

CONSENT FOR PREIMPLANTATION GENETIC DIAGNOSIS (PGD)

This consent form may contain words that you do not understand. Please ask the study doctor or the study staff to explain any words or information that you do not clearly understand.

You will receive a copy of this consent form for your records.

DESCRIPTION/PURPOSE

Preimplantation genetic diagnosis (PGD) is the chromosomal analysis of your eggs or embryos that may result from your in-vitro fertilization procedure. Genetic errors, such as changes in chromosome numbers, or aneuploidy, and changes in chromosome configuration will be studied. Aneuploid embryos are those with either a missing chromosome (monosomy) or an extra chromosome (trisomy). Aneuploidy occurs more frequently in eggs and embryos in women over 34 years of age. Changes in chromosome configuration include translocations. Couples in whom one or both partners have a known translocation can benefit. Translocation testing can determine the presence or absence of a certain chromosomal disorder, but cannot detect genetic disease nor predict congenital malformation. The study doctors and scientists at RBA hope to determine the genetic health of your eggs or embryos prior to transfer. The purpose is to select and replace only those embryos that appear to be chromosomally normal, so that there will be a reduced probability of losing the pregnancy or carrying a chromosomally abnormal baby to term.

The purpose of this procedure is to select and replace only embryos which do not have certain known chromosomal abnormalities.

OVERALL RATIONALE FOR UNDERTAKING THE STUDY

Chromosomes are the elements within every cell of your body that contain genetic information. Chromosomes are string-like structures found in the center of the cell, the nucleus. Inherited information is housed on the chromosomes. The traits are located in the genes that make up the chromosomes. Aneuploidies, or errors in development, occur more frequently in eggs and embryos in women over 34 years of age. Normally, there are 23 identical pairs in each cell, with a total of 46 chromosomes. Each patient provides 23 chromosomes, but in some children there is an extra chromosome: this is called trisomy. The most well-known trisomy is trisomy 21, also called Down's syndrome. Trisomic embryos usually do not implant, but if they do, this may lead to affected children. These extra chromosomes are usually formed during the final stages of egg ripening. The chromosomes mostly affected are chromosome numbers 13, 15, 16, 17, 18, 21, and 22.

A translocation is a change in chromosome configuration in which chromosomes are attached to each other or pieces of different chromosomes have been interchanged. An individual with a translocation is unaffected if there is no extra or missing chromosome material and if the break in the chromosome did not disrupt gene function. If there is no additional or missing chromosome material, the translocation is considered to be "balanced". A translocation is "unbalanced" if there is extra or missing material. Individuals with balanced translocations typically have no medical issues though some do have fertility problems such as reduced fertility. The concern regarding

having a balanced translocation is that, though the individual is healthy, the egg or sperm of that individual can have an unbalanced chromosome make-up that leads to the resultant embryo or pregnancy being unbalanced. The presence of an unbalanced translocation can lead to an embryo not implanting, a pregnancy being lost or a child being born with mental and physical problems. As such, individuals with a translocation may experience multiple pregnancy losses or have a child affected with physical and mental problems that may be lethal. Carriers of balanced translocations may still be affected by cryptic congenital malformations, unrelated or related to the inherited condition. The same may be the case when embryos or fetuses are diagnosed as balanced. The child may still be adversely affected during gestation (*congenital malformation*). It is postulated that there may be an increase in congenital malformations in individuals with balanced translocation.

There are two types of translocations. Approximately 1 in 625 individuals have a *reciprocal* translocation. These translocations involve any of the chromosomes. *Reciprocal* defines the translocation as one in which chromosomes have swapped material. Breaks occur anywhere in the chromosomes allowing for pieces to be interchanged between chromosomes. The second type of translocation is so-called *Robertsonian*. Approximately 1 in 900 individuals have this translocation. These translocations involve chromosomes 13, 14, 15, 21 and 22. These chromosomes have a unique structure in that they are primarily made of a bottom half of a chromosome. This translocation results from fusion of 2 of these chromosomes such that the two bottoms are attached.

OBJECTIVES

1. The study doctors and scientists at Reproductive Biology Associates (RBA) hope that chromosome testing of your eggs and embryos will increase your likelihood of becoming pregnant.
2. They also hope that the test will substantially reduce the chance of conceiving a baby with certain chromosomal abnormalities that occur more frequently in women over 34 years of age.
3. Observation

Chromosomes will be tested either by removing and testing polar bodies, small cells which are the by-product from the ripening process of the egg, or by removing and testing one or two cells from embryos which have divided satisfactorily. When testing for aneuploidy, or when the person with the translocation is the female partner, we may be able to analyze the *polar body*. By analyzing polar bodies, we obtain information only from the mother. Chromosome abnormalities that may occur after fertilization, when the sperm meets the egg, will not be detected. Polar bodies are discarded by the embryo itself during the normal process of fertilization. They contain exactly as many chromosomes as the egg itself. To test the polar body, an opening is made in the covering of the egg and the polar body is removed with a pipette. The polar body is then analyzed while the egg is placed in an incubator.

The chromosomes present in the cells (blastomeres) extracted from the divided embryo also provide a diagnosis for the genetic status and health of the embryo. The tests are performed

exclusively on the removed cell, and the embryo itself remains unaffected. *Blastomeres* are analyzed for abnormal chromosome number, when the male has the translocation and, in certain cases, when the female has the translocation. The PGD team of doctors, geneticists and embryologists will decide which method to use depending on the type of translocation, and other considerations.

Chromosome testing of blastomeres involves removing one cell (blastomere) with a micropipette from the embryo on day 3; at this stage the embryo usually has 6 to 10 identical cells, each with a full complement of chromosomal material. The embryo(s) remain in incubation while the cell is tested. Normally only a single cell is removed by biopsy from each embryo, as it is expected to be identical to all the other cells. In some cases, it may be necessary to remove a second cell according to circumstances. In either of the above cases, the analysis of the biopsied cell(s) uses a technique called fluorescence in-situ hybridization or FISH, which takes about one day. The cells are glued to a glass slide and heated and cooled and their DNA is 'labeled' with colored fluorescent dyes called probes, one for each chromosome sought (13, 15, 16, 17, 18, 21, 22, X and Y) or part of the chromosome depending on whether you are having PGD for a translocation. Once complete, the geneticist counts the colors and the position of the spots using a powerful microscope, thereby distinguishing normal and abnormal cells. After this process the cells are no longer viable in any way, and the slides on which they sit are placed in a deep freeze for future reference.

Some patients will wish to choose PGD for the purpose of sex selection only. RBA will consent to performing PGD for sex selection if the following criteria are met:

1. Patient(s) must agree to cryopreserve genetically normal embryos of the undesired sex.
2. Patient(s) must agree to transfer these cryopreserved embryos to the female partner's uterus within 5 years of their creation OR patient(s) must agree to donate these embryos to another couple either as a directed donation or anonymously.
3. Embryos may not be destroyed or discarded.

RBA reserves the right to charge appropriate fees for cryopreservation and storage of the embryos found to have the undesired sex. (See addendum for fees)

About 90% of abnormal embryos can now be detected using the techniques which will be applied to your eggs and embryos. Prenatal testing after the IVF cycle is still strongly advised, since this would confirm the prognosis. Because of the risks described below, your pregnancy will be carefully monitored as should all pregnancies after IVF procedures. Between 10 to 16 weeks, we will recommend chorionic villus sampling or amniocentesis, where samples of cells are taken from the fluids or placenta beside the developing fetus for similar testing. These tests will provide a comprehensive genetic analysis of the fetus. The fetus will also be monitored with ultrasound to detect its growth and development.

RISKS

1. The process takes up to 24 hours, and results will be brought immediately to the patient couple for consultation and discussion of the outcome. When possible, eggs or embryos diagnosed as abnormal, not chosen for replacement, will again be fully tested to confirm the original diagnosis. Complete testing will, in a majority of cases (more than 95%), eliminate all excess embryos. Most patients will have all embryos that are normal by PGD, and viable by microscopic appearance, transferred, and excess abnormal or non-viable embryos will be discarded. Occasionally (less than 5% of the time), there may be more embryos that test normal than are safe to transfer. In these cases the biopsied normal embryos will be allowed to grow in the laboratory for 1-2 more days. If they progress to the next stage of development, the blastocyst stage, they may be cryo-preserved at that time for later use by the couple. The slides containing the tested cells will be frozen for future reference and the patient-couple fully informed of the results upon request.
2. It should be understood that only a subset of the chromosomes that are present in each biopsied cell can be diagnosed. Sometimes chromosome anomalies are present in a cell, yet not in other cells of the same embryo and vice-versa, in a condition called mosaicism. Therefore, analysis of a single cell has limitations, and wrong diagnoses do happen, either false-positive or false-negative, in as much as ten percent (10%) of embryos. Geneticists are unable to detect other genetic abnormalities. Other congenital malformations or genetic disease cannot be tested using FISH. As with all IVF pregnancies, prenatal testing should be performed by chorionic villus sampling or amniocentesis to confirm the development of a normal fetus. At this time, PGD cannot be considered a substitute for these tests.
3. Physicians and scientists of RBA are uncertain of the risks involved in microsurgery on the embryos, but believe them to be acceptably low. Numerous animal studies and some human studies show that the microsurgery of the embryo needed to remove the cells, does not affect the normal development of the baby. This procedure, however, has been performed in a limited number of studies on human embryos, so the precise negative effects, if any, are unknown. In animal studies there have been no apparent problems and preliminary evidence with human eggs and embryos suggests that this is also true. Even though there have been more than 200 live births after PGD of aneuploidy world wide to date (May 2001) also involving biopsy, the biopsy procedure is still relatively new; and therefore the major risk is that the procedure will not be successful in spite of all best efforts. Although a rare occurrence (0.1%), it is possible that some or all egg(s) or embryo(s) may be accidentally damaged during biopsy. Furthermore, a relatively large number of the eggs or embryos may be abnormal providing a very limited number of embryos for replacement. It is possible that in some cases, none of the embryos may be normal, and embryo replacement should then not be performed. It is also possible that no reading is available on one or more embryos. While this is a disappointing outcome, it is likely that the cycle would have failed without PGD or an abnormal conception would have occurred.

4. Finally, the tests may fail in any individual case because of unforeseen technical malfunctions. It is therefore not possible to guarantee pregnancy after PGD for aneuploidy or translocation or even to promise that there will be benefits for any individual case.
5. For translocations, there is a risk of congenital abnormalities not produced by the translocation that may be higher than that found in the general population. Physicians and scientists of RBA are particularly concerned with these findings and emphasize the experimental nature of this procedure. You may have an increased risk of congenital malformation in your child when consenting to this procedure.
6. When any pregnancy is achieved after an IVF procedure, physicians and scientists of RBA strongly recommend a comprehensive genetic analysis of the fetus between 10-18 weeks. This can be chorionic villus sampling or amniocentesis, which provide samples of cells taken from the fluids of the placenta. The risks and benefits of this testing should be discussed with your obstetrician. The fetus should also be checked with ultrasound to monitor growth and development. Physicians and scientists of RBA sincerely request that these results be forwarded to our IVF Lab Director, Zolt Peter Nagy, Ph.D., telephone (404) 257-1840. Similarly, after birth and periodically thereafter, we will send you a questionnaire that will inform the team of the progress of your child.
7. If you participate in this study, you will not be exposed to any further risks of physical injury or discomfort other than those already connected with standard IVF procedures.

RBA (RBA) is collaborating with scientists and physicians in other centers who have performed PGD in over 1000 cycles so far. Some genetic testing may be done at RBA while some cases will sent to a collaborating center. RBA will notify you in advance if your testing must be done elsewhere.

ALTERNATIVE TREATMENT

Similar techniques can be used to test any embryos which might be affected by a genetic disease known to be present in your family. Such procedures are applied in order to select embryos which are not affected by the disease.

You can choose to have no genetic diagnosis performed on the eggs or embryos, and to have the embryos transferred. In that case, the only way of determining possible genetic deformities during early pregnancy would be by testing through chorionic villus sampling or amniocentesis from 10 weeks gestation onwards. You do not have to participate in this study in order to have IVF.

COSTS

You (or your insurance) will be responsible for the costs of the study procedure as described above. Fees for PGD for aneuploidies or translocation are in addition to the cost of the IVF cycle, over and above those related to your normal IVF and/or egg donation procedure. If the PGD of translocation procedure is paid for but not performed, your payment will be refunded.

VOLUNTARY PARTICIPATION/WITHDRAWAL

Your decision whether or not to have translocation testing will not prejudice your future relations with RBA and the treatment you are now undergoing at this site. If you decide to participate, you are free to discontinue participation at any time. By signing this consent you have not waived any other legal rights, which you would otherwise have as a patient, or a subject in a research study or released any party from liability for negligence. Your participation is voluntary and your refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled.

COMPENSATION FOR INJURY

In accordance with Federal regulations, we are obliged to inform you about RBA's policy in the event physical injury occurs. If, as a result of your participation, you experience physical injury from known or unknown risks of the research procedures as described, immediate medical care and treatment, including hospitalization if necessary, will be available. No monetary compensation, however, is available and you will be responsible for the costs of such medical treatment, either directly, or through your medical insurance and/or other forms of medical coverage. Further information can be obtained by calling Zolt Peter Nagy, Ph.D., at (404) 257-1840.

QUESTIONS

If you have any questions concerning your participation in this study, or if at any time you feel you have experienced a research-related injury or a reaction to the study procedure, contact IVF Lab Director, Zolt Peter Nagy., Ph.D., telephone (404) 257-1840 or Daniel B. Shapiro, M.D., telephone 404-257-1900.

Do not sign this form unless you have had a chance to ask questions and have received satisfactory answers to all of your questions.

CONSENT

I have read and I understand all of the preceding information describing this study and all my questions regarding the study and my participation in it has been answered to my satisfaction. I freely give my consent to participate in this study until I decide otherwise.

I understand that I will receive a copy of this consent form.

By signing this form I have not waived any of the legal rights which I otherwise would have as a participant in a research study.

Signature of female partner

Date

Signature of witness

Date

Signature of male partner

Date

Signature of witness

Date

Signature of Doctor

Date