

**CORK INSTITUTE OF TECHNOLOGY  
INSTITIÚID TEICNEOLAÍOCHTA CHORCAÍ**

**Semester 1 Examinations 2009/2010**

**Bioanalytical Science V**

**Module Code: BIOT7002**

**School: Science**

**Programme Title: BSc in Applied Biosciences with Biotechnology**

**Programme Code: CR-SBIBI-7**

**External Examiner(s): Prof. Gary Walsh**

**Internal Examiner(s): Dr. L. Goold  
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**Instructions: Attempt 2 questions from Section A and 2 questions from Section B**

**Duration: 2 hours**

**Sitting: Autumn 2010**

**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.

## SECTION A

- Q1. (a) Describe the nature of the stationary phase as well as the underlying principles of separation in (i) ion exchange chromatography and (ii) size exclusion chromatography.

(10 marks)

- (b) A chromatography separation yielded the following data for a given set of experimental conditions:-

**Column Length:** 20 cm. **Phase Ratio ( $V_s/V_m$ ):** 0.25

**Retention Times:-** non retained component = **30 seconds**

Component A = **1.5 minutes**

Component B = **2.0 minutes**

**Peak Base Widths:** Component A = **10 seconds**

Component B = **15 seconds**

Calculate the following:- (i) The capacity factor,  $k'$ , for Component A.

(ii) The partition co-efficient,  $K_d$ , for component B.

(iii) The resolution,  $R$ , between A and B

(iv) The length of column required to give a resolution value of 1.50

(10 marks)

- Q2. (a) Compare the processes of isocratic and gradient elution modes of HPLC analysis and explain why the latter mode might be required for successful separation when the former mode is unsatisfactory.

(8 marks)

- (b) Explain the following HPLC instrumental features and comment on their significance:

(i) instrument dead volume (ii) detector wavelength programming (iii) guard column.

(12 marks)

- Q3. (a) Construct a labelled block diagram of a gas chromatographic instrument and use the diagram to give a brief description of how a sample solution containing a mixture of volatile compounds is separated and analysed by this instrument.
- (10 marks)
- (b) An alcoholic beverage sample was analysed by gas chromatography for its ethanol content using the internal standard method of quantitation. Isopropanol was chosen as the internal standard and was added to the sample and standards of ethanol so that all solutions had a constant concentration of isopropanol. The following data were obtained for all solutions analysed:-

<b>%(v/v) Ethanol</b>	<b>Area of Ethanol Peak (integration counts)</b>	<b>Area of Isopropanol Peak (integration counts)</b>
2	660947	1812442
4	1269649	1800540
6	1861078	1799212
8	2247155	1652320
10	3081370	1803780
sample	1185944	1701250

- (i) Use a graphical method to accurately determine the %(v/v) of ethanol in the sample.
- (8 marks)
- (ii) Comment on the usefulness of the internal method of quantitation in gas chromatographic analysis.
- (2 marks)

## SECTION B

Q4. (a) Write a brief note on immunoassay validation (10 marks)

(b) Describe, the principle of **either**:

(i) a reagent excess non-competitive immunoassay

OR

(ii) a reagent limited competitive immunoassay

Use a diagram to illustrate your answer. (10 marks)

Q5. (a) Define & distinguish between each of the following immunoassay classification groups:

(i) Heterogeneous immunoassay (3 marks)

(ii) Homogeneous immunoassay (3 marks)

(b) Write an overview of isotopic & non-isotopic labels used in immunoassay systems (6 marks)

(c) Outline the principle of a particle agglutination immunoassay (8 marks)

Q6. (a) Outline the important experimental considerations in designing a Polyacrylamide Gel Electrophoresis (PAGE) system. (10 marks)

(b) Explain the system of Internal Quality Control (IQC) required in bioanalytical laboratory techniques (10 marks)