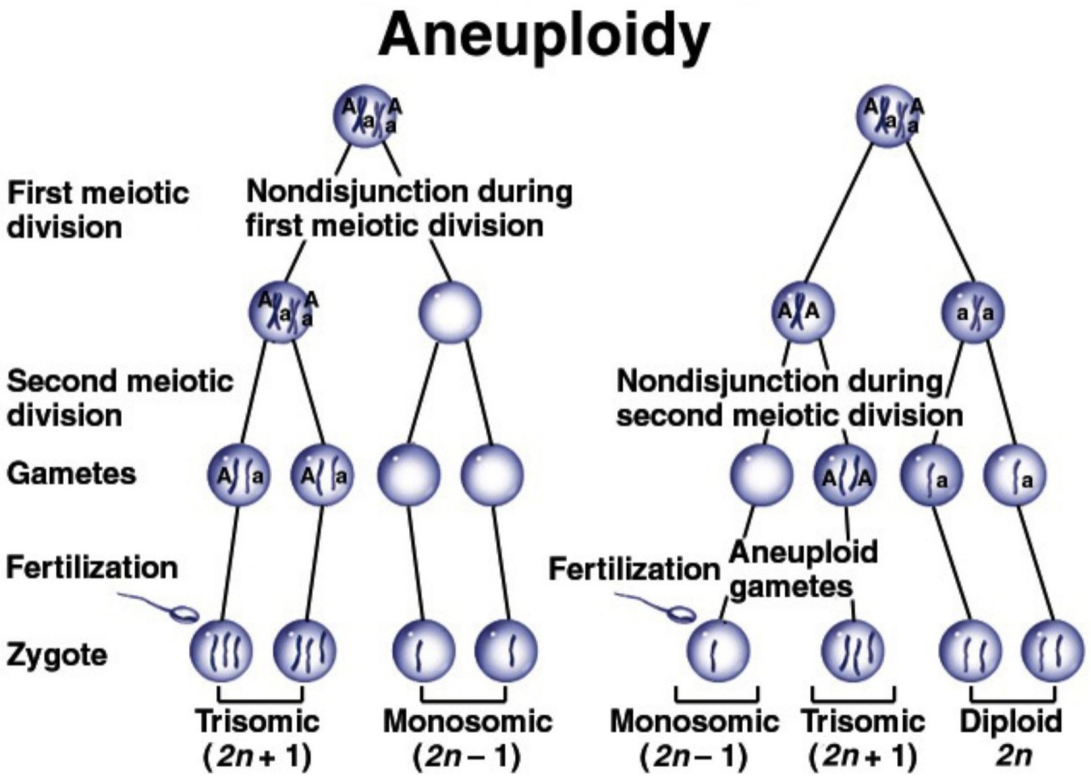
Bi 122 Final Review

Review Midterm content!

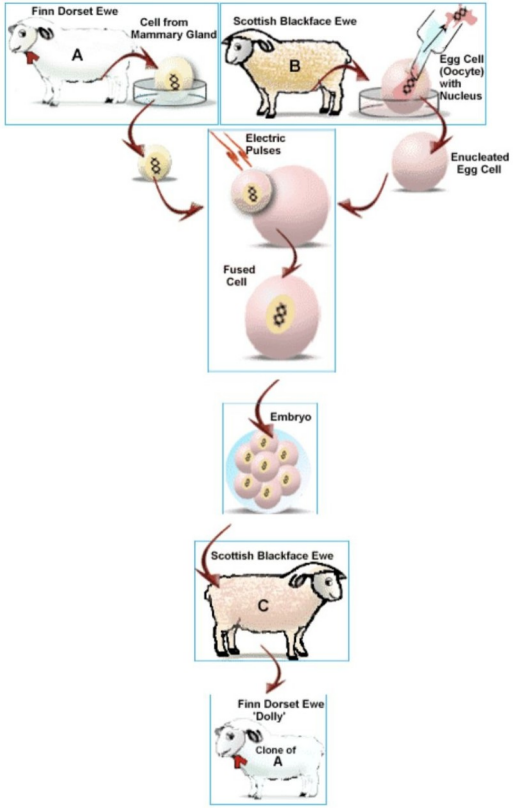
Polyploidy


Aneuploidy
First meiotic division
Nondisjunction during first meiotic division
Second meiotic division
Gametes
Fertilization
Zygote
Trisomic (2n + 1)
Monosomic (2n – 1)
Nondisjunction during second meiotic division
Fertilization
Aneuploid gametes
Monosomic (2n – 1)
Trisomic (2n + 1)
Diploid 2n

Genomic imprinting: Maternal alleles are unequal to paternal alleles

1. Mouse embryos with two maternal or paternal genomes fail to develop.
2. Human triploid abortuses are phenotypically different depending on whether the extra genome is maternal or paternal
3. Certain human characters are autosomal dominant, but manifest themselves only when inherited from one parent. For example, in some families, glomus tumors are inherited as an autosomal dominant character, but expressed only in people who inherit the gene from their father
4. Deletion of certain chromosomal regions produces a different phenotype when the deletion is present on the maternal or paternal chromosome.
5. Allele loss in many cancers preferentially involves the paternal allele.

Nuclear Cloning

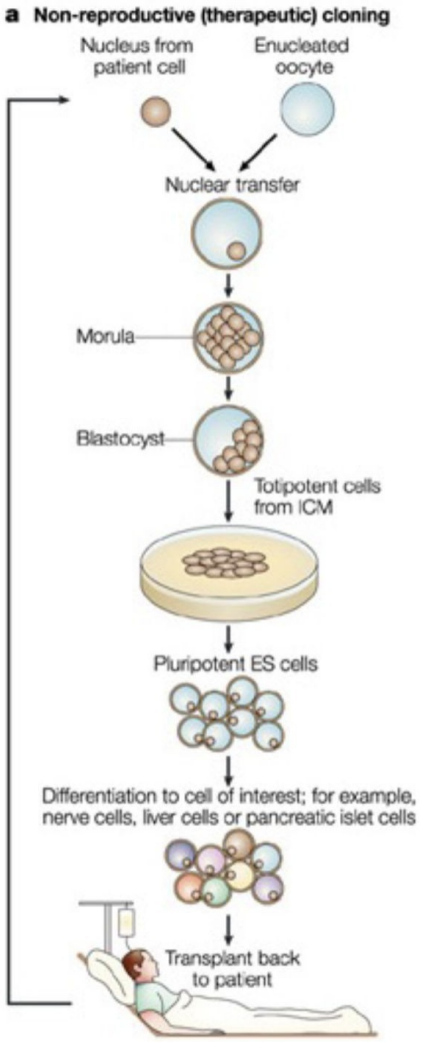


Nucleus removed
From egg cell

Cell with DNA fused
Into enucleated egg cell

Implantation into
The surrogate mother

Birth of clone of
Sheep A (Dolly)

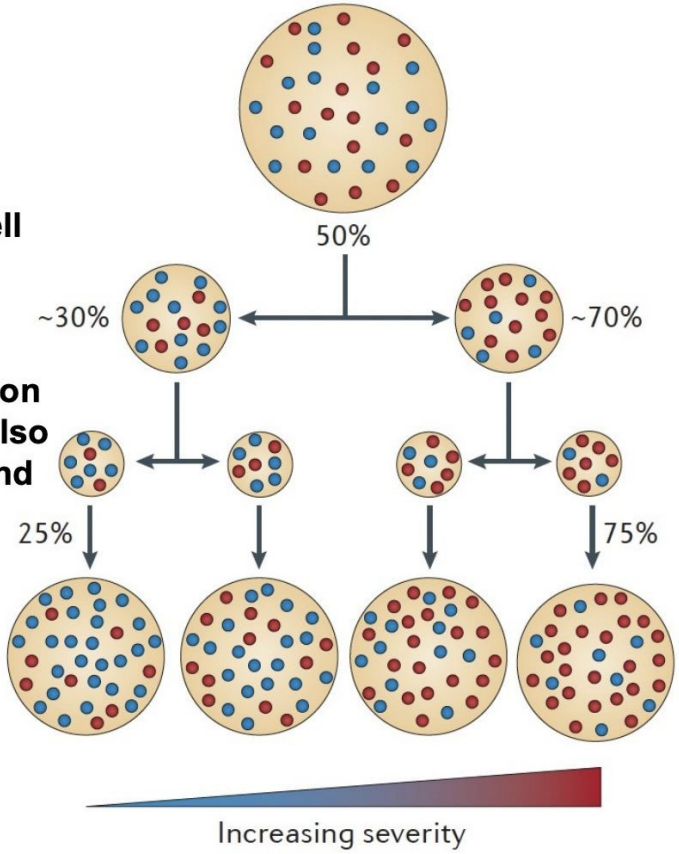


Homoplasmy: All mitochondrial DNA molecules are identical.

Heteroplasmy: Different mitochondrial genomes are present in the same cell

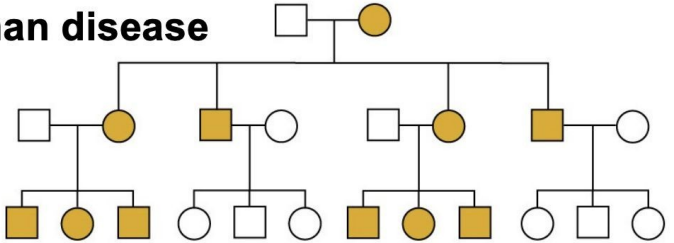
Different levels of heteroplasmy arise through random segregation during cell division.

In non-dividing cells mitochondria continue to undergo cycles of replication and degradation, a process which is also stochastic in terms of which are lost and which are replicated



Mitochondria and human disease

1. Maternal inheritance
2. Variable penetrance and expressivity
3. Variable age of onset



All offspring of diseased mother can express the disease phenotype, while none of the offspring of the diseased fathers do.

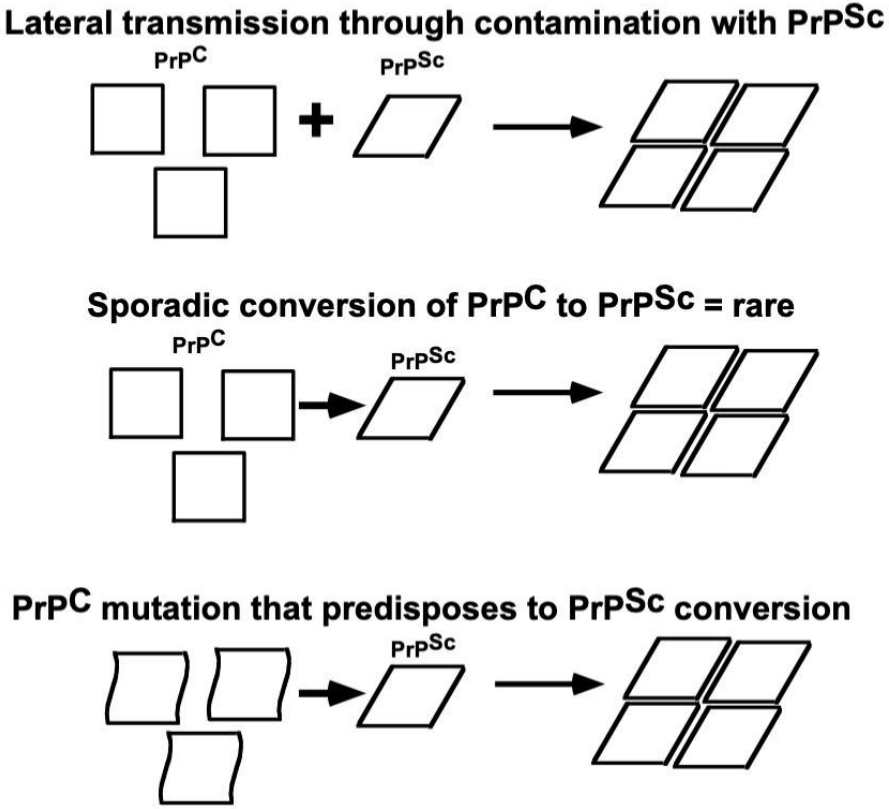
Individual mtDNA genotypes		Tissues Affected				
				Skeletal Muscle		
		Brain	Heart	Type I	Type II	Skin
I	20% mutant mtDNAs	+	–	–	–	–
II	40% mutant mtDNAs	+	+/–	–	–	–
III	60% mutant mtDNAs	+	+	+	–	–
IV	80% mutant mtDNAs	+	+	+	+/–	+/–

Problems with determining maternal inheritance in mtDNA diseases.

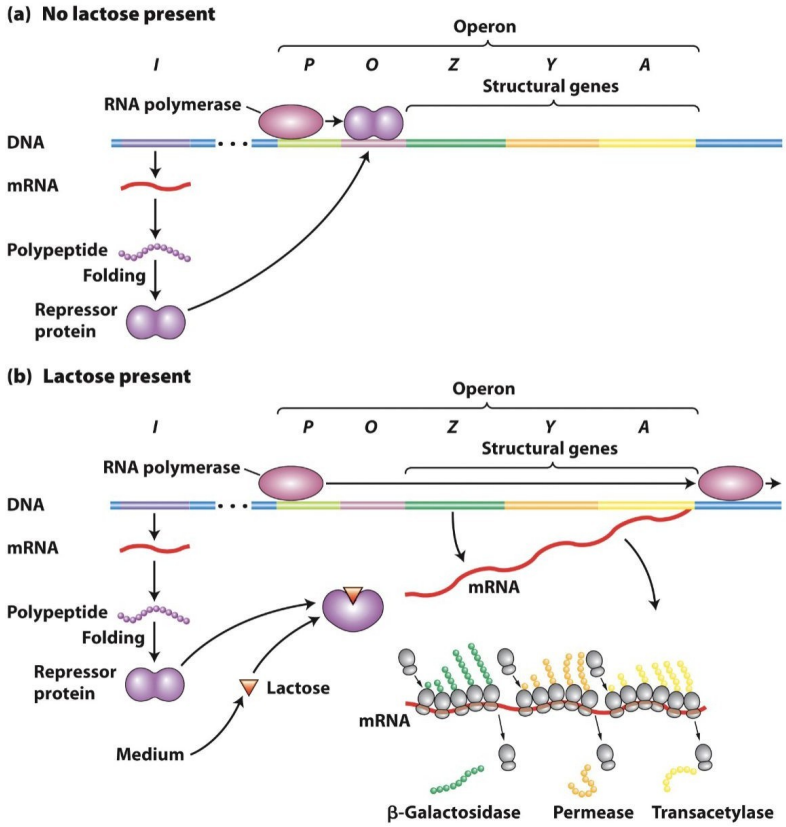
- Individuals within the pedigree will differ in phenotype (variable penetrance and expressivity) because of variability in mutant:wild type mtDNA ratio (heteroplasmy).
- Different organs within an individual can have different degrees of pathology because of heteroplasmy.

The proportion of mutant mitochondria determines the severity of the MERF phenotype and the tissues that are affected. Tissues with higher energy requirements (e.g., brain) are least Tolerant of mutant mitochondria. Tissues with low energy Requirements (e.g., skin) are only affected when the proportion Wildtype mitochondria is greatly reduced.

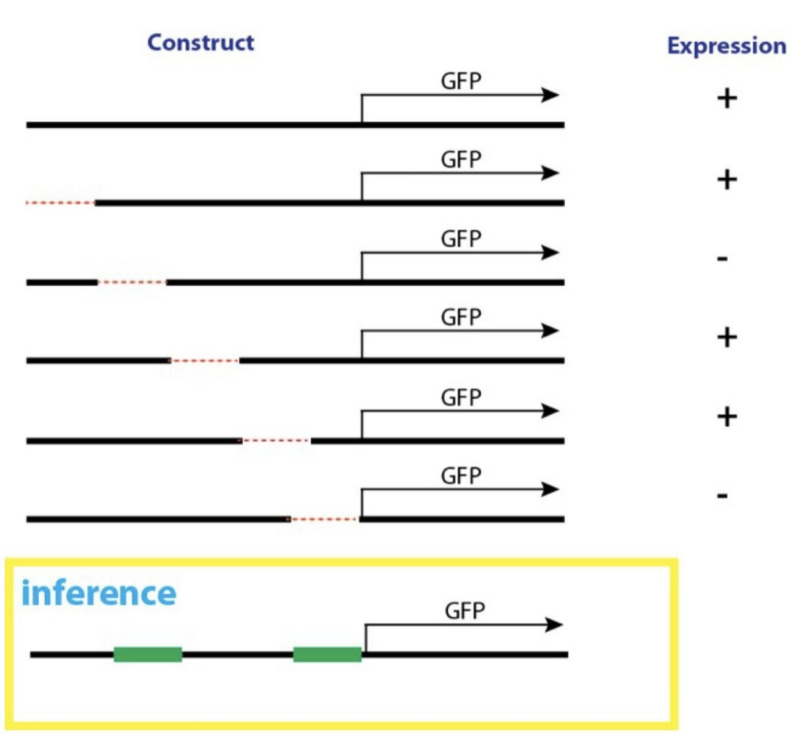
Prions

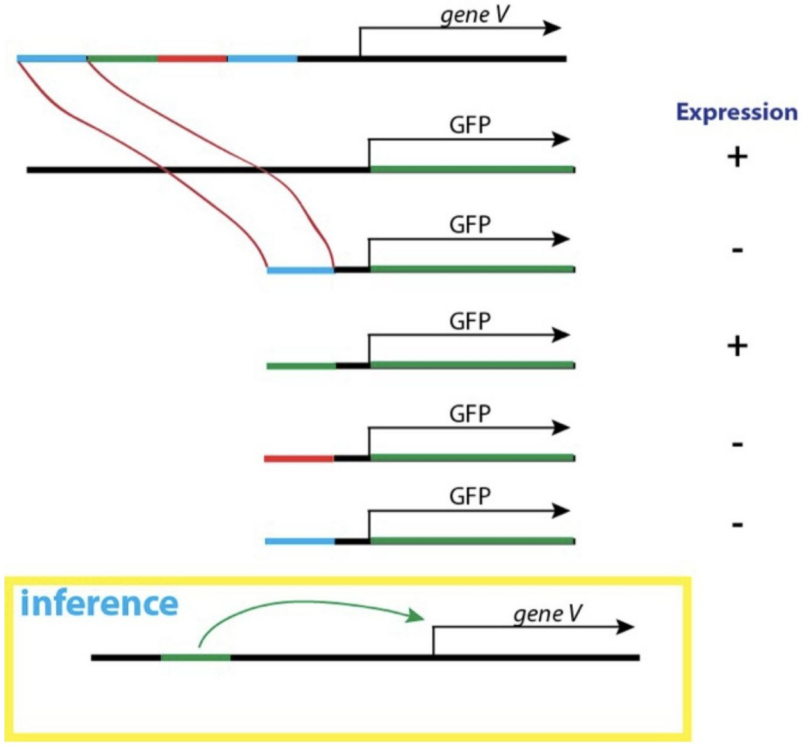

Lateral transmission through contamination with PrPSc
PrPC
PrPSc
Sporadic conversion of PrPC to PrPSc = rare
PrPC
PrPSc
PrPC mutation that predisposes to PrPSc conversion
PrPSc

Operons

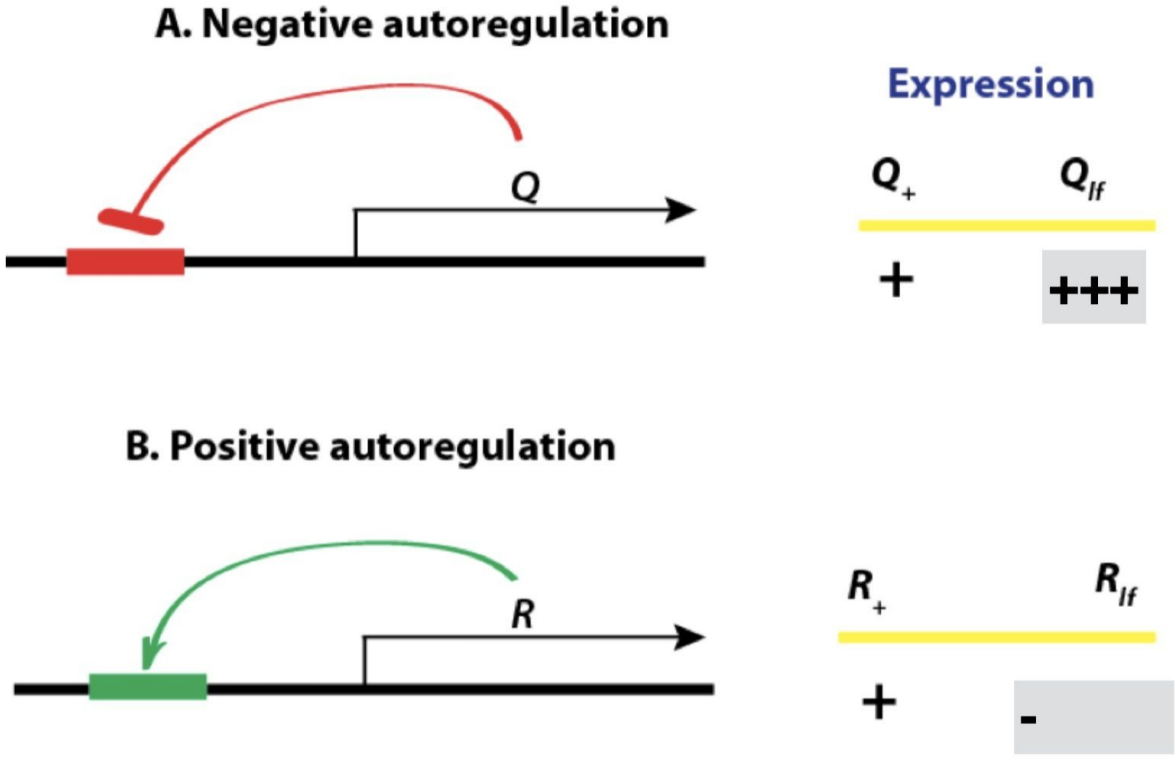

(a) No lactose present
Operon
I
P
O
Z
Y
A
Structural genes
RNA polymerase
DNA
mRNA
Polypeptide
Folding
Repressor protein
(b) Lactose present
Operon
I
P
O
Z
Y
A
Structural genes
RNA polymerase
DNA
mRNA
mRNA
Polypeptide
Folding
Repressor protein
Lactose
Medium
mRNA
β-Galactosidase
Permease
Transacetylase

Necessary vs Sufficient

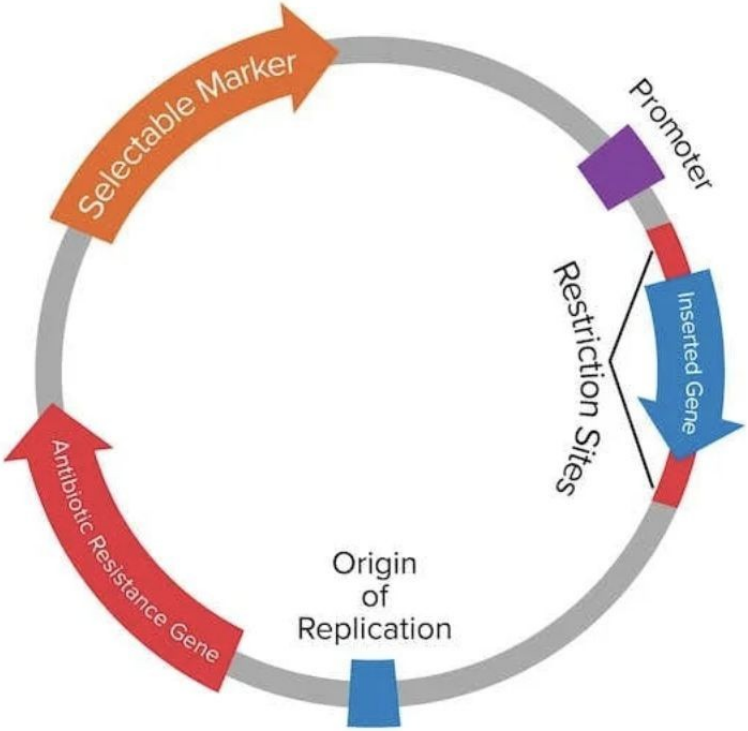




Positive vs Negative Gene Regulation

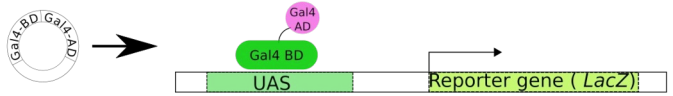


Plasmids

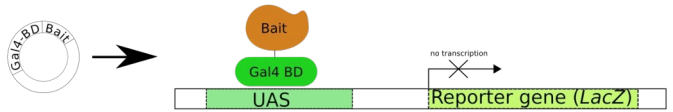


- Ori: origin of replication; where DNA replication begins
- Selectable marker: can include antibiotic resistance genes; helps us screen for bacteria containing the plasmid
- Restriction enzymes: area where plasmid is cut and gene is inserted
- Promoter: drives expression of inserted gene
- Screening: after plasmids are cloned and transformed into cells

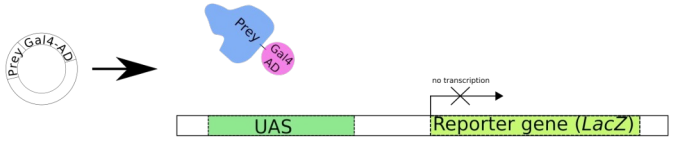
Yeast Two-Hybrid



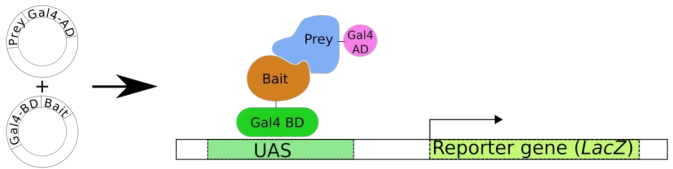
A. Regular transcription of the reporter gene



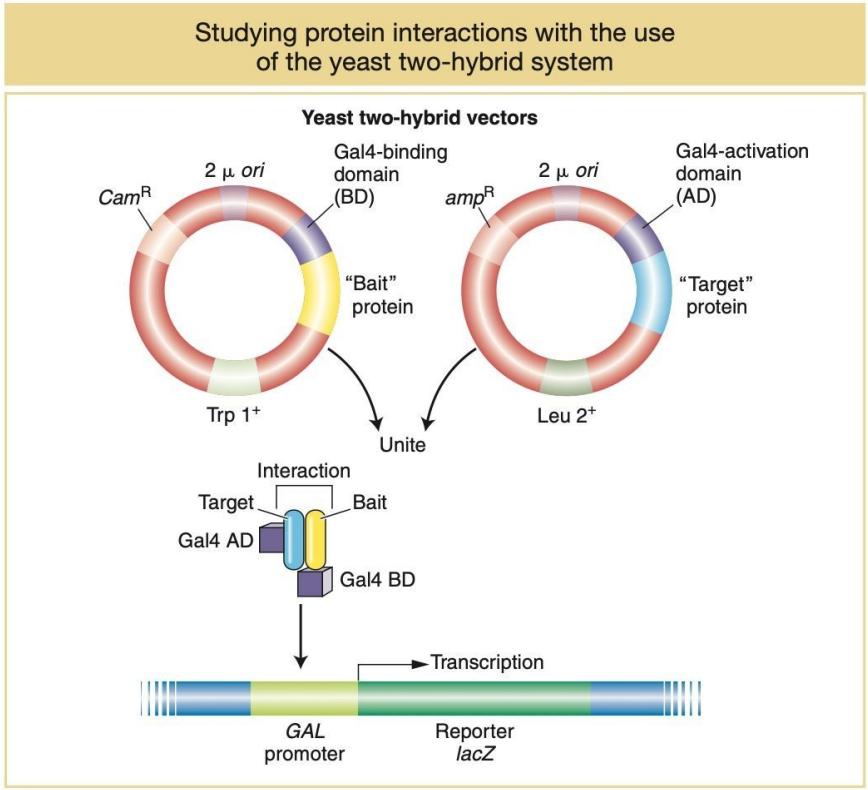
B. One fusion protein only (Gal4-BD + Bait) - no transcription



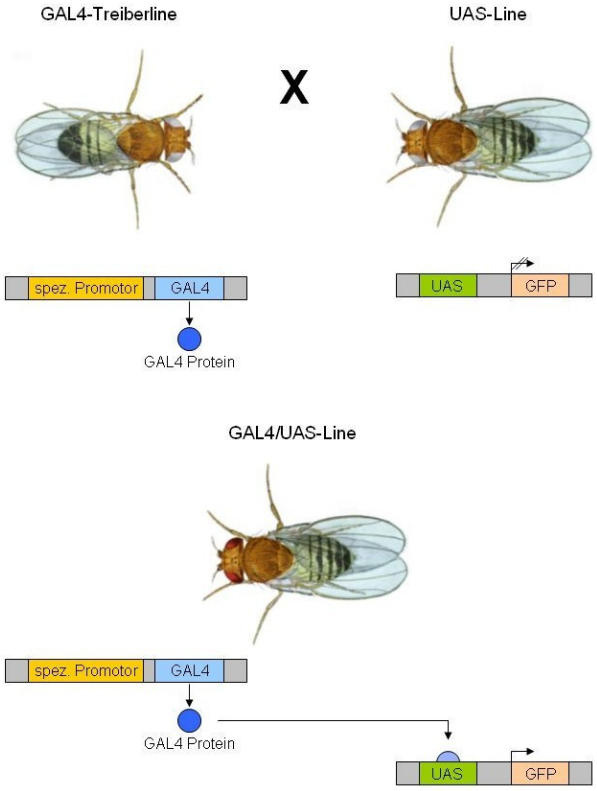
C. One fusion protein only (Gal4-AD + Prey) - no transcription



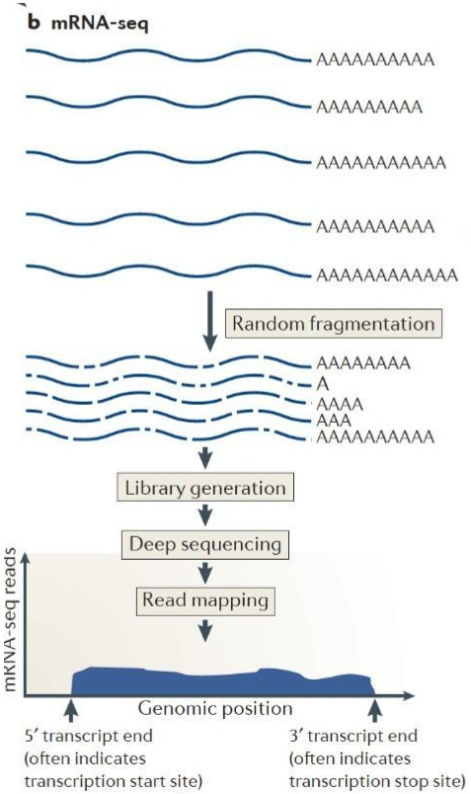
D. Two fusion proteins with interacting Bait and Prey

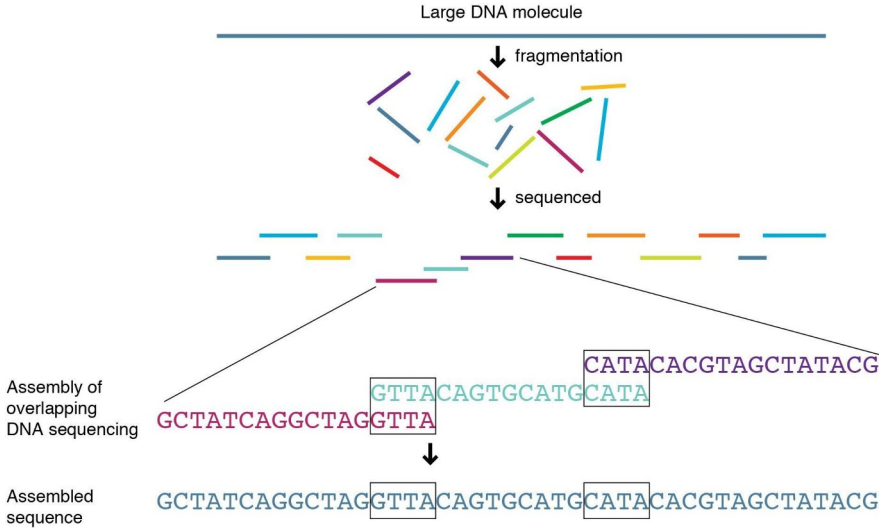


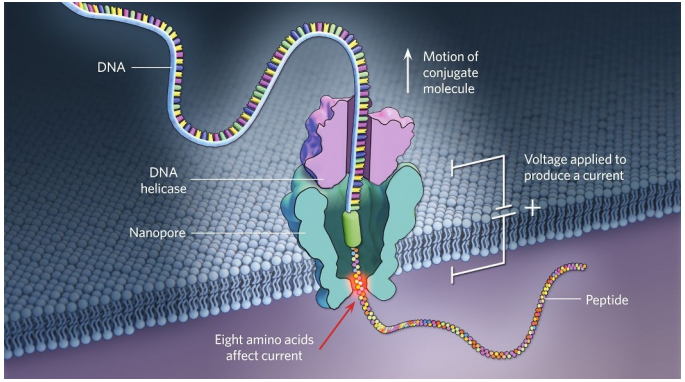
Gal4/UAS in other system



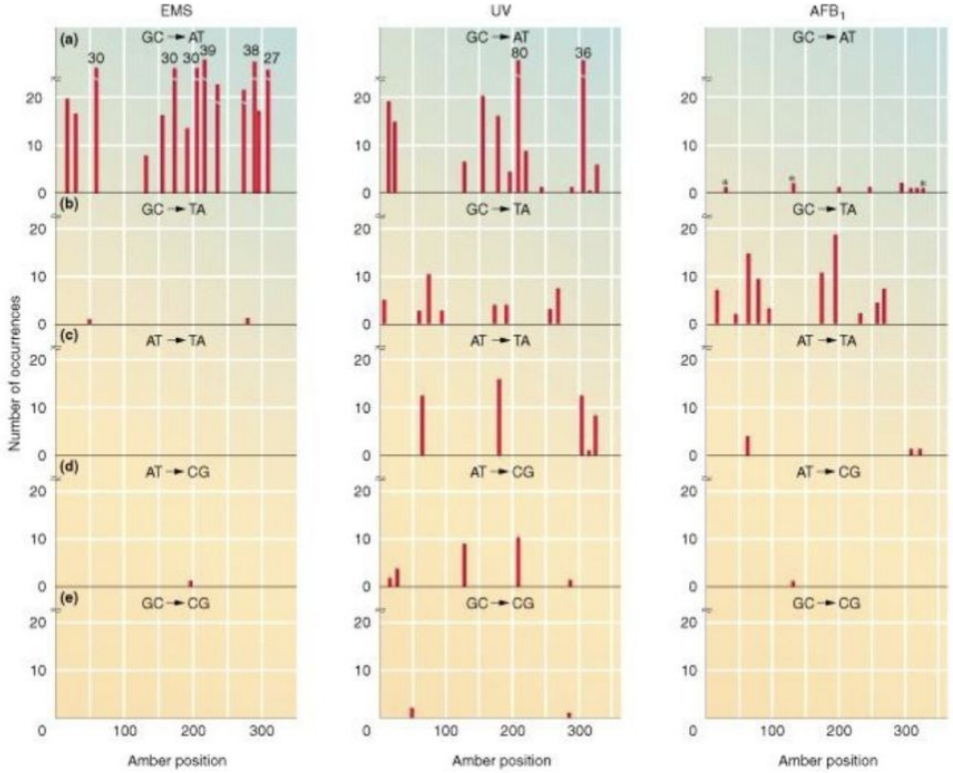
Sequencing


b mRNA-seq
AAAAAAAAAA
AAAAAAAAA
AAAAAAAAAAA
AAAAAAAAAA
AAAAAAAAAAAA
Random fragmentation
AAAAAAAA
A
AAAA
AAA
AAAAAAAAAA
Library generation
Deep sequencing
Read mapping
mRNA-seq reads
Genomic position
5′ transcript end
(often indicates
transcription start site)
3′ transcript end
(often indicates
transcription stop site)

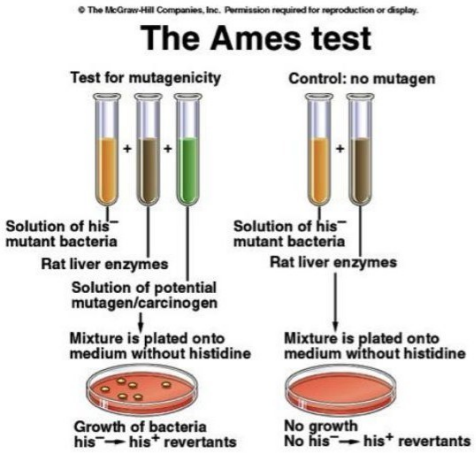

Large DNA molecule
fragmentation
sequenced
Assembly of
overlapping
DNA sequencing
GCTATCAGGCTAGGTTA
GTTACAGTGCATGCATA
CATACACGTAGCTATACG
Assembled
sequence
GCTATCAGGCTAGGTTACAGTGCATGCATACACGTAGCTATACG

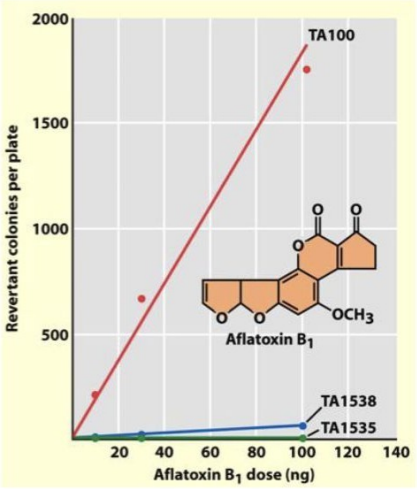

DNA
Motion of
conjugate
molecule
DNA
helicase
Voltage applied to
produce a current
Nanopore
Peptide
Eight amino acids
affect current

Mutagens


EMS
UV
AFB1
(a) GC → AT
(b) GC → TA
(c) AT → TA
(d) AT → CG
(e) GC → CG
30
30 30 39
38 27
80
36
Number of occurrences
Amber position
0
10
20
100
200
300

Mutagen Tests



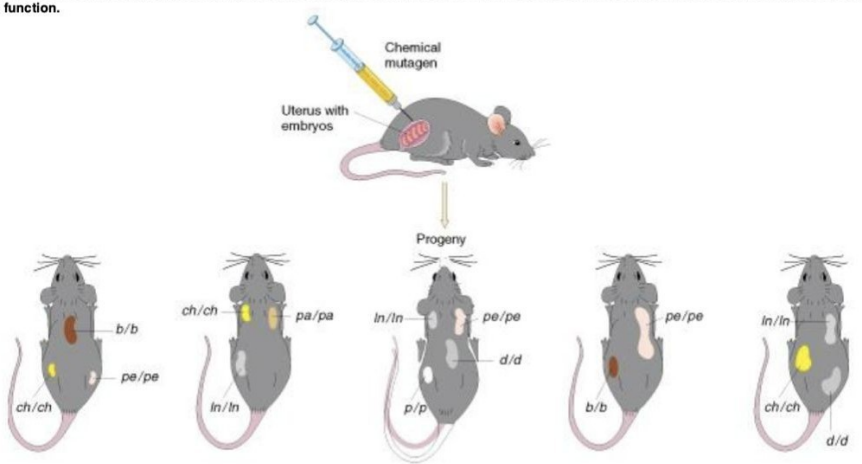


TA100 sensitive to base pair substitution, TA1535 and TA1538 sensitive to reversion through frameshift

1. The specific locus test

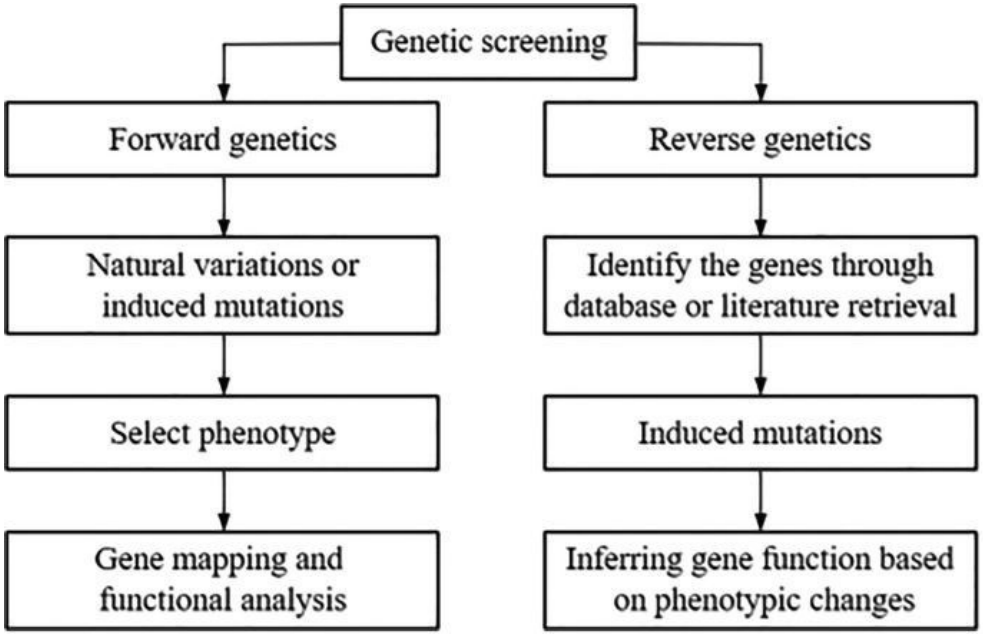
A. organism based tests

Parents homozygous for the markers listed are mated, giving rise to a female with offspring that are heterozygous at each of these loci. In the absence of loss-of-function mutations at any of these loci her offspring will all look wildtype. The combination of mitotic recombination and spontaneous mutation rate is given by the frequency of progeny with clones of homozygous mutant coat color. Increases above this background, in response to environmental stimuli, give a measure of mutagenicity. The use of multiple loci provides a way of averaging out problems associated with the fact that genes have different sizes, and that some are more sensitive than others to mutation-induced alteration In function.

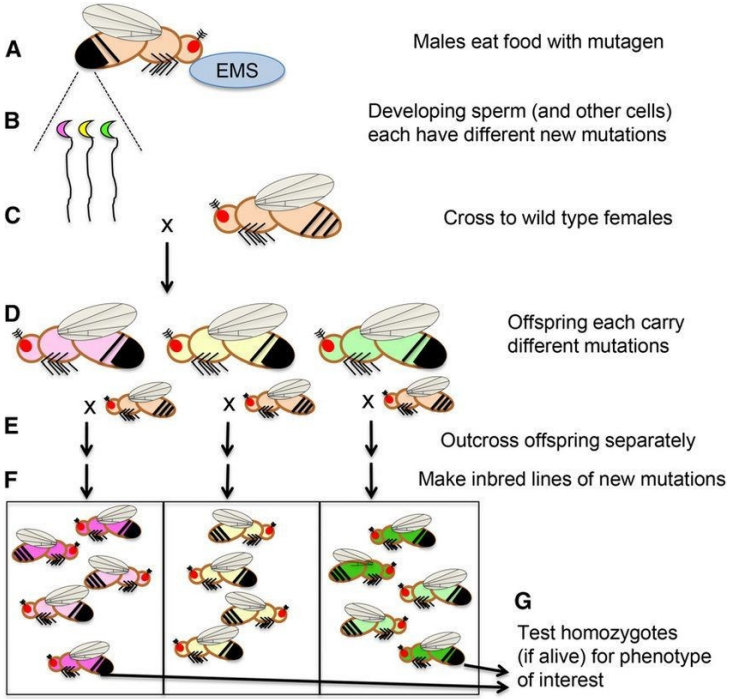


Genotype of progeny is *In/In*⁺ • *pa/pa*⁺ • *b/b*⁺ • *ch/ch*⁺ • *p/p*⁺ • *d/d*⁺ • *pe/pe*⁺
Phenotype of progeny is wild type with mutant sectors

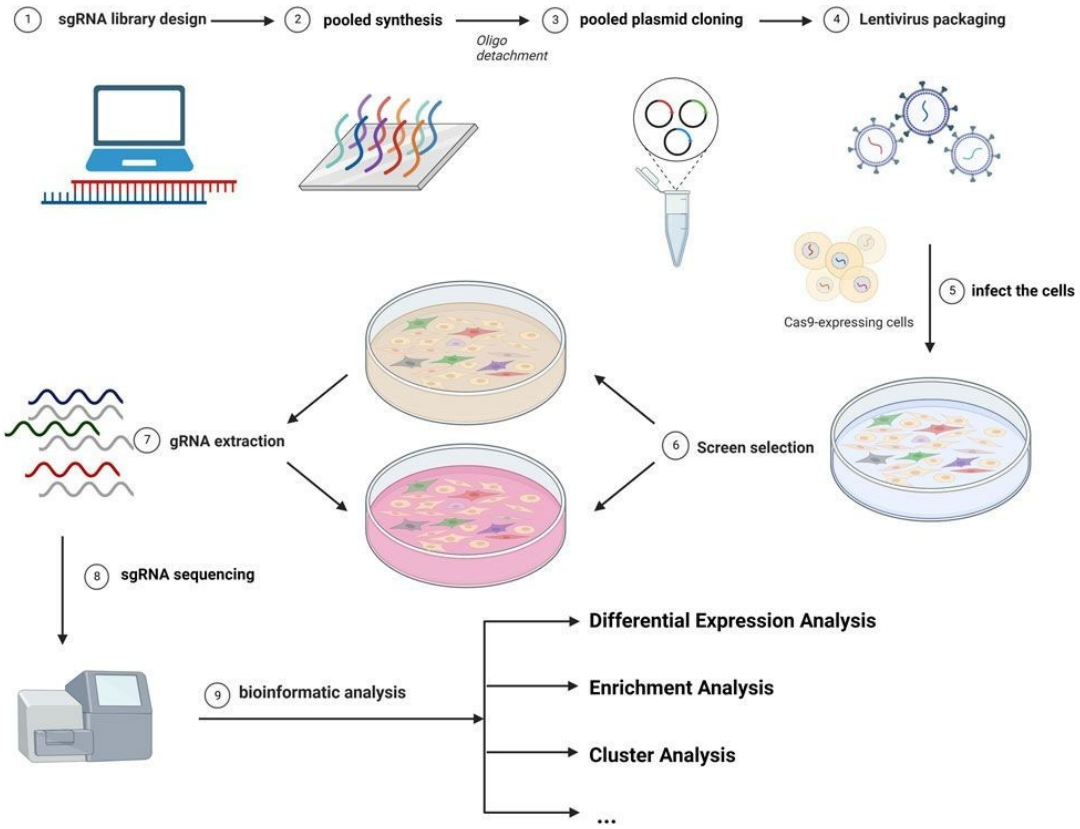
Genetic Screens



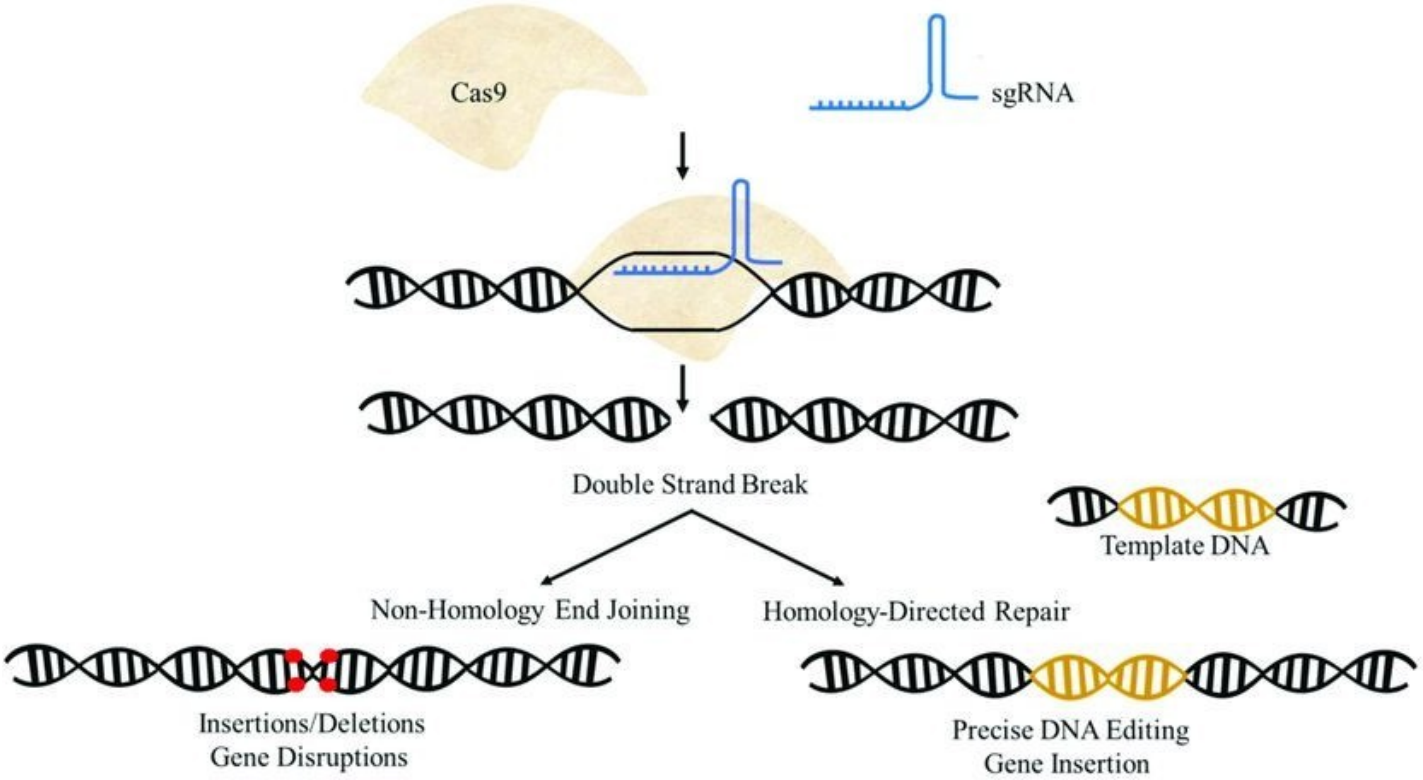
Forward Screen



CRISPR screening

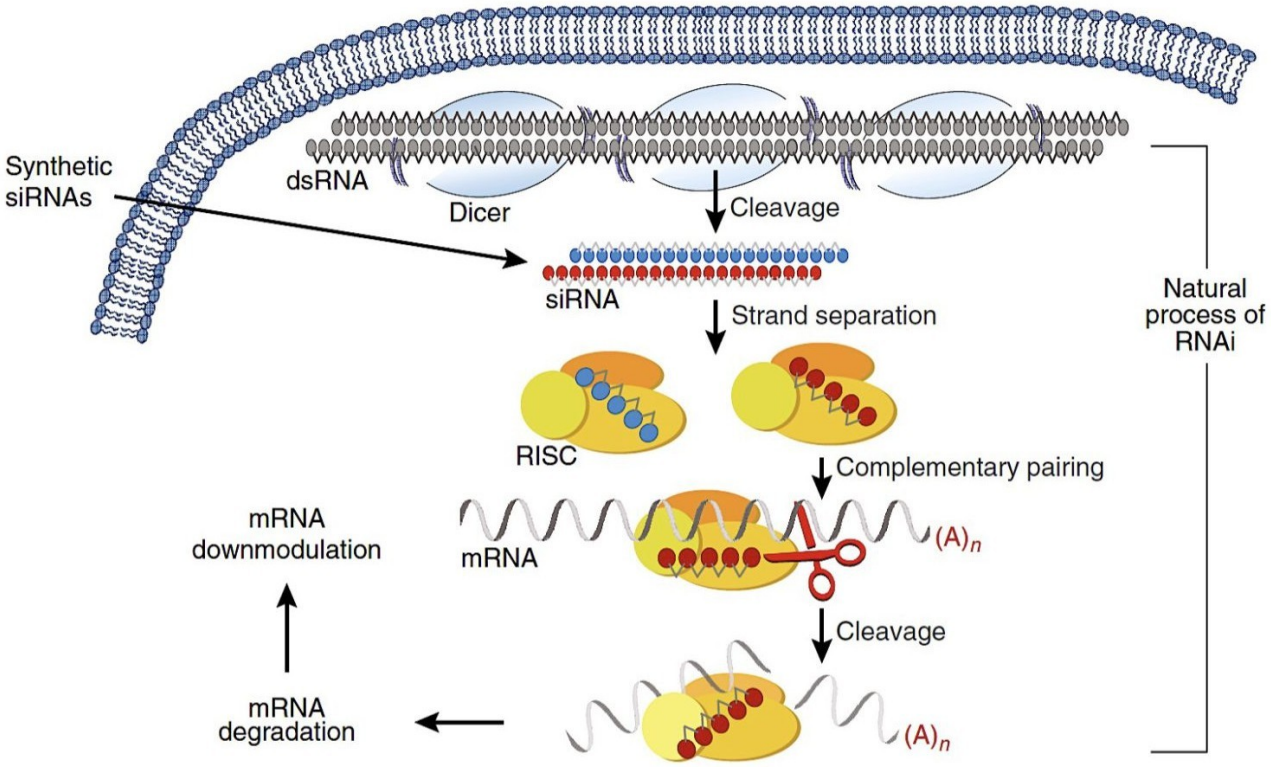


CRISPR Editing



Ahmad, A., Munawar, N., Khan, Z., Qusmani, A. T., Khan, S. H., Jamil, A., ... & Qari, S. H. (2021). An outlook on global regulatory landscape for genome-edited crops. *International journal of molecular sciences*, 22(21), 11753.

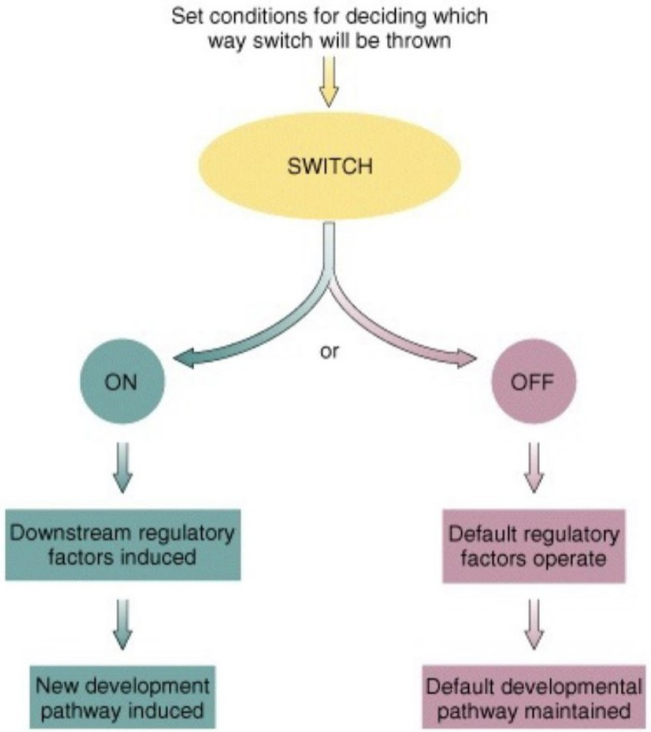
RNAi


Synthetic
siRNAs
dsRNA
Dicer
Cleavage
siRNA
Strand separation
RISC
Complementary pairing
mRNA
$(A)_n$
Cleavage
$(A)_n$
mRNA
degradation
mRNA
downmodulation
Natural
process of
RNAi

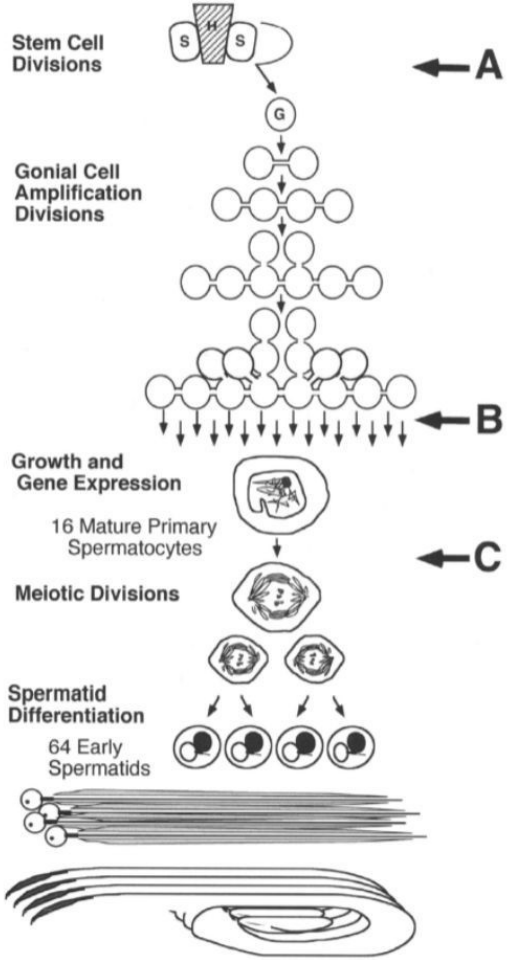
Oncogenes vs Tumor Suppressors

Oncogenes	Tumor suppressor gene
	Positive regulator of DNA repair
Positive regulator of cell growth	Negative regulator of cell growth
Activated forms promote malignancy	Inactivated forms promote malignancy
The mutated form is dominant	The mutated form is recessive

Epistasis Pathways

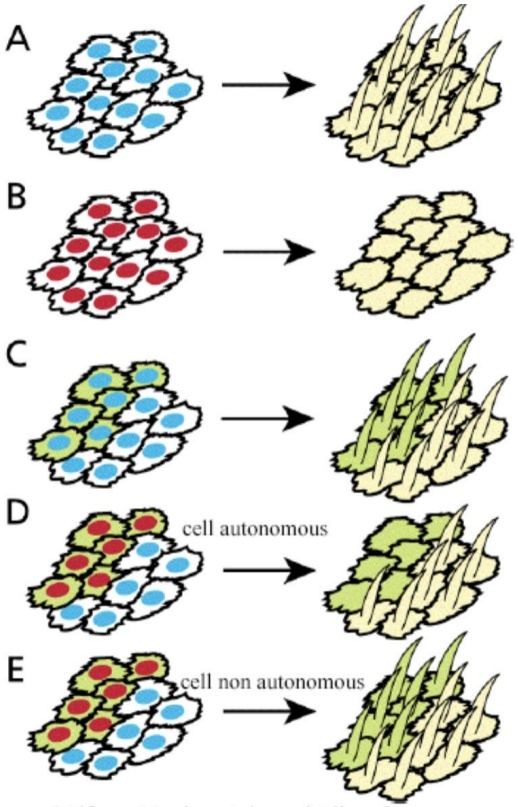


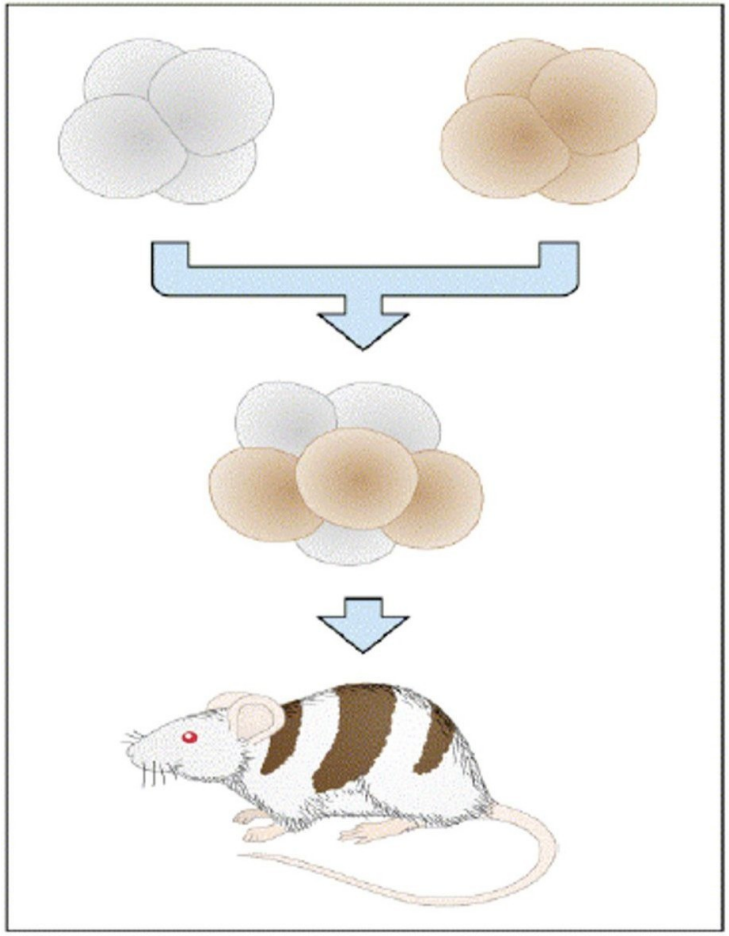
VS



gure 1. Overview of wild type spermatogenesis in *Dro-bhila*, showing three major cell fate transitions. Male

Mosaicism





Immunogenetics (Not covered)