

Genetics Notes Chapter 5

Intro.

- Many modern molecular technologies are based on **bacterial genetics**.
- Bacteria are **prokaryotes**, they don't have a nuclear membrane enclosing their DNA.
- Have a single circular chromosome made up of a double strand of DNA
- Also have some extra circular DNA called **plasmids** (much smaller than the chromosome)
- Can be attacked by viruses called **bacteriophages**.
- Viruses have genetic material such as DNA or RNA but they are not living because they cannot reproduce alone.
- Bacteria mostly reproduces asexually, however sometimes there are occurrences of genetic exchange which can happen in four ways.
- In **conjugation** two bacterial cells fuse together. A donor cell donates part or all of its genome, or a plasmid to the receiver cell.
- In **transformation** a bacterial cells takes up a piece of DNA from the external environment and incorporates it into its genome.
- Some phages can pick up a piece of DNA in one bacterial cell and inject it into another, this is called **transduction**.
- Phages can undergo their own recombination when they infect the same bacterial cell.

5.1

- Bacteria are small, easy to culture in **medium** or on agar gel.
- The process of pipetting bacteria and spreading them onto agar gel is called **plating**.
- When the cells divide they don't travel far but they become a clump called a **colony**.
- Each colony is made up of clones of a single genetic ancestor.
- Mutants are easily obtainable and can be used in genetically marking certain alleles.

5.2

- Two scientists, Legerberg and Tatum demonstrated that genetic recombination between bacteria was possible.
- They took a strain A- of E. Coli that could only form colonies on a plate supplied with biotin and methanine. They took a strain B- that could only form colonies on a plate supplied with threonine, leucine, and thiamine.
- When each strain was plated separately on gel with no nutrients then no colonies were formed.
- However, when mixed together and then plated on a no-nutrient plate colonies did grow. These cells are assumed to be the wildtype, created from genetic recombination of the parent cells.
- Another scientist tried to test to see if the bacteria needed physical contact to share and transmit genes or if it only picked up genetic material once another cell got rid of it.
- He took a U-shaped tube and a fine filter small enough to block bacteria but large enough to allow gene fragments. With strain A- in one arm and B- in another arm he let it sit and then tested to see if any of the cells later on were wild type. None were.
- This proved physical contact between the bacteria is necessary for genetic exchange, a process called conjugation.

- The transfer of genetic material in E. Coli is not reciprocal. One cell, the **donor**, gives part or all of its genome to another cell, the **recipient**.
- The "donor ability" is designated by being F+. Donor ability can be passed between cells through genetic transfer.
- The F (**fertility factor**) is controlled by a plasmid. This plasmid controls for "**pili**" projections on the outside of the bacteria that contact a recipient and draw it in closer.
- The F plasmid in the donor cell is replicated through "**rolling circle replication**." This copy is then rolled over to the recipient cell which makes another copy from that to make two strands.
- Some of these cells with the F plasmid incorporated into their genome are called **Hfr cells (High Frequency Recombination)** they attempt to donate a copy of their whole genome to any F- cell it encounters.
- A cell bearing a donor allele that had undergone conjugation is an **exconjugant**.
- HFR DNA transfer starts from a singular point called the **origin**. The being said one can see how late the F gene was transferred to a recipient by seeing how far the F gene is from the origin. The F factor enters the gene last from a linear fashion.
- One end of the integrated F factor is the origin where the transfer begins. The other end is the terminus at the end of F.
- The gene is homologous to most parts of the chromosome and so can be inserted just about anywhere.
- Fertility Factor exists in two states:
 - The **plasmid state**: as a free cytoplasmic element, easily transferred.
 - The **integrated state**: as a continuous part of a circular chromosome, F is only transmitted very late in conjugation.
- Bacterial chromosomes are mapped in this case by **minutes**. For example if it took 10 minutes for the F gene to transfer after some gene a+ then the gene a+ and the gene F would be 10 units apart.
- The full genome of the recipient cell during conjugation is called the "**endogenote**"
- The partial genome derived from the Hfr donor is the **exogenote**.
- At this stage the cell has one copy of the endogenote and one copy of the exogenote making it a **partial diploid** called a **merozygote**.
- Recombination due to conjugation results from a double-crossover-like event, this gives rise to reciprocal recombinants, only one of which survives.
- Although the F fragment is generally stable once inserted in the chromosome, sometimes it is liberated from the chromosome cleanly and reformed into a plasmid.
- Sometimes the break is not so clean and the plasmid takes part of the main chromosome's DNA with it before reforming into a plasmid. This is called an **F prime (F') plasmid**.
- Vectors carrying multiple resistances to antibiotics and medicines that are easily transferrable are called **R plasmids**.
- The resistances are most often located on the R plasmids' are **transposons** which are segments of DNA that can easily move amongst locations in the genome and insert itself.

5.3

- Some bacteria can uptake genetic material from its external surroundings.
- This genetic material can come from dead cells or secreted from live bacterial cells.

- If the DNA is of a different genotype of the recipient the genotype can become changed permanently, this process is called **transformation**.
- Transformation is different from conjugation, in conjugation cells receive DNA from another living cell through close contact. In transformation a cell receives DNA from the external environment taken in through the cell wall and plasma membrane.
- It has been found that the genome of eukaryotic cells can also be changed through transformation.
- Transformation can be used to map genes on bacterial chromosomes.
- When DNA is being extracted for transformation experiments it is likely that some parts of the chromosome will get broken up before being taken up by the recipient cell.
- The closer two genes are together the more likely they will be taken up and integrated on the same genetic fragment to cause a double transformation.
- Conversely, the farther they are apart the less likely they will be taken up on the same genetic fragment to cause a double transformation.
- Another possible situation, is that the two genes are on separate gene fragments, but both gene fragments get taken up and integrated by the recipient cell independently.
- This situation is much less likely to occur, and is equal to the product of the probability of each gene fragment being transformed independently.
- If the percentage of both genes getting integrated through transformation is larger than what the product rule dictates then it means that the two genes are linked.
- However, this process is complicated because one must take into account factors such as the fact that not all bacterial cells are competent enough to participate in transformation.

5.4

- A **bacteriophage** is a bacterial virus that parasitizes and kills bacteria.
- Two distinct phage genotypes can be crossed and their recombination frequencies can be used to map the viral genome.
- Phages can be used to study the way of bringing bacterial genes together for **linkage**.
- A phage is made up of a **nucleic acid** "chromosome" of either DNA or RNA and an **outer protein covering**.
- They are represented by symbols.
- During infection a phage attaches to a bacterium and injects its genetic material into the bacterial cytoplasm.
- The phage's genetic material then turns off the machinery that produces bacterial components and then turns it back on redirecting it to synthesize phage components instead.
- Ultimately, many phage descendants are produced all with copies of the phage chromosome.
- They eventually break through the cell wall of the bacteria in a process called **lysis**.
- The population of the phage progeny is called the **lysate**.
- After lysis phages then go on to attack neighboring cells in a vicious cycle of infection.
- The population of the phages and the number of lysed bacterial cells increase exponentially.
- This can eventually be seen by the naked eye as a clear area on the surface of a bacterial lawn on a plate (an area where no bacteria grow because of attack and lysis from the phages) this clear area is called a **plaque**.