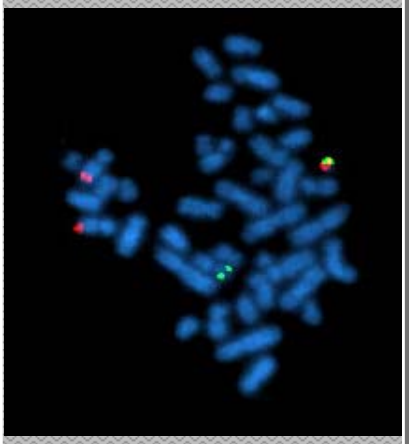

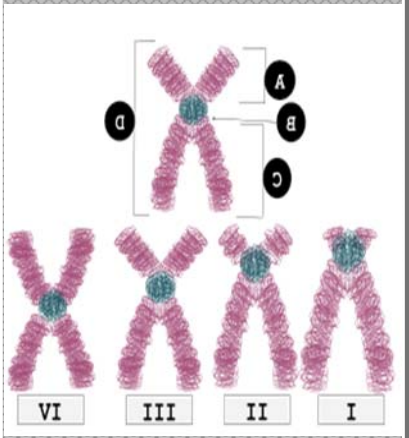

EMGEN Newsletter

Vol. 6, Issue 12

IN THIS ISSUE:

Eastern Mediterranean Health Genomics and Biotechnology Network (EMGEN) was created in 2004 with collaboration of representatives of selected centers of excellence in (health related) molecular biology, biotechnology & genomics in the Eastern Mediterranean region by recommendations and efforts of WHO/ EMRO. Sponsored by Iran Biotechnology Development Council.

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OVERVIEW OF PGD

The ratio of live births in some areas is low. In others, mostly in the developing countries where newborn and maternal mortalities are high, birth rates stay similarly high.

In the developed countries, the way of making healthy babies via a kind of method named as PGD has become medicalized.

Preimplantation Genetic Diagnosis (PGD) is a special way used before implantation to help recognition of genetic defects (mutation, translocation and etc.). In this method, only healthy embryos (without any genetic defects) are moved to the uteruses for implantation.

The methods can also be named preimplantation Genetic Profiling (PGP) since they are usually used on oocytes, polar body, blastomere or embryos before transferring the embryo to the women's womb. PGD establishes alternative post-conception diagnostic ways (Alpha-fetoprotein, Amniocentesis, Estriol, Chorionic villus sampling and etc.) which are often followed by the hard choice of gestation termination if genetic outcomes are undesirable.

History of PGD

Robert Edwards et al. announced the successful sexing of rabbit blastocysts and after that PCR was identified as a tool for sexing determination in patients with the X-linked disorder. After a few years in 2002 first 1000 babies were born by PGD and ART(Assisted reproductive technology).

Edwards won several prizes and awards such as:

1. He was selected as a Fellow of the Royal Society (FRS) in 1984
2. In 2001, he received the Albert Lasker Clinical Medical Research Award and etc.

Figure 1. Robert Edwards

History of Preimplantation Genetic diagnosis	
1967 Biopsy of mice embryo	**1968** Sexing of Rabbit Embryos
1989-90 First PGD Baby was Born (for an X-linked disorder)	**1993-2000** PGD for Aneuploidies ,Chromosomal Translocations and Late Onset Common Disorders
2002-2006 First Thousand PGD Babies were Born	**2009-2013** PGD for Chromosomal Disorders by Microarray-Based Technology: Next Generation Sequencing for Preimplantation Aneuploidy Testing

Nowadays both technology and medical genetics can identify whether an embryo will have hundred different genetic or chromosomal abnormalities. In addition, Preimplantation Genetic Screening (PGS) is a proper term for removing at least one cell from an embryo to test for aneuploidy

The most important goal of PGD and PGS is to determine whether the young embryo is influenced by chromosomal impairments, aneuploidy or mutation in target gene that then the implantation of abnormal fetus will be prevented. It is mandatory to maintain human embryos quality, during the testing and biopsy .

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1. Edwards, R. and R. Gardner (1967). Sexing of live rabbit blastocysts. *Nature* 214: 576-577.
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4. https://en.wikipedia.org/wiki/Robert_Edwards_(physiologist)#/media/File:Robert_Edwards.jpg

PGD vs. PGS

In Vitro Fertilization (IVF) or *Intracytoplasmic sperm injection* (ICSI) presents a way to entrance into the oocyte. The clinical method "preimplantation genetic testing" includes removal of at least one nucleus from polar bodies or embryos (blastomeres, trophectoderm cells) via sonication or laser. However, the term "preimplantation genetic screening" (PGS) is applied when the *in vitro embryo fertilizations* are screened for aneuploidy and chromosomal structure.

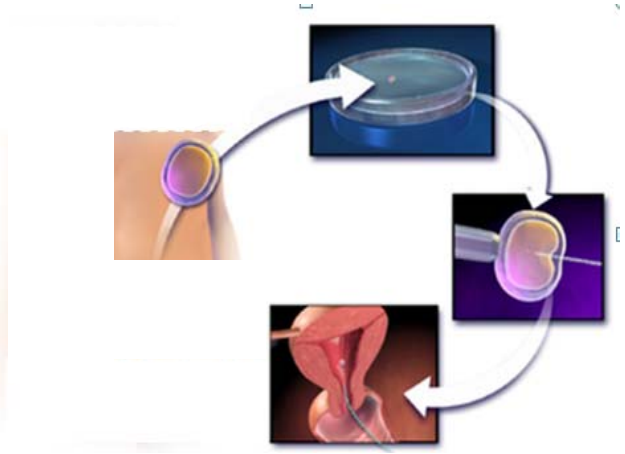
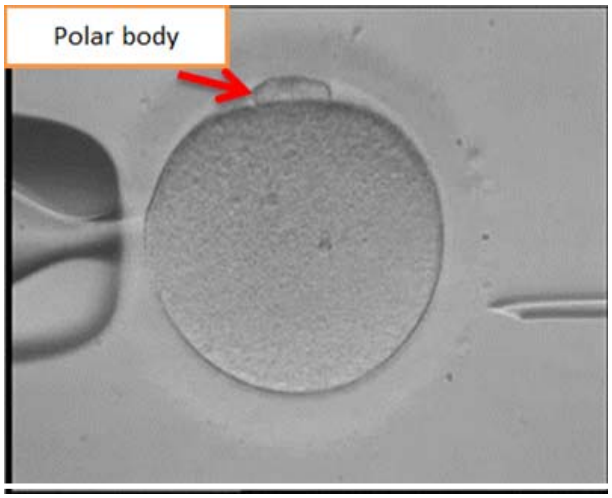


Figure 1. Oocyte fertilization via ICSI

Preimplantation genetic diagnosis step by step

Preimplantation genetic diagnosis (PGD) offers parents to have a child without any inherited genetic disorders or chromosome abnormalities (both in function or numerical disorders).

PGD is a medical chance that gives parents a healthy child without termination painful experience or abortion of an influenced gestation. Abortion is a public health issue, the psychological and physical risks or complications of abortion mostly in repeated conditions in one family, are so important and not inconsequential.



In 2006, a center in London presented data of 330 PGD cases containing 96 and 62 cycles for single-gene and X-linked disorders respectively. Only 58/330 cases were born alive. This result was identical to reported data by the European Society of Human Reproduction and Embryology PGD Consortium in 2014.

Figure 2. Only one fresh sperm per incubated egg is needed in *intracytoplasmic sperm injection* (ICSI) method.

PGD can be used for:

1. HLA matching (savior sibling)
2. Chromosome translocation
3. Aneuploidy screening
4. Severe single gene disorder
5. Cancer predisposition
6. Genetic infertilities
7. Sex discernment in X-linked disorders

PGD can potentially be used to detect a large number of monogenic disorders involving 3 categories, such as:

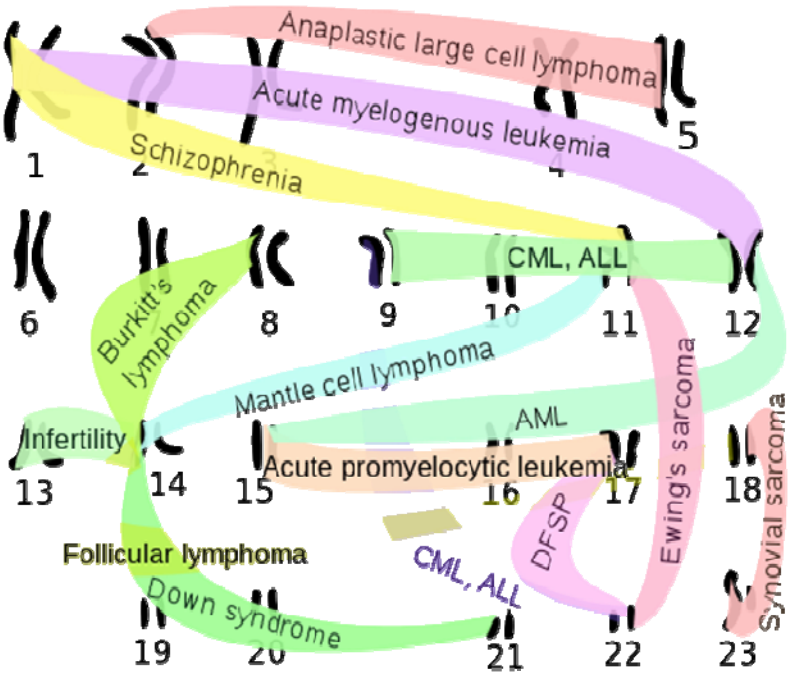


Figure 3. Chromosomal translocations in different cancers

1. **Group 1**

Cystic fibrosis, Beta-thalassemia, Sickle cell disease and Spinal muscular atrophy type 1(e.g.)

2. **Group 2**

Myotonic dystrophy, Huntington's disease, Charcot–Marie–Tooth disease (e.g.)

3. **Group 3**

Fragile X syndrome, Hemophilia A Hunter disease., and Duchenne muscular dystrophy (e.g.), and finally PGD is available for mitochondrial disorders.

Diagnosis of late-onset diseases and cancer predisposition syndromes are other diseases which can be detected via PGD testing to avoid embryo implantation with the same mutation back to the patients. When no HLA identical donor is available in the family with one affected child, PGD will be combine with HLA matching.

This strategy has given a chance to couples to select unaffected embryos with similar HLA typing to recipient child. In this procedure, an expert technician selects an embryo with good quality to have the chances for well ending pregnancy. At birth, hematopoietic stem cells from the newborn umbilical cord blood are used for transplantation.

PGD for Aneuploidy Screening (PGD-AS)

Reciprocal and *Robertsonian* translocations may be identified when couples come to the doctor because of infertility or recurrent pregnancy loss. Chromosome translocation (*reciprocal* or *nonreciprocal*), detected via cytogenetic or a karyotyping, is a chromosome structural abnormality affected by rearrangement or exchange of sections between non-homologous chromosomes. If the break or rearrangement in the chromosome does not delete or disrupt the gene function, Parents with a translocation are unaffected (but carrier).

Mostly, recurrent spontaneous miscarriages are caused by an abnormal chromosomal structure called as an aneuploidy occurring in chromosomes 13, 16, 18, 21, 22 and X, Y. When PGD is carried out for known aneuploidies screening in couples undergoing IVF procedures for infertility or recurrent spontaneous miscarriage, it has been compared with *ESHRE CRITERIA* as preimplantation genetic screening (PGS) or PGD-AS. Polar bodies possess 23 bivalent maternal chromosomes; therefore, they can be used for genotyping, genetic testing and aneuploidy screening of the oocyte without modifying embryo.

How PGD is performed?

There are different methods used to catch oocytes and sperms via ovarian hyper stimulation by drugs and assisted reproductive techniques such as IVF or ICSI respectively. PGD can be carried out on cells from different stages.

Types of Biopsy:

- Biopsy of polar bodies (day 0)
- Biopsy at cleavage stage or the eight-cell stage (day 3)
- Biopsy at blastocysts stage (day 5-6)

Nowadays biopsy at blastomere or trophectoderm stage is the first option for many PGD centers worldwide. Blastomere biopsy is a little useful way which embryos with 8 cells are developed *in vitro* on the third day after insemination.

Biopsy of embryo cells after the third day of development can include potential harmful results, however, this procedure is made possible by incubation in calcium and magnesium-free medium, then the gap is made in the zona pellucida via using the *Tyrode's* solution, sharp needles or laser technology.

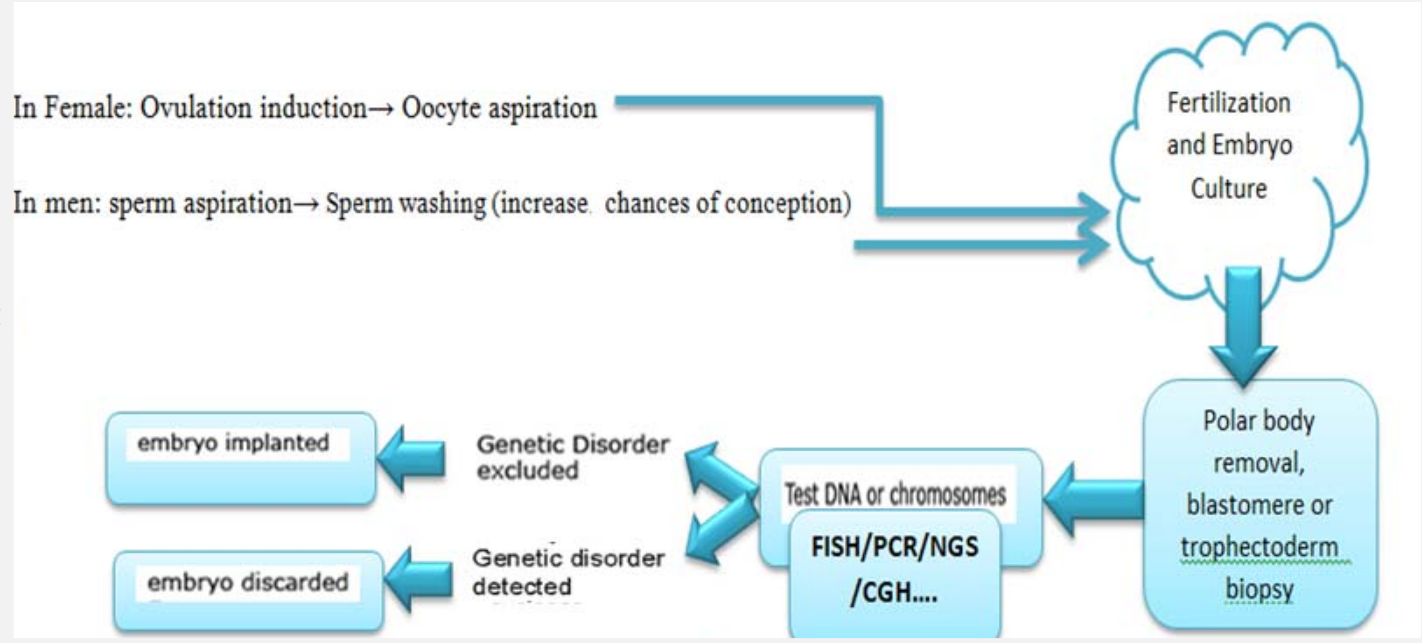
The trophectoderm cells biopsy can be used to select embryos that have a specific genetic disease (MT DNA mutation, AD/AR, X-Linked) to remove allelic dropout problems.

Allelic dropouts are common reasons for genotyping error, missing the healthy embryo and causing a mistake in heterozygous samples. Recently, trophectoderm biopsy is more useful than blastomere biopsy, because trophectoderm cells do become the placenta (a broad circular organ in the mammalian uterus during pregnancy) and less impact the inner cell mass. In this procedure as an alternative to removing blastomeres in PGD, several trophectoderm cells are picked up.

Disadvantages of trophectoderm biopsy

Nowadays all genetic analysis need at least 24 hours, because of incomplete answers, implantation cannot be done, so after the trophectoderm biopsy, cryopreservation technique can be done.

Conclusion



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9. https://en.wikipedia.org/wiki/Chromosomal_translocation
10. https://en.wikipedia.org/wiki/In_vitro_fertilisation#/media/File: Blausen_0060_AssistedReproductiveTechnology.png
11. https://en.wikipedia.org/wiki/Assisted_reproductive_technology#/media/File:In_vitro_fertilization.jpg
12. https://en.wikipedia.org/wiki/Chromosomal_translocation#/media/File:Chromosomal_translocations.svg

ALZHEIMER'S DISEASE: ARE WE CLOSE TO FINDING A CURE?

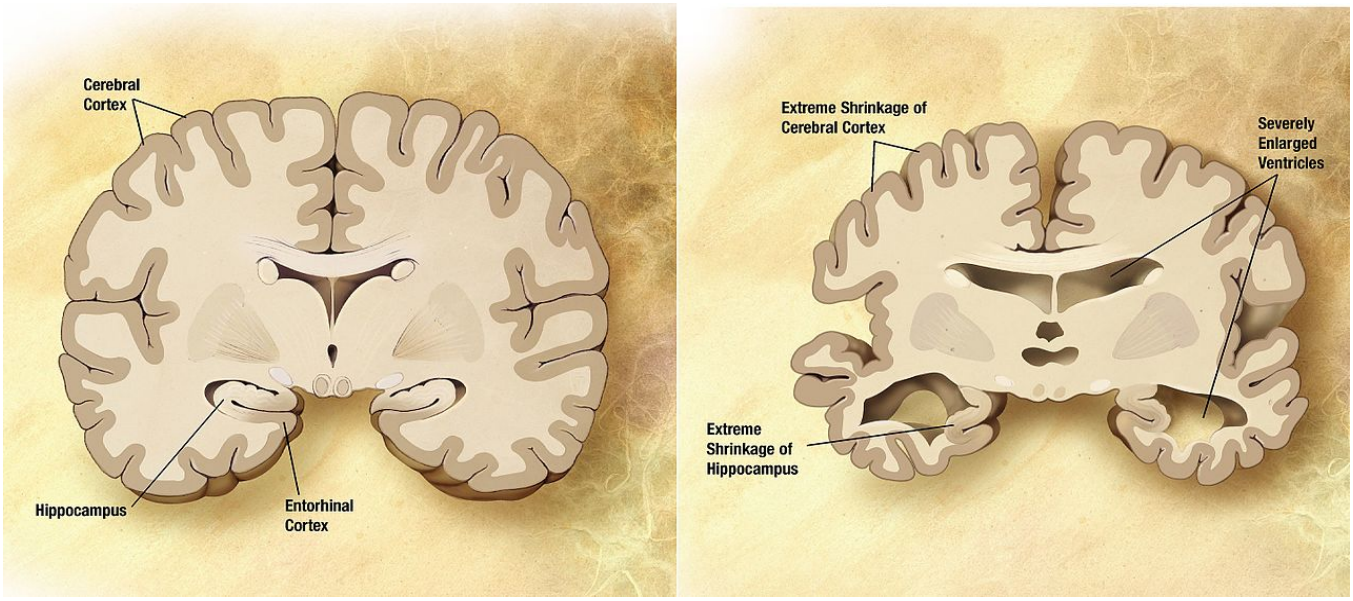

Figure 1. A normal aged brain (left) vs. the brains of people with Alzheimer's (right).

Alzheimer's disease (AD) is a chronic neurological disease that often begins gradually, and does not affect all memory parts; however, capacity of memory and behavioral changes are common and progressively get worse in these patients.

This illness often emerges in individuals older than age 65, but some types of the disease become visible earlier in adulthood meaning before age 50. Memory loss and thinking disorders are the most common signs of neurodegenerative disorder such as Alzheimer's disease. The dementia worsens over time until it disrupts all activities of daily living.

It may become hard to distinguish their families and names. Additionally, routine tasks can be disrupting.

Affected individuals more and more require help with all daily tasks such as dressing, preparing meals, doing laundry and personal care.

Following division by β-secretase, *APP* is separated by γ-secretase to produce Aβ. Presenilins 1 and 2 are the core enzymes of this complex with Nicastrin, Aph1, and *PEN-2*. Several mutations have been found in the *APP* gene. These lead to onset of early-onset familial Alzheimer's disease that happens in the zone of the *APP* gene (encodes the Aβ domain). Genetic testing is accessible for patients or family individuals without any symptoms. Most patients with early-onset Alzheimer are affected by inheritable gene mutations. Researchers have found that three genes are mutated in early-onset Alzheimer: ***APP*, *PSEN1***, or *PSEN2*. When functions of any of these genes are modified, large amounts of amyloid beta peptide are released in the brain.

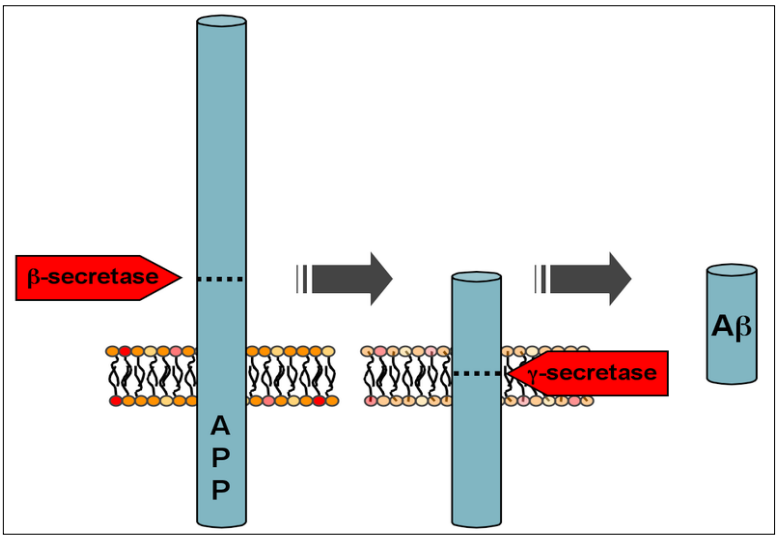

Figure 2. Cleavages of the amyloid precursor protein(APP)

Amyloid beta peptide can accumulate in the environment of brain to amyloid plaques which can lead to the loss of neurons and cause the memory, thinking and behavioral problems linked to AD and get worse over time.

Tau hypothesis

The tau hypothesis proposes that tau protein abnormalities to form neurofibrillary masses inner the nerve cell body. At the same time, these microtubules , damaging the organization of the cell's cytoskeleton which disrupts the neuron's transport function and biochemical communication.

Past studies have suggested that several ways could be decreased signs and symptoms in AD patients. These several ways are:

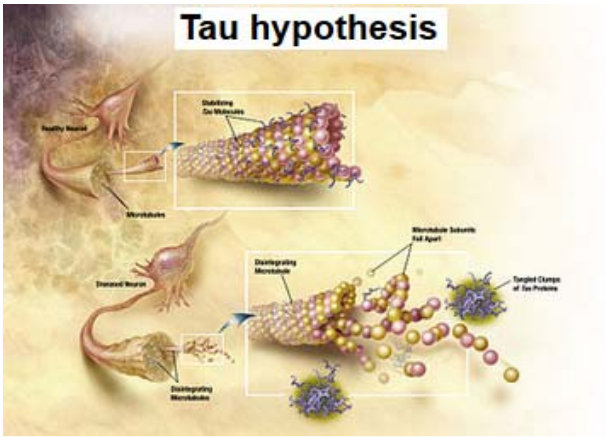

Figure 3. Tau hypothesis in Alzheimer's disease

- Neural stem cells genetically- modified to express neprilysin reduce pathology in Alzheimer transgenic models; Sleep deprivation impairs memory, tau metabolism, and synaptic integrity of a mouse model of Alzheimer's disease with plaques and tangles, (*Neurobiology of Aging*, published 18 February 2014)

- Beneficial effects of caffeine in a transgenic model of Alzheimer's Disease-like Tau pathology (Christa E. Müller, David Blum, et al., *Neurobiology of Aging*)
- Oral glycotoxins are modifiable causes of dementia and the metabolic syndrome in mice and humans (Helen Vlassara et al., published in the Proceedings of *the National Academy of Sciences,* 24 January 2014)

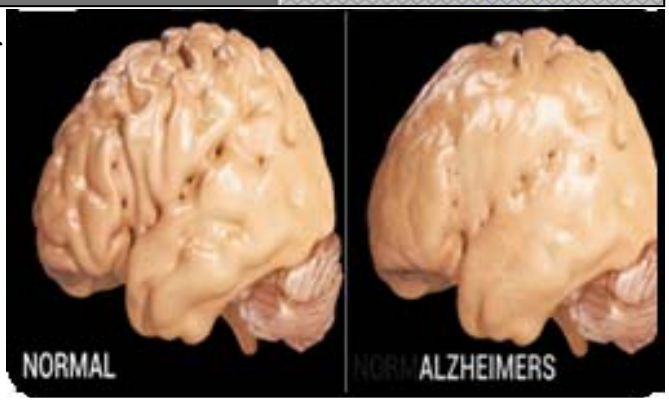

Figure 4. Cortical atrophy in Alzheimer's Disease

References:

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3. https://en.wikipedia.org/wiki/Alzheimer%27s_disease#/media/File:Alzheimer%27s_disease_brain_comparison.jpg
4. https://en.wikipedia.org/wiki/Beta-secretase_1#/media/File:APP_processing.png
5. https://en.wikipedia.org/wiki/Alzheimer%27s_disease#/media/File:TANGLES_HIGH.jpg
6. https://en.wikipedia.org/wiki/Alzheimer%27s_disease#/media/File:Alzheimer%27s_Disease.gif

IS GENE THERAPY AVAILABLE TO TREAT MY DISORDER?

Gene therapy is a new method that uses genes and their belongings to cure or inhibit disease. In the next years, this way may help doctors to treat a genetic or congenital disorder by putting a gene into a patient's cells or special clones . Researchers are testing several ways to gene therapy such as:

1. Replacing a mutated gene with a healthy one.
2. Knocking out a mutated gene.

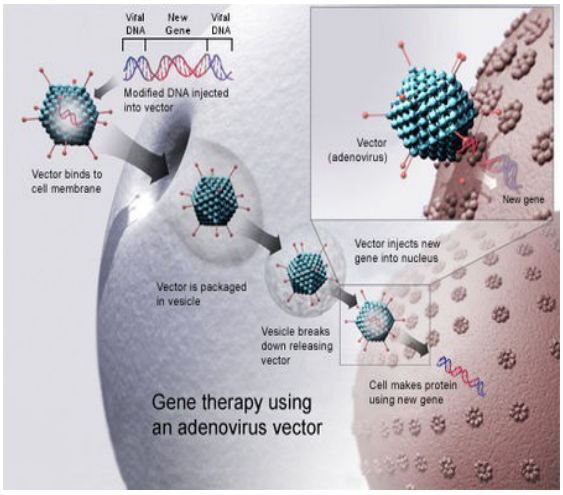


Figure 1. Gene therapy

Gene therapy can be classified into two groups:

1. somatic cell gene therapy (SCGT)
2. germline gene therapy (GGT)

In SCGT, the healing genes are moved into any cells other than germ cells types. In somatic gene therapy, therapeutic DNA with or without plasmid is applied to treat genetic disorders. However in germline gene therapy germline cells (sperm or eggs) are improved by healthy genes into their genomes. This way leads to all the organism's cells have the altered gene. In GGT method, evolution is heritable. Because of their side effects, wrong replacement, short effectiveness and etc. these techniques are precarious. Nowadays gene therapy is only being examined for the treatment of disorders in patients that have no other treatments like cancer, and HIV/AIDS.

Reference:

1. https://ghr.nlm.nih.gov/primer/therapy/availability
2. https://en.wikipedia.org/wiki/File:Gene_therapy.jpg

NATURE REVIEWS. GENETICS

ISSN: 1471-0056

Scope : Genetics, Genetics (clinical), Molecular Biology principles and applications in medicine, genetic technology, scientific breadth of modern genetics, environmental concerns and regulatory issues.

Impact Factor: 35.898

GENOME RESEARCH

ISSN:1549-5469

Scope :includes a new vision about the genome biology of all organisms, genomic medicine, genome structure and function, comparative genomics, molecular evolution, genome-scale quantitative and population genetics, proteomics, epigenomics.

Impact Factor: 14630

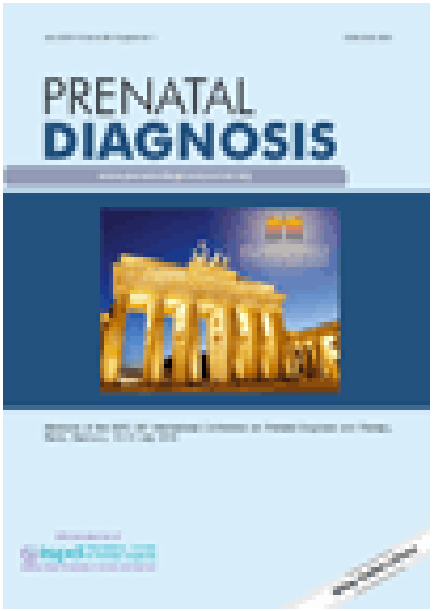
CIRCULATION CARDIOVASCULAR GENETICS

ISSN:1942-3268 (Electronic),1942-325X (Print)

Scope: includes about Cardiology and Cardiovascular Medicine, Genetics, Genetics (clinical), Molecular Biology,Medicine (miscellaneous)

Impact Factor: 6.728

PRENATAL DIAGNOSIS



ISSN: 1097-0223

Scope: prenatal cytogenetics, prenatal screening studies,preimplantation genetic diagnosis (PGD) ,fetal therapy and etc.

Impact Factor: 3.043

Announcements

1967 - 2017: 50th Anniversary of the ESHG
The European Human Genetics Conference 2017

Copenhagen, Denmark
May 27-30, 2017

ESHG
EUROPEAN SOCIETY OF HUMAN GENETICS

https://www.eshg.org/94.0.html

http://isv.variome.org/

Announcements

Frontiers of Immunology in Health & Disease, AWAJI, JAPAN

Awaji Yumebutai Conference Center, Japan

October 3-6, 2016

http://www.csh-asia.org/2016meetings/immune.html

http://www.eurovirology2016.eu/

Announcements

http://www.icecb.org/

http://www.icecb.org/

Announcements

https://www.eshg.org/830.0.html

http://lifescienceevents.com/reproductive17/

FLUORESCENCE IN SITU HYBRIDIZATION

Fluorescence in situ hybridization (FISH) was developed by Ward, D. C et al in 1980. FISH is a cytogenetic and molecular technique that permits genetic counselors and researchers to identify:

1. The positions of specific DNA
2. The Positions of genes and sequences
3. Chromosomal rearrangements
4. Aneuploidy
5. Numerical disorders
6. Specific RNA targets

In this method, fluorescent probes bind to only specific parts of the chromosome, so it can detect several abnormalities in a chromosomal structure by Fluorescence microscopy.

References:

1. Langer-Safer, P. R.; Levine, M.; Ward, D. C. (1982). Immunological method for mapping genes on Drosophila polytene chromosomes. *Proceedings of the National Academy of Sciences* **79** (14): 4381–5
2. https://en.wikipedia.org/wiki/Fluorescence_in_situ_hybridization

TURNER SYNDROME

Turner syndrome (TS) or gonadal dysgenesis is a chromosomal condition in a female with partly or entirely lacking an X chromosome. About half of girls with TS have only one copy of the X chromosome in their cells. Before the birth, prenatal ultrasound of baby girl with TS may display different features such as:

1. Abnormal fluid collections
2. Heart or kidney abnormalities
3. Several common and important signs of Turner syndrome:
4. Short fingers and toes
5. Short stature
6. Delayed growth
7. Short and webbed neck
8. Early loss of ovarian function

Only several females with Turner syndrome have the normal ovarian function. The chromosomal missing or abnormalities may be available in some cells in baby girls with TS, these individuals are also known as TS with mosaicism; therefore, in these individuals signs and symptoms are usually fewer than other TS. However, there isn't any specific treatment for TS, recognition is based on signs, symptoms, and genetic testing. Furthermore, human growth hormone injections and estrogen replacement therapy may reduce their problems.

Reference: https://en.wikipedia.org/wiki/Turner_syndrome

CLASSIFICATIONS OF CHROMOSOMES

Each duplicated chromosome is identified by two arms, named p (the shortest arm or petit) and q (the longest arm), centromere and telomere. They can be collected in to 4 groups: metacentric, sub metacentric, acrocentric or telocentric manner. These groups have special features which are classified in the following table:

4 major Types of chromosomes	Features	In human genome
Metacentric	The centromere is in the middle of the arms, and in these chromosomes the length of P is equal to q	Six chromosomes are considered metacentric: chromosomes 1, 3, 16,18, 19, and 20
Sub-metacentric	P and q arms are very close in length but not equal. Slight asymmetry exists in the length of the two sections .q>p	Twelve chromosomes are located in Sub-Metacentric group.
Acrocentric	The centromere is near telomere , and the p arm is so much shorter than q arm, this chromosome is named acro-centric.	The human genome involves six acro-centric chromosomes: 13, 14, 15, 21, 22 and the Y chromo-some. These chromosomes are located in D & G groups.
Telocentric	In this group centromere is located at the terminal end of the chromosome called as a Telomere.	Humans don't have any telocentric chromosomes.

Reference: https://en.wikipedia.org/wiki/Centromere#Acrocentric