

NCERT Solutions for Class-XII Biology

Chapter -9

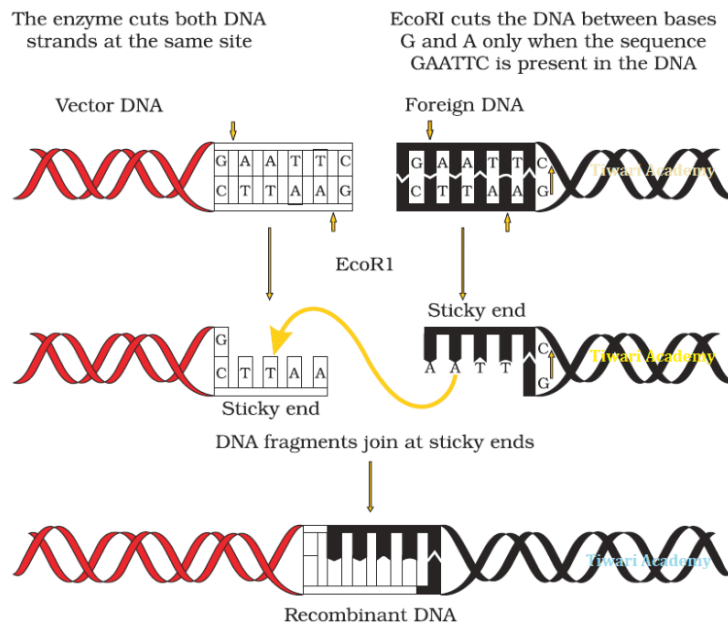
NCERT Biology Class 12

1. Can you list 10 recombinant proteins which are used in medical practice? Find out where they are used as therapeutics (use the internet).
1. The ten recombinant proteins which are used in medical practice are as follows:

Recombinant proteins	Uses in medical practice
<ul style="list-style-type: none">• Insulin• Tissue plasminogen activator• HBsAg protein• Human recombinant growth hormone somatostatin• Blood coagulation factor VIII• Blood coagulation factor IX• Erythropoietin• DNase I• Thyroid stimulating hormone• Monoclonal antibodies	<ul style="list-style-type: none">• For the treatment of Diabetes mellitus.• To treat acute myocardial infarction.• Subunit vaccine against hepatitis B.• To promote growth and treat dwarfism.• To treat haemophilia A.• To treat Haemophilia B.• To treat anaemia.• To treat cystic fibrosis.• To treat recurrent thyroid cancer.• To stimulate the host immune system.

Note: Monoclonal antibodies are recombinant antibodies produced by myeloma B – cells and have unique specificity for antigens used to treat diseases caused by specific antigens by stimulating host immune system against the attack.

2. Make a chart (with diagrammatic representation) showing a restriction enzyme, the substrate DNA on which it acts, the site at which it cuts DNA and the product it produces.
2. Action of restriction enzyme:



Steps in formation of recombinant DNA by action of restriction endonuclease enzyme-EcoRI

3. From what you have learnt, can you tell whether enzymes are bigger or DNA is bigger in molecular size? How did you know?
3. A DNA molecule is always bigger than a protein, in size because a protein is the product of one or more mRNAs whereas these mRNAs are produced from DNA. For example, human DNA houses approximately about 30,000 genes which are used to produce all the different types of proteins required by the cell. Even after this the only transcribed region is the euchromatin region and not the heterochromatic region of the genome. Thus, the DNA molecule is certainly larger than a protein which is only a fraction of the translated DNA.
4. What would be the molar concentration of human DNA in a human cell? Consult your teacher.
4. The molar concentration of human DNA in a human diploid cell is as follows:

$$\Rightarrow \text{Total number of chromosomes} \times 6.023 \times 10^{23}$$

$$\Rightarrow 46 \times 6.23 \times 10^{23} \Rightarrow 2.77 \times 10^{23} \text{ moles}$$
 Hence, the molar concentration of DNA in each diploid cell in humans is 2.77×10^{23} moles.
5. Do eukaryotic cells have restriction endonucleases? Justify your answer.
5. No, eukaryotic cells do not have restriction endonucleases. All restriction endonucleases have been isolated from various strains of bacteria till date. Even the restriction

endonucleases are named according to the genus, species, strains and isolation number of the bacterias. The first letter of the restriction endonuclease is the genus and the last two is the species of the organism. The subscript is first letter the strain of the organism, if already used the next letter is used. The Roman numbers in the end is the isolation number of the enzyme chronologically when the organism has more than one restriction endonuclease.

6. Besides better aeration and mixing properties, what other advantages do stirred tank bioreactors have over shake flasks?
6. The shake flask method is used for a small-scale production of biotechnological products in a laboratory. On the other hand, stirred tank bioreactors are used for a large-scale production of biotechnology products.

Stirred tank bioreactors have several advantages over shake flasks. Some of them are

- Small volumes of culture can be taken out from the reactor for sampling or testing.
- It has a foam breaker for regulating the foam.
- It has a control system that regulates the temperature and pH.

7. Collect 5 examples of palindromic DNA sequences by consulting your teacher. Better try to create a palindromic sequence by following base-pair rules.
7. A palindromic sequence is a sequence of double stranded DNA when read from either side of the double strand it is read the same. For example, “MADAM” that is, when we read it from either side, from forward as well as backward it is read the same. The same happens in case of a DNA double helix only difference is that it is read on two strands and when read from 3’ or 5’ on both strands it reads the same. The five palindromic sequences are:

1. 3’...CGTATACG...5’

5’...GCATATGC...3’

2. 3’...AAGCTT...5’ (restriction site of endonuclease Hind III)

5’...TTCGAA...3’

3. 3’...GAATTC...5’ (restriction site of endonuclease EcoRI)

5’...CTTAAG...3’

4. 3’...AGCT...5’ (restriction site of endonuclease Alu1)

5’...TCGA...3’

5. 3’...TTATTCGAATAA...5’

5’...AATAAGCTTATT...3’

8. Can you recall meiosis and indicate at what stage a recombinant DNA is made?
8. Meiosis is a process that involves the reduction in the amount of genetic material. It is two types, namely meiosis I and meiosis II.

During the pachytene stage of prophase I, crossing over of chromosomes takes place where the exchange of segments between non-sister chromatids of homologous chromosomes takes place. This results in the formation of recombinant DNA.

9. Can you think and answer how a reporter enzyme can be used to monitor transformation of host cells by foreign DNA in addition to a selectable marker?
9. When a reporter enzyme such as the Lac Z gene is present in the genome of a transformed cell it produces the enzyme β -galactosidase. The substrate on which β -galactosidase acts on is lactose. When lactose is present in the culture medium as the carbon source of the bacteria β -galactosidase breaks down lactose to produce the end product X – gal which gives a characteristic blue colour. This blue colour is visible with naked eye and by observing the culture plates it can be identified which of the host cells are transformed. When a gene of interest is incorporated within this Lac Z gene the gene becomes inactivated as it gets disrupted. This is known as Insertional inactivation. When such a plasmid transforms the host bacterial cell it does not develop any colour and thus can be identified. This process of identification is known as the blue–white screening process.
10. Describe briefly the following:
 - A. Origin of replication
 - B. Bioreactors
 - C. Downstream processing

10. (a) **Origin of Replication**

This is a sequence from where replication starts and any piece of DNA when linked to this sequence can be made to replicate within the host cells. Origin of replication is defined as the DNA sequence in a genome from where replication initiates. The initiation of replication can be either uni-directional or bi-directional. A protein complex recognizes the 'on' site, unwinds the two strands, and initiates the copying of the DNA.

(b) **Bioreactors**

Bioreactors are large vessels used for the large-scale production of biotechnology products from raw materials. They provide optimal conditions to obtain the desired product by providing the optimum temperature, pH, vitamin, oxygen, etc. Bioreactors have an oxygen delivery system, a foam control system, a PH, a temperature control system, and a sampling port to obtain a small volume of culture for sampling. The most commonly used bioreactor is the stirring type.

(c) **Downstream Processing**

Downstream processing is a method of separation and purification of foreign gene products after the completion of the biosynthetic stage. The product is subjected to various processes in order to separate and purify the product. After downstream

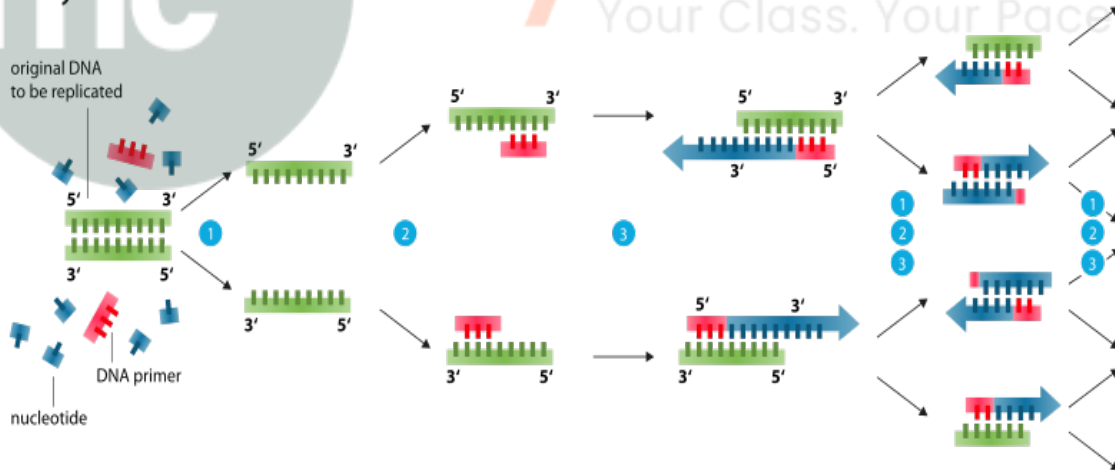
processing, the product is formulated and is passed through various clinical trials for quality control and other tests.

11. Explain briefly

- (a) PCR
- (b) Restriction enzymes and DNA
- (c) Chitinase

11. (a) PCR or the polymerase chain reaction is the invitro amplification of DNA for experimental purpose when the sample is in very low quantity. This process was first developed by Kary Mullis in 1985. The process uses template DNA which is first denatured by heating at 96°C followed by annealing of two primers at the 3' ends of the template DNA strands at a comparatively lower temperature. Now DNA polymerase is added along with the four deoxyribonucleoside triphosphates (dATP, dTTP, dCTP and dGTP) to elongate the DNA strand. Normal polymerases cannot withstand the high temperature of the PCR thus a special polymerase called the Taq polymerase is added. This Taq polymerase is thermostable in nature as it is isolated from the bacterium *Thermus aquaticus* which is found in the hot springs and thus is active in very high temperatures. The full process is maintained at a range of 72°C to 75°C and is allowed to run for any required number of cycles.

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Polymerase chain reaction - PCR



- 1 Denaturation at 94-96°C
- 2 Annealing at ~68°C
- 3 Elongation at ca. 72 °C

(b) Restriction enzymes are synthesised by eubacterias as a defence mechanism. These are specifically endonucleases and cleave double stranded DNA where they find their specific restriction sites. Restriction enzymes are of three types: type I, type II and type

III. Of which only type II restriction endonucleases restricts DNA within the recognition site and thus are used as molecular scissors for genetic engineering. Sites recognisable by the restriction enzymes are also present in the host genome but they are methylated for protection against their own enzyme, so that they do not undergo self digestion.

(c) Chitinase is an enzyme which is used to degrade the glycosidic bond present in chitin. Chitin makes up the fungal cell membrane exoskeleton. Thus when it is required to break open a fungal cell for experimentation Chitinase is used.

Note: Glycosidic bonds are those that occur between two glucose molecules.

12. Discuss with your teacher and find out how to distinguish between

(a) Plasmid DNA and Chromosomal DNA

(b) RNA and DNA

(c) Exonuclease and Endonuclease

12. (a) Plasmid DNA and Chromosomal DNA

Plasmid DNA	Chromosomal DNA
Plasmid DNA is an extra-chromosomal DNA molecule in bacteria that is capable of replicating, independent of chromosomal DNA	Chromosomal DNA is the entire DNA of an organism present inside chromosomes.

(b) RNA and DNA

RNA	DNA
1. RNA is a single stranded molecule.	1. DNA is a double stranded molecule.
2. It contains ribose sugar.	2. It contains deoxyribose sugar.
3. The pyrimidines in RNA are adenine and uracil.	3. The pyrimidines in DNA are adenine and thymine.
4. RNA cannot replicate itself.	4. DNA molecules have the ability to replicate.
5. It is a component of the ribosomes.	5. It is component of the chromosomes.

(c) Exonuclease and Endonuclease

Exonuclease	Endonuclease
It is a type of restriction enzyme that removes the nucleotide from 5' or 3' ends of the DNA molecule.	It is a type of restriction enzyme that makes a cut within the DNA at a specific site to generate sticky ends.



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