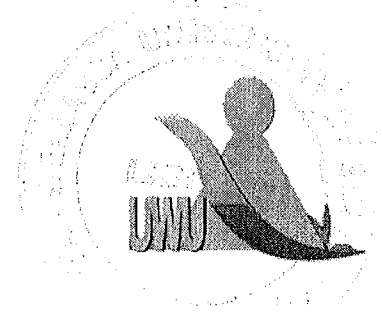


Uva Wellassa University, Sri Lanka
End Semester Examination – September 2011
SCT 319-2 Instrumental Methods in Biology



Time: Two (02) hours

Total five (05) questions.

Answer four (04) questions only.

Use of standalone scientific calculators is allowed.

All symbols carry standard meanings.

Use of standard symbols without a definition is allowed

Question 1

- (i) (25 marks) A particular analytical balance has the capability of reading five digits (including the decimal point). It is required to weigh ~ 0.1235 g of the plant sample for total digestion in a Teflon vessel (around 132.3 g). To weigh required amount of the plant sample, the Teflon vessel was weighed and tarred; the plant sample was transferred into the vessel for digestion. Briefly discuss errors associated in the weighing procedure. Indicate remedial measures you suggested.
- (ii) (25 marks) Define following terms as appeared in instrumental analysis:
- | | |
|--------------------|--------------------------|
| (a) Accuracy | (b) Authenticate samples |
| (c) Spike analysis | (d) Matrix effect |
- (iii) (25 marks) The following replicate data were obtained for the concentration of NO_x in the air near a paper mill: 1.96, 1.91, 1.88, and 1.94 ppm. Calculate (a) the absolute standard deviation, (b) the relative standard deviation, and, (c) the coefficient of variation for the data. Comment on the precision of the data.
- (iv) (25 marks) The mercury in samples of fish taken from the Mahaweli River was determined by atomic absorption method, and following results were obtained: (in ppm) 1.80, 1.58, and 1.64. Calculate at 95% confidence limits for the mean value.

Question 2

- (i) (20 marks) The wave length of characteristic yellow light emitted by sodium street lamps is 589 nm. For photons of this wavelength calculate the frequency wave number and the energy.
- (ii) (20 marks) Explain the meaning of atomic absorption, emission and fluorescence with the aid of energy level diagrams
- (iii) (20 marks) Explain the operational principle of hollow cathode lamp. What is the sputtering process?
- (iv) (20 marks) What is Lock and Key effect? Explain the principle of a diffraction grating used in AAS.

Question 3

- (i) (10 marks) What is the Beer's Law? The measured absorbance for a particular sample is 0.0699. What percentage of light is transmitted through this sample?
- (ii) (25 marks) The following values of absorbance, *A* were obtained for a series of standard Zn solutions:

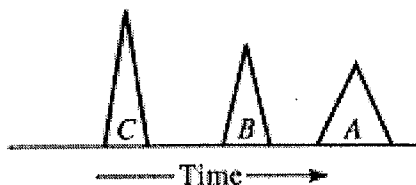
<i>A</i>	0.0	0.152	0.298	0.450	0.600	0.740	0.860	0.940
Zn,	0	2	4	6	8	10	12	14

Plot the calibration curve and determine the concentration of two unknown solutions with absorbance of 0.225 and 0.900 respectively.

- (iii) (25 marks) 5.0 cm³ aliquots of water were analyzed for Cd by adding various amounts of a standard solution of known Cd concentration, and making the resulting solution up to a volume of 10 cm³. Use the data below to determine the concentration of Cd in the original water samples by means of a standard addition plot.

<i>Added Cd ppm</i>	<i>Absorbance</i>
0.0	0.070
0.2	0.112
0.4	0.156
0.6	0.194

- (iv) (40 marks) Differentiate very briefly between reversed phase and normal HPLC methods. Answer this question using the polarities of stationary and mobile phases. Three compounds, A, B, & C were separated using normal phase HPLC. Order these compounds according to polarity. Predict the chromatograph you expected when these compounds are separated by reversed phase HPLC.



Question 4:

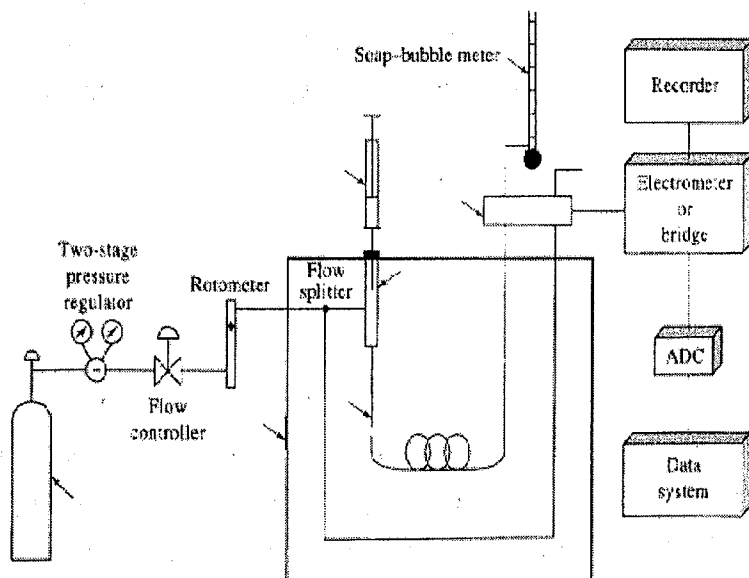
(a) (20 marks) Define following terms in chromatography:

- | | |
|---------------------------|--------------------------------|
| (i). Mobile phase | (ii). Retention time |
| (iii). Selectivity factor | (iv). Distribution coefficient |

(b) (40 marks) Substances A and B were found to have retention times of 16.40 and 17.63 min. respectively on a 30.0 cm column. An unretained species passed through the column in 1.30 min. The peak widths (at base) for A and B were 1.11 and 1.21 min., respectively. Calculate (a). Column resolution, (b). Average number of theoretical plates in the column.

(c) (20 marks) Write van Deemter equation and define each term in it. Write the simplified van Deemter equation as applied for capillary columns.

(d) (20 marks) A schematic diagram of a gas chromatograph (GC) is shown below. Name all components indicated by arrows. List two types of detectors used in GC.



Question 5:

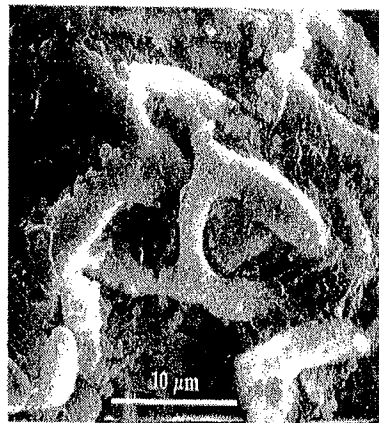
Part A

(40 marks) Use block diagrams to illustrate different modules of AAS, AFS, AES, and AES-ICP. List two non-metals detected by AES-ICP. Why are such detections not possible by AAS?

Part B

(a) (30 marks) State important interactions that occur when an electron beam strikes the specimen. Which of these interactions are important in Scanning Electron Microscopy (SEM)? What is a monochromatic electron beam? How is it generated? Note: a labeled diagram of electron column is required in the answer.

(b) (30 marks) Fungi sample was imaged by SEM. Both images represent the same fungi.

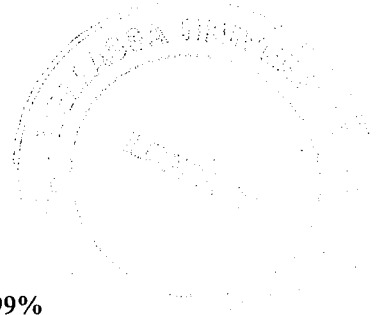


A



B

- i) For morphological analysis of fungi which image is recommended? Why? How was this image taken?
- ii) For compositional analysis of fungi which image is recommended? Why? How was this image taken?



t- Table

Degrees of freedom (N-1)	80%	90%	95%	99%
1	3.08	6.31	12.7	63.7
2	1.89	2.92	4.30	9.92
3	1.64	2.35	3.18	5.84
4	1.53	2.13	2.78	4.60
5	1.48	2.02	2.57	4.03
6	1.44	1.94	2.45	3.71
7	1.42	1.90	2.36	3.50
8	1.40	1.86	2.31	3.36
9	1.38	1.83	2.26	3.25
19	1.33	1.73	2.10	2.88
59	1.30	1.67	2.00	2.66
∞	1.29	1.64	1.96	2.58

z- Table

Confidence level, %	z
50	0.67
68	1.0
80	1.29
90	1.64
95	1.96
96	2.00
99	2.58
99.7	3.00
99.9	3.29