**Sample Abstract – Computational & Systems Biology**

**Abstract Title:** Coevolution-Based Inference of Amino Acid Interactions Underlying Protein Function

Protein function arises from a poorly understood pattern of energetic interactions between amino acid residues. Sequence-based strategies for deducing this pattern have been proposed, but lack of benchmark data has limited experimental verification. Here, we extend deep-mutation technologies to enable measurement of many thousands of pairwise amino acid couplings in several homologs of a protein family - a deep coupling scan (DCS). The data show that cooperative interactions between residues are loaded in a sparse, evolutionarily conserved, spatially contiguous network of amino acids. The pattern of amino acid coupling is quantitatively captured in the coevolution of amino acid positions, especially as indicated by the statistical coupling analysis (SCA), providing experimental confirmation of the key tenets of this method. This work exposes the collective nature of physical constraints on protein function and clarifies its link with sequence analysis, enabling a general practical approach for understanding the structural basis for protein function.