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Molecular Biology 585.407

DNA, Chromosomes, and the Nucleus,” Part 3



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Information: DNA, Chromosomes, and the Nucleus Part 3



DNA in the Genome

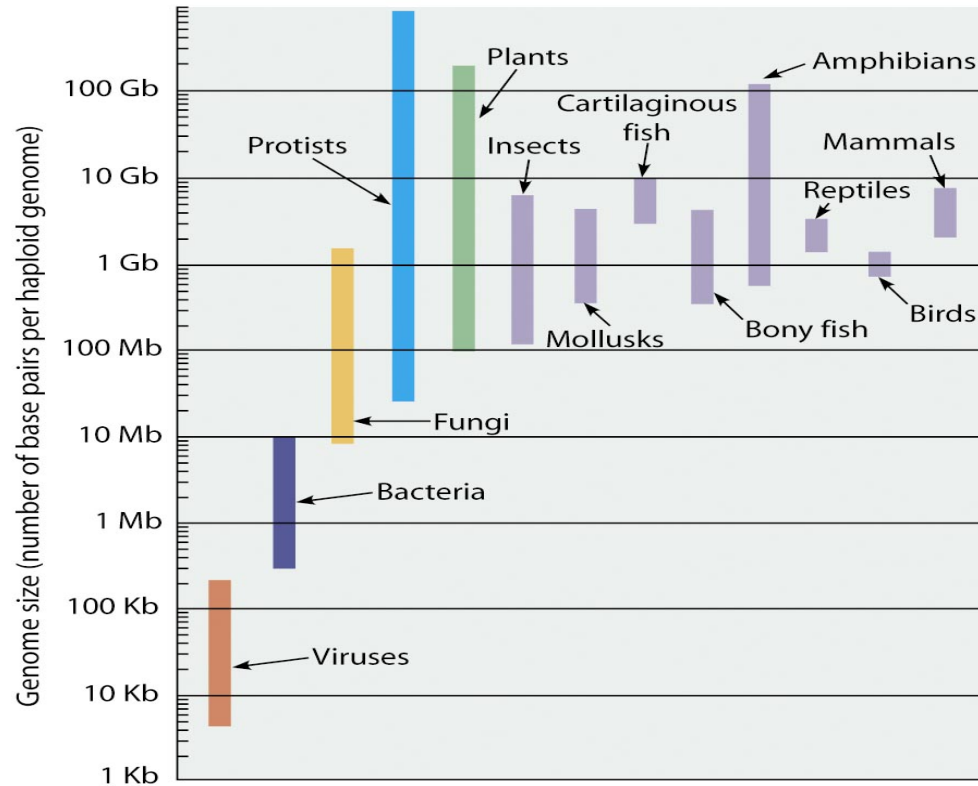
- The **genome** of an organism or virus consists of the DNA (or for some viruses, RNA) that contains one complete copy of **all the** genetic information of that organism or virus.
- For many **viruses** and **prokaryotes**, the genome resides in a single DNA molecule.
- **Eukaryotic** cells have a **nuclear** genome, a **mitochondrial** genome, and, in the case of plants and algae, a **chloroplast** genome as well.
- The nuclear genome generally consists of multiple DNA molecules dispersed among a haploid set of chromosomes.
- Sperm and egg cells each have a haploid set of chromosomes.



DNA Packaging

- The typical *E. coli* cell measures about 1 microns in diameter and 2 microns in length, yet it must accommodate a (circular) DNA molecule with a length of about 1600 microns.
- A human cell of average size contains enough DNA to wrap around the cell more than 15,000 times.
- DNA must be efficiently packaged into cells and yet remain accessible to the cellular machinery for both DNA replication and the transcription.





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Figure 18-11

Genome size

- Genome size increases with complexity of the organism.
- There are great variations in genome size among eukaryotic species that do not seem to correlate with any known differences in organismal complexity.
- Trillium is a member of the lily family, with no obvious need for exceptional amounts of genetic information. Its genome size is more than **20 x** that of peas and **30 x** that of humans. We have no idea what it does with all that DNA.
- Genome size is less important than the number and identity of functional genes.



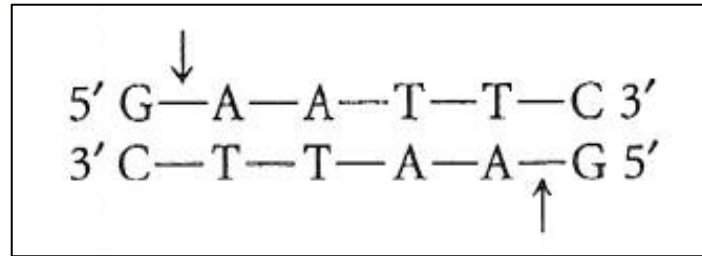
Restriction enzymes cleave DNA molecules at specific sites

- Most DNA molecules are far too large to be studied intact. Until the early 1970s, DNA was the most difficult biological molecule to analyze biochemically.
- Eukaryotic DNA seemed especially intimidating, given the size of most eukaryotic genomes, and no method was known for cutting DNA at specific sites to yield reproducible fragments.



Restriction enzymes

- **Restriction enzymes** have made it possible to cut DNA molecules at specific sites.
- Restriction enzyme generates a specific set of DNA pieces called **restriction fragments**. Each restriction enzyme cleaves double stranded DNA only in places where it encounters a specific recognition sequence, **called a restriction site**.



Restriction enzymes

- Arrows indicate where EcoRI cuts the DNA.
- Restriction enzymes cleave DNA into fragments ranging from a few hundred to a few thousand base pairs in length.
- Fragments are more amenable to further manipulation than the enormously long DNA molecules.



Table 18B-1

Some Common Restriction Enzymes and Their Recognition Sequences

Enzyme	Source Organism	Recognition Sequence*
<i>Bam</i> HI	<i>Bacillus amyloliquefaciens</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ G—G—A—T—C—C } 3' \\ 3' \text{ C—C—T—A—G—G } 5' \\ \uparrow \end{array} $
<i>Eco</i> RI	<i>Escherichia coli</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ G—A—A—T—T—C } 3' \\ 3' \text{ C—T—T—A—A—G } 5' \\ \uparrow \end{array} $
<i>Hae</i> III	<i>Hemophilus aegyptius</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ G—G—C—C } 3' \\ 3' \text{ C—C—G—G } 5' \\ \uparrow \end{array} $
<i>Hind</i> III	<i>Hemophilus influenzae</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ A—A—G—C—T—T } 3' \\ 3' \text{ T—T—C—G—A—A } 5' \\ \updownarrow \end{array} $
<i>Pst</i> I	<i>Providencia stuartii</i> 164	$ \begin{array}{c} \downarrow \\ 5' \text{ C—T—G—C—A—G } 3' \\ 3' \text{ G—A—C—G—T—C } 5' \\ \uparrow \end{array} $
<i>Pvu</i> I	<i>Proteus vulgaris</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ C—G—A—T—C—G } 3' \\ 3' \text{ G—C—T—A—G—C } 5' \\ \uparrow \end{array} $
<i>Pvu</i> II	<i>Proteus vulgaris</i>	$ \begin{array}{c} \downarrow \\ 5' \text{ C—A—G—C—T—G } 3' \\ 3' \text{ G—T—C—G—A—C } 5' \\ \uparrow \end{array} $
<i>Sal</i> I	<i>Streptomyces albus</i> G	$ \begin{array}{c} \downarrow \\ 5' \text{ C—T—C—G—A—C } 3' \\ 3' \text{ C—A—G—C—T—G } 5' \\ \uparrow \end{array} $

*The arrows within the recognition sequence indicate the points at which each restriction enzyme cuts the two strands of the DNA molecule.

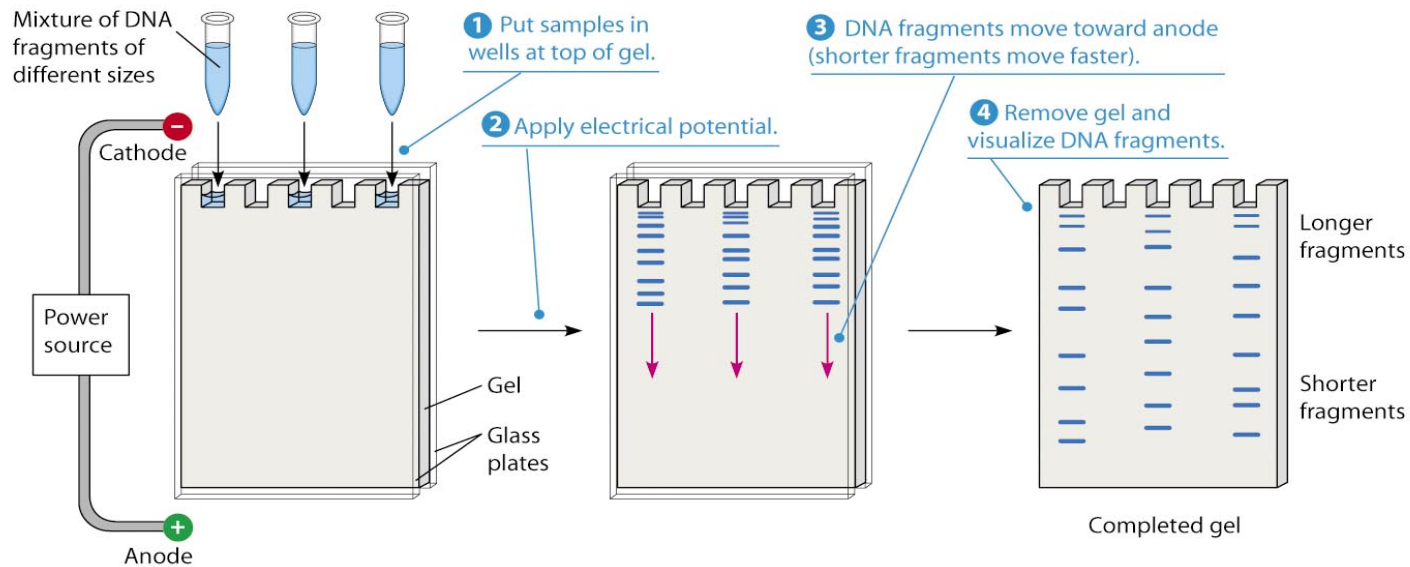


Table 18B-1

Separation of restriction fragments by gel electrophoresis

- To determine the number and lengths of DNA fragments and to isolate individual fragments for further study, one must be able to separate the fragments from each other.
- **Gel electrophoresis** is the technique of choice to determine the number and lengths of DNA fragments and to isolate individual fragments for further study.





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The nucleus

- The **nucleus** is the site within the eukaryotic cell where the chromosomes are localized and replicated and where the DNA they contain is transcribed.
- The nucleus is both the **repository** of most of the cell's genetic information and the **control center** for expression of information.

