
UNIVERSITI SAINS MALAYSIA

Second Semester Examination
Academic Session 2010/2011

April/May 2011

KAT 244 – Separation Methods
[Kaedah Pemisahan]

Duration : 3 hours
[Masa : 3 jam]

Please check that this examination paper consists of EIGHT printed pages before you begin the examination.

Instructions:-

Answer any **FIVE** (5) questions.

Answer to each question on a new page.

You may answer either in Bahasa Malaysia or in English.

If a candidate answers more than five questions, only the answers to the first five questions in the answer sheet will be graded.

In the event of any discrepancies, the English version shall be used.

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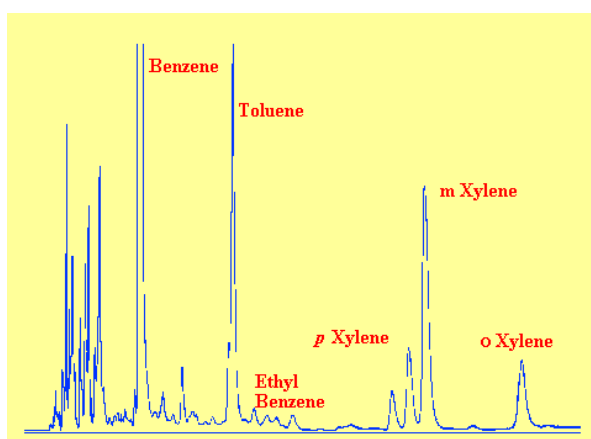
1. (a) Compare, in general terms, solvent extraction and solid phase extraction as a means for isolation of analytes of relatively low polarity from an aqueous sample.
- (10 marks)
- (b) Discuss thoroughly the basic foundation for chromatography and the different chromatographic modes by which analytical separations can be carried out. Your answer should address when, where, and for what kinds of samples the respective technique should preferably be used.
- (10 marks)
2. Compound HA has a partition coefficient of 3 between toluene and water. Suppose we take 100 mL of a 0.010 M solution of HA and extract this with toluene.
- (i) What fraction of HA remains in the aqueous phase if one extraction is done with 500 mL of toluene? For now, ignore any ionization of HA and presume that only simple HA extracts.
- (ii) What fraction remains in the aqueous phase for the single 500 mL toluene extraction if A^- which is the salt of a weak acid, and the pH of the aqueous layer is adjusted so that 50% of the HA species exist as HA and 50% as A^- ?
($K_a=1.0 \times 10^{-9}$, K is 3 for HA, and is 0 for A^- .)
- (20 marks)
3. (a) What are five principal sources of error in the extraction processes which result in the recovery of the analyte being less than 100 percent?
- (5 marks)
- (b) An uncharged organic dye, A, is present in 600.0 mL of polluted water at an unknown concentration. The sample is extracted with 75.00 mL of cyclohexane. After the extraction, the absorbance of the cyclohexane phase, measured at 635 nm in a 1.00-cm cell, is 0.870. The absorbance of a blank (pure cyclohexane) is 0.007. Calculate the concentration of A in the polluted water.
- (Molar absorptivity of A in cyclohexane is $68500 \text{ M}^{-1} \text{ cm}^{-1}$,
Partition coefficient, K of A between cyclohexane and water is 32.)
- (15 marks)

4. Derive van Deemter Equation and describe;

- (i) Why did the same molecules in the column travel at slightly different rates, even though the molecules themselves were identical?
- (ii) How could these effects be minimized or eliminated?

(20 marks)

5. The chromatogram below is the separation of benzene and its substituents by gas chromatography;



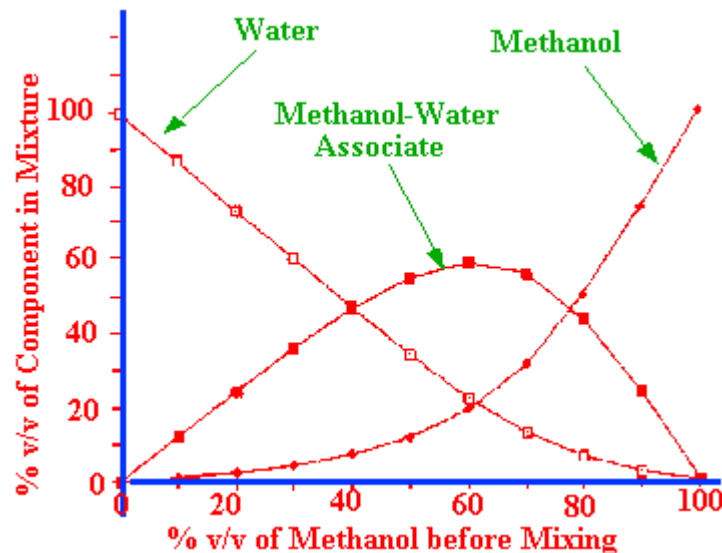
- (i) List some stationary phases suitable for the separation of the mixture as above based on the suitability to function as stationary phase and the ability in the separation.
- (ii) Describe the trend in the separation of the xylene isomers.

(20 marks)

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6. The figure below is the observation reported by Scott & Kucera (R.P.W.Scott and P.Kucera, *J.Chromatogr* **149**,(1978),93.). Explain the mobile phase interaction phenomena of methanol- water and relate with the interaction of the solutes (at least two) of your choice leading towards their separation in isocratic and gradient elution.

Explain why order of elution can be reversed by changing the solvent composition of the mobile phase?



(20 marks)

7. (a) With current demand on high performance with least expenditure, describe how could you acquire *The Ideal Detector*, for general use in high performance liquid chromatography (HPLC)? Does such a detector exist?

(5 marks)

- (b) Discuss the choice of detection wavelength in UV spectroscopic detection for HPLC. Some hints on issues to include in your discussion are solvent properties, signal-to-noise ratio, artifacts resulting from the wavelength selective element of the detector, etc.

(10 marks)

- (c) Describe how diode array detector (DAD) differs from single wavelength UV-VIS HPLC detector. What are the practical implications of this difference?

(5 marks)

TERJEMAHAN

Jawab **LIMA** soalan sahaja.

Jawab tiap-tiap soalan pada muka surat yang baru.

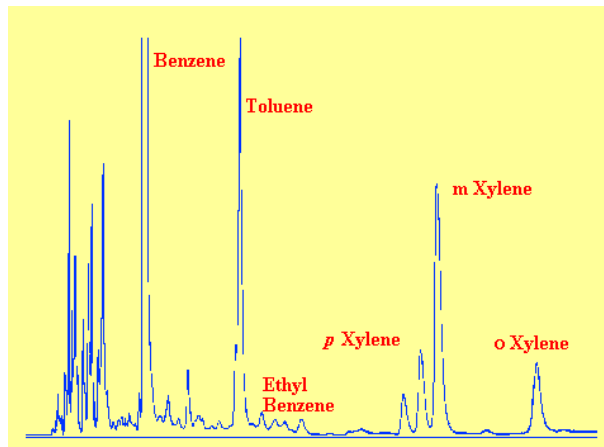
Anda boleh menjawab sama ada dalam Bahasa Malaysia atau Bahasa Inggeris.

Jika calon menjawab lebih daripada lima soalan, hanya lima soalan pertama mengikut susunan dalam skrip jawapan akan diberi markah.

Sekiranya terdapat sebarang percanggahan pada soalan peperiksaan, versi Bahasa Inggeris hendaklah diguna pakai.

1. (a) Bandingkan secara umum, pengekstrakan pelarut dan pengekstrakan fasa pepejal sebagai cara untuk memencilkan analit yang agak rendah kekutubannya daripada sampel akueus.
- (10 markah)
- (b) Bincangkan secara menyeluruh dasar asas bagi kromatografi dan perbezaan pelbagai mod kromatografi dalam mana pemisahan analisis dapat dilaksanakan. Jawapan anda patut menzahirkan bila, dimana, dan bagi jenis sampel bagaimana teknik tersebut patut digunakan.
- (10 markah)
2. Pekali pembahagian sebatian HA diantara toluena dan air adalah 3. Jika diambil sebanyak 100 mL daripada larutan HA 0.010 M dan diekstrakkannya dengan toluena.
- (i) Berapakah pecahan HA tertinggal didalam fasa akueus jika dilakukan pengekstrakan dengan 500 mL of toluena? Buat masa ini, abaikan sebarang pengionan HA dan anggapkan hanya HA sahaja terekstrak.
- (ii) Berapakah pecahan tertinggal didalam fasa akueus bagi pengekstrakan tunggal dengan 500 mL toluena jika A^- yang merupakan garam asid lemah, dan pH lapisan akueus dilaraskan supaya 50% daripada spesies HA wujud sebagai HA dan 50% sebagai A^- ?
- ($K_a=1.0 \times 10^{-9}$, K adalah 3 bagi HA, dan 0 bagi A^- .)
- (20 markah)
3. (a) Apakah lima punca utama ralat didalam proses pengekstrakan pelarut yang menyebabkan keputusan pemulihan analit kurang daripada 100 peratus?
- (5 markah)
- (b) Suatu pewarna organik takbercas, A, hadir didalam 600.0 mL air tercemar yang tidak diketahui kepekatannya. Sampel diekstrak dengan 75.00 mL sikloheksana. Sesudah pengekstrakan, penyerapan daripada fasa sikloheksana, diukur pada 635 nm didalam sel 1.00-cm, adalah 0.870. Penyerapan blank (sikloheksana tulen) adalah 0.007. Kiralah kepekatan A didalam air tercemar tersebut.
- (Keserapan molar A didalam sikloheksana adalah $68500 \text{ M}^{-1} \text{ cm}^{-1}$, Pekali pembahagian, K bagi A diantara siklohesana dan air adalah 32.)
- (15 markah)

4. Turunkan persamaan van Deemter dan jelaskan;
- (i) Kenapakah molekul yang sama didalam suatu turus bergerak pada kadar yang sedikit berbeza, walaupun semua molekul sama?
 - (ii) Bagaimana kesan ini dapat dikurangkan atau dihapuskan? (20 markah)
5. Kromatogram dibawah adalah pemisahan daripada campuran benzena dan produk hasil subsitusinya secara kromatografi gas;

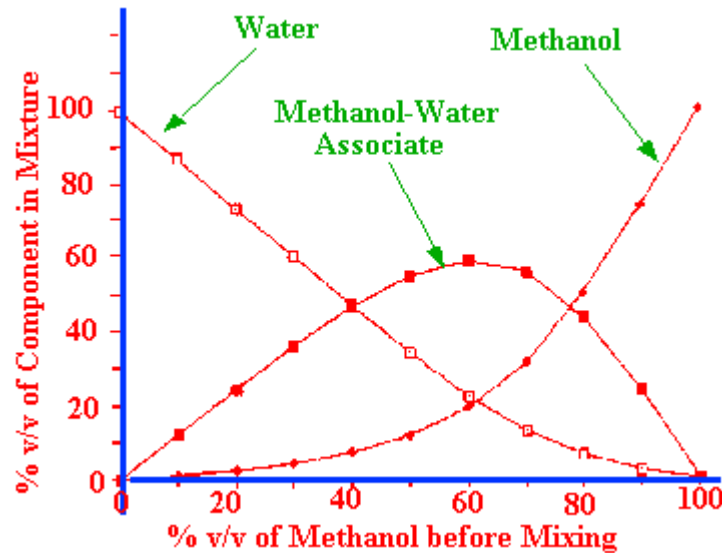


- (i) Senaraikan beberapa fasa pegun yang sesuai untuk pemisahan campuran seperti diatas berdasarkan kesesuaian dari segi fungsinya sebagai fasa pegun dan terangkan kemampuannya dari segi pemisahan.
- (ii) Jelaskan mengenai trend pemisahan isomer xylena. (20 markah)

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6. Gambarajah dibawah adalah pemerhatian yang telah dilaporkan oleh Scott & Kucera (R.P.W.Scott and P.Kucera, *J.Chromatogr* **149**,(1978),93.). Terangkan fenomena interaksi fasa bergerak methanol-air dan kaitkan dengan interaksi solut (sekurang-kurangnya dua) yang anda pilih yang boleh membawa kearah pemisahan isokratik dan pengelusan gradien.

Terangkan kenapa tertib pengelusan menjadi berlawanan dengan mengubah kandungan pelarut didalam fasa bergerak?



(20 markah)

7. (a) Untuk memenuhi permintaan semasa kepada keupayaan tinggi dengan perbelanjaan murah, jelaskan bagaimana anda mendapatkan suatu *Pengesan Ideal*, untuk penggunaan umum didalam kromatografi cecair keupayaan tinggi (HPLC)? Apakah pengesan begini wujud?

(5 markah)

- (b) Bincangkan mengenai pemilihan jarak gelombang pengesanan didalam pengesanan spektroskopi UV bagi HPLC. Beberapa petunjuk kepada isu yang patut dimasukkan didalam perbincangan termasuklah sifat pelarut, nisbah signal-bisingan, tinggalan hasil daripada unsure kepilahan jarak gelombang oleh pengesan, dll.

(10 markah)

- (c) Terangkan bagaimana pengesan susunan diod (DAD) berbeza daripada pengesan HPLC, UV-VIS jarak gelombang tunggal. Apakah implikasi praktikal terhadap perbezaan ini?

(5 markah)

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