

**CORK INSTITUTE OF TECHNOLOGY
INSTITIÚID TEICNEOLAÍOCHTA CHORCAÍ**

Semester 2 Examinations 2013 (Legacy Paper)

Module Title: Bioanalytical Science II
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Module Code: **BIOL 6004**

School : Science

Programme Title: Bachelor of Science in Applied Sciences – Year 1
 Bachelor of Science in Nutrition and Health – Year 1
 Bachelor of Science in Pharmaceutical Biotechnology – Year 1

Programme Code: **SBIOS_7_Y1**
 SNHSC_8_Y1
 SPHBI_8_Y1

External Examiner: Dr. Anne Nelson, Dr. Jerry Bird

Internal Examiner(s): Dr. M. Begley, Dr. M. Sheahan

Instructions: Answer **FOUR** questions as follows:

Section A: Answer *any two questions*

Section B: Answer Question 4 (compulsory) **and** either question 5 **or** 6

Use **separate answer book** for each section.

All questions carry **25 marks**.

Duration: **2 Hours**

Sitting: **Semester 2 2013**

Requirements for this examination: **Periodic Table**

<p>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.</p>
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Section A

Q1. Answer ALL parts

- (a) Distinguish between a *saturated solution* and a *standard solution*? (3 marks)
- (b) Give **three** different units which may be used to express concentration. (2 marks)
- (c) Give an example of a mobile phase and a stationary phase used in thin layer chromatography? (2 marks)
- (d) Where appropriate, show the relevant calculations in your answer:
 - (i) Which of the following solutions contains the most moles of Li^+ ions; 0.15M LiOH or 0.08M Li_2CO_3 ? Explain your answer. (2 marks)
 - (ii) An aqueous solution of potassium chloride (KCl) was prepared by dissolving 50g of KCl in enough water to make a 600cm^3 of solution. What is the molar concentration of the solution? (3 marks)
 - (iii) What is the concentration of a solution of sodium acetate (CH_3COONa) which was prepared by diluting 5cm^3 of 0.05M sodium acetate to 0.25L? (3 marks)
 - (iv) What is the significance of the retardation factor in thin layer chromatography? Use an appropriate sketch to explain how it may be calculated. (4 marks)
 - (v) List **six** criteria a reagent must satisfy for it to be suitable for use as a primary standard. (3 marks)
 - (vi) What volume of the 20% w/v saline solution is required to prepare a liter of a 5% w/v solution? Show your calculations. (3 marks)

Q2.

- (a) Distinguish between reverse phase and normal phase chromatography. (4 marks)
- (b) What do you understand by the letters HPLC? Highlight the main advantages of HPLC over gravity column chromatography. (6 marks)
- (c) The underwritten are important parameters in gas chromatography. Write a note on each parameter:
 - (i) Sample injection and injection port temperature
 - (ii) Column temperature
 - (iii) How component separation is achieved

(15 marks)

Q3.

- (a) Give the approximate wavelength range for the IR, UV and visible spectrum regions which are utilized in spectrophotometry. Briefly state why electronic transitions do not arise in the IR region. (5 marks)
- (b) What is a *chromophore*? (3 marks)
- (c) A series of potassium permanganate standards were prepared from a 100ppm stock solution. The absorbance of each standard was measured in a 1cm cuvette at the λ_{\max} ; and results are given in the table below. A 5cm³ aliquot of a potassium permanganate solution of unknown concentration was transferred to a 25cm³ volumetric flask and diluted to the mark; the absorbance of this solution was found to be 0.052.

Conc. of standards (ppm)	1	5	10	15	20
Absorbance	0.008	0.039	0.073	0.117	0.160

- (i) Use the data in the table to construct a standard calibration plot and determine the concentration of the unknown solution of KMnO₄. (6 marks)
- (ii) Write a brief note on the precautions that should be taken to minimize errors in the procedure. (6 marks)
- (iii) Give an equation for the Beer Lambert Law. Use the equation to calculate the absorptivity of the KMnO₄ (assume the pathlength is 1cm). Assign appropriate units to the absorptivity. (5 marks)

Section B

Q4. Answer ALL parts – each part is worth **2.5 marks**.

- (a) List three advantages with using plastic ware instead of glassware in a laboratory.
- (b) What kind of information should be included in a centrifuge maintenance log?
- (c) Explain how an agarose gel can separate DNA fragments of different lengths.
- (d) Why is a molecular weight marker (DNA ladder) run on every agarose gel?
- (e) What is a standard operating procedure (SOP)?
- (f) When should standard operating procedures (SOPs) be reviewed and revised?
- (g) Give one example of a chemical indicator, a biological indicator and a physical indicator that can be used for autoclave quality assurance.
- (h) Identify two possible sources of error when using an analytical balance.
- (i) Explain how the isoelectric point (pI) of an amino acid can be determined from its titration curve.
- (j) What type of cuvettes should be used for UV spectrophotometry? Why?

(25 marks)

Q5.

- (a) What is a buffer? List three applications of buffers. (5 marks)
- (b) Calculate the weight of Na_2HPO_4 ($M_r = 142\text{g}$) and KH_2PO_4 ($M_r = 136\text{g}$) required to make 100ml of a 0.05M phosphate buffer $\text{pH} = 6.4$ and $\text{pK}_a = 6.8$. (10 marks)
- (c) Calculate the weight of Na_2CO_3 ($M_r = 106\text{g}$) and NaHCO_3 ($M_r = 84\text{g}$) required to make 100ml of a 0.02M bicarbonate buffer $\text{pH} = 9.6$ and $\text{pK}_a = 10.25$. (10 marks)

Q6.

- (a) Explain your understanding of 'relative centrifugal force'. (5 marks)
- (b) A protocol requires a sample to be centrifuged for 5 mins at 5000g. The radius of the rotor is 7.2 cm. What rpm should the centrifuge be set at? ($\text{RCF} = 1.118 \times 10^{-5} \cdot r \cdot \text{rpm}^2$) (7 marks)
- (c) Give two reasons why it is important to ensure a centrifuge is balanced. (6 marks)
- (d) List three practices which should be followed when centrifuging hazardous material. (7 marks)