Experimental Report

Proposal number: 30-01-853	**structural characterisation of lectin and glycosyl hydrolases**
Beamline: BM30A	**Date(s) of experiment:** from: 16/07/2010 to: 17/07/2010

Objective & expected results.

We want to determine the molecular basis of the lectins-sugars interactions in order to better understand the necessary determinants for binding (lectins) or catalytic mechanism (glycosyl hydrolase). We need to obtain data at high resolution from relatively small crystals. We wanted to test crystallization conditions from crystallization robot in order to decide which are the ones best to optimize.

Results and the conclusions of the study:

We tried to dissect the mechanism of the family 93 of the glycoside hydrolase through the structure determination of protein-ligand complexes of the arabinofuranosidase Arb93A from *Fusarium graminearum*. We solved the structure of the proposed catalytic base E242A and of the wild-type with an inhibitor designed for arabinofuranosidases, figure1. The latest has shown ring distorsion in the clivage site and allowed us to proposed a conformationnal itinary for Arb93A.

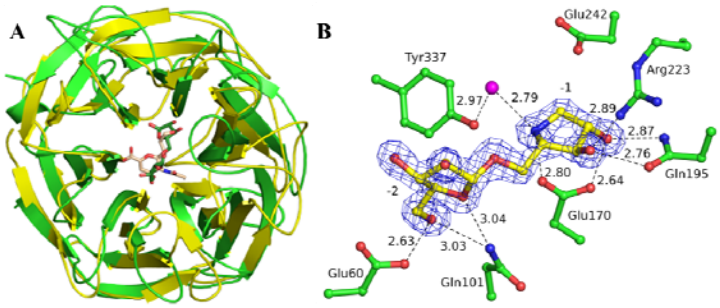

Figure 1: Crystal structure of the arabinanase Arb93A in complew with imino-sugar inhibitor at 1.6Å.

The opportunistic bacteria *Burkholderia ambifaria* is part of the Burkholderia cepacia complex responsible of nosocomial infections. It presents a fucose binding lectin homologue to the lectin RSL from *Ralstonia solanacearum* specific for fucosylated blood group antigens. We solved several structures of BambL in complex with different blood group epitopes. The lectin is trimeric and forms a 6 blade beta-propeller with two binding sites per monomer, figure 2. We have analysed the two different binding sites and tried to rationalised the binding of BambL.

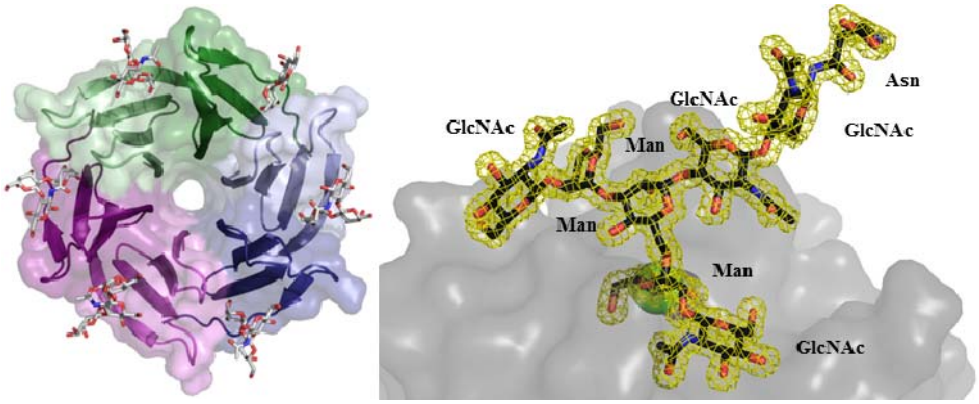

Figure2: Left: Overal fold of the BambL lectin in complex with H-type 1 antigen. Right. 2Fo-DFc electron density observed for the heptasaccharide bound to the lectin PelA contoured at 1sigma (0.47e Å3).

The plant lectin from *Platipodium elegans* recognised mannose and in particularly asymetrical N-Glycans. We solved the structure of PelA in complex with a symetrical heptaoligosaccharide at 1.6 Å which has hallowed us to better understand its preference for asymetrical ligands, figure 2.

Publication(s):

1-Goddard-Borger ED, Carapito R, Jeltsch JM, Phalip V, Stick RV, Varrot A. **α-l-Arabinofuranosylatedpyrrolidines as arabinanase inhibitors.** *Chem Commun*, 2011, 47(34):9684-6.

2-Audfray A, Claudinon J, Abounit S, Ruvoën-Clouet N, Larson G, Smith DF, Wimmerova M, Le Pendu J, Römer W, Varrot A, Imberty A. **The fucose-binding lectin from opportunistic pathogen Burkholderia ambifaria binds to both plant and human oligosaccharidic epitopes**. *J Biol Chem* 2012, doi 10.1074/jbc.M111.314831.

3-Guimarães Benevides R, Ganne G, da Conceição Simões R, Niemietz M, Unverzagt C, Chazalet V, Breton C, Varrot A, Sousa Cavada B, Imberty A. **A lectin from Platypodium elegans with unusual specificity and affinity for asymmetric complex N-glycans**. *J Biol Chem* 2012 (under revision).