Homology Modeling and Redox Partner Identification for *Pseudomonas aeruginosa* CYP168A1

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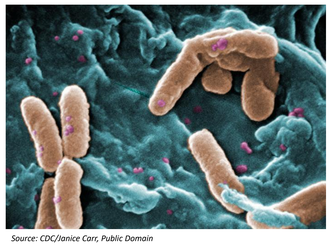
Background and Significance

Antibiotic resistance is a growing problem in medicine, primarily because of the extensive use of broad-spectrum antibiotics. One proposed method of overcoming and slowing the progression of antibiotic resistance is to develop species-specific inhibitors for problematic pathogens.



Source: CDC/Janice Carr, Public Domain

Pseudomonas aeruginosa is one of these pathogens, and It has a P450, CYP168A1, that is a possible target for inhibition. Importantly, recent reports[1] have identified a cytochrome P450 (CYP168A1) from *Pseudomonas* that is capable of producing lipids that resemble known immunomodulators and that has expression patterns correlating with pathogenicity.

Objectives

Firstly, we wanted to design a homology model for CYP168A1 to identify possible pockets within the closed structure that is different from the open one. These pockets are the site of future inhibitor design efforts.

Also, in some P450s, the native partner proteins affect the final product of enzymatic reactions within the protein, so, we set out to propose what these partner proteins are in *Pseudomonas*.

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Homology Modeling and Identification of Pockets for CYP168A1

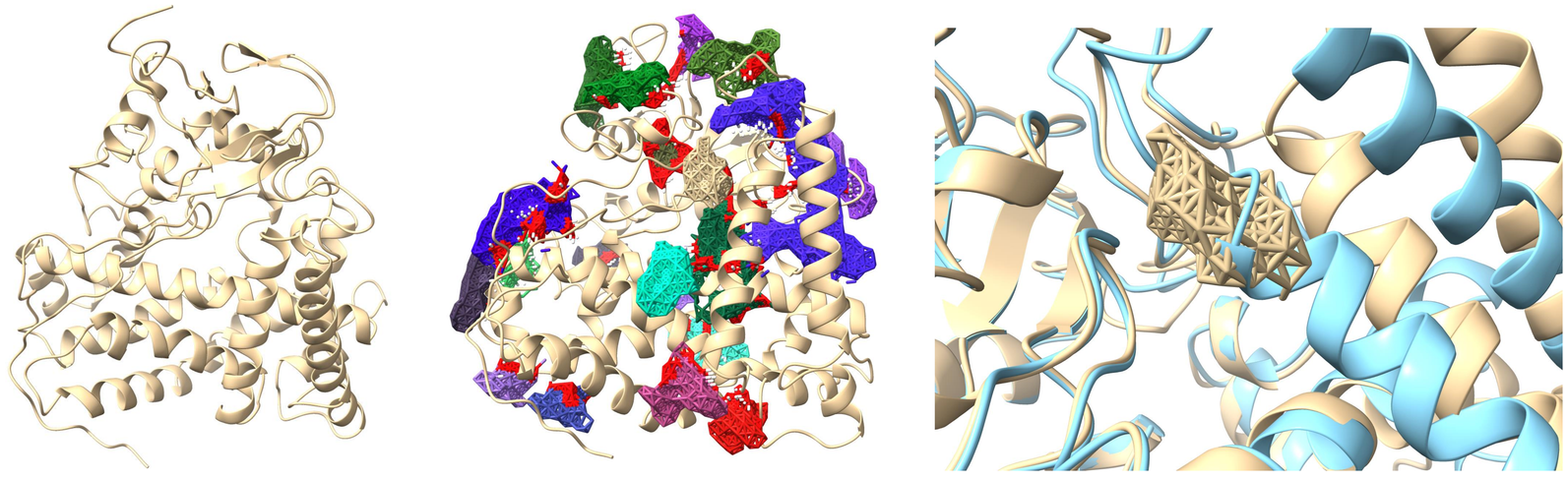

Figure 1: **The Process of Identifying Pockets in CYP168A1.** The process started by creating an initial homology model (left). Then, using autosite,[2] pockets were identified within the model (center). Finally, comparing the open and closed confirmations with the located pockets led to the discovery of possible locations for inhibitor design (right). Images created with Chimera X.[3]

Partner Protein Identification

To better characterize CYP168A1, we identified possible redox partner proteins within the reference genome of *Pseudomonas*. This was done via a protein BLAST of the reference genome for homologues of putidaredoxin, chloroplastic ferredoxin, adrenodoxin, and their corresponding reductases.

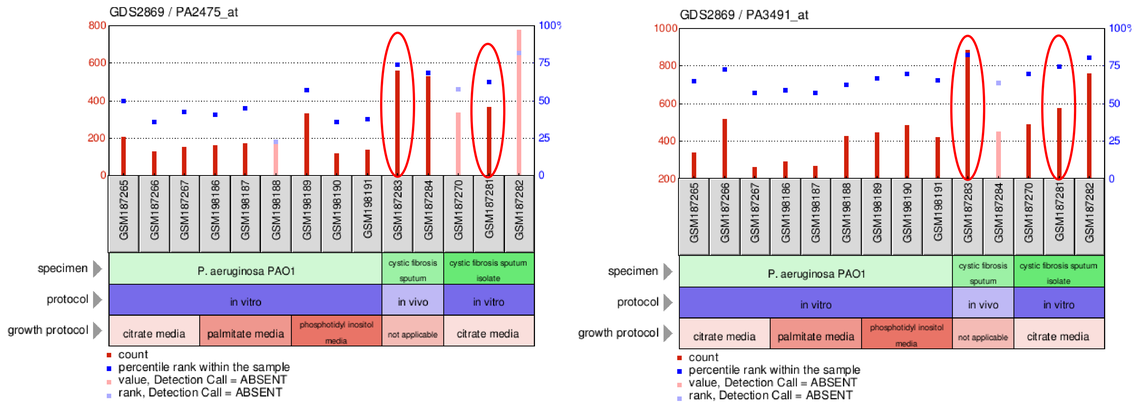

Figure 2: **Correlation data between CYP168A1 (left) and a possible partner protein (right).** To help reduce the search of possible partner proteins, I looked at the strength of expression in each protein in correlation with CYP168A1.[4] Charts were taken from *pseudomonas.com.*[5]

Future Plans

The next step for the homology modeling project is to refine the current models by relaxing their structure. This allows for a greater accuracy in identifying pockets within the molecule as well as designing possible inhibitors.

The next step in the partner protein identification project is to begin cloning the putative partners for expression and purification. After the proteins are purified, *in vitro* reactions can be performed investigating what changes in reactions occur with CYP168A1 in the presence of the partners.

References

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2. *Bioinformatics* **2016**, *32*, 3142–3149
3. *Protein Sci.* **2018**, *27*, 14-25; *Protein Sci.* **2021**, *30*, 70-82.
4. *Infect Immun*. **2007**, *75*, 5313-5324.
5. *Nucleic Acids Res.* ***2016***, *44*, D646–D653