What is CGH?

CGH or Comparative Genomic Hybridization is a technique developed to look at the total number of chromosomes in a single cell to check for extra or missing chromosomes.

Why array CGH?

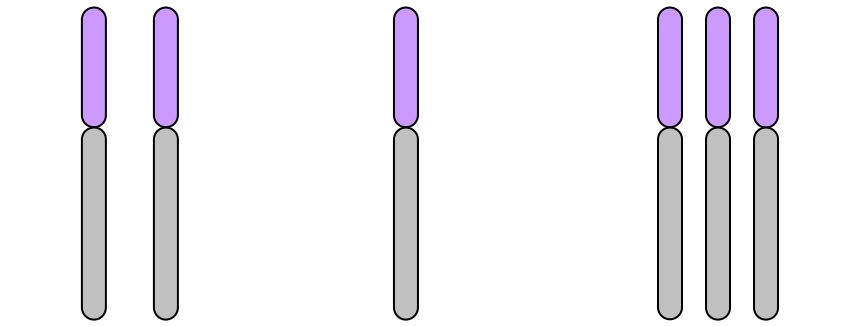
Array CGH is a significant advance on conventional CGH as it is a more refined, robust and is much quicker. Results are typically available within 24-48 hours allowing a fresh embryo transfer whereas conventional CGH can take at least 5 days, making it necessary to freeze the embryos and transfer at a later date.

What is a chromosome?

A chromosome is a highly complex structure containing genes that make us who we are, and they are found in every cell of our body (except red blood cells). One chromosome may carry over 1000 genes. Chromosomes are in pairs, and in humans there are 23 pairs.

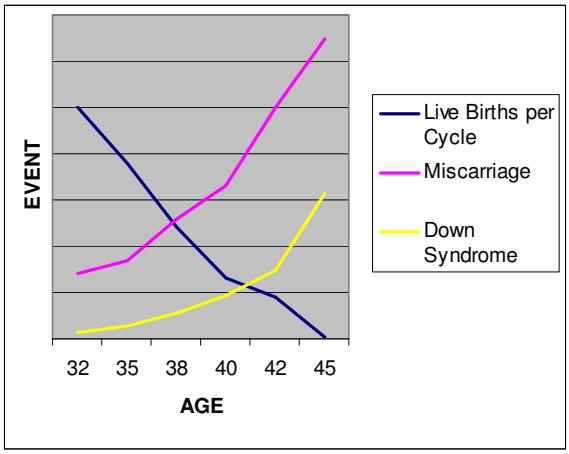
Why are chromosomes important in fertility?

Approximately 70% of embryos produced either through natural conception or IVF are lost before birth. The vast majority of embryos are lost within the first three months of pregnancy, most of these even before implantation. A major cause of embryo loss, including miscarriage, is a chromosome anomaly (known as 'aneuploidy') where there is either a loss of a chromosome or a gain.
Some of these anomalies are compatible with full term delivery, such as three copies of chromosome 21 (known as Down syndrome) or three copies of chromosome 18 (Edward Syndrome). Some are not compatible with full term delivery; and others cause the embryo to arrest its development before implantation.



Normal chromosome pair | **One chromosome missing** | **A gain of one chromosome**

The rate of aneuploidy in eggs also increases with a woman's age, with Down syndrome being the most commonly known. This table shows that as age increases, so does the risk of aneuploidy and chromosome-related miscarriage, causing a major reduction in live birth rates.
The incidence of Down syndrome rises from 1 in 900 at age 30, through 1 in 230 at age 37 to 1 in 20 at 46.



Why might my eggs have chromosome abnormalities?

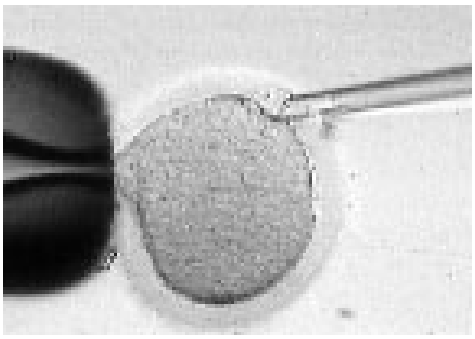
At least 85% of embryo aneuploidy can be traced back to the egg. Sperm chromosome anomalies have a much smaller impact, estimated to be around 7-8%. The remainder can arise randomly as the cells go through their early cell division process.
It has been demonstrated that more than 50% of human eggs have a chromosome anomaly. As this increases significantly with age, it is probably the main reason why women of advanced reproductive age have such difficulties conceiving, and also have a high incidence of miscarriage.

How can the egg be tested?

When eggs and sperm develop, they have 46 chromosomes (23 pairs) as do all cells in the body. Before they are ready to make an embryo they must halve this amount so that when they fuse together during fertilisation they will again have their full complement of 46.
The egg's ingenious way of doing this is to shunt out half of all the chromosomes in a tiny amount of cytoplasm – this cytoplasm is called the 'polar body'.

Mature human egg

This process of extruding the polar body gives us the very opportunity to study the chromosomes of the egg without damage. The polar body plays no role in fertilisation or embryo development and is therefore redundant.
The polar body is gently removed from the egg by a process known as 'polar body biopsy'.


Polar body biopsy

Why test eggs and not embryos?

When we test embryos, we usually take one cell away from the embryo on Day 3, leaving around 6-8 cells so the embryo can grow on. The problem with this approach is that it is possible for an embryo to have one abnormal cell but the rest of the embryo's cells remain normal. Conversely, it is possible that the cell examined is normal but several of the remaining cells could be abnormal, giving the impression the embryo has no chromosome anomalies whilst in fact it is unable to develop normally. This difference in cells is known as 'mosaicism' and is unfortunately relatively common in embryo development. Either of the above scenarios will give us a false result, hence the reason why we do not use Day 3 embryos for aneuploidy screening.

The hope for CGH- 'One Embryo-One Baby'

Embryologists in routine IVF practice cannot differentiate between chromosomally viable and aneuploid eggs/embryos, and hence aneuploid embryos will unwittingly and inevitably be transferred to the womb, thereby compromising IVF outcome. Conversely, it follows that by identifying the aneuploid status of the egg, embryologists have up to 85% chance of eliminating the chromosomally abnormal embryos before transfer.

It is hoped that this technology should:

i) lead to markedly improved IVF success rates;
ii) reduce the incidence of multiple pregnancies (by transferring fewer but 'competent' embryos);
iii) reduce the incidence of pregnancy wastage (miscarriage);
iv) reduce the cost of achieving a viable IVF pregnancy and,
v) reduce overall reproductive health care costs.

How will array CGH work for me?

IVF will be planned in the normal way:

Phase 1: A protocol of drugs will be prescribed for ensuring the most appropriate ovarian stimulation regimen. You will come to CARE for monitoring and at the appropriate time undergo egg collection. At this point you can return home whilst the work continues in the laboratory. The collected eggs will be examined for their maturity. Only mature eggs that have their polar body extruded will be used. Each of these mature eggs will undergo ICSI and then have its polar body removed, prepared and stored. Fertilised eggs will be developed in the embryology laboratory.

Phase 2: Because array CGH is a very expensive technology, and not every egg will fertilize we have designed a strategy that should minimise the cost of having to examine every single polar body from every single egg. Once fertilisation has occurred, we will send the prepared polar bodies of only those eggs that have fertilised for testing. Only those embryos that have come from an egg whose chromosomes were normal will be transferred or frozen for future use.

Please note that only a maximum of two embryos will be transferred, and in many cases we would counsel couples to have only a single embryo transferred.

Risks - general

There are no health risks associated with the programme that are not apparent in conventional IVF.

Array CGH is a new experimental technology and as such must still be considered to be in a test phase. There is insufficient data to provide any reliable guide to how effective this technique is in embryo selection. We acknowledge that there is therefore a risk of technical, equipment or analysis failure or malfunction which may affect the accuracy of assay results, or prevent the delivery of any results.

Risks – related to biopsy and analysis procedures

The procedure for this technique has been used for many years. Each CARE biopsy practitioner is licensed by the HFEA (Human Fertilisation and Embryology Authority). Accidental risk to the egg is less than 0.2%.

Misdiagnosis

It is estimated that the risk of a misdiagnosis (either positive or negative) is less than 5%. However we advise that a prenatal diagnosis (CVS or amniocentesis in early pregnancy) is still performed in the routine way for pregnant women, for further reassurance.

No normal eggs/embryos

There is approximately a 25% chance that all polar bodies tested show abnormal results, with none of the embryos therefore being available for transfer. If this occurs you will be offered a detailed review with your consultant to discuss the relevance and options. **CARE cannot under any circumstances transfer any embryo(s) shown to carry abnormal chromosomes.**

Indeterminate results

Occasionally it could arise that there are no results from any of the chromosomes from a polar body. In this case we do not know if the egg is chromosomally normal or abnormal. If this occurs for the only available embryo(s) for transfer, the relevance of this will be discussed between you and your consultant.

Pregnancies following CGH

There is no guarantee against miscarriage occurring even after screening all the chromosomes. Some miscarriages occur due to other factors such as immune conditions or undiagnosed genetic problems. If you do become pregnant following screening the pregnancy will still be subject to all risks and problems that can be associated with natural pregnancies including ectopic pregnancies and congenital abnormalities. The procedure itself should not affect any resulting pregnancy. Thousands of children have now been born following biopsy for other reasons.

Is Chromosome Screening regulated?

Yes, and it is mandatory for a UK clinic to hold an HFEA (Human Fertilisation and Embryology Authority) licence. CARE is the first organisation to be licensed by the HFEA for this method, and for all chromosomes to be examined.

Indeed, following CARE's success, the European Society of Human Reproduction (ESHRE) Task Force on Preimplantation Genetic Screening has formulated the view that, "*Polar body biopsy and 24-chromosome analysis would in theory be the best alternative*" - referring to techniques to assess chromosome status.

How do I consider if this is right for me?

CARE can offer you a detailed clinical consultation where the medical and scientific issues will be discussed, and you will also be offered counselling by a trained independent counsellor. We recognise that the issues are complicated and we urge all patients to feel free to discuss with our specialised team any aspect of the procedure or any concerns.

Are there any additional tests we will need before we proceed?

All the normal screening tests required before a normal IVF/ICSI cycle are also needed before an arrayCGH cycle and these are document in our separate "IVF/ICSI Cycle" Information document. In addition though, we will need to have information on karyotypes for both partners and we advise that the man considers having his sperm tested for both sperm aneuploidy and Sperm DNA Fragmentation. We advise these additional tests before treatments as abnormalities in these tests can be significant confounding variables for outcome of a treatment cycle and may affect the decision whether to proceed with arrayCGH.

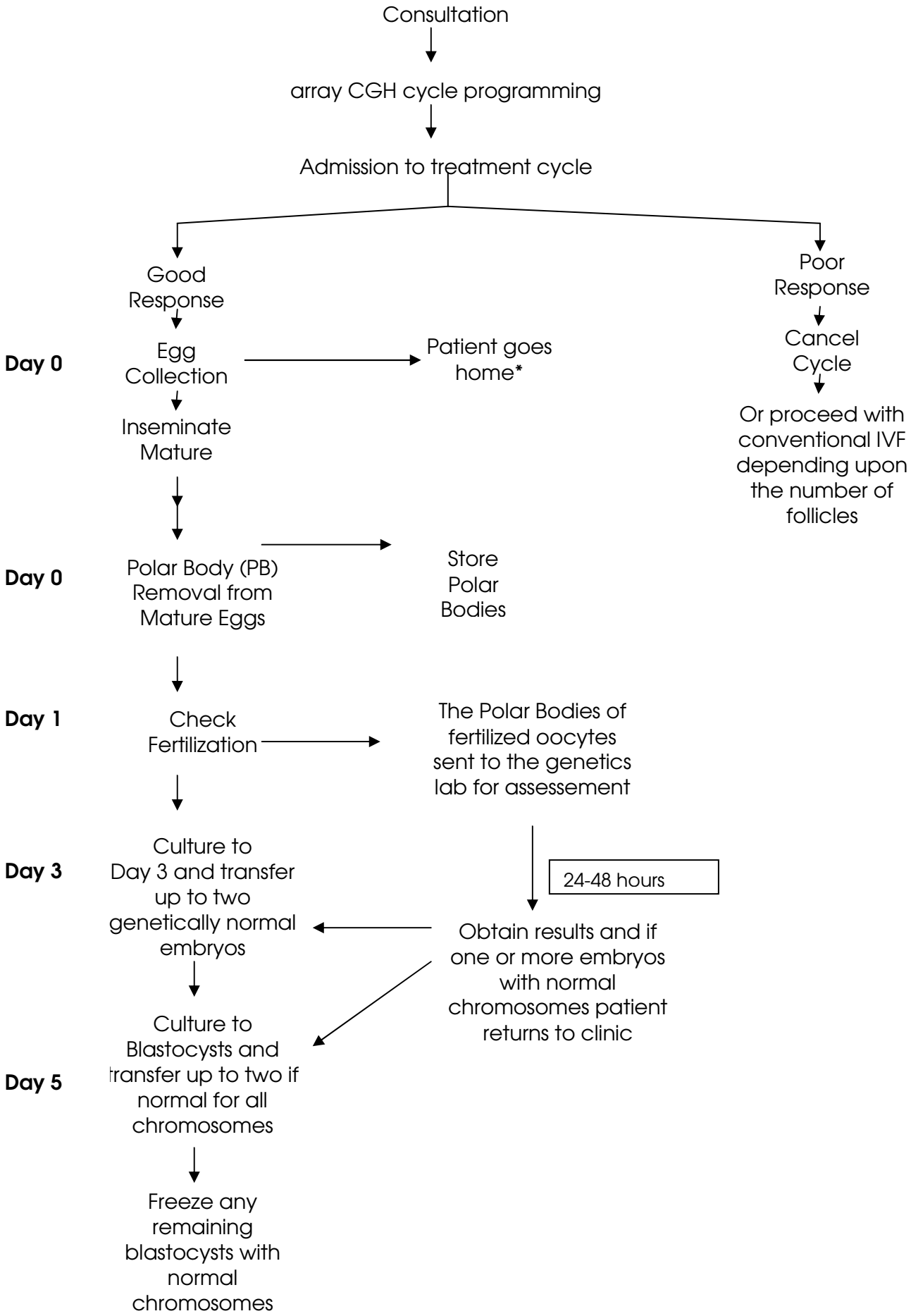
Costs

The cost of array CGH is additional to the cost of IVF with ICSI. Please see CARE fee schedule, available from CARE Reception or on the Fee Schedule section of www.carefertility.com , for current pricing. The fee schedule is structured to allow the testing of up to 8 eggs within the price of CGH. Additional eggs will be charged individually on top of this. If subsequent cells are tested on another day a further charge will apply.

PLEASE NOTE: SHOULD POLAR BODY BIOPSY BE UNDERTAKEN BUT ARRAY CGH IS NOT PERFORMED ON THE POLAR BODIES FOR ANY REASON, THE CHARGE FOR THE BIOPSY WILL STILL APPLY (PLEASE SEE CURRENT FEE SCHEDULE).

We recognise that there is a lot of information contained in this document. However, as this technology is very new it is important that we provide you with comprehensive information to help you make an informed decision. Please read the information carefully and do ask any questions of our consultants or senior clinical staff during your consultation or at any time.

CGH Flow Chart



Glossary

Aneuploid (aneuploidy)	An abnormal chromosome profile in a cell
Biopsy	The removal of a portion of cellular material for testing
Blastocyst	The stage 5 – 6 days after fertilisation when the zygote has reached about 30 or more cells
CGH	"Comparative Genomic Hybridization" – a technique used to analyse all the chromosomes of a single cell
Chromosome	Paired structures within each cell containing the genes of the individual
Embryo	The name given to the fertilised egg once it divides into two cells, and continues cell division
Euploid	A normal chromosome profile
Follicles	Sacs in the ovary that contain the eggs
Gamete	The sperm or egg
ICSI	"Intra Cytoplasmic Sperm Injection" – a method to directly inject a single sperm into an egg
Nucleus	The central part of almost all cells, which contains the chromosomes and genetic material
Oocyte	The female gamete; egg
Polar Body	The tiny part of an egg that contains half the egg's chromosomes but does not take part in the fertilization process
Zygote	The fertilized egg, containing the chromosomes from both the egg and the sperm