Carrier Reproduction

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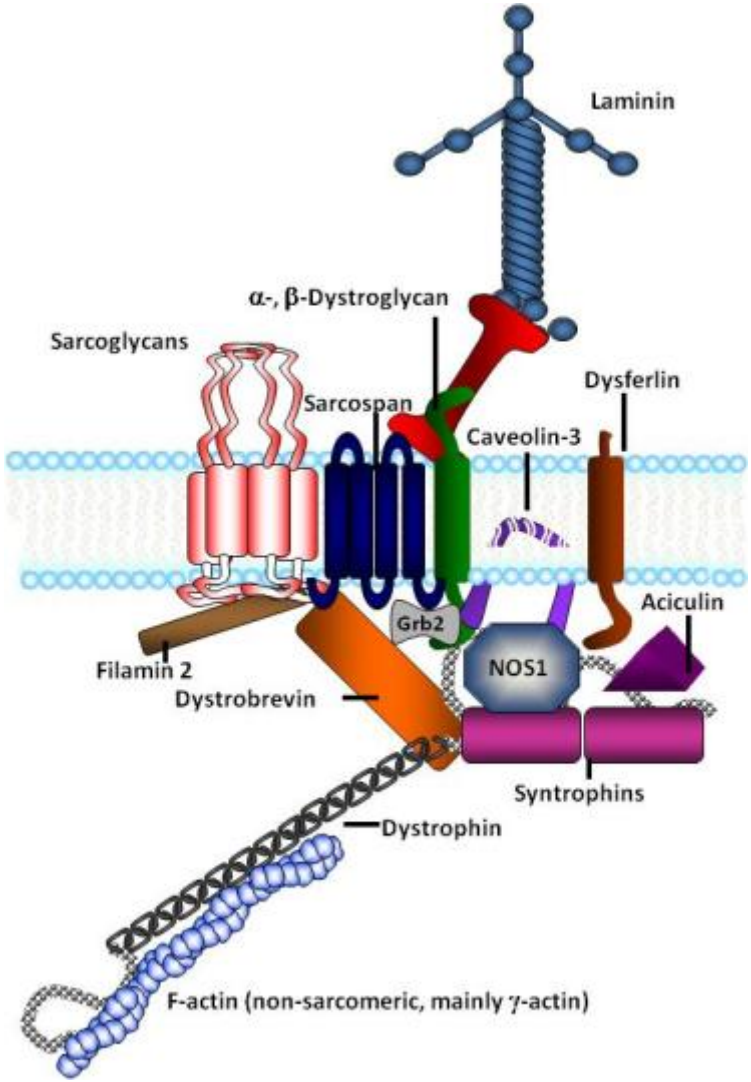
NEWCASTLE UPON TYNE

Duchenne Muscular Dystrophy

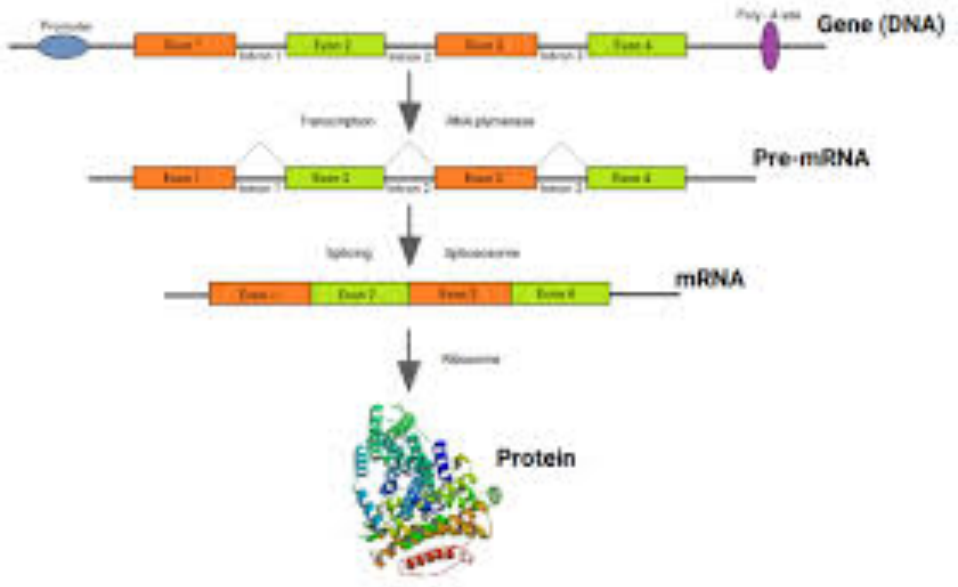
- Due to mutations in the dystrophin gene
- X linked disorder, predominantly affecting boys
- Prevalence 1 in 3600 - 1 in 6000 male births
- 10% female carriers affected, usually mildly



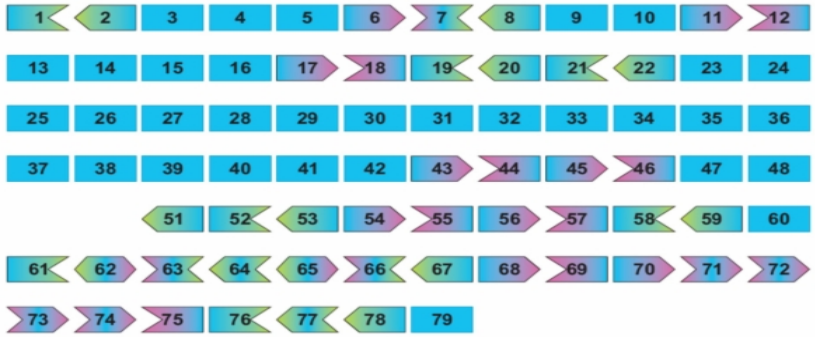
The dystrophin gene

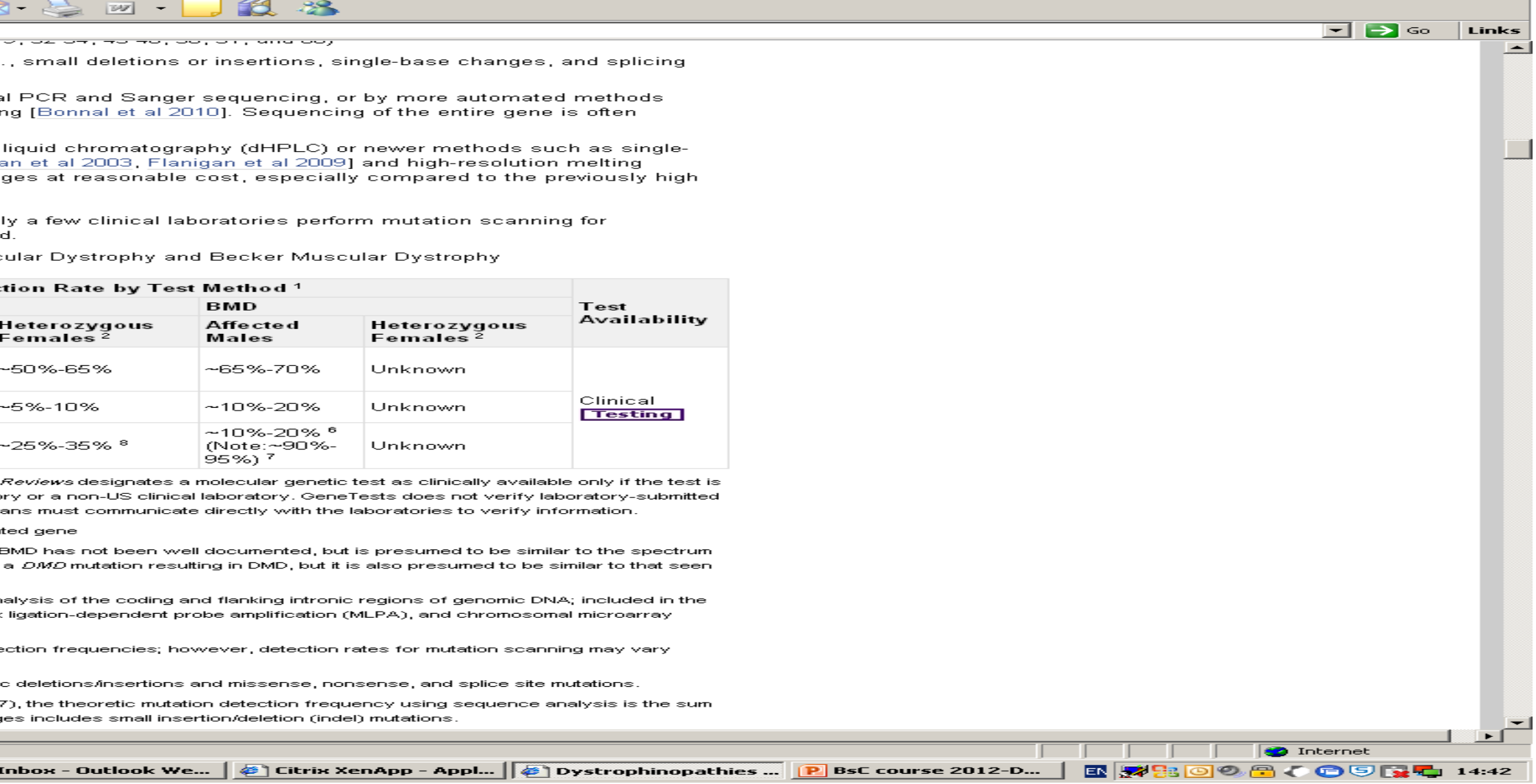
It is really, really big

- the largest known gene
  - 79 exons
- over 99.5% of pre-mRNA is discarded during processing



Exon skipping as a treatment for DMD





., small deletions or insertions, single-base changes, and splicing

al PCR and Sanger sequencing, or by more automated methods
ng [Bonnal et al 2010]. Sequencing of the entire gene is often

liquid chromatography (dHPLC) or newer methods such as single-
an et al 2003, Flanigan et al 2009] and high-resolution melting
ges at reasonable cost, especially compared to the previously high

ly a few clinical laboratories perform mutation scanning for
d.

cular Dystrophy and Becker Muscular Dystrophy

tion Rate by Test Method [1]

	BMD		Test Availability
Heterozygous Females [2]	Affected Males	Heterozygous Females [2]	
~50%-65%	~65%-70%	Unknown	Clinical Testing
~5%-10%	~10%-20%	Unknown	
~25%-35% [8]	~10%-20% [6] (Note:~90%-95%) [7]	Unknown	

Reviews designates a molecular genetic test as clinically available only if the test is
ory or a non-US clinical laboratory. GeneTests does not verify laboratory-submitted
ans must communicate directly with the laboratories to verify information.

ted gene

BMD has not been well documented, but is presumed to be similar to the spectrum
a *DMD* mutation resulting in DMD, but it is also presumed to be similar to that seen

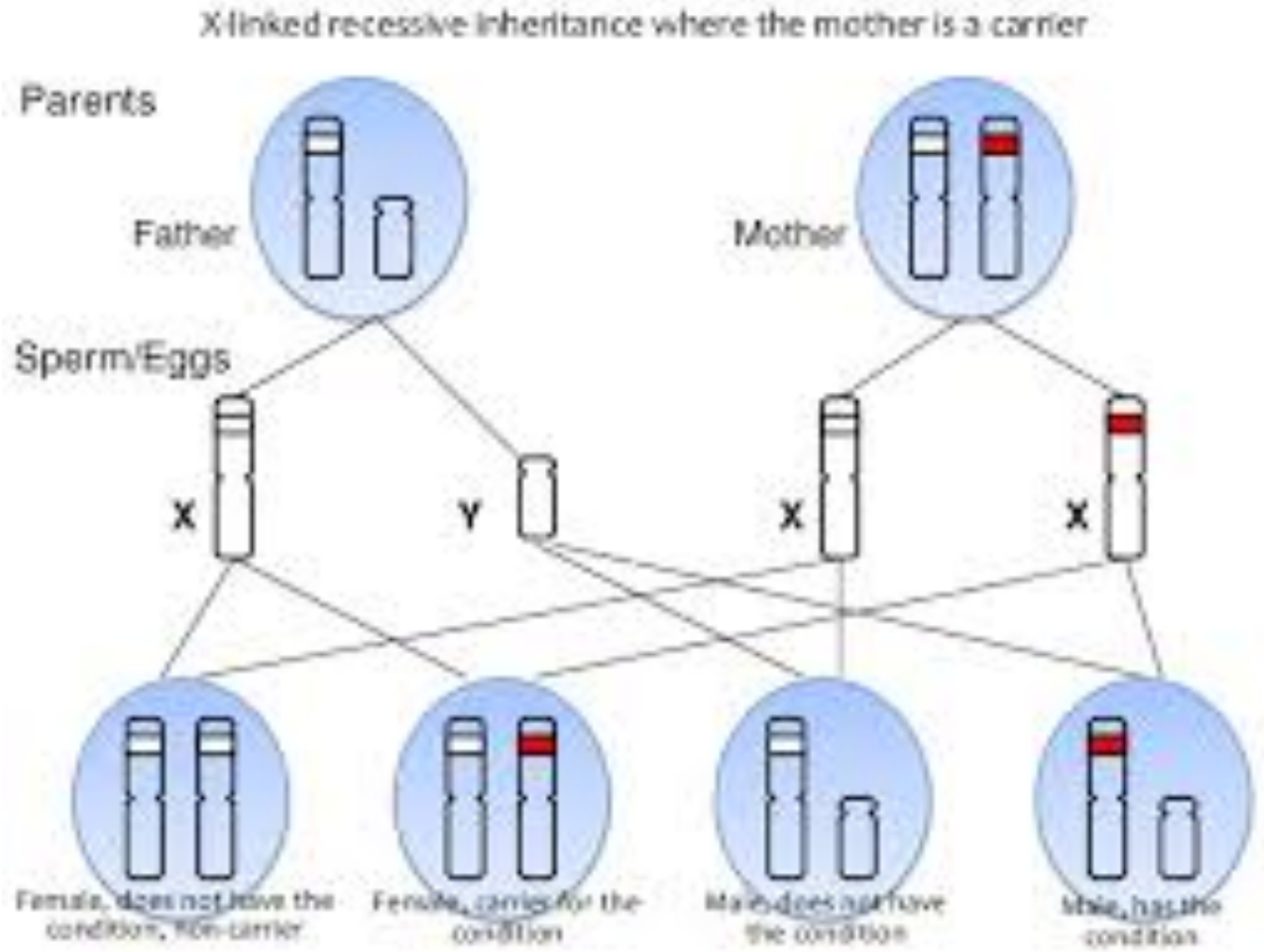
nalysis of the coding and flanking intronic regions of genomic DNA; included in the
ligation-dependent probe amplification (MLPA), and chromosomal microarray

ection frequencies; however, detection rates for mutation scanning may vary

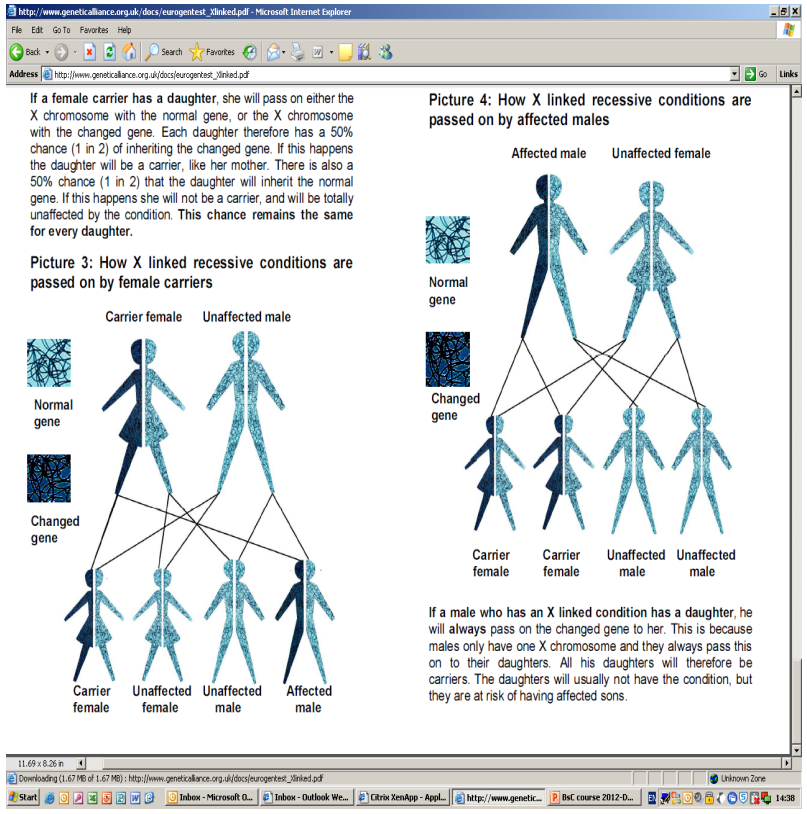
c deletions/insertions and missense, nonsense, and splice site mutations.

7), the theoretic mutation detection frequency using sequence analysis is the sum
ges includes small insertion/deletion (indel) mutations.

DMD is an X-linked disease



DMD is an X-linked disease

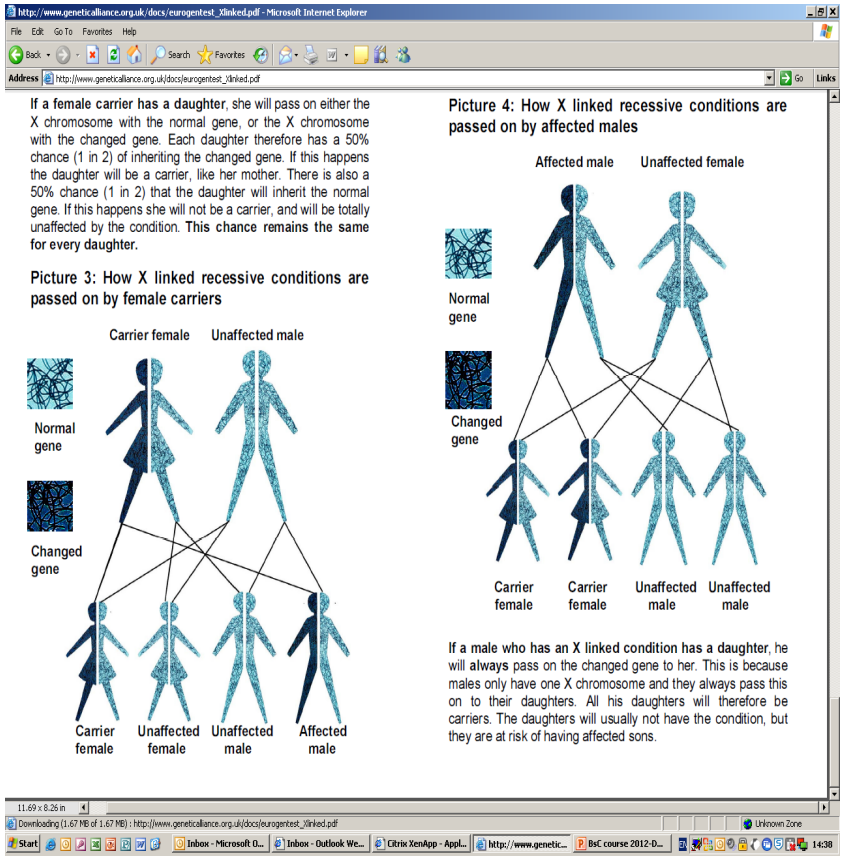


If a female carrier has a daughter, she will pass on either the X chromosome with the normal gene, or the X chromosome with the changed gene. Each daughter therefore has a 50% chance (1 in 2) of inheriting the changed gene. If this happens the daughter will be a carrier, like her mother. There is also a 50% chance (1 in 2) that the daughter will inherit the normal gene. If this happens she will not be a carrier, and will be totally unaffected by the condition. **This chance remains the same for every daughter.**

Picture 3: How X linked recessive conditions are passed on by female carriers

Picture 4: How X linked recessive conditions are passed on by affected males

If a male who has an X linked condition has a daughter, he will **always** pass on the changed gene to her. This is because males only have one X chromosome and they always pass this on to their daughters. All his daughters will therefore be carriers. The daughters will usually not have the condition, but they are at risk of having affected sons.



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Picture 3: How X linked recessive conditions are passed on by female carriers

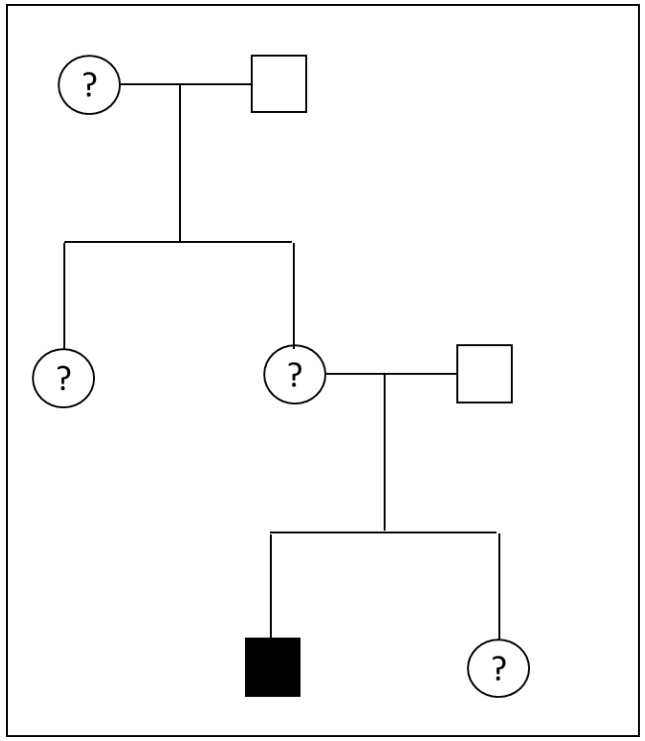
Carrier female
Unaffected male
Normal gene
Changed gene
Carrier female
Unaffected female
Unaffected male
Affected male

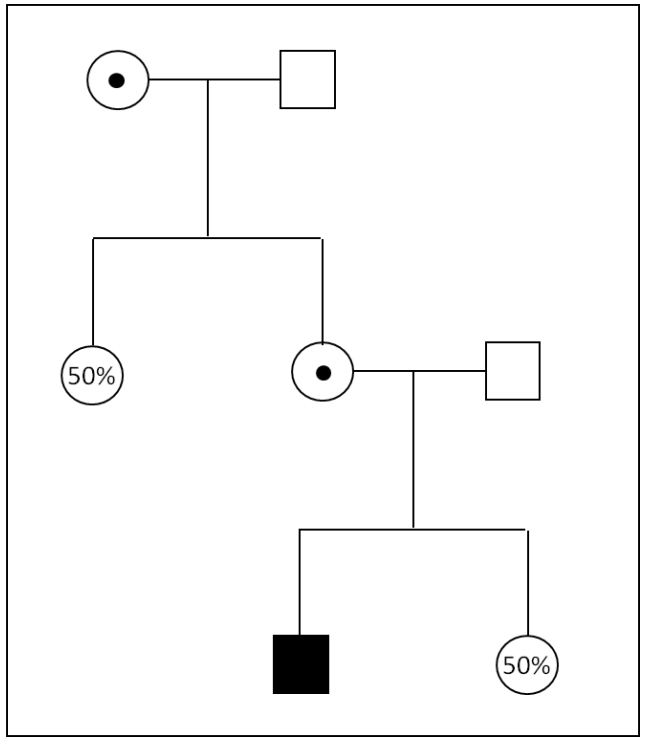
Picture 4: How X linked recessive conditions are passed on by affected males

Affected male
Unaffected female
Normal gene
Changed gene
Carrier female
Carrier female
Unaffected male
Unaffected male

If a male who has an X linked condition has a daughter, he will **always** pass on the changed gene to her. This is because males only have one X chromosome and they always pass this on to their daughters. All his daughters will therefore be carriers. The daughters will usually not have the condition, but they are at risk of having affected sons.

Recurrence risks?





- Sporadic patient
  - ✓ 1/3 *de novo* mutation (mother not a carrier)
  - ✓ 2/3 carrier mother

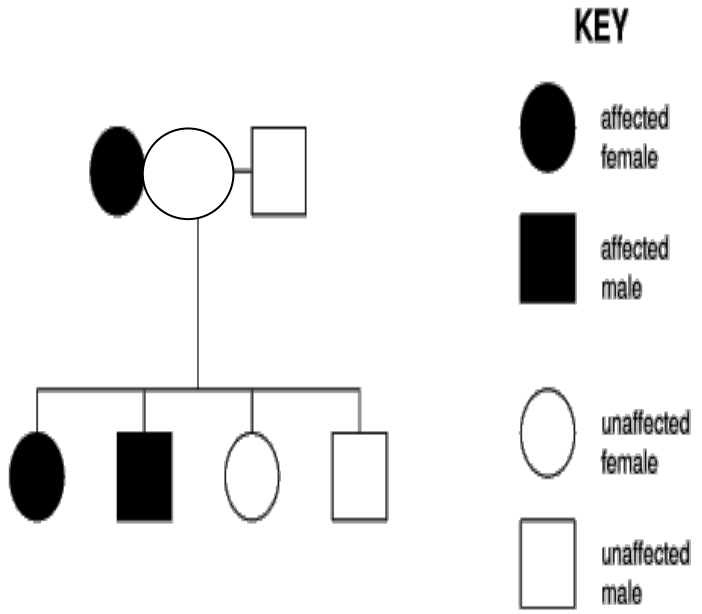
Germ line masaicism

DE NOVO MUTATIONS: occurring during mitosis/meiosis

Multiple affected offspring from apparently non carrier parents

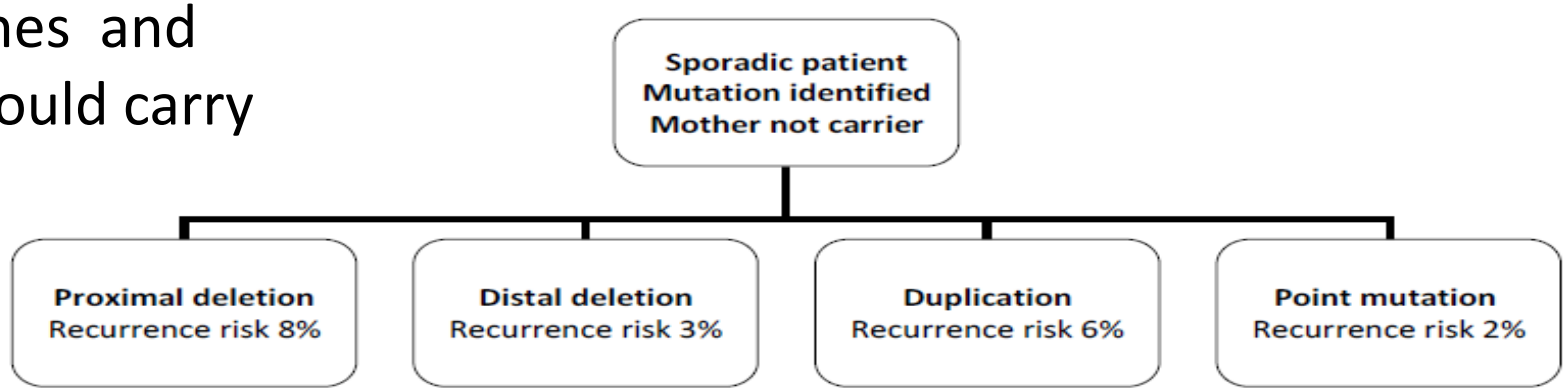
Reported in > 60 genetic diseases

Recurrence risk known only for a few



Germ line masaicism

- Distinct recurrence risks are calculated for proximal and distal deletions
- Higher recurrence risk for proximal deletions
- Proximal deletions arise earlier in embryogenesis than distal ones and therefore more cells would carry the mutation



Helderman-van den Enden et al, Clin Genet 2009

Reproduction options – Newborn screening

- Sickle cell disease
- Cystic fibrosis
- Congenital hypothyroidism
- Inherited metabolic diseases
  - ➢Phenylketonuria
  - ➢Medium-chain-acyl-CoA dehydrogenase deficiency
  - ➢Maple syrup urine disease
  - ➢Isovaleric acidaemia
  - ➢Glutaric Aciduria type 1
  - ➢Homocystinuria

Reproduction options for couples at risk of passing on an inherited disorder

https://www.england.nhs.uk/wp-content/uploads/2014/04/e01-med-gen-0414.pdf

- To remain childless / not having other children
- To adopt a child
- To pursue gamete donation (assisted conception techniques in which one or both parents would not be the biological parent of the child)
- To conceive naturally, and accept the risk of their child inheriting the genetic
- To conceive naturally and undergo conventional prenatal diagnosis (PND) following conception
- To undergo Non-Invasive Pre-Natal Diagnosis (limited to some inherited disorders) or Pre-Implantation Genetic Diagnosis (PGD) NIPD?

Reproduction options – Non-Invasive Prenatal Diagnosis (NIPD)

- Performed from 8 weeks of pregnancy (confirmed on scan)
- 20mls Blood taken from mother to look for cell free fetal DNA
- Results sometimes inconclusive result because insufficient Fetal DNA
- Can be used for chromosome abnormalities (Down syndrome, trisomy 18, or trisomy 13) - *available only privately*

Non-Invasive Prenatal Diagnosis (NIPD) for Duchenne Muscular Dystrophy

- NIPD for Duchenne muscular dystrophy is performed in two stages

  Stage 1: sample collected at week 8-9 of pregnancy. If the baby is predicted to be female no further testing will be performed.

  Stage 2: If the baby is predicted to be male the laboratory will go on to look at special markers to determine which copy of the X chromosome has been inherited from the mother. If the normal copy has been inherited the male baby is unaffected. If the altered copy has been inherited the male baby is affected.

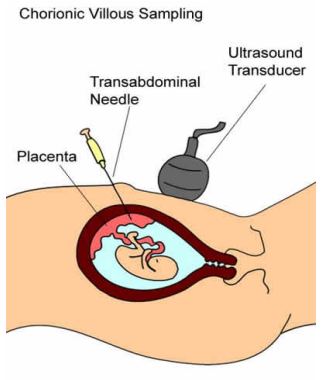
  The results for this can take up to 14 working days.

Non-Invasive Prenatal Diagnosis (NIDP) for Duchenne Muscular Dystrophy

- Require DNA from an affected child (or other affected family member), mum and dad.
- In same cases, NIPD is not possible depending on the type of the mutation.
- If NIPD for DMD is not possible, NIPD can be used to determine the sex of the baby but an invasive test would then be needed to find out if a male baby had DMD
- Free through NHS
- **For any enquiry on NIPD for DMD, speak to a genetic counsellor at the local genetics service**

Reproduction options – Pre-natal options

Chorionic Villus Sample



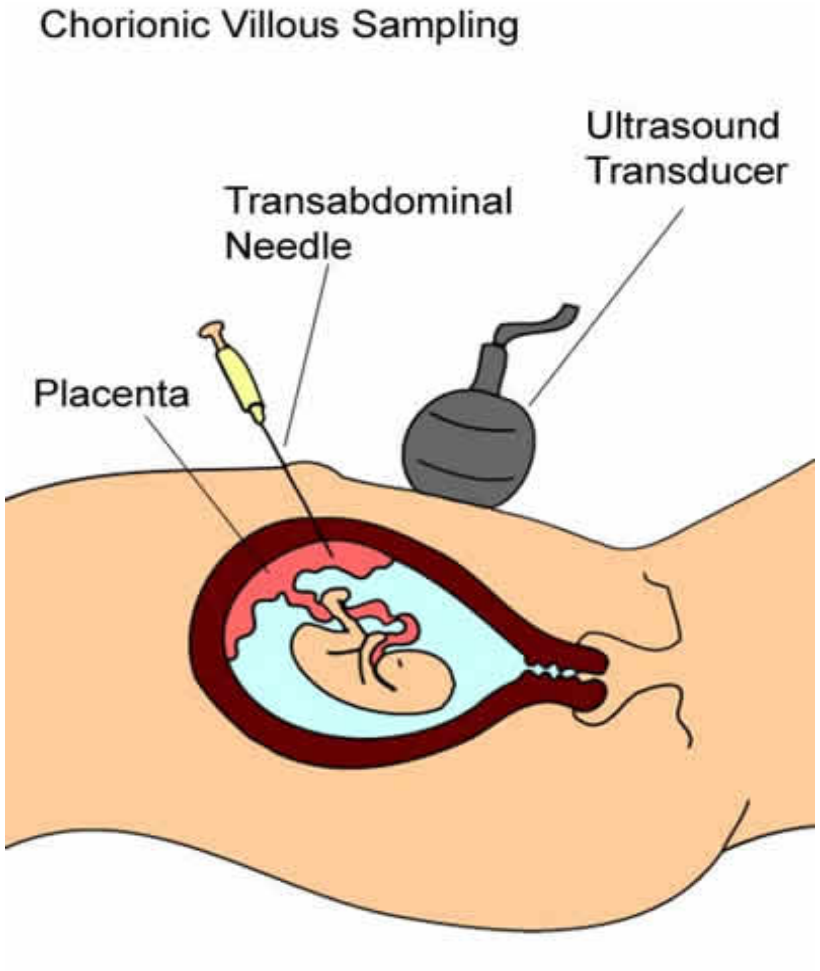
- Between 11th and 14th week of pregnancy
- Risk of miscarriage 1:100 women
- Other risks: infection, need to repeat the procedure

Amniocentesis

Between 15th and 20th week of pregnancy
Risk of miscarriage 1:100 women
Other risks: infection, need to repeat the procedure

Reproduction options – Pre-natal options

Chorionic Villus Sample



- Invasive test taking cells from the chorionic villi of the placenta.
- Result available within a week
- Risk of miscarriage is a small increase over that of the risk of any pregnant woman

Reproduction op

Embryo testing - Pre-Ir

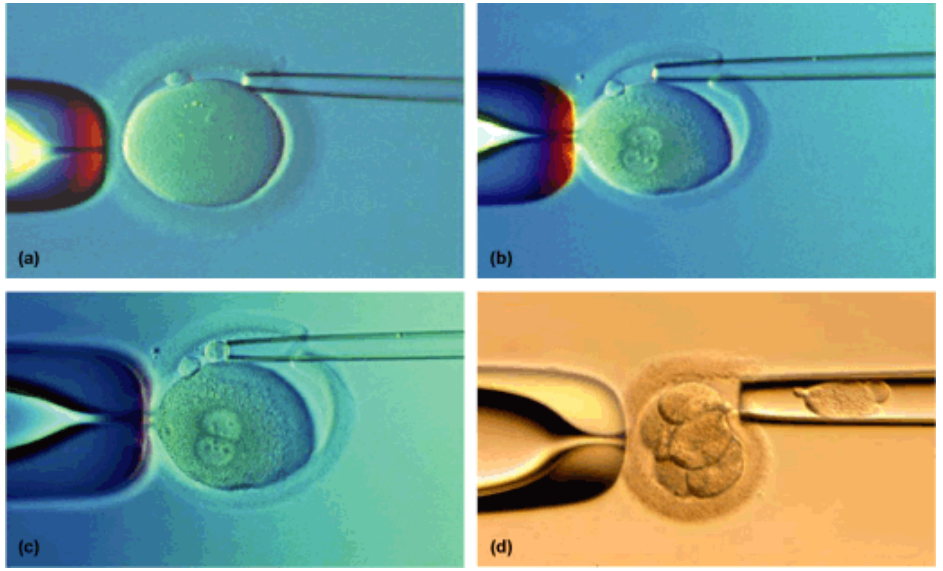
- Aim: allow couples at si ave a child that is genetically
- Offered to people with
- Only way for parents to l parents, without risking

https://www.england.nhs.uk/wp-content/uploads/2014/04/e01-med-gen-0414.pdf

Reproduction options – Pre-natal options

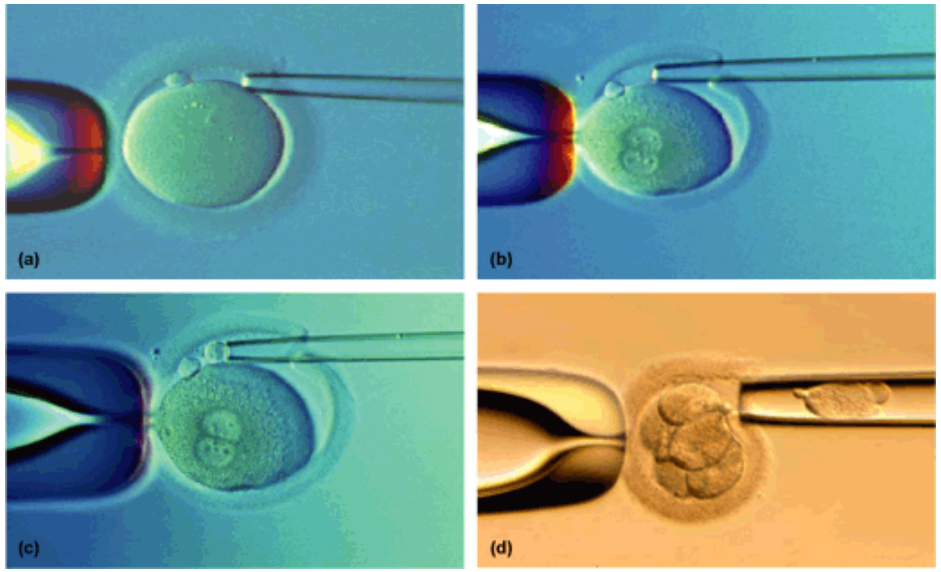
Embryo testing - Pre-Implantation Genetic Diagnosis

- It requires assisted conception
- It requires a highly skilled technical team and laboratory set up
- More expensive than the more common PND options
- Deliverable only at a very limited number of providers within a strictly regulated environment.



Pre-Implantation Genetic Diagnosis (PGD)

- It involves checking the genes or chromosomes of your embryos for a specific genetic condition
- It requires In-vitro fertilisation (IVF)
- The embryos are tested in the lab for the genetic defect that it is known in the family
- Embryos which have been tested and are free of the condition will be transferred into the womb
- Patients who meet the funding criteria are entitled to receive three NHS funded cycles of PGD



In-Vitro Fertilisation (IVF)

IVF is a common treatment for people who are unable to conceive naturally or for PGD

Fertility hormones are administered to stimulate the ovaries to produce several eggs

The eggs are then harvested and a single sperm is injected into a single egg

Once the embryo reaches 5 days a few cells are removed and tested in the laboratory

A good quality embryo that is not affected is then transferred to the woman's womb

If more than one embryo survives, they are frozen

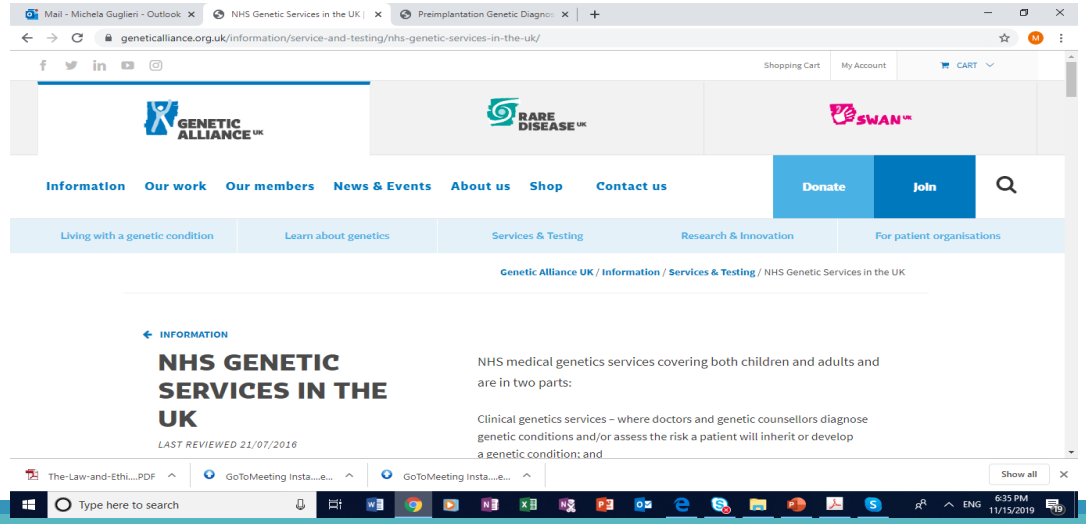
Only one embryo is transferred at a time as transferring more than one embryo increases the chance of having twins or triplets, which carries health risks

The frozen embryos can be used later if the first cycle is unsuccessful . If the couple have a successful outcome they can pay for transfer of another embryo at a later date but this is not funded by the NHS

https://www.hfea.gov.uk/treatments/explore-all-treatments/in-vitro-fertilisation-ivf/

How to get PGD treatment

- Referral to a Regional Clinical Genetics Service by the GP for advice about all options.
- If it's been agreed to proceed with PGD, referral to a specialist at a fertility clinic that has a licence from the Human Fertilisation and Embryology Authority to carry out PGD treatment.



PGD – who is eligible?

- Previous termination of pregnancy due to a serious genetic condition
- Having a child with a serious genetic condition and want to avoid this happening again
- Carrier/s of a serious genetic condition
- Carrier of a chromosomal disorder.

PGD – access criteria

- The couple must be at risk of having a child with a serious genetic condition.
- The couple must have been referred to the PGD provider by a NHS Clinical Genetics Service
- The risk of conceiving a pregnancy affected by a serious genetic condition must be 10% or more
- The couple must have received genetic counselling from a clinical geneticist or a registered genetic counsellor.

PGD – access criteria

- The female partner must be < 40 years of age at the time of treatment
- The female partner must have a BMI > 19 and <30.
- Both partners must be non smokers
- There must be no living unaffected child from the current relationship.
- The HFEA must have licensed the indication for PGD.
- The test must be included in the list of UK Genetic Testing Network (UKGTN) approved tests

PGD – rate of success and risks

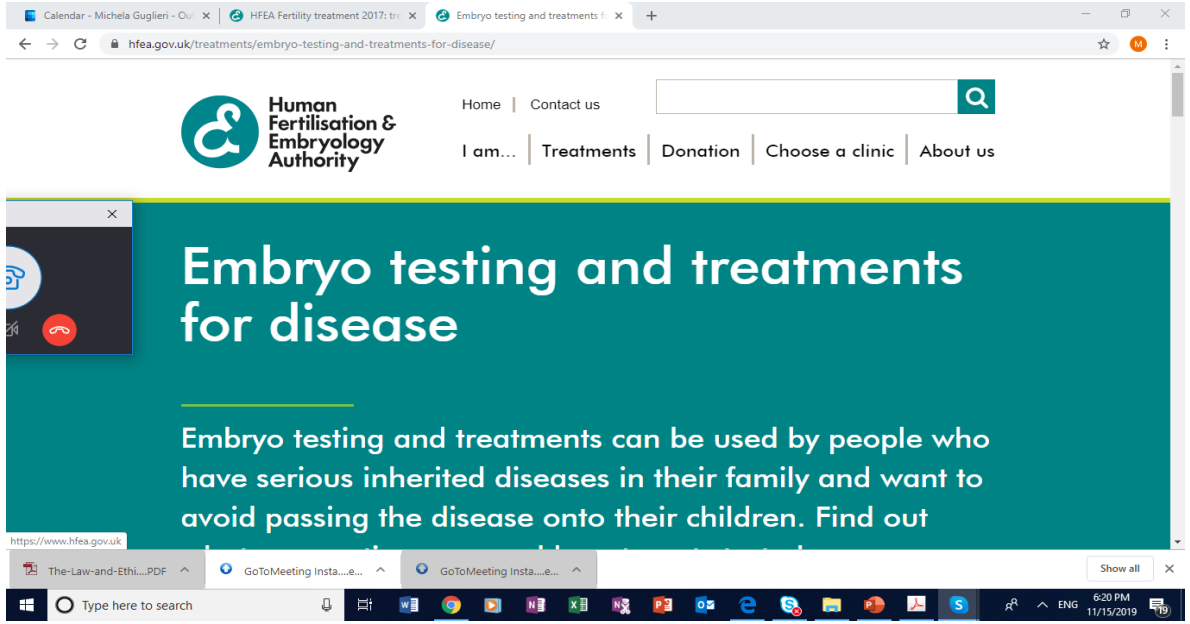
- Success depends on many factors, including the woman's age and whether there are any existing fertility problems.
- Current successful rate is reported = 38% per embryo transferred (2017)
- PGD is not 100% accurate so there's a small chance the tests may not work or may give the wrong information
- The PGD treatment itself is thought to be very safe
- There are risks from having IVF

Risks of IVF

Generally very safe

Potential risks include:

- Ovarian hyperstimulation syndrome(a severe reaction to fertility drugs)
- Having a multiple pregnancy or birth (twins, triplets or more), which can cause serious health problems to both mum and babies
- Having an ectopic pregnancy
- Possible birth defects (these are rare and research is still ongoing).



https://www.hfea.gov.uk/treatments/embryo-testing-and-treatments-for-disease/

https://www.hfea.gov.uk/treatments/explore-all-treatments/in-vitro-fertilisation-ivf/

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https://www.hfea.gov.uk/media/2894/fertility-treatment-2017-trends-and-figures-may-2019.pdf

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THANK YOU!