**Homework due at start of class on July 24**

Questions and answers provided by: Jennifer Barber, Emoniel Isakharov, Kaiser Valshon

*Read*: "High-throughput Screening for Potent and Selective Inhibitors of *Plasmodium falciparum* Dihydroorotate Dehydrogenase" (PubMed ID 15795226)

*Worksheet to hand in:*

1. The researchers in this paper noted that they screened compounds that had already been shown to have drug-like properties. Starting from large compound libraries, how to researchers narrow down which might have drug-like properties? What characteristics do they look for?

   - Drug design involves hit assays of hundreds of small molecules. This can get expensive so researchers can look at small molecules that are complementary in shape and charge to the biomolecular target to which they bind. In general a drug-like compound should be able to penetrate the cell membrane, be stable at the targeted PH, have the appropriate half life, not get exported from the cell, and be stable at the appropriate temperature. If you look at table 1, all the compounds look fairly similar in structure. So if you know one compound that inhibits an enzyme target, then you can test other compounds in that class. Example: compounds 1-3 are all naphthamide derivatives.

2. What properties of malarial DHODH make it a good drug target? (Think about how it relates to human DHODH.)

   - DHODH is poorly conserved, and the malarial form is very dissimilar to the human form. Therefore, drugs that have a certain level of activity on malarial DHODH may have a very different level of activity on human DHODH. Additionally, if the human DHODH is inhibited, we have a pyrimidine salvage pathway and our DNA can continue to replicate. Malaria lacks such a pathway, and so blocking their DHODH enzyme prevents them from replicating.

3. DHODH is an enzyme target. How does the researchers' use of HEPES in their materials & methods relate to this?

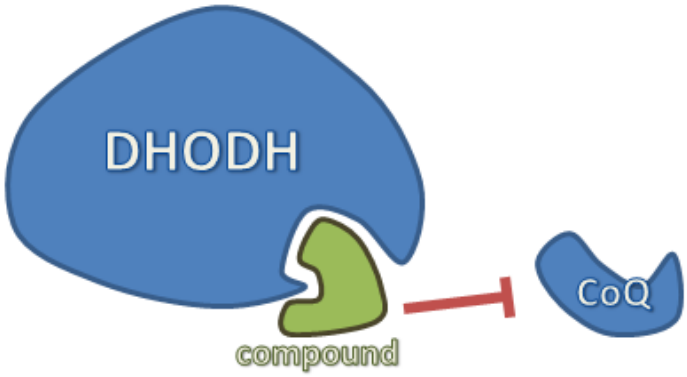
   - We basically want them to look this up and understand some of the importance behind the materials used in the drug-making process. HEPES, according to Wikipedia, is an effective buffering agent for maintaining enzyme structure and function at low temperatures. This would allow them to keep and utilize enzymes in mediums without loss of function due to non-drug effects, in order to keep confounding variables at a minimum.

4. What method did the researchers use in Fig 3 and why was it important for their results?

   - For this, we want students to touch on how these figures are based on absorbance and mass spectroscopy and how these are used to screen for inhibitors.

5. Draw a simplified picture of how the authors believe the compounds may be inhibiting their target. (You may wish to refer to Figure 4.)

   - Some depiction of competitive inhibition of CoQ binding to DHODH would be good. Example:




6. What happens to the parasite when its pyrimidine biosynthesis pathway is blocked?

   - A general answer is preferred, since we want them to get the bigger picture of what's going on with the enzyme in relation to the parasite's life cycle. Inhibiting the pyrimidine-biosynthesis pathway disrupts a crucial function in the parasite since it has no other way of making pyrimidines, which are necessary for replication, thus halting growth of the parasite.