

Biotechnology and its Applications

CASE STUDY / PASSAGE BASED QUESTIONS

1

Read the following and answer any four questions from 1(i) to 1(v) given below:

Crown gall is a neoplastic disease of most dicotyledonous plants and is caused by the soil bacterium *Agrobacterium tumefaciens*. A large extra chromosomal plasmid in these bacteria was found to be responsible for this disease. The plasmid is known as Ti plasmid. Bacteria free crown gall cells can be cultured in the absence of phytohormones. Ti plasmid is widely used in genetic engineering to deliver the desirable genes. The part of Ti plasmid transferred into plant cell DNA is called T-DNA. T-DNA with desired DNA segment is inserted into the chromosome of the host plant where it produces copies of itself.

Syllabus

Application of biotechnology in health and agriculture: Human insulin and vaccine production, stem cell technology, gene therapy; genetically modified organisms - Bt crops; transgenic animals; biosafety issues, biopiracy and patents.

Read the following and answer any four questions from 2(i) to 2(v) given below:

Golden rice was engineered from normal rice by Potrykus and Beyer in the 1990s. The typical golden colour is due to the production of β -carotene a precursor of vitamin A. Golden rice differs from its parental strain by the addition of three β -carotene genes. These included two genes from daffodil plant and third from a bacterium. The incorporation of these genes allows the rice plant to modify certain metabolic pathways in its cells to produce β -carotene.

- (i) Due to genetic modification golden rice plants produce and store β -carotene in
 - (a) stem
 - (b) seed
 - (c) leaves
 - (d) all of these.
- (ii) Transfer of genes to produce golden rice is achieved by
 - (a) *Agrobacterium*
 - (b) pBR322
 - (c) λ -phage
 - (d) gene gun.
- (iii) In golden rice two genes were taken from
 - (a) *Narcissus* sp.
 - (b) *Erwinia*
 - (c) *Coryza sativa*
 - (d) none of these.
- (iv) Golden rice is helpful to fight against disease caused by the deficiency of
 - (a) Vitamin B₁₂
 - (b) Vitamin C
 - (c) Vitamin A
 - (d) Vitamin D.
- (v) Golden rice was genetically engineered by
 - (a) Fire and Mello
 - (b) Potrykus and Beyer
 - (c) Banting and Best
 - (d) Kohler and Milstein.

Read the following and answer any four questions from 3(i) to 3(v) given below:

Plants having foreign genes in their genome through genetic engineering are called transgenic plants. Genes can be incorporated either through a vector or through direct introduction of DNA. Bt cotton is a genetically modified organism which is pest resistant. It contains gene *cry I Ac* and *cry II Ab* of *Bacillus thuringiensis*. It is used to control lepidopterans, coleopterans and dipterans. Bt cotton can resist cotton bollworm and produce higher yields. *Cry* gene produces *cry* protein or Bt toxin. It is an endotoxin which remains as protoxin in plants and converted to active toxin after getting ingested by the insects. Alkaline pH of the insect gut solubilises the protein crystals, the activated toxin creates pores to the mid guts wall of the insects which cause them to death.

- (i) Bt cotton crops are
 - (a) fungal resistant
 - (b) insect resistant
 - (c) drought resistant
 - (d) all of these.
- (ii) Cotton bollworms are killed by the protein encoded by the gene
 - (a) *cry I Ac*
 - (b) *cry I Ab*
 - (c) *cry II Ab*
 - (d) both (a) and (c).
- (iii) Which of the following is not an advantage of GM crops?
 - (a) GM plants enhance nutritional value of food.
 - (b) GM plants are more tolerant to abiotic stresses.
 - (c) GM plants have helped to reduce post harvest losses.
 - (d) GM plants can cause gene transfer to non-target plant species.
- (iv) *Bacillus thuringiensis* is a
 - (a) air borne bacteria
 - (b) soil borne bacteria
 - (c) soil borne fungus
 - (d) food borne bacteria.

- (v) **Assertion :** Cotton bollworms are killed by ingesting the leaves of the transgenic plant.
Reason : Bt toxin binds with the midgut wall of the insect and make pores in them.
- (a) Both assertion and reason are true and reason is the correct explanation of assertion.
 - (b) Both assertion and reason are true but reason is not the correct explanation of assertion.
 - (c) Assertion is true but reason is false.
 - (d) Both assertion and reason are false.

4

Read the following and answer any four questions from 4(i) to 4(v) given below:

Transgenic animals can serve as factories that in some cases, may produce large amount of proteins more efficiently. Transgenic mice have been engineered to express human antibodies by introducing large segment of human DNA encoding human immunoglobulin genes. In transgenic large animals such as cow or sheep proteins of pharmaceutical value can be produced in large quantities in milk which is later purified. Transgenesis can be used to alter many phenotypic properties including growth rate, fat composition, milk production, hair texture, etc.

- (i) The production of transgenic animals includes
 - (a) identification and separation of desired gene
 - (b) combining the desired gene with appropriate vector
 - (c) introduction of vector in cells, tissues or embryos
 - (d) all of these.
- (ii) In transgenic animals, *i.e.*, cow and sheep proteins of pharmaceutical value are produced in large quantities in the
 - (a) blood
 - (b) accumulated fat
 - (c) mammary glands
 - (d) none of these.
- (iii) Mouse is mostly preferred animal for studies on gene transfer because
 - A. short oestrous cycle
 - B. long gestation period
 - C. Short generation time
 - D. Production of one or two offspring per pregnancy
 - (a) both (A) and (C)
 - (b) both (A) and (B)
 - (c) only D
 - (d) both (C) and (D).
- (iv) Transgenic genes alter many phenotypic properties including
 - (a) growth rate
 - (b) fat composition
 - (c) milk production
 - (d) all of these.
- (v) **Assertion :** Transgenic mice have been engineered to express human antibodies.
Reason : Large segment of human DNA encoding human immunoglobulin have been transferred to mice.

 - (a) Both assertion and reason are true and reason is the correct explanation of assertion.
 - (b) Both assertion and reason are true but reason is not the correct explanation of assertion.
 - (c) Assertion is true but reason is false.
 - (d) Both assertion and reason are false.

Read the following and answer any four questions from 5(i) to 5(v) given below:

Transgenic cows have extra gene or genes inserted into their DNA. Firstly the genes for the desired product is identified and sequenced. Then a gene construct containing this desired gene is introduced into female cow cells by transfection. Transgenic bovine cells are selected and fused with bovine oocytes that have had all of their chromosomes removed. Once fused with the oocyte, the transgenic cells chromosomes are reprogrammed to direct development into an embryo which can be implanted into a recipient cow. The resulting transgenic cow only express the transgene in her milk. This is because expression of the transgene is controlled by a promoter specific to lactating mammary cells. The first transgenic cow was 'Rosie'.

- (i) The gene construct with desired gene is introduced into female cow cells by
 - (a) transformation
 - (b) transduction
 - (c) transfection
 - (d) transplantation.
- (ii) Production of transgenic cow fulfill the objective of
 - (a) increased milk production
 - (b) increased meat production
 - (c) molecular farming
 - (d) all of these.
- (iii) The name of first transgenic cow is
 - (a) Tracy
 - (b) Dolly
 - (c) Rosie
 - (d) ANDI.
- (iv) Transgenic cow is produced through the implantation of _____ containing transgene into recipient cow.
 - (a) ova
 - (b) embryo
 - (c) mammary cell
 - (d) both (a) and (b)
- (v) **Assertion :** Transgenes only express in the mammary glands of transgenic cow.
Reason : Transgenes are present in chromosomes of every cell in transgenic cow.
 - (a) Both assertion and reason are true and reason is the correct explanation of assertion.
 - (b) Both assertion and reason are true but reason is not the correct explanation of assertion.
 - (c) Assertion is true but reason is false.
 - (d) Both assertion and reason are false.

Read the following and answer any four questions from 6(i) to 6(v) given below:

European patent office, Munich granted patent for fungicidal use of neem oil, to firm of W.R. Grace & Co. It was challenged by Vandana Shiva and Ajay Phadke who had researched neem in India and it was shown that Grace & Co. had not unveiled any novelty factor in neem's properties. Ethics include rules of conduct by which a community regulates the behaviour and decides as to which activity is lawful and which is not.

- (i) _____ allow private, monopoly right over animals and plants.
 - (a) Bioethics
 - (b) Biopatents
 - (c) Bioweapons
 - (d) Either (a) or (b)
- (ii) Which of the following is not a criteria of granting patent?
 - (a) Novelty
 - (b) Utility
 - (c) Non-obviousness
 - (d) Prior art

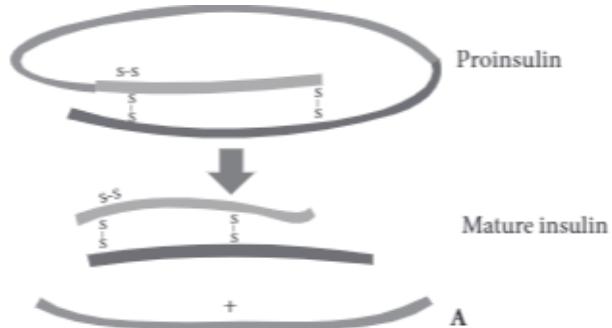
- (iii) On which plants patents have been granted?
- Piper nigrum*
 - Brassica campestris*
 - Punica granatum*
 - All of these
- (iv) Rules of conduct that may be used to regulate our activities in relation to biological world are included in _____.
- bioethics
 - biopiracy
 - biopatents
 - biowar
- (v) Assertion : Genes and cells should not be patented.
Reason : Genes and cells are not an inventions.
- Both assertion and reason are true and reason is the correct explanation of assertion.
 - Both assertion and reason are true but reason is not the correct explanation of assertion.
 - Assertion is true but reason is false.
 - Both assertion and reason are false.

7

Read the following and answer any four questions from 7(i) to 7(v) given below:

Insulin used to cure diabetes was earlier extracted from pancreas of slaughtered cattle and pigs. Insulin extracted from an animal source, though caused some patients to develop allergy or other types of reactions to the foreign protein. Human insulin consists of two short polypeptide chains : chain A and chain B, that are linked together by disulphide bridges. In mammals including humans, insulin is synthesised as a pro-hormone which contains an extra stretch called the C-peptide. This C peptide is not present in mature insulin and is removed during maturation into insulin.

- (i) Identify A in the given figure.



- (a) Polypeptide chain A
(b) Polypeptide chain B
(c) Polypeptide chain C
(d) None of these
- (ii) The following is a list of some stages involved in producing human insulin from genetically engineered bacteria.
- The bacteria are cultured in a fermenter for large scale production.
 - Recombinant insulin is extracted from the bacterial cells that expresses insulin gene.
 - The same restriction enzyme is used again to cut the bacterial plasmid for insertion of the human insulin gene.
 - Bacteria take up the plasmid carrying the insulin gene.
 - A restriction enzyme is used to cut human DNA to extract the insulin gene.
- Select the correct order of these stages.
- 1, 5, 3, 4, 2
 - 2, 4, 3, 5, 1
 - 4, 5, 3, 2, 1
 - 5, 3, 4, 1, 2

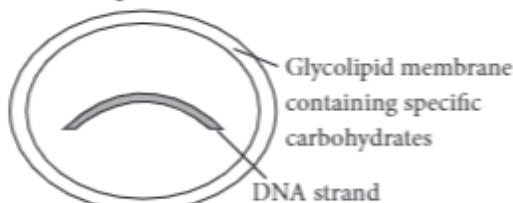
- (iii) To insert the insulin gene into bacterial DNA, both the bacterial plasmid and the human chromosome containing the insulin gene are treated with the same restriction enzyme. Using the same restriction enzyme ensures that
- DNA ligase is able to join the segments of human and bacterial DNA
 - the exact length of nucleotides matching the insulin gene is removed from the plasmid
 - both the bacterial and human DNA will contain sticky ends
 - Sticky ends in the cut plasmid and insulin gene are complementary.
- (iv) Why is the fermentor important for the production of human insulin by transgenic bacteria?
- It provides optimal conditions for the transgenic to multiply rapidly.
 - It facilitates the extraction and purification of insulin from the transgenic bacteria.
 - It maximise the rate of fermentation of the transgenic bacteria.
 - It provides the low-oxygen conditions that are important for insulin production.
- (v) A bacteriologist carries out his first attempt at engineering *E.coli* with the gene for human insulin. During the process, he realises that his stock of DNA ligase has depleted but decides to continue anyway. What is a likely consequence of his decision?
- Bacteria with the rDNA will not be able to form colonies in a fermenter.
 - The resulting plasmids are not able to enter the *E.coli* bacteria even after applying heat shock.
 - The resulting *E.coli* bacteria do not contain the human insulin gene.
 - The bacterial plasmids do not have sticky ends and are unable to accommodate the human gene.

8

Read the following and answer any four questions from 8(i) to 8(v) given below:

Gene therapy is the technique of genetic engineering to replace a faulty gene by a normal healthy functional gene. It is a way of treating genetic disease by introducing a piece of DNA into the cells of an affected individual. Liposomes can be used for gene therapy as they target the cells affected by a genetic disease.

- (i) A patient suffers from adenosine deaminase (ADA) deficiency, an autosomal recessive immune deficiency in which bone marrow lymphoblasts cannot replicate to generate immuno-competent lymphocytes. The treatment option that would cure the patient is
- germ-line gene therapy to replace one ADA gene copy
 - germ-line gene therapy to replace both ADA gene copies
 - somatic cell gene therapy to replace both ADA gene copies in circulating lymphocytes
 - somatic cell gene therapy to replace one ADA gene copy in bone marrow lymphoblasts.
- (ii) Which type of gene therapy is least effective in transferring DNA to the nucleus?
- | | |
|------------------|------------------------------|
| (a) Adenoviruses | (b) DNA attached to a ligand |
| (c) Liposomes | (d) Retroviruses |
- (iii) Liposomes can be used for gene therapy as they target the cells affected by a genetic disease.
The diagram shows the structure of a liposome.



What feature of a cell surface membrane allows the liposome to target cells affected by a genetic disease?

- (a) Carrier molecules (b) Phosphate groups
(c) Protein channels (d) Receptor molecules

(iv) Which of the following disorders would gene therapy be least effective?

(a) Cystic fibrosis (b) Huntington's disease
(c) Sickle cell anaemia (d) Type II diabetes

(v) Following are some reasons explaining why gene therapy is not yet fully an effective treatment for genetic diseases.

I. Lack of knowledge of DNA sequences
II. Lack of efficient and safe gene delivery system
III. Cost of the technology
IV. Some diseases involve multiple genes
V. Problems with appropriate gene regulation

Select the correct option.

(a) I, III and V only (b) II, IV and V only
(c) III, IV and V only (d) I, III and IV only

9

Read the following and answer any four questions from 9(i) to 9(v) given below:

Stem cells hold the potential for manifold applications in biotechnology based next generation therapeutics. Scientists are trying to formulate better and more personalised treatment modalities against some seemingly irremediable diseases, by harnessing body's own stem cells and stem cell niche. Stem cells of different origin and level of potency are being investigated for tissue regeneration, treatment of bone defect, drug testing, gene therapy and cell based therapy for muscle damage, spinal cord injury, cancer therapy, etc. The properties of embryonic and adult stem cells to either self-renew or differentiate into multiple cell lineages make them an attractive source for cell therapies, tissue engineering and as model system for drug screening.

- (i) Stem cells are widely used in medical research. Which property of stem cells makes them particularly useful in this research?

 - (a) They can be fused together to form a zygote.
 - (b) They can divide and eventually give rise to a whole organism.
 - (c) They can divide and be made to differentiate into various types of cell.
 - (d) They will continue to divide indefinitely.

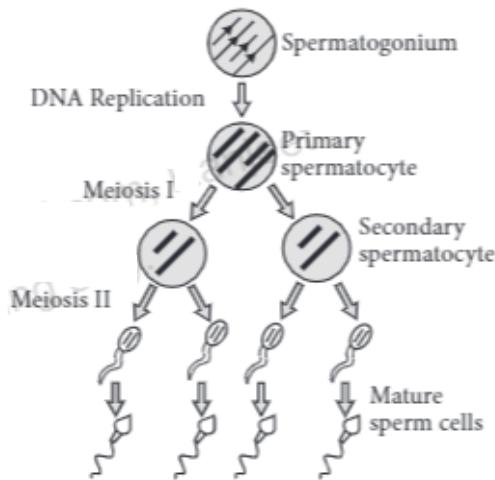
(ii) Stem cells can be divided into four main types. Which of the four types of stem cell can differentiate into a limited range of tissues?

 - (a) Adult stem cells
 - (b) Embryonic stem cells
 - (c) Fetal stem cells
 - (d) Zygotic stem cells

(iii) Which feature of stem cells obtained from blood in the umbilical cord enables their use in the treatment of a variety of blood cancers?

 - (a) They can differentiate into bone marrow cells.
 - (b) They can differentiate into any cellular component of blood.
 - (c) They can replace blood stem cells affected by cancer.
 - (d) They are totipotent.

- (iv) The search for pluripotent stem cells is intense. The spermatogonium is the diploid precursor of haploid sperm in the development pathway of mouse gametes as shown below.

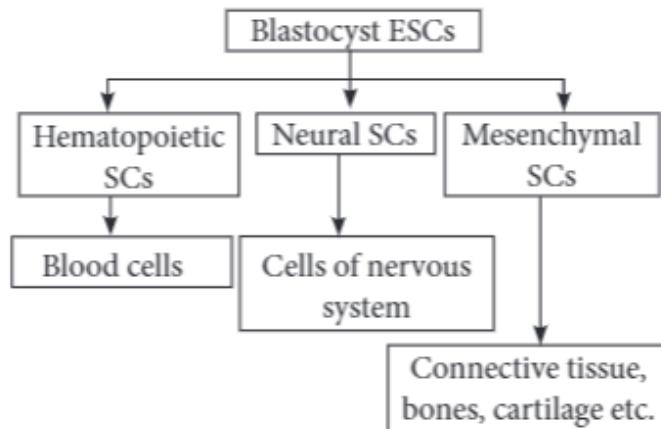


Scientists were interested to find out which stages of sperm development have stem cell capacity. Two experiments were carried out using sterile host males with testes that lacked germ cells. The results are shown in the table.

	Type of cells used	Result
Experiment 1	Secondary spermatocytes were injected	Fertility was not restored
Experiment 2	Spermatogonia were injected	Fertility was restored for the rest of the mouse's life

What property of a stem cell is missing in the secondary spermatocytes?

- (v) Which feature of embryonic stem cells (ESCs) is illustrated below?



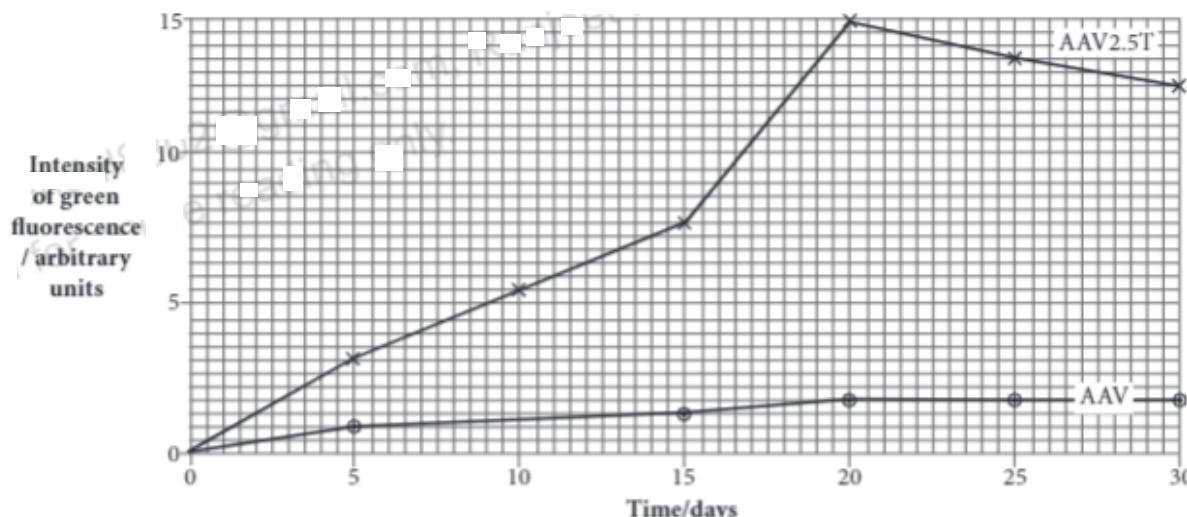
- (a) ESCs are capable of dividing. (b) ESCs are multipotent.
(c) ESCs are pluripotent. (d) ESCs show plasticity.

Read the following and answer any four questions from 10(i) to 10(v) given below:

One approach of gene therapy to treat cystic fibrosis uses viruses to deliver normal alleles of the CFTR gene into epithelial cells of the airways.

A team of researchers in the USA developed a new strain of non-pathogenic adeno-associated virus (AAV), AAV2.5T. Genes for the CFTR protein and the enzyme luciferase were inserted into the DNA of the viruses. Luciferase catalyses the production of a green fluorescent protein when luciferin is added.

The normal AAV strain and the AAV2.5T strain were added to cultures of epithelial cells from the airways. After adding luciferin, the number of cells that had taken up the viral genes was estimated using the intensity of the green fluorescence which developed. The result are shown in the given graph.



- (i) What could be the probable reason for inserting a gene for luciferase by researchers into the viral DNA?
 - (a) Infected cells are able to produce luciferase.
 - (b) It is easy to identify the infected cells that have taken up viral DNA under fluorescent lamp.
 - (c) The non-infected cells were easily identified under fluorescent lamp as they will glow.
 - (d) Both (a) and (b)
- (ii) Select the incorrect statement with respect to the graph given.
 - (a) Both AAV and AAV2.5T can infect epithelial cells.
 - (b) Intensity of green fluorescence increases more in AAV 2.5 T as compared to normal AAV.
 - (c) AAV infect cells more readily than AAV 2.5 T.
 - (d) None of these
- (iii) Why cDNA of the CFTR gene is used in therapeutic instead of normal CFTR gene?
 - (a) cDNA does not contain non-coding regions such as introns.
 - (b) cDNA can be transcribed and translated directly.
 - (c) There is no need of post-transcriptional modification such as splicing, etc.
 - (d) All of these
- (iv) There is a decrease in intensity of green fluorescence in cells infected with AAV 2.5T during the last 10 day. This is because
 - (a) Green fluorescent protein was broken down
 - (b) luciferin was used up
 - (c) infected cells die
 - (d) all of these.
- (v) Which of the following best describe gene therapy?
 - (a) Mutating the sequence of a particular gene.
 - (b) Replacing a healthy gene with a defective gene.
 - (c) Replacing a defective gene with a functional gene.
 - (d) Transferring a healthy gene from one species with healthy gene of other species.

ASSERTION & REASON

For question numbers 11-25, two statements are given-one labelled Assertion and the other labelled Reason. Select the correct answer to these questions from the codes (a), (b), (c) and (d) as given below.

- (a) Both assertion and reason are true and reason is the correct explanation of assertion.
- (b) Both assertion and reason are true but reason is not the correct explanation of assertion.
- (c) Assertion is true but reason is false.
- (d) Both assertion and reason are false

11. **Assertion :** Transgenic food may cause toxicity or produce allergy.

Reason : Transgenic plants have high nutrient content.

12. **Assertion :** Genetically modified microbes help in crop protection.

Reason : Transgenic bacteria control insects by producing endotoxins.

13. **Assertion :** Organs of pig such as heart, pancreas, etc., for human use can be grown through transgenic animals.

Reason : Transgenic pigs show improved growth and meat production.

14. **Assertion :** Mouse is the most preferred mammal for studies on gene transfers.

Reason : Mouse possesses features like short oestrous cycle and gestation period, relatively short generation time, production of several offspring per pregnancy, etc.

15. **Assertion :** *Agrobacterium tumefaciens* is called natural genetic engineer.

Reason : *Agrobacterium tumefaciens* infects all broad-leaved agricultural crops but does not infect cereal crops.

16. **Assertion :** ADA deficiency cannot be cured permanently by gene therapy.

Reason : The genetically engineered lymphocytes can survive only in culture conditions.

17. **Assertion :** PCR is routinely used for early diagnosis of HIV in suspected AIDS patients.

Reason : PCR can detect low amounts of DNA.

18. **Assertion :** Molecular probes are available for diagnosing genetic disorders, e.g., Duchenne muscular dystrophy, cystic fibrosis, Tay-sach's disease.

Reason : The molecular probes are usually double stranded pieces of DNAs, labelled with radio isotopes such as ^{32}P .

19. **Assertion :** In gene therapy, new gene is introduced only into the somatic cells.

Reason : Genetic modification is done to replace faulty genes by normal functional gene.

20. **Assertion :** Biopatents are awarded for biological entities and all products derived from them.

Reason : Patent on use of turmeric in wound healing was cancelled in 2008.

21. **Assertion :** *Agrobacterium tumefaciens* is popular in genetic engineering because this bacterium is associated with the roots of all cereal and pulse crops.

Reason : A gene incorporated in the bacterial chromosomal genome gets automatically transferred to the crop with which the bacterium is associated.

22. **Assertion :** A gene from *Bacillus thuringiensis* is incorporated in plant genome to increase their yield.

Reason : *Bacillus thuringiensis* has Bt toxin producing gene, which kills the larva of insects.

23. **Assertion :** GM crops can affect human health by causing allergic reactions.

Reason : Transgenes in commercial crops can endanger native species, e.g., the Bt toxin gene expressed in pollen might endanger pollinators like honeybees.

24. **Assertion :** GM foods are facing widespread resistance by the people.

Reason : GM foods have mutated genes which cause infections and allergies.

25. **Assertion :** Flavr-Savr tomato was the first transgenic commercial crop that entered the market.

Reason : Roundup variety of soybean was prepared through breeding.

HINTS & EXPLANATIONS

1. (i) (a): Transfer DNA (T-DNA) is the transferred DNA of Ti plasmid of some species of bacteria.
- (ii) (b): *Agrobacterium* infects all broad-leaved agricultural crops. It does not infect cereals.
- (iii) (a): Part of Ti plasmid transferred into the plant cell DNA, is called T-DNA. This T-DNA with desired DNA spliced into it, is inserted into the chromosomes of the host plant where it produces copies of itself, by migrating from one chromosomal position to another at random. But it no longer produces tumors.
- (iv) (d)
- (v) (a): Refer to answer 1(iii).
2. (i) (b): Golden rice is a transgenic variety of rice (*Oryza sativa*) which contains good quantities of β -carotene (provitamin A - inactive state of vitamin A). β -carotene is a principal source of vitamin A. Since the grains (seeds) of the rice are yellow in colour due to β -carotene, the rice is commonly called golden rice.
- (ii) (a): Foreign genes were inserted into the genome of a temperate rice variety by using *Agrobacterium tumefaciens* as the vector.
- (iii) (a): In golden rice, two genes for β -carotene were taken from daffodil (*Narcissus* sp.) and one gene from a bacterium *Erwinia*.
- (iv) (c): Golden rice is rich in β -carotene which is a precursor of vitamin A.
- (v) (b): Prof. Ingo Potrykus and Peter Beyer produced genetically engineered rice by introducing three gene associated with synthesis of carotene. The grains (seeds) of transgenic rice are rich in provitamin.
3. (i) (b): The genetically modified crop called Bt cotton contains Bt toxin genes. The gene cry I Ac and cry II Ab control cotton bollworms. Similarly, cry I Ab has been introduced in Bt corn to protect the same from corn borer.
- (ii) (d): cry I Ab has been introduced to Bt corn to protect them from corn borer. cry I Ac and cry II Ab control cotton bollworms.
- (iii) (d): Gene transfer to non-target plant species is a major disadvantageous concern of GM plants. It can even cause to produce 'superweeds'.
- (iv) (b)
- (v) (a): The Bt toxin proteins exist as inactive protoxins but once an insect ingests the inactive toxin it is converted into an active form of toxin due to the alkaline pH of the alimentary canal that solubilises the crystals. The activated toxin binds to the surface of midgut epithelial cells and create pores which cause cell swelling and lysis and finally cause death of the insect.
4. (i) (d)
- (ii) (c)
- (iii) (a): Mouse has short gestation period and it produces several number of offspring per pregnancy.
- (iv) (d)
- (v) (a)
5. (i) (c)
- (ii) (d): The two chief objectives of transgenic cow production are as follows : (i) increased milk and meat production and (ii) molecular farming.
- (iii) (c)
- (iv) (b): Transgenic bovine cells are selected and fused with bovine oocytes that have had all its chromosomes removed. Once fused with oocyte, the transgenic cells chromosomes are reprogrammed to direct development into embryo which is implanted into recipient cow.
- (v) (b)
6. (i) (b): A patent is the right granted by a government to an inventor to prevent others from commercial use of his invention. Biopatent system allows private, monopoly rights over cells, genes, animals and plants.
- (ii) (d): Patents are supposed to satisfy three criteria of novelty, non-obviousness and utility.
- (iii) (d): Patents have been taken out on plants such as black pepper, basmati rice, Indian mustard, Pomegranate, Turmeric and neem.
- (iv) (a)
- (v) (a): Genes, cells, micro-organisms, plants and animals are not inventions and hence must not be patented.
7. (i) (c): A represents polypeptide chain C which is removed prior to insulin formation.

(ii) (d)

(iii) (d): Each particular restriction enzyme produces unique sticky ends. Using the same enzyme for both the bacterial and human DNA will produce complementary sticky ends that can bind together by complementary base pairing. This would allow the human insulin gene to be inserted into the plasmid.

(iv) (a): The optimal temperature, pH, oxygen and nutrient conditions in the fermenter allow the bacteria containing the insulin gene to reproduce quickly and produce large quantities of it.

(v) (c): DNA ligase forms strong hydrogen bonds between the DNA bases on the human insulin gene and the bacterial plasmid, producing a continuous double stranded DNA loop. Without DNA ligase, the human insulin gene, despite being able to undergo complementary base pairing with the bacterial DNA at the sticky ends would not be securely inserted into the plasmid. Thus, the resulting *E.coli* bacteria would receive plasmids that lack the human insulin gene.

8. (i) (d)

(ii) (b): Viral delivery using adenoviruses and retroviruses are specific and ensure effectiveness. The viral vector recognises and binds to receptor on cell and enters cells. The vector is able to deliver genes to specific target tissues and is able to deliver DNA to the nucleus.

(iii) (d): The liposome's glycolipid membrane containing specific carbohydrates recognises and binds to receptor molecules on cell surface membrane of target cells. The liposome is thus able to deliver genes to specific target cells and is able to deliver DNA into the cell.

(iv) (d): Type II diabetes is a type of multifactorial and polygenic (complex) disorder where manifestation of disease may be dependent on the effects of multiple genes in combination with lifestyle and environmental factors.

(v) (b)

9. (i) (c): Stem cells are capable of dividing indefinitely and producing copies of themselves. In addition, stem cells can undergo differentiation giving rise to specialized cell types.

(ii) (a): Adult stem cells are tissue specific and can enter normal differentiation pathways to form only the specialized cell types of the tissue which they reside in.

(iii) (b): Umbilical cord blood stem cells are multipotent in that they have the ability to differentiate

into limited cell types to give rise to a range of specialized cells that have a specific function. Umbilical cord stem cells found in blood only can differentiate into any cellular component of blood, e.g., white blood cells (such as B lymphocytes, T lymphocytes, natural killer cells, macrophages and platelets etc.) vital for fighting infections and safeguarding the body, red blood cells – important for transporting oxygen to cells.

(iv) (d): Stem cell niche is used to describe the microenvironment in which stem cells are found. Since fertility was restored after the injection of spermatogonia, it suggests that spermatogonium displays the general properties of stem cells such as being capable of dividing indefinitely and producing copies of themselves (self-renewal), can undergo differentiation giving rise to specialised cell types (haploid sperm).

As fertility was not restored upon injection of secondary spermatocytes, it suggest that the secondary spermatocytes do not have a stem cell niche and do not display the properties of stem cell.

(v) (c): Embryonic stem cells are pluripotent cells. That is, they have the ability to develop into almost any kind of cell in the body (as seen from diagram, hematopoietic SCs, neural SCs and mesenchymal SCs) except extra-embryonic membrane (but not the trophoblast).

10. (i) (d): Luciferase catalyses the production of a green fluorescent protein when luciferin is added.

(ii) (c): AAV 2.5T infects cells more readily than AAV.

(iii) (d): cDNA is a DNA copy produced from the mRNA by reverse transcriptase.

(iv) (d)

(v) (c): Gene therapy refers to the insertion of genes into an individual cells and tissues to treat a disease. Essentially defective/mutant genes are replaced with healthy, functional ones.

11. (b): Plant in which foreign genes have been introduced through genetic engineering are called transgenic plants. Transgenic food from such plants may cause toxicity and or produce allergies. The enzyme produced by the antibiotic resistance genes can cause allergies because it is a foreign protein. GM plants enhance nutritional value of food, e.g., Golden rice.

12. (b)

13. (b)

14. (a): Mouse is the most preferred mammal for studies on gene transfers due to its many favourable features like (i) short oestrous cycle and gestation period, (ii) relatively short generation time, (iii) production of several offspring per pregnancy, (iv) convenient *in vitro* fertilisation, successful culture of embryos *in vitro*.

15. (b): Ti plasmid (tumor inducing) from *Agrobacterium tumefaciens* is effectively used as a vector for gene transfer to plant cells. In nature, it induces tumors in broad leaf crops, e.g., tomato, soybean, cotton, etc.

16. (d): As a first step towards gene therapy, lymphocytes, a kind of white blood cells, are extracted from the bone marrow of the patient and are grown in a culture outside the body. A functional ADA cDNA (using a retroviral vector) is then introduced into these lymphocytes, which are reinjected to the patient's bone marrow. But as these cells do not always remain alive, the patient requires periodic infusion of such genetically engineered lymphocytes. However, if the isolated gene from bone marrow cells producing ADA is introduced into cells at early embryonic stages, it can be permanent cure.

17. (b)

18. (c): DNA probes are stretches of single - stranded DNA used to detect presence of complementary nucleic acid sequence (target sequences) by hybridisation. They are usually labelled with radioisotopes, to enable their detection.

19. (b)

20. (c): Biopatents are awarded for biological entities and all products derived from them. 1995 patent on 'use of turmeric in wound healing' was cancelled in 1998.

21. (d): *A. tumefaciens* causes crown gall disease on a wide range of dicot (broad leaved) plants especially members of rose family by transferring its own DNA into plant cells. But in laboratory, the ability to move all sorts of genes into plants has made the microbe

the standard tool for investigating plant genetics and modifying crops.

22. (a): *Bacillus thuringiensis* is a bacterium found in soil worldwide. Several strains can infect and kill insects. Due to this property, through genetic engineering, gene coding for insecticide is incorporated into plant genome. Plant having this gene are insect resistant thus high yielding, e.g., Bt cotton is usually resistant to bollworm disease of cotton. It is the first genetically modified crop of India. This produces its own insecticide, as it contains a gene from soil bacterium *Bacillus thuringiensis*.

23. (b): The transgenic food may cause toxicity or produce allergies. The enzyme produced by the antibiotic resistance can cause allergies, because it is a foreign protein. The introduction of transgenic plants into agriculture is creating some issues. One of them is the potential risk of transgene in commercial crops endangering native or non-target species. For example the gene for Bt toxin expressed in pollen might endanger pollinators like honeybees.

24. (a): The GM crops are fast becoming a part of agriculture throughout the world because of their contribution to the increased crop productivity and to global food, feed and fiber security, besides their use in health-care and industry.

However, GM foods are facing widespread resistance by the people from all over the world. It is because transgenes in commercial crops can endanger native species. For example, the gene for Bt toxin expressed in pollen might endanger pollinators like honeybees.

These crops cause problems in human health by supplying allergens and transfer of antibiotic resistance markers. The GM crops may change the fundamental vegetable nature of plants as the genes from animals (e.g., fish or mouse) are being introduced into crop plants. GM foods also have a bad effect on environment and biodiversity.

25. (c): The Flavr-Savr tomato, was the first genetically modified fruit/vegetable to reach the market in USA. Roundup ready soybean is a genetically modified herbicide tolerant cultivar.