Synthesis of a Chiral Macrocyclic Dibenzodicyclohexanotetraamide-Containing Stationary Phase for Liquid Chromatography

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Received October 31, 1998

Allyloxy-substituted macrocyclic dibenzodicyclohexanotetraamide **2** was prepared by the following sequence. MonoBoc-protected chiral 1,2-cyclohexanediamine (**3**) was treated with isophthaloyl chloride followed by removal of the Boc group to form bisisophthalamide **5**. Compound **5** was then treated with allyloxyphthaloyl chloride to form the macrocyclic tetraamide **2** in a 56% yield. Chiral selector **2** was converted to its ethoxydimethylsilane derivative and heated in a suspension of silica gel and toluene to form the chiral macrocycle-containing silica gel phase **1**. This phase separated the enantiomers of (±)-α-methylbenzylamine and (±)-DL-α-aminobutyric acid methyl ester in a liquid chromatograph.

J. Heterocyclic Chem., **36**, 381 (1999).

1. Introduction.

With the rapid development of asymmetric syntheses, an increasing number of chiral organic compounds have been synthesized. This has stimulated the development of analytical methods for the analysis of the enantiomeric purity of these compounds. Many methods are available for the determination of enantiomeric purity of racemates [1]. Among these, chromatography on chiral stationary phases is widely accepted as a reliable and efficient method. Several reviews describing current technologies for chiral stationary phases have been published [2-5].

The separation of enantiomers of a racemic solute by chromatography using chiral station phases is based on the formation of transient diastereomeric complexes between enantiomers of a racemic solute and a chiral selector in the stationary phase. The difference in stability between the transient diastereomeric complexes leads to a difference in retention time; the enantiomer forming the less stable complex is eluted first and vice versa. A chiral selector can be a derivatized naturally existing substance, such as a protein (bovine serum albumin, enzyme, *etc.*), a cyclodextrin, a cellulose derivative, or a fully synthetic small molecule.

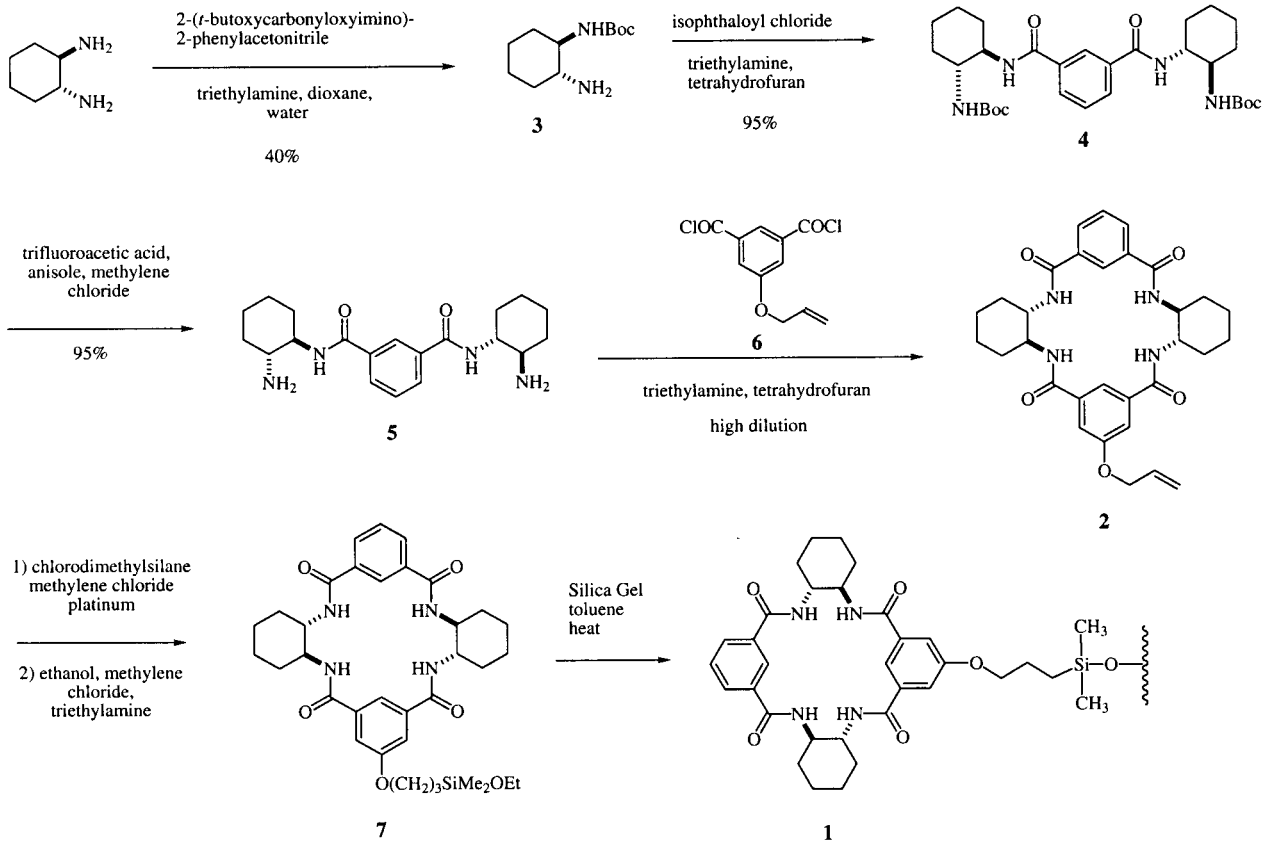
The development of chiral station phases based on synthetic chiral molecules relies on a sensible rationale: the greater the number of specific, discrete, and simultaneous interactions between chiral solute molecules and the chiral selector on the stationary phase, the greater the likelihood of effective chiral discrimination, and thus of chromatographic separation of enantiomeric solutes. Starting from this premise, a variety of chiral organic molecules have been prepared and incorporated into stationary phases. These chiral selectors have at least one of three types of functional groups in close proximity to the stereogenic center(s): 1) electron rich or electron deficient aromatic groups capable of donor-acceptor interactions; 2) polar hydrogen-bond and /or dipole stacking sites; and 3) bulky non-polar groups which provide steric repulsion. Many structurally and configurationally distinct multiple-interaction chiral stationary phases have been reported.

A number of chiral stationary phases have been developed in the past few years [6]. Among those studied, chiral stationary phases containing amide linkages provide excellent separations of the enantiomers of various polar solutes [7,8]. A chiral stationary phase derived from chiral *trans*-1,2-cyclohexanediamine separated the enantiomers of a broad range of polar compounds [9-12]. Yoon and Still reported that a cage molecule formed by macrocyclization of two molecules of isophthaloyl chloride and two molecules of enantiopure 1,2-cyclohexanediamine interacts strongly with short peptides [13]. This cyclic tetraamide should be an excellent candidate for the chiral selector of a chiral stationary phase. We herein report the synthesis of new chiral stationary phase **1** (Scheme 1) wherein the above mentioned macrocyclic tetraamide unit **3** is attached to silica gel. This stationary phase was used in the liquid chromatographic separation of the enantiomers of (±)-α-methylbenzylamine and (±)-DL-α-aminobutyric acid methyl ester.

Results and Discussion.

The synthesis of the allyloxy-substituted macrocyclic tetraamide **2** needed for the synthesis of chiral stationary phase **1** is shown in Scheme 1. Diamidodiamine **5**, a key synthon for **2**, was synthesized by first preparing monoprotected (*R,R*)-1,2-cyclohexanediamine (**3**). Enantiopure 1,2-cyclohexanediamine was treated with 2-(*tert*-butoxycarbonyloxyimino)-2-phenylacetonitrile in the presence of triethylamine to give a mixture of starting material, monoprotected diamine, and diprotected diamine [14]. The highest yield of monoprotected product **3** was 40%. Addition of one equivalent of di-*tert*-butyl dicarbonate [15] to the diamine gave the diprotected by-product and unreacted starting diamine.

Scheme 1
Synthesis of Chiral Phase **1** *via* Allyloxy-substituted Chiral Macrocyclic Tetraamide **2**



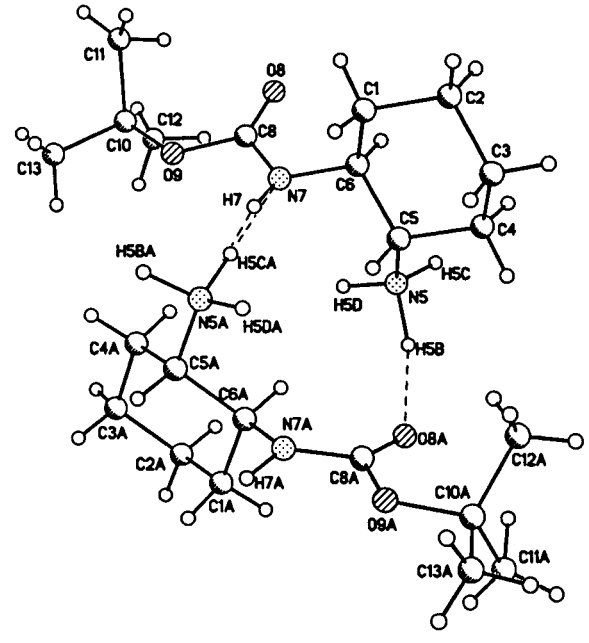


Figure 1. The solid state structure of **3** and a symmetry related molecule. The figure shows the three intermolecular interactions between the two molecules. The hydrogen bond joining H7 to N5A is partially hidden by the N5A-H5CA bond.

Many amines absorb carbon dioxide readily to form salts which are insoluble in most organic solvents. However, mono-*tert*-butylcarbonyl protected **3** does not form these byproducts even after being exposed to air for several years. An X-ray investigation (see later) indicates the presence of an intermolecular hydrogen bond in the solid state between the amine nitrogen and the carbamate hydrogen (see Figure 1).

Di-*tert*-butylcarbonyl-protected **4** was obtained in quantitative yield by treating **3** with isophthaloyl chloride and triethylamine. Removal of the two *tert*-butylcarbonyl groups of **4** was realized by adding **4** to a mixture of trifluoroacetic acid and ten equivalents of anisole. Anisole was used to scavenge the liberated *t*-butyl cations thus preventing alkylation of the aromatic ring [16]. A large excess of anisole was necessary to obtain the best yields. When only two equivalents of anisole were used, the yield of **5** decreased to 70%.

Compound **2** was obtained in a 56% yield by cyclizing diamidodiamine **5** and 5-(allyloxy)isophthaloyl chloride using a high dilution technique. Solutions of the two reactants in tetrahydrofuran were added *via* syringe pumps over a period of 8 hours to a solution of triethylamine in 100 ml of tetrahydrofuran at room temperature. The reac-

tion was pleasantly clean with no by-products except for some polymeric material. The success of this macrocyclization reaction depends on the purities of the two reactants. Compound **5**, which absorbs carbon dioxide from air, was used immediately after it was prepared for cyclization. The macrocyclization reaction failed when **5**, which had been stored for more than one day, was utilized.

hydrogen atoms are involved in hydrogen bonds linking N7 to N5A. In addition to these intermolecular interactions, there is a third intermolecular hydrogen bond (Figure 1). These three hydrogen bonds form a network of molecules. The hydrogen bond data are listed in Table 1. These intermolecular interactions likely contribute to the unusual stability of **3** in the solid state.

Table 1

Hydrogen Bond Data for **3**

Donor	Hydrogen	Acceptor	H...A(Å)	D...A(Å)	D-H...A(Å)	Symmetry Translation of A
N7	H7	N5	2.12	3.051	163	1/2 + x, 1/2 - y, 2 - z
N5	H5C	N7	1.91	3.051	176	-1/2 + x, 1/2 - y, 2 - z
N5	H5B	O8	2.11	3.151	148	1/2 + x, 1/2 - y, 2 - z

The 5-(allyloxy)isophthaloyl chloride was prepared by treating diethyl 3-hydroxyphthalate with allyl bromide and base. The resulting diacid was converted to the diacid dichloride using oxalyl chloride.

Macrocyclic tetraamide **2** could also have been prepared from the 5-(allyloxy)isophthalamide analog of **5** and isophthaloyl chloride. This sequence would require the removal of the *tert*-butylcarbonyl protecting groups in the presence of an electron rich benzene ring and a double bond. Since this could have caused unwanted by-products, the sequence shown in Scheme 1 was chosen.

Chiral stationary phase **1** was prepared by first hydrosilylation of **2** with chlorodimethylsilane using chloroplatinic acid [17] (Scheme 2). The resulting chlorosilane was treated with ethanol to form the macrocyclic tetraamide-containing ethoxysilane **7** [18]. This ethoxy compound was heated with silica gel to form chiral stationary phase **1** as reported for other silica gel bound macrocyclic ligands [19]. Elemental analysis of the thoroughly dried bonded phase showed that approximately 0.14 mmole (based on nitrogen, 0.16 mmole if based on carbon) of chiral selector was attached to each gram of chiral stationary phase **1**.

A structural study of **3** was initiated in an effort to find possible reasons for its unusual stability in the presence of carbon dioxide. The solid-state structure of **3** together with that of a symmetry related molecule are shown in Figure 1. The figure shows that **3** is disordered with both the expected and zwitterion forms of **3** being present in the unit cell. The X-ray study yields an averaged structure of the compounds. The average structure of **3** appears to have three hydrogen atoms on the amine nitrogen (N5) and one hydrogen atom on the carbamate nitrogen (N7). Actually, one half of the molecules have three hydrogen atoms bonded to N5 and no hydrogen atoms bonded to N7 resulting in a zwitterion, while the other half of the molecules have the expected two hydrogen atoms bonded to N5 and one hydrogen atom bonded to N7. Both partial

Enantiomers containing amino groups or hydroxy groups were used to conduct preliminary tests of the enantioselectivity exerted by chiral stationary phase **1**. The results show that the chiral stationary phase **1** has high enantioselectivity for enantiomers containing amine groups. Two pairs of such enantiomers were separated with values of more than 1.10, as illustrated in Figures 2 and 3. Some peak tailing was observed, which is mainly ascribed to the polarity of these enantiomers. Further silica surface deactivation would be required for separations of highly polar enantiomers. The separation mechanism for chiral stationary phase **1** is unclear at this time. Further work is needed to investigate the detailed chromatographic performance of chiral stationary phase **1**.

Table 2

Crystal Data and Experimental Data for **3**

Empirical formula	$C_{11}H_{22}N_2O_2$
Formula weight	214.31
Crystal size	0.4 x 0.4 x 0.15 mm
Crystal System	Orthorhombic
Space group	$P2_12_12_1$
Unit cell dimensions	a = 7.200(2) Å b = 7.501(2) Å c = 25.153 (5) Å α = 90% β = 90% γ = 90%
Volume	1358.5 $Å^3$
Density calculated	1.048 g/cm^3
Absorption coefficient	0.072 mm^{-1}
2θ max	50.0%
Limiting indices	$0 \le h \le 8, 0 \le k \le 8, 0 \le l \le 29$
Independent data	1412
Refinement method	Full-matrix least-squares on F^2
Data/restraints/parameters	1392/0/146
Goodness-of-fit on F^2	1.046
Final R indices (I>2σ(I))	R1 = 0.065, wR^2 = 0.148
Largest peak in difference map	0.171e$Å^{-3}$
Largest hole in difference map	-0.145e$Å^{-3}$

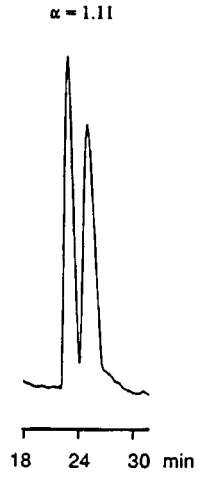


Figure 2. Liquid chromatogram of (±)-methylbenzylamine using **1**. Conditions: 28 cm x 250 μm i.d. capillary column containing bonded and end-capped silica (5 m, 80 Å) particles (chiral stationary phase6); methanol/water (95:5, v/v) mobile phase; uv (254 nm) detection

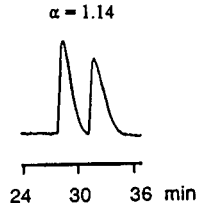


Figure 3. Liquid chromatogram of (Å)DL-α-aminobutyric acid methyl ester. Conditions are the same as in Figure 2 except for uv (215 nm) detection.

Table 3

Atomic Coordinates [x10^4] and Equivalent Isotropic Displacement Parameters [Å^2 x 10^3] for **3**. U(eq) is defined as one-third of the trace of the orthogonalized U_{ij} tensor.

	x	y	z	U(eq) [a]
Cl	2710(10)	-1547(7)	10341(2)	67(2)
HlA	2229(10)	-2449(7)	10101(2)	80
HlB	4047(10)	-1691(7)	10359(2)	80
C2	1896(12)	-1839(8)	10887(2)	83(2)
H2A	552(12)	-1857(8)	10864(2)	100
H2B	2302(12)	-2983(8)	11024(2)	100
C3	2493(10)	-385(9)	11261(2)	80(2)
H3A	3823(10)	-465(9)	11318(2)	95
H3B	1884(10)	-551(9)	11602(2)	95
C4	2019(10)	1450(8)	11045(2)	62(2)
H4A	679(10)	1577(8)	11027(2)	75
H4B	2489(10)	2352(8)	11286(2)	75
C5	2832(8)	1750(6)	10500(2)	48(1)
H5A	4189(8)	1745(6)	10530(2)	58
N5	2241(6)	3520(5)	10298(2)	56(1)
H5B	3135(6)	4600(5)	10511(2)	84
H5C	688(6)	3800(5)	10348(2)	84
H5D	2522(6)	3805(5)	9873(2)	84
C6	2264(7)	295(7)	10116(2)	48(1)
H6A	916(7)	370(7)	10069(2)	58
N7	3127(6)	571(5)	9598(1)	48(1)
H7	4448(6)	601(5)	9639(1)	58

Table 3 (continued)

	x	y	z	U(eq) [a]
C8	2238(7)	161(7)	9145(2)	49(1)
O8	638(5)	-252(6)	9105(1)	70(1)
O9	3453(5)	292(5)	8733(1)	59(1)
C10	2875(9)	35(10)	8185(2)	76(2)
C11	2212(15)	-1904(15)	8121(4)	132(4)
H11A	1076(75)	-1416(109)	8258(33)	197
H11B	2004(130)	-2362(103)	7770(13)	197
H11C	2627(123)	-2851(81)	8349(26)	197
C12	1423(20)	1400(20)	8038(4)	187(6)
H12A	631(128)	1581(171)	8340(28)	280
H12B	2069(153)	2487(100)	7958(50)	280
H12C	686(135)	1053(173)	7738(30)	280
C13	4668(10)	242(13)	7890(2)	100(3)
H13A	5580(80)	-616(68)	8000(25)	151
H13B	4369(87)	56(85)	7522(8)	151
H13C	5156(101)	1423(37)	7936(27)	151

[a] U(eq) for hydrogen atoms are regular isotropic displacement parameters.

EXPERIMENTAL

Proton and carbon nmr spectra were recorded in deuteriochloroform on a Varian 300 MHz spectrometer. All starting materials were used as obtained from commercial sources. (±)-Methylbenzylamine was purchased from Aldrich (Milwaukee, WI). (±) DL-α-Aminobutyric acid methyl ester was purchased from Sigma (St. Louis, MO). Fused silica capillary tubing was purchased from Polymicro Technologies (Phoenix, AZ). Untreated spherical silica having 5 μm diameter and 80 Å pores was purchased from Phase Separations (Norwalk, CT).

(1*R*,2*R*)-(–)-*N*-(*t*-Butoxycarbonyl)-1,2-cyclohexanediamine (**3**) (Scheme 1).

A solution of 2-(*tert*-butoxycarbonyloxyimino)-2-phenylacetonitrile (14.72 g, 60 mmoles) in 75 ml of dioxane was added dropwise to a mixture of (1*R*,2*R*)-(–)-1,2-cyclohexanediamine (7.01 g, 60 mmoles) in triethylamine/water/dioxane (1/4/4) at 0°. After stirring for 4 hours at room temperature, the mixture was diluted with 200 ml of water, and then extracted with ethyl acetate (4 x 200 ml). The combined ethyl acetate extracts were dried over sodium sulfate and evaporated *in vacuo* to give a brown solid. Flash chromatography (silica gel, 5% triethylamine in ethyl acetate) of the brown solid and recrystallization from ethyl acetate gave compound **3** as clear crystals (4.0 g, 30%); mp = 112-113°; R_f = 0.4 (1:10/triethylamine:ethyl acetate); $[\alpha]_D^{20}$ = -24.3° (c = 1.0, chloroform); ^{1}H nmr: δ 4.48 (br s, 1H), 3.20-3.14 (m, 1H), 2.36-2.28 (td, *J* = 9.9, 3.9 Hz, 1H), 2.03-1.94 (m, 2H), 1.73-1.69 (m, 2H), 1.45 (s, 9H), 1.39-1.07 (m, 6H); ^{13}C nmr: δ 156.2, 79.3, 57.7, 55.7, 35.3, 32.9, 28.4, 25.2, 25.1; ms: (CI) m/z 215 (MH^+).

Anal. Calcd. for $C_{11}H_{22}N_2O_2$: C, 61.65; H, 10.35. Found: C, 61.76; H, 10.24.

N,N'-Bis[2'-(*t*-butoxycarbonyl)-(1'*R*,2'*R*)-diaminocyclohexyl]-isophthalamide (**4**).

Isophthaloyl chloride (0.62 g, 3.0 mmoles) in 10 ml of dry tetrahydrofuran was added dropwise to a solution of mono-

protected **3** (1.29 g, 6.0 mmoles) in 100 ml of dry tetrahydrofuran with stirring over a period of 40 minutes at room temperature. The mixture was stirred for another 20 minutes and the white precipitate was filtered. Evaporation of solvent gave a white solid which was recrystallized from ethanol to yield a white powder (1.6 g, 95%) which was pure enough to be used in the next step. A small portion was further purified by recrystallization from ethanol, mp 232-234°; R_f = 0.24 (3:2/hexanes:ethyl acetate); $[\alpha]_D^{20}$ = -71.5° (c = 2.2, chloroform); ^{1}H nmr: δ 8.25 (s, 1H), 7.98 (d, *J* = 8.1 Hz, 2H), 7.44 (t, *J* = 8.1 Hz, 1H), 7.19 (d, *J* = 7.5 Hz, 2H), 4.72 (d, *J* = 8.7 Hz, 2H), 3.84-3.75 (m, 2H), 3.60-3.49 (m, 2H), 2.25-2.20 (m, 2H), 2.08-2.02 (m, 2H), 1.82-1.72 (m, 4H), 1.37-1.21 (m, 8H), 1.28 (s, 18H); ^{13}C nmr: δ 166.5, 157.2, 134.6, 130.3, 128.8, 125.5, 79.9, 56.3, 53.9, 32.8, 32.7, 28.5, 25.3, 24.7; ms: (fab) *m/z* 559 (MH^+), 581 (MNa^+).

Anal. Calcd. for $C_{30}H_{46}N_4O_6$: C, 64.49; H, 8.30. Found: C, 64.29; H, 8.10.

N,N'-Bis[2'-amino-(1'*R*,2'*R*)-cyclohexyl]isophthalamide (**5**).

Compound **4** (0.83 g, 1.49 mmoles) in 25 ml of methylene chloride was added dropwise to a mixture of trifluoroacetic acid (10 ml, 65 mmoles), anisole (6.02 g, 60 mmoles), and 10 ml of methylene chloride. After stirring for 30 minutes at room temperature, the methylene chloride and trifluoroacetic acid were carefully evaporated. The oily residue was treated with 10 ml of water and extracted with diethyl ether (2 x 20 ml) to leave an acidic aqueous solution. The combined ether extracts were washed with 5 ml of water and the aqueous wash mixture was combined with the above acidic solution. The combined aqueous solutions were treated with 10 ml of 30% ammonia solution followed by addition of 20 g of sodium chloride. The salt solution was extracted with methylene chloride (5 x 30 ml). The solution was dried over sodium sulfate and evaporated to give compound **5** as a white solid (0.51 g, 95%). This compound readily absorbs carbon dioxide from the air, mp 194-196°; R_f = 0.32 (1:1:8/ammonium hydroxide:methanol:ethyl acetate); $[\alpha]_D^{20}$ = -31.6° (c = 1.0, 0.1 *N* hydrochloric acid); ^{1}H nmr (dimethyl-d_6 sulfoxide): δ 8.29 (s, 1H), 7.93 (d, *J* = 7.8 Hz, 2H), 7.46 (t, *J* = 7.8 Hz, 1H), 6.92 (d, *J* = 8.1 Hz, 2H), 3.73-3.65 (m, 2H), 2.58-2.50 (m, 2H), 2.10-1.95 (m, 8H), 1.74-1.72 (m, 4H), 1.37-1.20 (m, 8H); ^{13}C nmr (dimethyl-d_6 sulfoxide): δ 167.2, 135.0, 130.4, 129.1, 125.5, 56.8, 55.5, 36.0, 32.7, 25.2; ms: (CI) *m/z* 359 (MH^+); hrms: (CI) *m/z* Calcd for $C_{20}H_{31}N_4O_2$ (MH^+): 359.2447; Found: 359.2455.

Ethyl 5-Hydroxyisophthalate.

Dry hydrogen chloride gas (3.87 g, 0.106 mole) was introduced into a solution of 5-hydroxyisophthalic acid (10.01 g, 0.055 mole) in 200 ml of absolute ethanol. The mixture was refluxed for 36 hours. Thin layer chromatography analysis showed that the majority of the diacid was converted to the diester. Ethanol was evaporated and the residue was dissolved in diethyl ether. The ether solution was washed with a saturated sodium bicarbonate solution and water. The aqueous solution was back-extracted with diethyl ether. The combined diethyl ether layers were dried over sodium sulfate and evaporated. The ester (12.5 g, 95%) was obtained as a white solid which could be recrystallized from diethyl ether, mp 100-102°; R_f = 0.58 (1:2:7/acetic acid:ethyl acetate:hexanes); ^{1}H nmr: δ 8.23 (s, 1H), 7.84 (d, *J* = 1.5 Hz, 2H), 7.11 (br s, 1H), 4.41 (d, *J* = 7.2 Hz, 4H), 1.41 (t, *J* = 7.2 Hz, 6H); ^{13}C nmr: δ 166.4, 156.7, 132.2, 122.8, 121.2, 61.9, 14.4; ms: (EI) *m/z* 193 (100), 238 (M^+, 60).

Anal. Calcd. for $C_{12}H_{14}O_5$: C, 60.50: H, 5.92. Found: C, 60.50; H, 5.71.

5-(Allyloxy)isophthalic Acid.

Potassium *t*-butoxide (30 g, 0.267 mole) was slowly added to a solution of the above diester (19.4 g, 81 mmoles) in 400 ml of dry tetrahydrofuran under a nitrogen atmosphere. After 30 minutes, allyl bromide (71 ml, 0.81 mole) was added dropwise. The resulting mixture was stirred overnight and treated with 15 g of a 50% sodium hydroxide solution and allowed to stir for 1 hour. The solvent was evaporated. The dry solid residue was dissolved in 50 ml of water and acidified with 2 *N* hydrochloric acid until the *p*H of the mixture reached 2. The white solid which precipitated was filtered and dried under vacuum. Recrystallization of the solid in aqueous ethanol gave 5-(allyloxy)isophthalic acid as white needles (13.5 g, 75%); R_f = 0.52 (1:2:8/acetic acid:ethyl acetate:hexanes); mp 226-228°; ^{1}H nmr (dimethyl-d_6 sulfoxide): δ 13.3 (br s, 2H), 8.09-8.07 (m, 1H), 7.68-7.66 (m, 2H), 6.11-6.01 (m, 1H), 5.46-5.27 (m, 2H), 4.72-4.70 (m, 2H); ^{13}C nmr (dimethyl-d_6 sulfoxide): δ 166.4, 158.3, 133.2, 132.6, 122.4, 119.3, 117.7, 68.6; ms: (CI) *m/z* 223 (MH^+).

Anal. Calcd. for $C_{11}H_{10}O_5$: C, 59.46: H, 4.54. Found: C, 59.62; H, 4.67.

5-(Allyloxy)isophthaloyl Chloride (**6**).

Oxalyl chloride (6.42 g, 0.05 mole) was added dropwise to a solution of the diacid (2.81 g, 0.0126 mole) in 100 ml of dry toluene. The mixture was stirred for 4 hours at 80° and then the toluene and excess oxalyl chloride were evaporated. The residual oil was distilled under high vacuum. The less colored oil was redistilled twice to give **6** (2.3 g, 70%), bp 210°/0.8-1.0 mm; ^{1}H nmr (dimethyl-d_6 sulfoxide): δ 8.45-8.44 (m, 1H), 7.91-7.90 (m, 2H), 6.10-5.99 (m, 1H), 5.51-5.37 (m, 2H), 4.70-4.67 (m, 2H); ^{13}C nmr (dimethyl-d_6 sulfoxide): δ 167.3, 159.4, 135.5, 131.7, 126.3, 123.3, 119.3, 69.9.

Anal. Calcd. for $C_{11}H_8Cl_2O_3$: C, 50.99: H, 3.11. Found: C, 50.81; H, 3.30.

Chiral Selector **2.**

Chiral diamine **5** (0.52 mg, 0.00145 mole) and dichloride **6** (0.376 mg, 0.00145 mole) were each dissolved in 20 ml of dry methylene chloride and the solutions were placed in different syringes. The solutions were added with a dual-channel syringe pump to a stirred solution of triethylamine (1 ml, 0.007 mole) and 100 ml of dry methylene chloride over a period of 8 hours. The mixture was then stirred for 20 minutes and filtered. The filtrate was evaporated and the residue was purified by flash chromatography eluting with (1:4) hexanes-ethyl acetate to give compound **2** as a white solid (0.44 g, 56%), mp 240-242°; $[\alpha]_D^{20}$ = -73° (c = 1.0, methylene chloride); R_f = 0.26 (1:4/hexanes:ethyl acetate); ^{1}H nmr: δ 7.79 (s, 2H), 7.74-7.60 (m, 4H), 7.58-7.48 (m, 2H), 7.43 (s, 1H), 7.26-7.19 (m, 1H), 7.14 (s, 2H), 6.12-5.99 (m, 1H), 5.49-5.30 (m, 2H), 4.60-4.58 (m, 2H), 3.90 (broad s, 4H), 2.31-2.27 (m, 4H), 1.92-1.85 (m, 4H), 1.52-1.41 (m, 8H); ^{13}C nmr: δ 169.3, 169.1, 158.5, 136.5, 135.1, 132.7, 129.8, 128.8, 125.7, 118.3, 118.2, 116.6, 69.3, 55.5, 32.2, 25.1; ms: (fab) *m/z* 545 (MH^+), 567(MNa^+); hrms: (fab) *m/z* Calcd. for $C_{31}H_{37}O_5N_4$ (MH^+): 545.2674; Found: 545.2750.

Anal. Calcd. for $C_{31}H_{36}N_4O_5$: C, 68.36: H, 6.66. Found: C, 68.48; H, 6.66.

Ethoxydimethylsilane-containing Macrocyclic Tetraamide **7.**

Allyloxy-substituted macrocycle **2** (0.17 g, 0.312 mmole) was dissolved in 3 ml of methylene chloride. To this solution were added chlorodimethylsilane (1.4 ml, 0.0125 mole) and dihydrogen platinum hexachloride hydrate (4 mg in 0.1 ml of dry tetrahydrofuran) [16]. The mixture was refluxed for 40 minutes and evaporated to dryness. The solid residue was dissolved in 3 ml of methylene chloride, and 2 ml of 1:1 ethanol-triethylamine was added dropwise to the solution at 0°. After stirring at room temperature for 30 minutes, the mixture was evaporated to dryness and the solid residue was chromatographed on silica gel (3:7 hexanes-ethyl acetate) to give **7** as a white solid (0.101 g, 50%), mp 202-204°; R_f = 0.41(1:4/hexanes:ethyl acetate); ^{1}H nmr: δ 7.81 (s, 1H), 7.74-7.70 (m, 4H), 7.56-7.44 (m, 2H), 7.40 (s, 1H), 7.27-7.20 (m, 1H), 7.11 (s, 2H), 4.00-3.92 (m, 6H), 3.70 (q, *J* = 7.2 Hz, 2H), 2.30- 2.26 (m, 4H), 1.92-1.81 (m, 6H), 1.58-1.32 (m, 8H), 1.21 (t, *J* = 7.2 Hz, 3H), 0.75-0.69 (m, 2H), 0.16 (s, 3H), 0.15 (s, 3H); ^{13}C nmr: δ 169.3, 159.1, 136.4, 135.0, 129.9, 128.7, 125.7, 118.0, 116.4, 71.0, 58.6, 55.5, 32.2, 25.1, 23.4, 18.8, 12.6, -1.87, -1.91; hrms: (fab) *m/z* Calcd. for $C_{35}H_{48}N_4O_6SiNa$ (MNa^+): 671.3240; Found: 671.3237.

Anal. Calcd. for $C_{35}H_{48}N_4O_6Si$: C, 64.79: H, 7.46. Found: C, 64.60; H, 7.24.

Chiral Stationary Phase **1.**

The ethoxysilane compound **7** (82 mg, 0.127 mmole) and silica gel (5 μm, 350 mg, dried at 270° for 24 hours under helium) were suspended in 20 ml of dry toluene. The mixture was stirred at 80° for 109 hours. The bonded phase was then filtered and washed successively with methanol, acetone, ethyl acetate, toluene, hexane and diethyl ether. Elemental analysis of the dried bonded phase (C: 6.95, N: 0.77) showed a loading of approximately 0.16 mmole of chiral selector per gram of stationary phase based on carbon; 0.14 mmole based on nitrogen; ir (potassium bromide): 1684, 1654 cm^{-1}.

X-ray Crystal Structure of **3.**

A suitable crystal of **3** was mounted on an automated Bruker P4 diffractometer and crystal and intensity data were obtained using MoKα radiation (λ = 0.71073Å). A summary of crystal data and experimental conditions are listed in Table 2. Positional and equivalent isotropic thermal parameters for all atoms are listed in Table 3.

The crystal structure was solved using direct methods. Nonhydrogen atoms were refined anisotropically. Positions for all hydrogen atoms bonded to carbon atoms were calculated based on known molecular geometry. Positions for hydrogen atoms bonded to nitrogen atoms were obtained from a difference map. The map showed three hydrogen atoms bonded to N5 in a tetrahedral arrangement. It was also possible to find the position for one hydrogen atom bonded to N7. This hydrogen atom, H7, and a hydrogen atom H5CA of a symmetry related molecule are involved in intermolecular hydrogen bonds between N7 and N5A of the two molecules (Table 1) but the hydrogen atoms are only about 1Å apart. This can only be explained by the presence of the disorder which has been described earlier. The structure was solved, refined and displayed using programs included in the program package provided by Bruker [20].

Packed Capillary Column LC Experiments.

A liquid chromatography pump (Varian 8500, Walnut Creek, CA) was used to deliver the mobile phase. The packed capillary column was directly connected to a manual liquid injector valve (Valco, 60-nL sample loop). A 30 cm x 100 μm i.d. fused silica capillary was used to connect the column outlet to a uv absorption detector (uv-vis Spectra 100, Thermo Separation Products, San Jose, CA). A Model SP 4270 integrator (Spectra-Physics, San Jose, CA) was used to record the chromatograms.

Column Preparation.

The bonded particles (chiral stationary phase **1**) were dried for 20 hours under a purge of helium. For end-capping, 1 ml of hexamethyldisilazane was dissolved in 3 ml of methylene chloride and the solution was transferred into a reaction vessel containing 0.1 g of dried chiral stationary phase. The mixture was sonicated for 5 minutes and the solvent was evaporated by purging with helium. The vessel was heated for 2 hours at 150% under a purge of helium. The cooled particles were then washed with 20 ml of methylene chloride and then with methanol. A carbon dioxide (SFC grade, Scott Specialty Gases Plumsteadville, PA) slurry packing method was used to prepare the packed fused silica capillary column [21].

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