

Module 13 Assignment

585.751.81 Immunoengineering

1. Read the following paper on models of T cell activation:
<https://www.nature.com/articles/nri3728> at least up to “Extensions of phenotypic models” on page 623. Describe each of the five possible models for T cell activation listed in the paper (in 2-3 sentences each). Which model best describes T cell activation and why? (50 points)
The paper describes five models for T cell activation:
 - **Occupancy model:** also known as affinity model, proposes that T cell activation is proportional to the number of T-cell receptors (TCRs) occupied by peptide-MHC (pMHC) complexes. This model states that TCRs become signaling-competent immediately upon pMHC binding. This model predicts that pMHC potency (EC_{50}) correlates directly with the dissociation time and the TCR-pMHC dissociation constant (K_d) and that the maximum response (E_{max}) is independent from the binding parameters which contradicts experimental data.
 - **Kinetic proofreading model** explains how T cells discriminate between ligands based on the dissociation time of the ligand-receptor interaction. It proposes that T cell activation is proportional to the fraction of TCRs that remain bound by pMHC complexes long enough to undergo biochemical modifications, like tyrosine phosphorylation, to reach a signaling-competent state. The delay between pMHC binding and TCR signaling, enables T cells to discern between pMHC complexes based on their dissociation times from the TCR, with the prediction that longer binding times lead to greater T cell activation, which is supported by observed correlations between EC_{50} and K_d .
 - **Kinetic proofreading with limited signaling model:** both the kinetic proofreading model and serial triggering models start with the same biochemical assumptions but differ in what triggers T cell activation. In serial triggering models, each TCR can signal only once per pMHC binding event, limiting continuous activation, particularly from pMHC complexes that dissociate slowly. The kinetic proofreading with limited signaling suggests that TCRs can only signal for a limited time after they become active, necessitating multiple pMHCs to bind sequentially for sustained T cell activation. Because of this limited time for signaling, even at high concentrations of pMHCs, each complex can only activate TCRs for a short period. This results in an optimal dissociation time for effective T cell activation, where pMHCs with too long dissociation times fail to sustain activation because they remain bound to non-signaling TCRs.
 - **Kinetic proofreading with sustained signaling model** allows signaling-competent TCRs to sustain signaling even after pMHC unbinding. This sustained signaling capability is supported by experimental evidence suggesting that TCRs, along with their associated signaling complexes, can remain active until they are either dephosphorylated by phosphatases or internalized by the cell. This model modifies the dynamics of T cell activation by allowing pMHC complexes with various dissociation times to produce maximal signaling at high concentrations, without the need for serial binding, thus allowing an optimal dissociation time that varies with pMHC concentration.
 - **Kinetic proofreading with negative feedback model** is an extension of the kinetic proofreading model and introduces a mechanism to regulate the activation of TCRs through negative feedback loops involving phosphorylation. This negative feedback is



mediated by phosphatases like SHP1 and other phosphatases. Such feedback results in T cell activation showing an optimal response function to the pMHC dose, which is modulated by the dissociation time between TCR and pMHC.

2. You are studying HIV and want to identify viral epitopes that may be recognized by CD8+ T cells to kill HIV-infected CD4+ T cells. Use the following database (http://www.iedb.org/home_v3.php) to search for linear peptide epitopes from the organism Human immunodeficiency virus 1 (the more common type of the HIV virus) that bind to HLA-A*02:01 (the most common HLA-A allele in humans) in human hosts. Once you submit the search, change the linear peptide length to 9 amino acids (as most HLA molecules have a strong preference for binding 9mers) on the left hand side. Export your results, pick the first 50 epitopes in your search results and input them into the netMHC artificial neural network prediction program using the PEPTIDE format (<https://services.healthtech.dtu.dk/service.php?NetMHC-4.0>). For this problem, look at the binding affinity of your peptides to HLA-A*02:01 allele in the HLA-A species/loci. (50 points)
- Please list/provide a screenshot of the peptides that you tested in the software and list the peptides that were predicted to be strong binders to the HLA molecule (those labeled "SB"). (15 points)
 - Are there any features in common between the peptides listed as "strong binders"? If so, what are those features and why are they conserved between the peptides? If you do not see any features in common or only have 1-2 strong binding peptides, answer the question more generally: what common features would you expect to see between peptides that bind strongly to a given HLA/MHC allele? (20 points)
 - Describe (in no more than a few sentences) how netMHC could be utilized in an immunoengineering context. (15 points)

