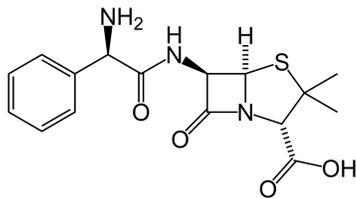# The Effects of Ampicillin on the Growth of Escherichia coli

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Abstract

Escherichia coli is a Gram-negative, rod-shaped bacterium that normally inhabits the intestines of humans and animals. Not all strains of E. *coli* are pathogenic, but ones that are contribute to one of the leading causes of foodborne illnesses. (2) Given this information, it would helpful to know if an antibiotic such as Ampicillin has any effect on the growth of E. *coli.* Ampicillin is a beta-lactam antibiotic that attacks Gram-positive and some Gram-negative bacteria. The amino group in Ampicillin allows it to penetrate the outer membrane of Gram- negative bacteria. It will then become an inhibitor of transpeptidase, which is needed for bacterial cell wall formation, and eventually leads to cell lysis. (1)

Figure 1: Ampicillin Structure (1)

Methods/Materials

Several preliminary experiments were used in order to determine if E. *coli* was sensitive to Ampicillin. Once this was successfully achieved varying amounts of liquid ampicillin were used to see if the IC50 could be determined. Throughout experiments, the amount of ampicillin added to samples ranged from 0-200 μL. Each sample mixture contained H20, low salt concentration LB and the inoculum, totaling 250 μL per sample. There were approximately 6-8 different samples used for each experiment. After making all sample mixtures, 200 μL of solution was loaded into wells to be measured in the Bioscreen C growth curve analyzer.

Results

Performing the experiment three times, a trend was discovered between the lag and log phase. Through graphing, as seen in Figure 2, the slope of the log phase was shown to be consistent throughout the varying amounts of ampicillin. However, the lag phase was most affected. As the amount of ampicillin was increased, the distance between the lag phases increased also in a correlating manner. At the higher concentrations, there appeared to be no growth because the lag phase was delayed so much. The graphical correlation of Ampicillin to the length of the lag phase can clearly be seen in Figure 2.
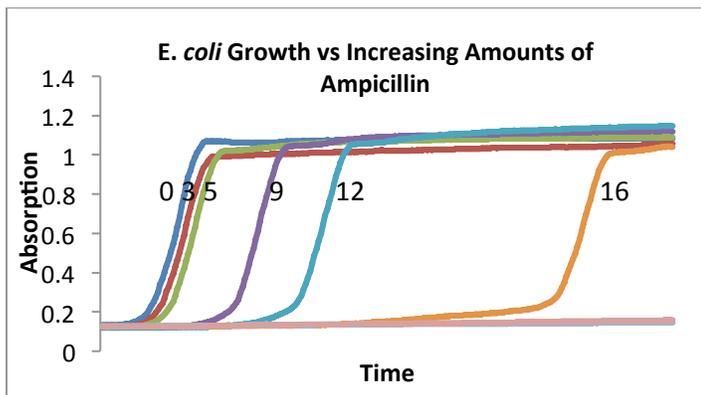
Figure 2: Measured absorptions as ampicillin is increased to display the effects on E. *coli* growth
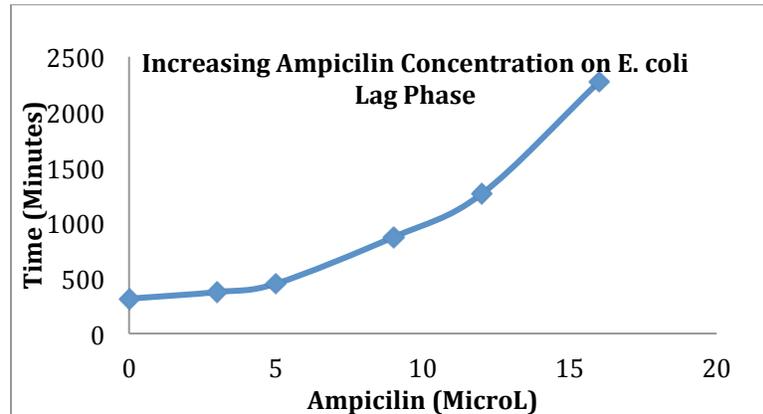
Figure 3: Displays increasing amounts of ampicillin (in 250microL mixtures) affects the length of the lag phase

Discussion

There were some difficulties with the experiment. When the experiments were first started we had focused on using serial dilutions of ampicillin to find the IC50. The titrations did not show a great enough effect on the E. *coli*. Later, we settled on using varying amounts of ampicillin to find a correlation between lag and ampicillin. From varying the amounts of ampicillin, we found the max and the slope of the log phase stayed the same. The lag phase was most affected for as the amount of ampicillin increased, the amount of distances between the lag phases increased also. After 16 μL, the lag phase cannot be measured as the time it takes for it to begin growing was outside the time frame of the experiment. The reason why the E. *coli* was most affected in the lag phase and nowhere else had to do with the cells trying to adapt to their environment. As their environment became more filled with ampicillin and less filled with water, the E. *coli* cells took more to adapt and mature. This caused the delay within the lag phases.

Conclusion

The lag phase of E. *coli* is most affected by the varying concentrations of ampicillin. In future experiments, we could test E. *coli* in different environments, like one similar to humans or farm animals, to see if the conclusion is the same. We also could see if there was a way to have the lag phase affected at lower concentrations by taking nutrients, like the Luria broth, away.

Acknowledgements

References

(1) Ampicillin/sulbactam. (2013, March 14). Retrieved December 3, 2013, from Wikipedia: en.wikipedia.org/Ampicillin/sulbactam

(2) Escherichia coli (E. coli). (2012, August 3). Retrieved December 3, 2013, from Centers for Disease Control and Prevention www.cdc.gov/ecoli/general/