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BlueFuse™ Multi Analysis Software for Preimplantation Genetic Diagnosis

A powerful software package built by clinical geneticists for clinical geneticists offering quick, efficient, integrated analysis of array and sequencing data.

Highlights

- **Clear Data Interpretation**
  Optimized, validated algorithms produce accurate, easy-to-visualize haploblock charts
- **Rapid, Consistent Reporting**
  Automated report generation from barcoded data
- **Scalable Software Platform**
  Single software package provides data and information management solutions

Introduction

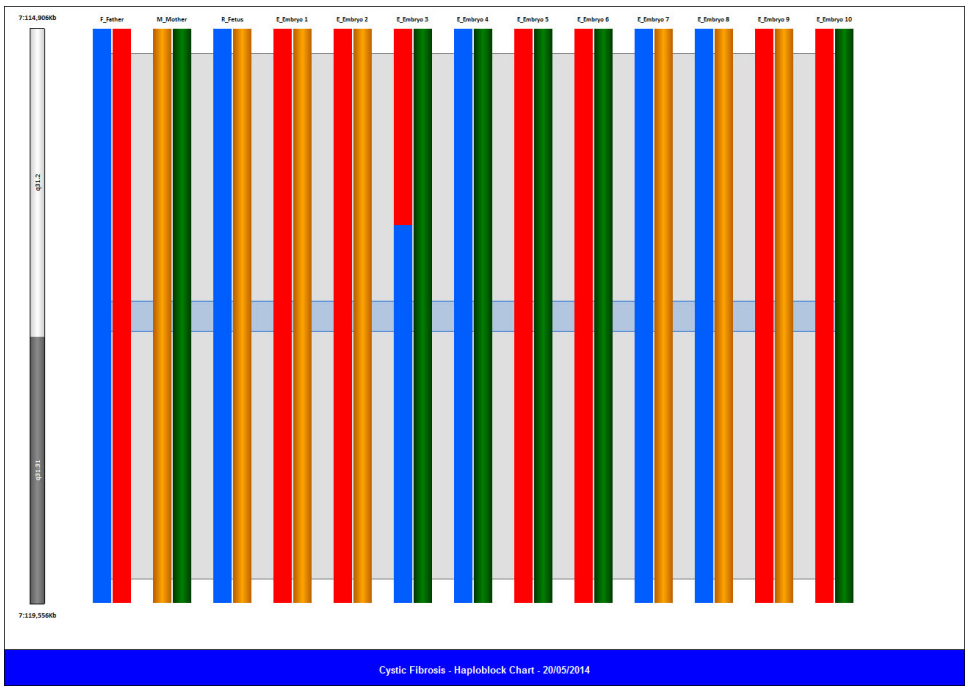
Preimplantation genetic diagnosis (PGD) enables screening of embryos for specific genetic conditions before an *in vitro* fertilization (IVF) procedure. Screening by PGD uses genetic markers located throughout the genome to assess the likelihood of an embryo carrying a mutated gene involved in a single-gene disorder. Karyomapping is one method currently available for performing PGD.

Karyomapping uses advanced array-based technology to provide a high-density view of the chromosomes in an embryo to establish the parental origin of inherited chromosomal segments, or haploblocks. Data generated through this reliable assay is analyzed using the BlueFuse Multi Analysis Software with karyomapping module. BlueFuse Multi Software provides optimized algorithms for tracking maternal and paternal inheritance patterns in the embryo for fast, reliable results and reports.

Visualize Single-Gene Inheritance

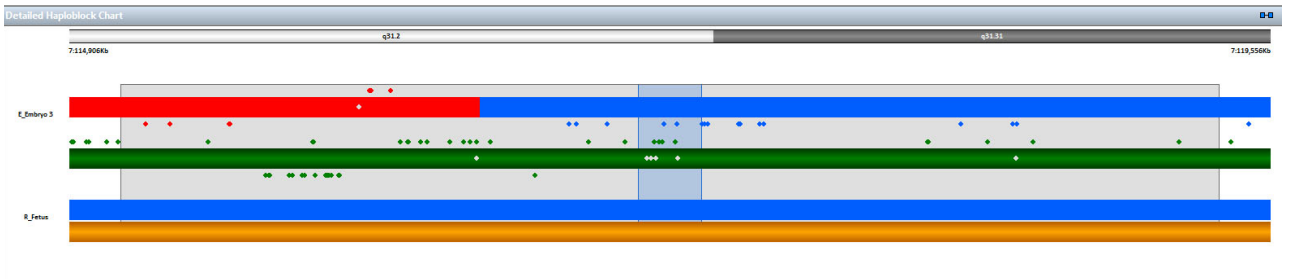
BlueFuse Multi Software with karyomapping module is a sophisticated multi-user database for storing, analyzing, and interpreting karyomapping results. From single-click batch import to automated

Figure 1: Haploblocks Showing Inheritance Patterns Surrounding the *CFTR* Region



Maternal and paternal inheritance patterns in the flanking regions around the CFTR gene (blue highlight) in the embryos.

Figure 2: Detailed SNP Information Used to Make an Inheritance Call



For each chromosome, key SNPs are displayed above the chromosome and non-key SNPs below the chromosome, enabling users to determine the accuracy of haploblock calling. SNPs failed to be called are displayed as white points in the middle of the chromosome.

reporting, BlueFuse Multi Software enables clinical laboratories to process large numbers of samples efficiently and store all cases in a central database.

The karyomapping module enables users to investigate and visualize the inheritance of single genes within embryos. Starting with data generated by the Infinium® HumanKaryomap-12 assay, a highly validated algorithm tracks haploblocks from the parents for quick, clear assessment of the genetic status of each embryo. Automatic report generation supports fast and easy data reporting.

Automated Algorithms for Rapid Interpretation

All PGD data are run through automated, validated algorithms to provide accurate, reliable results, independent of user preferences and settings. The proprietary karyomapping algorithm establishes which haploblocks, either maternal or paternal, the embryo has inherited across the genome*. Identifying the parental source allows detection of the likely phase and carrier status for each embryo for the gene, or region, of interest.

Clear Visualization for Data Confirmation

Key to karyomapping data interpretation is the ability to compare embryo inheritance to the status of a reference sample, such as a close relative of known disease status. The BlueFuse karyomapping module enables single-click zoom of a haploblock chart to show the inheritance pattern for all embryos of the gene, or region, of interest (Figure 1). Users can confirm the accuracy of the results by simply clicking each embryo number to see detailed information regarding key and informative SNPs in the haploblock (Figure 2). This clear, unambiguous data presentation enables users to make quick, informed decisions regarding the carrier status of each embryo. If needed, automated calls can be overridden manually.

Automated Report Generation

BlueFuse Multi Software automatically generates individual reports for each embryo as well as an overall case report. These reports are updated as changes are made to the case, ensuring that they are always up to date with the latest experiments.

Karyomapping Embryo Reports

A karyomapping embryo report is produced for every embryo. This simple report allows users to confirm that the assay has passed QC before performing the analysis. Information from this report is also available in the full karyomapping case report.

Karyomapping Case Reports

The karyomapping case report contains final phasing of all embryos along with biological and workflow details, information on all samples analyzed, the gene or region of interest, software version, and essential QC metrics (Figure 3). This report is easily exported, copied, or printed, and ready to pass on to a clinician.

Scalable Workflow

BlueFuse Multi Software supports multi-user access to the database at the same time to ensure the fastest workflows and ease of use. If installed with the recommended BlueFuse Server (optional), every user maintains write access to the database for full functionality. There are three levels of user access (Figure 4):

1. User—write samples and experiments to the database only
2. Supervisor—write, annotate, and delete samples and experiments to the database
3. Administrator—write, annotate, and delete samples and experiments to the database as well as change global database settings

* To learn more, download "A Technical Guide to Karyomapping Calling" from support.illumina.com/downloads/humankaryomap-12-v1-technical-guide-15052496.ilmn.

Figure 3: Karyomapping Case Report Example

Barcode	Array	Designation/Sample ID	Call Rate	AA	AB	BB	ADO	Mis-Call	X Heterozygous Rate	Y Call Rate
9322935132	R06C02	Father Father	0.98	0.33	0.29	0.38			0.01	0.94
9322935147	R01C01	Mother Mother	0.98	0.32	0.30	0.38			0.23	0.06
9322935147	R02C01	Reference Fetus	0.98	0.32	0.30	0.38	0.00	0.00	0.22	0.08
9322935147	R03C01	Embryo Embryo 1	0.87	0.37	0.21	0.42	0.09	0.00		
9322935147	R04C01	Embryo Embryo 2	0.97	0.33	0.29	0.38	0.00	0.00		
9322935147	R05C01	Embryo Embryo 3	0.93	0.35	0.25	0.40	0.10	0.00		

Region CFTR, Sample Embryo 1

Predicted Phase	M1, P2
Supporting Evidence	5 key SNPs support M1
	11 key SNPs support P2
Contrary Evidence	0 key SNPs oppose M1
	0 key SNPs oppose P2

Excerpts from a BlueFuse summary report. For each SNP, the report lists assay success rate in accurately calling genotypes at the global level. A detailed section shows additional SNP information from the flanking regions of the gene of interest.

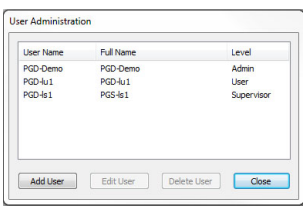
BlueFuse Server for Multi-User Write Access

The BlueFuse Server is an optional install that enables multiple users to write simultaneously to a central database. Without the BlueFuse Server, multiple users can log in with read-only access, but only one user can write to the database at any given time.

Complete Audit Trail Through Sample Sign-Off

It is critical that only high-quality data are used to generate the final reports that inform patient-physician decisions. Users inspect each experiment and provide "sign-off" indicating whether the experiment passed or failed. Each inspection includes a user name and time stamp to build an audit trail. The experiment is then passed to the next user and confirmed before sign-off by senior members of the team. BlueFuse Multi Software automatically tracks and stamps each interaction in the process. This trail cannot be deleted or edited, even by an administrator, providing a high-integrity audit trail of the sample (Figure 5).

Figure 4: Configuring User Level in the Database



BlueFuse ensures access control by assigning three user levels with different privileges and functions. The software supports multiple users at each level and administrators can reassign access levels any time.

Centralized Database

Experimental results analyzed using BlueFuse Multi Software are stored in a central database. Data are permanently available and can be revisited at any point in the future for comparison across multiple IVF cycles, which may have a treatment or counseling impact. Data storage also ensures that any follow-up questions can be readily answered.

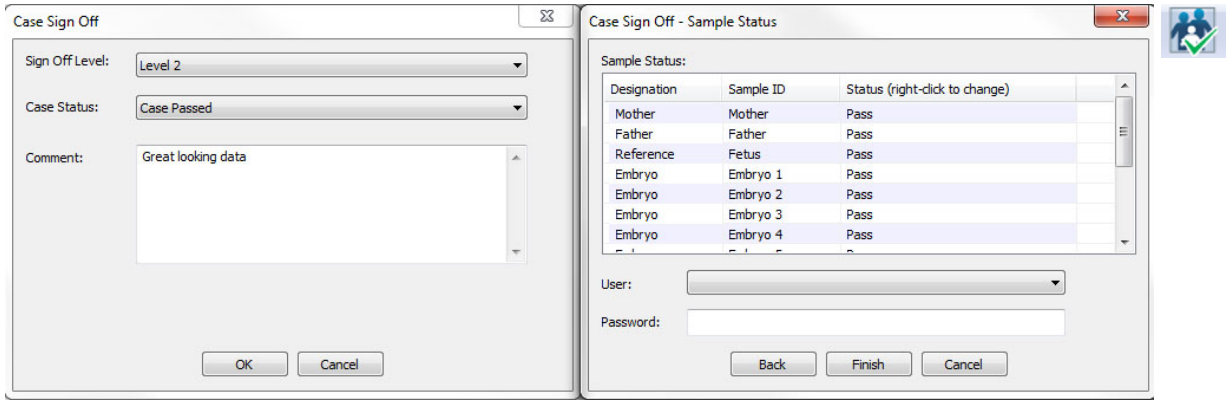
Integrated Framework

BlueFuse Multi Software offers a single framework for analyzing data for IVF and molecular cytogenetic applications. Developed by the clinical genetics community for the clinical genetics community, the clear intuitive interface ensures that the unique needs of these laboratories are met. In addition, the software supports analysis and visualization of sequencing and array data on the same platform. This integrated approach enables IVF laboratories to run a suite of technologies and expand their testing portfolios.

Summary

BlueFuse Multi Software with karyomapping module enables rapid, accurate visualization and reporting of PGD cases. Optimized algorithms combined with automated report generation allow laboratories to run multiple tests per day with confidence and minimal hands-on time. In addition, the integrated framework ensures that laboratories are able to scale rapidly to higher volumes and offer new tests with minimal staff training. Designated user login and profiles coupled with experiment sign-off requirements allows the lab to control and track data through the workflow and institute.

Figure 5: Sample Sign-Off Report



BlueFuse Software requires case sign-off for each report, establishing a clear audit trail for each sample.

Ordering Information

Access to BlueFuse Multi Software is available free of charge to users of Illumina cytogenetic and IVF technologies with unlimited licenses per site. The software is not available as an open platform to other vendors or LDTs. Download the software from www.cambridgebluegnome.com/software. For license information, contact techsupport@illumina.com.

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