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Role of Clinical Genetics in PGT

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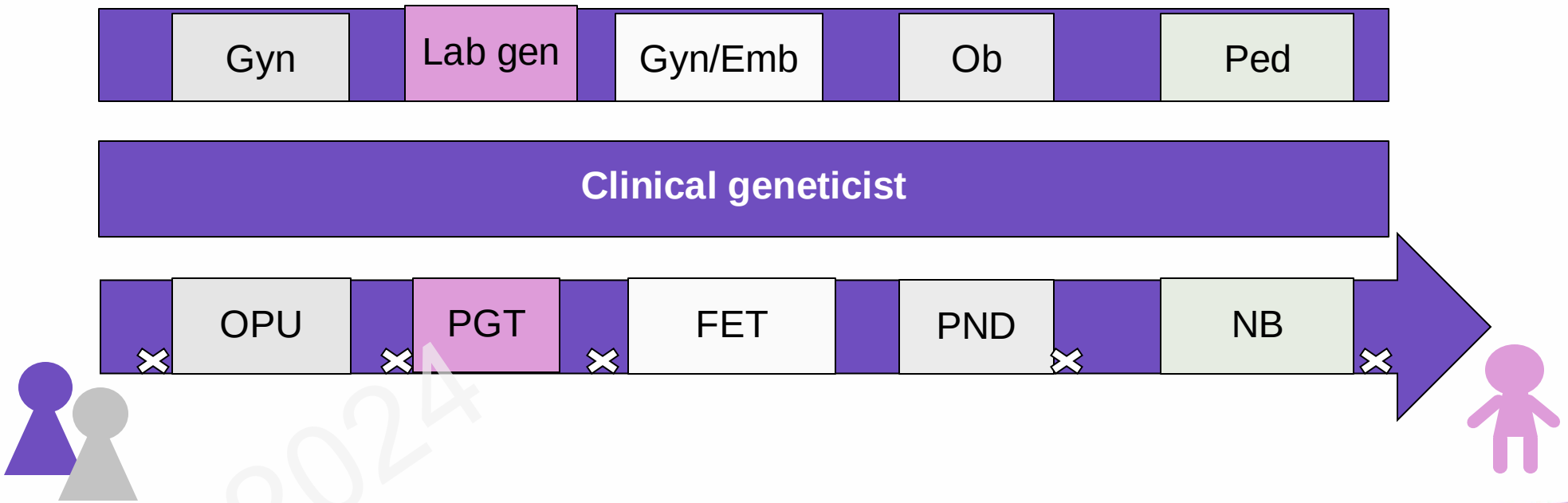
Role of clinical geneticist in PGT management



- Give five genetic counselling to each PGT patient at least
- Work together with specialists from 5 dept

Mostly asked Questions

- From patients
  - Can I do PGT?
- From Gyn
  - Multiple RCTs showed PGT-A did not work, why do it?
- From laboratory geneticist
  - Mosaic status

- 1. Indications of PGT

The International Glossary on Infertility and Fertility Care, 2017

The International Committee for Monitoring Assisted Reproductive Technologies (ICMART), ASRM, ESHRE, IFFS, FIGO etc.

A test performed to analyze the DNA from oocytes determining genetic abnormalities (polar bodies) or embryos (cleavage stage or blastocyst) for HLA-typing or for. These include:

- PGT-A PGT for aneuploidies
- PGT-M PGT for monogenic/single gene defects
- PGT-SR PGT for chromosomal structural rearrangements

Genetic Risks of PGT

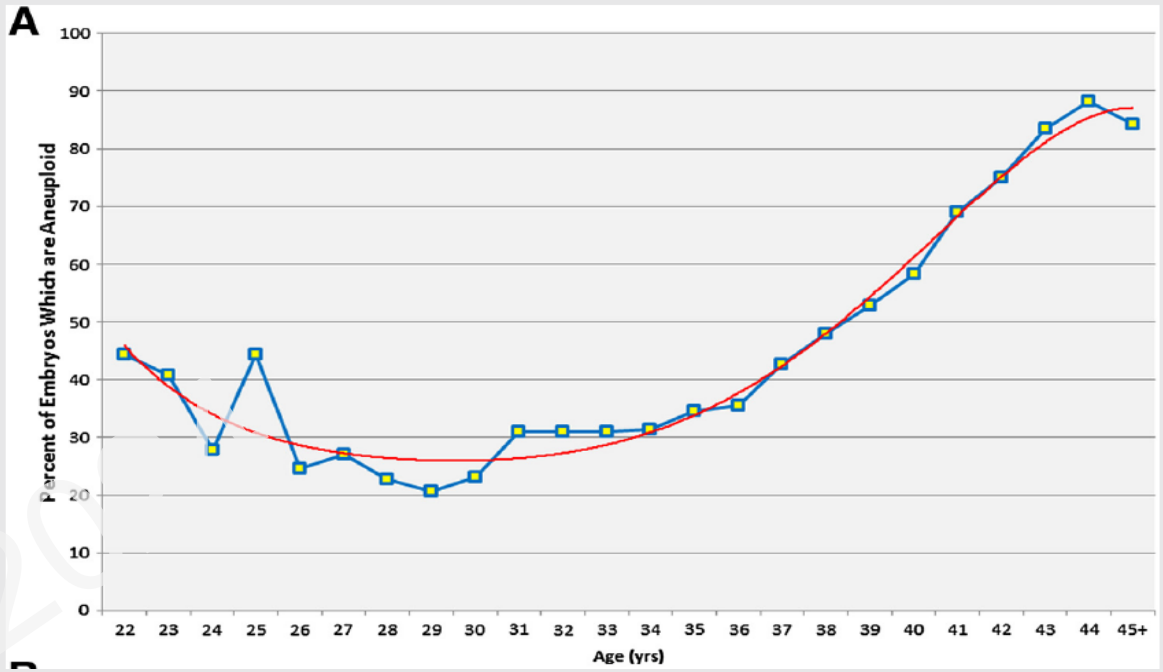
	Genetic Risk Observed	Harms	
		Maternal	New born
PGT-A	>20% for all ages	Implantation failure, biochemical pregnancy, miscarriage, induction of labor	
PGT-SR	~2/3	Implantation failure, biochemical pregnancy, miscarriage, induction of labor	
PGT-M	Mostly >25%	Induction of labor	Abnormal structure and function

Genetic Risks Management of PGT

	Risk Management		
	ESHRE 2005	Reproductive Medicine Branch, CMA 2017	Reproductive Medicine Branch, CPA 2018
PGT-A (observed risk%)	>36yr; 2 miscarriages; multiple implantation failures	>38yr; 3 miscarriages OR 2 miscarriages with 1 confirmed CNV; multiple implantation failures	>38yr; 2 miscarriages; multiple implantation failures
	>30%	>40%	>40%
PGT-SR PGT-M	>10% genetic risk; HLA matching	Genetic disease, chromosome rearrangement, CNV; HLA matching	+carry genetic susceptible mutation, such as BRCA1 or BRCA2,

Age versus aneuploidy

FIGURE 2



Prevalence of aneuploidy. (A) The prevalence of aneuploidy relative to the age of the female partner demonstrates the lowest risk in women from their middle to late twenties, with significantly higher rates in embryos obtained from both younger and older women ($P<1*10^{-6}$). The relationship between age and the rate of aneuploidy is a best fit at the 5th degree polynomial (regression line shown). (B) The relationship between maternal age and the probability that no euploid blastocysts will be available within a single cohort demonstrates a uniformly low risk between the maternal ages of 26 and 37 years. Higher risks are present in younger and older patients ($P<.0003$ or less).

Franasiak. Aneuploidy versus age. Fertil Steril 2014.

Franasiak, Fertil Steril, 2014

Personal opinion

- We may apply the same genetic risk standard to PGT indication
- Patient's decision should be taken into consideration of priority
- PGT manages genetic risk and should be used to improve health of both pregnancy and newborns.

- 2. Does PGT-A work?

Facts we know

- Euploidy embryo transfer has better outcomes than aneuploid transfer
- Theoretically PGT determines genetic abnormalities
- Multiple PGT-A RCTs question about whether euploidy screening can improve pregnancy outcomes after embryo transfer

Rubio C, et. al. *Fertil Steril* 2017
Ozgur K, et. al. *J Assist Reprod Genet* 2019
Tiegs AW, et. al. *Fertil Steril* 2021
Yan JH, et. al. *NEJM* 2021
Wang L, et. al., *Prenat Diagn* 2021

JOURNALS

PRENATAL DIAGNOSIS

ORIGINAL ARTICLE

PGT-A: The biology and hidden failures of randomized control trials

Li Wang, Xiaohong Wang, Min Li, Yun Liu, Xianghong Ou, Lei Chen, Xiaoguang Shao, Song Quan, Jinliang Duan, Wei He, Huan Shen, Ling Sun, Yuexin Yu, David S. Cram, Donald Leigh, Yuanqing Yao

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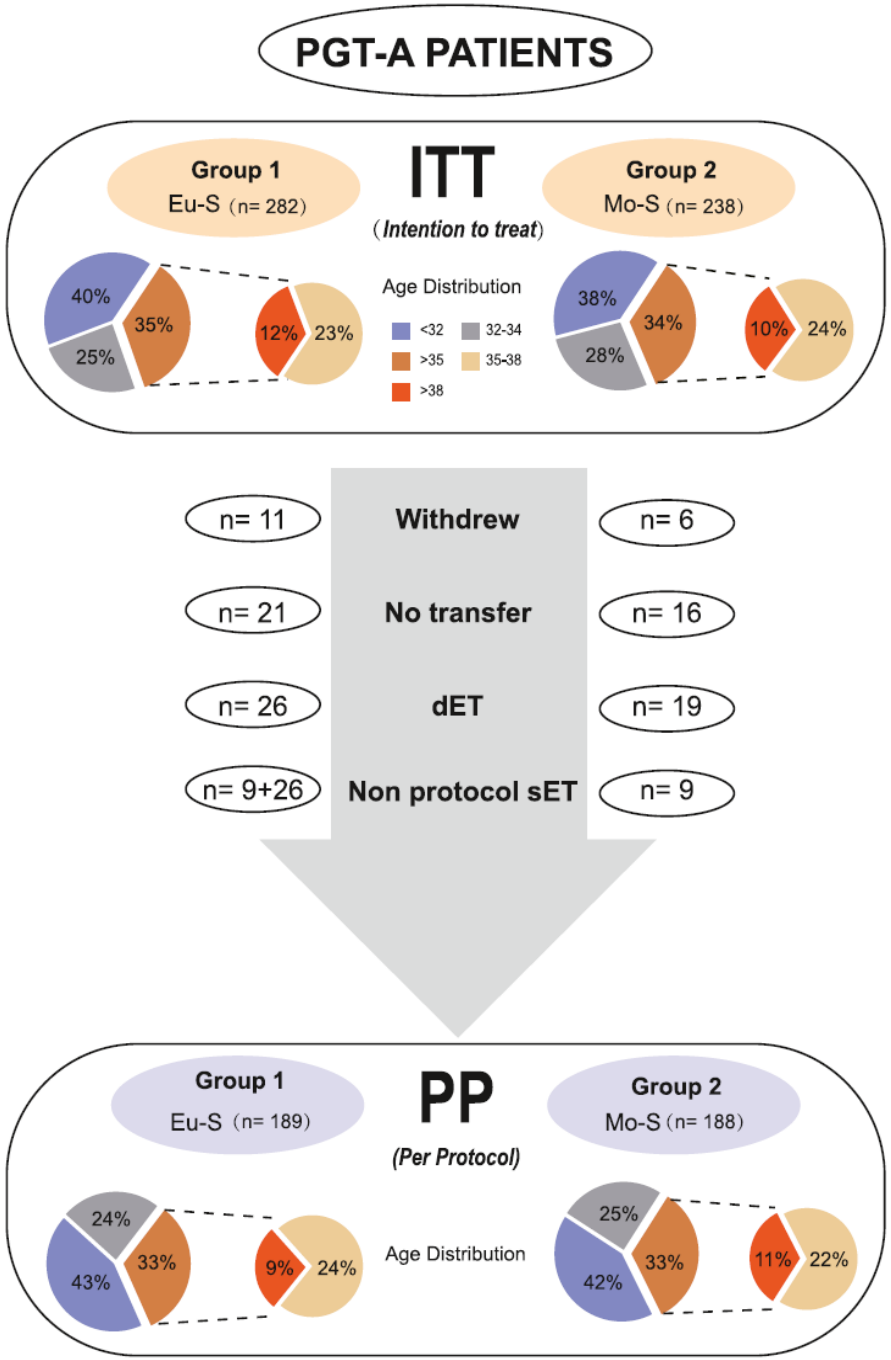
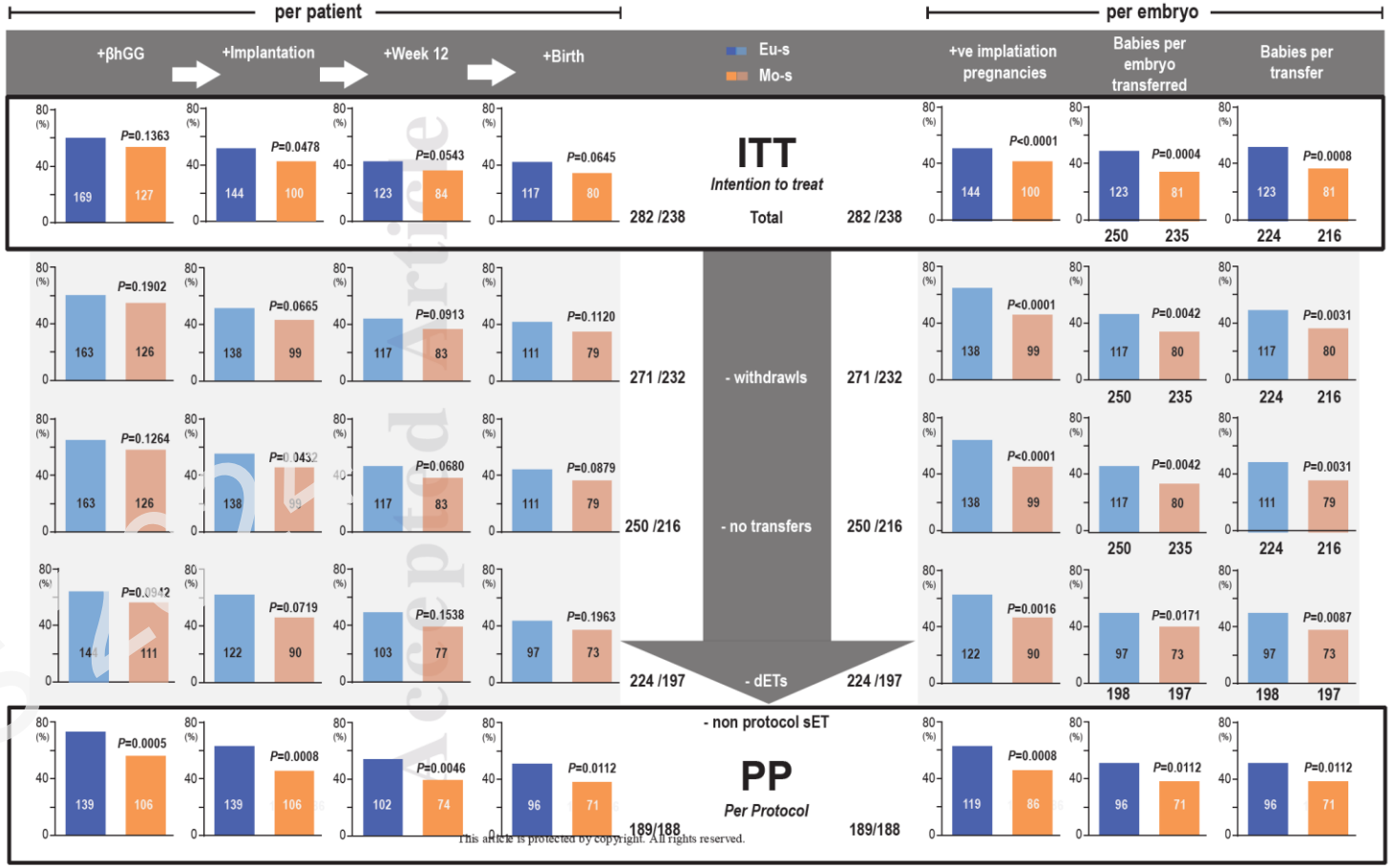
Li Wang, Xiaohong Wang, and Min Li contributed equally to this work.

Study Design

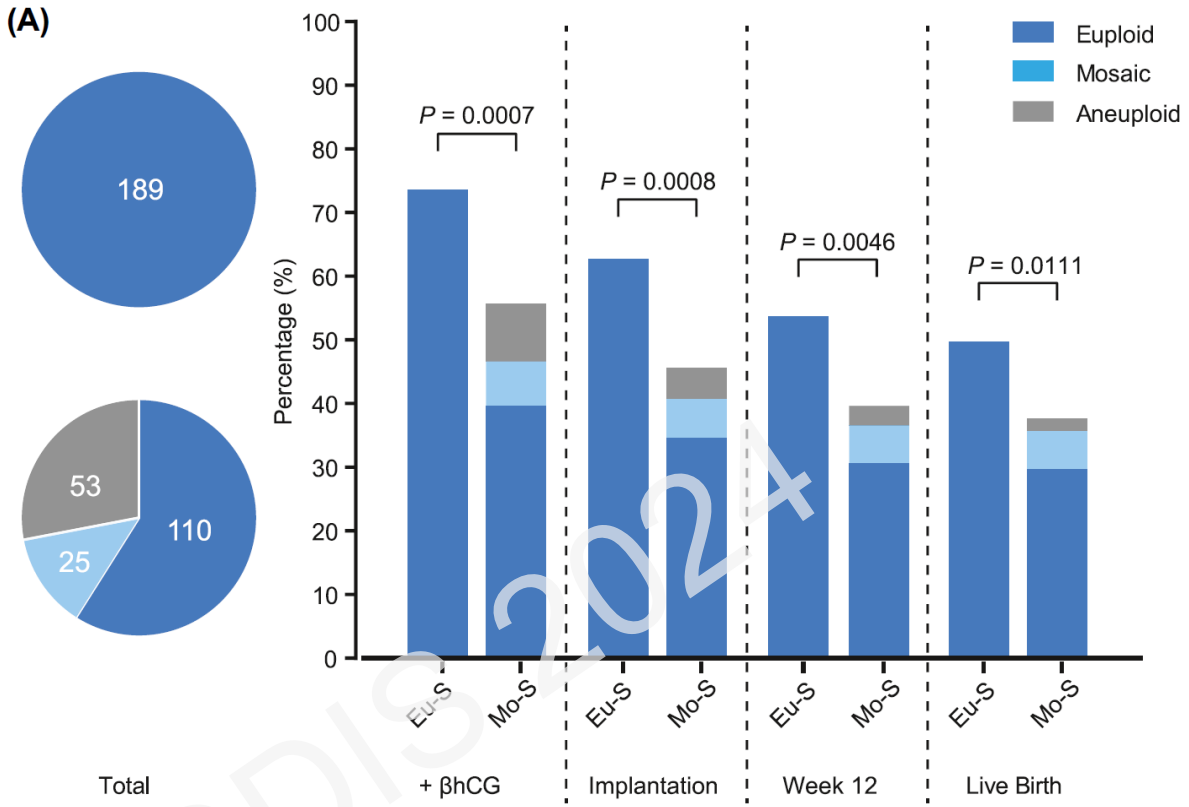
FIGURE 1 Study design. The maternal age distribution for ITT and PP groups is shown. Data for Mo-S group PP has been previously presented in Wang et al.[3] In group 1 Eu-S: 11 withdrew- 6 with natural pregnancies; 47 patients did not proceed with transfer: 15 no transfer yet and 6 had only a single embryo-still to accumulate while 26 patients with no euploids were carried through to the non-protocol transfer category. In group 2 Mo-S: 6 withdrew- 1 with natural pregnancy; 16 patients did not proceed with transfer yet and 3 had only a single embryo-still to accumulate. dET, double embryo transfer; Eu-S, euploid selection; ITT, intention to treat; Mo-S, morphology selection; PP, per protocol; sET, single embryo transfer [Colour figure can be viewed at wileyonlinelibrary.com]



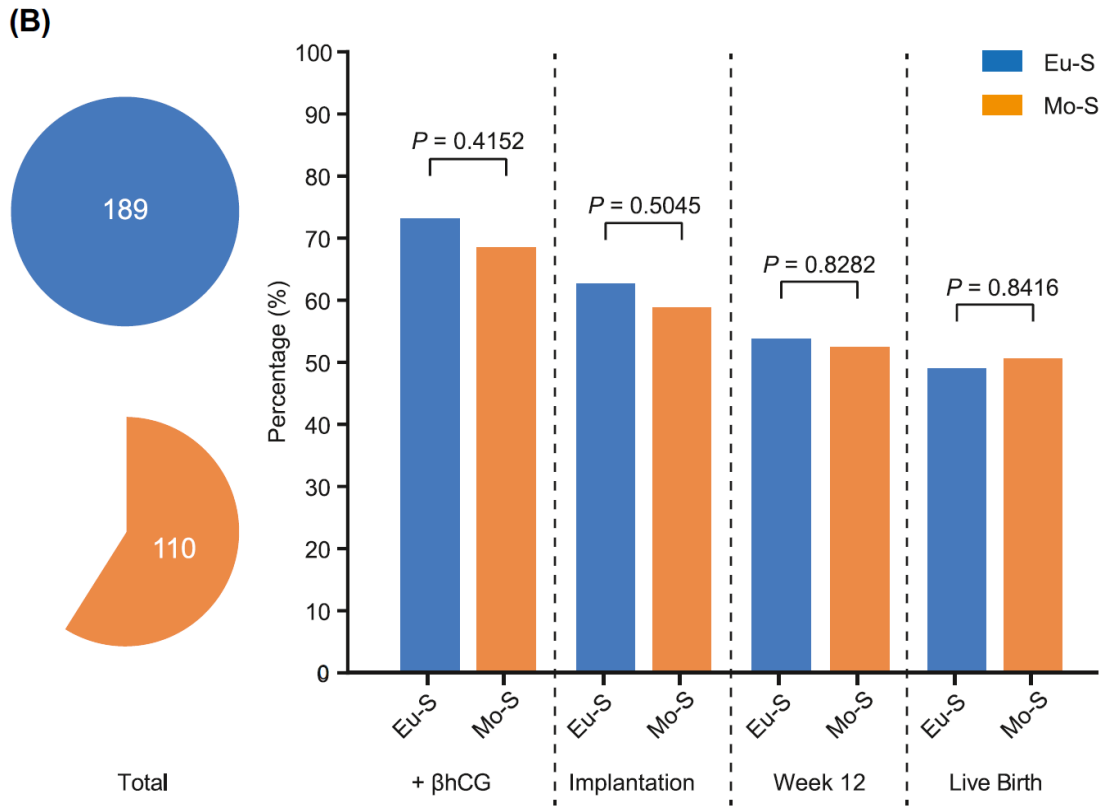
Progressive outcomes for Eu-S and Mo-S groups in transition from ITT to PP analysis



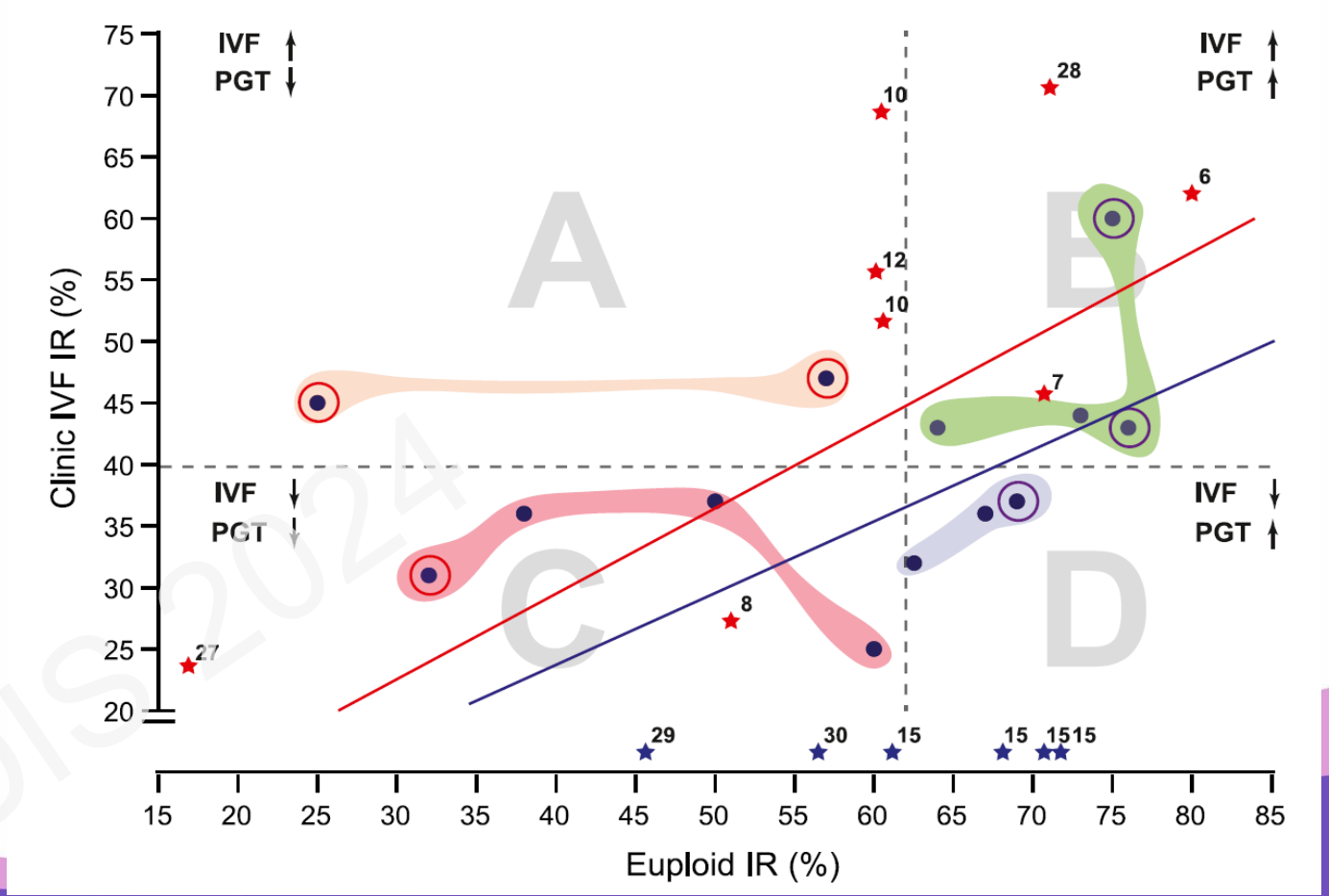
Per Protocol outcomes of single embryo transfers



Transfer outcomes for euploid sET for Eu-S versus Mo-S



Individual clinic implantation rate outcomes for routine IVF and single euploid embryo transfers



Effect of IVF IR on PGT outcome：

$$IR_{expected} = IR_{clinic} / ER_{observed}$$

IVF(IR)	Euploidy rate observed	Predicted PGT IR
30%	59%	0.508475
40%		0.677966
50%		0.847458

Does PGT work?

- Euploidy embryo selection is potentially beneficial for all IVF patients.
- Individual clinic performances play a key role in both routine IVF and successful application of adjunct processes such as PGT-A.
- Our findings question the need for using RCTs in assessing the true value of PGT-A given the more appropriate and more immediate alternatives of clinic self-assessment and non-selection studies.

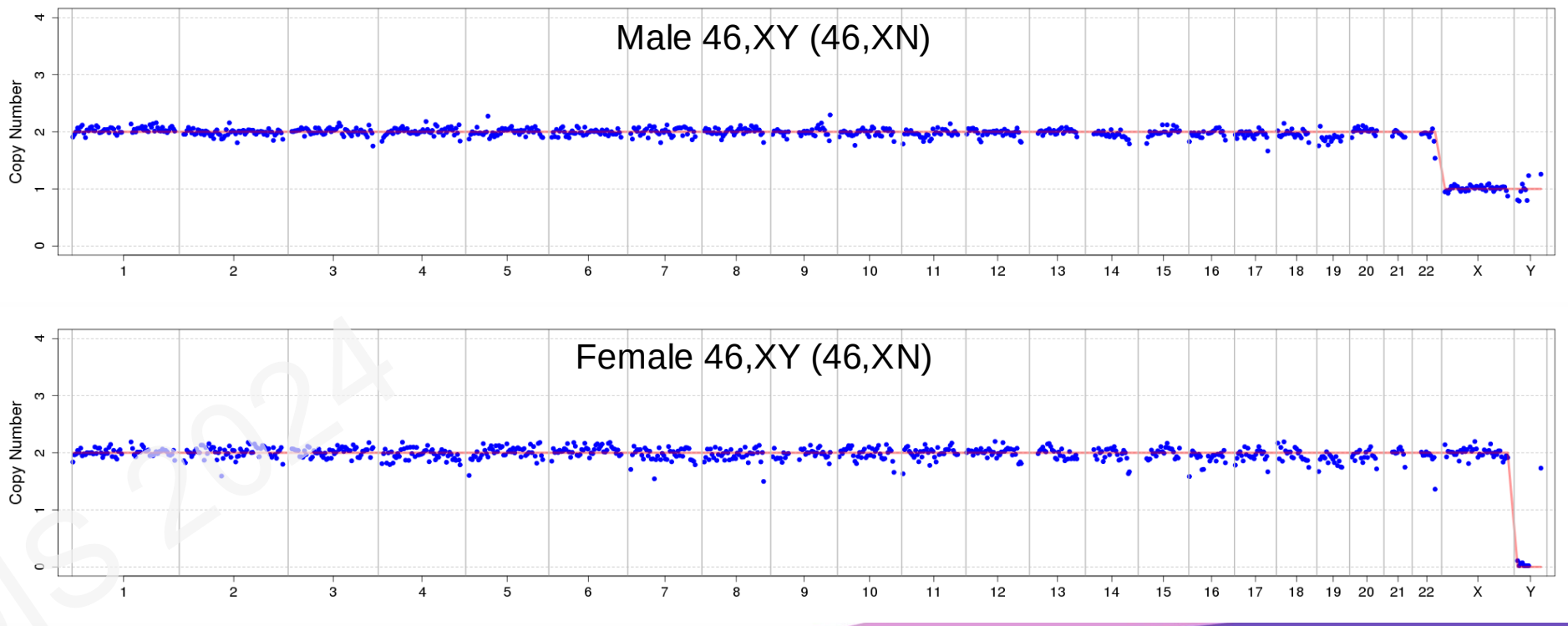
- 3. Mosaic status

Table 1 **CNV-Seq analysis of mosaicism in artificial blastocyst biopsy models**

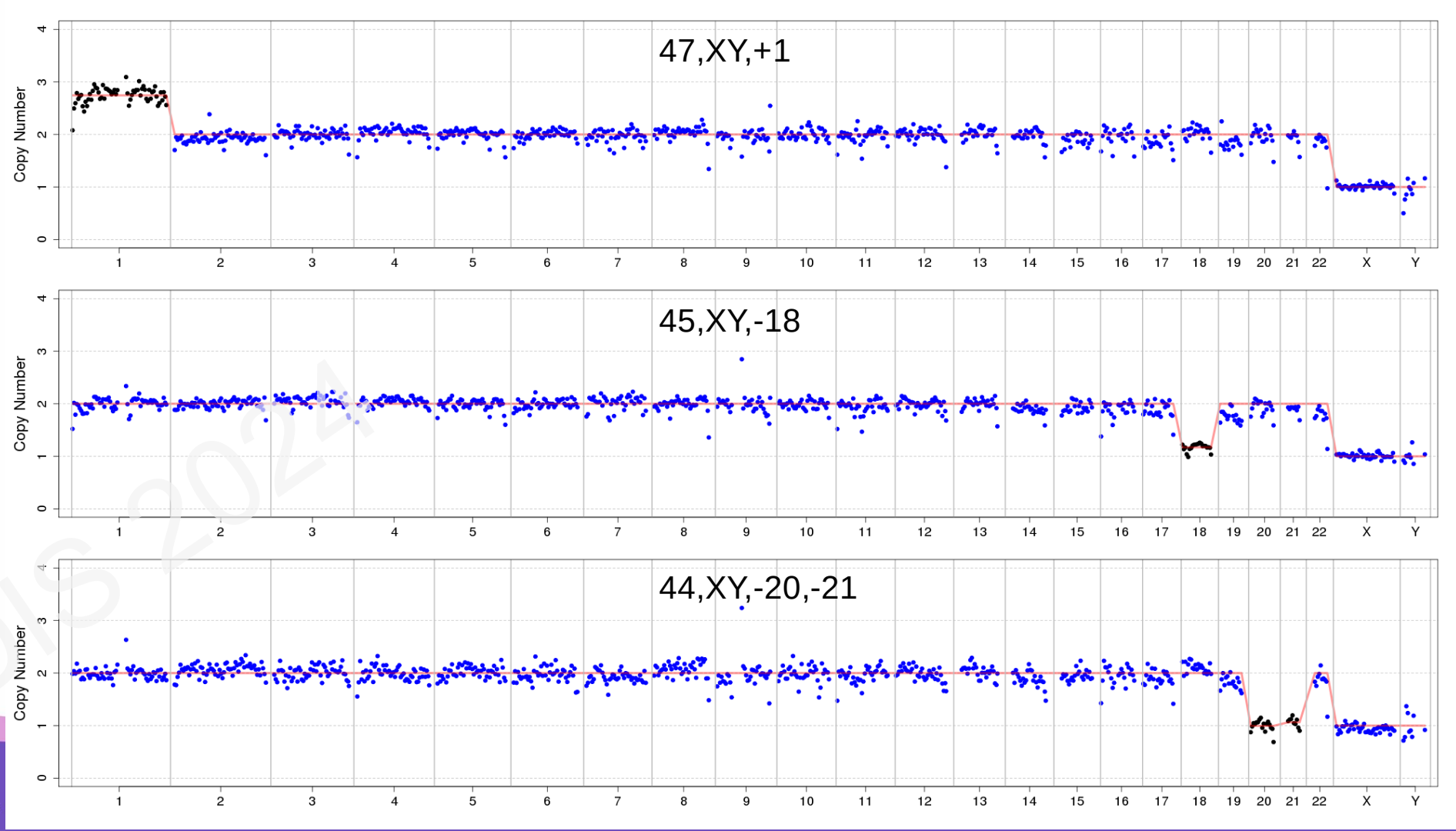
Mosaicism model					Actual levels of mosaicism by CNV-Seq (%) Replicate 1, Replicate 2, Replicate 3 (Replicate Mean)		
47, XY,+21		46,XX	Mosaicism		Trisomy 21	Monosomy X	
10 cells	+	0 cells	=	100%	90%, 87%, 93% (90%)	98%, 99%, 100 (99%)	
5 cells	+	5 cells	=	50%	54%, 54%, 50% (53%)	48%, 53%, 39% (47%)	
47,XX,+16	+	46,XX			Trisomy 16		
50 ng WGA	+	0 ng WGA	=	100%	87%, 88% (88%)		
25 ng WGA	+	25 ng WGA	=	50%	44%, 45%, 43% (44%)		
10 ng WGA	+	40 ng WGA	=	20%	17%, 16%, 17% (17%)		
44,XY,−12,−18	+	46,XX			Monosomy 12	Monosomy 18	Monosomy X
50 ng WGA	+	0 ng WGA	=	100%	95%, 96% (96%)	97%, 97% (97%)	98%, 98% (98%)
25 ng WGA	+	25 ng WGA	=	50%	46%, 46%, 47% (46%)	44%, 46%, 47% (46%)	46%, 47%, 47% (47%)
10 ng WGA	+	40 ng WGA	=	20%	16%, 17%, 17% (17%)	18%, 18%, 18% (18%)	16%, 17%, 17% (17%)

Wang L, et. al., Biol Reprod 2014a
Wang L, et. al., Biol Reprod 2014b
Ruttanajit T, et. al., Prenat Diagn. 2016

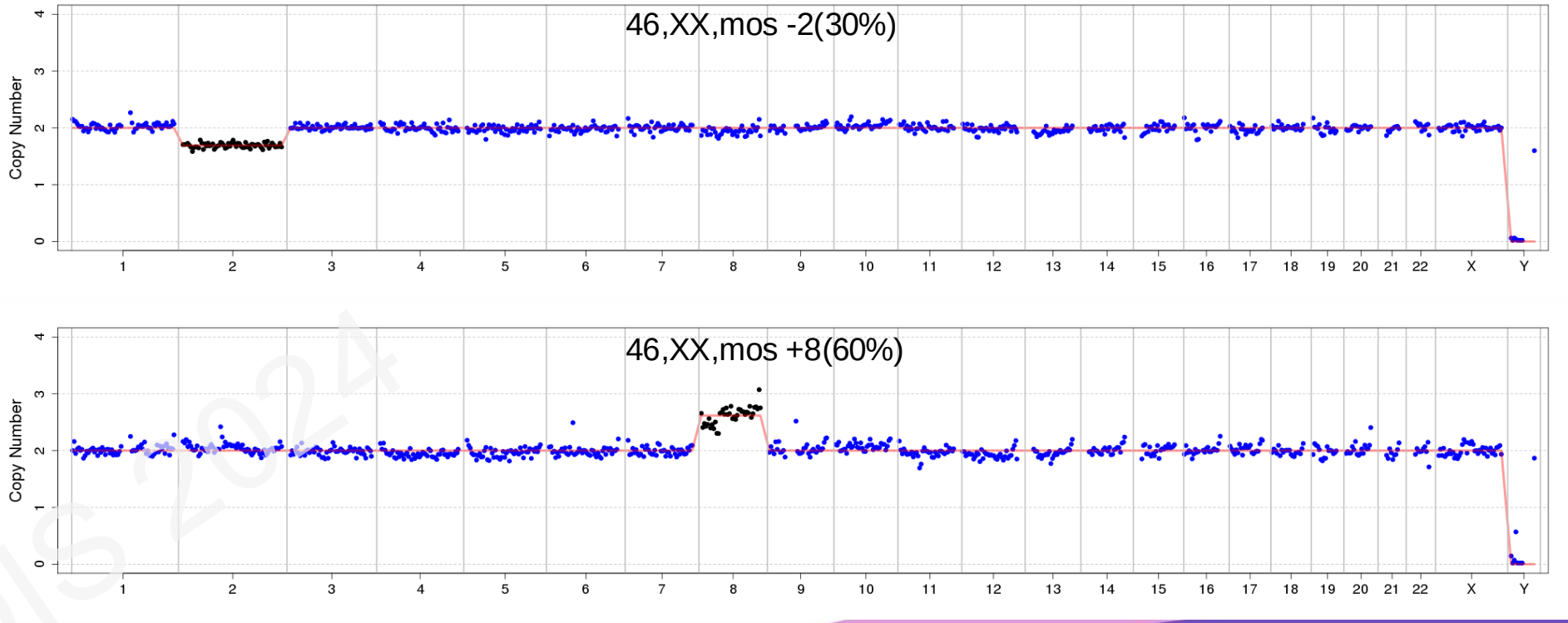
Euploidy (1.8<CN< 2.2)



Aneuploidy
Trisomy CN > 2.8，monosomy CN < 1.2



Mosaic trisomy (2.2<CN<2.8) , mosaic monosomy(1.8>CN>1.2)



Validate cut off value before reporting mosaic status:

- Use both male and female samples
- Use known aneuploidy or fragmental aneuploid DNA samples
- Validating CNV-Seq for measuring chromosome mosaicism, determines the cut-off value:
- - Mix aneuploidy and aneuploid DNA in proportion
- - or euploid and aneuploid cells are mixed proportionally

5G+ cloud-based molecular diagnostic consultation and referral system

- The molecular diagnosis process management cloud system standardizes the whole process of testing, and the traceability system runs through the whole process
- Provide online tools for data analysis to support variant analysis
- Assist with data interpretation
- Consultation and referral for intractable and rare diseases
- Digital sample bank, visual storage, standardized sample management, and efficient molecular diagnosis

Summarry

- Patients should have the right to choose PGT-A
- Patient makes the final decision of (which) embryo to be transferred based on the advice from the clinician and/or genetic counselor.
- To improve the clinical effect of PGT-A, it is necessary to refine whole process management of ART in individual clinic, rather than conducting more RCTs.
- Use artificial mosaic model to validate PGT-A platform and get cut off value of your own.

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