

Summary report for project MX-1099- Structural studies on retinoblastoma protein and its conformational changes upon phosphorylation:

1. Project objectives.

The project aims to get a structural information on retinoblastoma protein and its conformational changes upon ligand binding or phosphorylation.

2. Results.

Full-length Rb contains three recognized structural domains, the N-terminal domain (RbN), the central Rb pocket domain and the C-terminal domain (RbC). The structures of the individual subdomains are known, however how the different domains align and interact to form the full size molecule is not clear.

To answer this question, the fragment of Rb containing RbN and Rb pocket domains has been studied by SAXS. The *ab initio* model has been built. However, due to the fact that RbN and Rb pocket domains structures have high similarity, from the current *ab initio* model it was impossible to determine the domains orientation within the molecule. To overcome this difficulty, maltose binding protein (MBP) has been fused to the N-terminus of the Rb fragment containing RbN and Rb pocket domains, and the fusion protein has been expressed and purified. The analysis of the fusion protein SAXS data has allowed to obtain the orientation of the RbN domain. However, the orientation of the Rb pocket domain is still unknown. The additional SAXS measurements using different Rb constructs are needed to obtain the accurate data on the Rb pocket domain orientation and to proceed further for the studies on Rb phosphorylation.