

**Question 1:**

Crystals of Bt toxin produced by some bacteria do not kill the bacteria themselves because –

- (a) bacteria are resistant to the toxin
- (b) toxin is immature:
- (c) toxin is inactive:
- (d) bacteria encloses toxin in a special sac.

Answer

toxin is inactive:

In bacteria, the toxin is present in an inactive form, called prototoxin, which gets converted into active form when it enters the body of an insect.

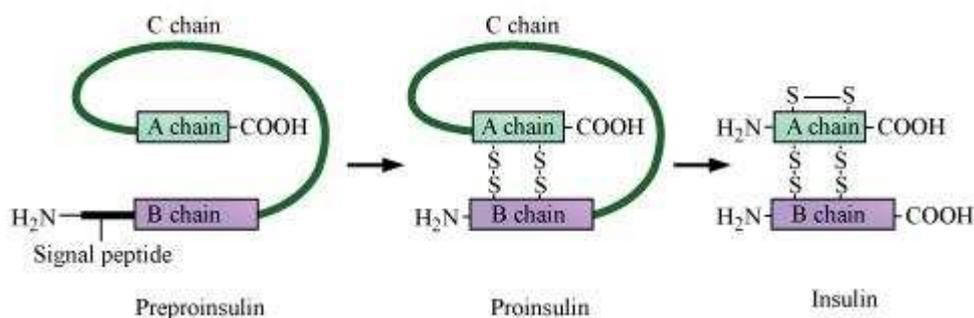
Question 2:

What are transgenic bacteria? Illustrate using any one example.

Answer

Transgenic bacteria contain foreign gene that is intentionally introduced into its genome. They are manipulated to express the desirable gene for the production of various commercially important products.

An example of transgenic bacteria is *E.coli*. In the plasmid of *E.coli*, the two DNA sequences corresponding to A and B chain of human insulin are inserted, so as to produce the respective human insulin chains. Hence, after the insertion of insulin gene into the bacterium, it becomes transgenic and starts producing chains of human insulin. Later on, these chains are extracted from *E.coli* and combined to form human insulin.

**Question 3:**



Compare and contrast the advantages and disadvantages of production of genetically modified crops.

Answer

The production of genetically modified (GM) or transgenic plants has several advantages.

(i) Most of the GM crops have been developed for pest resistance, which increases the crop productivity and therefore, reduces the reliance on chemical pesticides.

(ii) Many varieties of GM food crops have been developed, which have enhanced nutritional quality. For example, golden rice is a transgenic variety in rice, which is rich in vitamin A.

(iii) These plants prevent the loss of fertility of soil by increasing the efficiency of mineral usage.

(iv) They are highly tolerant to unfavourable abiotic conditions.

(v) The use of GM crops decreases the post harvesting loss of crops.

However, there are certain controversies regarding the use of genetically modified crops around the world. The use of these crops can affect the native biodiversity in an area. For example, the use of Bt toxin to decrease the amount of pesticide is posing a threat for beneficial insect pollinators such as honey bee. If the gene expressed for Bt toxin gets expressed in the pollen, then the honey bee might be affected. As a result, the process of pollination by honey bees would be affected. Also, genetically modified crops are affecting human health. They supply allergens and certain antibiotic resistance markers in the body. Also, they can cause genetic pollution in the wild relatives of the crop plants. Hence, it is affecting our natural environment.

Question 4:

What are Cry proteins? Name an organism that produces it. How has man exploited this protein to his benefit?

Answer

Cry proteins are encoded by cry genes. These proteins are toxins, which are produced by *Bacillus thuringiensis* bacteria. This bacterium contains these proteins in their inactive form. When the inactive toxin protein is ingested by the insect, it gets activated by the alkaline pH of the gut. This results in the lysis of epithelial cell and eventually the death of the insect. Therefore, man has exploited this protein to develop certain transgenic crops with insect resistance such as Bt cotton, Bt corn, etc.

**Question 5:**

What is gene therapy? Illustrate using the example of adenosine deaminase (ADA) deficiency.

Answer

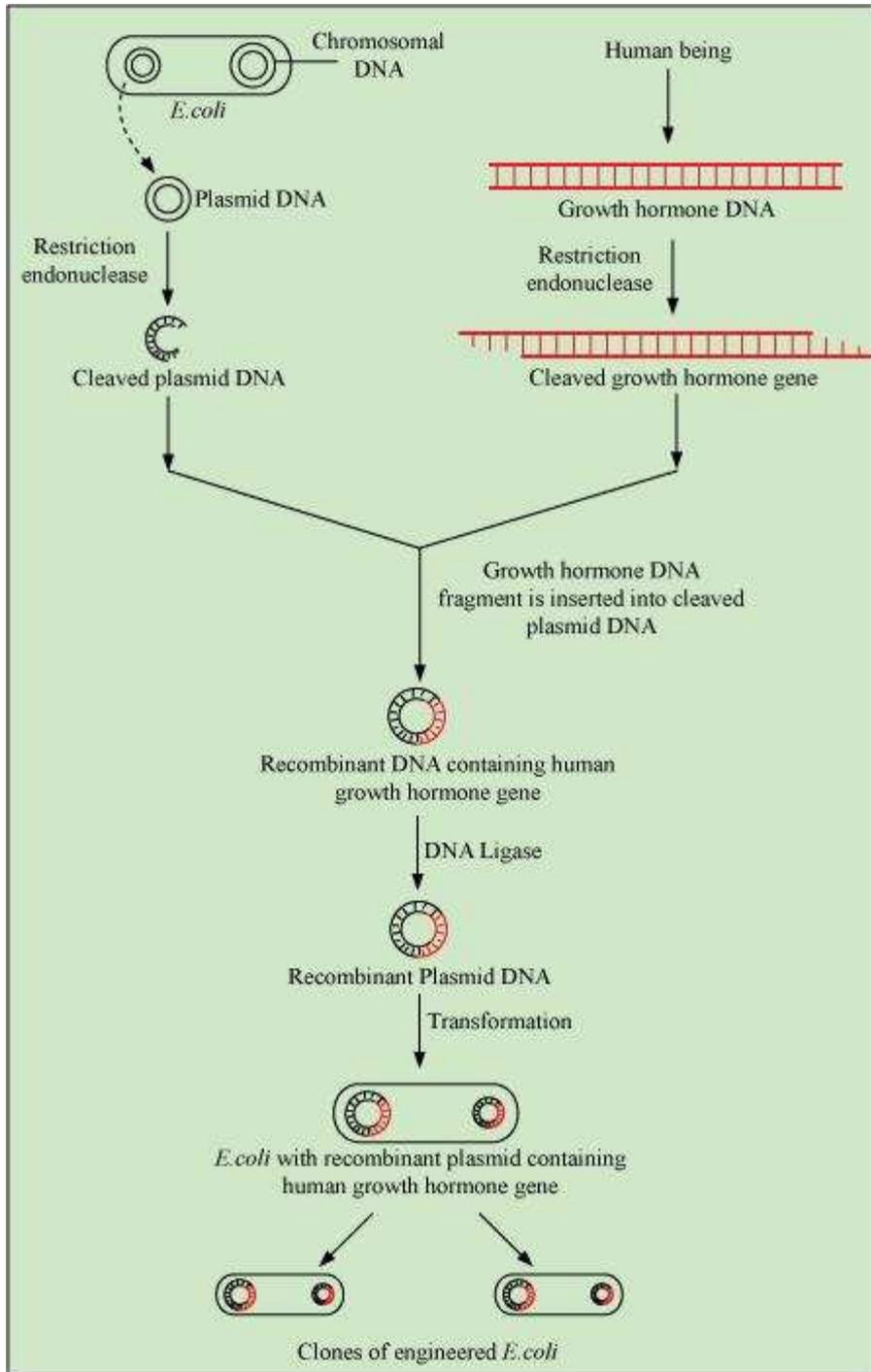
Gene therapy is a technique for correcting a defective gene through gene manipulation. It involves the delivery of a normal gene into the individual to replace the defective gene, for example, the introduction of gene for adenosine deaminase (ADA) in ADA deficient individual. The adenosine deaminase enzyme is important for the normal functioning of the immune system. The individual suffering from this disorder can be cured by transplantation of bone marrow cells. The first step involves the extraction of lymphocyte from the patient's bone marrow. Then, a functional gene for ADA is introduced into lymphocytes with the help of retrovirus. These treated lymphocytes containing ADA gene are then introduced into the patient's bone marrow. Thus, the gene gets activated producing functional T- lymphocytes and activating the patient's immune system.

Question 6:

Diagrammatically represent the experimental steps in cloning and expressing an human gene (say the gene for growth hormone) into a bacterium like *E. coli* ?

Answer

DNA cloning is a method of producing multiple identical copies of specific template DNA. It involves the use of a vector to carry the specific foreign DNA fragment into the host cell. The mechanism of cloning and transfer of gene for growth hormone into *E.coli* is represented below.



Question 7:



Can you suggest a method to remove oil (hydrocarbon) from seeds based on your understanding of rDNA technology and chemistry of oil?

Answer

Recombinant DNA technology (rDNA) is a technique used for manipulating the genetic material of an organism to obtain the desired result. For example, this technology is used for removing oil from seeds. The constituents of oil are glycerol and fatty acids. Using rDNA, one can obtain oilless seeds by preventing the synthesis of either glycerol or fatty acids. This is done by removing the specific gene responsible for the synthesis.

Question 8:

Find out from internet what is golden rice.

Answer

Golden rice is a genetically modified variety of rice, *Oryza sativa*, which has been developed as a fortified food for areas where there is a shortage of dietary vitamin A. It contains a precursor of pro-vitamin A, called beta-carotene, which has been introduced into the rice through genetic engineering. The rice plant naturally produces beta-carotene pigment in its leaves. However, it is absent in the endosperm of the seed. This is because beta-carotene pigment helps in the process of photosynthesis while photosynthesis does not occur in endosperm. Since beta-carotene is a precursor of pro-vitamin A, it is introduced into the rice variety to fulfill the shortage of dietary vitamin A. It is simple and a less expensive alternative to vitamin supplements. However, this variety of rice has faced a significant opposition from environment activists. Therefore, they are still not available in market for human consumption.

Question 9:

Does our blood have proteases and nucleases?

Answer

No, human blood does not include the enzymes, nucleases and proteases. In human beings, blood serum contains different types of protease inhibitors, which protect the blood proteins from being broken down by the action of proteases. The enzyme, nucleases, catalyses the hydrolysis of nucleic acids that is absent in blood.

Question 10:



Consult internet and find out how to make orally active protein pharmaceutical. What is the major problem to be encountered?

Answer

Orally active protein pharmaceuticals contain biologically active materials such as peptides or proteins, antibodies, and polymeric beads. It is administered orally into the body through various formulations. It involves the encapsulation of protein or peptide in liposomes or formulations using penetration enhancers. These proteins or peptides are used for treatment of various diseases and are also used as vaccines. However, the oral administration of these peptides or proteins has some problems related to it. Once these proteins are ingested, the proteases present in the stomach juices denature the protein. As a result, their effect will be nullified. Hence, it is necessary to protect the therapeutic protein from digestive enzymes, if taken orally. This is the reason for the proteins to be injected directly into the target site.

CHAPTER 12

BIOTECHNOLOGY AND ITS APPLICATIONS

POINTS TO REMEMBER

Biopesticides : Biological agents that are used to control weeds, insects and other pests.

Cry Gene : The Bt toxins are coded by a gene named Cry.

Cry Protein : The insecticidal protein which is produced by *Bacillus thuringiensis*.

Green Revolution : Substantial increase in crop yields due to use of high yielding varieties, use of fertilisers and pesticides, improved agricultural practices etc.

Genetically Modified Organisms (GMO) : The organisms which have altered genes in them. These are also known as transgenic organisms.

Molecular Diagnosis : Refers to early detection of diseases using recombinant DNA molecules and techniques like PCR and autoradiography.

RNA Interference (RNAi) : Process used to develop pest resistant plants. It involves silencing of a specific mRNA due to complementary double stranded RNA.

Sustainable Agriculture : It involves organic farming and other integrated management practices which maintain soil fertility while increasing crop productivity.

Uses of GM Plants : Tolerant to abiotic stress, Reduced dependence on chemical pesticides, less post harvest-loss, Efficient use of minerals, enhanced nutritional value.

Uses of Transgenic Animals : To study normal physiology and development, to study diseases, to get biological products, to test vaccine and chemical safety testing.

Gene Therapy : It is a technique of inserting genes into the cells and tissue of an individual to treat a hereditary disease.

- The first clinical gene therapy was given in 1990 to a four year old girl with adenosine deaminase (ADA) deficiency. ADA enzyme is required for proper functioning of immune system.

- This disorder is caused due to the deletion of the gene for adenosine deaminase enzyme. In some children ADA deficiency can be cured by bone marrow plantation. Lymphocytes from the blood of patient are grown in a culture. A functional ADA cDNA is then introduced into these lymphocytes using retroviral vector. The lymphocytes are transferred into the body of patients.
- As these cells are not immortal, the patient required periodic infusion of such genetically engineered lymphocytes.
- If a functional gene is introduced into a bone marrow cells at early embryonic stage, It could be a permanent cure of ADA deficiency.

Bt. Cotton : The soil bacterium *Bacillus thuringiensis* produced crystal protein called cry protein that kills certain insects larvae such as tobacco budworm, armyworm, beetles and flies.

- Bt toxin protein exists as inactive protoxins, but once an insect ingest this inactive toxin, it is converted into active form of toxin due to the alkaline pH of the gut which solubilise the crystal. This causes swelling and lysis of epithelial cells of midgut leading to death of insect larvae.
- Bt toxin genes were isolated from *Bacillus thuringiensis* and incorporated into the several crop plants such as cotton.
- The proteins encoded by the genes : cryIAc and cryIIAb control the cotton bollworms and cryIAb control corn borer.

Pest Resistant Plants : A nematode *Meloidegyne incognitia* infects tobacco plants and reduces their yield.

- Nematode specific genes were introduced into the host plant using *Agrobacterium* as a vector.
- The introduction of DNA was such that it produced both sense and antisense RNA in the host cells.
- These two RNAs being complementary to each other formed a double stranded RNA (dsRNA) making it inactive.
- of the nucleotide by the process called RNA interference (RNAi).
- The result was that the parasite could not survive in the transgenic host and the transgenic plant got protected for the parasite.

Three Critical Research Areas of Biotechnology

- (i) Providing best catalyst in the form of improved organism usually a microbe.
- (ii) Creating optimal conditions for a catalyst to act.
- (iii) Downstreaming processing technologies to purify the desirable product.

QUESTIONS

VSA (1 MARK)

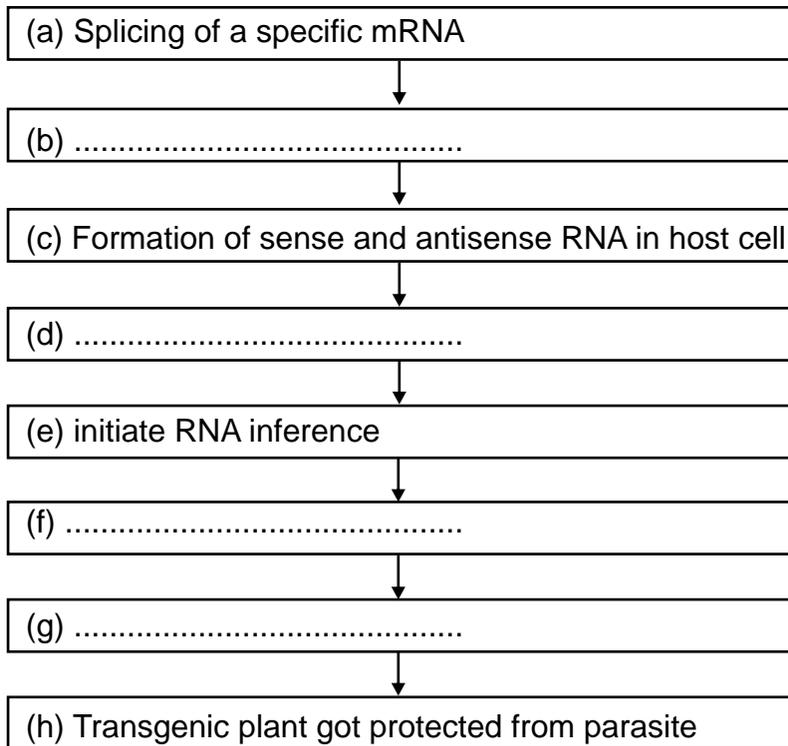
1. Name the technique based on the principle of antigen-antibody interaction used in detection of a virus (HIV).
2. Development of a transgenic food crop may help in solving the problem of night blindness in the developing countries, name this crop plant.
3. Which nematode infects the roots of tobacco plant and causes a great reduction in yield?
4. The first transgenic cow, produced human protein . enriched milk. Name the cow and the protein found in milk.
5. The insulin produced using recombinant DNA technology is more advantageous than the insulin extracted from pancreas of slaughtered cattle and pigs. How?
6. Name two pest resistant plants produced by using recombinant DNA technology.

SA-II (2 MARKS)

7. What are the two methods for correcting ADA deficiency in a child?
8. Some crop plants are modified genetically by manipulating their genes. How are they made beneficial?
9. GEAC is one of the organisation set up by Indian Government. Write its full form. Give its two objectives.
10. "Industrialised nations are exploiting the bioresources of under industrialised nations". Justify the statement with a suitable example.

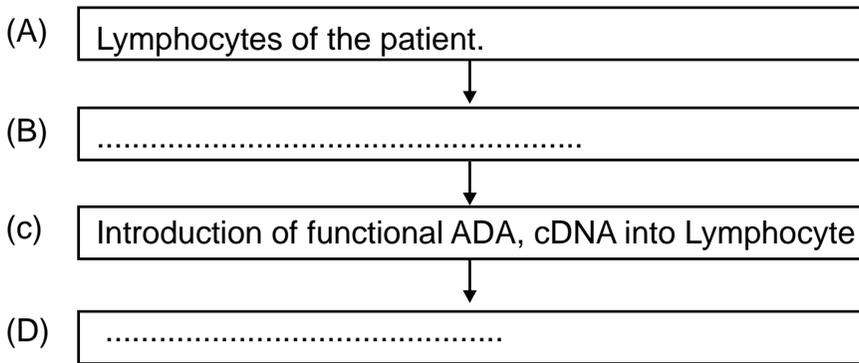
SA-I (3 MARKS)

11. Some multinational companies and other organisations are using bioresources for commercial benefits, without proper authentication and compensation to concerned authorities.
- (a) Give the term for this unauthorised act.
 - (b) Suggest any two ways to get rid of this.
12. A bacterium *Bacillus thuringiensis* produces a toxic protein named .cry protein. that is lethal to certain insects but not to bacterium
- (a) Why this toxin does not kill the bacteria?
 - (b) What type of changes occur in the gut of insects on consuming this protein?
 - (c) How man has exploited this protein for his benefit?
13. Given below is an incomplete flow chart showing the process of production of nematode resistant tobacco plants based on RNAi technique.
- (i) Write the missing steps in proper sequence
 - (ii) At which level RNAi silences the gene?



LA (5 Marks)

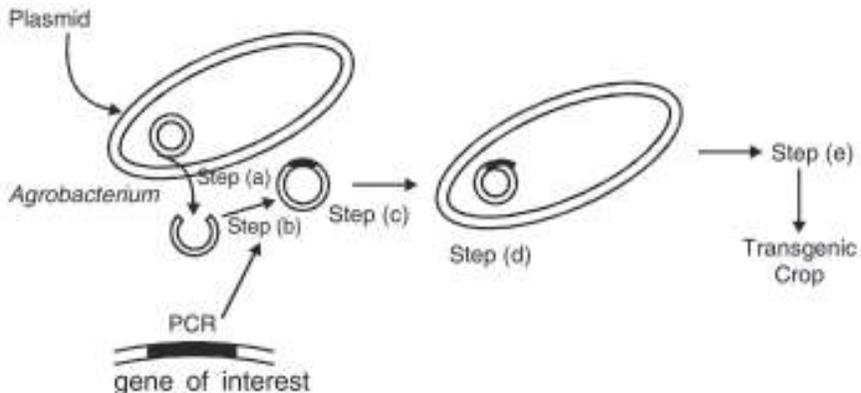
14. The clinical gene therapy is given to a 4 years old patient for an enzyme which is crucial for the immune system to function.



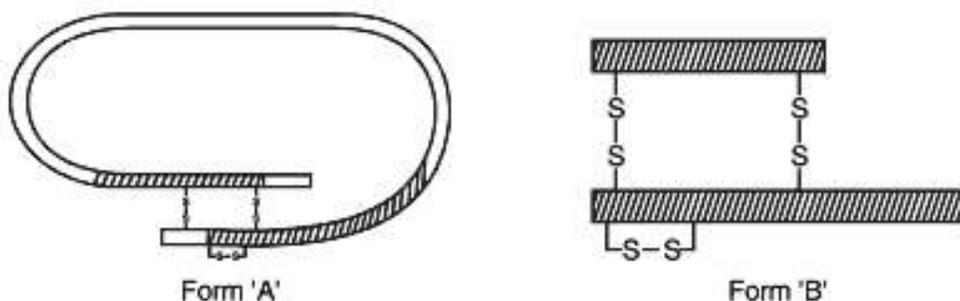
Observe the therapeutical flow chart and give the answer of the following:

- (a) Complete the missing steps (B) and (D)
- (b) Identify the disease to be cured.
- (c) Why the above method is not a complete solution to the problem?
- (d) Scientists have developed a method to cure this disease permanently. How?

15. In the given figure, *Agrobacterium* is utilized for the production of a transgenic crop. Explain the steps a, b, c, d and e shown in the figure.



16. In the given figure, Form (A) and Form (B) represents different forms of a proteinaceous hormone secreted by pancreas in mammals.



- (a) What type of bonding is present between chains of this hormone?
- (b) What are these form (A) and form (B). How these forms differ from each other?
- (c) Explain how was this hormone produced by Eli Lilly, an American company, using rDNA technology.

ANSWERS

VSA (1 MARK)

1. ELISA (Enzyme linked immuno - sorbent Assay)
2. Golden Rice
3. *Meloidegyne incognitia*.
4. Rosie, alpha-lactalbumin
5. Insulin obtained from animal source causes allergy.
6. Bt Cotton, Bt Corn, Bt Brinjal.

SA-II (2 MARKS)

7. Bone marrow transplantation having functional ADA enzyme and Enzyme replacement therapy.
8. More tolerant to abiotic stresses; pest resistant; reduction in post harvest losses; increased nutritional value of food.
9. GEAC: Genetic Engineering approval committee.

Objectives of GEAC are :

- (i) To make decisions regarding validity of GM research.
 - (ii) Safety of introducing GMO for public use.
10. • Industrialised nations are collecting and patenting the genetic resources of under industrialised country like India. An American Company got patent rights on Basmati rice.
- Valuable biomolecules obtained from bioresources are patented and used for commercial purposes.

SA-I (3 MARKS)

11. (a) Biopiracy
- (b) (i) Benefits of bioresources should be shared between developed and developing nations
- (ii) Laws should be developed to prevent unauthorised exploitation of them bioresources.
12. (a) Produced in inactive form as Prototoxins.
- (b) Prototoxin becomes active toxin in alkaline pH of gut of insects. Toxins bind to surface of midgut and cause perforation, swelling, lysis of cells ultimately leading to death.
- (c) Specific Bt toxin genes isolated from *Bacillus thuringiensis* and incorporated into several crop plants such as cotton and corn which become pest resistant against certain insects.
13. (i) (b) Using *Agrobacterium* as a vector, introduced into tobacco
- (d) dsRNA (double stranded RNA)
- (f) Silenced specific mRNA of the nematode
- (g) Parasite could not survive.
- (ii) RNAi silences the gene at translation level

LA (5 MARKS)

14. (a) Step (B) : Lymphocytes are grown in culture medium.
- Step (D) : Infusion of genetically engineered lymphocytes into patients.
- (b) Adenosine deaminase (ADA) deficiency.

- (c) As genetically engineered lymphocytes are not immortal, the patient requires periodic infusion of cells.
 - (d) If the gene isolated from bone marrow cells producing ADA is introduced into cells at early embryonic stages, it could be a permanent cure.
15. **Step (a)** Plasmid is removed and cut open with restriction endonuclease.
- Step (b)** Gene of interest is isolated from another organism and amplified using PCR
- Step (c)** New gene is inserted into plasmid
- Step (d)** Plasmid is put back into Agrobacterium
- Step (e)** Agrobacterium based transformation.
16. (a) Disulphide bonds
- (b) Form (A) : Proinsulin
Form (B) : Mature insulin.
Proinsulin contains an extra stretch called C . peptide which is absent in mature insulin.
- (c) Eli Lilly company prepared two DNA sequences corresponding to A and B peptide chains of human insulin and introduced them in plasmid E. coli to produce insulin chains. Chains A and B were produced separately, extracted and combined by creating disulphide bonds to form insulin.