Student Name : ______KEY_________ 2014-03-28

Student ID : ___________________________

Instructions:

Write neatly and clearly. Cross out with a single line any material you do not wish to have marked. Marks will be deducted for incorrect statements. Students must work independently and may not knowingly utilize resource materials or share resource materials with other students. Students may use pens, pencils, erasers and calculators only.

Electronic devices including cell phones, personal information managers and audio devices are prohibited.

Question	**Mark**	**Total Marks**
1		10
2		~~11~~ 9
3		8
4		9
5		5
6		9
7		11
Total		**~~63~~ 61**

1 – In the space provided, give a unique definition or description of the following biochemical terms and phrases: (10 marks)

(a) conservative mutation – a mutation to a residue with similar physiochemical properties. Less stringent functional role.

(b) primary transcript – RNA produced by DNA-dependent RNA polymerase that lacks post-transcriptional modifications.

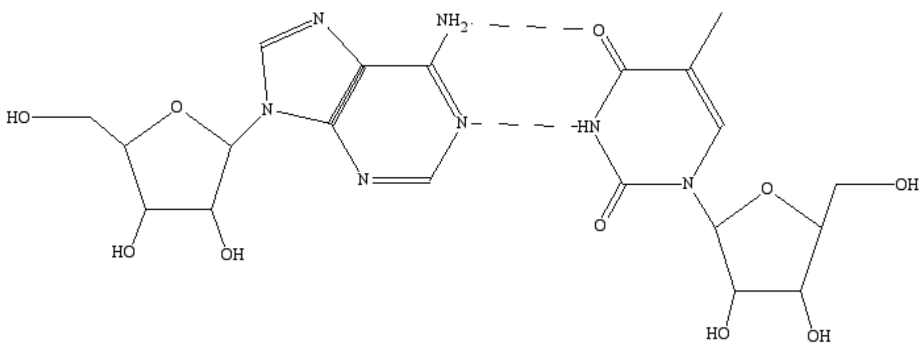
(c) hydrophobic effect – tendency of water to minimize contact with non-polar molecules

(d) homotropic activator – Substrate as effector molecule that increases activity or affinity.

(e) oligopeptide – a short polymer of up to ~20 residues connected by peptide bonds

2 – (2) Draw the following structures: (~~11~~ 9 marks)

(a) A stick diagram of a Watson-Crick AU base pair between ribonucleosides.

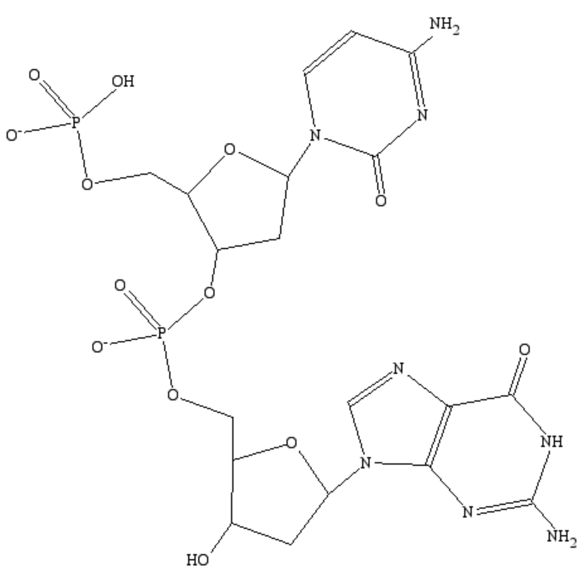


4 marks – 1 for each base

1 for ribonucleoside

1 for H-bonds

(b) A stick diagram of the deoxyribonucleotide: 5'-cytidyl-3',5'-guanylic acid.



5 marks

1 each for bases, 1 each for phosphate/connection, 1 for deoxyribose

3 – Fill in the blanks in the following statements concerning Michaelis-Menten enzymes: (8 marks)

(a) When the rate constant __k_2_ is very small compared to the rate constant k_{-1}, _K_M_ is a measure of the substrates binding affinity.

(b) When the rate constant k_2 is large compared to the rate constant _k_1_, the enzyme's efficiency is _greatest_.

(c) The _Steady-State_ assumption states that the concentration of the _Enzyme-Substrate Complex_ remains constant during initial kinetics experiments.

(d) The rates of enzyme catalyzed reactions are _independent_ of substrate concentration at high _substrate concentration_ .

4 - Michaelis-Menten Kinetics (9 marks):

Consider a Michaelis-Menten enzyme with $k_{cat} = 20\ s^{-1}$, $V_{max} = 0.01\ \mu M\ s^{-1}$ and $K_M = 0.2\ \mu M$.

(a) What is the initial velocity of the enzyme at a substrate concentration of 0.04 µM?

$v_o = (V_{max}\,[S]) / (K_M + [S]) = (0.01\ \mu M\ s^{-1})\,(0.04\ \mu M) / (0.2\ \mu M + 0.04\ \mu M) = 1.7\ nM\ s^{-1}$

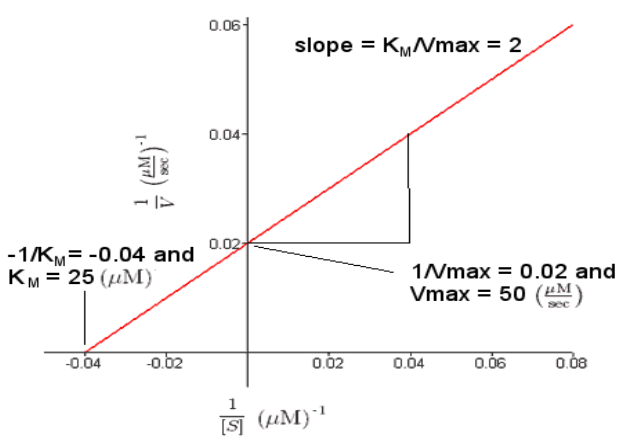
1 each for equation and answer

(b) What fraction of the enzyme binding sites are filled at an initial velocity of 6 nM s^{-1}?

All sites are filled (100%) when the enzyme's velocity is V_{max} (10 nM s^{-1}). At 6 nM s^{-1}, the enzyme is at 60% of its maximum velocity and 60% of the enzyme binding sites are filled.

1 each for equation and answer

(c) Derive the kinetic parameters V_{max} and K_M from the Lineweaver Burk plot below. Clearly indicate how the plot is used to determine these parameters.



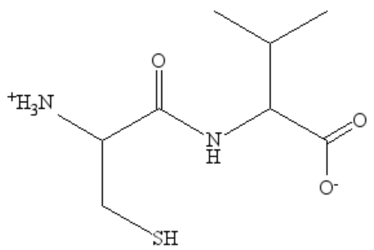
1 mark for each kinetic parameter identified and determined to max of 5 marks

5 – Consider the formation of the dipeptide Cys-Val from individual amino acids. (5 marks)

(a) Write a complete (overall) reaction equation for formation of the Cys-Val dipeptide.

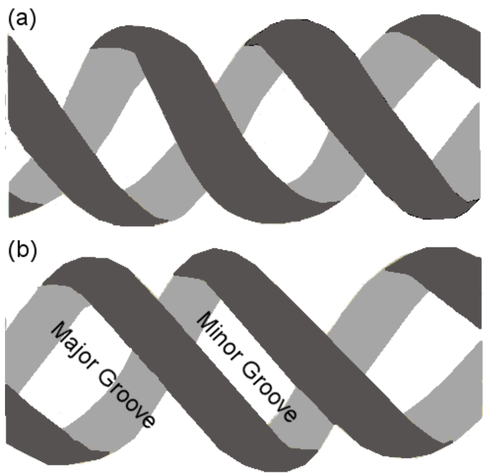
Cys + Val ↔ Cys-Val + H_2O (1 mark)

(b) Draw a stick diagram of the Cys-Val dipeptide at physiological pH.



1 mark – each sidechain, backbone and charges

6 – Consider the following backbone representations of two different dsDNA: (9 marks)



(a) Identify the conformation or name of each of dsDNAs in the figure to the left.

Panel A – A DNA 1 mark each

Panel B – B DNA

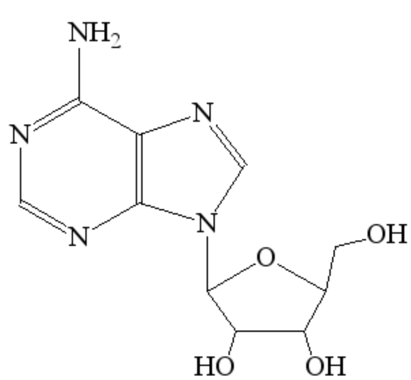
(b) Identify with an arrow both the major and minor grooves of the dsDNA shown in panel (b).

1 mark each

(c) How does the relative UV-absorbance of dsDNA change upon denaturation?

It increases at all wavelengths 1 mark

(d) Draw a stick diagram of any common ribonucleoside with the glycosidic bond in the *anti* conformation. Is this the lowest or highest energy conformation?



Lowest energy conformation.

1 mark – base, ribose, nucleoside, anti, lowest

7 – Answer the following questions regarding bacterial transcription: (11 marks)

(a) What are the roles of the σ^{70} polypeptide subunit of DNA-dependent RNA polymerase in procaryotic transcription?
- prevents DNA-dependent RNA polymerase from binding tightly to non-promoter DNA
- allows DNA-dependent RNA polymerase to bind tightly to promoter DNA
- dissociates from DNA-dependent RNA polymerase once initiation is complete and elongation begins

(b) Identify the conserved regions of the bacterial promoter.
- weakly conserved CAT sequence at transcription start site
- TATA (or Pribnow or -10 box) sequence involved in polymerase binding
- TTGACA (or -35 box) sequence involved in polymerase binding

(c) Bacterial transcription is rapid, yet error-prone. How do bacterial cells survive (*ie*. why are transcriptional errors not a problem)?
- multiple transcripts are produced so a correct copy is present
- mutations do not always change protein primary sequence
- mutations do not always affect protein function

(d) Complete the reaction below for extending an RNA by one C during transcription elongation by providing names (or abbreviations) for the missing substrate and product.

$(RNA)_n$ + _CTP_ → $(RNA)_{n+1}$ + _PP_i_

1 mark for each bullet point or blank