

**CORK INSTITUTE OF TECHNOLOGY**  
**INSTITIÚID TEICNEOLAÍOCHTA CHORCAÍ**

**Autumn Examinations 2015/2016**

**Module Title: Applied Enzymology**

**Module Code:** BIOL7001

**School:** Science

**Programme Title:** Bachelor of Science in Applied Biosciences and Biotechnology  
Bachelor of Science (Honours) in Pharmaceutical Biotechnology  
Bachelor of Science (Honours) in Herbal Science

**Programme Code:** SBIBI\_7\_Y3  
SHERB\_8\_Y3  
SPHBI\_8\_Y3

**External Examiner(s):** Dr. Brendan O Donnell  
**Internal Examiner(s):** Dr. Fiona O Halloran

**Instructions:** Answer Section A (compulsory) and TWO questions from Section B.

**Duration:** 2 Hours

**Sitting:** Autumn 2016

**Requirements for this examination:** Scientific calculator, graph paper

**Note to Candidates:** Please check the Programme Title and the Module Title to ensure that you have received the correct examination paper. If in doubt please contact an Invigilator.

## Section A – compulsory (40 Marks)

Q1.

(a) The kinetics of an enzyme was measured as a function of substrate concentration in the presence and absence of an inhibitor ( $I_1$ ). Using the data provided in Table 1:

- (i) Draw a Lineweaver-Burke plot of the results (15 Marks)
- (ii) Estimate  $K_m$  and  $V_{max}$  in the presence and absence of the inhibitor (10 Marks)
- (iii) Indicate the nature of inhibition and calculate  $K_i$  for the inhibitor (10 Marks)

**Table 1: Initial velocities calculated at varying substrate concentrations for the enzyme catalyzed reaction**

Substrate [S] mM	Vo ( $\mu\text{mol/min}$ )	
	No inhibitor	With Inhibitor ( $I_1$ @ 0.01mM)
0.3	10.1	3.33
0.5	14.3	5.56
1.0	23.5	10.0
3.1	32.8	16.4
5.5	40.5	27.5
9.0	41.3	35.2

(b) Initial velocities were measured under defined conditions for the reaction of sucrase with its substrate sucrose at six different substrate concentrations. Using the data provided below estimate the  $V_{max}$  and  $K_m$  for this reaction. Explain your answer in each case.

[Sucrose]mM	0.0025	0.015	0.05	0.20	3.5	10
Vo (mM/min)	15	36	58	62	70	71

(5 Marks)

## Section B. Answer two questions

Q2.

- (a) Using two examples of assays you have studied describe how enzymes can be used as analytical tools to determine the concentration of specific target analytes. (20 Marks)
- (b) Describe how fluorescence is used to monitor enzyme assays. List two disadvantages to using fluorescent-based assays. (10 marks)

Q3.

- (a) Describe the method used by *E. coli* to regulate the activity of the enzyme beta galactosidase. (15 Marks)
- (b) What is a zymogen? Give two examples of zymogens you have studied. (5 Marks)
- (c) Explain the action of a competitive enzyme inhibitor and using a graph describe its effect on the kinetics of an enzyme. Give one example of a competitive enzyme inhibitor and name the enzyme it inhibits. (10 Marks)

Q4.

Discuss the technique of enzyme immobilization under the following headings:

- |       |                          |            |
|-------|--------------------------|------------|
| (i)   | Principle                | (5 Marks)  |
| (ii)  | Method of immobilization | (10 Marks) |
| (iii) | Advantages               | (5 Marks)  |
| (iv)  | Disadvantages            | (5 Marks)  |
| (v)   | Applications             | (5 Marks)  |