

25-14 The advantage of voltammetry at ultramicroelectrodes include: (1) their low IR loss that permits their use in low-dielectric-constant solvents, such as hydrocarbons, (2) their small size that permits electrochemical studies on exceedingly small samples, such as the inside of living organs, and (3) their great speed of equilibration even in unstirred solution.

CHAPTER 26

26-1 (a) *Elution* is a process in which species are washed through a chromatographic column by additions of fresh solvent.

(b) The *mobile phase* in chromatography is the one that moves over or through an immobilized phase that is fixed in place in a column or on the surface of a flat plate.

(c) The *stationary phase* in a chromatographic column is a solid or liquid that is fixed in place. A mobile phase then passes over or through the stationary phase.

(d) The *distribution constant* K in chromatography is the ratio of the concentration of the analyte in the stationary phase to its concentration in the mobile phase when equilibrium exists between the two phases.

(e) The *retention time* for an analyte is the time interval between its injection onto a column and the appearance of its peak at the other end of the column.

(f) The *retention factor* k' is defined by the equation

$$k' = K_A V_S / V_M$$

where K_A is the distribution constant for species A and V_S and V_M are the volumes of the stationary and mobile phases respectively.

(g) The *selectivity factor* α of a column toward two species is given by $\alpha = K_B / K_A$ where K_B is the distribution constant for the more strongly held species and K_A is the corresponding constant for the less strongly held species.

(h) The *plate height* H of a chromatographic column is defined by the relationship

$$H = \sigma^2 / L$$

where σ^2 is the variance obtained from the Gaussian shaped chromatographic peak and L is the length of the column packing in cm.

(i) *Longitudinal diffusion* is a source of band broadening in a column in which a solute diffuses from the concentrated center of a band to the more dilute regions on either side. This movement is thus toward and opposed to the direction of flow of the mobile phase.

(j) *Eddy diffusion* is a phenomenon in which molecules of an analyte reach the end of a column at different times as consequence of traveling through the column by pathways that differ in length.

(k) The *resolution* R , of a column toward two species A and B is given by the equation $R_s = 2\Delta Z / (W_A + W_B)$ where ΔZ is the distance (in units of time) between the peaks for the two species and W_A and W_B are the widths (also in units of time) of the peaks at their bases.

(l) The *eluent* in chromatography is the fresh mobile phase that carries the analyte through the column.

- 26-2** The *general elution problem* arises whenever chromatograms are obtained on samples that contain species with widely different partition ratios. When conditions are such that good separations of the more strongly held species are realized, lack of resolution among the weakly retained species is observed. Conversely, when conditions are chosen that give satisfactory separations of the weakly retained compounds, severe band broadening and long retention times are encountered for the strongly bound species. The general elution problem is often solved in liquid chromatography by gradient elution; temperature programming serves the same purpose in gas chromatography.
- 26-3** The variables that lead to *zone broadening* include: (1) large particle diameters for stationary phases; (2) large column diameters; (3) high temperatures (important only in gas chromatography); (4) for liquid stationary phases, thick layers of the immobilized liquid; and (5) very rapid or very slow flow rates.
- 26-4** In gas-liquid chromatography, the mobile phase is a gas, whereas in liquid-liquid chromatography it is a liquid.
- 26-5** In liquid-liquid chromatography, the stationary phase is a liquid which is immobilized by adsorption or chemical bonding to a solid surface. The equilibria that cause separation are distribution equilibria between two immiscible liquid phases. In liquid-solid chromatography, the stationary phase is a solid surface and the equilibria involved are adsorption equilibria.
- 26-6** Variables that affect the selectivity factor α include the composition of the mobile phase, column temperature, composition of the stationary phase, and chemical interaction between the stationary phase and one of the solutes being separated.
- 26-7** In gas chromatography, the capacity factor is varied by changing the column temperature (temperature programming). In liquid chromatography, variation is accomplished by altering the composition of the solvent (gradient elution).
- 26-8** The number of plates in a column can be determined by measuring the retention time t_R and width of a peak at its base W . The number of plates N is then given by the equation $N = 16 (t_R / W)^2$.
- 26-9** In gas chromatography temperature increases result in shorter elution times but poorer resolution of similar species. Small temperature variations have little effect in liquid chromatography.
- 26-10** The minima observed in plots of plate height versus flow rate are caused by longitudinal diffusion, which in contrast to other broadening sources, goes on to a greater extent at low flow rates than at high. The rate of longitudinal diffusion is, however, orders of magnitude larger in a gaseous mobile phase than in a liquid. Thus, the phenomenon becomes noticeable at higher flow rates in gases than in liquids.

26-11 Gradient elution is a method of performing liquid chromatography in which the composition of the mobile phase is changed continuously or in steps in order to optimize separations.

26-12 $N = 16 (t_R/W)^2$ (Equation 26 - 17)

(a)	<u>N</u>	<u>N²</u>
A	$16(5.4/0.41)^2 = 2775.49$	7.703349×10^6
B	$16(13.3/1.07)^2 = 2472.04$	6.11099×10^6
C	$16(14.1/1.16)^2 = 2363.97$	5.58836×10^6
D	$16(21.6/1.72)^2 = 2523.32$	6.367093×10^6
	$\Sigma N = 10134.81$	$\Sigma N^2 = 25.76979 \times 10^6$

$$\bar{N} = 10134.81/5 = 2533.70 = \underline{2.53 \times 10^3}$$

$$(b) \quad s = \sqrt{\frac{25.76979 \times 10^6 - (10134.81)^2/4}{4-1}} = 174 = 0.2 \times 10^3$$

$$\bar{N} = \underline{2.5(\pm 0.2) \times 10^3}$$

$$(c) \quad H = 24.7 \text{ cm}/2534 \text{ plates} = 9.747 \times 10^{-3} = \underline{0.0097 \text{ cm}}$$

26-13 (a) $k' = (t_R - t_M)/t_M$ (Equation 26 - 8, page 680)

$$A \quad k'_A = (5.4 - 3.1)/3.1 = 0.742 = \underline{0.74}$$

$$B \quad k'_B = (13.3 - 3.1)/3.1 = 3.29 = \underline{3.3}$$

$$C \quad k'_C = (14.1 - 3.1)/3.1 = 3.55 = \underline{3.5}$$

$$D \quad k'_D = (21.6 - 3.1)/3.1 = 5.97 = \underline{6.0}$$

(b) Rearranging Equation 26-5 (page 680) yields

$$K = k'V_M/V_S$$

Substituting the equation in part (a) and the numerical data for V_M and V_S gives

$$K = [(t_R - t_M)/t_M] 1.37/0.164 = [(t_R - t_M)/t_M] 8.35$$

and

$$K_A = [(5.4 - 3.1)/3.1] 8.35 = \underline{6.2}$$

$$K_B = [(13.3 - 3.1)/3.1] 8.35 = \underline{27}$$

26-16 $N = 16(t_R/w)^2$ (Equation 26-17, p683)
 Letting methylcyclohexane be A, methylcyclohexene be B, and toluene be C.

	N_i	N_i^2
N_A	$16(10.0/0.76)^2 = 2770.08$	7.67336×10^6
N_B	$16(10.9/0.82)^2 = 2827.13$	7.99265×10^6
N_C	$16(13.4/1.06)^2 = 2556.92$	6.53786×10^6
	$\Sigma N_i = 8154.13$	$\Sigma N_i^2 = 22.20387 \times 10^6$

(a) $\bar{N} = 8154.13/3 = 2718 = \underline{2.72 \times 10^3}$ plates

(b) $s = \sqrt{\frac{22.20387 \times 10^6 - (8154.13)^2/3}{3-1}} = 142$ plates

$s_m = 142/\sqrt{3} = 82 = \underline{0.08 \times 10^3}$ (Equation a1 - 17, page A - 9)

$\bar{N} = 2.72 \times 10^3 (\pm 0.08 \times 10^3)$ plates

(c) $H = 40/2718 = 1.47 \times 10^{-2} = \underline{0.015}$ cm/plate

26-17 (a) Substituting into Equation 26-20 (page 688)

$R_s = 2(10.9 - 10.0)/(0.79 + 0.82) = 1.14 = \underline{1.1}$

(b) $R_s = 2(13.4 - 10.9)/(0.82 + 1.06) = 2.66 = \underline{2.7}$

(c) $R_s = 2(13.4 - 10.0)/(1.06 + 0.76) = 3.74 = \underline{3.7}$

26-18 (a) Proceeding as in part (d) of Example 26-1, we write

$\frac{N_1}{N_2} = \frac{(R_s)_1^2}{(R_s)_2^2} = \frac{2718}{N_2} = \frac{(1.14)^2}{(1.5)^2}$

$N_2 = 4706 = \underline{4.7 \times 10^3}$ plates

(b) From Solution 26-16(c), $H = 1.47 \times 10^{-2}$ cm/plate

$L = 4706 \times 1.47 \times 10^{-2} = 69.1$ cm = 69 cm

(c) Proceeding as in part (e) of Example 26-1, we write

$\frac{(t_R)_1}{(t_R)_2} = \frac{(R_s)_1^2}{(R_s)_2^2} = \frac{(1.14)^2}{(1.5)^2} = \frac{10.9}{(t_R)_2}$

$(t_R)_2 = 18.9 = \underline{19}$ min

26-19 (a) $k_1' = (10.0 - 1.9)/1.9 = 4.26 = \underline{4.3}$ (Equation 26-8)

$k_2' = (10.9 - 1.9)/1.9 = 4.74 = \underline{4.7}$

$k_3' = (13.4 - 1.9)/1.9 = 6.053 = \underline{6.1}$

(b) Rearranging Equation 26-5 and substituting numerical values for V_M and V_S gives

$K_1 = 4.26 \times 62.6/19.6 = 1.366 = \underline{14}$

$K_2 = 4.74 \times 62.6/19.6 = 1.51 = \underline{15}$

$K_3 = 6.05 \times 62.6/19.6 = 1.93 = \underline{19}$

(c) $\alpha_{2,1} = (10.9 - 1.9)/(10.0 - 1.9) = \underline{1.11}$ (Equation 26-11)

(d) $\alpha_{3,2} = (13.4 - 1.9)/(10.9 - 1.9) = \underline{1.28}$

26-20 (a) Variables that lead to band broadening: (1) very low or very high flow rates; (2) high viscosity mobile phases; (3) low temperatures; (4) large particle sizes for packing; (5) for liquid stationary phases, thick films; (6) long columns; (7) slow introduction of sample; (8) large samples.

(b) Variables that lead to band separation: (1) packings that produce partition coefficients that differ significantly; (2) increases the length of the column packing; (3) variations in mobile phase composition; (4) choice of an optimum temperature; (5) changes in pH of the mobile phase; (6) incorporation of a species in the stationary phases that selectively complexes certain analytes.

26-21 Slow sample introduction leads to band broadening.

26-22 (a) $k_M' = K_M V_S / V_M = 6.01 \times 0.422 = \underline{2.54}$ (Equation 26-5)

$k_N' = 6.20 \times 0.422 = \underline{2.62}$

(b) $\alpha = 6.20/6.01 = \underline{1.03}$ (Equation 26-10)

(c) Substituting into Equation 26-22 gives

$$N = 16(1.5)^2 \left(\frac{1.03}{1.03 - 1.00} \right)^2 \left(\frac{1.00 + 2.62}{2.62} \right)^2 = \underline{8.1 \times 10^4 \text{ plates}}$$

(d) $L = 8.1 \times 10^4 \times 2.2 \times 10^{-3} = 178 = \underline{1.8 \times 10^2 \text{ cm}}$

(e) Substituting into Equation 26-25 gives

$$(t_R)_N = \frac{16(1.5)^2 \times 2.2 \times 10^{-3}}{7.10} \left(\frac{1.03}{1.03 - 1.00} \right)^2 \frac{(3.62)^3}{(2.62)^2} = \underline{91 \text{ min}}$$