DNA Is The Stuff Of Life

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The research of Gregor Mendel dramatically changed our perception of heredity. His conclusion that a trait was controlled by a particulate ***factor*** suggested that some physical entity existed that controlled heredity. Mendel's 1st Law, the law of segregation, suggested the factor was somehow reduced when it was passed onto what we know now is the gamete. We also know that this reduction event occurs during meiosis. Mendel's 2nd Law, the law of independent assortment, implied that each trait was controlled by a unique factor. As significant as the discoveries of Mendel were, they did not consider the actual physical entity that controls heredity. A separate set of conclusions, many based on simple empirical scientific observation, lead to the eventual determination that these factors reside on chromosomes, and that DNA was the heredity material.

Genes Reside on Chromosomes

From 1879-1892, Flemming, Strasburger, Waldeyer, van Beneden, and Weismann made significant contributions to our concepts of chromosomes. Flemming (1882) observed structures in the nucleus of salamanders that bound dye, and these structures had a string like appearance. He termed the structures ***chromatin*** (or colored substance). He also developed the concept of cell division that he later termed ***mitosis***. The universality of this discovery is attributed to Strasburger who discovered the same process in plants. Waldeyer, in 1888, called the structures that divided during mitosis ***chromosomes*** (or colored bodies).

Weismann made the very critical observation that sperm and egg cells contain exactly half the number of chromosomes. van Beneden further observed that when a sperm cell fertilizes an egg, the result is the diploid chromosome number found in cells that undergo mitosis. He also determined that the sperm and egg cell each contribute equal numbers of chromosomes to the zygote. This final result demonstrated a link between chromosome and inheritance because it was generally known that offspring receive traits from both of their parents.

These concepts were echoed by Hugo deVries (1889) but from a different perspective. He had called the hereditary material ***pangenes***. He felt the pangene was larger than a single chemical molecule, yet still invisible. All of the pangenes that controlled all of the organisms traits were found within the cell of a diploid organism. He further suggested that reproductive cells receive half of the pangenes and that when reproductive cells unite the diploid number of pangenes is restored.

In retrospect, these conclusions accurately represented our current understanding of how genes are passed from generation to generation. But although completely accurate, deVries theory was somewhat forgotten when E.B. Wilson released his book *The Cell in Development and Inheritance*. Notice the relationship between the cell and inheritance in the title; the linkage we are so now familiar with was growing stronger. His book also discussed the relationship between chromosomes and heredity. Although the relationship was suggested, the linkage was most strongly demonstrated by Sutton and Boveri.

Walter Sutton and Theodor Boveri worked independently. Boveri observed (as had van Beneden) that male and female gametes contribute an equal number of chromosomes to the zygote. In 1902, he made the link between chromosomes and the factors that were described by Mendel. This is most evident in a quote of his that states "… the characters dealt with in Mendelian experiments are truly connected to specific chromosomes." Sutton described chromosomes as unique individual units that occur in pairs that separate during meiosis. These concepts of Doveri and Sutton are known as the ***Chromosomal Theory of Inheritance***. These studies set the stage for the intense study of chromosomes and their role in heredity.

The final link between heredity and chromosomes required definitive proof that a genetic factor was indeed located on a chromosome. The work of Thomas Hunt Morgan, and later Calvin Bridges, provided such proof. Morgan, in 1910, discovered a mutant line of *Drosophila* with white rather than the wild type red eye. He suggested by genetic experiments that the gene for eye color was located on the X chromosome. This is one of the two sex chromosomes in the fly. Bridges followed this up by cytological experiments in 1914 that coupled the presence of the X chromosome with a specific eye color. This demonstrated conclusively that genes indeed reside on chromosomes.

If chromosomes were the carriers of genetic information, then they must contain all of the factors that make an organism unique. Yet, the total number of chromosomes is vastly less than the number of traits. For chromosomes to actually contain the genetic material, they must be shown to contain many factors. This was conclusively shown by researchers in Morgan's laboratory. By mating among *Drosophila*, it was shown that a chromosome contains multiple genes that could be organized into a linear ***linkage group***. The discovery that the number of linkage groups that could be mapped in a species was equal to the number of chromosomes solidified the importance of chromosomes in heredity.

It was clear that genes resided on chromosomes, and chromosomes were the functional unit of heredity. Alfred Sturtevant (1925) demonstrated that the actual physical order of the genes was also essential. He discovered the concept of ***position effect*** by showing that altering the physical environment of a gene on a chromosome altered its function. This implied that the actual physical organization of the chromosome was essential for proper phenotypic expression. Therefore, it became clear that we could not simply look at a single gene to understand its expression, but rather we needed to consider the entire chromosomal region. In many regards, this pointed the way to genomics, a science where we considered the entire DNA complement.

DNA is the genetic material

DNA is the stuff of life. Embedded in DNA are the sequences that encode for all of the genes that determine what an organism will look like and how that organism will react to its environment. But the discovery that DNA was the master information molecule was spread over nearly 80 years. Early (1866) observations of Ernst Haeckel suggested that nucleus transmitted hereditary information to the next generation. Because of his professional stature in biology, this concept focused the work of others toward the nucleus.

The next milestone in determining the nature of the hereditary information was performed by Friedrich Miescher. He studied pus cells that he collected from bandages he collected from surgeries. The majority of these cells were white blood cells. The cells are distinctive because they are primarily composed of nuclei. In 1871, he called this nuclear material ***nuclein***. Miescher further characterized this material chemically (1874). First, he determined that nuclein was rich in phosphorus. Further analysis revealed that it contained acidic (DNA and RNA) and basic (histone proteins) portions. Even though a link was made between a chemical molecule and the hereditary material, further discoveries were necessary to make a definitive link between DNA and heredity.

The experiments of Fred Griffith (1928) set the stage for the final proof that DNA was the genetic material. Working with lethal and non-lethal strains of the *Streptococcus pneumoniae*, he was able to show that a factor from a lethal strain could convert a non-lethal strain to a lethal strain. Because his factor was able to convert one phenotype to another (the true nature of a gene), Griffith called this factor the ***transforming principle***. Avery, MacLeod, and McCarty (1944) studied this factor in depth. Their experiments definitively proved that ***the factor was DNA***, and not protein or RNA, the other two constituents in the nucleus.

In retrospect, the Avery, MacLeod and McCarty experiments were definitive. Others did not think so, and the search continued. The experiments by Hershey and Chase (1952) demonstrated to many scientists that DNA was the hereditary material. Using T2 bacteriophage, they showed that DNA and not protein entered the bacterial cell. Since new T2 phage particles were produced inside the cell, it was concluded that DNA was responsible for its development. Although quite conclusive at the time, scientists now realize that the fact that the DNA was actually contaminated with a bit of protein make these less than conclusive experiments.

Although we focus so much of our attention on DNA, it should be noted that RNA can also be the genetic material. But this is only the case for RNA viruses. Heinz Fraenkel-Conrat (1957) and researchers working with him performed these experiments. Using reconstruction experiments, they were able to able to interconvert strains of tobacco mosaic virus by the addition of the RNA isolated from an alternate strain.

Now that it was known that DNA was indeed the genetic material, it only remained to determine its structure. That work was completed by James Watson and Francis Crick (1953). They determined that DNA is double-stranded, the strands are oriented in an anti-parallel manner to each other, purines nucleotides are opposite pyrimidines nucleotides in the strand, guanine hydrogen bonds with cytosine, while adenine hydrogen bonds with thymine, and the structure is stabilized by hydrogen bonds and hydrophobic bonding between stacked bases.

It should be noted that Watson and Crick did not perform any experiments. Their conclusions were based on research of others. Erwin Chargaff noted that the concentrations of guanine and cytosine were always equal in DNA. Likewise, the concentrations of adenine and thymine were equal. Rosalind Franklin and Maurice Wilkins used X-ray crystallography to study DNA. From these structures, Watson and Crick deduced that DNA had repeating structures (nucleotides), the DNA was of a constant width, and was double-stranded. All of this data went into their model of the DNA structure.

DNA Is The Stuff Of Life

Gregor Mendel

- dramatically changed our perception heredity
- particulate ***factor*** concept
- suggested that some physical entity existed that controlled heredity

Mendel’s 1st Law, the law of segregation

- suggested the factor was reduced during gamete production
- reduction event occurs during meiosis

Mendel’s 2nd Law, the law of independent assortment

- implied that each trait was controlled by a unique factor

Mendel

- did not consider the actual physical entity that controls experiments

Other experiments determined

- Mendel’s factors (genes) reside on chromosomes
- DNA was the heredity material.

Genes Reside on Chromosomes

1879-1892

Flemming, Strasburger, Waldeyer, van Beneden, and Weismann developed the concept of chromosomes

Flemming (1882)

- observed structures in the nucleus of salamanders that bound dye
- structures had a string like appearance to them
    - termed the structures ***chromatin*** (or colored substance)
- also developed the concept of cell division
    - he later termed ***mitosis***.

Strasburger

- the universality of this mitosis
- same process in plants

Waldeyer (1888)

- called the structures that divided during mitosis ***chromosomes***
- ***chromosomes*** (=colored bodies)

Weismann

- sperm and egg cells contain exactly half the number of chromosomes

van Beneden

- when an egg cell is fertilized by a sperm cell
    - the resulting cell has the diploid chromosome number
- also the sperm and egg cell
    - each contribute equal numbers of chromosomes to the zygote

This final result is important

- links chromosomes and inheritance

Why??

- it was generally known that offspring receive traits from their parents.

Hugo deVries (1889)

- called the hereditary material ***pangenes***

- considered pangenes
    - larger than a single chemical molecule
    - yet still invisible

- pangenes
    - controlled all the organisms traits
    - found within the cell of a diploid organism

- reproductive cells
    - receive half of the pangenes

- when reproductive cells unite
    - the diploid number of pangenes is restored

- ***First linkage between inheritance and reproductive cells***

E.B. Wilson

- *The Cell in Development and Inheritance*
    - suggested a relationship between the cell and inheritance
    - overshadowed the work of deVries

The Chromosomal Theory of Inheritance

Walter Sutton and Theodor Boveri

- worked independently

Boveri (1902)

- observed male and female gametes contribute an equal number of chromosomes to the zygote
- linked chromosomes and the factors that were described by Mendel

  - "… the characters dealt with in Mendelian experiments are truly connected to specific chromosomes."

Sutton (1902)

- described chromosomes as
  - unique individual units
  - that occur in pairs
  - separate during meiosis

Chromosomal Theory of Inheritance

Chromosomes are the carrier of Mendelian factors and meiosis is the basis of separating the factors into gametes.

Linking Genes and Chromosome

Thomas Hunt Morgan and Calvin Bridges

Morgan (1910)

- discovered a mutant white eye *Drosophila*
- different than the wild type red eye
- performed genetic experiments
    - these suggested the gene for eye color was located on the X chromosome

Bridges (1914)

- studied Morgan's white eye mutant
- coupled the presence of the X chromosome with a specific eye color
    - this demonstrated conclusively that ***genes indeed reside on chromosomes***

Problem!!

- chromosomes carried the genetic information
- they must contain all of the genetic factors
    - but, the number of chromosomes is less than the number of traits.
- **it's essential** to show chromosomes contain many factors

Solution!!

- Morgan's laboratory
    - mated among *Drosophila* with several different contrasting phenotypes
    - multiple genes are organized into a linear ***linkage group***
    - number of linkage groups equals the number of chromosomes

All of the features necessary for a hereditary unit are found in chromosomes

Position Effect

Sturtevant (1925)

- if the physical environment of a gene is altered
    - the expression of the gene is affected
    - implies physical structure essential for phenotypic expression

In many ways, this conclusion is the first solid reason that we should take a genomcs approach to study gene expression.

DNA is the genetic material

Ernst Haeckel (1866)

- nucleus transmitted hereditary information to the next generation
- his professional stature in biology helped focus the work of others toward the nucleus

Friedrich Miescher

1871

- sudied pus cells
- collected these from bandages at surgeries
  - majority of the cells were white blood cells
  - white blood cells unique
    - primarily composed of nuclei
- called this nuclear material ***nuclein***

1874

- Miescher further characterized this material chemically
- determined that nuclein was rich in phosphorus
- it contained two portions
  - acidic (DNA and RNA)
  - basic (histone proteins) portions

Now it was necessary to make a definitive link between DNA and heredity.

Fred Griffith (1928)

- worked with lethal and non-lethal strains of the *Streptococcus pneumoniae*
- converted a non-lethal strain to a lethal strain
- conversion involved mixing dead lethal and live non-lethal strains

Transforming principle

- converts one phenotype to another (the true nature of a gene)

Avery, MacLeod, and McCarty (1944)

- studied this transforming principle in depth
- ***the factor was DNA***
    - <u>**not**</u> protein or RNA, the other two constituents in the nucleus.

Hershey and Chase (1952)

- demonstrated to many that DNA was the hereditary material.
- T2 bacteriophage
- new particle produced inside a bacterial cell
- Heshey and Chase showed that DNA and not protein entered the bacterial cell
    - DNA was responsible for T2 development
    - it must be the genetic material

- **Problem**: DNA was actually contaminated with a bit of protein
- these are less than conclusive experiments

RNA Is A Genetic Material

Heinz Fraenkel-Conrat (1957)

- RNA viruses exist
  - he interconverted strains of tobacco mosaic virus
  - RNA mediated the interconversion

Watson and Crick (1953)

Structure of DNA

- DNA is double-stranded
- strands are oriented in an anti-parallel manner to each other
- purines nucleotides are opposite pyrimidines nucleotides
    - guanine hydrogen bonds with cytosine
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- structure is stabilized by
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Watson and Crick

- did not perform any experiments
- based on research of others
- Erwin Chargaff noted :
    - the concentrations of guanine and cytosine were always equal in DNA
    - the concentrations of adenine and thymine were equal

- Rosalind Franklin and Maurice Wilkins:
    - used X-ray crystallography to study DNA

- Watson and Crick deduced that DNA
    - had repeating structures (nucleotides)
    - the DNA was of a constant width
    - DNA was double-stranded