EVERWIN MATRIC. HR. SEC. SCHOOL

STD-XII (A-E) BIO-ZOOLOGY STUDY MATERIAL

I. Genetic Material:

1. Oswald Avery, Colin Macleod and Maclyn Mccarty repeated Griffith expt in an in vitro system.

* R-strain –Rough, non virulent.
* S-strain –smooth, virulent.
* This experiment proved that the biochemical substance which can be the genetic material is DNA.
* The doubt was on protein, or RNA or DNA which had the possibility to be a genetic material.
* Steps involved were:

a. Protein part was destroyed and the denatured. S-strain (heat killed) was added to R-strain, virulent S-strain appeared.

b. RNA was destroyed and S-strain (heat killed) was added to rough strain again. S- Strain appeared.

c. DNA was destroyed and S-strain (heat killed) was added to Rough strain no S-Strain appeared, no transformation occurred. So, DNA is the genetic material.

2. Hershey and Chase experiment on T_2 bacteriophage (virus). Scientist and Alfred Hershey and Martha Chase also called as blender experiment.

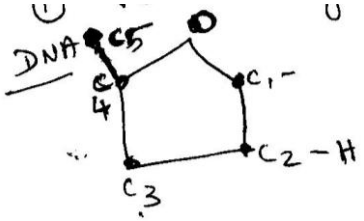
* There was a doubt that proteins can be the genetic material rather than DNA. This experiment is done to prove that DNA is the genetic material.
* Nucleic acid of DNA contains phosphorus.
* Proteins are made of amino acids and some amino acids like (methionine and cysteine contain sulphur).
* To prove it as DNA they take Phosphorus P^{32}.
* To prove it as protein they take Sulphur isotope S^{35}.
* Two different culture medium is taken one medium has bacteria growing in P^{32} and Bacteriophage (virus) infecting it. Another medium has bacteria growing in S^{35} and Bacteriophage (virus) infecting it.
* Bacteriophage (virus) which grew in S^{35} has labelled Sulphur and Bacteriophage (virus) which grew in P^{32} has labelled Phosphorus.
* The two labelled $S^{35} \propto P^{32}$ phages are allowed to infect unlabelled bacteria E.coli.
* The phages infect the bacteria and they are given a slight agitation before lysis with a blender.
* They found that P^{32} has entered the Bacterial cell, and S^{35} remained in the outside medium proving DNA is the genetic material.

Chemistry of Nucleic acids

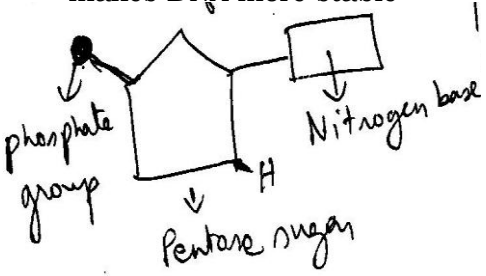
DNA and RNA

DNA is made (i) Pentose sugar (ii) Nitrogen base (iii) Phosphate group

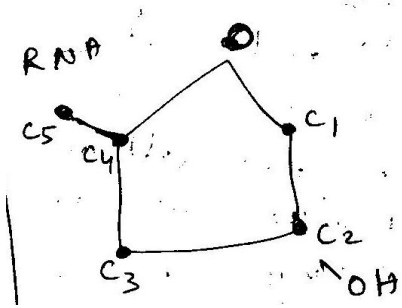
1. Pentose sugar



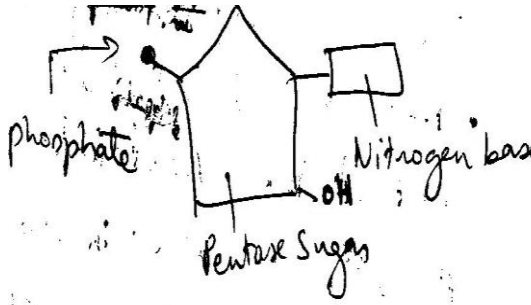
C2-only 'H' in DNA no 'OH' group makes DNA more stable



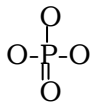
RNA



2. Basic structure of RNA



DNA and RNA Phosphate group



1. Phosphate group gives negative charge and acidic nature which retains it within cell or nuclear membrane
2. Oxygen molecules give negative charge
3. Release of H^+ ions

*from phosphate group gives acidic nature to nucleic acid

3. Nitrogen Base DNA	Nitrogen Base RNA:
Accepts H^+ ions gives basic nature	Accepts H^+ ions gives basic nature
* There are two types of nitrogen bases Purines and Pyrimidines	* There are two types of nitrogen bases purines and Pyrimidines
* Purines are Adenine, Gnanine	*Purines are Adenine and Curanine
* Pyrimidines are cytosine, Thymine	Pyrimidines: Pyrimidines are cytosine and uracil.
* Thymine gives more stability to DNA	Uracil makes RNA more unstable and reactive.
Purines two nitrogen rings	Purine two rings Pyrimidine are
* Pyrimidine one nitrogen ring	nitrogen ring

4. <u>DNA – Double standard</u>:	
Hydrogen bonds are found between Purines and Pyrimidines	RNA single standard only tarna seems to form Hydrogen bonds by pairing of complementary base pairs forming bonds (Hydrogen) only in certain regions.
A= T → two hydrogen bonds	
G≡C → three hydrogen bonds	
DNA is double stranded and it gives more stability	

<u>Packaging of DNA helix:</u>
haploid human
genonee (23) chromosomes
has 3.2 billion base pairs length of DNA double helix is 2.2m

<u>Packaging:</u>
Prokaryotes lacks (no) chromatin
Eukaryotes it is complex, chromatin is formed of repeating units called nucleosomes
Nucleosomes → proposed by Korn berg
* Four histones - proteins are present
* Each type of histone has two molecules
* The four histones are H2A, H2B, H3, H4
* Each histone with two molecules make a <u>octomere</u> (eight molecules)
  H2A-two, H3-two
  H2B-two, H4-two

*DNA is wrapped twice around the Nucleosome
*200bp are packed around a Nucleosome
*The Nucleosmes are connected by H1
*DNA is negatively charged and the histones are positively charged.
* Solenoids are formed of 6-Nuclesomes per turn.
*DNA is made of 40-folds of Solenoid
*Chromatin of interphase is 200-300nm (nanometer) in diameter

<u>DNA replication:</u>
*Semi conservation replication of DNA was postulated by Watson and Crick
*<u>Meselson</u> and <u>Stahl</u> proved this semi conservative replication of DNA postulated by Watson and circle was correct
* Two culture mediums with two different isotopes was used
* One set of Bacteria E-coli was grown in culture medium with heavy isotope ^{15}N and other light isotope ^{14}N (NH_4Cl)
* After many generations each formed DNA with purely Heavy ^{15}N and ^{14}N
* They were distinguished by cesium chloride density gradient centrifugation.
* Each type settled in two distinct separate bands

* Heavy culture was transferred to plain NH_4Cl and after the first replication made to undergo centrifugation and showed the band settling between heavy and light band, (intermediate position) proving it had one heavy and one light band.
* Again samples were taken after <u>second replication</u>, they found DNA settling at intermediate and light band position proving semi consecutive replication

Replication
Replication of DNA in prokaryotes
Enzymes:
DNA, I, II, III polymerase
DNA – I → Kornberg enzyme used for repair
DNA-II polymerase for repair
DNA-III main enzyme for DNA replication

Energy component for replication is → Deoxy nucleotide triphosphate initiation at ori-site
* DNA – unwinds
* Two strands are formed
* One leading strand → 3' → 5' polarity
* One lagging strand → 5' → 3' polarity
* Leading strand replication is continuous
* Lagging strand replication is discontinuous
* Leading strand is 3" to 5" because DNA polymerase can add bases from 5' and 3' end
* lagging strand replication is discontinuous causing formation okazali fragments.

Replication in Eukaryotes:
Enzymes are
* Helicase → unwinds the DNA strand
* Topoisomerase → keep the strand open till replication is completed
*Replication fork is formed
* DNA polymerase adds complementary nucleotides
* RNA primase – adds primer to help DNA polymerase to start replication
Exonuclease - removes RNA primers
* Gaps are filled by DNA polymerase
* Ligase seals, the gaps

Transcription – Synthesis of RNA
* Always only template strand with 3' to 5' used, it must has 3' to 5' polarity
* The other strand coding strand is not transcribed
* Monocistromic structural gene is seen in Eukaryotes (one gene one protein)
* Polycistronic structural gene is seen in prokaryotes (cluster of many related genes are presented) having many related genes, related proteins forming 'operon'

Transcription unit of DNA has
* Promoter region
* Structural gene
* Terminator

Promoter region:
* Prokaryotes have pribnow box
* Eukaryotes have Gold berg Hongness box or TATA box
TATA → Thymine, adnine rich region
* Structural gene
Prokaryote – polycistronic (operon)

Eukaryote – monocistronic
In prokaryotes:
RNA polymerase enzyme helps in transcription has two parts one is core enzyme and other sigma subunit, core enzyme (β', β, $\propto$,$\propto$) helps RNA synthesis.

Sigma helps recognition of promoter.
* The gene ends in terminator termination is by rho(ρ)
*Iniitiation factor is sigma
* And the termination factor is rho
* RNA polymerase associates with these factors to start and stop transcription.

Eukaryotes:
* There are three RNA polymerases
* RNA -I → transcribes rRNA
* RNA -II transcribes – mRNA (hnRNA)
hnRNA has (exon, intron)
RNA III for T-RNA
Intron → mobile gene, non coding sequence
* Introns are intervening sequence
* Exons are expressed sequence
* Introns have to be spliced and capping and failing has to be done to change hn RNA to mRNA which can come out of Nucleus
*Capping by methyl gnanosine triphospahte
*Tailing by polyadenyl residues
Capping and tailing protects MRNA from enzymes of cytoplasm.