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Chapter <

9

APPLICATIONS OF BIOTECHNOLOGY

CHAPTER SNAPSHOT

- 10.01 Applications in Medicine
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- 10.05 Transgenic Animals
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Evaluation

- 1. The first clinical gene therapy was done for the treatment of
 - (a) AIDS
- (b) Cancer
- (c) Cystic fibrosis
- (d) SCID

[Ans. (c) Cystic fibrosis]

- **2.** Dolly, the sheep was obtained by a technique known as
 - (a) Cloning by gene transfer
 - (b) Cloning without the help of gametes
 - (c) Cloning by tissue culture of somatic cells
 - (d) Cloning by nuclear transfer

[Ans. (d) Cloning by nuclear transfer]

- **3.** The genetic defect adenosine deaminase deficiency may be cured permanently by
 - (a) Enzyme replacement therapy
 - (b) periodic infusion of genetically engineered lymphocytes having ADA cDNA
 - (c) administering adenosine deaminase activators
 - (d) introducing bone marrow cells producing ADA into embryo at an early stage of development.

[Ans. (a) Enzyme replacement therapy]

- 4. How many amino acids are arranged in the two chains of Insulin?
 - (a) Chain A has 12 and Chain B has 13
 - (b) Chain A has 21 and Chain B has 30 aminoacids
 - (c) Chain A has 20 and chain B has 30 amino acids
 - (d) Chain A has 12 and chain B has 20 amino acids.

[Ans. (b) Chain A has 21 and Chain B has 30 amino acids]

- **5.** PCR proceeds in three distinct steps governed by temperature, they are in order of
 - (a) Denaturation, Annealing, Synthesis
 - (b) Synthesis, Annealing, Denaturation'
 - (c) Annealing, Synthesis, Denaturation
 - (d) Denaturation, Synthesis, Annealing

[Ans. (a) Denaturation, Annealing, Synthesis]

- 6. Which one of the following statements is true regarding DNA polymerase used in PCR?
 - (a) It is used to ligate introduced DNA in recipient cells
 - (b) It serves as a selectable marker
 - (c) It is isolated from a Virus
 - (d) It remains active at a high temperature

[Ans. (d) It remains active at a high temperature]

- 7. ELISA is mainly used for
 - (a) Detection of mutations
 - (b) Detection of pathogens
 - (c) Selecting animals having desired traits
 - (d) Selecting plants having desired traits

 [Ans. (b) Detection of pathogens]
- 8. Transgenic animals are those which have
 - (a) Foreign DNA in some of their cells
 - (b) Foreign DNA in all their cells
 - (c) Foreign RNA in some of their cells
 - (d) Foreign RNA in all their cells

[Ans. (a) Foreign DNA in all their cells]

- 9. Recombinant Factor VIII is produced in the _____ cells of the Chinese Hamster.
 - (a) Liver cells
- (b) blood cells
- (c) ovarian cells
- (d) brain cells.

[Ans. (c) ovarian cells]

- 10. Vaccines that use components of a pathogenic organism rather than the whole organism are called
 - (a) Subunit recombinant vaccines
 - (b) attenuated recombinant vaccines
 - (c) DNA vaccines
 - (d) conventional vaccines.

[Ans. (a) Subunit recombinant vaccines]

11. Mention the number of primers required in each cycle of PCR. Write the role of primers and DNA polymerase in PCR. Name the source organism of the DNA polymerase used in PCR.

Ans. The number of primers required in each cycle of PCR are two.

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Role of Primers and DNA Polymerase in PCR:

- (i) During PCR process in the second stage, each separated DNA stand is allowed to hybridize with a primer. This is called renaturation or primer annealing. The primer template is used to synthesize DNA by using Taq-DNA polymerase.
- (ii) Annealing is done by rapid cooling of the mixture allowing the primers to bind to the sequences in each of the two strands flanking the target DNA.
- (iii) During the third stage known as primer extension or synthesis, the Taq DNA polymerase extends each primer by copying the single stranded template.

Source organism of the enzyme DNA polymerase: Escherichia coli

12. How is the amplification of a gene sample of interest carried out using PCR?

- **Ans. (i)** The polymerase chain reaction (PCR) is an in vitro amplification technique used for synthesising multiple identical copies (billions) of DNA of interest.
 - (ii) Denaturation, renaturation or primer annealing and synthesis or primer extension, are the three steps involved in PCR.
 - (iii) The double stranded DNA of interest is denatured to separate into two individual strands by high temperature. This is called denaturation.
 - (iv) Each strand is allowed to hybridize with a primer (renaturation or primer annealing). The primer template is used to synthesize DNA by using Taq DNA polymerase.
 - (v) During denaturation the reaction mixture is heated to 95° C for a short time to denature the target DNA into single strands that will act as a template for DNA synthesis.
 - (vi) Annealing is done by rapid cooling of the mixture, allowing the primers to bind to the sequences on each of the two strands flanking the target DNA.
 - (vii) During primer extension or synthesis the temperature of the mixture is increased

to 75°C for a sufficient period of time to allow Taq DNA polymerase to extend each primer by copying the single stranded template. At the end of incubation both single template strands will be made partially double stranded. The new strand of each double stranded DNA extends to a variable distance downstream.

(viii) These steps are repeated again and again to generate multiple forms of the desired DNA. This process is also called DNA amplification.

13. What is genetically engineered insulin?

Ans. Production of insulin by recombinant DNA technology is called genetically engineered insulin. This involves the insertion of human insulin gene on the plasmids of *Escherichia Coli*. Insulin was the first pharmaceutical product of recombinant DNA technology.

14. Explain how "Rosie" is different from a normal cow.

Ans. In 1997, Rosie, the first transgenic cow produced human protein enriched milk, which contained the human alpha lactalbumin. The protein rich milk (2.4 gm/litre) was a nutritionally balanced food for new born babies than the normal milk produced by the cows.

15. How was Insulin obtained before the advent of rDNA technology? What were the problems encountered?

- **Ans.** In the early years, insulin isolated and purified from the pancreas of pigs and cows was used to treat diabetic patients. Due to minor differences in the structure of the animal insulin as compared to human insulin, it resulted in the occurrence of allergic reactions in some diabetic patients.
- 16. ELISA is a technique based on the principles of antigen-antibody reactions. Can this technique be used in the molecular diagnosis of a genetic disorder such as Phenylketonuria?
- **Ans.** NO. ELISA is based only on antigen/antibody reaction. Polymerase chain reaction technique analyses DNA. Therefore it is ideal to diagnose a genetic disorder such as phenyl ketonuria using PCR.

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ZOOLOGY LONG VERSION QUESTIONS (FOR PURE SCIENCE GROUP)

Q.No. 1 to 3 Refer Evaluation.

4. GEAC stands for

- (a) Genome Engineering Action Committee
- (b) Ground Environment Action Committee
- (c) Genetic Engineering Approval Committee
- (d) Genetic and Environment Approval Committee

[Ans. (c) Genetic Engineering Approval Committee]

- 5. Refer Evaluation Q.No.4
- 6. Refer Evaluation Q.No.5
- 7. Refer Evaluation Q.No.6
- 8. Refer Evaluation Q.No.7
- 9. Refer Evaluation Q.No.8
- 10. Refer Evaluation Q.No.9
- 11. Refer Evaluation Q.No.10
- 12. Refer Evaluation Q.No.11
- 13. Refer Evaluation Q.No.12
- 14. Refer Evaluation Q.No.13
- 15. Refer Evaluation Q.No.14
- 16. Refer Evaluation Q.No.15
- 17. Refer Evaluation Q.No.16
- 18. Refer Evaluation Q.No.17
- 19. Refer Evaluation Q.No.18
- 20. Refer Evaluation Q.No.19
- 21. Refer Evaluation Q.No.20
- 22. Refer Evaluation Q.No.21
- 23. Refer Evaluation Q.No.22
- 24. Refer Evaluation Q.No.23

25. What are the possible risks of GMOs?

- **Ans.** GMOs stands for Genetically modified organisms. The possible risks of GMO's include:
 - (i) Creating new or more vigorous pests and pathogens. Worsening the effects of existing pests through hybridization with related transgenic organisms.
 - (ii) Harming non-target species such as soil organisms, non-pest insects, birds and other animals.
 - (iii) Disrupting biotic communities including agro ecosystems.
 - (iv) Irreparable loss or changes in species diversity or genetic diversity within species.
 - (v) Creating risks for human health.
 - (vi) The release of GMOs into the environment could also have far reaching consequences. This is because the living GMOs proliferate, persist, disperse and sometimes may transfer their DNA into other organisms.
 - (vii) They could also displace the existing organism and create new species which may cause severe environmental damage.
- 26. Refer Evaluation Q.No.24
- 27. Refer Evaluation Q.No.25
- 28. Refer Evaluation Q.No.26
- 29. Refer Evaluation Q.No.27
- **30**. Refer Evaluation Q.No.28
- **31.** Refer Evaluation Q.No.29
- **32.** Refer Evaluation Q.No.30

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Additional Questions

\mathbf{C}	HOOSE THE CORRECT	T ANSWER 1 Mark	9.	Choose the option not applicable to ELISA,
	I. CHOOSE THE C	ORRECT OPTIONS		(a) It is a bio chemical produce to detect
		w Questions:		antigens in a sample.
1	Insulin was first isolated by			(b) The intensity of colour obtained in the test is proportional to amount of antigen.
1.				(c) It can detect DNA
	(a) Banting	(b) Allen		(d) ELISA is a test for detecting human
	(c) Wilmut	(d) Anderson		chorionic gonadotropin hormone
		[Ans. (a) Banting]		[Ans. (c) It can detect DNA]
2 .	_	is a protein with	10	The first living organism to be patented was
	aminoac		10.	The first fiving organism to be patented was
	(a) 120	(b) 133		(a) E.coli (b) Saccharomyces
	(c) 123	(d) 140		(c) Pseudomonas putida (d) Lactobacillus
		[Ans. (c) 123]		Ans. (c) Pseudomonas putida]
3 .	-	uced using	11	
	(a) Pigs	(b) yeast	11.	
	(c) E.coli	(d) viruses		(a) Patents (b) Copy rights (c) GI (d) trademarks
		[Ans. (b) yeast]	((d) tradefliarks [Ans. (c) GI]
4.	The two chains of	f Insulin molecule are		
	attached by		12.	All the following are the functions of insulin.
	(a) Covalent bonds	±		Except.
		(d) both a and b		(a) Insulin controls the level of glucose in the blood.
	[A	ns. (b) disulphide bonds		(b) Insulin facilitates the cellular uptake of
5 .	Best and Banting	isolated insulin from		glucose.
	pancreatic islets of _			(c) Insulin facilitates the utilization of glucose.
	(a) E.coli	(b) yeast		(d) Insulin breaks the glucose which guess
	(c) dog	(d) pig		energy.
		[Ans. (c) dog]		[Ans. (d) Insulin breaks the glucose
6 .	Interferons were disc	covered by		which guess energy]
	(a) Banting	(b) Flemming	13.	Pre-pro insulin is devoid of
	(c) Engvall			(a) C chain (b) disulphide bonds
	(d) Alick Isaacs and I	Lindemann		(c) COOH and NH ₂ group
	[Ans. (d) Alick Isaacs and Lindemann]			(d) Polypeptide chain
7 .	The first synthetic va	accine produced was		[Ans. (b) disulphide bonds]
	(a) DPT	(b) HbsAg	14.	Using recombinant E.Coli, mass production
	(c) Measles	(d) Polio		of hGH is carried out by
		[Ans. (b) HbsAg]		(a) genetic engineering
8.	Myeloid stem cells can differentiate into B and			(b) biotechnology
	T cells only, This is a case of			(c) fermentation technology
	(a) Totipotency	(b) Unipotency		(d) recombinant DNA
	(c) Oligopotency	(d) Multipotency		[Ans. (a) genetic engineering]
	. 01 /	[Ans. (c) Oligopotency]		

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Answer in one word *

1.	First	pharmaceutical	produc	t of	DNA
	recom	binant technology .		Ans.	insulin]

2. Trade name of Human Insulin ______
[Ans. humulin]

3. Number of amino acids in Insulin

[Ans. 51]

- **4.** Abundant protein present in human milk

 [Ans. alpha-lactalbumin]
- **5.** Where is Alpha-lactalbumin synthesized ______ [Ans. mammary glands]
- **6.** Where are the genes for formation of Factor VIII located _____ [Ans. X chromosome]
- 7. Antiviral proteins produced by virus infected cells ______ [Ans. interferons]
- **8.** Vaccines made using genetically modified pathogens _____

[Ans. attenuated recombinant vaccines]

9. Example of a DNA vaccine _____

[Ans. recombinant Hepatitis B vaccine]

10. Organism in which Hepatitis B vaccine is produced _____

[Ans. Saccharomyces cerevisiae]

11. Disease treated using Gene therapy

[Ans. cystic fibrosis]

12. Disease cured by gene therapy for the first time

[Ans. Severe Combined Immunodeficiency Disease (SCID)]

- **13.** Property of stem cells to differentiate into germ layers ______. [Ans. pluripotency]
- **14.** Ability of a single cell to divide and produce all the differentiated cells in an organism ______

[Ans. totipotency]

- **15.** Heating of DNA to separate the strands [Ans. denaturation]
- **16.** A name for genetically modified animals ______ [Ans. transgenic animals]

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- **17.** Technique by which Dolly was developed _____ [Ans. Nuclear transfer technique]
- **18.** Who created Dolly _____

[Ans. Ian Wilmut and Campbell]

19. Who coined the term biotechnology _____

[Ans. Karl Ereky]

VERY SHORT ANSWERS

2 Marks

- 1. Describe the chemical structure of insulin.
- Ans. Human insulin is synthesized by the β cells of Islets of Langerhans in the pancreas. It is formed of 51 aminoacids which are arranged in two polypeptide chains, A and B. The polypeptide chain A has 21 amino acids while the polypeptide chain B has 30 amino acids. Both A and B chains are attached together by disulphide bonds.
- 2. What is alpha lactalbumin?

Ans. Alpha lactalbumin is a protein composed of 123 amino acids. It is the most abundant protein in human milk.

- 3. What is significance of lactalbumin?
- **Ans.** It is a protein which is most abundant in human milk. It is synthesized by the mammary glands. It binds calcium and zinc ions and possesses bactericidal and antitumour properties.
- 4. What is hGH?
- Ans. hGH stands for Human Growth Hormone Somatostatin and Somatotropin are human growth hormones which are peptide hormones produced by the pituitary gland. They are produced by DNA recombinant technology and are used to treat growth disorders in children.
- **5.** What is factor VIII?
- **Ans.** Factor VIII is one of the factors required for natural blood clotting process in human and is located in the X chromosome. A genetic defect in the synthesis of factor **VIII** results in the disease haemophilia, a sex linked disease.

^{*} Only for quick revision not in pattern

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22. What is GI?

Ans. A geographical indication is a name or sign used on products which correspond to a specific geographical origin and possess qualities or a reputation that are due to that origin. Darjeeling Tea was the first GI tagged product in India.

23. What is Trademark?

Ans. Any specific symbol or words to identify a particular product or process of a company constitute trademark. This enables the public to distinguish between a trader's goods from similar goods of other traders.

SHORT ANSWERS

3 Marks

1. What is HbsAg?

Ans. Recombinant Hepatitis B vaccine is a subunit vaccine produced by cloning hepatitis B surface antigen (HbsAg) gene in the yeast *saccharomyces cerevisiae*. It is the first synthetic vaccine to be produced.

2. What is gene therapy? Mention the types.

Ans. It is a connective therapy to cure hereditary diseases and involves the transfer of a normal gene into a person's cells that carries one or more mutant alleles. It is of two types

- (i) Somatic cell gene therapy.
- (ii) Germ line gene therapy.

3. What is gene augmentation therapy?

Ans. Gene augmentation therapy involves insertion of DNA into the genome to replace the missing gene product.

4. Differentiate embryonic and adult stem cells.

Ans.

	S No.	Embryonic Stem Cell	Adult Stem Cells
	1.	They are pluripotent and can differentiate to form the germ layers	They are multipotent and can differentiate into various types of cells that are related.
	2.	They are isolated form blastocyst	Bone marrow is a rich source of adult stem cells.

5. What is a stem cell bank?

- **Ans. (i)** Stem cell banking is the extraction, processing and storage of stem cells, so that they may be used for treatment in the future, when required.
 - (ii) Amniotic cell bank is a facility that stores stem cells derived from amniotic fluid for future use.
 - (iii) Cord blood banking is the extraction of stem cells from the umbilical cord during childbirth.

6. What is the advantage of PCR and ELISA over conventional diagnostic methods.

Ans. Presence of pathogens like virus, bacteria, etc., is detected only when the pathogen produces symptoms in the patient. By the time the symptoms appear, concentration of pathogen becomes very high in the body. However very low concentration of a bacteria or a virus, even when the symptoms of the disease does not appear, can be detected by amplification of their nucleic acid. Thus PCR and ELISA help in early diagnosis.

7. What is the application of PCR in forensic science?

Ans. PCR technique can be used in the field of forensic medicine. A single molecule of DNA from blood stains, hair, semen of an individual is adequate for amplification by PCR. The amplified DNA is used to develop DNA fingerprint which is used as an important tool in forensic science. Thus, PCR is very useful for identification of criminals.

8. List the criteria for patenting.

Ans. The following criteria must be satisfied for patenting:

- (i) The invention must be novel and useful;
- (ii) The product must be inventive and reproducible;
- (iii) The patent application should provide the full description of the invention and the invention must be patentable.

9. Why is *E.Coli* not used for production of Interferons?

Ans. The yeast *Saccharomyces cerevisiae* is more suitable for production of recombinant interferons than *E.coli*, since *E.coli* does not possess the machinery for glycosylation of proteins.

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10. Can interferons be isolated from blood.

Ans. Yes but the amount of blood required for isolation of interferons is enormous. Thus they are produced by rDNA technology.

11. Name some diseases that can be treated using interferons.

Ans. Cancer, AIDS, multiple sclerosis, hepatitis C and herpes zoster.

12. Can DNA vaccine cause the disease?

Ans. The DNA vaccine cannot cause the disease as it contains only copies of a few of the genes of the pathogen.

13. Name the strategies used in gene therapy.

- **Ans. (i)** Gene augmentation therapy Insertion of DNA into the genome to replace the missing gene product
 - (ii) Gene inhibition therapy Insertion of the anti sense gene which inhibits the expression of the dominant gene.

14. What are the precautions to be taken in gene therapy?

- Ans. (i) The gene must be harmless to the patient.
 - (ii) Gene must be appropriately expressed.
 - (iii) Body's Immune system does not react to the foreign proteins produced by the new genes.

15. What is oligopotency?

- **Ans.** (i) Oligopotency refers to stem cells that can differentiate into few cell types.
 - (ii) For example lymphoid or myeloid stem cells can differentiate into B and T cells but not RBC.

16. GMO's are sometimes a threat to environment. Why are they produced?

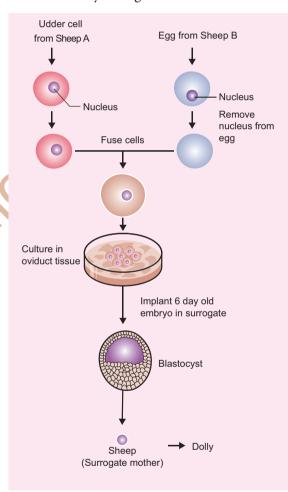
- Ans. (i) Genetically Modified Organisms (GMOs) also called Genetically Engineered organisms (GEOs) are created to play a role in agriculture, forestry, aquaculture, bioremediation and environmental management in developed and developing countries.
 - (ii) However, deliberate or inadvertent release of GMOs into the environment could have negative ecological effects under certain circumstances.

LONG ANSWERS

5 Marks

1. Explain the creation of Dolly.

Ans. (i) Dolly was the first mammal (Sheep) clone developed by Ian Wilmut and Campbell in 1997. Dolly, the transgenic clone was developed by the nuclear transfer technique and the phenomenon of totipotency. Totipotency refers to the potential of a cell to develop different cells, tissues, organs and finally an organism.



Cloning of dolly

(ii) The mammary gland udder cells (somatic cells) from a donor sheep (ewe) were isolated and subjected to starvation for 5 days. The udder cells could not undergo normal growth cycle, entered a dormant stage and became totipotent. An ovum (egg cell) was taken from another sheep (ewe)

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and its nucleus was removed to form an enucleated ovum. The dormant mammary gland cell/udder cell and the enucleated ovum were fused. The outer membrane of the mammary cell was ruptured allowing the ovum to envelope the nucleus. The fused cell was implanted into another ewe which served as a surrogate mother. Five months later dolly was born. Dolly was the first animal to be cloned from a differentiated somatic cell taken from an adult animal without the process of fertilization.

2. List the uses of transgenesis.

- **Ans. (i)** Transgenesis is a powerful tool to study gene expression and developmental processes in higher organisms.
 - (ii) Transgenesis helps in the improvement of genetic characters in animals. Transgenic animals serve as good models for understanding human diseases which

- help in the new treatments for diseases. Transgenic models exist for many human diseases such as cancer, Alzheimer's, cystic fibrosis, rheumatoid arthritis and sickle cell anemia.
- (iii) Transgenic animals are used to produce proteins which are important for medical and pharmaceutical applications.
- (iv) Transgenic mice are used for testing the safety of vaccines.
- (v) Transgenic animals are used for testing toxicity in animals that carry genes which make them sensitive to toxic substances than non-transgenic animals exposed to toxic substances and their effects are studied.
- (vi) Transgenesis is important for improving the quality and quantity of milk, meat, eggs and wool production in addition to testing drug resistance.

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