THE SEARCH FOR GENETIC MATERIAL

The Genetic Material is DNA

Although the discoveries of nucleon by Miescher and Mendel's principles of inheritance occurred nearly simultaneously, it took a considerable amount of time for the confirmation of DNA's role as the genetic material.

The following experiments serve as evidence to support this assertion:

Transforming Principle

The transformation experiments conducted by Frederick Griffith in 1928 provide crucial evidence for understanding the nature of genetic material. Griffith's experiments involved selecting two strains of the bacterium Streptococcus pneumoniae, also known as Pneumococcus: The S-III and R-II strains.

(i) The S-III strain, characterized by its smooth or capsulated appearance, possesses a mucous (polysaccharide) coat and forms smooth, shiny colonies when cultured. These strains are virulent and capable of causing pneumonia.

(ii) On the other hand, the R-II strain, distinguished by its rough or non-capsulated phenotype, lacks a mucous coat and forms rough colonies. These strains are non-virulent and do not cause pneumonia.

The experiment can be outlined in the following four stages:

(a) S strain bacteria are inoculated into mice, resulting in the death of the mice.

(b) R strain bacteria are injected into mice, leading to the survival of the mice.

(c) Heat-killed S strain bacteria are introduced into mice, and the mice survive.

(d) A mixture of heat-killed S strain bacteria and live R strain bacteria is administered to mice, causing the death of the mice.

Griffith successfully eliminated bacteria through the application of heat. Notably, he observed that injecting heat-killed S-strain bacteria into mice did not result in their demise. However, when he introduced a combination of heat-killed S-strain bacteria and live R-strain bacteria, the mice perished. Furthermore, he managed to isolate living S-strain bacteria from the deceased mice.

From these findings, Griffith concluded that the R-strain bacteria had undergone transformation facilitated by the heat-killed S-strain bacteria. This transformation likely occurred through the absorption of a transforming substance or principle by the rough-type bacteria from the heat-killed smooth bacteria. This absorption enabled the R-strain bacteria to synthesize a smooth polysaccharide coat and become virulent. Griffith inferred that this phenomenon was indicative of the transfer of genetic material. However, the biochemical composition of this genetic material remained undefined based on his experiments.

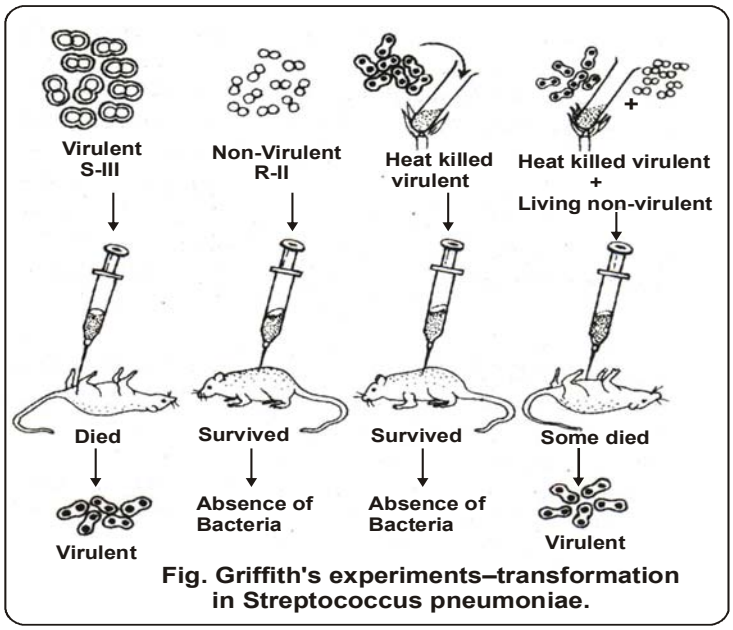

Fig. Griffith's experiments–transformation in Streptococcus pneumoniae.

Biochemical Characterization of The Transforming Principle

In 1944, Avery, MacLeod, and McCarty replicated Griffith's experiment in a controlled laboratory setting to pinpoint the nature of the transforming substance. Their findings conclusively identified this substance as DNA.

They meticulously isolated and purified biochemical components from the killed S-type bacteria, segregating them into three categories: DNA, carbohydrate, and protein. The DNA fraction was further subdivided into two segments: one treated with deoxyribonuclease (DNase) and the other left untreated. These four components were subsequently introduced into separate culture tubes containing R-type bacteria. Following a period of incubation, the contents were examined for bacterial growth.

Remarkably, only the DNA extracted from S-type bacteria was capable of converting the R-type bacteria into the S-type. This pivotal discovery led to the inference that the characteristic or gene of virulence resides within DNA. Consequently, Avery, MacLeod, and McCarty established that DNA is the chemical entity inherited by organisms, thereby forming the chemical or molecular foundation of heredity.

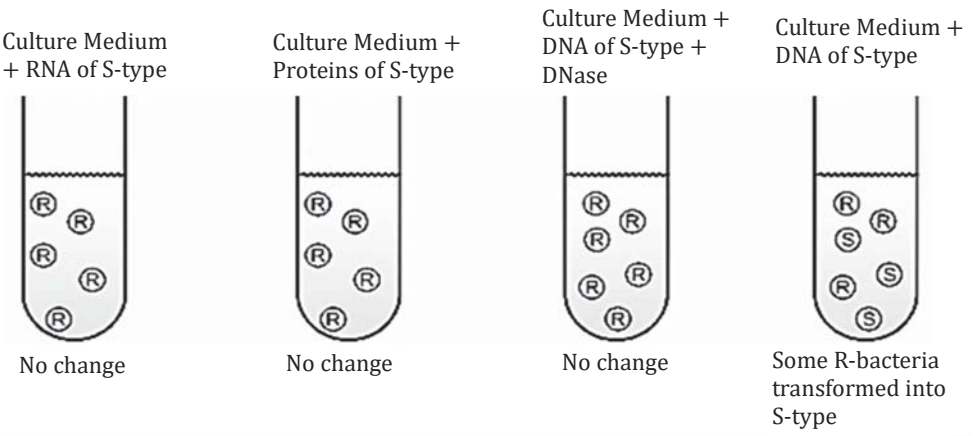

Figure: Avery et al.'s Experiment Demonstrating DNA as the Transforming Principle

Evidence from Experiments with Bacteriophage

The decisive evidence affirming DNA as the genetic material stemmed from the groundbreaking experiments conducted by Alfred Hershey and Martha Chase in 1952. Their investigations focused on the T_2 bacteriophage, a virus that infects the bacterium Escherichia coli and replicates within it. Comprising DNA and a protein coat, the T_2 phage presented an ideal subject for determining whether DNA or protein contains the information necessary for the generation of new virus particles.

To discern the functions of DNA and proteins, Hershey and Chase utilized radioactive tracers for labeling. DNA, known to contain phosphorus but not sulfur, was labeled with P^{32}. This labeling process involved growing bacteria infected with phages in a culture medium containing $^{32}PO_4$. Conversely, the protein coat of the phage, containing sulfur but not phosphorus, was labeled with S^{35}. Bacteria infected with phages were cultured in a separate medium containing $^{35}SO_4$ for this purpose. Subsequent to labeling, the experimental process involved three stages: infection, blending, and centrifugation.

(i) Infection: The labeled phages, both DNA and protein, were permitted to infect bacteria cultured under normal conditions in separate trials.

(ii) Blending: The bacterial cells were vigorously agitated in a blender to disrupt the interaction between the virus and the bacteria.

(iii) Centrifugation: The virus particles were isolated from the bacteria by subjecting the mixture to centrifugal force, which led to their separation through spinning in a centrifuge.

Following centrifugation, analysis revealed the presence of radioactive DNA labeled with P^{32} within the bacterial cells, while radioactive protein labeled with S^{35} was detected outside the bacterial cells, specifically in the surrounding medium. Moreover, labeled DNA was identified in the subsequent

generation of phages. Notably, bacteria infected with viruses containing radioactive proteins did not exhibit radioactivity, indicating that proteins did not enter the bacteria from the viruses. Consequently, these findings strongly support the conclusion that DNA serves as the genetic material transferred from the virus to the bacteria.

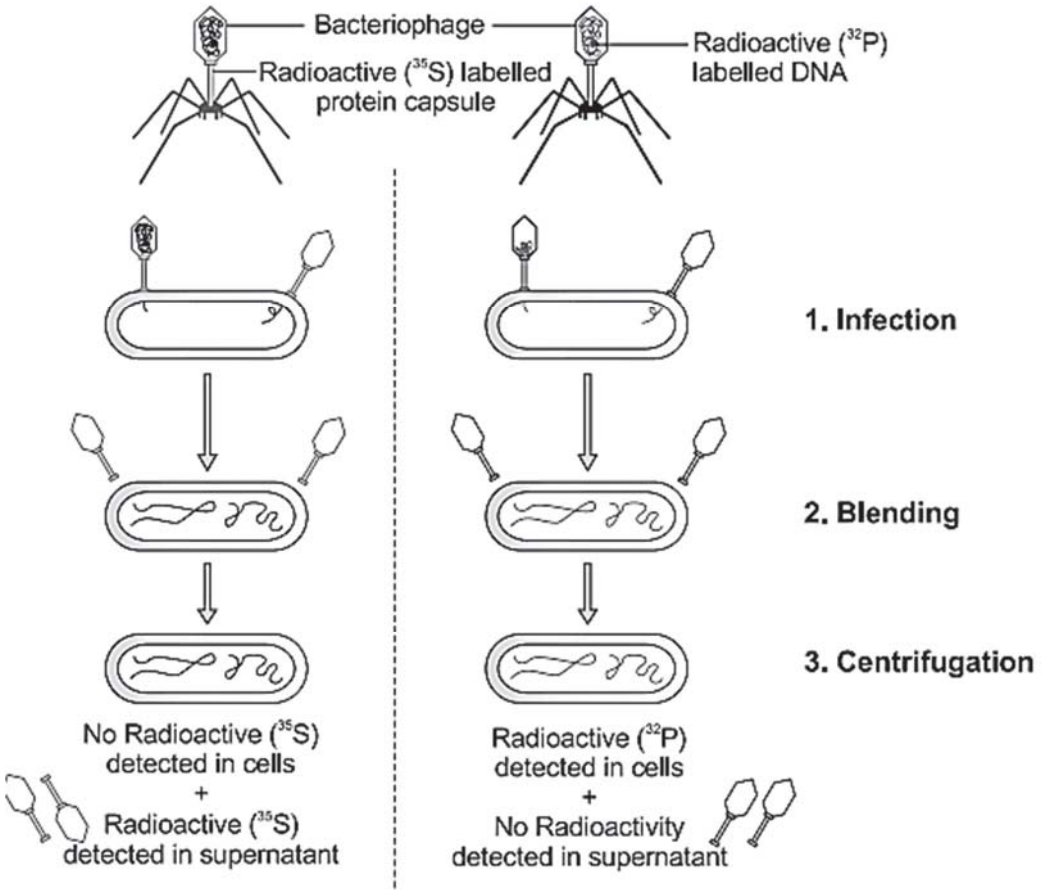

Fig. : The Hershey-Chase Experiment

Properties of Genetic Material (DNA versus RNA)

It has been firmly established that DNA serves as the primary genetic material. However, it has become evident that in certain viruses, such as Tobacco Mosaic viruses and QB bacteriophage, RNA acts as the genetic material.

For a molecule to function as genetic material, it must meet specific criteria:

(i) Replication: It should be capable of replicating itself.

(ii) Stability: It should possess chemical and structural stability.

(iii) Mutation: It should allow for gradual changes necessary for evolution.

(iv) Expression: It should be able to express itself as Mendelian characters.

Upon examination of each criterion, both DNA and RNA exhibit the ability to direct replication due to the rules of base pairing and complementarity. Other molecules in living systems, such as proteins, fail to fulfill this criterion.

Stability is another crucial property of genetic material. Griffith's "transforming principle" experiment demonstrated that heat, which killed the bacteria, did not destroy the properties of the genetic material. This stability can be attributed to DNA's complementary strands rejoining under appropriate conditions. Furthermore, RNA's 2'-OH group makes it chemically reactive and prone to degradation, rendering DNA more stable in comparison.

Both DNA and RNA are prone to mutation, with RNA mutating at a faster rate due to its instability. Consequently, viruses with RNA genomes have shorter lifespans and evolve more rapidly.

RNA has the ability to directly code for protein synthesis, facilitating easy expression of genetic traits. DNA, however, relies on RNA for protein synthesis, indicating that the protein synthesis machinery has evolved around RNA.

In summary, both RNA and DNA can function as genetic material, but DNA's greater stability makes it preferable for storing genetic information. However, for the transmission of genetic information, RNA is more efficient.