Preliminary Crystallographic Studies Of Human SET Protein

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The human SET protein is a potential marker for head and neck cancer by presenting altered expression levels in these tissues. SET is a multifunctional protein that has been shown to exhibit histone chaperone activity, interact with various factors such as DNA-binding proteins and regulate transcription, replication, and apoptosis. In this project we are focusing on the structural characterization of SET protein by X-ray crystallography including expression, purification and crystallization experiments. SET has been cloned in pET23a expression vector and expressed in *Escherichia coli* strain BL21 (codon plus). Cells were grown in Luria-Bertani (LB) medium supplemented with carbenicilin (50 μg/ml) and protein expression was induced by IPTG (500μM by 3h at 37^{o}C). SET was purified to homogeneity by affinity chromatography using a poly-L-lysine resin connected to ÄKTA *Prime* purification system using a gradient of NaCl in buffer containing Tris 100mM, EDTA 10mM, PMSF 100pM and protease inhibitor cocktail. The final product appeared as a single band in SDS-PAGE. The preliminary crystallographic studies consisted of crystallizing human SET protein using the sparse-matrix approach by the vapor diffusion technique. Crystals of SET were obtained in presence of PEG and NaCl. X-ray diffraction analysis of SET protein crystals are in progress.