

Name \_\_\_\_\_

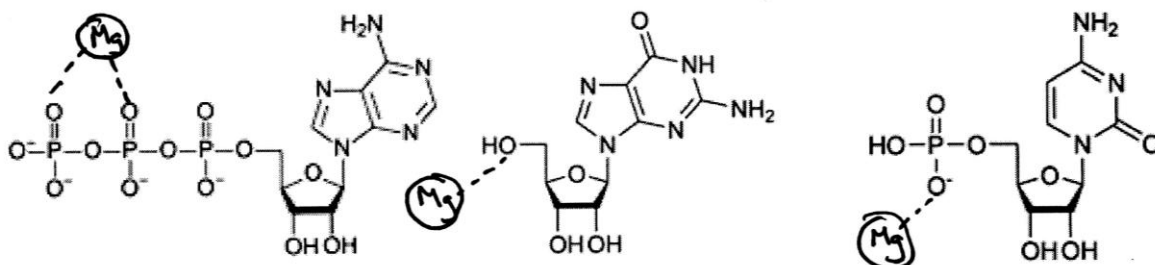
(12) **1.** Calcium levels in cells need to be regulated but also need to change rapidly in response to different stimuli. Describe how **three different** systems function to transport calcium across membranes.

1. Calcium-dependent ATPase transport systems. Relies on the hydrolysis of ATP to generate a proton gradient to import calcium into cells or into the ER.
2. Receptor mediated calcium channels. Uses the binding of an external signal molecule at an associated receptor to open a calcium channel for import into cells or export from the ER.
3. Sodium/calcium exchanges. Uses the uptake of 3 sodium ions to generate an ion gradient to export a calcium ion from the cell or the uptake of 2 sodium ions from the cell to export a calcium ion from the mitochondria.
4. Voltage sensitive calcium channels. Uses a charge difference to open a channel for calcium import into cells or into the mitochondria.

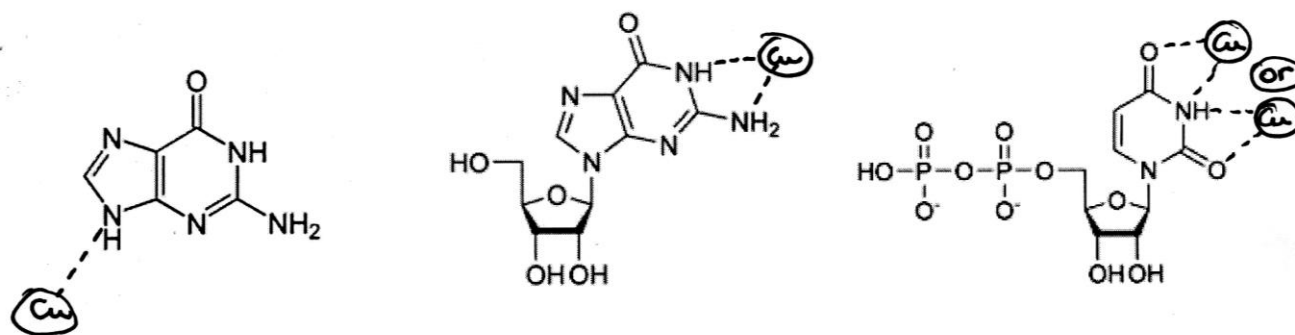
(18) 2. For each of the following compounds:

- (a) draw the most likely interactions that will occur between these compounds and  $Mg^{2+}$  and  $Cu^{2+}$ .

Mg complexes



Cu complexes



- (b) For any one of your complexes describe the features of the metal and ligand that make this the preferred binding mode.

*Mg(II) is a hard metal with a preference for hard donor atoms. Coordination to the  $\beta$ - and  $\gamma$ -phosphate oxygens of ATP provides a stable 6-membered chelate ring.*

*Cu (II) is a borderline transition metal with a strong preference for nitrogen donor atoms. N9 of the adenine ring has the lowest  $pK_a$  for proton displacement and either O2 or O4 of the uridine ring can help form a bidentate interaction.*

(20) 3. Chelation therapy is used for the treatment of acute iron poisoning.

(a) Describe three important properties of an ideal iron chelator and indicate why each property is important.

1. *selectivity for iron – minimize competition from other metal ions and avoid removal of essential metal ions during therapy*
2. *stable and excreted – does not break down to toxic by-products and does not release the bound iron prior to excretion*
3. *inexpensive and orally administered – Chelation therapy required a very high concentration of the compound to be administered*
4. *low inherent toxicity – since large quantities are needed the chelator must have very low toxicity*

(b) What causes most potent iron chelators to fail in human trials and suggest how improved chelators can be produced?

*Most human trials fail either because the chelator is toxic at the high levels needed or the chelator is ineffective at binding and competing with other binding sites in the body for removal of iron.*

*Stable compounds are needed that cannot be metabolized before excretion.*

*Designing compounds with high affinity iron sites is necessary, but knowledge of the physiological iron binding sites should be used to test new chelators for their ability to compete for iron binding.*

(20) 4. Provide short answers for **five and only five** of the questions listed below:

(a) How is iron uptake regulated in mammals?

*Iron regulatory proteins (IRPs) are the key iron sensors in mammalian cells. During low iron levels IRPs repress proteins that require iron for their function and stabilize the mRNA that encodes for transferrin to increase iron uptake. During high iron levels IRPs acquire an Fe/S cluster and no longer function as a repressor.*

(b) What are the most important properties of a good MRI contrast agent?

*MRI contrast agents work through the presence of unpaired electrons that cause a rapid relaxation of water protons. The best MRI agents contain binding sites with high affinities for transition or lanthanide metal ions with unpaired electrons as well as open ligand positions to interact with and relax water molecules.*

(c) How does bleomycin function as an anti-cancer drug?

*Bleomycin contains a planar bithiazole ring structure that can intercalate into DNA. An Fe (III) binding site on bleomycin provides a place for oxygen to bind and to generate superoxide and hydroxyl radicals that will cleave DNA.*

(d) How does calcium binding alter the target selection for calmodulin?

*The binding of two Ca(II) ions induces a conformational change in calmodulin that causes it to wrap around a specific helical segment in a set of target proteins leading to their activation. The binding of a second pair of Ca(II) ions causes an additional conformational change and leads to recognition of a different set of target proteins.*

(e) How does the release of excess zinc lead to cell death?

*High zinc inhibits glycolysis causing depletion of ATP leading to cell necrosis  
Zinc levels also activate protein kinases leading to oxidative stress  
Zinc can also cause activation of caspases which triggers apoptosis*

(f) Describe the role of metals in molybdenum cofactor assembly.

*Mg(II) is required to complex to ATP, the substrate for activation of several intermediates in cofactor assembly  
Cu(II) is required as a cofactor for several oxidative enzymes during maturation of the molybdenum cofactor  
Fe(III) is required as part of an iron/sulfur cluster used to convert GTP to a precursor molecule*

(g) How does cadherin function in cell adhesion?

*Proteolytic activation of cadherin leads to production of an N-terminal segment with a tryptophan at position 2 that inserts into a hydrophobic binding pocket of a cadherin molecule from an adjacent cell. This interaction is stabilized by a hydrogen-bond to the terminal amino group thus linking adjacent cells.*

(30) 5. Pick the response(s) that best match the characteristics described and list the corresponding letter(s) in the space provided. Some of these responses may be the correct answer for more than one question and some may not be the correct response for any question.

human copper transport protein	<u>G, R</u>	A. aconitase
primary calcium regulatory protein	<u>D</u>	B. cadherin
human zinc export protein	<u>F</u>	C. calcineurin
target for immunosuppressive therapy	<u>C</u>	D. calmodulin
mammalian iron transport protein	<u>Q</u>	E. calsequestrin
iron export protein	<u>G</u>	F. CDF protein
copper export protein	<u>O</u>	G. ceruloplasmin
calcium storage protein	<u>E</u>	H. cytochrome oxidase
iron storage protein	<u>K</u>	I. enterobactin
iron uptake protein in mammals	<u>Q</u>	J. ferrichrome
releases calcium during muscle contraction	<u>E</u>	K. ferritin
major target for copper transport	<u>H, P</u>	L. hemoglobin
detoxification protein for soft metals	<u>O</u>	M. hemosiderin
primary target for iron transport in humans	<u>L</u>	N. hephaestin
		O. metallothionein
		P. superoxide dismutase
		Q. transferrin
		R. transcuprein
		S. ZIP protein