Augustine Tseng
Undergrad
LSUHSC, New Orleans, Louisiana

Mentor: Bo Yang and Zenzhang Tom Wen
Louisiana State University Health Center

"Biofilm Formation by *Rothia mucilaginosa* and *Candida albicans* in Dual-Species Significantly Enhanced"

Background: The oral microbiota is highly diverse and dynamic, and interactions between the different major species are believed to play an important role in oral health and disease. *Streptococcus mutans,* a keystone pathogen of human dental caries (aka tooth decay, cavities) can facilitate the colonization of *Candida albicans,* also known to cause oral thrush, leading to rampant early childhood caries. *Rothia mucilaginosa* is one of the most abundant bacteria of the oral microbiota, although its role in oral health remains unclear. Little information is available concerning how *R. mucilaginosa*, interacts with other major species in the oral cavity. **Objective:** This study is designed to investigate if and how *R. mucilaginosa* interact with *S. mutans* and *C. albicans,* influencing biofilm formation. **Methods:** *R. mucilaginosa, S. mutans* and *C. albicans* were grown individually in trypticase soy broth and yeast extract (TSY), and when reaching optical density (OD_{600nm}) ~0.5, diluted 1:100, and mixed proportionally with TSY plus sucrose. Aliquots were then loaded to 96 well plates in pentaplicate. The bacterial cultures were incubated at 37°C, 5 %CO_2 for 24 hours. By the end of the experiment, the biofilms on the wells were stained with 0.1% crystal violet, properly washed, and following extraction using ethanol-acetone mix, the biofilms in absorbance at 575nm were measured using a spectrophotometer. For composition and proportion of the different bacterium in a community, aliquots of serial dilutions of the mixed cultures were spotted on trypticase soy broth agar plates, and following incubation overnight, the colony-forming units (CFU) were determined by counting. **Results and Discussion:** Among the three strains, *S. mutans* showed the fastest planktonic growth rate and achieved the highest culture density, while *R. mucilaginosa* was the slowest with the least density. When grown on the polystyrene surface in 96-well plates individually, *S. mutans* produced the highest quantity of biofilms, followed by *R. mucilaginosa* with robust biofilms, while *C. albicans* produced only limited biofilms. When grown in dual-species model, *C. albicans* and *S. mutans* displayed enhanced biofilm formation, compared to the respective single-species model, which is consistent with the literature. *C. albicans* and *R. mucilaginosa* together increased biofilms by >5-fold than they grew alone. No significant differences were observed when *S. mutans* was growing together with *R. mucilaginosa.* Interestingly, when *R. mucilaginosa*, *S. mutans* and *C. albicans* were grown in triple-species, significantly less biofilms were measured, as compared to the respective dual-species models. These results suggest that *C. albicans* and *R. mucilaginosa* in a community interact significantly influencing biofilm formation, although the underlying mechanisms await further investigation.