Volumetric Analysis of Cytochrome P450 Binding Pockets

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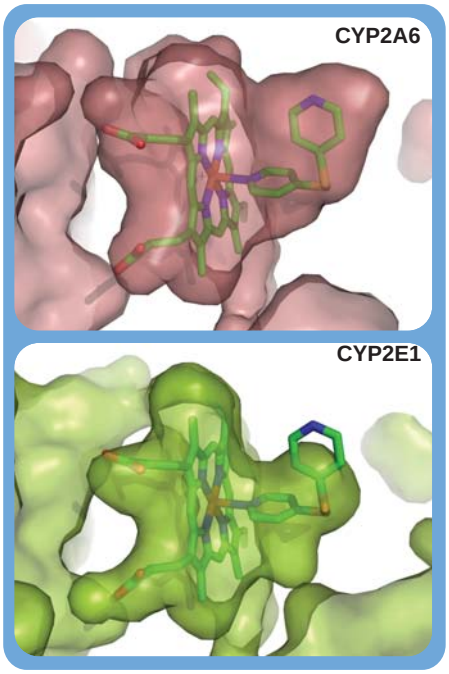
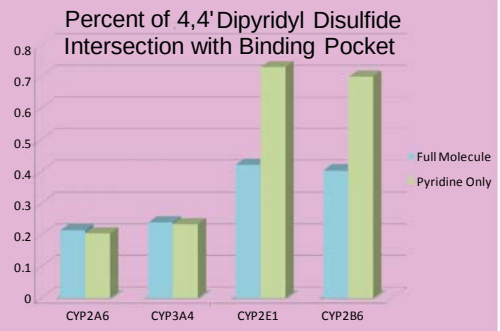
Background

Cytochrome P450s are a crucial family of proteins involved in the metabolism of multiple substrates involved in homeostasis, including hazardous chemicals (xenobiotics). One such cytochrome, CYP3A4, metabolizes over 50% of marketed pharmaceuticals in the liver,[1] partially inhibiting the delivery of drugs from their targets. To better understand how cytochromes interact with drugs, we are developing methods for examining similarities and variations in their binding sites. In this study, volumetric analysis was used to compare the bound position of 4,4'-Dipyridyl Disulfide, modeled on four out of the fifteen members of xenobiotic metabolizing cytochrome P450s. We observed that 4,4'-Dipyridyl Disulfide has little steric clash with CYP3A4 and CYP2A6 while exhibiting significant clash with CYP2E1 and CYP2B6. These observations demonstrate that volumetric analysis can identify differences in protein structure that influence drug selectivity.



Results

CYP2A6 and CYP3A4 had low volumes of intersection with 4,4'-Dipyridyl Disulfide while CYP2E1 and CYP2B6 had large volumes of intersection. These intersections indicate that 4,4'-Dipyridyl Disulfide can easily bind in the pockets of CYP2E1 and CYP2B6, and not in those of 2E1, as established experimentally.[3]

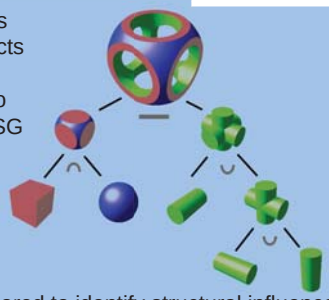


The pyridine ring in 4,4'-Dipyridyl Disulfide caused the highest point of intersection, indicating a geometric region contributing to specificity.

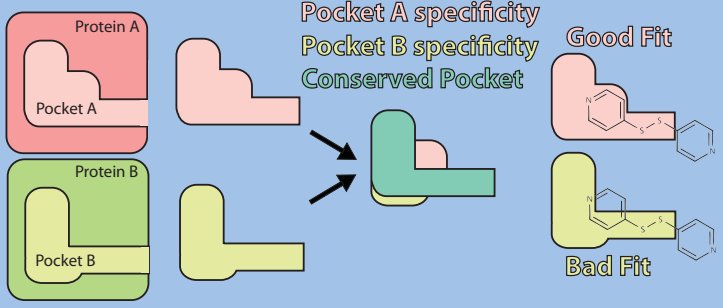
These preliminary observations represent a small-scale experiment that point to large-scale potential. By automating this analysis on a large sample, a profile of the cytochrome P450 binding sites could potentially be realized. Such a profile would enable more accurate drug repurposing and potentially decreased metabolization of pharmaceutical drugs by major cytochrome groups.

Volumetric Analysis

Our software represents proteins as geometric solids. These objects can be manipulated with CSG (Constructive Solid Geometry) to compare molecular surfaces. CSG provides volumetric unions, intersections, and differences between regions.



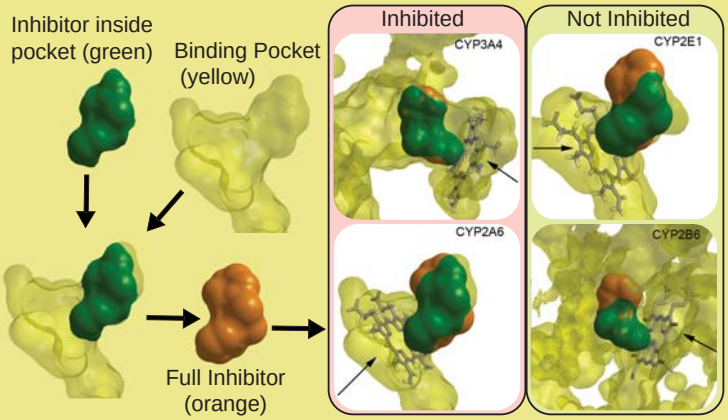
Molecular surfaces can be compared to identify structural influences on specificity. VASP (Volumetric Analysis of Surface Properties), created by Brian Y. Chen, can be used to isolate differences in binding pockets. These variations often directly influence to the specificity of the protein.[2]



Protein regions contributing to specificity can be identified and compiled to catalog potential influences on protein binding site specificity. These catalogs can be compared with those from other proteins to infer similarities and differences in binding affinity.

Experimental Method

Four members of the Cytochrome P450 family were examined: CYP2A6, CYP3A4, CYP2E1, and CYP2B6. They were aligned using PyMOL, a molecular visualization system. Once aligned, we computed the CSG intersection between the cytochrome binding pocket and that of 44DD, and measured the volume of the region where they overlap.



Non-inhibited cytochromes have a larger orange sections on the inhibitor, indicating a poor fit.

References

1. Denisov, Ilia. "Structure and Chemistry of Cytochrome P450." American Chemical Society. 105. (2005): 2253-2277. Print.
2. Chen BY, Honig B (2010) VASP: A Volumetric Analysis of Surface Properties Yields Insights into Protein-Ligand Binding Specificity. PLoS Comput Biol 6(8): e1000881. doi:10.1371/journal.pcbi.1000881
3. Fujita, Ken-Ichi. "SCREENING OF ORGANOSULFUR COMPOUNDS AS INHIBITORS OF HUMAN CYP2A6." DRUG METABOLISM AND DISPOSITION. 29.7 (2001): 983-989. Print.

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