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Measurement of the Dimerization Equilibrium Constants of Enantiomers

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The formation of homochiral and heterochiral dimers in solutions of enantiomers causes a number of remarkable phenomena, such as the enantiomeric enrichment of nonracemic mixtures on achiral chromatographic columns when chiral monomers and dimers exhibit different retention behavior. In this work, such effects on optical rotation and UV absorbance are analyzed theoretically and experimentally. In particular, it is demonstrated how optical rotation measurements can be used to estimate the dimerization equilibrium constants. It is shown how significant this is for polarimeter calibration, as well as for the determination of enantiomer concentrations in process streams, when optical rotation and UV absorbance measurements are combined. All experimental measurements refer to the enantiomers of the chiral 2,2′-dihydroxy-1,1′-binaphthyl in chloroform, at a temperature of 23 °C. The measured values of its dimerization equilibrium constants are 1.3 ± 0.5 and 3.1 ± 1.0 L/mol for the formation of homochiral and of heterochiral dimers, respectively.

1. Introduction

Enantiomers in solution may exhibit association, particularly by forming dimers. As an example, enantiomers carrying −OH groups may form dimers through hydrogen bonding when dissolved in apolar solvents. This is responsible for other remarkable phenomena, such as nonlinear behavior of optical rotation[1,2] and UV absorbance,[3] nonlinear effects in asymmetric catalysis,[4−7] as well as enantiomeric enrichment through chromatography on achiral stationary phases.[8−16] Among other examples, the enantiomeric purity of nonracemic mixtures of the enantiomers of the chiral 2,2′-dihydroxy-1,1′-binaphthyl (bi-naphthol in the following) in different solvents has been increased through chromatography on different silica gel based achiral columns.[12,15] As shown in Figure 1, in the case of the enantiomers of bi-naphthol in chloroform using an achiral Licrosphere 100 NH_2 column, we have recently been able to collect two fractions constituted of the pure enantiomer present in excess (reaching 100% e.e.) and of the racemic mixture, respectively.[17] An independent estimation of the dimerization equilibrium constants would be rather helpful in the context of preparative chromatography,[15,18] but this is not available in general and in particular for the bi-naphthol system. The dimerization equilibrium constants of a methylzinc alkoxide have been measured through vapor pressure osmometry, but because of the difficulties of the technique, the authors wish that a more sensitive method be developed.[5]

The goal of this paper is to present a method, which allows estimating with a rather good accuracy the chemical equilibrium constant of the dimerization reactions occurring between enantiomers in solution. This is based on using a polarimeter and analyzing its signal while accounting for the association behavior of the enantiomers in solution. Bi-naphthol in chloroform will be used as the model system through the whole paper, but the method is completely general. The enantiomerism of bi-naphthol

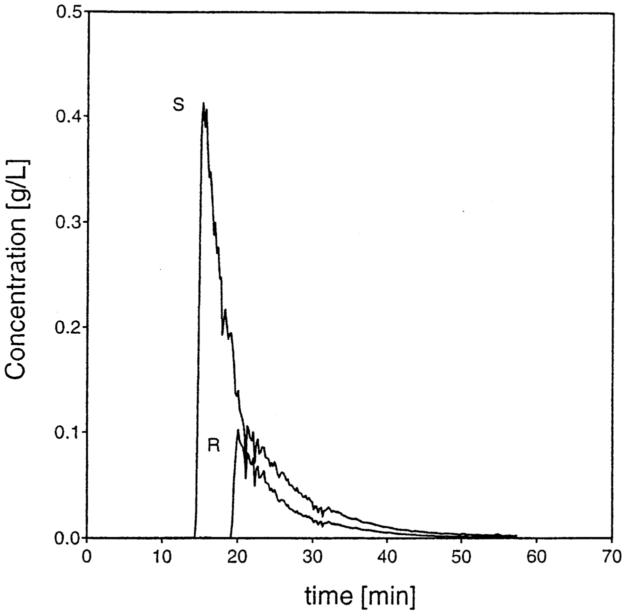

Figure 1. Experimental elution profiles on an achiral Licrosphere 100 NH_2 column for a 100 μL pulse of an 80/20 solution of (*S*)-(−)- and (*R*)-(+)-bi-naphthol at 30 g/L total concentration in chloroform. Concentration profiles have been obtained by sampling the outlet stream periodically (five samples per minute) and analyzing the samples using a HP 1100 liquid chromatograph (Hewlett-Packard, Palo Alto, CA) equipped with a Chiral NEA column (YMC, Kyoto, Japan) as discussed in detail elsewhere.[17]

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is that typical of all bi-phenyls and bi-naphthyls (such as 1,1′-bi-naphthyl) and is called atropisomerism.[19] Carrying two −OH groups, each enantiomer molecule has the possibility of forming hydrogen bonds with other molecules in solution provided that these are not shielded by the solvent. After presenting the experimental materials and methods, experimental evidence of association of bi-naphthol enantiomers in solution is provided. Then a theory is derived how to account for dimerization in analyzing polarimeter and UV signals. Applying this new approach allows us to interpret the experimental data in a consistent way and to estimate the dimerization equilibrium constants. Finally, the general value and potential applications of these findings are discussed.

2. Materials and Methods

Materials. Enantiomers of 2,2′-dihydroxy-1,1′-binaphthyl (bi-naphthol) were purchased from Fluka Chemie (Buchs, Switzerland). Enantiomeric purities of (*R*)-(+)-bi-naphthol and (*S*)-(−)-bi-naphthol were 99.8% and 99.9%, respectively; enantiomeric purities of bi-naphthol enantiomers have been checked through HPLC on a chiral NEA column (25 × 0.46 cm i.d., spherical 5 μm particles, 30 nm average pore size) purchased from YMC (Kyoto, Japan) using a mixture of 90% acetonitryl and 10% methanol as mobile phase.[17] Racemic 1,1′-binaphthyl (98% purity) was purchased from Acros organics (Geel, Belgium). Chloroform (99.8% purity, with less than 0.01% water) and methanol (99.8% purity) were purchased from J. T. Baker (Deventer, The Netherlands); ethanol (99.8% purity) was purchased from Fluka Chemie (Buchs, Switzerland). The Tröger's base enantiomers were purchased from Fluka Chemie (Buchs, Switzerland); enantiomeric purities of the (+) and (−) enantiomers after recrystallization from ethanol were above 99.8%, as checked through HPLC using a Chiralcel OJ (Daicel, Japan) column (25 × 0.46 cm i.d.) with a mixture of 50% ethanol and 50% *n*-hexane as mobile phase.[20]

Methods. Optical rotation was measured by a Polarimeter Jasco OR-990 (Jasco International Co. Ltd, Tokio, Japan), equipped with a tapered flow cell having a 0.25 dm path length. The detector uses a 150 W Hg−Xe lamp having the line spectra on a wide wavelength range, i.e., from 350 to 900 nm. The detection limit of the polarimeter allowed us to determine the enantiomeric enrichment for pulse experiments at concentrations larger than 30 g/L. A UV detector Jasco UV-970 (Jasco International Co. Ltd, Tokio, Japan) was also used for UV absorbance measurements, which was provided with a preparative cell, with a selected wavelength equal to 308 nm. All measurements have been carried out at 23 ± 0.2 °C.

The ^{1}H NMR spectra and the diffusion measurements were performed on a Bruker AVANCE 400 MHz spectrometer equipped with a microprocessor controlled gradient unit and a multinuclear probe with an actively shielded Z-gradient coil. For the NMR studies, the molecules were dissolved in deuteriochloroform ($CDCl_3$) and measured at 296 K without spinning. The shape for the gradients was rectangular, their length was 5 ms, the strength (G) was varied in the course of the experiments, from 0.56 to 21.28 G/cm, every 0.56 G/cm, and the diffusion time was equal to 70 ms. The concentrations were 5.0 g/L, for the pure (*S*)-(−)-bi-naphthol enantiomer, 3.9 g/L for the racemic mixture (2.0 and 1.9 g/L of R and S enantiomer respectively), and 7.0 g/L for 1,1′-binaphthyl.

3. Experimental Evidence of Dimerization

3.1. NMR Investigation. To the aim of elucidating the aggregation state of bi-naphthol, as pure enantiomers or in a racemic mixture, a NMR investigation has been carried out in a $CDCl_3$ solution. The ^{1}H NMR spectra of bi-naphthol, either racemic mixture or pure enantiomer *S*, are reported in Figure 2 parts a and b, respectively. These figures show clearly that the differences in the chemical shifts as well as in the coupling constants are within the experimental error, except for the hydroxyl protons signal for the pure *S* enantiomer, which is broader than that of the racemic mixture. This can be due to a faster exchange with some traces of water that might be present in the $CDCl_3$ solution. These spectra are consistent with those reported previously, which when superimposed are also coincident within the experimental error.[15] However, these provide no conclusive evidence about the existence or nonexistence of dimers in solution nor of any diastereomeric interaction in the racemic solution. Possibly a very fast equilibrium between monomeric and dimeric species takes place in solution, so that each of the obtained proton spectra represents an average.

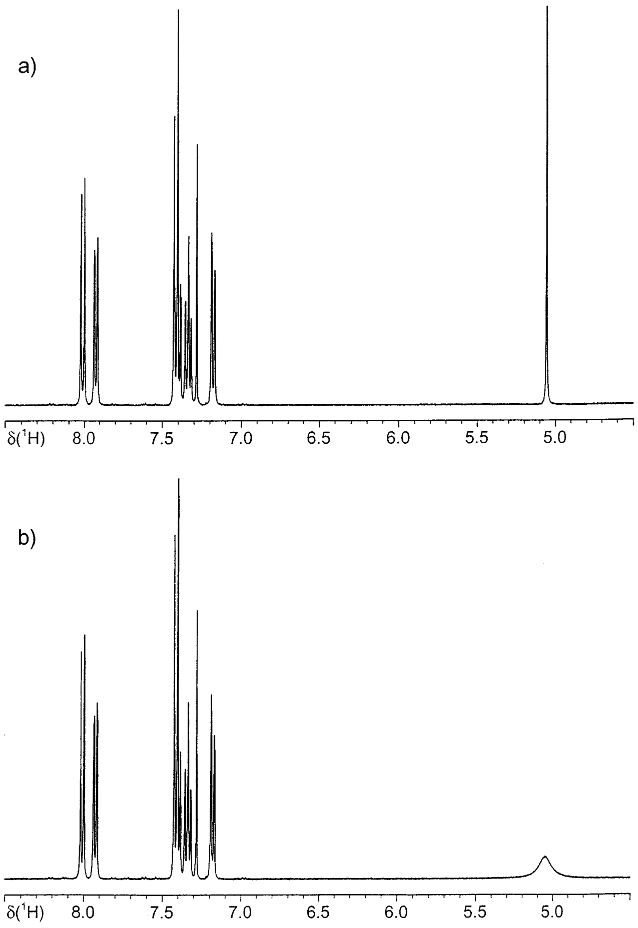

Figure 2. ^{1}H NMR spectra of bi-naphthol, racemic (a), and pure (*S*)-(−) enantiomer (b) in a $CDCl_3$ solution.

To clarify whether the bi-naphthol, pure enantiomer, or racemic mixture is present in $CDCl_3$ solution as a dimer, its diffusion coefficient was measured by NMR, using the FT pulsed field gradient spin−echo (FT-PGSE) experiment,[21,22] and compared to that of 1,1′-binaphthyl. In fact, the latter has no hydroxyl group available for hydrogen bonding and is supposed to be present in solution as a monomer, but its molecular dimensions and chemical structure are similar to those of 1,1′-bi-2-naphthol. The results of this experiment are illustrated in Figure 3, where the relative variation of the NMR signal intensities, i.e., $\ln(I/I_0)$, is plotted as a function of the square of the gradient amplitude, G. The experimental data were fitted by a linear releationship. The slope of the fitting lines has been

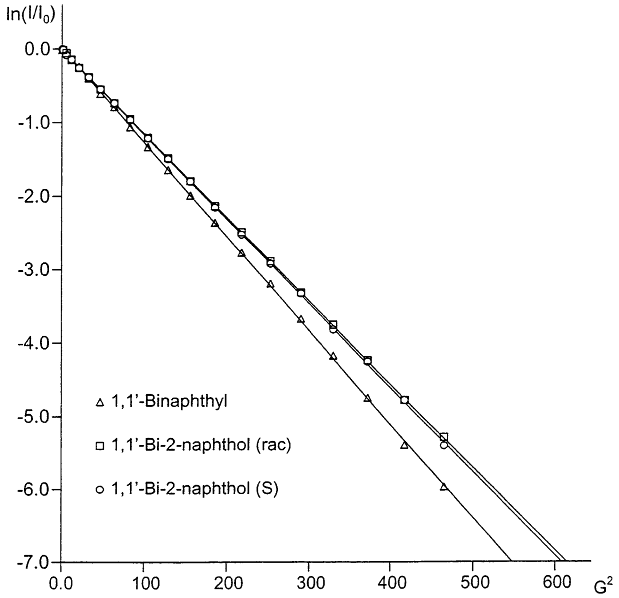

Figure 3. Plot of $\ln(I/I_0)$ of 1,1′-binaphthyl (△), racemic bi-naphthol (◇), and pure (*S*)-(−)-bi-naphthol enantiomer (□) vs the square of the gradient amplitude, G^2, in G^2/cm^2.

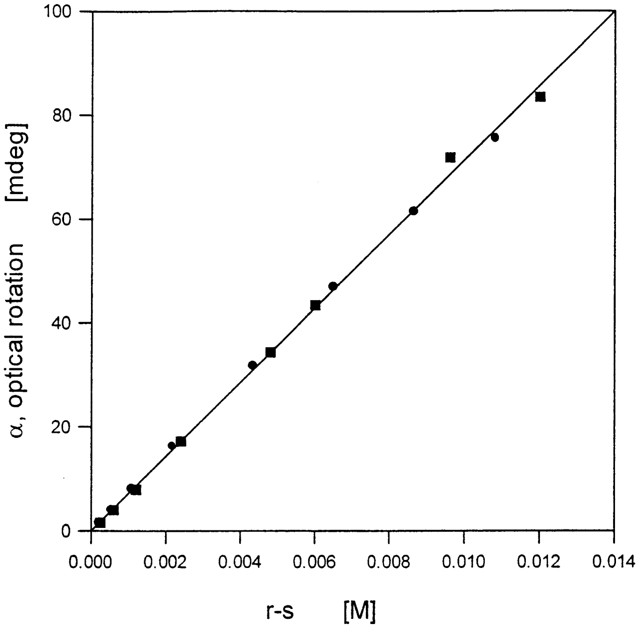

Figure 4. Optical rotation vs difference of the nominal enantiomer concentrations ($r - s$) for different mixtures of the Tröger's base enantiomers in ethanol. Enantiomer concentrations *r* and *s* are in the ranges 0.00024−0.01430 M, and 0.00007−0.00350 M, respectively. Symbols: (■) $q = \infty$; (●) $q = 4$; the line linearly interpolates all the experimental data.

evaluated to be −0.0112 for the *S* enantiomer, −0.0111 for the mixture of the *S* and *R* enantiomers, and −0.0125 for 1,1′-binaphthyl. These values are directly proportional to the diffusion coefficient, which is inversely proportional to the molecule diameter according to the Stokes−Einstein equation.[22]

Figure 3 shows that racemic and pure *S* enantiomer solutions of bi-naphthol in $CDCl_3$ have the same slope and therefore the same diffusion coefficient, i.e., 1.055×10^{-5} and 1.061×10^{-5} cm^2/s, respectively, within the experimental error of $\pm 6 \times 10^{-8}$ cm^2/s. On the contrary, 1,1′-binaphthyl has a larger absolute value of the slope and , hence, a larger diffusion coefficient of 1.178×10^{-5} cm^2/s and a smaller molecular diameter (about 10% smaller). Because this is not justified by the small differences in molecular structure, these results can be explained by concluding that bi-naphthol is present in solution not only in monomeric form but also as dimers. The *R* enantiomer exhibits of course the same behavior as the *S* enantiomer. These measurements prove that dimerization occurs and indicate that this involves a significant fraction of the molecules in solution. A more precise evaluation of the dimerization degree is not possible with this technique, because of the complex geometry of the molecules. In fact, it is difficult to estimate properly the molecular volume of monomers and dimers of bi-naphthol to be used in the Einstein equation for the estimation of the diffusion coefficient. It is worth noting that the assumption that 1,1′-binaphthyl enantiomers do not dimerize is not necessary to reach the conclusions above. In fact, if 1,1′-binaphthyl forms no dimers, the results above show that bi-naphthol does; if on the contrary 1,1′-binaphthyl does dimerize, the results above prove that bi-naphthol does also but to a larger extent. In both cases, we can conclude that bi-naphthol enantiomers forms indeed dimers in solution.

3.2. Polarimetric Effects. Let us consider a solution of the *R* and *S* enantiomers at the molar concentrations *r* and *s*, respectively. It is worth noting that such a composition can also be defined through the overall molar concentration $c = r + s$ and the nominal enantiomeric ratio $q = r/s$. Inverting these definitions yields

$$r = \frac{qc}{q + 1} \tag{1}$$

$$s = \frac{c}{q + 1} \tag{2}$$

Therefore, the nominal enantiomeric excess is

$$e.e. = \frac{r - s}{r + s} = \frac{q - 1}{q + 1} \tag{3}$$

The optical rotation measured by the polarimeter is in principle proportional to the difference ($r - s$), irrespective of the specific values of *c* and *q*. This standard behavior is illustrated in Figure 4, where several values of the optical rotation of different solutions of the enantiomers of the Tröger's base in ethanol are plotted as a function of ($r - s$); despite two sets of sample solutions having two different *q* values have been considered, all measurements lay on the same straight line. A different behavior is shown in Figure 5, where the optical rotation of five different groups of solutions of bi-naphthol enantiomers in chloroform (each having constant *q* value, namely, 1.49, 2.35, 3.76, 8.61, and ∞, and different overall concentration *c*) is plotted versus ($r - s$). The five sets of points belong to the same straight line (broken line in the figure) only at very low concentration, whereas at higher concentration, these lay on five distinct curved lines. This behavior can be explained by considering that bi-naphthol enantiomers associate, forming dimers that have optical activity as well and perturb the normal linear response of the polarimeter. The larger the difference $r - s$, the larger this nonlinear effect, which is evident in our case only beyond the threshold $r - s \approx 0.01$ M. This effect has already been reported in the literature and explained qualitatively;[1,2] in the next section, we will discuss it in a quantitative way.

4. Theory and Application

4.1. Dimerization. In the case of the two enantiomers *R* and *S* in solution, three dimerization reactions may take place

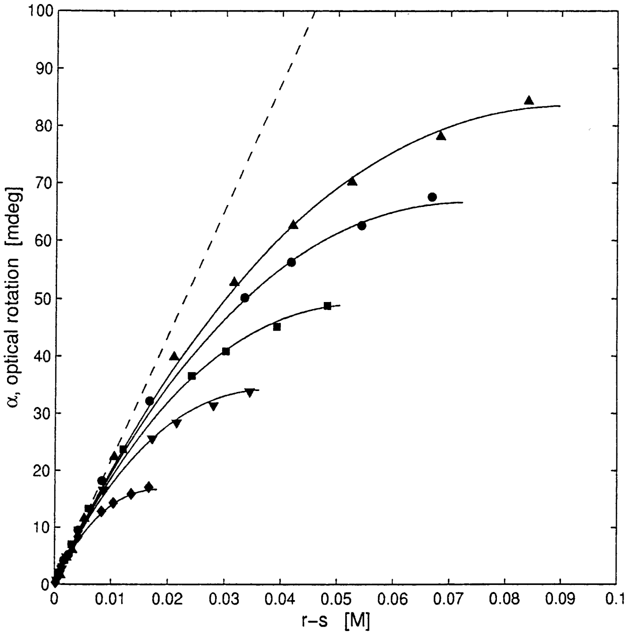

Figure 5. Optical rotation vs difference of the nominal enantiomer concentrations ($r - s$) for different mixtures of the bi-naphthol enantiomers in chloroform. The solid curves are calculated using eq 18 with the parameter values $K_{\text{homo}} = 1.3$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = -11000$ mdeg L/mol; $K_{\text{hetero}} = 3.1$ L/mol. The dashed line is the tangent to the experimental data in the origin. Enantiomer concentrations r and s are in the ranges 0.00084−0.06300 M and 0.00014−0.01400 M, respectively. Symbols: (◆), $q = 1.49$; (▼), $q = 2.35$; (■), $q = 3.76$; (●), $q = 8.61$; (▲), $q = \infty$.

according to the following scheme, thus leading to both the homochiral dimers (*RR* and *SS*) and the heterochiral dimer (*RS*):

$$R + R \rightleftharpoons RR \tag{4}$$

$$S + S \rightleftharpoons SS \tag{5}$$

$$R + S \rightleftharpoons RS \tag{6}$$

The homochiral *RR* and *SS* dimers are also enantiomers, whereas *RR* or *SS* and the heterochiral *RS* dimer are diastereomers. Because we assume that all reactions are fast, due to symmetry, only the two reaction equilibrium constants, i.e., $K_{\text{homo}} = K_{\text{RR}} = K_{\text{SS}}$ and $K_{\text{hetero}} = K_{\text{RS}}$, are needed to describe the reacting system.

When a solution of the *R* and *S* enantiomers is prepared at the molar concentrations r and s, respectively, the following material balances apply because of dimers' formation:

$$r = m_R + 2d_{RR} + d_{RS} \tag{7}$$

$$s = m_S + 2d_{SS} + d_{RS} \tag{8}$$

where m_R and m_S and d_{RR}, d_{SS}, and d_{RS} represent actual monomer and dimer molar concentrations, respectively. Assuming equilibrium of the dimerization reactions, and ideal behavior of the liquid phase, i.e., $d_{RR} = K_{\text{homo}} m_R^2$, $d_{SS} = K_{\text{homo}} m_S^2$, and $d_{RS} = K_{\text{hetero}} m_R m_S$, these can be recast as a one-to-one mapping between overall concentrations and monomer concentrations:

$$r = m_R + 2K_{\text{homo}} m_R^2 + K_{\text{hetero}} m_R m_S \tag{9}$$

$$s = m_S + 2K_{\text{homo}} m_S^2 + K_{\text{hetero}} m_R m_S \tag{10}$$

These can be inverted, yielding only one pair of positive values (m_R, m_S), which can be cast in the form of a function of the concentrations r and s, depending on the values of the two equilibrium constants as follows:

$$m_R = m_R(r, s; K_{\text{homo}}, K_{\text{hetero}}) \tag{11}$$

$$m_S = m_S(r, s; K_{\text{homo}}, K_{\text{hetero}}) \tag{12}$$

Thus summarizing, five species are present in solution, namely, the enantiomers *R* and *S* and the dimers *RR*, *SS*, and *RS*. However, thanks to the three dimerization equilibria, only two parameters are needed to characterize the composition of the solution; these can be either the two overall concentrations (r, s), or the monomer concentrations (m_R, m_S), or alternatively also the two parameters (c, q). The equations above allow us to obtain from one pair of indepedent variables all of the others.

Two special cases are worth considering, which are important in the following developments.

Case I. The first special case refers to a mixture where only one enantiomer, say *R*, is present at the nominal concentration r. It follows that no heterochiral dimers can form, and accordingly, the simplified version of eq 9 applies. This can be explicitly inverted, thus yielding the monomer concentration as a function of r and K_{homo}:

$$m_R = \frac{2r}{1 + \sqrt{1 + 8K_{\text{homo}} r}} \tag{13}$$

If only the enantiomer *S* is present, the same equation is obtained with m_S and s replacing m_R and r, respectively.

Case II. The second special case deals with the situation where both enantiomers are present, as in the general case, but the two equilibrium constants are not independent, namely, $K_{\text{hetero}} = 2K_{\text{homo}}$. Then, only one parameter characterizes the three dimerization reactions at equilibrium, i.e., K_{homo}.[18] This condition is equivalent to assuming that the enthalpy of formation of the homochiral and of the heterochiral dimers is the same, whereas their absolute molar entropies differ by a factor $R \ln 2$ as it can be demonstrated through statistical mechanics arguments.[23] In this special case, where $K_{\text{hetero}} = 2K_{\text{homo}}$, eqs 9 and 10 can be explicitly inverted, thus yielding

$$m_R = \frac{2r}{1 + \sqrt{1 + 8K_{\text{homo}}(r + s)}} \tag{14}$$

$$m_S = \frac{2s}{1 + \sqrt{1 + 8K_{\text{homo}}(r + s)}} \tag{15}$$

which represent a special form of eqs 11 and 12.

4.2. Optical Rotation. *4.2.1. Theory.* The optical rotation of the solution represents the measurement of the combined effect of all optical active species present. In this case, these are the two monomeric enantiomers *R* and *S* and the two homochiral dimeric enantiomers *RR* and *SS*. The heterochiral dimer is not optically active and plays no role here. Each pair of enantiomers contributes to the optical rotation of the solution to an extent proportional to the difference between the concentration of the two enantiomers themselves. Therefore, because optical rotation is additive, its instrumental reading is given by the following relationship:

$$\alpha = \mu(m_{\text{R}} - m_{\text{S}}) + \delta(d_{\text{RR}} - d_{\text{SS}}) \tag{16}$$

where the response parameters μ and δ for monomers and dimers, respectively, will in general be different, in principle also in their sign.[2,7] Of course the operating conditions of the polarimeter (see section 2.2) are kept constant for all the measurements.

By substituting the chemical equilibrium relationships, i.e., $d_{RR} = K_{\mathrm{homo}} m_R^2$, and $d_{SS} = K_{\mathrm{homo}} m_S^2$, eq 16 can be recast in terms of the concentrations of the two enantiomers R and S as

$$\alpha = \mu(m_R - m_S) + \delta K_{\mathrm{homo}}(m_R^{\ 2} - m_S^{\ 2}) \tag{17}$$

It is clear that such a relationship cannot be cast in the form of a linear function of $(r - s)$, unless there is no dimerization, i.e., either for $K_{\mathrm{homo}} = K_{\mathrm{hetero}} = 0$ or at very low concentration, where the quadratic terms in the previous equation become negligible. This result is sufficient to explain the different behavior of Tröger's base and bi-naphthol illustrated in Figures 4 and 5; the enantiomers of the former do not associate, whereas those of the latter do. Moreover, from eqs 11 and 12 it follows that the optical rotation is a function of the nominal enantiomer concentrations and of four parameters:

$$\alpha = \alpha(r, s; K_{\mathrm{homo}}, K_{\mathrm{hetero}}, \mu, \delta) \tag{18}$$

The last equation indicates that the equilibrium constants K_{homo} and K_{hetero}, as well as the response parameters μ and δ, can be determined by fitting optical rotation values measured for different values of r and s, i.e., different overall concentration c and enantiomeric ratio q, using eq 18.

Case I. In this case, the last equation can be significantly simplified. In fact, knowing that only one enantiomer and the corresponding dimer are present and using eq 13, eq 17 simplifies to

$$\alpha = \mu m_R + \delta K_{\mathrm{homo}} m_R^{\ 2} = \frac{2r}{1 + \sqrt{1 + 8K_{\mathrm{homo}} r}}\left(\mu + \frac{2r\delta K_{\mathrm{homo}}}{1 + \sqrt{1 + 8K_{\mathrm{homo}} r}}\right) \tag{19}$$

This is remarkable in that it involves only three parameters out of the four present in eq 18, namely, the response coefficients μ for monomers and δ for dimers and the homo-dimerization equilibrium constant K_{homo}.

Case II. The case where $K_{\mathrm{hetero}} = 2K_{\mathrm{homo}}$ exhibits a qualitatively different behavior. In fact, substituting eqs 14 and 15 into eq 17 yields the following relationship:

$$\alpha = \frac{q - 1}{q + 1}(\mu m + \delta K_{\mathrm{homo}} m^2) \tag{20}$$

where m represents the overall molar concentration of the enantiomers in monomeric form and is given, using again eqs 14 and 15, by

$$m = m_R + m_S = \frac{2c}{1 + \sqrt{1 + 8K_{\mathrm{homo}} c}} \tag{21}$$

It is worth noting that $c = m(1 + 2K_{\mathrm{homo}} m)$ and that the extent of dimerization is $(c - m)/c = 2K_{\mathrm{homo}} m/(1 + 2K_{\mathrm{homo}} m)$. Equation 20 establishes that in case II the optical rotation is given by the product of the ratio $(q - 1)/(q + 1)$, which according to eq 3 represents the nominal e.e. of the solution depending only on q, with the term in brackets, which is a rather complex function of the nominal overall concentration c, but can be cast as a simple quadratic relationship in terms of the actual overall monomer concentration m. Furthermore, for measurements such as those reported in Figure 5, eq 20 indicates that plotting $\alpha(q + 1)/(q - 1)$ vs m yields a single curve, i.e., a parabola, whatever the enantiomeric ratio q is. This exhibits the same functional dependence as eq 19, with c replacing r, and depends on the same three parameters, i.e., μ, δ, and K_{homo}.

4.2.2. Parameter Estimation. According to the analysis above, and particularly to eq 18, the optical rotation depends on the two equilibrium constants K_{homo} and K_{hetero} and on the response parameters μ and δ. In principle, these could be estimated altogether by fitting all of the optical rotation measurements, e.g., those in Figure 5, using eq 18. In practice, it is more convenient to estimate the four parameters independently, by exploiting the limit behavior of optical rotation under special conditions and by adopting the following three step procedure.

Step 1. At very low concentration, the extent of dimerization is negligible, and optical rotation is proportional to $(r - s)$, with a proportionality constant given by μ, whatever the value of the nominal enantiomeric ratio. This is demonstrated by eq 16 and illustrated in Figure 5. By linearly regressing all of the low concentration data, the dashed straight line through the origin in Figure 5 is obtained, whose slope is the response parameter μ.

Step 2. For a pure enantiomer, i.e., case I above, optical rotation depends only on three parameters, i.e., μ, δ, and K_{homo}, according to eq 19. Because μ is known at this stage, the other two parameters can be estimated by using only the pure enantiomer measurements, i.e., those where $q = \infty$. This is done by setting a value of K_{homo}, calculating m_{R} from r using eq 13, estimating the best-fit parameter δ as the coefficient of the second-order term in the quadratic relationship of eq 19, and determining the corresponding error, i.e., the sum of the squares of the difference between the measured value of α and the calculated right-hand side of eq 19. Absolute errors are used, to give more weight to the high concentration measurements, which are more sensitive to the values of K_{homo} and of δ. The K_{homo} value corresponding to the smallest error is the desired dimerization equilibrium constant.

Step 3. The last parameter, i.e., the equilibrium constant of the heterochiral dimerization K_{hetero}, is estimated by fitting all optical rotation measurements involving both enantiomers at different overall concentration c and enantiomeric ratio q, with $0 < q < \infty$, using the general eq 18. Also in this case, and for the same reasons as in step 2, the absolute error is used.

4.2.3. Application to Bi-naphthol. Following the procedure outlined in the previous section, first the response parameter for the enantiomers in monomeric form has been estimated through linear regression of the low concentration measurements to be $\mu = 2190 \pm 130$ mdeg L/mol. Confidence limits in this section are calculated for a confidence level of 90% with an estimated variance of the polarimeter measurement error of 1 mdeg. The corresponding straight line has been drawn as a dashed line in Figure 5. Second, the pure enantiomer measurements, corresponding to $q = \infty$ in Figure 5, have been used to estimate the homo-dimerization equilibrium constant K_{homo}, as well as the response factor for the dimers δ. The results of the nonlinear regression are illustrated in Figure 6a, where the sum of the squares of the absolute errors between experimental and calculated values is plotted vs K_{homo}. The best fit values are $K_{\mathrm{homo}} = 1.3 \pm 0.5$ L/mol, and $\delta = -11000 \pm 200$ mdeg L/mol. Rather interestingly, the values of μ and δ have opposite sign. It is worth noting that, though not necessarily general, the same behavior was observed previously in the case of the enantiomers of 1-ethyl-1-methylsuccinic acid and its dimers[2]. Finally, using the optical rotation measurements in binary solutions, the best fit between experimental data and values calculated with eq 18

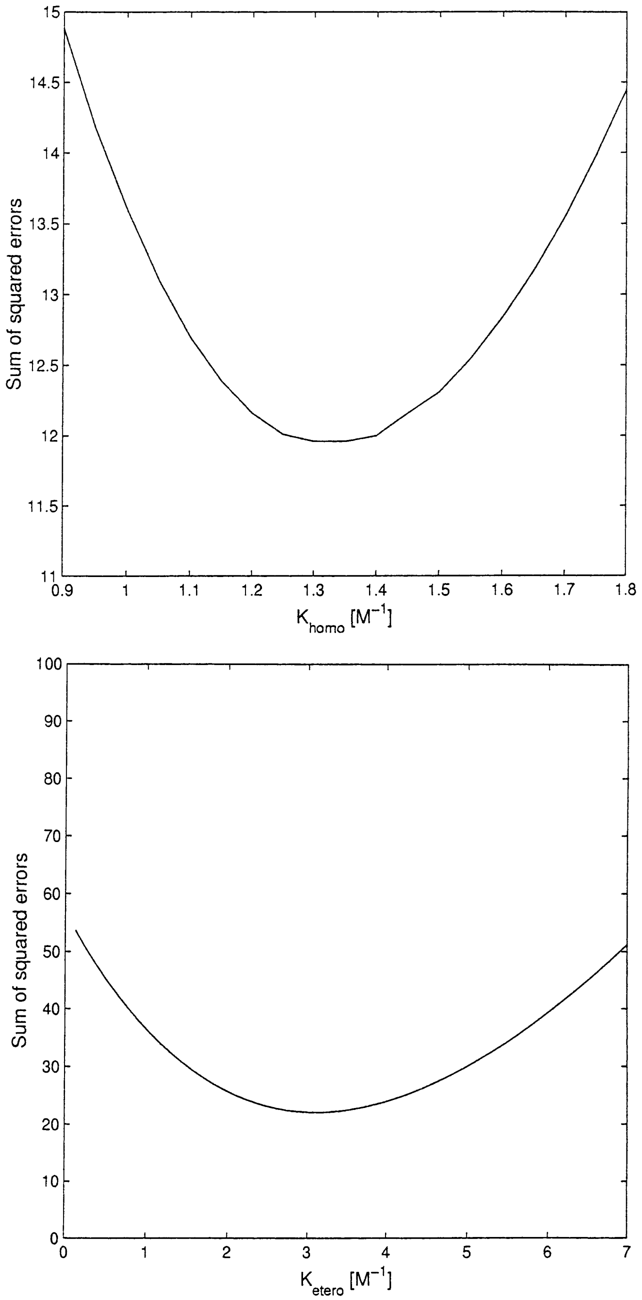

Figure 6. Estimation of the dimerization equilibrium constants of binaphthol. Sum of the squares of the absolute errors between experimental and calculated values of the optical rotation as a function of the relevant constant. (a) Estimation of K_{homo} (step 2 of the procedure in section 4.2.2; only data with $q = \infty$ are considered). (b) Estimation of K_{hetero} (step 3 of the procedure in section 4.2.2; all experimental data with $q < \infty$ are considered).

is obtained for the value $K_{\text{hetero}} = 3.1 \pm 1.0$ L/mol, as illustrated in Figure 6b. This indicates that the heterochiral dimer is about 2 times more stable than the homochiral dimer; the Gibbs free energy changes upon dimerization at 296 K are $\Delta G_{\text{homo}} = -0.65$ kJ/mol and $\Delta G_{\text{hetero}} = -2.78$ kJ/mol. It follows that the difference in the reaction enthalpy between the homo- and hetero-dimerization is $\Delta H_{\text{homo}} - \Delta H_{\text{hetero}} = \Delta G_{\text{homo}} - \Delta G_{\text{hetero}} + T(\Delta S_{\text{homo}} - \Delta S_{\text{hetero}}) = -0.65 + 2.78 - RT \ln 2 = 0.42$ kJ/mol.

The experimental optical rotation data (symbols) and the fitted relationship (eq 18; solid lines) are compared in Figure 5. It can be readily observed that the model accuracy is rather satisfactory for all sets of data, in the whole range of concentrations explored; it is worth noting that the highest enantiomer concentration values that could be reached were limited by their solubility in chloroform.

4.3. UV Absorbance. *4.3.1. Theory and Parameter Estimation.* Both monomers and dimers contribute to the total absorbance of the solution, as theoretically discussed in the literature with reference to a single species yielding a single dimer[3]. In the present case, there are five species, and because enantiomers are indistinguishable in terms of UV signal, the total absorbance is given by

$$A = M(m_R + m_S) + D(d_{RR} + d_{SS}) + Ed_{RS} \qquad (22)$$

where M, D, and E are positive constants, in general different. By substituting the chemical equilibrium relationships, i.e., $d_{RR} = K_{\text{homo}}m_R^2$, $d_{SS} = K_{\text{homo}}m_S^2$, and $d_{RS} = K_{\text{hetero}}m_Rm_S$, this can be recast in terms of the concentrations of the two enantiomers R and S as

$$A = M(m_R + m_S) + DK_{\text{homo}}(m_R^2 + m_S^2) + EK_{\text{hetero}}m_Rm_S = A(r, s; K_{\text{homo}}, K_{\text{hetero}}, M, D, E) \qquad (23)$$

where eqs 11 and 12 have been used. Once the equilibrium constants, i.e., K_{homo} and K_{hetero}, are known, the values of the parameters M, D, and E can be estimated by fitting UV absorbance measurements for different values of r and s using the last equation.

As in the case of optical rotation, the three parameters can be estimated sequentially. First, M can be obtained by linearly regressing low concentration measurements, where the second and third terms in eq 23 are negligible. Then, D is estimated by simple quadratic regression using single enantiomer measurements by noting that in this case, i.e., case I, where $m_S = 0$, eq 23 reduces to a second-order polynomial in m_R. Finally, binary mixture UV absorbance data can be used to estimate the last parameter E through the complete form of eq 23.

Also in this case a particular behavior is observed in the special case II, where $K_{\text{hetero}} = 2K_{\text{homo}}$. In fact, by substituting eqs 14, 15, and 21 into eq 22, the following relationship in the overall monomer concentration m is obtained:

$$A = Mm + \left[\frac{D(1 + q^2) + 2Eq}{(1 + q)^2}\right]K_{\text{homo}}m^2 \qquad (24)$$

This depends on both m and q, unless the condition $D = E$ applies and eq 23 reduces to the simple quadratic relationship in m:

$$A = Mm + K_{\text{homo}}Dm^2 \qquad (25)$$

It is worth noting that, using eq 21, the last equation can be used to obtain the absorbance as a function of the overall concentration c, thus obtaining a relationship that is also independent of q.

4.3.2. Application to Bi-naphthol. In Figure 7, the experimental absorbance of the same samples considered in Figure 5 (except those where the UV signal was already overloaded, i.e., where $c \gtrsim 0.06$ mol/L) is plotted as a function of c, i.e., the overall molar concentration. For the sake of clarity, only the data corresponding to $q = 1.49$ and 8.61 are plotted; in fact for the same c, the experimental absorbances at different values of q are very similar. In the same figure, the dotted line is the tangent to the experimental points in the origin; as pointed out above, its slope is given by the parameter M in eq 23, whose value is $M = 32.8 \pm 0.6$ V L/mol. Confidence limits in this

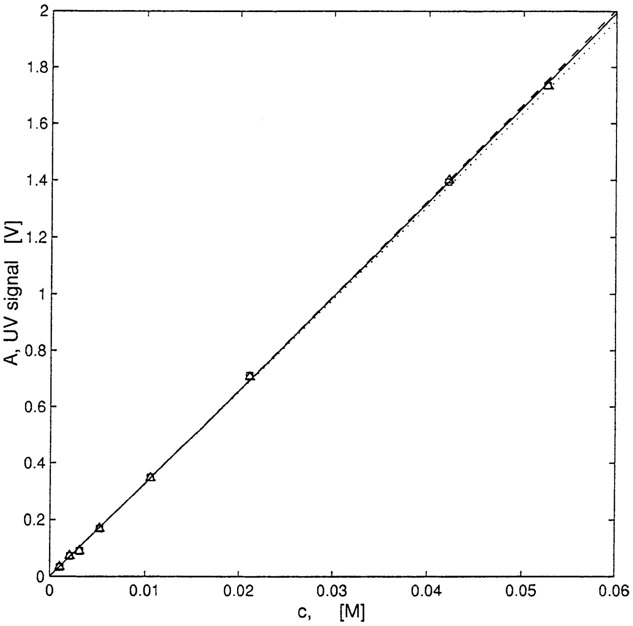

Figure 7. UV absorbance measurements. Experimental values of the UV signal A vs c, together with the interpolating function of eq 23 corresponding to the parameter values $K_{\mathrm{homo}} = 1.3$ L/mol; $M = 32.8$ V L/mol; $D = 71$ V L/mol; $E = 156$ V L/mol; $K_{\mathrm{hetero}} = 3.1$ L/mol. The dotted line is the tangent to the experimental data in the origin. Symbols: (□) and dashed line, $q = 1.49$; (△) and solid line, $q = 8.61$.

section are calculated for a confidence level of 90% with an estimated variance of the absorbance measurement error of 0.01 V. Using single enantiomer measurements first, and then binary measurements, the values $D = 71 \pm 5$ V L/mol and $E = 156 \pm 11$ V L/mol are estimated. Two remarks are worth making, with reference to Figure 7. First, the calculated curves corresponding to the two experimental q values are in satisfactory agreement with the experimental data; hence, they overlap one with the other. Second, the effect of dimerization on UV absorbance is rather weak; in fact, the data at all values of overall concentration c are very close to the straight dotted line. As a consequence, the accuracy in the estimation of the parameters D and E is lower than that in estimating M, as reflected by the amplitude of the corresponding confidence limits reported above.

5. Discussion and Conclusions

5.1. Estimation of the Dimerization Equilibrium Constants. The equilibrium constants for the dimerization of bi-naphthol enantiomers have been estimated through optical rotation measurements, using a method, which is fully general and is based on eq 18. It is worth analyzing the role of the coefficients μ and δ, and particularly of their sign. For the sake of simplicity, but without loss of generality, this is illustrated in Figure 8 with reference to a situation where $K_{\mathrm{hetero}} = 2K_{\mathrm{homo}}$, i.e., case II above; in this case, the simpler explicit eq 20 can be used, and plotting $\alpha(q + 1)/(q - 1)$ vs m yields a single curve, i.e., a parabola, whatever the enantiomeric ratio q is. Curve a corresponds to the case under investigation where μ and δ have opposite sign, whereas curve e applies to systems where they have the same sign (positive in the figure). The straight line d is obtained when no dimerization occurs. It is worth noting that curve a crosses the horizontal axis; that is, there is one value of m or of the overall concentration c according to eq 21, where the solution behaves like a racemic mixture; that is, no optical rotation is observed. In the case where $K_{\mathrm{hetero}} = 2K_{\mathrm{homo}}$, these values are simply $m_0 = -\mu/(K_{\mathrm{homo}}\delta)$ and $c_0 = m_0(1 + 2K_{\mathrm{homo}}m_0)$.

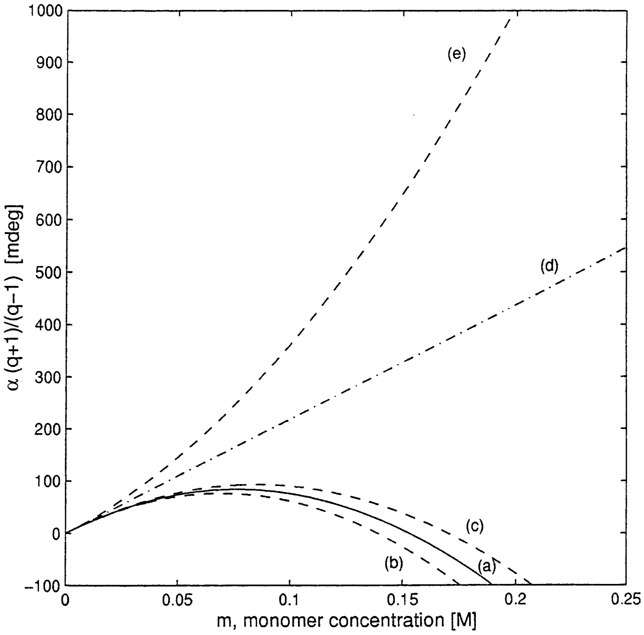

Figure 8. Polarimetric effects due to dimerization. Plot of the quadratic relationship $\alpha(q + 1)/(q - 1)$ vs m of eq 20, for different values of the parameters. Curve (a), $K_{\mathrm{homo}} = 1.3$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = -11000$ mdeg L/mol; (b), $K_{\mathrm{homo}} = 1.43$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = -11000$ mdeg L/mol; (c), $K_{\mathrm{homo}} = 1.17$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = -11000$ mdeg L/mol; (d), $K_{\mathrm{homo}} = 0$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = 0$ mdeg L/mol; (e), $K_{\mathrm{homo}} = 1.3$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = 11000$ mdeg L/mol.

If such a large concentration level could be reached, which is not the case for bi-naphthol in chloroform because of solubility limitations, the accuracy of the proposed estimation method would be higher than when small concentration values are used, i.e., values corresponding to the left-hand side of the parabola with respect to its maximum. This is illustrated in Figure 8 by curves b and c, which have been calculated using the same values of μ and δ as for curve a, but with a value of the equilibrium constant K_{homo} differing by ±10% with respect to the value used for curve a. The three curves are very close to each other until the proximity of the maximum but are wide apart when they approach the intersection with the $\alpha = 0$ axis. In fact, it can easily be shown that $d \ln c_0 = d \ln m_0 = -d \ln K_{\mathrm{homo}}$; that is, the estimation of the equilibrium constant K_{homo} is affected by the same relative error as the measurement of the concentration where optical rotation is zero.

5.2. Optical Purity. Optical purity, o.p., is defined as the ratio between the measured optical rotation of the sample mixture with given values of c and q and that of the enantiomerically pure substance, i.e., of a sample with the same value of c but $q = \infty$.[2,7] Optical purity and enantiomeric excess coincide by definition in the case of nonassociating enantiomers, as can be seen by analyzing eqs 3, 7, 8, and 16. Using eqs 3 and 20 shows that optical purity coincides with the nominal enantiomeric excess also in the case of associating enantiomers where the special condition $K_{\mathrm{hetero}} = 2K_{\mathrm{homo}}$ applies. In fact, e.e. $= f(q)$ and $f(q) \rightarrow 1$ when $q \rightarrow \infty$, and from eq 20, $\alpha = f(q)\, g(c)$; hence, o.p. $= f(q) =$ e.e. as was to be proved. In the general case of associating enantiomers where $K_{\mathrm{hetero}} \neq 2K_{\mathrm{homo}}$, optical purity can be larger or smaller than enantiomeric excess, depending on the specific values of the dimerization equilibrium constants and of the response coefficients μ and δ. This is shown in Figure 9, where beside the diagonal two curves are drawn; they represent optical purity for a system having the same values of K_{homo}, μ, and δ as bi-naphthol but a value of the hetero-

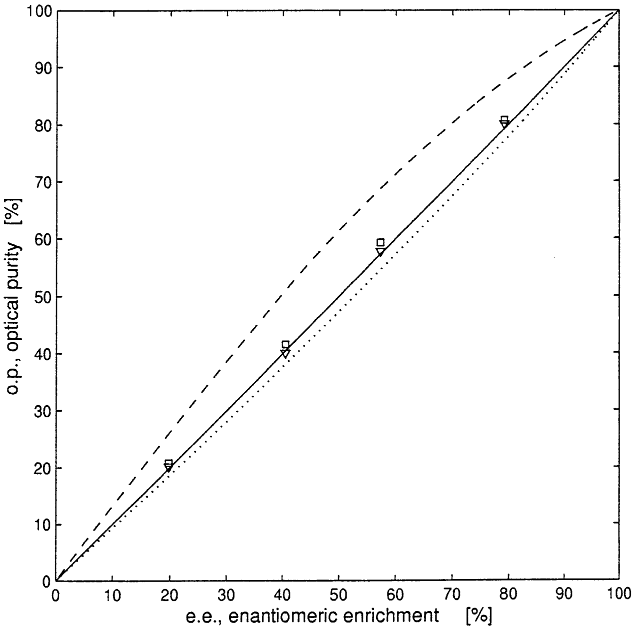

Figure 9. Optical purity vs enantiomeric enrichment. Symbols refer to different overall concentration values, $c = r + s$: (□) $c = 0.021$ M; (▽) $c = 0.084$ M. Lines are calculated with $K_{\mathrm{homo}} = 1.3$ L/mol; $\mu = 2190$ mdeg L/mol; $\delta = -11000$ mdeg L/mol; dashed line: $K_{\mathrm{hetero}} = 25$ L/mol; dotted line: $K_{\mathrm{hetero}} = 0.25$ L/mol.

dimerization equilibrium constant K_{hetero} that is larger (dashed curve, $K_{\mathrm{hetero}} = 25$ L/mol) or smaller (dotted curve, $K_{\mathrm{hetero}} = 0.25$ L/mol) than twice K_{homo}. In the former case, the curve is above the diagonal, whereas in the latter case, it is below. In the same figure, also the measured optical rotation of bi-naphthol corresponding to $c = 0.021$ M (squares) and 0.084 M g/L (triangles) of overall nominal concentration are reported. Accordingly, the experimental points are above the diagonal, although only slightly. It is also worth noting that, as expected when considering eq 18, points corresponding to the same value of e.e., i.e., to the same nominal enantiomeric ratio q, but to different overall concentration c do not coincide. This is particularly evident in Figure 9 where e.e. = 40% ($q = 2.35$) and 58% ($q = 3.76$).

This analysis leads to the conclusion that experimental results reported previously, particularly those about the enantiomers of 1-ethyl-1-methylsuccinic acid, where o.p. differs from e.e., can be interpreted as being due to dimerization of enantiomers in solution, with different values of the reaction enthalpy for the formation of the heterochiral and of the homochiral dimers.[2,7]

5.3. Measurement of Enantiomer Concentrations. The results obtained so far are relevant also to the use of a polarimeter and a UV detector to determine the concentration of two enantiomers in solution, i.e., the overall molar concentrations r and s according to our notation. The calibration of polarimeter and UV when no dimerization occurs is rather straightforward and the two signals can be easily combined, thus yielding the desired concentration values.[20] In the presence of association, eqs 18 and 23 should be used instead, after estimating the dimerization equilibrium constants following the method described above, as well as the response coefficients μ and δ for the polarimeter and M, D and E for the UV detector. In the general case eqs 18 and 23 constitute a system of two algebraic equations in the unknowns r and s that has to be solved numerically. In the particular case where $K_{\mathrm{hetero}} = 2K_{\mathrm{homo}}$ and $D = E$, the simplified eqs 20 and 25 can be explicitly inverted yielding

$$m = \frac{-M + \sqrt{M^2 + 4K_{\mathrm{homo}}DA}}{2K_{\mathrm{homo}}D} \tag{26}$$

$$q = \frac{\mu m + \delta K_{\mathrm{homo}}m^2 + \alpha}{\mu m + \delta K_{\mathrm{homo}}m^2 - \alpha} \tag{27}$$

From these, c is obtained as $c = m(1 + 2K_{\mathrm{homo}}m)$ and then r and s through eqs 1 and 2.

5.4. Concluding Remarks. The analysis and the method presented in this paper lead to two rather significant results. On one hand, they allow us to estimate the reaction equilibrium constants of the hetero-chiral and homo-chiral dimerization reaction between pairs of enantiomers in solution, i.e., a rather useful result per se. The method has been applied to the enantiomers of bi-naphthol in chloroform, and suggestions of how to control and improve the accuracy of the estimation have been discussed. On the other hand, this approach is necessary to allow for a correct interpretation of optical rotation and UV absorbance measurements, in particular when determining the concentrations of two enantiomers in solution. Overlooking dimerization reactions or neglecting their quantitative effect may lead to major errors, as illustrated in Figure 8 where it is shown that even a nonracemic mixture of enantiomers which associate may exhibit zero optical rotation. The proposed method is based on optical rotation and UV measurements, which are rather accurate and not time-consuming. In fact, the required measurements do not take more time than what is required for a standard calibration.

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Notation

A = UV absorbance, [V]
c = overall molar concentration, $c = r + s$, [M]
d_{ii} = molar concentration of dimers ($ii = RR, SS, RS$), [M]
D = UV absorbance coefficient for homochiral dimers, [VL/mol]
e.e. = enantiomeric excess, [-]
E = UV absorbance coefficient for heterochiral dimers, [VL/mol]
G = spectrometer gradient strength, [G/cm]
ΔG = Gibbs free energy change, [kJ/mol]
ΔH = Enthalpy change, [kJ/mol]
I/I_0 = ratio of NMR signal intensities, [-]
K_{r} = dimerization equilibrium constant (r = *homo*, *hetero*), [L/mol]
m = overall molar concentration of enantiomers in monomeric form, $\mathrm{m} = \mathrm{m_R} + \mathrm{m_S}$, [M]
m_{i} = molar concentration of enantiomers in monomeric form ($i = R, S$), [M]
M = UV absorbance coefficient for enantiomers in monomeric form, [VL/mol]
o.p. = optical purity, [−]
q = nominal enantiomeric ratio of R and S enantiomers, q = r/s, [-]
r = overall molar concentration of the R enantiomer, [M]
R = gas constant, R = 0.008314 kJ/(mol·K)
s = overall molar concentration of the S enantiomer, [M]

ΔS = Entropy change, [kJ/(mol·K)]

greek letters

α = optical rotation (instrumental reading), [mdeg]
δ = polarimeter response coefficient for enantiomers in monomeric form, [mdeg·L/mol]
μ = polarimeter response coefficient for homochiral dimers, [mdeg·L/mol]

subscripts and superscripts

0 = conditions corresponding to $\alpha = 0$
homo = homochiral dimerization
hetero = heterochiral dimerization
R = R monomer (enantiomer)
RR = RR dimer (enantiomer)
RS = RS dimer
S = S monomer (enantiomer)
SS = SS dimer (enantiomer)

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