How is antibiotic resistance of *E. coli* affected by evolutionary history and nutrient limitation?

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**Abstract**

Antibiotic resistance is increasingly common in bacteria, making it crucial that we understand how it changes across different conditions. We conducted minimal inhibitory concentration (MIC) tests using ampicillin, erythromycin, tetracycline, and tobramycin in order to determine the impact of evolutionary history and carbon or nitrogen limitation on Long Term Evolution Experiment strains of *E. coli*, which are grown without antibiotics and evolved over thousands of generations to identify differences in evolution. We found that evolutionary history has a significant impact on antibiotic resistance while nutrient limitation does not. Additionally, one strain used in our experiment had different levels of antibiotic resistance compared to others in the population despite having a shared ancestor and the same environmental conditions.

**Methods**

Decreasing concentrations of each antibiotic were pipetted onto 96-well plates containing the same concentration of bacteria in each well. The MIC was determined as the well with the lowest antibiotic dilution without any visible growth.

**Results**

A: Median MIC values were plotted. The MIC values of strains descended from A-2 were significantly lower than those from all other ancestors (three-way ANOVA, *p*<.05). A-1-derived strains had significantly higher MIC than A-4 and A-5-derived strains. Nutrient limitation does not have a significant effect on MIC.

B: The mean MIC for each strain was plotted. Large vs small A-2 clones did not have significant differences (*p*=0.5) when compared using ANOVA tests. REL11335 (A-2 strain with higher MICs) was an outlier, causing it to be significantly different than other strains (*p*<.05).

**Conclusions**

- Nutrient limitation does not have a significant impact on antibiotic resistance.
- Evolutionary history does significantly impact antibiotic resistance.
- Clones evolved in the same conditions from the same lineage can have different levels of antibiotic resistance.