Abstract
This thesis was worked up as a part of a project that is concerned with the manner of interaction of 14-3-3 proteins with forkhead transcriptional faktor FoxO4. 14-3-3 proteins are proteins interacting with a considerable amount of cell proteins. They affect activity and cell localization through this interaction. Transcriptional factor FoxO4 plays significant role in regulation of a cell cycle, apoptosis, respond to oxidative stress and many other cell proceses. Transcriptional aktivity of FoxO4 is regulated through phophorylation by protein kinases B. This phosphorylation induce inhibition of FoxO4/DNA binding, induce FoxO4/14-3-3 binding and thus relocalization of this complex from a cell core to cytoplasma. One of main goals of this project is to explain molecular mechanisms of these interactions.
Main goal of this thesis was to create cDNA of a mutational protein FoxO4 containing just two tryptophan residues by site directed mutagenesis and after gaining this cDNA to express and pyrity this mutational FoxO4. This mutational protein was going to be a part of a model system for conformational changes studies. Besides expression and purification of 14-3-3 protein that is also part of our model system was performed.