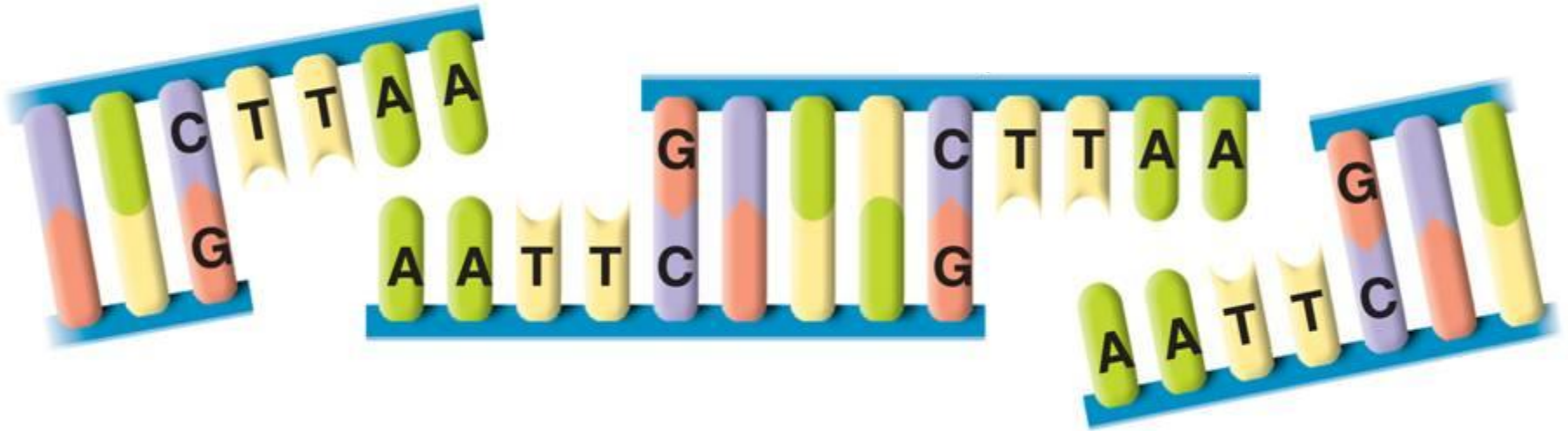


# 13-2 Manipulating DNA



# The Tools of Molecular Biology



**How do scientists make changes to DNA?**

**Scientists use their knowledge of the structure of DNA and its chemical properties to study and change DNA molecules.**

Scientists use different techniques to:

- extract DNA from cells
  - cut DNA into smaller pieces
  - identify the sequence of bases in a DNA molecule
  - make unlimited copies of DNA
- 
- In **genetic engineering**, biologists make changes in the DNA code of a living organism.

## DNA Extraction

DNA can be extracted from most cells by a simple chemical procedure.

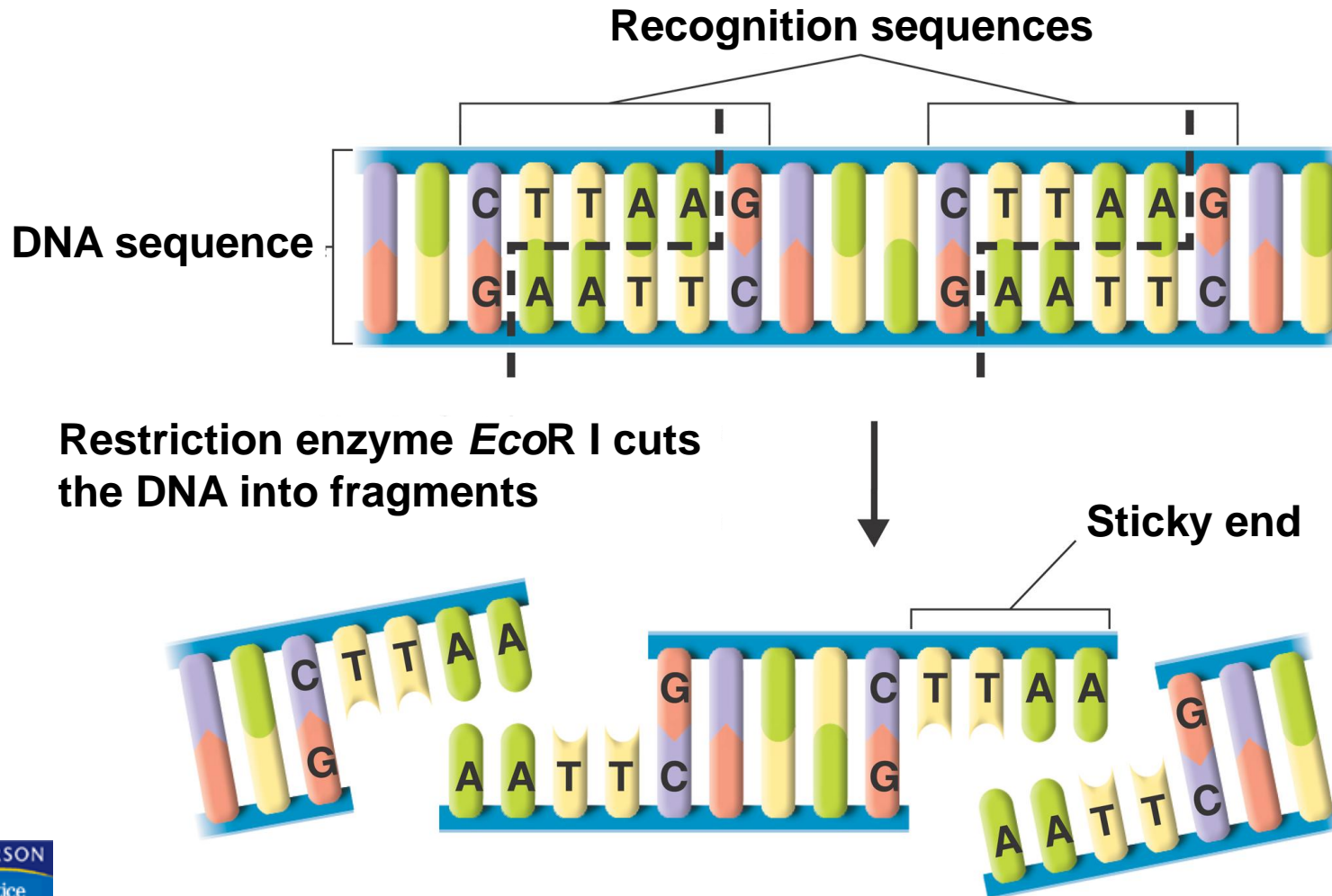
The cells are opened and the DNA is separated from the other cell parts.

## Cutting DNA

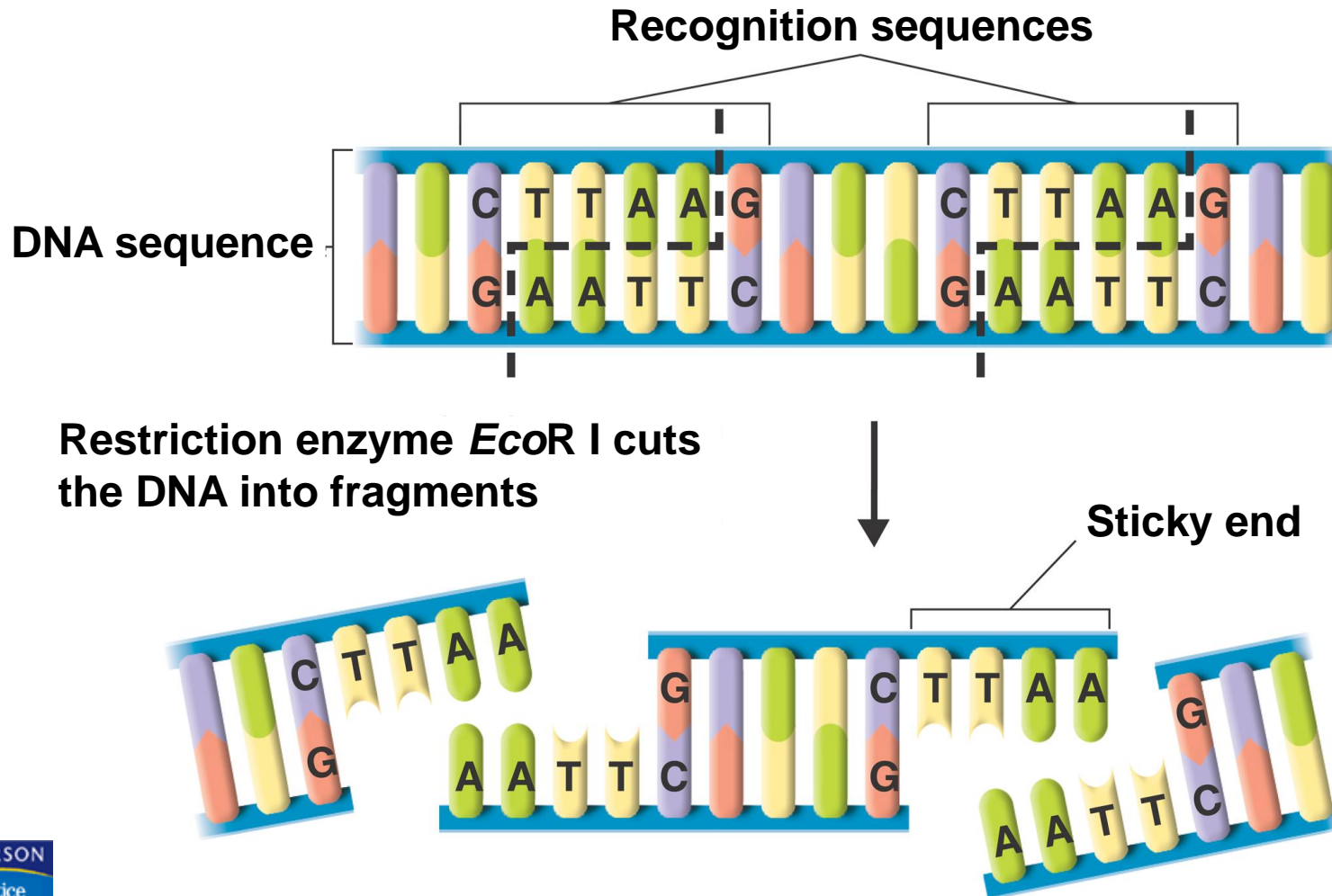
Most DNA molecules are too large to be analyzed, so biologists cut them into smaller fragments using restriction enzymes.



Each **restriction enzyme** cuts DNA at a specific sequence of nucleotides.



A restriction enzyme will cut a DNA sequence only if it matches the sequence precisely.



## Separating DNA

In **gel electrophoresis**, DNA fragments are placed at one end of a porous gel, and an electric voltage is applied to the gel.

When the power is turned on, the negatively charged DNA molecules move toward the positive end of the gel.

Gel electrophoresis can be used to compare the genomes of different organisms or different individuals.

It can also be used to locate and identify one particular gene in an individual's genome.



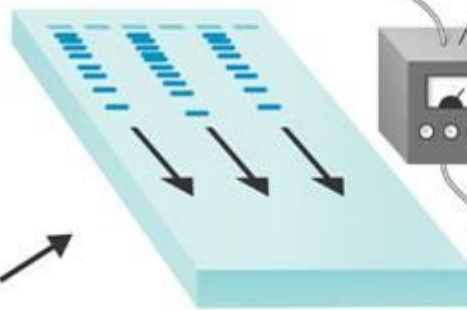
**DNA plus restriction enzyme**



**Mixture of DNA fragments**



**Gel**



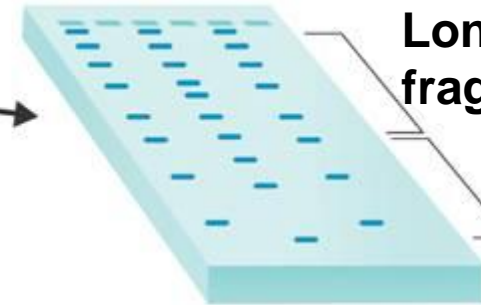
**Power source**



+

**Longer fragments**

**Shorter fragments**

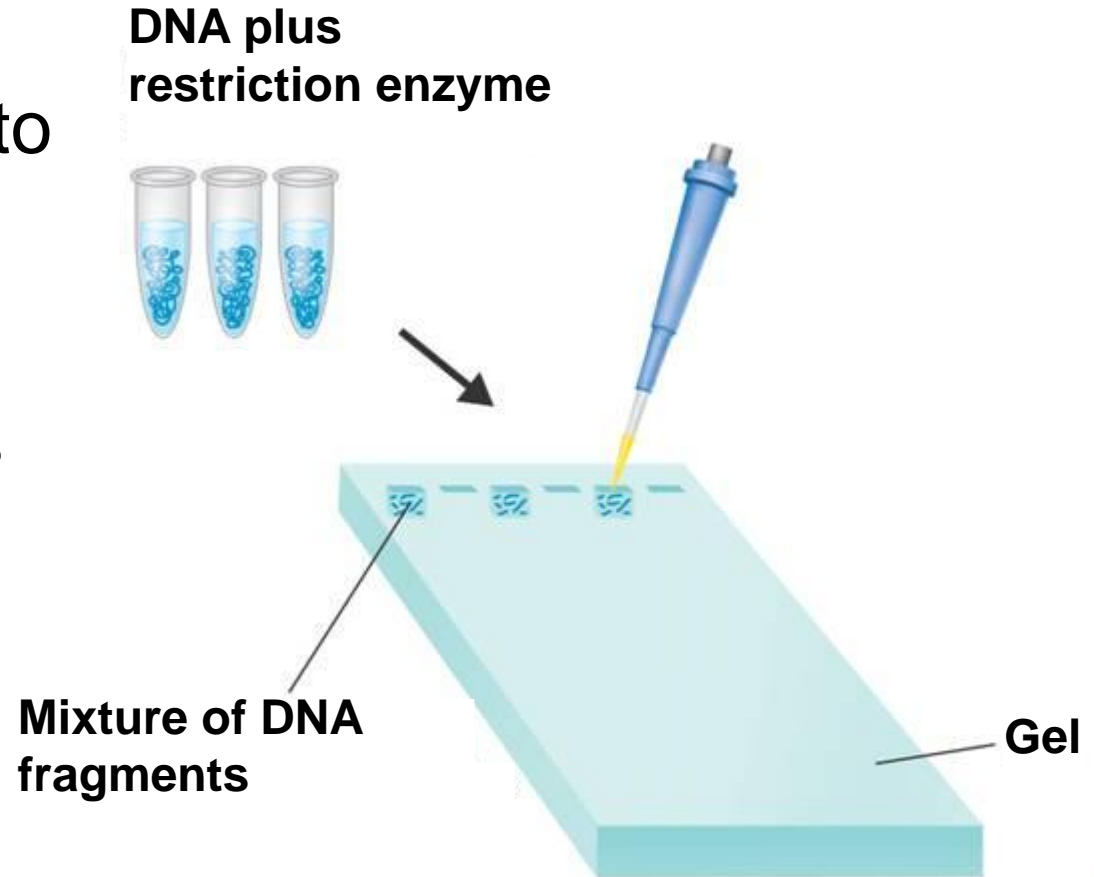


## **Gel Electrophoresis**



First, restriction enzymes cut DNA into fragments.

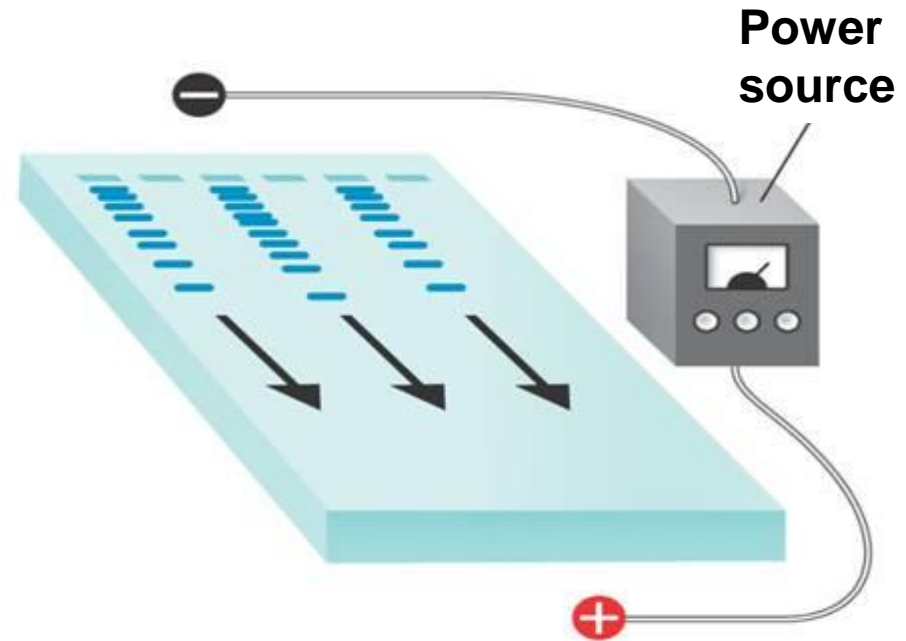
The DNA fragments are poured into wells on a gel.



## Gel Electrophoresis

An electric voltage is applied to the gel. This moves the DNA fragments across the gel.

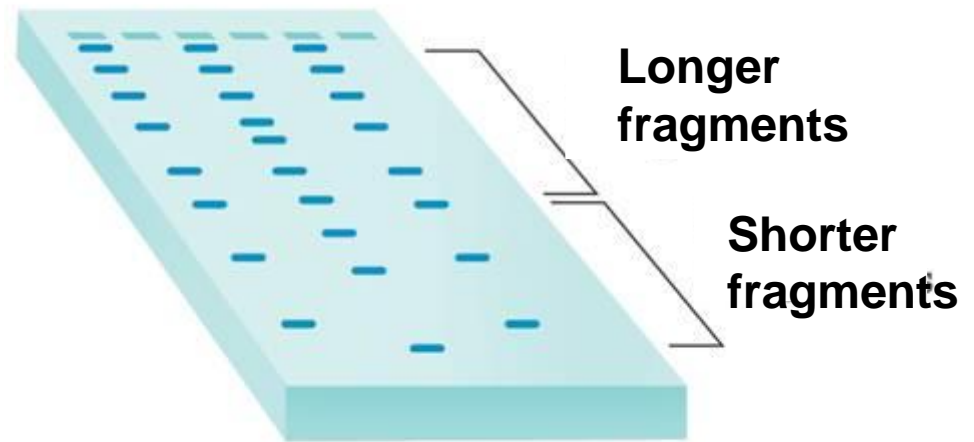
The smaller the DNA fragment, the faster and farther it will move across the gel.



### Gel Electrophoresis

Based on size, the DNA fragments make a pattern of bands on the gel.

These bands can then be compared with other samples of DNA.



## Gel Electrophoresis

## Using the DNA Sequence

Knowing the sequence of an organism's DNA allows researchers to study specific genes, to compare them with the genes of other organisms, and to try to discover the functions of different genes and gene combinations.



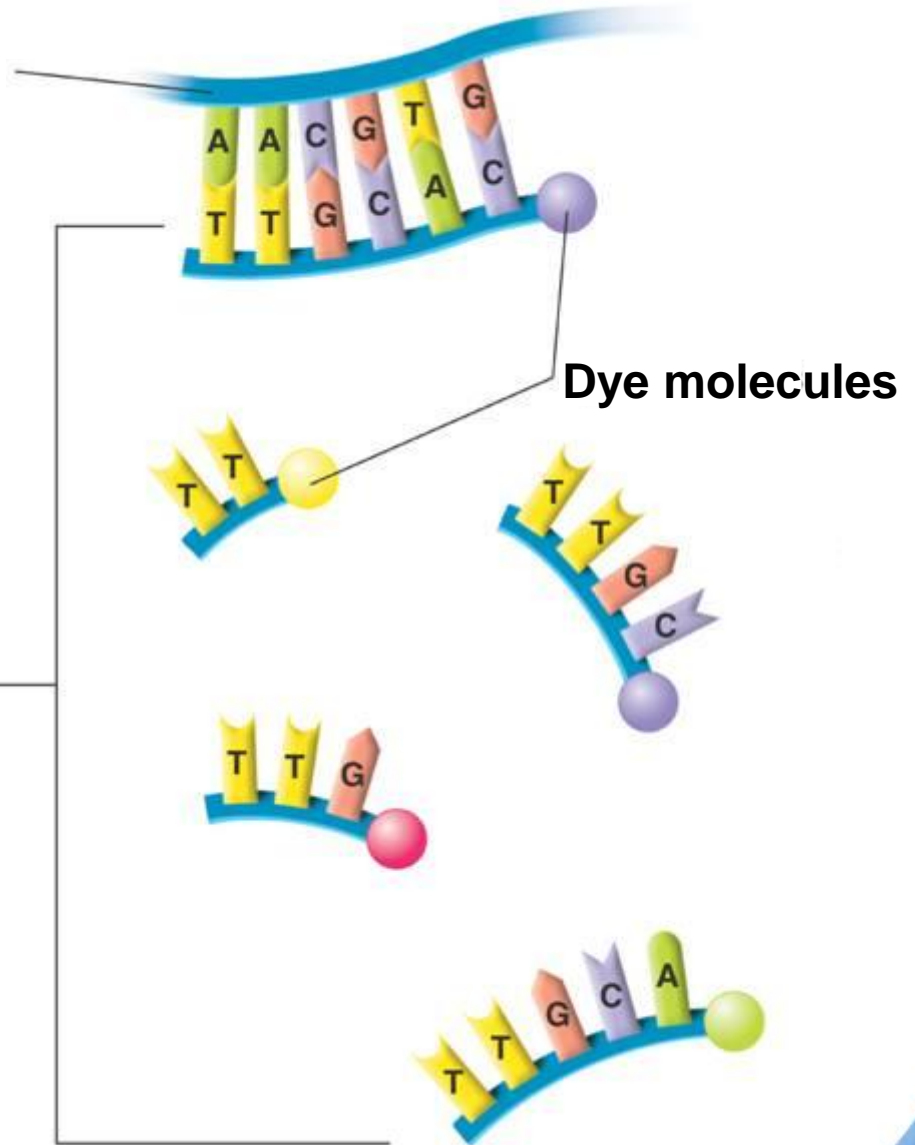
## Reading the Sequence

In DNA sequencing, a complementary DNA strand is made using a small proportion of fluorescently labeled nucleotides.

# DNA Sequencing

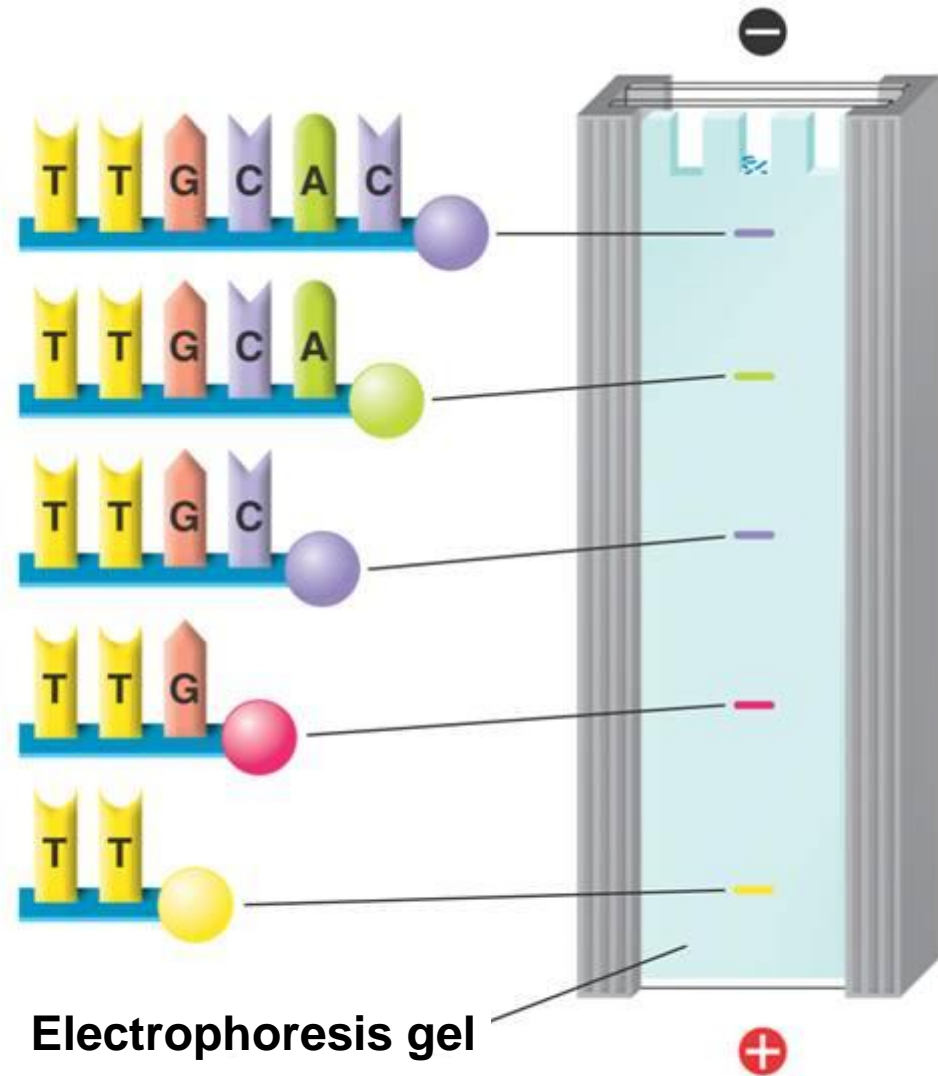
DNA strand with unknown base sequence

DNA fragments synthesized using unknown strand as a template



Base sequence  
as “read” from  
the order of the  
dye bands on  
the gel from  
bottom to top:

**T G C A C**



Each time a labeled nucleotide is added, it stops the process of replication, producing a short color-coded DNA fragment.

When the mixture of fragments is separated on a gel, the DNA sequence can be read.

### Cutting and Pasting

Short sequences of DNA can be assembled using DNA synthesizers.

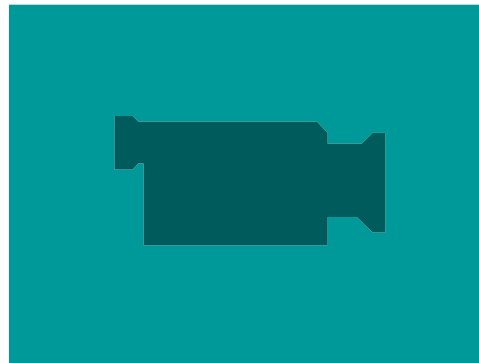
“Synthetic” sequences can be joined to “natural” sequences using enzymes that splice DNA together.



These special enzymes are known as **restriction endonucleases**

These enzymes also make it possible to take a gene from one organism and attach it to the DNA of another organism.

Such DNA molecules are sometimes called **recombinant DNA**.



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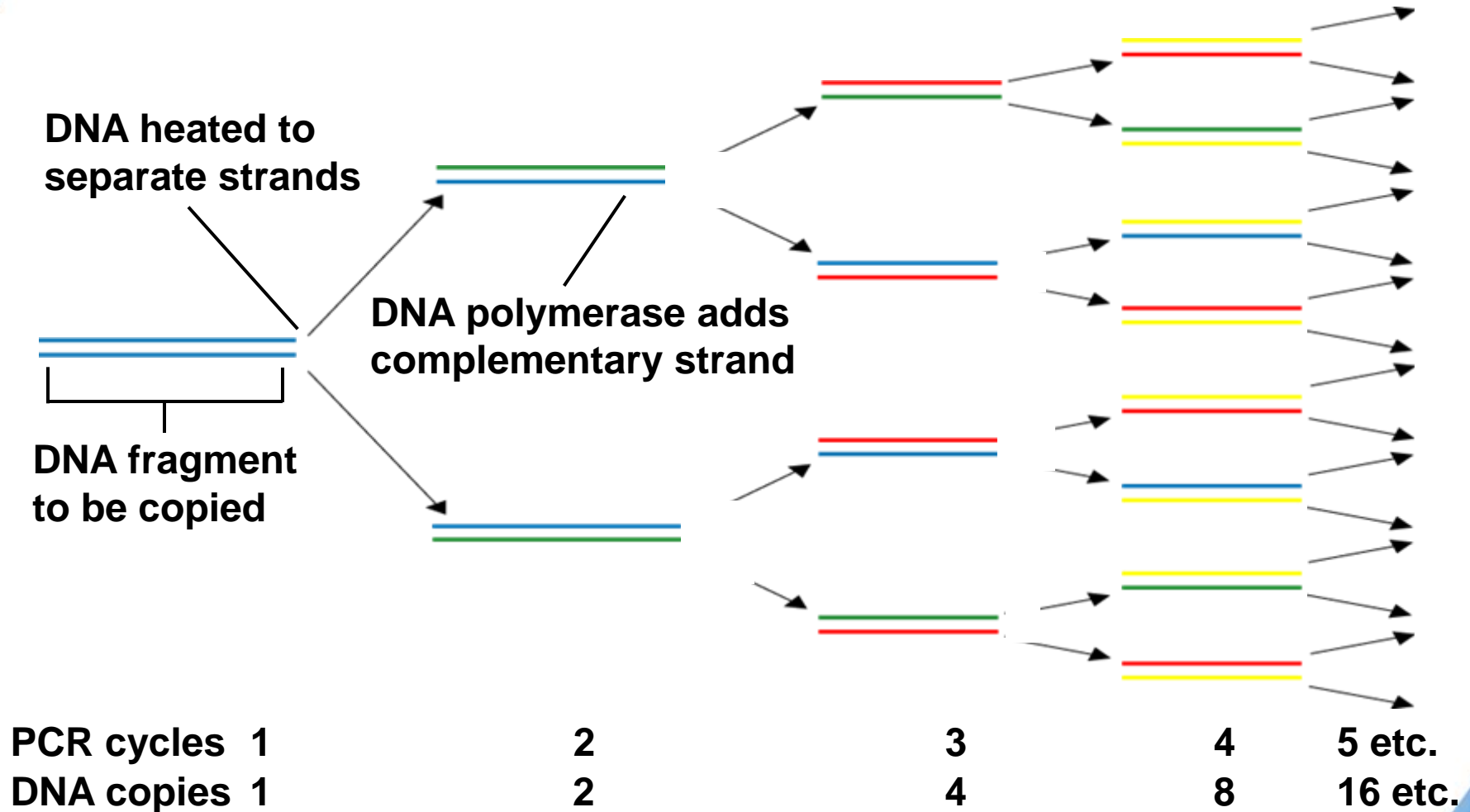
## Making Copies

- **Polymerase chain reaction (PCR)** is a technique that allows biologists to make copies of genes.
- A biologist adds short pieces of DNA that are complementary to portions of the sequence.

DNA is heated to separate its two strands, then cooled to allow the primers to bind to single-stranded DNA.

DNA polymerase starts making copies of the region between the primers.

# Polymerase Chain Reaction (PCR)



## 13-2 Section QUIZ

Continue to:

**Section QUIZ**

- or -

Click to Launch:



## 13-2 Section QUIZ

1 Restriction enzymes are used to

a. extract DNA.

b. cut DNA.

c. separate DNA.

d. replicate DNA.

## 13-2 Section QUIZ

**2** During gel electrophoresis, the smaller the DNA fragment is, the

a. more slowly it moves.

b. heavier it is.

c. more quickly it moves.

d. darker it stains.

## 13-2 Section QUIZ

**3** The DNA polymerase enzyme Kary Mullis found in bacteria living in the hot springs of Yellowstone National Park illustrates

- a. genetic engineering.
- b. the importance of biodiversity to biotechnology.
- c. the polymerase chain reaction.
- d. selective breeding.



## 13-2 Section QUIZ

- 4 A particular restriction enzyme is used to
- a. cut up DNA in random locations.
  - b. cut DNA at a specific nucleotide sequence.
  - c. extract DNA from cells.
  - d. separate negatively charged DNA molecules.

**5** During gel electrophoresis, DNA fragments become separated because

- a. multiple copies of DNA are made.
- b. recombinant DNA is formed.
- c. DNA molecules are negatively charged.
- d. smaller DNA molecules move faster than larger fragments.