

# Cork Institute of Technology

## Higher Certificate in Science in Applied Biosciences-Award

(NFQ – Level 6)

Summer 2007

### Bioanalytical Science 2

(Time: 3 Hours)

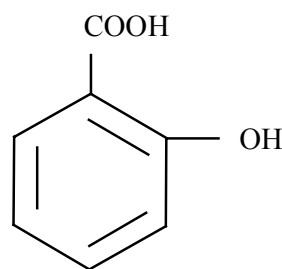
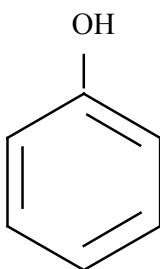
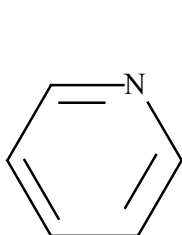
Answer Five questions. Question 1 is compulsory. TWO questions from Section B, ONE from Section C and a fifth questions from Section B or C

Examiners: Dr. R. Hourihane  
Ms. A. Ward  
Mr. C. O' Farrell  
Prof. R. Fitzgerald

## Section A

**Q1. Attempt Ten of the following. All questions carry equal marks.**

- (i) Explain the abbreviations LSC and GSC in chromatography.
- (ii) Which of the following compounds would you expect to fluoresce?

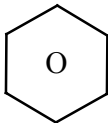
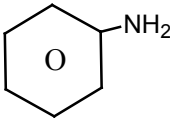


- (iii) List three types of band broadening in chromatography. How is each dependant on flow rate?
- (iv) Radiation from any area of the electromagnetic spectrums shares three fundamental properties. List these properties.
- (v) Calculate the molar concentration of 1.00ppm solution of  $\text{Na}^+$  ions.
- (vi) List three deviations from Beer's Law.
- (vii) Draw a simple diagram of the structure of an IgG molecule
- (viii) Define each of the following: (a) monoclonal antibody (b) polyclonal antibody
- (ix) Draw a diagram illustrating the principle of the Ouchterlony precipitation assay
- (x) Outline one method used to assess the accuracy of a bioanalytical assay
- (xi) List the five main classes of immunoglobulins.

- (xii) Name the 4 components that make up a spectrophotometer.
- (xiii) Name the components and sketch a typical setup for a monochromator.
- (xiv) What type of light source and what region of the electromagnetic spectrum are they optimised for:
1. Laser
  2. Deuterium lamp
  3. Tungsten Filament
  4. Heated Inert Solids
- (xv) A laboratory centrifuge operates at a rotational speed of 10,000rpm.
1. What is the magnitude of the centripetal acceleration on a red blood cell at a radial distance of 6.00cm from the centrifuges axis of rotation?
  2. How does this acceleration compare to g?
- (xvi) Name the different types of centrifuge instruments available.

## Section B

- Q2. (a) (i) Distinguish between a chromophor and auxochrom in ultra violet spectroscopy.
- (ii) Consider the data in the following table.

	Compound	$\frac{\lambda}{\text{nm}}$	$\epsilon$
(1)	$\text{CH}_2 = \text{CH}_2$	180	103
	$\text{CH}_2 = \text{CHNH}_2$	220	104
(2)		255	23
		280	143

- Name and explain the Spectral change (s) the  $\text{NH}_2$  substituent is causing in 1 and 2 above.
- What is the  $\text{NH}_2$  in each scenario?
- Identify two other substitutes which may cause similar effects. (8 marks)

- (b) (i) List at least three characteristics of an ideal solvent for use in spectroscopy.
- (ii) Explain the effect on the resulting Spectrum of changing from a non polar solvent to a polar solvent. (5 marks)
- (c) A solution containing 1.00mg of sodium in 250mL of water was observed to transmit 65% of the incident light compared to the appropriate blank.
  - (i) What is the absorbance of the solution at this wavelength?
  - (ii) What would be the transmittance value of the solution of sodium which is half as concentrated? (7 marks)

Q3. (a) Atomic absorption spectroscopy (AAS) is a low temperature flame method.

- (i) Identify the steps in the flame process from sample introduction (aspiration) to the measurement of absorbance.
- (ii) What is the typical flame temperature in AAS?
- (iii) Identify typical sample types and sample pre-treatment required by the method.
- (iv) Identify an emission method which complements (AAS). (8 marks)
- (b) Flame methods are subjected to interferences.  
List all four interference, discuss two in detail. (6 marks)
- (c) Background correction is essential in most atomic methods.
  - (i) What is meant by background correction?
  - (ii) Name at least three background correction methods.
  - (iii) Discuss two methods of background correction in detail. (6 marks)

Q4. Attempt three of the following:

- (i) Draw a fully labelled energy level diagram detailing the process of fluorescence.  
Include the two radiation less process in your diagram.
- (ii) Write a note detailing the method of ion exchange chromatography.  
In your discussion mention typical types of mobile and stationary phases used, as well as sample types.  
What is meant by the exchange capacity? Diagram required.
- (iii) Illustrate graphically how the molar conductivity of both a strong and weak electrolyte varies with concentration ( $\sqrt{c}$ ).
  - (a) Explain the shape of the graph in each case.
  - (b) Which graph cuts the y axis, what is this value called?  
(Identify by name and symbol)

- (iv) What is meant by the term ion selective electrode (ISE)?  
List four main types.  
The pH electrode is a member of one of these categories. Discuss the pH electrode mentioning and explaining two errors associated with pH measurements.
- (v) What is the column efficiency required to give complete separation to two components with retention times of 25 and 26 seconds?  
Assume the width of both peaks is the same. (20 marks)

- Q5. (a) (i) Using a labelled energy level diagram, illustrate the processes of absorption and emission, of electromagnetic radiation from the ultraviolet (UV) region.  
(ii) Give the energy equation related to these processes.  
(iii) Name the types of transitions illustrated in (i) above. (6 marks)
- (b) A food sample was analysed using Ultraviolet (UV) absorption spectroscopy according to recognised procedures. To this end a series of standard solutions were prepared from a 1000ppm stock solution. Their absorbencies were measured at 305nm. The food sample was treated in a similar manner to the standard solutions. The results are shown in the table below.

Abs @ 305nm	Ions /ppm
0	Blank (H <sub>2</sub> O)
0.197	5
0.360	10
0.550	15
0.925	25
1.121	30
2.605	Food Sample
0.751	Diluted Food Sample

As can be seen from the data the food sample's absorbance reading is outside the range of the standards.

The sample was diluted to 25% of the original concentration and reanalysed.

- (i) Construct an appropriate calibration plot.  
(ii) Determine the concentration of the original undiluted food sample.  
(iii) How would the suggested dilution be achieved?  
(iv) The stock solution of 1000ppm was subjected to serial dilution. What does this mean?  
Suggest how to carry this out in the present analysis. (14 marks)

## Section C

- Q6. Outline, using diagrams for illustration, the principle of each of the following immunoanalytical techniques.
- (i) Immunoaffinity Chromatography
  - (ii) Rocket Immunoelectrophoresis
  - (iii) Single Radial Immunodiffusion (20 marks)
- Q7. (a) Write a brief overview of internal quality control and external quality assessment schemes used in the bioanalytical laboratory. (10 marks)
- (b) Write a note on the use of control charts for plotting analytical data. (7 marks)
- (c) List three potential sources of error commonly encountered in analytical testing. (3 marks)