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Organelle inheritance

Reading: chapter 16
Problem set posted

Non Mendelian Inheritance

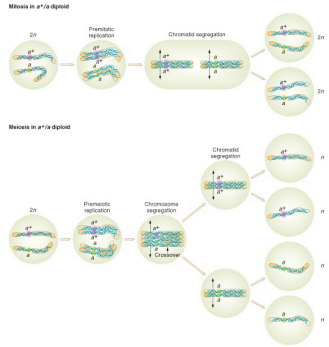
Inheritance patterns studied so far can be explained by Mendel.

In today's lecture, we will consider examples of inheritance that cannot be interpreted using Mendelian principles.

Outline

Non-Mendelian inheritance
Focus on mitochondria
- Mitochondria and their genome
- Digression into endosymbiont theory
- Inheritance in yeast
- Inheritance mammals
  - Digression into difference between maternal inheritance and maternal effect
  - Mitochondrial diseases
  - Using mitochondria to study history
    - Recent history
    - Early human history

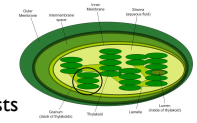
Centromeres ensure the proper segregation of genes during mitosis and meiosis.



Molecules that are not linked to chromosomes bearing nuclear centromeres behave differently during mitosis and do not obey Mendel's rules.

DNA and RNA plasmids
Protein structure (Prions)
Organelle chromosomes

Mitochondria



Chloroplasts

Organelle Genetics

Mitochondria and Chloroplasts
Involved in producing energy for the cell though either oxidative phosphoryation or photosynthesis
Each contain their own small genome

We will focus on mitochondria here, but chloroplast inheritance is also non-Mendelian.

Inheritance patterns vary depending on organism.
Focus on *Saccharomyces cerevisiae* and mammals.

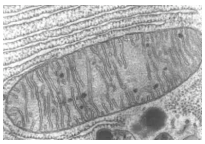
Mitochondria

Use oxygen to produce energy efficiently (aerobic metabolism).
--muscle cells are loaded with them.

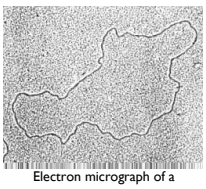
Contain own small genome
~17,000 bp circular DNA in humans

Encodes 2 rRNAs and 22 tRNAs for protein synthesis
13 proteins for energy metabolism

99.9% of the mitochondrial proteins encoded by nuclear genes.

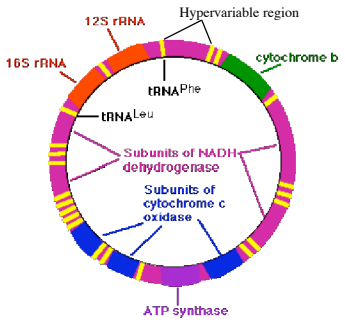


Electron micrograph of a mitochondrion



Electron micrograph of a mitochondrial DNA

Map of human mtDNA

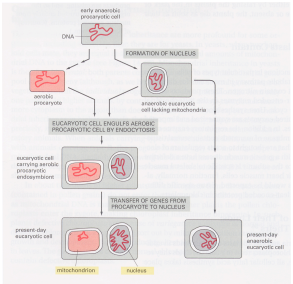


Nuclear vs Mitochondrial DNA (human)

Nuclear DNA	**Mitochondrial DNA**
Linear	Circular
3×10^6 kbp	17 kbp
>30,000 genes	37 genes
Cell contains two copies of each gene	Cell contains 1000s of copies

Digression:
How did mitochondria (and chloroplasts in plants) arise?

The endosymbiont theory was proposed by Lynn Margulis in the 1980s.
It proposed that an anaerobic cell engulfed aerobic bacteria, and the two cells developed a symbiotic relationship. With time most bacterial genes ended up in the nucleus, but a few genes remained in what became mitochondria.



Evidence for Endosymbiont Theory

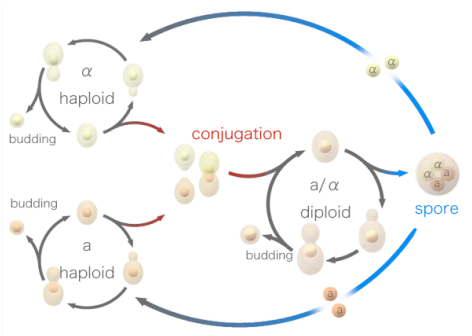
Similar lipid compositions in membranes of bacteria and mitochondria

Bacterial and mitochondrial genomes circular and lack associated histones

Protein synthesis in bacteria and mitochondria similar

rRNAs similar in bacteria and mitochondria

S. cerevisiae life cycle



Yeast mt genetics

•20-40 copies of mtDNA/cell

•During mating, mitochondria of haploids fuse.

•During mitosis and meiosis, mitochondria undergo fission and each cell inherits mitochondria.

•mitochondria essential, BUT mtDNA not necessary.

•In absence of electron transport and oxidative phosphorylation, yeast can ferment carbon sources such as glucose, but not carbon sources like glycerol, which can only be metabolized oxidatively.

Boris Ephrussi identified petite mutants

•Wild-type strains grow quickly and give rise to large or grande colonies.

•Petite mutants grow slowly and give rise to small or petite colonies.

•But any mutation that affects cell health can lead to slow growth. Ephrussi was able to distinguish mutants defective in mt function by growing on glycerol, which can't be fermented. Mutants defective in mt function grow slowly in glucose, but fail to grow in glycerol.

Different types of petites

Nuclear (segregational) petites

pet^- X pet^+

↓

diploid (grande)

↓

2 pet^-
2 pet^+ (grande)

Mitochondrial petites (e.g. ρ^o petites)

ρ^o petites: no mtDNA

ρ^o X grande

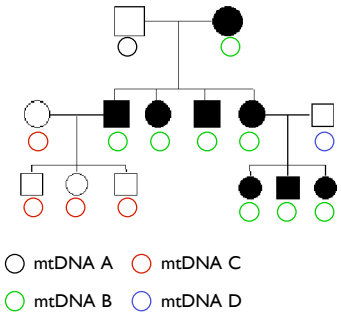
↓

diploid (grande)

↓

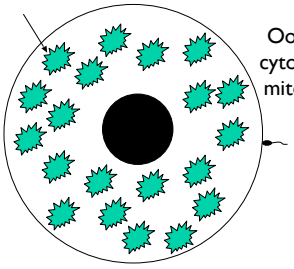
All grande
4:0 grande:petite

In animals, the rules for mitochondrial inheritance are different. mtDNA is inherited maternally.



Why?

mitochondria



Oocytes contribute cytoplasm, and hence mitochondria during fertilization

An important digression
Maternal inheritance is different from maternal effect!

m/+ ♀ X m/+ ♂ → 1/4 m/m (but wild type)

In the F1, wild-type RNA or protein is provided in the oocyte.

m/m ♀ X +/+ ♂ → m/+ (mutant)

In the F1 no wild-type product provided in oocyte.

Unlike maternal inheritance, maternal effect results from the inheritance of nuclear gene products. The genotype of the mother and not the zygote is what determines phenotype.

Human genetic diseases can result from mtDNA mutations.

•mtDNA mutations that eliminate mt function not tolerated unless wild-type DNA present (heteroplasmy).

•Human cell 100 mitochondria, 1000 mtDNAs.

Mitochondrial mutations and associated diseases

Disease	mutation
CPEO	3243 tRNALeu(UUR)
Cardiomyopathy with or without encephalopathy	4269 tRNAIle
	4317I tRNAIe
	3260 tRNALeu(UUR)
	3243 tRNALeu(UUR)
KSS	Deletion/duplication, 3243 tRNALeu(UUR)
	8344 tRNALys
MELAS	3243 tRNALeu(UUR)
	11084 ND4
	3271 tRNALeu(UUR)
MERRF	8344 tRNALys
	8356 tRNALys
NARP	8993 ATPase 6
Pearson's syndrome	
LHON	ND4 (11778)
	ND1 (3460)
	ND6 (14484)
Leigh's syndrome	8344 tRNALys
	8993 ATPase 6
Diabetes mellitus and deafness	3243 tRNALeu(UUR)

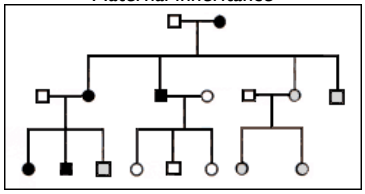
Problems with determining maternal inheritance in mtDNA diseases.

•Individuals within the pedigree will differ in phenotype (variable penetrance and expressivity) because of variability in mutant:wild type mtDNA ratio (heteroplasmy).

•Different organs within an individual can have different degrees of pathology because of heteroplasmy.

Hallmarks of mtDNA genetic diseases

Maternal inheritance



Affects tissues require lots of ATP

Defective mt function in affected tissue

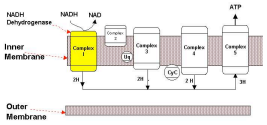
Mitochondria contain mutant DNA

LHON

Leber Hereditary Optic Neuropathy

Degeneration of retinal ganglion cells

Missense mutations in NADH dehydrogenase genes



Can have individuals that are homoplasmic for mutation--some are affected but some are not.

Defect thought to be caused by production of free radicals

CPEO

Chronic Progressive External Ophthalmoplegia

Slow paralysis of the extraocular muscles

$tRNA^{Leu}$ mutation

Both cardiomyopathy, diabetes and MELAS (mitochondrial myopathy, encephalopathy, lactic acidosis, and stroke) patients have the same tRNA lesion!!

Cardiopathy and MELAS can be associated with other mt mutations.

The use of mtDNA to study history

Between 1976 and 1983, the military of Argentina kidnapped, jailed, and killed more that 10,000 dissidents. Along with their parents, many infants and toddlers disappeared.

In 1977, the grandmothers of these children began to hold vigils in the main square of Buenos Aires to inform others that about the disappearance of their grandchildren. This group became known as the "Grandmothers of the Plaza de Mayo." They contacted many organizations for help, including the American Association for the Advancement of Science (AAAS).

First, different alleles of the HLA locus were used.

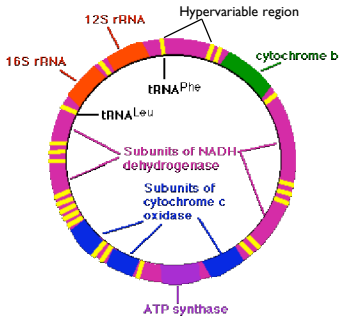
Compared the human lymphocyte antigens (HLAs) on white blood cells.

Children should share one allele with one of the paternal grandparents and the other with one of the maternal grandparents.

Mary Claire King, then a geneticist at Cal, taught Argentine medical workers to analyze HLA markers on white blood cells, and the workers typed HLAs from family members of the missing children.

The probability that a tested child belonged to the family claiming him or her on the basis of eye-witness accounts of abduction varied from 75-99%.

Hypervariable region can often be used to distinguish individuals



Then, mtDNA analysis was used.

By the mid-1980s, the simpler cases had been solved.

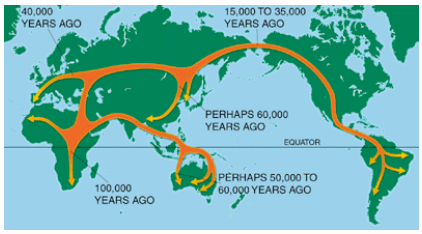
Professor King turned to her former mentor and colleague Allan Wilson, a molecular biologist at Cal who was studying mtDNA.

PCR amplification and sequencing of the highly variable region of mtDNA made it possible to match a child with his or her maternal grandmother.

The probability that two unrelated individuals would be identical for this 131 bp region is almost nil.

In addition, the high copy number of the mtDNA aided in the identification of the remains of individuals killed by the military regime.

Human migrations can be studied using Y chromosome and mtDNAs



Analysis of human mtDNA led to the Mitochondrial Eve Hypothesis

In the 1980s, Allan Wilson pioneered the use of mtDNA to study human evolution.

In two papers published in 1987 and 1991, he and his colleagues at Cal proposed that we all come from a population of humans that lived in Africa approximately 200,000 years ago.

Evidence for Mitochondrial Eve hypothesis

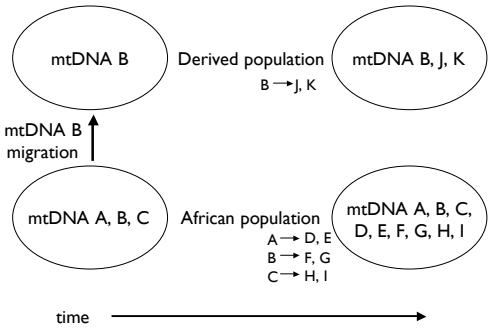
Compared sequences from the highly variable region of mtDNA.

Greater sequence differences among native Africans than any other group, which included Europeans, American Indians, Papua New Guineans, Asians, aboriginal Australians and individuals of Middle Eastern origin.

The diversity of Africans is equivalent to the diversity of all groups combined.

These observations led Wilson to propose that the African population has had the longest time to evolve variation and thus humans originated in Africa.

Model for understanding mtDNA diversity argument



But when did Mitochondrial Eve live?
(arguments from Wilson paper)

2.8% of the mitochondrial base pairs differed in the population of sub-Saharan Africans studied.

Chimpanzees and humans diverged about 5 million years ago, and human and chimpanzee mtDNAs differed at 15% of the base pairs of the mtDNA.

Adjusting the data to account for multiple substitutions at the same base pair, they calculated that mtDNA has been diverging at a rate of 13.8% per million years.

Assuming that this "molecular clock" is ticking at a constant rate, they determined that a "Mitochondrial Eve" from whom we are all derived lived 2.8/13.8 or 0.20 million years ago. Although there has been much argument about the assumptions and statistical methods used, most evolutionary geneticists accept that the women carrying our ancestral mtDNA lived in sub-Saharan Africa approximately 200,000 years ago.

Many mtDNAs /cell

2 for nuclear DNAs; 1000s for mtDNAs

After death DNA slowly degrades

Can often amplify mtDNA but not nuclear DNA from preserved tissues and skeletons.

In 1883, the last existing Quagga died in an Amsterdam zoo.

More than 100 years later, analysis of mtDNA from museum Quagga tissue revealed that Quaggas were closely related to the extant plains zebra.

Did Neanderthals interbreed with modern humans?

Two theories for interactions between modern humans and archaic Homo species.

The replacement theory: As early *Homo sapiens* migrated out of Africa, they entered regions occupied by archaic Homo species and competed with them for resources. *H. sapiens* won the competition, leading to the extinction of archaic species.

The regional continuity theory: *H. sapiens* interbred with the other species to generate modern humans.

Neanderthals were one of these archaic species, coexisting with modern humans for over 70,000 years until their extinction 28,000 years ago.

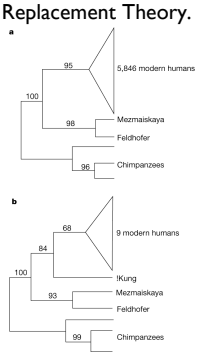
In 1997 and then again in 2000 scientists were able to amplify by PCR nucleotides from the highly variable region of mtDNA sequences from two Neanderthals, one from western Germany and one from the Caucasus.

mtDNA analysis favors the Replacement Theory.

3x as many differences between human and Neanderthal sequences than between pairs of humans.

The types and positions of the differences were distinct in the two groups.

The line that gave rise to Neanderthals and modern humans diverged between 365,000 and 853,000 years ago. The two Neanderthal sequences were much more closely related, and the scientists estimated that the ancestor to the two Neanderthals existed 150,000 to 350,000 years ago. The investigators argued that these differences support the replacement theory.



Two trees showing relationships of mtDNAs from humans, Neanderthals (Mezmaiska & Feldhoffer), and chimps. a and b are different statistical approaches using different human data sets.