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High-Resolution H/D Exchange Measurements to Analyze Protein Structures in Solution: A Mass Spectrometry Approach to Structure-Based Drug Design. Patricia C. Weber, K. Molnar, S. Coales, M. Southern, P. Griffin, Y. Hamuro, ExSAR, Monmouth Junction, NJ.

The solution structure of proteins can now be analyzed in segments over the entire sequence using mass spectrometry to detect amide proton hydrogen/deuterium exchange. In this talk use of H/D exchange to determine the intrinsic foldedness of proteins, protein:protein and protein:ligand interaction sites, and solvent- or ligand-dependent conformational changes will be presented. As an example, the ligand binding domain (LBD) of the peroxisome proliferator-activated receptor subtype γ (PPAR γ) nuclear receptor (NH) was studied to determine the conformational changes accompanying formation of modulator-receptor complexes. The PPAR γ system was chosen because it is a representative NH, the ligand binding properties have been studied by other structural biology methods, and well-characterized ligands are available. PPAR γ is also a diabetes drug target, and an improved model of modulator-dependent structural changes will aid discovery of new anti-diabetics and drugs to treat metabolic syndrome, a common diabetes precursor. The specific mechanism of conformational signaling where changes in LBD structure induced by the initial ligand binding event persist during subsequent formation of the multiprotein signaling complexes is a fascinating structural biology question that remains largely unanswered. The solution conformation of the LBD free and complexed with several classes of modulators was determined by high-resolution amide proton H/D exchange. The unique insights into the dynamical properties of the PPAR γ LBD in solution provided by this MS-based structural biology method will be presented, along with comparisons to data from X-ray crystallography and NMR.