Survey of antibiotic resistance in soil samples from Yavapai County

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Introduction

Antibiotic resistance (AMR) is an important health topic. It represents one of the top threats to human, environmental, and animal health. It has been named as one of the leading causes of deaths worldwide, with a documented one million globally in 2019. To date, however, there are few studies that surveille the Arizona landscapes to determine the current AMR load in the wild. The primary focus of this research was to survey the local soils to collect data across Yavapai County, and in particular, the Prescott area. Factors that may impact the spread of AMR include seasonal rainfall (monsoons), natural flow of water, and proximity to human activity. Samples were taken from soils between Dewey-Humbolt and Williamson Valley.

Research Question: How does the proximity to human populations influence the amount of AMR in a soil sample?

Hypothesis: We hypothesized that the samples collected closer to areas with heavy human influence would show higher levels of resistance.

Methods/ Materials

Sample sites: Sites were selected for accessibility both for human and wildlife traffic. These sites included: A) Watson Lake, B) Stone Ridge golf resort, C) Granite Mountain, D) Dewey Humboldt

Sample collection: Collection was performed each week to obtain a wide range of samples. They were collected in a sterile container to avoid contamination from other sources. Once the samples were collected, they were brought to the lab for processing to minimize the amount of time the sample spent in the collection vessel.

Sample processing: Samples were processed by using distilled water at a 1:3 ratio soil to water. After an incubation of 10 minutes, 100 ul of the soil water was plated on TSA plates and spread with a cell spreader. Antibiotic disks were placed on the plates, and then they were incubated at 37C for 24 hours.

Antibiotic assay: Kirby-Bauer assay was performed to examine the number of antibiotics the soil microbes were resistant to as well as the degree to which they were resistant. These antibiotics included: Cefoxitin, Erythromycin, Cefazolin, Rifampin, Pipercillin, Clindamycin, Tetracycline, Bactracin.

Analysis: Measurements of the zone of inhibition were examined and recorded.

Results

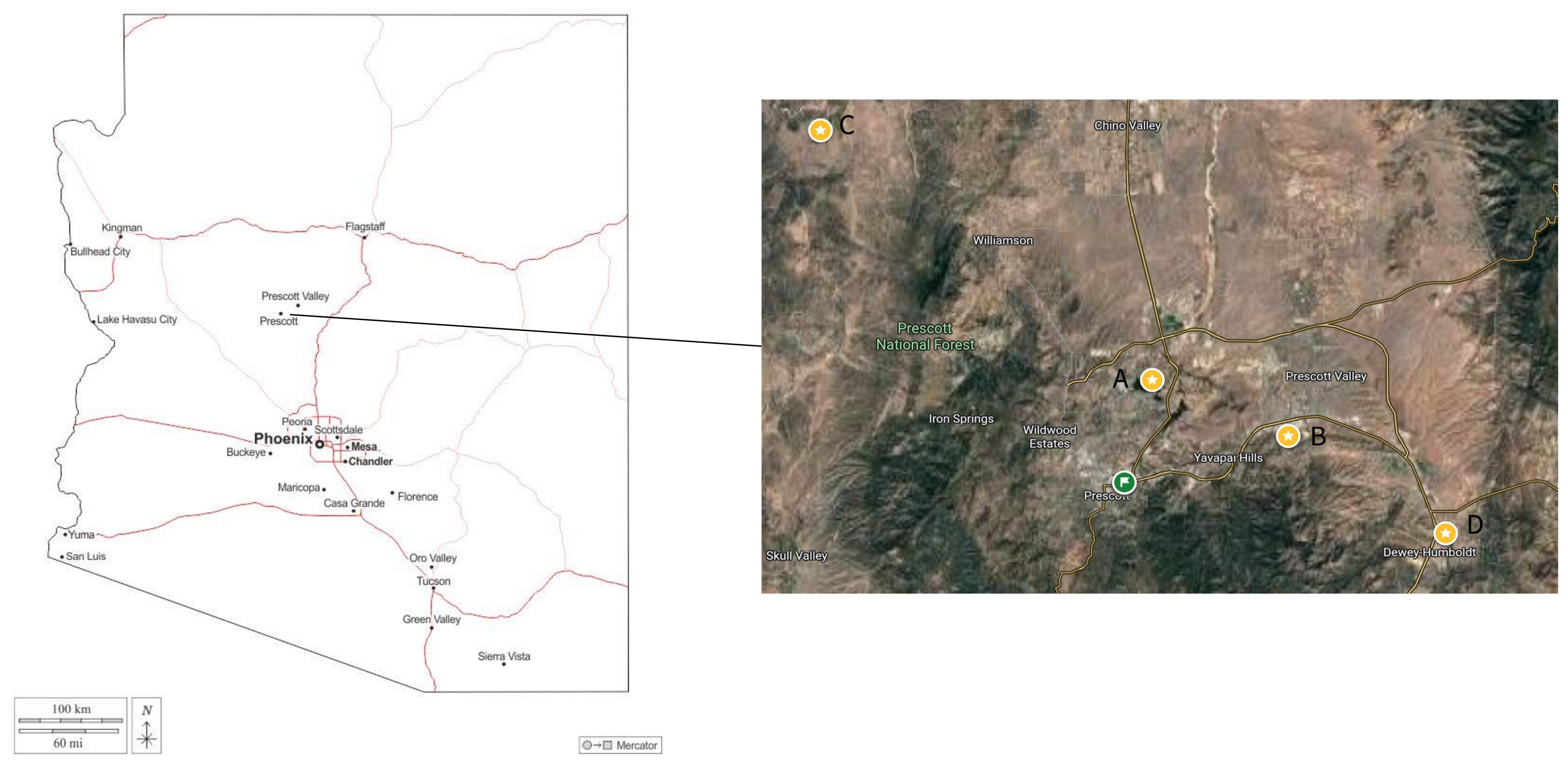
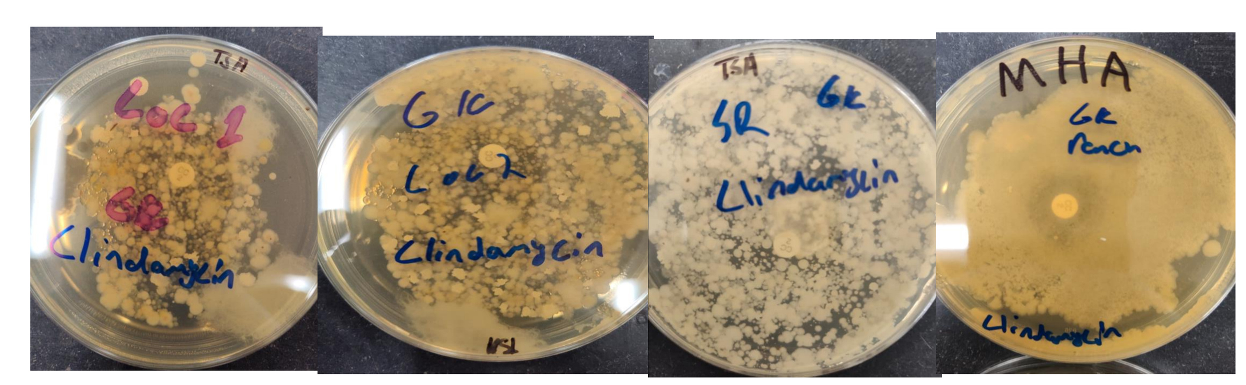
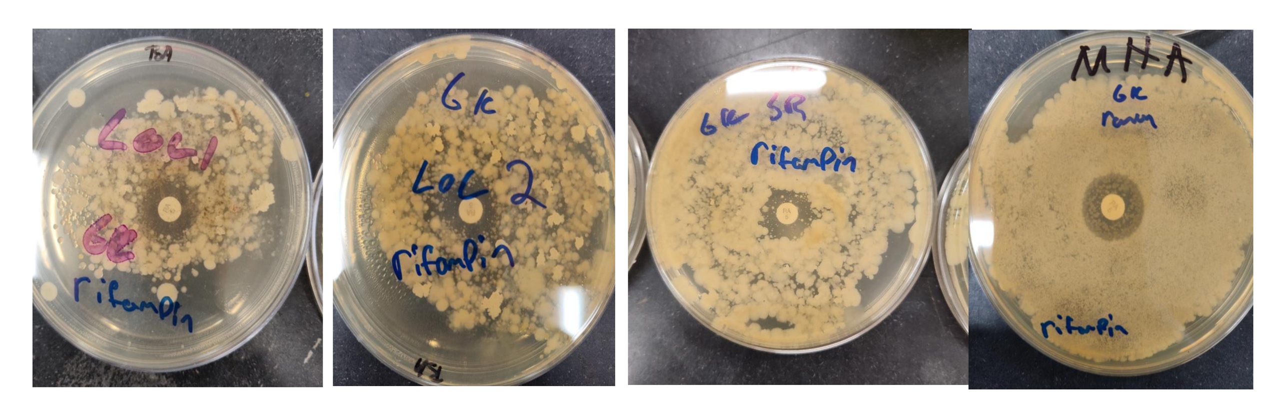


Fig. 1) Map of Arizona. Zoom in locations of soil sample collection sites. On the map, the stars represent each location collection site. A = Watson Lake; B = Stone Ridge Golf resort; C = Granite Mountain; D = Dewey-Humbolt. The flag represents Yavapai College.



A. Clindamycin



B. Rifampin

Fig. 2) Representative pictures of the Kirby-Bauer assay, illustrating results from two different antibiotics. The pictures posted above are only a representative sample of the antibiotics tested in this project. The first (A) is clindamycin, a broad-spectrum, very common antibiotic used in human health, agriculture, and in livestock. Clindamycin is among the most common antibiotics used in hospital settings and in over-the-counter antibiotic creams and ointments. In (B) the photos show that the antibiotic rifampin is much more effective against the bacteria in the local soil samples. The results pictured here in (B) are similar to the results from the other antibiotics tested. The average zone of inhibition for those that displayed resistance was about 7-10 mm in diameter.

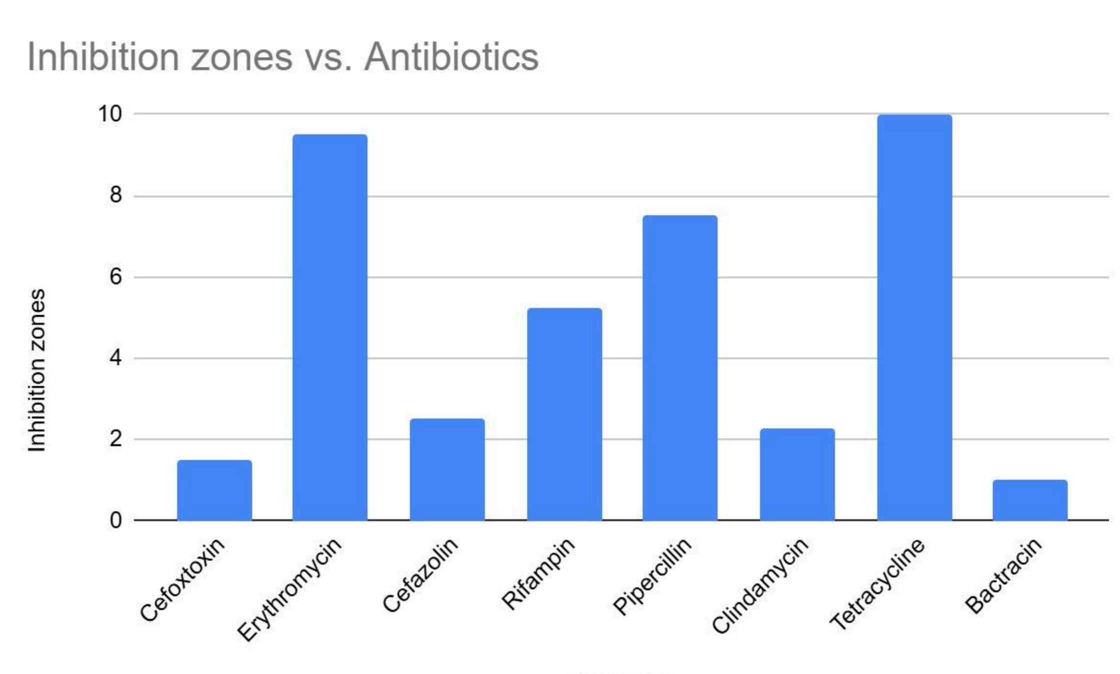


Fig.3) Graph of average zones of inhibitions for each antibiotic. Erythromycin and tetracycline show the greatest zones of inhibition, indicating few resistant bacteria. Clindamycin, cefoxitin, and bacitracin all having very small average zones of inhibition indicating more resistance to those antibiotics.

Conclusions

After completing this initial portion of the data collection database, it is apparent from the soil samples tested that the soil exhibits different microbial resistance. The differences in them could be narrowed down to whether it is farmland or a high-traffic public space. This plays a crucial role in how the soil develops and in how the bacteria within it build resistance. This is based on the clear zones of inhibition in each picture. Some types of soil with antibacterial properties have been added to the soil; this could have happened through wind and other elements transporting them passively.

Higher AMR was detected in areas of higher human influence, such as areas closer to where humans live, work or play.

Future Directions

- Continue to further the database for Yavapai College by expanding the data collection area to all of Northern Arizona
- Further analyze the types of bacteria that grow and become resistant to specific types of antibiotics
- Explore the various types of antibiotics
- DNA sequence the microbes to find what microbes are living in our soils as well as what genes are responsible for the AMR.

References

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