

Abstract No. Qasb0230

**X-ray Crystallographic Investigations on the Structure and Function of  $\beta$ 1,4-Galactosyltransferase**

P.K.Qasba (NCI), B.Ramakrishnan (SAIC at Frederick /NCI) and R.Velavan (NCI)

Beamline(s): X9B

**Introduction:** Structure and function studies on the  $\beta$ 1,4-galactosyltransferases (Gal-T1) have been carried out by single crystal structure determination on the various mutants of Gal-T1 in the presence and absence of its substrates. From our previous crystal structure studies on Gal-T1 and its complex with  $\alpha$ -lactalbumin (LA) it has been shown that Gal-T1 undergoes conformational changes upon its substrate binding from open to closed form. Further more, these conformational changes are essential not only for the binding of its modulator protein  $\alpha$ -lactalbumin but also for its enzymology.

**Methods and Materials:** In order to understand this conformational dynamics we have under taken studies using both single crystal structure determination and enzyme kinetic studies on several mutants of Gal-T1

**Results:** The crystal structure of the mutant W314A in the absence of the substrate at 1.9 D resolution revealed that the mutant prefers an open conformation. On the contrary, the crystal structure of the mutant M344H at 2.4 D resolution revealed that it readily undergoes conformational change from open to close form in the presence of UDP compared to the wild-type Gal-T1. Also, this structure reveal the binding of sulfated-GlcNAc to Gal-T1, whose enzymology is least known. Further crystal structure analysis on the mutants such as His347E, R228K\_Y286L and M344H crystallized in the presence of different substrates are underway.

**Conclusions:** These studies are expected to enhances our understanding on the structure and function of Gal-T1 which is an important enzyme for the synthesis of oligosaccharide chains of glycoproteins and glycolipids.

**Acknowledgments:** This project has been funded in whole or in part with Federal funds from the National Cancer Institute, National Institutes of Health, under Contract No. NO1-CO-12400.