# Effectiveness of *Escherichia coli* Nissle in preventing cariogenic environments caused by *Streptococcus mutans*

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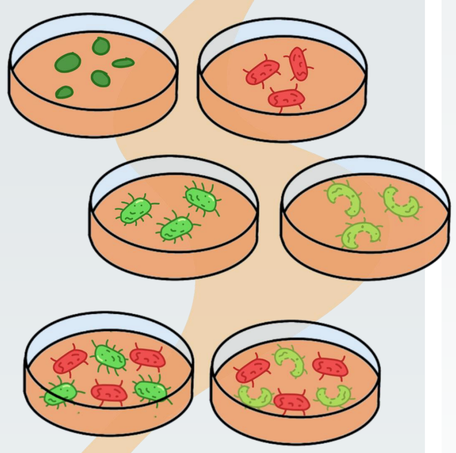
## INTRODUCTION

- **Dental caries** are a disease characterized by deteriorated enamel.
- *Streptococcus mutans* are highly cariogenic oral bacteria that can catabolize a large variety of carbohydrates to secrete organic acids and sticky polysaccharides called glucans that promote bacteria adsorption to the tooth (i.e. biofilm).
- The resulting biofilm matrix acts as a diffusion barrier, sustaining the acidic environment that supports cariogenic bacteria and causes enamel demineralization.
- This study investigates the competitiveness of *Escherichia coli* Nissle against *S. mutans* while it possesses a lysing circuit—the mechanism by which mammalian proteins can be released.

***Escherichia coli* Nissle (EcN)** and its engineered variants are evaluated in their ability to survive against *S. mutans* and inhibit its acid and biofilm production, as eventually they will be delivered to white spot lesions via fibrous scaffolds to facilitate remineralization and disinfection.

## METHODS

**Growth curves:** Brain Heart Infusion (BHI) media, EcN, sfGFP-tagged EcN, its self-lysing variant (SLIC), and *S. mutans* were pipetted into single-culture and co-culture wells in a 96-well plate. At 37°C, a plate reader records the plate's fluorescence and absorbance values for 24 hr.

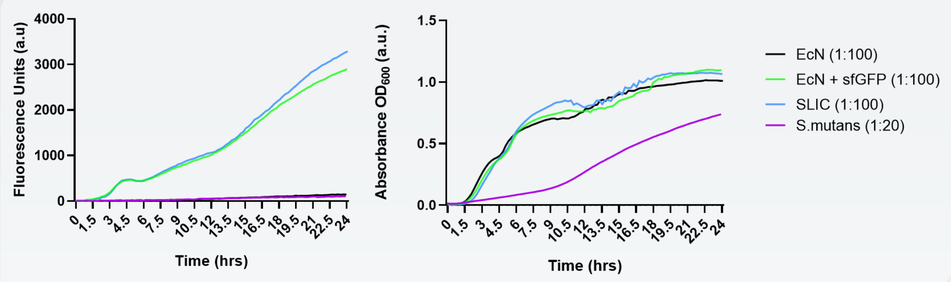



**Biofilm deposition:** After aspirating the cells and media, the wells are stained with 0.1% crystal violet solution. The plate reader reads the absorbance at 585 nm.

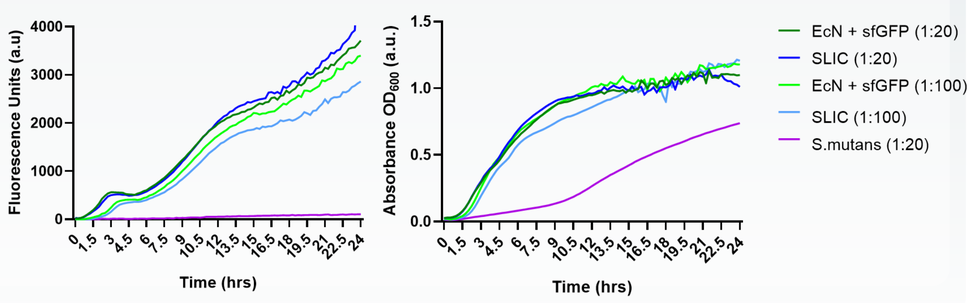
**pH:** The pH of corresponding Falcon tubes is each measured with a pH probe at 0, 4.5, and 24 hr.

## RESULTS

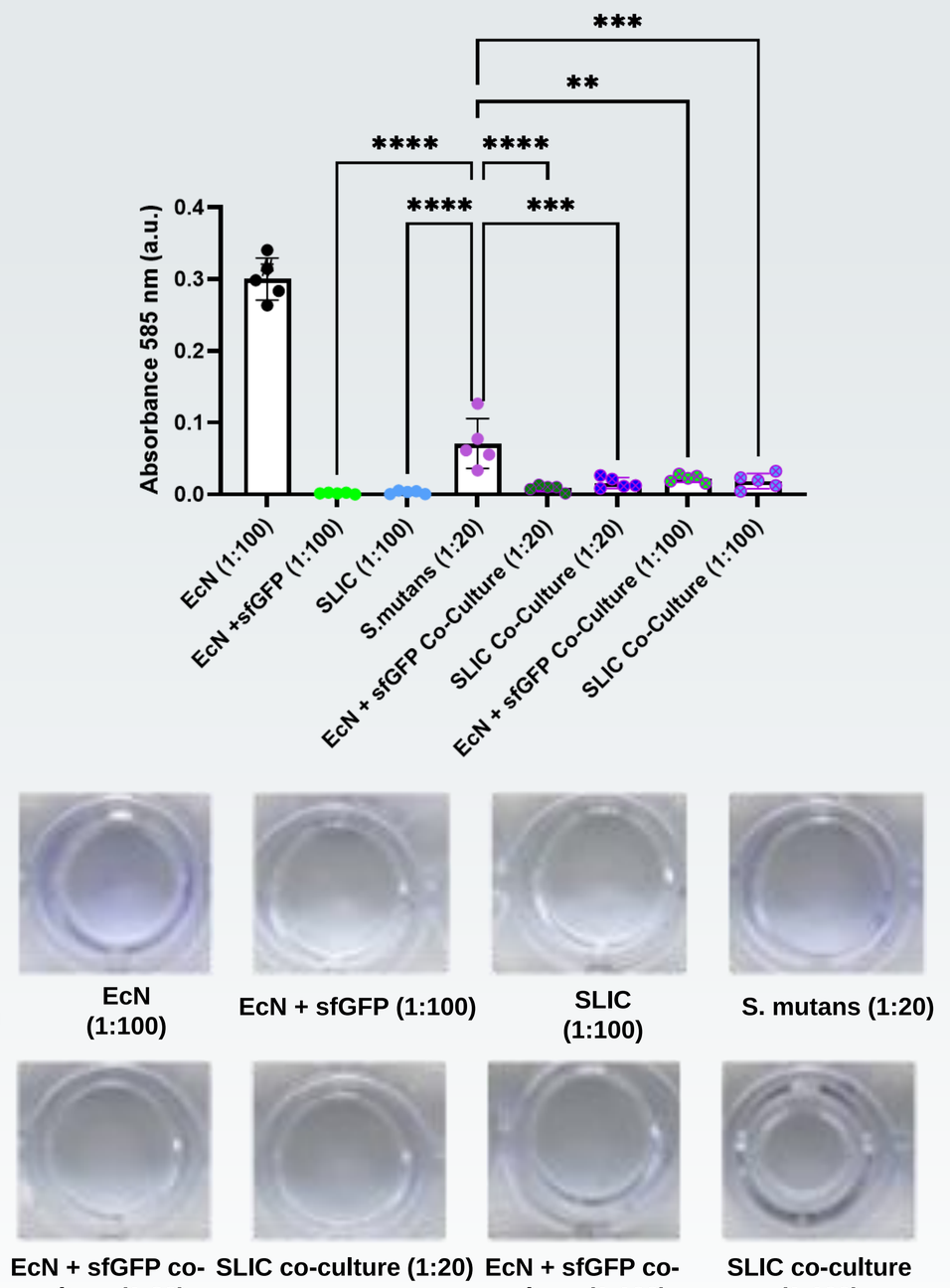
### Single culture growth curves over 24 hours




### Co-culture growth curves over 24 hours



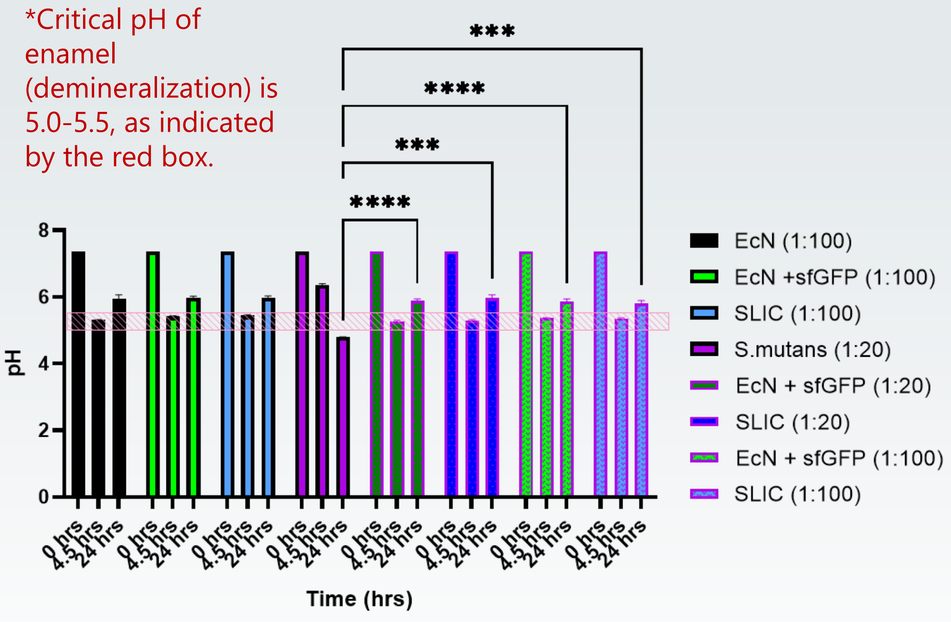

### Biofilm formation after 24 hours





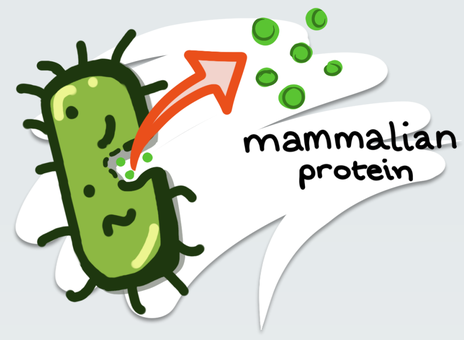


### pH over 24 hours

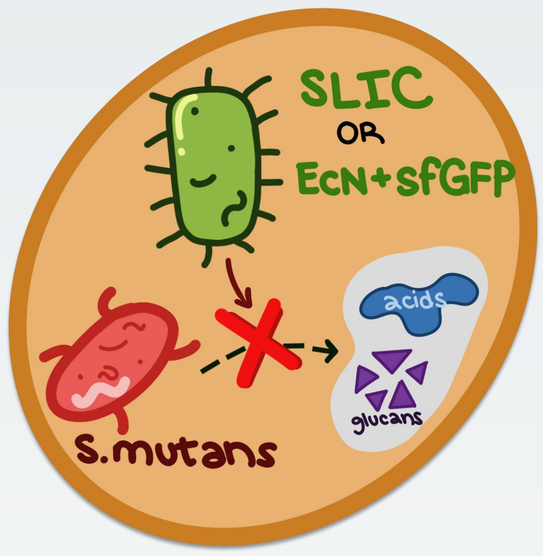



## CONCLUSION

- SLIC's survival is not significantly impeded by the presence of *S. mutans*, showing potential as probiotic that can release assistive proteins through self-lysis.




- Engineered EcN seems to reduce the biofilm production and acidogenicity of *S. mutans.*




- Further studies will assess EcN's survivability in conditions that more closely resemble the oral cavity (anaerobic and in artificial saliva).
- In addition to competing with *S. mutans*, this probiotic will be engineered to expel amelogenin to further induce enamel regeneration.

## REFERENCES

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