

Biotechnology and its Applications

Topic 1: **Biotechnological Applications in Agriculture and Medicine**

Previous Years' Examination Questions

1 Mark Questions

1. Name any two techniques that serve the purpose of early diagnosis of some bacterial/viral human diseases.
[Foreign 2011]
2. Name the source organism of the gene cry IAc and its target pest. [Foreign 2011]
3. Name a molecular diagnostic technique to detect the presence of a pathogen in its early stage of infection. [Delhi 2010]

4. What is the host called that produces a foreign gene product? what is this product called? [Foreign 2010]
5. Name the cry genes that control cotton bollworm and corn-borer respectively. [All India 2009C]
6. Suggest any two techniques which can help in early detection of bacterial/viral infections much before the symptoms appear in the body. [All India 2008]
7. What is the significance of the process of RNA interference (RNAi) in eukaryotic organisms? [Foreign 2008]
8. State the principle on which ELISA works. [Foreign 2008]
9. How does silencing of specific mRNA in RNA interference prevent parasitic infestation? [Delhi 2008C]
10. How are tobacco plants benefitted when nematode-specific genes are introduced into them using certain vectors? Name the vectors used? [Delhi 2008C]

2 Marks Questions

11. (a) State the role of DNA ligase in biotechnology.
(b) What happens when *Meloidegryne incognitia* consumes cells with RNAi gene? [Delhi 2012]
12. (a) Mention the cause and the body system attacked by ADA deficiency in humans.
(b) Name the vector used for transferring ADA-DNA into the recipient cells in humans. Name the recipient cells. [All India 2012]
13. Explain the process of RNA interference. [Delhi 2011]
14. Why is the introduction of genetically engineered lymphocytes into an ADA deficiency patient not a permanent cure? Suggest a possible permanent cure. [Delhi 2010]
15. How did Eli Lilly synthesize the human insulin? Mention the difference between this insulin and the one produced by the human pancreas. [All India 2010]
16. Why do the toxic insecticidal proteins secreted by *Bacillus thuringiensis* kill the insect and not the bacteria itself? [Foreign 2010]
17. How do Eli Lilly synthesize the human insulin? Mention the difference between this insulin and the one synthesized by the human pancreas. [Delhi 2010C]
18. How is Bt cotton made to attain resistance against bollworm? [Delhi 2010C]
19. Highlight any four advantages of genetically modified organisms (GMOs). [Foreign 2009]
20. List the three molecular diagnostic techniques that help detect pathogens from suspected patients. Mention one advantage of these techniques over conventional methods. [Delhi 2009C]
21. Expand ELISA. On what principle is ELISA test based? List two ways by which an infection can be detected by this test. [All India 2009C]
22. State the principle on which ELISA technique is based. How does it help in early detection of a disease? [Delhi 2008C]
23. List four applications of genetically modified plants. [All India 2008C]
24. How is gene therapy being used in treating ADA deficiency patients? [All India 2008C]

3 Marks Questions

25. Name the genes responsible for making Bt cotton plants resistant to bollworm attack. How do such plants attain resistance against bollworm attacks. Explain. [Delhi 2012]
26. (a) Tobacco plants are damaged severely when infested with *Meloidegryne incognitia*. Name and explain the strategy that is adopted to stop this infestation.

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- (b) Name the vector used for introducing the nematode specific gene in tobacco plant. [All India 2012]
27. How does RNA interference help in developing resistance in tobacco plant against nematode infection? [Delhi 2010]
28. (a) How has biotechnology helped in producing *Meloidegryne incognitia* resistant tobacco plant?
(b) Why does this nematode die on eating such a GM plant? [Delhi 2010C]
29. (a) Explain the effect of deletion of the gene for ADA in an individual.
(b) How does the gene therapy help in this case? [All India 2010C]
30. Plasmid is boon to biotechnology. Justify this statement quoting the production of human insulin as an example. [All India 2009]
31. Name the source and the types of *cry* genes isolated from it for incorporation into crops by biotechnologists. Explain how have these genes brought beneficial changes in the genetically modified crops. [All India 2009]
32. How did Eli Lilly company go about preparing the human insulin? How is the insulin thus produced different from that produced by the functional human insulin gene? [Foreign 2009]
33. What are *cry* proteins? Name an organism that produces it. How has man exploited this protein to his benefit? [Delhi 2009C]
35. Name the process involved in the production of nematode-resistant tobacco plants, using genetic engineering explain the strategy adopted to develop such plants. [Foreign 2011]
36. Describe the various stages involved in gene transfer for the commercial production of human insulin by Eli Lilly. [Foreign 2011]
37. One of the main objectives of biotechnology is to minimize the use of insecticides on cultivated crops. Explain with the help of a suitable example, how insect resistant crops have been developed using techniques of biotechnology. [Delhi 2009]
38. (a) How is mature insulin different from proinsulin secreted by pancreas in human?
(b) Explain how was human functional insulin produced using rDNA technology.
(c) Why is the functional insulin thus produced considered better than the ones used earlier by diabetic patients. [Delhi 2009]
39. How is a transgenic tobacco plant protected against *Meloidegryne incognitia*? Explain the procedure. [All India 2009]
40. Expand the name of the enzyme ADA. Why is the enzyme essential in the human body? Suggest a gene therapy for its deficiency. [All India 2009]

5 Marks Questions

34. (a) Name the source from which insulin was extracted earlier. Why is this insulin no more in by diabetic people?
(b) Explain the process of synthesis of insulin by Eli Lilly company. Name the technique used by the company.
(c) How is the insulin produced by human body different from the insulin produced by the above mentioned company? [All India 2011]
41. (a) How is a transgenic tobacco plant protected against *Meloidegryne incognitia*?
(b) Explain the procedure of making such plants. [Foreign 2009]
42. What is ADA deficiency? Describe three methods to cure it. [All India 2009]
43. (a) What is plasmid?
(b) What is meant by ADA deficiency? How is gene therapy a solution to this problem? Why is it not a permanent cure? [Delhi 2008; Foreign 2008]

44. (a) Name the nematode that infests and damages tobacco roots.
 (b) How are transgenic tobacco plants produced to solve this problem?
 [All India 2008]
45. Explain the steps involved in the production of genetically engineered insulin. Why is insulin thus produced

preferred to the one produced from non-human sources? [Foreign 2008]

46. (a) Why is *Bacillus thuringiensis* considered suitable for developing GM plants?
 (b) Explain how it has been used to develop GM crops. [Foreign 2008]

Explanations

- (i) DNA recombinant technology.
 (ii) ELISA.
- Source—*Bacillus thuringiensis*
 Target— Cotton bollworms
- Polymerase Chain Reaction (PCR).
- Transgenic organisms or genetically modified organism.
 - Recombinant proteins.
- Cotton bollworm – *cry IAc* and *cry IIAb*
 Corn borer – *cry IAb*
- Polymerase Chain Reaction (PCR) and Recombinant DNA Technology.
- RNA interference (RNAi) acts as cellular defense in all eukaryotic organisms.
- ELISA is based on the principle of antigen-antibody interaction.
- It can be prevented by using RNA interference (RNAi) process which is checked by silencing of specific mRNA due to a complementary dsRNA. dsRNA binds and prevents translation of the mRNA (silencing).
- Nematode-specific genes were introduced into the host plants which produce both sense and anti-sense RNA in the host cells. These two RNAs are complementary to each other and forms a double RNA (dsRNA) that initiate RNAi and silence the specific mRNA of the parasite cannot survive in transgenic host, so prevents the plants from pest.

Vector used is *Agrobacterium*.

- (a) DNA ligase enzyme is used to join two DNA fragment from their ends.
 (b) When *Meloidogyne incognita* (parasite) consumes cells with RNAi gene, parasite cannot survive and this prevents infestation. The introduced RNAi gene DNA forms both sense and anti-sense RNA. Two strands being complementary to each other bend and form dsRNA, leading to RNAi. Thus, the mRNA of nematode is silenced and the parasite cannot survive there. This produces *Meloidogyne incognita* resistant tobacco plants.
- (a) ADA is caused due to deletion of gene for adenosine aminase.
 Immune system of body is affected due to this.
 (b) Retroviral vector is used to transfer ADA-DNA into the recipient cells of human.
 Recipient cells—Lymphocytes.
- Process of RNA interference (RNAi) is related with silencing of a specific mRNA.
 - A complementary double-stranded RNA binds to the mRNA and prevents its translation.
 - This complementary RNA could be from an infection by viruses having RNA genomes or mobile genetic elements (transposons) that replicate via an RNA intermediate.

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- (iii) Using *Agrobacterium* vectors, nematode specific genes were introduced into the host plants.
- (iv) It produces both sense and anti-sense RNA in the host cells.
- (v) These two RNAs are complementary to each other formed a double-stranded (dsRNA) that initiated RNAi, silencing the specific mRNA of the nematode.
- (vi) Due to this, parasite could not survive in a transgenic host expressing interfering RNA.
- 14.** The genetically engineered lymphocytes have a life span. Hence, the patient requires periodic infusion of genetically engineered lymphocytes, so the cure is not permanent. The cure can be permanent, if the gene isolated from marrow cells producing ADA is introduced into the cells at early embryonic stages.
- 15.** Insulin production by Eli Lilly company :
- DNA sequences to the two polypeptide A and B of insulin are synthesized *in vitro*.
 - They are introduced into plasmid DNA of *E.coli*.
 - This bacterium is cloned under suitable conditions.
 - The transgene is expressed in the form of polypeptides A and B, secreted into the medium.
 - They are extracted and combined by creating disulphide bridge to form human insulin.
- Differences between insulin produced by rDNA and insulin produced by pancreas.

Insulin Produced by rDNA	Insulin Produced by Pancreas
It produces only A and B polypeptides.	It has three polypeptides A, B and C before maturing.

- 16.** *Bt* toxin does not kill bacteria because it exists as an inactive pro-toxin.
- When *Bt* toxin is ingested by an insect, it is converted into its active form when exposed to the alkaline pH in the gut. The activated toxin binds to the surface of the epithelial cells of the

midgut and creates pores. This causes swelling and lysis of cells of insect body.

- 17.** Refer to Ans. 15.
- 18.** Specific *Bt* cotton genes obtained from *Bacillus thuringiensis* are used in plants like cotton. The toxin is coded by a gene called *cry*, encoded by the genes *cry IAc* and *cry II Ab* that control the cotton bollworms.
- 19.** Advantages of GMOs.
- Tolerant against biotic stresses, such as cold, drought, salt, heat.
 - Reduces relevance on chemical pesticides.
 - Reduces post harvest losses.
 - Increase efficiency of mineral usage by plants.
- 20.** Molecular diagnostic techniques.
- Polymerase chain reaction.
 - Recombinant DNA technology.
 - Enzyme Linked Immuno-Sorbent Assay (ELISA).

The advantage of these techniques is that these help in early detection and treatment of diseases, which is not possible by the conventional diagnosis methods.

- 21.** ELISA – Enzyme Linked Immuno Sorbent Assay.
- ELISA is based on antigen-antibody reaction.
- By ELISA :
- The presence of antigens (proteins, glycoproteins, etc.) are detected.
 - Antibodies produced against the pathogen are detected.

22. Refer to Ans. 21.

23. Refer to Ans. 19.

- 24.** ADA is caused due to the deletion of gene for adenosine deaminase.

Lymphocytes from patient's blood were grown in a culture and functional ADA and DNA was introduced in these lymphocytes using a retroviral vector.

Lymphocytes were transferred into the patient's body. Periodic infusion of such

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genetically engineered lymphocyte is done because these cells are mortal.

For permanent cure, gene isolated from the bone marrow cells producing ADA at early embryonic stage can be possible cure.

25. *cry* gene.

Bt toxins are initially inactive pro-toxins but after ingestion by the insect, their inactive toxin becomes active due to the alkaline pH of the gut which solubilize the crystals. The activated toxin binds to the surface of midgut epithelial cells thus creating pores which cause swelling and lysis, leading to death of the bollworm. This way *Bt* cotton attains resistance against bollworm.

26. (a) Infestation of tobacco plant can be stopped by using RNA interference (RNAi) process.

Process of RNAi : Refer to Ans. 13 of Topic 1.

(b) Vector is *Agrobacterium*.

27. Refer to Ans. 13.

28. (a) Refer to 11(b).

(b) Due to RNAi process, specific mRNA of nematode is silenced. The result is that the parasite could not survive in a transgenic host expressing, specific interfering RNA.

29. (a) Effect of deletion of the gene for ADA in an individual : This leads to ADA deficiency disorder, adenosine deaminase (ADA) enzyme is crucial for immune system to function.

(b) Gene therapy in case of ADA deficiency : Refer to Ans. 24.

30. Plasmid is an autonomously replicating extra chromosomal circular DNA found in bacterial cells. Because it can replicate with in a bacterial cell, it is used as a vector in rDNA technology.

Production of Insulin :

(i) Eli Lilly, an A company produced two DNA seq *in vitro* coding for the polypeptides of insulin.

(ii) These DNA s were introduced into the plasm *oli*.

(iii) The *E.coli* cells produced the two poly peptides, which were isolated and combined by disulphide bridges to form human insulin.

31. The source is *Bacillus thuringiensis*.

Types of *cry* genes – *cry I Ac*, *cry II Ab*, *cry I Ab*

Method of changes caused by *cry* genes in GM crops :

(i) The *cry* genes code for certain crystal proteins that have *Bt* toxin.

(ii) *Bt* toxin exists as inactive pro-toxin and gets converted into active form (toxin) in the alkaline pH of the gut of the insect.

(iii) The activated toxin binds to the epithelial cells lining the surface of the midgut and creates pores leading to swelling and lysis of the cells and ultimately cause death of the insect.

(iv) This way GM crops show resistance against insect pests.

32. Refer to Ans. 15.

33. *cry* protein (crystal protein) is a toxin coded by a gene *cry* and is poisonous to some insects. Thus, giving resistant characters to the plants.

Bacillus thuringiensis produces *cry* protein.

cry protein producing gene is transferred to the plant to provide resistance against insect larvae. Man has developed several transgenic crops by introducing these genes from bacteria to crop plants such as *Bt* cotton, *Bt* corn, etc.

34. (a) Pancreas of slaughtered pigs and cattle animals. Insulin obtained from these sources caused some allergy or some other reactions to the foreign protein.

(b) Production of human insulin by Eli Lilly; Refer to Ans. 15.

The company used rDNA technology for this.

(c) Refer to Ans. 15. for difference.

35. The process is called RNA interference (RNAi) which involves silencing of a specific mRNA.

Strategy adopted to develop such plants. Refer to Ans. 13.

36. Refer to Ans. 15.

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37. Development of insert resistance crops using techniques to biotechnology :
- The bacterium *Bacillus thuringiensis* produce proteins, called *Bt* toxins, which kill the larvae of some insects.
 - This *Bt* toxin gene has been isolated and cloned in bacteria.
 - These genes are then introduced in crop plants, which could be expressed in plants to provide resistance to the insects, i.e., they function as biopesticides, so there is no need for insecticides.
 - The *Bt* toxins are insect group specific and coded by *cry* genes. The protein encoded by genes *cry IAc* and *cry IIAb* control cotton bollworm while *cry IAb* controls corn borer.
38. (a) The proinsulin has an extra stretch called the C-peptide along with two short polypeptide chain A and B while the mature insulin has only two polypeptide chains A and B.
- (b) From the human cell, DNA containing insulin gene is isolated. The two DNA sequences corresponding to A and B chains of human insulin is introduced into the plasmids of *E. coli* to produce insulin chains A and B. The extracted chains A and B were combined by creating disulphide bonds to form human insulin (humulin).
- (c) The insulin prepared by rDNA technology does not produce sensitive allergic and immunological reaction. Whereas those used earlier, produced allergic reactions and other complications to the foreign protein as earlier extracted from the pancreas of slaughtered cattle or pigs.
39. Transgenic tobacco plants were produced based on the process of RNA interference (RNAi). Refer to Ans. 13 for procedure.
40. ADA-Adenosine Deaminase. It is required for the proper functioning of immune system. Gene therapy for ADA deficiency : Refer to Ans. 24.
41. (a) Transgenic tobacco plant is protected against *Meloidegryne incognitia* by a process called RNA interference (RNAi).
- (b) Refer to Ans. 13 for procedure.
42. ADA deficiency is caused due to the deletion of gene for adenosine deaminase.
- Methods to cure :
- 1st method** In some cases, it can be cured by bone marrow transplantation and enzyme replacement therapy but it is not fully curative.
 - 2nd Method** Lymphocytes from patient's blood were grown in a culture and functional ADA, cDNA was introduced in these lymphocytes using a retroviral vector. The lymphocytes were transferred into the patient's body. Periodic infusion of such genetically engineered lymphocytes is done because these cells are mortal.
 - 3rd method** This is a permanent method. Genes isolated from the bone marrow cells producing ADA at early embryonic stage can be a possible cure.
43. (a) Plasmid is an extra chromosomal, self-replicating, usually circular, double stranded DNA molecule found naturally in many bacteria.
- (b) ADA deficiency occurs due to the deletion of gene for adenosine deaminase enzyme. The enzyme is crucial for immune system functioning.
- In gene therapy**, lymphocytes from the blood of the patients are grown in a culture outside the body. A functional ADA, cDNA is then introduced using a retro-viral vector into the lymphocytes. These lymphocytes are then returned to the patient.
- Because these cells are not immortal, the patient requires periodic infusion of such genetically engineered lymphocytes.
44. (a) *Meloidegryne incognitia*
- (b) By using the process of RNA interference (RNAi). For procedure. Refer to Ans. 13.
45. Genetically engineered Insulin :
- A mature/functional insulin molecule consists of two polypeptide chains, coded by two genes.

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- (ii) Eli Lilly, an American company prepared two genes or DNA sequences coding for the two polypeptide chains.
- (iii) The genes were introduced into the plasmid of bacterium *E. coli*.
- (iv) The bacterium produces the polypeptides which are secreted in the medium.
- (v) They are isolated from the medium and combined by creating disulphide bonds into functional insulin.

Because insulin produced from non-human sources causes allergy or other reactions to the foreign protein, genetically engineered insulin is preferred for therapy.

- 46.** (a) *Bacillus thuringiensis* produces a protein, which is toxic to the larvae of insects like the bollworm, fly, beetle, etc.

Bt toxin gene is cloned from the bacteria and has been expressed in plants to provide resistance to insects without the need for chemical insecticides.

- (b) (i) *cry* gene coding for proteins has been isolated using restriction enzymes. They are cloned in the vectors and then introduced into desired crop plants. The different types of *cry* genes that code for insect-specific *cry* proteins are : *CryI Ac* and *cry IIAb* that control cotton bollworm. *cryI Ab* controls corn borer.
- (ii) The transgenic plants, i.e., *Bt* cotton, *Bt* corn, *Bt* rice, produce the protein in their cells and express to the insect pest resistance.

Topic 2: **Transgenic Animals and Ethical Issues**

Previous Years' **Examination Questions**

1 Mark Questions

1. Name the Indian variety of rice patented by an American company. [Delhi 2008]
2. What was the speciality of the milk produced by the transgenic cow Rosie? [All India 2008]
3. A multinational company outside India tried to sell new varieties of turmeric without proper patent rights. What is such an act referred to? [All India 2008]
4. What does the organization GEAC check with reference to genetic engineering? [Foreign 2008]

2 Marks Questions

5. How is 'Rosie' considered different from a normal cow? Explain. [All India 2011]

6. Biopiracy should be prevented. State why and how. [All India 2011]
7. Name the first transgenic cow developed and explain the improvement in the quality of the product produced by it. [Foreign 2010]
8. Describe the responsibility of GEAC, set up by the Indian Government. [Delhi 2009]
9. Highlight any four advantages of Genetically Modified Organism (GMOs). [Foreign 2009]

5 Marks Question

10. What are transgenic animals? Explain any four ways in which such animals can be beneficial for humans. [Foreign 2008]

Explanations

1. Basmati Rice.
2. The transgenic cow, Rosie, produced human protein enriched milk (2.4 gm/L). It contained the human alpha-lactalbumin and was nutritionally a more balanced product for human babies.
3. This is called biopiracy. It refers to the use of bio-resources by multinational companies and other organization without proper

authorization from the countries and people concerned.

4. GEAC. (Genetic Engineering Approval Committee) make decisions regarding the validity of GM research and the safety of introducing GM organisms for public services.
5. 'Rosie' is a transgenic cow. The milk of this cow contained the human alpha-lactalbumin and is nutritionally more balanced product for human babies than normal cows.

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6. Biopiracy should be prevented because :

- (i) The countries and people concerned are not given adequate compensatory payment.
- (ii) The countries/people concerned also lose their right to grow and use in breeding experiments to improve the other varieties of the same species.

7. Refer to Ans. 5.

8. Indian Government has set up organizations such as GEAC (Genetic Engineering Approval Committee) which will make decisions regarding the validity of GM research and the safety of introducing GM-organisms for public services.

9. Advantages of Genetically modified organisms.

- (i) Crops become more tolerant to abiotic stresses like cold, heat, drought, salinity, etc.

- (ii) Dependence on chemical pesticides is reduced as they are made pest resistant.
- (iii) Post harvest losses are much reduced.
- (iv) Food produced from GMOs has high nutritional value.

10. Animals that have had their DNA manipulated to possess and express an extra (foreign) gene are called transgenic animals.

Uses to transgenic animals for humans.

- (i) Useful to study gene regulation, their effect on the normal functions of the body and its development.
- (ii) Study of genes which are responsible for diseases in human and their treatment.
- (iii) Useful biological products can be produced by introducing the portion of DNA which codes for a particular product into transgenic animals.
- (iv) Transgenic mice are developed to test the safety of vaccines before being used in humans.