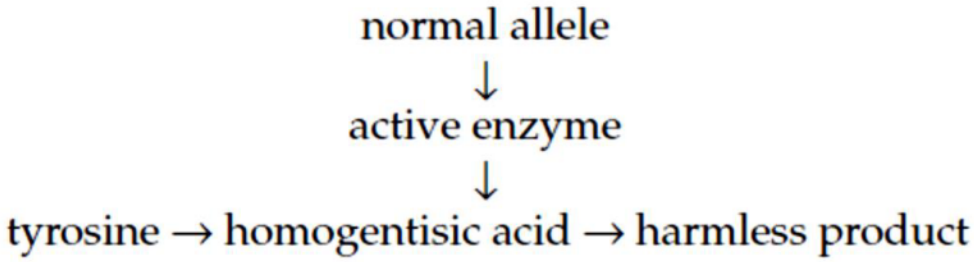
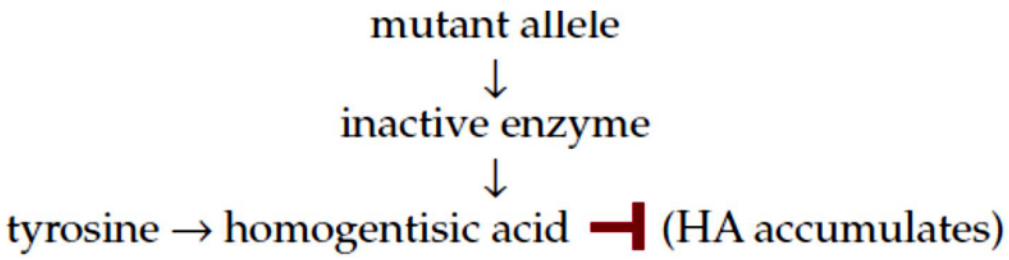
First observation for inheritance being related to a metabolic pathway

Archibald Garrod was an English physician. He observed that many children exhibited a symptom - the urine turns dark brown immediately after urination, i.e. when exposed to air. Further analysis indicated that the frequency of the disease was more common in children of consanguineous (within the family marriage e.g. Cousin-Cousin, uncle-niece) marriages. Simultaneously, Garrod found the concepts involved in rediscovery of Mendelian inheritance inspiring. A pedigree chart allowed Garrod to determine that the couples had a recessive allele causing the child to be homozygous recessive.

Biochemical composition of the urine was investigated by Garrod and observed the following pattern for normal individuals or heterozygous individuals.



So in homozygous recessive cases the reaction is blocked.



This was an indication that inheritance is responsible for turning the urine black. Garrod speculated this fact, but was unable to identify the enzyme or the gene. However an exact confirmation required several years of study. In 1958, the enzyme was identified –homogentisic acid oxidase and in 1996 the gene was identified.

Chromosomes contain both proteins and DNA: What is the evidence that which chemical component carries the genetic information?

Chromosome is a combination of two chemicals: DNA and Proteins:

The chromosome is a dynamic structure in the sense that it condenses and expands during various stages of the cell cycle. Chromosome is a mixture of two different components (i) DNA and (ii) proteins in higher quantity compared to DNA. In fact, DNA is bound to proteins. This unique combination accounts for dynamicity of the structure. What we see or represent for a

chromosome is the most condensed state of chromatin fibers (DNA fiber). This can be visually seen during the metaphase of the cell division (Fig 1).

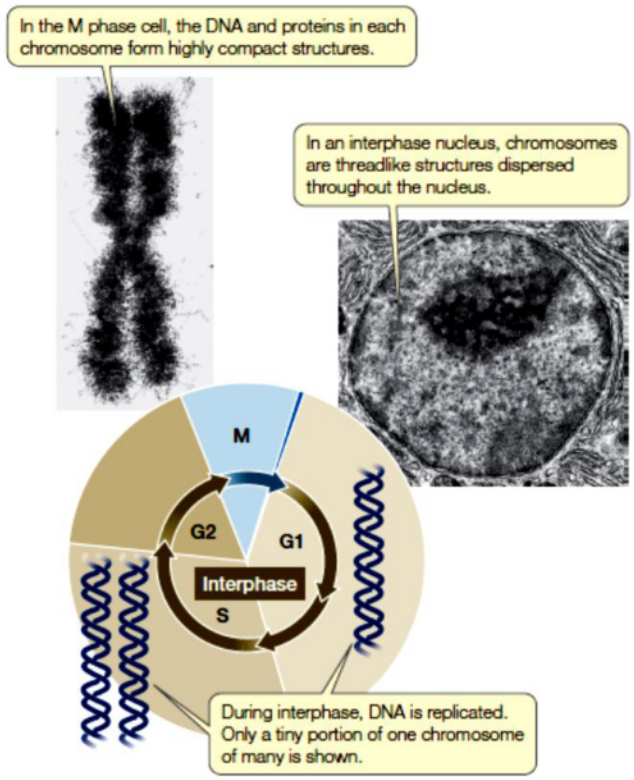


Figure 1: Dynamicity of the chromatin fibers – they expand and contract according to the stage of the cell cycle: M=Mitotic phase (Division phase), G1 and G2 are gap phases and synthesis of the raw materials occurs during the Synthesis (S phase). [Figure adapted from Sadava et al, Life: The science of Biology, 9th edition, Page 211, Fig 11.8]

The circumstantial evidences and logic to assume that DNA is the genetic material:

Living creatures exhibit a great diversity. Similar traits are observed in living forms of the same kind (species), while differences are observed between different species. It means that the genetic composition of living forms of one kind differs from that of the other kind. This exactly means their amount will differ. So can we prove it? This theory was proven by Robert Feulgen, who developed a red colored dye which binds to DNA. It stains DNA material red inside the nucleus. So the intensity of the red color is an approximate estimate of the DNA it contains. This dye is known as Feulgen stain. The Feulgen staining techniques presented the following information: It was in the right position (inside the nucleus) and the color intensity varied between two species.

The need of more cause and effect evidence!

The Feulgen staining only provided a circumstantial evidence. We should prove with a cause and effect situation that DNA carries the genetic information and not the proteins. How to do it?

Frederick Griffith was a physician from England. He was working with bacteria, which are visible only under a microscope. Pneumonia was taking many lives during his time. So he wanted to develop a vaccine for pneumonia. He found that there exists two forms (strains) of the bacteria which causes pneumonia, *Streptococcus pneumonia.* These strains are: Smooth (S) and Rough (R) forms.

Smooth forms care capable of causing the disease, while rough forms do not cause the disease. The reason is that the smooth forms are hidden inside a proteinaceous cover, so it can cheat the firewall (The immune system), while rough forms are not able to utilize that trick, as they lack the protein coat. Hence the firewall will definitely catch and eliminate them. Now he planned and executed the experiments as illustrated in figure 2.

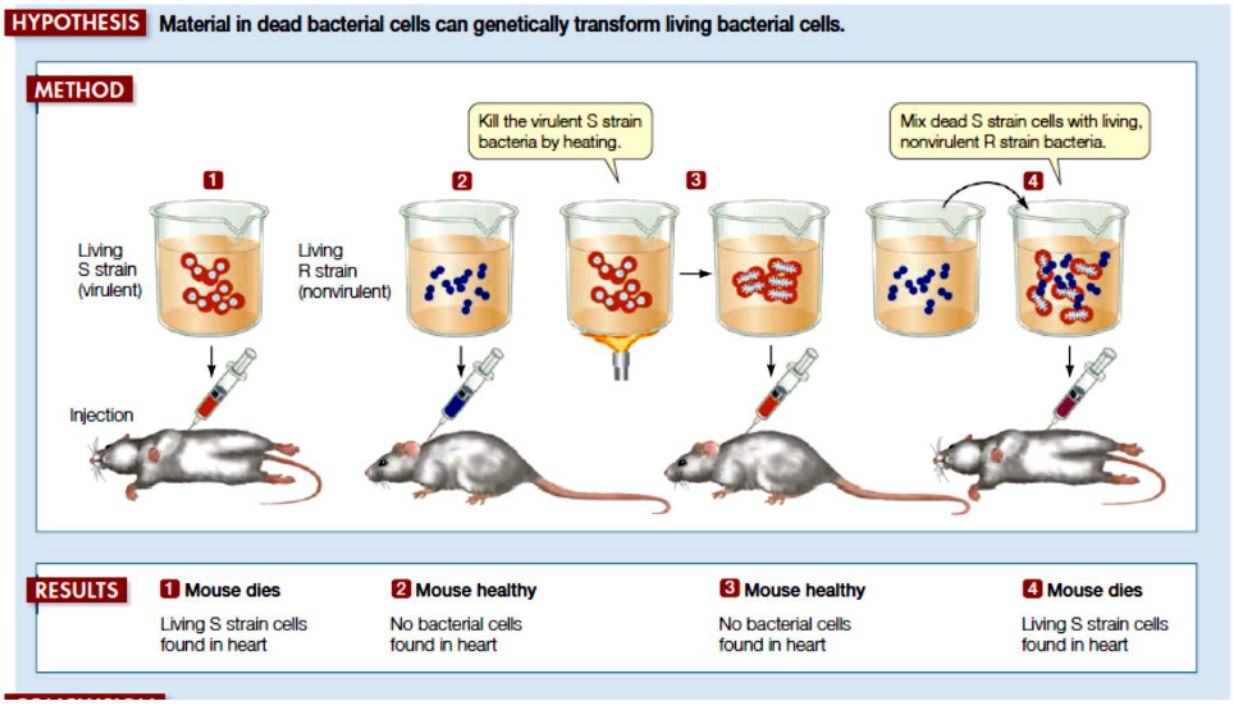


Figure 2: The experiments conducted by Griffith with the bacteria and mouse. [Figure adapted from Sadava et al, Life: The science of Biology, 9th edition, Page 268, Fig 13.2].

The above figure illustrates that in the experiment 4the material present in the S form transforms the material present in the R form. So we can say that some transforming principle is responsible for this change from an R form to S form of bacteria. How to identify this transforming principle? The scientific group led by Oswald Avery of Rockefeller University cracked this problem. Their experiment is illustrated in Figure 3.

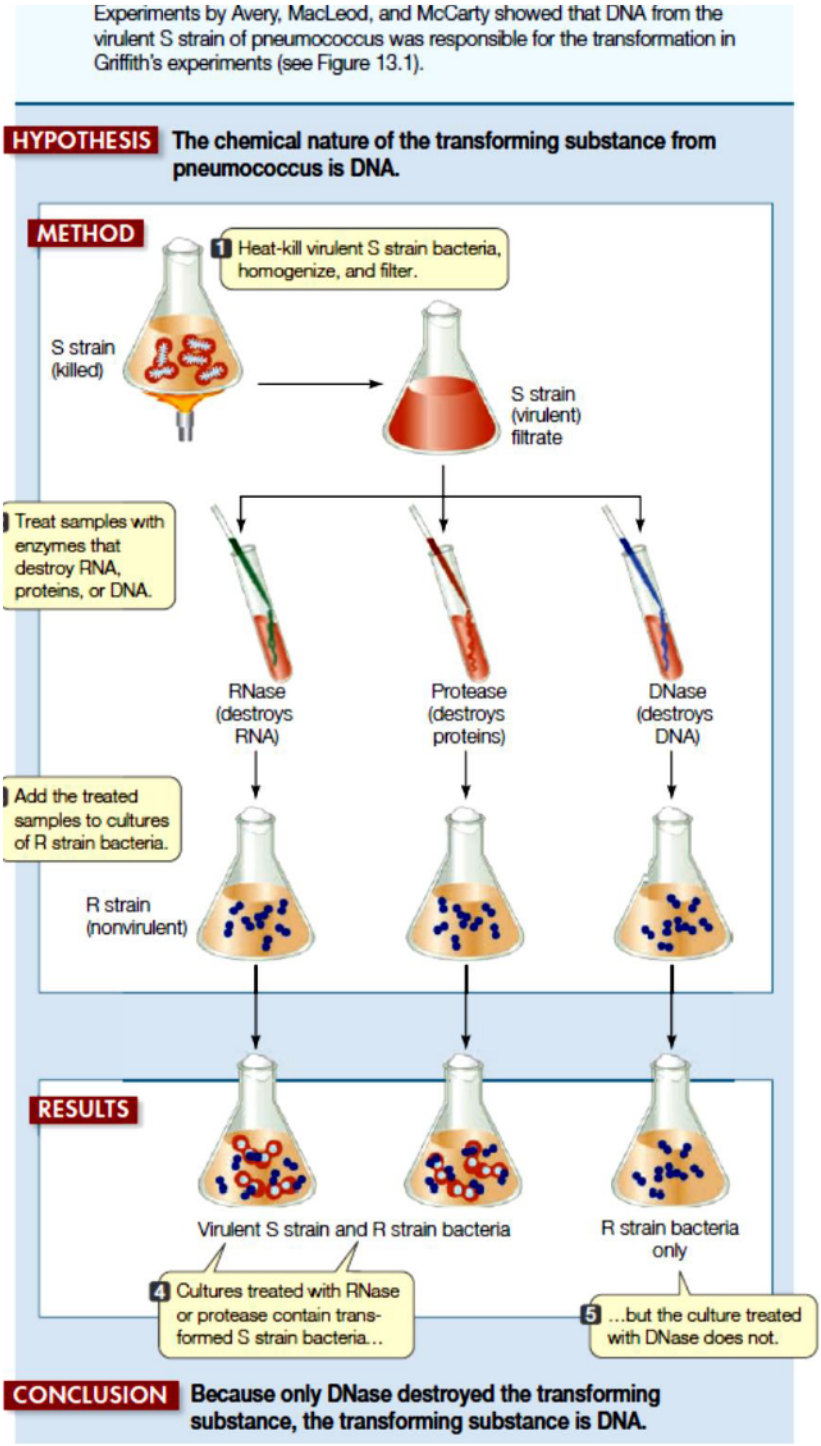


Figure 3: Identifying the "transforming principle". [Figure adapted from Sadava et al, Life: The science of Biology, 9th edition, Page 269, Fig 13.1]

The above work was published without much impact in 1944 because, many were not aware of the fact that DNA is complex enough to give the diverse output. Moreover many were still wondering whether microscopic small creatures, like bacteria, has genes in it.

The impact of this work was intensified after another experimental work published in 1952 by Alfred Hershey and Martha Chase at Carnegie Laboratory of Genetics. They were trying to determine whether DNA or protein contains the genetic material by using a bacteriophage (a virus that attacks and kills the bacteria). Why they have selected a virus? Because virus is composed of just two components that we are trying to sort, the protein cover and the DNA inside it (Figure 4).

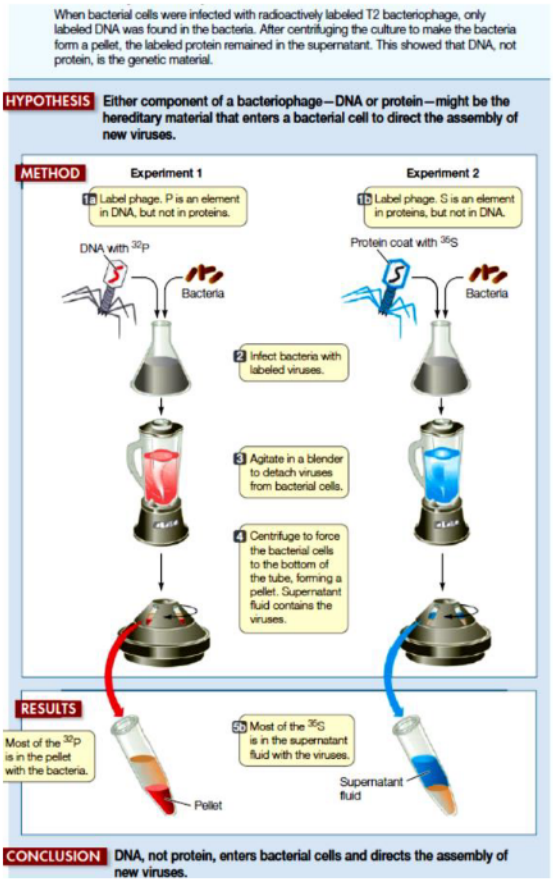


Figure 4: Hershey and Chase experiment. [Figure adapted from Sadava et al, Life: The science of Biology, 9th edition, Page 271, Fig 13.4]