Blood serum proteins serve various functions, including transport of lipids, hormones, vitamins. They are responsible for maintaining acid-base balance, oncotic pressure, plasma viscosity, and functioning of the immune system. There are several hundred different proteins in the blood serum, which total concentration varies within the limits of 6.6-8.7 g/dl, but only a small amount is determined for laboratory diagnostics. One of the serum protein is butyrylcholinesterase (BChE, EC 3.1.1.8), which exists predominantly in the form of a glycosylated tetramer (G4) with a mass of 340 kDa. Four identical subunits assemble into a tetramer by the interaction of a proline-rich peptide with the BChE tetramerization domain at the C-terminus. Our results suggest that BChE interacts with plasma proteins and form much larger complexes than predicted from mass of tetramer. In order to investigate and isolate such complexes we developed a strategy to find protein-protein interactions by combined native size-exclusion chromatography (SEC) with affinity chromatography using resin that binds BChE. Moreover, to confirm specificity of protein complexes we performed also fractionation of blood serum proteins by density gradient ultracentrifugation followed by co-immunoprecipitation using anti-BCHE monoclonal antibodies. The proteins isolated in complexes with BChE were identified by mass spectroscopy.

Acknowledgement

This project was supported by the Polish National Science Center (NCN), grant No. 2017/01/X/NZ4/01522