



Use this document to frame questions for your group before you research information on MDH and CS interactions. The Introduction to Protein-Protein Interaction gives a modestly deep insight into how a protein interactions work. After this we will study how protein interactions are assayed and then study the specific interactions between MDH and CS.

First read this document, then with your group – discuss and answer each of the following questions:

- 1) Review the functional groups of each of the 20 common amino acids and look at the structure of the peptide backbone. Recognize the portions of each amino acid/protein-main chain that will interact in a hydrogen bond, polar contact, salt bridge, hydrophobic interactions. Review and recognize these now as we will be assuming you remember these along with the single and three letter abbreviations for the amino acids for the rest of the semester.
- 2) Proteins that interact require two types of complementarity. What are these two and how do you imagine that induced fit might be something to consider when looking for the interface of two proteins? Consider the kinetics and reaction coordinate as part (but not all) of your answer.
- 3) What are the differences between a transient and a permanent protein interaction? What might be the differences in forces between the two types of interacting proteins?
- 4) What is the purpose of interacting proteins in the TCA? Can you find a reason that MDH and CS might be somehow linked (aka substrate shuttling)?
- 5) In several locations in the document water is discussed – for both the free proteins, during the interaction docking process and for the proteins after docking. What energetic role does water play through these processes? Why does it matter if water is solvating the surface of a free protein and only partially found in the interface?
- 6) If you were to look at the interface of two docked proteins, what chemical features might you expect that maintain the interaction? Be specific here please.
- 7) Protein interaction is thought of in kinetic terms. What are the reactants and products of a protein pair that interact? How can you shift the equilibrium from reactant to product? Define K_d and K_a for protein interaction.
- 8) Reiterate the factors that influence (both positive and negative) protein-protein interactions.
- 9) What are the most common amino acids involved in protein-protein interactions in the interface. And how do they interact with what? What is a “hot spot”? Do amino acids only interact with other amino acids?
- 10) How would post-translational modification alter the forces maintaining an interface between two interacting proteins?
- 11) Site directed mutagenesis is a technique used to change the DNA/plasmid coding region of a protein resulting in a changed, deleted or inserted amino acid. How might you use this technique to show the importance of an amino acid in a protein-protein interaction? What exactly is alanine scanning and how is that used to define sites of interaction? Why alanine?