Self-association of band 3 protein from erythrocyte membranes in solutions of a nonionic detergent, Ammonyx-LO

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Band 3, the anion exchange protein of the human erythrocyte membrane, was solubilized and purified in solutions of a nonionic detergent, Ammonyx-LO, similar to the method of Yu and Steck (1). The self-association of purified band 3 was studied, in detergent solutions of pH 8.0 and an ionic strength of approx. 50 mM, by analytical ultracentrifugation. Protein concentration was between 20 and 500 µg/ml.

In sedimentation velocity runs, band 3 showed a single boundary the s-value of which strongly and reversibly increased with increasing protein concentration. This is indicative of an association equilibrium. The occurrence of self-association and the existence of an association equilibrium of band 3 was confirmed by sedimentation equilibrium experiments. By mathematical analysis of the concentration distributions, monomers, dimers and tetramers of band 3 were identified as components of the association equilibrium.

Since nonionic detergents are thought to have little influence on protein-protein interactions among membrane proteins (2), our results strongly support the view that, in the erythrocyte membrane, band 3 protein is in a monomer/dimer/tetramer association equilibrium (3). They disagree with the findings that detergent-solubilized band 3 is a stable dimer (4,5) or a mixture of stable dimers and tetramers (6).

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