

CLUTCH

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CONCEPT: GENETIC CLONING

• The ability to _____ genes is the basis for all modern genetic advancements

1. Amplifying the DNA of interest to obtain many copies

- **Polymerase Chain Reaction (PCR)** is the main method used to amplify DNA

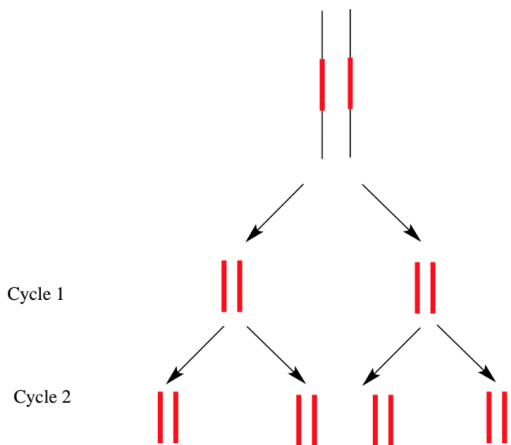
- Heat the DNA strands to a high temperature to separate the double helix

- Lower the temperature to **anneal** primers to the DNA sequence of interest

- Raise the temperature to allow the polymerase to find the primers and replicate the DNA

- Can be used to create and amplify cDNA from RNA

EXAMPLE:

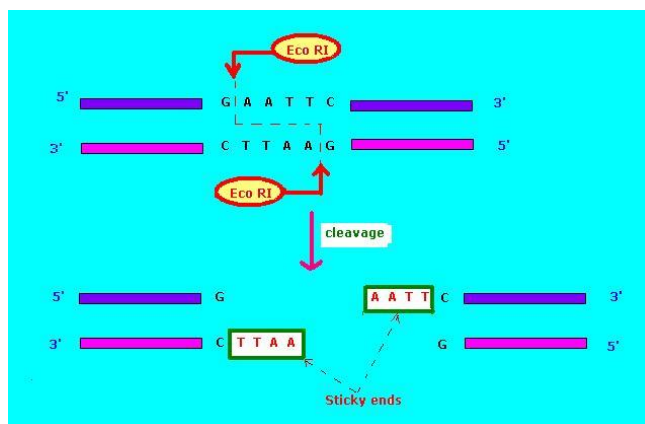


2. Cutting the DNA of interest into small _____

- **Restriction enzymes** are proteins used to chop DNA at specific sequences

- Can create **blunt ends** with no sequence overhangs or **sticky ends** with sequence overhangs

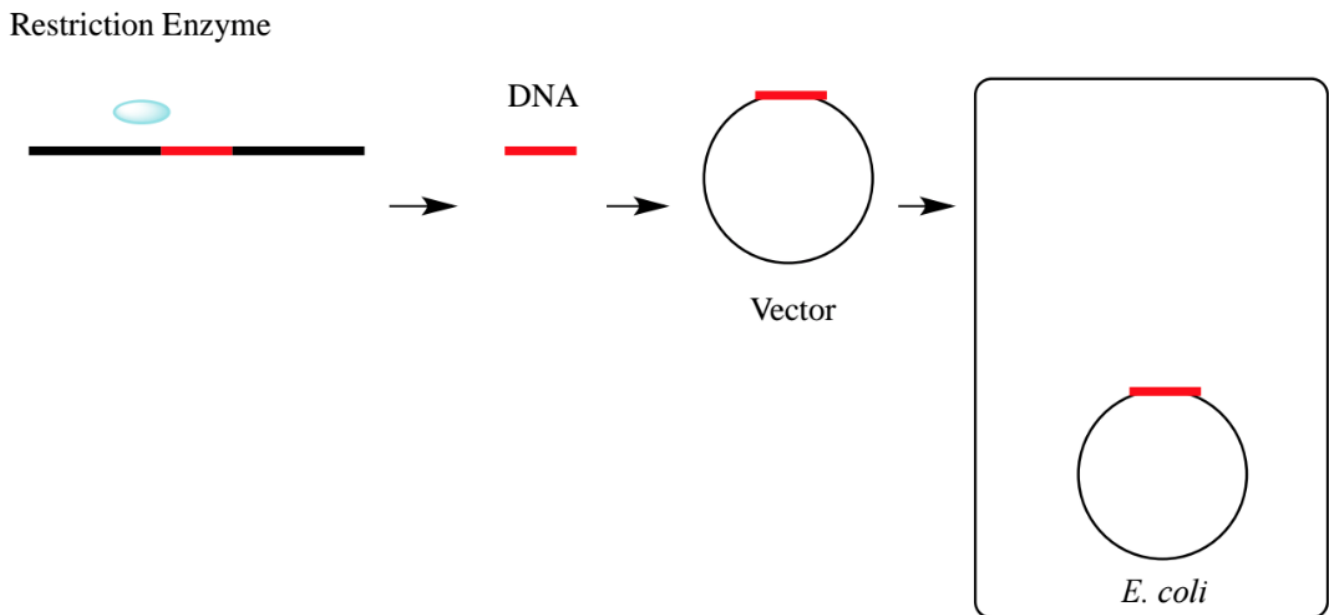
EXAMPLE:



3. Pasting the DNA into a _____ (called **recombinant DNA**)

- **DNA ligase** is the enzyme that seals the DNA of interest into a plasmid
 - Restriction enzymes create *sticky ends* that are overhangs to which the seal is made
- A **vector** is a bacterial plasmid in which the DNA sequence of interest is placed
 - For large DNA inserts, **BACs** or **YACs** are used (bacterial or yeast artificial chromosomes)
 - **T_i plasmids** are often used for plant cells

EXAMPLE:



4. The recombinant DNA is then placed in bacteria (*E. coli*) or another organism (ex: virus)

- A **transgenic organism** is created by adding a *transgene*
 - A **transgene** is a new gene introduced into an organism

PRACTICE:

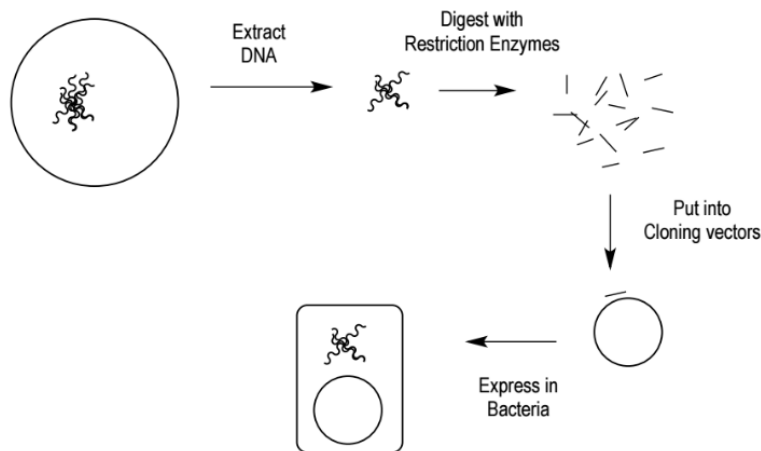
1. The purpose of polymerase chain reaction is to do what?
 - a. Create RNA templates
 - b. Create fluorescent probes
 - c. Amplify a short DNA sequence
 - d. Isolate proteins

2. Which of the following lists the steps of genetic cloning in the proper order?
 - a. PCR → Restriction Enzyme Cutting → Ligate into a Vector → Placed into Organism
 - b. Restriction Enzyme Cutting → PCR → Ligate into a Vector → Placed into Organism
 - c. Ligate into a Vector → PCR → Restriction Enzyme Cutting → Placed into Organism
 - d. PCR → Ligate into a Vector → Restriction Enzyme Cutting → Placed into Organism

CONCEPT: METHODS FOR ANALYZING DNA AND RNA

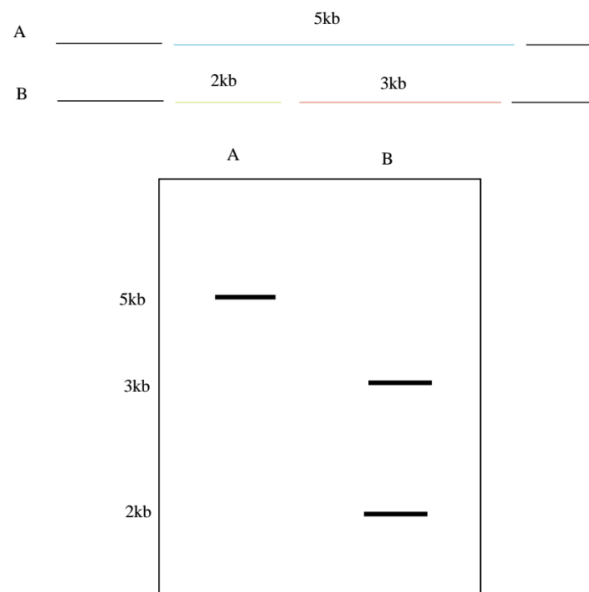
- Geneticists use a variety of _____ to study DNA and RNA
 - **Libraries** are collections of bacterial clones with genetic sequences in them
 - **Genomic libraries** is a collection of small fragment of the entire genome
 - **cDNA libraries** are collections of small fragments of complementary DNA (represents mRNA)

EXAMPLE: Genomic Libraries



- Blotting is a method used to separate and _____ DNA or RNA
 - **Southern blots** separate genomic DNA
 - **Northern blots** separate and visualize RNA sequences

EXAMPLE:



PRACTICE:

1. In terms of molecular biology, what is a library?
 - a. A place where molecular biology textbooks are stored
 - b. A collection of clones containing short genetic sequences
 - c. A collection of clones containing a large collection of proteins
 - d. A collection of clones with different mutant phenotypes

2. If you had a sample of RNA to analyze, which of the following techniques would you most likely use?
 - a. Southern Blots
 - b. SDS-PAGE Blots
 - c. Northern Blots
 - d. Western Blots