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CHROMATOGRAPHIC STUDY OF OPTICAL RESOLUTION

IX*. OPTICAL RESOLUTION OF MONOVALENT COMPLEX CATIONS ON AN ANION-EXCHANGE COLUMN

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SUMMARY

The principle has been established of the optical resolution of a monovalent complex cation on an anion-exchange column using an eluent containing a divalent chiral selector anion. Based on the expressions of two equilibria, ion association and ion exchange, general equations have been derived for the retention volume, the difference in the retention volumes and the separation factor for two enantiomers. These equations have been verified by experimental data for actual complexes eluted with the eluent containing antimony *d*-tartrate.

INTRODUCTION

The chromatographic resolution of racemic cations is usually carried out on a cation-exchange column using an eluent containing chiral selector anions. Many examples of a complete resolution by this technique have been reported¹. In contrast, there are few papers^{2,3} which describe the optical resolution of racemic cations through a column packed with the chiral selector-anionic form of an anion-exchange resin***. This type of chromatography was first attempted by Yoshino *et al.*² who succeeded in the resolution of racemic $[\text{Co}(\text{en})_3]^{3+}$ on an anion-exchange resin saturated with optically active *d*-tartrate or antimony *d*-tartrate (hereafter abbreviated as *d*-tart²⁻ and $[\text{Sb}_2\text{-}d\text{-tart}_2]^{2-}$ respectively). They also achieved optical resolution of racemic $[\text{Co}(\text{EDTA})]^-$ by elution through a cation-exchange column saturated with optically active *A*- $[\text{Co}(\text{en})_3]^{3+}$. However, since their interest was focused on finding a substitute for an optically active resin, no consideration on the detailed mechanism of an optical resolution was made either from the stereochemical point of view or from

* Part VIII: see ref. 6.

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*** In ref. 3 the resolution of the octahedral complex cations was achieved by preferential adsorption on an anion-exchange column in the *d*-tartrate form.

the chromatographic separation theory. Thus, only a partial resolution was obtained. A similar attempt was made by Gaál and Inczédy⁴ who reported a complete resolution of racemic aspartic acid on a cation-exchange column saturated with optically active Λ -[Co(en)₃]³⁺. They assumed a preferential ion association of one enantiomeric form of aspartate anion over the other towards Λ -[Co(en)₃]³⁺. However, although they gave no detailed discussion on the separation mechanism, the experimental conditions they used do not seem to be reasonable for the enantiomeric separation based on preferential ion association.

Here we reconsider the separation of metal cations using an anion-exchange resin. This technique was explored as part of the Manhattan Project, and many examples of separations were reported after World War II⁵. In this method, metal ions are adsorbed on an anion-exchange column as negatively charged complex anions and eluted with an eluent containing anionic ligands. It consists of two equilibria, complex formation and ion exchange, and the separation is achieved owing to the difference in their degrees of complex formation. The theory of separation was established along this line. A similar situation can be imagined for the optical resolution of racemic cations on an anion-exchange column.

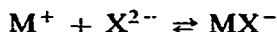
Consider the case where monovalent complex cations are to be separated into enantiomers with divalent chiral selector anions. Complex cations form ion pairs with chiral selector anions. Since the ion pair thus formed has a single negative charge, it can be retained on an anion-exchange resin. Although the enantiomeric separation on an anion-exchange column is, in principle, carried out in the same way as the metal ion separation, the former has disadvantages compared with the latter. In the metal ion separation, complex formation, such as $M^{2+} + 4Cl^- \rightleftharpoons [MCl_4]^{2-}$, is an inner-sphere association and its formation constant is large. However, in the present enantiomeric separation, we are dealing with an outer-sphere association and its formation constant is relatively small. Also, while the charge of the anionic complex in the metal ion separation is higher than that of the anionic ligand, the charge of the ion pair in the present enantiomeric separation is lower than that of the selector anion. Therefore, it is fairly difficult for the complex to be retained on an anion-exchange column. Poor resolution on an anion-exchange column is expected. For good resolution, it is necessary to have a large retention volume for each enantiomer. The dissociation of the ion pair should be depressed with an eluent containing the proper concentration of a chiral selector anion. However, if the concentration is too high, the chiral selector anion will replace the adsorbed ion pair, which results in a decrease in the retention volume. There should be an optimum concentration of the eluent. The situation can be formulated in a quantitative way. This work describes the theory of optical resolution on an anion-exchange column and the experimental data which support the theory.

THEORETICAL

Suppose a monovalent complex cation M^+ is loaded on to an anion-exchange resin of a divalent anionic form X^{2-} and eluted with an eluent containing the anionic ligand X^{2-} . The complex cation M^+ is associated with the anion X^{2-} to form a negatively charged ion pair MX^- which is retained on an anion-exchange resin because of its negative charge. Now, let us consider how the complex M^+ is eluted with

varying concentrations of X^{2-} in an eluent. In the low concentration range, the increase in the concentration in X^{2-} causes an increase in the formation of the ion pair MX^- , which results in an increase in the retention volume of the complex ion. However, above a certain concentration of X^{2-} , another trend becomes predominant. The ion-pair formation reaches a limit. The increased numbers of the anion X^{2-} are directed mainly to exchanging with the adsorbed MX^- . This results in a decrease in the retention volume of the complex ion. Considering two equilibria, ion-pair formation and ion exchange, the situation can be described in a quantitative way.

As the ion-pair formation equilibrium we have



where the association constant β is defined by

$$\beta = \frac{[MX]}{[M][X]} \quad (1)$$

For the ion-exchange equilibrium on an anion-exchange resin, we have



where R is the functional group of the resin. Here, the ion-exchange equilibrium constant K^X involving MX^- and X^{2-} is defined by

$$K^X = \frac{(MX)^2[X]}{[MX]^2(X)} \quad (2)$$

The symbol in parentheses denotes the concentration in the ion-exchange resin, and the symbol in square brackets the concentration in the eluent.

Since the complex ion exists in two forms, M^+ and MX^- , the distribution ratio D_M is given by

$$D_M = \frac{(MX)}{[MX] + [M]} \quad (3)$$

If the total concentration of the complex ion is assumed to be low both in the resin and the eluent phases, *i.e.* $(X) \gg (MX)$ and $[X] \gg [MX]$, the concentration of the divalent anion adsorbed in the resin is, to a good approximation, equal to half of the ion-exchange capacity Q (mequiv./ml), *i.e.*

$$(X) = \frac{1}{2} \cdot Q \quad (4)$$

Using eqns. 1-4, the distribution ratio D_M is expressed by

$$D_M = \frac{\sqrt{K^X Q [X] / 2}}{[X] + \frac{1}{\beta}} \quad (5)$$

The adjusted retention volume, V , for the elution of the complex ion can be related to D_M by

$$V = D_M V_r$$

where V_r is the volume of the resin in the column. Substituting V for D_M in eqn. 5, we obtain

$$V = \frac{K \sqrt{[X]}}{[X] + \frac{1}{\beta}} \quad (6)$$

where $K = V_r \sqrt{K^X Q/2}$. As is easily seen from $dV/d[X] = 0$, the retention volume V has a maximum equal to $K\beta/2$ at an eluent concentration $[X]$ of $1/\beta$.

Now that the general equation for elution has been obtained, chromatographic conditions for enantiomeric separation can be examined based on this general equation. The complex ion M^+ exists in two enantiomeric forms, Δ - M^+ and Λ - M^+ . We have to discriminate the quantities related to them by putting the symbols Δ and Λ as affixes such as V_{Δ} , V_{Λ} , β_{Δ} and β_{Λ} . Fig. 1 shows a plot of the retention volumes V_{Δ} and V_{Λ} against $[X]$ according to eqn. 6, where β_{Λ} is assumed to be greater than β_{Δ} . It must be noted that the maximum in V appears at different values ($1/\beta_{\Delta}$ and $1/\beta_{\Lambda}$) of $[X]$ for the two enantiomers.

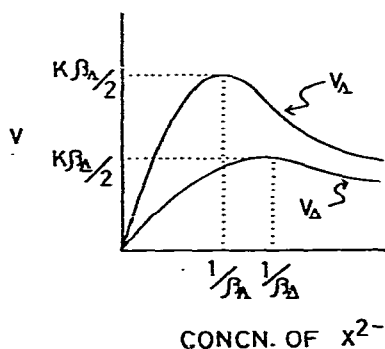


Fig. 1. Theoretical curves of retention volumes, V_{Δ} and V_{Λ} , as a function of eluent concentration $[X]$.

If K in eqn. 6 is assumed to be unchanged for the two enantiomers, the difference in the retention volumes ΔV can be expressed by

$$\Delta V = V_{\Lambda} - V_{\Delta} = \frac{K \left(\frac{1}{\beta_{\Delta}} - \frac{1}{\beta_{\Lambda}} \right) \sqrt{[X]}}{\left([X] + \frac{1}{\beta_{\Delta}} \right) \left([X] + \frac{1}{\beta_{\Lambda}} \right)} \quad (7)$$

where the product $K(1/\beta_A - 1/\beta_A)$ can be regarded as constant and described as K' . Thus, eqn. 7 can be rewritten as

$$\Delta V = \frac{K' \sqrt{[X]}}{\left([X] + \frac{1}{\beta_A}\right) \left([X] + \frac{1}{\beta_A}\right)} \quad (8)$$

The plot of ΔV against $[X]$ is shown in Fig. 2. Here again, a maximum is obtained. It can be proved mathematically that this maximum in ΔV lies at a smaller value of $[X]$ than either of the two maxima in V for Δ and Λ .

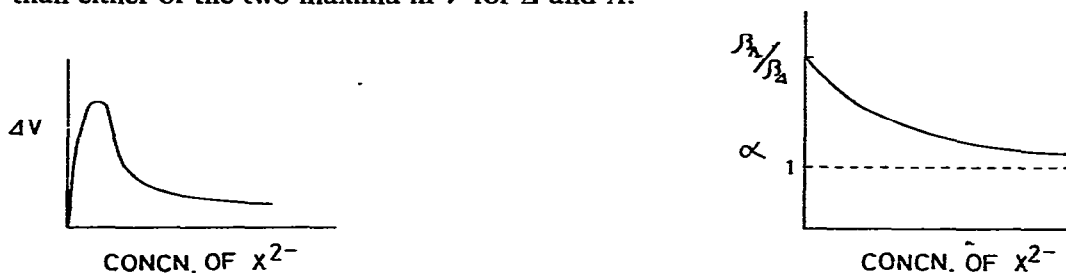


Fig. 2. Theoretical curve of the difference (ΔV) in the retention volumes, V_A and V_A , as a function of eluent concentration $[X]$.

Fig. 3. Theoretical curve of the separation factor as a function of eluent concentration $[X]$.

The separation factor α is defined as the ratio of the retention volumes V_A and V_A . It is expressed as

$$\alpha = \frac{V_A}{V_A} = \frac{\beta_A \cdot \beta_A [X] + 1}{\beta_A \beta_A [X] + 1} \quad (9)$$

Inspection of this equation reveals that $\alpha = \beta_A/\beta_A$ at $[X] = 0$ and reaches unity at $[X] = \infty$. The plot of α against $[X]$ is shown in Fig. 3.

EXPERIMENTAL

Sample complexes

Three monovalent complexes, *cis*(O),*cis*(N)-[Co(en)(gly)₂]Cl·2H₂O (A), *cis*(O),*trans*(N)-[Co(NH₃)₂(gly)₂]Cl·H₂O (B) and *cis*- α -[Co(N₃)₂(trien)]I (C) were used as samples. The preparations of these complexes were described previously⁶.

Eluent

The eluent used was water containing potassium antimony *d*-tartrate, K₂[Sb₂-*d*-tart₂], at various concentrations.

Column

A large column (60 × 2.0 cm I.D.) was used for complex A, and a small

column (40 × 1.5 cm I.D.) for complexes B and C. Each column was packed with the $[\text{Sb}_2\text{-}d\text{-tart}_2]^{2-}$ form of QAE-Sephadex A-25 anion exchanger.

Procedure

A sample solution was prepared by dissolving 20 mg of complex A and 10 mg of complexes B and C in 2 ml and in 1 ml of eluent, respectively. The flow-rate was set at 0.38 ml/min for complex A and 0.16 ml/min for complexes B and C using a peristaltic pump. The detector was operated at the first absorption band of each complex. Circular dichroism and ultraviolet spectra of the eluate for each run proved that complete resolution was achieved.

RESULTS AND DISCUSSION

To check the validity of the theory, chromatographic separation was studied for three actual complexes, *cis*(O),*cis*(N)-[Co(en)(gly)₂]⁺ (A), *cis*(O),*trans*(N)-[Co(NH₃)₂(gly)₂]⁺ (B) and *cis*-α-[Co(N₃)₂(trien)]⁺ (C) using eluents containing K₂[Sb₂-*d*-tart₂] at various concentrations. Typical elution curves obtained with three different eluent concentrations are shown for complex A in Fig. 4. Comparison of these three elution curves reveals that the separation of the two peaks is greater with 0.01 M than with the other two concentrations, 0.0025 and 0.01 M. There seems to be an optimum concentration of the eluent for the best separation of the two peaks. A similar situation is seen for complexes B and C. This is as predicted by the theory.

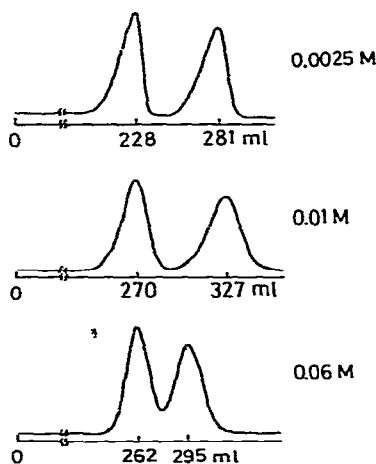


Fig. 4. Typical elution curves of *cis*(O),*cis*(N)-[Co(gly)₂(en)]⁺ with three concentrations of K₂[Sb₂-*d*-tart₂] solution.

Fig. 5 shows plots of the adjusted retention volumes for the enantiomers of the three complexes against the eluent concentrations. These plots show a close resemblance to those in Fig. 1. As expected from the theory, there is a maximum in the curve of retention volume against eluent concentration in all three cases. At low eluent concentration, a large fraction of the complex ion exists as cations, with only a very small fraction existing as negatively charged ion pairs. Under these circumstances, an in-

crease in the eluent concentration results in an increase in the number of the ion pairs retained by the anion-exchange resin. Thus, it brings about an increase in the retention volume. However, this trend soon reaches a limit. Above a certain eluent concentration, an increase in the numbers of the selector anion no longer brings about an increase in the numbers of the ion pair, but is directed towards expelling the adsorbed ion pairs, so that the retention volume of the complex ion will begin to decrease as the eluent concentration increases. The same trend can be found in Kraus and Nelson's review⁵ in which for many metal ions there is a maximum value for the retention volume when plotted against the concentration of the anionic ligands.

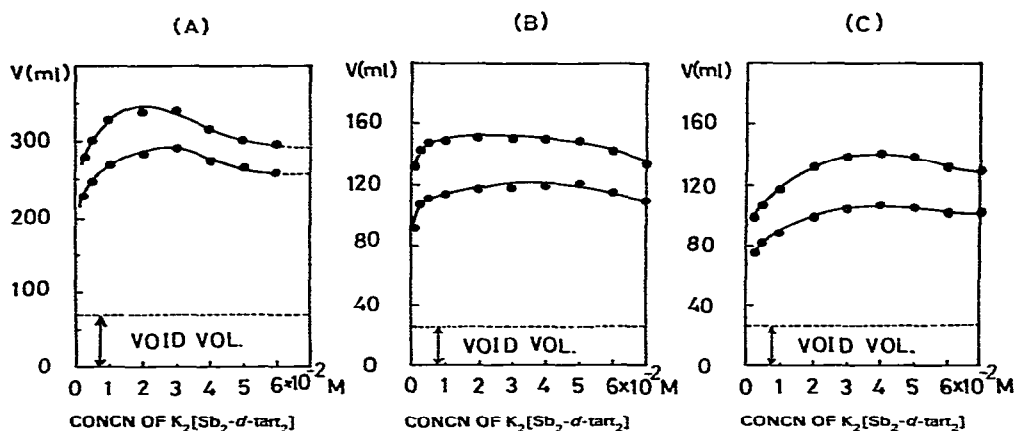


Fig. 5. Dependence of the retention volume on the concentration of $K_2[Sb_2-d-tart_2]$ for the complexes $cis(O),cis(N)-[Co(gly)_2(en)]^+$ (A), $cis(O),trans(N),cis(NH_3)-[Co(gly)_2(NH_3)_2]^+$ (B) and $cis-\alpha-[Co(N_3)_2(trien)]^+$ (C).

Fig. 6 shows plots of the peak separation, *i.e.* the difference in the retention volumes of the two enantiomers (ΔV) against the concentration of the chiral selector anion. Here again, a maximum is found in the curve. As mentioned in the Theoretical section, the concentration which gives a maximum value of ΔV is definitely lower than that which gives a maximum value of the retention volume. A careful look at these three curves, A, B and C, will reveal that while A and B have their maxima at very low eluent concentrations, C has its maximum at a high eluent concentration. The same situation can be seen in the plots of the retention volume in Fig. 5. Here again, the maximum lies at a higher eluent concentration in C, than in A and B. Since the eluent concentration which gives the maximum retention volume is equal to the reciprocal of the ion-pair formation constant, the above fact means that the tendency towards ion-pair formation in complex C is smaller than that in the other two complexes.

Fig. 7 shows the trend of the separation factor α with increasing eluent concentration. As predicted from the theory separation factor decreases with increasing eluent concentration. Here it must be noticed that the separation factor is always greater in C than in A and B. It is also worth noting that the separation factor in C will not decrease to unity so soon with increasing eluent concentration, contrasting with the cases in A and B. These facts can be understood from eqn. 9. As seen in this equation, the separation factor is a product of two terms, β_A/β_B and $(\beta_A[X] +$

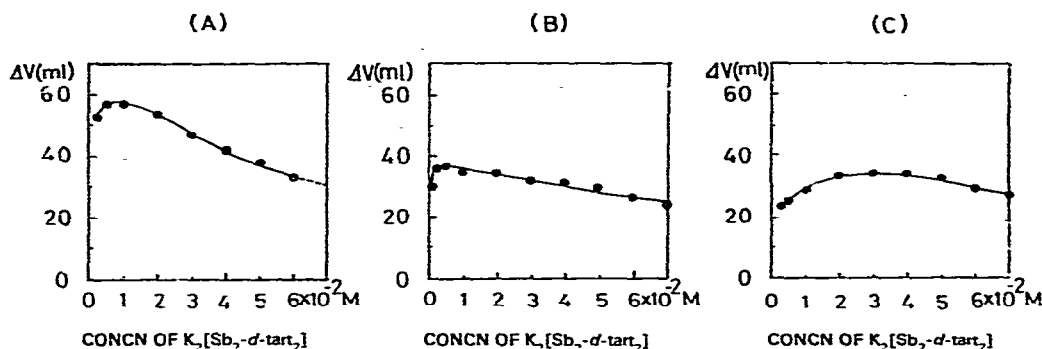


Fig. 6. Dependence of the difference (ΔV) in the retention volumes, V_A and V_A on the concentration of $K_2[Sb_2-d-tart_2]$ solution A, B and C.

$1)/(\beta_A[X] + 1)$. At $[X] = 0$, the second term becomes unity, so that the separation factor is just the ratio of two ion-pair formation constants. At $[X] = \infty$, the second term becomes β_A/β_A , which is just the reciprocal of the first term, so that the separation factor is unity. Between these two extreme cases, the second term varies from 1 to β_A/β_A . In case β_A and β_A are small, the second term will not decrease so soon to β_A/β_A as $[X]$ increases. Thus, the separation factor also will not decrease so soon to unity. This corresponds to the case in C. It can therefore be concluded that although the degree of ion-pair formation is small in complex C, discrimination between the two enantiomers is large.

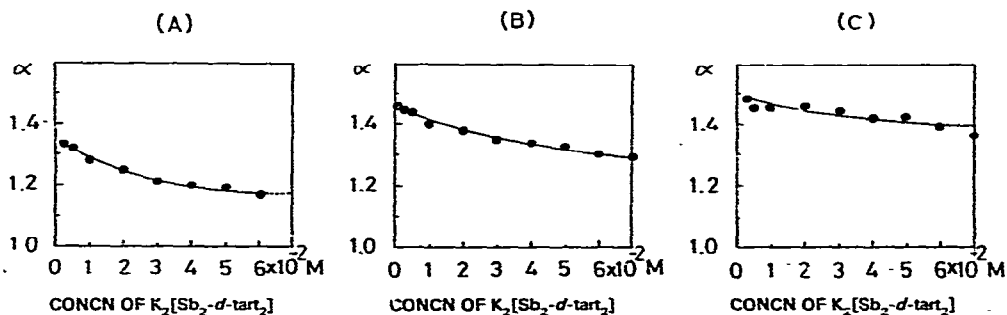


Fig. 7. Dependence of the separation factor α on the concentration of $K_2[Sb_2-d-tart_2]$ for complexes A, B and C.

Finally, the advantage of this "ambush-type" chromatography over ordinary chromatography must be mentioned. If racemic cations are resolved on a cation-exchange column, a large amount of a chiral selector must be used as an eluent. Therefore, if we want to obtain the resolved enantiomers as crystals, we have to isolate them from the eluate containing a large amount of the chiral selector. In contrast, in the "ambush-type" chromatography, a fairly dilute solution of the chiral selector is used, so that it is much easier to obtain the resolved enantiomers in pure form.

REFERENCES

- 1 Y. Yoshikawa, *Coord. Chem. Rev.*, 28 (1979) 205.
- 2 Y. Yoshino, H. Sugiyam, S. Nagaito and H. Kinoshita, *Sci. Pap. Coll. Gen. Educ., Univ. Tokyo*, 16 (1966) 57.
- 3 R. D. Gillard and P. R. Mitchell, *Transition Met. Chem. (Weinheim)*, 1 (1976) 223.
- 4 J. Gaál and J. Inczédy, *Talanta*, 23 (1976) 78.
- 5 K. A. Kraus and F. Nelson, in W. J. Hamber (Editor), *The Structure of Electrolyte Solutions*, Wiley, New York, 1959, p. 340.
- 6 S. Yamazaki and H. Yoneda, *J. Chromatogr.*, 219 (1981) 29.