21ST

PGDIS CONFERENCE

PREIMPLANTATION GENETIC DIAGNOSIS INTERNATIONAL SOCIETY

PGDIS

6-8 May 2024

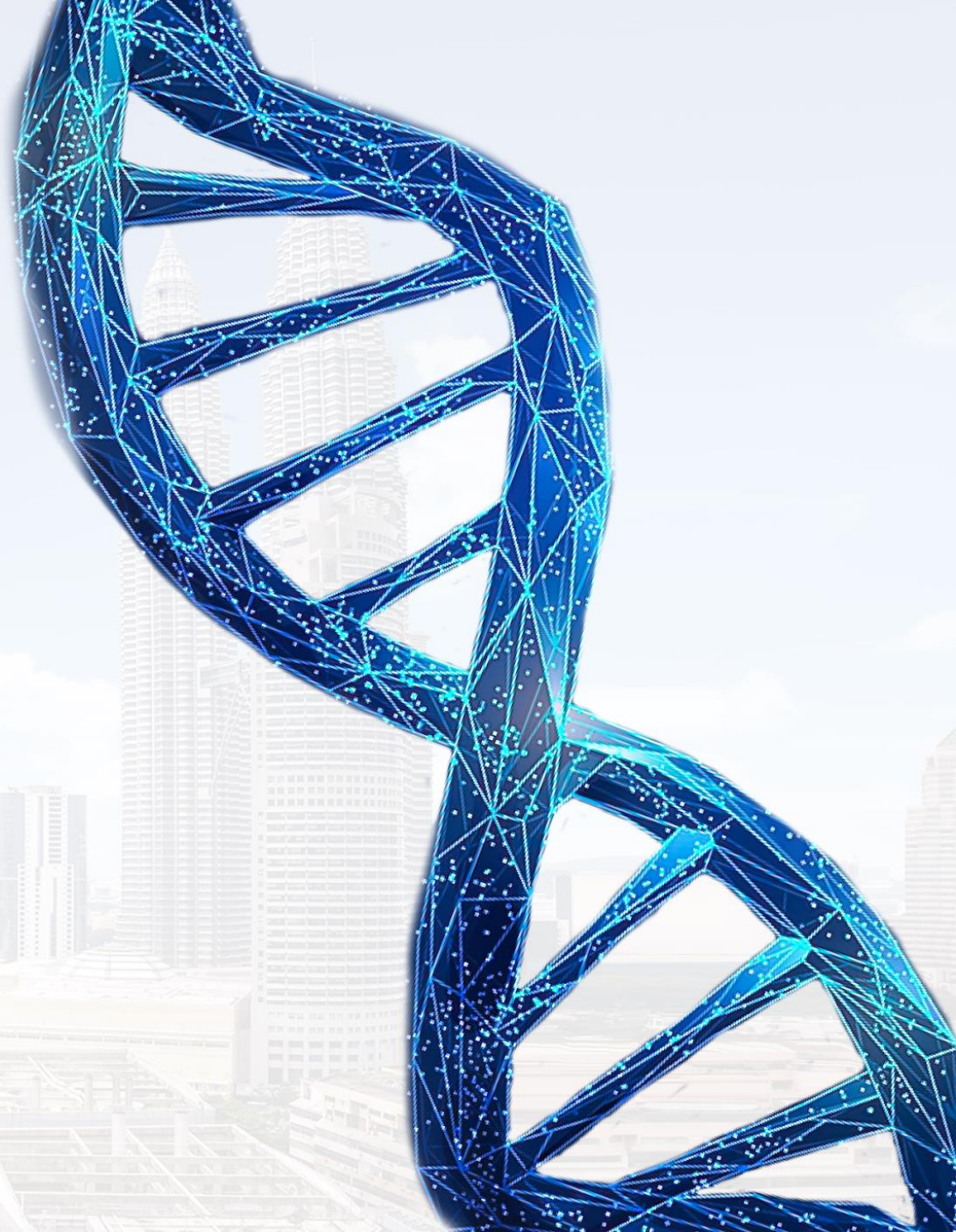
Kuala Lumpur

Malaysia

PGT and BEYOND...

21ST PGDIS CONFERENCE

Noninvasive PGT: 8 years of clinical applications



Sijia Lu

7th, May, 2024

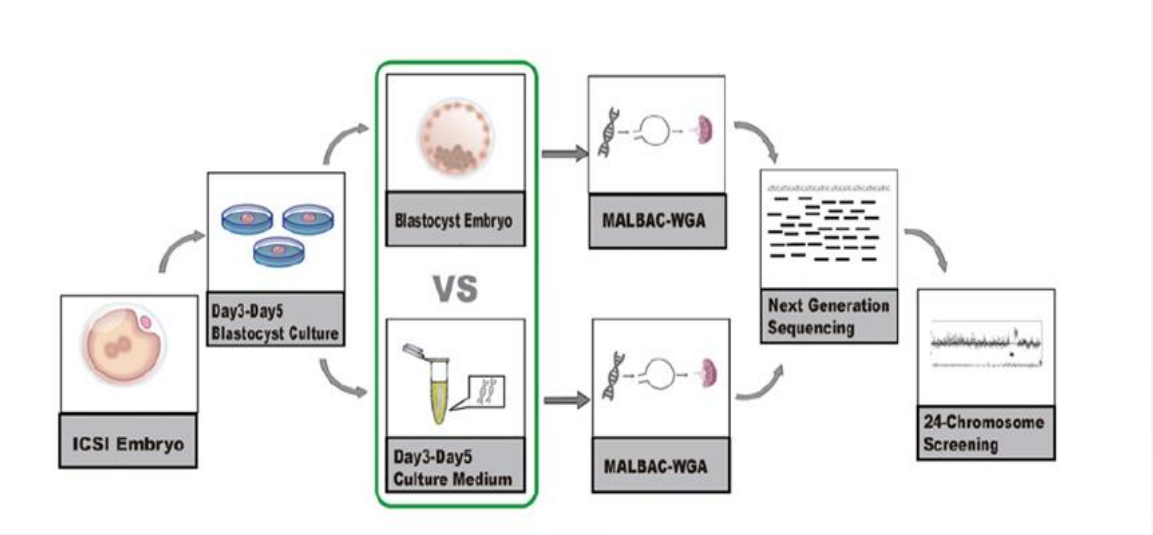
Disclosure

Cofounder and CEO of Yikon Genomics

PGT and BEYOND...

Main Contents

Noninvasive chromosome screening (NICS) Development



NICS Development

Performance	Number	Ratio（%）
Sensitivity	15/17	88.2%
Specificity	21/25	84.0%
PPV	15/19	78.9%
NPV	21/23	91.3%

PPV: pos. predictive value； NPV: neg.predictive value

NICS baby born in Wuxi, China

Table 2. Clinical outcome of the first seven patients subjected to NICS

Patient no.	Maternal age	Clinical indications	Transfer cycles	Clinical outcome
P01	30	Reciprocal translocation 46,XY,t(14;15)	1	Singleton pregnancy—live birth
P02	28	Azoospermia	1	Singleton pregnancy—live birth
P03	34	Inversion 46,XY,inv(9)	1	Singleton pregnancy—live birth
P04	32	Reciprocal translocation 46,XX,t(1;18)	2	Implantation failure
P05	26	Recurrent pregnancy loss	1	Singleton pregnancy—live birth
P06	32	47,XYY	2	Singleton pregnancy—live birth
P07	29	Recurrent implantation failure	1	Singleton pregnancy—following up

Xu J et al. Proc Natl Acad Sci U S A. 2016 Oct 18;113(42):11907-11912.

NICS Accuracy Validations

- **Sample size:**52 donated frozen blastocysts
- **Validation method:** Whole embryo as a gold standard
- **Conclusion:** NiPGT-A is more reliable than TE-biopsy PGT-A in frozen-thawed embryos.

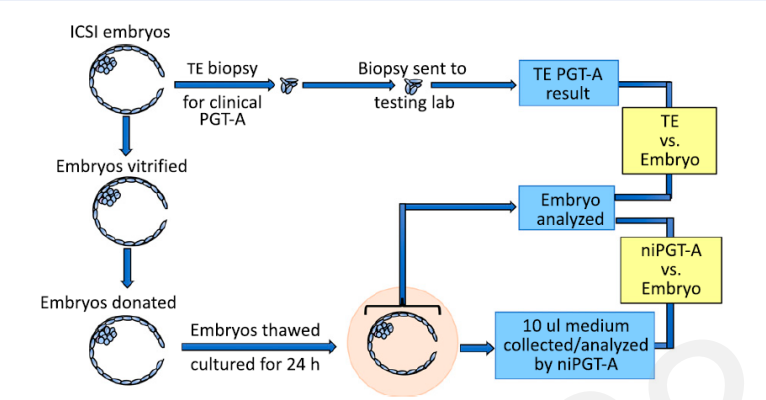


Fig. 2. Workflow of sample processing for PGT-A analysis of TE biopsy, embryo, and spent culture media.

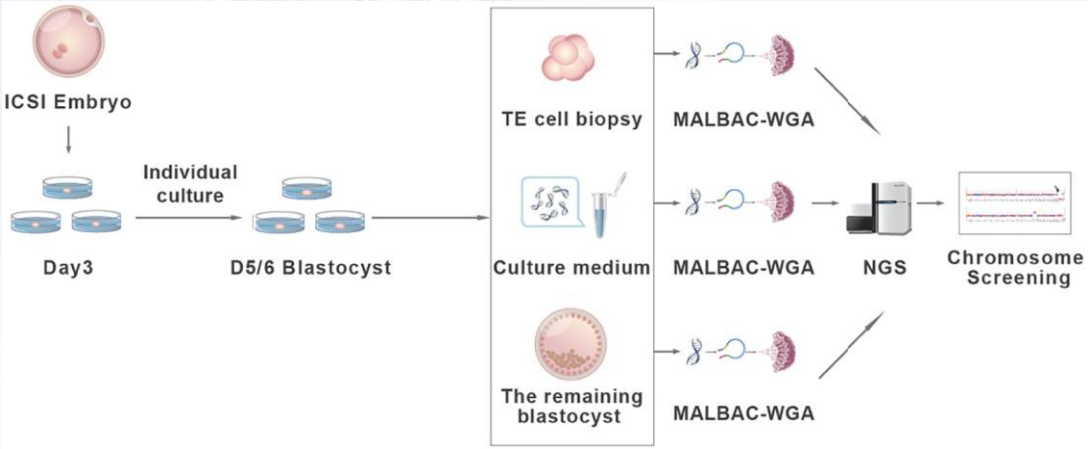
Table 2. Comparison of the performance of niPGT-A versus TE biopsy for PGT-A

Performance characteristic	niPGT-A (n = 48)	TE-biopsy (n = 50)
FPR	20.0% (3/15)	50.0% (9/18)
FNR	0.0% (0/33)	0.0% (0/32)
PPV	91.7% (33/36)	78.0% (32/41)
NPV	100.0% (15/15)	100.0% (18/18)
Sensitivity	100.0% (33/33)	100.0% (32/32)
Specificity	80.0% (12/15)	50.0% (9/18)
% Concordance for embryo ploidy	93.8% (45/48)	82.0% (41/50)
% Concordance for chromosome CNs	83.3% (40/48)	62.0% (31/50)

niPGT-A and TE biopsy results were compared with those of the embryo. Sequencing threshold was set at 60% mosaicism.

Huang L, et al. Proc Natl Acad Sci U S A. 2019 Jul 9;116(28):14105-14112.

- **Sample size:**265 donated embryos
- **Validation method:** Whole embryo as a gold standard
- **Conclusion: Accuracy of NICS** comparable to TE-PGT



Assay	Sensitivity % (95% CI)	Specificity% (95% CI)	NPV (95% CI)	PPV (95% CI)
TE-PGT	89.6(81.9-94.2)	80.0(73.1-85.5)	92.8(87.2-96.0)	72.9(64.2-80.1)
NICS	86.5(78.2-91.9)	73.1(65.8-79.4)	90.0(83.6-94.1)	65.9(57.2-73.6)
P	0.6291	0.1524	0.5144	0.2677

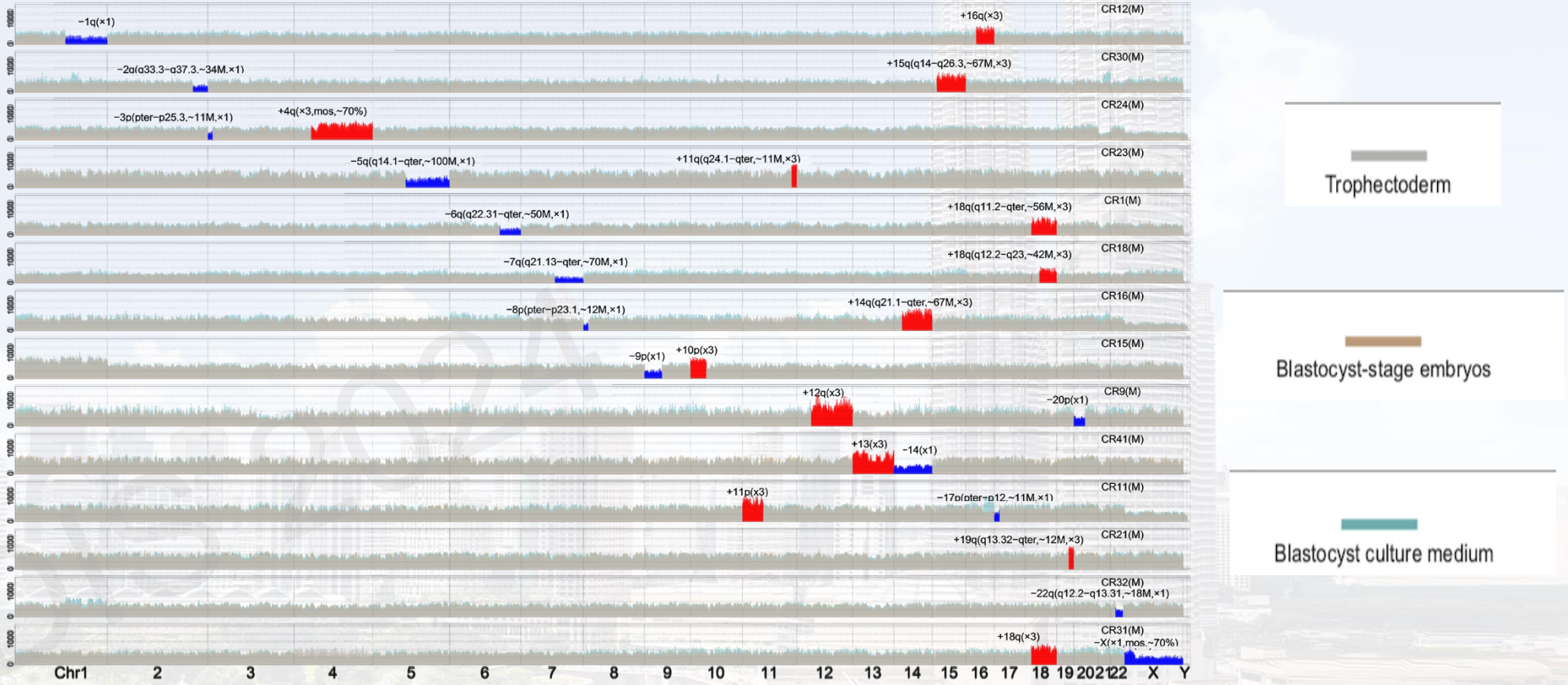
Chen L et al. Front. Cell Dev. Biol9:708322.

NICS Accuracy Validations

PGT-SR

- **Sample size:**41 frozen donated blastocysts
- **Validation method:** Whole embryo as a gold standard

NICS CNV are highly concordant with biopsied based PGT-SR



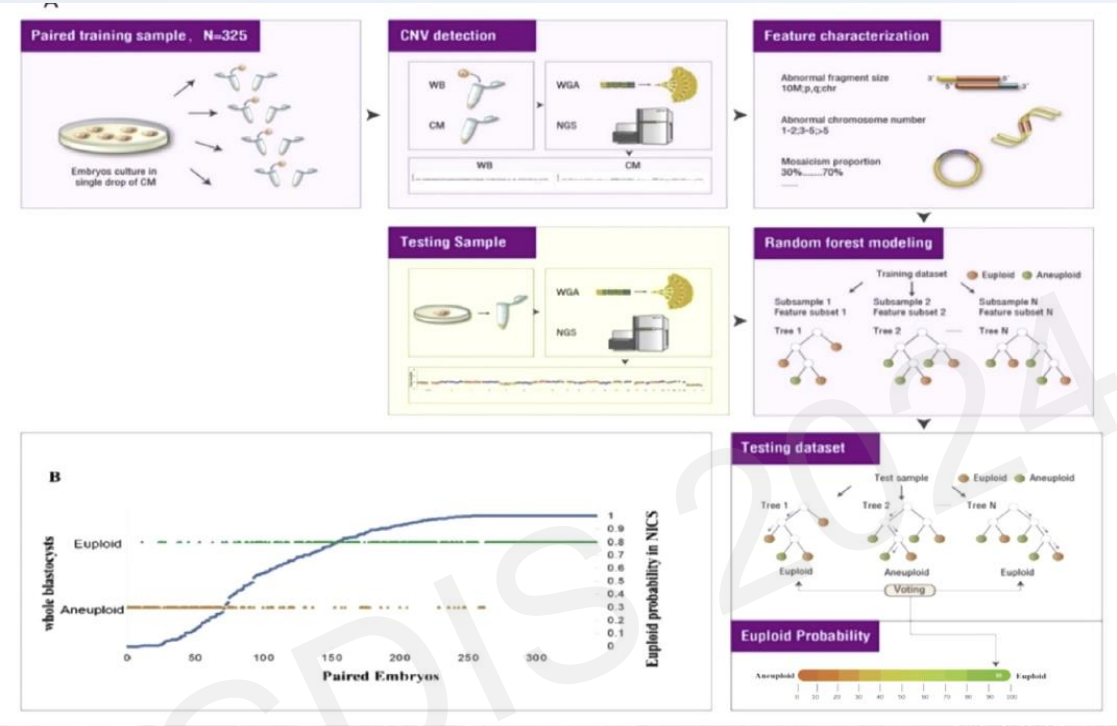
Comparison of results from TE and SCM to embryos: red highlights the duplication and blue highlights the deletion related to translocation.

Jiao J., et al., Hum Reprod. 2019 Jul 8;34(7):1369-1379.

NICS Accuracy Validations

NICS AI Grading Strategy

- 345 paired blastocyst culture medium and whole blastocyst samples
- Embryos were graded as A, B or C according to their euploidy probability levels



NICS AI grading system using the machine learning method.

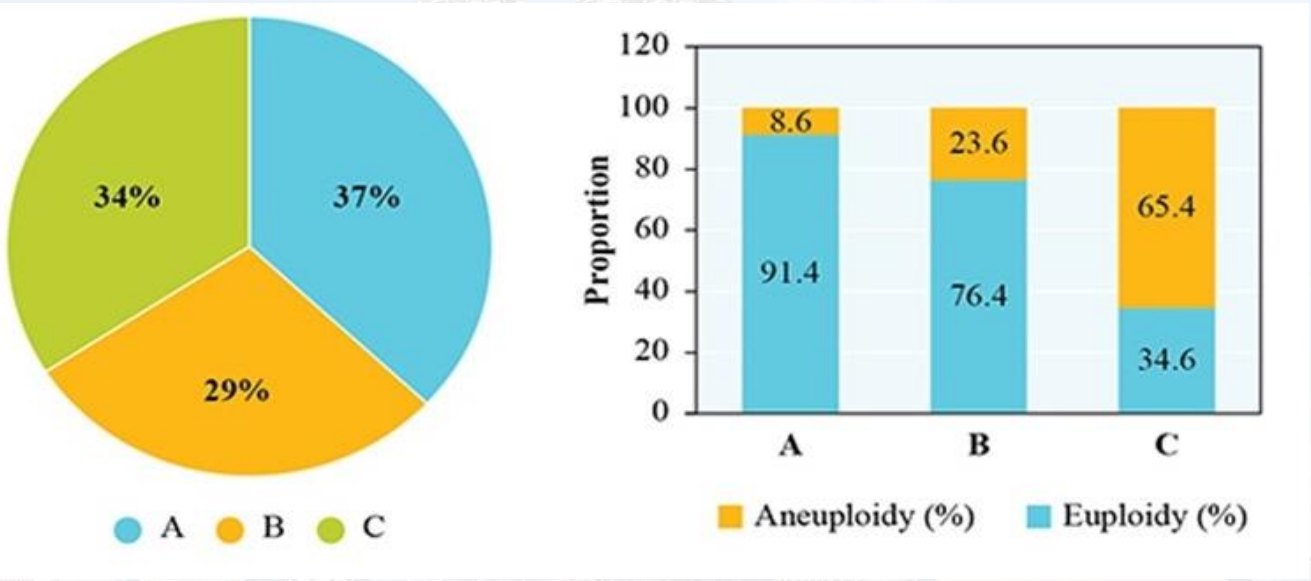


Figure A： NICS grading distribution of grade A, B and C

Figure B: Grade A 91.4% Euploid; Grade B 76.4% Euploid ;
Grade C 34.6% Euploid

Chen L et al. Reprod Biomed Online. 2022 Jul;45(1):26-34.

Clinical Studies and Applications

RPL and RIF patients

Patients with RPL (≥3 times) or RIF (≥3 times).

A total of 52 euploid embryos were transferred in 43 RPL or RIF patients.

Table 3 Pregnancy outcomes with NICS

	Chromosomal rearrangement	Normal karyotype	Total
Total ET			
Cycles	25	25	50
Patients	22	21	43
(SET/DET)	25/0	23/2	48/2
Transferred euploid blastocysts	25	27	52
Biochemical pregnancies	68% (17/25)	76% (19/25)	72.0% (36/50)
Clinical pregnancies	52% (13/25)	64% (16/25)	58.0% (29/50)
Miscarriages	15.4% (2/13)	6.2% (1/16)	10.3% (3/29)
Deliveries	11	15	26
Singleton/twins	11/0	14/1	25/1
Babies born (male/female)	11 (6/5)	16 (9/7)	27 (15/12)
Birth weight (g, mean ± SD)	3283.7 ± 412.4	3174.7 ± 391.5	3217.5 ± 403.4

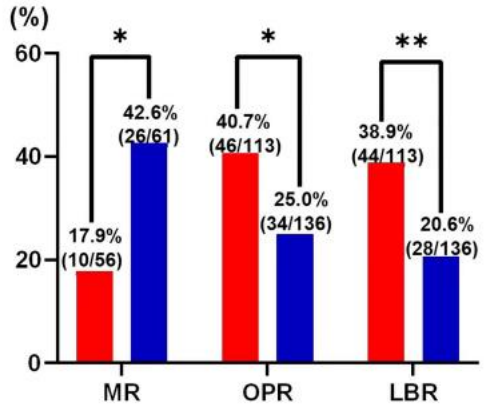
DET, double embryos transfer; ET, embryo transfer; NICS, non-invasive chromosome screening; SET, single embryo transfer

A total of 27 healthy babies were delivered.

Fang R., et al., J Transl Med. 2019 Mar 8;17(1):73.

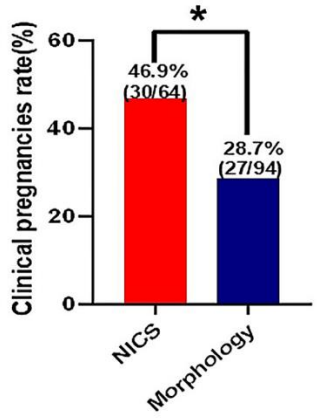
Patients with subclinical RPL and RIF

A total of 273 women with a history of RPL or RIF



Patients with RPL≥2 times or once with abnormal product of conception.

Reduce the incidence of miscarriage
Increase ongoing pregnancy rate



Patients with RIF≥2 times

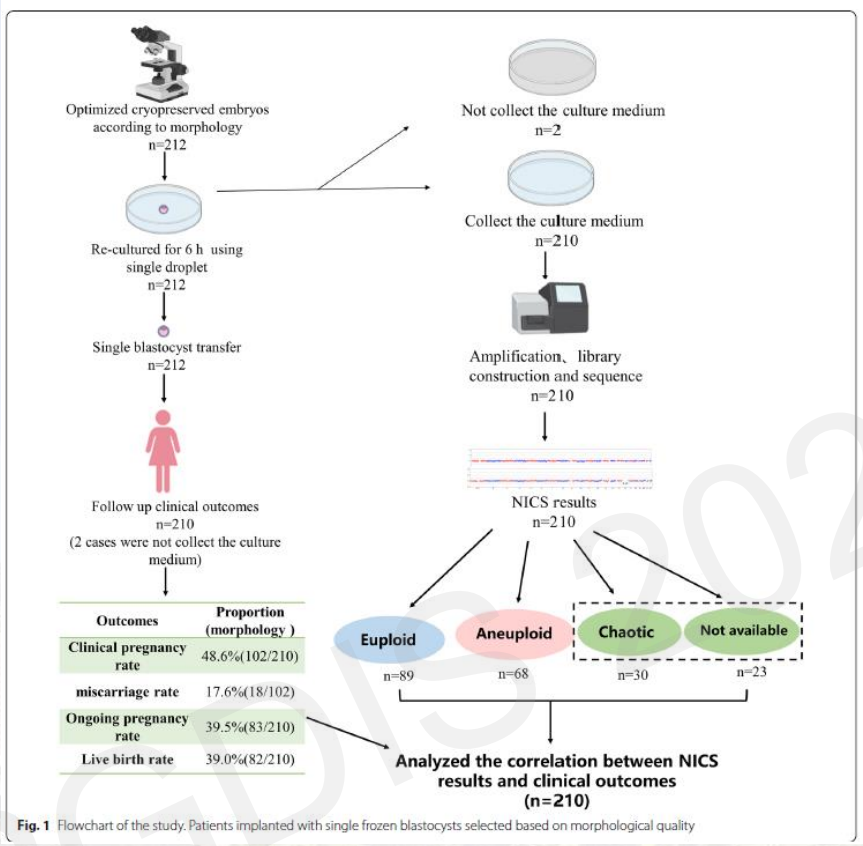
Increase the clinical pregnancy rate

Xi H., et al., Front Endocrinol . 13:896357.

Clinical Study and Applications

Frozen-thawed cycles

- 212 frozen-thawed single-blastocyst transfers based on morphological grades
- SCM collection in preincubation for 6 h after thawing



Outcomes	Proportion (morphology)
Clinical pregnancy rate	48.6%(102/210)
miscarriage rate	17.6%(18/102)
Ongoing pregnancy rate	39.5%(83/210)
Live birth rate	39.0%(82/210)

Fig. 1 Flowchart of the study. Patients implanted with single frozen blastocysts selected based on morphological quality

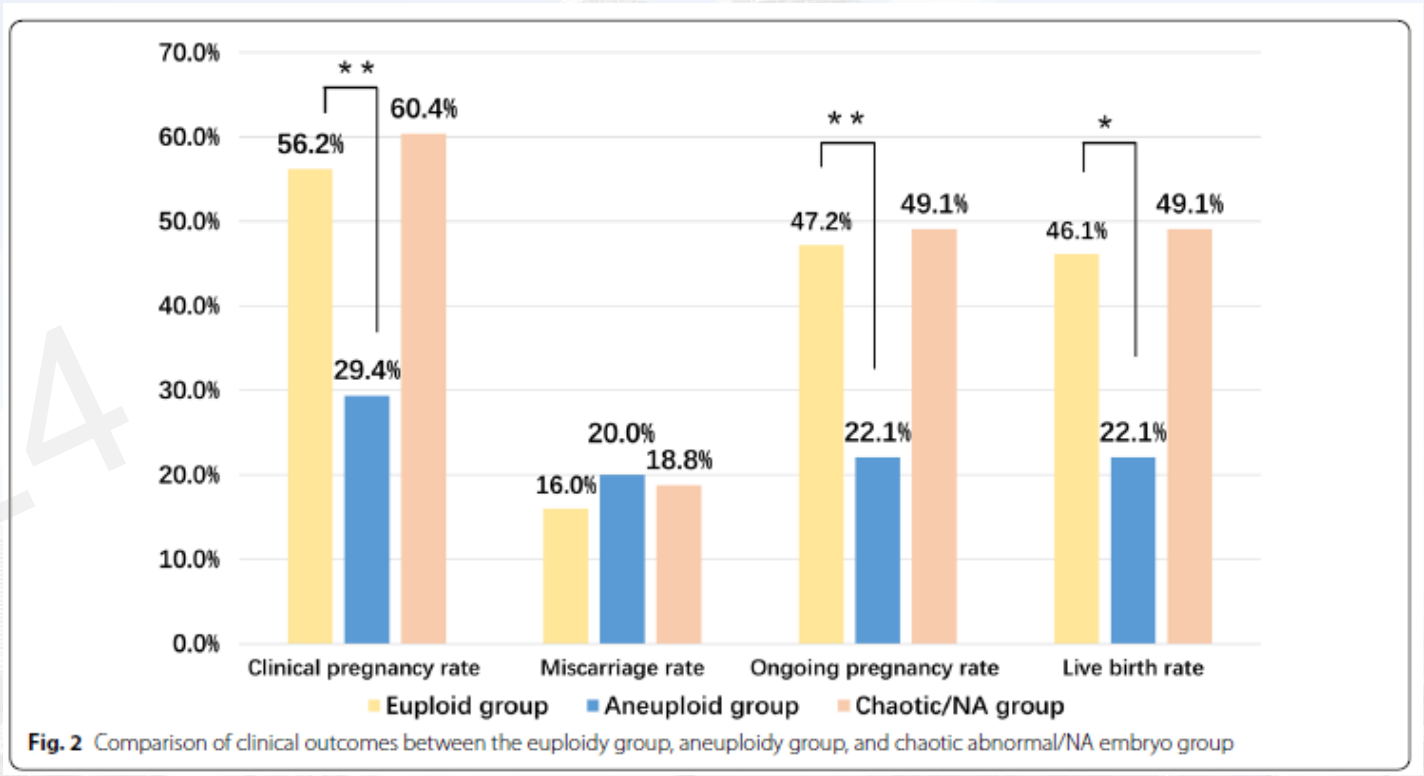


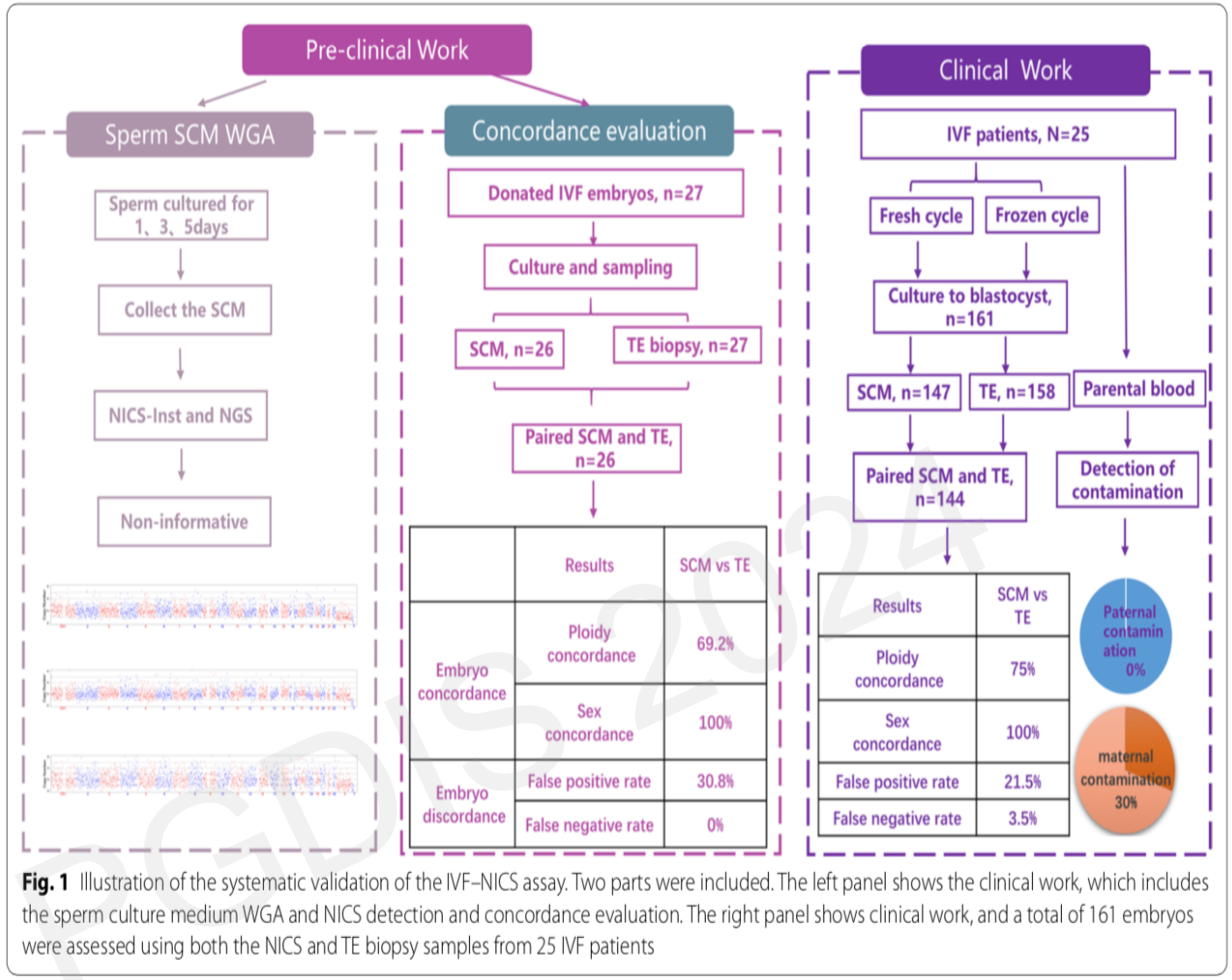
Fig. 2 Comparison of clinical outcomes between the euploidy group, aneuploidy group, and chaotic abnormal/NA embryo group

The pregnancy rates were significantly higher in both the euploidy and N/A groups compared to the aneuploidy group.

Chen R et al. J Transl Med. 2022 Dec 3;20(1):553.

Clinical Study and Applications

Conventional IVF



	Results	SCM vs TE
Embryo concordance	Ploidy concordance	69.2%
	Sex concordance	100%
Embryo discordance	False positive rate	30.8%
	False negative rate	0%

Results	SCM vs TE
Ploidy concordance	75%
Sex concordance	100%
False positive rate	21.5%
False negative rate	3.5%

Fig. 1 Illustration of the systematic validation of the IVF–NICS assay. Two parts were included. The left panel shows the clinical work, which includes the sperm culture medium WGA and NICS detection and concordance evaluation. The right panel shows clinical work, and a total of 161 embryos were assessed using both the NICS and TE biopsy samples from 25 IVF patients

- Failed sperm DNA amplification in the current amplification system
- No paternal contamination was observed in conventional IVF SCM.
- IVF NICS performances ≈ ICSI.

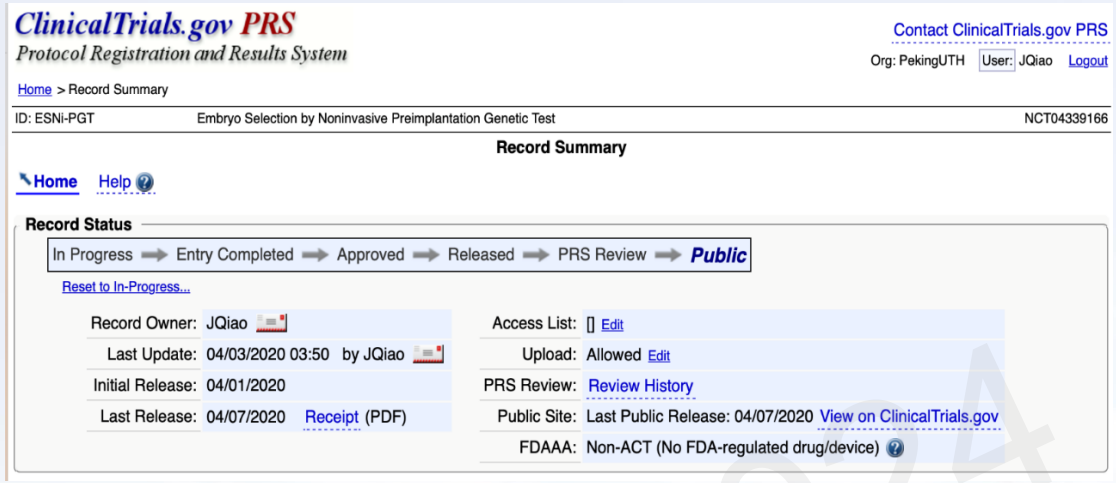
	Concordance	Sensitivity	Specificity	PPV	NPV
IVF-Adjusted	75%(108/144)	91.38%(53/58)	63.95%(55/86)	63.1%(53/84)	91.67%(55/60)
ICSI-Adjusted	74.58%(88/118)	84.21%(48/57)	65.57%(40/61)	69.57%(48/69)	81.63%(40/49)
P-Value	0.94	0.24	0.84	0.40	0.12

Xie P., et al., J Transl Med. 2022 Sep 4;20(1):396.

Clinical Study and Applications

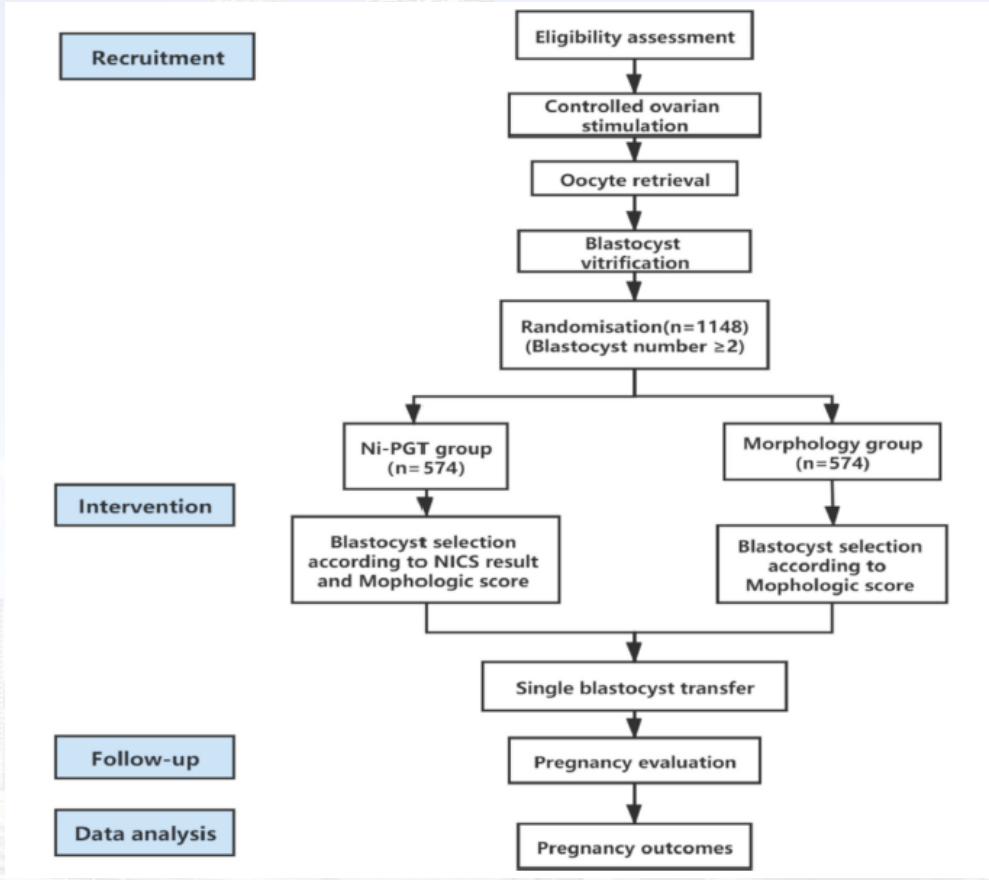
Embryo Transfer Prioritization

- ESNi-PGT (Embryo Selection by Noninvasive Preimplantation Genetic Test，NCT04339166)



1148 couples aged 35~42 (women) are planned to be enrolled from 13 IVF centers in China mainland.

finished recruting patients in 2023
result unblinded April 2024.



A Double-blind, randomised controlled trial

Huang J, et al. BMJ Open 2022;12:e057254.

Clinical Usage/Benefits

- Clinical NICS usage: >50000 tests from ~230 centers in the past 5 years as a supplement to PGTA.
- For all IVF cycles? Probably not for now.
- For patients with increased risk of embryo anneuploidy (in China, ≥2 failed implantations and ≥1 miscarriage, or >35 maternal age)
- Reanalyze frozen embryos (D5 thawed for 6-8 hours, <5% no call, >93% concordance)
- For embryos with poor morphology
- Embryo biopsy limited by expertise or regulation.

Limitations and Future directions

- Accuracy: Susceptible to sample contamination (cumulus, polar body, external contaminations): recommend to check SNP, especially for embryos with euploid results.
  NOT a diagnostic test as of now.

- Sensitivity: D5 no call rate: ~10-15%, ~80% concordance, D6 embryos are more accurate, <5% no call rate, ~90% concordance... But should D5 embryos be cultured to D6?

- Standardization: automation

- PGTM and PGTSR?

- Need more research on the origin of contamination. (methylation? SNP? transcriptomics?)

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- *Dr. Wang Xiuxia of Shengjing Hospital, Shenyang*
- ……

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THANK YOU

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