Identification of New Signaling Components that Govern Biofilm Formation in *P. aeruginosa*

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**Introduction**

- *Pseudomonas aeruginosa* is an opportunistic pathogen.
- *P. aeruginosa* is often difficult to treat due to its ability to produce biofilms, in which cells are surrounded by a self-produced extracellular matrix of polysaccharides, lipids, and proteins.
- These biofilms offer a barrier to protect the bacteria from natural host defenses and treatments such as antibiotics.

**Methods**

**Visual Assay**

Once colonies with the transposon insertion were grown, colony morphology could be observed to assess the amount of biofilm formation.

**Transposon Mutagenesis**

By mating *E. coli* with a moderately wrinkled *P. aeruginosa*, strain DamrZ, a transposon was inserted. Using selective media with IRG, an antibiotic that transposon carried resistance to, and gentamycin, an antibiotic lethal to *E. coli*, *P. aeruginosa* colonies with the transposon insertion was selectively grown.

**Sequencing**

Following the visual assay for the transposon-inserted mutants, mutants are purified using PCR to find the location of the transposon insertion.

**Results**

- PA_30280, trxA1
- thioredoxin reductase
- Thioredoxin reductase plays a role in DNA synthesis and defense against oxidative stress
- PA14_71530 PurU2
- Formyltetrahydrofolate Deformylase
- Activated by host responses in infections
- CSS 136
- Upstream
- PA14_21970–PA3249
- (transcriptional regulator)
- No known interaction with biofilm production
- intergenic region 21960 and 21970

**Future Directions**

In the future, we will work to quantify the amount of biofilm being produced by using a Congo Red dye assay and by measuring cyclic diGMP levels in the cell.

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