GENETICS

Sheet no.14

Writer:

Leen Aburumman &

Layan Al-zoubi

Corrector:

Nermeen Abuhalaweh

Doctor:

Mohammad Al shboul

Non- Mendelian patterns of inheritance.

- Some disorders do not follow Mendelian patterns of inheritance.
- These disorders are clearly genetic (inherited) and their inheritance is classified as non-Mendelian.
- We now understand why some of these disorders do not follow Mendelian patterns and examples include: mitochondrial inheritance, unstable trinucleotide repeats, and imprinting.

MITOCHONDRIAL DNA (MTDNA)

- Most genetically inherited diseases are caused by mutations in the nuclear genome, however, there is a small but significant number of diseases that are caused by mitochondrial genome mutations.
- Mitochondria are double-membrane-bound organelles that are present in all nucleated eukaryotic cells and are responsible for the production of cellular energy required for the body to function through metabolic processes, in particular, oxidative phosphorylation
- The mitochondrion has been called the 'powerhouse' of the cell, as produce most of the energy in ATP form.
- Most organs in our body need mitochondria. The most energy dependent organs are the brain, heart, skeletal muscle, kidney, endocrine glands and bone marrow and these are the organ systems commonly affected in mitochondrial diseases -either single organ defects or multi organs defects, that's why mitochondrial diseases are difficult to diagnose clinically, usually we must do genetic testing for mitochondrial genome after excluding every other mutation in the nuclear genome.

MITOCHONDRIAL INHERITANCE:

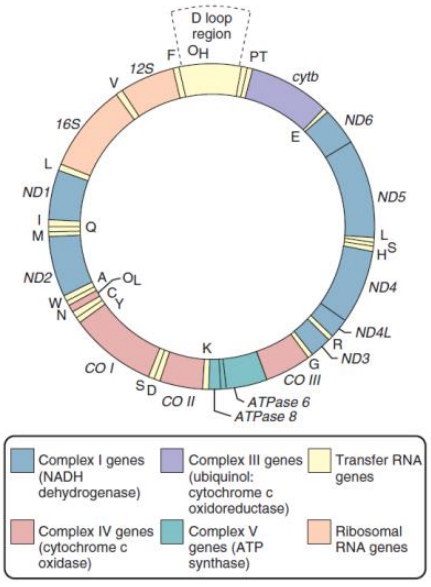
- Multiple copies of mtDNA exist within each cell and the total amount can vary between a few hundred and many tens of thousands of copies, depending on the cell type – cells need of energy, the higher the number mitochondria in the cell, the more effect of the mitochondrial disease on it -.
- If we compare between mtDNA and nuclear DNA we will find that:
    - mtDNA (37 genes) is much smaller than the nuclear DNA (20000-25000 genes codes for proteins)
    - mtDNA is usually a circular and double-stranded, nuclear genome is double-stranded DNA molecule that is linear

- mtDNA is found in the mitochondria, whereas nuclear genome is in the nucleus.
- mtDNA encodes genes for mitochondrial function and energy metabolism while the nuclear genome encodes a wide variety of proteins for cellular structure, function and regulation.
- mtDNA replicate independently of the cell cycle and it has high mutations rate, while nuclear genome replicates in the S phase of interphase and have a wide variety of proteins that fix defects that happen during replication.
- mtDNA does not contain histones, while nuclear DNA is wrapped around them.
- mtDNA Is very compact, containing little repetitive DNA, and codes for essential enzymes involved in ATP production

- There are a few exceptions where mtDNA is linear, generally in lower eukaryotes such as yeast and some other fungi.
- mtDNA differs greatly in size among organisms.
- In animals, it is typically 16–18 kb (around 0.3% of total genome),
- Mutations in either the mtDNA or the nuclear genome underlie the largest collection of inborn errors of metabolism and diseases, these diseases have different age of onset and severity even in the same family, and there is growing evidence that a gradual decline in mitochondrial activity is associated with aging and age-associated disorders.

MITOCHONDRIAL DNA (MTDNA)

- It carries relatively 37 genes only 24 of which code for RNA.
- 2 rRNA and 22 tRNA genes, these are used for translation, which means that the proteins that are encoded by the mitochondria are translated separately from the rest of the genome, So the mitochondria use its own compartments to produce the proteins.
- Ponder the figure, mtDNA is circular, double-stranded, and encodes for tRNA and 13 proteins.



- mRNA: Mammalian mitochondria contain their own genome (mtDNA), which encodes a total of 13 proteins that are all core components of oxidative phosphorylation. **This does not mean that there are only 13 mitochondrial diseases,** there are hundreds of them and the majority of mitochondrial diseases are caused by *nuclear genome*.
- Nuclear genes: around 1300 (>99%) proteins are nuclear encoded and imported into the organelle mitochondria.

Mitochondrial Inheritance

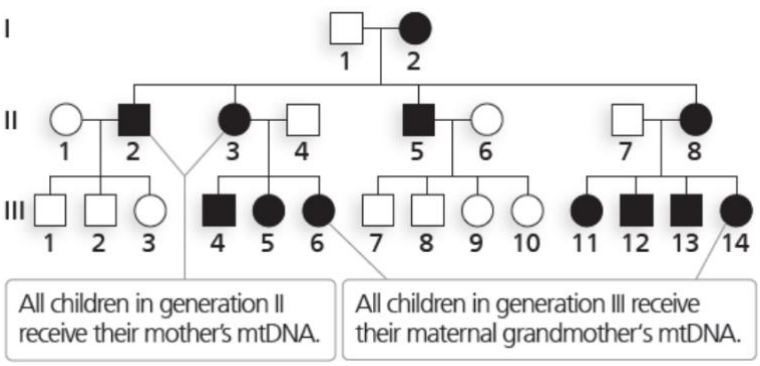
- In humans, at fertilization, the ovum contributes significantly more cytoplasm to the zygote than does the sperm.
- The sperm mitochondria degenerate upon penetration of the ovum.
- Mitochondria in offspring are exclusively maternal in origin, while the nuclear DNA is formed by equal contribution from both parents.

The pattern of inheritance of organelles

- Mutations in mitochondrial genes are also the cause of several single gene disorders.
- Mutation rate in mt is 10 times more than in nuclear DNA due to?

- Limited repairing mechanisms of the mtDNA or completely lack of them.
- Oxidative phosphorylation in the mitochondria produces a lot free oxygen radicals that can damage the DNA by a rate that exceeds any repair system.

- Mother's children all inherit the trait.
- Father's children never inherit the trait!
- In the following pedigree, note that the mother in I is affected and all of her children in II are affected as well, all of the daughters of II have transfer the disease to all of their children in III. So, if the mother is affected, all of her children will also be affected. While males cannot transfer the disease to their children – note II,2 and II, 5.
- The type of inheritance here is termed *mitochondrial/ maternal/ cytoplasmic inheritance*.

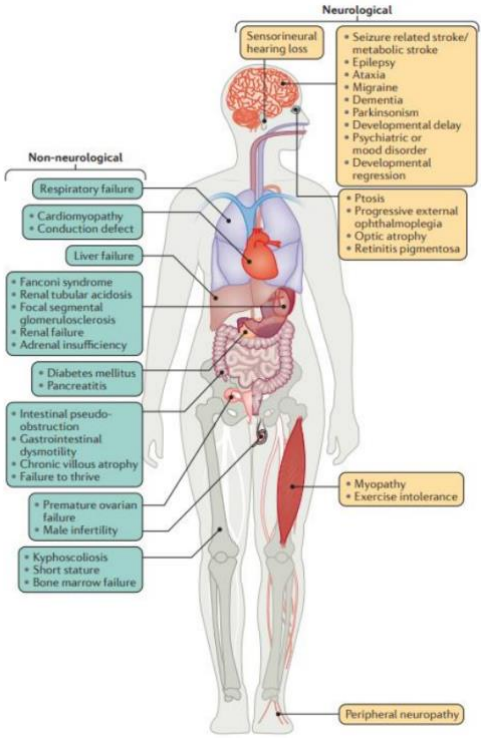


The types of mitochondrial disease Inheritance

- The pathophysiology of mitochondrial diseases is complex and involves genetic mutations in mitochondrial DNA (mtDNA) and nuclear DNA (nDNA).
- Autosomal and X-linked inheritance for nDNA depending on its location, can be dominant or recessive, so not all mitochondrial diseases are maternal.
- Maternal or mitochondrial inheritance for mtDNA mutations.
- de novo mutations which is actually responsible for a good number of new diseases (25%), where the disease occurs in an affected child with no family history and both parents are not responsible for this mutation.

Mitochondrial diseases

- Mitochondria are under the dual control of mitochondrial DNA and nuclear DNA, not caused by a single gene.
- More than 40 known types can be caused by any pattern of inheritance; heterogenous group of disorders.
- Mitochondrial disease is a difficult disorder to identify because it can take many forms depending on the affected location and range from mild to severe causing a wide range of symptoms.



- Such disorders are heterogeneous, genetically inherited and can affect an individual at any age and in any body organ, including nerves, the brain and other major organs.
- Mitochondrial disease is a chronic (there is no cure for the disease, only supportive therapy for the symptoms that may arise), genetic disorder that occurs when the mitochondria of the cell fail to produce enough energy for cell or organ function - dysfunction of mitochondrial respiratory chain-.
- Mitochondria is important for on function of the majority of cells. The higher tissues' needs for energy, the higher the defects and the severity of the severity of the disease. For example, the nervous system, usually the main symptoms of mitochondrial diseases are related to the nervous system. other examples include the muscles – needs high energy, the eyes, the heart…etc. So, the disease symptoms will be mostly in these organs as they are the most affected by the disease.

How Common is Adult Mitochondrial Disease?

- Mitochondrial dysfunction can arise because of defects in either mitochondrial DNA or nuclear mitochondrial genes, and can present in childhood or adulthood in association with vast clinical heterogeneity, with symptoms affecting a single organ or tissue, or multisystem involvement.
- These include
  - Type II Diabetes Mellitus,
  - Parkinson Disease,
  - Atherosclerotic Heart Disease,
  - Stroke,
  - Alzheimer Dementia, and
  - Cancer.
- It is clear that mitochondria are involved because their function is measurably disturbed. Even autoimmune diseases such as multiple Sclerosis, Systemic Lupus Erythematosus, and Rheumatoid Arthritis appear to have mitochondrial components.

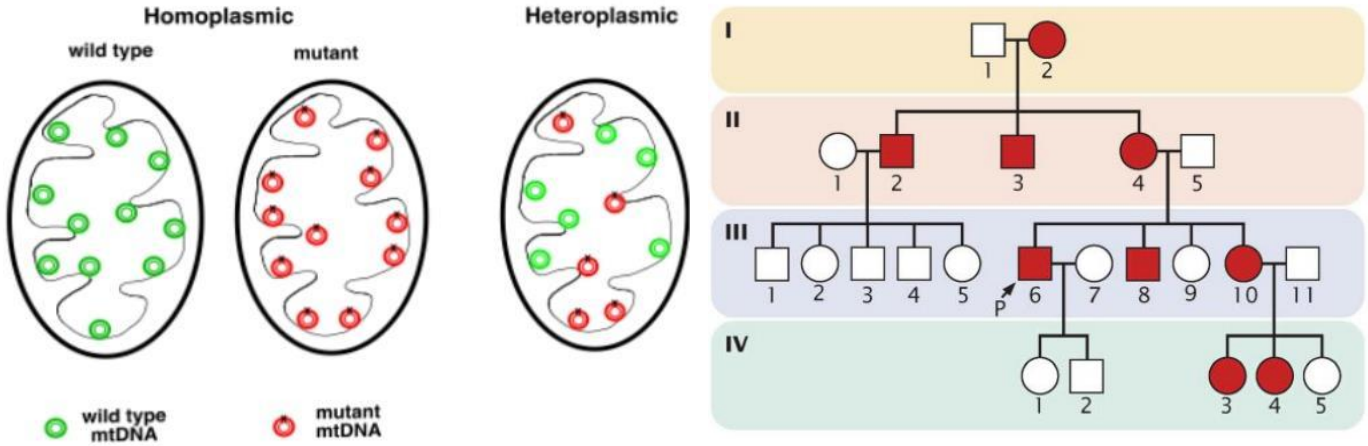
Examples of Diseases Due to Mutations and Deletions in Mitochondrial DNA

Abbreviation	MIM No.	Designation
LHON	535000	Leber's hereditary optical neuropathy (Missence M)
MELAS	540000	Mitochondrial encephalomyopathy with lactic acidosis and stroke-like episodes
	540050	Lactic acidosis with stroke-like signs (Single base M)
MERRF	545030	Myoclonic epilepsy and ragged red fibers (Single base M)
MMC	590050	Maternally inherited myopathy and cardiomyopathy
NARP	551500	Neurogenic muscular weakness with ataxia and retinitis pigmentosa
CEOP	258470	Progressive external ophthalmoplegia
KSS	530000	Kearns-Sayre syndrome (ophthalmoplegia, pigmental degeneration of the retina, and cardiomyopathy)
PEAR	557000	Pearson syndrome (bone marrow and pancreatic failure)
ADMIMY	157640	Autosomal dominant inherited mitochondrial myopathy with mitochondrial deletion in the D loop (type Zeviani)

- Note that the name of the disease indicates what organ will be affected (muscles, eye, nervous system), so from the name of the disease you should be able to know if it is mitochondrial or not.

- This will help us in the exam =), the doctor said that he would ask about the diseases but we don't have to memorize them – the diseases in the previous two slides -, only know if it is mitochondrial or not from what the name indicates 😐.
- A typical human cell — including the egg cell — contains only one nucleus but hundreds to thousands of mitochondria. A single cell can contain both mutant mitochondria and normal mitochondria, and the balance between the two will determine the cell's health.
- This helps explain why the symptoms of mitochondrial disease can vary so much from person to person, even within the same family.
- Imagine that a woman's egg cells (and other cells in her body) contain both normal and mutant mitochondria, and that some have just a few mutant mitochondria, while others have many. A child conceived from a "mostly healthy" egg cell probably won't develop disease, and a child conceived from a "mostly mutant" egg cell probably will.
- Also, the woman may or may not have symptoms of mitochondrial disease herself. This can explain why a healthy woman can give birth to affected children, in this case, this woman is considered a carrier, because her mitochondria are mixed, normal and affected, but the number of the affected mitochondria are not enough to cause the disease.
- These diseases also can arise in a sporadic fashion, meaning they may occur with no family history.

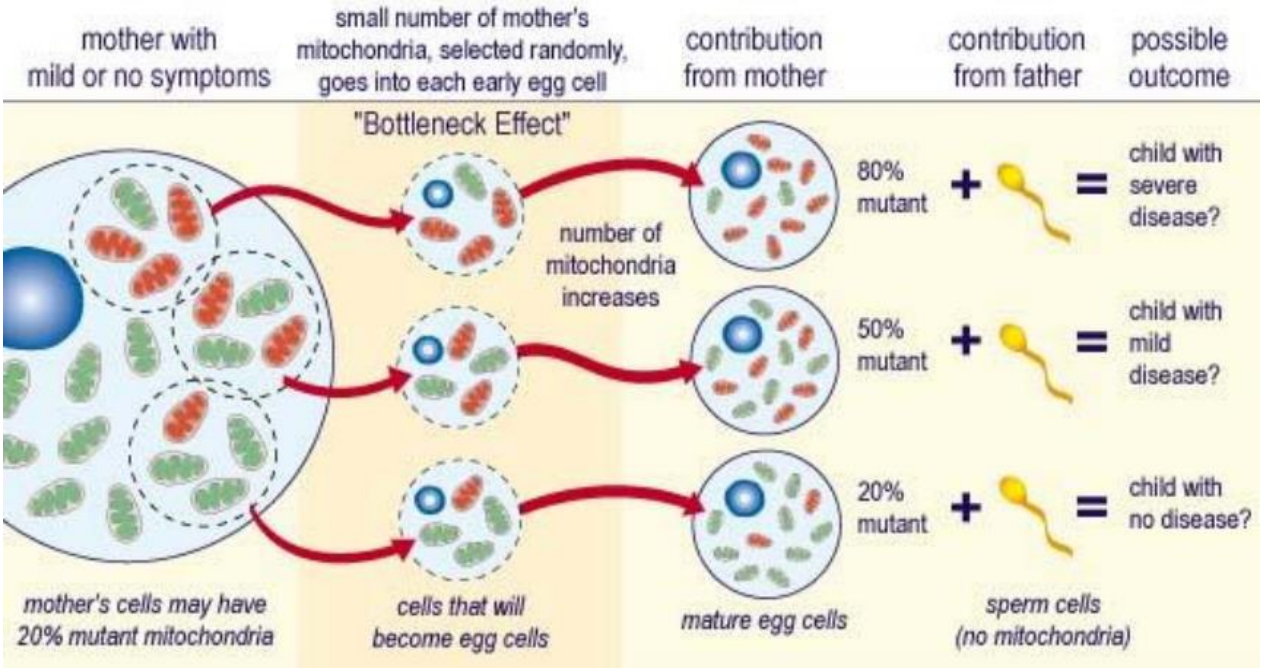
Heteroplasmy VS. Homoplasmy



- Heteroplasmy: A cell that has some mitochondria that have a mutation in the mtDNA and some that do not – wild type and mutated-.

- The proportion of mutant mtDNA molecules determines both the penetrance and severity of expression of some diseases.
- Homoplasmy: A cell that has a uniform collection of mtDNA: either completely normal mtDNA or completely mutant mtDNA.
- In nDNA, we say heterozygous or homozygous, but because there is high number of mtDNA we say heteroplasmy or homoplasmy.
- A unique feature of mtDNA is that, at cell division, the mtDNA replicates and sorts randomly among mitochondria. In turn, the mitochondria sort randomly among daughter cells. Therefore, in cells where heteroplasmy is present, each daughter cell may receive different proportions of mitochondria carrying normal and mutant mtDNA.

MATERNAL INHERITANCE OF MITOCHONDRIAL DNA MUTATIONS



Red mitos are mutant, green are normal. ponder the figure while studying =)

- The severity of the condition is dependent on the number of disabled mitochondria present in the egg.
- An egg with a large number of disabled mitochondria would result in a child with severe abnormalities, similar to the first child in the picture above.
- A child with 50% mutant mitochondria will have milder symptoms than the first one.
- An egg with only a few -below the threshold- disabled mitochondria would result in an individual only mildly affected or even does not have symptoms at all.

- So, the level of defeat actually depends on the number of defective mitochondria inside the cell and how much this cell depends on mitochondria to do its function. for example, in a skin and a muscle cells with 40% defective mitochondria, the muscle cell will show some phenotypes that the skin cell will not.

Trinucleotide Repeats

- It is a common disease in humans, caused by repeating 3 nucleotides over and over in our genome - although normally we have some hundreds or thousands of repetitions-.
- Some disorders were observed to increase in severity from one generation to another,
- and/or the age of onset of symptoms became earlier in successive generations.
- This was termed **anticipation** and the mechanism was a mystery since mutations were presumed to be inherited in a stable manner from one generation to another.
- Furthermore, in some disorders the sex of the parent who passed on the disorder seemed to influence the severity or age of onset of symptoms. If it was inherited from the father, it will cause the disease and if from the mother, it will not and the other way around.
- This too was a puzzle because in Mendelian traits maternal and paternal DNA was assumed to be equivalent.
- Anticipation and parent of origin effects are now known to be due to a novel type of dynamic mutation known as unstable trinucleotide repeats.
- An example on these diseases is Huntington disease, a neuromuscular disease that is inherited from the father. Whereas myotonic dystrophy, a muscular disease, is inherited from the mother. This is very important when counseling for future pregnancies.
- This suggests that the trinucleotide expansion happens during oogenesis or spermatogenesis, depending on the gene that is involved.
- Anticipation usually depends on whether the disease is inherited from the mother or father.

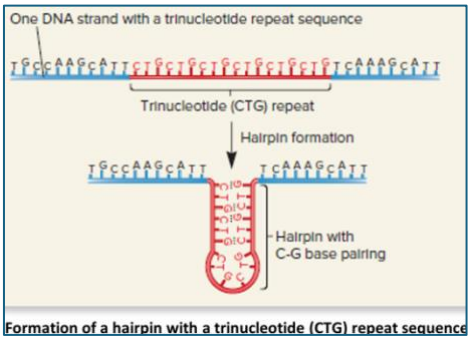
- **Please study the following table carefully as the doctor focused on the outlined diseases** - except SBMA-**.**
- Different repeats cause different diseases.

TABLE 19.5
TNRE Disorders

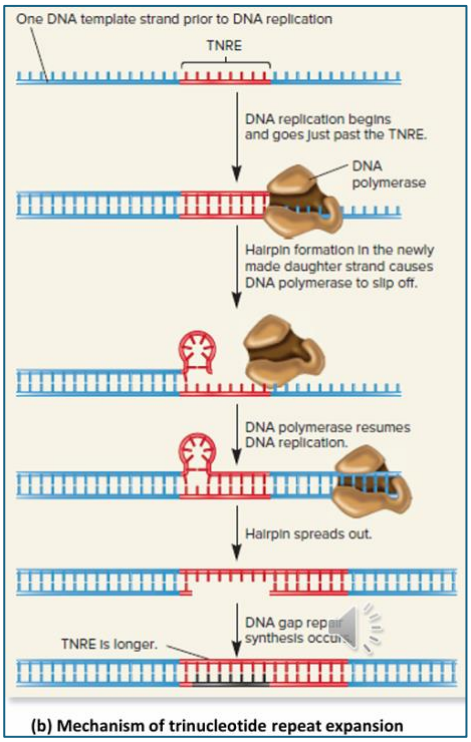
Disease*	SBMA	HD	SCA1	FRAXA	FRAXE	DM
Repeated Triplet	CAG	CAG	CAG	CGG	GCC	CTG
Location of Repeat	Coding sequence	Coding sequence	Coding sequence	5′-UTR	5′-UTR	3′-UTR
Number of Repeats in Unaffected Individuals	11–33	6–37	6–44	6–53	6–35	5–37
Number of Repeats in Affected Individuals	36–62	27–121	43–81	>200	>200	>200
Pattern of Inheritance	X-linked	Autosomal dominant	Autosomal dominant	X-linked	X-linked	Autosomal dominant
Disease Symptoms	Neuro-degenerative	Neuro-degenerative	Neuro-degenerative	Mental impairment	Mental impairment	Muscle disease
Anticipation†	None	Male	Male	Female	None	Female

*SBMA, spinal and bulbar muscular atrophy; HD, Huntington disease; SCA1, spinocerebellar ataxia; FRAXA and FRAXE, fragile X syndromes; DM, *dystrophia myotonica* (myotonic muscular dystrophy).
†Indicates the sex of the parent from whom the disease is usually inherited when anticipation occurs.

- Now the question is how does trinucleotide repeat expansion occur?
- A triplet repeat can form a hairpin, also called a stem-loop.
- The most important thing to find in these repeat expansions is C and G that can allow the formation of a hairpin like structure as presented in the figure.
- During DNA replication, this loop can lead to an increase in the length of a DNA region if it occurs in the newly made daughter strand.
- DNA polymerase may temporarily slip off the template strand.
- Next, DNA polymerase back onto the template strand and resumes DNA replication from the end of the hairpin.
- When this occurs, DNA polymerase is synthesizing most of the hairpin region twice.
- So, the whole point of this process is to increase the repeat expansions from a generation to another.



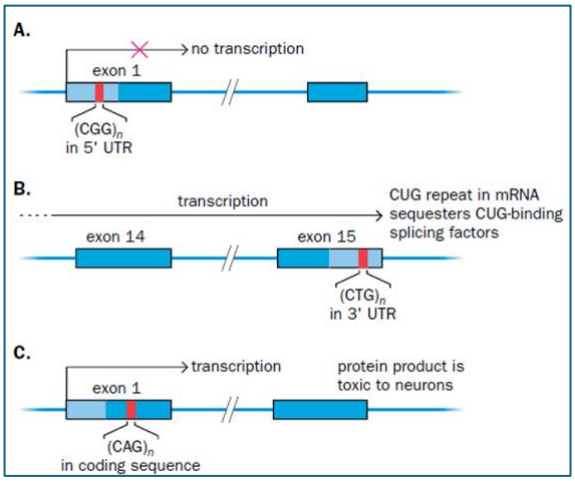
Formation of a hairpin with a trinucleotide (CTG) repeat sequence



(b) Mechanism of trinucleotide repeat expansion

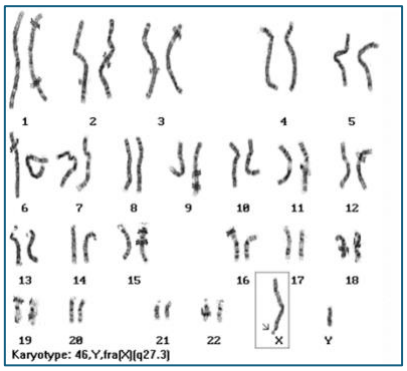
- Trinucleotide repeat expansions are associated with some diseases, especially neurologic and musculoskeletal disorders such as:
    1. Fragile X Mental Retardation syndrome.
    2. Huntington disease.
    3. myotonic dystrophy.
    4. spinocerebellar ataxia.
    5. Kennedy disease.
    6. Joseph disease.
    7. Friedreich Ataxia.

- **Three mechanisms by which dynamic mutations may be pathogenic:**



- In fragile X syndrome, the expanded repeat in the 5' UTR of the gene triggers methylation of the promoter and prevents transcription (No proteins).
- (B) In myotonic dystrophy, the expanded repeat in the 3' UTR causes the mRNA transcript to prevent the correct splicing of several unrelated genes.
- (C) In Huntington disease, the gene containing the expanded repeat is transcribed and translated as normal, but the protein product has an expanded polyglutamine (CAG) tract that renders it toxic (Precipitation of abnormal proteins in the brain tissue).

- **Fragile X Syndrome:**



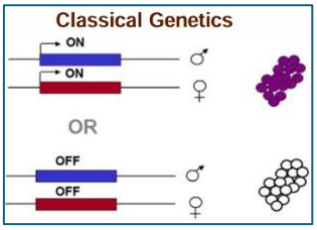
- A fragile site close to the telomere at the end of the long arm at Xq27.3.
- Appears as a gap or unstained region in the chromosome and it is responsible for the mental retardation, the hyperactivity and the other symptoms associated with this disease.
- Most common cause of inherited mental retardation in males (X linked).
- Phenotype in males includes moderate mental retardation, large head, long face, prominent forehead and chin, protruding and larger ears, large testes after puberty, speech delay, and loose joints. Behavior abnormalities include hyperactivity, hand flapping, hand biting, temper tantrums and sometimes autism spectrum disorder.

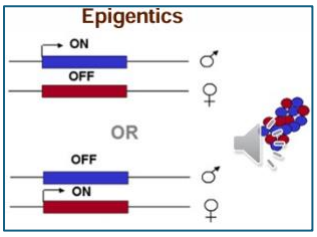
- Keep in mind that these repeat expansions happen in the 5' UTRs and their presence prevents the transcription of the proteins; loos of function mutation.
- The normal triplet repeats range is (5-45) repeats.
- Approximately 50% of female carriers of a full mutation have mental retardation that is usually less severe than in affected males.
- About 30% of males who carry a premutation will develop Fragile X-associated tremor/ataxia syndrome (FXTAS) which is characterized by late-onset, progressive cerebellar ataxia and intention tremor.
- About 20% of females who carry a premutation will develop premature ovarian failure (POF) that is associated with symptoms as early menopause, irregular menstrual cycle, infertility and elevated levels of FSH so the premutation stage is critical because both males and females develop symptoms at a specific age.

Number of Triplet Repeats (Normal Range 5–44)
Males
45–54 (intermediate alleles)
55–200 (premutation)
200–2000 (full mutation)
Females
45–54 (intermediate alleles)
55–200 (premutation)
200–2000 (full mutation)

- Genetic features:
  - Atypical X-linked inheritance showing parent of origin effect.
  - In affected males associated with a fragile site at Xq27.3 in 10-40% of metaphase spreads, however, this cytogenetic testing is no longer used for diagnostic testing.
  - Amplified 'CGG' trinucleotide repeat as well as abnormal methylation (hypermethylation) of the FMR-1 gene. The normal protein product, FMRP, is an RNA-binding protein that seems to function as a nucleocytoplasmic shuttling protein and it binds several mRNAs including its own. It also seems to affect cytoskeletal structure, synaptic transmission and neuronal maturation.
  - The FMR-1 gene mutation results in gene silencing and the loss of function results in suppression of translation of proteins from its RNA targets.
  - The presence of such repeats in the 5' UTRs prevents the translation, especially the cytosine ones because they are prone to methylation that silences them giving rise to a loss of function mutation.

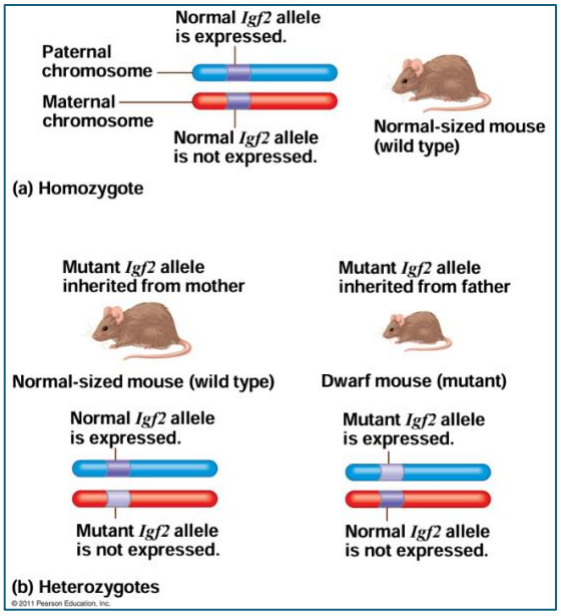
Epigentics





- Cells normally have two copies (alleles) of autosomal genes on chromosomes other than the X and Y.
- One allele is inherited from the mother (maternal allele) and one is inherited from the father (paternal allele).
- For most genes, both copies are expressed by the cell. However, a small class of genes is "monoallelically" expressed (In such cases, one allele is expressed and the second one is silenced and this is *epigenetics*).
- We have two types of monoallelic expression: The first one being the random type and it happens in immune cells causing immune diversity, and the second is genomic imprinting that depends on the parental origin of the gene.
- Genomic imprinting: A small class of genes shows monoallelic expression, where a single allele in a cell is preferentially expressed and the functionally silenced gene is said to be imprinted.
- Offspring expresses either the maternally-inherited or the paternally-inherited allele but not both (parent-of-origin expression).
- Imprinted genes - functionally haploid.
- In most cases of genomic imprinting, the copy of a gene inherited from one parent is transcriptionally inactive in all or most of the tissues in which the copy from the other parent is active.
- The term "imprinting" signifies that whatever silences the maternal or paternal copy of an imprinted gene is not encoded in its DNA sequence but due to epigenetic alteration.
- Genomic imprinting involves the transmission of epigenetic information, in the form of DNA methylation marks, from gametes to offspring, with the result that a set of genes (both protein coding genes and non-coding RNA genes) are expressed from only one of the two chromosomes in cells.
  - At present, over 100 genes have been confirmed to be imprinted in humans.
  - These genes tend to have important roles in development and the loss of imprinting is implicated in several genetic diseases and types of cancer in humans.
- DNA methylation (Mainly cytosine) is the main mechanism by which the expression is modified. In addition to hyperacetylation and microRNA.

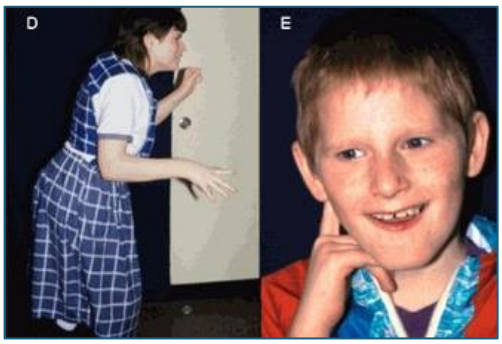
- Now let's discuss this example to elaborate more on genetic imprinting; we have the Igf2 gene that is responsible for growth, development and the prevention of apoptosis so a mutation in this gene causes dwarfism or short stature.
- This paternal allele of this gene is what's normally expressed while the maternal one is silenced.
- Notice that we have 2 scenarios; the homozygous and heterozygous.
- In the homozygous one both alleles are normal and the maternal one is silences and thus we have a normal mouse.
- In the heterozygous scenario, we have a normal allele and a mutant one. Our main problem here is when the mutant allele is the paternal one because it produces a dwarf mouse, however if the maternal allele is the mutant one then it won't be problem since it's silenced either way.



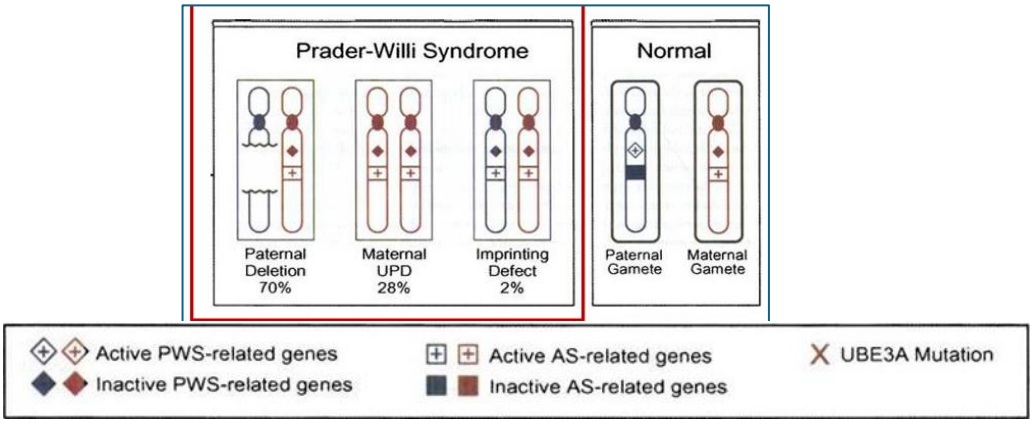
- **Genomic imprinting is essential for normal development:**
- Many common cancers, paternally-expressed imprinted genes enhance growth/proliferation; some are 'oncogenes'
- Hypomethylation (20-60% less) => chromosome instability + oncogene activation
    - IGF2 (Insulin-like Growth Factor 2)
- Function: IGF2 promotes fetal growth and development by stimulating cell proliferation and inhibiting apoptosis. It plays a vital role in prenatal growth by acting as a mitogen.
- Consequences of Dysregulation:
    - Beckwith-Wiedemann Syndrome (BWS): a disorder characterized by overgrowth, increased risk of embryonal tumors such as Wilms' tumor, and other developmental abnormalities.
    - Overexpression of IGF2 is associated with BWS, leading to overgrowth and an increased risk of childhood tumors.
    - Cancer: Aberrant IGF2 expression is linked to several cancers, including colorectal, liver, and breast cancers. Elevated IGF2 levels can promote tumor growth and survival

- Maternally-expressed Imprinted genes constrain growth/proliferation; some are 'tumor suppressors'.
    - Hypermethylation => tumor suppressor genes silenced (oncogene activation).
    - E.g. CDKN1C.
- Function: CDKN1C encodes a cyclin-dependent kinase inhibitor that controls cell cycle progression by inhibiting cyclin/CDK complexes.
- Consequences of Dysregulation:
    - Beckwith-Wiedemann Syndrome (BWS): Loss of function or reduced expression of CDKN1C is associated with BWS, a disorder characterized by overgrowth, increased risk of embryonal tumors such as Wilms' tumor, and other developmental abnormalities.
    - Cancer: As a tumor suppressor, reduced expression or mutations in CDKN1C can lead to uncontrolled cell proliferation and contribute to cancer development.
- **Genomic imprinting and neurodevelopmental disorders:**
- Three disorders that are the result of either direct or indirect deregulation of imprinted genes: Prader-Willi syndrome, Angelman syndrome and Rett syndrome.
- Genes associated with both Prader–Willi syndrome and Angelman syndrome map to the long (q) arm of chromosome 15 (15q11-13).
- These two syndromes are associated with the same chromosome, but they give rise to completely different features.
- Prader-Willi Syndrome - obesity, increased appetite, mental retardation, short stature. (abbreviated PWS)
- Angelman Syndrome - uncontrollable laughter, jerky movements, seizures and other motor and mental symptoms. (abbreviated AS) Syndrome.
- Defects in genomic imprinting lead to Prader-Willi syndrome (PWS) and Angelman syndrome (AS).
- Prader-Willi syndrome occurs when the paternal allele(s) that would normally be expressed are "missing."
    - In a normal child the PWS allele(s) are only expressed from the paternal chromosome 15, and the PWS allele(s) on the maternal chromosome 15 are inactive.

- Angelman syndrome occurs when the maternal alleles that would normally be expressed are “missing.”
    - In a normal child the AS allele(s) are only expressed from maternal chromosome 15, and the AS allele(s) on the paternal chromosome 15 are inactive.

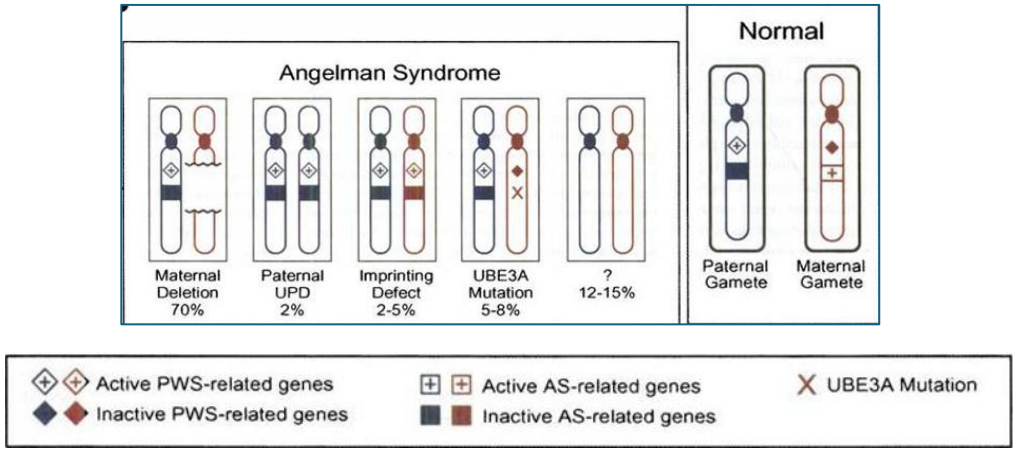


- So PWS occurs because we’re missing a paternal allele and AS occurs because a maternal one is missing.
- So basically in the case of syndrome development, the normally active allele is missed and the other one is now active. However, there are other scenarios that can cause these two syndromes.
- Genetic Causes of Prader-Willi Syndrome:
- You can notice in the figure below that we have the paternal chromosome in blue and maternal in red, and you can see that normally the alleles responsible for PWS are active on the paternal while inactive on the maternal, also you can see that the UBE3A gene (represented as x) is active on the maternal whereas it’s inactive on the paternal -This gene is responsible for neurological function and development.
- Paternal Deletion (70-75% of cases): Loss of a segment of the father's chromosome 15.
- Maternal UPD-Uniparental disomy- (20-25% of cases): Inheritance of two maternal chromosome 15s with no paternal contribution (Both are inactive).
- Imprinting Defects (1-3% of cases): Errors in the expression of paternal genes due to imprinting control mutations or epigenetic changes (Both are inactive).



➢ Genetic Causes of Angelman Syndrome:

- Maternal Deletion (about 70% of cases): Loss of a segment of the mother's chromosome 15, including the UBE3A gene.
- Paternal UPD (1-3% of cases): Inheritance of two paternal chromosome 15s with no maternal contribution, leading to absence of maternal UBE3A expression.
- Imprinting Defects (about 3% of cases): Errors in the expression of maternal genes due to imprinting control mutations or epigenetic changes.
- UBE3A Mutations (about 10% of cases): Direct mutations in the UBE3A gene inherited from the mother.
- A very small percentage is from unknown causes.



في الروحِ إصرارٌ وفي أعماقنا أملٌ

لا يعتري خطواتنا يأسٌ ولا سأمُ

هنيئًا لكم دفعة القدس إنهاء سنواتكم الثلاث الأولى في كلية الطب... وفّقكم الله، ونفع بكم، وجعلكم ممن تُنصر بهم الأمة.

Motivation is what gets you started but discipline is what keeps you going.

Check the link :P