UNIVERSITY OF TORONTO

FACULTY OF ARTS AND SCIENCE

BIOCHEMISTRY 210H1F

December Examinations 2005

Duration: 3 hours

Examiner: Dr. R. Baker

Candidates may use simple, non-programmable calculators (supplied by candidates).

This examination is worth 50% of the final grade for the course. There are 25 multiple choice questions worth 1.2 marks each for a total of 30 marks and 5 short answer questions, worth a total of 20 marks.

For the multiple choice questions you are to choose ONE answer for each question and fill in the appropriate circle on the computer card with a soft lead pencil (**not pen!**). Marks are not deducted for wrong choices but no marks are given if more than one circle is filled in per question.

*****CAREFULLY fill in the appropriate circles on the computer card for your name and student number WITH PENCIL. Please also print your name and number on the card.**

We cannot help you if you make transcriptional errors. It is thus not a good idea to leave entering your answers on the card to the last few minutes of the exam.

When not entering answers on your card you should cover the answer card with your question paper.

****Please note that the examiners make every effort to ensure there is only one suitable answer for each multiple choice question. However if you are convinced that a particular question has two answers, you should select the answer that seems MOST appropriate.**

Answer all the multiple choice questions and all the short answer questions.
Place your short answers in the examination booklet provided.
Please print your name and student number on your examination booklet.

The Assignment (15% of the final grade for the course) is located at the end of this exam. Please use a separate booklet for the Assignment.

Multiple Choice Questions (30 Marks)

1. Which ONE of the following is **INCORRECT**?

a) Trypsin is a hydrolase which can break a peptide bond within a protein on the carboxyl side of an arginine residue, because of the binding of the arginine side chain within a pocket at the active site.

b) Alcohol dehydrogenase is an oxidoreductase that has specificity for a number of alcohols, and when this enzyme uses ethanol as substrate and NAD^+ as coenzyme the enzyme will produce the aldehyde $H_2C{=}O$ and $NADH + H^+$.

c) Hexokinase is a transferase, which can utilize a number of hexose substrates including α-D-glucose, α-D-mannose and α-D-fructose (which is a ketohexose in its straight chain form).

d) During the breakdown of glycogen in liver, phosphoglucomutase, an isomerase, produces a reducing sugar that can enter glycolysis.

2. Which ONE of the following concerning the active site is **INCORRECT**?

a) The active sites of RNase A and chymotrypsin can handle macromolecules as substrates by binding specific components found within the large substrate.

b) If hexokinase did not show its characteristic change in conformation after the binding of α-D-glucose, likely ATP would be hydrolyzed to ADP + inorganic phosphate + H^+ at the active site during the enzyme reaction.

c) The active site of an enzyme usually makes up a small part of the total volume of the enzyme and the active site can bind substrates or competitive inhibitors.

d) ASA is an irreversible inhibitor of the enzyme COX, and ASA promotes an acetylation reaction that chemically alters the arachidonate binding site in the enzyme.

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3. Which ONE of the following concerning enzyme regulation is **INCORRECT**?

a) The metabolic pathway for the production of isoleucine from threonine is controlled at the first enzyme in the path by an allosteric inhibitor in a process known as feedback inhibition.

b) If activation of trypsinogen results in the release of the hexapeptide: $^{+}NH_3$-Lys-$(Asp)_4$-Val-COO^{-}, this indicates the removal of this peptide from the N-terminus of trypsinogen during activation in the duodenum by the enzyme trypsin.

c) The inhibition of E_1 in the pyruvate dehydrogenase (PDH) complex is an example of covalent modification that can be modulated allosterically.

d) The covalent modification of glycogen synthase results in an inactive phosphorylated form of this enzyme which can be reactivated by the action of protein phosphatase 1.

4. The reaction below is catalyzed by lactate dehydrogenase (LDH)

$$CH_3\text{-}\underset{\underset{O}{\|}}{C}\text{-}COO^{-} + NADH + H^{+} \xrightarrow{LDH} CH_3\text{-}CHOH\text{-}COO^{-} + NAD^{+}$$

This reaction can be studied because NADH shows an absorption of ultraviolet light at a wavelength of 340 nm (while NAD^{+} shows no absorbance at this wavelength). Thus you can follow the reaction by measuring light absorption at 340 nm in a test tube or cuvet. This shows the loss of NADH with time.

Which ONE of the following concerning this assay of the LDH reaction is **INCORRECT**?

a) The reaction, noted above, shows a conversion of the substrate pyruvate into the product lactate.

b) If pH and temperature are optimal and the concentrations of substrate and coenzyme are sufficient, there should be a linear loss of NADH with time, if the concentration of enzyme is not too high.

c) Increasing the substrate concentration should increase the enzyme rate observed, so that as [ES] maximizes, kcat [ES] will approach a maximal value called Vmax.

d) Increasing the concentration of enzyme in the reaction will likely affect K_M, which is defined in terms of the reaction kinetics shown in question 5 as $(k_2 + k_3)/k_1$

5. The Michaelis-Menten equation was derived from the enzyme reaction:

$$E + S \underset{k_2}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_3} E + P$$

Following the derivation of the Michaelis-Menten equation, which ONE of the following **cannot** be derived from the steady state equation? (E_T is total enzyme)

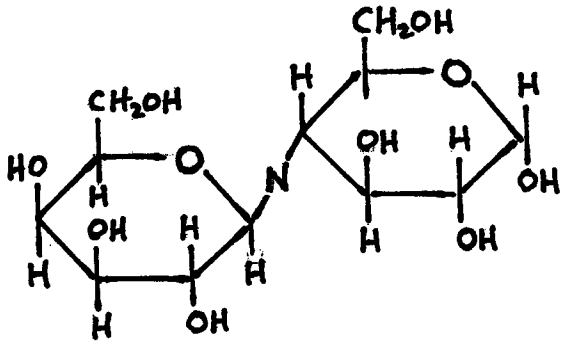
a) $[E] = [ES]\, K_M/[S]$

b) $[S] = K_M\, [ES]/[E]$

c) $[S] = \dfrac{K_M}{k_3\, [E_T] + v}$

d) $[ES] = \dfrac{[E_T]\,[S]}{[S] + K_M}$

6. In studies of the intestinal enzyme lactase an inhibitor I, shown below, was used:



Which ONE of the following is **INCORRECT**?

a) If I is added to lactase, increasing concentrations of lactose should ultimately allow the reaction to approach closely the same maximum velocity that is seen with this enzyme in the absence of I.

b) In the Lineweaver-Burk plot the value for the negative x-axis intercept should be larger in the presence of I, compared with the intercept in the uninhibited reaction.

c) In the Lineweaver-Burk plot the value for the y axis intercept should be the same for both the inhibited and the uninhibited enzyme reaction.

d) If lactase is first incubated with a high concentration of lactose, the subsequent addition of I (in small concentration) should then have little effect on the enzyme activity.

7. Which ONE of the following matchups is **INCORRECT**?

a) Action of di-isopropyl fluorophospate on acetylcholinesterase: irreversible inhibition

b) Action of α-D-glucose-6-phosphate on hexokinase: allosteric product inhibition

c) Action of CTP on aspartate transcarbamoylase: allosteric feedback inhibition

d) Action of AMP on phosphofructokinase 1: non-competitive inhibition

8. Which ONE of the following concerning Ribonuclease A (RNase A) is **INCORRECT**?

a) Given that the enzyme can be denatured in the presence of urea, you would expect that RNase A would be more stable than RNase S in the presence of urea, as measured by the hydrolytic rates of the two enzymes.

b) If the short RNA substrate 5'AAGACAGA3', is hydrolyzed by RNase A, you would expect the formation of two products, with the larger product having a 3' cytidine monophosphate terminus.

c) If single stranded DNA binds to the active site of RNase A, the enzyme mechanism will be blocked because the 2',3' cyclic nucleoside monophosphate intermediate formed using the DNA substrate will not interact with water.

d) If a molecule of RNase A is chemically modified using iodoacetate and then converted into RNase S, either His 119 will be modified in the component S protein or His 12 will be modified in the component S peptide.

9. If water is not available to take part in the chymotrypsin reaction, which ONE of the following would **most** likely occur?

a) The carboxylate product of the reaction would accumulate at the active site.

b) The first tetrahedral intermediate would be unable to access the oxyanion hole.

c) The acyl-enzyme intermediate would accumulate.

d) The binding of the side chain of Phe, Tyr or Trp residues at the active site of chymotrypsin would be prevented.

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10. Which ONE of the following is INCORRECT?

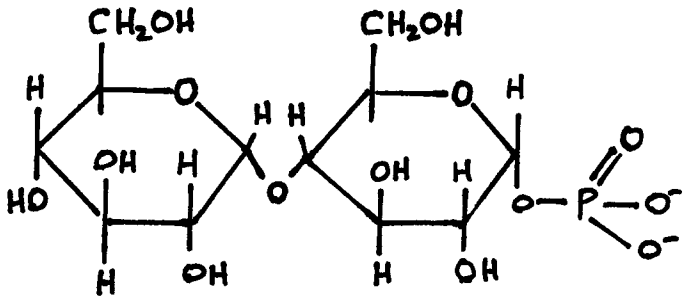
a) The cyclization of D-glucose creates two possible anomers (hemiacetals) which can be distinguished by the orientation of the -OH group above or below the pyran ring at the anomeric carbon.

b) The enzymatic hydrolysis of the α (1→4) glycosidic linkage of β-maltose will produce two monosaccharides, both of which are reducing sugars.

c) If, because of a rare mutation, a patient has inactive maltase, it follows that after a heavy starch meal, she/he may experience gas and intestinal bloating, as well as diarrhea.

d) β-D-Fructose-1,6-bisphosphate is the substrate for the glycolytic enzyme aldolase which creates two triose phosphate products (both are non-reducing sugars) by hydrolytic cleavage of the hemiketal substrate.

11. Which ONE of the following is **CORRECT** concerning the compound X shown below?



a) X is a reducing sugar and has an α (1→4) glycosidic linkage joining two hexoses in pyranose configuration.

b) X is produced during the attack of phosphorylase a on glycogen.

c) X is composed of α-D-galactose in glycosidic linkage with the monosaccharide product of glycogen phosphorylase a

d) The complete hydrolysis of 100 grams of X to monosaccharides will require 5.3 g of water. (C =12, H=1, O=16)

12. Which ONE of the following concerning lipids is **INCORRECT**?

a) Tripalmitin is a neutral glyceride with low polarity, has a fatty acid/glycerol molar ratio of 3:1, contains non-essential fatty acids and is a common component of saturated animal fat.

b) As the polyunsaturated fatty acid linoleic acid is a metabolic precursor of arachidonic acid, it follows that consuming most of your fat calories from corn, soybean, sunflower or safflower oils, is not considered a wise dietary decision.

c) 1,2-diacylglycerol is a neutral glyceride with one free -OH group and it can activate a protein kinase known as PKA found in platelets and liver cells.

d) Although dietary cholesterol is now not considered a major source of the cholesterol found in LDL (or oxidized LDL), dairy products and red meat are still considered unhealthy dietary choices.

13. Which ONE of the following is **INCORRECT**?

a) The outer shell of chylomicrons consists of a bilayer of phospholipid and cholesterol, with a structure similar to lipid bilayers found in phospholipid liposomes and biological membranes.

b) Cholesterol and its steroid derivatives usually have a common cyclic structure consisting of 3 six member carbon rings and 1 five member carbon ring.

c) Pancreatic lipase breaks down dietary triacylglycerols with the assistance of amphipathic bile salts (such as the salt of cholic acid), generating two molecules of fatty acids per triacylglycerol and one molecule of 2-monoacylglycerol.

d) The cells lining the duodenum (enterocytes) take up the released fatty acids and 2-monoacylglycerol from triacylglycerol digestion and use them to regenerate triacylglycerols that are subsequently packaged in lipoproteins known as chylomicrons.

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14. Which ONE of the following concerning LDL is **INCORRECT**?

a) As both LDL and VLDL have apoprotein B-100, this is evidence that LDL is derived from VLDL following the attack of lipoprotein lipase.

b) LDL is enriched in cholesterol esters, because the hydrolysis of triacylglycerols in VLDL and IDL leaves cholesterol esters as the principal lipids of the small LDL.

c) Lipoprotein lipase is a hydrolase, and with the help of the apoprotein apo C-II on the surface of chylomicrons and VLDL, can hydrolyze triacylglycerol to glycerol and free fatty acids.

d) Elevated serum cholesterol values are principally due to elevations in serum LDL, and this elevated LDL level is caused by the inability of lipoprotein lipase to completely hydrolyse triacylglycerols in LDL.

15. Considering Nat, the patient we discussed who suffered a TIA, which ONE of the following is **INCORRECT**?

a) Nat was not immediately given ASA when he arrived at the Emergency Room, mainly because the physician (Dr Nimmo) was concerned that Nat would develop gastric bleeding.

b) An atherosclerotic plaque was found in one of his carotid arteries and was quite likely caused by the entry of LDL, oxidized by oxidants in cigarette smoke, into the artery wall.

c) The TIA itself was likely caused by an embolus that broke away from a larger platelet-containing thrombus formed at the surface of the atherosclerotic plaque.

d) Activated platelets release arachidonic acid (because of phospholipase A_2 attack) and this 20:4 is converted into the eicosanoid TXA_2 which can stimulate other platelets via their thromboxane receptor.

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16. Which ONE of the following is **INCORRECT**?

a) If the $\Delta G^{o'}$ value for the triose phosphate isomerase reaction, using dihydroxyacetone phosphate as substrate, is 7.6 kJ/mol, at equilibrium under standard conditions the concentration ratio of [dihydroxyacetone phosphate]/ [D-glyceraldehyde-3-phosphate] is less than 0.1.

b) The hydrolysis of ATP to form ADP, inorganic phosphate and H^+ is a reaction that has a positive ΔS value and a negative ΔH value.

c) If the formation of ATP in the reaction catalyzed by creatine kinase has a K'eq value of 191, this indicates that the reaction has a $\Delta G^{o'}$ value of -13 kJ/mol.

d) If an enzyme could catalyze the following reaction:
phosphoenolpyruvate + creatine $\rightarrow$ pyruvate + phosphocreatine
under standard conditions the $\Delta G^{o'}$ value for this reaction should be negative with a K'eq value less than 1000.

17. Which ONE of the following reactions in glycolysis does **not** produce a compound with a phosphate group that has a $\Delta G^{o'}$ of hydrolysis that equals or exceeds -30 kJ/mol.

a) Enolase

b) Glyceraldehyde-3-phosphate dehydrogenase

c) Phosphofructokinase-1

d) Pyruvate kinase

18. Which ONE of the following, comparing aerobic glycolysis with fermentation (anaerobic) is INCORRECT?

a) While the fermentation (anaerobic) pathway has the lyase pyruvate decarboxylase, there is no lyase found in the aerobic glycolysis pathway.

b) Fermentation (anaerobic) does not give a net production of NADH, as this reduced coenzyme is used in the conversion of acetaldehyde to ethanol.

c) Both fermentation (anaerobic) and aerobic glycolysis have at least one oxidoreductase.

d) Pyruvate decarboxylase likely is a more favourable reaction, under standard conditions, than is phosphoglycerate mutase (using 3-phosphoglycerate as substrate).