PGT-A

Preimplantation Genetic Testing for Aneuploidy (PGT-A) of GeneMind

Easy-to-use

Automated and simplified sequencing and data analysis workflow

Fast

Quick test from sample preparation to final report within 24 hours

Flexible

Compatible with common used NGS library prep kits and bioinformatics tools

High sensitive

Accurately detect different types of mutations even in a low copies
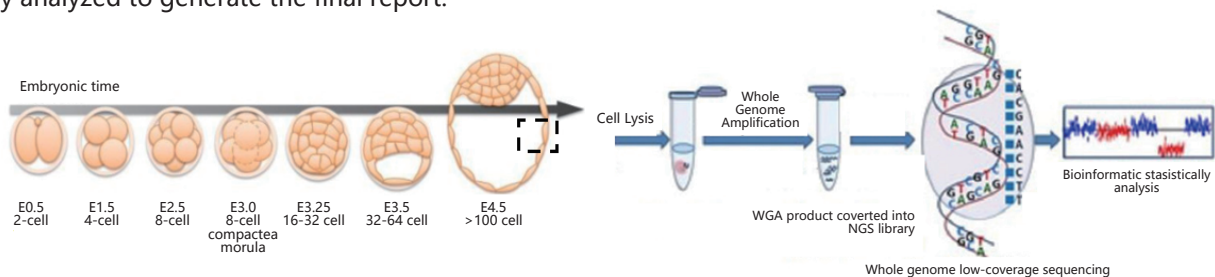
◆ Introduction of PGT-A

Single-cell whole genome amplification (WGA) and whole genome low-coverage sequencing technologies are used to detect chromosomal copy number variations (CNVs) by sequencing the genomic DNA from 5-10 embryo cells. The differences in the number of respective chromosomes are statistically analyzed using the data analysis software to evaluate whether any CNVs are detected in the whole cell genome.

Preimplantation genetic testing for aneuploidy identifies chromosomal abnormalities in embryo samples for the research of in vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI). These genetic insights may increase the probability of success for future pregnancies and healthier future generations.

Using GeneMind single-cell amplification kits, PGT-A library preparation sets and together with GenoLab M* / FASTASeq 300*/ SURFSeq 5000* next-generation sequencing system could accurately detect different mutations including 23 pairs of chromosomal aneuploidy abnormalities, microdeletions / microduplications over 4 Mb in the whole region, and microdeletions / microduplications over 1 Mb in the known region.

◆ Detection Process

Picogram quantities of genomic DNA is extracted from embryo cells and then subjected to do the WGA. The amplification products can be sequenced by the high-throughput gene sequencing platform and then automatically analyzed to generate the final report.

Embryonic time

| E0.5 2-cell | E1.5 4-cell | E2.5 8-cell | E3.0 8-cell compactea morula | E3.25 16-32 cell | E3.5 32-64 cell | E4.5 >100 cell |

Cell Lysis — Whole Genome Amplification — WGA product coverted into NGS library — Bioinformatic stasistically analysis

Whole genome low-coverage sequencing

◆ Workflow on GenoLab M* Sequencing System

| 4 h WGA | 2.5 h Library preparation | 0.5 h Library pooling and loading | 15 h Sequencing | 2 h Analysing and reporting |

◆ Workflow on FASTASeq 300* Sequencing System

| 4 h WGA | 2.5 h Library preparation | 0.5 h Library pooling and loading | 7 h Sequencing | 2 h Analysing and reporting |

◆ Workflow on SURFSeq 5000* Sequencing System

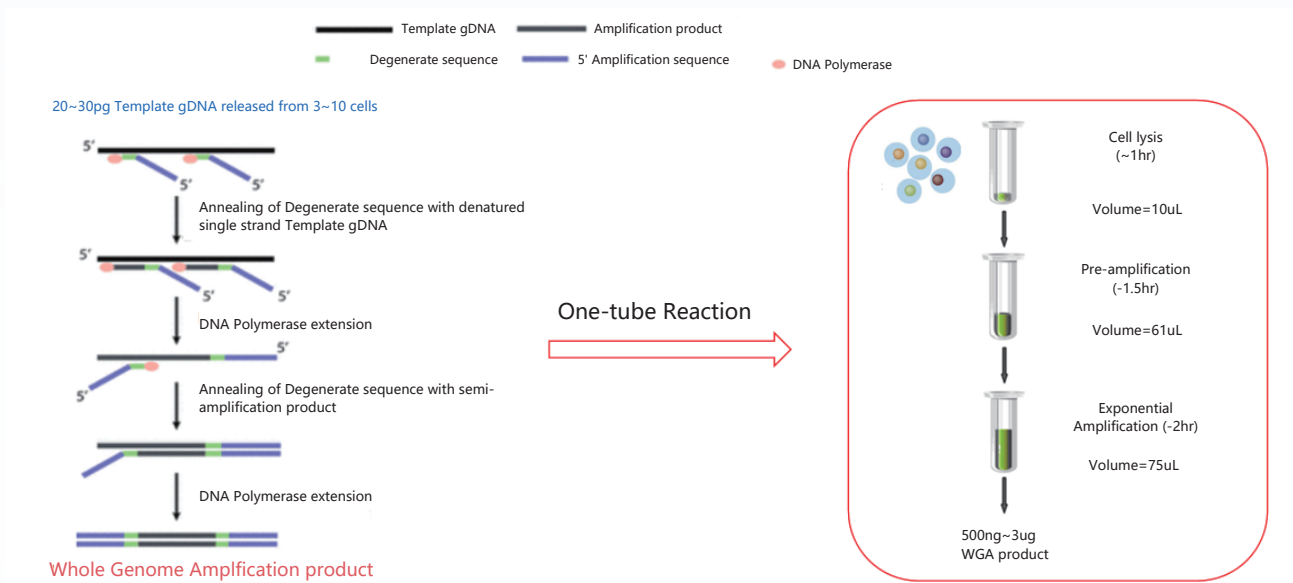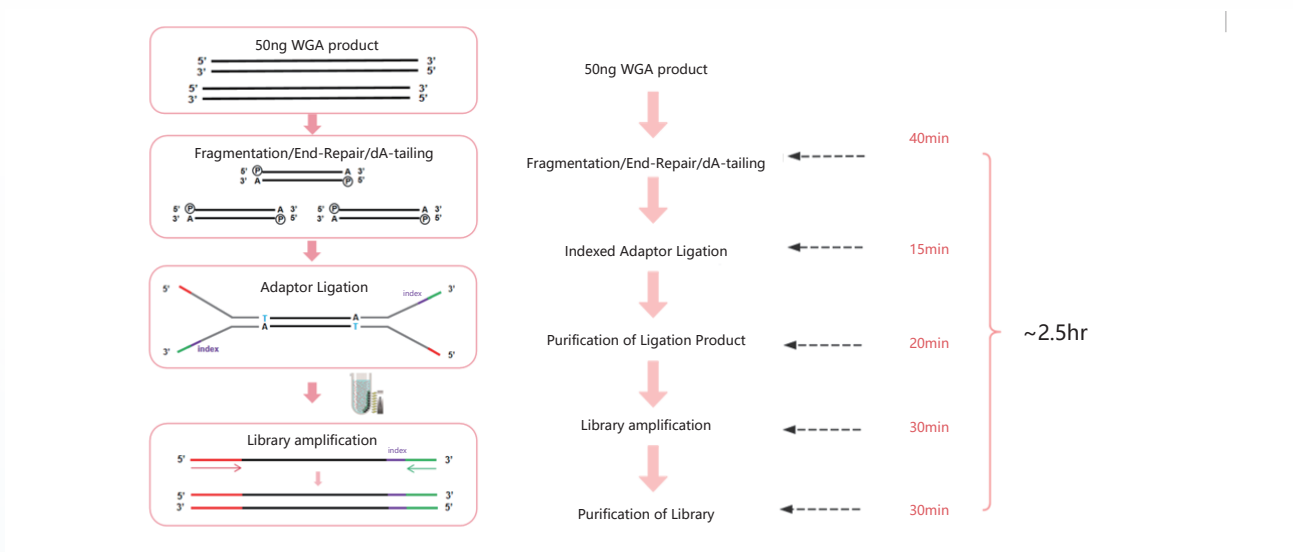| 4 h WGA | 2.5 h Library preparation | 0.5 h Library pooling and loading | 14 h Sequencing | 2 h Analysing and reporting |

◆ Single-cell Genome Amplification Process

Template gDNA Amplification product
Degenerate sequence 5' Amplification sequence
DNA Polymerase

20~30pg Template gDNA released from 3~10 cells

Annealing of Degenerate sequence with denatured single strand Template gDNA

DNA Polymerase extension

Annealing of Degenerate sequence with semi-amplification product

DNA Polymerase extension

Whole Genome Amplification product

One-tube Reaction

Cell lysis (~1hr)
Volume=10uL

Pre-amplification (-1.5hr)
Volume=61uL

Exponential Amplification (-2hr)
Volume=75uL

500ng~3ug WGA product

◆ Library Construction Process

50ng WGA product

Fragmentation/End-Repair/dA-tailing

Adaptor Ligation
index

Library amplification
index

50ng WGA product

Fragmentation/End-Repair/dA-tailing — 40min

Indexed Adaptor Ligation — 15min

Purification of Ligation Product — 20min

Library amplification — 30min
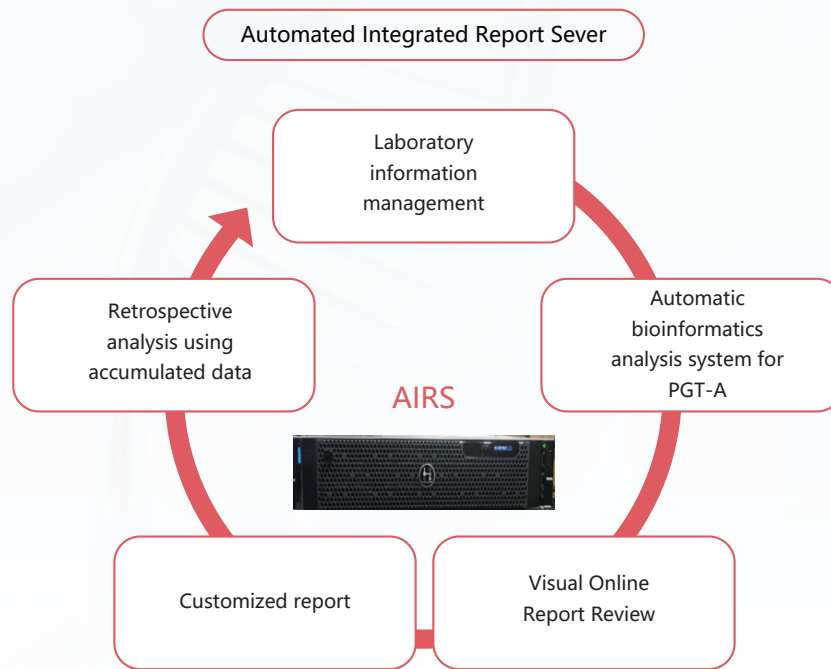
Purification of Library — 30min

~2.5hr

◆ GeneMind Data Analysis System

No.	Sample No.	Data quality control	Unique reads	Coverage rate(%)	GC Content(%)	SD	Report result	Scatter map of the genome
1	sample173	Qualified	22625948	25.15	39.18	0.115	46,XX,-1(q43-q44,~9.9Mb)	
2	sample174	Qualified	17696734	20.09	39.05	0.116	46,XX,-17(p13.3-p11.2,~4.2...	
3	sample175	Qualified	19900590	24.46	38.84	0.082	46,XX,-17(p11.2-p11.2,~4.3...	
4	sample171	Qualified	16683910	21.33	39.05	0.093	46,XX,-1(q24.1-q32.1,~33.3...	
5	sample172	Qualified	9991814	13.52	40.01	0.116	46,XX	
6	sample167	Unqualified	17852069	5.32	37.86	0.281	46,X(Y,-13)(q21.33-q22.1,~4...	
7	sample168	Unqualified	18342578	3.48	39.5	0.379	45,X(O,-X,+8)(q13.1-q24.3,~...	
8	sample164	Qualified	21338151	24.1	39.1	0.116	45,X(O,-X,+8)(q13.1-q24.3,~...	
9	sample162	Qualified	20095130	24.5	38.7	0.114	45,X(O,-X,+8)(q13.1-q24.3,~...	
10	sample165	Unqualified	7267853	7.22	38.33	0.346	46,X+3(mos,~34.0%),+7(...	
11	sample169	Qualified	16807340	20.9	39	0.077	46,XX,-11(q23.1-q25,~14.7...	
12	sample152	Qualified	72415400	49.08	38.4	0.121	46,XX,-11(q23.1-q25,~14.7...	
13	sample163	Qualified	15806675	19.56	38.88	0.116	45,X(O,-X,+8)(q13.1-q24.3,~...	

Chromosomal aneuploidy data analysis and report software installed in on-site server

The chromosomal aneuploidy data analysis software can accurately detect chromosomal aneuploidy, chromosomal deletions or duplications over 1 Mb in the known region as well as more than 30% of chimeric abnormalities.

Automated Integrated Report Sever

Laboratory information management

Automatic bioinformatics analysis system for PGT-A

AIRS

Retrospective analysis using accumulated data

Customized report

Visual Online Report Review

◆ Data Performance

WGA is performed using the GenoVerse Single-cell WGA Kit V1.0, and the library of WGA products is constructed using the Preimplantation Genetic Testing For Aneuploidy Detection Library preparation Kit V1.0.

The obtained library is sequenced and analyzed using the GenoLab M* / FASTASeq 300*/ SURFSeq 5000* next-generation sequencing system and the GeneMind chromosomal aneuploidy data analysis software, respectively.

The data analysis results show that reads are quite evenly distributed in the whole genome (Figure 1); The chromosomal aneuploidy abnormalities can be accurately identified even in a quite low copies (Figure 2); Some microduplications as low as 1 Mb can be successfully detected as well (Figure 3).

The results show that GenoVerse Single-cell WGA Kit V1.0 has good uniformity and could be used for PGT-A analysis when combined with Preimplantation Genetic Testing For Aneuploidy Detection Library preparation Kit
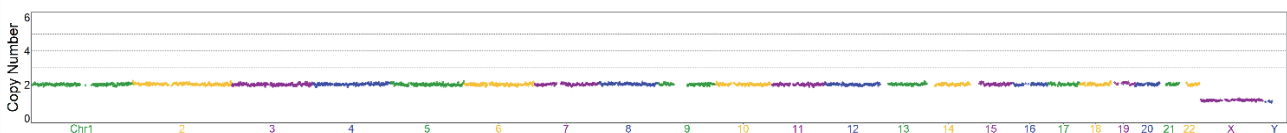

Figure 1. Scatter plot of whole genome copy number — negative samples
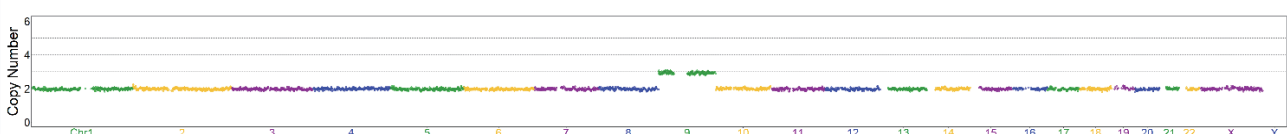

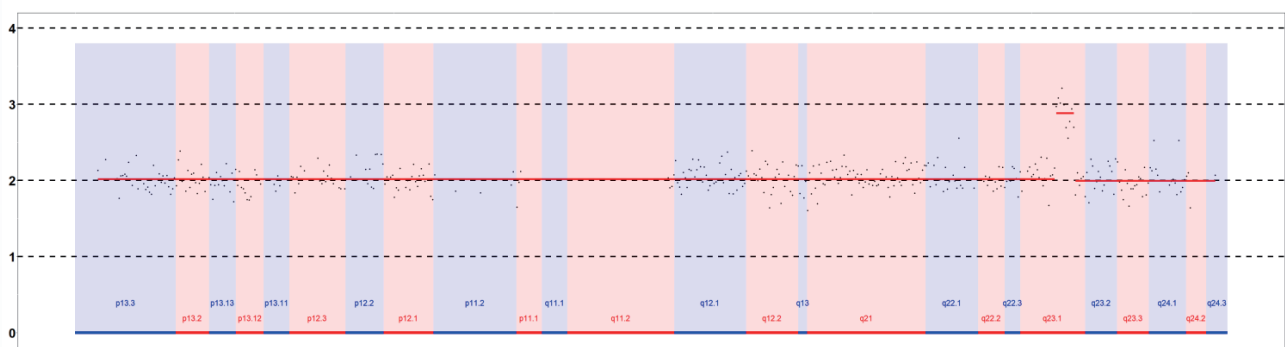Figure 2. Scatter plot of whole genome copy number — T9 samples

Figure 3. Scatter plot of Chr16 copy number — dup (16) (q23.1) samples

In addition, the chromosomal aneuploidy data analysis software includes the CNV interpretation system, which can automatically annotate CNV results and help end-users further understand the relation between the clinical symptoms and CNVs. The CNV interpretation system provides information about microdeletions/micro-duplications from databases such as OMIM, Decipher, ClinVar, ClinGen, dbVar, and MedGene. The CNV results will be ranked automatically according to ACMG guidelines, evaluates its clinical significance, and provides interpreta-

◆ GeneMind PGT-A Solution

PARAMETER			
Sample volume	5-10 embryo cells		
Library preparation	WGA		
Method	Whole genome sequencing		
Read length	SE75 for GenoLab M* and FASTSeq 300*　　SE50 for SURFSeq 5000*		
Sequencing platform	GenoLab M*	FASTASeq 300*	SURFSeq 5000*
No. of samples/run	1 FCM : 32 2 FCM/1 FCH : 64 1 FCM+1 FCH: 96 2 FCH: 128	1 FCM : 12 1 FCH : 32	1 FCH : 160 2 FCH : 320
Turn-around-time	24 h	16 h	23 h
Reads requirement/sample	GenoLab M*/FASTASeq 300*: Unique reads per sample ≥ 3.5M; SURFSeq 5000*　Unique reads per sample ≥ 4M		
Report generation	Local analysis and report system		

◆ About GeneMind

GeneMind Biosciences Co., Ltd. was founded in 2012. The headquarters is located in Luohu, Shenzhen. Its facility includes more than 10,000 square meters of research and development labs and GMP production line.

Specializing in the independent R&D and manufacturing of molecular diagnosis technology platform centred on DNA sequencing system, GeneMind Biosciences is devoted to constructing a precision medicine ecosystem to serve human life and health through collaboration with genetic testing service providers and medical institutions.

GeneMind has launched the high-throughput sequencing platform GenoLab M*, FASTASeq 300* and SURF-Seq 5000* through independent research and innovation, and offers the "equipment-reagent-flowcell-software" total platform solution. GeneMind is one of the few technology companies with core upstream technology and independent brand of gene sequencing in the world.

*Unless otherwise informed, GeneMind sequencing platform and related sequencing reagents are not available in the USA, Canada, Australia, Japan, Singapore, Western Europe, Southern Europe and Nordic countries yet.