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Microwave-assisted Synthesis of Novel Chiral Receptors Derived from Deoxycholic Acid and Their Molecular Recognition Properties

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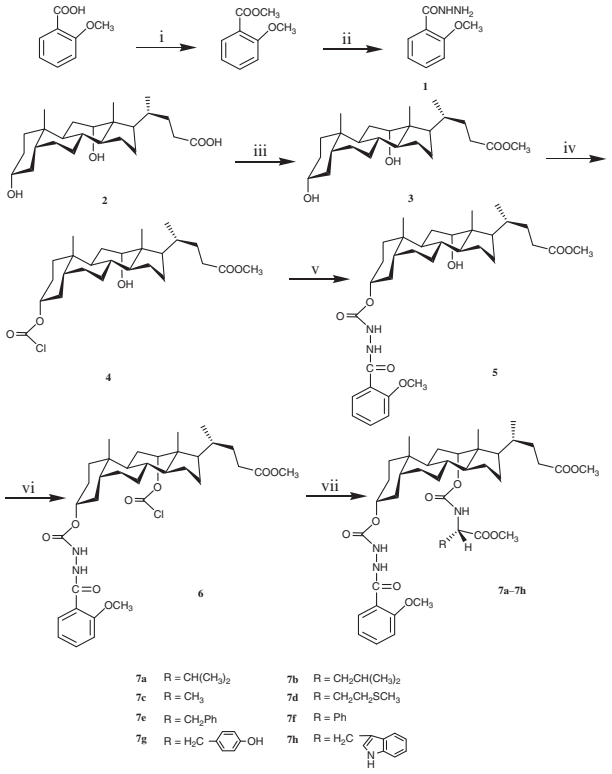
Under microwave irradiation (MWI), novel chiral receptors derived from deoxycholic acid were synthesized by using deoxycholic acid methyl ester as the spacer, and arylhydrazine and amino acids as the arm. Selective recognition properties of these receptors for aromatic amines and D/L-amino acids have been investigated by UV–vis spectral titration and ^{1}H NMR spectral study. The results indicate this type of receptors can form a 1:1 supramolecular complex with an aromatic amine and a 1:2 supramolecular complex with D/L-tryptophan.

Using artificial receptors to build bionic enzyme models has exhibited potential applications in the fields of biomimetic catalysis, drug synthesis, chiral separation, molecular devices, biosensors, and so on;[1–4] synthesis and molecular recognition properties of different types of artificial receptors have also become the focus of modern bioorganic chemistry research.[5–8] Bile acids, because of their rigid structures and asymmetric features, have been widely used as ideal building blocks for the construction of molecular tweezers.[9,10] Since it is well known that biological activity and chemical properties of organic species are highly dependent on stereochemistry, great efforts have been made to design and synthesize chiral receptors for enantioselective recognition of chiral organic species.[11–13]

Moreover, with the development of "green chemistry" in recent years, microwave-assisted techniques have become increasingly important in the field of organic synthesis not only because of the advantage of minimal use of organic solvents, but also because of reduced reaction times, fewer by-products, and a simplified work under microwave irradiation (MWI).[14,15] In this paper, we report an efficient microwave-assisted method to synthesize novel chiral unsymmetrical arylhydrazide-type molecular tweezers based on deoxycholic acid. In this type of receptor structure, three NH groups can be found, which can form hydrogen bonds with substrates in the molecular recognition process. The binding ability of this kind of molecular tweezers for recognition of substrates, which also possess the NH group, such as aromatic amines and D/L-amino acids, has been investigated; some target molecular tweezers showed interesting enantioselective recognition of D/L-amino acids, which have never been reported.

Molecular tweezers **7a**–**7h** were synthesized through seven steps (Scheme 1). Intermediate **1** was prepared following a reported procedure;[16] then, deoxycholic acid (**2**) was methylated to methyl ester **3** (yield 87%) by using methyl iodide, followed by treatment with triphosgene to give compound **4**. After microwave irradiating in K_2CO_3 with intermediate **1** (yield 85%) and anhydrous pyridine, compound **5** (yield 91%) was synthesized, which subsequently reacted with triphosgene to give compound **6**. We found that the yield of compound **5** was highest when irradiating microwave at 450 W for 10 min. Then, the L-amino acid methyl ester hydrochloride and anhydrous pyridine were added to compound **6**, and the mixture was irradiated continually for 8–10 min at 300 W. TLC was used to monitor the reaction progress until it was complete. Target molecular tweezers **7a**–**7h** (yield 85%–90%) were obtained and were purified furthermore by column chromatography on silica gel H with dichloromethane/acetone as the eluent.



Scheme 1. The synthetic route of molecular tweezers **7a**–**7h**. Reagents and conditions: (i) CH_3OH, H_2SO_4; (ii) $N_2H_4 \cdot H_2O$; (iii) CH_3I, K_2CO_3, THF, MWI; (iv) triphosgene, CH_2Cl_2, pyridine, MWI; (v) pyridine, K_2CO_3, intermediate **1**, MWI; (vi) triphosgene, CH_2Cl_2, pyridine, MWI; (vii) L-amino acid methyl esters hydrochloride, pyridine, MWI.

A series of experiments were carried out to make a better comparison between MWI and conventional heating in the synthesis of intermediate **5** and compounds **7a**–**7h**. The results are shown in Table 1.

The supports remarkably affect the yields of the products. It was found that when using silica gel H and neutral Al_2O_3 as the support and microwave irradiating for 10 min, only a small amount of compound **5** was obtained, while using K_2CO_3 as the

Table 1. Synthetic comparison of intermediate **5** and compounds **7a**–**7h** between microwave irradiation and conventional heating

Compd.	Conventional method		Microwave method		t_c/t_{mw}[a]
	t/min	Yield/%	t/min	Yield/%	
5	2880	75	10	91	288
7a	2100	68	20	89	105
7b	2100	52	18	86	117
7c	2400	65	20	87	120
7d	2100	58	20	86	105
7e	2280	70	19	90	120
7f	2340	68	18	89	130
7g	2400	63	18	87	133
7h	2400	42	19	85	126

[a]t_c, conventional method time; t_{mw}, microwave method time.

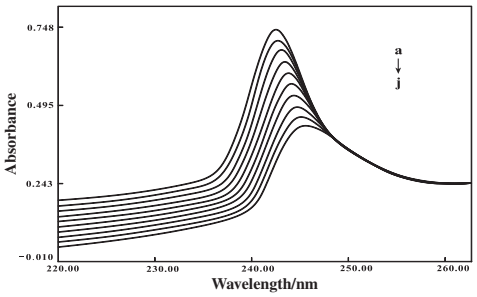


Figure 1. UV–vis spectral changes of compound **7e** (1×10^{-4} mol L^{-1}) in the presence of *p*-methoxyaniline: (a) 0, (b) 0.4×10^{-4}, (c) 0.8×10^{-4}, (d) 1.2×10^{-4}, (e) 1.6×10^{-4}, (f) 2.0×10^{-4}, (g) 2.4×10^{-4}, (h) 2.8×10^{-4}, (i) 3.2×10^{-4}, and (j) 3.6×10^{-4} mol L^{-1} with λ_{max} at 242 nm.

support and under the same microwave condition, the yield of intermediate **5** reached 91%.

UV–vis spectral titration was used to investigate the recognition abilities of compound **7a**, **7e**, **7g**, and **7h** for aniline, *p*-methoxyaniline, *m*-methoxyaniline, *o*-methoxyaniline, D/L-tryptophan, D/L-histidine, and D/L-tyrosine by monitoring the absorbance changes. When *p*-methoxyaniline (0–3.6×10^{-4} mol L^{-1}) was added to the solution of compound **7e** (1×10^{-4} mol L^{-1}) in $CHCl_3$, the absorbance band at 242 nm decreased regularly. The UV–vis plot of **7e** for *p*-methoxyaniline is shown in Figure 1. The titration data were analyzed by using the Hildebrand–Benesi equation. The plots of $1/[G]_0$ versus $1/\Delta A$ gave a straight line (Figure 2), which showed that compound **7e** could form a complex with *p*-methoxyaniline and the supramolecular complex consisted of 1:1 host and guest molecules. The association constants (K_a) were calculated according to the intercept and the slope of the line. Job's plots also indicated the 1:1 complex formation when aniline, *p*-methoxyaniline, *m*-methoxyaniline, and *o*-methoxyaniline were added to the solution of **7a**, **7e**, **7g**, and **7h**, respectively. Association constants (K_a) and free energy change ($-\Delta G^0$) for the inclusion complexes of **7a**, **7e**, **7g**, and **7h** with aniline, *p*-methoxyaniline, *m*-methoxyaniline, and *o*-methoxyaniline are listed in Table 2. Interestingly, when different volumes of the guest D/L-tryptophan solution was added into defined amounts of hosts **7a**, **7e**, **7g**, and **7h**, though the UV absorbance of the hosts decreased, the absorption peak shape became narrow

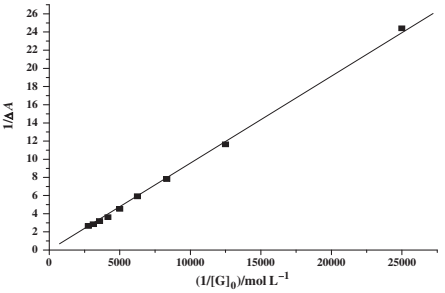


Figure 2. Typical plot of $1/\Delta A$ versus $1/[G]_0$ for the inclusion complex of molecular tweezer **7e** with *p*-methoxyaniline in $CHCl_3$ at 25 °C.

Table 2. Association constants (K_a) and Gibbs free energy changes (ΔG^0) for the inclusion complexes of *p*-methoxyaniline with molecular tweezers **7a**, **7e**, **7g**, and **7h** in $CHCl_3$ at 25 °C

Host	Guest	K_a/L mol^{-1}	$-\Delta G_0$/kJ mol^{-1}
7a	aniline	874.29	16.78
	o-methoxyaniline	39.29	9.10
	m-methoxyaniline	620.73	15.93
	p-methoxyaniline	714.48	16.28
7e	aniline	2242.54	19.12
	o-methoxyaniline	592.87	15.82
	m-methoxyaniline	687.20	16.19
	p-methoxyaniline	332.20	14.38
7g	aniline	366.82	14.63
	o-methoxyaniline	813.30	16.67
	m-methoxyaniline	834.91	16.66
	p-methoxyaniline	279.29	13.95
7h	aniline	1697.09	18.42
	o-methoxyaniline	661.54	16.09
	m-methoxyaniline	389.61	14.78
	p-methoxyaniline	488.55	15.34

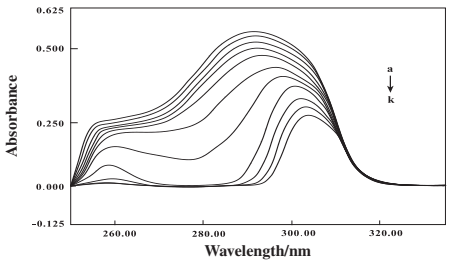


Figure 3. UV–vis spectral changes of compound **7e**: (a) 1.5×10^{-3}, (b) 1.0×10^{-3}, (c) 0.8×10^{-3}, (d) 0.6×10^{-3}, (e) 0.5×10^{-3}, (f) 0.4×10^{-3}, (g) 0.3×10^{-3}, (h) 0.2×10^{-3}, (i) 0.15×10^{-3}, (j) 0.12×10^{-3}, and (k) 0.1×10^{-3} mol L^{-1} in the presence of L-tryptophan (1×10^{-3} mol L^{-1}) with λ_{max} at 288–304 nm.

(Figure 3) and the plots of $1/[G]_0$ versus $1/\Delta A$ gave a curve. Both Job's plots and the plots of molar ratio versus absorbance indicated the formation of a 1:2 supramolecular complex (Figure 4); the association constants (K_a) were obtained from the nonlinear curve fitting of A versus C_R/C_M. The association constants (K_a) and K_D/K_L for the inclusion complexes of **7a**, **7e**, **7g**, and **7h** with D/L-tryptophan are displayed in Table 3.

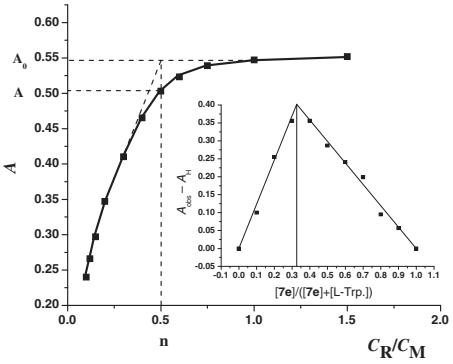


Figure 4. Job's plot and typical plot of A versus C_R/C_M for the inclusion complex of molecular tweezer **7e** with L-tryptophan in DMSO at 25 °C, C_R: the concentration of molecular tweezer **7e**; C_M: the concentration of L-tryptophan.

Table 3. Association constants (K_a) for the inclusion complexes of D/L-tryptophan with molecular tweezers **7a**, **7e**, **7g**, and **7h** in DMSO at 25 °C

Host	Guest	K_a/L mol^{-1}	K_D/K_L
7a	D-tryptophan	48849.34	4.41
	L-tryptophan	11074.15	
7e	D-tryptophan	15451.57	2.83
	L-tryptophan	5466.05	
7g	D-tryptophan	23549.89	1.72
	L-tryptophan	13656.81	
7h	D-tryptophan	91768.81	2.90
	L-tryptophan	31665.74	

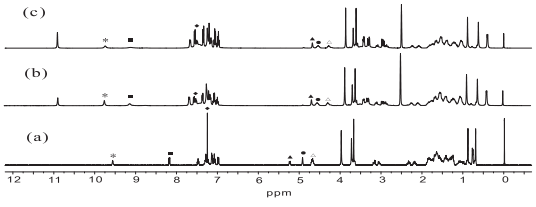


Figure 5. Portion of ^{1}H NMR spectra of compound **7e** in DMSO-d_6 in the presence of increasing amount of L-tryptophane: (a) +0 equiv; (b) +1 equiv; (c) +2 equiv (*, CH_3O–PhCONH; ■, 3α-OCONH; ◆, ArH; ▲, 12α-OCONH; ●, 12β-H; △, 3β-H + NCH).

As shown in Table 3, these receptors showed good chiral recognition for D-tryptophan; the maximum K_D/K_L of compound **7a** was 4.41 for D/L-tryptophan. However, when different amounts of guests D/L-histidine and D/L-tyrosine were added into defined amounts of hosts **7a**, **7e**, **7g**, and **7h**, the UV absorbance showed almost no change. The result clearly indicated that receptors **7a**, **7e**, **7g**, and **7h** selectively bind D/L-tryptophan.

In order to ascertain the binding site of the receptor upon interaction with the guest molecule, the mode of binding of L-tryptophan with compound **7a**, **7e**, **7g**, and **7h** was investigated by ^{1}H NMR spectroscopy (see the Supporting Information). For example, as shown in Figure 5a, free compound **7e** displayed two sharp signals in the downfield region at 9.56 and 8.17 ppm, which were assigned to the amide NH attached to the phenyl ring and the amide NH attached to the 3α-moiety of deoxycholic acid methyl ester. Moreover, other three sharp signals were found at 5.23, 4.91, and 4.67 ppm, which belong to the amide NH attached to the 12α-moiety of deoxycholic acid methyl ester, CH in the 12β-moiety, and CH in the 3β-moiety, respectively. Upon addition of 1.0 equiv of L-tryptophan, the amide NH attached to the phenyl ring and the amide NH attached to the 3α-moiety of deoxycholic acid methyl ester were downfield shifted to 0.18 and 0.95 ppm, while the amide NH attached to the 12α-moiety of deoxycholic acid methyl ester, CH in the 12β-moiety, and CH in the 3β-moiety were upfield shifted to 0.55, 0.37, and 0.39 ppm, respectively. What's more, all the protons of the phenyl rings were downfield shifted in varying degrees (Figure 5b), which showed that the π–π stacking interactions also contributed to the recognition process. When the added L-tryptophan was increased up to 2.0 equiv, the signal of the amide NH attached to the 3α-moiety of deoxycholic acid methyl ester significantly broadened and weakened (Figure 5c). The change of chemical shift indicated that both the amide NH and phenyl rings of compound **7e** should bind with L-tryptophan.

In conclusion, novel chiral unsymmetrical arylhydrazide-type molecular tweezers based on deoxycholic acid were developed and the receptors showed remarkable selectivity and affinity for D/L-tryptophan and aromatic amines. It might lead to potential applications in analysis and separation of chiral amino acids and aromatic amines, or in transport of amino acid drugs and aromatic amine drugs.

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Supporting Information is available electronically on J-STAGE.

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