*Review Article*

# Pre-implantation Genetic Diagnosis: A Review

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**Abstract:**
*Preimplantation genetic diagnosis was developed nearly a quarter-century ago as an alternative form of prenatal diagnosis that is carried out on embryos. Initially offered for diagnosis in couples at risk for single gene genetic disorders, such as cystic fibrosis, spinal muscular atrophy and Huntington's disease, preimplantation genetic diagnosis (PGD) has most frequently been employed in assisted reproduction for detection of chromosome aneuploidy from advancing maternal age or structural chromosome rearrangements. Major improvements have been seen in PGD analysis with movement away from older, less effective technologies, such as fluorescence in situ hybridization (FISH), to newer molecular tools, such as DNA microarrays and next generation sequencing. Improved results have also started to be seen with decreasing use of Day 3 blastomere biopsy in favour of polar body or Day 5 trophectoderm biopsy. Discussions regarding the scientific, ethical, legal and social issues surrounding the use of sequence data from embryo biopsy have begun and must continue to avoid concern regarding eugenic or inappropriate use of this technology.*

**Keywords:** preimplantation genetic diagnosis, inherited genetic disorders, biopsy for PGD

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**Introduction:**
Pre-implantation genetic diagnosis (PIGD) is the genetic profiling of the embryos prior to implantation (as a form of embryo profiling), and sometimes even of oocytes prior to fertilization. PGD is considered in a similar fashion to prenatal diagnosis. The world's first PGD was performed in 1990 by Handyside, Kontogianni and Winston at the Hammersmith Hospital in London[1]. The term Pre-implantation genetic screening (PGS) refers to set of techniques for testing whether the embryos obtained through In Vitro Fertilization (IVF)/ Intra Cytoplasmic Sperm Insemination (ICSI) have abnormal chromosomes number.

The PGD allows studying the DNA of eggs or embryos to select those that carry certain mutation for genetic diseases. It is useful when there are previous chromosomal or genetic disorders in the family and within the context of IVF program[2]. Here a concise review was done on PGD/ PGS regarding its current status, both domestically and globally, as well as its future challenges.

**Historical Aspect:**
Pre-implantation genetic diagnosis (PIGD) was first introduced in 1990 by selecting female embryos in order to prevent the birth of male patients affected with X-Linked recessive disorders[3]. It is well recognized by the clinical community that it is indicated in preventing monogenic inherited disorders with severe morbidity and mortality[4].

**Indications and Applications:**
PGD is used primarily for genetic disease prevention, by selecting only those embryos that do not have a known genetic disorder. PGD may also be used to increase chances of successful pregnancy, to match a sibling in Human leukocyte antigen (HLA) type in order to be a donor, to have less cancer pre-deposition, and for sex selection[5-8].

**Monogenic Disorders:**
It is also available for large numbers of monogenic disorders that is, disorders due to a single gene only (autosomal recessive, autosomal dominant or X-linked) or of chromosomal structural aberrations (such as balanced translocation). The most frequently diagnosed autosomal recessive disorders are cystic fibrosis, beta-thalassemia, sickle cell disease and spinal muscular atrophy type-1. The most common dominant diseases are myotonic dystrophy, Huntington's disease and Charcot-Marie-Tooth disease and in the case of X-linked diseases, most of the cycles are performed for Fragile X syndrome, haemophilia A and Duchenne muscular dystrophy.

**HLA Matching:**
Human leukocyte antigen (HLA) typing of embryos, so that the child's HLA matches a sick sibling,

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availing for cord-blood stem cell donation[9]. The child is in this sense a "Savior sibling" for the recipient child. HLA typing has meanwhile become an important PGD indication in those countries where the law permits it[10].

**Cancer Predisposition:**
A more recent application of PGD is to diagnose Late-onset diseases and cancer predisposition syndromes. Since affected individuals remain healthy until the onset of the disease, frequently in the fourth decade of life, there is debate on whether or not PGD is appropriate in these cases.

**Sex Discernment:**
Pre-implantation genetic diagnosis provides a method of prenatal sex discernment even before implantation, and may therefore be termed preimplantation sex discernment. Potential applications of preimplantation sex discernment include:

- A complement to specific gene testing for monogenic disorders, which can be very useful for genetic diseases whose presentation is linked to the sex, such as, for example, X-linked diseases.
- Sex selection: Most clinics perform it only for "family balancing", which is where a couple with two or more children of one sex desire a child of the other, but half do not restrict sex selection to family balancing. In India, this practice has been used to select only male embryos although this is illegal[11].

**PGD to Rescue Siblings:**
In recent and highly publicized applications, parents of children with fatal disorders have undergone IVF and PGD to select embryos that can provide bone marrow transplants for the sick child. This use of PGD has a less-exacting predecessor. In the 1980s, several families conceived offspring with the hope of having a child that could provide bone marrow for his or her sibling.

**Timing of Biopsy:**
Despite the timing of biopsy included polar body, cleavage-stage embryos, morula stage embryos, it is now more recognized that biopsy during the blastocyst stage by aspirating the trophectoderm cells are safer. The human blastocyst contains approximately 130 cells distributed between the inner cell mass, which will develop into the fetus proper, and the surrounding trophectoderm cells, which will become the placenta and fetal membranes.

**Technical Aspect:**
PGD is a form of genetic diagnosis performed prior to implantation. This implies that the patient's oocytes should be fertilized in-vitro and the embryos kept in culture until the diagnosis is established. It is also necessary to perform a biopsy on these embryos in order to obtain material on which to perform the diagnosis. Generally, PCR-based methods are used for monogenic disorders and FISH for chromosomal abnormalities and for sexing those cases in which no PCR protocol is available for X-linked disease.

**Ethical Issues:**
PGD has raised ethical issues, although this approach could reduce reliance on fetal deselection during pregnancy. The technique can be used for prenatal sex discernment of the embryo, and thus potentially can be used to select embryos of one sex in preference of the other in the context of "Family balancing". It may be possible to make other "social selection" choices in future that introduce socio-economic concerns. Only unaffected embryos are implanted in a women's uterus, those that are affected are either discarded or donated to science[12].

PGD has the potential to screen for genetic issues unrelated to medical necessity, such as intelligence and beauty, and against negative traits such as disabilities. The medical community has regarded this as counterintuitive and controversial suggestion[13]. The concept of a "designer baby" is closely related to the PGD technique, creating a fear that increasing frequency of genetic screening will move toward a modern eugenics movement[14].

**Discussion:**
Preimplantation genetic diagnosis (PGD) is a form of prenatal diagnosis that is performed on early embryos created by in vitro fertilization (IVF). In comparison to other established methods of prenatal diagnosis, such as chorionic villus sampling and amniocentesis, PGD is not performed on an outgoing intrauterine pregnancy in the late first or early second trimester, but on embryos developing in the IVF laboratory prior to transfer to the uterus.

Despite some misconception to the contrary, PGD is not a therapeutic procedure for embryo; there are no change to the DNA or any other genetic-related structures. It is solely a diagnostic procedure that can identify whether a specific embryo carries a single gene disorder for which the couple is at-risk or a chromosomal abnormality that could lead to either failed implantation, subsequent miscarriage or the birth of a child with physical and/or developmental disability.

In developed countries, genetically determined disorder accounts for up to one third of admission to pediatric wards and are a significant cause of childhood deaths. The Human Genome Project and related advances in molecular biopsy becoming the promising means for the long term curative treatment of many severe genetic disorders[15]. The

current approach for controlling these disorders remains prevention, including application of prenatal diagnosis (PND) which is an accepted procedure in most populations[16]. PGD aims to provide an accurate, rapid result as early in pregnancy as possible. A prerequisite involve obtaining fetal material promptly and safely. Current methods include trophoblast sampling and amniocentesis.

Fetal cells and free fetal DNA are also present in the circulation of the pregnant mother and provided a potential source for "non-invasive" fetal sampling, but reliable protocols have yet to be established for clinical application[17,18]. As data have accumulated from chromosomal analysis of human preimplantation embryos, it has become apparent that there is higher rate of chromosomal abnormalities in cleavage stage embryos and blastocyst detected by FISH[19,20,21]. Results show that only a minority (<35%) of human embryos derived from IVF have a normal chromosome complement in all cells, with the remaining embryos observed to be abnormal non-mosaic, mosaic[22,23].

The positive outcome of any PGD cycle, that is, the birth of the healthy unaffected baby, depends upon success at each of the multiple stages of the assisted reproductive procedure, as well as an accurate genetic diagnosis. Generally about 70% of all oocytes collected will fertilize and about 70% of all these will develop to the cleavage stage, of which not all will be suitable for biopsy. PGD is successful in about 80-90% of successfully biopsied embryos and about half of these are diagnosed as suitable for transfer (unaffected).

Reported pregnancy rates vary, but rarely surpass about one third of all cycles initiated[24,25]. The safety of PGD for children born is a major concern, but initial evaluation of about 250 babies born worldwide after PGD indicated that the procedure had no adverse consequence on early development[26,27]. There is also public concern about the use of PGD to prevent the birth of children with the severe genetic disorders, there are few countries which has begun to offer PGD for "social" sexing. Thus, it is imperative to establish appropriate ethical guidelines and legislation as soon as possible.

**Conclusion:**

The initial concept of PGD appeared fairly simple; especially following the development of PCR based DNA methodologies. However, a decade of practical application has proven that this is not the case, and although experience, research efforts, and some technological advances have led to many improvements. PGD remains a technically challenging, multistep, labour intensive procedure which requires the close collaboration of a team of specialists. Efforts continue to ameliorate and simplify protocols, particularly for genetic analysis and to develop methods for more disorders, but present technologies still limit wider application.

**References:**

1. Handyside AH, Kontogianni EH, Hardy K, Winston RM. Pregnancies from biopsied human preimplantation embryo sexed by Y-specific DNA amplification. Nature. 1990; 344 (6268): 768-70.

2. Latham SR. The once and future debate on human embryonic stem cell research. Yale J Health Policy Law Ethics. 2009; 9 (Suppl): 483-94.

3. Kuliev A, Rechitsky S. Preimplantation genetic testing: current challenges and future prospects. Expert Rev Mol Diagn. 2017; 17 (12): 1071-88.

4. Geraedts JP, De Wert GM. Preimplantation genetic diagnosis. Clin Genet. 2009; 76 (4): 315-25.

5. Sullivan-Pyke C, Dokras A. Preimplantation genetic screening and preimplantation genetic diagnosis. Obstet Gynecol Clin North Am. 2018; 45 (1): 113-25.

6. Handyside AH. 'Designer babies' almost thirty years on. Reproduction. 2018; 156 (1): F75-F79.

7. Iews M, Tan J, Taskin O, Alfaraj S, AbdelHafez FF, Abdellah AH, et al. Does preimplantation genetic diagnosis improve reproductive outcome in couples with recurrent pregnancy loss owing to structural chromosomal rearrangement? A systematic review. Reprod Biomed Online. 2018; 36 (6): 677-85.

8. Mastenbroek S, Twisk M, van der veen F, Repping S. Preimplantation Genetic Screening: A systematic review and meta-analysis of RCTs. Hum Reprod Update. 2011; 17 (4): 454-66.

9. Verlinsky Y, Rechitsky S, Schoolcraft W, Strom C, Kuliev A. Preimplantation diagnosis for Fanconi anemia combined with HLA matching. JAMA. 2001; 285 (24): 3130-3.

10. Baruch S, Kaufman D, Hudson KL. Genetic testing of embryos: practices and perspectives of US in vitro fertilization clinics. Fertil Steril. 2008; 89 (5): 1053-8.

11. Fragouli E, Wells D. Current status and future prospects of noninvasive preimplantation

genetic testing for aneuploidy. Fertil Steril. 2018; 110 (3): 408-9.

12. Braude P, Pickering S, Flinter F, Ogilvie CM. Preimplantation Genetic Diagnosis. Nat Rev Genet. 2002; 3 (12): 941-53.

13. Robertson JA. Extending Preimplantation Genetic Diagnosis: the ethical debate. Ethical issues in new uses of Preimplantation Genetic Diagnosis. Hum Reprod. 2003; 18 (3): 465-71.

14. Savulescu J. Procreative beneficence: why we should select the best children. Bioethics. 2001; 15 (5-6): 413-26.

15. Savulescu J, Kahane G. The moral obligation to create children with the best chance of the best life. Bioethics. 2009; 23 (5): 274-90.

16. World Health Organization. Community control of genetic and congenital disorders. Available at: https://apps.who.int/iris/handle /10665/119571. [Accessed on March 8, 2018]

17. Cheung LP. Patient selection for assisted reproductive technology treatments. Hong Kong Med J. 2000; 6 (2): 177-83.

18. Rutherford AJ, Subak-Sharpe R, Dawson KJ, Margara RA, Franks S, Winston RM. Improvement of invitro fertilization after treatment with buserelin, an agonist of luteinising hormone releasing hormone. Br Med J (Clin Res Ed). 1988; 296 (6639): 1765-1768.

19. Ray PF, Ao A, Taylor DM, Winston RM, Handyside AH. Assessment of the reliability of single blastomere analysis for preimplantation diagnosis of the delta F508 deletion causing cystic fibrosis in clinical practice. Prenat Diagn. 1998; 18 (13): 1402-12.

20. Rechitsky S, Strom C, Verlinsky O, Amet T, Ivakhnenko V, Kukharenko V, et al. Allele drop out in polar bodies and blastomeres. J Assist Reprod Genet. 1998; 15 (5): 253–7.

21. Kanavakis E, Traeger-Synodinos J. Pre-implantation genetic diagnosis in clinical practice. J Med Genet. 2002; 39 (1): 6-11.

22. Ruangvutilert P, Delhanty JD, Serhal P, Simopoulou M, Rodeck CH, Harper JC. FISH analysis on day 5 post-insemination of human arrested and blastocyst stage embryos. Prenat Diagn. 2000; 20 (7): 552–60.

23. Wells D, Delhanty JD. Comprehensive chromosomal analysis of human preimplantation embryos using whole genome amplification and single cell comparative genome hybridization. Mol Hum Reprod. 2000; 6 (11): 1055-62.

24. Cieslak J, Ivakhnenko V, Wolf G, Sheleg S, Verlinsky Y. Three-dimensional partial zona dissection for preimplantation genetic diagnosis and assisted hatching. Fertil Steril. 1999; 71 (2): 308-13.

25. Sermon K, Lissens W, Messiaen L, Bonduelle M, Vandervorst M, Van Steirteghem A, et al. Preimplantation genetic diagnosis of Marfan syndrome with the use of fluorescent polymerase chain reaction and the automated laser fluorescence DNA sequencer. Fertil Steril. 1999; 71 (1): 163-6.

26. Strom CM, Levin R, Strom S, Masciangelo C, Kuliev A, Verlinsky Y. Neonatal outcome of preimplantation genetic diagnosis by polar body removal: the first 109 infants. Pediatrics. 2000; 106 (4): 650-3.

27. Eldar-Geva T, Srebnik N, Altarescu G, Varshaver I, Brooks B, Levy-Lahad E, et al. Neonatal outcome after preimplantation genetic diagnosis. Fertil Steril. 2014; 102 (4): 1016-21.

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