J. Org. Chem. **2000,** *65,* 7588−7594

Chiral Recognition between a Substituted Cyclooctatetraene Dianion and a Half Crown Ether Substituted with Ibuprofen

Cheryl D. Stevenson* and Daniel K. Cashion

Department of Chemistry, Illinois State University, Normal, Illinois 61790-4160

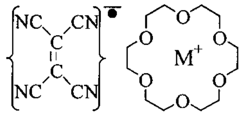
Stevenson@xenon.che.ilstu.edu

Received June 28, 2000

(*S*)-Verbenol was substituted onto cyclooctatetraene (COT) via an ether linkage. In tetrahydrofuran (THF), Cs^+ or Na^+ counterions are tightly ion associated with the verbenoxy-COT dianion. A cosolvent, consisting of an ibuprofen unit connected to a half crown ether, was added to the verbenoxy-COT^{2-},M^+_2 solutions. The intimate interaction between the chiral cosolvent (ibuprofoxymethoxyethoxyethane) and the ion-associated counterion (either Na^+ or Cs^+) forces a chiral recognition between the verbenoxy moiety and the ibuprofoxy moiety. When a molar excess of the cosolvent is present in the dianion THF solution, separation of the cosolvent associated with the verbenoxy-COT^{2-},M^+_2 complex from the uncomplexed cosolvent allows partial resolution of the enantiomers of ibuprofoxymethoxyethoxyethane.

Introduction

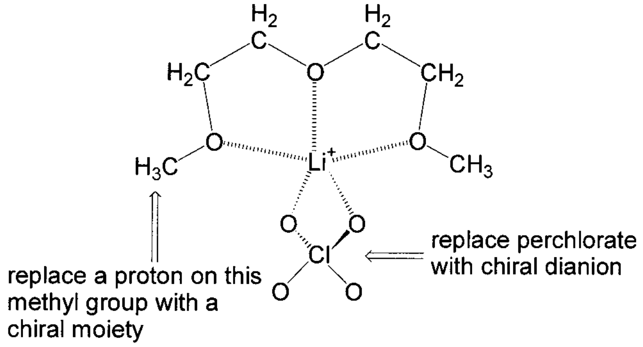
Shortly after the discovery of the amazing affinity the crown ethers have for the alkali metal cation, attempts were made to separate the cations from the anions in tightly ion-associated anion radical complexes with agents such as 18-crown-6. Indeed, EPR studies have shown that the addition of 18-crown-6 to an organic anion radical−alkali metal complex results in the loss of the metal hyperfine structure. However, ion association still persists, but the entire crown−metal cation is involved in the ion associated complex, e.g., Structure I.[1]



I

High level ab initio calculation also predicts that when lithium perchlorate is dissolved in diglyme, the lithium cation is tightly ion associated to the perchlorate anion and simultaneously ligated to the oxygens of the diglyme.[2]

The presence of the anion lowers the Li^+−diglyme binding energy. However, the dissociation energy for [diglyme−$LiClO_4 \rightarrow LiClO_4$ + diglyme] is still predicted to be over 45 kcal/mol, and the distance between the diglyme oxygens and the Li^+ is relatively short (about 2.0 Å, see Structure II).[2,3] It follows that if a chiral hydrocarbon replaces one of the methyl protons of the diglyme system, this chiral moiety could be forced into close proximity to the anion. Further, if the anionic ligand were also chiral, then chiral recognition should take place. We were motivated to investigate the possibility of utilizing such chiral recognition to partially resolve mixtures of enantiomers.



II

The cyclooctatetraene (COT) system is ideal to serve as the anionic part of the proposed complex. Two electrons can be added to COT, and the resulting dianion[4] associates more strongly to alkali metal cations than do the corresponding monoanions.[5] If the COT were attached to a chiral appendage, it seems reasonable that it would recognize the chiral appendage on the diglyme unit.

The possibility of chiral recognition between the substituted diglyme and the substituted COT dianion is further suggested by recent NMR studies.[5] The ^7Li NMR signals for the *R* and *S* enantiomers of the dilithium salts of the *sec*-butoxycyclooctatetraene dianion in a chiral solvent {*S*,*S*-(+)-2,3-dimethoxy-1,4-bis(dimethylamino)-butane} were shown to be considerably different indicating solvent−ion pair chiral recognition. The equilibrium controlling the ion pair dissociation of the double tight ion associated species to form the monosolvent separated

---

(1) (a) Eastman, M. P.; Ramirez, D. A.; Jager, C. D.; Watts, M. T. *J. Phys. Chem.* **1976**, *80*, 182. (b) Watts, M. T.; Lu, M. L.; Chen. R. C.; Eastman, M. P. *J. Phys. Chem.* **1973**, *77*, 2959.

(2) Baboul, A. G.; Redfern, P. C.; Sutjianto, A.; Curtiss, L. A. *J. Am. Chem. Soc.* **1999**, *114*, 968.

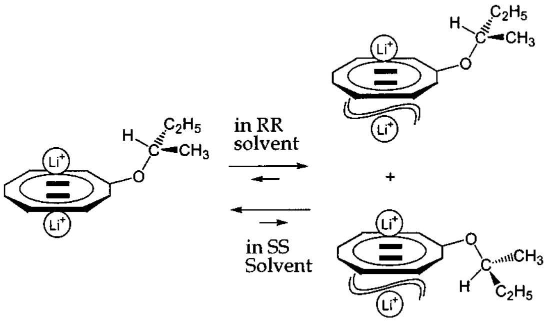
(3) Sutjianto, A.; Curtiss, L. A. *J. Phys. Chem. A* **1998**, *102*, 968.

(4) (a) Strauss, H. L.; Katz, T. J.; Fraenkel, G. K. *J. Am. Chem. Soc.* **1963**, *85*, 2360. (b) Stevenson, C. D., Concepcion, J. G. *J. Phys. Chem.* **1972**, *85*, 2176.

(5) (a) Stevenson, C. D.; Fico, Jr., R. M.; Brown, E. C. *J. Phys. Chem. B* **1998**, *102*, 2841. (b) Cox, R. H.; Harrison, L. W.; Austin, W. K., Jr. *J. Phys. Chem.* **1973**, *77*, 200.

10.1021/jo000972p CCC: $19.00

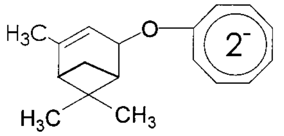
species is shifted to the right in the case of the *S* dianion. The equilibrium (reaction 1, double wavy line indicates a molecule of solvent) can be shifted either to the left or the right by varying the chirality of the solvent or of the dianion.[5]



(1)

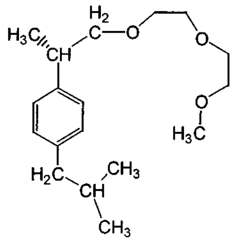
This necessarily means that the association of the solvent with the COT^{2-},Li^{+2} complex is dependent upon the enantiomer of the solvent, and it raises the possibility of using an enantiomerically substituted COT dianion to resolve the enantiomers of a cosolvent (a coordinating solvent added to the primary solvent). The cosolvent, such as a substituted diglyme, has a much higher affinity for the ion-associated cations than does a simple ethereal solvent (e.g., tetrahydrofuran). This allows analogous chiral recognition in the presence of a bulk nonchiral solvent system. The chiral recognition might be enhanced by utilizing a relatively large, rigid (bicyclic) substituent on the COT dianion. The verbenoxy unit has all of the desired properties.

Verbenol is a bark beetle pheromone and is readily available as a pure enantiomer. Since it is an alcohol, it can be readily attached to COT via an ether linkage.[6] In a THF solution of the dianion of verbenoxy-COT (III), containing a molar excess of the racemic cosolvent, one of the two enantiomers of the cosolvent should be preferentially coordinated to the counterion. This counterion is, in turn, tightly associated with the enantiomerically pure verbenoxy-COT^{2-}. The unassociated enantiomer of the cosolvent might then be washed from the mixture with hexane. This should result in some separation of the isomers of the cosolvent.

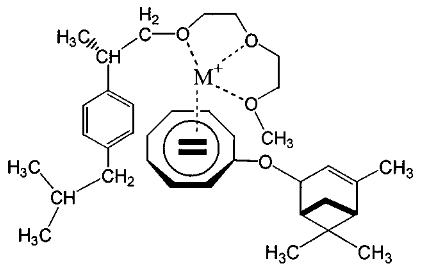


III (verbenoxy-COT dianion)

The cosolvent of choice should contain a large chiral hydrocarbon unit, which is, in turn, attached to a diglyme moiety. The experiment would be of further interest if the cosolvent were bonded to a molecule of pragmatic interest. Ibuprofoxymethoxyethoxyethane (IV) has all of the desirable traits. If the equilibrium constants for the solvation competition reactions (reactions 2 and 3) differ from unity, then the two-electron reduction of III in the presence of a mixture of enantiomers of IV will predominantly yield a dianion, which is preferentially solvated by one enantiomer of the cosolvent. Consequently, separation of the ion pair complexed cosolvent (V) from the free cosolvent may result in a partial chiral resolution of the enantiomers of IV. The two cosolvent ion pairs are diasteriomeric possibly rendering the thermodynamic parameters controlling reactions 2 and 3 different. It seems possible that the two chiral moieties in the solvent−ion pair complex (V) could avoid a steric interaction by being on opposite sides of the complex. Could this minimize the chiral recognition and result in a chiral perturbation upon reactions 2 and 3 is unobservable? We were motivated to answer this question.



IV (ibuprofoxymethoxyethoxyethane)



V

The first report of diasteroselective solvation involves an organolithium compound [lithium 2-methoxy-((*R*)-1-phenylethyl)((*S*)-1-phenylethyl)amine] in (*R*)- and (*S*)-2-methyl-THF.[7] In this case, the two diasteriomers proved to be analytically different with respect to ^{13}C NMR chemical shifts. In this elegant study the authors used their NMR results to show that the two enantiomers of the organolithium compound have different affinities for the (*R*) and (*S*) enantiomers of the 2-methyl-THF.[7]

Results and Discussion

The reduction of verbenoxy-COT with a molar deficient amount of lithium metal in HMPA (hexamethylphosphoramide) leads to a solution that yields a strong well resolved EPR signal corresponding to an anion radical that is free of ion association,[8] and EPR analysis exhibits a quartet ($a_{H(odd)} = 5.64$ G, 3 Hs) of pentets ($a_{H(even)} = 0.68$ G, 4 Hs), which is consistent with the published quantum mechanical model for monosubstituted cyclooctatetraene anion radicals (Figure 1).[9] When the reduction is carried out in THF, where both the anion radicals and dianion are more strongly ion associated,[10] only a very weak EPR

---

(6) (a) Krebs, A. *Angew. Chem.* **1965**, *77*, 1966. (b) Stevenson C. D.; Davis, J. P.; Reiter, R. C., *J. Phys. Chem.* A **1999**, 103, 5343.

(7) Hilmersson, G.; Ahlberg, P.; Davidsson, O. *J. Am. Chem. Soc.* **1996,** *118,* 3539.

(8) (a) Stevenson, C. D.; Echegoyen L.; Lizardi, L. R. *J. Phys. Chem.* **1972**, *76*, 1439. (b) Levin, G.; Jagur-Grodzinski, J.; Szwarc, M. *J. Am. Chem. Soc.* **1970**, *92*, 2268. (c) Szwarc, M. In *Ions and Ion Pairs in Organic Reactions*; Szwarc, M., Ed.; John Wiley and Sons: New York, 1974; Vol. 2, Chapter 1.

(9) (a) Stevenson C. D.; Davis, J. P.; Reiter, R. C. *J. Phys. Chem.* A **1999**, *103*, 5343. (b) Hammons, J. H.; Hrovat, D. A.; Borden, W. T. *J. Am. Chem. Soc.* **1991**, *113*, 4500.

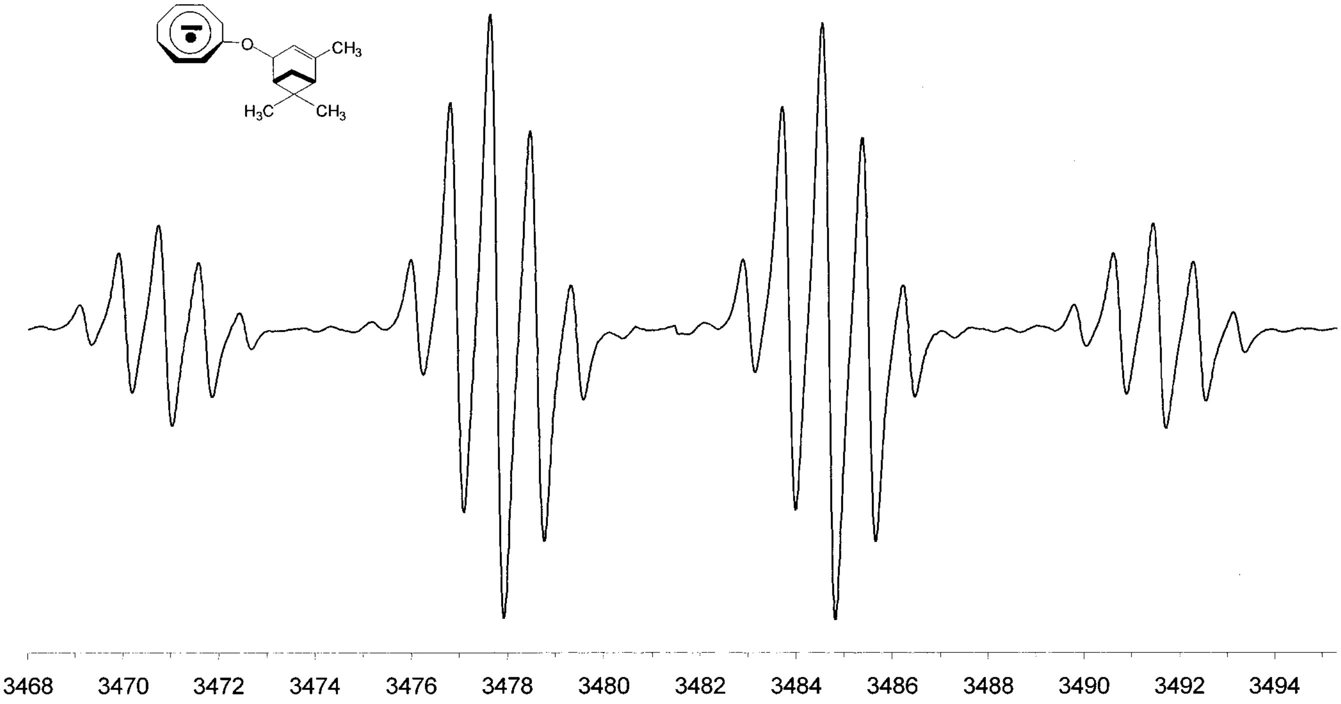
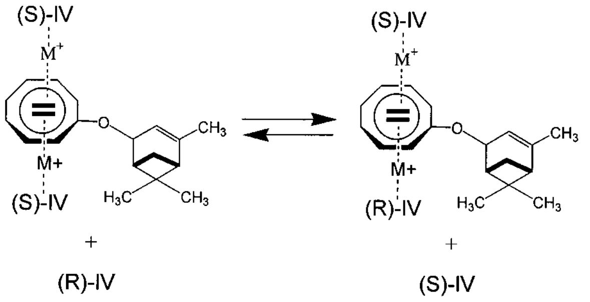
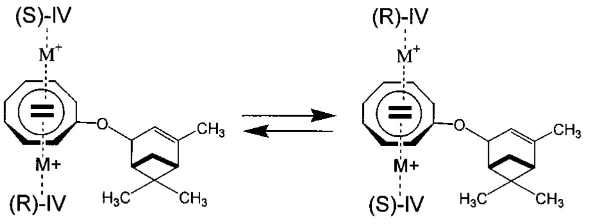


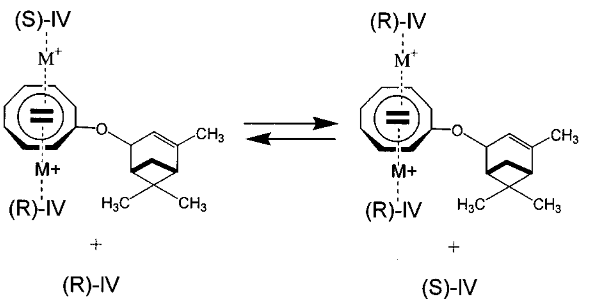
Figure 1. X-band EPR spectrum (*y*-axis in Gauss) of the anion radical of verbenoxycyclooctatetraene in HMPA at ambient temperature. The modulation amplitude was 0.05 G.



(2)

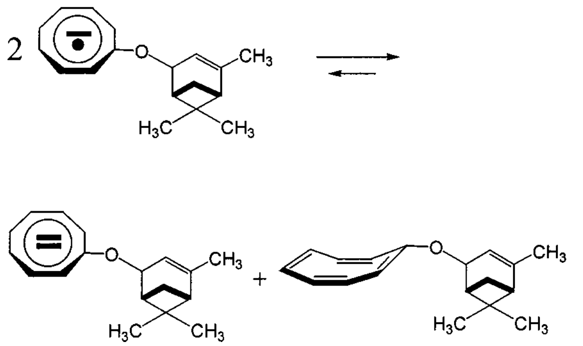


(3)



(4)

signal is observed. This is a consequence of tight ion association forcing the disproportionation equilibrium (reaction 5) far to the right.[4a,10]



(5)

(*S*)-Ibuprofen was reduced to the corresponding alcohol with lithium aluminum hydride. Since enantiomerically pure (*R*)-Ibuprofen is not available, racemic ibuprofen was similarly reduced with lithium aluminum deuteride. The (*S*)-ibuprofenol and racemic dideuterioibuprofenol were attached to methoxyethoxyethane via ether linkages. The differing masses of the (*S*)-IV and the racemic IV-d_2 allow for unambiguous GC-mass spectral analysis.

Mixtures of the (*S*)-IV and racemic IV-d_2 were used as the cosolvent during the two electron reductions of verbenoxy-COT in THF with either sodium or cesium metal. Since the cosolvents were present in molar excess (more than two moles of cosolvent per mole of verbenoxy-COT dianion), only a fraction of the cosolvent can be associated with the dianion. Hence the equilibria described in reactions 2 and 3 were established. After removal of the THF under reduced pressure, the uncomplexed cosolvent mixture was separated from the dianion−cosolvent−metal complexes via dissolution in hexane. This solution was subsequently labeled Phase I.

The dianion−cosolvent−metal complexes proved to be, as expected, insoluble in hexane. A solution of I_2 in

---

[10] Ion association strongly promotes disproportionation and renders the concentration of anion radical small; see: (a) Stevenson, C. D.; Catlett, D. L.; Sedwick, J. B.; Wiedrich, R. *J. Phys. Chem.* **1983**, *87*, 578. (b) Smentowski, F. J.; Stevenson, C. D. *J. Phys. Chem.* **1969**, *73*, 340. (c) Smentowski, F. J.; Stevenson, C. D. *J. Am. Chem. Soc.* **1967**, *89*, 5120.

Table 1. Metal Used for the Reduction, Ratio of the Total Amount of Deuterated Cosolvent over the Total Amount of Protiated Cosolvent in the Original Mixture, Q, and the Separation Factor (α)

metal used	total D/total H in original mix	X_I	X_{II}	Q	α
Na	1.004 ±0.004	1.079 ±0.015	0.990 ±0.005	0.502 ±0.004	0.846 ±0.030
Cs	0.976 ±0.003	0.999 ±0.005	1.053 ±0.006	0.488 ±0.002	1.11 ±0.02

hexane was then added to the complexed dianion salt, whereby it was reoxidized to the neutral state ($C_{10}H_{14}-O-COT^{2-},2M^{+}:2IV + I_2 \rightarrow C_{10}H_{14}-O-COT + 2MI + 2IV$). This reoxidation freed the cosolvent, which was dissolved in hexane and labeled Phase II.

When Na served as the reducing agent ($M^+ = Na^+$ in reactions 2 and 3), GC-mass spectral analysis revealed that Phase I is enriched in IV-d_2 and depleted in the perprotio-IV, while the reverse is true in Phase II, Table 1. This means the *R* isomer is enriched in Phase I and depleted in Phase II relative to the *S* isomer. The interaction between *S* enantiomer of the cosolvent and the disodium salt of the (*S*)-verbenoxy-COT dianion must be more favorable than the analogous interaction with the *R* enantiomer of the cosolvent.

Let S_{hI}, S_{hII}, R_{hI}, and R_{hII} represent the relative amounts of perprotiated (*S*)- and (R)-cosolvents in Phases I and II, respectively. Likewise, let S_{dI}, S_{dII}, R_{dI}, R_{dII} represent the relative amounts of dideuterated (*S*)- and (*R*)-cosolvents in Phases I and II. The mass spectral data yields the ratio of dideuterated to perprotiated material in each phase [$X_1 = (S_{dI} + R_{dI})/S_{hI}$ and $X_2 = (S_{dII} + R_{dII})/S_{hII}$]. Keep in mind that there is no perprotiated *R* isomer; hence, R_{hI} and $R_{hII} = 0$. The separation factor (α) is defined by the ratio of *S* enantiomer to *R* enantiomer in Phase I divided by that ratio in Phase II[12] and is given by eq 6.

$$\alpha = \frac{[(S)\text{-IV}]/[(\text{R})\text{-IV}] \text{ in Phase I}}{[(S)\text{-IV}]/[(R)\text{-IV}] \text{ in Phase II}} = \frac{(S_{hI} + S_{dI})(R_{dI})}{(S_{hII} + S_{dII})(R_{dII})} \tag{6}$$

The ratio of dideuterated to perprotiated (*S*) cosolvent is the same in both phases ($S_{dI}/S_{hI} = S_{dII}/S_{hII} = Q$), which is the same as that in the original mixture of racemic dideuterated and *S* perprotiated cosolvents. This value, *Q*, is easily obtained from the composition of the original mixture. Substituting the expressions for X_I, X_{II}, and Q in eq 6 leads to an expression which can be directly evaluated from the mass spectral data, eq 7.

$$\alpha = \frac{(S_{hI} + S_{dI})/(X_I S_{hI} - S_{dI})}{(S_{hII} + S_{dII})/(X_{II} S_{hII} - S_{dII})} = \frac{(X_{II} - Q)}{(X_I - Q)} \tag{7}$$

Using the data from our most successful separation (Table 1) in eq 6 yields a separation factor of $\alpha = 1.11 \pm 0.02$ when Cs^+ serves as the counterion and $\alpha = 0.846 \pm 0.030$ when Na^+ serves as the counterion. The data show that there is no doubt that there is chiral recognition between the verbenoxy group on the COT dianion and the ibuprofoxy moiety on the cosolvent. The separation factors deviate from unity, although it is physically possible for the two chiral moieties to remain on opposite sides of the cyclooctatetraenyl ring system as diagrammed in Structure V.

Apparently the molecular dynamics in the cosolvent−ion pair complex does result in some physical interaction between the two chiral moieties. This chiral recognition is of sufficient magnitude to render the free energy changes of reactions 2 and/or 3 finite in value. At this point it is impossible to delineate which equilibria (reactions 2−4) is the largest contributor to α, but most likely all three have nonzero values for $\Delta G°$. The net value for $\Delta\Delta G°$, which represents the difference in the overall abilities of the (*R*) and (*S*) cosolvents to associate with the $C_{10}H_{14}-O-COT^{2-},2M^+$ complex, is simply defined as $\Delta\Delta G° = -RT \ln \alpha$.[12] With this in mind $\Delta\Delta G° = -48$ and 77 cal/mol for the Cs and Na systems, respectively.

The separations described above establish the existence of chiral recognition between the ion pair and the cosolvent. More insight into the nature of this interaction may be possible through nuclear Ovehauser experiments. The chiral recognition between the cosolvent and the ion-associated dianion could possibly be greatly augmented by placing a second verbenoxy group on the COT ring system, perhaps in the number four position. There are some synthetic challenges involved in this proposed experiment, but we are currently addressing the problem.

The contrasting values for α in the sodium and cesium systems came as a surprise. The subtle interactions between two chiral moieties, the counterion, and the THF are very complex. Further, the large number of atoms involved rules out the use of sophisticated quantum mechanical treatments. Nevertheless cosolvent−ion pair chiral recognition has been achieved, and it is of sufficient magnitude to allow the partial physical separation of the enantiomers of ibuprofen. Further, the results of the partial separations allow useful insight into the nature of the solvent−ion pair interaction.

The use of the verbenoxy-COT dianion or diverbenoxy-COT dianion as a stationary phase in a chromatographic column represents another experimental challenge that we are currently addressing. Indeed according to Linder,[12] a $\Delta\Delta G°$ value of 29 cal/mol require 15000 theoretical plates to generate a complete separation of a pair of enantiomers. Our $\Delta\Delta G°$ values are larger in magnitude than this.

Experimental Section

Verbenoxycyclooctatetraene. The synthesis of verbenoxy-COT was based on Kreb's preparation of *tert*-butoxycyclooctatetraene.[6] Verbenol (5.0 g, 33 mmol), from Aldrich Chemical Co., was refluxed in 20 mL of tetrahydrofuran (THF) with a molar excess of potassium metal to form the alkoxide. The potassium alkoxide salt in THF was then added to 4.3 g of bromocyclooctatetraene (23 mmol) in 20 mL of THF at −78 °C. The reaction mixture (Scheme 1) was maintained at −78 °C for 24 h. After 24 h, the reaction mixture was allowed to warm to room temperature, and the THF was distilled from the reaction vessel. Verbenxoy-COT, a viscous yellow oil, was purified by vacuum (10^{-3} Torr) distillation in 14% yield and collected from 140 to 150 °C. The structure was verified by NMR, mass spectral, and C, H, and O analysis (expected: 85.0% C, 8.66% H, 6.30% O; found: 85.6% C, 8.17% H, 6.23% O).

Ibuprofoxymethoxyethoxyethane: (±)-Ibuprofenol-d_2 (4.7 g, 24 mmol) was obtained in nearly 100% yield by the reduction

---

[11] Stevenson, C. D.; McElheny, D. J.; Kage, D. E.; Ciszewski, J. T.; Reiter, R. C. *Anal. Chem.* **1998**, *70*, 3830.

[12] Linder, W. In *Chirality- From Weak Bosons to the α-Helix*; Janoschek, R., Ed.; Springer-Verlag: New York, 1991; Chapter 9.

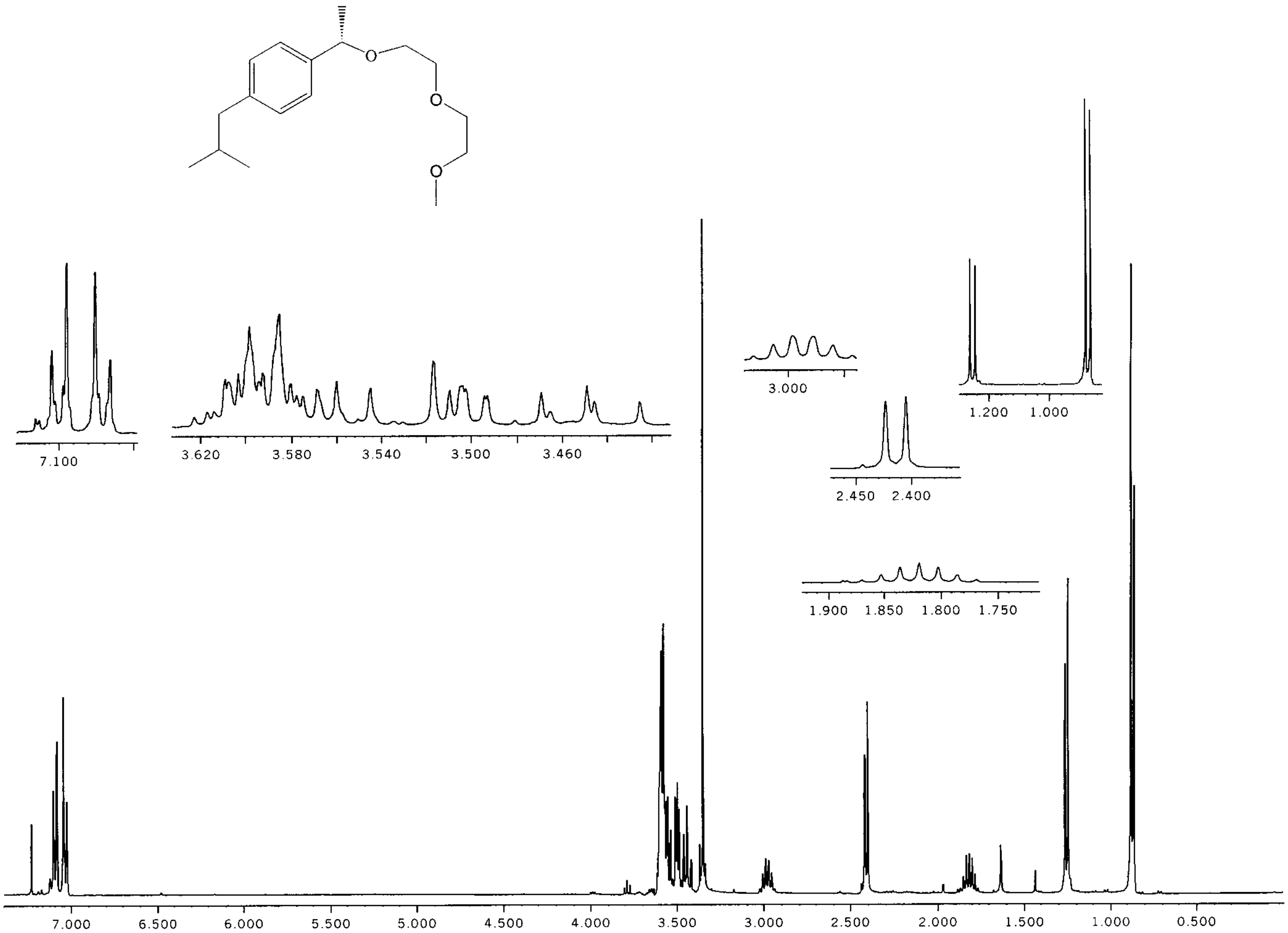
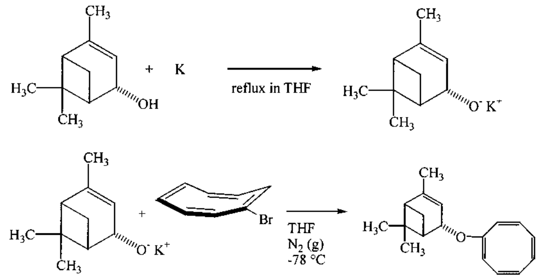
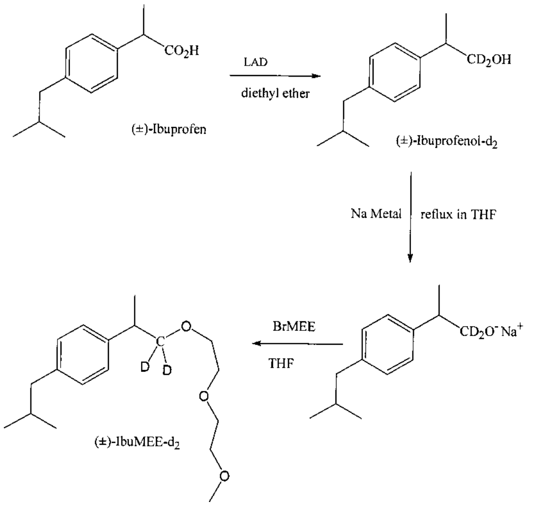


Figure 2. ^{1}H NMR (400 MHz) spectrum (*y*-axis in ppm) of the *S* cosolvent [(*S*)- ibuprofoxymethoxyethoxyethane] in $CDCl_3$.

Scheme 1



Scheme 2



of 5.0 g of (±)-4-isobutyl-α-methylphenylacetic acid (ibuprofen) (24 mmol) with 1.0 g of $LiAlD_4$ (98 atom % D) (32 mmol) at 0 °C in anhydrous diethyl ether. After 1 h, the reaction mixture was allowed to come to ambient temperature. Wet ether (20 mL) was then added to the reaction mixture, followed by the addition of 10 mL of a 5% HCl solution. The ether solution was separated and concentrated under reduced pressure. The resulting (±)-ibuprofenol-d_2 (4.7 g, 24 mmol) was refluxed with a molar excess of sodium metal in THF for approximately 36 h to form the respective sodium alkoxide. The sodium alkoxide was not isolated but was added to a separate reaction vessel containing 13.0 g of 1-bromo-2-(2-methoxyethoxy)ethane (BrMEE, 71 mmol) in THF and left to react at 50 °C for 24 h (Scheme 2). The THF was distilled from the reaction vessel while remaining under an N_2 (g) atmosphere. Deionized water (20 mL) was added to the reaction mixture, followed by addition of a 5% NaOH solution until the reaction mixture was basic to litmus. The resulting mixture was extracted with diethyl ether several times and dried with $MgSO_4$. Most of the unreacted BrMEE was then removed under reduced pressure to increase the efficiency of the following chromatographic step. The resulting (±)-ibuprofoxymethoxyethoxyethane-d_2 [(±)-ibuMEE-d_2, 0.8 g, 2.7 mmol] was purified via alumina flash chromatography and recovered in ~11% yield. Deionized water (6 mL) was added to 200 mL of alumina gel (activated, neutral, Brockman I) in a slurry of 10% ethyl acetate in hexane. The addition of 6 mL of water to each 100 mL of dry alumina was necessary to reduce the activity of the alumina. The R_f values of (±)-ibuprofenol-d_2, (±)-ibuMEE-d_2, and BrMEE under these chromatographic conditions were 0.11, 0.32, and 0.51, respectively. (*S*)-(+)-ibuMEE was synthesized using $LiAlH_4$ in place

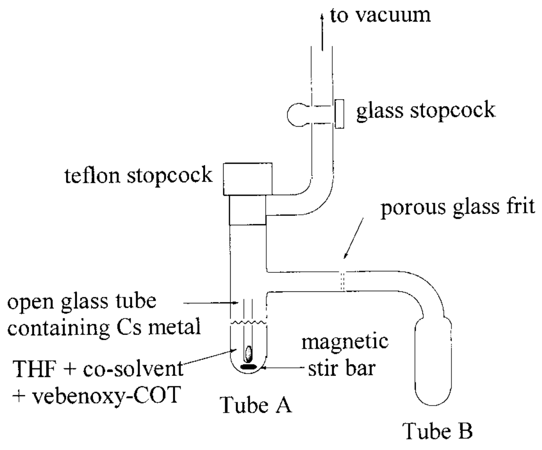


Figure 3. Apparatus used for the partial separations of the (*R*) and (*S*) enantiomers of the cosolvent using Cs metal as the reducing agent. When Na served as the reducing metal, a sidearm was attached to tube A. The sodium metal was then distilled into tube A (forming a mirror) while the contents of tube A were maintained at liquid nitrogen temperature.

of $LiAlD_4$ in an analogous manner. The structure was verified by NMR (Figure 2), mass spectral, and C and H analysis (expected: 73.97% C, 9.58% H, 16.45% O; found: 74.0% C, 9.55% H, 16.4% O).

Separations. Experiments involving the cesium cation were carried out in an apparatus such as that shown in Figure 3. In a typical experiment, a cosolvent mixture was prepared by dissolving 150 μL (0.508 mmol) of (±)-ibuMEE-d_2 and 150 μL (0.508 mmol) of (*S*)-(+)-ibuMEE in 10 mL of hexane distilled from NaK_2. This cosolvent mixture in hexane was dried over calcium hydride under an argon atmosphere. Into bulb A, Figure 3, was placed 20 μL (0.085 mmol) of verbenoxy-COT. Under an argon atmosphere, the cosolvent mixture in hexane was then placed in bulb A. The apparatus was then attached to a vacuum line, and the hexane was removed with stirring under vacuum. The apparatus was removed from the vacuum line and opened under an inert argon atmosphere. A sealed glass ampule containing 0.27 g of Cs metal (2.0 mmol) was then opened and placed in bulb A. The apparatus was reattached to the vacuum line, and 10 mL of THF were distilled into bulb A from NaK_2. The mixture in bulb A was then maintained at −78 °C and allowed to stir for 24 h. The solution in bulb A changed color to a dark burgundy, signifying that the verbenoxy-COT had been reduced to the dianion,

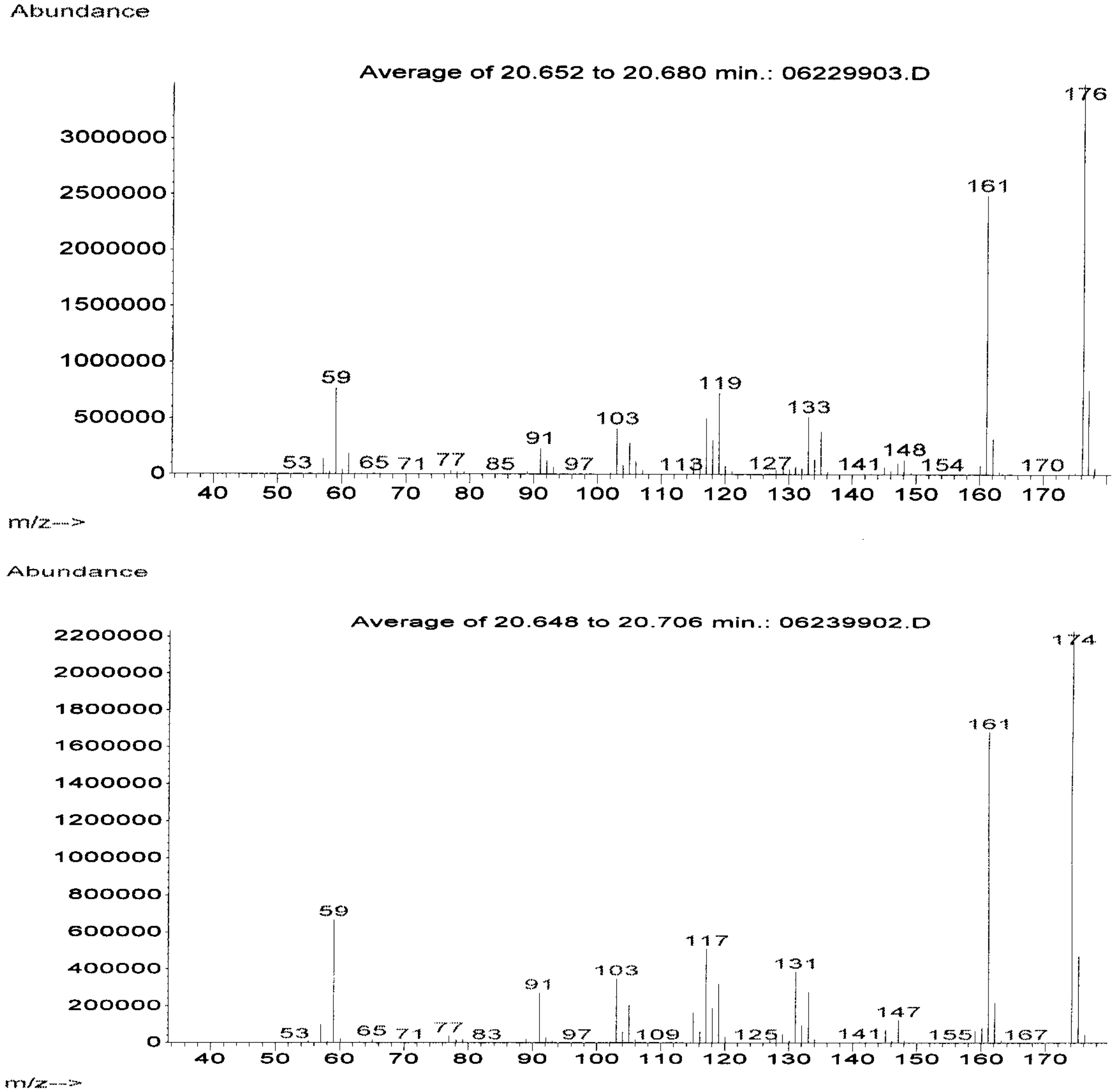


Figure 4. (Upper) Mass spectrum of dideuterioibuprofoxymethoxyethoxyethane (IV-d_2). Note the intense peak at $m/z = 176$ corresponding to $C_{13}H_{16}D_2^+$. The fragility of the ether linkages renders the molecular ion unobservable. (Lower) Mass spectrum of ibuprofoxymethoxyethoxyethane (IV-h_2). These spectra were obtained using a GC-mass spectrometer. The elution time for the material rendering the mass spectral data appears above the respective spectrum.

reaction 2. The verbenoxy-COT dianion appeared as a solid dark burgundy salt at the bottom of tube A. The THF was then removed under vacuum while the mixture was maintained at ≤−40 °C. Hexane (10 mL) was then distilled from NaK_2 into tube A, and the mixture was stirred for several minutes. Allowing the solid salt mixture to settle to the bottom of tube A for several minutes resulted in a clear hexane phase over the solid salt. This clear hexane phase was decanted from the solid verbenoxy-COT dianion salt and poured through the porous glass frit into tube B. An additional 10 mL of dry hexane was distilled into tube A, and the above hexane extraction process was repeated. The resulting mixture in tube B was labeled Phase I. A saturated solution of I_2 in 10 mL of hexane was then added to tube A, reoxidizing the dianion to the neutral state. ($C_{10}H_{14}-O-COT^{2-},2M^{+}:2IV + I_2 \rightarrow C_{10}H_{14}-O-COT + 2MI + 2IV$). This reoxidation freed the cosolvent in tube A. The resulting mixture in tube A was labeled Phase II.

Samples of the original ibuMEE mixture, Phase I, and Phase II were then subjected to GC mass spectral analysis. The mass spectrometer was operated in single ion mode to detect only fragment ions of m/z of 174.10 ± 0.9 and 176.20 ± 0.9. These ions were chosen because the $m/z = 174$ peak is extremely small in the spectrum of the dideuterated material (Figure 4). The ratio of the fragment ions 176.20:174.10 in each sample is the same as the ratio of ibuMEE-d_2:ibuMEE in each sample. Multiple (5−10) injections of each sample were made, and an average ratio ± standard deviation for each sample was recorded. These ratios were then used to calculate the separation factor (α) obtained from each experiment.

Instrumentation. Typical X-band EPR acquisition parameters; data were obtained at 9.8 GHz. Modulation frequency: 60.00 kHz, modulation amplitude: 0.05 G.

Acknowledgment. We thank the National Science Foundation (Grant 9617066) for support of this work.

JO000972P