CORK INSTITUTE OF TECHNOLOGY
INSTITIÚID TEICNEOLAÍOCHTA CHORCAÍ

Semester 1 Examinations 2013/2014

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. Gillian Gardiner
Internal Examiner(s): Dr. Fiona O Halloran

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

Duration: 2 Hours

Sitting: Winter, 2013

Requirements for this examination: Scientific calculator

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
Answer **eight** of the following (each question carries equal marks)

Q1.

(a) Distinguish between noncompetitive and uncompetitive enzyme inhibition.

(b) Describe, using a graph, the relationship between reaction rate and substrate concentration in an enzyme catalyzed reaction.

(c) Name the three amino acids that constitute the catalytic triad within the active site of serine proteases.

(d) The K_{cat} values determined for acetylcholine esterase and fumarase are 14,000 s^{-1} and 800 s^{-1} respectively. Which is the most efficient enzyme? Explain your answer.

(e) An enzyme with a K_m of 5 mM yields 15 µmol of product per minute in the presence of saturating substrate concentration. A non-competitive inhibitor, at 5 µM, lowers the activity to 10 µmol/min. Calculate the K_i for the inhibitor.

(f) What is meant by the term 'zymogen'?

(g) Using a graph, explain the effects that a competitive enzyme inhibitor has on Km and Vmax values.

(h) An enzyme that follows Michaelis-Menten kinetics has a K_m of 5 µM. The initial velocity is 0.2 µmol/min at a substrate concentration of 100 µM. What is the initial velocity when substrate concentration is equal to 0.1 mM?

(i) Define the Arrhenius equation, listing what each component of the equation represents.

(j) List five methods that can be used to disrupt cells in enzyme extraction methods

(40 Marks)

Section B. Answer <u>two</u> questions

Q2.

(a) Describe, using Cleland plots, the difference between ordered sequential, random sequential and ping pong mechanisms in multi-substrate reactions. Name one example in each case of an enzyme that operates using this mechanism.

(15 marks)

(b) Describe in detail the catalytic mechanism of action of chymotrypsin.

(15 marks)

Q3.

(a) Using an example you have studied, describe what is meant by a coupled enzyme assay.

(8 marks)

(b) What is the difference between 'fixed-time' and 'Kinetic' enzyme assays

(10 marks)

(c) Describe how the compounds NADH and NAD+ can be used to monitor enzyme catalyzed reactions spectrophotometrically.

(12 marks)

Q4.

(a) What is an allosteric enzyme?

(15 marks)

(b) Describe in detail how the activity of Phosphofructokinase is regulated in the glycolytic pathway of mammals.

(15 marks)