Bicontinuous microemulsion (BμE) as a membrane-mimic to stabilize and enable structural studies of membrane proteins

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Bicontinuous microemulsion (BμE) phase of Winsor-III (WIII) systems formed by an aerosol-OT (AOT)/alkyl ethoxylate mixed surfactant system to stabilize membrane proteins (MP) in aqueous environment as a biomimetic analog of cell membranes. α-synuclein (ASYN) and bacteriorhodopsin (BR) MP were readily incorporated into bicontinuous microemulsions (BμEs) formed by two microemulsion systems: water/heptane/Aerosol-OT (AOT)/CK-2,13 and water/dodecane/sodium dodecyl sulfate (SDS)/1-pentanol. (CK-2,13 is an alkyl ethoxylate possessing two alkyl tail groups of carbon chain length 2 and 13 and an average degree of ethoxylation of 5.6.) MP were encapsulated in BμEs through preparation of Winsor-III systems at optimal salinity, with the anionic surfactants AOT and SDS providing the driving force for extraction. Dissolution of ASYN in BμEs greatly increased the former's α-helicity, similar to ASYN's behavior in the presence of biomembranes, while BμE- and vesicle-encapsulated BR possessed similar secondary structure. Small-angle neutron scattering (SANS) results clearly demonstrated the direct interaction of MP with the surfactants, resulting in a decrease of surface area per volume for surfactant monolayers due to decreased surfactant efficiency. The SANS signal for ASYN was isolated through the use of neutron contrast matching for the surfactants through partial deuteration of water and oil, one of the first reports of contrast matching for BμEs in the literature. The SANS results of the contrast-matched sample reflected similar aggregation for ASYN in BμEs as was reported previously for vesicles and SDS solution. Further, antimicrobial peptide melittin when encapsulated in bicontinuous microemulsions formed using three-phase (Winsor-III) systems, melittin's helicity increases greatly due to penetration into the surfactant monolayers, mimicking its behavior in biomembranes. But, the threshold melittin concentration required to achieve these trends is lower for the microemulsions. The extent of penetration was decreased when the interfacial fluidity of the microemulsions was increased. These studies together demonstrate the potential use of BμEs as MP host systems for conducting biochemical reactions such as the conversion of sunlight into adenosine triphosphate by BR and studying the fundamental behavior of MP, such as the role of ASYN dysfunction in Parkinson's disease, as well as for isolation and purification of MP via Winsor-III-based extraction.