ZO1104 Apiary management
Introduction to genetics

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DIOPLÓMA I BEACHADÓIREACHT
DIPLOMA IN APICULTURE

Aims and Learning outcomes

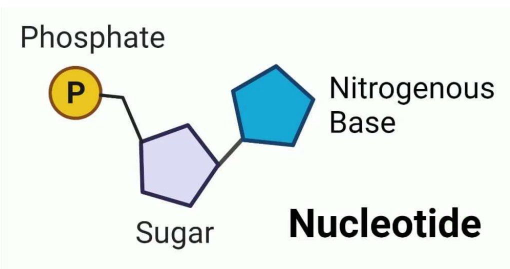
Aim : To introduce a basic understanding of genetics

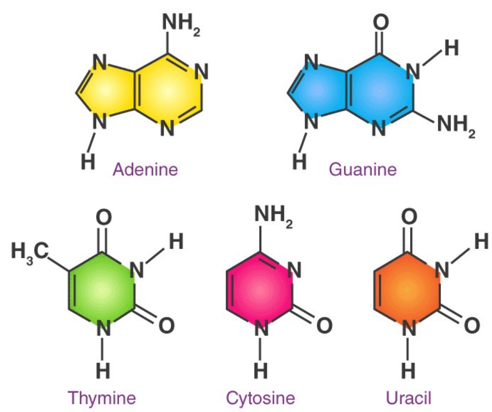
1. To gain an understanding of the terminology involved in genetics
2. Enhance understanding of the central dogma of molecular biology
3. Enhance understanding of the haplo-diploid nature of the honey bee
4. Gain an understanding of the need for genetic diversity

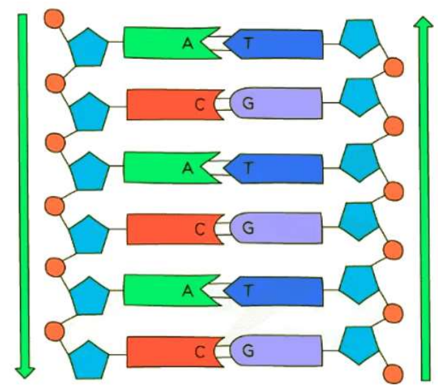
DIA

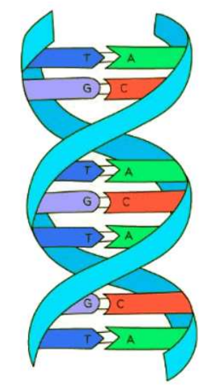
What is DeoxyriboNucleic Acid?

DNA is a long chain of similar molecules linked together (a polymer)





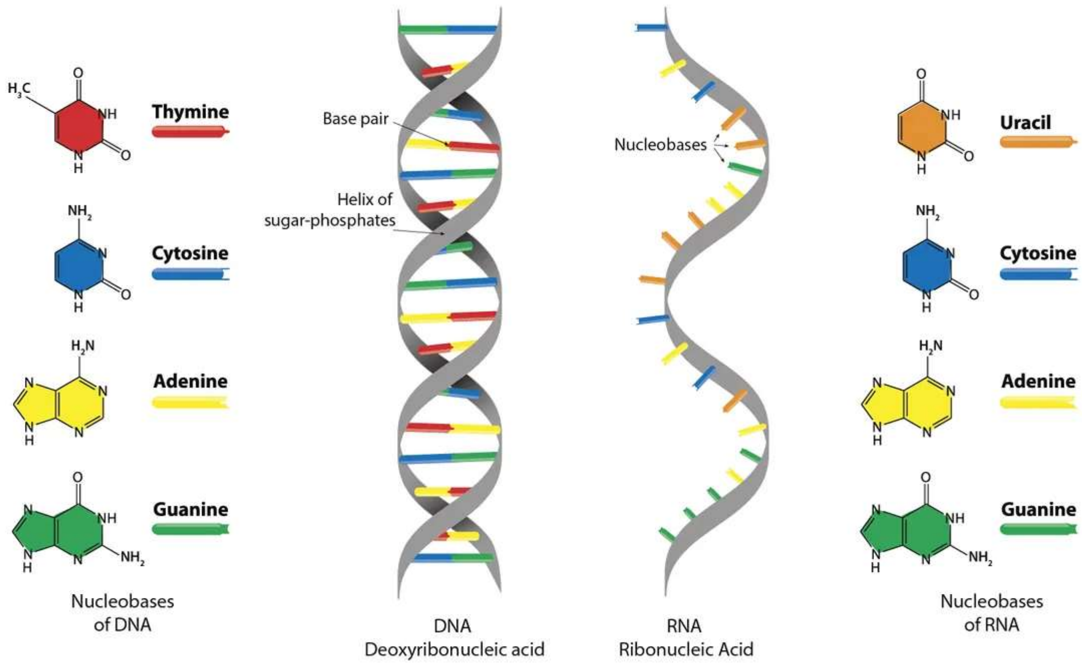




These similar molecules are called nucleotides (a.k.a. ‘Bases’)

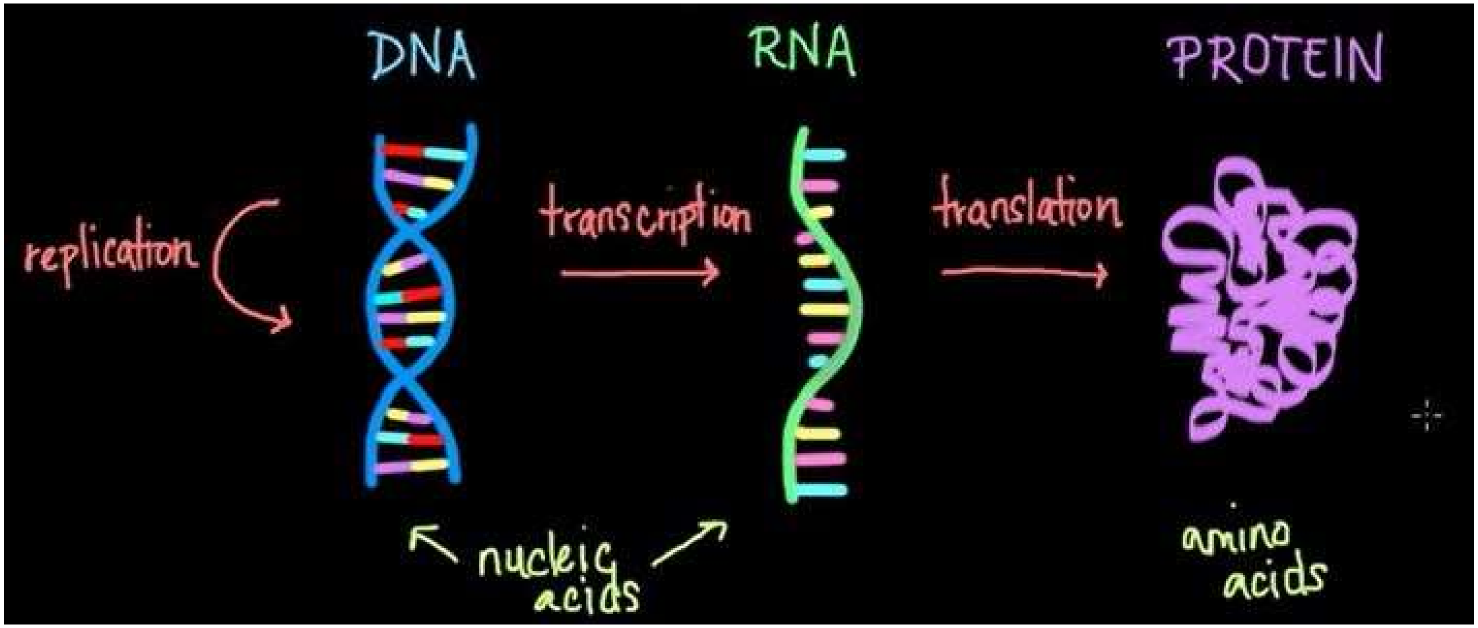
Rosalind
Franklin

DNA and RNA compared



The central dogma of molecular biology

"DNA, becomes RNA, becomes Protein"



Somatic vs germline cells

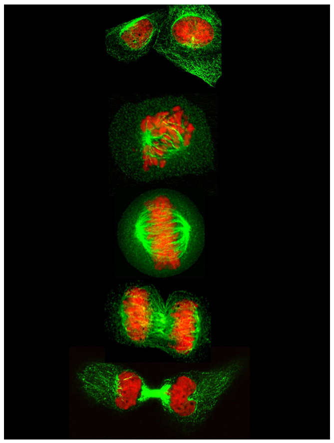
For our purposes:

Somatic cells are all the cells that go to make up the body of an organism

Germline cells are those which will eventually go on to form gametes (eggs and sperm)

These contain the copies of DNA which are inherited by the offspring

Part 1- DNA replication

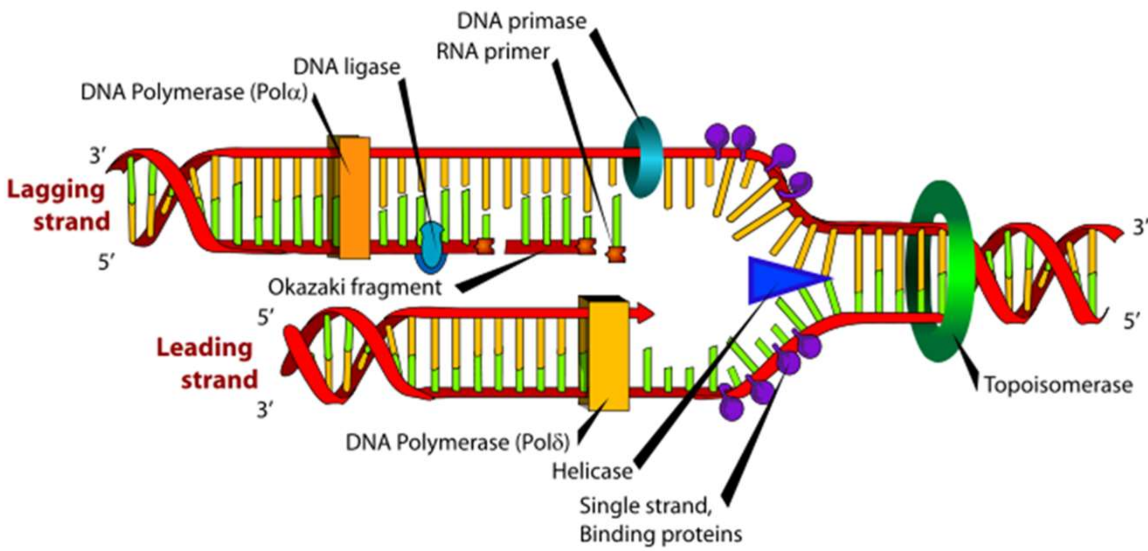


Each new somatic cell must have a copy of the DNA

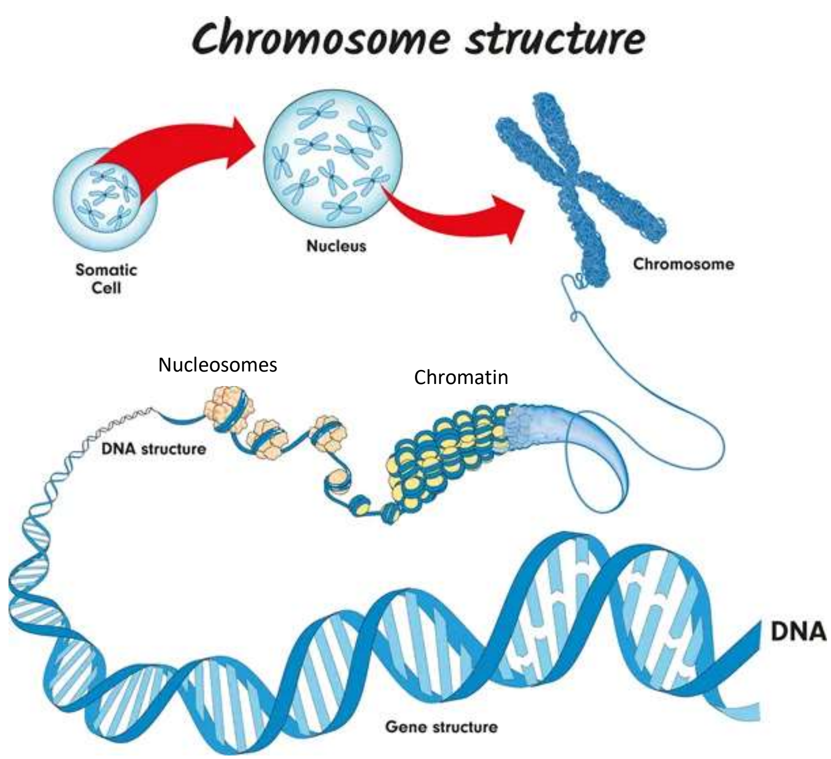
DNA replication is constantly going on in organisms for new cell production

Very, very low rate of inaccuracy (One base in every billion to 100 billion replications)

But…….there are an awful lot of replications



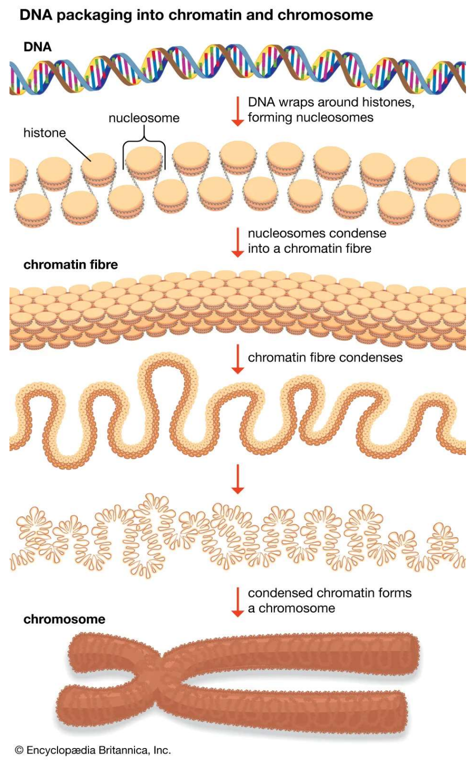
How chromosomes are formed from DNA



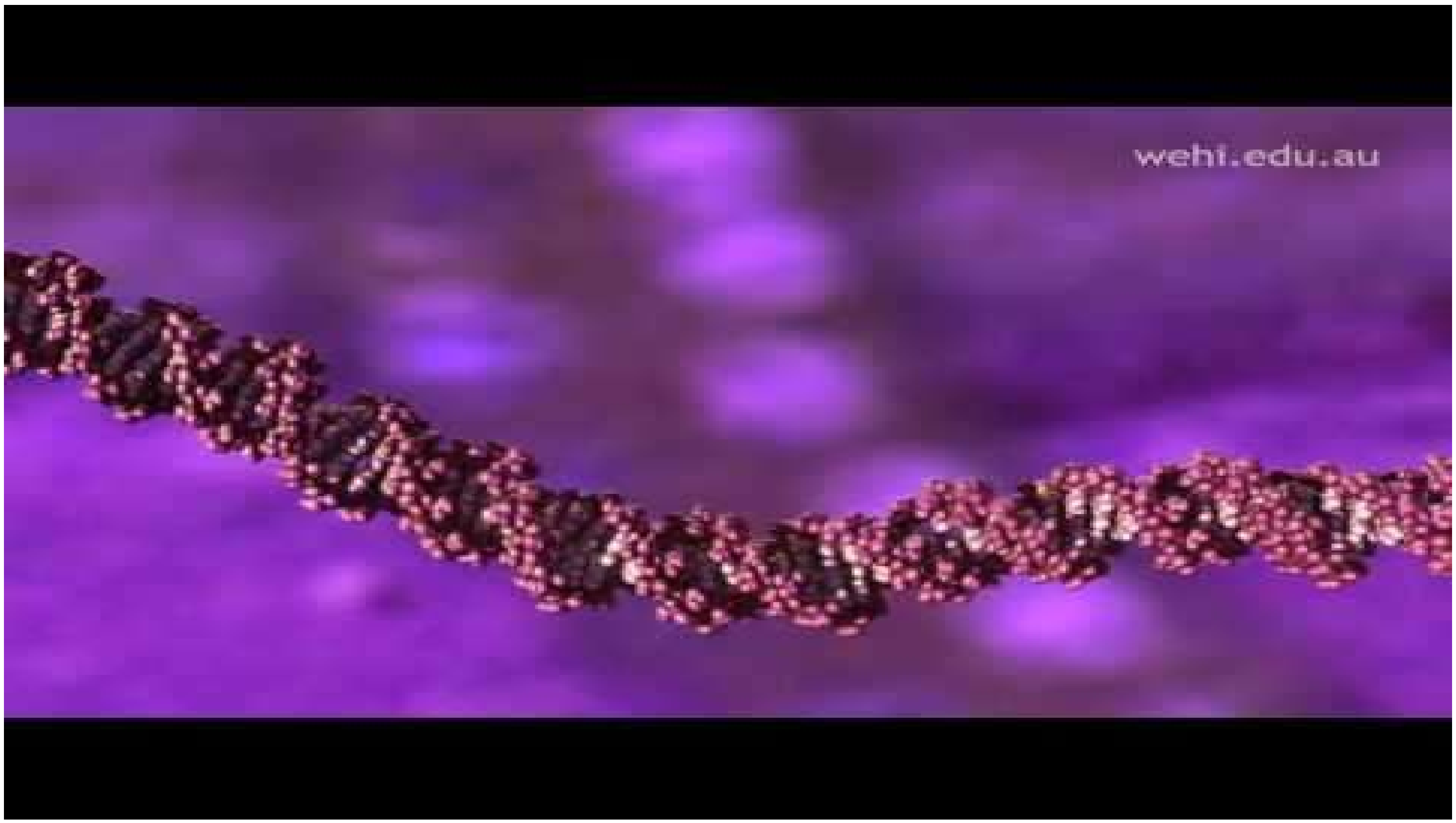
DNA strand wrap around proteins called histones.

This structure then wraps around itself (condenses) to form chromatin.

The chromatin then condenses further to form a chromosome

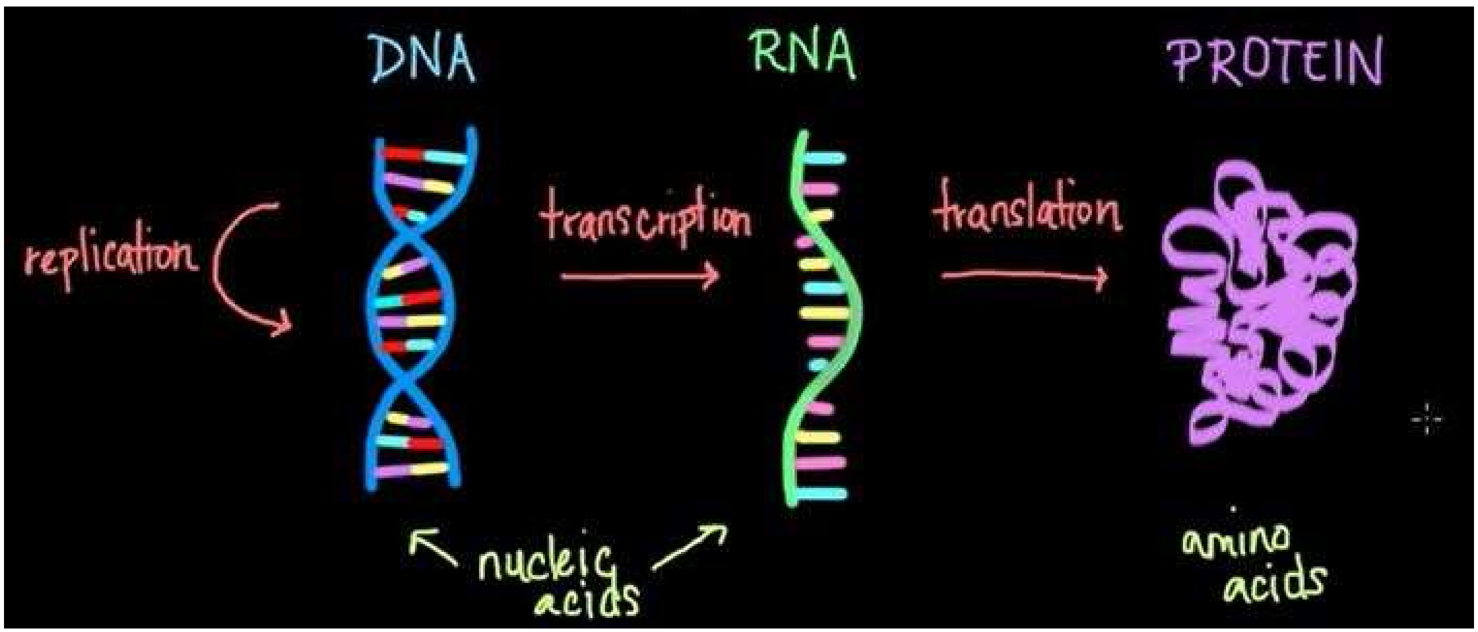


Chromosome construction



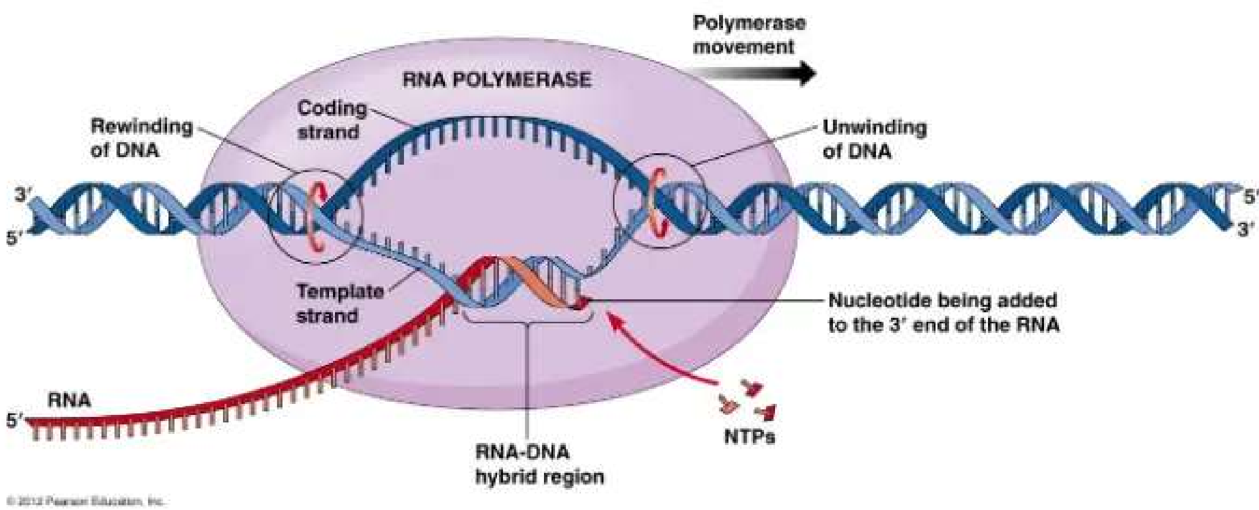
https://www.youtube.com/watch?v=OjPcT1uUZiE

Parts 2 and 3 – Transcription and Translation

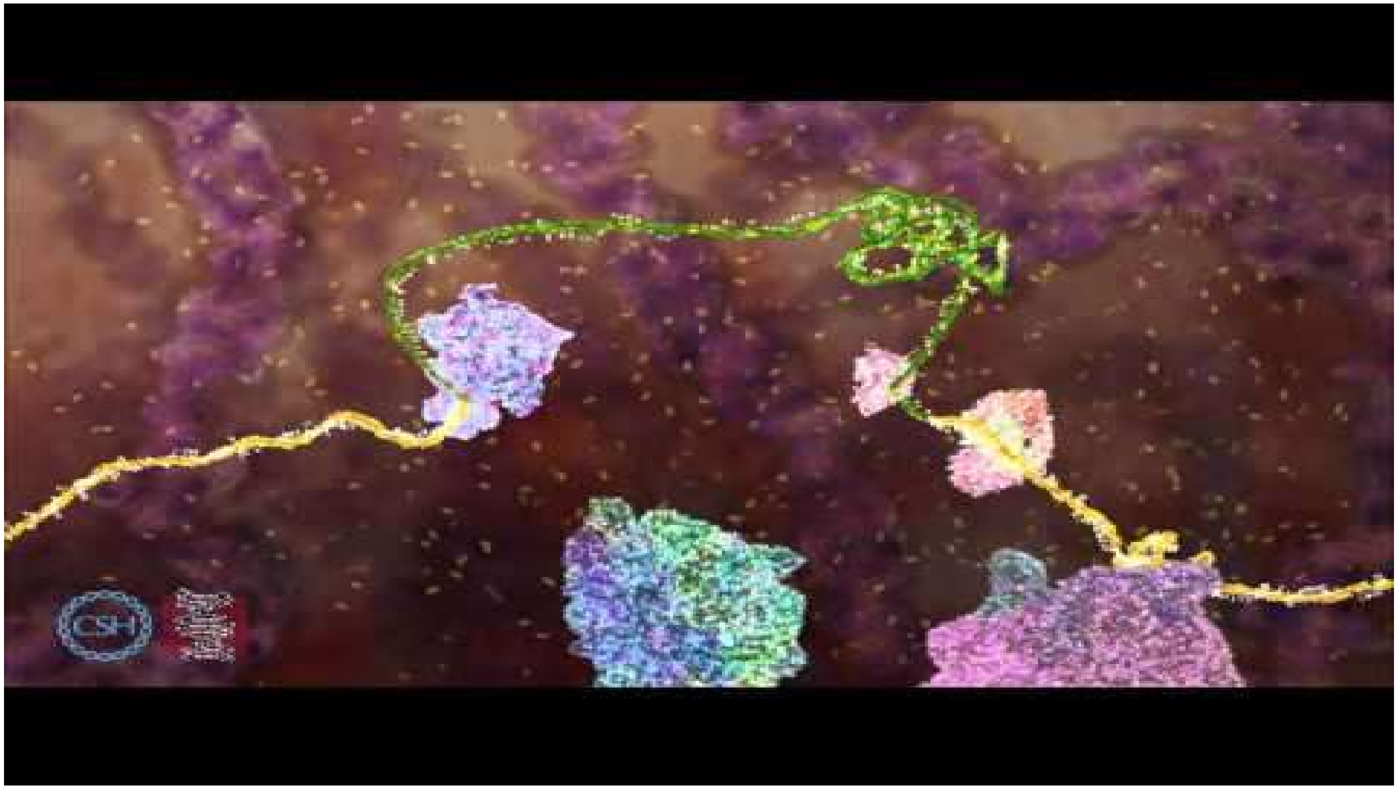


Production of RNA

The DNA strand is unwound and a copy of the coding strand is constructed



DNA transcription and translation



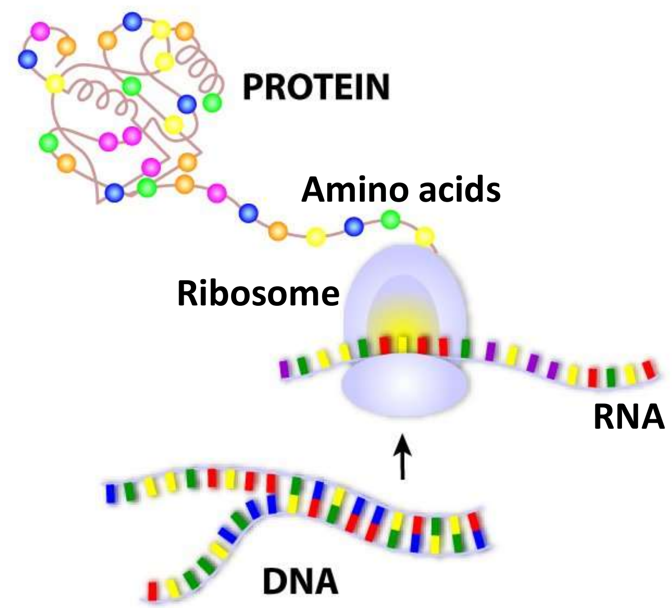
https://www.youtube.com/watch?v=9kOGOY7vthk

Amino acids are the building blocks of proteins

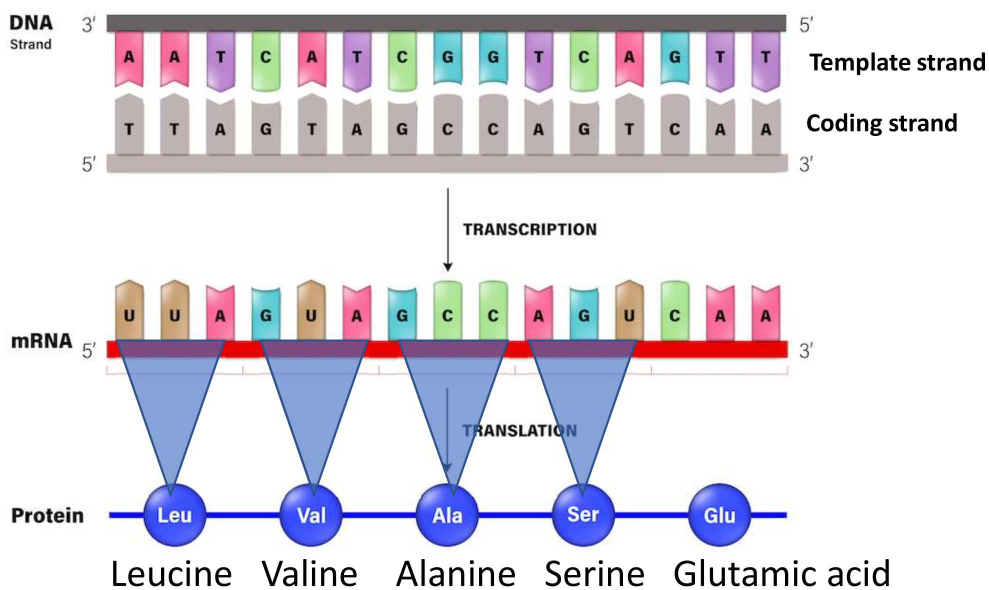
Proteins create part of the phenotype – How an organism looks and acts

Types of protein

- Structural – eg: collagen
- Enzymes – required for cell chemical reactions
- Messenger – eg: hormones to transit signals
- Immunity – eg: antibodies
- Transport and storage - eg: haemoglobin



Codons provide the codes for amino acids



second base in codon

first base in codon	U	C	A	G	third base in codon
U	UUU Phe	UCU Ser	UAU Tyr	UGU Cys	U
	UUC Phe	UCC Ser	UAC Tyr	UGC Cys	C
	UUA Leu	UCA Ser	UAA stop	UGA stop	A
	UUG Leu	UCG Ser	UAG stop	UGG Trp	G
C	CUU Leu	CCU Pro	CAU His	CGU Arg	U
	CUC Leu	CCC Pro	CAC His	CGC Arg	C
	CUA Leu	CCA Pro	CAA Gln	CGA Arg	A
	CUG Leu	CCG Pro	CAG Gln	CGG Arg	G
A	AUU Ile	ACU Thr	AAU Asn	AGU Ser	U
	AUC Ile	ACC Thr	AAC Asn	AGC Ser	C
	AUA Ile	ACA Thr	AAA Lys	AGA Arg	A
	AUG Met	ACG Thr	AAG Lys	AGG Arg	G
G	GUU Val	GCU Ala	GAU Asp	GGU Gly	U
	GUC Val	GCC Ala	GAC Asp	GGC Gly	C
	GUA Val	GCA Ala	GAA Glu	GGA Gly	A
	GUG Val	GCG Ala	GAG Glu	GGG Gly	G

The 21 amino acids

Genes and their alleles

A gene is any series of bases (nucleotides) that provide the code for a protein.
An allele is a version of a gene. Some make a difference to the gene's output, some don't

'Stop' codons

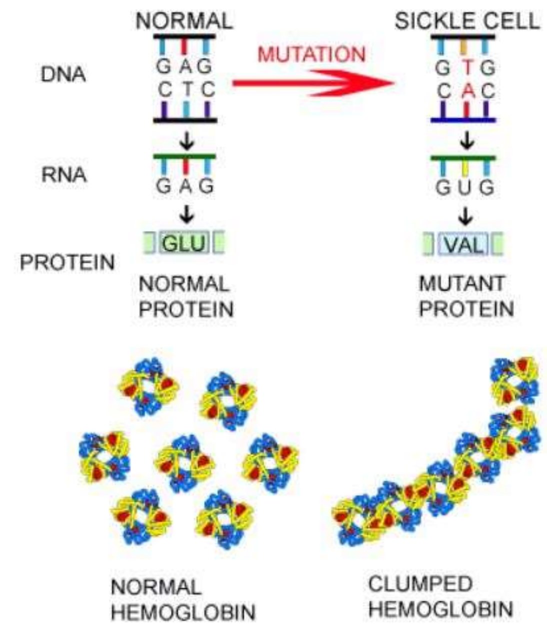
Part of the gene sequence for haemoglobin protein

………ACTGAATCCGGATGATGAGACTTTATAAGAATAA……… Normal gene

………ACTGAATCCGGATGATGTGACTTTATAAGAATAA……… Sickle cell version (allele)

Gene sequence starts back here….

…and the genome keeps going this way



What's in a genome?

The genome of an individual organism (eg: you, your sibling, a worker bee, a queen) is all the nucleotide pairs (base pairs) that make up genes and includes all the DNA that doesn't code for proteins such as regulatory genes (genes that control other genes)

The genome of a species (eg: *Homo sapiens, Apis mellifera*) is a representative genome. It's an artificial construct, created from the genomes of one or multiple representative individuals from the species.

Eg: The *Apis mellifera* genome was created from the DNA of multiple drones of a single partially inbred queen from a US apiary. The human genome is a mix of 12 US individuals

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The two genomes

Animals have two genomes:

One nuclear genome

One mitochondrial genome

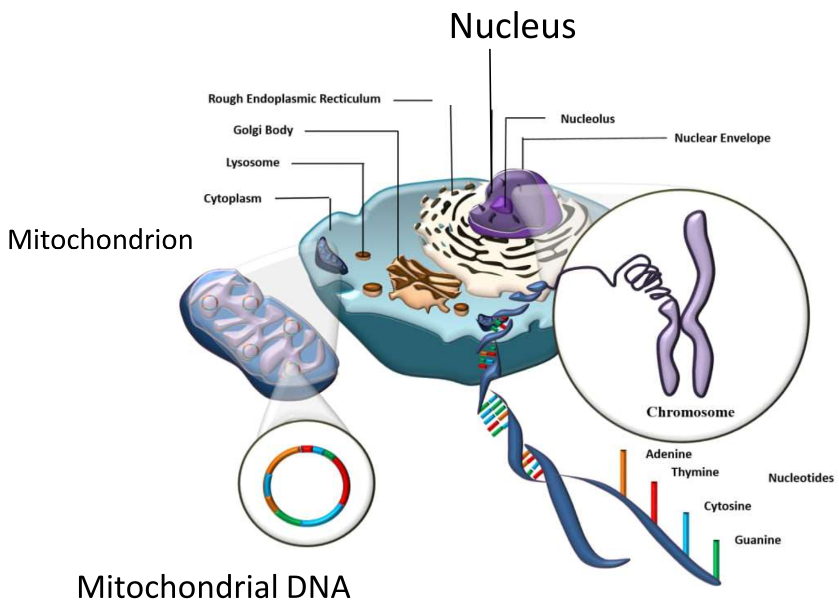

Image adapted from Bioethics Advisory Committee

A cell contains a nucleus where the DNA in the chromosomes (nuclear genome) are located

AND

lots of mitochondria which have their own DNA (mitochondrial genome)

What's in a genotype?

1. 'Genotype' and 'Genome' are often used interchangeably. This can lead to confusion
2. A genotype is an artificial construct and consists of a specific part of the nuclear and/or mitochondrial genome.
3. Each genotype is dependent on what you are looking at and is usually only part of the genome:
    - Nuclear genome = Genomic genotype (often, confusingly, just called the genotype)
    - Mitochondrial genome = Mitotype (or mitochondrial genotype)
    - A haploid genome = Haplotype
    - SNPs , a part of the nuclear genome = SNP genotype
    - Microsatellites, a part of the nuclear genome = Microsatellite genotype

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Honey bee Mitochondrial DNA genotype variants

Introduction to Ploidy

Ploid = chromosome

Ploidy = how many sets of chromosomes exist in an organism

Organisms can be diploid (2) or haploid (1)

Diploid (2n) = two sets of chromosomes.

Usually one from male and one from female

Haploid (n) = Only one set of chromosomes.

Usually from the female

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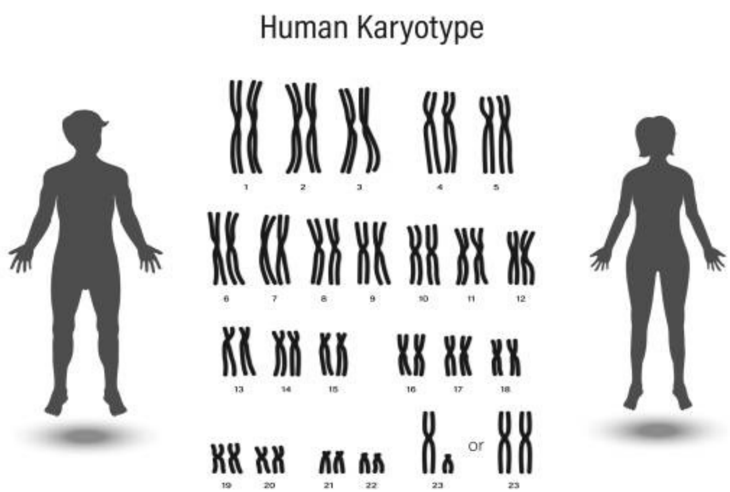
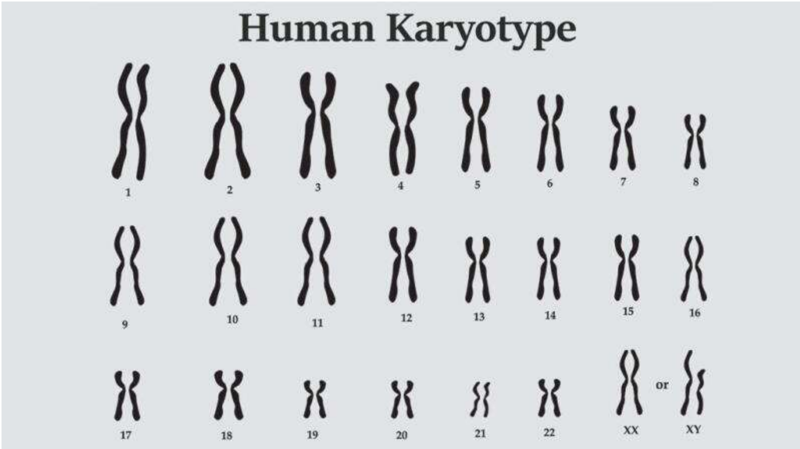
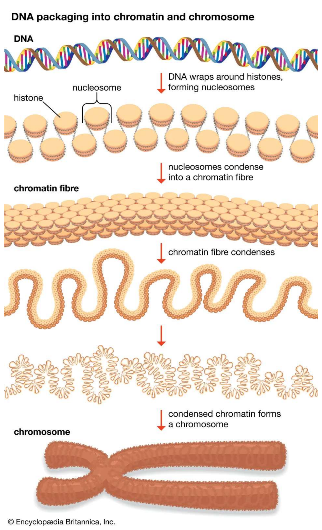
Let’s be clear about what is a chromosome

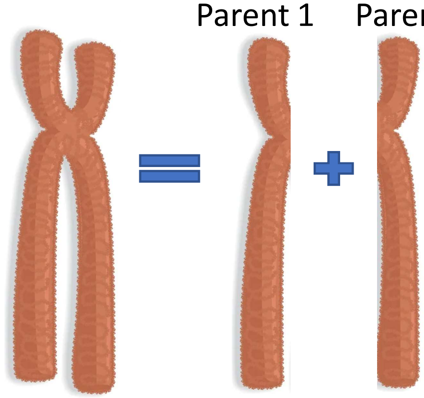




X-shaped chomosomes, as usually depicted, are in fact a chromosome pair, in the process of replication (very misleading)

Let’s be clear about what is a chromosome





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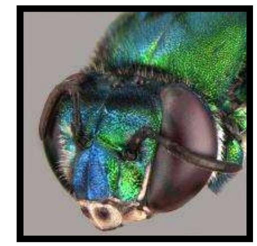
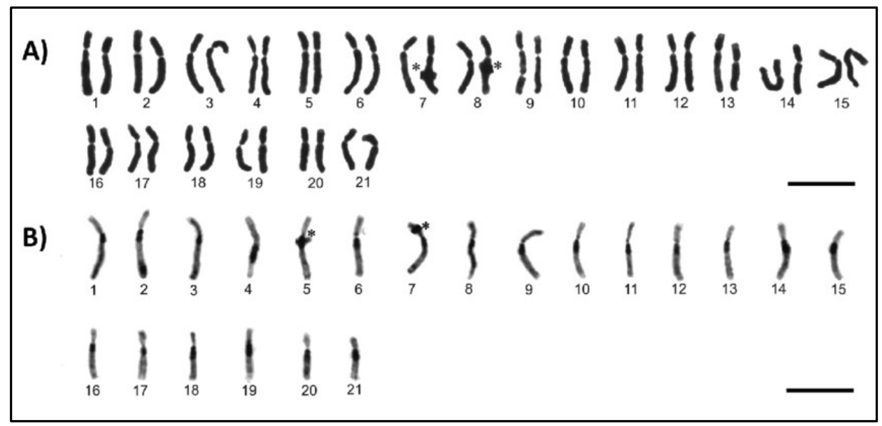
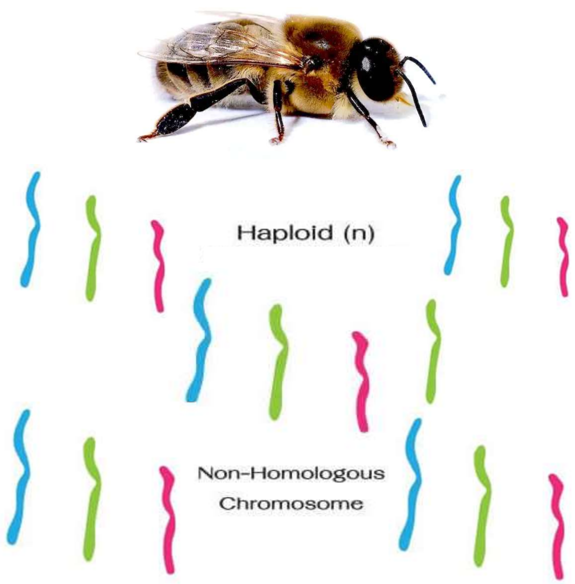


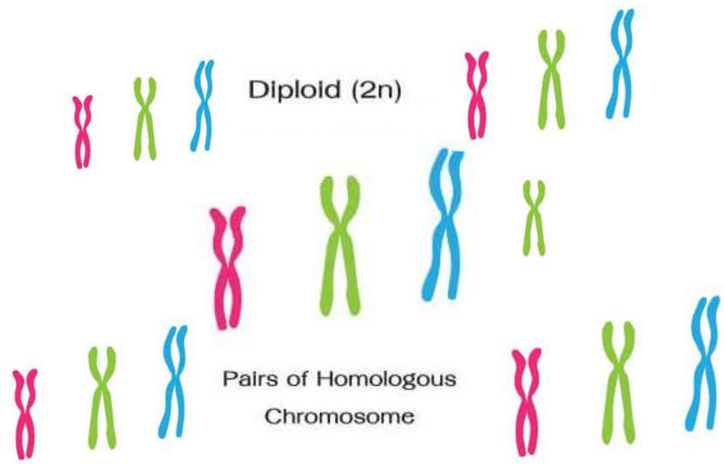


Karyotype of *Euglossa townsendi* (Orchid bee)
A)Female   B) Male

Adapted from Fernandes et al 2013

Honey bee ploidy





Queen and worker
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Drones have 16
chromosomes.
One of each type

Workers and queens have a total of 32 chromosomes. Two of each of the 16 chromosome types (i.e. a homologous pair, one from each parent)

Effect of diploid females and haploid drones

1. Unique mating behaviour gives rise to patrilines
2. Haplodiploidy
   - Females are diploid, 16 pairs of chromosomes (32)
   - Males are haploid, 16 single chromosomes
   - Purges lethal recessive traits from the population.
3. Behaviour & phenotype of drones inherited from the queen (and her father, the drone’s grandfather)

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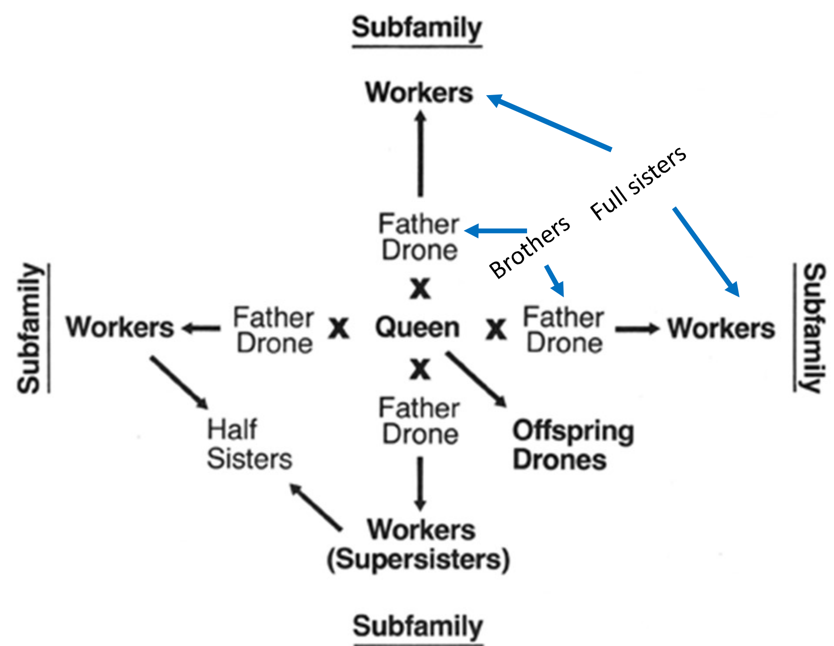
The degrees of worker sisterhood

Workers fathered by the same drone and same queen are super-sisters.

Workers from the same queen who were fathered by two unrelated drones, are half sisters.

Workers from the same queen and brother drones are full sisters



Adapted from Harbo and Rinderer BEEKEEPING IN THE UNITED STATES AGRICULTURE HANDBOOK NUMBER 335 Revised October 1980 Pages 49 – 57

https://www.beesource.com/threads/breeding-and-genetics-of-honey-bees.365903/

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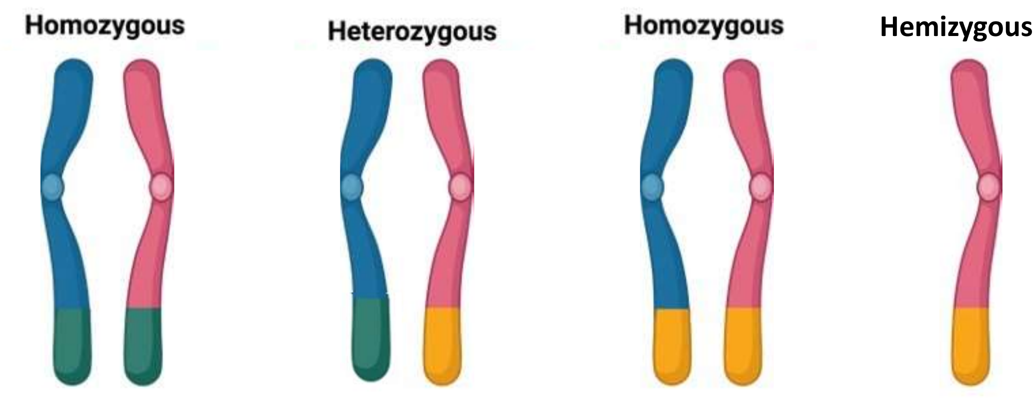
Heterozygous, homozygous and hemizygous

Homo = same
Hetero = different
Hemi = half
Zygous = of the zygote

Refers to the alleles of genes rather than the whole chromosome



Diploid: With same allele on each chromosome

Diploid: With different allele on each chromosome

Diploid: With same allele on each chromosome

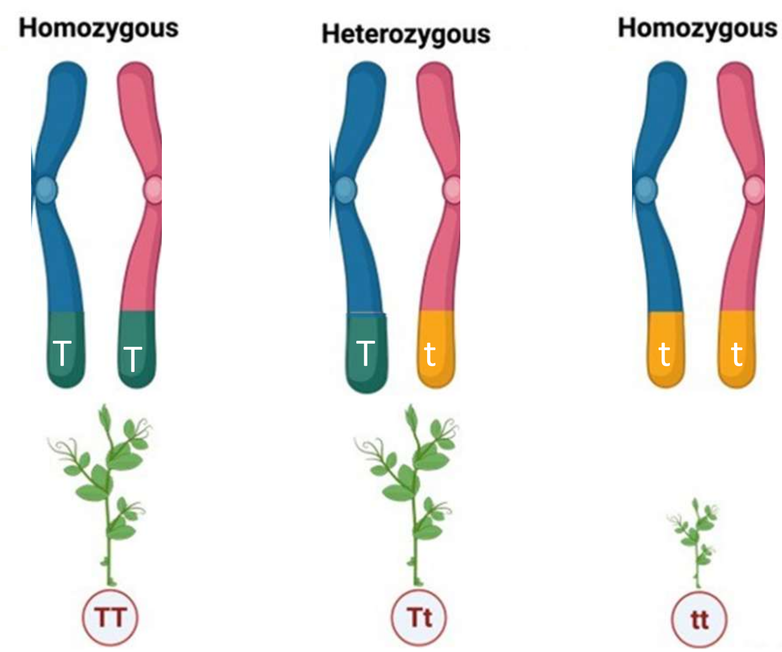
Haploid: Only one allele, because only one chromosome

Dominant vs recessive genes (or alleles)

For any given gene, dominant alleles will be preferentially expressed over the recessive allele

In the example to the right:
T = tall (the dominant allele)
t = short (the recessive allele)

In **homozygous TT** the plant will be tall
In **heterozygous Tt**, the dominant allele T will be expressed so the plant will also be tall.
In **homozygous tt**, there is no dominant allele present so the plant will be short



DIA

Dominant and recessive:
How alleles and zygosity interact

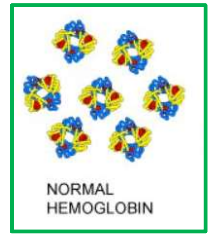
‘Stop’
codons

Part of the gene sequence for haemoglobin protein

Ch1a…ACTGAATCCGGATGAT**GAG**ACTTTATAAGAA**TAA**……

Ch1b…ACTGAATCCGGATGAT**GTG**ACTTTATAAGAA**TAA**……

Dominant allele
+
Recessive sickle cell allele

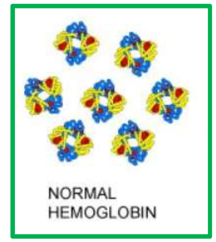


Heterozygous dominant

Ch1a…ACTGAATCCGGATGAT**GAG**ACTTTATAAGAA**TAA**……

Ch1b…ACTGAATCCGGATGAT**GAG**ACTTTATAAGAA**TAA**……

Dominant allele
+
Dominant allele

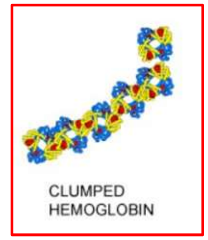


Homozygous dominant

Ch1a…ACTGAATCCGGATGAT**GTG**ACTTTATAAGAA**TAA**………

Ch1b…ACTGAATCCGGATGAT**GTG**ACTTTATAAGAA**TAA**………

Recessive sickle cell allele
+
Recessive sickle cell allele

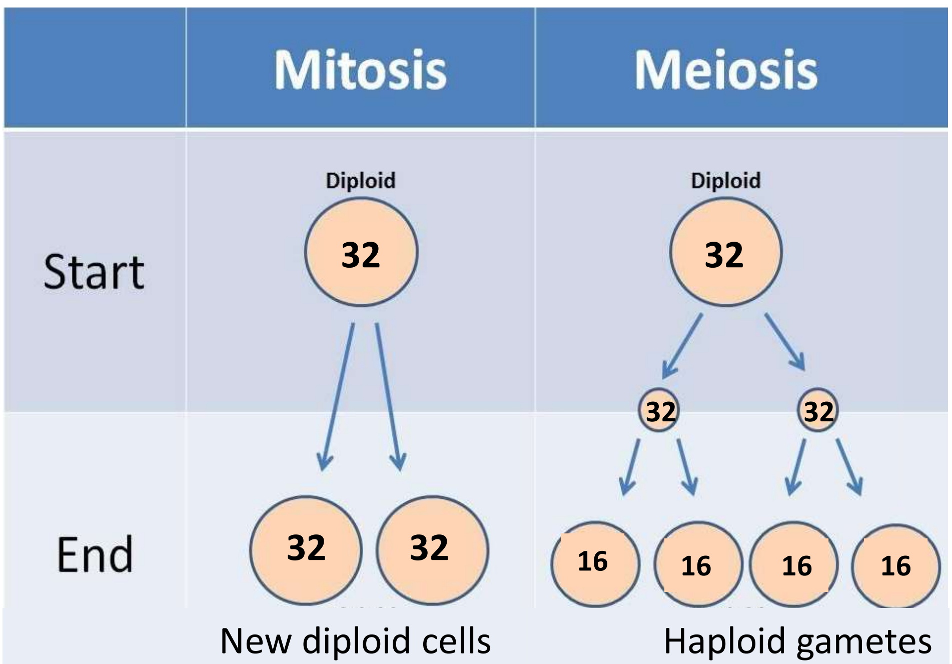


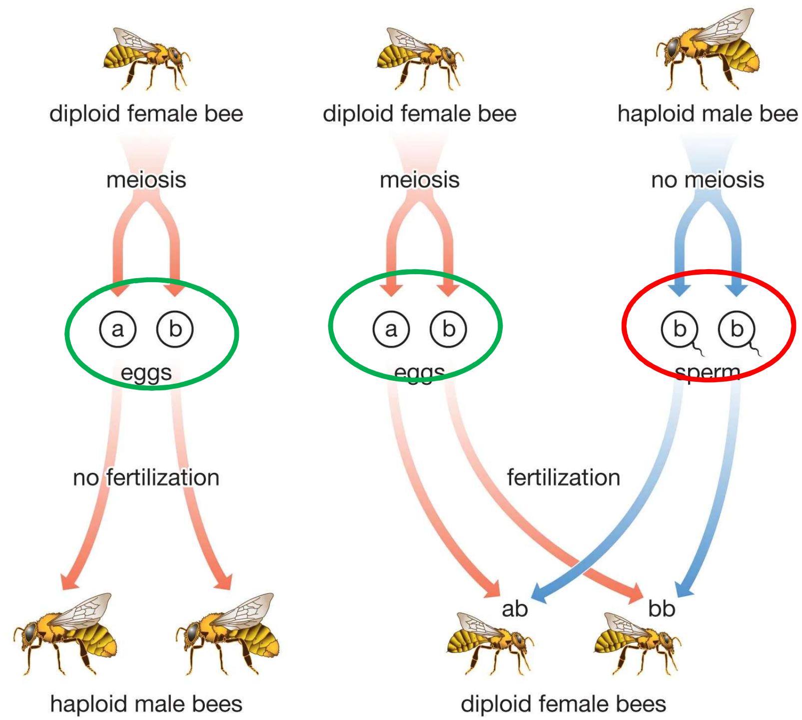
Homozygous recessive

Haplo-diploidy in honey bees

Somatic
Cell duplication

Germline
Gamete production



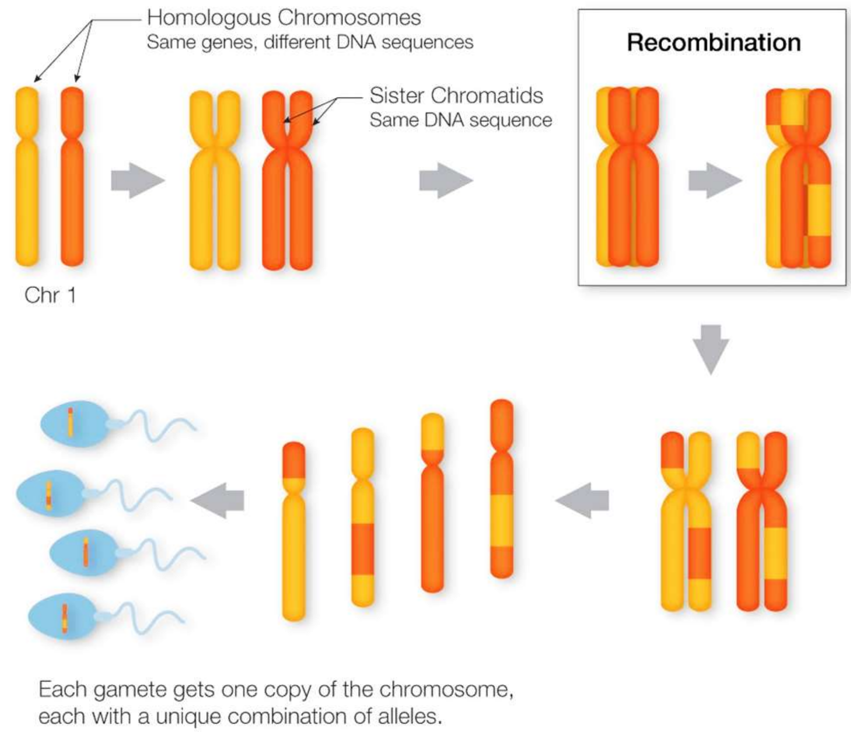


Recombination

Diploid meiotic recombination is inextricably linked to sexual reproduction

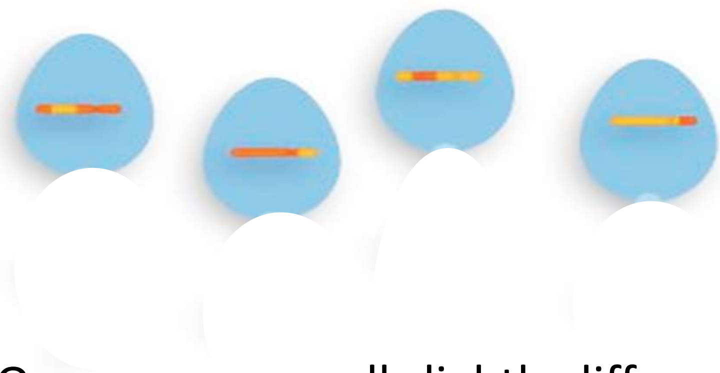
Homologous chromosomes – One from each parent

Several social insects in the order Hymenoptera have genome-wide recombination rates that greatly exceed those of other insects, plants, and mammals



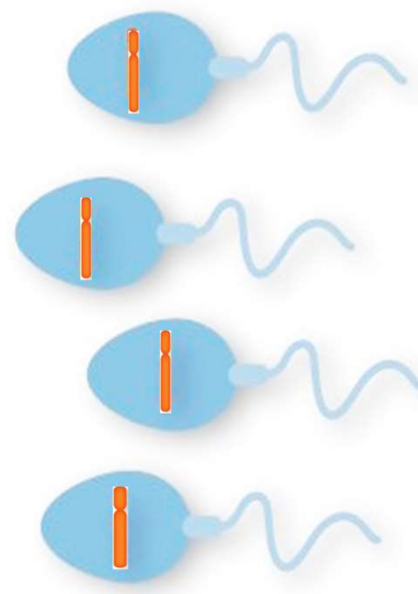
Recombination in haplo-diploid species

Recombination in honey bee egg formation
5x most other animals



Queen eggs are all slightly different genotypes

Recombination does not occur in drone tissue during the formation of sperm (spermatogenesis).



A drone’s sperm are all the same genotype

Sex determination & the role of sex alleles

- Honey bees do not have a gender specific chromosome.
- The genome, instead, contains an area called the **sex determining locus**.
- This is a region of DNA which harbours the '**c**omplementary **s**ex **d**eterminer' gene also known as the ***csd*** gene or sex allele.

- **The way that these alleles combine, creates a primary signal of sex determination**.

  Heterozygosity generates two complementary proteins which combine to form **an active protein** that initiates female development.

  Hemizygosity or homozygosity results in a **non-active CSD protein** and defaults to male development.

- If the individual is homozygous at the *csd*, a diploid male develops, but these are removed by workers at the first larval instar and are therefore non-viable (Woke 1963).

- **In diverse populations, different combinations of sex alleles are more numerous, so <u>extensive evidence of diploid drones is an indicator of reduced genetic diversity</u>.**

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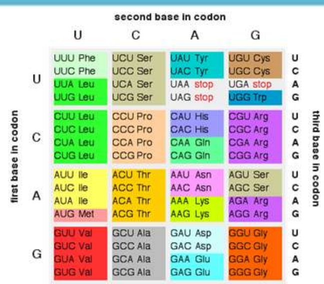
Complimentary Sex Determination (*csd*) alleles



Comparison of the *CSD* amino acid sequences from *ligustica* and *carnica* samples

AUU AUU AGC AGC

IISSLSNKTIHNNNNYKYNYNNNYNNNNNYNNYNNTNYKKLYYNINYI

IISSLSNKTIHDNNNYKYNYNNNNNNYKNYNNY_______KKLYYNINYI

Kolics et al 2020 An Alternative, High Throughput Method to Identify Csd Alleles of the Honey Bee

DIA

The effect of zygosity in *csd* alleles

Heterozygous *csd* alleles (different alleles from queen and drone) produces females (workers and queens)

Hemizygous *csd* allele (single allele in a haploid individual) produces drones

Homozygous *csd* alleles (same allele from queen and drone) produces diploid drones

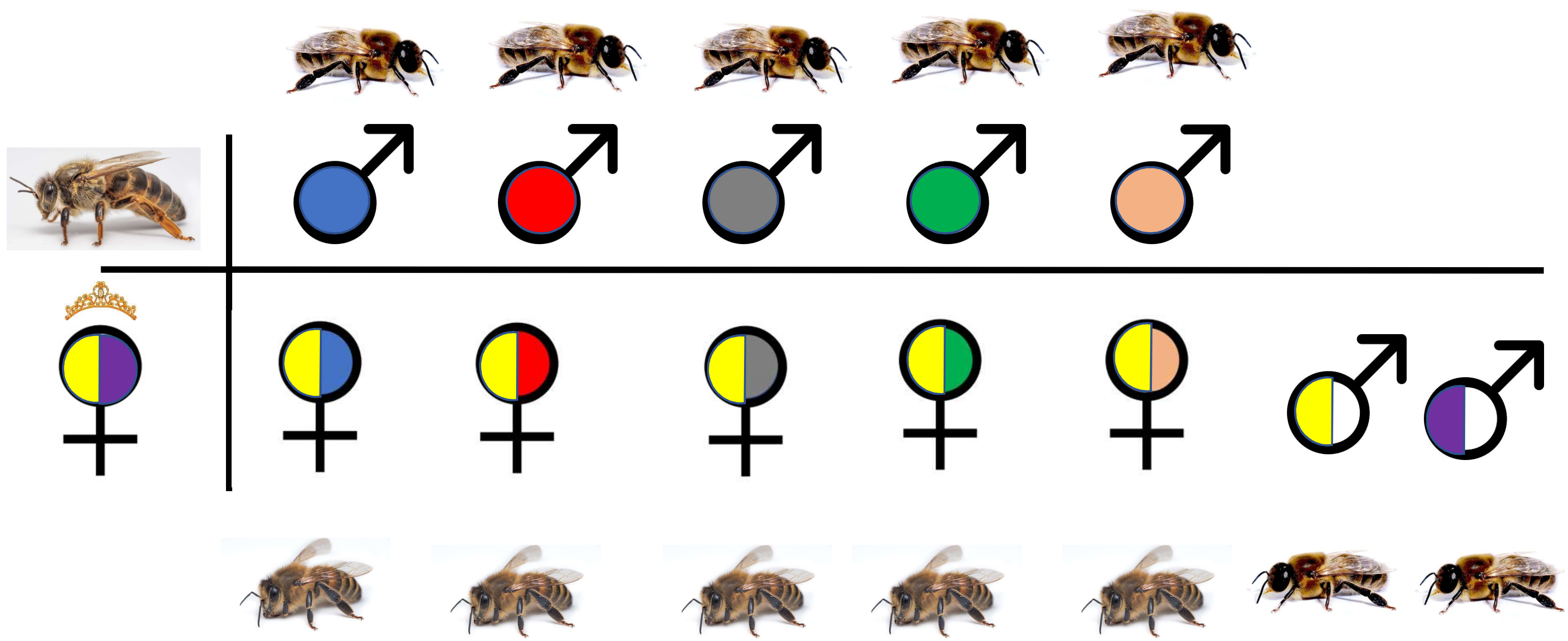
Diploid drone larvae are removed from the colony by workers

DIA

Healthy matings result from *csd* variation

Colours represent different *csd* alleles in the queen and drone gametes

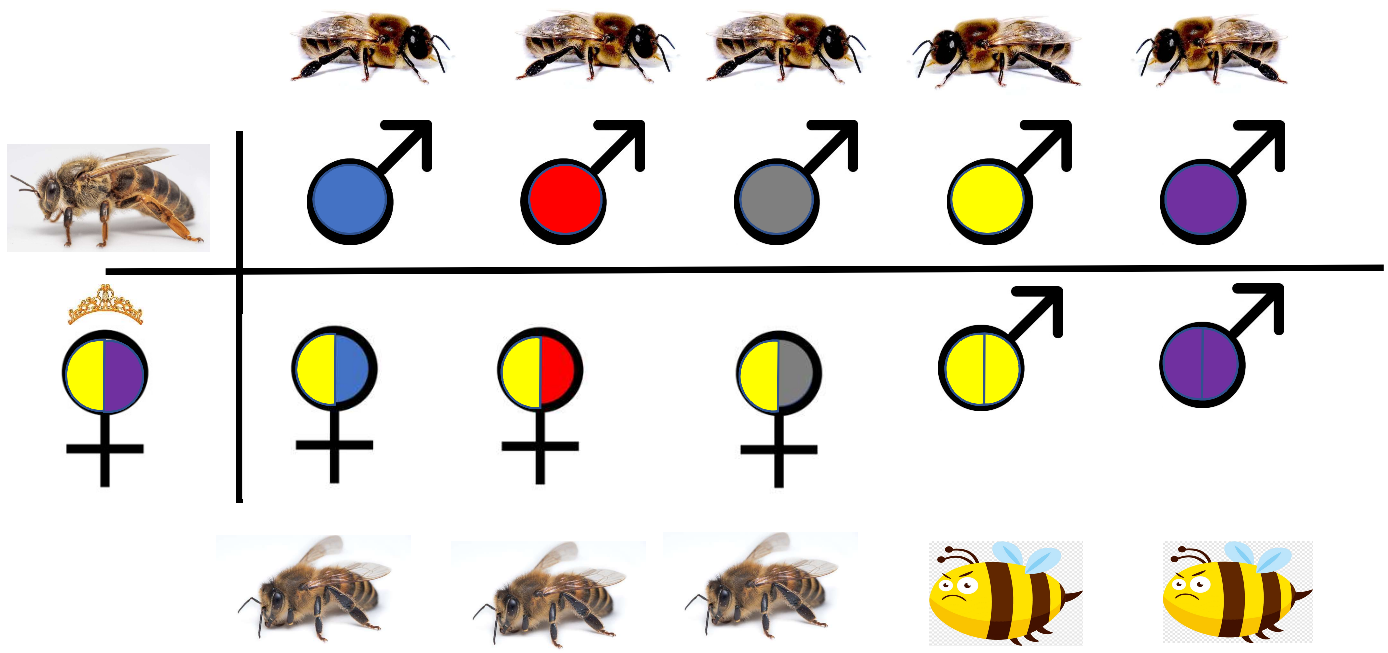
If the queen mates with drones that have different *csd* gene alleles from her, all the brood should be healthy



Adapted from H. Mooney 2018

Unhealthy matings result from lack of *csd* variation

Colours represent different *csd* alleles in the queen's and drone's gametes



If the queen mates with drones containing the same csd allele as her, some fertilised eggs will produce homozygous diploid drones.

Adapted from H. Mooney 2018

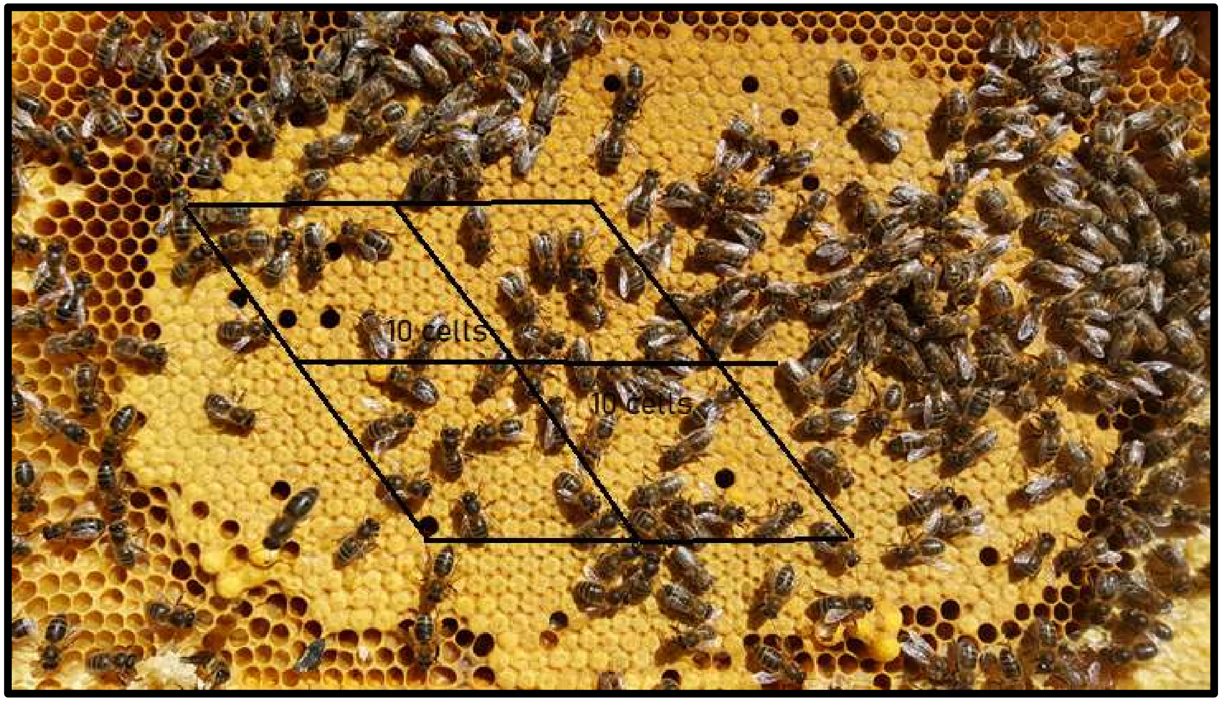
Too many diploid drones reduces brood viability (BV)

BV can be calculated

Make a template that covers 200 cells

(eg:10 by 20 cells)

1. For any 200 cells count the number of empty cells.
2. Subtract the number of empty cells from 200
3. Divide the resulting number by 2 to calculate the percentage of capped cells
4. Over 90% is ideal



This figure shows a 400 cell grid.
Four 10 x 10 rhomboids

DIA

Brood viability allows a calculation of *csd* alleles

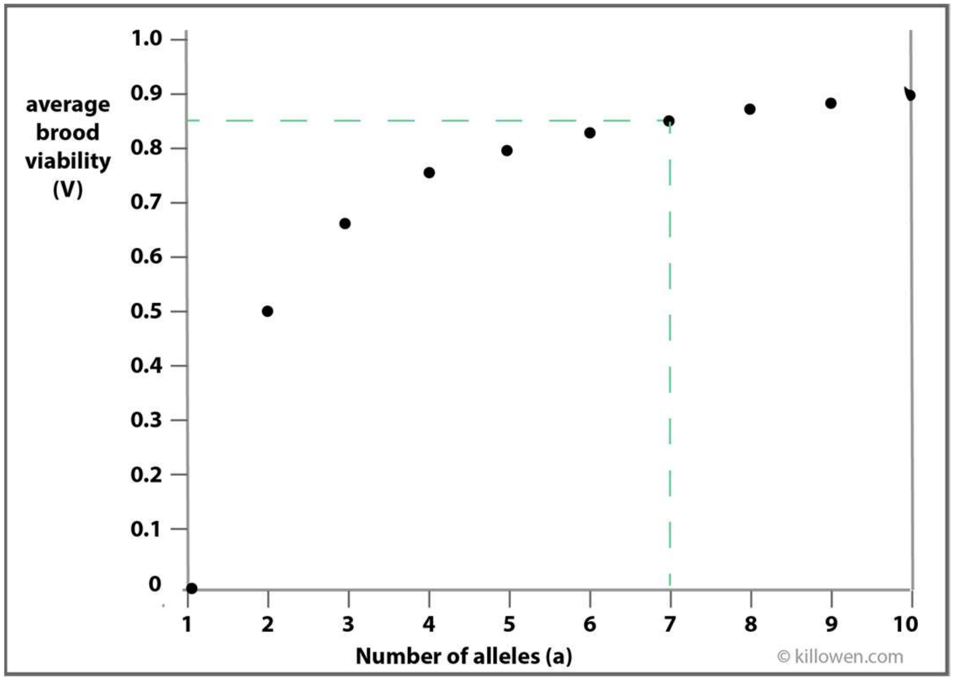
"the average brood viability *(V)* in a population of honey bee colonies is a function of the number of *csd* alleles in the population is given by:

$V = 1 - 1/a$ where *(a)* is the number of alleles"

Page and Marks 1982, THE POPULATION GENETICS OF SEX DETERMINATION IN HONEY BEES: RANDOM MATING IN CLOSED POPULATIONS

1. Calculate your BV
2. Read the number of *csd* alleles off the graph

https://www.killowen.com/genetics2B.html



In this example, an 85% brood viability indicates the presence of 7 *csd* alleles

DIA

Why genetic diversity is so important

Colony task diversification and protection against pathogens

<u>Diversity in foraging ability</u>

Foraging trait for nectar, water, pollen or propolis is heritable
(but also environmentally mediated)

<u>Social immunity against predators and pathogens requires many traits</u>

Hygienic behaviour (undertakers, Varroa specific hygiene (VSH)

Grooming behaviour (allo- and auto-) *(others- and self-)*

Propolis collection and use

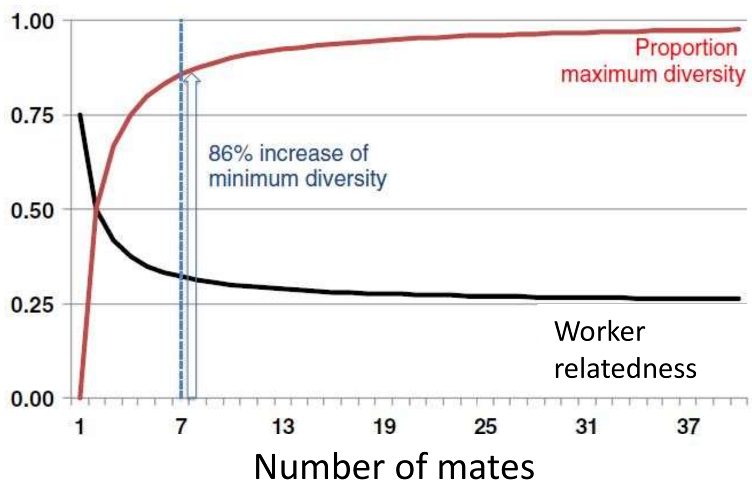
Defensive behaviour such as guarding and balling

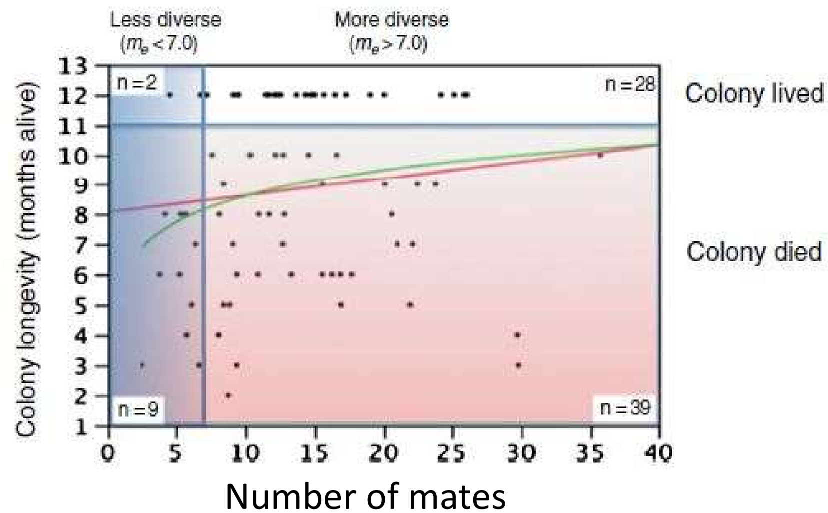
DIA

Why genetic diversity is so important

<u>Colony longevity</u>

Queen with more than 7 mates appears to leave the colony 2.86 times more likely to live





Tarpy et al 2013 Genetic diversity affects colony survivorship in commercial honey bee colonies

Aims and Learning outcomes

Aim : To introduce a broad understanding of genetics

1. To gain an understanding of the terminology involved in genetics
2. Enhance understanding of the central dogma of molecular biology
3. Enhance understanding of the haplo-diploid nature of the honey bee
4. Gain an understanding of the need for genetic diversity

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