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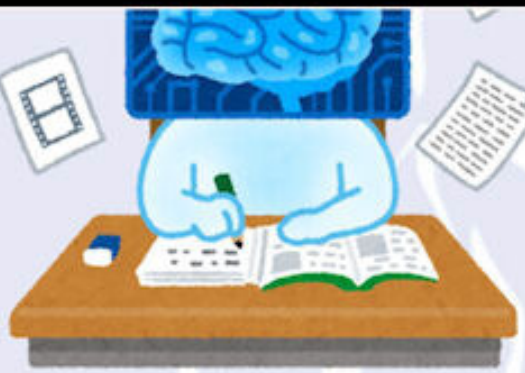
Science & Tech Preview



SCIENCE & TECHNOLOGY

Prelims & Mains

For Civil Services Exams

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Chapter 1 Biotechnology

Meaning

The word 'biotechnology' is made up of two words: 'bio' and 'technology'. 'Bio' means life, and 'technology' means applying or harnessing of science for a specific purpose. Therefore, the term 'biotechnology' refers to the modification or use of any living organism for any useful purpose.

The term 'biotechnology' is largely believed to have been coined in 1919 by the Hungarian engineer, Károly Ereky. In the late twentieth and early twenty-first centuries, biotechnology has expanded to include new and diverse sciences, which we will discuss in this chapter. Even after so many advances, the Recombinant DNA Technology forms the basis of biotechnology. Let us first understand this technology.

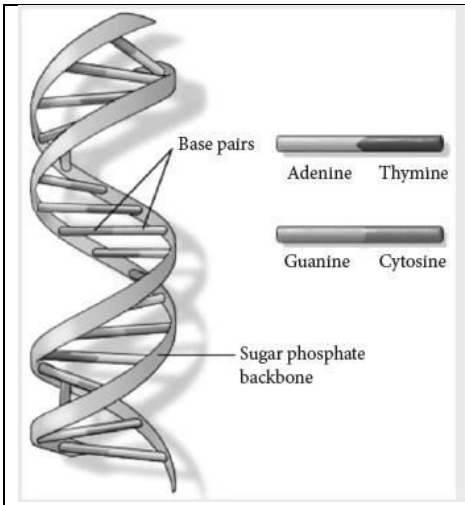
Recombinant DNA Technology

The recombinant DNA technology is a technology through which a foreign gene of an organism is inserted into a host organism to produce desired qualities in the host organism. Such a foreign gene may be acquired even from an organism that is unrelated to the host organism.

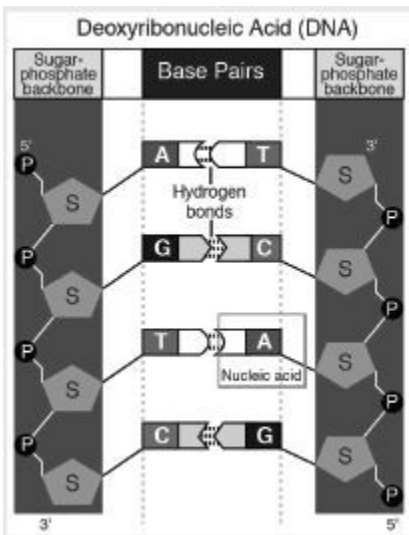
What Is DNA?

Deoxyribonucleic acid (DNA) is the genetic material typically found in all living cells of humans as well as animals and plants. It contains hereditary data passed on from parents to their children, which is unique to each person (except in the case of identical twins). DNA is a double helix structure (helix structure means shape of spiral staircase) and consists of base pairs formed from four bases- adenine (A), guanine (G), cytosine (C), and thymine (T).

Each base is attached to a sugar phosphate backbone (structure formed from phosphate and a sugar molecule). A sugar molecule, phosphate molecule, and a base together form a nucleotide. The nucleotides are arranged in two long strands to form a double helix structure, as shown in the figure.



When the base pairs are formed, the bonding between the bases causes the DNA strands to spiral around each other, forming a double helix shape. Each base pair is made up of two bases. Adenine always pairs with Thymine, and Cytosine always pairs with Guanine to form a base pair.



DNA has the capability to replicate itself. It follows the pattern in a DNA strand to create identical DNA. In other words, DNA replicates by duplicating the sequence of bases.

DNA and RNA

DNA, or deoxyribonucleic acid, is like a genetic blueprint of guidelines that a living organism must follow to exist and remain functional. RNA, or ribonucleic acid, helps

carry out this blueprint's guidelines. It is complementary to DNA, helping carry out the tasks encoded in DNA.

DNA is more stable and holds more complex information for longer periods of time. On the other hand, RNA is more flexible and capable of performing different tasks. DNA is found in the nucleus of a cell (nuclear DNA) and mitochondria (mitochondrial DNA). Mitochondria is the part of the cell responsible for generation of energy. It has a fixed double helix structure. The RNA does not have a fixed location in a cell and adopts different structures depending on the role it is to play—as messenger RNA (mRNA), transfer RNA (tRNA), or ribosomal RNA (rRNA).

Messenger RNA (mRNA): Messenger RNA (mRNA) transcribes genetic information from the DNA found in a cell's nucleus and then carries this information to other cell organelles, specifically ribosome (site of protein manufacture). The full range of messenger RNA, or mRNA, molecules expressed by an organism is transcriptome. An organism's transcriptome varies depending on many factors, including the stage of development and environmental conditions. In contrast with the genome, which is characterised by its stability, the transcriptome actively changes.

Transfer RNA (tRNA): Transfer RNA (tRNA) is found in a cell's cytoplasm (fluid that fills the inside of the cell and surrounds all the cell organelles) and is closely related to mRNA as its helper. tRNA transfers amino acids, the core components of proteins, to the mRNA in a ribosome.

Ribosomal RNA (rRNA): Ribosomal RNA (rRNA) is also found in a cell's cytoplasm. In the ribosome, it takes mRNA and tRNA and translates the information they provide. From this information, it 'learns' whether it should create protein.

DNA genes are expressed or manifested through the proteins which are produced with the help of RNA. Traits (phenotypes) come from which proteins are made and which are switched on or off. The information found in DNA determines which traits are to be created, activated, or deactivated, while the various forms of RNA do the work.

Steps in rDNA Technology

The following are the basic steps involved in Recombinant DNA technology:

1. Insert is isolated. The DNA fragment that contains the gene of interest and needs to be inserted into a host organism is called the Insert.
2. This DNA fragment is inserted into a carrier DNA molecule and a recombinant DNA

molecule is thus generated. The carrier DNA molecule is called the vector. The rDNA thus generated has the ability to self-replicate within a host cell.

3. The third step involves a process called transformation. In this, the rDNA generated in the earlier step is transferred into an E. coli (Escherichia coli) bacteria host cell.
4. The host cells carrying the rDNA are selectively allowed to multiply. This trend generates more rDNA molecules.

This leads to generation of a large amount of rDNA, and hence, gene cloning is achieved. To successfully carry out the process, we need an insert and vector and a method to precisely cut and join the DNA molecules. The joining of the DNA molecules together is called ligation.

Tools Used in rDNA Technology

The tools generally used in rDNA technology are as follows:

Restriction Enzymes: Restriction enzymes act as the molecular scissors. In other words, these enzymes can be used to precisely cut a DNA molecule at the required location. These enzymes are derived from bacteria. In bacteria, these enzymes are naturally present as part of its defense mechanism called the Restriction-Modification System. These restriction (restriction endonucleases) and modification enzymes (methylases) are unique to each bacterial species.

Restriction-modification System

The Restriction-Modification System in bacteria consists of two components: a restriction enzyme and a modification enzyme.

The restriction enzyme selectively recognises a particular sequence of DNA. Once it recognises such a sequence in a DNA, it digests the DNA fragment containing that sequence. In other words, it restricts that particular sequence of DNA from propagation by digesting the DNA fragment. This helps the bacteria to protect itself from any foreign DNA (such as virus).

The modification enzyme, on the other hand, recognises a particular sequence and adds a methyl group to one or two bases in the DNA sequence. This is called methylation. A methyl group is derived from methane, containing one carbon atom bonded to three hydrogen atoms— CH_3.

Once methylation is done, that particular sequence cannot be digested by the bacteria. This enzyme adds methyl group only to Bacteria's own DNA and excludes foreign DNA from methylation. As the foreign DNA is not protected by methylation, it

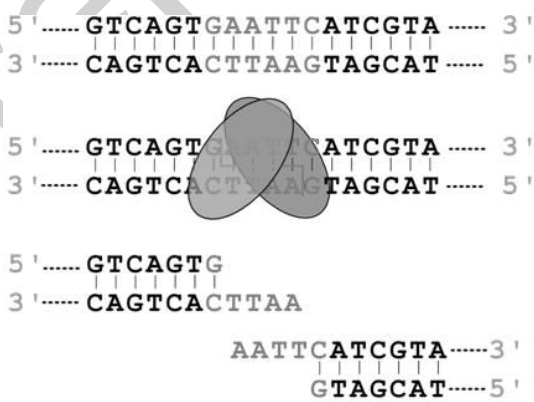
can be easily digested. In simple terms, the modification enzyme selectively protects a Bacteria's DNA and leaves foreign DNA, which is then digested by the restriction enzyme.

Each species of bacteria have their own set of restriction-modification enzymes. The three main classes of restriction endonucleases are named type I, type II, and type III. Of these three types, only type II restriction enzymes are used in rDNA technology as they can be used to recognise and cut the DNA in the required specific sequence.

These type II enzymes are sourced from different microbial sources. The genetic engineers choose a particular enzyme based on the requirement (i.e., the location where the DNA needs to be cut). There are more than a thousand restriction enzymes available at present, from which a restricted enzyme can be chosen.

The Type II restriction enzymes are named after the bacterial species from which they are isolated. EcoRI, which is isolated from the E.coli bacteria, is the most commonly used restriction enzyme. As the name indicates, E stands for the genus, co stands for the species, and 'R' refers to the strain RY 13. The roman numeral 'I' indicates that this is the first enzyme to be isolated from this strain of bacteria.

Some of the restriction enzymes (Type II) and their sources are given in the table below after the diagram. The location at which a restriction enzyme can cut the DNA is indicated by the arrow symbols in the recognition sequence. Any DNA that matches this recognition sequence (DNA in which base pairs are similarly arranged) will be digested by the restriction enzyme of the bacteria. For instance, EcoRI cuts the GAATTC sequence, as shown in the figure. The enzyme is shown in grey colour and oval shape.



Restriction enzyme	Microbial source	Recognition sequence
Alu I	Arthrobacter luteus	↓ 5'A-G-C-T 3' 3'T-C-G-A 5' ↑
BamHI	Bacillus amyloliquefaciens	↓ 5'G-G-A-T-C-C 3' 3'C-C-T-A-G-G 5' ↑
EcoRI	Escherichia coli	↓ 5'G-A-A-T-T-C 3' 3'C-T-T-A-A-G 5' ↑
EcoRII	Escherichia coli	↓ 5'C-C-T-G-G 3' 3'G-G-A-C-C 5' ↑

What Are 5′ and 3′ in a DNA Sequence?
The DNA direction is indicated by using 5′ and 3′ which are called "five prime" and "three prime". These numbers indicate the location of a phosphate group (phosphate group is attached to 5′ carbon) and the hydroxyl group (attached to 3′ carbon) attached to the sugar molecule. In other words, 5′ and 3′ are used to indicate how the base pairs are arranged in a DNA strand. They are written in a 5′- 3′ pattern as DNA synthesis always happens from 5′ to 3′.

DNA ligase: DNA Ligase is another enzyme that plays an important role in the rDNA technology. It is used to connect two strands of DNA together by forming a covalent bond between the two. The phosphate group on one DNA strand is connected with the deoxyribose group on the other DNA strand. Deoxyribose is the five-carbon sugar molecule that helps form the phosphate backbone of DNA molecules. The bonds that are formed as a result are called the phosphodiester bonds. One of the most frequently used DNA ligases is isolated from the bacteriophage T4 (bacteriophages are viruses that infect bacterial cells).

Alkaline phosphatase: Generally, in ligation, a 5′phosphate group of one strand joins with the other strand containing a 3′hydroxyl group. The presence of a phosphate group is essential to carry out ligation. In some cases, the same DNA fragments' phosphate group may join with its own hydroxyl group. This is called self-ligation,

which needs to be avoided to generate the required rDNA. Hence, the DNA fragments are treated with an enzyme called alkaline phosphatase. This removes the phosphate groups from the DNA fragments. This ensures that the fragments cannot ligate within themselves and are forced to ligate with other fragments containing the 5'phospahte groups.

Vectors: Vectors are DNA molecules that serve as a vehicle to carry the required foreign DNA sequence into a host cell. The vector must contain an origin of replication (a particular sequence in a DNA molecule at which replication is initiated) so that it can replicate independently within the host. This ensures that the foreign insert carried by the vector is automatically replicated. Along with this, a vector should also have a selectable marker. Selectable marker is a gene that is used to identify the host cells containing the vector.

The procedure that involves transformation of cells using rDNA technology is around 1% efficient. We need to identify the cells where the procedure is successful and isolate them. Selectable markers are used to identify these cells.

For instance, genes that confer antibiotic resistance and enzymes such as β-galactosidase can turn the substrates blue in the host cell colony. Similarly, a gene that has Green Fluorescent Protein exhibits green fluorescence when viewed under UV light. Let's consider that we have used such a vector to make the required rDNA. To check whether the multiplication of the required genes is successful, we can observe the host cells under UV light. If we find more genes exhibiting fluorescence, this means the presence of the vector DNA has increased, which indicates that the multiplication of the foreign DNA is successful. Using selective markers, the transformed cells can be easily identified. These transformed cells can then be selected for growth and division.

APPLICATIONS OF BIOTECHNOLOGY

1. Green Biotechnology

The use of biotechnology in the field of agriculture is called green biotechnology. One of the important applications under green biotechnology is the development of transgenic plants.

What are transgenic plants? In a transgenic crop plant, one or more genes are artificially inserted instead of the plant acquiring such genes through pollination. The inserted gene sequence (transgene) may come either from another unrelated plant or from a completely different species. Plants containing transgenes are often called

genetically modified (GM)crops.

For instance, *Bacillus thuringiensis* (Bt) cotton is a transgenic crop developed with the help of particular genetic engineering technology popularly known as Bollgard technology. In the Bollgard-I technology, 'Cry1Ac' gene of Bt is introduced to make the crop pest-resistant. After the introduction of this gene, the cotton crop starts producing its own pesticide. The Bt trait has been believed to save the cotton plant from the pest, which is popularly known as 'ball worm'. Bollgard-II technology is a superior double- gene technology—Cry1Ac and Cry 2Ab, which provide protection against bollworms and Spodoptera caterpillar, respectively, are introduced to the cotton plant.

Bacillus thuringiensis (Bt)
Bacillus thuringiensis (Bt) is a soil-dwelling bacterium, commonly used as a biological pesticide. It also occurs naturally in the gut of moths and butterflies, on leaf surfaces, etc. It is deliberately used in flour mills and in grain-storage facilities.

Why should we make transgenic crop plants? A plant breeder tries to accumulate a combination of genes in a crop plant so as to make it useful and more productive as far as possible.

Desirable genes may provide the following features:

1. **Increased crop production:** Transgenic plants, which have been developed to produce higher yields, are tolerant to diseases, drought, etc., such crop attributes have facilitated increased crop production.

2. **Improved nutritional value:** Transgenic plants with higher nutritional value have been produced. For instance, golden rice is a transgenic variety of rice, with genes for the synthesis of beta-carotene (β-carotene) taken from daffodil plant and inserted into rice. The β-carotene is converted into vitamin A. Thus, golden rice helps in preventing nutritional blindness in people, which occurs due to the deficiency of vitamin A, and is also responsible for the normal functioning of the immune system. The golden rice is called so because the rice grain is pale yellow in colour, instead of pearly white. The colour is due to the presence of β-carotene.

3. **Increased shelf life:** Transgenic plants have been developed with longer shelf life, making storage and transportation easier.

4. **Environmental benefits:** Transgenic varieties rely on reduced consumption of

pesticides. Consequently, there is fewer pesticide residues in foods, reduction of pesticide leaching into groundwater, and minimization of the farm worker's exposure to hazardous products.

Comparison with traditional plant breeding

Traditional plant breeding has been limited to artificially crossing plants within the same species or closely related species to bring different genes together through selective or mutation breeding techniques. For example, a gene for protein in soya bean could not be transferred to a completely different crop such as wheat using traditional techniques.

Transgenic technology enables plant breeders to bring useful genes together in one plant from a wide range of living sources. Thus, it expands the possibilities beyond the limitations imposed by traditional cross-pollination and selection techniques.

How are transgenic plants developed? Transgenic plants are the plants that have been genetically engineered, a breeding approach that uses recombinant DNA techniques to create plants with desired characteristics. Genetic engineering is the process by which scientists modify the genome (the complete set of genes or genetic material present in a cell or organism) of an organism. The creation of genetically modified organisms requires recombinant DNA.

This technology provides the means for identifying and isolating the genes controlling specific characteristics in one kind of organism and for moving copies of those genes into another different organism, which will also have those characteristics. This powerful tool enables the plant breeders to generate more useful and productive crop and animal varieties containing new combinations of genes.

In other words, transgenic plants are obtained through recombinant DNA technology or genetic engineering. Recombinant DNA technology is a technology through which a foreign gene of an organism is inserted into a host organism to produce desired qualities in the host organism. Such a foreign gene may be acquired even from an organism that is unrelated to the host organism.

Pest Resistant Plants: Green Biotechnology is also used for the development of pest-resistant plants. For instance, a process called RNA interference (RNAi) was used to develop a pest resistant tobacco plant. In this process, a specific mRNA (messenger RNA) is prevented from undergoing translation (translation is a process of creating proteins from messenger RNA or mRNA). RNA interference can also be induced

experimentally by using a double stranded RNA molecule. The double stranded RNA molecule binds to the specific mRNA, and hence prevents its translation. This is called silencing of mRNA. This technique was used to develop a pest resistant tobacco plant.

A nematode (roundworm) known as Meloidegyne incognitia infects the roots of tobacco plants, thereby causing a reduction in the yield. Nematode-specific genes introduced into the host plant produce a double stranded RNA (dsRNA) that initiates RNAi. The dsRNA binds to the mRNA of the pest and silences it. As a result, the pest cannot survive in the host plant. In other words, the pest resistant tobacco plant is developed in such a way that the parasite could not survive in these tobacco plants.

Red Biotechnology

The use of biotechnology in the field of medicine is called red biotechnology. Red biotechnology is used in the following medical processes:

1. **Gene therapy:** It is a subdivision of red biotechnology that deals with the diagnosis and treatment of genetic diseases and other diseases that are related to genetic makeup of an individual. Modern life science considers genetic makeup important in diseases even associated with heart or cancer. In gene therapy, the treatment revolves around the manipulation or replacement of defective genes. It may also involve insertion of missing genes.

2. **Pharmacogenomics:** This field is a combination of genetics and pharmaceuticals. Pharmacogenomics analyses how genetic makeup affects an individual's response to drugs. It deals with the influence of genetic variation of drug response in patients by correlating gene expression with the efficacy or toxicity of a drug.

   Pharmacogenomics offers new possibilities in the design and production of drugs and how they can be adapted to each individual and their genetic makeup. Specifically, applications in pharmacogenomics have resulted in the development of custom drugs, accurate dosages for different individuals, etc.

3. **Genetic testing (or genetic screening):** Genetic testing allows diagnosis of genetic vulnerabilities leading to inherited diseases and can also be used to determine a child's parentage (genetic mother and father) or in general, a person's ancestry.

   Genetic testing, in a broader sense, includes biochemical tests for the possible presence of genetic diseases or changes that are associated with inherited disorders, or mutant forms of genes associated with increased risk of developing genetic

disorders. Since genetic testing revelations may lead to psychological problems, genetic testing is often accompanied by genetic counselling.

4. **Drug administration:** Biotechnology has contributed to the discovery and manufacturing of pharmaceutical drugs as well as drugs that are the product of biotechnology, which are called biopharmaceutics.

   Biopharmaceutics examines the interrelationship of the physical/chemical properties of the drug, the dosage form (drug product), and the manner of drug administration on the rate and extent of drug absorption. Thus, biopharmaceutics has helped in enhancing the therapeutic effect of a drug.

   Biotechnology has also helped in the field of **pharmacokinetics**. It is described as what the body does to a drug, refers to the movement of drug into, through, and out of the body—the time course from its absorption to excretion.

5. **Virotherapy:** It is a medical treatment that uses biotechnology to convert viruses into therapeutic agents. The viruses are genetically modified to treat diseases. There are three main branches of virotherapy: anti-cancer oncolytic viruses, viral vectors for gene therapy, and viral immunotherapy.

6. **Molecular Diagnosis:** Molecular diagnosis refers to the process of identifying a disease by studying molecules such as DNA, RNA, and proteins in a tissue. Molecular diagnosis helps in early detection of disease, even before the symptoms have appeared. Early diagnosis of disease is essential for effective treatment of a disease. In other words, detecting a disease only after the symptoms manifest may make the treatment difficult. By the time symptoms manifest, the concentration of the disease-causing pathogen (virus, bacteria, etc) is already very high in the body. Conventional methods such as urine or serum analysis cannot detect bacteria at low concentrations.

   Using biotechnology, techniques such as Polymerase Chain Reaction (PCR) and Enzyme-linked Immuno-sorbent Assay (ELISA) make the early detection of diseases possible.

   PCR

   In PCR, DNA from the sample is amplified and observed for the presence of pathogens. How does this work? A segment of DNA is collected from the patient and heated, causing the DNA to separate into two strands. This is called DNA denaturation. An enzyme known as 'Taq polymerase' (obtained from a bacteria called Thermus

aquaticus) synthesises DNA from each strand. In other words, from a single DNA, two DNAs are produced. The DNA duplication continues, and in a few rounds, 1000s of DNA are produced. In other words, very low concentrations of pathogens are amplified and detected using PCR. It is generally used to test for HIV, gene mutations, or genetic disorders in suspected patients.

ELISA

Enzyme-linked Immuno-sorbent Assay (ELISA) test is used to diagnose a disease by identifying and measuring antibodies in blood. Antibodies are proteins produced in the body to fight against antigens. Antigen is a substance that is not recognised by the immune system. Each type of infection is characterised by a specific antigen. The immune system tries to fight against the infection by producing antibodies. In other words, the presence of antibodies indicates the presence of infection. In ELISA, the antigen (related to the condition for which the patient is being tested) is added to the blood sample. If antibodies are present in the blood against this particular antigen, they bind together, and the colour of the sample changes. The change in colour determines the presence of a particular pathogen. ELISA can be used to diagnose infections such as HIV, Lyme disease, rotavirus, syphilis, Zika virus, etc.

Genetically Engineered Insulin

Animal-sourced insulin (extracted from the pancreas of slaughtered cattle and pigs) was used to manage diabetes for many years before the advent of genetically-engineered insulin. Using genetic engineering, insulin is being produced from yeast and bacteria. A small piece of DNA called plasmid is extracted (this acts as the vector) from a bacteria (such as E. coli) or yeast cell and cut using the restriction enzymes. The plasmid is then modified by inserting the human insulin gene into the gap within the plasmid. The modified plasmid is then introduced into a host (new yeast or bacteria cell). The cell divides in the host and makes insulin. Large amounts of insulin can be manufactured by using this process.

Can Biotechnology Control Dengue and Chikungunya?

Trials are underway to develop transgenic Aedes aegypti variety in Jalna, Maharashtra, which will help in controlling dengue and chikungunya.

How Will This New Variety Reduce Disease?

Aedes aegypti vector mosquitoes, responsible for spreading dengue and chikungunya, among other diseases, are engineered through advanced biotechnology to be self-limiting—in other words, genetically modified so that their offspring will die.

Why Are Transgenic Mosquitoes Certain to Work?

The 'friendly Aedes' (modified Aedes aegypti) has already been trademarked by Oxitec (the research unit of Oxford University). These are transgenic male mosquitoes with a self-limiting gene inserted through advanced genetics. Banking upon the male's natural instinct to mate with a wild female, the OX513A strain is inherited by the offspring, causing the larvae to die before they mature to become adult mosquitoes.

White Biotechnology

The use of biotechnology in industry is regarded as white biotechnology. It helps to improve industrial processes and create new industrial products.

The uses of white biotechnology are mentioned below:

1. Biotechnology is used to develop microorganisms that can increase the rate of fermentation of organic matter in order to convert it into alcohol, acids, and biomass.
2. It is used to enhance oil recovery from its well. Genetically modified organisms by consuming dense hydrocarbons can reduce the surface tension of the oil to a greater extent and hence facilitate easy recovery of oil.
3. It is used to produce microorganisms, which can act as preservatives for perishable products.
4. It is used to produce biofuels, a renewable source of energy.

Use of Biological Processes for Extraction of Natural Resources

The following biological processes can be used for the extraction of natural resources:

1. **Biosorption:** It is a property of certain types of inactive, dead, microbial biomass to bind and concentrate heavy metals from even very dilute aqueous solutions.

2. **Biomining**: It is an approach for the extraction of desired minerals from ores with the help of living organisms. Biomining can be undertaken through microbes (microbial mining) or plants (phytomining).

3. **Bioleaching:** Microorganisms are used to leach out the minerals, rather than the traditional methods of extreme heat or toxic chemicals, which have a deleterious effect on the environment. Bioleaching is a sub-type of biomining. It is widely used as an extractive metallurgy technique which converts metal into soluble salts in aqueous media.

4. **Phytomining**: Phytomining is an approach in which mining is done with the help of plants. For instance, some plants absorb copper compounds through their roots. As a result, copper compounds remain concentrated in their roots. The plants can be

burned to produce ash that contains copper.

Blue Biotechnology

Blue biotechnology deals with the aquatic environment (along with marine organisms) to generate new sources of energy, develop new drugs, extract useful resources, or develop new varieties of marine organisms.

Uses of blue biotechnology Biotechnology can be used to develop microorganisms to clean water bodies. For instance, oil spills can occur both over land as well as over water bodies. Oil spills over water bodies are more dangerous as the oil layer prevents the penetration of sunlight, leading to reduction of photosynthesis activity. Consequently, availability of oxygen in the water bodies is reduced, leading to the death of marine animals.

Oil zapper is a mixture of five types of bacteria which feed on hydrocarbon compounds present in the crude oil. The hazardous hydrocarbon waste generated by oil refineries is known as oil sludge.

Oil zapper converts hydrocarbons into CO_2 and H_2O. Oil Zapper consists of Oil Zapping bacteria, which are immobilised and packed into polythene bags. The shelf-life of oil zapper is three months. Oil zapper is used to clean oil spills.

Biotechnology can create transgenic aquatic organisms with desirable features. Transgenic aquatic organisms are those in which a foreign gene is added to the organisms to produce the desired qualities.

Uses of Biotechnology in Environment

Bioremediation

It refers to the cleaning of environment with the help of living organisms. Living organisms range from microorganisms to different species of plants. For example, bacteria help in the decomposition of organic waste, and certain plant species such as mustard helps in the absorption of poisonous elements such as selenium.

Bioremediation usually takes a longer time period. However, bioremediation effectively discriminates between pollutants and the required nutrients.

Strategies of bioremediation are mentioned below:

In situ bioremediation techniques

It refers to the treatment of waste at its site. These techniques not only assist in the degradation of adsorbed fuel residuals but also assist in the degradation of volatile organic compounds. In situ bioremediation techniques include biosparging, bioventing, bioaugmentation, and bioculture.

1. **Biosparging:** It is an in-situ remediation technology that uses indigenous microorganisms to biodegrade organic constituents in the saturated zone. In biosparging, air (or oxygen) and nutrients are injected at high pressure to increase the biological activity of the indigenous microorganisms and to enhance their decomposition activity.

2. **Bioventing:** It is an in-situ remediation technology that uses microorganisms to biodegrade organic constituents adsorbed in soils in the unsaturated zone. Bioventing enhances the activity of indigenous bacteria and simulates the natural in situ biodegradation of hydrocarbons in the soil by inducing air or oxygen flow at low pressure into the unsaturated zone and, if necessary, by adding nutrients. In conventional bioventing systems, oxygen is delivered by an electric blower to subsurface wells.

Saturated zone requires injection of air and nutrients at high pressure, and unsaturated zone requires injection of air and nutrients at low pressure.

3. **Bioaugmentation:** In this technology, the microorganisms are imported to the contaminated site to carry out the degradation of organic waste. For instance, oil zapper (explained earlier).

4. **Bioculture:** It is a bacterial formulation to improve waste degradation in septic tanks and eliminate odours due to organic buildup. Bioculture refers to the use of blend of bacteria that collectively produce enzymes for the degradation of fats, oils, proteins, starch, and carbohydrates. Bioculture is specifically used for sewage treatment.

Ex-situ bioremediation techniques

Ex-situ refers to the transfer of contaminated material for treatment to some other site. Ex-situ bioremediation techniques include land farming and biopile.

1. **Land farming:** In this technique, the contaminated soil is spread over a prepared bed.

The soil is periodically tilled to stimulate the growth of microorganisms for the degradation of organic waste.

2. **Biopile:** It is a hybrid of land farming and composting. Excavated soils are spread over a prepared bed, formed into compost piles and enclosed for treatment.

   Moisture, heat, nutrients, oxygen, and pH are controlled to enhance biodegradation. An irrigation/ nutrient system is used to pass air and nutrients through the soil. Soil piles can be up to the height of 20 feet. They may be covered with plastic to control runoff, evaporation, and to promote solar heating.

   Treatment time is typically three to six months, after which the excavated material is either returned to its original location or disposed off.

   The treatment area is generally covered or contained with an impermeable lining to minimise the risk of contaminants leaching into the uncontaminated soil.

Bioremediation Techniques

1. **Phytoremediation:** It means using plants to remove contaminants from soil and water. Neem plant is used for phytoremediation as it absorbs poisonous elements and reduces the growth of harmful microorganisms.

2. **Phytoextraction:** It is a subprocess of phytoremediation in which plants remove dangerous elements or compounds from soil or water, mostly heavy metals, metals that have high density and are toxic to organisms even at relatively low concentrations.

3. **Mycoremediation:** It involves the use of fungus such as *mycelia* to decontaminate an area. *Mycorrhiza* is another type of fungus which is used for bioremediation. It also has other important uses in agriculture.

   '*Mycor*'–'*rhiza*' literally means 'fungus'–'root'. It exists in a mutually beneficial relationship with plant roots. These fungi develop on plant roots and extend far into the soil. Thereafter, these fungi act as extensions of root systems and are, in fact, more effective in nutrient and water absorption than the roots themselves. Mycorrhiza also protects plants against pathogens and toxic substances present in the soil. The fungus also facilitates restoration and helps in revegetation of disturbed mined lands.

What Is Synthetic Biology?

It is an emerging science through which new life forms can potentially be made in labs

and existing life forms, such as bacteria and other microbes, are altered to produce specific proteins or chemically useful products.

Possible Benefits of Synthetic Biology

Synthetic biology in microbial systems holds promise for the production of drugs, vaccines, fuel components, and other chemicals. Microorganisms have also been constructed to act as sensors that can detect a toxin in vitro (outside a living organism) or in vivo (inside a living organism).

DRAWBACKS OF BIOTECHNOLOGY

1. Biotechnology can be used to develop Weapons of Mass Destruction (WMD). Biological weapons of mass destruction are cheap and easy to build. Moreover, these weapons have devastating effect only on living organisms and do not affect infrastructure.

2. Biotechnology can bring back certain extinct forms of life, which may lead to some unpredictable and harmful consequences. For instance, the smallpox virus can be regenerated and left in the environment to infect people.

3. Biotechnology may have a negative effect on biodiversity. At present, few plant and animal species are the focus of research, leading to ignorance of other species. The focus on few species may lead to their growth and can have a negative effect (even extinction) on remaining species.

4. Biotechnology is used to develop plant varieties with terminator genes. A terminator gene in a genetically modified crop plant stops the plant from releasing fertile seed. Hence, the farmer is again required to purchase the seeds in the next cropping season. The practice of incorporating terminator gene trait in some seed varieties is adopted by multinational companies (MNCs) to enhance their sale of seeds. This terminator trait may cross-pollinate with local varieties and may affect the continuity of agriculture.

CLONING

It is a process of asexual reproduction in which the offspring or the progeny is an exact replica of the single parent donor who has contributed the genetic material. Cloning is possible because each cell is equipped with genetic information of an organism, which has the ability to develop into full organism.

In contrast, in sexual reproduction, the progeny inherits genetic material in an equal amount from both the parents.

Cloning in animals is used to produce duplicates of animals. The first successfully cloned animal was a sheep called Dolly in 1997 at Roslin Institute of Technology, Scotland. Since then, a large number of animals have been cloned.

The following are India's achievements in animal cloning:

1. **Samrupa:** In 2009, the world's first cloned buffalo calf, named Samrupa, was developed by National Dairy Research Institute (NDRI) in Karnal, Haryana. But unlike Dolly, the first mammal cloned 13 years ago, who lived for seven years, Samrupa succumbed to a lung infection just five days after it was born.

2. **Garima:** It was the world's second cloned buffalo at NDRI in Karnal, Haryana. It was developed in 2009 and survived for more than two years. It died because of heart failure in 2011.

3. **Cirb Gaurav:** In 2016, the scientists at the Central Institute for Research on Buffaloes (CIRB) in Hisar, Haryana, cloned a buffalo offspring named 'Cirb Gaurav'.

Human Cloning

The process of creating a genetically identical copy of a human being, human cell, or human tissue is called human cloning.

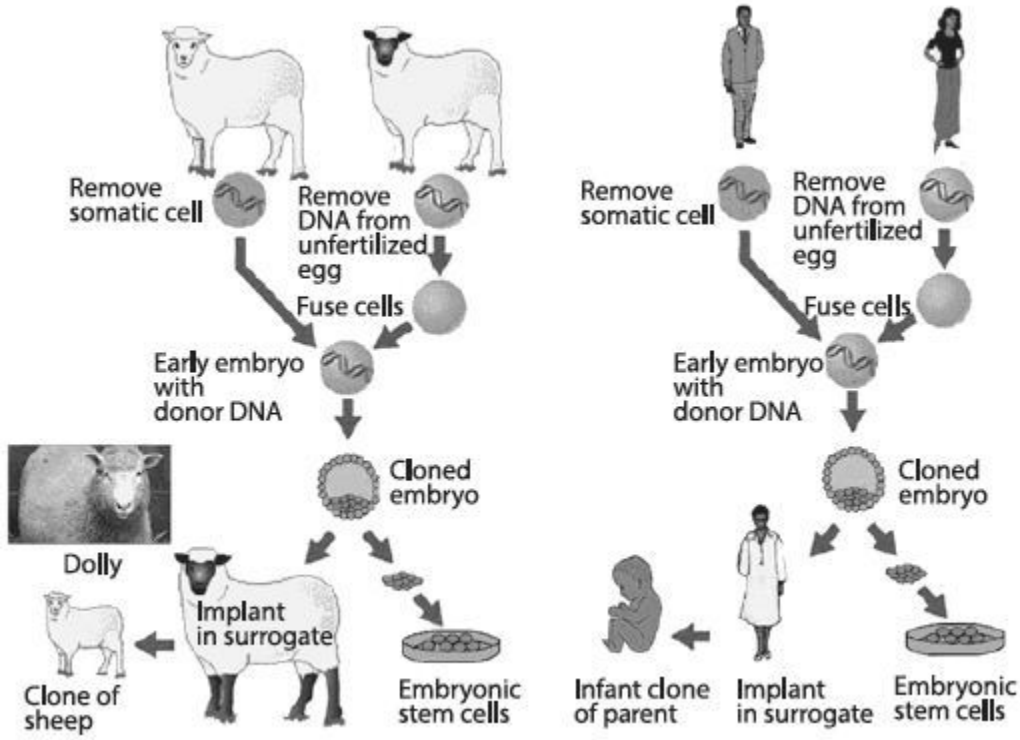
Process of Cloning

Human cloning is performed by somatic cells (any cells in the body other than sperm and egg, the two types of reproductive cells) by a nucleus transfer to an enucleated egg (an egg cell whose nucleus has been removed). The egg so obtained is then stimulated by electric shock and chemicals to initiate division. Within a week, this single cell becomes a ball of mass, having around 150 unspecialised cells. This stage of development is called 'blastocyst'. The blastocyst is inserted into the uterus of a surrogate mother to complete the process of embryonic development.

There are two types of cloning: reproductive and therapeutic. The difference between the two is listed below:

1. In reproductive cloning, the newly created embryo is placed back into the uterus, where it can develop into an individual. Reproductive cloning is the production of a genetic duplicate of an existing organism. A human clone would be a genetic copy of an existing person.

2. Therapeutic cloning involves the replication of human embryos in order to harvest stem cells for medical uses. In therapeutic cloning, an embryo is created in a similar manner, but the resulting 'cloned' cells are stored in the lab; they are not implanted into a female's uterus. We will learn about the stem cells later in this chapter.



Concerns over Reproductive Human Cloning

Reproductive human cloning is opposed on various ethical grounds.

1. It may undermine society's respect for human life. It may happen that clones are treated as secondary race or even as slaves.

2. It may affect social institutions such as marriage and family. A single parent may go for reproductive cloning.

   Children born out of reproductive cloning may be treated secondary in the family. A cloned son of a man would be his identical twin. This would create a set of very complicated family relations. Thus, the institution of family may be affected.

3. Reproductive Cloning (RC) may create a global security concern. Nations or even terrorist organizations may create cloned armies.

4. The RC may emphasize infusing desirable traits in clones. This may promote the

concept of 'designer babies', babies who are genetically engineered to exhibit desirable characteristics.

5. Experiments and research on cloning require working on embryos. This is opposed because, according to some religious organizations, life begins at conception.

How Is RC Different from Surrogacy?

1. **Process:** In case of cloning, a somatic cell is taken from a donor to create an embryo. This child is born of a single parent and carries his/her DNA only.

   In an In Vitro Fertilization (IVF) or test tube baby, an egg fertilized by a sperm (creating a zygote) is transferred into the uterus. It creates a progeny similar to normal conception. The child carries the DNA of both his/her parents.

2. **Uniqueness of progeny:** Biologically, a child from IVF is a unique human (unless he/she has an identical twin), while a cloned child is genetically identical to his/her parent.

3. **Lifespan:** Shortened lifespan has been reported in many cases of animal cloning. The progeny born out of IVF leads a normal life.

4. **Ethical issues:** The ethical issues with IVF are not questioned as much as RC. Both IVF and RC help infertile couples and same-sex couples with their parenting rights.

   This, in turn, has led to the commercialization of IVF and creation of 'contract mothers'. Many conservatives also believe that it is a commodification of children, where the social perception of motherhood and fatherhood changes and turns the baby into a commodity.

Stem Cells

Stem cells are the raw materials for other body cells. They are considered raw materials because all other cells with specialised functions are generated from these cells.

In other words, stem cells are unspecialised cells that have not yet developed into mature, specialised cells. These cells have the ability to develop into different, specialised body cells.

Stem cells have the following two important properties:

1. **Ability of self-renewal:** The ability of stem cells to go through numerous cycles of cell

division while maintaining the undifferentiated state is called self-renewal.

Under the right conditions in the body or a laboratory, stem cells divide to form more cells called daughter cells.

These daughter cells either become new stem cells (self-renewal) or become specialised cells (differentiation) with a more specific function. No other cell in the body has the natural ability to generate new cell types.

2. **Ability to specialise:** The stem cells have the capacity to differentiate into specialised cell types. They have the ability to specialise in various body cells types such as blood cells, brain cells, heart muscle, or bone.

Sources of Stem Cells

Stem cells are obtained from the following sources:

1. **Embryonic stem cells:** These stem cells come from embryos three to five days after conception. At this stage, an embryo is called a blastocyst and has around 150 cells.

   These stem cells are pluripotent stem cells, meaning they can divide into more stem cells or become any type of cell in the body. Thus, these stem cells can be used to regenerate or repair diseased tissues and organs.

2. **Adult stem cells:** These stem cells are found in small numbers in most adult tissues, such as bone marrow or fat. Compared to embryonic stem cells, adult stem cells have a limited ability to specialise in various cells of the body.

   For instance, bone marrow stem cells may create bone or heart muscle cells, but not nerve cells. The research involving adult stem cells is undergoing clinical trials to test its usefulness and safety in people. For example, adult stem cells are currently being tested in people with neurological or heart disease.

   Scientists have also successfully transformed regular adult cells into stem cells using genetic reprogramming. By altering the genes in the adult cells, researchers can reprogram the cells to act like embryonic stem cells. These stem cells are called induced pluripotent stem cells.

   However, researchers are yet to know if these reprogrammed cells will cause adverse effects in humans.

3. **Perinatal stem cells:** Perinatal means relating to time, usually a number of weeks, immediately before and after birth. Researchers have discovered stem cells in amniotic fluid in addition to umbilical cord blood stem cells. These stem cells also have the ability to change into specialised cells. Clinical trials are undertaken to understand the potential of amniotic fluid stem cells.

Totipotent, pluripotent and multipotent Stem Cells

We have learned that stem cells have the ability to develop into specialised body cells. On the basis of their ability to develop into specialised cells, stem cells can be divided into three types- Totipotent cells, Pluripotent cells, and Multipotent cells.

Totipotent cells: Totipotent is made up of two words- 'Toti' means 'entire or whole' and 'potent' word is used to reflect 'capability or dominance' of something. Thus, Totipotent stem cells are the type of stem cells that can be specialised into an entire range of cell types in the body, including embryonic and placental cells. Thus, these cells are capable of developing into a complete organism.

Therefore, these cells have the maximum ability to specialise into any cell type. Embryonic cells within the first two cell divisions after fertilization are the only cells that are totipotent.

Pluripotent cells: The word 'Pluri' means 'many'. Pluripotent cells are the stem cells that can be specialised into all the cell types that make up the body. However, they cannot be specialised into embryonic and placental cells. Thus, the ability of Pluripotent cells to specialise is less than the Totipotent cells' as the Pluripotent cells cannot specialise into embryonic and placental cells. Pluripotent stem cells can be are sourced from embryonic stem cells.

Multipotent cells: The word 'Multi' means 'more than one or a few'. Multipotent cells can be developed into more than one cell type, but their ability to specialise is limited to pluripotent cells. Multipotent stem cells can be obtained from adult stem cells and umbilical cord blood stem cells.

Totipotent, Pluripotent, and Multipotent stem cells can also be understood through one more approach. Tutipotent stem cells can be obtained from embryos within the first two cell divisions after fertilization. As embryo develops, Pluripotent cells can be obtained from the embryo. Once an embryo develops into specialised cells and takes birth, then only multipotent stem cells can be obtained.

Uses of Stem Cells

Stem cells have the following applications:

1. **Increased understanding of how diseases occur:** By watching stem cells mature into cells in bones, heart muscles, nerves, and other organs and tissues, researchers and doctors may better understand how diseases and conditions develop.

2. **Generate healthy cells to replace diseased cells (regenerative medicine):** Stem cells can be guided to become specific cells that can be used to regenerate and repair diseased or damaged tissues in people. People who might benefit from stem cell therapies include those with spinal cord injuries, Type 1 diabetes, Parkinson's disease, Alzheimer's disease, heart disease, burns, cancer, and osteoarthritis.

3. **Test new drugs for safety and effectiveness:** Before using new drugs in people, some types of stem cells are useful to test the safety and quality of investigational drugs.

   For testing new drugs, the cells are programmed to acquire properties of the type of cells to be tested. For instance, nerve cells could be generated to test a new drug for a nerve disease. Tests could show whether the new drug had any effect on the cells and whether the cells were harmed.

Ethical Issues in the Use of Stem Cells

Embryonic stem cells are obtained from early-stage embryos—a group of cells that forms when a woman's egg is fertilised with a man's sperm. Thus, the use of embryos to obtain stem cells is opposed by some religious bodies which believe that life begins immediately after fertilization.

Problems with the Use of adult Stem Cells

Adult stem cells have limited ability to specialise, which limits how adult stem cells can be used to treat diseases.

Adult stem cells are also more likely to contain abnormalities such as toxins or errors acquired by the cells during replication.

Organoids

Organoids are tiny, self-organized, 3D tissue cultures, grown from stem cells in a controlled environment. In other words, organoids are tiny, functional versions of a

fully grown organ. Their size ranges from 200 microns to a few millimeters. Organoids can be derived from all three types of stem cells. Organoids resembling the brain, lungs, intestine, liver, kidney, heart, eyes, inner ear, pancreas, salivary glands, and stomach, among others, can be developed.

Applications of Organoids

Organoids find numerous applications in the fields of medical research, diagnosis, pathology (study of diseases), and treatment of diseases. These applications include:

1. Medical Research: Organoids can contribute significantly to embryology by demonstrating how specific organs are developed from the embryo. Earlier, scientists used to rely on the studies on mice and other animals and extrapolate these studies to understand how human embryonic development takes place.

2. Research on diseases: Organoids can be used to understand how a disease affects a particular organ system. For instance, to study how Microcephaly (a neurological condition in which an infant's head is significantly smaller) affects the patients, cerebral organoids can be cultured from a patient and studied.

3. Pharmaceuticals: Organoids can be used in testing of drugs and clinical trials. Organoids successfully replaced host animals like mice and primates in certain cases for drug testing. They can also improve the efficiency of the testing processes in certain cases where the diseased animals may not show a response similar to humans.

For instance, in the case of COVID-19, mice infected with SARS-COV-2 were not exhibiting the immune response seen in humans. As a result, it becomes difficult to draw reliable conclusions by using mice for drug testing. To overcome this issue, mini lungs (organoids) are developed and exposed to SARS-COV-2. The organoids showed an aggressive immune response. During the testing, virus infected blood vessel organoids was observed. This helped explain how the virus reaches other organs through bloodstream. Subsequently, COVID drug trials on mini lungs were used to isolate the compounds which reduced viral levels in mini lungs.

Organoids can also help to improve the efficacy and safety of certain drugs and reduce any allergic reactions. They can also be used in developing patient-specific drug interventions that will significantly reduce anaphylactic responses (severe potentially life-threatening allergic reactions) to drug administration.

4. Treatment of genetic diseases: Further, Organoids can also be used in the

treatment of genetic diseases. For instance, they can be used in tissue transformation and transplantation to mitigate the effect of genetic anomalies. Researchers have shown that when intestinal cells are transformed into insulin producing beta cells and transplanted into a diabetic mouse, these cells regulate the blood sugar levels of the mouse.

Limitations to the Use of Organoids

1. Despite the advantages, there are some ethical and legal concerns around the use of organoids. For instance, scientists are worried about the unethical use of organ culture to produce organs of commercial interest which may not fulfill the necessary criteria. This can potentially boost illegal organ trade and even the trade of organs with sub-optimal functioning.

2. Organoids can exhibit only the basic functions of an organ, as they are still at the initial stages of organ development. For example, cerebral organoids lack the manifestation of consciousness and, therefore may be useless in the treatment of certain psychological disorders. Thus, there is a significant limitation on the use of organoids for medical treatments.

3. The immune responses of the organoids and the actual organs may vary, as organoids are grown in controlled environments. This means their acquired immunity levels are lower than those exhibited by the actual human organs. These differences should be taken into account while conducting drug trials.

DNA Fingerprinting

DNA fingerprinting refers to identifying a complete (or partial) set of genetic information of a particular individual. It is essentially a DNA-based identification system that relies on genetic differences among individuals or organisms. A sample of blood, saliva, semen, vaginal lubrication, or other appropriate fluid or tissue from personal items can be used for DNA fingerprinting.

Like fingerprints, every human has unique DNA; unlike fingerprints which can be surgically altered, one cannot change the DNA. The DNA fingerprinting is also known as DNA analysis or DNA profiling.

What is DNA?

Deoxyribonucleic acid (DNA) is the genetic material typically found in all living cells of humans as well as animals and plants. It invariably contains hereditary data passed on from parent to children, which is unique to each person (except in the case of identical

twins). This makes DNA profiling a reliable and unique personal identification tool.

Applications of DNA fingerprinting

1. **Identification of criminals:** DNA analysis of hair, bodily fluids, skin, etc., obtained from a crime scene is used to compare with the DNA analysis of suspects to identify the actual criminals.

2. **Claim over dead body:** DNA fingerprinting is used to identify the unrecognizable dead body.

3. **Paternity:** Paternity can be established with certainty with DNA analysis.

4. **Effective drugs:** Drugs can be developed which are more effective for a particular genetic disposition.

5. **Treatment of genetic vulnerability:** Genetic vulnerabilities can be identified beforehand and prevented. For instance, if a person has a genetic tendency for hypertension, then preventive steps can be taken to prevent the occurrence of hypertension.

6. **Wildlife management:** The more the genetic makeup of plant and animal populations is understood, the better conservation and management plans can be formulated.

Concerns Over DNA fingerprinting

1. The information about gene pool can lead to a preference for designer babies and ignore the genes of undesirable characteristics, thus reducing genetic diversity.

2. Genetic privacy of individuals will be violated. It can have multiple repercussions. For instance, a person with a particular genetic vulnerability may be looked down upon socially.

3. Genetic information can be misused for commercial purposes. For instance, medical institutions will start offering medical packages based on DNA analysis.

4. Genetic information may be used to create weapons of mass destructions, leading to ethnic cleansing of a particular community.

DNA Barcoding

DNA barcoding is a technique used to identify and classify species using a short, standardized segment of genetic material. This method has revolutionized the field of taxonomy, making species identification faster, more accurate, and more accessible.

How DNA Barcoding Works

DNA barcoding involves several key steps:

1. **Sample Collection**: A specimen is collected from the environment. This could be a plant leaf, an insect leg, or a sample of water containing microbial life.
2. **DNA Extraction**: The genetic material is extracted from the sample using chemical or mechanical means.
3. **PCR Amplification**: The target DNA region is amplified using polymerase chain reaction (PCR) to produce enough copies for analysis.
4. **Sequencing**: The amplified DNA is sequenced to determine the exact order of nucleotides.
5. **Data Analysis**: The sequence is compared to a reference database to identify the species.

Applications of DNA Barcoding

DNA barcoding has numerous applications across various fields:

- **Biodiversity Studies**: It helps in cataloging and monitoring biodiversity, especially in regions with rich but poorly documented flora and fauna.
- **Conservation Biology**: DNA barcoding aids in identifying endangered species and monitoring illegal wildlife trade.
- **Food Safety**: It is used to authenticate food products and detect adulteration or mislabeling.
- **Environmental Monitoring**: DNA barcoding can assess ecosystem health by analyzing the presence and abundance of different species in environmental samples.
- **Medical Research**: It helps in identifying pathogens and understanding their genetic diversity.

Advantages of DNA Barcoding

DNA barcoding offers several advantages over traditional taxonomic methods:

- **Speed and Efficiency**: It is much faster than morphological identification, which can be time-consuming and require expert knowledge.
- **Accuracy**: Genetic identification is highly accurate, reducing the chances of misidentification.
- **Universality**: A single standardized method can be applied to a wide range of organisms.

Challenges and Limitations

Despite its benefits, DNA barcoding faces some challenges:

- **Database Limitations**: The accuracy of DNA barcoding depends on the comprehensiveness of reference databases. Gaps in the database can lead to unidentified or misidentified species.
- **Cost and Accessibility**: The cost of sequencing and the need for specialized equipment can be prohibitive, especially in developing regions.
- **Genetic Variation**: In some cases, the chosen barcode region may not vary enough to distinguish closely related species.

Future Perspectives

The future of DNA barcoding looks promising with advancements in sequencing technologies and bioinformatics. Next-generation sequencing (NGS) and metagenomics are expanding the scope of DNA barcoding, allowing for the identification of multiple species from a single environmental sample. As databases grow and technology becomes more accessible, DNA barcoding will continue to play a crucial role in biological research and conservation efforts.

Conclusion

DNA barcoding is a powerful tool that has transformed species identification and classification. Its applications span various fields, contributing significantly to our understanding of biodiversity and ecosystem health. Despite some challenges, ongoing advancements in technology and methodology promise to enhance the efficacy and reach of DNA barcoding in the future.

Gene Therapy

Gene therapy is a medical technique that manipulates genes to treat or prevent a disease. Gene therapy research is focused on the following approaches:

- Replacing a gene responsible for disease with a healthy gene.
- Inactivating or 'knocking out' a gene that is functioning improperly.
- Introducing a new gene into the body to help fight a disease.

In the future, this technique may allow doctors to treat a disorder by manipulating a gene into a patient's cells instead of using drugs or surgery. Gene therapy is a treatment option for a number of diseases (including inherited disorders, some types of cancer, and certain viral infections).

Use of Vector to Introduce a Gene

Usually, a gene that is inserted directly into a cell does not function on its own.

Instead, a carrier called vector is genetically engineered to deliver the gene. Certain viruses such as retrovirus are often used as vectors because they can deliver the new gene by infecting the cell. The viruses are modified not to cause disease when inserted into people.

The vector can be injected or given intravenously (by IV), directly into a specific tissue in the body, where it is taken up by individual cells. Alternately, a sample of the patient's cells can be removed and exposed to the vector in a laboratory setting. The cells containing the vector are then returned to the patient. If the treatment is successful, the new gene delivered by the vector will make a functioning protein.

Concerns Over Gene Therapy

1. **Short-lived nature of treatment:** Before gene therapy can become a permanent cure for a condition, the therapeutic DNA introduced into the target cells must remain functional and the cells containing the therapeutic DNA must be stable. Problems with integrating therapeutic DNA into the genome and the rapidly dividing nature of many cells prevent it from achieving long-term benefits. Patients undergoing gene therapy often require multiple treatments.

2. **Immune response:** Depending upon the number of times a foreign object is introduced into our body, the immune system is stimulated to attack the invader. As a result, the gene therapy might activate the response of our immune system. Even our immune system reduces the effectiveness of gene therapy.

3. **Multi-gene disorders:** Some commonly occurring disorders such as heart disease, high blood pressure, Alzheimer's disease, arthritis, and diabetes are affected by variations in multiple genes, which complicate the use of gene therapy.
   Presently, the technique remains risky. It is not yet proven to be safe and effective. Gene therapy is currently being tested only for the treatment of diseases that have no other cures.

DNA Vaccines or Third Generation Vaccines

DNA vaccines are also called third-generation vaccines. These vaccines are made up of a small, circular piece of bacterial DNA (called plasmid) or virus that has been genetically engineered to produce one or two specific proteins (antigens) from a pathogen.

In the DNA vaccine, a piece of bacterial DNA (plasmid) carrying antigens is directly given to us, and our body absorbs the DNA into our genetic system. Our body then

replicates the production of plasmid- carrying antigen. This release of antigen by our own DNA activates our immune system. Like any vaccine, the immune system will then recognize the bacteria or virus in the future—hopefully preventing illness.

What are the advantages of DNa Vaccines?

1. **Require short time span for development:** Quick changes can easily be brought in gene-based vaccine than bacteria- or virus-based vaccine. Such changes are important to deal with strains of bacteria or viruses that are constantly mutating.

2. **DNA vaccines are easy to transport and store:** DNA is a very stable molecule and does not need to be stored at low temperatures, making transportation and storage cheaper and easier than conventional vaccines.

3. **Less risk to those who are making the vaccine:** Conventional vaccines require raising up the infectious bacteria or virus. Thus, there is a risk (even though small) to those who make the vaccines, whereas making DNA vaccines is less risky.

What are the Disadvantages of DNA Vaccines?

So far, no DNA vaccine has been licensed for the use in humans. Although some DNA vaccines are now in clinical trials, none are licensed for use.

First-generation Vaccines

These vaccines consist of infectious organisms, either in mild or dead form. The first-generation vaccines are still widely used today.

Live and mild/attenuated forms of infectious organisms produce both humoral (antibody) and cellular immune responses. For example, an oral polio virus vaccine uses polio virus in mild form. When we take the vaccine, our body reacts as if it is affected by an actual virus. Consequently, our immune system gets activated and T-killer cells attack the polio virus. Thereafter, when there is any actual attack of polio virus, then our immune system is already developed to handle such an attack.

The only problem with these vaccines is that the actual pathogen in these vaccines may take a dangerous form.

Dead pathogen vaccines do generate an antibody response but they do not generate cellular responses (no T-killer response). Depending on the disease, antibody production may or may not be enough to ward off infection. The advantage of using killed pathogen vaccine is that there is no chance of infection from the vaccine.

T-killer cells are T-lymphocytes (a type of white blood cell); these cells kill other cells that are infected (particularly with viruses) or cells that are damaged in other ways.

Second-generation Vaccines

The second-generation vaccines were created in order to minimise the risks of pathogen revert to a dangerous form.

The way these vaccines work is that they do not contain the whole organism, but rather contain only subunits. Subunits may consist of the toxins that the pathogen uses for infecting the body. A great example of second-generation vaccine is DTP vaccine. The second-generation vaccines can generate antibody response but not T-killer response.

CRISPR-Cas9

CRISPR stands for Clustered Regularly Interspaced Short Palindromic Repeats. This is the name given to unique DNA structures found in bacteria and other microorganisms, which constitute a key part of their 'immune system'. If a virus enters bacteria, the CRISPR immune system fights back by destroying the genome of the virus. It cuts the invading virus's DNA, and as a result, the genetic material which is necessary for the replication of the virus is destroyed. Hence the CRISPR immune system protects bacteria from the viral infection.

Moreover, the bacteria also store some of the invading virus's DNA. When there is an attack by any virus in the future, the bacteria produce an enzyme called Cas9 (CRISPR-associated) endonuclease or Cas9 enzyme. The Cas9 matches the fingerprints stored earlier with that of the new invading virus. If the fingerprints match, the new virus is the same as the earlier virus that invaded the bacteria. The Cas9 then acts as a molecular scissors and snips the invading virus's DNA. The location at which Cas9 needs to cut the DNA is specified by the RNA sequence of CRISPR. CRISPR-Cas9 finds applications in gene editing where it is used as a gene-editing tool.

Gene-editing tool: The CRISPR-Cas9 gene editing tool comprises two components: a mechanism to identify a specific target and a molecular scissor. A short RNA sequence binds to a specific target of the DNA and the Cas9 enzyme cuts the DNA at the location where RNA sequence is bound. The natural DNA repair mechanism kicks in after the DNA is cut. This mechanism is utilised to make the required changes to the DNA, to add or remove genetic material. For instance, during this auto-repair process, scientists may intervene to supply a desired gene sequence that binds with broken

DNA. In simple terms, to draw an analogy, CRISPR-Cas9 works like the 'Find and Replace' function in a word processing program such as Microsoft word.

Applications of CRISpR-Cas9

CRISPR-Cas9 can be used in a wide range of applications such as industry, medicine, and research. For instance, industrial processes that use bacteria (e.g., fermentation based industries such as dairy, distilleries) can use the CRISPR system to make the bacterial cultures resistant to any particular viral attack, and hence increase productivity.

CRISPR technology is also used in medical research to understand how a particular gene affects an organism. Scientists can change the gene of interest and study how it affects the organisms. Further, this tool can be used to silence a particular gene—delete, correct, or insert a new gene.

In medicine, the tool may be used in the treatment of genetic diseases. For instance, in persons with sickle cell anemia, a mutation causes the red blood cells to be sickle shaped. This hinders their capability to carry out oxygen throughout the body. In such cases, by changing a single gene, the mutation can be reversed. Thus, the disease can be cured using gene editing. Along with genetic diseases, infectious diseases can also be treated using gene-editing technologies. For instance, specific antibiotics which selectively target only disease-causing bacteria can be developed to treat infectious diseases.

The He Jiankui affair

In 2018, a Chinese scientist He Jiankui claimed that he created the world's first genetically edited babies. This raised international furore and a series of questions on the ethical and legal issues surrounding the use of gene editing and use of tools such as CRISPR-Cas9.

What Did the Scientist Claim?

The scientist claimed to have carried out assisted reproduction in a couple, where one of them had HIV. During the in-vitro fertilisation, he used the CRISPR-Cas9 tool to disable a gene CCR5 in the embryo. CCR5 encodes a protein, and it allows HIV to enter and infect the host cells. By disabling this, the doctor claimed to have made HIV-resistant babies. He also claimed to have implanted a gene-edited embryo in another woman.

Why Is it Illegal and Unethical?

There is an international consensus that gene editing should not be used on Human embryos until sufficient international protocols are developed to prevent its misuse. Moreover, research still needs to be carried out on the long-term health impacts if such gene modifications are done in babies.

In this case, though it is believed that HIV enters and infects the host through gene CCR5, there were no clinical trials carried out anywhere in the world to test the same. Hence, the efficacy of such procedures and the side effects are not known. Further, the CCR5 gene plays an important role in protecting against viral infections such as west nile viral infection. Without this gene, babies are more susceptible to such virus infections. Moreover, the X4 form of HIV uses a different protein to enter the cells (not CCR5). Hence, babies may not be resistant to HIV from this source of infection.

Scientists and policymakers argue that if gene editing on human embryos is allowed, it may lead to several unintended consequences. For instance, people may choose to edit embryos for very specific qualities such as desirable physical attributes, intelligence, and so on. As genes pass on from one generation to another, this may inadvertently alter the genome of future generations. Hence, guidelines need to be established to ensure that the genetic engineering tools are used in very specific and rare cases such as treating diseases with no alternative treatments.

What Was the Outcome in He Jiankui's Case?

The Chinese court sentenced the doctor to three years in prison for violating medical regulations and ethics. The court claimed that the doctor flouted the medical regulations and ethics in the pursuit of "fame and profit".

Food fortification

Food fortification refers to the process of adding certain important vitamins and minerals to staple foods to improve their nutritional content.

The common staple foods that are fortified include wheat, rice, milk, oil, and salt. The commonly added vitamins and minerals are iron, zinc, iodine, vitamin A and vitamin D, to name a few. These nutrients may not be originally present in these food items or may have been lost during their processing. Adding these nutrients improves the nutritional content of food and helps address the malnutrition in the population.

The following are the benefits of food fortification:

1. Since nutrients added to staple foods are consumed by a large section of the population, food fortification helps in addressing nutrition deficiencies effectively at overall nation level.
2. The small quantities of nutrients are added, which do not pose any health risks on account of their consumption. In other words, fortified foods are safe to be consumed.
3. It is a cost-effective method and does not need any major changes in the existing food habits of the people. This makes it a socio-culturally acceptable method to deliver the required nutrients.
4. The food characteristics such as taste, texture, or aroma are not altered.

Some of the commonly used fortified foods and their benefits are given below:

Type of Fortified Food	Commonly added nutrients	Benefits
Salt	Iodine	Iodine deficiency occurs because crops are grown on iodine- deficient soils. Fortification of salt with iodine addresses various iodine deficiency related disorders effectively as salt is a food item that is consumed daily and almost universally. Iodine deficiency disorders include goitre, brain damage, hypothyroidism, intellectual disability, etc.
Double Fortified Salt	Iron and Iodine	Double Fortified Salt (DFS) formulations can provide 100% of dietary iodine requirements and 30 to 60% of dietary iron requirements.

Milk	Vitamin A and D	Milk is a natural source of vitamins A and D. However, these vitamins are lost when the milk is processed. Hence the lost vitamins are compensated by adding these vitamins back to milk.
Edible oil	Vitamin A and D	Vitamins A and D are added to edible oils to address the micronutrient deficiencies. Fortified oil can provide around 25% to 30% of the daily dietary requirements of vitamins A and D.
Rice	Iron, Folic acid (man-made version of Vitamin B9) and Vitamin B12	Fortification of rice can be done by adding the vitamins and minerals in the post-harvest phase. Since rice is an important staple food of around 65% of the Indian population, the essential nutrients can easily reach the large population. Further, rice is also supplied by the Government's public distribution system and is easily accessible across the country. This makes it a preferred food to deliver the essential micronutrients.
Wheat Flour	Iron, Folic acid, Vitamin B12, Zinc, Vitamin A, Other B-Complex Vitamins such as Thiamine (B1), Riboflavin (B2), Niacin (B3) and Pyridoxine (B6)	Wheat is also an important staple food of Indians, especially in the wheat-growing regions of north, west and central India. Similar to rice, it is an effective way to deliver nutrients and prevent diseases such as anemia.

Genetically Modified Organisms

Genetically Modified Organisms (GMOs) are organisms whose genetic materials have been altered using genetic engineering techniques to provide the organisms with certain special characteristics. GMOs can include plants, animals, and even microorganisms.

We have learned that genetic modification can lead to various benefits. GMO research in animals is at the nascent stage. However, it has attained some success in plants. Many genetically modified plant varieties have been developed.

Some of the popular genetically modified crops are mentioned below:

1. **Golden rice:** We have already discussed Golden Rice in this chapter. At present, research on Golden Rice is taking place. It is not yet commercially cultivated.

2. **Bt cotton:** We have already discussed that Bt cotton is a transgenic crop in which 'Cry1 AC' gene of Bacillus thuringiensis is introduced to make the pest-resistant crop. After the introduction of this gene, the cotton crop starts producing its own pesticide. The Bt trait is believed to save the cotton plant from the pest popularly known as bollworm.

3. **Bt brinjal and Bt mustard:** Bt brinjal and Bt mustard have been developed on the lines of Bt cotton. These crops are also transgenic in nature in which 'Cry1 AC' gene of Bacillus thuringiensis is introduced to make the pest-resistant crop.

Position of Genetically modified Crops in India

At present, commercial cultivation of edible, genetically modified (GM) crops such as Bt brinjal and Bt mustard is not allowed because of the following reasons:

1. Presently, research is not enough to understand the impact of Bt food crops on human health. The regular consumption of such varieties may have long-term repercussions on health.

2. Bt trait food crop may cross-pollinate with local wild weeds to make them superweeds, which would then require a large amount of pesticides for their elimination.

3. Moreover, there is a strong opposition from some groups for the cultivation of GM crops. Farmer groups oppose the cultivation of GM crops because promotion of sale of GM crops would hamper the sale of non-GM crops. NGOs such as Greenpeace stringently oppose the cultivation of GM crops.

On the other hand, commercial cultivation of non-edible Bt crops is allowed. For instance, Bt cotton crop is cultivated at many places in India.

GM Mustard (DMH-11)

Mustard is a self-pollinating crop. Hence, it is difficult to develop a hybrid of mustard. In other words, it is difficult to cross pollinate a mustard crop. In 2016, researchers of the Delhi University have genetically modified an Indian mustard (Varuna) and an East-European mustard to cross- pollinate them. After cross-pollinating these genetically modified mustard varieties, the new variety of mustard developed was named Dhara mustard hybrid (DMH)-11.

Researchers have sought permission for the following:
1. To commercially release DMH-11 and
2. To use the two GM parental lines, Indian and East-European, for developing new hybrids.

Benefits claimed from DMH-11include:
1. DMH-11 yields about 30% more than the traditional reference mustard variety.
2. It will help in boosting edible mustard oil production; thus, reducing the huge import bills for edible oil.
3. GM mustard is resistant to herbicides.

Views of Government on GM Mustard

Our government has given the required permission to the researchers to develop GM mustard. The Genetic Engineering Appraisal Committee (GEAC) is the government agency responsible for granting permission for research on genetically engineered organisms and products.

Views of Supreme Court on GM Mustard

The Supreme Court has stayed permission to develop GM mustard. It holds that the approval for GM mustard has been given without consulting people. Moreover, it holds that denying citizens a voice in this matter is all the more serious, considering that no labeling regime is in place in India. Without proper labeling, citizens will not know whether they consume food made from GM mustard.

Genetic Engineering Appraisal Committee

The Genetic Engineering Appraisal Committee (GEAC) was formed under the Environment Protection Act, 1986, and functions under the Ministry of Environment, Forest and Climate Change (MoEFCC). It gives approval for the cultivation of GM crops.

The functioning of GEAC has the following loopholes:

1. The GEAC is not an autonomous body as it functions under MoEFCC. Thus, while approving for genetically modified crops, the GEAC may come under pressure from MoEFCC.
2. Moreover, the GEAC does not possess independent research capabilities. It gives approval based on the research report furnished by the MNCs that seek approval for the cultivation of genetically modified crops.

There is a need to setup Biotechnology Regulatory Authority of India (BRAI), which

would be an autonomous body. Moreover, the BRAI shall possess independent research facilities to verify the claim made by the MNCs for a particular GM crop. These research facilities shall also be able to assess long- term impact of GM crop on health and environment.

INTERDISCIPLINARY DEPENDENCY OF BIOTECHNOLOGY

Biotechnology is dependent on many disciplines and techniques for accumulation of data, its organization and analysis. Some of these disciplines are mentioned below:

Biological Engineering

Biological Engineering or Bioengineering is an interdisciplinary area focusing on the application of engineering principles to analyse biological systems and solve problems relating to biological systems with human-designed machines, structures, processes, and instrumentation.

In many cases, currently available knowledge is inadequate to support the engineering design of biological processes. Hence, fundamental knowledge of biology and its potential applications remain a focus of biological engineering.

Examples of bioengineering include:

- Artificial hips, knees, and other joints.
- Ultrasound, MRI, and other medical imaging techniques.
- Using engineered organisms for chemical and pharmaceutical manufacturing.

Biomimetics

Biomimetics, also known as biomimicry, is the usage and implementation of concepts and principles from nature to create new materials, devices, and systems.

This adaptation of methods and systems found in nature into man-made products is desirable because living organisms have evolved into well-adapted structures and materials over geological time through natural selection. Moreover, human beings have looked at nature for answers to problems throughout their existence. Nature has solutions to many problems such as self-healing abilities, environmental exposure tolerance and resistance, harnessing solar energy, etc.

A simple example of biomimetics is the inspiration to develop Velcro tape from the hooks on Burdock burrs.

Velcro tape

Bionics

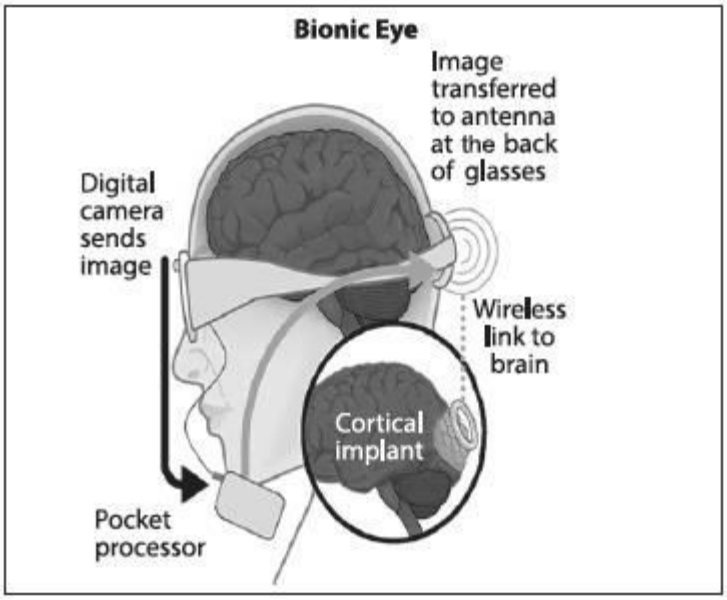
Bionics can mean different things to different people. It started as a term for the application of principles of biology to engineering. Now it is used to describe a method to engineer organs that can replace diseased or non-functional human organs. In the future, it could also be a way to design machines that can mimic biological behaviour. Thus, the term bionics is related to biomimetics. Bionics is distinct from bioengineering (or biotechnology), which is based on the use of actual living organisms.

Under the traditional method, well-fitting limbs such as wooden leg or a glass eye were made for persons who had lost legs or eyes. Bionic technology does not stop at making well-fitting prostheses. The term 'bio' in the word 'bionic' refers to artificial eyes or limbs such as legs or arms, which can carry out functions like normal limbs.

Example of bionic eye

Bionic eye is an artificial device that has been developed to restore vision. The device includes, among other things, a small video camera, a transmitter mounted on a pair of glasses, and an implant in the brain, which works as a wireless link between the transmitter and the brain.

This new surgically implanted assistive device provides an option for patients who have lost their sight, for which there are no approved treatments.



Bioinformatics

The recent flood of data from biology and the need for its organization have given rise to a new field, namely, bioinformatics, which combines the elements of Biology and Computer Science. Think about the data generated by more than three billion nitrogen pairs in a human body which form the gene sequences of an individual!

Bioinformatics begins with conceptualizing biological processes and systems and then applying 'informatics' techniques (derived from disciplines such as Applied Mathematics, Computer Science, and Statistics) to understand and organise the formation associated with these processes and systems on a large scale.

DEPARTMENT OF BIOTECHNOLOGY

The Department of Biotechnology was set up under the Ministry of Science and Technology in 1986. It replaced the National Biotechnology Board, which was earlier set up in 1982. The department is involved in the planning, promotion, and coordination of research and development in the area of biotechnology. The responsibilities of the department are as follows:

- Creation of policy frameworks
- Promote innovation and excellence
- Support research activities in the applications of biotechnology in fields such as:
  - agriculture and nutritional security
  - healthcare and medical biotechnology

- environmental safety
- new generation biofuels
- animal and aquatic sciences

■ Ensure that the benefits of biotechnology reach the community at large.

Project "Genome India"

The Genome India Project was initiated in January 2020 by the Department of Biotechnology. The project aims to build a reference genome by collecting 10,000 genetic samples from Indian citizens. Once the data is collected, whole genome sequencing and analysis will be carried out.

The project is significant for a number of reasons. The data is expected to increase the understanding of diseases that are affecting the Indian population. This will play a major role in developing the predictive diagnostic markers. Different subpopulations of patients may respond to the same therapy differently. Predictive markers help in identifying the possible response from a particular subpopulation of patients to specific therapies. This will allow us to identify the patients who will most likely to benefit from a given treatment. At the same time, it will help in avoiding administering ineffective treatments to other patient groups, where the treatment will not have the intended effect. This will also form the foundation for the advancement of next-generation personalised medicine.

The information generated from the project can also be used in preventive medicine. For instance, population with more susceptible risk factors for a certain diseases can be identified, and necessary interventions at community level can be made for the preventive measure from the disease.

Further, the existing genome research is dominated by western countries. For instance, more than 95% of the genome samples on which research is carried out are based on the white Caucasian genome. These genomes are not representative of the overall global population. Moreover, the genetic diversity of the Indian population varies greatly given the long and diverse history of the Indian subcontinent. Hence, the Indian project aims to add to the available research in a significant way, as the scale and diversity of Indian population are huge and unique. For instance, though the initial migrations to the Indian subcontinent were from Africa, there were periodic migrations and intermingling of races throughout the history of the subcontinent. This is called 'horizontal diversity'. On the other hand, certain groups practiced endogamy and restricted marriages outside that group. As a result, certain traits and diseases have passed within very specific groups. This is called 'vertical diversity'.

Understanding and studying these diversities will act as the basis of personalized healthcare in the future.

The Genome India Project is a collaborative effort of 20 leading institutions. It is led by the Centre for Brain Research, Indian Institute of Science (IISC), Bengaluru. The data collection will be carried out through hospitals, where the investigators will collect blood samples from participants and add the information to biobanks.

What Is a Genome?
Genome, in simple terms, can be defined as all the genetic matter in an organism. It is an organism's complete set of DNA and all its genes. Hence, a genome contains within it all the information that are needed to build and maintain an organism.

Human Genome Project
In 2003, the Human Genome Project, an international project aimed at decoding the entire human genome, was completed. DNA molecules are made of paired strands. The human genome is an estimated to contain around 3 billion of base pairs, which form estimated 30,000 genes in a human body and reside in the 23 pairs of chromosomes. The final version of the human genome sequence is published by the International Human Genome Sequencing Consortium. The Human Genome Project is a collaborative effort between the United States, France, the United Kingdom, China, Japan, and Germany.

Human microbiome Initiative
Human microbiome refers to the diverse communities of microorganisms present in the human body. These microorganisms play an important role in physiological processes, metabolism, and immune system in humans. Different types of microorganisms are present in human body based on factors such the organs in which they are located (different organs possess different microorganisms), genetics, age, dietary habits, and even geographic location. The detailed study of these microbes will lead to an increased understanding of a wide range of diseases and their effects.

The human microbiome initiative is a project launched by the Department of Biotechnology in 2019 that aims to collect the saliva, stool, and skin swabs from 20,000 Indians across various ethnic groups and geographical regions. The project aims to map the influence of diet, age, lifestyle, and geography on the gut microbiome in 17 different endogamous groups (groups that marry within themselves) across the country. The objectives of the project include:

- To generate baseline gut microbiome data

- To understand the relationship between diet and gut microorganisms
- To understand the association between Ayurvedic phenotype and microbiome.

Brahma–The Indian Brain Template

BRAHMA is the name given to the Indian Brain template developed by scientists in 2020 based on a research study of two years. In simple terms, a brain template is like a map that provides a standard reference coordinate system to analyse the structure of the brain. The National Brain Research Centre initiated the project in 2018 and successfully completed the construction of the first high-resolution Indian population specific brain template in 2020.

The template is developed using the MRI scans of healthy Indians and shows intrinsic details of the anatomy of the Indian brain. So far, scientists have relied on the brain templates of other countries during brain surgeries, diagnosis, and treatment of brain-related disorders. There could be significant variations in the key brain regions among different racial types. Hence, several countries developed their own brain templates to serve as a reference point for effective surgeries and treatment. For instance, China and Canada have a brain template of their population. The development of Indian brain template, therefore, is expected to serve as a better guide to neuroscientists and surgeons.

Manav: Human atlas Initiative

Ayurvedic phenotype

In Ayurveda, a person's Prakriti, that is, a person's nature is determined by the proportion of three doshas called Vata, Pitta, and Kapha. These doshas are the primary functional energies in the human body. According to Ayurveda, the state of these doshas determine the health of an individual. If the three doshas are balanced, an individual is healthy. Any imbalance in the doshas causes ill health.

Each person is characterised by a predominant dosha type and accordingly, will exhibit different phenotypes. In other words, certain characteristics and diseases are associated with Vata, Pitta, or Kapha phenotypes, respectively. For instance, Vata Prakriti individuals are characterized by dry skin and hair and lean phenotypes. They are generally susceptible to fatigue, insomnia, nervous system related disorders, etc. On the other hand, Pitta Prakriti individuals tend to develop inflammation related disorders such as ulcers. The Kapha Prakriti individuals have a tendency to be heavy and are susceptible to obesity and respiratory disorders.

Ayurvedic healing hence focuses on treating the imbalances of doshas. The treatment is unique to each dosha type.

Manav—Human Atlas initiative aims to collate and present all the relevant macro and micro data with reference to the entire human body in a single place. The objective of the project is to provide reliable information at a single point to researchers, students, teachers and medical professionals. This will help improve our understanding of the working of the human body, trace causes of various diseases and changes to the tissues and cells at different stages of diseases and design better therapeutic agents to treat diseases.

MANAV is derived from the Sanskrit word 'Manav', which means Human. The Atlas would be created by mapping the molecular level details of each cell, tissue, and organ in the human body. In simple terms, all the relevant information will be sourced from existing scientific literature and public databases and presented in the Atlas. The project aims to understand and capture the information on human physiology in two stages: normal (healthy) and diseased stages. The final year students in the selected disciplines will be trained in the curation of information and annotation using specialised tools.

The project was launched in 2019 under public–private partnership by the Department of Biotechnology and Persistent Systems (a technology services private company),

Indian Institute of Science Education and Research (IISER), Pune, and National Center for Cell Sciences (NCCS), Pune.

Practice Questions

1. Which of the following benefits may be reaped from transgenic crops?
1. Increased shelf life
2. Higher crop production
3. Higher nutritional value
Select the correct answer using the codes given below:
(a) 1 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

2. Consider the following statements with reference to 'gene testing':
1. Gene testing can be used to determine a person's ancestry.
2. Gene testing may help in preventing genetic disorders.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

3. Which one of the following techniques can be used to establish the paternity of a child?
(a) Protein analysis
(b) Chromosome counting
(c) Quantitative analysis of DNA
(d) DNA fingerprinting

4. The application of biotechnology to make industrial processes more efficient is called:
(a) Green biotechnology
(b) Blue biotechnology
(c) White biotechnology
(d) Red biotechnology

5. Consider the following statements regarding stem cells:

1. In our day to day lives, stem cells regularly replace dead cells from our body tissues.
2. Stem cells help in understanding the occurrence of degenerative diseases.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

6. Consider the following statements regarding traditional plant breeding:
1. Traditional plant breeding cannot be carried among interspecies.
2. Traditional plant breeding is carried through recombinant DNA technology.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

7. The term 'Cirb Gaurav' is used in the context of:
(a) A cloned monkey offspring
(b) First cloned human offspring
(c) A cloned buffalo offspring
(d) First cloned sheep offspring

8. Which of the following are ex-situ bioremediation techniques:
1. Bioaugmentation
2. Biopile
3. Land farming
Select the correct answer using the codes given below:
(a) 1 and 2 only
(b) 2 and 3 only
(c) 3 only
(d) 1, 2, and 3

9. Consider the following statements regarding golden rice:
1. The golden yellow colour of the rice is due to the presence of high protein in the rice.
2. The golden rice is fortified with proteins in order to eliminate protein deficiency.
Which of the statements given above is/are correct?
(a) 1 only

(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

10. Consider the following statements regarding cloning:
1. World's first cloned animal was Dolly, the sheep.
2. The first case of human cloning was recorded in Germany in 2004.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

11. Consider the following statements with regard to DNA analysis:
1. DNA fingerprinting has become an important test to establish the paternity and identity the criminals of rape cases.
2. Dried blood and semen are adequate for DNA analysis.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

12. The term 'virotherapy' refers to:
(a) A virus that is used to treat leukemia
(b) A medical treatment to kill foreign virus that may cause some disease
(c) A medical treatment that converts viruses into therapeutic agents
(d) A medical treatment that is used to cure any genetic disorder

13. Consider the following statements:
1. If scientists could locate and extract the DNA out of a lock of Einstein's hair, another Einstein could be produced by cloning.
2. The DNA extracted from the cell of an embryo at an early stage of development can be transferred to denucleated egg, which in turn can be implanted into the uterus of a surrogate mother to give birth to an identical offspring.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2

(d) Neither 1 nor 2

14. Consider the following statements:
1. Phytomining is an approach in which mining is done with the help of plants.
2. Biosorption is an approach in which extraction of desired minerals from ores is done with the help of living organisms.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

15. Stem cell therapy (SCT) is not useful for the treatment of which one of the following ailments?
(a) Kidney-related ailments
(b) Hypertension
(c) Liver damage
(d) Vision impairment

16. The term 'biomimetics' is sometimes seen in the news with reference to:
(a) Implementation of concepts from nature to create new materials, devices, and systems.
(b) Use of rare earth metals to create artificial body parts which can function like real parts.
(c) Implementation of cloning technique to create exact replica of physical devices.
(d) Use of biotechnology principles in day-to-day life for a comfortable living.

17. With reference to the latest developments in stem cell research, consider the following statements:
1. The only source of human stem cells is the embryo at the blastocyst stage.
2. The stem cells can be derived without causing destruction to blastocysts.
3. The stem cells can automatically regenerate themselves.
Which of the statements given above is/are correct?
(a) 1 and 2 only
(b) 1, 2, and 3
(c) 1 only
(d) 3 only

18. With reference to 'biological engineering', consider the following statements:

1. It is an interdisciplinary area focusing on the application of engineering principles to analyse biological systems and solve problems related to biological systems.
2. Greater fundamental knowledge of biology and its potential applications are pre-requisites for biological engineering.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

19. With reference to the rDNA technology, consider the following statements:
1. The restriction enzymes attach themselves to a DNA base of the bacteria and restricts the DNA from being digested.
2. The modification enzymes derived from bacteria are used to cut a DNA at a particular location.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

20. Why is 'alkaline phosphatase' used in rDNA technology?
(a) To act as a vehicle to carry the required gene
(b) To prevent self-ligation
(c) To connect two DNA strands together
(d) To cut a DNA strand precisely

21. Consider the following statements with reference to CRISPR-Cas9:
1. CRISPR-Cas9 is a part of bacteria's immune system that protects it from viral infections.
2. CRISPR-Cas9 acts as a molecular scissors, and is used to cut DNA precisely.
3. The location at which CRISPR needs to cut, the DNA is specified by the RNA sequence of Cas9.
Which of the statements given above is/are correct?
(a) 1 and 2 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

22. Which of the following statements is/are incorrect with reference to the Genome India project?
1. Project Genome India aims to collect genome data from around 1,00,000 Indian citizens to carry out Indians specific genome sequencing.
2. India was a part of the Human Genome Project completed in 2003.
3. The Genome India Project is being led by the Indian Institute of Science, Bengaluru.
Select the correct answer using the codes given below:
(a) 1 and 2 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

23. Which of the following pairs is/are correctly matched?

Type of Food	Fortified with
1. Rice	Folic acid
2. Edible oil	Vitamin D
3. Salt	Iron

Select the correct answer using the code given below:
(a) 1 and 2 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

24. Which of the following pairs is/are incorrectly matched?

Project	Focus area
1. BRAHMA	The Human Atlas Initiative of India
2. MANAV	The Indian Brain Template
3. SAMRUPA	The Genome India Project

Select the correct answer using the codes given below:
(a) 2 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

25. At the present level of technological advancement, which of the following statements is/are correct with reference to the organoids derived from stem cells?

1. The organoids can be derived from only embryonic stem cells but not from induced pluripotent stem cells.
2. Organoids can be used to study human immune responses to various infections such as Covid-19.
3. The organoids can exactly replicate the human immune system and completely replace the need for humans in clinical trials.
Select the correct answer using the codes given below:
(a) 2 only
(b) 1 and 3 only
(c) 1 and 2 only
(d) 1, 2, and 3

26. Consider the following statements:
1. The Genetic Engineering Appraisal Committee was formed under the provisions of the Environment Protection Act, 1986.
2. The GEAC functions as an advisory body under the Ministry of Agriculture.
3. The GEAC is responsible to grant approval for the cultivation of genetically modified crops.
Which of the statements given above is/are correct?
(a) 1 and 2 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

27. The process in which new materials and systems are created artificially by adapting the methods and systems found in nature is called
(a) Bioprospecting
(b) Biomimetics
(c) Bionics
(d) Bioinformatics

28. Which of the following genetically modified crops are presently not allowed for cultivation in India?
1. HT Bt cotton
2. Bt Mustard
3. Bt Brinjal
4. Bt Soyabean
Select the correct answer using the code given below:
(a) 2, 3, and 4 only

(b) 1, 2, and 3 only
(c) 1, 3, and 4 only
(d) 1, 2, 3, and 4

29. With reference to gene editing, consider the following statements:
1. DNA Ligase is the enzyme used to connect two strands of DNA together in gene editing.
2. Methylation is used to prevent self-ligation in DNA molecules during gene editing.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

30. Which of the following is/are the benefits of the 'Genome India Project'?
1. The project will aid in developing personalised medicine in India.
2. The project aims to map the influence of diet, age, lifestyle, and geography on the gut microbiomes and the health of Indians.
Select the correct answer using the codes given below:
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

Perfecting Past Prelims

1. Mon 863 is a variety of maize. It was in the news for which of the following reasons? (2010)
(a) It is a genetically modified dwarf variety resistant to drought.
(b) It is a genetically modified variety that is pest-resistant.
(c) It is a genetically modified variety with ten times higher protein content than the regular maize crop.
(d) It is a genetically modified variety used exclusively for biofuel production.

2. Genetically modified 'golden rice' has been engineered to meet human nutritional requirements. Which one of the following statements best qualifies golden rice? (2010)
(a) The grains have been fortified with genes to provide three times higher grain yield per acre than other high-yielding varieties.

(b) Its grains contain pro-vitamin A, which upon ingestion is converted into vitamin A in the human body.
(c) Its modified genes causing the synthesis of all the nine essential amino acids.
(d) Its modified genes cause fortification of rice grains with vitamin D.

3. A genetically engineered form of brinjal, known as the Bt-brinjal, has been developed. The objective of this is (2011)
(a) To make it pest-resistant
(b) To improve its taste and nutritive qualities
(c) To make it drought-resistant
(d) To make its shelf life longer

4. What are the reasons for the people's resistance to the introduction of Bt brinjal in India? (2012)
1. Bt brinjal has been created by inserting a gene from a soil fungus into its genome.
2. The seeds of Bt brinjal are terminator seeds, and therefore the farmers have to buy the seeds before every season from the seed companies.
3. There is an apprehension that the consumption of Bt brinjal may have an adverse impact on health.
4. There is some concern that the introduction of Bt brinjal may have an adverse effect on biodiversity.
Which of the statements given above is/are correct?
(a) 1, 2 and 3 only
(b) 2 and 3 only
(c) 3 and 4 only
(d) 1, 2, 3, and 4

5. With reference to 'stem cells', frequently in the news, which of the following statements is/are correct? (2012)
1. Stem cells can be derived from mammals only.
2. Stem cells can be used for screening new drugs.
3. Stem cells can be used for medical therapies.
Select the correct answer using the codes given below:
(a) 1 and 2 only
(b) 2 and 3 only
(c) 3 only
(d) 1, 2, and 3

6. Other than resistance to pests, what are the prospects for which genetically

engineered plants have been created? (2012)
1. To enable them to withstand drought.
2. To increase the nutritive value of the produce.
3. To enable them to grow and do photosynthesis in spaceships and space stations.
4. To increase their shelf life.
Select the correct answer using the codes given below:
(a) 1 and 2 only
(b) 3 and 4 only
(c) 1, 2 and 4 only
(d) 1, 2, 3, and 4

7. Recombinant DNA technology (Genetic Engineering) allows genes to be transferred: (2013)
1. Across different species of plants.
2. From animals to plants.
3. From microorganisms to higher organisms.
Select the correct answer using the codes given below:
(a) 1 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

8. Mycorrhizal biotechnology has been used in rehabilitating degraded sites because mycorrhiza enables the plants to: (2013)
1. Resist drought and increase absorptive area.
2. Tolerate extremes of pH.
3. Resist disease infestation.
Select the correct answer using the codes given below:
(a) 1 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

9. Consider the following techniques/phenomena:
1. Budding and grafting in fruit plants
2. Cytoplasmic male sterility
3. Gene silencing
Which of the above is/are used to create transgenic crops? (2014)
(a) 1 only

(b) 2 and 3
(c) 1 and 3
(d) None

10. The Genetic Engineering Appraisal Committee is constituted under the: (2015)
(a) Food Safety and Standards Act, 2006.
(b) Geographical Indications of Goods (Registration and Protection) Act, 1999.
(c) Environment (Protection) Act, 1986.
(d) Wildlife (Protection) Act, 1972.

11. What is the application of somatic cell nuclear transfer technology? (2017)
(a) Production of biolarvicides.
(b) Manufacture of biodegradable plastics.
(c) Reproductive cloning of animals.
(d) Production of organisms that are free of diseases.

12. With reference to agriculture in India, how can the technique of 'genome sequencing', often seen in the news, be used in the immediate future? (2017)
1. Genome sequencing can be used to identify genetic markers for disease resistance and drought tolerance in various crop plants.
2. This technique helps in reducing the time required to develop the new varieties of crop plants.
3. It can be used to decipher the host–pathogen relationships in crops.
Select the correct answer using the codes given below:
(a) 1 only
(b) 2 and 3 only
(c) 1 and 3 only
(d) 1, 2, and 3

13. In the context of the developments in bioinformatics, the term 'transcriptome', sometimes seen in the news, refers to (2016)
(a) A range of enzymes used in genome editing.
(b) The full range of mRNA molecules expressed by an organism.
(c) The description of the mechanism of gene expression.
(d) A mechanism of genetic mutations taking place in cells.

14. With reference to the Genetically Modified mustard (GM mustard) developed in India, consider the following statements: (2018)
1. GM mustard has the genes of a soil bacterium that give the plant the property of

pest resistance for a wide variety of pests.
2. GM mustard has the genes that allow plant cross-pollination and hybridization.
3. GM mustard has been developed jointly by the IARI and Punjab Agricultural University.
Which of the following statements given above is/are correct?
(a) 1 and 3 only
(b) 2 only
(c) 2 and 3 only
(d) 1, 2, and 3

15. With reference to the recent developments in science, which one of the following statements is not correct? (2019)
(a) Functional chromosomes can be created by joining segments of DNA taken from cells of different species.
(b) Pieces of artificial functional DNA can be created in Laboratories.
(c) A piece of DNA taken out from an animal cell can be made to replicate outside a living cell in a laboratory.
(d) Cells taken out from plants and animals can be made to undergo cell division in laboratory petri dishes.

16. 'RNA interference (RNAi)' technology has gained popularity in the last few years. Why? (2019)
1. It is used in developing gene silencing therapies.
2. It can be used in developing therapies for the treatment of cancer.
3. It can be used to develop hormone replacement therapies.
4. It can be used to produce crop plants that are resistant to viral pathogens.
Select the correct answer using the codes given below.
(a) 1, 2, and 4
(b) 2 and 3
(c) 1 and 3
(d) 1 and 4 only

17. Consider the following statements: (2020)
1. Genetic changes can be introduced in the cells that produce eggs or sperms of a prospective parent.
2. A person's genome can be edited before birth at the early embryonic stage.
3. Human induced pluripotent stem cells can be injected into the embryo of a pig.
Which of the statements given above is/are correct?
(a) 1 only

(b) 2 and 3 only
(c) 2 only
(d) 1, 2, and 3

18. With reference to recent developments regarding 'Recombinant vector Vaccines' or DNA vaccines, consider the following statements: (2021)
1. Genetic engineering is applied in the development of these vaccines.
2. Bacteria and viruses are used as vectors.
Which of the statements given above is/are correct?
(a) 1 only
(b) 2 only
(c) Both 1 and 2
(d) Neither 1 nor 2

19. Bollgard I and Bollgard II technologies are mentioned in the context of (2021)
(a) Clonal propagation of crop plants
(b) Developing genetically modified crop plants
(c) Production of plant growth substances
(d) Production of biofertilizers

20. Consider the following statements: DNA Barcoding can be a tool to:
1. assess the age of a plant or animal.
2. distinguish among species that look alike.
3. identify undesirable animal or plant materials in processed foods.
Which of the statements given above is/are correct?
(a) 1 only
(b) 3 only
(c) 1 and 2
(d) 2 and 3

Answer Keys

Practice Questions

1. (d)	2. (c)	3. (d)	4. (c)	5. (c)
6. (d)	7. (c)	8. (b)	9. (d)	10. (a)
11. (c)	12. (c)	13. (b)	14. (a)	15. (b)
16. (a)	17. (d)	18. (c)	19. (d)	20. (b)
21. (a)	22. (a)	23. (d)	24. (d)	25. (a)
26. (c)	27. (b)	28. (d)	29. (a)	30. (a)

Perfecting Past Prelims

1. (b)	2. (b)	3. (a)	4. (c)	5. (b)
6. (c)	7. (d)	8. (d)	9. (b)	10. (c)
11. (c)	12. (d)	13. (b)	14. (b)	15. (a)
16. (a)	17. (d)	18. (c)	19. (b)	20. (d)

Solutions

Practice Questions

2. (c) Statement 1 is correct: Gene testing or genetic genealogy involves the examination of DNA variations which provide clues about where a person's ancestors might have come from.

Statement 2 is correct: Genetic testing is a type of medical test that identifies changes in genes. The results of a genetic test can confirm or rule out a suspected genetic condition or help determine a person's chance of developing or passing on a genetic disorder.

5. (c) Statement 1 is correct: Stem cells are responsible for the repair of damaged tissues, and the replacement and regeneration of tissues that turn over rapidly, such as the skin, blood, or the lining of the intestine.

Statement 2 is correct: Embryonic stem cells possess the property of pluripotency i.e., they have the potential to divide into different cell types. By isolating specific cell types related to the degenerative diseases, the diseases can be studied and in the long-run may also be cured.

6. (d) Statement 1 is incorrect: Traditional plant breeding, unlike animal breeding, can

be carried inter-species.

Statement 2 is incorrect: Traditional plant breeding involves techniques like grafting, natural pollination, etc. Recombinant DNA technology is used to create transgenic or genetically modified varieties.

8. (b) Biopiles and land farming are ex-situ bioremediation techniques. Bio augmentation is in situ biological treatment, involving mainly microbes, to clean up the hazardous contaminants in soil and water.

9. (d) Statement 1 is incorrect: The yellow colour of 'golden rice' is due to the presence of β-carotene (pro-vitamin A) and xanthophylls (yellow-coloured pigment).

Statement 2 is incorrect: Golden rice is rich in β-carotene (pro-vitamin A) that is effective in curtailing vitamin A deficiency (VAD). It is not fortified with proteins.

13. (b) Statement 1 is incorrect: A lock of hair is mostly comprised of protein and not cellular material, so it will not carry any human genomic DNA.

Statement 2 is correct: This is the process used in cloning.

14. (a) Statement 2 is incorrect: Biosorption is a property of certain types of inactive, dead, microbial biomass to bind and concentrate heavy metals from even very dilute aqueous solutions.

15. (b) Hypertension is not related to particular damaged cell or tissue. It is the medical condition related to the overall functioning of a body.

17. (d) Statement 1 is incorrect: Stem cells can also be obtained from body cells (bone marrow, adipose tissue, blood, etc.) and other sources such as umbilical cord.

Statement 2 is incorrect: Stem cells can be obtained from the blastocyst by only removing the embryo from the womb and taking the cells which were destined to form tissues of the growing foetus.

19. (d) Statement 1 is incorrect: The modification enzymes attach themselves to a DNA base of the bacteria and prevents the DNA from being digested.

Statement 2 is incorrect: The restriction enzymes derived from bacteria are used to cut

a DNA at a particular location.

20. (b) The vector (vehicle which carries required DNA sequence) is treated with alkaline phosphatase to prevent self-ligation.

21. (a) Statement 3 is incorrect: The location at which Cas9 needs to cut, the DNA is specified by the RNA sequence of CRISPR.

22. (a) Statement 1 is incorrect: Project Genome India aims to collect genome data from around 10,000 Indian citizens, not a lakh.

Statement 2 is incorrect: The Human Genome Project is a collaborative effort between the United States, France, the United Kingdom, China, Japan and Germany.

23. (d) Double Fortified salt is fortified with iron along with iodine.

24. (d) Pair 1 is incorrectly matched: BRAHMA project is to identify Indian Brain Template.

Pair 2 is incorrectly matched: MANAV is the Human Atlas Initiative.

Pair 3 is incorrectly matched: SAMRUPA is the world's first cloned buffalo calf by the National Dairy Research Institute.

25. (a) Statement 1 is incorrect: Organoids can be derived from induced pluripotent stem cells as well.

Statement 3 is incorrect: One of the major limitations of organoids is that they may not replicate the exact human immune response. As they are grown in controlled environment, their immune levels are lower than the acquired immunity levels exhibited by the actual human organs.

26. (c) Statement 2 is incorrect: The GEAC functions under the Ministry of Environment, Forest and Climate Change.

28. (d) The HT Bt stands for Herbicide-tolerant Bt Cotton. It adds another genetic modification to the Bt cotton crop to make the crop resistant to herbicide. As a result, farmers can spray herbicides to kill the weeds without harming the cotton plant. However, GEAC has not approved the use of HT Bt cotton in India.

29. (a) Statement 2 is incorrect: Methylation is part of the self defense mechanism in bacteria. DNA fragments are treated with an enzyme called alkaline phosphatase to prevent self-ligation in DNA molecules during gene editing.

30. (a) Statement 2 is incorrect: The Human Microbiome Initiative (not the Genome India project) aims to map the influence of diet, age, lifestyle, and geography on the gut microbiomes.

Perfecting Past Prelims

1. (b) MON 863 is a genetically modified variety of maize. It was in the news because it was genetically modified to resist pest.

4. (c) Statements 1 and 2 are incorrect.

Statement 3 is correct: Bt toxin trait works against certain insects that have alkaline pH digestive tract. A similar effect was feared in the digestive tracts of higher mammals such as humans.

Statement 4 is correct: It is feared that cross pollination of Bt gene with the local weeds may produce a new variety of super weeds that would be immune to weedicides.

5. (b) Statement 1 is incorrect: Stem cells can be derived from both plants and animals.

6. (c) Statement 3 is incorrect: GM plants are not modified to grow and photosynthesise in spaceships and space stations. There is an absolute absence of gases in space, which are the primary requirements to carry out photosynthesis.

7. (d) Statement 1 is correct: Genetic engineering has enabled the transfer of genes across different plant species such as daffodil plant gene in golden rice. Statements 2 and 3 are correct: The Bt gene of Bacillus thuringiensis is transferred to various crops for making them pest-resistant. This signifies both gene transfer from animals to plants and from lower organisms to higher ones.

9. (b) Statement 1 is incorrect: Budding and grafting in fruit plants are traditional methods of breeding, and are not used to create transgenic crops.

14. (b) Statement 1 is incorrect: Unlike most GM crops, GM Mustard do not have genes of soil bacterium. It is produced by cross pollinating Indian mustard with East

European mustard.

Statement 3 is also incorrect: GM Mustard has been prepared by researchers of Delhi University.

Statement 2 is correct.

15. (a) Functional chromosomes can be created by joining segments of DNA taken from cells of different species.

This question can be solved through elimination technique.

Let us start from option (d). It is easiest. The statement given in answer option can be eliminated on the basis of cloning technique. In the process of cloning, cells taken out from plants and animals undergo cell division in laboratory petri dishes.

Options (b) and (c) can also be eliminated. Synthetic biology is used to create pieces of artificial functional DNA. It can also be used to replicate DNA outside a living cell.

16. (a) 1, 2, and 4

Statement 1 is correct: As RNA transcribes genetic information from the DNA found in a cell's nucleus and then carries this information to other cell organelles, RNAi is used in developing gene silencing therapies.

Statement 2 is correct: Cancer is abnormal growth of body cells which is related to DNA present in them. As DNA are expressed through RNA, RNAi can be used in developing therapies for-the treatment of cancer.

Statement 3 is incorrect: Genetic information has not been intrinsically related to our hormone system. Thus, RNAi cannot be used to develop hormone replacement therapies.

Statement 4 is correct: RNA can impact expression of DNA which may make plant vulnerable to viral pathogens. Thus, RNAi can be used to produce crop plants that are resistant to viral pathogens.

17. (d) 1, 2 and 3

Statement 1 is correct: Genetic changes are possible in parent cells. Thus, Genetic changes can be introduced in the cells that produce eggs or sperms of a prospective parent.

Statement 2 is correct: A person's genome can be edited before birth at the early embryonic stage.

Statement 3 is correct: Human induced pluripotent stem cells can be injected into the embryo of a pig. This statement is based on a recent successful scientific experiment and is thus, part of current affairs.

20.(d) DNA barcoding is a system for species identification focused on the use of a short, standardized genetic region acting as a "barcode" in a similar way that Universal Product Codes (UPCs) are used by supermarket scanners to distinguish commercial products.

Statement 1 is incorrect: DNA barcoding has got no association with determining the age of an organism. So, Statement 1 is not correct.

Statement 2 is correct: DNA barcoding is a system for fast and accurate species identification that makes ecological system more accessible by using short DNA sequence instead of whole genome.

Statement 3 is correct: In recent times, DNA barcoding has emerged as the most potent tool for detection of adulteration of food with unwanted plant and animal material. So, statement 3 is correct.