Pre-implantation Genetic Testing for Aneuploidies (PGT-A)

Information for patients
Welcome

This booklet informs you how a PGT-A cycle is managed at Leeds Fertility (LF).

You can find further information at www.leedsfertilityclinic.co.uk and at our regular Open Evenings Presentation, which is also available to view online (see website or Reception notice board for up-coming dates and details).

How to contact us:

Please see page 28 for urgent and non-urgent contact details.
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What is PGT-A?

Pre-implantation genetic testing for aneuploidies (PGT-A) (formerly known as pre-implantation genetic screening (PGS)) is a test used alongside IVF. It is a selection tool to increase the chance of a healthy on-going pregnancy. It is not a guarantee of a pregnancy.

The success of IVF depends on a number of factors including a woman’s age, fertility diagnosis, number of embryos produced, and embryo quality. PGT-A provides important information about chromosomal health of embryos before transfer so we can select the embryo most likely to lead to a successful pregnancy.

A few cells are removed from each embryo created in an IVF cycle and the number of chromosomes in each embryo cell are counted. An embryo with a normal number of chromosomes can therefore be chosen to be transferred, and this is thought to give a higher chance of creating a healthy baby. By excluding the abnormal embryos, PGT-A helps to prevent the possible transfer of embryos which would end up not implanting, miscarrying or resulting in the birth of a child affected by an abnormal number of chromosomes. However, this screening may also result in no embryos being found that are normal so no transfer can take place.

In the UK, it is illegal to use this testing to select embryos on the basis of their sex for social/family reasons.
Situations where PGT-A may help

PGT-A may be offered to couples who have a higher chance of having a chromosomal problem in their embryos, as they are most likely to benefit.

These are:

- Couples with a history of recurrent miscarriages.
- Couples who have had several failed IVF attempts.
- Couples who have had previous pregnancies affected by chromosome abnormalities.
- Women over the age of 35 years requesting PGT-A.

Theoretically, PGT-A may reduce these risks, although this has not been conclusively proven. Overseas clinics have begun to offer PGT-A to all patient groups, outside of the above criteria, to help improve outcomes.

The PGT-A Process

There are a number of steps involved in the PGT-A process, and the aim of this booklet is to provide you with information about PGT-A itself; please refer to additional information provided by Leeds Fertility about IVF and frozen embryo transfer (FET) cycles.
The PGT-A Process step by step

1. Appointment at Leeds Fertility (LF)
   A doctor will take a detailed fertility history and arrange fertility tests to check if IVF is suitable. PGT-A will be discussed. Confirmation that PGT-A is still desired will be confirmed at a private appointment with a Consultant and a checklist and consent form are completed.

2. PGT-A Nurse Consultation
   The PGT-A checklist and consent are checked and standard IVF consents are completed. The IVF cycle is planned. PGT-A registration costs are paid together with payment for IVF.

3. IVF
   In vitro fertilisation (or intracytoplasmic sperm injection (ICSI) if needed) is performed and the resulting embryos are incubated.

4. Embryo Biopsy
   You will be seen at Leeds Fertility by an embryologist on Day 5 to discuss proceeding with PGT-A, and pay per embryo biopsied. An embryologist carefully removes a small cell sample from each embryo and the embryos are then frozen.

5. PGT-A
   Samples are sent to the PGT laboratory, testing is performed, and results are sent to LF. You will meet with a doctor and embryologist to discuss the results.

6. Frozen Embryo Transfer (FET)
   An unaffected embryo is transferred. Remaining unaffected embryos can be kept frozen for future use.
How do I access PGT-A-IVF treatment?

Referral & funding

Whether you are a new patient or an existing patient who has already had IVF treatment at Leeds Fertility, you may wish to use PGT-A alongside an IVF cycle. PGT-A and IVF involving eggs, sperm and embryos require a special license from the Human Fertilisation and Embryology Authority (HFEA) and there are limited clinics available with the required facilities (Please refer to Leeds Fertility IVF patient information booklet).

There is no NHS funding for PGT-A, and therefore a fee will be required for all patients undergoing PGT-A. The cost includes IVF (an additional cost for ICSI will be charged if required), registration for PGT-A and a frozen embryo transfer cycle. The cost does not include drugs. Once your embryos have reached day 5 (the blastocyst stage), you will then be charged for the biopsy and PGT-A per embryo tested as an additional cost. If you do not proceed with the biopsy and PGT-A, and have a fresh embryo transfer, this cost will be incorporated into the initial payment (Please refer to Leeds Fertility Price List).

Understanding genetics

DNA, or deoxyribonucleic acid, is the hereditary material that provides important instructions for the human body. Our DNA is divided up into genes. Genes are the instructions that tell the body how to make proteins that allow the body to develop, grow and function.

DNA is tightly wound and compacted in structures called chromosomes, which are found in the nucleus of our cells.
Healthy people typically have 23 pairs of chromosomes in each cell.

One chromosome in each pair is inherited from the mother (purple) and the other from the father (blue).

What happens if an embryo has an incorrect number of chromosomes?

Research has shown that many embryos have an incorrect number of chromosomes in their cells, a condition known as aneuploidy (extra or missing chromosomes). If a chromosome is lost, or one of them is duplicated, the genetic instructions no longer make sense and the embryo is unable to form a healthy baby.

Most aneuploid embryos fail to implant in the uterus or miscarry during pregnancy. However, there are very few situations where a pregnancy can reach full term and a baby is born e.g. Down’s Syndrome.
PGT-A is a technique that can be used to distinguish between embryos that may have a chromosomal change (affected embryos) and healthy (unaffected) embryos, and select only unaffected embryos to be transferred to the womb.

Comparing normal and abnormal numbers of chromosomes

In this example there is an extra chromosome number 10. When there is an extra chromosome, this is known as a trisomy. Down’s Syndrome is when there is an extra chromosome 21. Sometimes, a chromosome is missing and this is known as monosomy.

An embryo with missing or duplicated chromosomes may grow at a normal rate in its early stages. It is therefore possible to select an embryo for transfer that looks normal but actually contains faulty chromosomes.

This is an important reason why IVF treatment is not always successful.
What information does PGT-A provide?

PGT-A provides information about the make-up of each embryo and helps with the selection of the healthiest one(s). Embryos can be categorized in three ways based on their chromosomal health: euploid, aneuploid or mosaic.

<table>
<thead>
<tr>
<th>Number of chromosomes per cell</th>
<th>Euploid</th>
<th>Aneuploid</th>
<th>Mosaic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>Normal</td>
<td>Abnormal</td>
<td>Mixed</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(Some normal and some abnormal)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Mainly normal</td>
</tr>
<tr>
<td>Likelihood of producing a successful pregnancy</td>
<td>High</td>
<td>Very unlikely</td>
<td>Reduced, but possible</td>
</tr>
<tr>
<td>Recommended for transfer</td>
<td>Yes</td>
<td>No</td>
<td>Considered if no euploid embryos available</td>
</tr>
</tbody>
</table>
What is the relationship between chromosomal aneuploidy and the mother’s age?

The risk of having an embryo affected by a chromosomal aneuploidy is linked to the age of the female partner. The older the woman is, the higher the chance of producing abnormal embryos and having an affected pregnancy. There is a matching fall in the chance of the embryo attaching (implanting).

- For women in their early thirties, about 35% of embryos are aneuploid.
- Over the age of forty years, it is typical for at least 75% of embryos to be aneuploid.

What is the background behind PGT-A?

In the past, the test used in the laboratory to find out whether cells contained the correct number of chromosomes was a technique that had limited value as it only examined one or two cells from very early embryos and looked at only a few chromosomes.

Early studies looking at the value of PGT-A using these older techniques did not show there was a benefit in selecting a healthy embryo in terms of pregnancy rates. Technology has since improved, with the biopsy being performed at a later stage in embryo development, and more cells being sampled, and the genetics laboratory is now able to use tests that detect ALL of the chromosomes and are therefore more effective at selecting an embryo which has the best potential for the birth of a healthy baby.
More recent research indicates that the chances of an embryo with a confirmed normal number of chromosomes producing a healthy baby is more than 25% higher than when an embryo is selected for transfer based on the look of the embryo (morphology). PGT-A may also reduce the time it takes to get pregnant meaning that fewer IVF cycles are needed. It may also reduce the number of IVF pregnancies that miscarry since we know the commonest cause of miscarriage is chromosomal fault. On the other hand, not all studies using modern techniques have found chromosome testing to be of benefit. There is also data suggesting that PGT-A may not be helpful to patients with less than three embryos suitable to biopsy, although this is always a discussion the embryologist will have with you on the day of biopsy. Further robust clinical and laboratory trials are ongoing to work out whether PGT-A can significantly increase live birth rates.

Potential benefits of PGT-A

- In most cases normal and abnormal (aneuploid) embryos look identical. Without genetic screening, an embryologist cannot tell the difference between them and it is possible that chromosomally abnormal embryos could be inadvertently transferred to the uterus.

- It is believed that most abnormal embryos either do not implant or miscarry. By ensuring that embryos with a normal number of chromosomes are prioritized for transfer, the chance of conceiving a healthy child may increase with PGT-A.

- PGT-A happens in the laboratory and does not affect how you feel during a regular IVF cycle.
Alternatives to PGT-A

If a couple have no fertility problems, they can conceive naturally and then consider testing the baby during the pregnancy (prenatal testing using chorionic villous sampling (CVS) or amniocentesis).

Another new test available in early pregnancy is the Non-Invasive Prenatal Test (NIPT) which is a simple blood test for the mother which can detect the most common chromosome abnormalities in the baby.

However, it does mean that a couple may be faced with the very difficult decision about whether to continue the pregnancy if an abnormality is found. Furthermore, these methods will identify pregnancies affected by abnormalities such as Down’s Syndrome, but not increase the chances of a successful IVF cycle.

The IVF process

During IVF treatment, hormones taken by injections, cause the ovary to make several eggs at the same time.

These are taken out of the woman’s body in a minor surgical procedure. (Please refer to Leeds Fertility IVF patient information booklet).
The IVF Process step by step

1. Egg production stimulated by hormone therapy

2. Egg retrieved from ovary

3. Sperm sample provided

4. Sperm and egg mixed together (insemination in standard IVF) or sperm injected into egg (ICSI)

5. Fertilisation

6. Embryos incubated

7. Day 5 “blastocyst” embryo

8. Cells sent to PGT lab for genetic analysis

9. Blastocyst embryos frozen

10. PGT lab informs LF which embryos contain the correct number of chromosomes (e.g. embryos 3 and 6)

11. Embryo with correct number of chromosomes (e.g. embryo 6) is thawed and introduced into the womb
The eggs are put together with her partner’s sperm in the laboratory for fertilisation to take place. This is when the sperm enters the egg and the contents of both are combined to create a unique being. This may be using standard insemination where thousands of sperm are mixed with an egg (IVF) or directly injecting one sperm into an egg (ICSI). The laboratory allows the fertilised eggs, now called embryos, to grow under observation for five days, or sometimes six days.

**Embryo biopsy**

On their fifth or sixth day of development the embryo is called a “blastocyst”. Each blastocyst is assessed and those that have developed normally will be biopsied. This means that a few trophoectoderm (TE) cells will be removed from the blastocyst. These cells are “extra-embryonic” and produce tissues such as the placenta. The inner cell mass (ICM) is the group of cells that becomes the baby. There are over 100 trophoectoderm cells in a blastocyst so the removal of very few of these cells rarely impacts the embryo.

During a PGT-A cycle ALL embryos are cultured to blastocyst regardless of their quality on day 3. If embryos do not reach blastocyst stage, then it is likely these are abnormal embryos and unlikely to result in a successful pregnancy.
Embryos that do not reach a suitable blastocyst stage cannot be biopsied for technical reasons.

Embryos that are slower to develop into a blastocyst can be cultured on to day 6 and if they reach a suitable stage can be biopsied then.

**What happens during an embryo biopsy**

On day 5 you will be invited to meet with the embryologist to discuss the biopsy stage. At any point patients can choose to proceed with a fresh embryo transfer without biopsy and PGT-A. In certain circumstances, where there are very few embryos available the embryologist may advise against biopsy and PGT-A testing and recommend a fresh embryo transfer. All patients will be started on the hormone progesterone after their egg collection, so that if a fresh transfer is appropriate your womb lining will be ready to accept the embryo, and this will take place after meeting with the embryologist on day 5. If a biopsy is planned, you may stop the progesterone medication and you will be asked to pay a setup fee and per embryo and freezing costs at this stage.
If there are embryos that are not ready to biopsy on day 5, the embryologist will check again on Day 6. If there are more embryos to biopsy, you will be sent an invoice to pay for these additional embryos. (see Leeds Fertility price list). You will be given a date for your results appointment which is usually 2-3 weeks later.

Straight after the biopsy procedure the embryos will be frozen to await the result of the test. **There will be no embryo transfer in that cycle** unless you no longer wish to have the PGT-A testing and remove consent prior to embryo biopsy.

The cells removed by biopsy are analysed at a specialised PGT-A laboratory who then inform Leeds Fertility which embryos (if any) contain the correct number of chromosomes.

**Abstaining from unprotected intercourse**

It is critical that you avoid sexual intercourse or use barrier contraception once treatment has started until the pregnancy test.

This is required to prevent a natural pregnancy which could interfere with the PGT-A process and confuse interpretation of results.
Embryo storage

Human embryos generally cope well with freezing and thawing (80% will survive) and this does not affect their ability to grow into a healthy child, regardless of how long they are frozen.

(Please refer to Leeds Fertility Frozen Embryo Transfer cycles information booklet).

What happens to the biopsied cells?
The cells to be tested will be destroyed during the process of the analysis. This will usually occur within 5 days of the biopsy and the DNA inside them will be retained for a minimum of one year. Leeds Fertility are keen to know how your pregnancy progresses, especially if any causes for concern are found. If for any reason the PGT-A test is not performed the sample will be destroyed within 60 days of receipt, as stipulated by standard laboratory rules.

PGT-A analysis of embryos
The main test used for PGT-A at Leeds Fertility is called Next Generation Sequencing (NGS). It examines the full set of chromosomes of the embryos. NGS is cheaper than more old-fashioned methods and is able to detect embryos in which not all the cells are identical. This is called mosaicism. (See Page 11.)
Results

About three weeks after the biopsy the genetics laboratory will report back to Leeds Fertility with you results. You will meet with a doctor and embryologist to go through the results. If there is a healthy embryo to transfer, you will also meet with a nurse for the FET nurse consultation to plan your FET cycle. You will need to pay for the FET cycle at this stage. (Please refer to Leeds Fertility FET patient information booklet). In most circumstances, the FET cycle will occur in the next cycle. If there are no embryos suitable for transfer, you will be offered a consultation with a member of the medical team to discuss the cycle and the future treatment options for you. Legally we also need your consent to allow the abnormal embryos to perish or be used for training. This is included in the PGT-A consent form but we will confirm with you at your results appointment that you give your consent for this to happen.

In most cases, some embryos will be normal and some will be abnormal. However, there is a chance that none of the embryos will be normal, in which case there will be no embryo transfer. Additionally, there may be a technical failure for a small percentage (less than 5%) of embryos and no result is produced. This can be due to the chromosomes breaking up before testing, or technical errors. Further testing of these embryos may be possible.

Follow-up

If a miscarriage occurs, we ask that chromosome studies be carried out on the pregnancy tissue passed, especially if the miscarriage happens at a later stage.
All results from genetic testing of the pregnancy or the child up to the age of one year should be sent to the Leeds Fertility PGT-A coordinator. This information will remain confidential and will be used to cross check outcomes of the PGT-A program.

**PGT-A does NOT diagnose specific genetic disorders**

PGT-A will only look at the numbers of chromosomes in the embryo and cannot detect genetic disorders. If either partner carries a specific genetic disorder that could impair the normal development of a child affected by the disorder, you will be referred for Pre-implantation Genetic Testing for single gene disorders (PGT-M) or Pre-implantation Genetic Testing for structural chromosomal changes (PGT-SR). (Please refer to Leeds Fertility PGT-M/SR patient information booklet).

PGT-A does NOT screen against genetic disorders and does NOT guarantee a child will not be born with a genetic disorder (e.g. Cystic Fibrosis) rather than a chromosomal disorder (e.g. Down’s Syndrome).

**Antenatal Screening**

PGT-A is a “pre-implantation” test which carries a small risk of misdiagnosis. Therefore, if you do become pregnant, conventional “prenatal” cytogenetic analysis is still highly recommended. The purpose of the PGT-A is to decrease the risk of transferring an embryo with too many or too few chromosomes. However, technical limitations mean that the detection and transfer of a chromosomally normal embryo cannot be 100% guaranteed.

Chorionic villous sampling (CVS), amniocentesis and ultrasound scans are used to confirm the chromosome content of the pregnancy.
Non-invasive prenatal test (NIPT), which is a blood test in early pregnancy can also be used. This may not detect all the chromosomes, so is not as comprehensive as the PGT-A or standard CVS/amniocentesis tests.

**Treatment Risks**

**Risks of PGT-A**
As with all artificial reproductive techniques it is important to understand the risks that are involved when using these tests. Below is a list of potential risks for you to consider before embarking on IVF with PGT-A.

**Risk of an unsuccessful outcome and miscarriage**
By performing PGT-A and having a normal embryo transferred does not, unfortunately, guarantee a positive pregnancy test, a viable early pregnancy or a healthy live birth. PGT-A has large financial and emotional costs, and counselling is available for either partner at any stage of the treatment should you wish.

As with all pregnancies, there is also a risk of miscarriage. PGT-A testing and replacement of a chromosomally normal embryo does not eliminate the risk of miscarriage; it can only serve to reduce the risk.

**Risks of the cell biopsy procedure**
It is not yet known whether embryos that have been biopsied have the same chance of implanting as embryos that have not been tested. The biopsy itself may lower the chance of the embryo implanting. However, any reduction in success may be more than made up for by identifying a normal embryo with no reason not to implant. If an embryo is damaged by the procedure, it may not produce an embryo suitable for transfer.
The risk of damaging an embryo is less than 1%. Several thousand healthy babies have now been born from IVF with PGT-A. These babies do not have any more congenital abnormalities (birth defects) than occur naturally in the general population (3-5%).

**Risk of preparation of biopsied cells**

After the biopsy procedure, the cells are placed in a small test tube. The cells are no longer viable in any way after this process and can only be used for PGT-A. A fraction of the cells may not yield a test result (<5%), some may not contain any genetic material, cells may be lost during the highly technical fixation process of the test, or may have suboptimal fixation meaning they cannot be used for this complex analysis.

**Risk of the test giving the wrong result**

No scientific test is 100% perfect. There is a small chance that the test will report normal embryos as abnormal. This is called a false positive result and happens up to 15% of the time. The consequence is that a normal embryo is nor transferred when it could have resulted in a healthy baby.

The chance of the test reporting an abnormal embryo as normal is lower (5%). The consequence of this is that an abnormal baby may result. It is for this reason that prenatal testing during pregnancy is recommended, even after PGT-A, to double-check.

**Risk of having nothing available for biopsy**

There is a chance that no embryos will be suitable for biopsy as they have not reached the correct stage of development. In these cases, it is highly likely that the embryos that have stopped developing are chromosomally abnormal and would not produce a viable pregnancy in any case.
Risk of no normal embryos

The test may find that none of the embryos are normal, in which case there may be no embryo transfer procedure.

The likelihood that this will happen is influenced by a variety of factors; the most significant are advancing female age and having a small number of embryos to work with.

If there are no normal embryos where all the cells have the correct number of chromosomes (euploid), we will consider transferring embryos that have a mixture of normal and abnormal chromosome numbers, whereby the majority of the cells are normal (low level mosaicism), as these have been shown to produce healthy pregnancies although at a lower rate than with euploid embryos. We will discuss this with you when we give you the biopsy results. We will not transfer embryos with high level mosaicism or aneuploid embryos.

Risk of no diagnosis/partial diagnosis

Some embryos may have no diagnosis, due to the absence of chromosomes in the sample, or technical difficulties in the fixation process. In addition, sometimes the analysis may not be clear for one of the chromosomes tested. This will be discussed with you individually if this does occur.

Current PGT-A techniques are unable to provide a guarantee that the child will have a normal set of chromosomes after IVF and PGT-A.
Glossary

- **Aneuploidy**: extra or missing whole chromosomes.
- **Biopsy**: the removal of one or several cells for examination or testing.
- **Blastocyst**: This is a particular stage of development of an embryo which should be reached by day 5-6 after egg collection. A blastocyst has 50-60 cells and they have begun to separate into those that will form the baby and those that will form the placenta (afterbirth). A small area of fluid separates the two types of cells. Shortly after this stage the embryo will hatch and should implant into the lining of the womb.
- **Chromosomes**: structures of tightly wound and compacted DNA which allow for the genetic code to be stored within cells.
- **Deoxyribonucleic acid (DNA)**: is the building block of genes; made up of a sugar phosphate backbone and four nucleotide bases.
- **Eggs**: A woman’s lifetime supply of eggs is present in the ovary at birth. They reduce in number and quality with time. They pass on the woman’s half of the genetic instructions to the embryo / baby.
- **Embryo**: Once the fertilised egg begins to cleave (multiply its cells) it is called an embryo.
- **Euploidy**: Correct number of chromosomes in a cell (46 chromosomes).
- **Fertilisation**: Fertilisation is when the genetic material from the egg and sperm combine to create a new and unique cell which may go on to develop into an embryo and then a baby.
- **Genes:** unique stretches of DNA which use a four letter alphabet of nucleotide bases to encode for protein creation.

- **Miscarriage:** Any positive pregnancy test which does not reach 24 weeks of pregnancy and the potential for a live-born child is a miscarriage. Miscarriage is as common after IVF as it is after natural conception. Bleeding in early pregnancy is not always bad news, especially if there is no cramping pain. Unfortunately, some pregnancies miscarry without any outward signs (bleeding) and are not identified until a scan is done.

- **Monosomy:** one chromosome missing from the normal number (45 chromosomes).

- **Mosaicism:** a mixture of euploid (normal chromosome number) and aneuploid (abnormal chromosome number) cells in an embryo.

- **Preimplantation genetic testing for structural chromosome changes (PGT-SR):** the process of testing embryos for chromosome rearrangement prior to transfer.

- **Preimplantation genetic testing for monogenic / single gene disorders (PGT-M):** the process of testing embryos for a specific single gene disorder prior to transfer.

- **Preimplantation genetic testing for aneuploidy (PGT-A):** the process of testing embryos for the number of chromosomes prior to transfer.
• **Sperm:** The sperm develop in the man’s testes and continue to do so throughout adult life. They do not suffer the same deterioration with age as the woman’s eggs, as they are constantly being replaced. They pass on the man’s half of the genetic instructions to the embryo / baby.

• **Trisomy:** An extra chromosome present (47 chromosomes). Down’s Syndrome is also known as Trisomy 21 because the cells contain an extra chromosome 21.
Contact us

By post
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By Email
• leedsth-tr.leedsrmuenquiries@nhs.net

Online
• Web: www.leedsfertilityclinic.co.uk

By telephone

*Mon-Fri 08.00-17.00*
• For all NHS appointments: 0113 206 3100
• For clinical queries: 0113 206 3102

*Sat-Sun 08.00-12.00*
• Clinical queries only: 0113 206 3102

In an Emergency

*During working hours*
• Please call appointments or clinical queries as needed on the above numbers

*Outside working hours*
• Please call Leeds Teaching Hospitals Switchboard on 0113 243 3144 and request to be put through to the Duty Nurse / Dr for Leeds Fertility
• If necessary, attend your local A&E department.