1. Define mutagensis and discuss the difference between oncogenes and tumor suppressor genes. Provide at least two examples of each.

Mutagenesis is a change or alteration in genes. Oncogenes will promote cancer and may become activated when mutated. Tumor suppressor genes inhibit cancer formation and may be inactivated when mutated. Example oncogenes are: bcr-abl, bcl-2, ret, HER2, N-myc and c-Myc. Example tumor suppressors are: APC, BRCA-1, BRCA-2, CDKN2A, VHL, p53 and pRb.

2. Discuss drug resistance in the context of cancer chemotherapy, focusing on how it occurs and the effect it can have on the chemotherapy regimen.

Drug resistance in cancer chemotherapy arises because the cancer cells within any tumor are not truly identical. Therefore, cells will have different levels of sensitivity to any given drug. The most sensitive cells will die first while more resistant cells will survive and proliferate in a process called clonal selection. The chemotherapy regimen will then have to be changed to reflect the new resistance of the majority of the cancer cells to a given drug. Most of the time, chemo drugs are dosed in combination in an attempt to prevent large amounts of resistance from forming.

3. What are the major types of genetic mutation that can lead to formation of cancer? What specific type of mutation is responsible for the formation of the Philadelphia chromosome?

The major types of mutation are point mutations, changes in gene expression (most likely due to mutations in promoter regions), chromosomal rearrangement and inherited mutations. The Philadelphia chromosome is formed from a chromosomal rearrangement.

4. Please draw the structure of cisplatin below. Why should cisplatin only be reconstituted with saline?

Cisplatin undergoes an aquation step where the chlorine atoms are replaced with water molecules. This process will occur spontaneously in low ionic strength solutions such as water or the inside of cells. Once the water has liganded to the platinum atom, the molecule has an overall positive charge which will prevent it from passing through lipid membranes. Therefore, cisplatin must be reconstituted in saline (at high salt concentrations) to prevent premature aquation and allow the best amount of cell membrane permeability.
5. What is the reactive portion of busulfan? What is the mechanism of action?

Busulfan has a very good leaving group via the sulfur moiety which activates the main-chain carbon for nucleophilic attack by DNA. This attack and loss of the sulfur group is shown below. Be aware that because the molecule is symmetric, attack can occur at both ends.

![Busulfan structure diagram]

6. Dacarbazine is a prodrug used to treat Hodgkin’s lymphoma and must be activated initially by Cytochrome P450s. After O-dealkylation, a tautomerization step occurs which activates the molecule for nucleophilic attack. On the pathway below, please draw the reactive form of the molecule and indicate the driving force for the reaction.

![Dacarbazine structure diagram]

The driving force for the nucleophilic attack by DNA is the release of nitrogen gas. \(N_2\) is a very stable molecule and will dissociate quickly to help push the reaction to completion as well.
7. Cyclophosphamide is an important cancer prodrug. Name the class of chemotherapy agents to which it belongs. Explain how the prodrug activation changes the reactivity of the molecule. Show the initial product of the prodrug activation pathway.

Cyclophosphamide is an alkylating agent. The phosphoramide is electron withdrawing so the tertiary nitrogen cannot form the aziridine initially. After P450-mediated hydroxylation at the carbon shown below, subsequent tautomerization and beta-elimination will result in activation of the tertiary nitrogen which can then form the aziridine ring and covalently adduct molecules.

![Diagram of cyclophosphamide activation](image)

8. What type of cancers are nitrosoureas used for commonly? Draw the chemically active portion of the molecule below.

Nitrosoureas are used to treat brain cancers commonly due to their large amounts of CNS penetration.

![Diagram of nitrosoureas](image)

9. 5-FU is an important cancer drug. Name the class of chemotherapy agents to which it belongs and the enzyme that 5-FU inhibits. Also explain how the compound must be activated before inhibiting cancer cells.

5-FU is part of the class of drugs called antimetabolites and it inhibits thymidylate synthetase. This enzyme is used in the formation of the nucleic acids so 5-FU must be phosphorylated before being able to bind to the enzyme active site.

10. What class of cancer drugs does gemcitabine belong to and what type of cancer is it commonly used to treat? Additionally, it has two mechanisms of action. Describe both. Why does it have such a long half-life compared to similar drugs such as cytarabine?

Gemcitabine is an antimetabolite used to treat pancreatic cancer. It acts by inhibiting ribonucleotide reductase and the phosphorylated form will also compete with other nucleic acids for DNA incorporation. The reason that the half-life is so long is that gemcitabine also inhibits the main enzyme responsible for its metabolism (cytidine deaminase).
11. Despite having the same intracellular target, vincristine and paclitaxel have different mechanisms of action. Explain both mechanisms of action and how they differ as well as any important toxicological differences between the drugs.

Both of these drugs target microtubules to inhibit proper tubulin function during mitosis however vincristine inhibits tubulin polymerization while paclitaxel causes over-stabilization of tubulin polymers. It is important to note that paclitaxel has a major toxicity with myelosuppression which is not as large an issue with vincristine.

12. What is the name of the drug shown below? Draw a dashed line showing what bond is broken to form the active metabolite. Also explain the metabolism and clearance of the active metabolite and any potential hazards.

The drug is irinotecan and is converted to the following metabolite (SN-38). The metabolite is glucuronidated by UGT1A1 which is polymorphic. In patients with low levels of the enzyme, the AUC of the metabolite for a given dose can be much higher which may lead to toxicity if the dosing regimen is not reduced.