Lecture 4

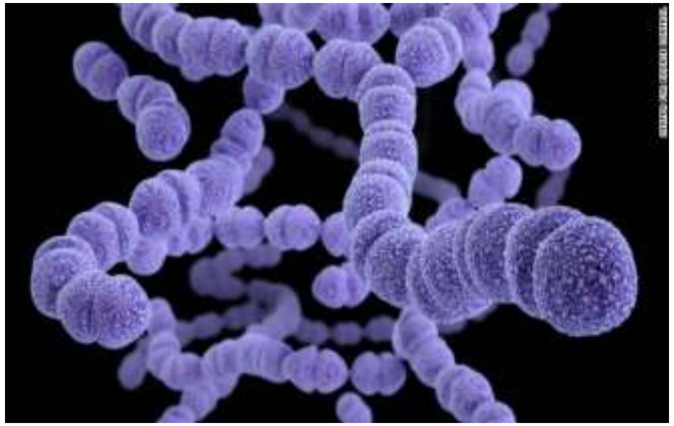
How did we get from Mendel's traits and alleles to DNA?

Like the one ring of power in Tolkien's "Lord of the Rings," **deoxyribonucleic acid (DNA)** is the master molecule of every cell.

https://science.howstuffworks.com/life/cellular-microscopic/dna.htm

(In 1928) while developing a vaccine for pneumonia…

Frederick Griffith
(1879-1941)
British medical officer and bacteriologist



- *Streptococcus pneumoniae* is a bacterium
- It causes pneumonia in mammals
- Griffith was trying to develop a vaccine
- By this time, some "biomolecules" were known: DNA, RNA, proteins
- But which of them is the genetic material?

Shown above is a digitally colorized image of Streptococci.
https://bioweb.uwlax.edu/bio203/f2013/schaefer_rya2/classification.htm

Different strains of *S. pneumoniae*

What are strains? Same organism having different stable phenotypes

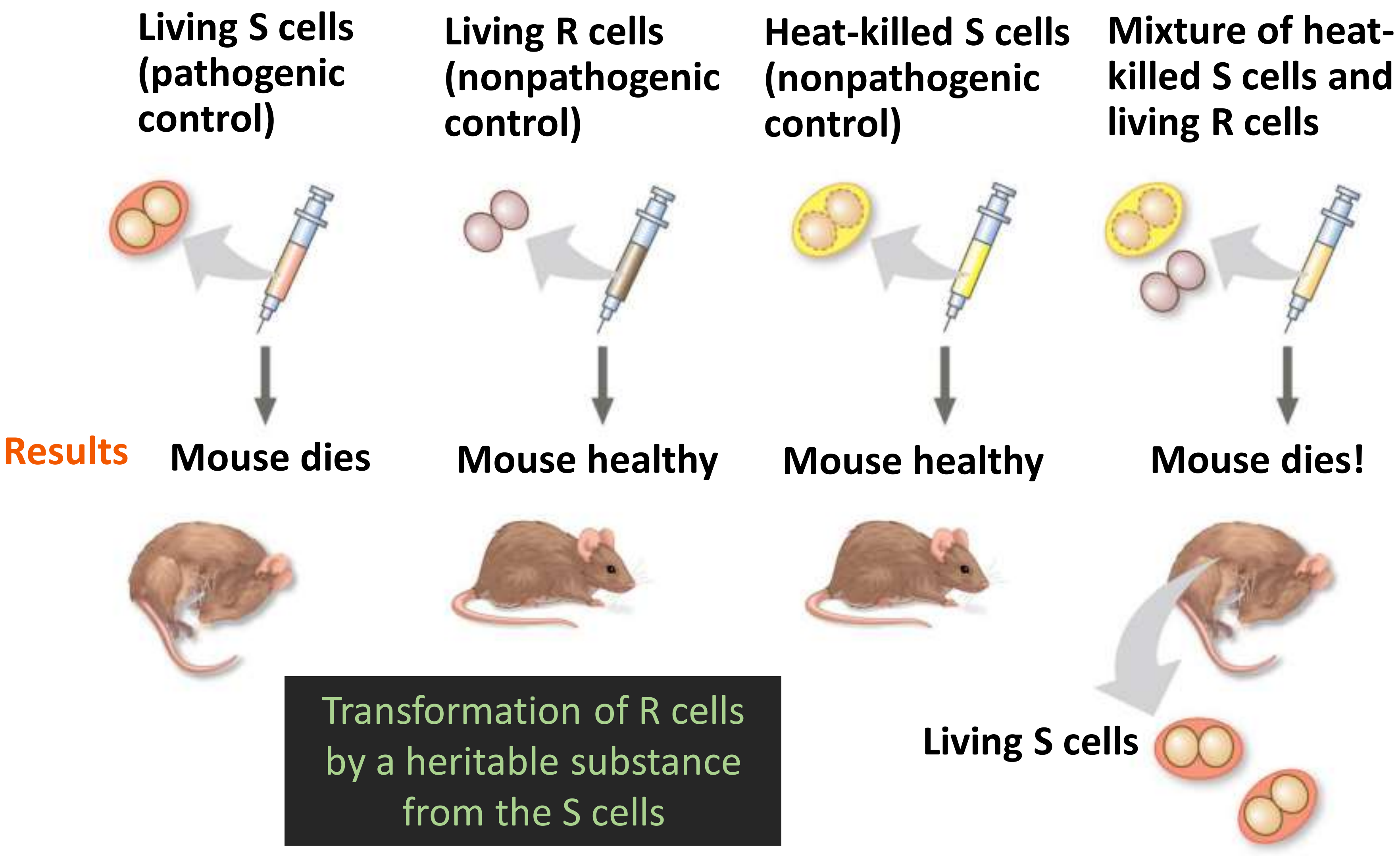
Strain S – appears smooth
- Has an outer capsule that protects from the defense system of mice
- Pathogenic strain
- Causes pneumonia in mice

Strain R – appears rough
- Lacks the outer capsule
- Non-pathogenic strain
- Does NOT cause pneumonia in mice

http://www.planetsrk.com/community/threads/5-movies-before-fan-in-which-shah-rukh-khan-played-double-roles.32656/

Strain S CANNOT become Strain R and vice versa

Unlike Mendel, who showed inheritance between generations, Griffith showed transfer of a trait between bacteria



Nature of the transforming factor?


Oswald Avery


Colin MacLeod


Maclyn McCarty

Images: Wikipedia

Heat killed pathogenic bacteria

DNA	RNA	Protein
inactivate	inactivate	inactivate
Test for its ability to transform	Test for its ability to transform	Test for its ability to transform

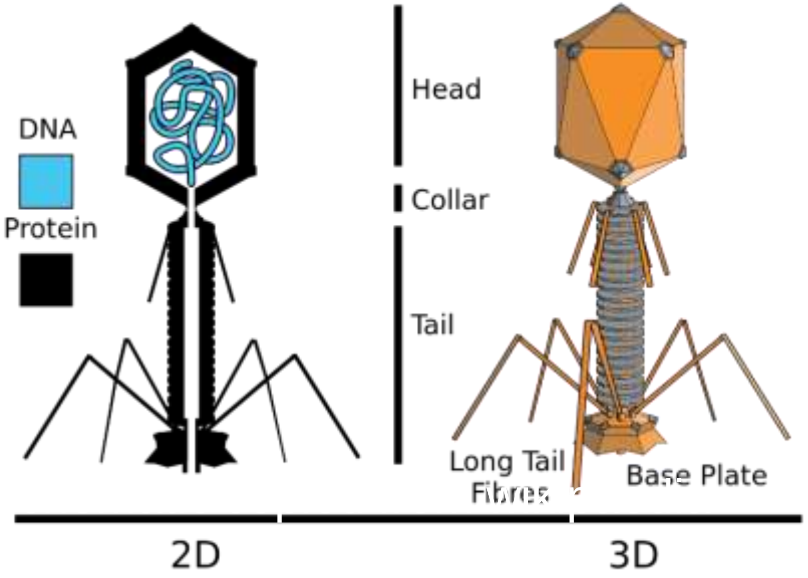
Question: based on a previous lecture, how would you separate DNA, RNA and proteins from each other?

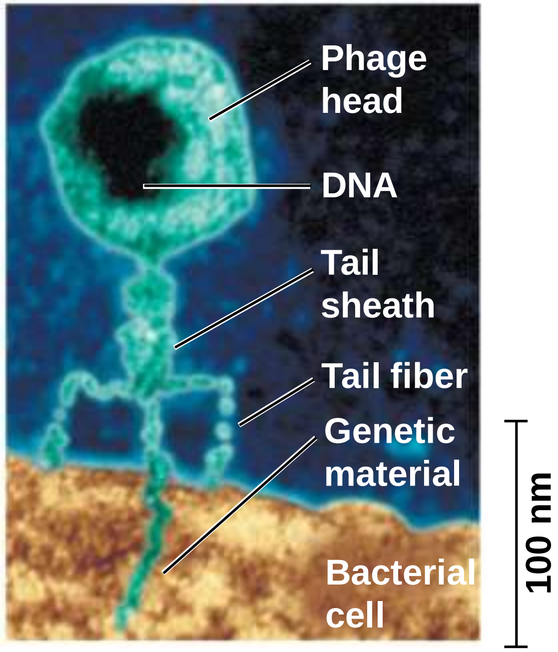
Passing on genetic information by a virus

Phages – viruses that infect bacteria

Phage T2

- Is attached to its host cell
- Is injecting the genetic material
- Head and tail parts remain outside the host

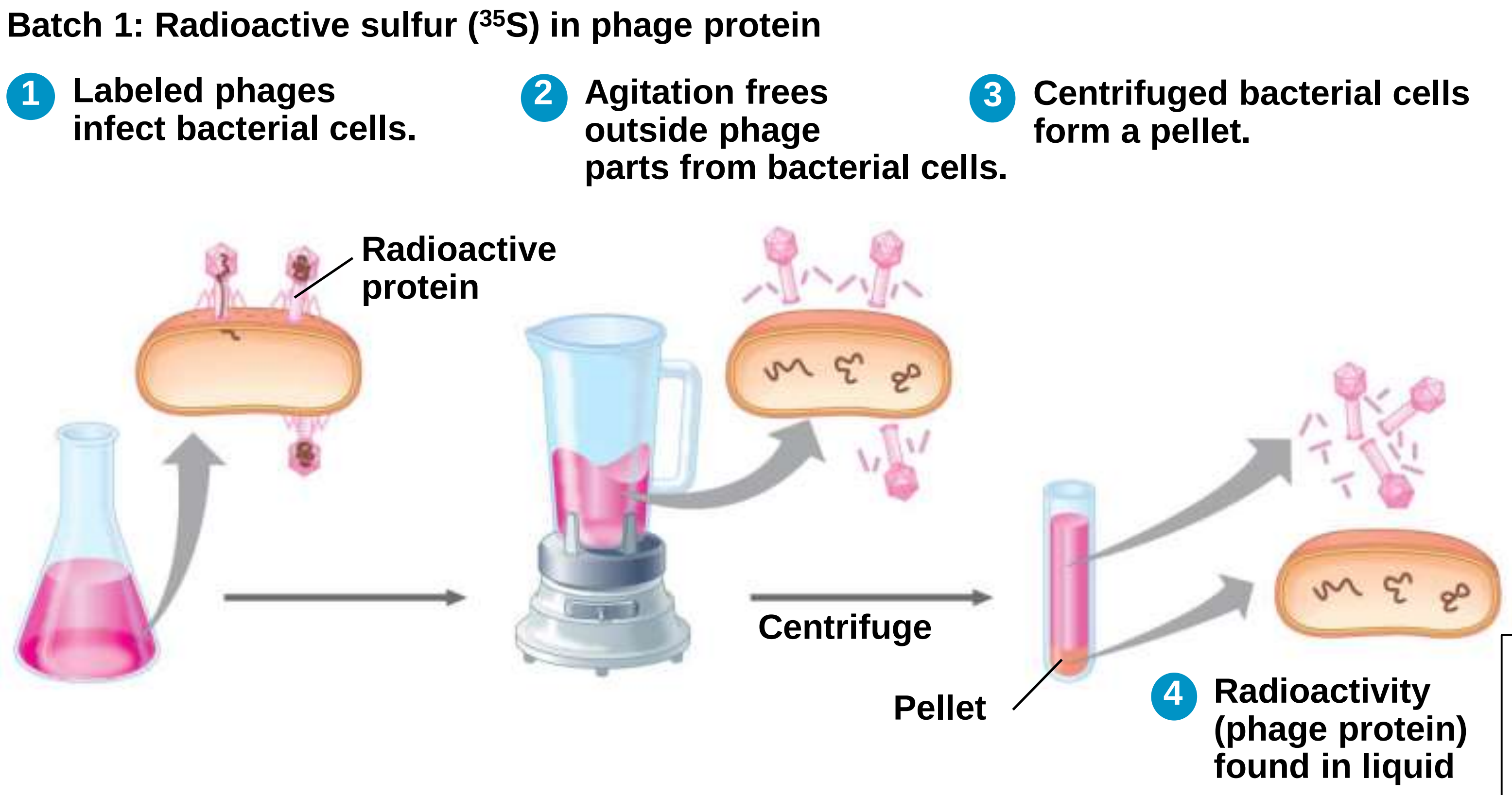




Colorized transmission electron micrograph

In 1952, Alfred Hershey & Martha Chase asked "What is the genetic material of T2 phage ?"

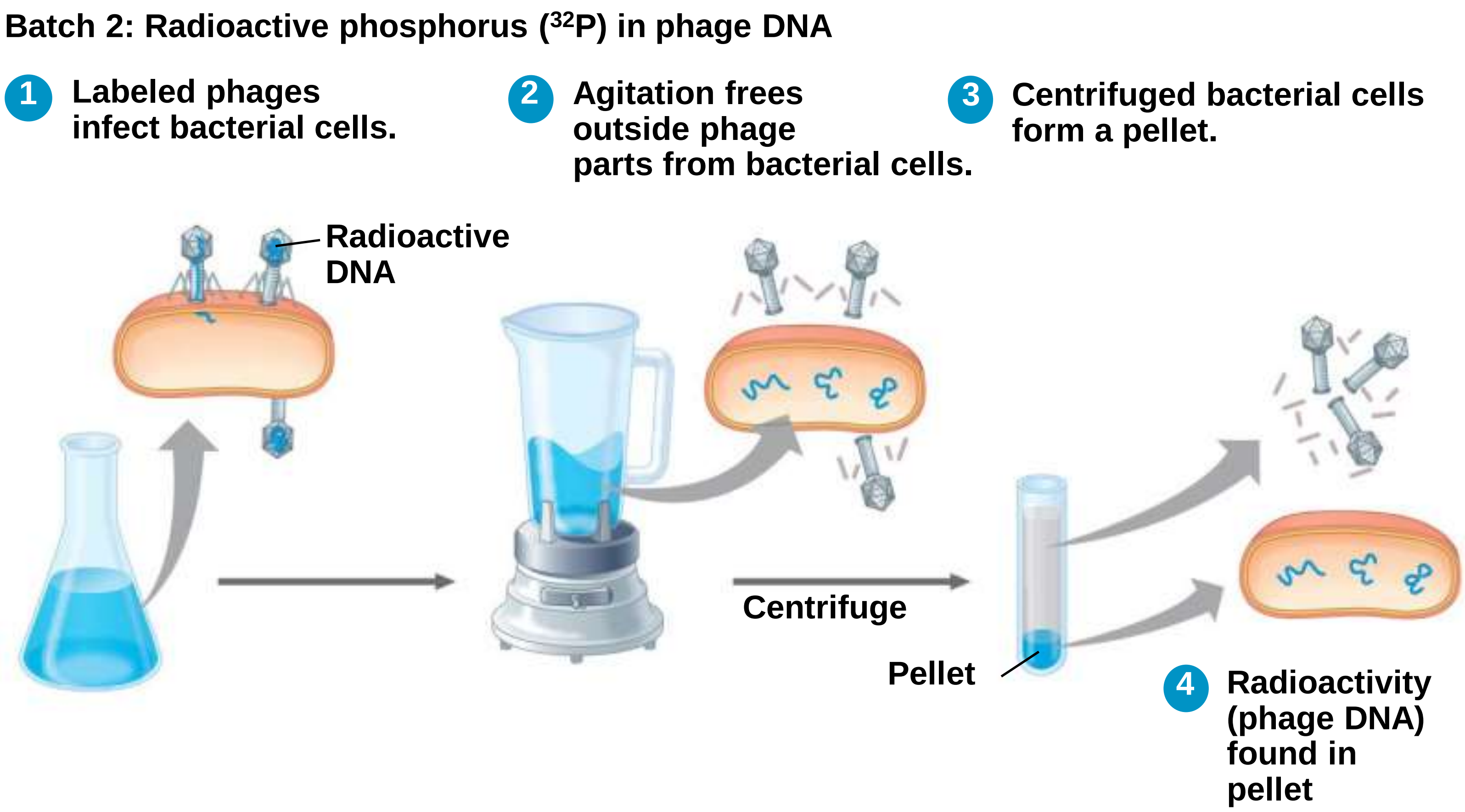
Taking advantage of S in proteins, P in DNA
Expectation: genetic material will be found in the host cells



Radioactive sulfur was not found in the bacterial pellet

Hershey-Chase experiment

DNA is the genetic material



Radioactive phosphorus was found in the bacterial pellet

Hershey-Chase experiment

The chemical composition of DNA was known: it is like Coca Cola (but carries genetic information)

Coke	DNA	Solubility
Water	**Present in water**	-
Sugar (sucrose)	**Sugar (deoxyribose)**	VERY High
Phosphate (PO_4^- acid)	**Phosphate**	moderate
Caffeine	**Nitrogenous bases (A,T, C, G)** You will see their structures in the upcoming slides.	extremely low

Next big question: what is the structure of DNA?

The base composition of DNA was known (Chargaff)

Source of DNA	Base percentage			
	Adenine	Guanine	Cytosine	Thymine
Sea urchin	32.8	17.7	17.3	32.1
Salmon	29.7	20.8	20.4	29.1
Wheat	28.1	21.8	22.7	27.4
E. coli	24.7	26.0	25.7	23.6
Human	30.4	19.6	19.9	30.1
Ox	29.0	21.2	21.2	28.7

Erwin Chargaff's observations

What was already known: DNA is a polymer consisting of A, C, G and T (referred to as nucleotide bases)

- **Observation #1**: Base composition of one organism differs from that of another

  Example: Adenine base constitutes 30.4% of human DNA but only 24.7% of *E. coli* (*Escherichia coli*)

  Implication: DNA captures the molecular diversity among species

- **Observation #2**: No. of A $\simeq$ No. of T; No. of G $\simeq$ No. of C

  Example: human DNA: A = 30.4%, C = 19.9%, G = 19.6%, T = 30.1%

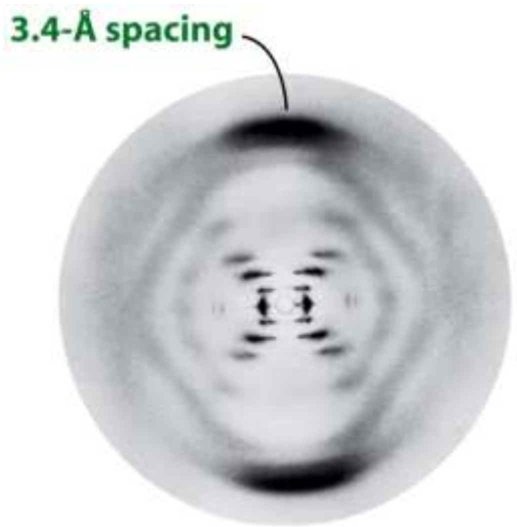
  Implication?

Hunt to elucidate the structure of DNA

Linus Pauling's hypothesis

- Three chains, twisted around each other in ropelike stands (turned out to be wrong)

Maurice Wilkins'/Rosalind Franklin's X ray crystallographic data

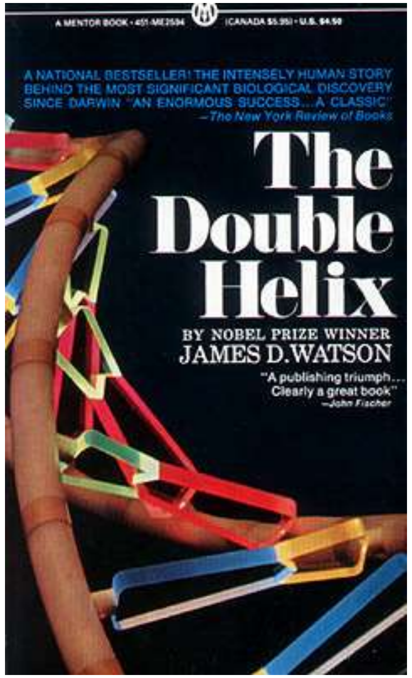


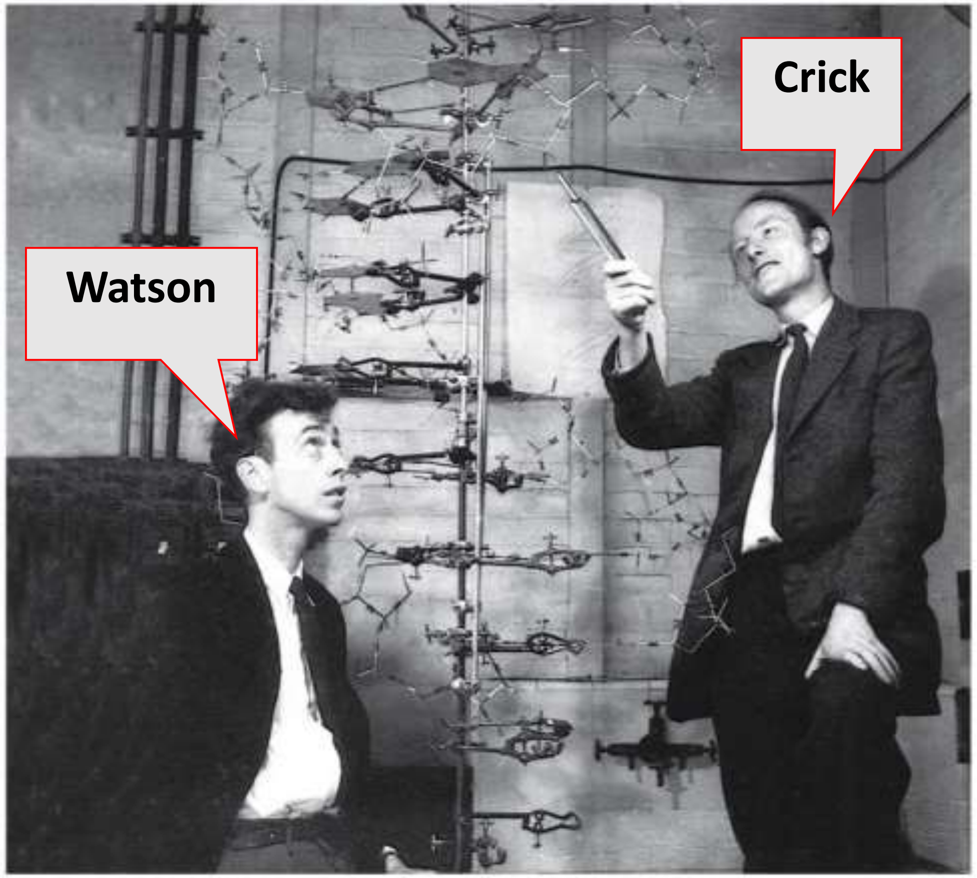
- Nucleotides are 3.4 Aº apart in the chain
- Structure repeats at 34 Aº interval

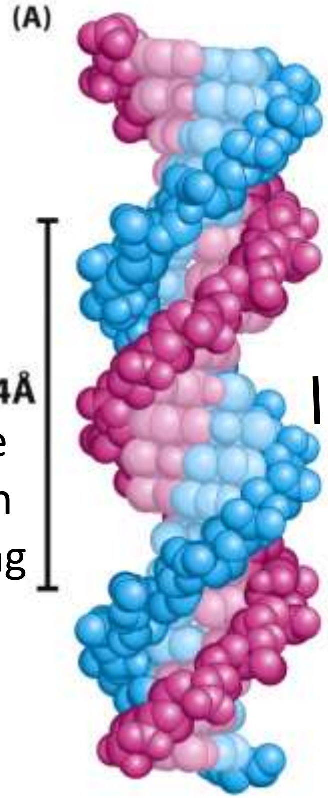
Erwin Chargaff's rule : %A = %T and %G = %C

Watson-Crick Model of Double-Helical DNA

1962 Nobel Prize







34Å

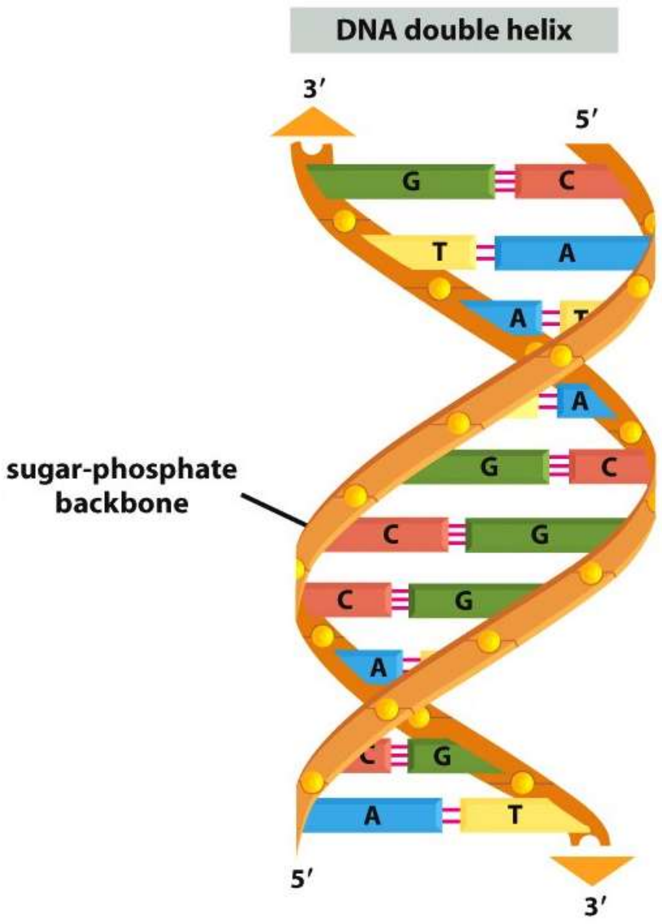
Distance between repeating unit of helix

3.4 Å

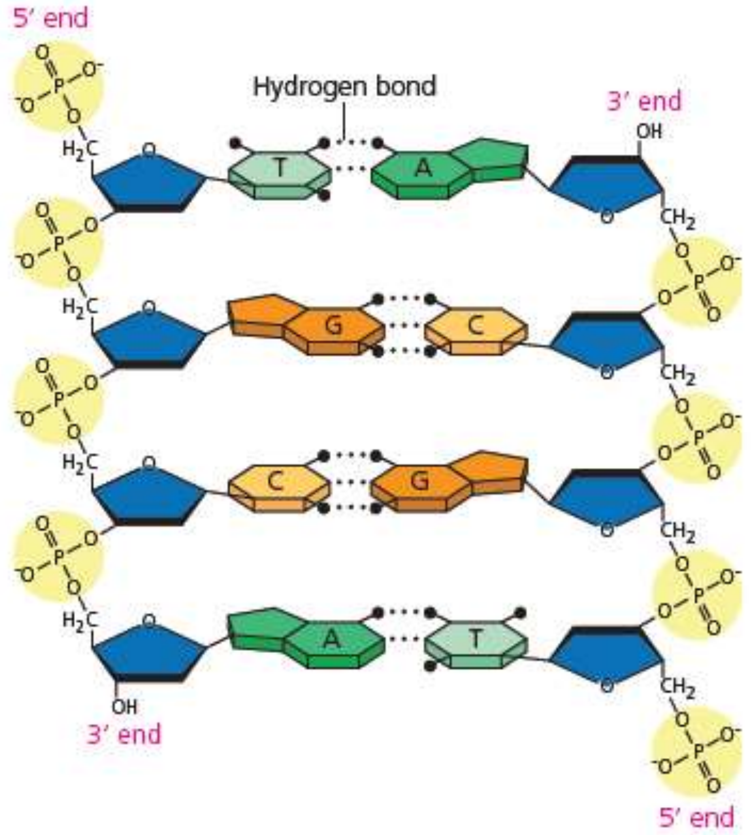
Distance between bases

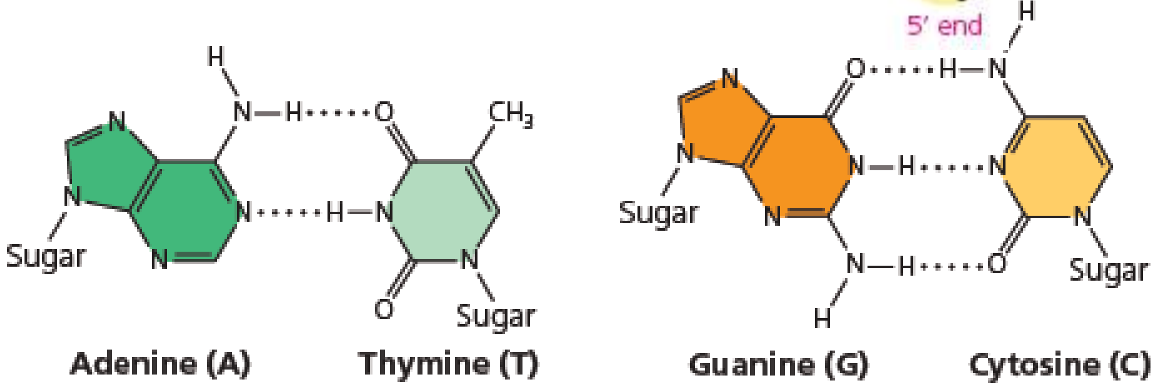
Watson and Crick played with chemical models to come up with a structure that matched the data

Features of the structure of DNA



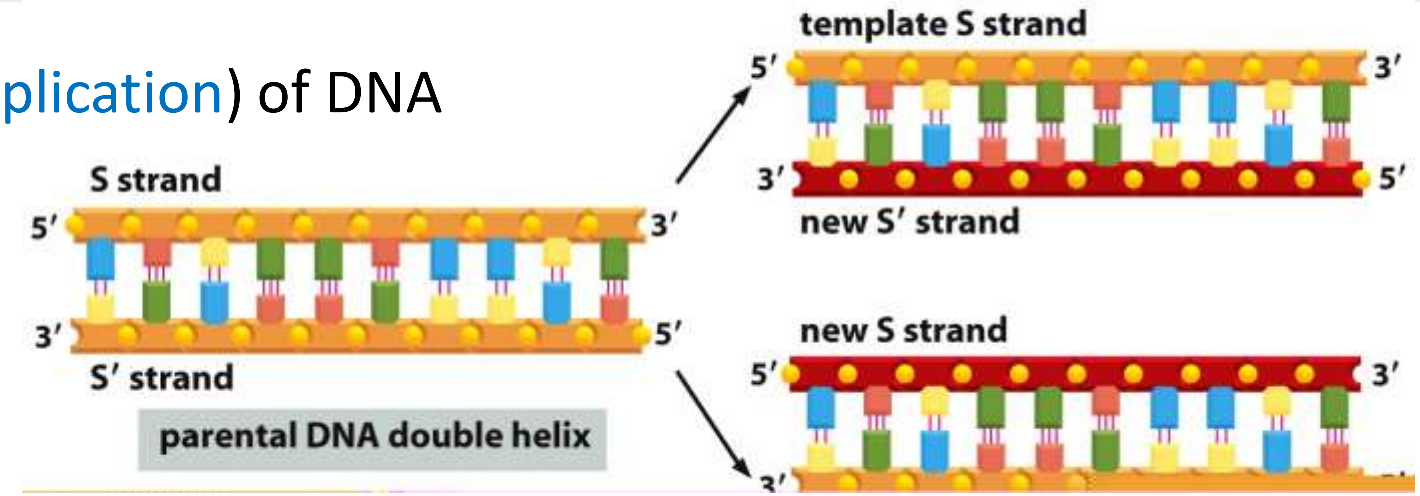
Two strands: anti-parallel

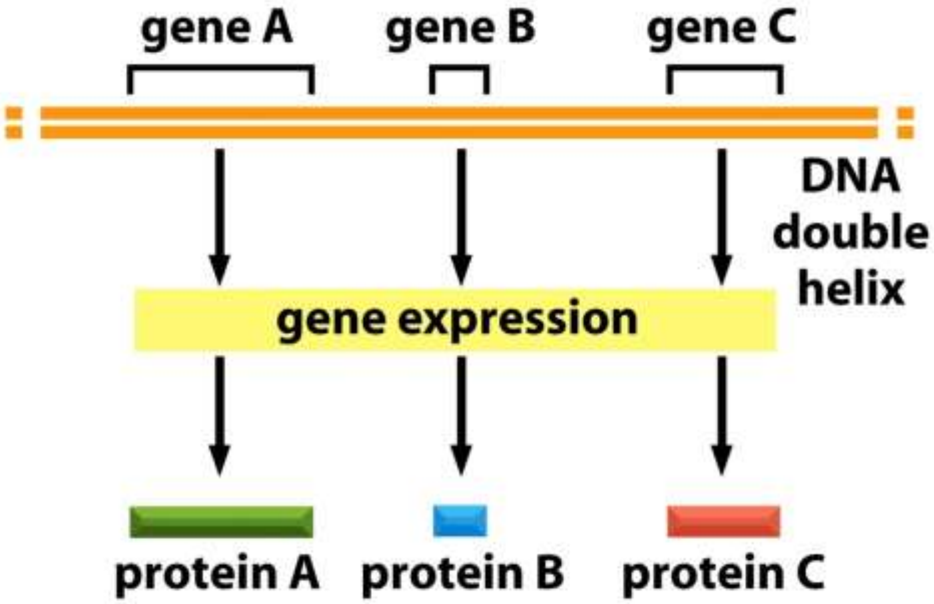




DNA and heredity: DNA is passed to offspring and it carries information to give phenotypes

Duplication (replication) of DNA

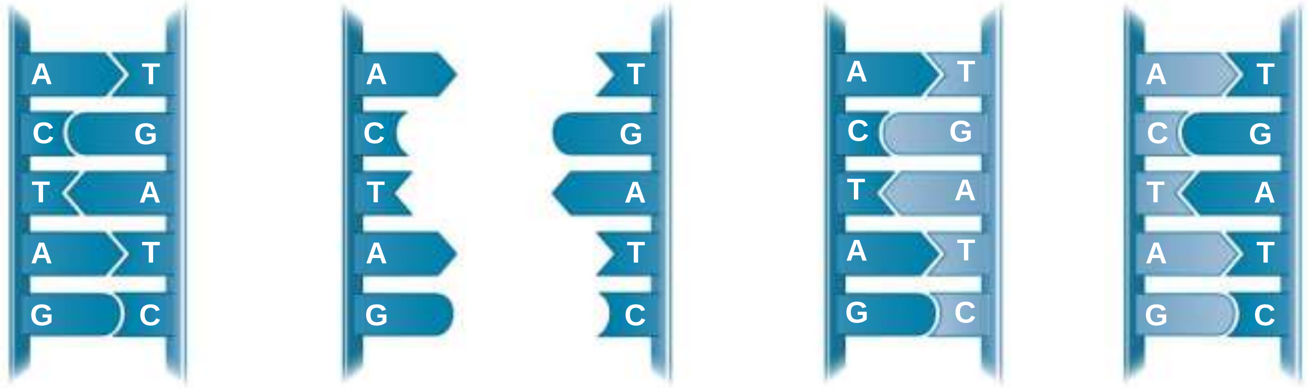




Genes on DNA: You will learn a lot more about this in the next lecture.

The structure of DNA gave clues as to how it might be replicated

Dark blue: parental strand
Light blue: daughter (new) strand



(a) Parental molecule

(b) Separation of parental strands into templates

(c) Formation of new strands complementary to template strands

Replication model envisaged by Watson and Crick
Semi-conservative replication model
Other proposals: conservative model, dispersive model

Meselson and Stahl: experimental evidence for the model of DNA replication

Matt Meselson and Frank Stahl

www.netxplica.com/loja/slides/biologia.11/PPT_BIO11_01/index.html

Designed an innovative experiment

Question: does DNA replication follow a semi-conservative mechanism?

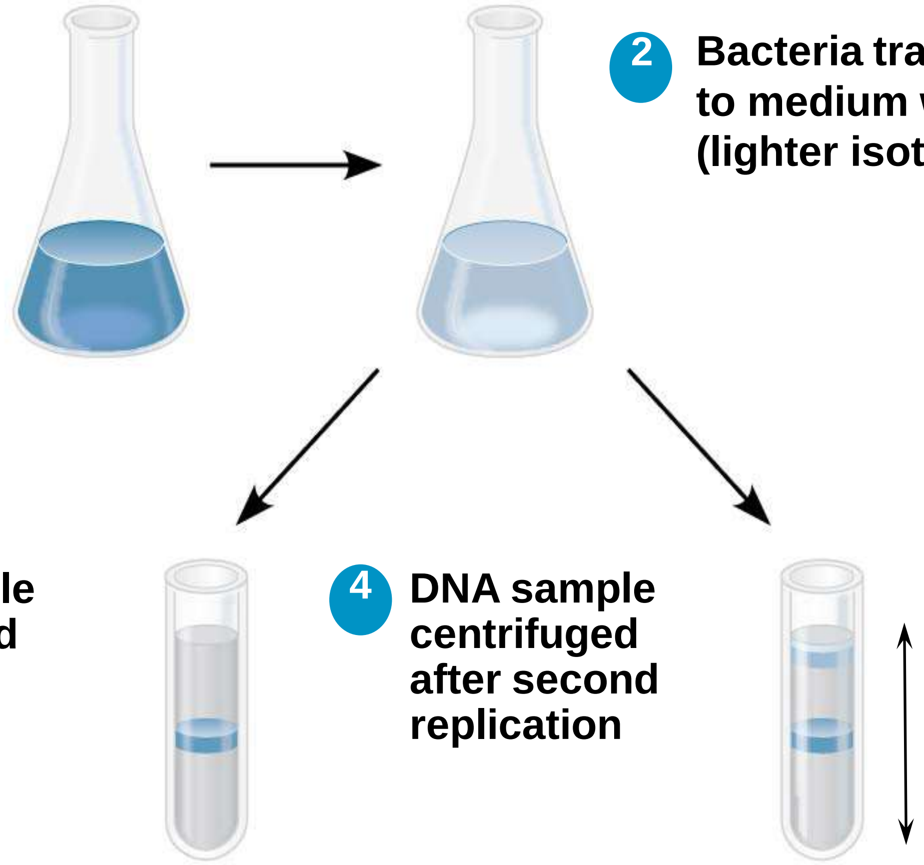
Novelty: exploiting the availability of a heavy isotope of nitrogen

Meselson and Stahl experiment: Design

Experiment

1. Bacteria cultured in medium with ^{15}N (heavy isotope)

2. Bacteria transferred to medium with ^{14}N (lighter isotope)



Results

3. DNA sample centrifuged after first replication

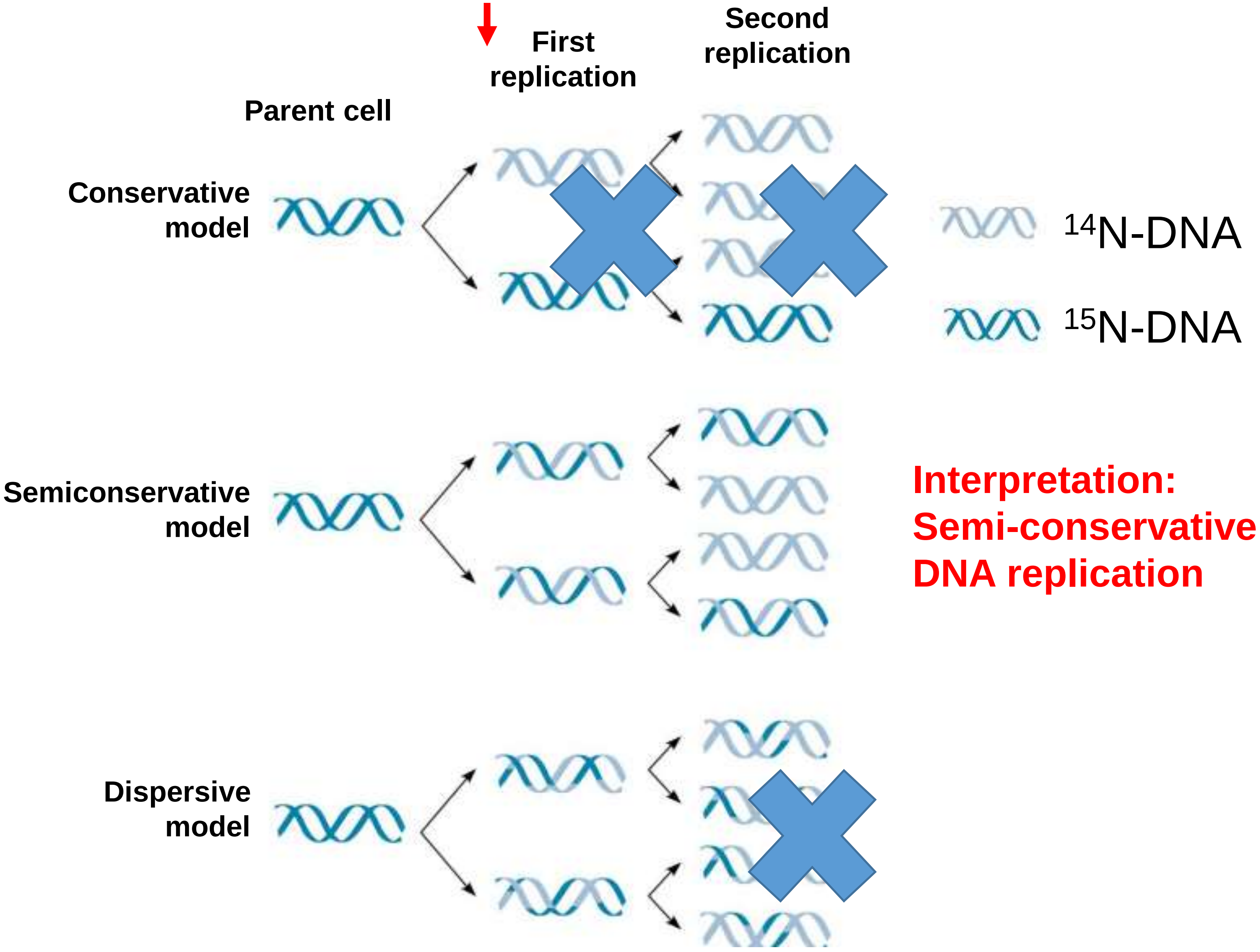
4. DNA sample centrifuged after second replication

Less dense

More dense

Meselson and Stahl experiment: Expectations and Results

Transfer to medium containing ^{14}N at this point

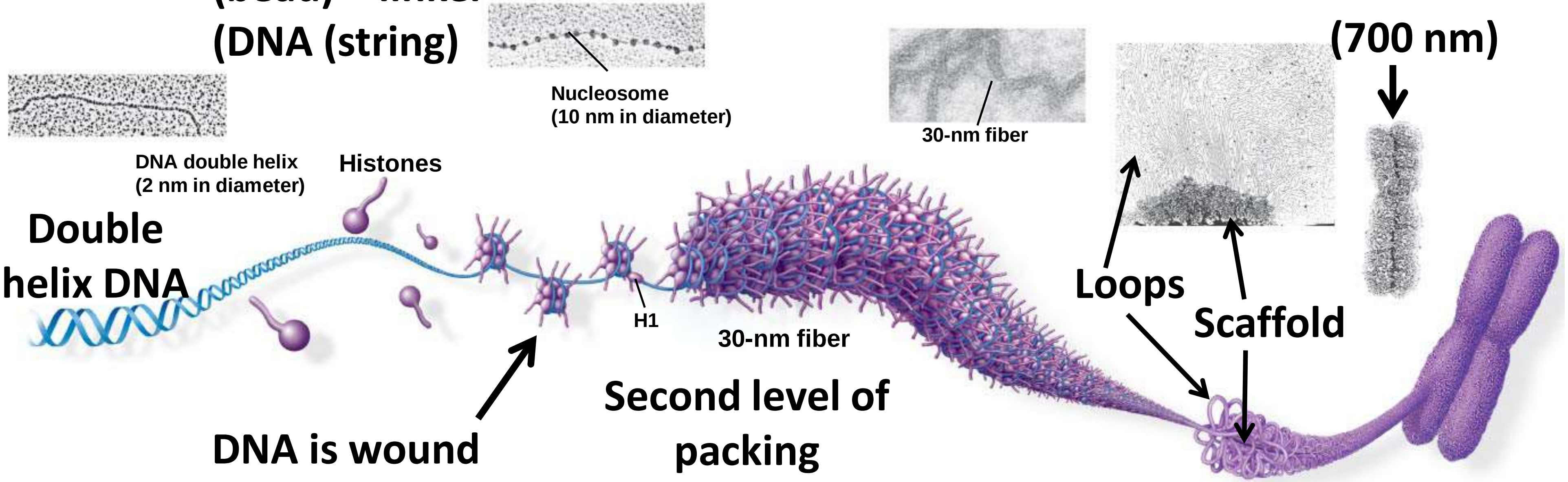


Interpretation: Semi-conservative DNA replication

The DNA polymer is long and needs to be compacted

Nucleosome (bead) + linker (DNA (string)

Two chromatids (700 nm)



Double helix DNA

Loops

Scaffold

DNA is wound around histones

Second level of packing (involves histone tails)

Looped domains (300 nm fibers)

Metaphase (replicated) chromosome (1400 nm)

Human chromosomes: end to end they are ~ 2 meters long

3 billion base pairs with 0.34 nm between each base = 1 meter per single genome (2 copies of each chromosome)

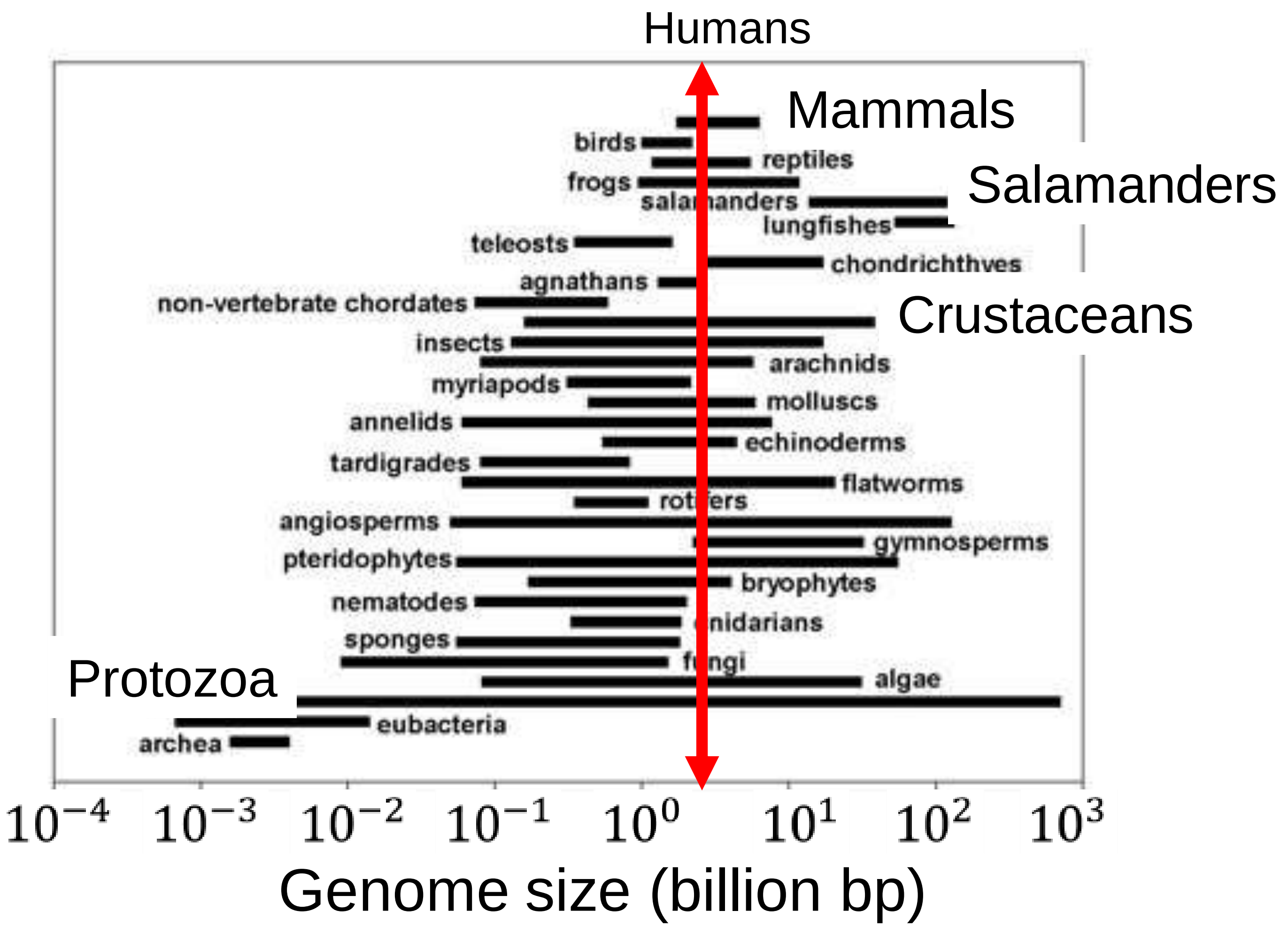
Eukaryotic genomes are organized into chromosomes

Humans have 23 pairs of chromosomes: 22 pairs of autosomes and one pair of sex chromosomes (X and Y) [similar to Mendel's pea plants]

The image below shows a spread of human chromosomes, each 'painted' with a different color; on the right these are organized into a karyotype

Karyotypes can tell us about diseases such as cancer (chromosome aberrations/translocations), disorders such as Down's syndrome (Trisomy 21) and sex determination (XX vs XY)

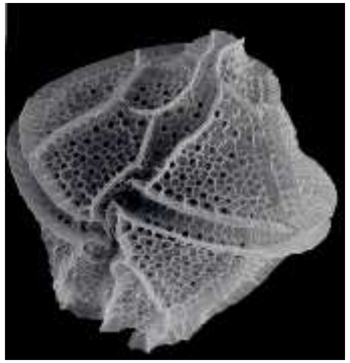
Do humans have the largest genome?



Does genome size correlate with “complexity”?

Human
3 billion bp
(base pairs)

Onion
~16 billion bp



An alga
~98 billion bp

Marbled lungfish

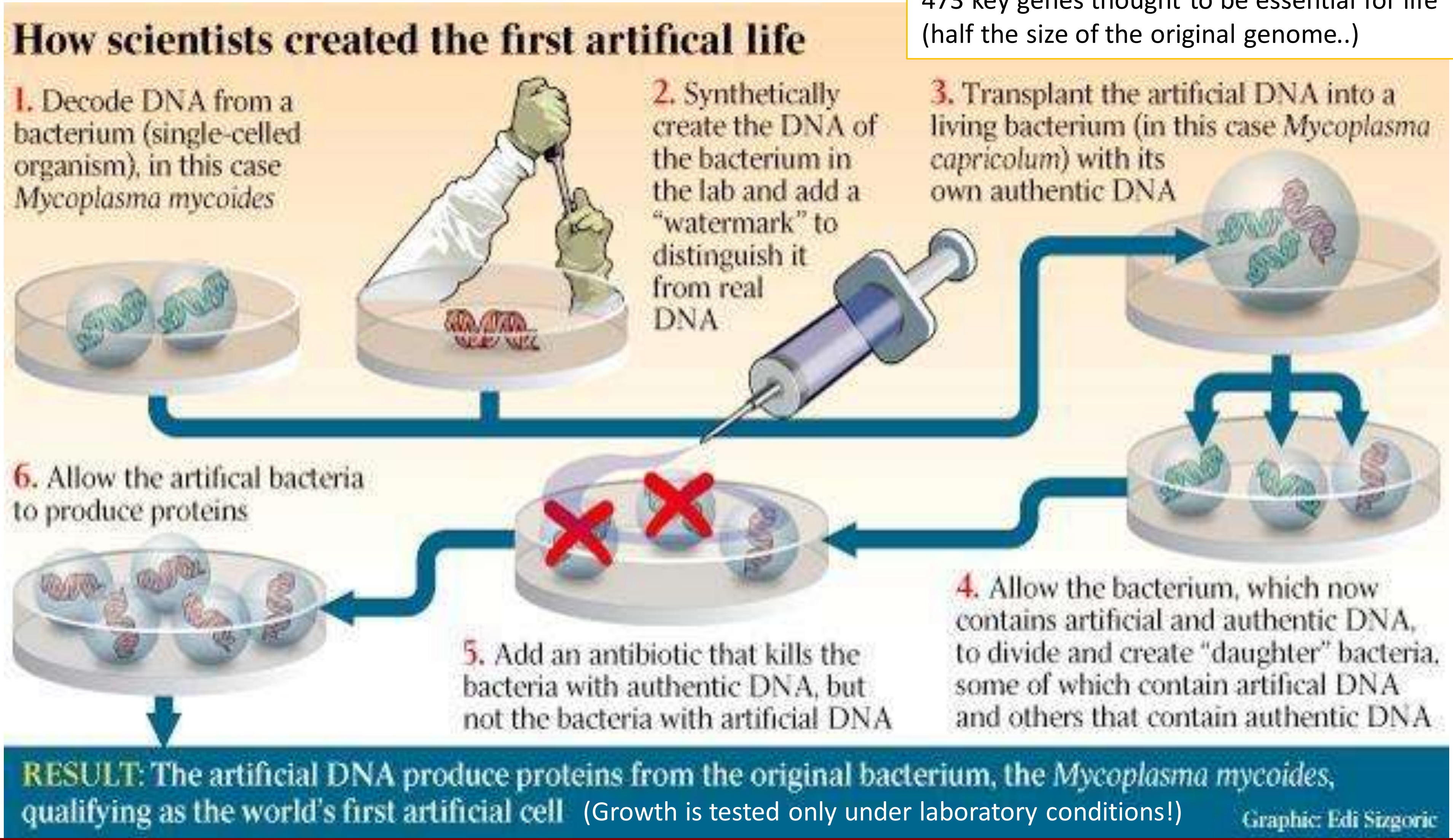
~130 billion bp

Onion: genome size is for *Allium cepa*; may be different for other onion varieties
Can. J. Genet. Cytol. (1983) 25:554 (PMID 6671147)

Can you make an organism with a synthetic instruction manual (genome)?

Science (2016) 351:1414

The synthetic genome in each cell contained only 473 key genes thought to be essential for life (half the size of the original genome..)



(Growth is tested only under laboratory conditions!)