







#### 4.1 Ineffectiveness of freshly extracted allicin against majority of tested bacteria species

The freshly extracted allicin did not live up to the postulations made at the beginning of this study. Although it undoubtedly has some antibacterial effect, it was not to the extent expected. There could be many possible reasons for this result. As stated above, allicin freshly extracted in a laboratory has varying quality. The quality depends on the extraction method; if mistakes were made during extraction, then naturally the quality will suffer. If the concentration of allicin compared to other compounds is low, then the purity of allicin will decrease exponentially. As mentioned previously, allicin has a short half-life; therefore, some may have dissipated before being introduced into the bacteria. Lastly, when infusing the sample discs, some of the allicin may have spilled out of the disc. This could possibly lead to false results regarding ZOI. To prevent some of these issues in the future, many improvements can be implemented. Instead of crushing the garlic with a mortar and pestle to make a garlic paste, it can be blended to ensure complete obliteration of the cellular structure of garlic. The purity of allicin can be checked using mass spectroscopy. By using this technique, the chances of using allicin of a weak concentration is reduced. A last step would be to buy pure allicin that was extracted through a patented and sophisticated extraction procedure. This step should be used as a last resort. The objective of this study was to examine the antibacterial properties of freshly crushed garlic. However, trials for future medical applications, this may be a better option to determine the importance of the potency of allicin.

#### 4.2 Mutagenesis in *P. aeruginosa*

As predicted, mutagenesis was most probably induced in a sample of *P. aeruginosa*. The disparity between the control and the mutated samples was significant enough to reach the conclusion that *P. aeruginosa* mutated to become resistant to allicin, although which specific genes caused this mutation is yet to be determined. Samples from this have been preserved, so that an in-depth examination of the genetic structure of *P. aeruginosa* can be done in the future. An interesting occurrence during this portion of the study was that the bacteria had a much higher sensitivity to allicin in this experiment than in the first trials at the beginning of the study. A possible reason for this is that even though allicin was extracted in a similar method, some variable changed when TLC was not used to create a more pure or potent form of allicin. Nevertheless, the results showed strong evidence that allicin had an inhibitory effect on *P. aeruginosa* and that the bacteria can be mutated to form resistance.

#### 4.3 Future directions

In the future, as antibiotic-resistance increases, allicin will become an important part in the treatment of bacterial infections and diseases. Although not truly effective in its freshly ex-

tracted form, pure allicin will perhaps be effective against many bacterial species. In fact, studies have been done in which allicin and an antibiotic have been combined to form a very potent antibacterial medication. [8] Studies such as this show that there is room for improvement and innovation regarding allicin in the field of biotechnology. In terms of this study, by examining the mutated strain of *P. aeruginosa*, the mode of action for the antibacterial effect of allicin may be elucidated. In the future, allicin can hopefully be used against both antibiotic-receptive and antibiotic-resistant bacterial illnesses. By creating a new pharmaceutical product made out of a natural source, the human microbiome will be a healthier and better functioning body.

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#### REFERENCES

- [1] Biljana, and Svetlana Cekovska. "Extracts from the history and medicinal properties of garlic." *Pharmacognosy reviews* 4.7 (2010): 106-110.
- [2] Peir, Sossain Kou, and Ali Gorji. "Garlic: a review of potential therapeutic effects." *Avicenna Journal of Phytomedicine* 4.1 (2014): 1-14.
- [3] "What Is Allicin?" *AllicinFacts*. Natural Health Publications Limited, n.d. Web. 13 July 2015.
- [4] "Future of Allicin." *AllicinFacts*. Natural Health Publications Limited, n.d. Web. 13 July 2015.
- [5] Ankri, Serge, and David Mirelman. "Antimicrobial properties of allicin from garlic." *Microbes and infection* 1.2 (1999): 125-129.
- [6] Williams, David Michael, and Chandra Mohan Pant. *PROCESS FOR THE PRODUCTION OF ALLICIN*. Neem Biotech Ltd., assignee. Patent 7,179,632. 20 Feb. 2007. Print.
- [7] Leng, Bing-Feng, et al. "Allicin Reduces the Production of Alpha-Toxin by *Staphylococcus aureus*." *Molecules* 16.9 (2011): 7958-7968.
- [8] Cai, Yun, et al. "Antibacterial activity of allicin alone and in combination with Beta-lactams against *Staphylococcus* spp. and *Pseudomonas aeruginosa*." *The Journal of antibiotics* 60.5 (2007): 335-338.
- [9] Mills, Ben. *Cysteine-to-allicin-2D-skeletal.png*. 2007. Photograph. Wikimedia Commons. Web. 17 Jul 2015. <<https://commons.wikimedia.org/wiki/File:Cysteine-to-allicin-2D-skeletal.png>>.