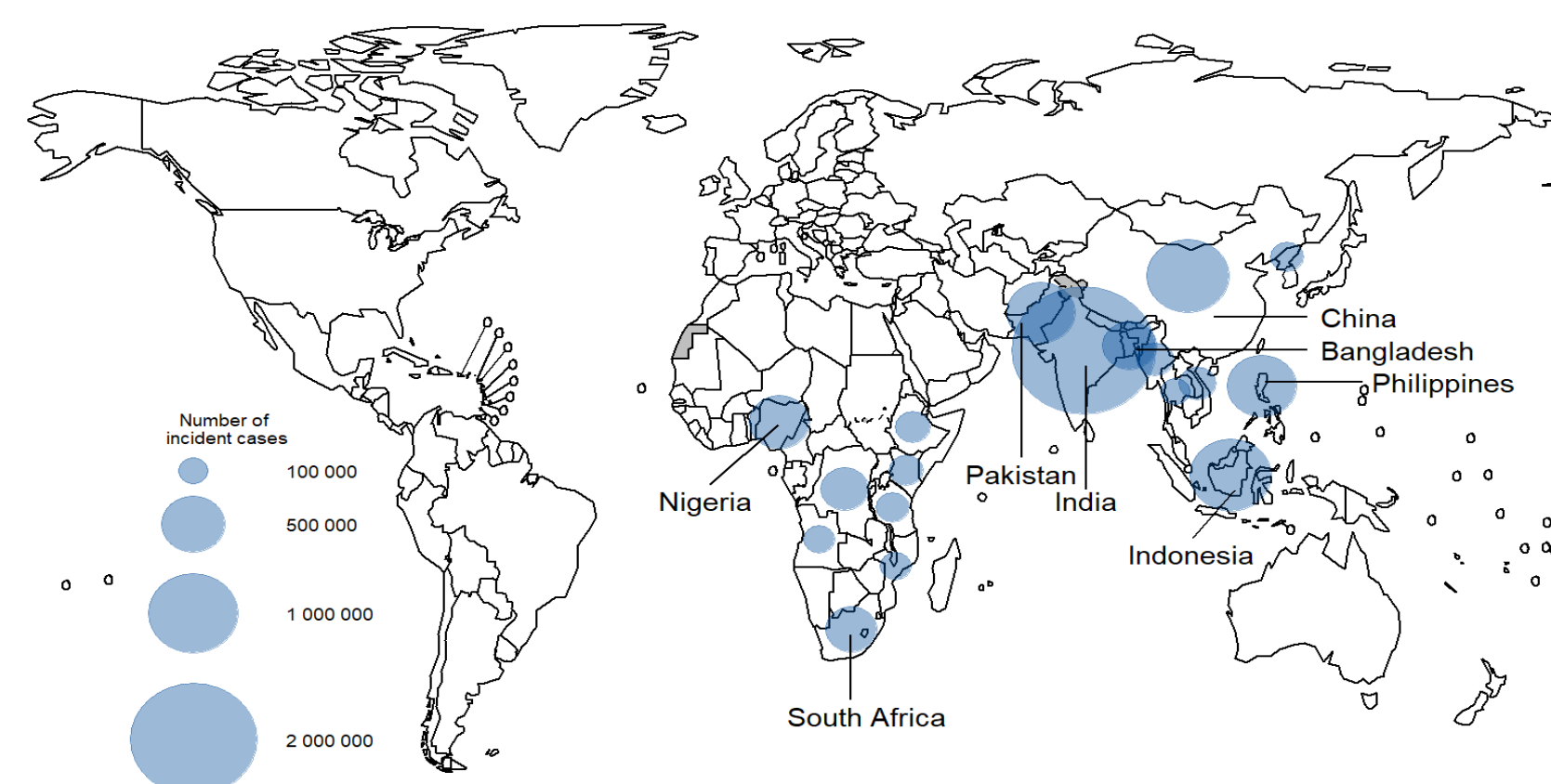


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Mentors: Professor Scarlet Shell, Julia Ryan

Spread of Tuberculosis

Tuberculosis (TB) remains a major threat to public health

- Caused by the bacterium, *Mycobacterium tuberculosis*, that targets the lungs.
- 2nd leading infectious killer after COVID-19.
- Affects people around the world **disproportionally**. Patients lacking access to 2nd line drugs are left with very limited treatment options for multidrug-resistant TB (MDR-TB).
- Only about **2 in 5 people** with drug resistant TB accessed treatment in 2022.



World Health Organization TB report 2022

Goal: Use *Mycobacterium smegmatis*, a nonpathogenic related species, to determine factors that allow the mycobacterium species to resist and tolerate many antibiotics during infection.

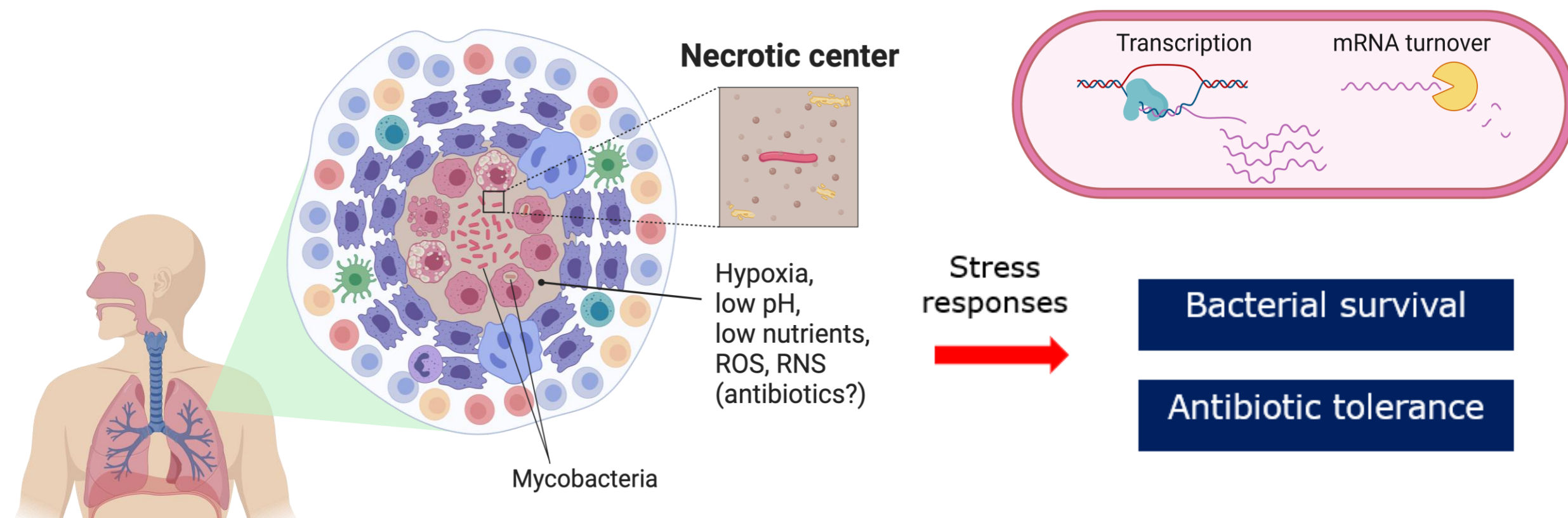


Ending the TB epidemic by 2030 is part of the health targets of the U.N. Sustainable Development Goals.

Researching drug-resistant TB will lead to new ways to ensure healthier lives and promote well-being for all ages.

Mycobacterial Survival

Mycobacteria theorized to adapt to stress by regulating gene expression.



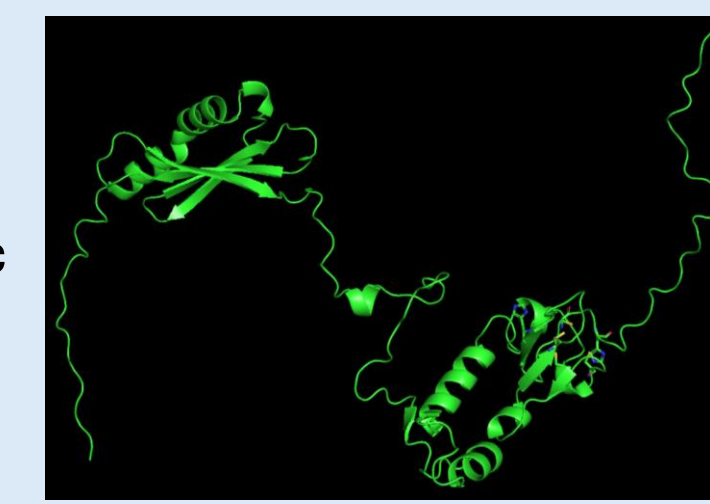
Mtb cells able to withstand a variety of environmental stressors during human infection.

Image credits: Opeyemi Isaac, Ibbtoye, Diego Vargas Blanco

New innovative methods are needed to understand infections as bacterial outbreaks increase with antimicrobial-resistant microbial strains.

MIC Growth Assays

- Researching **protein 5691**, conserved throughout mycobacteria species to phenotypically characterize *M. smegmatis*.



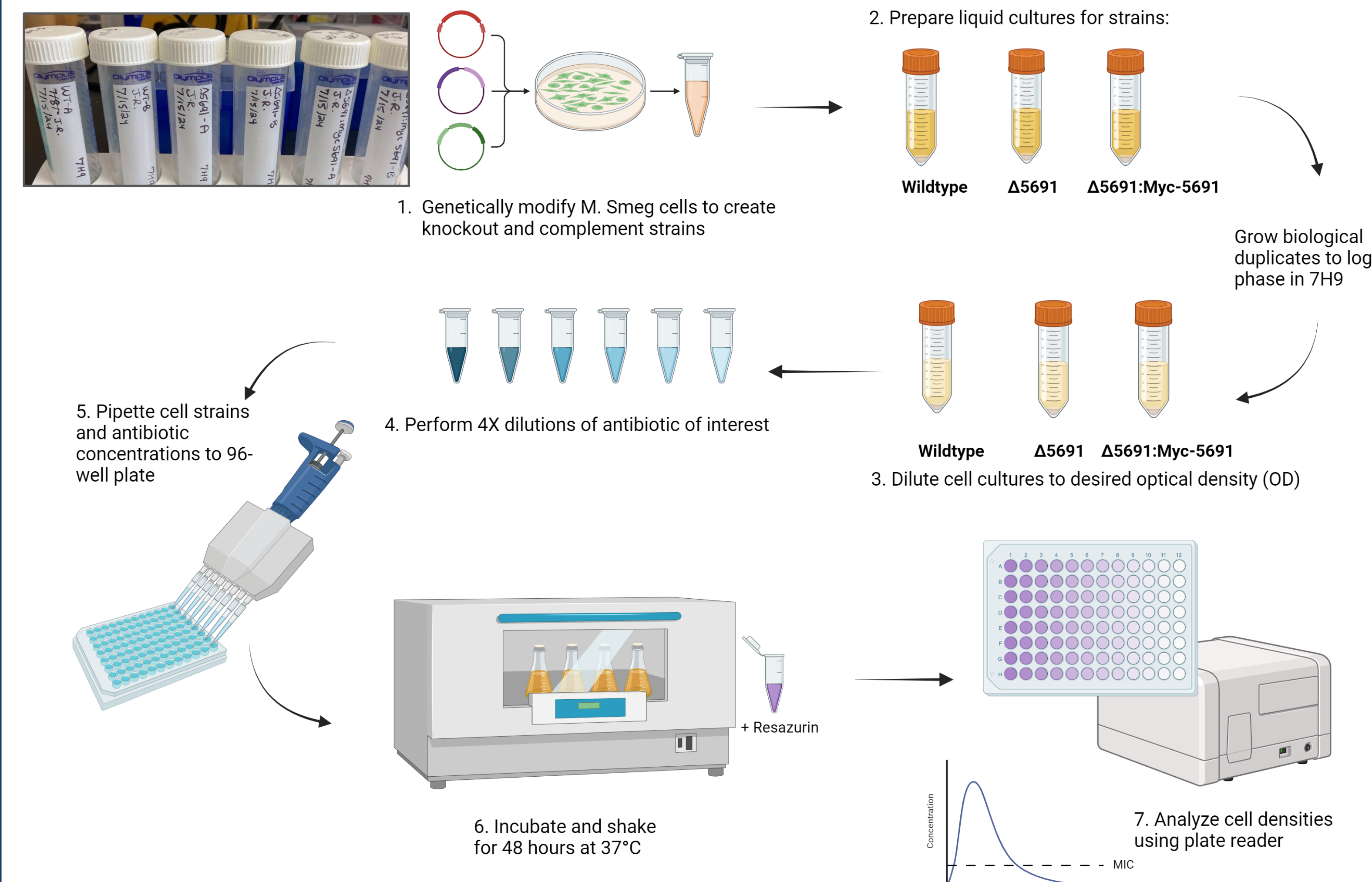
5691 AlphaFold. Image Credit: Julia Ryan

- Determine **in vitro** susceptibility and resistance of pathogenic microorganisms against antimicrobial agents.

Wildtype	Normal Growing Cells
$\Delta 5691$	"Knockout" - 5691 deleted from genome*
$\Delta 5691$:Myc-5691	Complement - 5691 added back into genome

Investigate **Minimum inhibitory concentration (MIC)** of antibiotics that interfere with translation to prevent growth of Wildtype, $\Delta 5691$, and $\Delta 5691$:Myc-5691.

Preparing MIC Growth Assays



Findings: Identify possible effects of 5691 on ribosomal translation related to the rRNA processing that has been previously observed in $\Delta 5691$.

Results and Conclusions

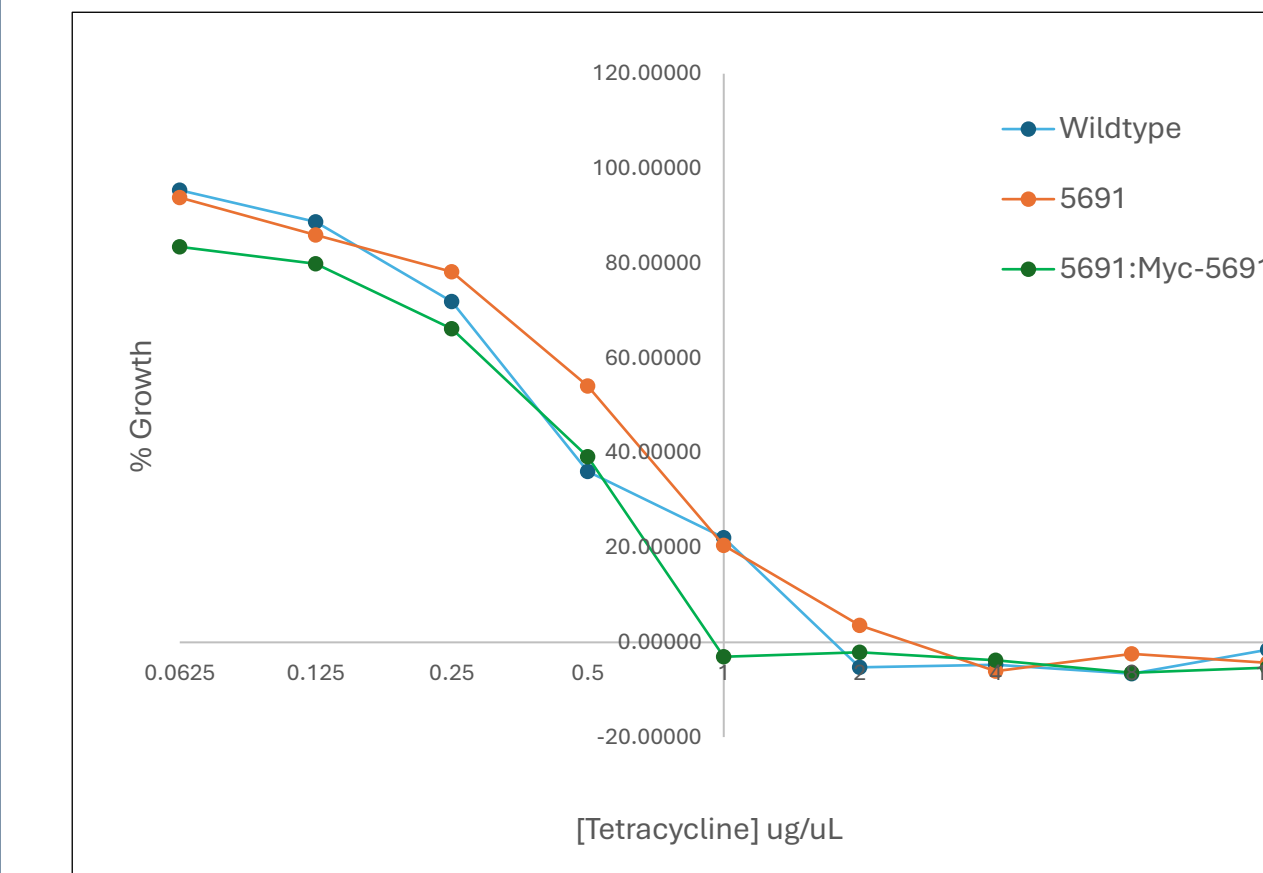


Figure 1. MIC cells grown in Tetracycline

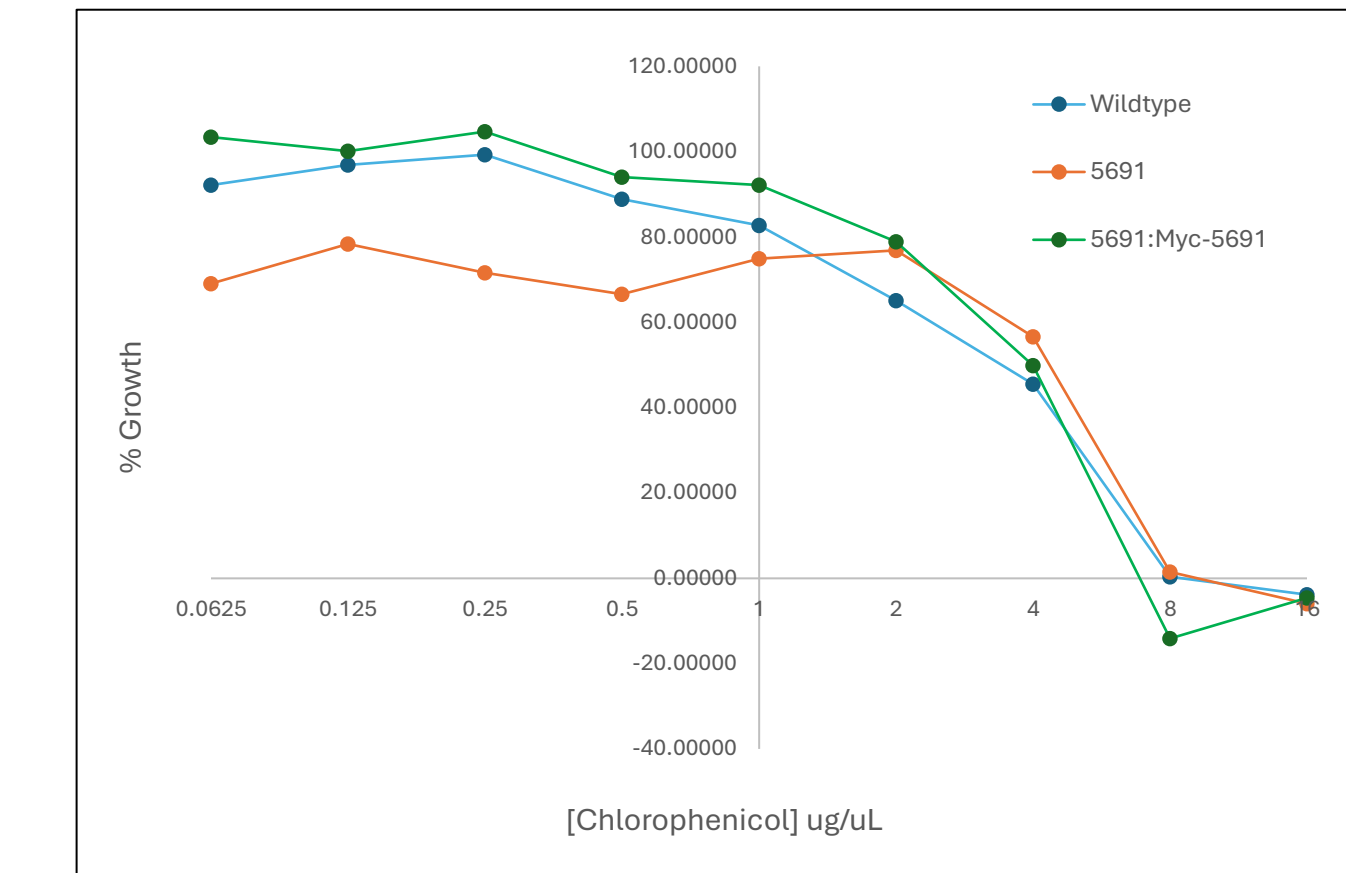


Figure 2. MIC cells grown in Chloramphenicol

Different growth patterns observed in the Knockout strain, when 5691 was removed.

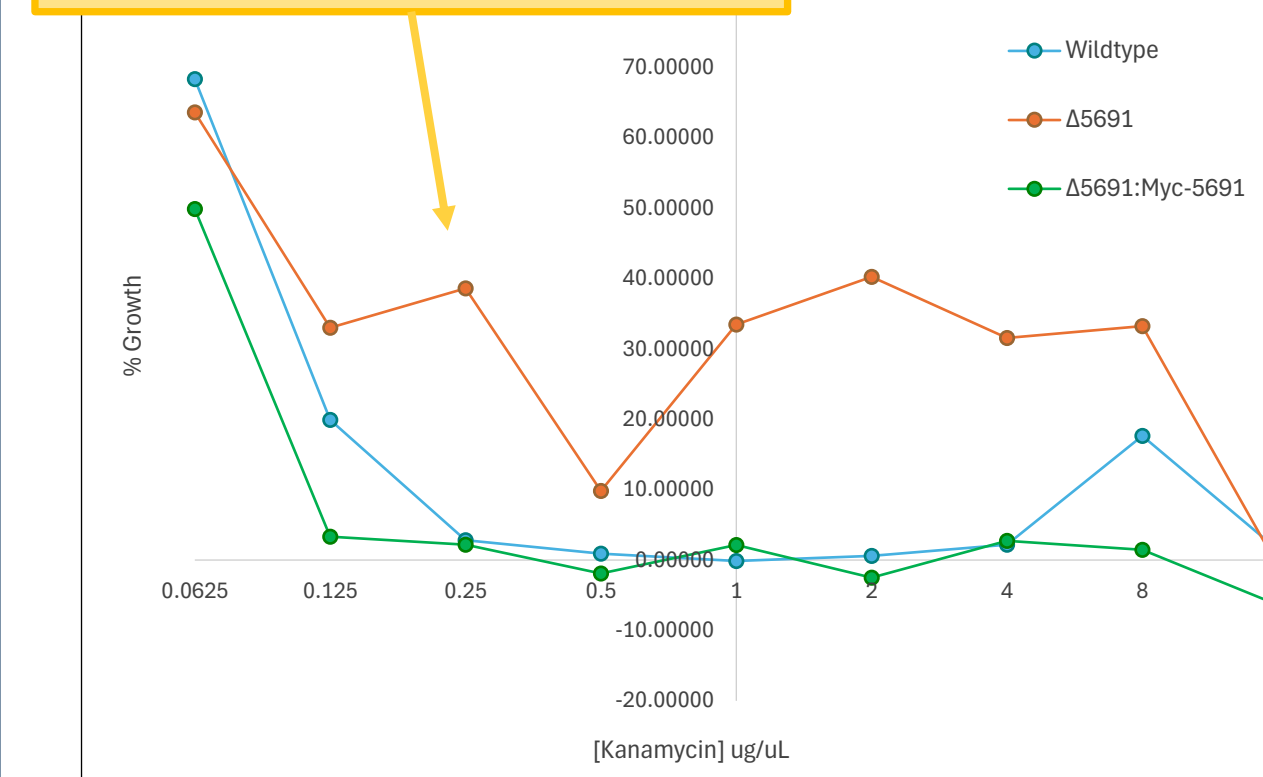


Figure 3. MIC cells grown in Kanamycin

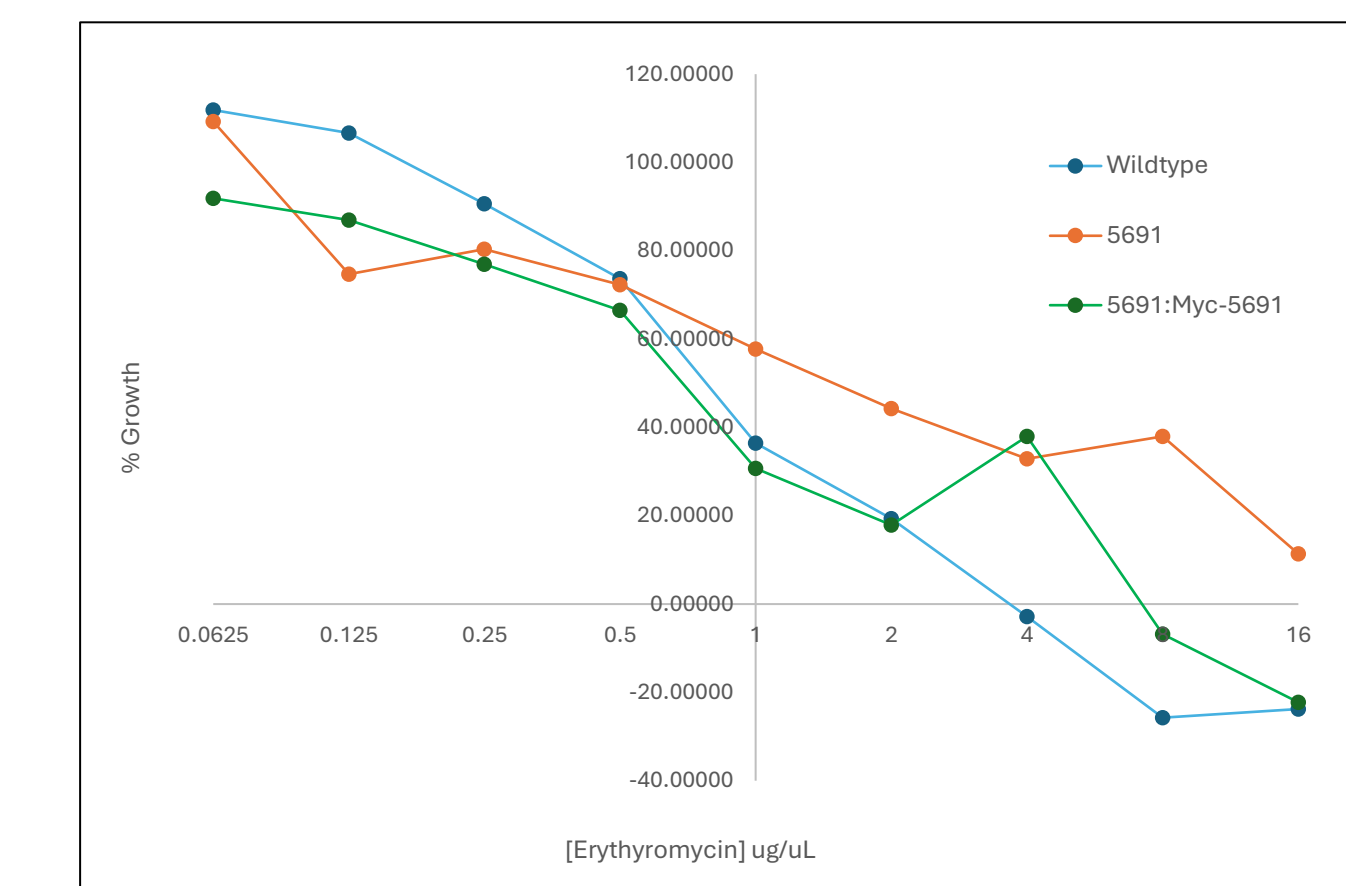
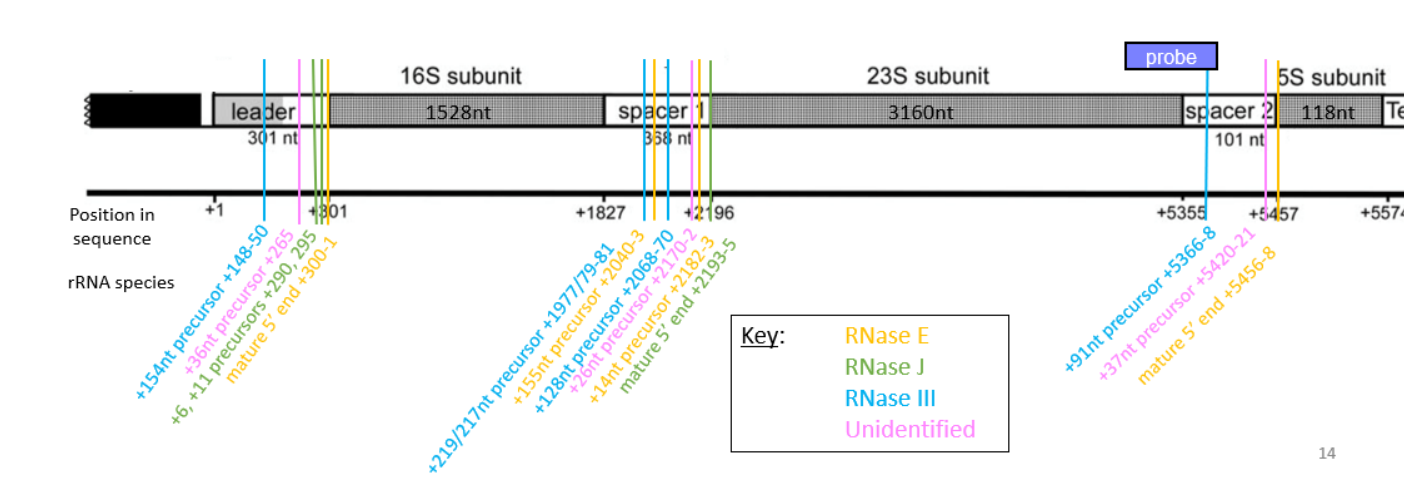


Figure 4. MIC cells grown in Erythromycin

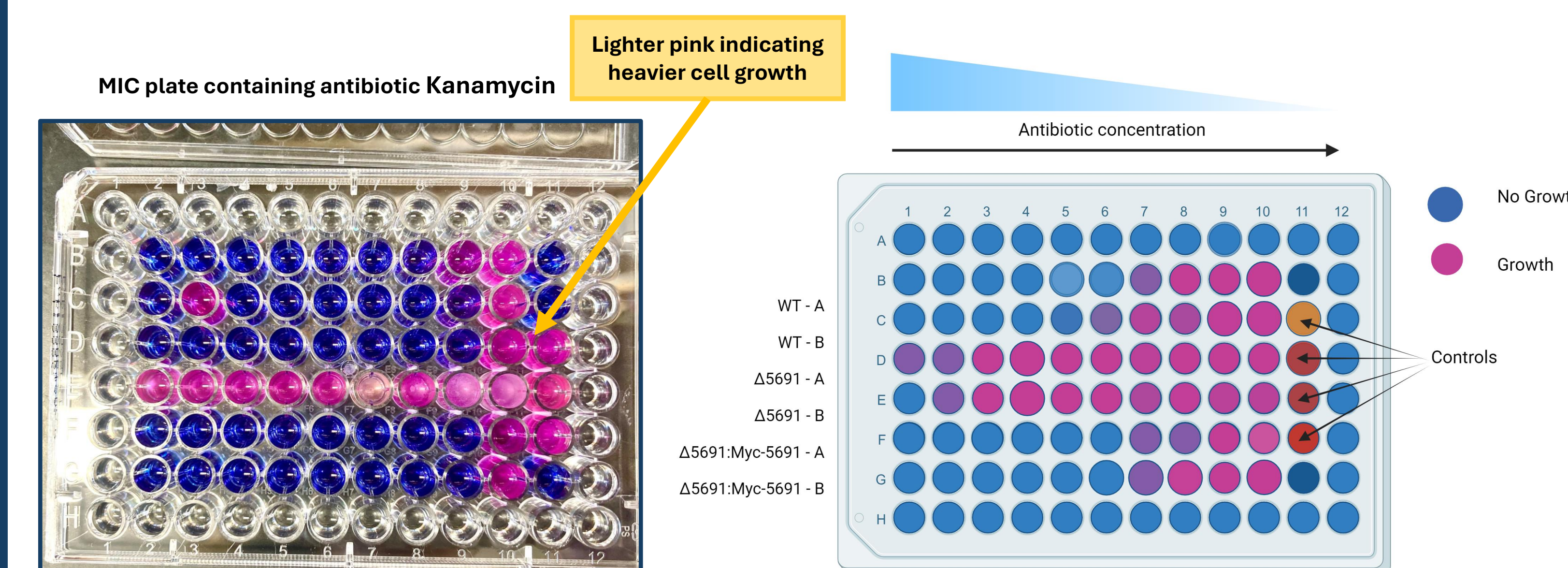
Results: Administering antibiotics from the class of Aminoglycosides, as seen in Kanamycin, have the greatest effect on inhibiting *M. Smeg* growth in the absence of 5691.

Future Work

- Analyze **genetic sequences** within RNA code to isolate genes that code for proteins involved with the RNase E complex.
- Important for cellular RNA degradation and mycobacterium survival.



Analyzing Cell Growth



Classroom Connections



- Construct **models** of transcription and translation for genetic disorders to explain the role of DNA and RNA coding for proteins that carry out essential functions of life.
- Perform **hands on lab inquiries** to identify mutations that result in genetic disorders.
- Investigate how **CRISPR gene editing technology** can be used to change the phenotype of an individual.

Acknowledgements

- A special thank you to Scarlet Shell and the members of the **Shell Lab** and all their help and encouragement including my mentor, Julia Ryan.
- Thank you to the members of the **STEM Education Center** including Kathy Chen, Donna Taylor, and Erin Solovey for making this work accessible to students.

This work is supported by the National Science Foundation under Grant No. EEC-205507.