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*Original Research*

# Clinical Application of Preconception Expanded Carrier Screening in Assisted Reproductive Technology Populations in Central China

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**Abstract**

**Background**: To investigate the clinical application of preconception expanded carrier screening (PECS) in couples undergoing assisted reproductive technology (ART) in Central China. **Methods**: We conducted a retrospective study at the Reproductive Medicine Center of Shiyan Renmin Hospital from August 2022 to November 2023. PECS results from infertile couples were analyzed to determine carrier rates for pathogenic single-gene variants and the proportion of at-risk couples (ARCs). Identified ARCs received genetic counseling and reproductive guidance, and pregnancy outcomes were assessed. **Results**: A total of 4853 patients (2082 couples) underwent PECS for 22 monogenic diseases. Among them, 81.58% (3959/4853) carried no pathogenic variants, whereas 18.42% (894/4853) carried at least one variant. The carrier rates for one, two, and three variants were 16.73%, 1.61%, and 0.08%, respectively. The genes with the highest carrier frequencies were gap junction protein beta 2 (*GJB2*) (3.05%), cytochrome P450 family 21 subfamily A member 2 (*CYP21A2*) (2.64%), hemoglobin subunit alpha 1/2 (*HBA1/HBA2*) (2.12%), survival of motor neuron 1 (*SMN1*) (2.12%), and ATPase copper transporting beta (*ATP7B*) (2.10%). ARCs were identified in 2.31% (48/2082) of couples. Of these, 1 couple chose preimplantation genetic testing for monogenic disorders (PGT-M), and another opted for preimplantation genetic testing for aneuploidy (PGT-A). 26 couples proceeded with routine ART, which resulted in 14 live births. Outcomes among the remaining ARCs included 4 canceled embryo transfer cycles, 3 failed pregnancies, 3 early miscarriages, and 2 elective terminations. **Conclusions**: PECS provides a valuable approach to identify single-gene disease risks in the infertile population of Central China. It facilitates informed reproductive decision-making and supports the prevention of affected offspring through targeted prenatal diagnosis.

**Keywords:** assisted reproductive technology; carrier screening; prenatal diagnosis; preimplantation genetic testing; genetic counseling

## 1. Introduction

Currently, more than 7000 monogenic diseases have been identified, with a collective incidence exceeding 1 in 100 [1,2]. Many of these conditions are fatal, teratogenic, or disabling, exhibit complex inheritance patterns, and present diverse clinical manifestations. Fewer than 5% have effective treatments, most of which entail high costs. Consequently, strategies to reduce the birth of children affected by monogenic diseases represent a significant public health priority [3]. Carrier screening tests asymptomatic individuals to identify pathogenic (P) or likely pathogenic (LP) variants in genes associated with common autosomal recessive (AR) disorders and represents an established strategy to prevent the transmission of these diseases [4].

High-risk couples, often termed at-risk couples (ARCs), are traditionally defined as those in which both partners carry P or LP variants (P/LP) in the same AR gene or in which the woman is a carrier of an X-linked (XL) condition [5,6]. In this study, the definition was extended to include couples in which the female partner carries a mitochondrial disorder or either partner carries pathogenic chromosomal microdeletions or microduplications. These couples face a substantial risk of having offspring with severe genetic disorders. Preconception expanded carrier screening (PECS) enables risk detection prior to pregnancy. Identified ARCs can subsequently receive personalized reproductive guidance through professional genetic counseling. From a health-economic perspective, PECS offers a favorable balance between screening yield and cost, thereby helping minimize the risk of genetic disease in offspring [7,8].

Carrier screening is estimated to prevent approximately 1.25% of birth defects. For instance, Franasiak et al. [9] reported that, after screening for 117 diseases, expanded carrier screening (ECS) results influenced the reproductive decisions of 0.21% of patients. PECS tests phenotypically normal individuals to identify specific genetic disease-related variants they may carry. [10] This process

clarifies individual genetic risk and helps estimate the corresponding risk for future offspring. Consequently, it provides opportunities for early intervention, treatment planning, and informed reproductive decision-making. Accordingly, PECS is recognized as a primary preventive measure against birth defects [11,12].

The American College of Medical Genetics and Genomics (ACMG) recommends screening for AR diseases with a carrier frequency $\geq$1/200 and for XL diseases with an incidence $\geq$1/40,000 among individuals planning pregnancy. However, existing data are limited. Indeed, carrier frequencies for many severe diseases fall below this threshold, and disease incidence varies considerably across populations. In particular, research on the prevalence and carrier frequency of monogenic diseases in the Chinese population remains insufficient [13,14]. Moreover, the widespread implementation of PECS faces several obstacles globally and in China. The first major barrier is cost and accessibility. In most regions of China, PECS is not covered by medical insurance, requiring out-of-pocket payment. This contributes to a low screening acceptance rate of only 35.83%, far below the over 90% observed in Western countries [15]. The lack of national infrastructure and policy support further limits availability to a limited number of medical centers. Second, technical and interpretation challenges exist. Current PECS panels often fail to cover non-coding regions and copy number variations (CNVs), and variants of uncertain significance (VUS) are excluded from risk assessments [16]. For genes such as survival of motor neuron 1 (*SMN1*), in which CNVs represent the primary pathogenic mechanism, technical limitations may restrict detection to single nucleotide variants (SNVs), leading to missed diagnoses [17,18]. Additionally, the inability to identify "high-risk carrier" configurations (e.g., two *SMN1* copies on the same chromosome) introduces a false-negative risk estimated at 0.051% [19]. A third challenge is the lack of uniformity in laboratory standards and panel designs. ECS panel compositions vary significantly worldwide; one report noted that only three diseases overlapped across 16 different laboratories [20]. In China, disparities in detection platforms and panel design across regions can bias comparisons of carries rates. Furthermore, substantial regional differences in carrier rates exist within China. For example, methylmalonic aciduria and phenylketonuria (PKU) are more common in the north, whereas *G6PD* deficiency and thalassemia are more prevalent in the south [18], which makes universal panels suboptimal for regional genetic landscapes [21]. Finally, public a nd clinical awareness is insufficient. Surveys indicate that 34.46% of Chinese respondents have never heard of ECS, and only 19.94% understand its significance. Many patients decline screening because they assume that the absence of family history indicates low or no genetic risk, while some partners refuse testing due to concerns about discrimination or doubts about result accuracy [22]. Therefore, the determination of an appropriate screening scope requires careful consideration of regional differences in carrier frequency. Carrier rates and ARC proportions are directly influenced by the size and composition of the ECS panel. Current panels do not comprehensively capture the spectrum of genetic variation across all regions, which suggests a need for customized panels tailored to specific populations. Guided by these principles, and in consideration of the scope of the study, data availability, technical feasibility, and cost-effectiveness, we selected 22 diseases for our initial research design. Future studies may expand to include additional diseases and genetic markers, particularly those with higher prevalence in target populations. This study aimed to characterize the disease spectrum and assess the optimal ECS panel size for infertile populations in central and western China.

## 2. Materials and Methods

### *2.1 Study Subjects*

This retrospective study included 4853 patients who underwent assisted reproductive technology (ART) at our center between August 2022 and November 2023. The cohort comprised 2733 females and 2120 males. Among them, 689 patients were analyzed individually, whereas the remaining 4164 patients, comprising 2082 couples, were included in the couple-based analysis. All patients were informed of the purpose and significance of preconception carrier screening during preoperative assessment and provided written informed consent prior to testing.

### *2.2 Study Methods*

Inclusion criteria: Participants were enrolled in accordance with relevant guidelines from the ACMG, the American College of Obstetricians and Gynecologists (ACOG), the National Society of Genetic Counselors, the Perinatal Quality Foundation, and the Society for Maternal-Fetal Medicine [23,24,25]. Eligible individuals were infertile couples under 35 years of age.

Exclusion criteria: Individuals with incomplete baseline or follow-up data, or those who declined participation, were excluded.

All participants received comprehensive information about the study and provided voluntary informed consent.

### *2.3 Screening Mode*

Couples underwent either sequential or simultaneous carrier screening. The screening approach was determined following consultation between the patients and their physician.

### *2.4 Genetic Testing and Bioinformatics Analysis*

All PECS were performed using a commercial testing service (Yikon Genomics Co., Ltd., Beijing, China). The technical workflow followed established protocols for reproductive genetic carrier screening. Detailed methodology is described below.

### 2.5 Sequencing Platform and Amplification Technology

PECS was performed using a next-generation sequencing (NGS) platform based on multiple annealing and looping-based amplification cycles (MALBAC). This technology integrates NGS with MALBAC for single-cell whole-genome amplification, which achieves high coverage uniformity (93%) and reduces allele dropout, thereby improving the accuracy of variant detection. Sequencing was conducted on clinical-grade, high-throughput instruments (Illumina NovaSeq or MGI DNBSEQ series), which have been widely validated for high sensitivity and specificity in reproductive genetics.

### 2.6 Target Region Capture

Targeted enrichment was performed using a customized liquid-hybridization capture strategy. Probes were designed to cover the complete coding sequences and $\pm$10 bp of flanking intronic regions of 26 core genes associated with the 22 screened diseases. Known pathogenic intronic sites documented in the Clinical Variation database maintained by NCBI (ClinVar) were also included. Capture specificity was $\geq$95%. Detectable variant types included single-nucleotide polymorphisms (SNPs), small insertions/deletions (InDels, $\leq$10 bp), and CNVs, such as *SMN1* exon deletions and *HBA1/HBA2* deletions (-SEA/-$\alpha$3.7).

### 2.7 Sequencing Depth

To ensure reliable detection of low-frequency variants, the average sequencing depth for target regions was set at $\geq$150$\times$, with $\geq$92% of bases covered at $\geq$20$\times$. These parameters were established based on platform performance and clinical requirements for carrier screening.

### 2.8 Quality Control

Strict quality control was applied at all stages. Genomic DNA extracted from peripheral blood was required to have a concentration $\geq$50 ng/µL and an OD260/280 ratio of 1.8–2.0; degraded or contaminated samples were excluded. Library fragments were size-selected to 200–300 bp, with adapter contamination $\leq$0.5%. Raw sequencing data met the following quality thresholds: Q30 $\geq$90% and Q20 $\geq$95%.

### 2.9 Bioinformatics Analysis Pipeline for Variant Calling

Bioinformatics analysis was performed using an automated pipeline combined with standard ECS protocols. Low-quality reads (Q $<$20), adapter-contaminated sequences, and polymerase chain reaction (PCR) duplicates were removed. Clean reads were aligned to the human reference genome (GRCh37/hg19) using BWA-MEM (v0.7.17) (Broad Institute, Cambridge, MA, USA) and processed with SAMtools. SNVs and InDels were called with GATK HaplotypeCaller (v4.2.6.1; Broad Institute, Cambridge, MA, USA), with a variant allele frequency (VAF) threshold $\geq$0.01. CNVs were detected using a depth-based normalization method to identify gene-level deletions and duplications. Variants were annotated using ANNOVAR, which integrates data from public databases (ClinVar: https://www.ncbi.nlm.nih.gov/clinvar/, gnomAD East Asian subset: https://gnomad.broadinstitute.org/, HGMD Professional: https://digitalinsights.qiagen.com/products-overview/clinical-insights-portfolio/human-gene-mutation-database/) and a local database of over 1000 Chinese individuals of reproductive age.

### 2.10 Variant Interpretation

Variants were classified according to the ACMG/AMP 2015 guidelines. P variants were defined based on criteria such as loss-of-function and supporting evidence from ClinVar. LP variants required supportive computational predictions and a low population frequency ($<$0.01% in the East Asian gnomAD dataset). VUS were excluded from carrier frequency calculations but were disclosed during genetic counseling. All P/LP variants underwent independent review by two senior analysts, with discrepancies resolved through consensus among technical and clinical staff.

### 2.11 Observational Indicator

The overall carrier rate was calculated as the number of individuals carrying at least one P/LP variant for the target diseases divided by the total number of screened individuals. The ARC rate was defined as the proportion of screened couples identified as at high genetic risk. All couple-level metrics used the number of couples as the denominator, whereas individual-level metrics used the number of individuals. Proportions were treated as binary data. 95% confidence intervals (CIs) were calculated based on the binomial distribution, using the normal approximation method when applicable (sample proportion $\geq$1%, n $\geq$5); otherwise the Clopper-Pearson exact method was used. CIs were rounded to two decimal places and computed with R 4.3.0. (version 4.3.0; R Foundation for Statistical Computing, Vienna, Austria) Clinical outcomes for ARCs were recorded.

## 3. Results

### 3.1 Overall PECS Results and Carrier Frequencies

Among the 4853 individuals tested by PECS, 81.58% (3959/4853) had no P variants. The overall carrier rate for the target genes was 18.42% (894/4853; 95% CI: 17.33–19.51%). Among carriers, 16.73% (812/4853; 95% CI: 15.80–17.69%) carried one variant, 1.61% (78/4853; 95% CI: 1.28–1.99%) carried two, and 0.08% (4/4853; 95% CI: 0.02–0.21%) carried three (Fig. 1). A total of 474 (53.02%) of the 894 carriers proceeded to assisted reproduction.

Among the 894 identified carriers, 363 (40.60%) were male and 531 (59.40%) were female. Their demographic characteristics are summarized in Table 1.

**Table 1. Demographic characteristics of the screened cohort.**

| Characteristic | Female (n = 2733) | Male (n = 2120) |
|---|---|---|
| Age (years) | 30.58 ± 3.03 (median: 31, range: 20–35) | 32.26 ± 3.89 (median: 32, range: 22–59) |
| Duration of infertility (years) | 3.64 ± 2.50 (median: 3, range: 1–21) | 3.66 ± 2.55 (median: 3, range: 1–23) |
| Body mass index (kg/m$^2$) | 23.43 ± 3.66 (median: 22.89, range: 13.84–41.53) | 24.11 ± 3.70 (median: 23.35, range: 15.00–41.80) |
| Type of infertility | | |
| Primary infertility | 1539 (56.31%) | 1419 (66.93%) |
| Secondary infertility | 1194 (43.69%) | 701 (33.07%) |

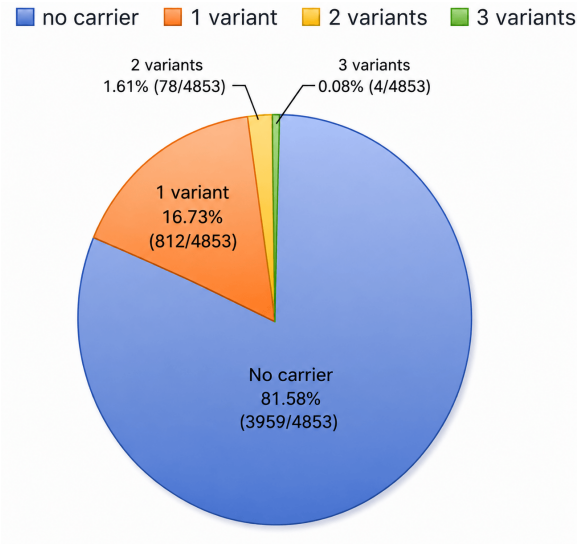


**Fig. 1. Distribution of carrier status by variant count.**

The screening panel encompassed 22 monogenic diseases (Table 2). This included an expanded panel for 20 conditions (covering 26 gene regions), along with targeted screening for Fragile X syndrome (Fragile X messenger ribonucleoprotein 1, *FMR1*) and spinal muscular atrophy (SMA; *SMN1*). The reported detection rates and population carrier frequencies refer to the Chinese population.

Genes were selected based on clinical severity, carrier frequency in the target population, and the availability of reliable detection methods. Identification of pathogenic variants in these genes enables subsequent interventions, including genetic counseling, preimplantation genetic testing for monogenic disorders (PGT-M), and prenatal diagnosis, to prevent the birth of affected offspring. Carrier rates differed by sex for some conditions; for example, gap junction protein beta 2 (*GJB2*) variants were carried by 3.40% of males versus 2.78% of females (Tables 3A,3B). For Table 3A, the denominator was 2120, which represents all male samples; for Table 3B, the denominator is 2733, which represents all female samples.

Pathogenic variants were identified across all screened individuals and ranked by carrier frequency (Table 4). The *GJB2* gene had the highest carrier rate (3.05%; 148/4853; 95% CI: 2.57–3.53%), followed by cytochrome P450 family 21 subfamily A member 2 (*CYP21A2*) (2.64%; 128/4853; 95% CI: 2.20–3.08%).

### *3.2 Clinical Utility of PECS in Couples Undergoing In Vitro Fertilization (IVF)*

PECS identified ARCs and supported informed reproductive decision-making. Among 2082 couples tested, 48 were identified as ARCs, yielding a detection rate of 2.31% (48/2082; 95% CI: 1.67–2.95%). Stratified by genetic etiology, ARC subtypes comprised 21 couples (43.75%) with chromosomal microdeletions or microduplications, 11 couples (22.92%) with AR risks, 8 couples (16.67%) with mitochondrial disorders, and 8 couples (16.67%) with XL risks. Thus, chromosomal abnormalities were the most common risk category.

Following genetic counseling, 28 ARC couples underwent ovarian stimulation. Their subtype distribution was 11 AR-related, 5 XL-related, 7 with microdeletions or microduplications, and 5 with mitochondrial disorders. 4 patients discontinued the cycle due to a lack of viable embryos.

Of the remaining 24 couples, 1 AR couple (both carriers of a pathogenic hemoglobin subunit beta, *HBB* variant) underwent PGT-M, resulting in a healthy live birth confirmed by prenatal diagnosis. Another couple, whose female was a carrier of pathogenic ATPase copper transporting beta (*ATP7B*) and a 1q21.1 microdeletion, opted for PGT-A, with embryo transfer pending.

The remaining 22 couples underwent conventional ART. During follow-up, 14 couples achieved live births. Of the 28 ARC couples who proceeded to treatment, a total of 15 healthy live births were achieved—14 among the couples who underwent conventional ART and 1 from the couple who underwent PGT-M (Table 5). 7 of these pregnancies underwent prenatal diagnosis via amniocentesis, all with normal results. 1 pregnancy was terminated after amniocentesis identified a high-risk dystrophin duchenne muscular dystrophy (DMD) variant (classified as pathogenic). This finding was a direct consequence of a PECS-identified pathogenic variant. As the variant is XL recessive, male fetuses who inherit it would develop DMD. Thus, the PECS result was causally linked to the pregnancy termination. Another pregnancy was terminated after prenatal testing revealed right-hand and digestive tract malformations in the fetus. The PECS panel identified heterozygous pathogenic variants in *HBA1/HBA2* (NM_000558.3/NM_000517.4) and a heterozygous gene deletion variant in steroid sulfatase (*STS*) (NM_000351.4)

**Table 2. Components of the genetic testing panel.**

| Disease | Gene | Mode of inheritance | Detection rate | Population carrier rate |
|---|---|---|---|---|
| Hereditary Hearing Loss | *GJB2* (NM_004004.5) | AR | >78% | 1/22 |
| | *SLC26A4* (NM_000441.1) | AR | >93% | 1/39 |
| | *MTRNR1* (NC_012920) | maternal inheritance | >89% | 1/438 (female) |
| Thalassemia | *HBA1* (NM_000558.3)<br>*HBA2* (NM 000517.4) | AR | >99% | 1/48 |
| | *HBB* (NM 000518.4) | AR | >99% | 1/313 |
| Duchenne muscular dystrophy (DMD) | *DMD* (NM_004006.2) | XL recessive | >80% | 1/1175 (female) |
| Hemophilia A | *F8* (NM_000132.3) | XL recessive | >60% | 1/1500 (female) |
| Fragile X Syndrome | *FMR1* (NM_002024.5) | XL | >98% | 1/1915 (female) |
| XL ichthyosis | *STS* (NM 000351.5) | XL recessive | >85% | 1/1915 (female) |
| Spinal muscular atrophy (SMA) | *SMN1* (NM 000344.3) | AR | >98% | 1/46 |
| PKU | *PAH* (NM_000277.1) | AR | >86% | 1/48 |
| | *PTS* (NM_000317.2) | AR | >85% | 1/136 |
| Methylmalonic Acidemia (MMA) | *MMACHC* (*NM_015506.2*) | AR | >90% | 1/77 |
| | *MMUT* (NM_000255.3) | AR | >80% | 1/151 |
| Congenital Adrenal Hyperplasia (CAH) | *CYP21A2* (NM 000500.7) | AR | >93% | 1/36 |
| Hepatolenticular Degeneration | *ATP7B* (NM_000053.3) | AR | >93% | 1/40 |
| Paget's disease | *GAA* (NM_000152.5) | AR | >72% | 1/65 |
| Xp11.22 Microduplication Syndrome | *HUWE1* (NM_031407.6) | XL | 100% | <1/100,000 (female) |
| Pelizaeus-Merzbacher Disease | *PLP1* (NM_000533.4) | XL recessive | >60% | 1/25,000 (female) |
| MECP2 Duplication Syndrome | *MECP2* (NM_004992.3) | XL recessive | 100% | <1/100,000 (female) |
| Int22h1/Int22h2-Mediated Xq28 Duplication Syndrome | *Xq28* (154.896–155.335 Mb) | XL | 100% | <1/100,000 (female) |
| Xg28 (MECF2) Microdeletion Associated Rett Syndrome | *Xq28* (154.021–154.097 Mb) | XL dominant | 100% | 1/10,000 (female) |
| 22q11.2 Microdeletion Microduplication Syndrome | *TBX1* (NM_080647.1) | autosomal dominant | 100% | 1/2300 |
| 1q21.1 Microdeletion Microduplication Syndrome | *GJA5* (NM 005266.6) | autosomal dominant | 100% | <1/100,000 |
| 16p11.2 Microdeletion Syndrome | *SH2B1* (NM_001145795.1) | autosomal dominant | 100% | <1/100,000 |
| 16p11.2Microdeletion Microduplication Syndrome | *TBX6* (NM_004608.3) | autosomal dominant | 100% | <1/100,000 |
| 17q12 Microdeletion Syndrome | *HNF1B* (NM_00458.3) | autosomal dominant | 100% | <1/100,000 |

AR, autosomal recessive; XL, X-linked; PKU, phenylketonuria.

**Table 3A. Pathogenic variant carrier status in male patients (n = 2120).**

| AR | | | Autosomal dominant | | | XL | | |
|---|---|---|---|---|---|---|---|---|
| Gene | Number | 95% CI | Gene | Number | 95% CI | Gene | Number | 95% CI |
| *GJB2* | 72 (3.40%) | 2.63 to 4.17 | *TBX6* | 3 (0.14%) | 0.03 to 0.41 | *STS* | 3 (0.14%) | 0.03 to 0.41 |
| *HBA1/2* | 52 (2.45%) | 1.79 to 3.11 | *TBX1* | 2 (0.09%) | 0.01 to 0.34 | *FMR1* | 1 (0.05%) | 0.00 to 0.26 |
| *CYP21A2* | 49 (2.31%) | 1.67 to 2.95 | *GJA5* | 2 (0.09%) | 0.01 to 0.34 | | | |
| *SMN1* | 40 (1.89%) | 1.31 to 2.47 | | | | | | |
| *ATP7B* | 38 (1.79%) | 1.23 to 2.35 | | | | | | |
| *PAH* | 30 (1.42%) | 0.91 to 1.92 | | | | | | |
| *MMACHC* | 27 (1.27%) | 0.80 to 1.75 | | | | | | |
| *SLC26A4* | 25 (1.18%) | 0.72 to 1.64 | | | | | | |
| *HBB* | 17 (0.80%) | 0.47 to 1.28 | | | | | | |
| *GAA* | 17 (0.80%) | 0.47 to 1.28 | | | | | | |
| *MMUT* | 16 (0.76%) | 0.43 to 1.21 | | | | | | |
| *PTS* | 5 (0.24%) | 0.08 to 0.55 | | | | | | |

CI, confidence interval.

**Table 3B. Pathogenic variant carrier status in female patients (n = 2733).**

| AR | | Autosomal dominant | | XL | | Maternal inheritance | |
|---|---|---|---|---|---|---|---|
| Gene | Number/95% CI | Gene | Number/95% CI | Gene | Number/95% CI | Gene | Number/95% CI |
| *CYP21A2* | 79 (2.89%)/2.26 to 3.52 | *TBX1* | 5 (0.18%)/0.06 to 0.42 | *STS* | 5 (0.11%)/0.02 to 0.32 | *MTRNR1* | 8 (0.29%)/0.13 to 0.57 |
| *GJB2* | 76 (2.78%)/2.16 to 3.40 | *GJA5* | 4 (0.15%)/0.04 to 0.37 | *FMR1* | 2 (0.07%)/0.01 to 0.26 | | |
| *ATP7B* | 64 (2.34%)/1.77 to 2.91 | *TBX6* | 5 (0.07%)/0.01 to 0.26 | *DMD* | 1 (0.04%)/0.00 to 0.21 | | |
| *SMN1* | 63 (2.31%)/1.75 to 2.87 | | | *F8* | 1 (0.04%)/0.00 to 0.21 | | |
| *SLC26A4* | 52 (1.90%)/1.39 to 2.41 | | | | | | |
| *HBA1/2* | 51 (1.87%)/1.36 to 2.38 | | | | | | |
| *PAH* | 44 (1.61%)/1.14 to 2.08 | | | | | | |
| *MMACHC* | 31 (1.13%)/0.73 to 1.53 | | | | | | |
| *HBB* | 28 (1.02%)/0.64 to 1.40 | | | | | | |
| *GAA* | 24 (0.88%)/0.55 to 1.32 | | | | | | |
| *MMUT* | 20 (0.73%)/0.44 to 1.15 | | | | | | |
| *PTS* | 12 (0.44%)/0.23 to 0.78 | | | | | | |

in this patient. Both variants were categorized as P according to ACMG/AMP criteria. The -$\alpha$3.7 deletion is associated with $\alpha$-thalassemia, and the *STS* deletion is associated with XL ichthyosis. However, neither condition is known to include limb or digestive tract malformations; therefore, the PECS findings were not associated with the fetal anomalies that led to termination. 3 patients experienced early miscarriage, and 3 did not conceive after embryo transfer. The overall study flow and clinical outcomes of ARC couples are summarized in Fig. 2. Briefly, among 4853 patients (2082 couples) screened by PECS, 48 ARCs were identified. 20 couples did not pursue further intervention after testing, whereas 28 entered ovarian stimulation. These couples subsequently received PGT-M, PGT-A, or conventional ART, with corresponding pregnancy outcomes.

Table 5 summarizes the carrier status and pregnancy outcomes for the 11 identified at-risk couples (ARCs). 8 couples achieved healthy live births. 2 patients experienced early miscarriage, and 1 patient canceled the embryo transfer due to a lack of viable embryos.

Table 6 presents the characteristics and pregnancy outcomes of 5 female carriers of XL diseases. 1 of these women was identified as a carrier of a pathogenic *DMD* variant.

Table 7 summarizes the data of patients with chromosomal microdeletions and microduplications. Prenatal diagnosis by amniocentesis was performed in only 1 carrier (of a *T-box transcription factor 6 TBX6* variant), which showed no abnormalities and resulted in a healthy live birth.

Table 8 presents the characteristics and pregnancy outcomes of 5 female carriers of maternally inherited diseases. All individuals were identified as carriers of the pathogenic MTRNR1 variant associated with hereditary hearing loss. Among them, 1 patient achieved a healthy live birth via rescue intracytoplasmic sperm injection (RICSI), 2 patients experienced miscarriage, and 2 individuals had no viable embryos for transfer.

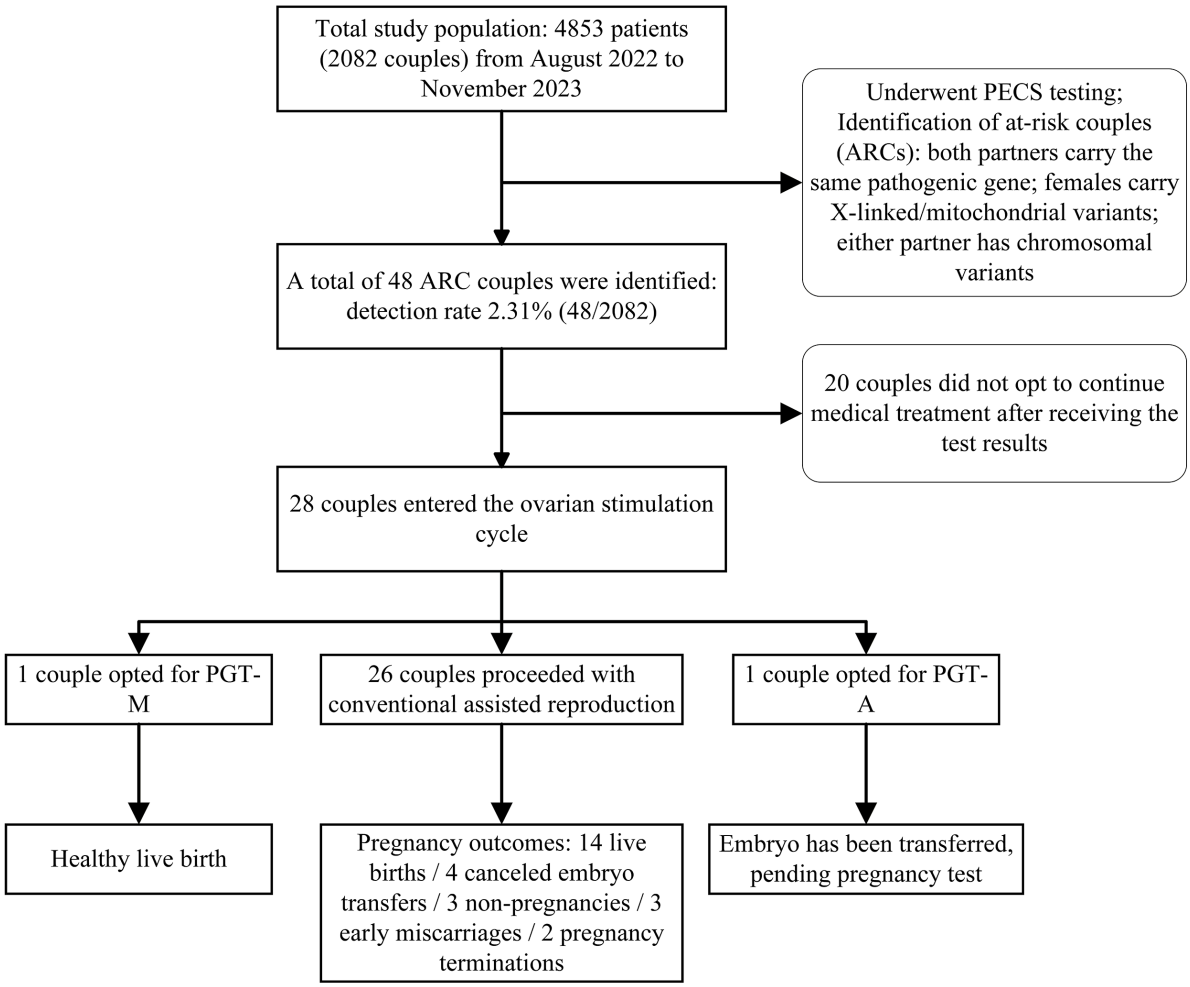


**Fig. 2. Flow diagram of the study cohort and clinical outcomes of at-risk couples (ARCs).** PECS, preconception expanded carrier screening; PGT-M, preimplantation genetic testing for monogenic disorders; PGT-A, preimplantation genetic testing for aneuploidy.

## 4. Discussion

In Europe, 2–4% of childbearing-age couples without a family history are estimated to be at increased risk of having a child with a recessive genetic disorder [26]. Carrier frequencies, however, vary substantially across populations. For instance, in southern China, 27.49% of individuals without a family history have been reported to carry at least one pathogenic variant, with carrier frequencies varying widely across ethnic groups (4.15–81.35%) [27]. Chen et al. [28] conducted ECS in 300 couples undergoing ART, targeting 330 genes associated with 342 AR or XL diseases (including both SNVs and CNVs), and identified that 64.83% of patients carried at least one pathogenic variant. Another study that focused on 201 recessive diseases in a Chinese infertile population reported a carrier rate of 46.73% [29]. In the present study, among 4853 individuals screened for 22 diseases, 81.58% (3959/4853) exhibited no pathogenic variants, and the overall carrier rate was 18.42% (894/4853).

To contextualize our findings, we compared them with domestic and international reports. Domestically, Xi et al. (2020) [29] reported a carrier rate of 46.73% among 2923 Chinese ART patients using a panel that covered 201 genes and 135 diseases, which is significantly higher than the 18.42% observed in our study. Internationally, Arjunan et al. (2021) [30] reported carrier rates of 57.4–60.6% in a U.S. cohort screened with a 176-gene panel, and Peyser et al. (2019) [31] documented a rate of 29.4% in 4232 infertility patients using a 102-gene panel. The lower carrier rate observed in our study is primarily attributable to the smaller panel size (22 diseases vs. 135–201 diseases in Chinese studies and 102–176 genes internationally), as panel breadth directly influences the probability of carrier detection. Furthermore, our cohort was recruited from Central China, whereas the study by Xi et al. (2020) [29] included patients from Shanghai; genetic differences among regional Han subgroups may also contribute to these differences.

Regarding the detection rate of ARCs, we reported 2.31% (48/2082), which is comparable to the 2.26% reported by Xi et al. (2020) [29] but higher than international estimates, including 1.3% in a U.S. study with a 176-gene panel, 1.2% in a U.S. fertility center, and 1.9% in an

**Table 4. Number of carriers and positive carrier rate for each pathogenic gene.**

| Disease | Gene | Carrier frequency | | |
|---|---|---|---|---|
| | | N | % | 95% CI |
| Hereditary Hearing Loss | *GJB2* | 148 | 3.05% | 2.57 to 3.53 |
| Congenital Adrenal Hyperplasia | *CYP21A2* | 128 | 2.64% | 2.20 to 3.08 |
| Thalassemia | *HBA1/HBA2* | 103 | 2.12% | 1.75 to 2.50 |
| SMA-1 | *SMN1* | 103 | 2.12% | 1.75 to 2.50 |
| Hepatocerebral Degeneration | *ATP7B* | 102 | 2.10% | 1.73 to 2.48 |
| Hereditary Hearing Loss | *SLC26A4* | 77 | 1.59% | 1.28 to 1.95 |
| PKU | *PAH* | 74 | 1.52% | 1.22 to 1.88 |
| MMA | *MMACHC* | 58 | 1.20% | 0.90 to 1.55 |
| Thalassemia | *HBB* | 45 | 0.93% | 0.69 to 1.23 |
| Pompe Disease | *GAA* | 41 | 0.84% | 0.61 to 1.13 |
| MMA | *MMUT* | 36 | 0.74% | 0.52 to 1.01 |
| PKU | *PTS* | 17 | 0.35% | 0.21 to 0.54 |
| Hereditary Hearing Loss | *MTRNR1* | 8 | 0.16% | 0.07 to 0.30 |
| XL Ichthyosis | *STS* | 8 | 0.16% | 0.07 to 0.30 |
| 16p11.2 Microdeletion/Microduplication Syndrome | *TBX6* | 8 | 0.16% | 0.07 to 0.30 |
| 22q11.2 Deletion/Duplication Syndrome | *TBX1* | 7 | 0.14% | 0.06 to 0.27 |
| 1q21.1 Microdeletion/Microduplication Syndrome | *GJA5* | 6 | 0.12% | 0.04 to 0.24 |
| Fragile X Syndrome | *FMR1* | 3 | 0.06% | 0.04 to 0.24 |
| Pelizaeus-Merzbacher Disease | *PLP1* | 2 | 0.04% | 0.00 to 0.08 |
| Pseudohypertrophic Muscular Dystrophy | *DMD* | 1 | 0.02% | 0.00 to 0.05 |
| Hemophilia A | *F8* | 1 | 0.02% | 0.00 to 0.05 |

Australian nationwide study using a 1281-gene panel. This discrepancy likely reflects our expanded ARC definition, which incorporates mitochondrial disorders and chromosomal microdeletions or microduplications, whereas most international studies consider only AR and XL conditions [30,31]. Notably, despite the smaller panel, our ARC rate closely aligns with that reported by Xi et al. (2020) [29], indicating that our panel effectively captures high-prevalence pathogenic genes in the Central Chinese ART population (e.g., *GJB2*, *CYP21A2*, *HBA1/HBA2*, *SMN1*, *ATP7B*, each with a carrier rate >2%).

In terms of balancing detection efficiency and cost-effectiveness, our 22-disease panel represents an optimized design. It was tailored to include conditions with high carrier frequencies in the Chinese population (e.g., *GJB2*: 3.05%, *CYP21A2*: 2.64%) and severe clinical outcomes, ensuring detection of clinically relevant ARCs while controlling costs (≈$80 per test). In contrast, larger panels (≥100 genes) provided limited additional benefit. Xi et al. (2020) [29] demonstrated that screening the top 31 genes (carrier rate >0.5%) identified 94% of ARCs, and Ben-Shachar et al. (2019) [23] observed that expansion beyond 18 core genes only marginally increased ARC detection. By excluding low-prevalence genes, our panel reduces provider burden and patient anxiety while maintaining clinical utility, consistent with the tandem-reflex strategy aimed at minimizing non-effective tests [30].

The observed differences in carrier rates may also reflect variations in screening depth and population genetic structure. In our cohort, 16.73% (812/4853) carried 1 variant, 1.61% (78/4853) carried 2, and 0.08% (4/4853) carried 3. The lower overall carrier rate relative to prior studies may be attributable to the narrower panel, as well as differences in testing methods and variant-reporting criteria. As a single-center retrospective analysis, our findings may be subject to regional factors, sample selection bias, and facility-specific limitations. Multicenter prospective studies are therefore required to validate the generalizability of these results. Furthermore, although genetic counseling and reproductive advice were provided to high-risk couples, individual decision-making varies and may not fully reflect post-screening choices across the broader patient population.

Research indicates that ARC detection rates attributable to AR diseases in the general population range from 0.10% to 0.20% [32]. In our study, the detection rate for AR ARCs was 0.53% (11/2082), which exceeds that reported in the general population.

With respect to specific genes, the carrier rate of *STS* in the general female population in the United Kingdom was reported to be approximately 0.14% [33], whereas it was relatively higher in our infertile cohort. In addition, 2 patients in our study carried *FMR1* variants (0.10%, 2/2082). *FMR1* premutation carriers have an increased risk of preterm birth or primary ovarian insufficiency, which may significantly impact female offspring [34,35,36,37,38]. 1 patient was a carrier of a pathogenic *DMD* variant; although her partner tested negative, she had a 50% risk of

**Table 5. Clinical, genetic characteristics, and pregnancy outcomes of high-risk couples.**

| Partner 1 pathogenic variant | Partner 2 pathogenic variant | Gene | Pathology | ART | Prenatal diagnosis | Results |
|---|---|---|---|---|---|---|
| HBA2 (NM_000517.4): -$\alpha$4.2 | HBA1/HBA2 (NM_000558.3/NM_000517.4): -$\alpha$3.7 | *HBA2* | Thalassemia | IVF | / | Live Healthy-Birth |
| HBB (NM_000518.4): c.126_129delCTTT (p.F42Lfs*19) | HBB (NM_000518.4): c.316-197C>T | *HBB* | Thalassemia | PGT-M | Negative | Live Healthy-Birth |
| HBB (NM_000518.4): c.217dupA (p.S73Kfs*2); ATP7B (NM_000053.3): c.3809A>G (p.N1270S) | HBB (NM_000518.4): c.126_129delCTTT (p.F42Lfs*19) | *HBB* | Thalassemia | IVF | Negative | Live Healthy-Birth |
| HBA2 (NM_000517.4): -$\alpha$4.2 | HBA2 (NM_000517.4): -$\alpha$4.2 | *HBA2* | Thalassemia | IVF | / | Live Healthy-Birth |
| GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3); CYP21A2 (NM_000500.7): c.955C>T (p.Q319*) | *GJB2* | Hereditary Hearing Loss | IVF | / | Live Healthy-Birth |
| GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | *GJB2* | Hereditary Hearing Loss | IVF | / | No viable embryo |
| GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | *GJB2* | Hereditary Hearing Loss | IVF | / | Miscarriage |
| SMN1 (NM_000344.3): (EX7-8DEL) | SMN1 (NM_000344.3): (EX7-8DEL) | *SMN1* | SMA-1 | IVF | Negative | Live Healthy-Birth |
| SMN1 (NM_000344.3): (EX7-8DEL) | SMN1 (NM_000344.3): (EX7-8DEL) | *SMN1* | SMA-1 | IVF | Negative | Live Healthy-Birth |
| CYP21A2 (NM_000500.7): c.955C>T (p.Q319*) | CYP21A2 (NM_000500.7): Gene fusion (CH-3) | *CYP21A2* | CAH | IVF | Negative | Live Healthy-Birth |
| CYP21A2 (NM_000500.7): c.955C>T (p.Q319*); GJB2 (NM_004004.5): c.235delC (p.L79Cfs*3) | CYP21A2 (NM_000500.7): c.955C>T (p.Q319*) | *CYP21A2* | CAH | IVF | / | Miscarriage |

ART, assisted reproductive technology; IVF, *in vitro* fertilization.

**Table 6. XL disease carriers information.**

| Pathogenic variant | Gene | Pathology | ART | Prenatal diagnosis | Results |
|---|---|---|---|---|---|
| STS (NM_000351.4): gene deletion mutation, Heterozygous | *STS* | XL ichthyosis | IVF | / | Live Healthy-Birth |
| STS (NM_000351.4): gene deletion mutation, Heterozygous | *STS* | XL ichthyosis | IVF | / | Live Healthy-Birth |
| STS (NM_000351.4): gene deletion mutation, Heterozygous HBA1/HBA2 (NM_000558.3/NM_000517.4): -$\alpha$3.7, Heterozygous | *STS* | XL ichthyosis | ICSI | / | Induction on labor (Right Hand and Digestive Tract Malformations) |
| STS (NM_000351.4): gene deletion mutation, Heterozygous | *STS* | XL ichthyosis | IVF | / | Not pregnant |
| DMD (NM_004006.2): del (EX51-55DEL) | *DMD* | DMD | IVF | Positive | Induction on labor (DMD) |

STS, steroid sulfatase; ICSI, intracytoplasmic sperm injection.

**Table 7. Patients with microdeletions and microduplications.**

| Pathogenic variant | Gene | Pathology | ART | Prenatal diagnosis | Result |
|---|---|---|---|---|---|
| TBX6 (NM_004608.3): gene deletion mutation, Heterozygous | *TBX6* | proximal 16p11.2 microdeletion syndrome | IVF | Negative | Live Healthy-Birth |
| TBX6 (NM_004608.3): gene duplication mutation, Heterozygous | *TBX6* | proximal 16p11.2 microduplication syndrome | ICSI | / | Miscarriage |
| TBX1 (NM_080647.1): gene duplication mutation, Heterozygous | *TBX1* | 22q11.2 microduplication syndrome | ICSI | / | Live Healthy-Birth |
| GJA5 (NM_005266.6): gene deletion mutation, Heterozygous ATP7B (NM_000053.3): c.2333G>T (p.R778L), Heterozygous | *GJA5 ATP7B* | distal 1q21.1 microdeletion syndrome Wilson's Disease | PGT-A | / | In preparation |
| TBX1 (NM_080647.1): gene duplication mutation, Heterozygous | *TBX1* | 22q11.2 microduplication syndrome | IVF | / | Not pregnant |
| TBX1 (NM_080647.1): gene duplication mutation, Heterozygous CYP21A2 (NM_000500.7): c.1069C>T (p.R357W), Heterozygous | *TBX1 CYP21A2* | 22q11.2 microduplication syndrome CAH | ICSI | / | Ongoing |
| TBX1 (NM_080647.1): gene deletion mutation, Heterozygous | *TBX1* | 22q11.2 microdeletion syndrome | ICSI | Negative | Ongoing |

**Table 8. Maternal inheritance disease carriers informations.**

| Pathogenic variant | Gene | Pathology | Prenatal diagnosis | ART | Results |
|---|---|---|---|---|---|
| MTRNR1 (NC_012920.1): m.1555A>G, Homoplasmy (Mutation Ratio 100%) | *MTRNR1* | Hereditary Hearing Loss | / | RICSI | Live Healthy-Birth |
| MTRNR1 (NC_012920.1): m.1555A>G, Homoplasmy (Mutation Ratio 100%) | *MTRNR1* | Hereditary Hearing Loss | / | IVF | No viable embryo |
| MTRNR1 (NC_012920.1): m.1555A>G, Heteroplasmy (Mutation Ratio 71%) | *MTRNR1* | Hereditary Hearing Loss | / | IVF | Miscarriage |
| MTRNR1 (NC_012920.1): m.1555A>G, Homoplasmy (Mutation Ratio 100%) | *MTRNR1* | Hereditary Hearing Loss | / | IVF | No viable embryo |
| MTRNR1 (NC_012920.1): m.1555A>G, Heteroplasmy (Mutation Ratio 68%) | *MTRNR1* | Hereditary Hearing Loss | / | IVF | Miscarriage |

RICSI, rescue intracytoplasmic sperm injection.

transmitting the variant to male offspring. At 16 weeks' gestation, amniocentesis confirmed a *DMD* variant in the fetus, leading to pregnancy termination.

21 patients carried chromosomal microdeletions or microduplications, corresponding to a detection rate of 1.0% (21/2082). Previous studies have reported that the incidence of such findings in fetuses of chromosomally normal pregnant women can reach 1–1.7% [39]; examples include Williams, DiGeorge, and Prader-Willi syndromes. Furthermore, chromosomal microdeletions or microduplications account for approximately 12% of unexplained intellectual disability, congenital malformations, and developmental delay [40]. 8 women carried mitochondrial DNA (mtDNA) mutations, which corresponds to a detection rate 0.29%, 8/2733. mtDNA mutations are associated with a broad range of disorders, including neuromuscular disease, diabetes, vision loss, and hearing impairment [41,42,43,44,45]. All 8 patients in our cohort had mitochondrially encoded 12S RNA *(MTRNR1)*-related hereditary deafness.

It is important to emphasize that PECS reports include only variants with well-defined phenotypes that are covered by the screening panel. VUS are not reported, which means that PECS cannot capture all potential genetic risks, especially those associated with emerging diseases or poorly characterized genotypes. Moreover, a negative PECS result does not fully exclude the risk of inherited disorders, given the complex inheritance patterns of many conditions and the limited gene coverage of any panel. Therefore, comprehensive pretest genetic counseling is essential to explain the purpose, scope, and limitations of screening, as well as the potential need for additional tests [46]. Clinicians must have sufficient expertise in genetic counseling to accurately interpret results and guide patients on risk management and future reproductive options.

Although ECS provides valuable information for reproductive planning, it also raises ethical considerations. A key concern is that ECS may be perceived as encouraging selective reproduction, bordering on eugenics. It should be clarified that the goal of ECS is not to eliminate individuals with disabilities, but to empower prospective parents through informed, non-directive counseling. Academic perspectives vary: some support widespread ECS to reduce the burden of severe genetic disease, whereas others caution against potential societal pressures, stigmatization, and inequitable access. Organizations such as ACOG and ACMG emphasize the principles of voluntariness, informed consent, and non-directive counseling. Therefore, any implementation strategy must balance public health benefits with individual rights. Further interdisciplinary research incorporating perspectives from medical ethics, sociology, and health economics is needed to guide the responsible integration of ECS into clinical practice.

*Limitations*

Several limitations of the present study should be acknowledged. First, this research relies on a single-center retrospective analysis, meaning the findings may be inherently influenced by regional characteristics, sample selection biases, and the specific limitations of the medical facilities involved. Therefore, large-scale, multi-center prospective studies are necessary to further validate the generalizability and reliability of these results across broader populations. Second, the initial research design restricted the expanded carrier screening (PECS) panel to 22 specific diseases. This targeted scope dictates that mutations with unclear clinical significance were not reported, and a negative screening result cannot completely eliminate the overall risk of offspring inheriting genetic diseases, given the complex, multigenic nature of many inherited disorders. Finally, although comprehensive genetic counseling and reproductive advice were provided to all identified high-risk couples, their final reproductive choices remain highly subjective. Consequently, the clinical outcomes observed here may not fully reflect the standardized reproductive decision-making processes of all patients following genetic screening.

## 5. Conclusions

This retrospective study evaluated the clinical value of PECS in an ART population in Central China. Among 4853 patients (2082 couples) screened for 22 monogenic diseases, 18.42% carried at least 1 pathogenic variant. The most frequently implicated genes were *GJB2*, *CYP21A2*, *HBA1/HBA2*, *SMN1*, and *ATP7B*. The ARC detection rate was 2.31%, encompassing AR, XL, chromosomal microdeletion or microduplication, and mitochondrial subtypes. Through tailored genetic counseling and subsequent interventions, including PGT-M, PGT-A, and conventional ART, most ARCs achieved favorable pregnancy outcomes, with 14 couples delivering healthy infants. In summary, PECS provides a valuable tool for identifying genetic risks in this population, supports informed reproductive decision-making, and contributes to the prevention of affected offspring. These findings support the implementation of a focused, population-tailored carrier screening strategy in Central China.

## Availability of Data and Materials

The datasets generated during and analyzed during the current study are available from the corresponding author on reasonable request.

## Author Contributions

ZS and YZ designed the research study. BS, HL and CC performed the research. XP, KF and ZW have participated in the acquisition and preprocessing of key research data, the analysis and interpretation of experimental data

for this study, which constitutes a substantial contribution to the work as required. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript. All authors have participated sufficiently in the work and agreed to be accountable for all aspects of the work.

## Ethics Approval and Consent to Participate

This study protocol was formulated in accordance with the requirements of the Declaration of Helsinki of the World Medical Association, and the protocol was approved by the Ethics Committee of Shiyan Renmin Hospital (approval number: SYRMYY-2025-107). All patients or their families/legal guardians gave their informed consent for inclusion before they participated in the study.


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## Conflicts of Interest

The authors declare no conflicts of interest.

## References


[1] Bell CJ, Dinwiddie DL, Miller NA, Hateley SL, Ganusova EE, Mudge J, et al. Carrier testing for severe childhood recessive diseases by next-generation sequencing. Science Translational Medicine. 2011; 3: 65ra4. https://doi.org/10.1126/scitranslmed.3001756

[2] Schürmann PJL, van Breda Vriesman SPE, Castro-Alpízar JA, Kooijmans SAA, Nieuwenhuis EES, Schiffelers RM, et al. Therapeutic application of mRNA for genetic diseases. Wiley Interdiscip Rev Nanomed Nanobiotechnol. 2025; 17: e70019. https://doi.org/10.1002/wnan.70019

[3] Baird PA, Anderson TW, Newcombe HB, Lowry RB. Genetic disorders in children and young adults: a population study. American Journal of Human Genetics. 1988; 42: 677–693.

[4] Lee E, Orton K, Kwan E, Faga C, Le T, Shaheen R, et al. Couple-Based Carrier Screening: How Gene and Variant Considerations Impact Outcomes. Genes (Basel). 2025; 16: 671. https://doi.org/10.3390/genes16060671

[5] Peterlin B, Peterlin A. Carrier screening and pregnancy. Best Practice & Research. Clinical Obstetrics & Gynaecology. 2025; 100: 102601. https://doi.org/10.1016/j.bpobgyn.2025.102601

[6] Achterbergh R, Lakeman P, Stemerding D, Moors EHM, Cornel MC. Implementation of preconceptional carrier screening for cystic fibrosis and haemoglobinopathies: a sociotechnical analysis. Health Policy (Amsterdam, Netherlands). 2007; 83: 277–286. https://doi.org/10.1016/j.healthpol.2007.02.007

[7] Dondorp W, De Wert G, Pennings G, Shenfield F, Devroey P, Tarlatzis B, et al. ESHRE Task Force on Ethics and Law 21: genetic screening of gamete donors: ethical issues. Human Reproduction (Oxford, England). 2014; 29: 1353–1359. https://doi.org/10.1093/humrep/deu111

[8] Delatycki MB, Alkuraya F, Archibald A, Castellani C, Cornel M, Grody WW, et al. International perspectives on the implementation of reproductive carrier screening. Prenatal Diagnosis. 2020; 40: 301–310. https://doi.org/10.1002/pd.5611

[9] Franasiak JM, Olcha M, Bergh PA, Hong KH, Werner MD, Forman EJ, et al. Expanded carrier screening in an infertile population: how often is clinical decision making affected? Genetics in Medicine. 2016; 18: 1097–1101. https://doi.org/10.1038/gim.2016.8

[10] Hussein N, Henneman L, Kai J, Qureshi N. Preconception risk assessment for thalassaemia, sickle cell disease, cystic fibrosis and Tay-Sachs disease. The Cochrane Database of Systematic Reviews. 2021; 10: CD010849. https://doi.org/10.1002/14651858.CD010849.pub4

[11] Zhou J, Li G, Zeng Y, Qiu X, Zhao P, Huang T, et al. Screening primary carnitine deficiency in 10 million Chinese newborns: a systematic review and meta-analysis. Orphanet Journal of Rare Diseases. 2024; 19: 248. https://doi.org/10.1186/s13023-024-03267-x

[12] Abulí A, Boada M, Rodríguez-Santiago B, Coroleu B, Veiga A, Armengol L, et al. NGS-Based Assay for the Identification of Individuals Carrying Recessive Genetic Mutations in Reproductive Medicine. Human Mutation. 2016; 37: 516–523. https://doi.org/10.1002/humu.22989

[13] Committee Opinion No. 690 Summary: Carrier Screening in the Age of Genomic Medicine. Obstetrics and Gynecology. 2017; 129: 595–596. https://doi.org/10.1097/AOG.0000000000001947

[14] Aminbeidokhti M, Qu JH, Belur S, Cakmak H, Jaswa E, Lathi RB, et al. Preconception Genetic Carrier Screening for Miscarriage Risk Assessment: A Bioinformatic Approach to Identifying Candidate Lethal Genes and Variants. medRxiv. 2023. https://doi.org/10.1101/2023.05.25.23290518 (preprint)

[15] Zhang J, Wang Y, Ma D, Sun Y, Li Y, Yang P, et al. Carrier Screening and Prenatal Diagnosis for Spinal Muscular Atrophy in 13,069 Chinese Pregnant Women. The Journal of Molecular Diagnostics: JMD. 2020; 22: 817–822. https://doi.org/10.1016/j.jmoldx.2020.03.001

[16] Goldberg JD, Pierson S, Taber KJ. Expanded carrier screening: What conditions should we screen for? Prenatal Diagnosis. 2023; 43: 496–505. https://doi.org/10.1002/pd.6306

[17] Luo M, Liu L, Peter I, Zhu J, Scott SA, Zhao G, et al. An Ashkenazi Jewish SMN1 haplotype specific to duplication alleles improves pan-ethnic carrier screening for spinal muscular atrophy. Genetics in Medicine. 2014; 16: 149–156. https://doi.org/10.1038/gim.2013.84

[18] Tan L, Qi Y, Zhao P, Cheng L, Yu G, Zhao D, et al. Clinical application value of pre-pregnancy carrier screening in Chinese Han childbearing population. Molecular Genetics & Genomic Medicine. 2024; 12: e2425. https://doi.org/10.1002/mgg3.2425

[19] Hendrickson BC, Donohoe C, Akmaev VR, Sugarman EA, Labrousse P, Boguslavskiy L, et al. Differences in SMN1 allele frequencies among ethnic groups within North America. Journal of Medical Genetics. 2009; 46: 641–644. https://doi.org/10.1136/jmg.2009.066969

[20] Chokoshvili D, Vears D, Borry P. Expanded carrier screening for monogenic disorders: where are we now? Prenatal Diagnosis. 2018; 38: 59–66. https://doi.org/10.1002/pd.5109

[21] Hou W, Fu X, Xie X, Zhang C, Zhang M, Xiao R, et al. Tailoring monogenic disease carrier screening panels for Chinese populations: The importance of considering regional differences. Eu-

ropean Journal of Medical Genetics. 2025; 74: 105002. https://doi.org/10.1016/j.ejmg.2025.105002
[22] Yang J, Chen M, Wu H, Shen W, Han J, Fu Y, et al. Knowledge and attitudes toward expanded carrier screening between the medical staff and general population in China. European Journal of Obstetrics, Gynecology, and Reproductive Biology. 2021; 263: 198–204. https://doi.org/10.1016/j.ejogrb.2021.06.022
[23] Ben-Shachar R, Svenson A, Goldberg JD, Muzzey D. A data-driven evaluation of the size and content of expanded carrier screening panels. Genetics in Medicine: Official Journal of the American College of Medical Genetics. 2019; 21: 1931–1939. https://doi.org/10.1038/s41436-019-0466-5
[24] Lazarin GA, Haque IS, Nazareth S, Iori K, Patterson AS, Jacobson JL, et al. An empirical estimate of carrier frequencies for 400+ causal Mendelian variants: results from an ethnically diverse clinical sample of 23,453 individuals. Genetics in Medicine: Official Journal of the American College of Medical Genetics. 2013; 15: 178–186. https://doi.org/10.1038/gim.2012.114
[25] Edwards JG, Feldman G, Goldberg J, Gregg AR, Norton ME, Rose NC, et al. Expanded carrier screening in reproductive medicine-points to consider: a joint statement of the American College of Medical Genetics and Genomics, American College of Obstetricians and Gynecologists, National Society of Genetic Counselors, Perinatal Quality Foundation, and Society for Maternal-Fetal Medicine. Obstetrics and Gynecology. 2015; 125: 653–662. https://doi.org/10.1097/AOG.0000000000000666
[26] Capalbo A, Fabiani M, Caroselli S, Poli M, Girardi L, Patassini C, et al. Clinical validity and utility of preconception expanded carrier screening for the management of reproductive genetic risk in IVF and general population. Human Reproduction. 2021; 36: 2050–2061. https://doi.org/10.1093/humrep/deab087
[27] Zhao S, Xiang J, Fan C, Asan, Shang X, Zhang X, et al. Pilot study of expanded carrier screening for 11 recessive diseases in China: results from 10,476 ethnically diverse couples. European Journal of Human Genetics: EJHG. 2019; 27: 254–262. https://doi.org/10.1038/s41431-018-0253-9
[28] Chen SC, Zhou XY, Li SY, Zhao MM, Huang HF, Jia J, et al. Carrier burden of over 300 diseases in Han Chinese identified by expanded carrier testing of 300 couples using assisted reproductive technology. Journal of Assisted Reproduction and Genetics. 2023; 40: 2157–2173. https://doi.org/10.1007/s10815-023-02876-y
[29] Xi Y, Chen G, Lei C, Wu J, Zhang S, Xiao M, et al. Expanded carrier screening in Chinese patients seeking the help of assisted reproductive technology. Molecular Genetics & Genomic Medicine. 2020; 8: e1340. https://doi.org/10.1002/mgg3.1340
[30] Arjunan A, Torres R, Gardiner A, Kaseniit KE, Wootton J, Ben-Shachar R, et al. Evaluating the efficacy of three carrier screening workflows designed to identify at-risk carrier couples. Prenatal Diagnosis. 2021; 41: 896–904. https://doi.org/10.1002/pd.5900
[31] Peyser A, Singer T, Mullin C, Bristow SL, Gamma A, Onel K, et al. Comparing ethnicity-based and expanded carrier screening methods at a single fertility center reveals significant differences in carrier rates and carrier couple rates. Genetics in Medicine: Official Journal of the American College of Medical Genetics. 2019; 21: 1400–1406. https://doi.org/10.1038/s41436-018-0331-y
[32] Ropers HH. On the future of genetic risk assessment. Journal of Community Genetics. 2012; 3: 229–236. https://doi.org/10.1007/s12687-012-0092-2
[33] Brcic L, Underwood JF, Kendall KM, Caseras X, Kirov G, Davies W. Medical and neurobehavioural phenotypes in carriers of X-linked ichthyosis-associated genetic deletions in the UK Biobank. Journal of Medical Genetics. 2020; 57: 692–698. https://doi.org/10.1136/jmedgenet-2019-106676
[34] Schwartz CE, Dean J, Howard-Peebles PN, Bugge M, Mikkelsen M, Tommerup N, et al. Obstetrical and gynecological complications in fragile X carriers: a multicenter study. American Journal of Medical Genetics. 1994; 51: 400–402. https://doi.org/10.1002/ajmg.1320510419
[35] Allingham-Hawkins DJ, Babul-Hirji R, Chitayat D, Holden JJ, Yang KT, Lee C, et al. Fragile X premutation is a significant risk factor for premature ovarian failure: the International Collaborative POF in Fragile X study–preliminary data. American Journal of Medical Genetics. 1999; 83: 322–325.
[36] Rifé M, Nadal A, Milà M, Willemsen R. Immunohistochemical FMRP studies in a full mutated female fetus. American Journal of Medical Genetics. Part a. 2004; 124A: 129–132. https://doi.org/10.1002/ajmg.a.20342
[37] Elizur SE, Lebovitz O, Derech-Haim S, Dratviman-Storobinsky O, Feldman B, Dor J, et al. Elevated levels of FMR1 mRNA in granulosa cells are associated with low ovarian reserve in FMR1 premutation carriers. PloS One. 2014; 9: e105121. https://doi.org/10.1371/journal.pone.0105121
[38] Pouresmaeili F, Fazeli Z. Premature ovarian failure: a critical condition in the reproductive potential with various genetic causes. International Journal of Fertility & Sterility. 2014; 8: 1–12.
[39] Wapner RJ, Martin CL, Levy B, Ballif BC, Eng CM, Zachary JM, et al. Chromosomal microarray versus karyotyping for prenatal diagnosis. The New England Journal of Medicine. 2012; 367: 2175–2184. https://doi.org/10.1056/NEJMoa1203382
[40] Yu S, Bittel DC, Kibiryeva N, Zwick DL, Cooley LD. Validation of the Agilent 244K oligonucleotide array-based comparative genomic hybridization platform for clinical cytogenetic diagnosis. American journal of Clinical Pathology. 2009; 132: 349–360. https://doi.org/10.1309/AJCP1BOUTWF6ERYS
[41] Wallace DC. A mitochondrial paradigm of metabolic and degenerative diseases, aging, and cancer: a dawn for evolutionary medicine. Annual Review of Genetics. 2005; 39: 359–407. https://doi.org/10.1146/annurev.genet.39.110304.095751
[42] Schon EA, DiMauro S, Hirano M. Human mitochondrial DNA: roles of inherited and somatic mutations. Nature Reviews. Genetics. 2012; 13: 878–890. https://doi.org/10.1038/nrg3275
[43] Taylor RW, Turnbull DM. Mitochondrial DNA mutations in human disease. Nature Reviews. Genetics. 2005; 6: 389–402. https://doi.org/10.1038/nrg1606
[44] Suzuki T, Nagao A, Suzuki T. Human mitochondrial tRNAs: biogenesis, function, structural aspects, and diseases. Annual Review of Genetics. 2011; 45: 299–329. https://doi.org/10.1146/annurev-genet-110410-132531
[45] Guan MX. Mitochondrial 12S rRNA mutations associated with aminoglycoside ototoxicity. Mitochondrion. 2011; 11: 237–245. https://doi.org/10.1016/j.mito.2010.10.006
[46] Lv Y, Yan Y, Ma Y, Ye X, Wang S, Xing L. Effect of Family-Centered Group Health Education Among Patients Undergoing Preimplantation Genetic Testing: A Randomized Controlled Study. Clinical and Experimental Obstetrics & Gynecology. 2025; 52: 40748. https://doi.org/10.31083/CEOG40748