

PREIMPLANTATION GENETIC DIAGNOSIS FACT SHEET

Preimplantation Genetic Diagnosis (PGD) is a relatively new procedure and is performed in conjunction with in vitro fertilization (IVF). PGD helps in detecting the genetic abnormalities either at gene level or at chromosome level before implantation thereby avoiding the transfer of genetically affected embryos. At Strong Fertility Center, We offer PGD for three different conditions; PGD for single gene defects, PGD for aneuploidy screening and PGD for chromosome translocation.

PGD for aneuploidy screening (PGD-AS)

This is the most common type of PGD analysis to test the embryos using a panel of chromosomes that are commonly involved in miscarriages or trisomic pregnancies. Embryos screened this way may have a higher rate of implantation, lower spontaneous loss and a reduced risk of trisomic offspring (e.g., Down's syndrome).

- A normal human cell contains 46 chromosomes or 23 pairs of chromosomes. These chromosomes are string like structures that resides in the center of each cell or nucleus and carry the genetic information.
- 22 pairs of chromosomes are called autosomes which are same in men and women. 23rd pair of chromosome is called sex chromosomes. Women normally have two of the same sex chromosomes, called the X chromosome, while men normally have 2 different sex chromosomes, known as X and Y chromosomes.
- Sperm and eggs contain half of the total number of 46 chromosomes or 23 chromosomes each.
- A normal fertilized embryo is derived by the fusion of 23 chromosomes from eggs and 23 chromosomes from sperm.
- Abnormal cell division in sperm or eggs might result in greater or fewer chromosomes than the normal 23. Therefore, any embryo that is derived from these sperm or eggs will carry extra or missing chromosomes, an abnormal condition referred as *aneuploidy*.
- In approximately, 70% of recurrent miscarriages abnormal numbers (*aneuploidy*) of chromosomes are identified.
- Some of the most common chromosome abnormalities found in miscarriages are trisomy 16 (3 copies of chromosome 16); trisomy for chromosome 22, 21, 15, 18 or 13; triploidy (3 copies of all the chromosomes); and abnormalities of the sex chromosomes.
- Chromosomally abnormal embryos usually fail to implant in the uterus and if implants may not develop normally and pregnancies miscarry and in some instances pregnancy can develop to term and give birth to trisomic babies (e.g., Down's Syndrome).
- The percentage of chromosomally abnormally embryos that each couple produces varies depending on their clinical status.

- Factors such as advanced maternal age (>35 years), the number of failed IVF cycles, miscarriages during normal conception and quality of sperm all influence the proportion of embryos that are abnormal.
- Any deviation from having 2 copies of each chromosome is considered abnormal. If only one of two chromosomes is identified the embryo is considered *monosomy* and if 3 chromosomes is identified it is considered *trisomy*. Both these conditions are abnormal and not suitable for embryo transfer.
- For PGD-AS embryos created by IVF are cultured in the laboratory for 3 days to grow ideally to an 8-cell stage. At that point one or two cells are removed by making a hole on the outer shell of the embryo. This procedure is called embryo biopsy. The biopsied embryo is returned back to culture until the result for that cell/cells are obtained usually on 5th day after egg retrieval.
- Removal of one or two cells from an eight cell embryo does not compromise the embryonic development.
- It is important that the embryo to be biopsied should have at least 5-cell and minimal fragmentation on the 3rd day of embryonic development. If the embryo contains too few cells, a biopsy might jeopardize the viability of the embryo.
- The nuclear material from the biopsied cell is sent to a reference lab. The reference lab performs a procedure called Fluorescent in situ hybridization (FISH) to determine the chromosome status of each cell.
- Our reference lab uses a mixture of FISH probes in one or two sequential hybridizations. Possible chromosomes to be tested include 13, 15, 16, 18, 21, 22, X and Y. Abnormalities in these chromosomes are found commonly in miscarriages and abnormal live births.
- A normal cell should show 2 copies of FISH signals for each of the numbered chromosomes, and either 2X signals for females or 1X and 1Y signals for males.
- There may be situations where the result cannot be obtained or is incorrect. Some of these situations include:
 - If the cell is lost during fixation or nucleus is ruptured no result will be obtained.
 - Occasionally the chromosomes in the nucleus might not have spread properly and lying on top of each other which would underestimate the number of chromosomes and might lead to a false result.
 - The FISH probe may fail to bind to a chromosome suggesting a missing chromosome.
 - If embryo biopsy happens when a particular cell is in the middle of cell division then the result appears to be two nuclei and four sets of chromosomes, FISH analysis from these cells can be difficult to interpret.
- A number of factors influence the success of a PGD/IVF cycle. Current data suggests that PGD cycles are most successful when 8 or more embryos are created by IVF and at least 5 are of good quality as graded by the embryology lab based on the number of cells, fragmentation and uniformity in cell size.

- PGD usually does not increase the pregnancy rates but it reduces the miscarriages and the incidence of trisomic pregnancies.
- PGD is most successful in the patients who have more than 3 embryos that do not show abnormalities and at least two of them are developed to the blastocyst stage.

PGD for single gene mutation:

This is indicated for the patients where both partners carry a gene for an autosomal recessive disorder or one partner may carry a gene for a dominant disorder and this increases the risk of conceiving a child with severe genetic disorder, e.g., Duchenne's muscular dystrophy, Cystic Fibrosis, Tay Sachs etc. Currently this list includes more than fifty disorders. This procedure involves prior testing to detect the gene mutation in the patient and partner so that a gene probe can be produced. There are several possible reasons for failure to achieve a successful identification of the gene or to achieve pregnancy. These include:

- Possible failure to amplify the gene in question or degraded nuclear material that does not allow for a clear DNA signal.
- PGD for single gene mutation can only detect the embryos that are carrying the mutation irrespective of the implantation potential of the embryo.

PGD for chromosomal translocation or structural abnormalities:

Chromosomal translocations involve a rearrangement of the chromosome material so that some of the genetic material from one chromosome is located on another one. This is known as a "balanced" translocation where all of the normal genetic material is present.

- If one partner carries a balanced chromosome rearrangement, he or she can produce sperms or eggs that can contain extra or missing segment of a particular chromosome material. These may produce conceptions which contain "unbalanced" translocation where there is either excess or deficient total genetic material. This can result in failed early embryonic development, recurrent pregnancy loss or birth of a child with mental and/or physical defects.
- Not all translocations or structural rearrangements can be tested by PGD. The reference lab would need to analyze the karyotype from the blood of both partners prior to start the IVF cycles to check the feasibility of performing the PGD for that anomaly.
- Although medical evidences show that PGD in couples who carry chromosomal translocations helps in reducing the pregnancy loss but it can not eliminate the miscarriages due to other factors.
- PGD for translocation can be carried out in conjunction with aneuploidy screening using the limited number of FISH probes.

IMPORTANT SUMMARY POINTS ABOUT PGD

- PGD-AS is a screening, NOT diagnostic test. It allows for screening out of the most common aneuploid birth defects and may reduce miscarriage rate.
- PGD of any type may not increase pregnancy rates and in some cases may reduce the chances for pregnancy due to decreased embryos available for transfer.
- PGD may reduce the opportunity for cryopreservation of embryos.
- PGD may result in NO embryos for transfer.
- In cases of low embryo number or poor embryo progression, plan for PGD may be cancelled.