Apolipoprotein E3 mutants linked with development of Type III Hyperlipoproteinemia alter the protein's thermodynamic properties

Dimitra Georgiadou[#], Angeliki Chroni[¶], Alexander Vezeridis[$], Vassilis I. Zannis[$] and Efstratios Stratikos[#]

[#] Protein Chemistry Laboratory, IRRP, National Centre for Scientific Research Demokritos, Agia Paraskevi, Athens 15310, Greece

[¶] Institute of Biology, National Centre for Scientific Research Demokritos, Agia Paraskevi, Athens 15310, Greece

[$] Molecular Genetics, Departments of Medicine and Biochemistry, Whitaker Cardiovascular Institute, Boston University School of Medicine, Boston, Massachusetts 02118

Abstract

Apolipoprotein E is a major lipid carrier protein in humans, plays important roles in lipid homeostasis and protection from atherosclerosis. ApoE is characterized by structural plasticity and thermodynamic instability and can undergo significant structural rearrangements as part of its biological function. Mutations in the LDL binding region 136-150 of apoE have been linked with lipoprotein disorders such as Type III Hyperlipoproteinemia in humans and experimental animals. In this study we examined whether three such mutations, namely R136S, R145C and K146E affect the thermodynamic stability and conformation of apoE3. Circular dichroism spectroscopy revealed that the mutations do not alter the secondary structure of the protein. However, all three mutants have altered thermodynamic stability compared to wild-type as evidenced by thermal and chemical denaturation experiments. Furthermore, all three variants are able to clarify DMPC emulsions, but with subtle changes in kinetics that suggest differences in equilibrium distribution of conformations. Overall, our findings suggest that single amino acid changes in the functionally important region 136-150 of apoE3 can affect the molecule's stability and structural lability in solution. We propose that these thermodynamic alterations be taken into account when evaluating the pathogenic potential of apoE3 variants.