Article

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J. Comb. Chem., **1999**, 1 (1), 105-112• DOI: 10.1021/cc980014p • Publication Date (Web): 18 December 1998

A Combinatorial Approach to Recognition of Chirality: Preparation of Highly Enantioselective Aryl-Dihydropyrimidine Selectors for Chiral HPLC

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Received September 8, 1998

A parallel library of 108 4-aryl-1,4-dihydropyrimidine (DHPM) enantiomers, which are potential selectors for chiral HPLC separations, was synthesized using the single-step Biginelli multicomponent condensation. The individual compounds were screened by observing the enantioselectivity for resolution on a "brush-type" L-(3,5-dinitrobenzoyl)leucine-based chiral stationary phase, and separation factors α up to 12 were achieved. The best candidates from the library contained an ortho-substituted aromatic group at C4 carbon atom of the pyrimidine ring and an alkyl substituent at N1 nitrogen atom. Resolution of the enantiomers of the lead compound, 4-(9-phenanthryl)-DHPM **8**, using semipreparative chiral HPLC followed by attachment to monodisperse macroporous aminomethacrylate beads, provided the novel polymer based chiral stationary phase with good enantioselectivities in the resolution of several π-acidic aryl-dihydropyrimidines and derivatized profens. In addition, 3,5-dinitrobenzamido derivatives of α-amino acids could be resolved under normal phase HPLC conditions with separation factors up to 8.

Introduction

The continuing trend to replace racemic drugs, agrochemicals, flavors, and some other products with their single enantiomers is driven by increasingly restrictive regulatory requirements due to the awareness that individual enantiomers have different interactions with biological systems. There are several methods to obtain enantiomerically pure compounds:[1] (i) syntheses based on chiral starting materials from natural sources (pool of chiral building blocks); (ii) enantioselective reactions; (iii) separation of a racemic mixture using methods such as crystallization via diastereoisomers, enzymatic or chemical kinetic resolution, and chromatographic separation. The synthesis of enantiomerically pure compounds often leads to products enriched to a certain degree with one enantiomer rather than a completely pure compound. Therefore, an additional purification step is required if the product is needed in an enantiomerically pure state (100% ee).

The liquid chromatographic method of separation of enantiomers using chiral stationary phases (CSP) emerged about one decade ago as a method useful for analytical assays in clinical testing as well as preparative and production scale separations due to its high efficiency and ease of operation in any scale. Preparative chromatography permits pharmacological and toxicological studies to already be carried out before an asymmetric synthesis has been developed and provides both enantiomers that are required for comparative biological testing. Therefore, a large number of CSPs have been developed to meet the challenge of enantiomer separations, and numerous CSPs are now commercially available.[2] Most commercial CSPs contain either chiral polymers or small-molecule selectors supported by porous silica beads. Optically active polymers, such as modified cellulose, polyacrylates, and proteins, have been used successfully for a variety of enantioseparations of small molecules.[2] Unfortunately, the mechanism of separation for these CSPs is not completely understood, which makes it difficult to develop new media of this type. In contrast, bonded natural and synthetic chiral selectors such as antibiotics, substituted cyclodextrins, crown ethers, and "Pirkle-type" selectors have several advantages including well-defined molecular structures and enantiomer "recognition" models.

The separation of enantiomers on a CSP requires the formation of diastereoisomeric adsorbate "complexes" between the analyte and the CSP. Enantiomer recognition with Pirkle's "brush-type" stationary phases is achieved as a result of three simultaneous attractive interactions (donor−acceptor interactions such as hydrogen bonding, π-stacking, dipole−dipole interactions, etc.) between the selector and one of the enantiomers being separated, with at least one of the interactions being stereochemically dependent[3] (the Dalgliesh's three-point model[4]). Compared to all others, Pirkle-type systems afford the most flexibility in the development of many different chiral stationary phases for the separation of a broad range of analyte types.[5,6] The majority of the original chiral selectors for "brush-type" CSPs was derived from natural sources. Selectors prepared from quinine[7] and amino acids, such as phenyl glycine and leucine,[8] are the most common examples. Further development of highly selective CSPs requires the design of new types of synthetic receptors [9] that also make use of compounds outside the pool of natural chiral building blocks.

A CSP is a complex system that involves the solid support, the selector, and the linker connecting the two. Ideally,

10.1021/cc980014p CCC: $18.00
Published on Web 12/18/1998

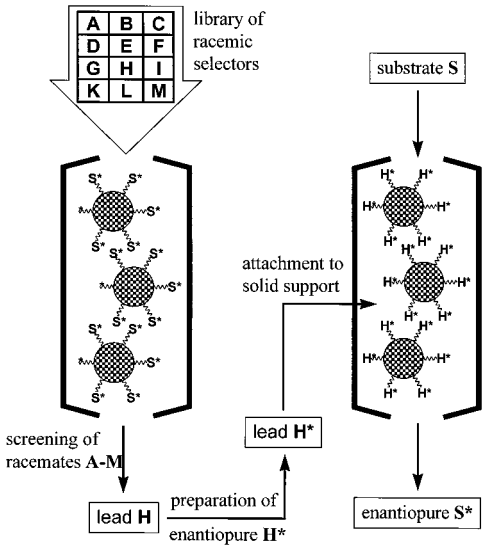


Figure 1. Concept of combinatorial approach to the preparation of chiral stationary phases.

interactions that lead to enantioseparation are maximized while nonspecific interactions are completely suppressed. For example, we have recently designed a polymer-bound "brush-type" chiral stationary phase with improved performance based on our advanced size monodisperse porous organic polymer beads. We demonstrated that separation media based on an organic polymer support provide greatly enhanced enantioselectivities and reduced retention times when compared to analogous silica-based chiral stationary phases as a result of substantially decreased nonspecific interactions.[10] Similarly, a study of common linkers led to the design of binding chemistry with minimized effects on enantiomer recognition.[11]

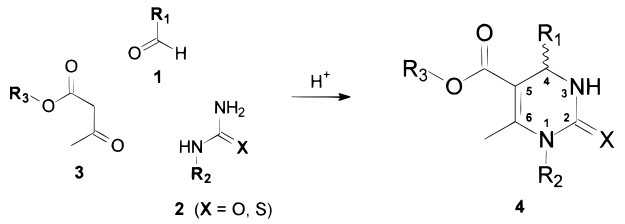
Combinatorial chemistry[12] is a powerful tool for the rapid preparation of large numbers of different compounds with numerous applications in the development of new drugs and drug candidates,[12] metal complexing ligands,[13] polymers,[14] materials for electronics,[15] sensors,[16] and peptidic ligands for affinity chromatography[17] and in supramolecular chemistry.[18] Such an approach provides the diversity needed for the discovery of lead compounds and allows prompt optimization.

In this article, we describe one possible approach to use combinatorial chemistry as a tool for the rapid development of novel, highly selective chiral stationary phases for HPLC (Figure 1). This approach involves an application of the so-called principle of reciprocity[19] which has been used by Pirkle et al. for the development of CSPs capable of separating important classes of pharmaceuticals, such as β-adrenergic blockers[20] and nonsteroidal antiinflammatory drugs (α-arylpropionic acids, profens).[9b]

Results and Discussion

Multicomponent condensation strategies[21] bring together a substrate and two or more reactants and provide durable core structures and highly variable side chains in a single step from simple starting materials. Therefore, these strategies are important for combinatorial syntheses of small-molecule libraries. The Biginelli dihydropyrimidine synthesis, first

Scheme 1



reported more than 100 years ago,[22] is one class of multicomponent condensation that has been recently adopted to solid[23] and fluorous phase[24] syntheses. This reaction involves a one-pot cyclocondensation of β-keto esters, aldehydes, and ureas, providing heterocycles that have a well-established pharmacological potential[25−27] (Scheme 1). For our purpose, it was essential that a variety of substituted compounds be obtained in which an aromatic substituent R_1 is appended to a chiral center also flanked by polar or hydrogen-bonding functionalities. We applied the classical solution three-component Biginelli condensation protocol to obtain a small parallel library of 108 individual 4-aryl-1,4-dihydropyrimidine (DHPM) enantiomers **4**.[28]

In a typical reaction, a urea was mixed with 1 equiv of an aldehyde and 1.5 equiv of a β-keto ester in the presence of catalytic HCl in ethanol, and the mixture was stirred for 3 h under reflux. The product was isolated from the crude reaction mixture by simple crystallization and provided the racemic DHPM in 20−60% yield and >80% purity (according to ^{1}H NMR).[29] The reactions were done on a 7−15 mmol scale to provide 0.5−1 g of the final DHPMs. Most of the substituents around the DHPM ring system were varied by simply choosing from a large set of aromatic aldehydes, ureas/thioureas, and acetoacetates/acetoacetamides shown in Figure 2. Most of these compounds are commercially available or could be easily prepared. With this small library of diverse racemic DHPMs in hand, a basis was set to establish a correlation between the structural features of our DHPMs and the enantioselectivity achieved in a chiral separation process.

Screening. To perform such a correlation, our library was "screened" with a CSP based on a (*S*)-(3,5-dinitrobenzoyl)-leucine selector **6**, which has been extensively used for the enantioseparations of various classes of compounds, including benzodiazepinones and succinimides.[30,31] This "inverse" selector was immobilized on monodisperse macroporous poly((*N*-methyl)aminoethyl methacrylate-*co*-methyl methacrylate-*co*-ethylene dimethacrylate) beads **5** via a tertiary amide bond (CSP **7**, Scheme 2). The physical and chemical properties of these polymeric beads were previously optimized for the "classical" preparation of highly selective chiral stationary phases.[11]

The screening results of our DHPM library using CSP **7** under standard normal phase conditions with CH_2Cl_2 as the mobile phase are summarized in Tables 1 and 2 and in Figures 5 and 6. While some racemic DHPMs in the library are not resolved at all (separation factor $\alpha = 1.0$), rather high α values of up to 5.2 were achieved for the top candidates such as 4-(9-phenanthryl)-DHPM **8** (entry 41). Figure 3 shows the chromatographic separation of this racemate.

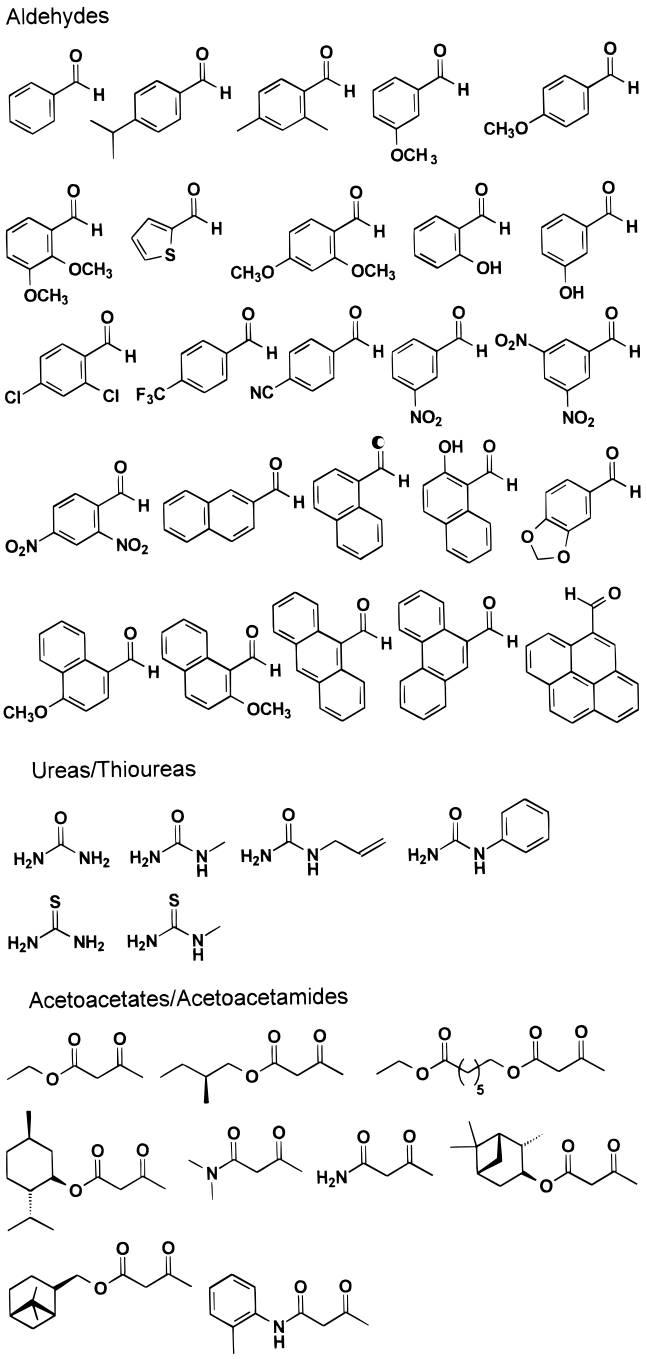


Figure 2. Building blocks for the DHPM library.

Inspection of the data in Tables 1 and 2 reveals some of the structural requirements necessary for enantiomer recognition. By comparing N-alkylated DHPMs either at positions 1 or 3 of the ring shown in Scheme 1 (and thus, different hydrogen-bonding sites), several enantioseparation effects were noted. First, as expected, only DHPMs containing a hydrogen-bonding donor at position 3 next to the chiral center were separated. Remarkably, DHPMs with nonsubstituted nitrogen atoms at positions 1 and 3 (Table 1, entries 1, 8, 15, 21, 23, 37, 43; Table 2, entries 1, 3, 5, 9) resulted in separations with longer retention times and decreased separation factors α. This is most likely due to simultaneous hydrogen bonding at both nitrogen sites, and, therefore, increased nonspecific interactions between the selector and the DHPM. Replacement of the hydrogen atom at the N1 nitrogen atom with an alkyl substituent eliminates the nonspecific interaction and improves the separation.

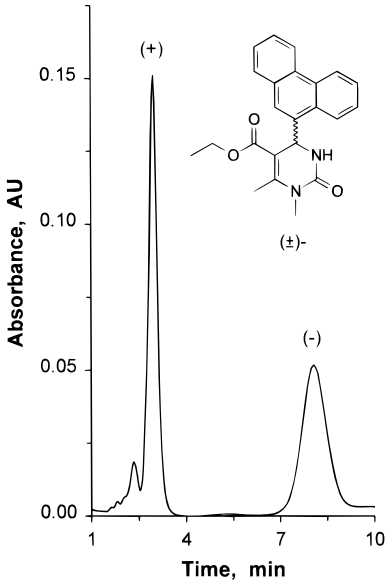
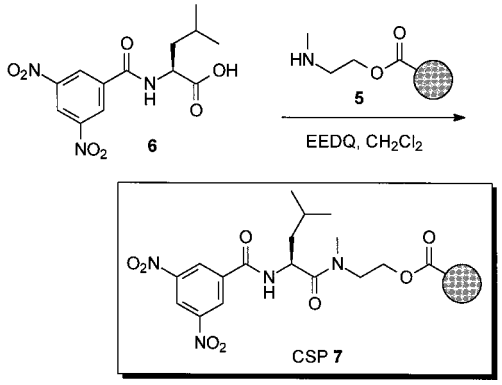


Figure 3. Separation of DHPM (±)-**8** (entry 44) on chiral stationary phase **7**. Conditions: column, 150 × 4.6 mm i.d.; mobile phase, dichloromethane; flow rate, 1 mL/min.

Scheme 2



If a chiral aliphatic substituent is introduced at the exocyclic ester bond, only moderate changes in enantioselectivity occur (e.g., Table 1, entry 30 versus 24). Interestingly, the presence of an additional hydrogen-bonding site, such as an exocyclic secondary amide bond, does not seem to affect the separation process (Table 1, entry 34 versus 24).

Not surprisingly, increasing the π-basicity of the aromatic group at C4 results in higher separation factors due to a stronger interaction with the π-acidic 3,5-dinitrobenzoyl group of CSP **7** used for the screening. However, the substitution pattern as well as the π-basicity of the aromatic group plays an essential role. DHPMs with ortho-substituted aromatic groups show much higher enantioselectivities compared to meta- and para-substituted groups. For example, the observed enantioselectivity for the 1-naphthyl-DHPM (entry 24) is almost twice that of the corresponding 2-naphthyl derivative (entry 22). Nevertheless, addition of a second ortho substituent in the aromatic ring of the DHPMs leads to a dramatic deterioration of enatioseparation as observed with the 9-anthryl- (entry 40) or 2-methoxy-1-naphthyl-substituted (entry 39) DHPMs.

Table 1. Separation of Aryl-DHPMs **4** (X = O) on CSP **7**[a]

	DHPM derivative[b]			HPLC data	
entry	R_1	R_2	R_3	k_2'	α
1	phenyl	H	ethyl	2.77	1.0
2	phenyl	methyl	ethyl	0.49	1.0
3	4-^{i}PrPh	methyl	ethyl	0.33	1.0
4	2,4-Me_2Ph	methyl	ethyl	0.76	1.5
5	3-MeOPh	methyl	ethyl	0.57	1.3
6	4-MeOPh	methyl	ethyl	0.69	1.2
7	2,3-$(MeO)_2$Ph	methyl	ethyl	0.16	1.2
8	2,4-$(MeO)_2$Ph	H	ethyl	1.81	1.1
9	2,4-$(MeO)_2$Ph	methyl	ethyl	0.35	2.9
10	3,4-(OCH_2O)Ph	methyl	ethyl	0.58	1.0
11	"2-(OH)Ph"[c]	methyl	ethyl	0.85	1.0
12	3-(OH)Ph	methyl	ethyl	2.90	1.0
13	2,4-Cl_2Ph	methyl	ethyl	0.33	1.0
14	3-NO_2Ph	methyl	ethyl	0.73	1.0
15	3,5-$(NO_2)_2$Ph	H	ethyl	3.32	1.0
16	3,5-$(NO_2)_2$Ph	methyl	ethyl	1.08	1.0
17	2,4-$(NO_2)_2$Ph	methyl	ethyl	0.13	1.0
18	4-(CN)Ph	methyl	ethyl	0.66	1.0
19	4-(CF_3)Ph	methyl	ethyl	0.66	1.0
20	2-thiophene	methyl	ethyl	0.71	1.0
21	2-naphthyl	H	ethyl	3.60	1.0
22	2-naphthyl	methyl	ethyl	0.89	1.7
23	1-naphthyl	H	ethyl	4.26	1.4
24	1-naphthyl	methyl	ethyl	1.86	3.2
25	1-naphthyl	allyl	ethyl	0.95	2.3
26	1-naphthyl	phenyl	ethyl	0.87	2.6
27	1-naphthyl	3,5-Me_2Ph	ethyl	0.72	2.7
28	1-naphthyl	methyl	2*S*-Me-1-Bu	1.63	3.5[d]
29	1-naphthyl	methyl	(1*S*,2*S*,5*S*)-myrtanyl	1.47	3.6[d]
30	1-naphthyl	methyl	(1*R*,2*S*,5*R*)-menthyl	1.17	4.1[d]
31	1-naphthyl	methyl	(1*S*,2*S*,3*S*,5*R*)-3-pinanyl	1.15	2.8[d]
32	1-naphthyl	methyl	H_2N	3.56	1.0
33	1-naphthyl	methyl	Me_2N	0.83	1.7
34	1-naphthyl	methyl	(2-MePh)HN	1.86	3.5
35	"2-OH-1-naphthyl"[c]	methyl	ethyl	3.24	2.1
36	5-NO_2-1-naphthyl	methyl	ethyl	1.92	2.4
37	4-MeO-1-naphthyl	H	ethyl	6.77	1.9
38	4-MeO-1-naphthyl	methyl	ethyl	3.18	5.2
39	2-MeO-1-naphthyl	methyl	ethyl	0.46	1.3
40	9-anthryl	methyl	ethyl	0.54	1.0
41	9-phenanthryl	methyl	ethyl	3.52	5.2
42	9-phenanthryl	methyl	2*S*-Me-1-Bu	3.50	5.2[d]
43	1-pyrenyl	H	ethyl	9.56	1.6
44	1-pyrenyl	methyl	ethyl	5.81	4.8

[a] Conditions: column, 150 × 4.6 mm i.d.; mobile phase, CH_2Cl_2; flow rate, 1 mL/min; UV detection at 254 nm. [b] See Scheme 1 for substituents R_1-R_3. [c] DHPM is a bicyclic, oxygen-bridged structure, see ref 33. [d] Separation factor of diastereoisomeric enantiomeric pairs.

To further elucidate this ortho-substitution effect, we performed a single-crystal X-ray structure analysis of the racemic 4-(9-phenanthryl)-DHPM **8** (entry 41) which exhibits the highest enantioselectivity (α = 5.2) among the series of 2-oxo-DHPM derivatives shown in Table 1. The solid-state structure of this compound depicted in Figure 4 shows a half-boat-like ("sofa") conformation[32] with the 9-phenanthryl group in a quasi-axial or quasi-flagpole position and the α,*β*-unsaturated exocyclic ester in a *s-cis* conformation (Figure 4a). The sterically demanding "ortho-substituted side" of the 9-phenanthryl group is positioned *anti-periplanar* to the C4−C5 bond, whereas the "meta-substituted side" is *syn-periplanar* (Figure 4b). This essentially creates a cleft-like

Table 2. Separation of Aryl-DHPMs **4** (X = S) on CSP **7**[a]

	DHPM derivative[b]			HPLC data	
entry	R_1	R_2	R_3	k_2'	α
1	phenyl	H	ethyl	1.42	1.2
2	phenyl	methyl	ethyl	0.29	1.0
3	1-naphthyl	H	ethyl	2.17	1.8
4	1-naphthyl	methyl	ethyl	1.10	4.7
5	4-MeO-1-naphthyl	H	ethyl	2.80	2.7
6	4-MeO-1-naphthyl	methyl	ethyl	2.29	8.8
7	9-phenanthryl	methyl	ethyl	2.57	8.0
8	9-phenanthryl	methyl	$EtOCOC_6H_{13}$	1.69	11.7
9	1-pyrenyl	H	ethyl	5.43	3.1
10	1-pyrenyl	methyl	ethyl	7.86	9.8

[a] Conditions: column, 150 × 4.6 mm i.d.; mobile phase, CH_2Cl_2; flow rate, 1 mL/min; UV detection at 254 nm. [b] See Scheme 1 for substituents R_1-R_3.

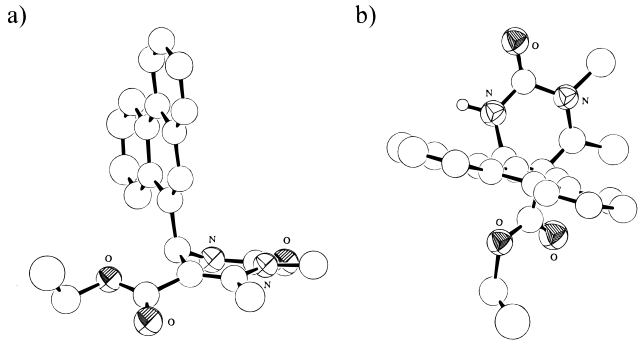


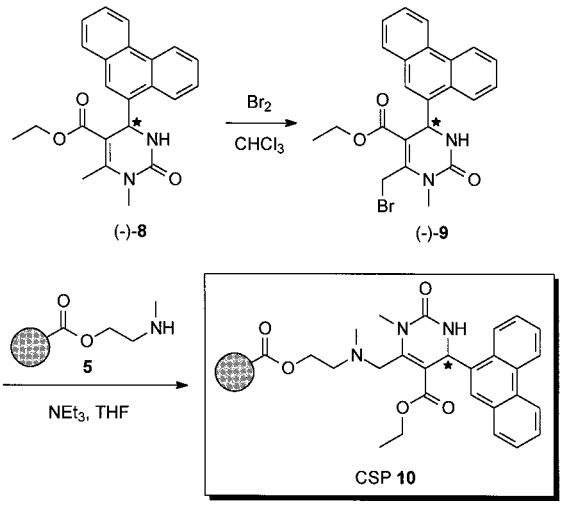
Figure 4. ORTEP representation of DHPM (±)-**8** (entry 44; see structure in Figure 3). The hydrogen atoms are not shown for clarity.

structure between the aromatic and heterocyclic ring systems. This is in contrast to the recently published conformational analysis of 4-aryl-DHPMs containing a nonalkylated N1 position and an ortho-substituted phenyl at C4.[33] In this structure, the aromatic group was nearly bisecting the DHPM ring (*synclinal* to C4−C5) with the ortho-substituent *syn-periplanar* to C4−H.

Finally, another important effect on enantioselectivity was observed when a thiourea was used instead of a urea input (see results in Table 2). In general, the selectivity factor for the 2-thio analogues increased 2-fold compared to the corresponding 2-oxo-DHPMs shown in Table 1. The compound exhibiting the highest selectivity was a 4-(9-phenanthryl)-DHPM (α = 11.7; see entry 8 in Table 2). Obviously, the lower electronegativity of the sulfur atom leads to a decrease in the extent of undesired nonspecific interactions between the analyte and selector.

Preparation of the Chiral Stationary Phase. In the next step, the best candidate from the series had to be isolated in enantiomerically pure form and attached to a macroporous support. Several methods have been described for the resolution of racemic DHPMs.[34] Most of these techniques involve multistep preparations of diastereoisomeric DHPMs followed by fractional crystallization. Although we prepared several diastereoisomeric DHPMs by including a chiral acetoacetate in the Biginelli reaction (see entries 28−31 and 42, Table 1) and also tried to form diastereoisomeric salts after hydrolysis of the exocyclic bond, the diastereoisomers could not be resolved by crystallization even by using a large variety of solvent mixtures. Since our HPLC method used for the screening was very successful and resulted in an

Scheme 3

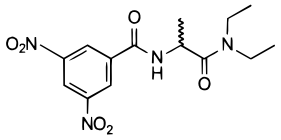
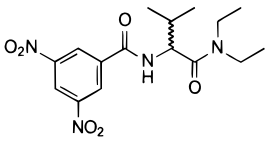
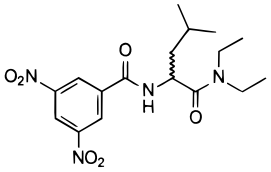
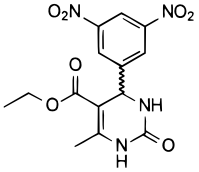
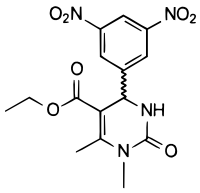
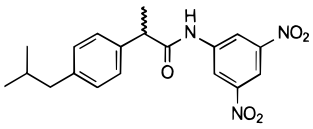
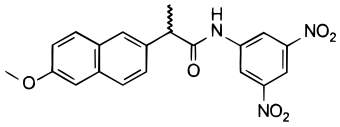


excellent separation of the individual enantiomers, it was scaled-up to a semipreparative size. A 60 × 0.8 cm i.d. stainless steel column packed with polymer-bound (*S*)-(3,5-dinitrobenzoyl)leucine (CSP **7)** identical to that utilized for the screening was used to obtain the desired milligram quantities of 2-oxo-4-(9-phenanthryl)-DHPM (−)-**8** (>98% ee), which was selected as an example for the implementation of the chiral separation medium.

To attach the isolated selector to the amino functionalized macroporous polymethacrylate support **5**, a suitable reactive group had to be made available. Although hydrolysis of the ester group of substituted DHPM is a tempting route to provide a reactive handle, this was not used because it requires harsh conditions and leads to a number of light-sensitive byproducts detected by both TLC and NMR in the product of this reaction that turned colored over time. A similar observation was made by Kappe[34] for other DHPM derivatives. A much more convenient route to introduce a reactive site in DHPM is the functionalization of the methyl group at the C6 carbon atom by a simple bromination.[35] Unfortunately, this reaction affords very low yields for 2-thio-DHPM. In contrast, the 6-bromomethyl-DHPM (−)-**9** was obtained in 95% yield using bromine at room temperature in chloroform without any racemization in the product, as determined by HPLC. Coupling of (−)-**9** to the amino functionalized support **5** gave the chiral stationary phase **10** (Scheme 3), which contains 0.20 mmol/g of the DHPM selector, according to its elemental analysis for nitrogen.

Separation of Enantiomers with the Dihydropyrimidine Chiral Stationary Phase. CSP **10** was packed into a 150 × 4.6 mm i.d. HPLC column, and a variety of analytes were tested under normal phase HPLC conditions. In general, CSP **10** exhibits excellent separation factors up to values of about 8 for a variety of racemic α-amino acid derivatives (Table 3). These selectivities are slightly higher than those determined in the reciprocal experiments, although a direct comparison is impossible due to the extra functionality introduced for binding the selector onto the polymer support. Unexpectedly, the substituted alanine derivatives are separated with much higher enantioselectivities than the corresponding leucine-based analytes. Figure 7 shows an example of a typical separation of 3,5-dinitrobenzamidoalanine-*N*,*N*-diethylamide enantiomers obtained using chiral stationary phase **10**. Once again the selectivity is good as a separation factor α of 7.7 is measured. Even though CSP **10** was designed for the separation of derivatized amino acids, other classes of compounds can also be resolved. For example, Table 3 shows that several π-acidic aryl-dihydropyrimidines and derivatized profens can be separated with reasonably high enantioselectivities.

Table 3. Separation of Analytes on CSP **10**[a]

entry	analyte	k_2'	α
1		6.75	7.66
2		4.70	3.99
3		6.94	5.74
4		10.18	1.24
5		1.31	1.34
6		3.02	1.32
7		3.52	1.25

[a] Conditions: column, 150 × 4.6 mm i.d.; mobile phase, hexane/dichloromethane; flow rate, 1 mL/min; UV detection at 254 nm.

CSP **10** can also be utilized under reversed phase conditions. Figure 8 shows the effect of water content in the mobile phase on the observed separation factor α for the separation of 3,5-dinitrobenzamidoalanine-*N*,*N*-diethylamide enantiomers. The value of α decreases with the amount of water introduced into the mobile phase. This is likely due to the suppression of the hydrogen-bonding interactions between the CSP and analyte. However, rather good enantioselectivities can still be obtained with up to 50% water (α = 3.5). Therefore, CSP **10** is a versatile phase capable of enantioseparations in either reversed phase or normal phase mode.

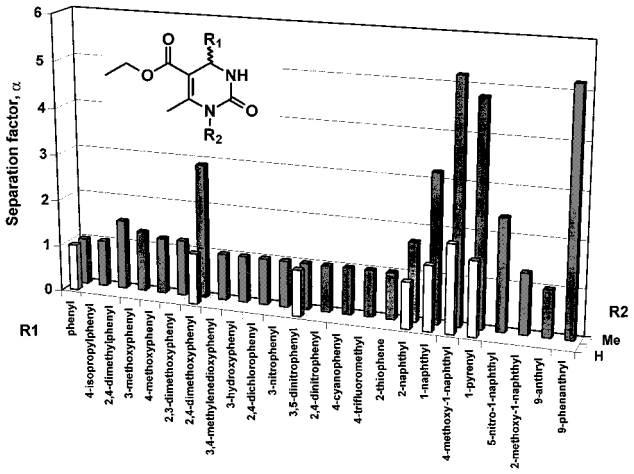


Figure 5. Selectivity factors for the separations of sublibraries of racemic ethyl 6-methyl- and ethyl 1,6-dimethyl-2-oxo-4-substituted-1,2,3,4-tetrahydropyrimidine-5-carboxylates.

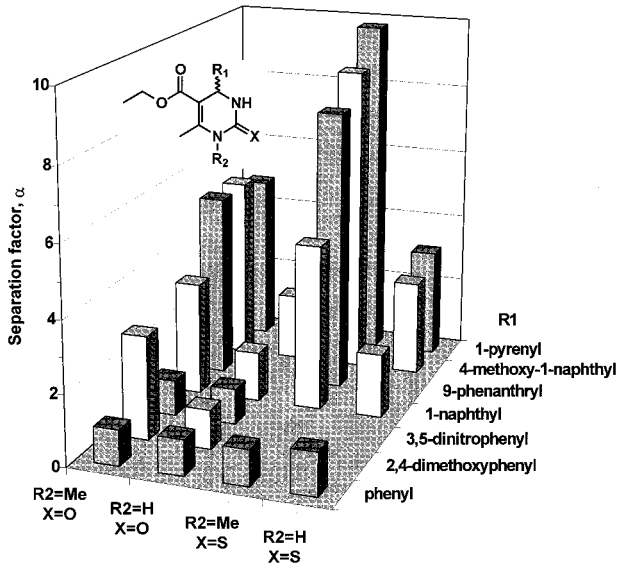


Figure 6. Selectivity factors for the separations of sublibraries of racemic ethyl 6-methyl-2-oxo-, ethyl 1,6-dimethyl-2-oxo-, ethyl 6-methyl-2-thio-, and ethyl 1,6-dimethyl-2-thio-4-substituted-1,2,3,4-tetrahydropyrimidine-5-carboxylates.

Conclusion

In addition to typical drug discovery, combinatorial chemistry is a powerful tool in molecular recognition as evident by this demonstration aimed at new selectors for chiral HPLC.[38] The strategy, based on the principle of reciprocity, is general and may be used with a broad array of libraries of potential racemic selectors. Although we have chosen a simple Biginelli three-component condensation reaction to prepare our library of selectors, many existing libraries of organic compounds could also be screened as potential selectors for chiral recognition. When such a study is carried out with structurally related families of compounds, a better understanding of chiral recognition may be generated.

Experimental Section

General. NMR (^{1}H, ^{13}C) spectra were recorded on Bruker AMX-300 and 400 spectrometers at room temperature. Chemical shifts, δ, are quoted in ppm downfield from internal

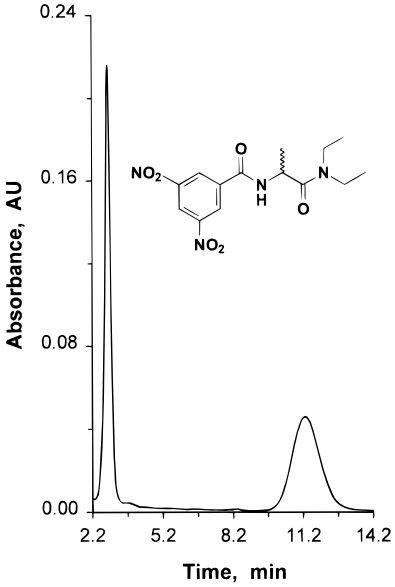


Figure 7. Separation of 3,5-dinitrobenzamidoalanine-*N*,*N*-diethylamide enantiomers on chiral stationary phase **10**. Conditions: column, 150 × 4.6 mm i.d.; mobile phase, dichloromethane; flow rate, 1 mL/min.

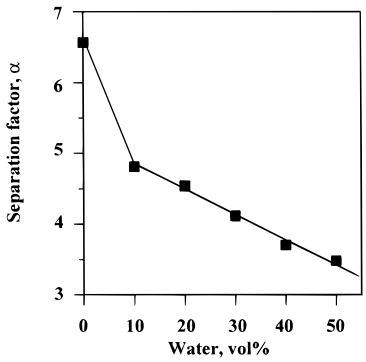


Figure 8. Effect of the water content in the mobile phase on the separation factor α for the reversed phase separation of 3,5-dinitrobenzamidoalanine-*N*,*N*-diethylamide enantiomers. Conditions: CSP **10**; column, 150 × 4.6 mm i.d.; mobile phase, acetonitrile-water; flow rate, 1 mL/min.

tetramethylsilane with coupling constants, *J*, in hertz. Optical rotations were measured with a Perkin-Elmer 241 polarimeter at room temperature using a 10 cm path length cell.

General Procedure for the Biginelli Reactions. A solution of aldehyde **1** (7.4 mmol), urea **2** (7.4 mmol), and acetoacetate or acetoacetamide **3** (11.1 mmol) in ethanol (4 mL) was treated with 1 drop of concentrated hydrochloric acid and heated to reflux for 3 h. The resulting solution was cooled to 0 °C, and the product **4** was collected by filtration and recrystallized from ethanol. Alternatively, the solvent was removed under vacuum, and the residue was purified by column chromatography on silica gel using ethyl acetate/hexane.

Ethyl 1,6-Dimethyl-2-oxo-4-(9-phenanthryl)-1,2,3,4-tetrahydropyrimidine-5-carboxylate ((±)8, Entry 41, Table 1). Compound (±)-**8** was prepared according to the general procedure with 9-phenanthrene carboxaldehyde (1.00 g, 4.8 mmol), *N*-methyl urea (0.36 g, 4.8 mmol), and ethyl acetoacetate (0.94 g, 7.2 mmol) to afford **8** (1.25 g, 70%) as a white solid: mp 166−170 °C; ^{1}H NMR (400 MHz, $CDCl_3$)

δ 0.90 (t, $J = 7.1$ Hz, 3H), 2.72 (s, 3H), 3.26 (s, 3H), 3.87−4.02 (m, 2H), 5.58 (s, 1H), 6.28 (s, 1H), 7.55−7.69 (m, 5H), 7.82 (d, $J = 6.7$ Hz, 1H), 8.08−8.10 (m, 1H), 8.62−8.79 (m, 2H); ^{13}C NMR (75 MHz, $CDCl_3$) δ (13.8, 13.9), (16.3, 16.5, 16.6, 16.8), (30.0, 30.2, 30.3, 30.4), (49.6, 49.8), (59.8, 60.0), 102.3, (122.4, 122.5), (122.9, 123.0), (123.4, 123.5), (124.6, 124.7), 126.5−127.3, (126.2, 126.3), (128.7, 128.9), 129.4, 130.2, 131.1, 131.3, 135.6, 150.6, 153.6, 166.3; MS (EI) *m*/*z* 374, 345, 301, 197, 178. Anal. Calcd for $C_{23}H_{22}N_2O_3$ (374.43): C, 73.78; H, 5.92; N, 7.48. Found C, 74.00; H, 6.12; N, 7.23.

Compound (±)-**8** was separated by semipreparative HPLC on CSP **7** using a 60 × 0.7 cm i.d. column. More retained enantiomer (−)-**8**: $[\alpha]_D$ −10 ($c = 0.10$, MeOH). Less retained enantiomer (+)-**8**: $[\alpha]_D$ +11 ($c = 0.10$, MeOH).

X-ray Structure of (±)-8. A colorless platelike crystal of (±)-**8** having approximate dimensions 0.30 mm × 0.28 mm × 0.05 mm was obtained from a CH_2Cl_2 solution containing 1 equiv of *N*-methyl urea. The crystal was mounted on a glass fiber using Paratone N hydrocarbon oil. All measurements were made on a Siemens SMART CCD area detector with graphite monochromated Mo Kα radiation ($\lambda = 0.710\ 69$ Å). Cell constants and an orientation matrix, obtained from a least-squares refinement using the measured positions of 2726 reflections in the range $3.00 < 2\theta < 45.00°$, corresponded to an A-centered orthorhombic cell with dimensions $a = 21.460(2)$ Å, $b = 17.336(2)$ Å, $c = 11.246(1)$ Å, $\alpha = 90.000(0)°$, $\beta = 90.000(0)°$, $\gamma = 90.000(0)°$.The data were collected at a temperature of -100 ± 1 °C. Frames corresponding to an arbitrary hemisphere of data were collected using ω scans at 0.3° counted for a total of 10.0 s per frame. Data were integrated by the program SAINT (Siemens Industrial Automation, Inc., 1995) to a maximum 2θ value of 46.5°. A total of 8255 reflections was collected, of which 1711 were unique ($R_{int} = 0.063$).

The structure was solved by direct methods and expanded using Fourier techniques. All non-hydrogen atoms were refined isotropically. Assignment of the "solvent" region as methylurea was based on analysis of the shape of the region of electon density and on the known identities of compounds included in the synthesis and workup. Hydrogen atoms were included in the calculated positions, except for those on the disordered ester group of the molecule, but not refined. The final cycle of full-matrix least-squaresd refinement was based on 762 observed reflections ($I = 3.00\sigma(I)$) and 146 variable parameters and converged with R (R_w) = 0.081 (0.076).

Tables of the positional and thermal parameters are available as Supporting Information.

Ethyl 1-Methyl-6-bromomethyl-2-oxo-4-(9-phenanthryl)-1,2,3,4-tetrahydropyrimidine-5-carboxylate ((−)-9). To a solution of (−)-**8** (the more retained enantiomer from the HPLC separation, 0.35 g, 0.93 mmol) in chloroform (2 mL) at room temperature was slowly added a solution of Br_2 (50 μL, 0.97 mmol) in chloroform (2 mL). After 1.5 h the solvent was removed, and the residue was purified by column chromatography on silica gel using ethyl acetate/methylene chloride (1:9) to yield (−)-**9** as a yellow solid (0.42 g, 99%): $[\alpha]_D$ −531 ($c = 0.10$, MeOH); ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.93 (t, $J = 7.1$ Hz, 3H), 3.43 (s, 3H), 3.88−4.12 (m, 2H), 4.60 (br s, 1H), 5.49 (br s, 1H), 6.23 (d, $J = 3.0$ Hz, 6H), 6.48 (d, $J = 2.7$ Hz, 1H), 7.47−7.72 (m, 5H), 7.81 (d, $J = 7.8$ Hz, 1H), 8.02 (d, $J = 7.8$ Hz, 1H), 8.59 (t, $J = 8.7$ Hz, 2H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 14.2, 24.9, 30.7, 49.5, 61.1, 105.1, 122.8, 123.0, 123.9, 125.0, 126.7, 127.1, 127.2, 127.4, 129.3, 129.5, 130.6, 131.5, 131.6, 134.8, 148.8, 154.0, 165.0; MS (EI) *m*/*z* 452, 373, 345, 327, 299, 275, 197, 178; Anal. Calcd for $C_{23}H_{21}BrN_2O_3$ (453.33): C, 60.94; H, 4.67; N, 6.18. Found: C, 61.08; H, 5.01; N, 5.85.

Preparation of CSP 7. To a suspension of poly((*N*-methyl)aminoethyl methacrylate-*co*-methyl methacrylate-*co*-ethylene dimethacrylate) beads[11] **5** (18 g) in methylene chloride (250 mL) were added (*S*)-(3,5-dinitrobenzoyl)-leucine[8] **6** (13 g, 40 mmol) and 2-ethoxy-1-ethoxycarbonyl-1,2-dihydroquinoline (EEDQ) (9.88 g, 40 mmol). After the mixture was stirred for 24 h at room temperature, the beads **7** were filtered and repeatedly washed with tetrahydrofuran (THF) and methylene chloride and then dried under vacuum. The selector content in the beads is 0.33 mmol/g based on elemental analysis of nitrogen.

Preparation of CSP 10. To a suspension of amino functionalized beads[11] **5** (1.50 g) in THF (15 mL) was added a solution of ethyl 1-methyl-6-bromomethyl-2-oxo-4-phenanthrene-1,2,3,4-tetrahydropyrimidine-5-carboxylate (−)-**9** (0.40 g, 8.8 mmol) in THF (10 mL). After the mixture was stirred at room temperature under nitrogen overnight, the beads **10** were filtered and repeatedly washed with THF, water, and methanol and then dried under vacuum. The selector content in the beads is 0.20 mmol/g based on elemental analysis of nitrogen.

Chromatography. The chiral stationary phases **7** and **10** were slurry packed at a constant pressure of 15.0 MPa into 150 × 4.6 mm i.d. stainless steel columns. Preparative separations were conducted using a 60 × 0.7 cm i.d. column packed with CSP **7**. A Waters HPLC system consisting of two 510 HPLC pumps, a 717 plus autosampler, a 486 UV detector, and a Jasco OR-990 chiral detector, controlled by Millennium 2010 software, was used for all of the chromatography.

Normal phase and reversed phase chiral separations were carried out using CH_2Cl_2 and acetonitrile/water, respectively, as the mobile phases. The separation factors α (selectivity) were calculated using the following equation

$$\alpha = k'_2/k'_1 \tag{1}$$

where k'_1 and k'_2 are the retention factors of the enantiomers defined as

$$k'_i = (t_R - t_0)/t_0 \tag{2}$$

where t_R and t_0 represent the retention times of the compound and 1,3,5-tri-*tert*-butylbenzene (void volume marker), respectively. The racemic analytes *N*-(3,5-dinitrobenzoyl)-α-amino acid alkyl amides and 3,5-dinitroanilides were prepared by methods similar to those reported elsewhere.[36] ^{1}H NMR and IR spectra are in agreement with the assigned structures shown in Table 3.

Acknowledgment. Support of this research by a grant of the National Institute of General Medical Sciences,

National Institutes of Health (GM-44885), is gratefully acknowledged. We thank Dr. Frederick Hollander (University of California at Berkeley) for the X-ray measurement. P.M. thanks the Swiss National Science Foundation for a post-doctoral fellowship.

Supporting Information Available. Crystallographic data for compound (±)-**8**, including positional and thermal parameters, and ^{1}H NMR spectra and melting points of compounds given in Tables 1 and 2 (15 pages). See any current masthead page for ordering and Internet access instructions.

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