

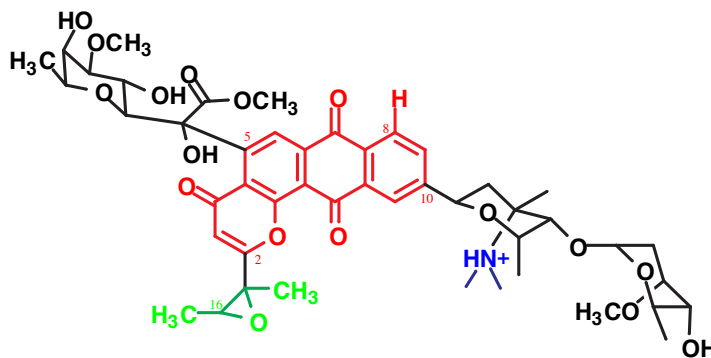
CHEM 4170

Problem Set 3 Answers

1. Why are DNA-targeted drugs only used to treat cancer and not as, say, antibacterial agents?

The chemical structure of our DNA is not significantly different from that of the DNA found in bacterial, yeast, or cancer cells. Thus, it is generally not possible to selectively target the DNA in "enemy" cells. DNA plays several central roles in the cell. Thus, agents that react with DNA tend to react with DNA in both normal cells and target cells. DNA damage in normal cells causes significant toxicity

2. Suggest how the compound shown below binds to DNA. Suggest a general binding "mode" and the specific type of weak force interactions that are like to occur.

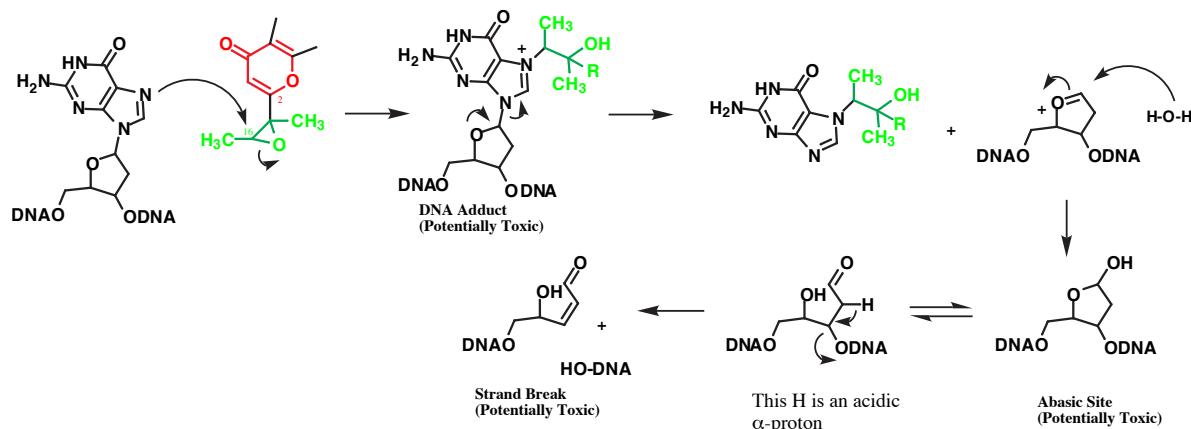


Altromycin B

The planar aromatic portion of the molecule in red will intercalate into duplex DNA. In addition, at physiologicval pH the amine residue in blue will be protonated and positively charged, and will bind to DNA via electrostatic interactions. In addition, other substituents on the sugar residues may form various hydrogen bonds with DNA.

3. Altromycin B alkylates DNA at the N7-position of 2'-deoxyguanosine in double-stranded DNA. Show this reaction and how it ultimately leads to a DNA strand break.

As we discussed in the context of affinity-labeling agents, epoxides are moderately electrophilic species that can survive in an aqueous environment, but react effectively with nucleophilic residues on biological macromolecules when they are held in close proximity to the nucleophile via noncovalent bonding. We also discussed that the N7-position of guanine is the most nucleophilic site in DNA.



4. Altromycin does *not* alkylate single-stranded DNA. Why?

As noted above, the molecule binds noncovalently to DNA. This creates a "high local concentration" of the epoxide near the DNA nucleophile. This high effective molarity increases the rate at which the epoxide reacts with the DNA nucleophile. Single-stranded DNA does not provide a structure to which the drug can bind... no binding, no high local concentration, no efficient reaction.

5. Eneidyne is able to abstract hydrogen atoms from DNA. Do a quick calculation involving bond enthalpies to help rationalize this observation. Data: C-H bond in deoxyribose is ~85 kcal/mol and a typical Ph-H bond is about 103 kcal/mol.

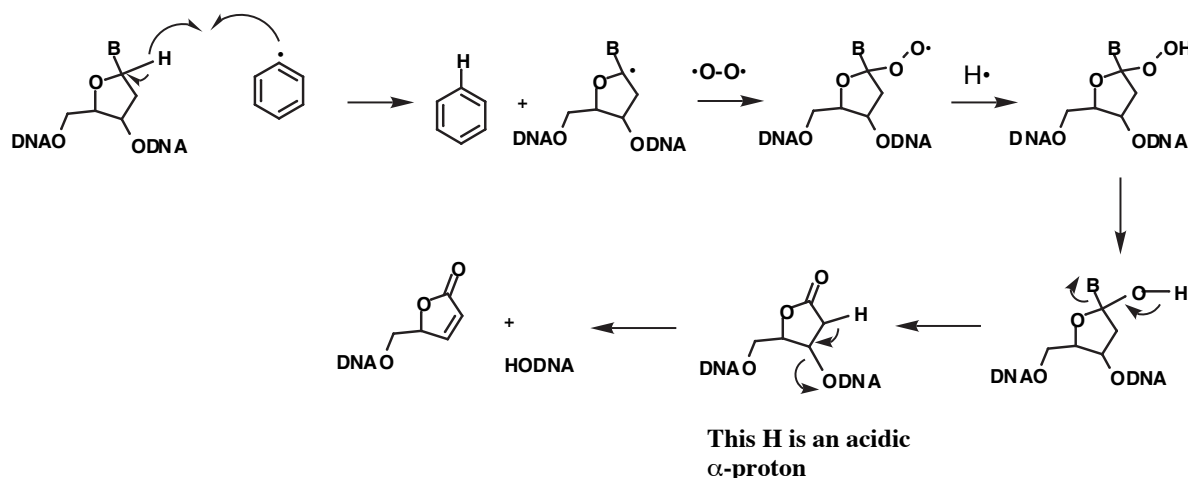
Bonds broken (costs energy) C-H 85 kcal/mol

Bonds formed (releases energy) Ph-H 103 kcal/mol

This quick calculation, estimates that the reaction is exothermic by 18 kcal/mol

6. Show how abstraction of a hydrogen atom from the 2'-deoxyribose sugar residue of DNA lead to a strand break.

This mechanism is slightly different than the one that we went through in class. Both mechanisms are correct, but the one that I show you here is probably the prevalent mechanism.



7. I gave you a paper in class that described the sequence-specific DNA-binding molecules developed by Dervan and coworkers. I have also posted a couple of papers on the website that describe this work. (a) Describe why it is possible, in principle, that these compounds could be used to treat any disease that can be treated by an enzyme inhibitor? (Unlike the typical DNA-damaging drugs mentioned in Question 1). (b) However, some medicinal chemists believe that these compounds will *never* be useful drugs. This shared by some drug experts is based upon a simple rule that is known to all CHEM 317 students. Look closely at the structure of these compounds and explain why.

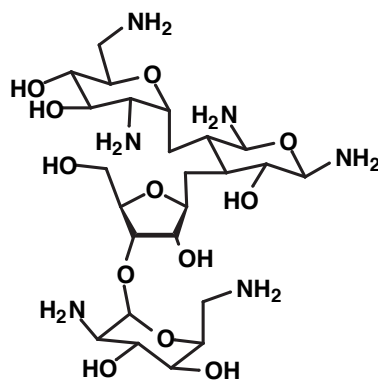
(a) Look at the handout that I gave in class. Preventing expression of a single gene should, in principle, have the same effect as blocking the action of the gene product (the protein) using a traditional small molecule drug. Importantly, Dervan's compounds do not cause non-specific DNA damage and should not cause the sort of cytotoxic or cytostatic effects shown by this type of damage.

(b) Dervan's compounds do not obey the "rule of 5's".

8. Draw a diagram that depicts the flow of information in the cell. Show the point in this flow of cellular information at which "antigene", "antisense" and "traditional" therapeutics elicit their activity. Briefly describe the antisense approach. What is the major drawback to current antisense drugs?

See the handout that I gave you in class. The diagram that I'm asking for here was shown on page one of the handout.

9. I posted two articles about RNA-small molecule interactions on the course website. Read them. The drug neomycin binds to the 16S subunit of the procaryotic ribosome and blocks bacterial protein synthesis. Electrostatic interactions are probably very important in the binding of neomycin to its target. (a) What are the specific functional group interactions that are involved here? (b) Take a look at the research paper I gave you by Tor and coworkers. I handed a copy out in class and also posted it on the course website. Neomycin contains multiple amine residues that can be protonated at pH 7. Following protonation of one amino group, it becomes more and more difficult to protonate each subsequent amine residue. Explain why.



(a) Neomycin contains multiple amine residues that can be protonated at pH 7. These cations can form electrostatic bonds with the negatively charged phosphate residues in the RNA molecules that make up the ribosome.

(b) Formation of polycations (or polyanions) is usually difficult because like charges repel each other. Thus, formation of the second, second, third and fourth ammonium cations on neomycin requires lower and lower pH values because it is not favorable to pack a lot of positive charges into such a small area in a single molecule.

10. How do the magainins punch holes in cell membranes? How do gramicidins punch holes in cell membranes?

See the handout that I gave on the last day of class. Gramicidin forms a β -helical dimer in which two of the molecules stack on each other. The gramicidin dimer has a channel down the center through which ions can flow. The magainins form channels by a different mechanism in which 6-8 of α -helices assemble into a "bundle" that places hydrophobic residues of the helices in the lipid bilayer and leaves a channel down the center of the bundle through which ions and water can flow.

11. Provide clear definitions for the following terms:

Pharmacophore - **Within a series of drugs that interact with the same biological target the pharmacophore is the smallest structural unit (the "core" fragment), common to the series, that retains significant biological activity.**

Receptor - **A protein that selectively binds to a biological signaling molecule (the autocoid). Upon binding of the autocoid, the receptor is activated and amplifies the signal.**

Autocoid - **Natural ligand for a receptor.**

Competitive Antagonist - **A molecule that blocks binding of the autocoid to a receptor by binding at the same site where the autocoid binds.**

Enzyme - **A protein that catalyzes a chemical reaction.**

Substrate - **A molecule that is converted to a new product via enzymatic catalysis.**

Competitive Inhibitor - **A molecule that blocks enzymatic conversion of its substrate to product by binding to the active site of the enzyme.**