



Technical University of Mombasa

Faculty of Applied and Health Sciences

DEPARTMENT OF PURE AND APPLIED SCIENCES

UNIVERSITY EXAMINATION FOR THE DEGREE OF BACHELOR OF
TECHNOLOGY IN INDUSTRIAL MICROBIOLOGY AND BIOTECHNOLOGY
BTMBT 12J

ABT 4206: BIOCHEMISTRY TECHNIQUE AND INSTRUMENTATION I

SEMESTER EXAMINATION

DECEMBER 2013 SERIES

2 HOURS

Instructions to candidates:

This paper consist of **FIVE** questions

Answer question **ONE** (compulsory) and any other **TWO** questions

QUESTION ONE

- a) Given a solution of $0.002\text{mol}^{-1} \text{HNO}_3$, Calculate
- (i) $[\text{H}^+]$ (1mark)
 - (ii) $[\text{OH}^-]$ (1mark)
 - (iii) pH (1mark)
 - (iv) pOH (1mark)
- b) Differentiate between the following terms
- (i) Chromophore and Auxochrome (1mark)
 - (ii) Hyperchromic shift and hypochromic shift (1mark)
 - (iii) Kosomotropic salt and chastropic salt (1mark)
- c) Compare and contrast between the following terms

- (i) Atomic Absorption spectroscopy (AAS) and atomic emission spectroscopy (AES) **(4marks)**
 - (ii) Continuous and discontinuous methods of enzyme assays **(4marks)**
- d) A solution contains 0.10mol L^{-1} acetic acid and 0.10mol L^{-1} sodium acetate. If the pK_a of acetic acid is 4.76 calculate the pH of the solution before and after adding 0.05mol L^{-1} NaOH and compare the value with the pH of the solution of 0.05mol L^{-1} NaOH **(6marks)**
- e) (i) Outline SIX ways of locating the separated components during electrophoresis **(3marks)**
- (ii) Using specific examples differentiate between Anionic and cationic exchanges **(4marks)**
- (iii) Explain the role of plaster of Paris (hydrated calcium sulphate) in thin layer chromatography (TLC) **(2marks)**

QUESTION TWO

- a) Discuss the factors that may lead to alteration of results during atomic absorption spectroscopy (AAS) analysis and the ways of overcoming them **(12marks)**
- b) A fluorescent compound gives a reading of 10 in a fluorimeter at concentration of $20\mu\text{mol L}^{-1}$ if the cuvette has a light path of 1cm and the compound has an extinction coefficient of 5270 what fluorescent reading would be expected with a concentration of $40\mu\text{mol L}^{-1}$ **(4marks)**
- c) Illustrate the absorption spectral analysis of oxidized and reduced form of Nicotinamide adenine dinucleotide (NAD) **(4marks)**

QUESTION THREE

Discuss the classification of chromatographic techniques **(20marks)**

QUESTION FOUR

- a) Outline the advantages of instrumental methods of biochemical analysis **(6marks)**
- b) Given the equilibrium constant (K_{eq}) and density of water at 25°C to be $1.8 \times 10^{-16}\text{mol L}^{-1}$ and 1000kg m^{-3} respectively show that the pH of pure water is 7.0 if the atomic masses of H = 1 and O = 16 **(8marks)**
- c) 3.46g of K_2HPO_4 and 2.27g of KH_2PO_4 were dissolved in 250ml of deionized water. Calculate the pH of the resulting solution ($\text{pK}_a = 7.2$, K = 39, H = 1, P = 31, O = 16) **(6marks)**

QUESTION FIVE

- a) Outline the characteristics of a good spectrophotometer **(5marks)**
- b) 20 μ L of glutamate decarboxylase extract was treated with gintamate in a total volume of 3ml and it gave carbon dioxide as shown in the table below

<i>Time (min)</i>	<i>Volume (ml)</i>
0.00	0.00
0.50	0.20
1.00	0.40
1.50	0.61
2.00	0.80

Calculate the enzyme activity in $\mu\text{mol min}^{-1} \text{ml}^{-1}$ **(6marks)**

- c) Different proteins of different molecular weights were precipitated using different acetone concentration (% v/v) as shown in the table below.

<i>Molecular weight (Dalton)</i>	<i>Acetone concentration (% V/V)</i>
39811	7.5 – 12.5
31623	17.5 – 22.5
25119	27.5 – 32.5
19953	37.5 – 42.5
15849	47.5 – 52.5
12589	57.5 – 62.5

Two proteins of unknown molecular were precipitated by 22.5 – 27.5 and 32.5 – 37.5 (% v/v) of Acetone. Approximate the molecular weights of these unknown proteins

(9 marks)