

**CORK INSTITUTE OF TECHNOLOGY
INSTITIÚID TEICNEOLAÍOCHTA CHORCAÍ**

Semester 2 Examinations 2016/2017

Module Title: Bioanalytical Techniques

Module Code: BIOT7002

School: Science & Informatics

**Programme Title: Bachelor of Science in Applied Biosciences & Biotechnology
Bachelor of Science (Hons) in Pharmaceutical Biotechnology
Bachelor of Science (Hons) in Nutrition & Health Science
Bachelor of Science (Hons) in Herbal Science**

**Programme Code: SBIOS_7_Y2
SPHBI_8_Y2
SNHSC_8_Y2
SHERB_8_Y2**

**External Examiner(s): Professor Nora O'Brien
Internal Examiner(s): Anne Ward, Dr. Craig Murphy**

Instructions: Answer FOUR questions only. All questions carry equal marks

Duration: 2hrs

Sitting: Summer 2017

Requirements for this examination:

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.

- Q1. (a) Illustrate and explain the principle of separation in gel filtration chromatography (10 marks)
- (b) How would you determine the K_{av} of a gel filtration gel? (5 marks)
- (c) Ferritin (M_w 450,000), transferrin (M_w 80,000) and ferric citrate were separated by gel filtration chromatography. The column had a length of 30 cm and a 0.8 cm diameter. Eluate fractions of 0.2 mL were collected. The maximum of each peak came at the following fraction numbers; ferritin (4), transferrin (34) and ferric citrate (75).
Assuming that ferritin is eluted at the V_o and ferric citrate is eluted at V_t , calculate K_{av} for transferrin. (5 marks)
- (d) Briefly describe the principle and application of dialysis (5 marks)

- Q2. . (a) Define each of the following terms;
- (i) Centrifugal Force (3 marks)
 - (ii) Relative Centrifugal Force (RCF) (3 marks)
 - (iii) Revolutions per minute (RPM) (3 marks)
- (b) Outline each of the following types of centrifugal separation technique:
- (i) Differential Centrifugation (5 marks)
 - (ii) Density Gradient Centrifugation (5 marks)
- (c) What is the adjusted run time (in minutes) needed to achieve the following requirements in a centrifugation separation experiment, given that;

$$T_s = 1 \text{ h}$$

$$RCF = 9,000 \text{ g}$$

$$RCF_s = 14,100 \text{ g}$$

(6 marks)

Q3 (a) Describe with the aid of a diagram the principle of ion exchange chromatography. (10 marks)

(b) Outline the method of HPLC under the following headings:

- (i) Principle of Reverse-Phase Chromatography (5 marks)
- (ii) Stationary and Liquid Phases (5 marks)
- (iii) HPLC detection systems (5 marks)

Q4. In relation to Affinity Chromatography:

- (a) Briefly, describe the principle of the separation technique. Use a diagram to illustrate your answer (10 marks)
- (b) Outline TWO important applications of affinity chromatography (5 marks)
- (c) Outline the experimental procedure in terms of column preparation, adsorption & elution (6 marks)
- (d) What type of matrix can typically be used for this type of chromatography (4 marks)

Q5. . (a) Write a short overview of the principle of separation in each of the following techniques:

- (i) SDS- PAGE (5 marks)
- (ii) Isoelectric focusing (5 marks)

(b) In the design of a Polyacrylamide Gel Electrophoresis system what are the important experimental considerations? (12 marks)

(c) List three methods of analysis post-electrophoresis (3 marks)

Q6. (a) Outline the principle of separation in gas chromatography (8 marks)

(b) List the main types of stationary phase used in gas chromatography (5 marks)

(c) What type of detector systems are commonly used in gas chromatography? (5 marks)

(d) What is the principle of analysis in Infra-Red Spectroscopy? (7 marks)