12. Leutwyler, S.; Even, U.; Jortner, J. *J. Chem. Phys.* **1983**, *79*, 5769.
13. Bree, A.; Zwarich, R. *J. Chem. Phys.* **1969**, *51*, 912.
14. Pacansky, J.; Koch, W.; Miller, M. D. *J. Am. Chem. Soc.* **1991**, *113*, 317.
15. Pople, J. A.; Krishnan, R.; Schlegel, H. B.; DeFrees, D.; Binkley, J. S.; Frisch, M. J.; Whiteside, R. F.; Hout, R. F.; Hehre, W. J. *Int. J. Quantum Chem. Symp.* **1981**, *15*, 269.
16. Foresman, J. B. *Exploring Chemistry with Electronic Structure Methods: A Guide to Using Gaussian*; Gaussian: Pittsburgh, 1993.
17. Boo, B. H.; Seo, H. Y.; Kang, S. K.; Hong, D. S.; Nak, H.-K. to be submitted for publication.
18. Longuett-Higgins, H. C.; Pople, J. A. *J. Chem. Phys.* **1957**, *27*, 192.
19. Infrared and Raman spectra of **FR** were also measured in our laboratory. The spectral assignments are under way.
20. Decker, C. *Chim. Acta* **1979**, *35A*, 1303.

Chiral Recognition Models for the Liquid Chromatographic Resolution of Enantiomers on (S)-Naproxen-Derived Chiral Stationary Phase Bearing Both π-Acidic and -Basic Sites

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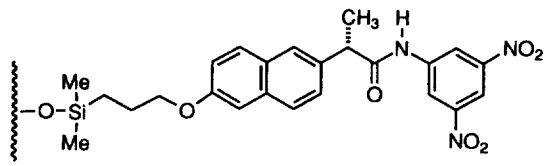
As an effort to elucidate the chiral recognition mechanisms exerted by the (S)-naproxen-derived CSP bearing both π-acidic and π-basic sites, a homologues series of π-basic N-acyl-α-(1-naphthyl)alkylamines and π-acidic N-(3,5-dinitrobenzoyl)-α-amino esters were prepared and resolved. Based on the chromatographic resolution trends of the homologues series of analytes on the (S)-naproxen-derived chiral stationary phase, we proposed chiral recognition mechanisms which demonstrate that the intercalation of the substituent in the analyte molecule between the strands of bonded phase does significantly influence the enantioselectivity for resolving N-acyl-α-(1-naphthyl)alkylamines but the intercalation process is not involved in resolving N-(3,5-dinitrobenzoyl)-α-amino esters.

Introduction

Liquid chromatographic resolution of enantiomers on chiral stationary phases (CSPs) has been known to be a very convenient and accurate technique in solving the problems related to stereochemistry including the determination of optical purity of chiral drugs.[1] Therefore, there have been significant efforts of developing new CSPs for the liquid chromatographic resolution of enantiomers. For example, various CSPs named Pirkle-type have been developed and successfully employed in determining the optical purity of chiral compounds.[2] Pirkle-type CSPs are known to resolve racemates by forming energetically different two transient diastereomeric π-π complexes with two enantiomers.[3] In order to utilize the effective formation of π-π complexes with two enantiomers, Pirkle-type CSPs have been usually designed to contain a π-acidic or π-basic aryl functional group and utilized in resolving π-basic or π-acidic racemates.[4] In relation to these, CSPs bearing both π-acidic and π-basic aryl functional group are quite interesting in that they can be used for resolving both π-basic and π-acidic racemates. However, up to date, a few CSPs containing both π-acidic and π-basic aryl functional groups have been reported.[5]

Recently, we reported the preparation of CSP **1** starting from (S)-naproxen.[6] CSP **1** which actually contains both π-basic and π-acidic aryl functional group has been successfully applied for resolving either π-acidic or π-basic racemates. However, the chiral recognition mechanism expected to be exerted by CSP **1** for resolving π-acidic or π-basic racemates have not been systematically studied yet. In the previous study, we only presumed that the 6-alkoxy-2-naphthyl group of CSP **1** acts as a π-basic interaction site for resolving π-acidic racemates while the 3,5-dinitrophenyl group of the CSP plays a role as a π-acidic interaction site for resolving π-basic racemates.[6]

In this study, we wish to propose chiral recognition models which can be applied for explaining the resolution behaviors of π-basic N-acyl-α-arylalkylamines and π-acidic N-(3,5-dinitrobenzoyl)-α-amino esters based on the chiral recognition trends for resolving a homologous series of π-acidic and π-basic racemates on CSP **1**.



CSP 1

Table 1. Resolution of the two enantiomers of N-acyl-α-(1-naphthyl)alkylamines **2** on CSP **1**[a]

Anal	R	Y	k_S'[b]	k_R'[c]	α[d]	Conf.[e]
2a	CH_3	CH_3	12.07	17.46	1.45	R
b	CH_3	CH_2CH_3	15.68	21.45	1.37	R
c	CH_3	$(CH_2)_2CH_3$	14.56	20.70	1.42	R
d	CH_3	$(CH_2)_3CH_3$	14.10	20.03	1.42	R
e	CH_3	$(CH_2)_5CH_3$	12.00	18.22	1.52	R
f	CH_3	$(CH_2)_6CH_3$	11.02	17.09	1.55	R
g	CH_3	$(CH_2)_8CH_3$	9.36	15.22	1.63	R
h	CH_3	$(CH_2)_{10}CH_3$	8.45	14.28	1.69	R
i	CH_3	$(CH_2)_{12}CH_3$	7.62	13.18	1.73	R
j	CH_3	$(CH_2)_{14}CH_3$	6.80	12.14	1.79	R
k	CH_3	$(CH_2)_{16}CH_3$	6.50	11.80	1.82	R
l	CH_2CH_3	CH_3	10.50	12.01	1.14	R
m	$(CH_2)_2CH_3$	CH_3	9.92	10.78	1.09	R
n	$(CH_2)_3CH_3$	CH_3	9.43	10.09	1.07	R
o	$(CH_2)_4CH_3$	CH_3	9.01	9.01	1.00	
p	$(CH_2)_6CH_3$	CH_3	7.98	7.98	1.00	
q	$(CH_2)_8CH_3$	CH_3	7.20	7.20	1.00	
r	$(CH_2)_{12}CH_3$	CH_3	6.45	5.75	1.12	S
s	$(CH_2)_{14}CH_3$	CH_3	5.99	5.24	1.14	S

[a] For the chromatographic conditions, see the experimental part. [b] Capacity factor of the (S)-enantiomer. [c] Capacity factor of the (R)-enantiomer. [d] Separation factor defined as the ratio of the capacity factor of the second eluted enantiomer to that of the first eluted enantiomer ($\alpha = k_2'/k_1'$). [e] Absolute configuration of the second eluted enantiomer.

Experimental

HPLC analyses were performed with a system which consists of a Waters Model 510 pump, a Rheodyne Model 7125 injector with a 20 mL sample loop, a Youngin Model 710 absorbance detector with a 254-nm UV filter and a Youngin D520B computing integrator. All chromatographic data were obtained using 2-propanol-hexane (90 : 20, v/v) as the mobile phase with a flow rate of 2.0 mL/min at 18 ℃. The column void volume was measured by injecting 1,3,5-tri-*tert*-butylbenzene.[7]

A stainless-steel HPLC chiral column (250 mm×4.6 mm I.D.) packed with CSP **1** was available from prior study[6] and was used unchanged. Analytes were also available from prior studies or were prepared as described previously.[4a,6,8] Unnatural α-amino acids were prepared by the literature procedure.[9]

Results and Discussion

As an effort to elucidate the chiral recognition mechanism exerted by CSP **1** for resolving π-acidic or π-basic racemates, we prepared a homologous series of N-acyl-α-(1-naphthyl)alkylamines **2** and N-(3,5-dinitrobenzoyl)-α-amino esters **3** and resolved them on CSP **1**. The resolution trends for separating two enantiomers of a homologous series of racemic analytes on a certain Pirkle-type CSP have been often utilized as mechanistic probes to investigate the origins of enantioselectivity. For example, the strong dependence of the enantioselectivity of a homologous series of racemic analytes upon the length of a simple alkyl substituent in the analyte molecule has been considered as a clear demonstration that the alkyl substituent of one enantiomer intercalates between the strands of bonded phase during the chiral recognition and consequently differently influences the adsorption of enantiomers.[10]

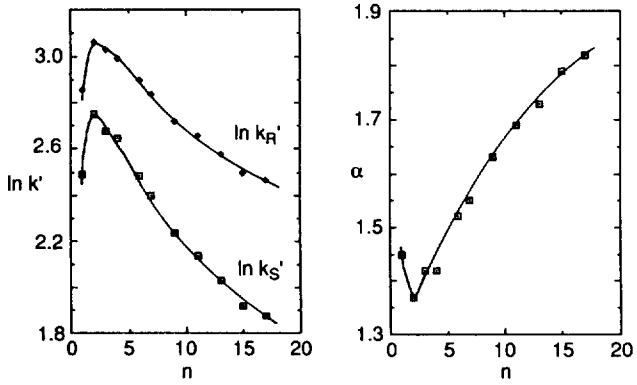
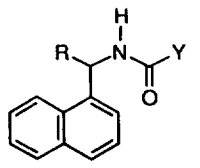


Figure 1. The dependence of the capacity factors (ln k') of the two enantiomers and the enantioselectivity (α) on the length of the N-acyl chain [Y : -$(CH_2)_n$-H] of the analyte denoted by n in the abscissa for resolving a series of N-acyl-α-(1-naphthyl)ethylamines (**2**a-k) on CSP **1**. The chromatographic conditions are described in the experimental.



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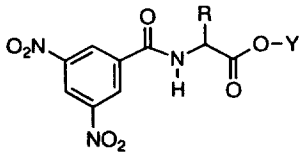

3

The results for resolving a homologous series of N-acyl-α-(1-naphthyl)alkylamines **2** on CSP **1** are summarized in Table 1. Even though some parts of the data for resolving N-acyl-α-(1-naphthyl)ethylamines were previously reported,[6] for the purpose of comparison, all data shown in Table 1 were newly collected under the same condition in this study. The elution orders shown in Table 1 were determined from the configurationally known samples.

The trends for resolving a homologous series of N-acyl-α-(1-naphthyl)ethylamines (**2**a-k) are consistent with those we already discussed in the previous paper[6] and are graphically shown in Figure 1. As shown in Figure 1, the capacity factors, k', for the two enantiomers decrease continuously except for the initial increase as the length of the N-acyl chain (denoted by n) increases. The decreasing trends of the capacity factors shown in Figure 1 are understood based on the well known fact that the increase in the lipophilicity of analytes generally diminish the retention of analytes on the column in the normal phase chromatography. The exceptional initial increase in the capacity factors might be a consequence of conformational factors as discussed previously.[10c] Even though the capacity factors of the two enantiomers decrease as the length of the N-acyl chain length of analytes

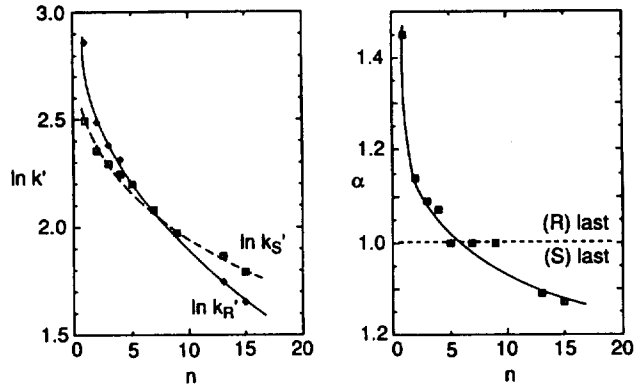

Figure 2. The dependence of the capacity factors (ln k') of the two enantiomers and the enantioselectivity (α) on the length of the alkyl substituent [R : -$(CH_2)_n$-H] at the chiral center of the analyte denoted by n in the abscissa for resolving a series of N-acetyl-α-(1-naphthyl)alkylamines (2l-s) on CSP **1**. The chromatographic conditions are described in the experimental.

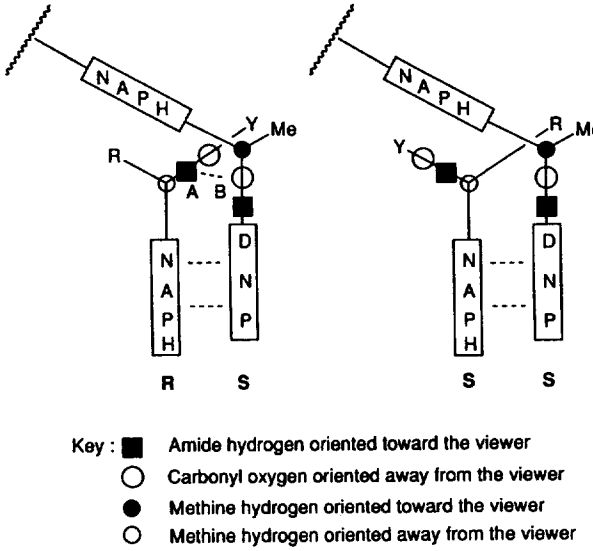

Figure 3. The proposed chiral recognition model for resolving N-acyl-α-(1-naphthyl)alkylamines **2** on CSP **1**. The (R,S)-complex is more stable than the (S,S)-complex because of the hydrogen bonding between the amide N-H hydrogen (A) of the (R)-analyte and the carbonyl oxygen (B) of the CSP.

increases, those of the less retained (S)-enantiomers decrease more rapidly than those of the more retained (R)-enantiomers and consequently the separation factor (α) defined as the ratio of the capacity factor of the more retained enantiomer to that of the less retained enantiomer increases as shown in Figure 1.

In contrast, the trends for resolving N-acetyl-α-(1-naphthyl)alkylamines (2l-s) on CSP **1** shown in Figure 2 indicates that the capacity factors of the initially more retained (R)-enantiomers decrease more rapidly than those of the (S)-enantiomers as the length of the alkyl substituent at the chiral center of the analyte increases and finally the (S)-enantiomers are retained longer on the column after the alkyl substituent at the chiral center of the analyte reaches at

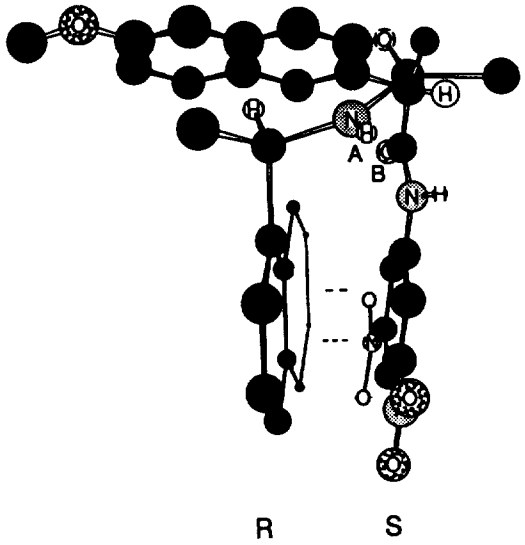

Figure 4. The Chem 3D ball-and-stick representation of the (R,S)-complex of Figure 3.

a certain length. In consequence, the separation factor (α) defined as above decreases continuously to 1.00 and then increases with the inversion of elution order as the length of the alkyl substituent at the chiral center of the analytes increases.

Those chromatographic resolution behaviors shown in Figure 1 and 2 suggest that unfavorable steric interactions are occurring to a greater extent during the chiral recognition for the less retained (S)-enantiomers of N-acyl-α-(1-naphthyl)ethylamines as the length of the N-acyl chain increases, presumably as a consequence of the intercalation of the N-acyl chain between adjacent strands of bonded phase. Similarly, unfavorable steric interactions are imagined to occur to a greater extent during the chiral recognition for the (R)-enantiomers of N-acetyl-α-(1-naphthyl)alkylamines as the alkyl substituent at the chiral center of analytes increases in length.

Based on these assumptions, one conceivable chiral recognition model which explains the trends shown in Figure 1 and 2 is proposed from study of the CPK molecular models as shown in Figure 3. It is difficult to clearly portray the three-dimensional complexes in the simplified two-dimensional schematic drawing such as shown in Figure 3 and therefore, the three-dimensional ball-and-stick model representation shown in Figure 4 may be helpful for the better understanding. The conformations of CSP **1** and the analyte shown in the model are presumed to be of relatively low energy and hence preferentially populated.[5d,10a] As shown in the models of Figure 3 and 4, CSP **1** and the analyte interact each other through the face-to-face π-π complexation between their respective π-acidic 3,5-dinitrophenyl (DNP) and the π-basic 1-naphthyl group (NAPH). Simultaneously, the 1-naphthyl group (NAPH) of the analyte presents its edge to the face of the 6-alkoxy-2-naphthyl group (NAPH) of the CSP, undergoing the face-to-edge π-π interaction, which has been known to be an important associative force between aromatic rings.[10c] Additionally the amide N-H hydrogen (A) of the (R)-analyte interacts with the carbonyl oxygen (B) of the CSP

Table 2. Resolution of the two enantiomers of N-(3,5-dinitrobenzoyl)-α-amino esters **3** on CSP **1**[a]

Anal	R	Y	k_S'[b]	k_R'[c]	α[d]	Conf.[e]
3a	$CH_2CH(CH_3)_2$	CH_3	12.84	26.74	2.08	R
b	$CH_2CH(CH_3)_2$	CH_2CH_3	10.48	22.07	2.11	R
c	$CH_2CH(CH_3)_2$	$(CH_2)_2CH_3$	9.99	20.93	2.10	R
d	$CH_2CH(CH_3)_2$	$(CH_2)_3CH_3$	9.17	19.24	2.10	R
e	$CH_2CH(CH_3)_2$	$(CH_2)_5CH_3$	8.21	17.72	2.16	R
f	$CH_2CH(CH_3)_2$	$(CH_2)_7CH_3$	6.97	15.64	2.24	R
g	$CH_2CH(CH_3)_2$	$(CH_2)_9CH_3$	7.07	15.19	2.15	R
h	$CH_2CH(CH_3)_2$	$(CH_2)_{11}CH_3$	6.45	13.86	2.15	R
i	$CH_2CH(CH_3)_2$	$(CH_2)_{13}CH_3$	6.16	13.40	2.18	R
j	$CH_2CH(CH_3)_2$	$(CH_2)_{15}CH_3$	5.82	12.73	2.19	R
k	CH_3	CH_2CH_3	12.42	21.24	1.71	R
l	$(CH_2)_2CH_3$	CH_2CH_3	11.76	22.02	1.87	R
m	$(CH_2)_4CH_3$	CH_2CH_3	10.61	19.00	1.79	R
n	$(CH_2)_6CH_3$	CH_2CH_3	9.29	17.38	1.87	R
o	$(CH_2)_7CH_3$	CH_2CH_3	9.70	17.96	1.85	R

[a] For the chromatographic conditions, see the experimental part. [b] Capacity factor of the (S)-enantiomer. [c] Capacity factor of the (R)-enantiomer. [d] Separation factor defined as the ratio of the capacity factor of the second eluted enantiomer to that of the first eluted enantiomer ($\alpha = k_2'/k_1'$). [e] Absolute configuration of the second eluted enantiomer.

through the hydrogen-bonding. In consequence the diastereomeric (R,S)-complex shown in Figure 3 should be more stable than the (S,S)-complex.

With the simultaneous interactions between the CSP and the analyte as shown in Figure 3, the alkyl chain (denoted by Y in the model) of the N-acyl group of the (R)-analyte is oriented to the direction of the methyl group of the CSP and that of the (S)-analyte is oriented alongside the tether of the CSP. In this event, the acyl alkyl chain (Y) of the (S)-analyte intercalates between the strands of the bonded phase and consequently the retention time of the (S)-analyte decrease more rapidly than that of the (R)-analyte as the length of the alkyl chain (Y) of the analyte increases and the separation factor (α) increases continuously.

Similarly, the alkyl substituent (denoted by R in the model) at the chiral center of the (R)-analyte is oriented alongside the tether of the CSP and eventually intercalates between the strands of the bonded phase as shown in Figure 3. Consequently, the retention time of the initially more retained (R)-analyte decreases more rapidly than that of the (S)-analyte as the alkyl substituent (R) at the chiral center of the analyte increases in length. When the alkyl substituent (R) at the chiral center of the analyte reaches to a certain length, the retention time of the initially more retained (R)-analyte becomes equal to that of the (S)-analyte and no separation of the two enantiomers is observed. After that, the retention time of the (R)-analyte becomes shorter than that of the (S)-analyte as the alkyl substituent (R) at the chiral center of the analyte increases in length and consequently the (S)-enantiomers are retained longer, resulting in the inversion of elution order.

The results for resolving N-(3,5-dinitrobenzoyl)-a-amino

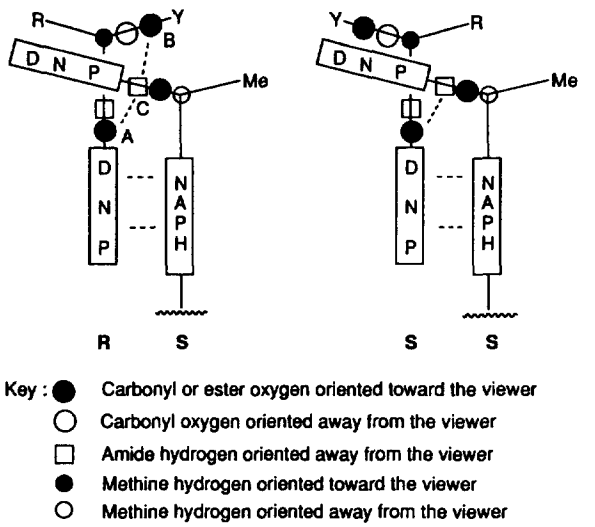

Figure 5. The proposed chiral recognition model for resolving N-(3,5-dinitrobenzoyl)-α-amino esters **3** on CSP **1**. The (R,S)-complex implying the additional hydrogen bonding between the ester alkoxy hydrogen (B) of the (R)-analyte and the amide N-H hydrogen (C) of the CSP is more stable than the (S,S)-complex.

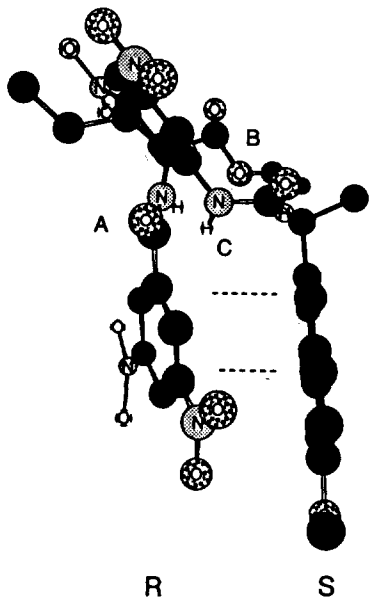

Figure 6. The Chem 3D ball-and-stick representation of the (R,S)-complex of Figure 5.

esters **3** on CSP **1** summarized in Table 2, however, do not show any notable trends in the separation factors, indicating that the intercalation of either the ester alkyl chain (Y) or the alkyl substituent (R) at the chiral center of **3** between the strands of the bonded phase is not involved in the chiral recognition processes. To explain those resolution results for N-(3,5-dinitrobenzoyl)-α-amino esters **3** on CSP **1**, we propose a chiral recognition model which does not involve any intercalation processes as shown in Figure 5 and 6.[11] In the model shown in Figure 5 and 6, CSP **1** shows the same conformation as drawn in Figure 3 and 4 and the analyte

is presumed to be in its lowest energy conformation as described previously.[10b] As shown in Figure 5 and 6, the 6-alkoxy-2-naphthyl group (NAPH) of the CSP is utilized as a π-basic site for the face-to-face π-π-interaction with the π-acidic 3,5-dinitrophenyl group (DNP) of the analyte. Simultaneously, the edge of the 3,5-dinitrophenyl group (DNP) of the analyte confronts the face of the 3,5-dinitrophenyl group (DNP) of the CSP, invoking the face-to-edge π-π interaction and the carbonyl oxygen (A) of the 3,5-dinitrobenzoyl group of the analyte interacts with the amide N-H hydrogen (C) of the CSP through the hydrogen bonding. In this event, the ester alkoxy oxygen (B) of the (R)-analyte can also interact with the amide N-H hydrogen (C) of the CSP whereas that of the (S)-analyte is not in the position of such an interaction. In consequence, the (R,S)-complex shown in Figure 5 should be more stable than the (S,S)-complex and the (R)-enantiomers should be retained longer than the (S)-enantiomers on the column. In the model shown in Figure 5, it should be also noted that both of the ester alkyl chain (denoted by Y) and the alkyl substituent (denoted by R) at the chiral center of the analyte are far from the connecting tether of the CSP and, therefore, lengthening either the ester alkyl chain (Y) or the alkyl substituent (R) at the chiral center of the analyte does not invoke any unfavorable interactions with the CSP and does not influence the enantioselectivities.

In conclusion, in this study, we proposed chiral recognition models which can explain the chromatographic behaviors for resolving either π-basic N-acyl-α-(1-naphthyl)alkylamines **2** or π-acidic N-(3,5-dinitrobenzoyl)-α-amino esters **3** on CSP **1**. In the proposed model, simultaneous non-covalent bonding interactions such as a face-to-face π-π interaction, a face-to-edge π-π interaction and a hydrogen bonding interaction are thought to occur between the CSP and the analyte, especially the hydrogen bonding interaction being enantioselective. Even though the models shown in Figure 3 and 5 are successful in explaining the chromatographic resolution behaviors, they might be verified or modified as more spectroscopic and/or crystallographic data for the diastereomeric complex formed between the CSP and the analyte are obtained.

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References

1. (a) Krstulović, A. M. Ed. *Chiral Separations By HPLC: Applications to Pharmaceutical Compounds*; Ellis Horwood: Chichester, England, 1989. (b) Ahuja, S. Ed. *Chiral Separations by Liquid Chromatography*; ACS Symposium Series 471, American Chemical Society: Washington, DC. 1991. (c) Taylor, D. R.; Maher, K. *J. Chromatogr. Sci.* **1992**, *30*, 67.
2. Perrin, S. R.; Pirkle, W. H. in *Chiral Separations by Liquid Chromatography*; Ahuja, S., Ed.; ACS Symposium Series 471, American Chemical Society: Washington, DC 1991, chap. 3.
3. Pirkle, W. H.; Pochapsky, T. C. *Chem. Rev.* **1989**, *89*, 347.
4. (a) Pirkle, W. H.; Hyun, M. H. *J. Org. Chem.* **1984**, *49*, 3043. (b) Hyun, M. H.; Kim, M. H. *J. Liq. Chromatogr.* **1990**, *13*, 3229. (c) Hyun, M. H.; Hwang, S.-Y.; Ryoo, J.-J. *Chem. Lett.* **1994**, 1021.
5. (a) Hyun, M. H.; Pirkle, W. H. *J. Chromatogr.* **1987**, *393*, 357. (b) Oi, N.; Kitahara, H.; Mathumoto, Y.; Nakajima, H., Horikawa, Y. *J. Chromatogr.* **1989**, *462*, 382. (c) Tambute, A.; Siret, L.; Caude, M.; Begos, A.; Rosset, R. *Chirality* **1990**, *2*, 106. (d) Pirkle, W. H.; Welch, C. J.; Lamm, B. *J. Org. Chem.* **1992**, *57*, 3854. (e) Hyun, M. H.; Min, C.-S. *Chem. Lett.* **1994**, 1463.
6. Hyun, M. H.; Jin, J. S.; Ryoo, J.-J.; Jyung, K. K. *Bull. Kor. Chem. Soc.* **1994**, *15*, 497.
7. Pirkle, W. H.; Welch, C. J. *J. Liq. Chromatogr.* **1991**, *14*, 1.
8. Pirkle, W. H.; Welch, C. J.; Hyun, M. H. *J. Org. Chem.* **1983**, *48*, 5022.
9. Barger, G.; Weichselbaum, T. E. *Org. Syn. Col. Vol. II*, John Wiely and Sons, New York, 1959, p 384.
10. (a) Pirkle, W. H.; Hyun, M. H.; Bank, B. *J. Chromatogr.* **1984**, *316*, 585. (b) Pirkle, W. H.; Murray, P. G.; Burke, J. A. *J. Chromatogr.* **1993**, *641*, 21. (c) Hyun, M. H.; Cho, S. M., Ryoo, J.-J.; Kim, M. S. *J. Liq. Chromatogr.* **1994**, *17*, 317.
11. A similar chiral recognition model has been proposed for resolving N-(3,5-dinitrobenzoyl)-α-amino esters on a CSP derived from 3,5-dimethylanilide derivative of (S)-naproxen. Hyun, M. H.; Cho, Y. J.; Ryoo, J.-J.; Jyung, K. K. *J. Chromatogr. A*, accepted for publication.