Crystallization and structural studies of an aldo-keto reductase from opium poppy

M.A. Torres[1], M. Dastmalchi[1], C. Comfort[1], P. Facchini[1], K.K.S. Ng[1]

[1] University of Calgary, Calgary, Canada

Plants are the source of many widely used pharmaceuticals produced via a plethora of specialized metabolic pathways. Benzylisoquinoline alkaloids (BIA) are a large and diverse group of pharmacologically active compounds found in a variety of plants, including the opium poppy. Among the most important BIAs are morphine and its immediate biosynthetic precursor codeine. In plants, codeine is reduced from codeinone by the enzyme codeinone reductase (COR). However, COR also catalyzes alternative reactions that yield unwanted compounds, such as neopine. The accumulation of less desirable products from these side-reactions seriously compromises the efficiency of current synthetic biology efforts to develop fermentation-based production systems in yeast and bacteria. To understand the molecular structural mechanisms responsible for substrate recognition, and to inform protein engineering and systems biology efforts to minimalize unwanted alternate reactions, we have crystallized COR and measured diffraction data to 2.9 Å resolution (completeness 96.1%, R_{sym}=0.127, redundancy=3.0) at SSRL beamline 9-2. Molecular replacement calculations using chalcone reductase (52% sequence identity) as a search model indicate six copies in the asymmetric unit (space group $P2_1$; a=78.4 Å, b=90.1 Å, c=143.7 Å, β=94.2°; 47.1% solvent). Some initial results from differential scanning fluorimetry suggest directions to facilitate the further optimization of crystal quality and the cocrystallization of complexes with substrates and cofactors.