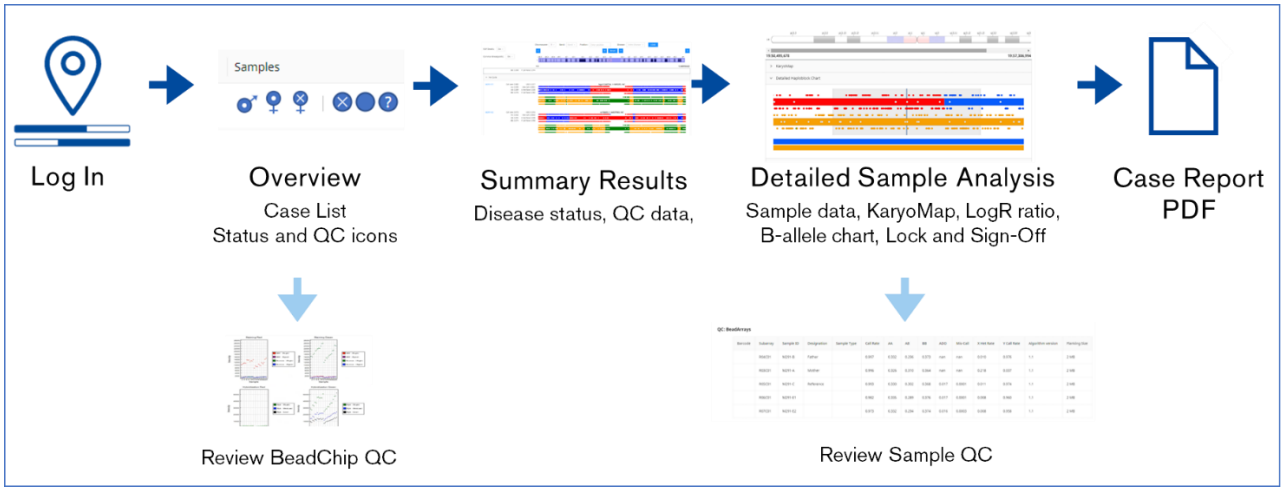


Figure 2: Data review and reporting in the kMap software.

Getting Started with kMap.

kMap Components

kMap-Uploader Software: The Uploader Software is a set of Microsoft Windows™ executables that must be installed on the scanning instrument (iScan or NestSeq 550) or a computer with access to the KaryoMap v2 BeadChip scan data. Installation and usage are described in *Appendix 4*.

kMap Analysis Software: The kMap analysis software is a cloud-based solution for the management and analysis of data generated through the KaryoMap v2 solution. kMap software can be securely accessed from any networked computer using an internet browser and a personal login.

Use of both software components are regulated by the Terms and Conditions outlined at https://eu-central-1.kmap.vitrolife.com/terms for users within the EU and https://ap-southeast-2.kmap.vitrolife.com/terms for users in other regions (also available in the kMap webpage footer).

Accessing kMap

The kMap software is available and free-to-use for all KaryoMap v2 customers. Each customer will be provided with a kMap account for their Laboratory or Group, which they can self-administer to add additional Users or adjust software Settings. Two different levels of User accounts can be configured to control access to specific software features between Users. Further details on the User and Group options are explained in the *kMap Account* section. Once each User has their own login details, they can access their kMap Group account from https://kmap.vitrolife.com. We strongly recommend Users to keep their password secure and log out from the system after completing a session.

The menu links ensure easy navigation through the software and are shown in the top right-hand side of the browser on all pages (Table 1).

Table 1: kMap navigation menus

Menu	Navigation Links
HOME	View the kMap Dashboard
PLANNING	Accessioning, Laboratory Planner
DATA	Case List, Bead Arrays, Report Builder, Log File
ACCOUNT	Profile, Manage Users [Level 2 Users only], Settings, Support, Logout

Search

The text box on the top right-hand corner of every page offers global search functionality for Accessioning and Result data within the specific Group's database.

1. Enter text and press enter to search.
2. The result page is split by the result type (Analyses, Samples, Cases).
3. The search term is highlighted, and links provide access to the source pages of the data.

Analyses, Samples and Cases have sort functionality built-in for all the fields. The sort function is indicated by the sort tool in the column headers. Sorting is done ascending or descending.

kMap Account

User Management

Each Group account can be self-administered to create or modify User profiles for team members in kMap. To allow traceability of changes to the data as part of GxP compliance, each User should always log in with their own credentials. The account levels are:

- **Level 1** for standard Users performing sample preparation and initial data analysis.
- **Level 2,** same as Level 1, with additional privileges to:
    - Administration of Users
    - Modify System Settings
    - Sign-Off samples for reporting

1. Navigate to *ACCOUNT* > *Manage Users* (only available to Level 2 Users)
2. Review the list of Users. Search , sort and filter options are available in the column headers.
3. Click + New to create a new User or click to edit an existing User account.
4. Select a User with the checkbox and select Delete to remove a User.

User Profile Configuration

1. Creating or editing a User profile allows the input of:
    a. First name
    b. Last name
    c. E-mail address used for login and receiving system notifications
    d. Set or Update a password
    e. Select profile Role privileges, Level 1 (Restricted) or Level 2 (Admin).
    f. Apply Technical Support restrictions (see below).
2. Click Save to apply changes.

Technical Support Access

A special type of User profile can be created by a Level 2 User (account admin), for allowing external support (including Vitrolife representatives) or for the (temporary) sharing of data. Technical Support User profiles are configured such that privacy-sensitive data is obfuscated (made un-readable) when this User logs in.

1. Technical Support accounts are controlled by the database owner (account admins) and should only be created when Technical Support is required. In most cases support can be given without needing to share access to your account.
2. The account admin is responsible for sharing the Technical Support login information and deleting the User profile or updating the password once the required support has been provided.
3. When creating a new User, or editing and existing one:
    a. Use the selector to enable Technical Support restrictions Technical Support YES NO.
    b. Click Save to apply changes.
4. The data fields in **Table 2** will not be readable to the Technical Support User profile, which also applies to the system Logs visible to the User:

Table 2: Obfuscated data fields are applied when Technical Support restrictions are enables for a User profile.

Parameter	Obfuscated data fields
Software User	First name, last name, email address
Subject	First name, last name, date of birth, partner first name, partner last name, notes
Data	GTC file (not available for download)

Note: User profiles with Level 1 permissions and the *Technical Support* option **enabled** will not be able to Manage Users or disable (self-administer) the *Technical Support* restrictions.

5. System IDs (Case, Subject, Cycle, Sample and Plate IDs) are **not obfuscated** to allow support for specific cases or analysis and therefore should be chosen to not contain any personal, sensitive information in routine use of kMap.
6. When the support case is resolved, the account can be deleted, or the password changed until required again.

User Profile

Profile

All Users (Level 1 and Level 2) can set or update options on their own profile page with the following steps (Figure 3).

1. Log into kMap.
2. Navigate to *ACCOUNT* > *Profile*.
3. Click the area under "Profile" to upload a profile image (maximum size 2 Mb).
4. Select to enable e-mail notifications for analysis status and system errors, using the address used to log into kMap.
5. Select system language (where available), and then click to Save.
6. Click to change the password, re-enter, and then click to Save.
7. Click **Access Keys** to view the unique access credentials for this database for performing data uploads to the kMap software. Never share the Access Key with anyone outside your laboratory/group. See *Appendix 4* for kMap-Uploader software installation instructions.

Chart Settings

1. Select the default Preferred Charts to be displayed (currently KaryoMap only).
2. Select the default Expanded charts to be displayed in expanded or hidden view by default (Detailed Haploblock Chart, LogR Chart and B-allele Chart).

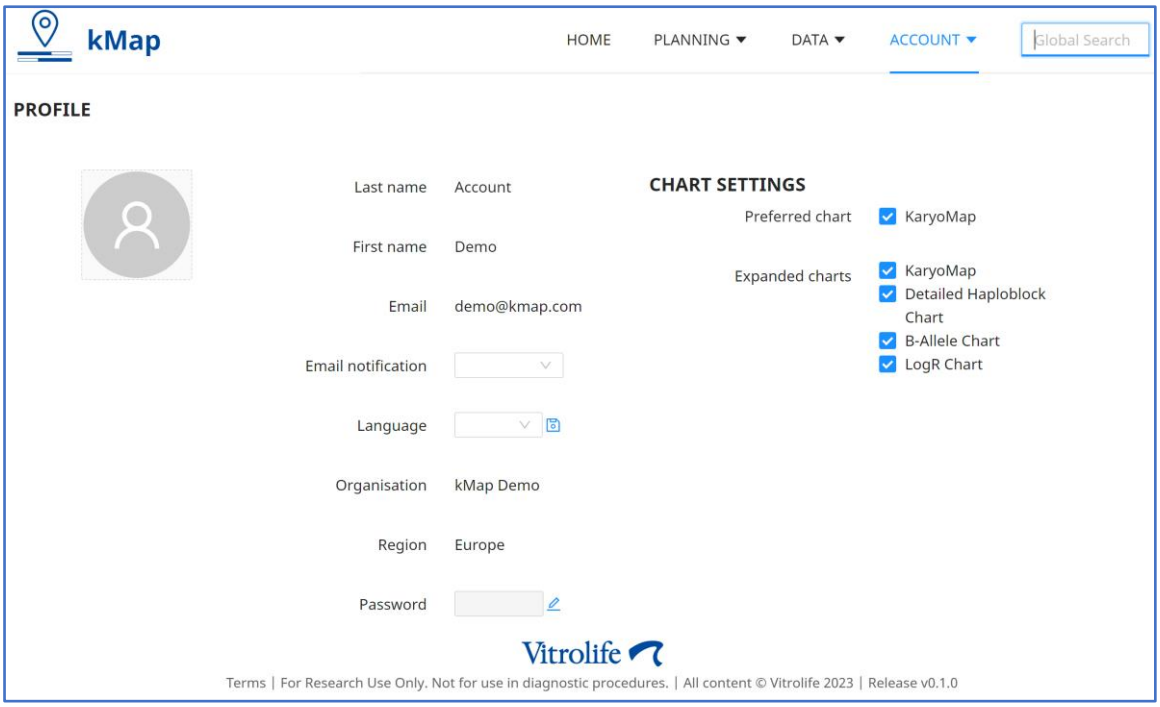


Figure 3: kMap User Profile page

System Logs

For traceability, actions performed within kMap are recorded in the system log. This log can be reviewed and downloaded from the *DATA* > *Log file* menu.

1. The time of the event in universal time, the action and the Username are recorded.
2. Use the column header tools to search, sort and filter results.
3. Click Export to TSV to export and archive the system logs in tab-delimited.tsv format.

Detailed logs for the data analysis are displayed separately on the *Case View* page in the *Log* tab for the respective *Analysis ID*. Critical errors and QC failures will also be displayed as *Alerts* on the *HOME* page.

Settings

Navigate to the system *Settings* page by clicking *ACCOUNT* > *Settings*. The *Settings* page allows Users to customise the "Report Settings" and adjust the "Analysis Settings" as required.

The settings should be reviewed before using the kMap software. Changes made on the *Settings* page via the *ACCOUNT* menu will apply to all Users in the Group, and all KaryoMap datasets analysed thereafter. Previously analysed data will not be affected by changes to the settings parameters until the case(s) are re-analysed.
The tool tip symbol provides a short description for all available Settings; hover the cursor over the icon to reveal the text. Modification of system Settings can only be performed by Level 2 Users.

1. Press Save after modifying any values.
2. Modified analysis Settings will be applied to all analyses initiated after saving new values.

- Click Restore defaults to revert parameters to Vitrolife default values, except for Report Settings, Brute Force and MFA protection settings.

3. Click Cancel to discard any changes and return to the previous page.

Report Settings

The settings described in Table 3 will be applied as default values when using the *Report Builder* tool, although Users have the option to modify the values during the generation of individual reports.

Table 3: Report Settings

Parameter	Description	Default Values	Allowed Values
Logo	Upload an image file to be visible at the top of the PDF Report. Maximum size 2 Mb.	None	png, gif or jpeg files
Address	The laboratory/group address to be visible at the top of the PDF Report.	None	Any text
Disclaimer	A line of text visible in the footer of the PDF Report, e.g. a specific disclaimer.	None	Any short text
Report introduction	A block of text visible on the first page (case summary) of the PDF Report, e.g. description of the test and reporting criteria.	None	Any text
Charts Included	Use the selector (tick box) to include the KaryoMap, LogR Chart and/or B-allele Chart.	No	Yes/No

Analysis Settings

Table 4 describes the settings that are required for the Analysis algorithm. It is recommended to establish the settings suitable for your Group, especially the flanking region size.

Table 4: Analysis Settings

Parameter	Description	Default Values	Allowed Values
Flanking Region Info	The size (in Mb) of genomic regions (DNA sequences) on either side of the region of interest.	2	0.5 - 5.0
Scanning Hardware	The Instrument type used for scanning the Infinium KaryoMap v2 BeadChips.	iScan	iScan, NextSeq 550
BeadArray type	The BeadArray product name.	KaryoMap v2	KaryoMap v2

Miscellaneous Settings

Table 5 describes additional settings relating to the kMap user interface and may be configured according to the Group needs.

Table 5: Miscellaneous Settings

Parameter	Description	Default Values	Allowed Values
Enable Brute Force Protection for all users	Enable protections to limit the number of unsuccessful log in attempts before applying a temporary block of the username and notifying the Group's Level 2 users.	No	Yes / No
Enable MFA for all users	Enable Protections requiring all Group Users to setup Multi-Factor Authentication for access to kMap	No	Yes / No
PDF Guide Key 1	Choose an appropriate ID to be displayed on the laboratory Planner PDF (first line).	Case ID	Case ID, Sample ID, Tube ID, Cycle ID, Requisition or Blank
PDF Guide Key 2	Choose an appropriate ID to be displayed on the laboratory Planner PDF (second line).	Sample ID	
PDF Guide Key 3	Choose an appropriate ID to be displayed on the laboratory Planner PDF (third line).	Tube ID	
Session Timeout	Number of seconds of inactivity before the browser session will expire, requiring password to re-entry.	600	60-3600

Account Security

Brute Force Protection

When enabled, the Brute Force Protection will:

1. Warn the user when incorrect login credentials have been submitted.
2. Display the number of login attempts remaining before the user is locked-out.
3. Lock-out the user account for 5 minutes after three failed login attempts.
    a. A countdown will show the lock-out time remaining.
    b. Repeated login attempts during a lock-out period (including with correct credentials) will restart the lock-out period. Please do not refresh the login page during this time.
4. Display failed login attempts by username in System Logs.

Notify Group Account Level 2 users by e-mail of the failed login resulting in a lock-out, and the user's IP address.

Note: E-mail notifications are dependent on the Level 2 user settings in *ACCOUNT > Profile*.

Brute Force Protection is applied at the Group Account level (for all users) but enforced at the username level. Therefore, if one user is temporarily locked-out, a different user of the same Group Account will still be able to log in with correct credentials.

Multi-Factor Authentication

When enabled for the Group Account, each user will be directed to their *ACCOUNT > Profile* page immediately or upon next login. Each user is required to setup the Multi-Factor Authentication (MFA) before access is granted to eMap (this includes access to the *Settings* page). There are two options for MFA setup. One or both can be configured for each user:

1. MFA Authenticator App

a. The user will need to download and setup an MFA Authenticator App (e.g Microsoft Authenticator, Google Authenticator, or other) on a smartphone or tablet with camera access.
b. In eMap, the MFA Authenticator App option will show Not Enabled by default, indicating verification setup is required.
c. Click the configure icon to display a unique QR code and verification entry:
Enter code Verify Cancel
d. Generate a verification code using your Authenticator App:
    i. If using e.g. Microsoft Authenticator on a smartphone, select the "Verified IDs" tab and then "Scan QR code". Scan the eMap on-screen QC code with the device camera, then select the Authenticator tab to view the current verification code. Enter the code into eMap and click Verify
    ii. If using e.g. Google Authenticator on a smartphone, select "Add a code" and then "Scan a QR code". Scan the eMap on-screen QC code with the device camera to generate a response code. Enter the response code in eMap and click Verify
e. In kMap, the MFA Authenticator App option will show Enabled to confirm setup is complete.
f. Clicking the delete icon will remove this MFA setup. If the user has no enabled MFA options and MFA is enabled at the group Account level, the user will be prompted to complete setup again before eMap access is granted.

2. MFA e-mail
a. Click the configure icon to send an authentication code to the e-mail address that associated with the eMap user account. Enter code Verify Cancel
b. b. Enter the MFA code contained in the e-mail and click Verify.
c. c. In eMap, the MFA e-mail option will show Enabled to confirm setup is complete.
d. d. Clicking the delete icon will remove this MFA setup. If the user has no enabled MFA options and MFA is enabled at the group Account level, the user will be prompted to complete setup again before eMap access is granted.

3. Logging into eMap:
e. Upon login the user will be presented with the MFA approval screen.
f. If multiple MFA methods have been configured, select [Authenticator App] or [E-mail] from a list.
    i. Authenticator App: Enter the MFA code displayed in the app on your smartphone or tablet, then click Verify MFA Code.
    ii. E-mail: Click Send MFA Code, enter the code received and click Verify MFA Code.
g. Selecting the Trust device checkbox will add the current device (computer) to a trusted list for 30 days. During this time, MFA will not be required for this User-Device combination when logging in to eMap.

4. MFA management
a. If a user cannot use an existing MFA configuration to log in to eMap, another Level 2 user of the same Group Account can reset the affected user's MFA setup though the ACCOUNT > Manage Users menu.
b. From the Manage Users table, click Edit on the affected user.
c. Click Reset MFA, then read and confirm a warning message.
d. The affected user will be prompted to setup MFA again, before access to the Group Account is granted.

Home Dashboard

Upon logging into kMap, Users will be presented with a dashboard (Figure 4) providing an overview of the following:

- **Alerts**: Important recent system messages.
- **Recent Cases**: Expandable view of cases with Analysis ID, Analysis name, Reference sample name, Reference type, and all Samples in the case with a quick overview of individual sample statuses.
- **Recent BeadChip**: Expandable view of BeadChips associated with each Plate ID with Sample details on each subarray for the BeadChips.
- **Statistics**: Summary plots of Signed-Off samples over time.

Return to the dashboard at any time by clicking on *HOME* in the top right-hand side of the browser.

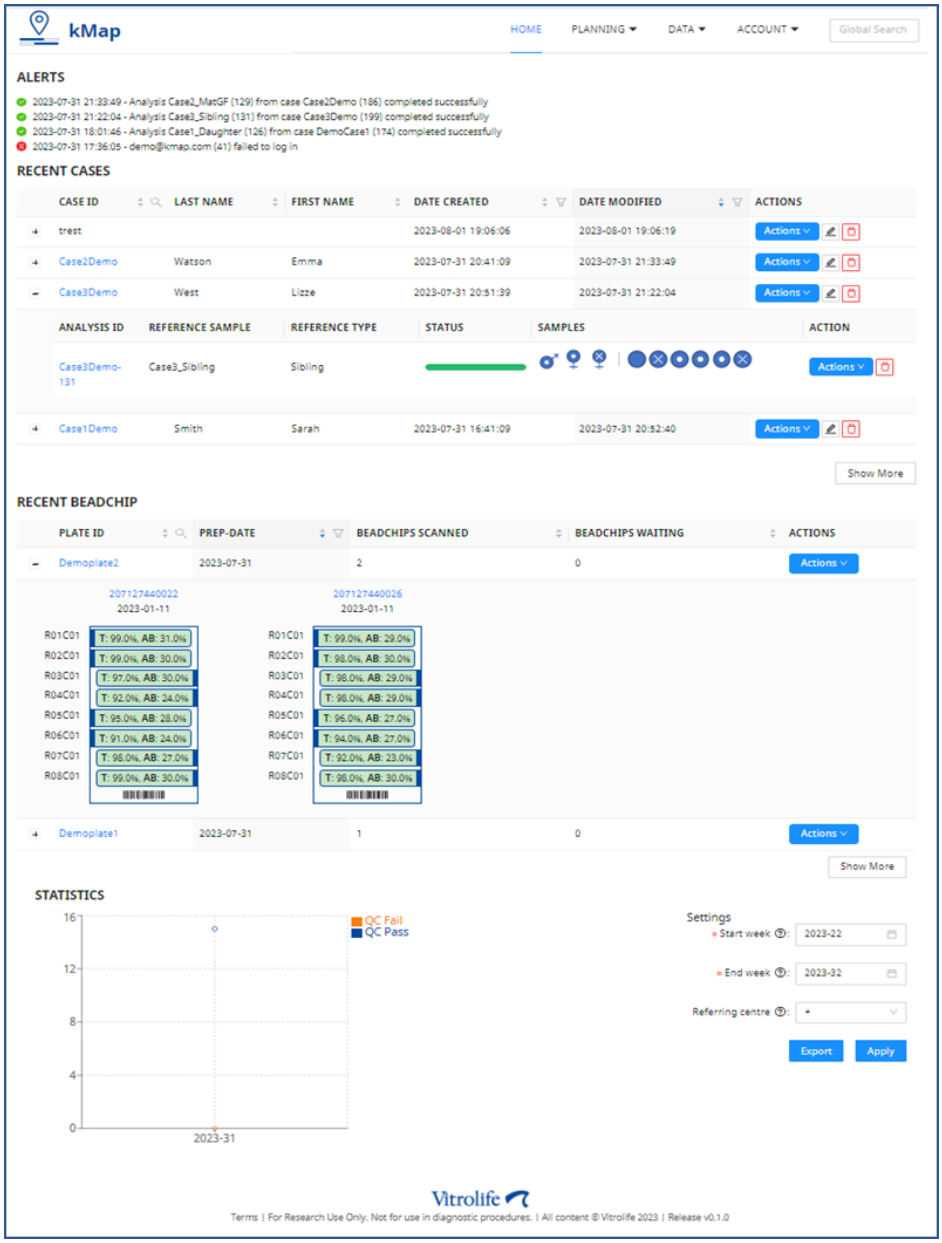


Figure 4: HOME dashboard view

Alerts

The *HOME* dashboard will display the five most recent *Alerts*.

Recent Cases

The *HOME* dashboard will display the five most recent *Cases*.

1. Click on Show More underneath the table to view the full lists of *Cases* or Select "Case List" from the DATA menu. This will take you to the "CASE LIST" page where twenty cases will be listed per page with the most recent cases in the top of the list.
2. Use the column header tools to search , sort and filter results.
3. Expand a specific *Case* by clicking on the + icon, to show analyses and status icons for all samples associated with that analysis and Case.
4. See section *4 Visualisation of Results* for case level results view, status updates, icons and Actions.

Recent BeadChip

The *HOME* dashboard will display the five most recent *Plate IDs*.

1. Click on Show More underneath the tables to view the full lists of *Plate IDs* or Select "Bead Arrays" from the DATA menu. This will take you to the "BEAD ARRAYS" page where twenty plates will be listed per page with the most recent plate in the top of the list.
2. Use the column header tools to search , sort and filter results.
3. Expand a specific Plate ID by clicking on the + icon, to show all the BeadChips and associated samples processed from that Plate.
4. Each BeadChip will display the eight subarray positions and the total Call Rate (T) and heterozygous Call Rate (AB) for each position. Hovering the mouse over a sub array will display the respective sample name.
5. See section *4 Visualisation of Results* Bead Array QC status, icons and Actions.

Statistics

A statistics chart will be presented to show the sample throughput by QC status per week.

1. The charts show data for samples that have been Signed-Off for reporting. See *Sample Sign-Off*.
2. Select a timeframe Start week and End week using the calendar tool. The week is displayed as <Year>-<Week Number>.
3. **Optional**. Select a Referring Centre to filter results. The list of Referring Centres is populated using entities made during Subject *Accessioning*.
4. Click Apply to save the settings and update the charts.
5. Click Export to save a tab-delimited .tsv file containing the data required to re-draw the charts in other applications.

2 PLANNING

Planning for a KaryoMap run is performed in two stages.

1. **Accessioning**: Add case and sample details to the kMap software.
2. **Laboratory Planner**: Assign BeadChips and sub-array positions for up to 96 samples, grouped by a unique Plate ID.

Accessioning

Enter Sample Data Manually

The following sections describe actions on the *Accessioning* page, under the kMap *PLANNING* menu. A full list of existing cases can also be viewed under *Case list* in the *DATA* menu; click on a Case row to navigate to the *Accessioning* page. For data fields, hover over the icons to see a description for each field.

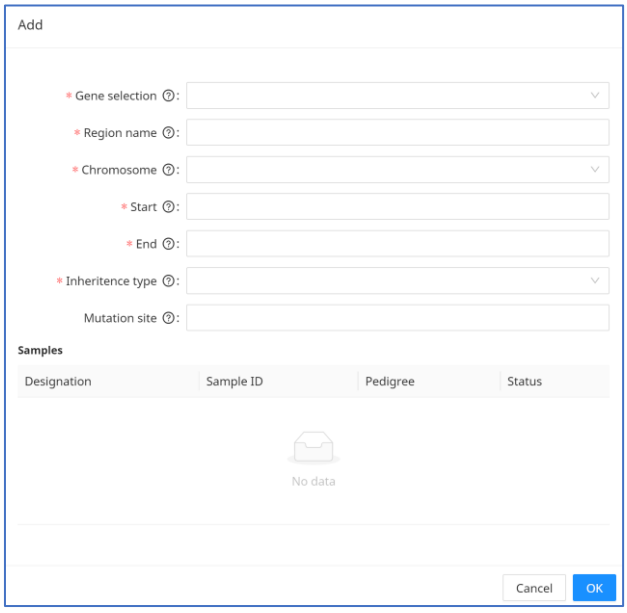
Add a New Case

Accessioning

1. Enter a unique identifier in the Case ID field.
2. **Optional**. Add Referring Centre, Referred by and Notes.
   Note: Data privacy obligations applicable to the account owner's organisation should be followed.
3. BeadChip Type is set to KaryoMap v2 by default.
4. Click Save at the bottom of the Accessioning section to add the new Case to the system.

Gene / Region

1. Under the Gene / Region section, click Add to add a gene region and a pop-window will open as below:

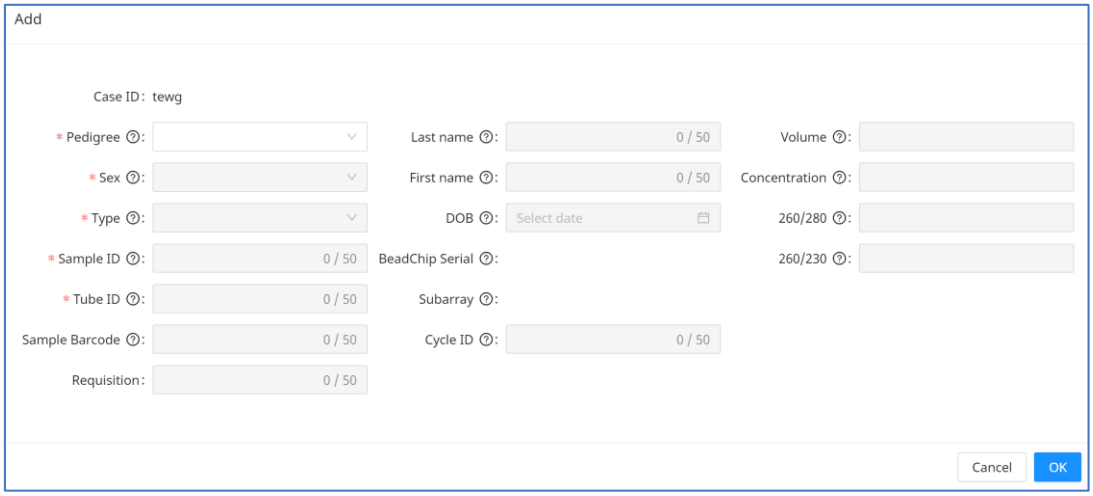


2. In the *Gene selection* field, type the HGNC gene nomenclature to select the region of interest from a list of commonly analysed genes.

3. The Region name, Chromosome, Start and End values will be automatically filled in by the kMap software using the HGNC gene information.
   Note*:* KaryoMap v2 uses the hg38 human reference database for genomic locations.
   Note: It is the responsibility of the user to ensure that the region start and end positions are suitable for the region of interest that is being analysed.
4. Alternatively, you can define your own specific region to be analysed by defining a new name and location or by modifying the pre-filled region data.
5. For *Inheritance type*, select one of the following from the drop-down menu (Autosomal recessive, Autosomal dominant, X linked recessive and X linked dominant).
6. **Optional**. Add mutation site information (genomic location of the mutation).
7. Click OK at the bottom of the pop-up window to add the *Gene / Region* to the Case.
8. Click Cancel at the bottom of pop-up window if you don't wish to add the *Gene / Region* to the Case.
9. The selected Gene / Region is listed in a table below the *Gene / Region* section and can be edited or deleted by clicking on the edit icon or the delete icon respectively.
10. If you do not wish to add a region, then you can select *No Region specified* by clicking on the check box below the *Gene / Region* header.

Samples

1. Under the samples section, click Add to add a sample and a pop-window will open as below:

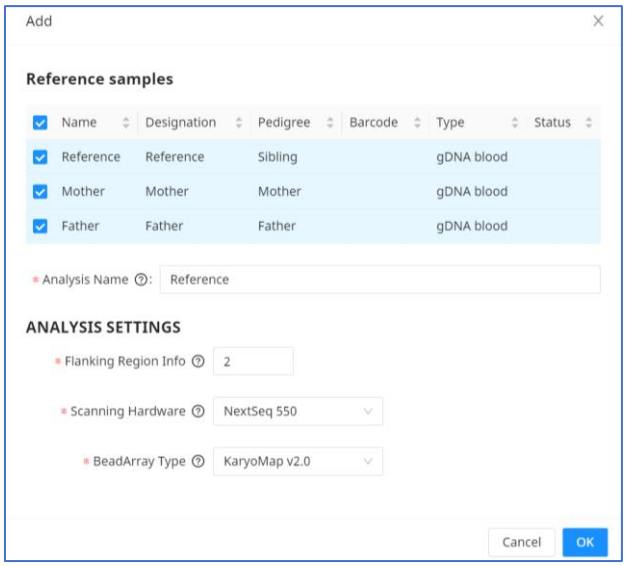


2. Enter the *Pedigree*, select one of the following from the drop-down menu (Father, Mother, Maternal Aunt, Maternal Grandfather, Maternal Grandmother, Maternal Uncle, Paternal Aunt, Paternal Grandfather, Paternal Grandmother, Paternal Uncle, Sample, Sample reference and Sibling)
3. The software will auto fill the relevant fields for the appropriate Pedigree selected.
4. For *Sex*, if not selected by the software, select one of the following from the drop-down menu (Female, Male and Unknown)
5. For sample *Type*, select one of the following from the drop-down menu (Blastomere, Genomic DNA, Other and Trophectoderm).
6. Enter the sample *Name* and *Tube ID* in the relevant fields.
7. **Optional**. Add *Sample Barcode*, *Requisition, Last name, First Name, DOB*, *Cycle ID*, *Volume*, *Concentration 260/280* and *260/230*)
8. In the Regions section, choose the Status of the sample from the Inheritance type drop-down list.
9. Click OK at the bottom of the pop-up window to add the new Sample to the Case.
10. Click Cancel at the bottom of pop-up window if you don't wish to add the new Sample to the Case.

11. All the samples in the case are listed in a table below the *Samples* section and can be duplicated by clicking on the duplicate icon , edited by clicking on the edit icon or deleted by clicking on the delete icon .

Analyses

1. Before result data can be added to a sample, this sample needs be part of an analysis.
2. Analyses can be set up manually by clicking Add in the Analyses section and a pop-window will appear with the all the trio samples (Father, Mother and Reference) in the case as below:



3. In the pop-up window, select the Father, Mother and the Reference sample required for the analysis.
4. The *Analysis Name* is auto populated with the name of the Reference sample.
   Note*:* The user can change the analysis name as required.
5. The *Flanking Region Info* by default is set to the value defined in the Group's settings, the users can use any value between 0.5 and 5 for this analysis.
6. The Scanning hardware by default is set to the value defined in the Group's settings, users can click on the drop-down menu to choose NextSeq 550 or iScan for this analysis.
7. The *BeadArray Type* is set to KaryoMap v2.
8. Click OK at the bottom of the pop-up window to add the new Analysis to the Case.
9. Click Cancel at the bottom of Add pop-up window if you don't wish to add the new Analysis to the Case.
10. All the Analyses in a case are listed in a table below the *Analyses* section. Analysis data can be reanalysed by clicking on the reanalyse icon or deleted by clicking on the delete icon .

Edit an Existing Case

On the kMap home page, under "Recent Cases" select the case you would like to edit or use the global search to find the case and choose "Accessioning" from the Actions tab. A full list of existing cases can also be viewed under *Case list* in the *DATA* menu; click on a Case row to navigate to the *Accessioning* page.

1. In the Accessioning page of the case, click Edit to update the Case ID, Referring Centre, Referred by, BeadChip Type and Notes.

Table 6: Sample Information Table values

Value	Description
Pedigree	**Required.** Pedigree is the genetic relationship of the DNA samples in relation to the test samples used in the Karyomapping case. Pedigree can be one of the following: Father, Mother, Maternal Aunt, Maternal Grandfather, maternal Grandmother, Maternal Uncle, Paternal Aunt, Paternal Grandfather, Paternal Grandmother, Paternal Uncle, Sample, Sample Reference and Sibling
Sex	**Required.** Sample gender.
Type	**Required.** Input must match one of the following values: Trophectoderm, Genomic DNA, Other.
Sample ID	**Required.** A unique identifier of the Sample, containing only alphanumeric characters or dashes [-]. Duplicate Sample IDs cannot be added to the same Plate during Laboratory Planning.
Tube ID	**Optional, recommended.** The short ID written on the sample tube. This value is displayed by default on the Laboratory Planner PDF to assist sample-to-BeadChip sub-array tracking during sample preparation.
Sample Barcode	**Optional.** A secondary unique identifier of the Sample. Barcodes must be unique within the kMap database.
Requisition	**Optional.** An identifier for the sample batch, such as a job requisition number or work order number.
Last Name	**Optional.** Last name of the Father, Mother or Reference sample. *Please observe applicable data protection regulations.*
First Name	**Optional.** First name of the Father, Mother or Reference sample. *Please observe applicable data protection regulations.*
DOB	**Optional.** Date of birth of the Father, Mother or Reference sample. *Please observe applicable data protection regulations.*
BeadChip Serial	**Optional.** Serial number of the BeadChip on which the sample was processed. **Note**: this field can only be auto populated while uploading a sample via Fast Track option.
Subarray	**Optional.** BeadChip Subarray ID on which the sample was processed. **Note**: this field can only be auto populated while uploading a sample via Fast Track option.
Cycle ID	**Optional.** Identifier for the IVF Cycle from which the test samples were prepared.
Volume	**Optional.** Volume of the sample processed on the KaryoMap v2 BeadChip
Concentration	**Optional.** DNA concentration of the sample processed on the KaryoMap v2 BeadChip
260/280	**Optional.** 260/280 ratio of the samples.
260/230	**Optional.** 260/230 ratio of the samples.

Import Data Using an Accessioning File

Case Accessioning

Case data including Case ID, Gene/Region and Sample information can alternatively be entered into the system by importing from a predefined tab-delimited .txt file. The import function is convenient for data already stored in a LIMS or similar system. See *Appendix 2* for rules concerning valid characters and required fields.

1. Use the template file *kMap_CaseAccessioning_Template.txt.*
2. [Header] section

a. Enter Date (in the format YYYY-MM-DD) and Case ID information in the [Header] section.
b. Optional: enter the details of the Parents including, Mother First Name, Mother Last Name, Mother DOB, Father First Name, Father Last Name, Father DOB.
c. Optional: Enter details of Consultant and Referring Centre.
d. Optional: Add any Notes
e. Array Type should be KaryoMap v2

3. [Gene Region] section
    a. Enter Region Name (OMIM nomenclature), Chromosome, Start, End, Inheritance Type, Mother Status, Father status, and Reference status. Mutation Site is optional.
    Note: the status of Trio samples should be logical otherwise import will not work.
4. [Trio] Section
    a. Enter information about Father, Mother and Reference. Pedigree, Sex, Type, Sample_ID and Tube_ID are mandatory.
    b. Optional: Sample_Barcode, Requisition, Volume_uL, Concentration_nguL, 260/280 and 230/280.
5. [Data] section.
    a. Enter sample information Type, Sample_ID and Tube_ID are mandatory.
    b. Optional: Sample_Barcode, Requisition, Volume_uL, Concentration_nguL, 260/280 and 230/280.
6. Once all the case and sample details are added, Save the *kMap_CaseAccessioning.txt* file on to your computer.
7. Click Import on the *Accessioning* page and browse to the import file.

Import Summary

1. Imported data is validated against existing Case records. If no matches are found, a new Case ID will be created using the information provided (
2. Figure **5**).
3. Click OK to confirm the import and add data to kMap. Click Cancel to stop the import and exit without adding data to kMap.
4. When using the import option “Analysis” will be automatically created by the kMap software.
Note: where there is more than one Father, Mother or Reference, the kMap software will use the first uploaded samples to create the Analysis.

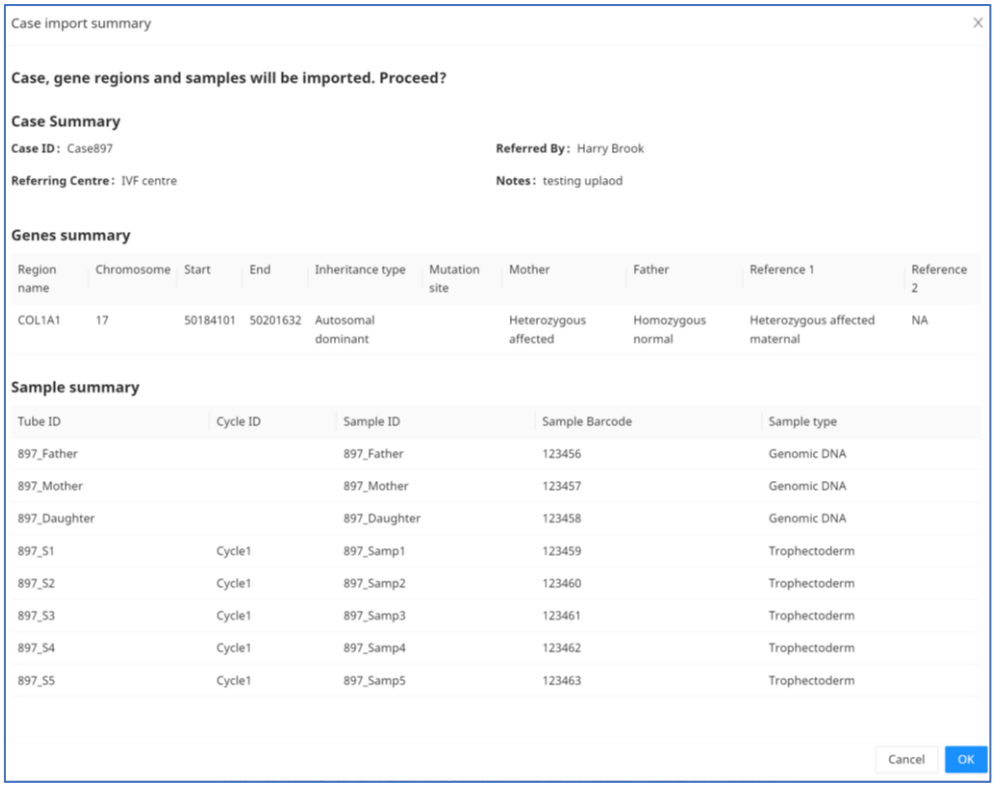


Figure 5: Case Accessioning import dialogue

Sample Accessioning

Sample data for a pre-existing case can alternatively be entered into the system by importing from a predefined tab-delimited .txt file. The import function is convenient for data already stored in a LIMS or similar system. See *Appendix 2* for rules concerning valid characters and required fields.

1. Use the template file *kMap_SampleAccessioning_Template.txt.*
2. Enter Date (in the format YYYY-MM-DD) and Case ID information in the [Header] section.
3. Array Type should be KaryoMap v2.
4. Enter Sample information in the [Data] section. Tube_ID, Sample_ID and Sample_Type is mandatory.
5. Once all the sample details are added, Save the *kMap_CaseAccessioning.txt* file on to your computer.
6. Click Import on the *Accessioning* page and browse to the import file.

Import Summary

1. Imported data is validated against existing Case records. (
2. Figure **5**).
3. Click OK to confirm the import and add data to kMap. Click Cancel to stop the import and exit without adding data to kMap. (Figure 6)

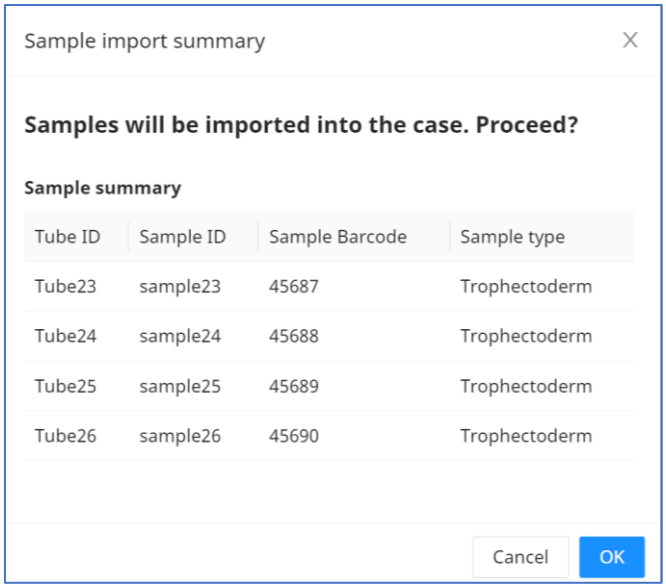


Figure 6: Sample Accessioning import dialogue

Laboratory Planner

To plan a Sample Preparation batch, accessioned samples must be assigned to a Plate ID using the Laboratory Planner tools. The assigned plate well corresponds to the respective sub-array position on a KaryoMap v2 BeadChip. For example, the sample added to well A1, shall be configured to be processed on the R01C01 of the first BeadChip. The output is a Laboratory Planner PDF which can be used for sample-to-BeadChip position (Sub-array) tracking during the KaryoMap v2 assay workflow.

Select *Laboratory Planner* under the *PLANNING* menu to get started.

The *Available Plates* section displays a list of saved Plates in the system with the Plate ID, Preparation Date, number of BeadChips processed and the User who created the Plate (Figure 7).

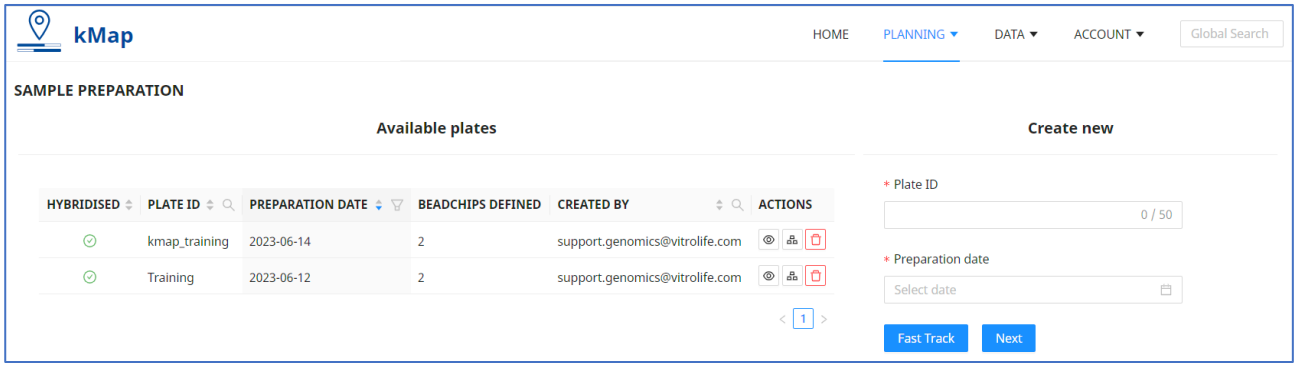


Figure 7: Laboratory Planning - Create New Plate

Create a New Plate - Plate Builder

1. Under *Create New,* enter a unique Plate ID and the Preparation Date as shown in Figure 7. The Plate ID must contain alphanumeric characters or dashes [-] only.
2. Click NEXT to proceed.

On the Plate Definition page, under the section of Available Samples the data is arranged by Case ID, and within a case the trio samples (father, mother and reference) are listed directly, while the test samples are listed under a separate list named after the Cycle ID. The list displays samples from accessioned Case-Samples that have not yet been assigned to a Plate (Figure 8). Clicking on a Case ID will expand the view to display the associated trio samples and the Cycles, each with a list of Test Samples. The most recently accessioned Cases are listed first. Alternatively, the search bar can be used to locate a specific Case. If no samples are 'available' for a Case, then the Case-Sample entry will no longer appear in the list.

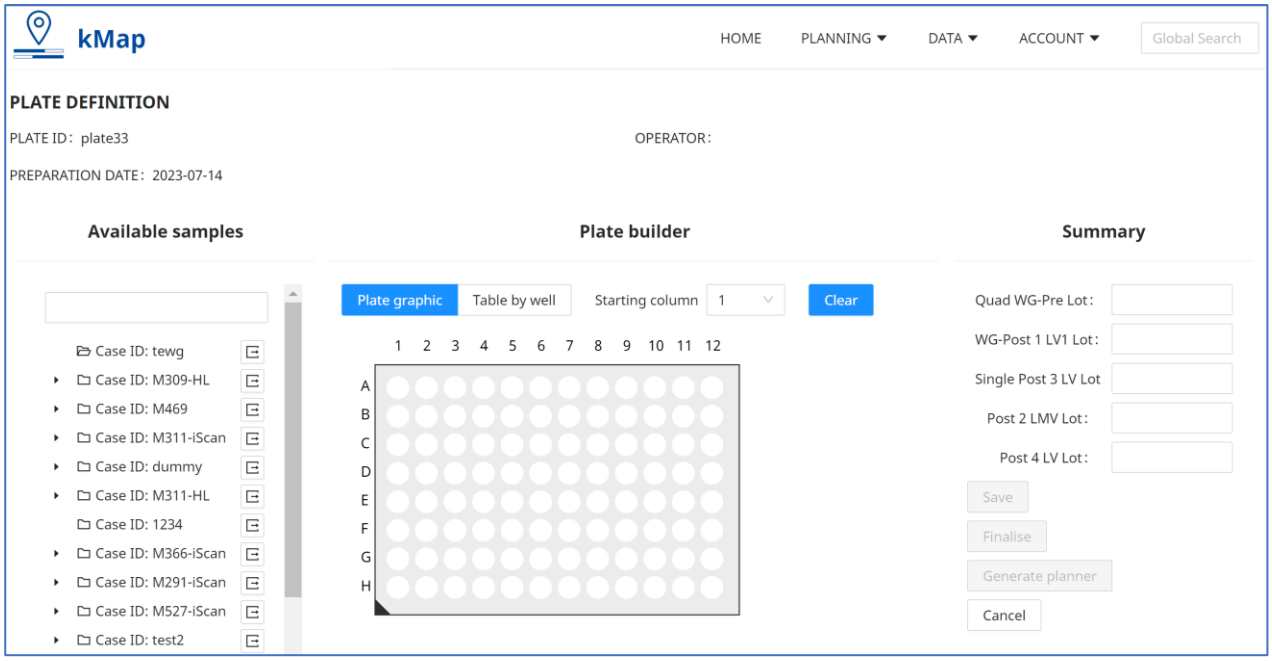
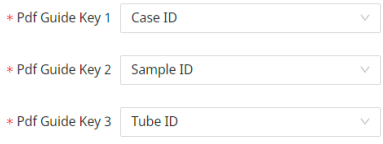
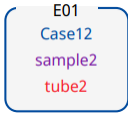


Figure 8: Plate Builder

3. Add each sample from the list of *Available Samples* to an empty well of the plate graphic in the *Plate Builder* section. A minimum of 8 and a maximum of 96 samples can be added to each Plate. Assigned wells are shown with solid fill; hover the cursor over the well to reveal the Sample information.
    a. Individual samples can be added to specific wells using the drag-and-drop feature with the left-mouse button.
    b. The Case autofill button ⇥ will add all samples of the Case to the next available well positions in the Plate.
4. Use the Starting Column selector to configure the Autofill ⇥ feature to start from column 1 to 12 of the plate as required.
5. Click Clear plate to remove all sample assignments from the Plate.
6. Use the view selector Plate graphic | Table by well to view additional sample accessioning information including, Case ID, Sample ID, Sample Barcode, Requisition, Tube ID and Well Position.
7. When the required samples have been added to the plate, enter the BeadChip Serial number and the KaryoMap v2 BeadChip kit box Lot numbers.
8. Select SAVE to save the plate well allocation for sample preparation.
   Note: after saving you can still edit the plate and add more samples.
9. Select Finalise to confirm the plate well allocation for sample preparation.
   Note: after finalising, the plate will be locked, and you will not be able to make any changes to the plate.
10. Select Generate planner to produce a Laboratory Planner PDF for sample-to-BeadArray position tracking during the Hybridisation step in the KaryoMap v2 protocol.
    In the wells of the plate graphic of the PDF guide, three rows of IDs can be displayed. Choose the types of IDs most appropriate for your laboratory on the *Settings* page.



Example Key selection defined in Settings



Example Laboratory Planner PDF Plate Graphic well

Modify an Existing Plate

1. The Laboratory Planner PDF can be accessed by clicking the button under *Actions,* for *Available Plates* (Figure 7).
2. Click on an available plate to return to the Plate Builder and review planning information.
3. To delete a plate, click the button.
   A dialogue box will be displayed with a checkbox option to also delete all samples associated with the Plate.

Upload Case Data - FastTrack

The Fast Track import function allows test sample *Accessioning* and *Laboratory Planning* steps to be performed using a **single import file**. This method is well suited to high-throughput Users or where data is already stored in a LIMS or similar system. It may also be used as a tool to instruct kMap if a different system is configured to perform Laboratory Planning. With the Fast track option, only one case can be uploaded at a time, so each case will need a separate FastTrack import file. See *Appendix 2* for rules concerning valid characters and required fields.

1. Use the template file *kMap_FastTrack_Template.txt*
2. [Header] section
   a. Enter Date (in the format YYYY-MM-DD) and Case ID information in the [Header] section.
   b. Optional: enter the details of the Parents including, Mother First Name, Mother Last Name, Mother DOB, Father First Name, Father Last Name, Father DOB.
   c. Optional: Enter details of Consultant and Referring Centre.
   d. Optional: Add any Notes
   e. Array Type should be KaryoMap v2
3. [Gene Region] section
   a. Enter Region Name (OMIM nomenclature), Chromosome, Start, End, Inheritance Type, Mother Status, Father status, and Reference status. Mutation Site is optional.
   Note: The status of Trio samples should be logical otherwise import will not work.
4. [Trio] Section
   a. Enter information about Father, Mother and Reference. Pedigree, Sex, Type, Sample_ID and Tube_ID are mandatory.
   b. Enter experiment information including Plate_ID, Preparation_Date, Well_Position, BeadChip_Serial, Subarray.
   c. Optional: Sample_Barcode, Requisition, Volume_uL, Concentration_nguL, 260/280, 230/280, reagent Lot information for Quad_WG-Pre_Lot, WG_Post_1_LV1_Lot, Single_Post_3_LV_Lot, Post_2_LMV_Lot and Post_4_LV_Lot.
5. [Data] section.
   a. Enter sample information including Type, Sample_ID and Tube_ID are mandatory.
   b. Enter experiment information including Plate_ID, Preparation_Date, Well_Position, BeadChip_Serial, Subarray.
   c. Optional: Sample_Barcode, Requisition, Volume_uL, Concentration_nguL, 260/280, 230/280, reagent Lot information for Quad_WG-Pre_Lot, WG_Post_1_LV1_Lot, Single_Post_3_LV_Lot, Post_2_LMV_Lot and Post_4_LV_Lot.
6. Once all the case and sample details are added, Save the *kMap_FastTrack.txt* file on to your computer.
   Note: Ensure all date fields are stored as text with the format YYYY-MM-DD. Opening a .txt file in Microsoft Excel may automatically update the date fields to an unsupported format to e.g. DD/MM/YYYY.
7. Click Fast Track on the *Plate Builder* page and browse to the import file.

8. Imported data is validated against existing Case and sample records. A summary table is shown confirming the cases and samples (Figure 9).

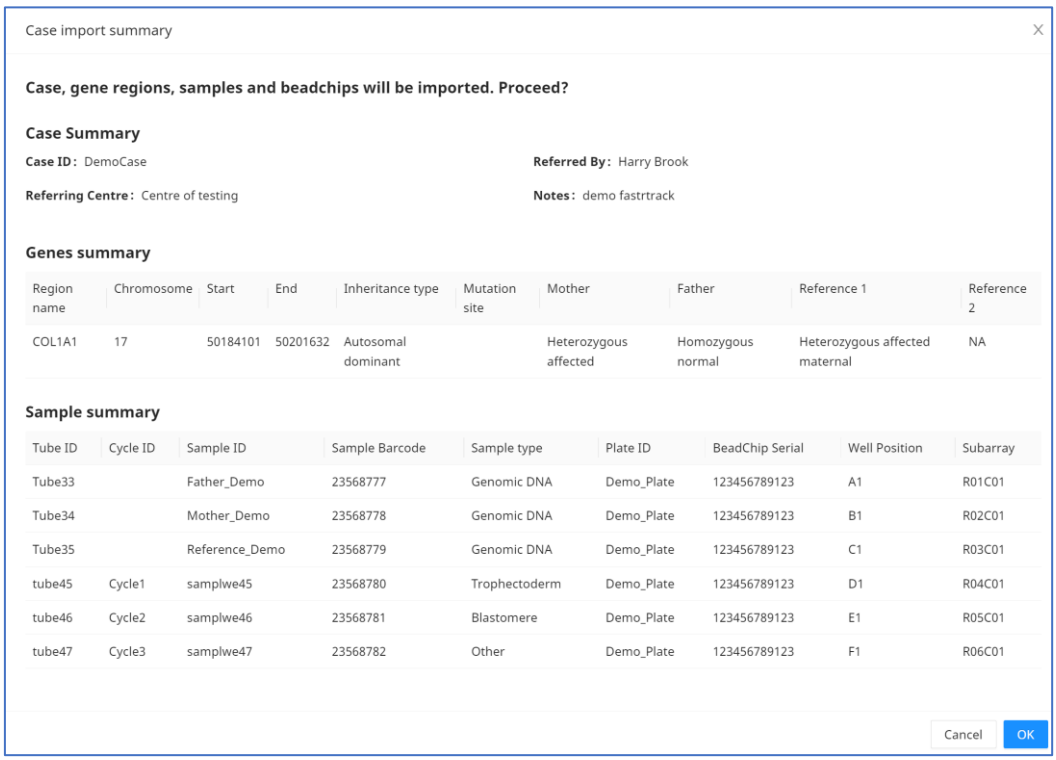


Figure 9: Fast Track import summary

9. Click OK to confirm the import and add data to kMap. Click Cancel to stop the import and exit without adding data to kMap.
10. The kMap software will create a case, plate and record all the associated data as uploaded.

3 UPLOADING DATA

After processing the samples on the KaryoMap v2 BeadChips, the BeadChips are scanned on an iScan or a NextSeq 550 instrument. The scan folders contain the results files in the format "*BeadChipSerial_SubarrayID.gtc*"
The results files (.gtc) required for data analysis should be manually uploaded to the Group's kMap account.

Upload of Scan Data

To upload the results files (.gtc) to the kMap software please do the following.

1. Open the *kMap-Uploader.exe* software and use the Browse button select up to four scan folder locations to be uploaded to the kMap software for analysis (Figure 10).
2. Once the scan folders are selected, type in your kMap account password in the field "kMap Passwd:" **Note**: Your computer can be set up to store the kMap-Uploader password to avoid having to type in the password every time. See Appendix 4 for more details.
3. Press Upload and monitor the *Log* area for success or failure messages. A *kmap-upload.log* file will also be saved in the same location as the kMap-upload.exe software.
4. Further settings are defined in the *Advanced Configurations* section, which can be expanded by clicking ▲. Note that the absolute paths will require updating if not using system defaults. Details of each setting are described in
5. **Table 7**.

Table 7: kMap-Uploader.exe Advanced Configurations

Action	Description
kMap Configuration	Absolute path to the JSON file with connection credentials.
kMap-Uploader SW	Absolute path to the kMap-upload-cmd.exe software.
Overwrite	Force the upload of already uploaded files.

6. During the upload, it is normal for the window to be unresponsive and may show a spinning wheel or "Not Responding" status. The speed of the upload is dependent on the internet connection; upload of data from a single KaryoMap v2 BeadChip should be completed in < 5 minutes. The *Log* window will populate after the upload attempt has completed.
7. Do not attempt to upload data into your Group's account simultaneously from the same or a different computer.
8. Click Quit to close the application.
   Any changes to the *Advanced Configuration* settings will be saved to kM*ap_upload.cache* in the same location, and restored upon next use.
   Clicking the Windows close button ✕ in the top right-hand corner will not save changes and settings will revert to the default values upon next use.

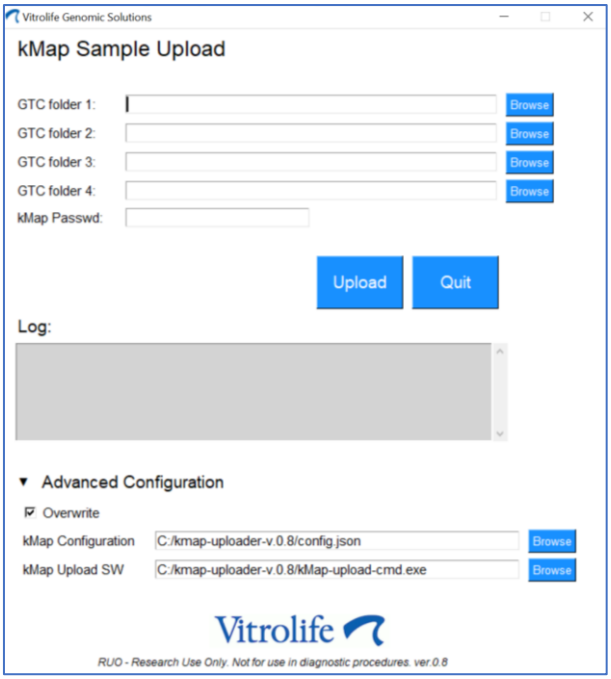


Figure 10: kMap-Uploader.exe User Interface

4 ANALYSIS

Results Overview

Data analysis by the kMap software is automatic and the software will display case level data and sample level data. The kMap software will also generate QC data for each sample. The KaryoMap v2 results can be reviewed by:

1. **Case List**: In the case list each case is listed by the case ID and is sub-divided into **Analyses** according to the trio datasets used to perform phasing (Maternal, Paternal and Reference sample data). The user will be able to access the case level data from the case list.
2. **Plate List**: In the plate list each plate is listed by the Plate ID and is sub-divided into individual **BeadChip Array** by serial number. The user will be able to access the sample level QC data from the plate list.

Case List

Each Case has an Actions menu, an edit button and a delete button (**Figure 11**). The Actions menu gives access to experiment-specific details. Actions are listed in Table 8.

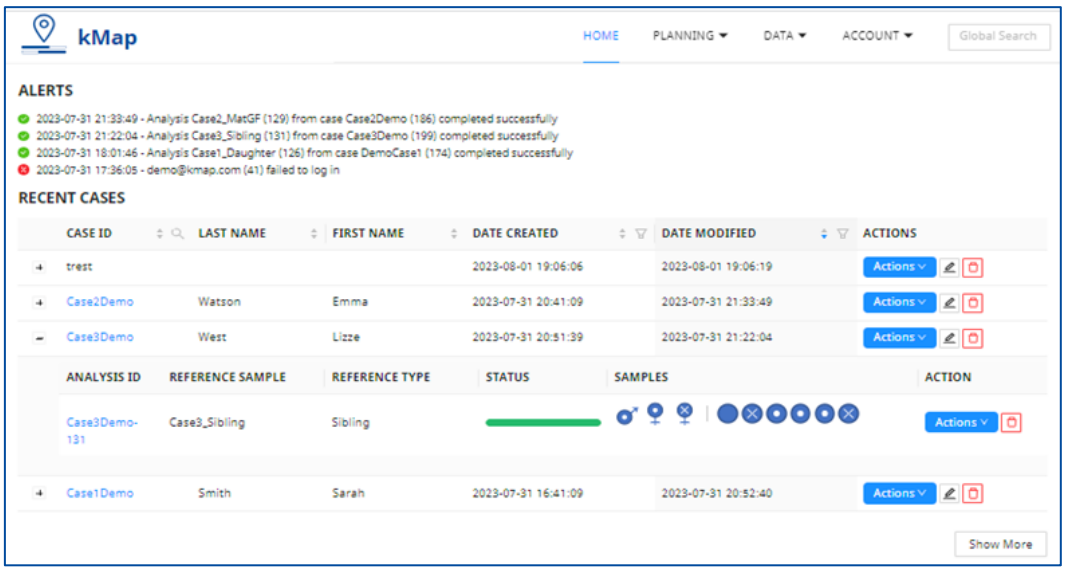


Figure 11: Recent Cases list with expanded Analyses list.

Table 8: Case actions menu

Action	Description
Accessioning	Open the *Accessioning* page, to view or edit details for this Case.
Planner	Open the *Plate Definition* page, showing the sample preparation plate well, BeadChip Array position assignment, and reagent lot information for this Plate ID.
Files	Open page listing all data files available to download for this experiment.
View	Open the Analysis page to view result data for this Case.

1. Click to view and edit the Accessioning details for this Case.
2. Click the icon next to a Case ID to reveal Analyses performed for the selected Case.
    a. Each Analysis also contains an Actions menu with *View* and *Files* options to navigate to the result page or the download page for the case data.
    b. Click the Analysis ID to view the result data for this Analysis.
    c. Click on an individual sample icon to navigate directly to the *Detailed Sample View* for this sample.
    d. Hover the cursor over a sample icon to reveal the Sample ID and most severe inheritance status (see **Error! Reference source not found.**).
    e. Click to delete result data relating to this specific Analysis.
3. Click delete and confirm at the Case ID level to delete all associated sample and result data from the system.

Case Status

Once the scan data (GTC files) is uploaded to the kMap software, the following steps are performed:

1. Upon successful upload, the Karyomapping analysis is automatically initiated using the parameters defined in the *Settings of this analysis.*
2. The sample data is automatically linked to the *Run Definition* that was created in kMap software during the Laboratory Planning steps.
3. The run status will be displayed and updated accordingly (as in Table 9).

Table 9**:** Case status types.

Case status	Identifier	Description
Analysis waiting		The case has been set up and kMap is waiting for the scan data to be uploaded for analysis.
Trio samples are being processed for this analysis.		Trio samples are being processed for this analysis. Data for test samples are not available.
Analysis completed for trio samples		Analysis completed for trio samples. Data for test samples are not available.
Samples are being processed for this analysis.		Analysis completed for trio samples. Data for test samples are being processed for this analysis.
Analysis completed		Analysis completed for all samples (trio and test samples) and data is ready for review.
Analysis failed trio analysis		Analysis failed trio analysis. One or more trio samples did not pass the QC criteria for analysis.
Analysis failed sample(s) analyses		Analysis completed for trio samples, but analysis for one or more test sample has failed.

Sample Icons

When viewing data under Case ID > Analysis ID, the icons representing the samples highlight the outcome of the data analysis. Sample icons are described in **Error! Reference source not found.**.

1. Hover the cursor over the sample icon to display the Sample ID and a written inheritance status.
2. Click on the sample icon to open the *Detailed Sample View* page showing the KaryoMap, Detailed Haploblock chart, LogR chart and B-allele chart for the Sample.

Table 10: kMap sample status icons

Icon	Status	Description
	Waiting (Paternal)	Data for the Trio sample has not yet been received.
	Waiting (Maternal)	Data for the Trio sample has not yet been received.
	Waiting (Reference)	Data for the Trio sample has not yet been received.
	Waiting (Test Sample)	Data for the sample has not yet been received.
	Unaffected (Paternal)	The paternal sample has been accessioned with “unaffected” status, or no gene/region has been specified.
	Carrier (Paternal)	The paternal sample has been accessioned with “carrier” status, for one or more genes/regions. See rules for order of severity.
	Affected (Paternal)	The paternal sample has been accessioned with “affected” status, for one or more genes/regions. See rules for order of severity.
	Unaffected (Maternal)	The maternal sample has been accessioned with “unaffected” status, or no gene/region has been specified.
	Carrier (Maternal)	The maternal sample has been accessioned with “carrier” status, for one or more genes/regions. See rules for order of severity.
	Affected (Maternal)	The maternal sample has been accessioned with “affected” status, for one or more genes/regions. See rules for order of severity.
	Unaffected (Reference or Sample)	**Reference sample**: Accessioned with “unaffected” status, or no gene/region has been specified. Test sample: The phasing result is “unaffected”.
	Carrier (Reference or Sample)	**Reference sample**: Accessioned with “carrier” status, for one or more gene/regions. See rules for order of severity. Test sample: The phasing result is “carrier” for one or more genes/regions. See rules for order of severity.
	Affected (Reference or Sample)	**Reference sample**: Accessioned with “affected” status, for one or more gene/regions. See rules for order of severity. Test sample: The phasing result is “affected” for one or more genes/regions. See rules for order of severity.
	Unknown status	**Trio sample**: Accessioned with “unknown” status, for one or more genes/regions. Test sample: The phase could not be predicted for one or more genes/regions. See Case analysis warnings for more information.
	Analysis failure	The sample could not be analysed, check analysis logs for more information.
		**Test sample**: At least one of the QC criteria was not met. The phase could not be predicted for one or more genes/regions.

Icon order of severity

Each sample icon

For example, a sample that is a carrier for disease region 1, and affected for disease region 2, shall be displayed with the “Affected” icon and written status.

Plate List

Each Plate has an Actions menu that gives access to experiment-specific details (Figure 12). Actions are listed in

Table 11.

Action	Description
Planner	Open the *Plate Definition* page, showing the sample preparation plate well, BeadChip Array position assignment, and reagent lot information for this Plate ID.
Files	Open page listing all data files available to download for this experiment.
QC	Display the Infinium control probe data for BeadChips in this experiment.

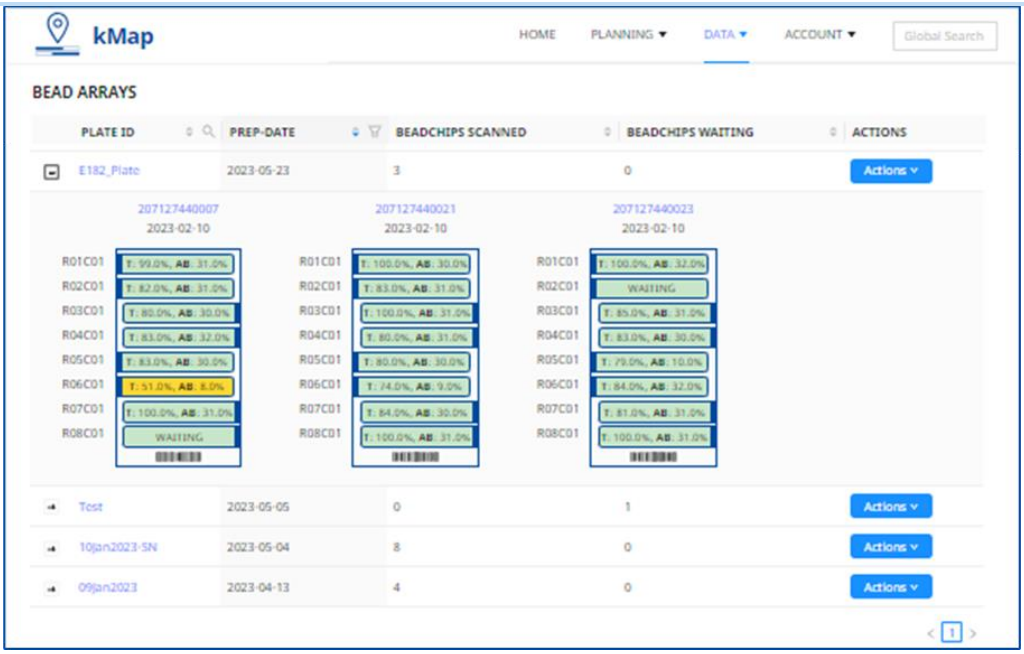


Figure 12: Recent Plates list with expanded BeadChip Array list and action menu.

Action	Description

Planner	Open the *Plate Definition* page, showing the sample preparation plate well, BeadChip Array position assignment, and reagent lot information for this Plate ID.
Files	Open page listing all data files available to download for this experiment.
QC	Display the Infinium control probe data for BeadChips in this experiment.

Table 11: Plate actions menu

1. Click the + icon next to a Plate ID to reveal BeadChip Array information for this experiment.
2. The BeadChip diagram displays the sample layout, with total (T) and heterozygous (AB) Call Rate percentages.
3. The sub-array fill colour signifies the total Call Rate QC status, where:
    a. Green = PASS (≥80% for bulk genomic DNA; ≥60% for amplified DNA)
    b. Amber = FAIL (<80% for bulk genomic DNA; <60% for amplified DNA)
4. Hover the cursor over a sample sub-array to reveal the Sample ID.
5. The BeadChip serial number and scan date are displayed above each BeadChip diagram.

BeadChip Array QC

All Infinium BeadChips are equipped with a set of internal control probes that are designed to support quality control of the assay's stringent performance criteria, and to demonstrate its robustness. The control probe data is presented in a format similar as in the *GenomeStudio* software application (Illumina Inc).

For guidance on interpreting control probe data, please refer to the *Evaluation of Infinium Genotyping Assay Controls Training Guide*, available from the Illumina website (www.illumina.com) or contact your Vitrolife Support representative for further details.

The Infinium Control Probe data can be assessed on the *BeadChip Array QC Metrics* page (Figure 13). Click *QC* under the Actions menu on any Plate or BeadChip Array list to access the *BeadChip Array QC Metrics* page.
QC data can be viewed by Case ID, Plate ID, or BeadChip Serial, additional samples can be included by selection more the one of these categories.

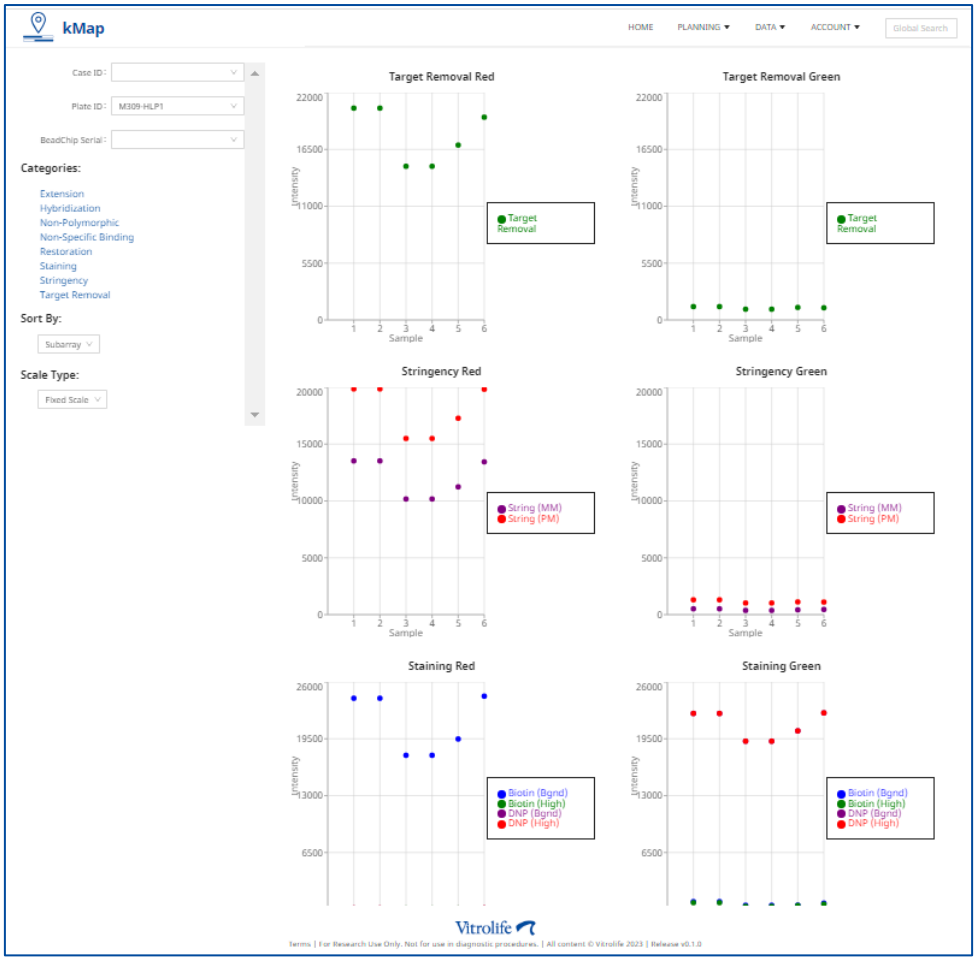


Figure 13: BeadChip Array QC metrics page

Visualisation of Results

The following sections describe features of the *Case Result* pages, containing the results of phased, linkage-based analysis for each Analysis (trio) included in the Case. The Case Result pages can be reached by clicking on any Case ID, Analysis ID, sample result icon or “Actions > View” menu from the list of recent cases (Figure 14).

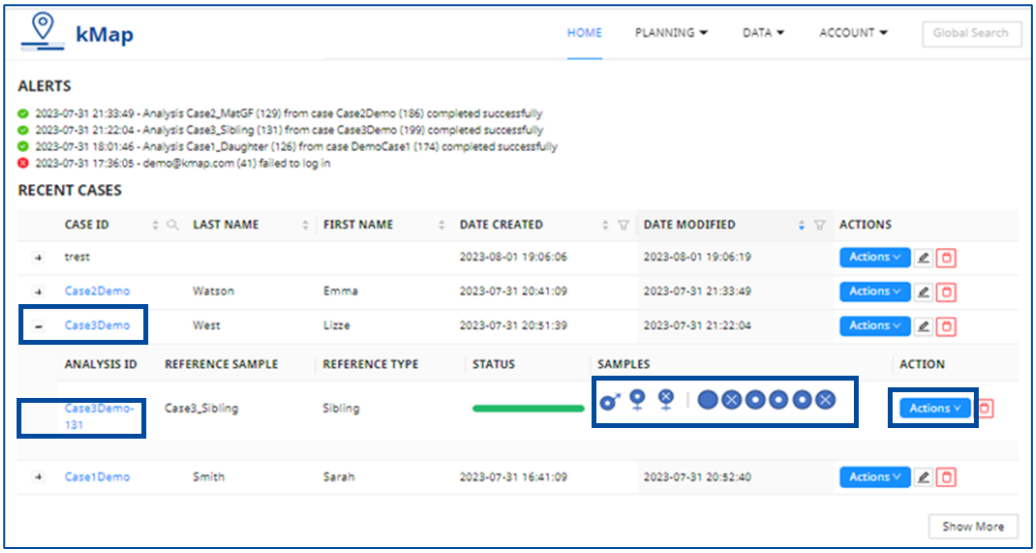


Figure 14: Recent Cases list with expanded Analyses list.

Results by Analysis

Each unique combination of trio samples (Maternal, Paternal and Reference DNAs) defined at Case Accessioning constitutes a separate Analysis. The trio data is used to identify a unique set of informative markers that are used for phasing the test samples. For example, analysing all test samples within a Case against two separate References requires two independent analyses. The results relating to a specific Analysis can be selected by clicking on the Analysis ID at the top of the *Case Result* page (Figure 15).

Case level data for each analysis can be visualised in separate tabs under the following categories (Figure 15):

1. **Case View** - Visualisation of the haplotype representation for all the samples in the case.
2. **Sample Detail View** - Detailed results for each sample (charts, QC data).
3. **Report: Summary** - Case level summary report.
4. **Report: Samples** - Sample level summary report.
5. **Log** - Detailed log of the steps in the data analysis with timestamp.

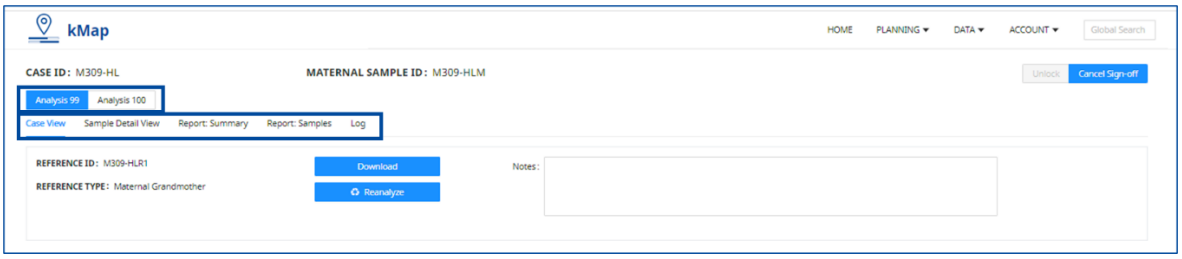


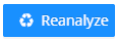
Figure 15: Cases with multiple Analyses shall display tabs by Analysis ID, to view results per analysis Trio.

Case View

The *Case View* page will display the haploblock charts for all the samples in the case along with some QC metrics. The Case View page contains the following sections:

Case information

1. Case ID and the maternal sample ID will be displayed at the top of the page.
2. The *Case View* page will have the option to *Lock* and *Sign off* a case. A case should first be locked and then only it will be available for *Sign off*.
3. The analysis for which the results are displayed will be highlighted in blue colour.
4. The Reference ID and the Reference type will be listed along with a window to add any notes.
5. Case results files can be downloaded by going to the download page by clicking 

.
6. The Analysis can be re-analysed by clicking 

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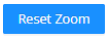
Warnings

The *Case View* page will list all Warnings related to the case.

Haplotype Navigation

1. Using the “SNP Details” drop-down menu the display of Key and Non-key SNPs can be switched off or switched on. **Note**: the default setting is to display the Key and Non-Key SNPs.
2. Using the “Two Region View” drop-down menu the software can be set up to display up to two regions (side by side). If a Karyomapping case is set-up for two or more regions, the first two regions will be displayed side by side.
    - To navigate to the haploblocks for any region within the genome, choose a Chromosome, Band, Position (as per the hg38 coordinates) or the Region name (HGNC nomenclature).
3. The default haplotype display is zoomed to the gene and both flanking regions. Haplotype display can be zoomed in by clicking the  icon and zoomed out by clicking the  icon.
    - The haplotype display can be reset to the full chromosome view by clicking 

and to the default gene by clicking on  while ensuring that the correct region is still selected
4. The haplotype display can also be navigated by using the arrow icons  and  .

Haplotype display

1. The case view page displays the haplotype chart for the Parental samples (Father and Mother) and the Reference sample in a separate drop-down menu. Below that, the haplotype charts for all the samples in the order of accessioning will be displayed in a separate drop-down menu (Figure 16).
2. For each sample the QC data (Call Rate, AA, BB, AB, ADO, Mis-call, X-Het Rate and Y-Call Rate) will be displayed alongside the haplotype charts.
3. By clicking on the “Sample ID” highlighted in blue colour, the user can navigate to the *Sample Detail View* page.

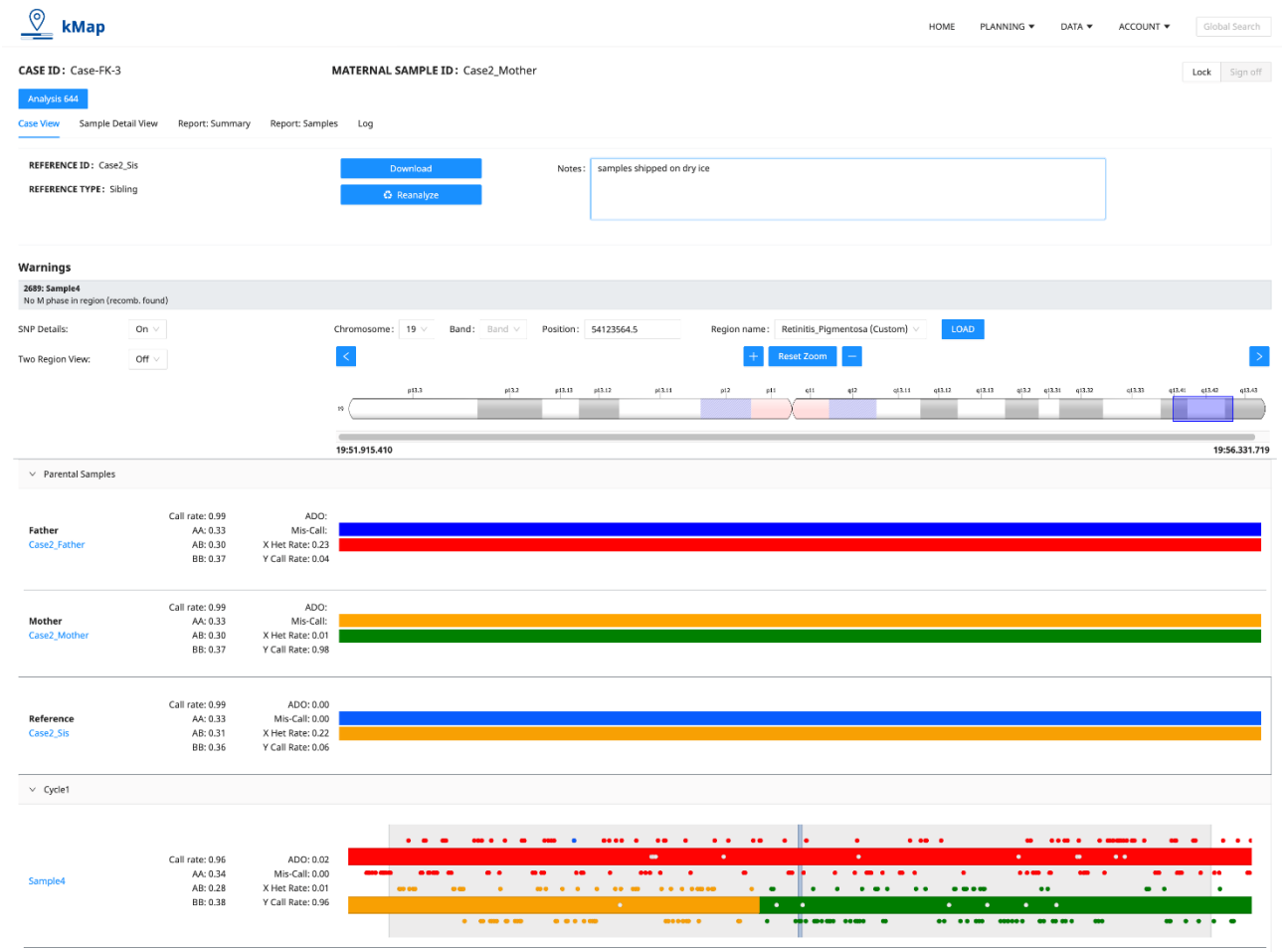


Figure 16: Overview of the Case View page.

Sample Detail View

1. The *Sample Detail View* page lists the sample ID and the Sample Name at the top of the page along with a table including the Call Rate, AA, AB, BB, ADO, Mis-Call, X-Het Rate, Y Call Rate, Known/Predicted Status and Overall Result (Figure 17).

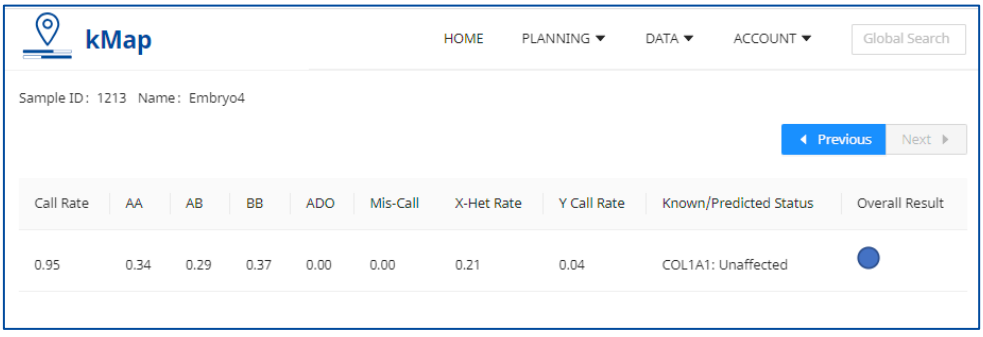


Figure 17: Sample results table – Sample Detail View page.

2. The Previous Next icons can be used to navigate through all the samples in a case.

3. The *Sample Detail View* page displays the KaryoMap chart, Haploblock Reference, Detailed Haploblock Chart, LogR Chart and B-allele chart for a sample. All the charts can be expanded or minimised by clicking the drop-down option next to their name (Figure 18).
4. The Detailed Haploblock Chart by default will be zoomed to the region of interest and flanking regions.
    - Reload the page after zooming or navigating in order to get back to the default view, or click Reset Zoom to load a full-chromosome view.
5. The Detailed Haploblock, LogR and B-allele charts can be accessed for each chromosome by clicking on the respective Chromosome number displayed above the KaryoMap chart.
6. To display the LogR and B-allele charts for the whole genome, click on the Full Genome icon.
   Note: When this option is chosen, Detailed Haploblock Chart will not be displayed.

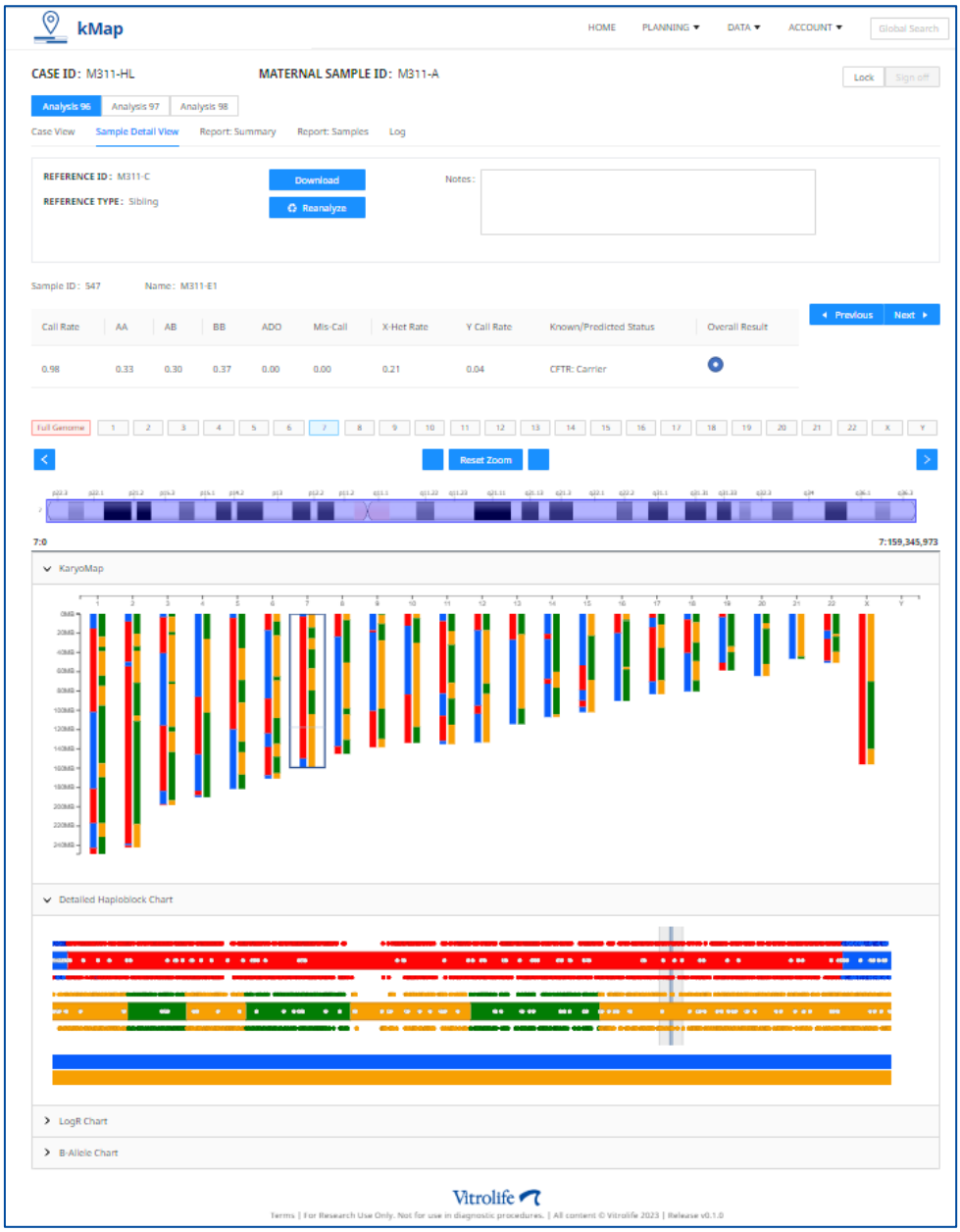


Figure 18: Sample Charts – Sample Detail View page.

Report: Summary

The *Report: Summary* page displays the "Analysis Report" for all the test samples in the case (Figure 19 and Figure 20). The Report: Summary contains the following sections:

Warnings

This section lists all the warnings associated with all the samples in the case.

Case Summary

1. The case summary displays the Case ID, Analysis ID, Referred By and Referring Center information.
2. Sample ID, First name and Last name of the mother is displayed in the table **Maternal Details**.
3. Sample ID, First name and Last name of the father is displayed in the table **Paternal Details**.
4. Sample ID, Pedigree and Sex of the Reference sample is displayed in the table **Reference Details.**
5. **Results for each sample including the** Sample ID, Case ID, Sample barcode, BeadChip Serial, Subarray, Sample Type, predicted status, QC and Result is displayed in the table **Sample Details**.
6. **Sign-off History**: displays all the events and any notes associated with the sign-off with timestamp and the user details.

Analysis Settings

Analysis settings summarises the settings used by the kMap algorithm. These settings can be reviewed in the group account *Settings* page. The various settings are:

1. Array Type: KaryoMap v2
2. Scanning Hardware: iScan or NextSeq 550
3. Manifest File: KaryoMap-v2-1_A1.bpm
4. Flanking Size: 0.2 to 5 Mb (default is 2 Mb)
5. Analysis Version: The analysis version of the kMap algorithm.

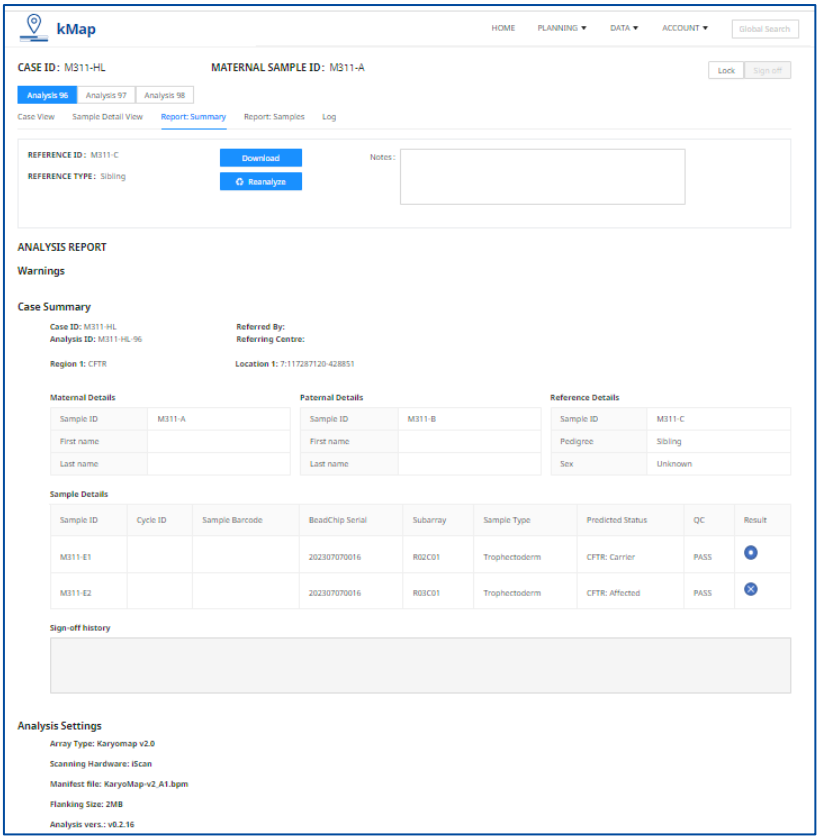


Figure 19: Report: Summary page – Case Summary, Sample Details, Sign-off and Analysis Settings

QC: BeadArays

The *QC: BeadArrays* section lists the BeadArray data for all the samples in the case including the BeadChip Serial, Subarray, Sample ID, Pedigree, Sample Type, QC status, Call Rate, AA, AB, BB, ADO Mis-call, X-Het rate and Y-Call Rate.

QC: DNA

The *QC: DNA* section lists the sample level information of DNA samples used in the Karyomapping assay. Sample ID, Designation, Sample Type, Sample Volume (µL), Concentration (ng/µL), 260/280 and 230/280.

Region Statistics

The region statistics are displayed for each region being investigated. The region statistics consist of

1. **Region Details**: Chromosome, Band, Start, End and Size
2. **Disease Status**: Disorder Type, Maternal Status, Paternal, Status and Reference Status.
3. **Available SNPs**: A table displays total number of Platform SNPs available for the selected region (or gene) and lists the number of Maternal and Paternal informative SNPs available for analysis. Available SNPS are split across the Left Flanking Region (3’), Main Region and Right Flanking Region (5’).

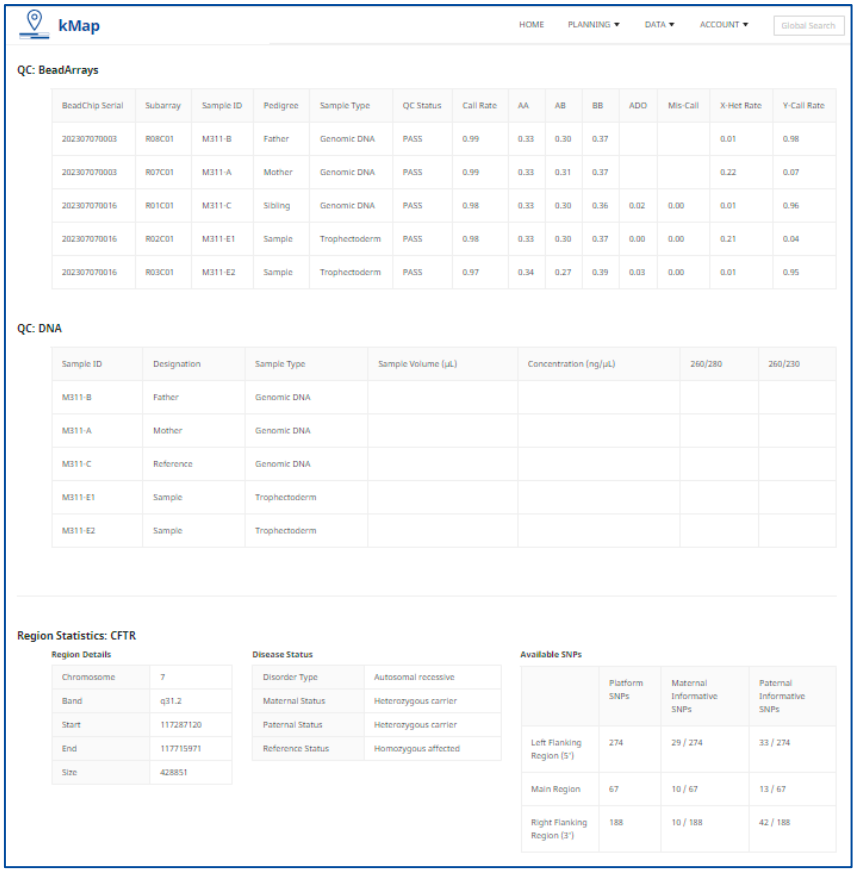


Figure 20: Report: Summary page – QC: BeadArrays, QC: DNA and Region Statistics

Report: Samples

The *Report: Sample* page displays the results for all the samples in the analysis, data is listed for each sample in the order of their accessioning. For each sample there is a header, Sample results table and a Region 1 table. If there is more than one region, each region will have a Region table (Figure 21).

The header includes the following details, Case ID, Sample ID, Maternal First Name, Maternal Last Name, BeadChp Serial and Subarray.

Sample Result tables displays the Call Rate, AA, AB, BB, ADO, Mis-Call, X-Het Rate, Y-Call Rate and Result.

The Region table lists the region name and the genomic coordinates. The Predicted Phase and Predicted Status is displayed along with Supporting Evidence (Key SNPs supporting the phase) and Contrary Evidence (Key SNPs supporting the phase).

Supporting Evidence for Paternal phasing is displayed in a table with the number of Key and Non-key SNPs supporting the phasing for Paternal-P1 (highlighted in Blue) and Paternal-P2 (highlighted in Red) in the Left Flanking Region (3'), Main Region and Right Flanking Region (5').

Supporting Evidence for Maternal phasing is displayed in a table with the number of Key and Non-key SNPs supporting the phasing for Maternal-M1(highlighted in Yellow) and Maternal-M2 (highlighted in Green) in the Left Flanking Region (3'), Main Region and Right Flanking Region (5').

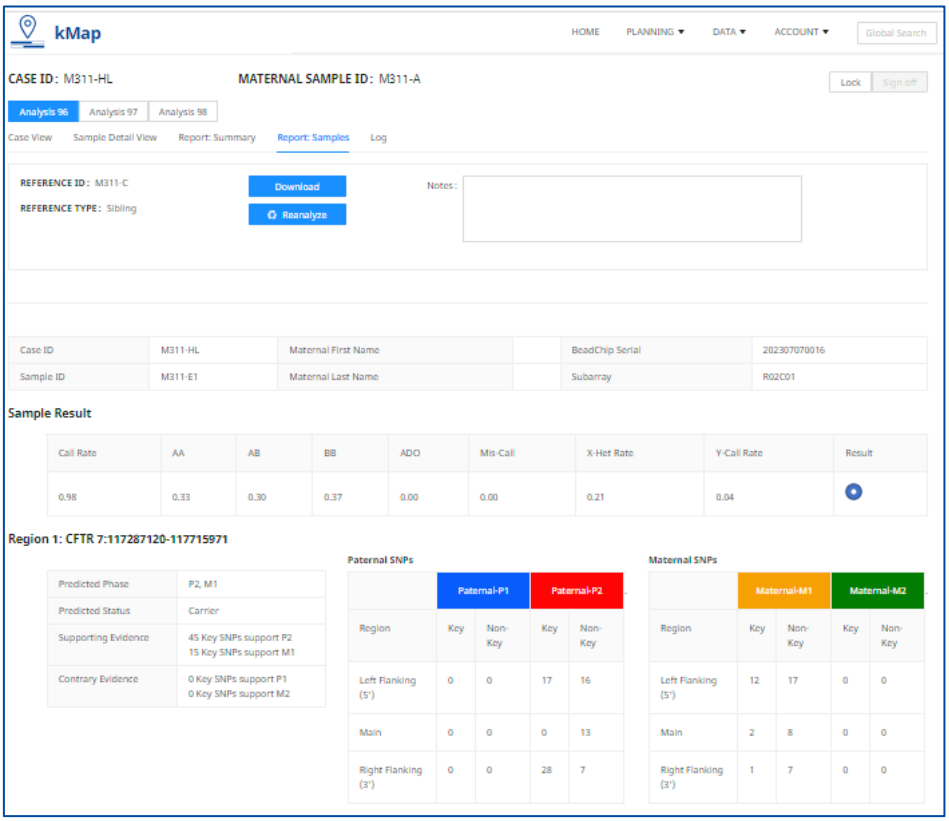


Figure 21: Report: Samples page.

Log

The *Log* page displays all the steps in the analysis and is required for troubleshooting (Figure 22).

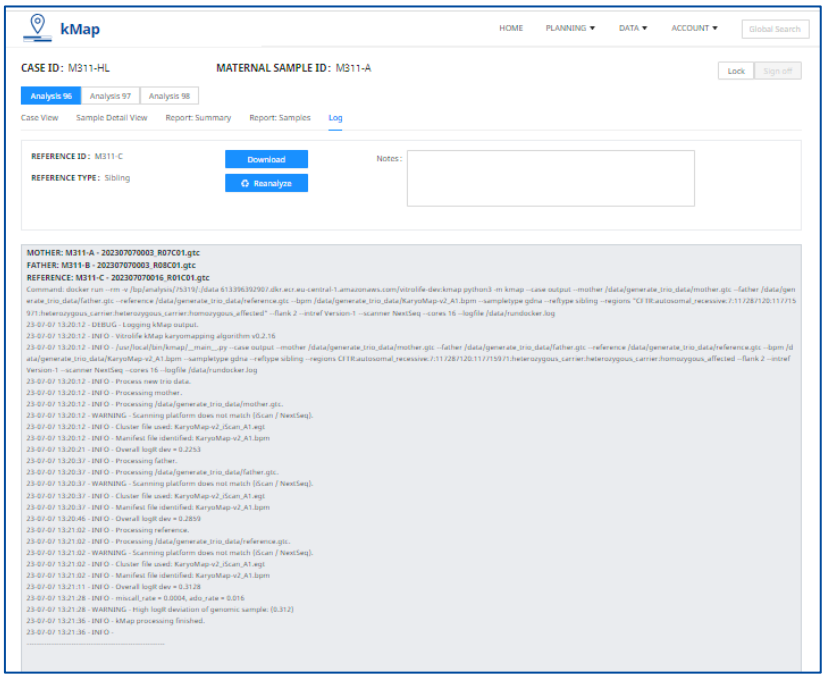


Figure 22: Log page.

Case Sign-Off

There are two levels to the Sign-Off procedure, Lock and Sign-Off. Locking a case prevents further changes being made to the analysis and protects the case pending final review. A case must be Unlocked to be edited.

Lock a Case

1. Level 1 and Level 2 Users can Lock and Unlock cases.
2. Click “Lock” from the Lock Sign off icons at the top of the *Case View* page.
3. The case will be locked and the Unlock Sign off icon will update to include “Unlock” and “Sign off” values.
4. To Unlock the sample, click Unlock Sign off .
5. All Locking and Unlocking actions are recorded in the system *Logs* with the User’s email address.

Sign-Off a Sample

1. Only Level 2 Users can Sign-Off cases.
2. A case must already be Locked, before it can be Signed-Off.
3. Click “Sign off” from the Unlock Sign off icons at the top of the *Case View* page.
4. A pop-up of the Sign-Off dialogue window will appear.
    a. Use the free-text box to enter Comments.
    Note: The Comments entered will appear on the Case Report PDF.
    b. Enter Username and password (Level 2 only).
    c. Click OK to confirm the Sign-Off or click Cancel to exit without saving.
5. Signed-Off cases are available for Reporting.
6. To cancel a Sign-Off, click Cancel Sign-off then Yes to confirm. The action will be recorded in system *Logs* with the User’s email address. The case will return to “Locked” status.

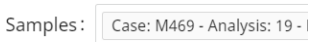
5 REPORTING

The Signed-Off case analysis results can be reported in a PDF document for archiving and distribution. Reports are generated with samples grouped by Analysis ID. This section describes the formatting and customisation of kMap Reports and how to build Reports.

Add Samples to a Report

The *Report Builder* tool allows selection of samples for reporting. Only Signed-Off samples will be available for selection.

1. Navigate to *DATA* > *Report Builder*.
2. Using the *Samples by Maternal Sample ID* or *Samples by Case* list on the left-hand side, select an Analysis-ID, then:
    a. Drag-and-drop or click on individual samples to add to the *Samples* bar, or;
    b. Use the Analysis-ID autofill button to add all samples for that Cycle to the *Samples* bar.
3. Click the "×" next to the Sample ID to remove a sample from the *Samples* bar and the Report PDF.



4. Addition of samples from different analyses will result in the creation of one Report PDF document for each analysis.
5. Once the required analyses are selected, click Create Report to proceed.

Create Report

The *Report Preview* page is shown after clicking Create Report on the *Report Builder* page. The *Report Preview* page is divided into two sections, Report Settings and Preview.

Report Settings

The Report settings can be pre-configured under *ACCOUNT* > *Settings*. The saved values will be automatically loaded here but may also be edited for each Report batch as required.

1. Add a company *Logo* and *Address*.
2. Enter a *Disclaimer* statement to appear in the footer of each page of the Report.
3. Enter a *Report introduction* to appear on the Case summary page of the Report only.
4. Use the selector checkboxes to include the charts:
    a. KaryoMap
    b. LogR Chart
    c. B-allele Chart
5. Select a page margin: Small or Normal.
6. Select a paper size: A4, Legal or Letter.

Preview

1. A list of Analysis IDs selected for reporting are shown under the heading *Preview* as tabs.
2. To generate a report firstly select the Analysis ID for the analysis of interest in the Preview section.

3. Click Generate report to open the PDF preview window.
4. A single Analysis for a Case will be included in the PDF report. See *Report Format.*
   Note: If multiple Analysis have been added to the report builder a report will need to be generated for each individual Analysis ID.
5. Click Download report to download the PDF analysis reports.
6. The PDF file name format will be <CaseID>_kmap.report.pdf
7. Clicking the download button within the preview window  will download the Case Report as a PDF but will not include the filename format described above.

Report Format

Each page of the Report contains the following elements in the footer section:

1. Custom disclaimer statement defined in *Report Settings*
2. Page number "*n* of *N*"
3. RUO statement
4. Printed (generated) date
5. Username (User's email address used for login)

The Report consists of two main parts, the first is an overview of the reported case (Case Overview) and the second is a summary section for each reported sample (Detailed Sample Summary). The first pages of the report contain the header "kMap Case Report" with the logo and address of the group.

Case Overview

Description

The *Description* section will display any introduction added in the settings page or the report builder page.

Warnings

The *Warnings* section will display all warnings related to the case.

Case Summary

The *Case summary* section will list the region(s) being investigated, Case level information (Maternal, Paternal and Reference details) from the *Accessioning* page The Maternal and Paternal details tables will consist of the headings described in Table 12. The Reference Details table will consist of the headings described **Table 13**.

Table 12: Parental details summary table

Parameter	Description
Sample ID	Sample identifier assigned during Accessioning
First Name	Sample first name assigned during Accessioning
Last Name	Sample last name assigned during Accessioning

Table 13: Reference details summary table

Parameter	Description
Sample ID	Sample identifier assigned during Accessioning
Sex	Reference sample sex
Pedigree	Reference sample pedigree assigned during Accessioning

Sample Details

The *Sample Details* table will summarise the results for all the samples in the analysis as described in **Table 14**.

Table 14: Sample details summary table

Parameter	Description
Sample ID	Sample identifier assigned during Accessioning
Cycle ID	Cycle ID identifier assigned during Accessioning
Sample Barcode	Sample identifier assigned during Accessioning
Subarray	The subarray position on the bead array chip for this sample
Sample Type	Sample type assigned during Accessioning
Predicted Status	The predicted status of the sample for the region(s) of interest
QC	QC Status (PASS or FAIL), according to QC thresholds
Result	kMap icon of the summary result for a quick overview

Sign-off History

A record of the Sign-off history for the case is provided. This includes the date of the sign off, user ID that signed off the case.

Analysis Settings

A record of the Analysis Setting for the reported case is provided (**Table 15**).

Table 15: Analysis Settings

Parameter	Description
Array Type	The type of bead array chip the samples were processed with
Scanning Hardware	The instrument type that was used to scan the BeadChip
Manifest file	The bead pool manifest (bpm) file used for the analysis
Flanking Size	The size of the flanking region used for analysis
Analysis vers	The version of the kMap analysis algorithm used

QC BeadArrays

The QC: BeadArrays table summarises the BeadArray QC status of all the samples in the analysis (

Report Section	Description
BeadChip Serial	Serial number of the BeadChip that the sample was processed with
Subarray	Subarray of the BeadChip the that the sample was processed with
Sample ID	Sample identifier assigned during Accessioning
Pedigree	Reference sample pedigree assigned during Accessioning

Sample Type	Sample Type assigned during Accessioning
QC Status	QC Status (PASS or FAIL), according to QC thresholds
Call Rate	The fraction of SNPs with a successfully called genotype
AA	The fractions of SNPs with an AA genotype
AB	The fractions of SNPs with an AB genotype
BB	The fractions of SNPs with an BB genotype
ADO	The estimated fraction of SNPs affected by allele dropout
Mis-Call	The estimated fraction of SNPs affected by genotype errors
X-Het Rate	The number of Heterozygous SNPs over the number of Homozygous and Heterozygous SNPs together on the X chromosome
Y-Call Rate	Number of SNP probes reported on the Y chromosome over the Number of SNPs on the Y chromosome

).

Table 16: QC: BeadArrays

Report Section	Description
BeadChip Serial	Serial number of the BeadChip that the sample was processed with
Subarray	Subarray of the BeadChip the that the sample was processed with
Sample ID	Sample identifier assigned during Accessioning
Pedigree	Reference sample pedigree assigned during Accessioning
Sample Type	Sample Type assigned during Accessioning
QC Status	QC Status (PASS or FAIL), according to QC thresholds
Call Rate	The fraction of SNPs with a successfully called genotype
AA	The fractions of SNPs with an AA genotype
AB	The fractions of SNPs with an AB genotype
BB	The fractions of SNPs with an BB genotype
ADO	The estimated fraction of SNPs affected by allele dropout
Mis-Call	The estimated fraction of SNPs affected by genotype errors
X-Het Rate	The number of Heterozygous SNPs over the number of Homozygous and Heterozygous SNPs together on the X chromosome
Y-Call Rate	Number of SNP probes reported on the Y chromosome over the Number of SNPs on the Y chromosome

QC: DNA

The QC: DNA table summarises the DNA quality of the input samples if the data is entered during Accessioning (**Table 17**).

Table 17: QC: DNA

Report Section	Description
Sample ID	Sample identifier assigned during Accessioning
Designation	The sample designation assigned during Accessioning
Sample Type	Sample Type assigned during Accessioning
Sample Volume (µL)	Volume of samples used on the assay
Concentration (ng/µL)	The DNA concentration of the sample
260/280	The ratio of absorbance at 260 nm and 280 nm for the sample
260/230	The ratio of absorbance at 260 nm and 230 nm for the sample

Region Statistics

An overview of the statistics for the region(s) of interest analysed in the cases are described in this section. These include the **Region Details**, **Disease Status**, and **Available SNPs**.

- **Region Details**: The genomic position and size of the region of interest.
- **Disease Status**: The disorder type and the status of the trio assigned during Accessioning.
- **Available SNPs**: The number of SNPs available on the platform for the region of interest and flanking region. The number of maternal/paternal informative SNPs for the region of interest and flanking region.

Detailed Sample Summary

The individual sample result pages that follow the case report summary detail the results for each sample in the analysis. The header of the *detailed sample summary* page includes Case ID, Sample ID, Maternal First Name, Maternal Last Name, BeadChip Serial, Subarray. If the sample information does not fit on a single page each additional page will repeat the header information to ensure sample tracking.

Sample Result

The *Sample Result* table summarises the results for the individual sample as in **Table 18**.

Table 18: Sample Results

Report Section	Description
Call Rate	The fraction of SNPs with a successfully called genotype
AA	The fractions of SNPs with an AA genotype
AB	The fractions of SNPs with an AB genotype
BB	The fractions of SNPs with an BB genotype
ADO	The estimated fraction of SNPs affected by allele dropout
Mis-Call	The estimated fraction of SNPs affected by genotype errors
X-Het Rate	The number of Heterozygous SNPs over the number of Homozygous and Heterozygous SNPs together on the X chromosome
Y-Call Rate	Number of SNP probes reported on the Y chromosome over the Number of SNPs on the Y chromosome
Result	kMap icon of the summary result for a quick overview

Region table

The results for the region(s) analysed in the case are summarised three individual tables:

- **Region Summary**: The predicted phase and status are presented, and the supporting and contrary evidence summarised.
- **Paternal SNPs information**: The number of P1 and P2 key and Non-Key SNPs in the region and both left (5') and right (3') flanking regions.
- **Maternal SNPs information**: The number of M1 and M2 key and Non-Key SNPs in the region and both left (5') and right (3') flanking regions.

KaryoMap chart

If the option to include the KaryoMap chart was selected a genome view haploblock chart is presented below the region summary.

LogR Chart and B-allele Chart

If the option to include the LogR Chart or the B-allele Chart option was selected, a second page is presented in landscape orientation with the header information included at the top of the page for sample tracking.

6 EXPORT DATA

Raw Input Files

The *Raw Input Files* section gives quick access to download the GTC files for all samples in the case.

Result Files

Result files for each Karyomapping analysis can be downloaded from the *Results Files* section. Results for each Analysis is grouped under a separate drop-down with the analysis name. The results files may be used to configure other applications to generate alternate Report formats. See *Appendix 3* for a list of data fields that are exported.

UI Files

The Analysis outcome of the algorithm for each Karyomapping analysis can be downloaded from the *UI Files* section. The Analysis outcome files for each Analysis is grouped under a separate drop-down with the analysis name.

7 TECHNICAL SUPPORT

For technical assistance, contact Vitrolife Genomics Support:

Email support.genomics@vitrolife.com

Website www.vitrolife.com

APPENDIX 1: FREQUENTLY ASKED QUESTIONS

1. Is my data safe?

kMap has been designed to ensure data security. Data protection is provided at multiple levels:

- The hardware and software infrastructure are set up by Amazon Web Services (AWS) to a level that reaches HIPAA and HITECH compliance and industry-recognised certifications and audits such as ISO 27001, FedRAMP, and the Service Organization Control Reports (SOC1, SOC2, and SOC3).
- All data is encrypted during transfer and at rest. Ask your representative about further protection mechanisms.
- By using upload keys and passwords, only the specified Group of Users have access to the data. Vitrolife personnel or the third-party provider managing the system, cannot log into your account without the User granting them access.
- If identifiable information is a concern, the kMap system can be used with non-identifiable IDs and without providing personal information such as first and last names.
- By default, Vitrolife does not have access to the data entered into the system.
- Please refer to the terms and conditions of the software and the contracts you have with Vitrolife for additional information.

2. Which browser should I use?

The kMap analysis software has been tested to be accessible using the following systems and browsers (earliest version tested):

Operating System	Browser
Microsoft Windows 10	Chrome 115.0.5790.102 Microsoft Edge 115.0.1901.183 Firefox Browser115.0.2
Microsoft Windows 11	Chrome 115.0.5790.102 Microsoft Edge 115.0.1901.183 Firefox Browser115.0.2
Apple OSX	Chrome 115.0.5790.102 Safari 16.5.2 Firefox 115.0.2

The Microsoft ”Internet Explorer” browser is outdated and not supported by kMap.

3. Why does the view not show the Case or Sample results as I expect?

Refresh your browser page to make sure you are viewing the most recent data.

4. My iScan is not set up for generating GTC files, how can I generate GTC files and analyse them in kMap?

You will need to generate GTC files using the “*Beeline*” software from Illumina. Please use the KaryoMap v2 Manifest (KaryoMap-v2-1_A1.bpm) and cluster files (KaryoMap-v2-1_iScan_A1.egt) along with the IDAT files created by the iScan to generate GTC files.

5. Is there a LIMS integration?

As there are many different laboratory data systems on the market and no standardised interfaces defined, kMap has the option to import Case and Sample data from simple tab-separated text files (Case Accessioning, Sample Accessioning and Fast Track, see section *2 Planning*) and export all results of a run to a tab-separated text file format (see section *6 Export Data*). This way the integration with a LIMS can be made with simple custom scripts.

6. Can I use GTC files generated with HumanKaryomap-12 beadChips for Trio samples and process them with GTC files generated with KaryoMap v2 BeadChips for test samples in a Karyomapping case?

No, you cannot analyse samples processed with HumanKaryomap-12 BeadChips and KaryoMap v2 BeadChips together in the same case. Data from HumanKaryomap-12 will not be recognised by the kMap software.

7. Can I process GTC files generated on an iScan and GTC files on a NextSeq together in the same case for Karyomapping analysis with kMap software?

No, you cannot process GTC files generated on an iScan and NextSeq 550 together in the same Karyomapping analysis. The kMap software will display an error.

APPENDIX 2: DATA IMPORT FORMATS

Note for all import files, the **date format** must be stored as text with the format *YYYY-MM-DD.* When opening template files in Microsoft Excel or similar, the date format may be automatically updated and will require converting to text before saving the file as a tab-delimited ".txt" file.

Case Accessioning

Case data including Case ID, Gene/Region and Sample (Trio and test samples) information can be entered into the system by importing from a predefined tab-delimited file (*kMap_CaseAccessioning.txt*).

Table 19: kMap_CaseAccessioning.txt requirements

Parameter	Description
[Header]	
File_Type	ca
File_Version	1
File_Date	Date the file was created, stored as text with the format YYYY-MM-DD
Case ID	Unique identifier for the Case
Mother First Name	Optional.
Mother Last Name	Optional.
Mother DOB	Optional. Date of Birth of the Subject, stored as text with the format YYYY-MM-DD
Father First Name	Optional.
Father Last Name	Optional.
Father DOB	Optional. Date of Birth of the Subject, stored as text with the format YYYY-MM-DD
Consultant	Optional.
Referring Centre	Recommended. Used to filter sample statistics on the HOME page
Notes	Optional.
Array Type	KaryoMap v2
[Gene Region]	
Region_Name	OMIM nomenclature of the gene / region of interest
Chromosome	Chromosome number 1 to 22 or X
Start	Genomic coordinate for the start of the region based on hg38 reference genome
End	Genomic coordinate for the end of the region based on hg38 reference genome
Inheritance_Type	autosomal dominant, autosomal recessive, x linked dominant, x linked recessive, unknown
Mutation_Site	Genomic coordinate of the mutation site as per the hg38 reference genome
Father_Status	heterozygous carrier, homozygous affected, homozygous normal, hemizygous affected, hemizygous normal
Mother_Status	heterozygous carrier, homozygous affected, homozygous normal
Reference_Status	heterozygous carrier paternal, heterozygous carrier maternal, homozygous affected, homozygous normal, unknown
[Trio]	
Pedigree	Pedigree is the genetic relationship of the DNA samples in relation to the test samples used in the karyomapping case. Pedigree can be one of the following: father, mother, matern aunt, matern

	grand father, matern grand mother, matern uncle, patern aunt, patern grand father, patern grand mother, patern uncle, sample, sample reference and sibling
Sex	Male, Female, Unknown
Type	Sample type. Allowed values are gDNA, trophectoderm, blastomere, other
Sample_ID	Unique identifier for the Sample; alphanumeric characters and dashes only [-]
Tube_ID	Unique identifier for the Sample tubes; alphanumeric characters and dashes only [-]
Sample_Barcode	Optional. Must be unique to kMap database
Requisition	Optional.
Volume_uL	Optional. The volume of sample (µL) used in the KaryoMap v2 assay
Concentration_nguL	Optional. The concentration of sample (ng/µL) used in the KaryoMap v2 assay
260/280	Optional. The 260/280 ration of the DNA sample used in the KaryoMap v2 assay
260/230	Optional. The 260/230 ration of the DNA sample used in the KaryoMap v2 assay

[Data]	
Type	Sample type. Allowed values are gDNA, trophectoderm, blastomere, other
Sample_ID	Unique identifier for the Sample; alphanumeric characters and dashes only [-]
Tube_ID	Unique identifier for the Sample tubes; alphanumeric characters and dashes only [-]
Sample_Barcode	Optional. Must be unique to kMap database
Requisition	Optional.
Cycle ID	Optional. Identifier of the Cycle containing Samples
Volume_uL	Optional. The volume of sample (µL) used in the KaryoMap v2 assay
Concentration_ nguL	Optional. The concentration of sample (ng/µL) used in the KaryoMap v2 assay
260/280	Optional. The 260/280 ration of the DNA sample used in the KaryoMap v2 assay
260/230	Optional. The 260/230 ration of the DNA sample used in the KaryoMap v2 assay

Sample Accessioning

Sample (test samples) data can be added to a pre-defined karyomapping case by importing from a predefined tab-delimited file (*kMap_SampleAccessioning.txt*).

Table 20: kMap_SampleAccessioning.txt requirements

Parameter	Description
[Header]	
File_Type	sa
File_Version	1
File_Date	Date the file was created, stored as text with the format YYYY-MM-DD
Case ID	Unique identifier for the Case. Must be an existing case ID in the kMap software
Array Type	KaryoMap v2

[Data]	
Type	Sample type. Allowed values are gDNA, trophectoderm, blastomere, other
Sample_ID	Unique identifier for the Sample; alphanumeric characters and dashes only [-]
Tube_ID	Unique identifier for the Sample tubes; alphanumeric characters and dashes only [-]
Sample_Barcode	Optional. Must be unique to kMap database
Requisition	Optional.

Cycle ID	Optional. Identifier of the Cycle containing Samples
Volume_uL	Optional. The volume of sample (µL) used in the KaryoMap v2 assay
Concentration_nguL	Optional. The concentration of sample (ng/µL) used in the KaryoMap v2 assay
260/280	Optional. The 260/280 ration of the DNA sample used in the KaryoMap v2 assay
260/230	Optional. The 260/230 ration of the DNA sample used in the KaryoMap v2 assay

Fast Track Import

If Laboratory Planning and BeadChip subarray tracking are to be performed using an alternate software or system (e.g. LIMS), Accessioning and Laboratory Planning data can be imported together using a single file for each Karyomapping case. The import file *kMap_FastTrack.txt* contains all relevant fields for Accessioning and Laboratory Planning. Upon importing a Fast Track file, the User can continue with directly uploading the scan data to the kMap software.

Table 21: kMap_FastTrack.txt requirements

Parameter	Description
[Header]	
File_Type	fa
File_Version	1
File_Date	Date the file was created, stored as text with the format YYYY-MM-DD
Case ID	Unique identifier for the Case
Mother First Name	Optional
Mother Last Name	Optional
Mother DOB	Optional. Date of Birth of the Subject, stored as text with the format YYYY-MM-DD
Father First Name	Optional
Father Last Name	Optional
Father DOB	Optional. Date of Birth of the Subject, stored as text with the format YYYY-MM-DD
Consultant	Optional
Referring Centre	Recommended. Used to filter sample statistics on the HOME page
Notes	Optional
Array Type	KaryoMap v2
[Gene Region]	
Region_Name	OMIM nomenclature of the gene / region of interest
Chromosome	Chromosome number 1 to 22 or X
Start	Genomic coordinate for the start of the region based on hg38 reference genome
End	Genomic coordinate for the end of the region based on hg38 reference genome
Inheritance_Type	autosomal dominant, autosomal recessive, X linked dominant, X linked recessive, unknown
Mutation_Site	Genomic coordinate of the mutation site as per the hg38 reference genome
Father_Status	heterozygous carrier, homozygous affected, homozygous normal, hemizygous affected, hemizygous normal
Mother_Status	heterozygous carrier, homozygous affected, homozygous normal
Reference_Status	heterozygous carrier paternal, heterozygous carrier maternal, homozygous affected, homozygous normal, unknown
[Trio]	

Pedigree	Pedigree is the genetic relationship of the DNA samples in relation to the test samples used in the karyomapping case. Pedigree can be one of the following: father, mother, matern aunt, matern grandfather, matern grandmother, matern uncle, patern aunt, patern grandfather, patern grandmother, patern uncle, sample, sample reference and sibling
Sex	Male, Female, Unknown
Type	Sample type. Allowed values are gDNA, trophectoderm, blastomere, other
Sample_ID	Unique identifier for the Sample; alphanumeric characters and dashes only [-]
Tube_ID	Unique identifier for the Sample tubes; alphanumeric characters and dashes only [-]
Sample_Barcode	Optional. Must be unique to kMap database
Requisition	Optional
Plate_ID	The Plate ID the Sample is assigned to after Laboratory Planning
Preparation_Date	The date the samples were prepared, as defined during Plate ID creation.
Well_position	The well positions of the plate in which the samples were processed
BeadChip_Serial	The serial number (Barcode) of the KaryoMap v2 BeadChip
Subarray	The subarray position of the KaryoMap v2 BeadChip on which a sample is processed. R01C01, R02C01, R03C01, R04C01, R05C01, R06C01, R07C01, R08C01
Volume	Optional. The volume of sample (µL) used in the KaryoMap v2 assay
Concentration	Optional. The concentration of sample (ng/µL) used in the KaryoMap v2 assay
260/280	Optional. The 260/280 ration of the DNA sample used in the KaryoMap v2 assay
260/230	Optional. The 260/230 ration of the DNA sample used in the KaryoMap v2 assay
Quad WG-Pre Lot	Lot Number of Reagent box, INF® HD Assay Kt Quad WG-Pre 16/48 SMP (11300762)
WG-Post 1 LV1 Lot	Lot Number of Reagent box, Infinium® HD Assay Kit WG-Post1 LV1 (11300771)
Single Post 3 LV Lot	Lot Number of Reagent box, Infinium® Assay Kit Single Post 3 LV (15023551)
Post 2 LMV Lot	Lot Number of Reagent box, Infinium® Assay Kit Post 2 LMV (15023542)
Post 4 LV Lot	Lot Number of Reagent box, Infinium® Assay Kit Post 4 LV (15023544)

[Data]	
Type	Sample type. Allowed values are gDNA, trophectoderm, blastomere, other
Sample_ID	Unique identifier for the Sample; alphanumeric characters and dashes only [-]
Tube_ID	Unique identifier for the Sample tubes; alphanumeric characters and dashes only [-]
Sample_Barcode	Optional. Must be unique to kMap database
Requisition	Optional
Plate_ID	The Plate ID the Sample is assigned to after Laboratory Planning
Preparation_Date	The date the samples were prepared, as defined during Plate ID creation.
Well_position	The well positions of the plate in which the samples were processed
BeadChip_Serial	The serial number (Barcode) of the KaryoMap v2 BeadChip
Subarray	The subarray position of the KaryoMap v2 BeadChip on which a sample is processed. R01C01, R02C01, R03C01, R04C01, R05C01, R06C01, R07C01, R08C01
Cycle ID	Optional. Identifier of the IVF Cycle Number
Volume_	Optional. The volume of sample (µL) used in the KaryoMap v2 assay
Concentration	Optional. The concentration of sample (ng/µL) used in the KaryoMap v2 assay
260/280	Optional. The 260/280 ration of the DNA sample used in the KaryoMap v2 assay
260/230	Optional. The 260/230 ration of the DNA sample used in the KaryoMap v2 assay
Quad WG-Pre Lot	Lot Number of Reagent box, INF® HD Assay Kt Quad WG-Pre 16/48 SMP (11300762)
WG-Post 1 LV1 Lot	Lot Number of Reagent box, Infinium® HD Assay Kit WG-Post1 LV1 (11300771)
Single Post 3 LV Lot	Lot Number of Reagent box, Infinium® Assay Kit Single Post 3 LV (15023551)

Post 2 LMV Lot	Lot Number of Reagent box, Infinium® Assay Kit Post 2 LMV (15023542)
Post 4 LV Lot	Lot Number of Reagent box, Infinium® Assay Kit Post 4 LV (15023544)

APPENDIX 3: EXPORT SAMPLE METADATA

Two set of files are available for download for each sample, “Stats file” and “Calls file”.

Parameter	Description
Stats file	
# header line	kMap algorithm version number
Case	The accessioned Case ID
File_Name	Default file name will be “<BeadChip-Serial>-<Subarray>gtc”
Pedigree	Pedigree is the genetic relationship of the DNA samples in relation to the test samples used in the karyomapping case. Pedigree can be one of the following: Father, Mother, Maternal Aunt, Maternal Grandfather, Maternal Grandmother, Maternal Uncle, Paternal Aunt, Paternal Grandfather, Paternal Grandmother, Paternal Uncle, Sample, Sample reference and Sibling
Sample_Name	Father, Mother, Reference, or Sample
Sample_ID	Sample ID as accessioned in kMap software
Sample_Type	gDNA, trophectoderm, blastomere, other
Ref_ID	Sample ID for the reference as accessioned in kMap software
Manifest_Name	Default: KaryoMap-v2-1_A1.bpm
Scanner_Name	Serial number of the scanning hardware as detected from the GTC file
Scanner_Type	iScan or NextSeq 550 as detected from the GTC file
Scan_Date	Date of BeadChip Scanning as detected from the GTC file
Analysis_Date	Date of karyomapping analysis by the kMap Software
Total_Calls	The total number of SNPs for which a genotype could be determined for the sample (out of the total number of SNPs on the BeadChip)
No-Calls	The number of SNPs for which no genotype could be determined for the sample.
Call-Rate	The fraction of called SNPs in the sample over the total number of SNPs on the BeadChip
Flank-Setting	Flanking region size as set by the user (default is 2, can be set between 0.5 to 5)
Miscall-Rate	The fraction of SNPs that most likely have false genotypes over the numbers of SNPs that can be assessed for correct genotypes
ADO-Rate	The fraction of SNPs that most likely have false genotypes because of Allele Drop Out (ADO) over the numbers of SNPs that can be assessed for ADO
Median-LogR	The median of the normalized measure of signal intensity for each SNP marker that produced data on the autosomes
Std-LogR	The standard deviation of the normalized measure of signal intensity for each SNP marker that produced data on the autosomes
Std-LogR-all	The standard deviation of the normalized measure of signal intensity for each SNP marker that produced data on all chromosomes
QC	QC status (PASS or FAIL)
X_Het-Rate	The fraction of heterozygous SNPs over the total number of SNP calls on the X chromosome. This is ~0.25 for female and ~0.0 for male samples
Y_Call-Rate	The fraction of SNPs called over the total number of SNPs available on the Y chromosome. This is ~0.0 for female and ~0.95 for male samples
Assigned Sex	The gender of the sample as assigned by the kMap software
AA-Calls	Total number of AA calls in the sample
BB-Calls	Total number of BB calls in the sample
AB-Calls	Total number of AB calls in the sample
Frequency(AA)	The fraction of AA calls over the total number of called SNPs
Frequency(AB)	The fraction of BB calls over the total number of called SNPs
Frequency(BB)	The fraction of AB calls over the total number of called SNPs

Warnings	Any warning related to the karyomapping case analysis
Region1_Name	The name of the gene / region of interest along with the chromosome, start and stop positions as per the hg38 human reference genome
Region1_Predicted_Phases	Predicted phasing of the sample in this region
Region1_Predicted_Status	Predicted status of the sample in this region
Region1_Supp	Supporting evidence for the predicted phasing (number of SNPs)
Region1_NonSupp	Opposing evidence for the predicted phasing (number of SNPs)
Region1_SNP_P1	Number of SNPs reported in the gene and flanking regions for haplotype P1 in this region
Region1_SNP_P2	Number of SNPs reported in the gene and flanking regions for haplotype P2 in this region
Region1_SNP_M1	Number of SNPs reported in the gene and flanking regions for haplotype M1 in this region
Region1_SNP_M2	Number of SNPs reported in the gene and flanking regions for haplotype M2 in this region
Disease_Status	Status of the test sample looking at all regions defined

Calls File	
snp	Name of the SNP probe
chrom	Chromosome name
chromnum	Chromosome number
position	Position of the SNP probe on the chromosomes
genomic_position	Genomic coordinates of the SNP as per the hg38 reference genome database
call	Genotype call for the SNP probe
gtscore	Genotype score for the SNP probe
baf	B-allele frequency data
logr	LogR Ratio data
is_key	Value is ”1” if it is a Key SNP
snp_phase	Phase of the SNP: 0 - no phase assigned, 1 - patern. phase 1, 2 - patern. phase 2, 3 - maternal phase 1, 4 - matern. phase 2
mat_block	Maternal haploblock phase (0/1/2) the SNP is part of
pat_block	Paternal haploblock phase (0/1/2) the SNP is part of
snp_description	Result of this SNP: No-Call, Not-informative, M1-Non-Key, M1-Key, M2-Non-Key, M2-Key, P1-Non-Key, P1-Key, P2-Non-Key, or P2-Key

APPENDIX 4: INSTALLING THE KMAP-UPLOADER SOFTWARE

The data output files (.gtc) required for analysis with the kMap software should be manually uploaded to the Group's kMap account. The kMap-Uploader software is provided to support this data upload into the kMap system.

Install kMap-Uploader Software

To install the kMap-Uploader software on a Microsoft Windows™ computer, follow the steps below.

1. Download and extract the software .zip package provided by your Vitrolife Support representative.
   The .zip folder contains:

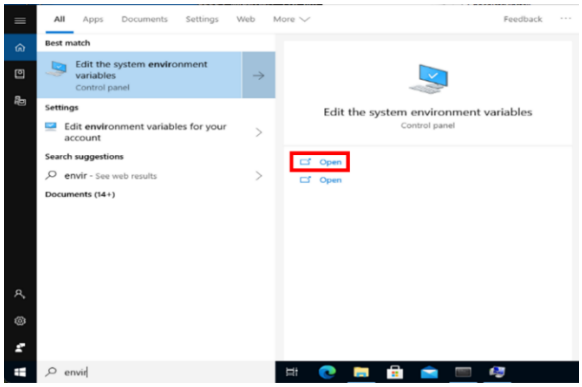
Config file name	Description
kMap-Uploader.exe	User interface to perform data uploads.
kMap-upload-cmd.exe *config.json* (User-supplied)	*Internal use* software to connect to kMap User account, validate input, transfer, and validate files.
.kMap_upload.cache	*Internal use* file, contains default data for *Advanced Configuration* settings

2. Copy the entire *kMap-Uploader* folder containing the software into the default location "*C:\Program Files (x86)*". Ensure the location is protected from unauthorised access (Administrator permission may be required). Custom locations may be used. Update the file paths accordingly in the kMap-Uploader (*kMap-Uploader.exe*) advanced configuration settings. Contact Vitrolife Support for assistance.
3. **Optional**. Add a shortcut to the *kMap-Uploader.exe* software on the Desktop.
4. Prepare the unique Access Key file:
   a. Copy and paste the entire personal **Access Keys** text from the kMap *ACCOUNT* > *Profile* page, into a text editor (e.g. Notepad++).
   b. Save with the file name *config.json.*
   c. Place the *config.json* file in the *kMap-Uploader* folder in the default location "*C:\Program Files (x86)\kMap-Uploader*".
   d. A log file is created when the kMap-Uplaoder software is started. The default location of the log file is ""*C:\Program Files (x86)\kMap-Uploader\kMap-Upload.log*".
5. Review the kMap_Upload.log file or share it with Vitrolife Support to troubleshoot any issues.

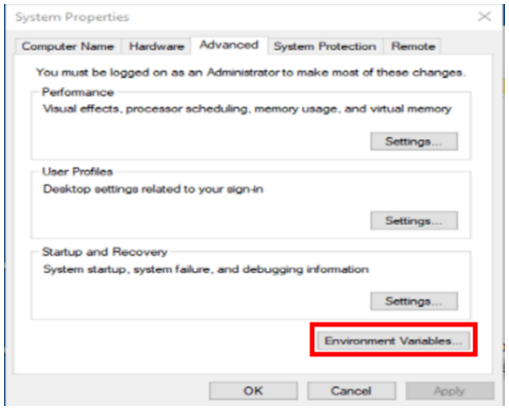
Store kMap-Uploader password

Your computer can be set up to store the kMap-Uploader password to avoid having to type in the password every time and to avoid displaying the password in sensitive environments. Please follow the steps below to set up your computer.

1. In the windows search box type “Edit the system environment variables”, navigate to the application and open it.
   Note: you can also access “Edit the system environment variables” from “Control Panel”



- In the System Properties click “Advanced” / “Environmental Variables” and a new dialogue box will open.



- Create new variable “KMAP_PASSWORD” by clicking “New” and add the required information. The new variable will be displayed in the list “User variables for IEUser” (or the respective current username).

2. Click ok and exit the programme. Your computer is now set up to remember the kMap password for kMap-Uploader.
   NOTE: For increased security the password will not be visible in the software UI when the password has been stored.