

Biochemical Engineering

Chemical Engineering - Handling of chemicals on a large scale

Biochemical engineering - Handling of biological systems on a large scale

- Biological systems are stable for a very narrow range of time.
- Two chemical systems on interaction doesn't change in mass or energy which isn't always true in the case of biological systems.
- Basically, biological systems grow by themselves and we have a very narrow range during which they are stable.
- Biology just tells us what happens in a process, while chemistry explains the reason behind the happening of it.
- Alcohol and medicines are some of the best examples of biochemical engineering
- Biomass is protein rich product (70% protein)

→ Plants

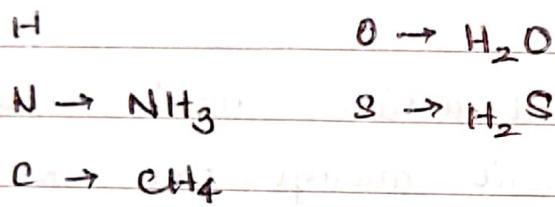
Autotrophic carbon Assimilation : Method to produce biomass

→ Plants assimilate carbon in themselves, collect water, minerals etc. to produce biomass.

Milner's Experiment (1953) - Proposed to make biomolecules from simple gases NH_3 , CH_4 , H_2O , H_2S

Miller hypothesized that there are certain env. in state which should be provided for the synthesis of the biomolecules.

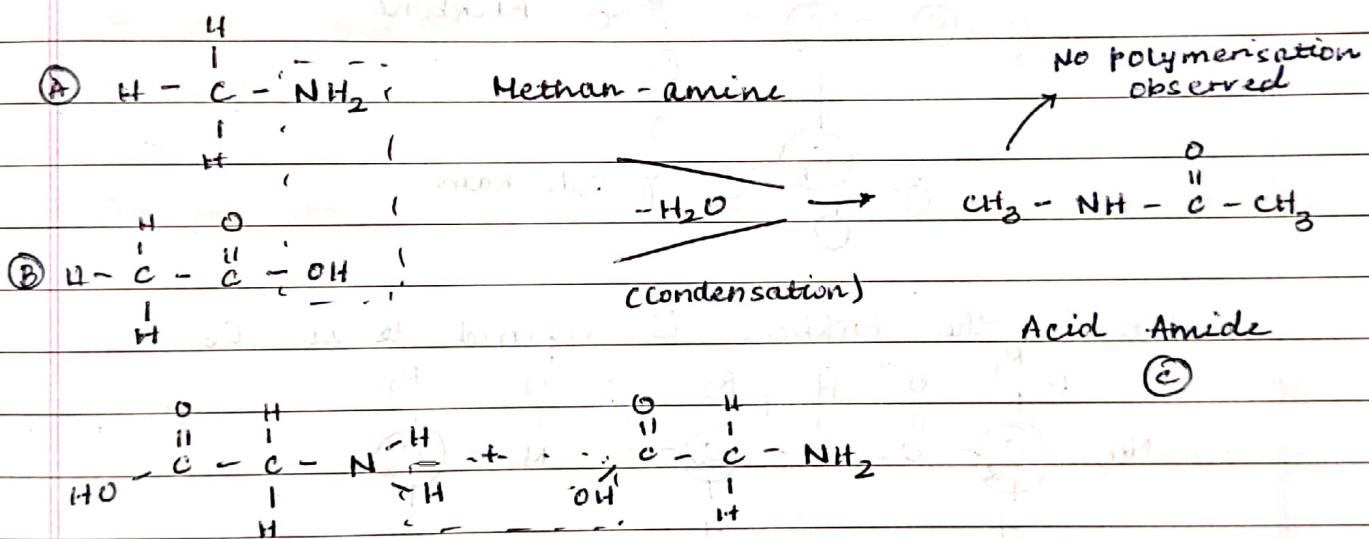
After several trials, he put a lightning arc for the formation of biomolecules in an abiotic method.



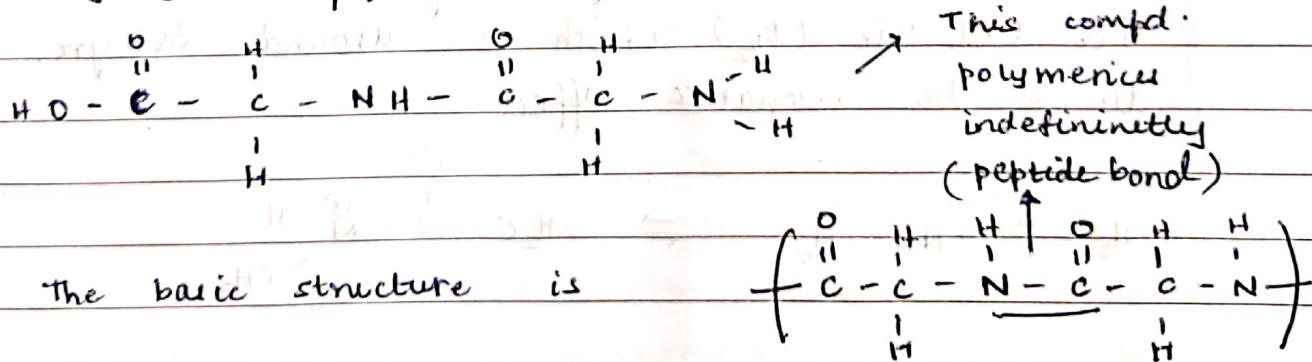
The way Miller analysed proteins is called primordial soup (70% of dried biomass)

Proteins

- They are very complex (~7000 atoms in an avg molecule)
- They are entangled in a compact form (tube rep.)
- Some parts are helices, some are flat and some are neither
- Residue Name: "Residue Name"
- Proteins are polymers (there are diff. no. of monomeric units that have been polymerised to form protein)
- There are various chains that are entangled, passing and crossing each other
- With change in structure, the fn. changes.
- Every protein is a poly-peptide



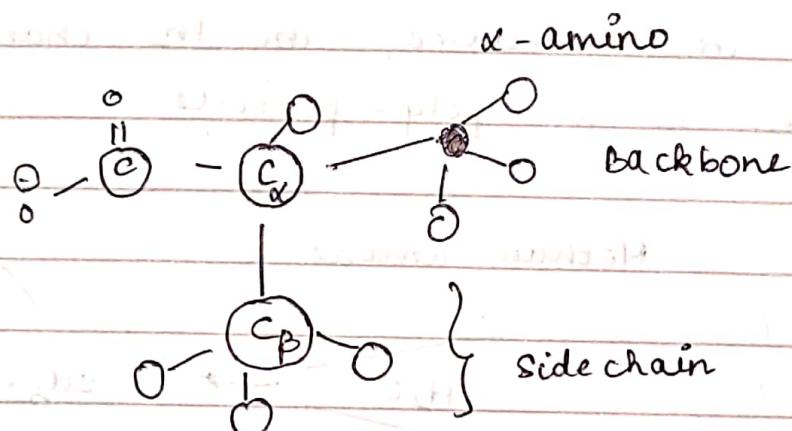
(Hydrolysis) $\text{H}_2\text{O} \uparrow \downarrow -\text{H}_2\text{O}$ (condensation)



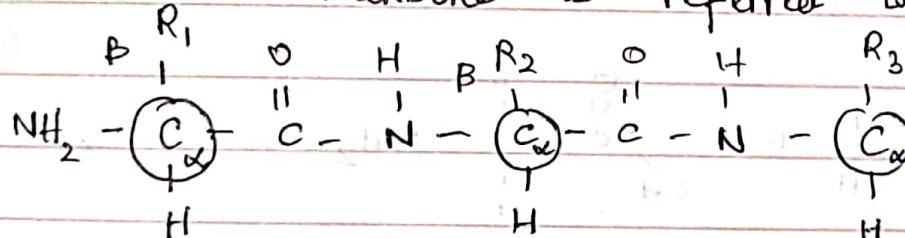
There are $-\text{CH}_2-$ groups which are sandwiched b/w $-\text{CO-NH-}$ groups, this $-\text{CH}_2-$ causes the different

monomeric units.

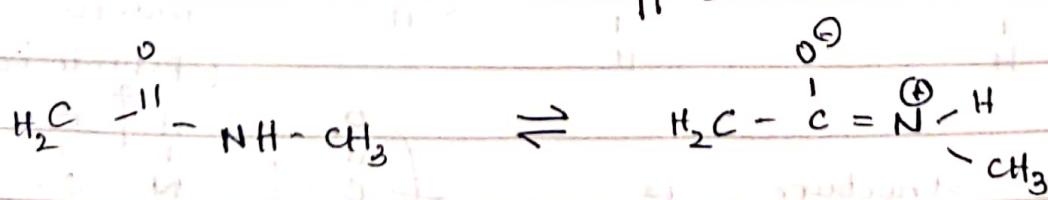
- There are 20 naturally occurring amino acids. This cont. C-N chain is called protein backbone, remaining extensions from backbone is called sidechain
- The difference in two proteins is mainly in the side chains and in its main chain the no. of monomeric units might change but structure is same.



Carbon in the backbone is referred to as C_α .



The peptide bond is very strong, this is clear from their half life ($t_{1/2}$) which is around 350 yrs, due to the resonance effect.



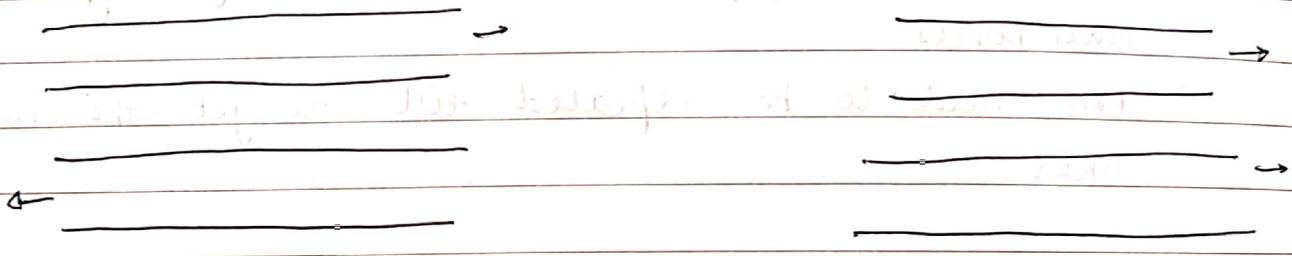
- In a chain, the two terminals must have one $-NH_2$ (N-terminus) and one $-COO^{\ominus}$ (C-terminus)
- The primary structure of protein, is the sequence in which the R groups are present from N-terminus to C-terminus
- Edman Degradation is to add the protein to an acid followed by heating resulting in breakage of the peptide bond.
 - ① More than the peptide bond, it might effect the other bonds
 - ② It needs to be repeated till we get the monomeric unit

The polymeric chain of the protein is getting degrading

- We perform Mass Spectroscopy which divides the polymer into fragments based on mass to charge ratio and the compare these fragments with available data to get the order.
- FASTA format:
 - ① comment
 - ② Amino Acid letterseach line with a max of 80 char

The $C=O$ and NH are in the $C-N$ are always in the same plane. This is the reason when each of these planes are viewed with respect to $C\alpha$, the $C-N$ planes are in different orientation

- These orientations lead to formation of coils.
- These coils are compressed due to H-bonding
- These coils are called helices.
- Most of the amino acids are ~~L-isomers~~ Right handed helices
 - The i -th residue makes H-bond with $(i+4)$ th residue.
 - The reason for this is most of the naturally occurring amino acids are L-isomers
- β -sheets : They formed sheets because of H-bonding

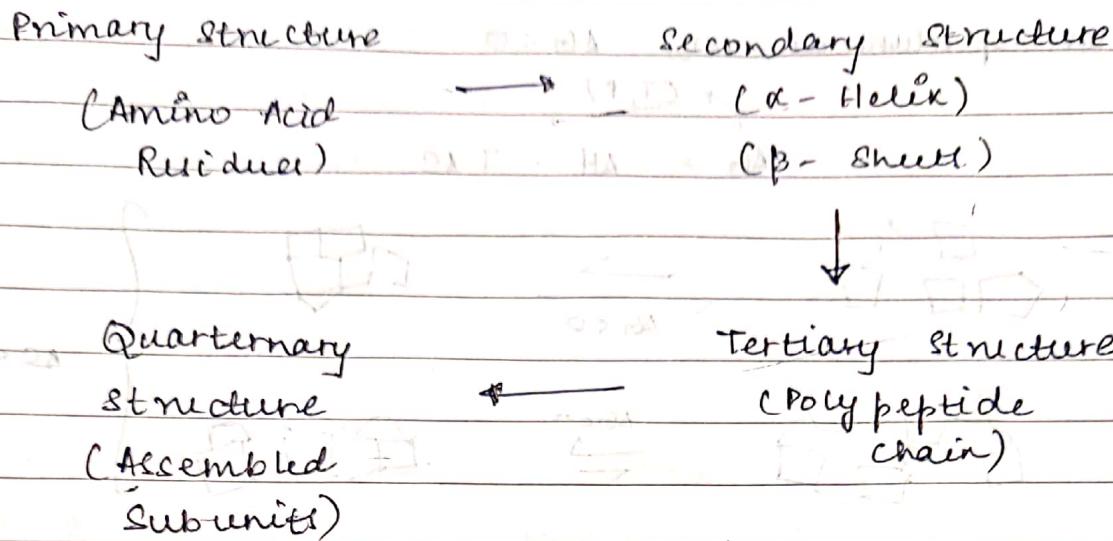


Anti-parallel sheet and Parallel sheet

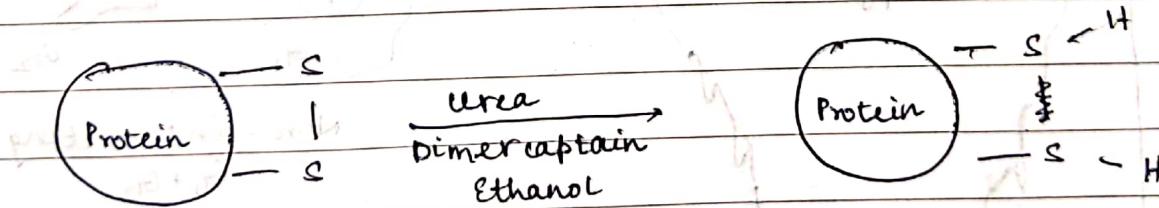
These β -sheets form α - β -Barrel

- Tertiary structure — Globular
- Fibrous
- Quaternary structure — Assembled subunits

(Poly peptide)



- Corresponding to a chain, there is a unique structure
- Alphafold is an AI system developed by DeepMind that predicts a protein's 3D structure from its amino acid sequence
- Mutation: change in sequence of amino acid or changing a particular amino acid(s) leading to change in structure of the protein
- Protein folding (Anfinsen's expt.)



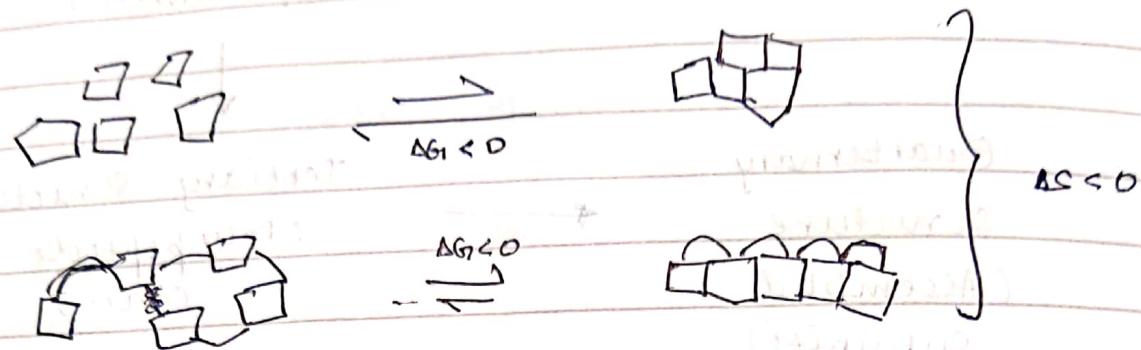
Corresponding to a chain, folding is unique

- Protein's fn. is due to the sidechain, not majorly due to the backbone.

(Chancery of 1999)

For spontaneous rxn: $\Delta G_1 < 0$
 $\rightarrow CT, P$

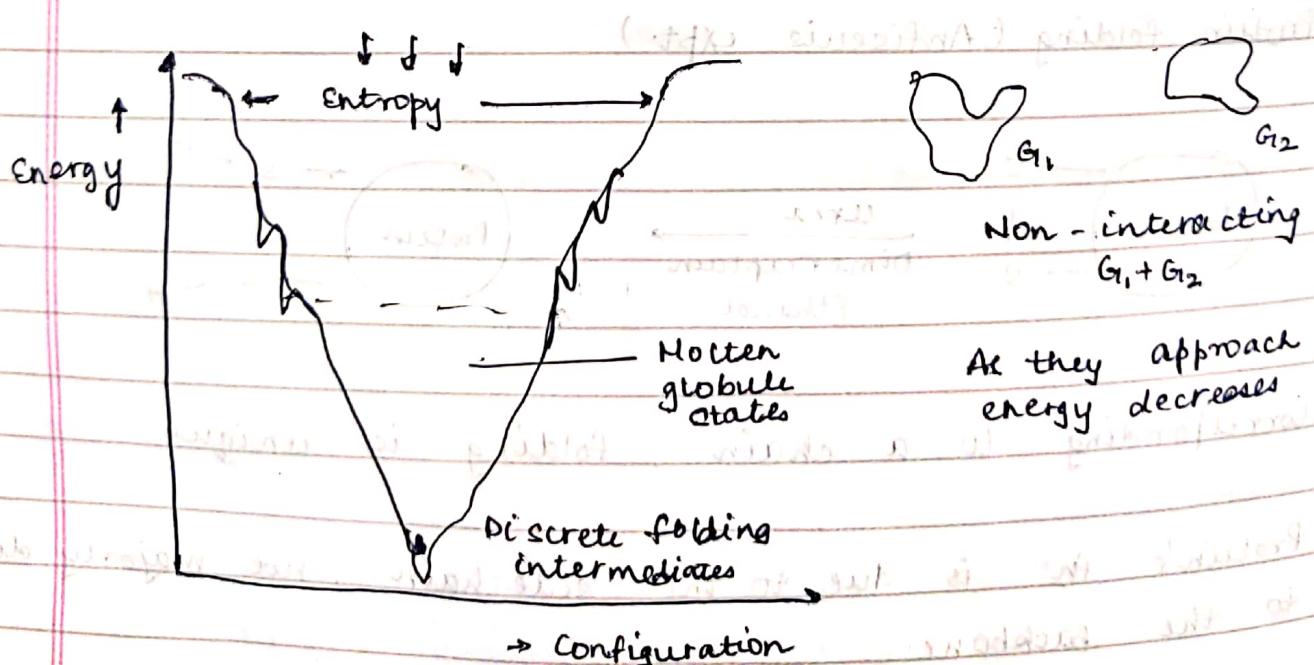
$$\Delta G_1 = \Delta H - T \Delta S$$



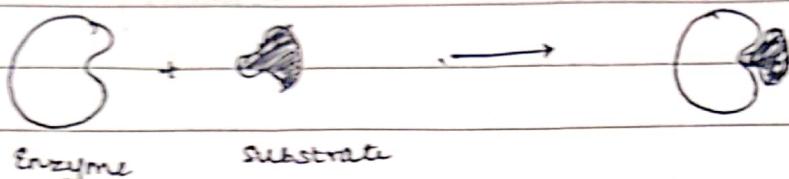
In case of no links, the decrease in ΔS is more compared to with links

For stability, the decrease in ΔH must be greater than ΔS decrease which is in the case with link and hence protein polymerise and fold.

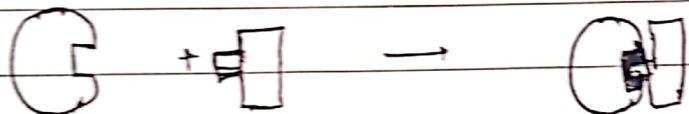
$$\Delta G_{\text{folding}} = G_{\text{folded}} - G_{\text{unfolded}} < 0$$



→ Lock and Key Mechanism - Displays that protein is rigid and until and unless we find the perfect key there is no interaction



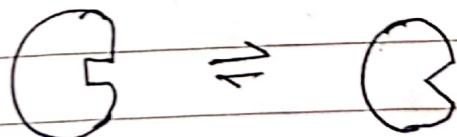
→ Induced fit Mechanism - Protein initially is normal and as the substrate approaches the protein, the protein automatically restructures itself so as to fit the substrate



This mechanism is the major reason for side-effects in drugs.

Enzymes are specific while chemical catalysts aren't, this property of proteins is called specificity.

→ Conformation Selection Mechanism



using one of the conformer to fit the substrate

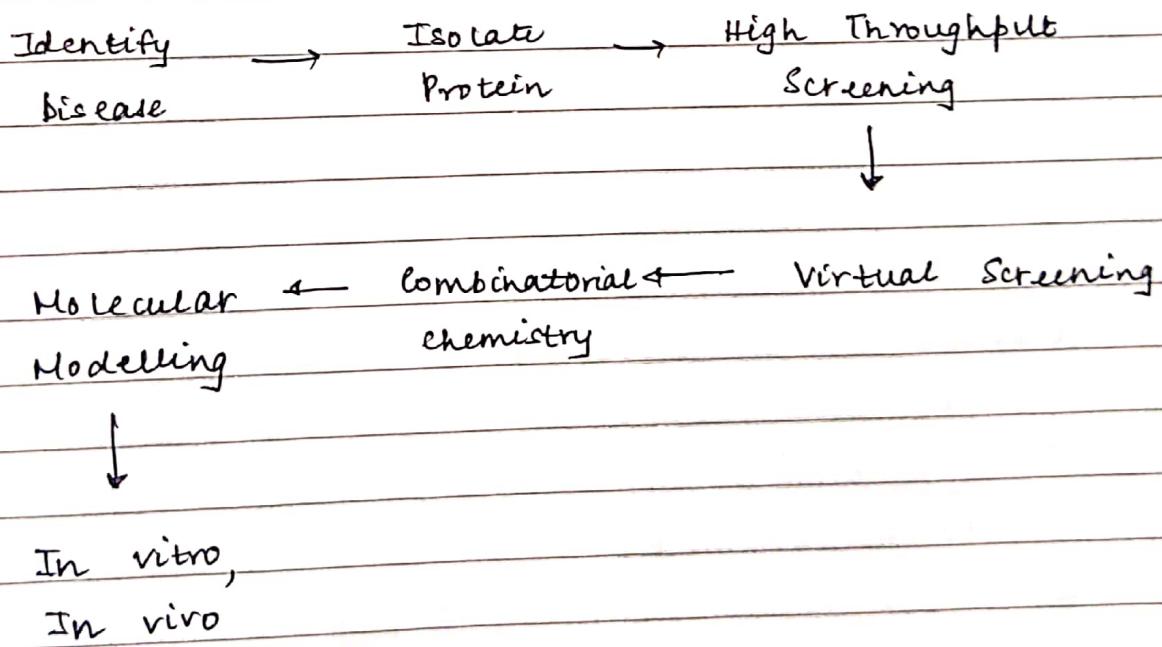
→ Protein Substrate Interaction

- ① Geometric Match
- ② Electrostatic Match

} → Thermodynamic stability

Sequence mismatch - mismatch with amino acid sequence, with side chains of different lengths and polarities.

- Diseases are identified from symptoms
- Proteomics is the study of proteomes
- Proteome is collection of all proteins in an organism
- High Throughput Screening - Screening up to 100k compounds a day for activity against a target protein
- Combinatorial chemistry - Rapidly producing vast no. of compds.
- In vitro - Experimenting ^{with} human samples on a petri dish
- In vivo - Experimenting inside the human body
- In silico - Computational



Molecular docking - ship comes to the dock and stays there, similarly the molecule comes and stays and the position of the molecule depends on the stability of the conformer

$$\text{energy} = U_{\text{bond}} + U_{\text{angle}} + U_{\text{dihedral}} + U_{\text{nonbond}}$$

Energy = sum of bond energy + angle energy + dihedral energy + nonbond energy

Each of these terms is proportional to the square of the distance between atoms.

For example, consider the interaction between two atoms separated by a distance r .

$$U(r) = \frac{A}{r^6} - \frac{B}{r^3}$$

$$U(r) = \frac{A}{r^6} - \frac{B}{r^3}$$

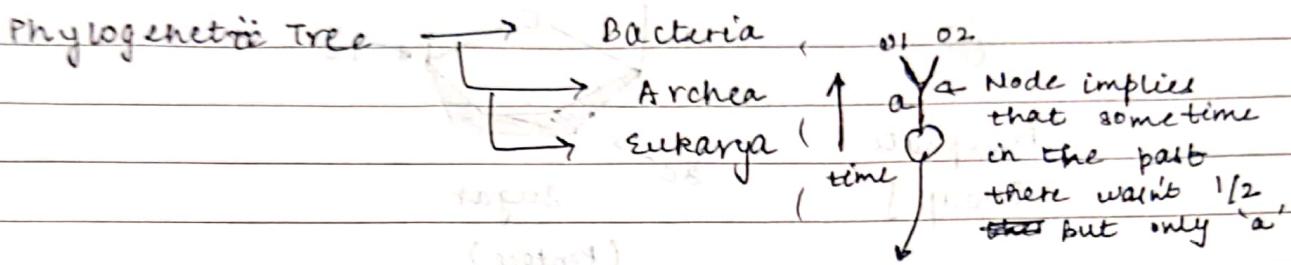


minimum at
large r

At large r , $U(r) \approx 0$ and $\frac{dU}{dr} \approx 0$
At small r , $U(r) \approx -\infty$ and $\frac{dU}{dr} \approx \infty$

Analysis of DNA and RNA

Phenotype means external appearance, protein arrangement themselves in a certain manner leading to different functionalities causing different phenotypical features



Today this organism
doesn't exist

Prokaryotes - Simple / Elementary Cells

Eukaryotes - Found in Multicellular bodies and are complex

Prokaryotes

→ Simple / Elementary cells → Complex cells

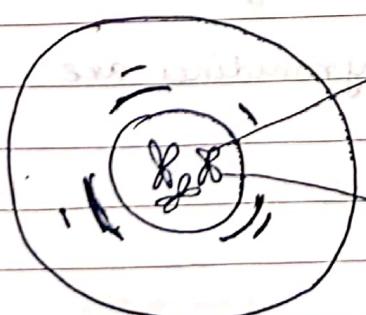
→ Nucleus is ill-defined

(Nuclear mass is floating around)

Eukaryotes

→ Nucleus is bounded

by nuclear membrane making it compact



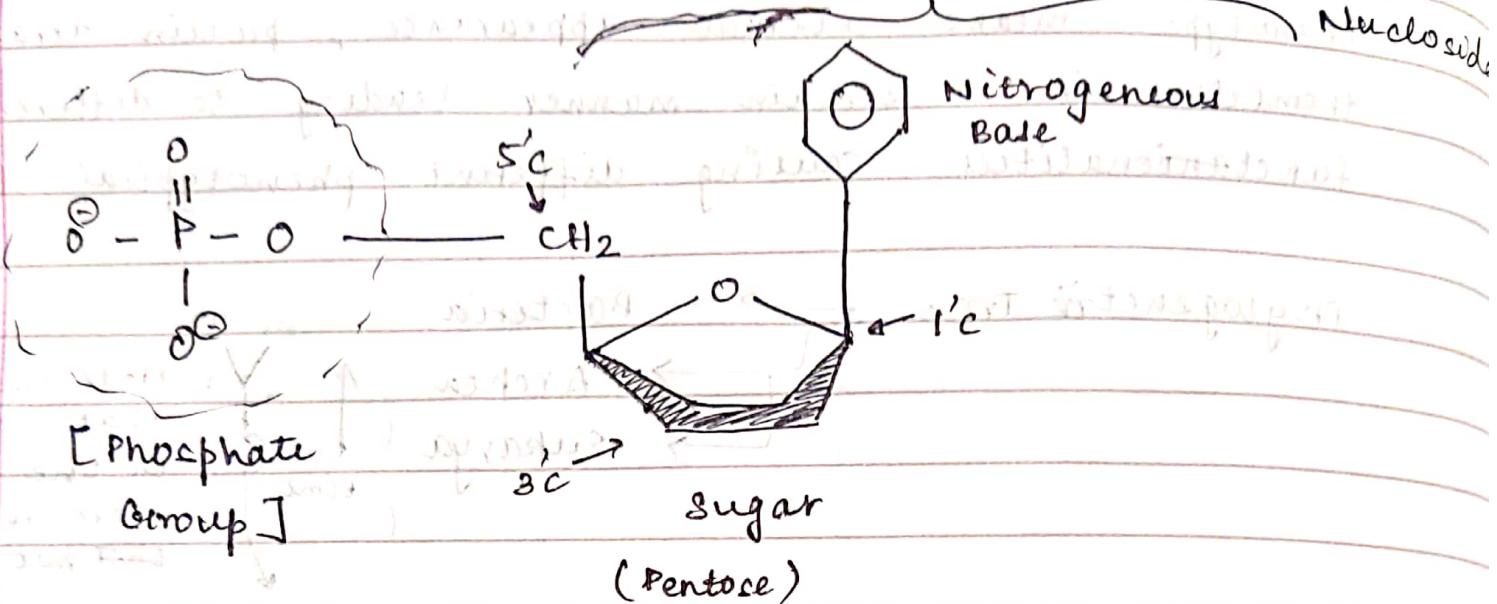
Chromosomes

(Compact Collection
of DNA)



(DNA)

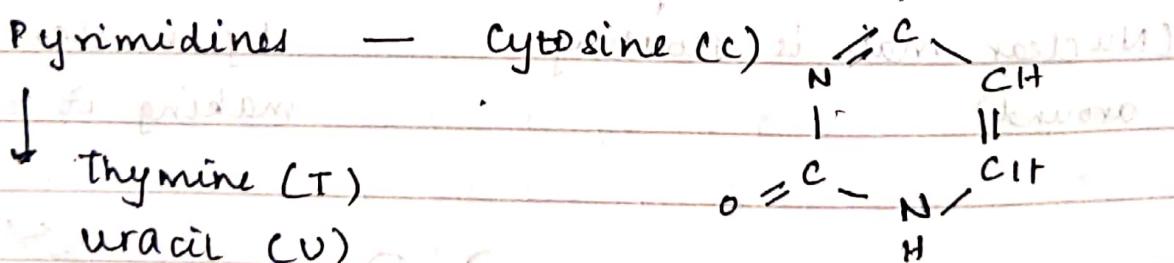
Nucleotide: Nucleic acid monomeric unit



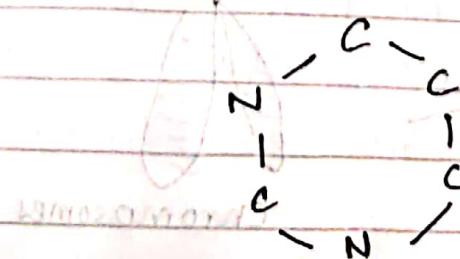
Nucleotide is a collection of phosphate, sugar and nitrogenous ^{bare} groups while nucleoside is a combination of pentose sugar and nitrogenous base and nucleo base is the N -base \rightarrow heterocyclic

Nucleic acid is the polymerisation of nucleotides

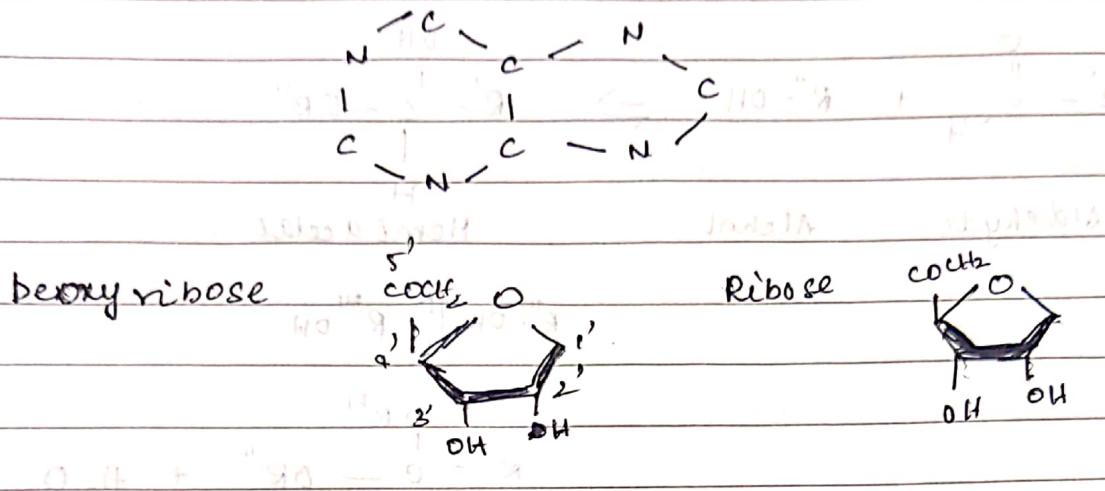
Nucleobases are of two categories pyrimidines (6-member rings) and purines (9-member rings)



The backbone structure of pyrimidines are



The backbone structure of purines



The difference b/w deoxyribose and ribose is the 2'-C's -OH group

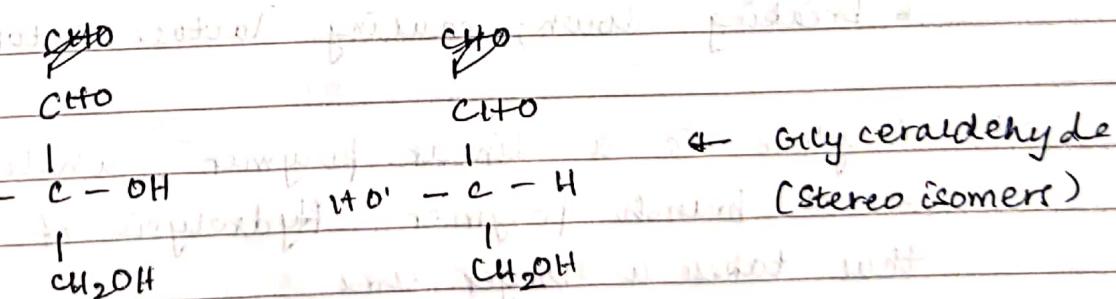
A detour to sugars

① Aldose - Aldehyde

Ketose - Ketone

② All sugars are polyalcohols (lot of -OH groups?)

③ Min. no. of C-atoms = 3



D-isomer

L-isomer

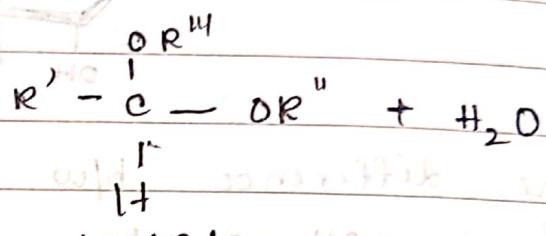
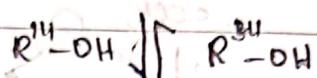
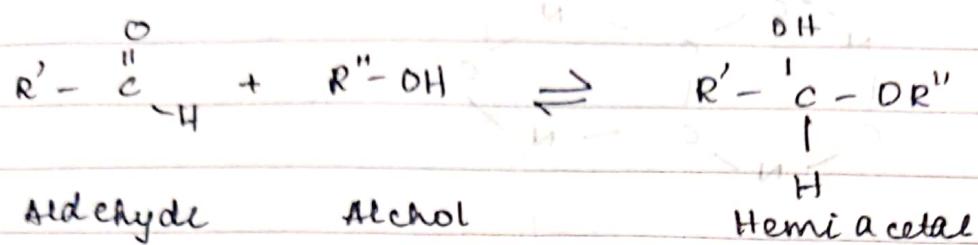
Glucose (aldohexose)

sugar with
aldehyde
group

Fructose is sugar sweetest
among all

Sucrose → Disaccharide of
glucose & fructose

→ Cyclisation of sugar



In α -Ribose sugar and α -D-Ribofuranose, cyclisation happens along in the chain itself

→ Lactose Intolerance: Lactose is broken down into mono saccharides by hydrolysis in presence of lactase. Due to age or genetic factors, there is absence of lactase leading to no breaking down; causing lactose intolerance.

→ Amylose is a linear polymer, while amylopectin is a branch polymer. Hydrolysis of starch thus takes a longer time.

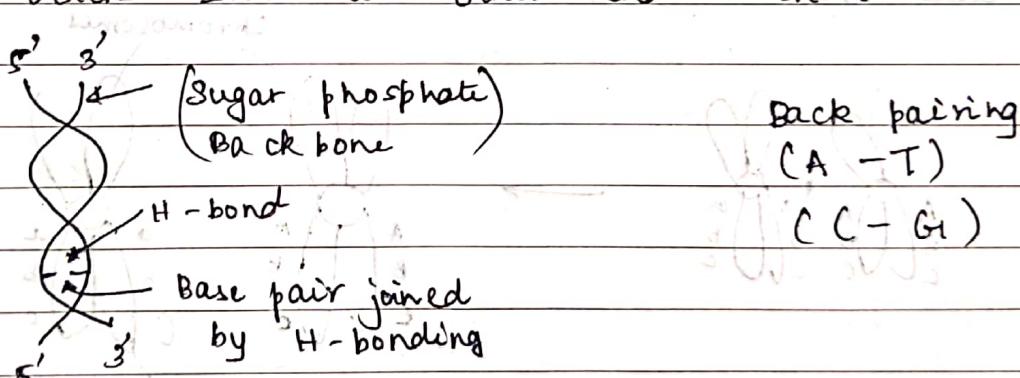
→ Cellulose is a crystalline highly branched polymer. The enzyme that breaks down cellulose is cellulase which is absent in humans (but present in cows).

- lignin is a complex mixture of organic compounds.
Hemicellulose is not so well defined and has some organic compds.
- Nucleotide Polymerization: Sugars, phosphates form the primary chain; for every DNA, the nucleoside would be different.

5' terminus } End points of nucleotide
3' terminus } polymer

In a DNA/RNA, we start with 5' end at 3'

- The double helix structure of DNA is not unique. DNA has two helices and one is complementary of the other. One starts with 5' ends at 3' and other starts at other end with 5' ends at 3'.



- Supercoiling: The DNA is under a lot of strain, as it is coiled and on increasing the strain, they supercoil i.e. coiling of the already coiled DNA. This strain can be induced by twisting or with linkage no.

$$Lk = Tw + Wr$$

Superhelical Density

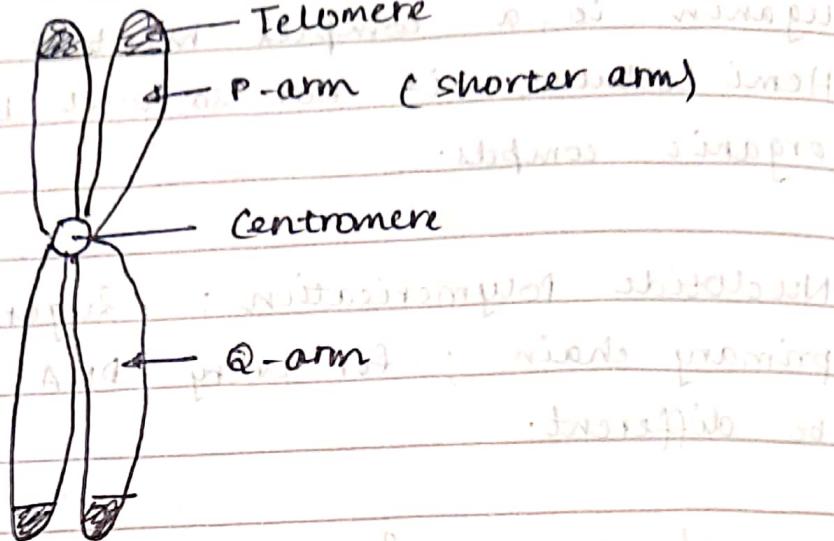
$Lk = 1$

$$\Rightarrow \frac{1 Lk}{Lk_0}$$

Superhelical?

$Lk = 6$

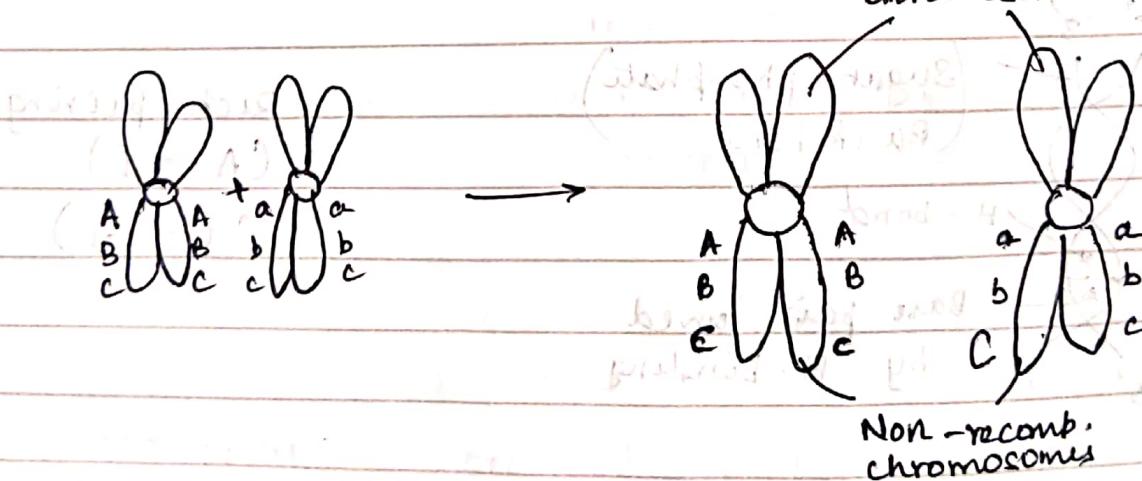
Linkage no. cutting at 6



structure of Chromosomes

Human cells are eukaryotic and in every cell there are the same no. of chromosomes (46 no., 23 pairs) except for sperms and eggs (23 no.) \rightarrow Fertile to give 23 pairs

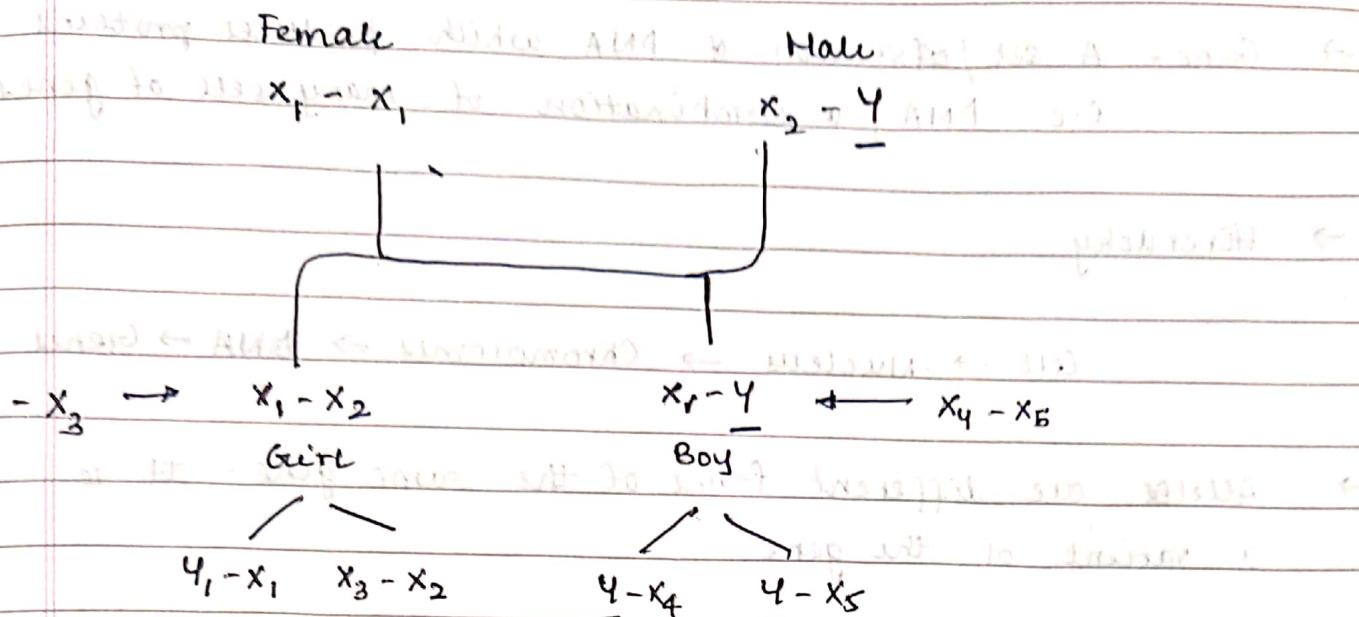
Cross Over \rightarrow leads to Recombinant chromosomes



Cross over leads to various different features.
Locus keeps on updating from generations

X - chromosome - 900 genes

Y - chromosome -

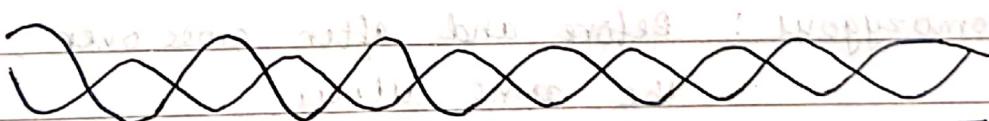


Great grandfather, Grandfather, Father, Son } Have same Y-chrom

The probability of a new born being a boy or a girl are equal. The ratio of boy to girl $\rightarrow 1:1$

DNA \rightarrow Exons (Synthesise protein)

\downarrow Intron (Useless part of DNA)
Synthesise protein \rightarrow Don't synthesise protein
 \rightarrow Junk "DNA"



Short Tandem Repeats

- ① Repeat sequences of length 2-9 base pairs/repeat
- ② variable Number of tandem repeats - repeat sequence length of 10-100 base pairs/repeat
- ③ They are for the stability of the DNA
- ④ They aren't doing anything "today", later might result in a completely diff organism

locus - Address/location of a part in a chromosome

Date _____
Page _____

→ Gene - A set/section of DNA which produces protein
i.e. DNA is a combination of many sets of genes

→ Hierarchy

Cell → Nucleus → Chromosomes → DNA → Gene

→ Alleles are different forms of the same gene. It is a variant of the gene

Consider a gene ACGT and SRT CA. Different alleles can be formed by SRT

Allele 1 : ACGT CA ACGT

Allele 2 : ACGT CA CA ACGT

Allele 3 : ACGT CA CA CA ACGT



Same gene with different allele

resulting in diff phenotypical features

→ Homozygous : Before and after cross over, they have the same alleles

Heterozygous : Before and after cross over, they have different alleles

→ Half DNA comes from each parent, however a full gene comes from both parents, but only one of them is dominant

→ Punnet square

B → dominant

b → inactive

| | | | |
|---|----|----|----------------------------------|
| | B | b | = neither dominant nor recessive |
| B | BB | Bb | P (Blue eyes) = 1/4 |
| b | Bb | bb | P (Brown eyes) = 3/4 |

The four blood groups are different in allele

A - antigen - A allele (dominant)

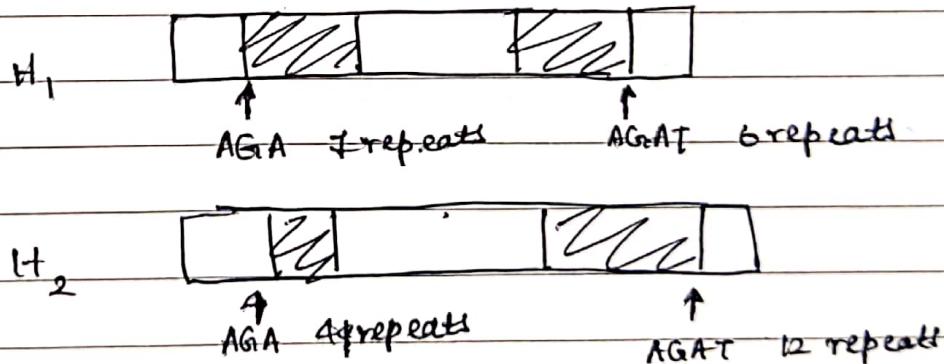
B - antigen - B allele (recessive)

A, B - antigen - A, B alleles

No antigen - O

→ DNA profiling

All have same genes, But different DNA's



Applications - Crime Scene Forensics

→ Bacteria and viruses replicate faster and even one mistake leads to a new branch of the phylogenetic tree

→ Protein synthesis

Transcription - Process of synthesis of a m-RNA
from DNA

translation - Process of polymerisation

Double stranded RNA ~~is~~ converts to 2 single stranded

Codone is collection of 3 base pairs, they decide which base pair is a suitable one

G, C - negative - G A

T, A - positive - T A

positive and E

Anti-codon just being used start the



Anticodon from the first AAs



Second amino acid - anticodon

Two new has been added, main has stopped
growing out of every two or three seconds

→ Metabolome -

→ The moment bacteria enters our body, they start producing chemicals

→ Viruses enter the cell as they are so small, they use their RNA to do reverse transcription and make DNA, and that DNA becomes an integral part of our cell. Thus, viral infections are difficult to address

⇒ Polymerase Chain Reaction (PCR) - Polymerase is an enzyme

→ Cloning - When the first cell is formed , they replicate to perform various functions , if we make multiple copies of the first cell , we have same multi genomic information in all aspects . This is called artificial cloning

Natural cloning - Identical twins

 [Monozygotic Twins - Share same placenta

 [Dizygotic Twins - Different placenta

