Inclusion Compounds

One-Pot Optical Resolution of Oligopeptide Helices through Artificial Peptide Bundling

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Molecular recognition through helix–helix interactions is an interesting subject in relation to biologically important peptide bundling.[1] Although the design of peptide bundling

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(coiled-coil) based on natural peptide motifs has been studied extensively,[2–4] secondary structure recognition between helical macromolecules has never been utilized in artificial host–guest chemistry for the one-pot separation of right- and left-handed helices in solution.[5,6] Herein, we report the first example of one-pot optical resolution of helical oligopeptides (*rac*-**2** and *rac*-**3**) through stereoselective helix bundling by using a cyclodimeric zinc porphyrin host **1** bearing a guest-binding chiral cavity with two helical peptidic units (Figure 1).

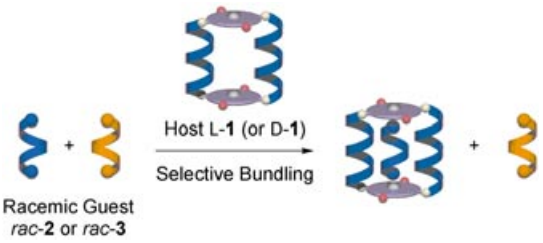
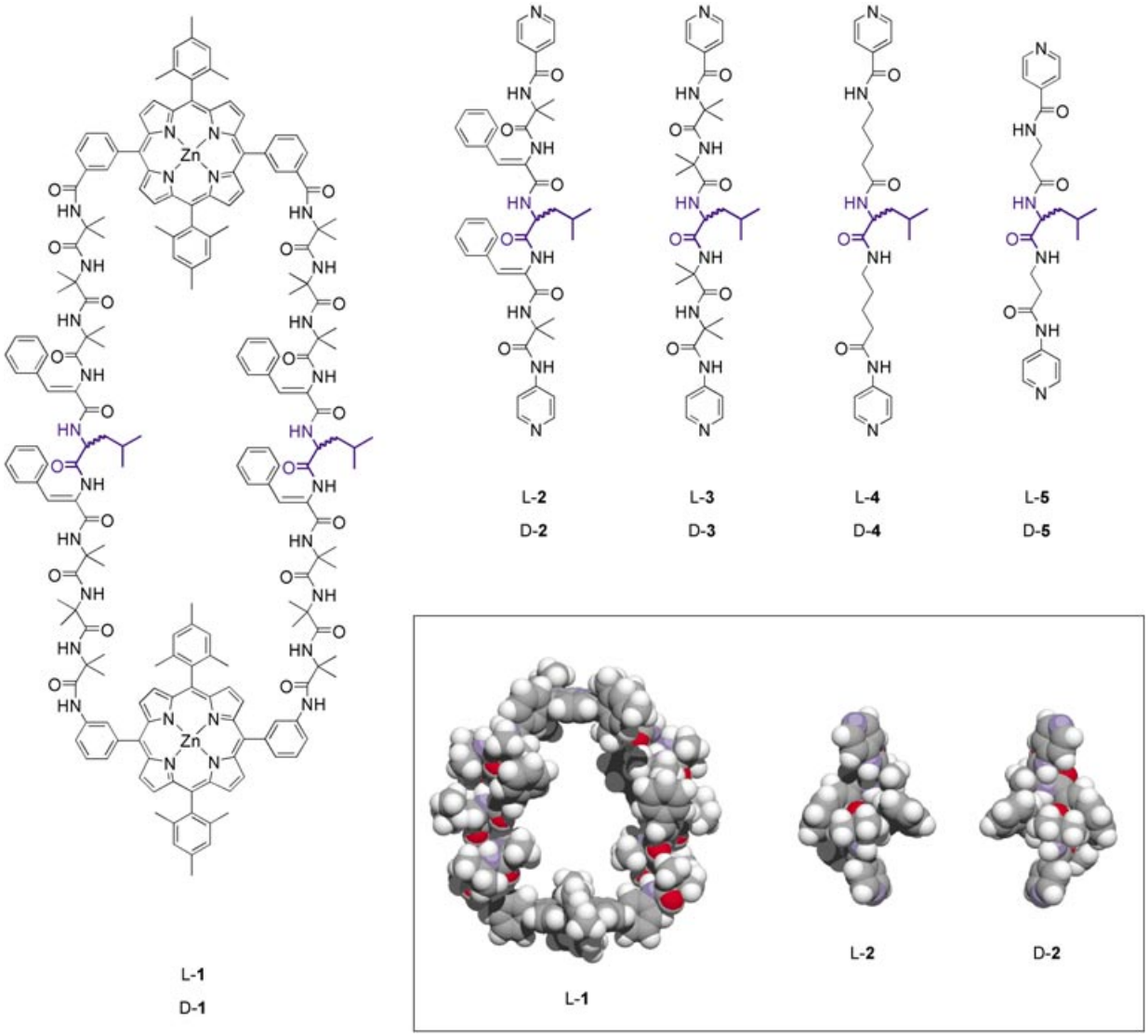


Figure 1. Schematic representation of helix-sense-selective host–guest complexation.

The oligopeptide parts in compounds **1**–**3** contain α-aminoisobutyric acid (Aib) units (Scheme 1).[7] Poly(Aib) is known to adopt a dynamic helical structure, which switches back and forth between right- and left-handed conformations.[8] Recently, we have found that a cyclodimeric zinc porphyrin with dynamic oligo(Aib) units can detect the helical sense of oligopeptides.[9] Although this chiroptical sensor lacks a stereogenic center, it becomes optically active through stereochemical interactions with helical guests upon inclusion in its cavity. In contrast, host **1** and guests **2** and **3** (Scheme 1) possess leucine residues (Leu) as a chiral source, which can induce, depending on their configurations, either a right- or left-handed helicity in the oligopeptide chains. Therefore, the stabilities of the host–guest complexes may be affected by the conformational matching among the three single-handed oligopeptide helices. Compounds **1** and **2** also contain two dehydrophenylalanine (ΔPhe)[10] units adjacent to the Leu residue whose benzylidene groups would have a steric influence on the host–guest interaction.



Scheme 1. Structures of helical oligopeptide host **1** and guests **2**–**5**, as well as molecular models of L-**1**, L-**2**, and D-**2**.

Cyclic host **1** was obtained by metalation with $Zn(OAc)_2$ of a precursor free-base porphyrin cyclic dimer, synthesized by coupling of 5,15-bis(3-aminophenyl)-10,20-dimesitylporphyrin with a nonapeptide containing Leu and ΔPhe residues, followed by macrocyclization with 5,15-bis(3-carboxyphenyl)-10,20-dimesitylporphyrin.[7] The CD spectrum (Figure 2a, black curve) of the L-leucine-containing host (L-**1**) in $CHCl_3$ displays an exciton-coupled CD band centered at 280 nm, as a result of the ΔPhe units. The oligopeptide units in L-**1** adopt a right-handed helical conformation, as evident by the sign of the split Cotton effect.[10] A characteristic exciton-couplet is also observed at the Soret absorption band of the zinc porphyrin moieties (400–440 nm), which indicates a clockwise-twisted geometry of the two facing zinc porphyrin units.[9,11] As expected, the CD spectrum of D-**1** bearing left-handed helical units (Figure 2b, black curve) is a perfect mirror-image of that of L-**1**.

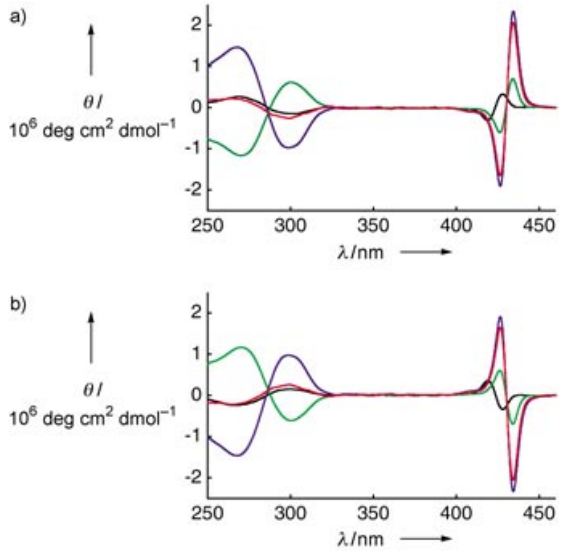


Figure 2. Complexation of **1** with **2** at [**2**]/[**1**] = 10:1 in $CHCl_3$ at 25 °C. a) CD spectra of L-**1** in the absence (black curve) and presence of L-**2** (blue curve), D-**2** (green curve), and *rac*-**2** (red curve). b) CD spectra of D-**1** in the absence (black curve) and presence of D-**2** (blue curve), L-**2** (green curve), and *rac*-**2** (red curve).

Spectroscopic titration in $CHCl_3$ and Job plots[7] suggest that L-**1** forms a stable 1:1 inclusion complex with L-**2**, a pyridine-anchored right-handed helical pentapeptide containing L-Leu, with an association constant K_{assoc}(L⊃L) of $14.2\times10^6\,M^{-1}$ (Table 1, entry 1). In contrast, the association constant for the complexation of left-handed helical guest D-**2** with L-**1**, is much smaller (K_{assoc}(L⊃D) = $2.9\times10^6\,M^{-1}$, Table 1, entry 1). The large difference between these association constants (K_{assoc}(L⊃L)/K_{assoc}(L⊃D) = 4.9:1) demonstrates that right-handed L-**2** is favored over left-handed D-**2** in the complexation with L-**1**. Accordingly, host D-**1** (which has left-handed helical units) shows a much larger affinity toward left-handed D-**2** than right-handed L-**2** (Table 1, entry 5), and the observed K_{assoc}(D⊃D)/K_{assoc}(D⊃L) ratio of 4.9:1 is identical to that observed for L-**1**⊃**2** (Table 1, entry 1).

Inclusion complexes L-**1**⊃L-**2** and L-**1**⊃D-**2** also display different CD spectral profiles (Figure 2a). Mixing L-**1** with the favorable guest L-**2** (10 equiv) results in the exciton-couplet at the Soret absorption band of the zinc porphyrin moieties being red-shifted from 424 to 431 nm (because of the coodination with the pyridyl terminus of L-**2**[9]) and significantly enhanced (Figure 2a, black→blue curves), whereas mixing of L-**1** with the unfavorable left-handed D-**2** (10 equiv) results in a red-shifted but much less enhanced CD band (Figure 2a, black→green curves). When D-**1** is used as the host, an analogous CD enhancement is observed upon complexation with D-**2** (Figure 2b, blue curve) rather than with L-**2** (green curve). These observations indicate that the twisted geometry of the zinc porphyrin units in the host is likely stabilized by the inclusion of a favorable guest in its cavity to form a peptide bundle. Interestingly, mixing of L-**1** or D-**1** with racemic guest *rac*-**2** (10 equiv) also results in a large enhancement of the CD signal (Figure 2, black→red curves), the extent of which is 87% of those observed for the favorable host–guest combinations (Figure 2, blue curves). This result suggests the occurrence of optical resolution of *rac*-**2** in solution by selective binding with **1** under competitive conditions (Figure 1). To prove this[7] inclusion complex L-**1**⊃**2**, formed upon mixing L-**1** with *rac*-**2** (10 equiv) in $CHCl_3$, was isolated by size-exclusion chromatography (SEC) with $CHCl_3$ as eluent. Decomplexation of L-**1**⊃**2** in THF[12] followed by SEC with THF as eluent allowed isolation of guest **2** in 78% yield based on L-**1**. HPLC analysis on a chiral stationary phase (Daicel ChiralPak AD-H, Figure 3b) with *rac*-**2** as reference (Figure 3a) demonstrated that the **2** thus isolated was considerably enriched in the L isomer, with a L-**2**/D-**2** ratio of 86:14 (72% *ee*; Table 1, entry 1). However, the use of D-**1** in place of L-**1** for the competitive complexation with *rac*-**2**, followed by an analogous chromatographic analysis (Figure 3c), resulted in a L-**2**/D-**2** ratio of 10:90 (80% *ee*; Table 1, entry 5).

To explore the origin of stereochemical guest selection we investigated the complexation of L-**1** with leucine-containing

Table 1: Association constants (K_{assoc}) for the complexation of helical host **1** with guests **2**–**5** in $CHCl_3$ at 25 °C, upon spectroscopic titration, and enantiomer ratios of **2** extracted from **1**⊃**2**, upon decomplexation.

Entry	Host	Guest	K_{assoc}(L⊃L) [M^{-1}]	K_{assoc}(L⊃D) [M^{-1}]	K_{large}/K_{small}	L-**2**/D-**2** extracted from **1**⊃**2**[a]
1	L-**1**	**2**	14.2×10^6	2.9×10^6	4.9	86:14
2	L-**1**	**3**	4.0×10^6	1.2×10^6	3.3	–
3	L-**1**	**4**	2.1×10^5	2.7×10^5	1.3	–
4	D-**1**	**5**	3.2×10^5	3.6×10^5	1.1	–
5	D-**1**	**2**	2.7×10^6 [= K_{assoc}(D⊃L)]	13.3×10^6 [= K_{assoc}(D⊃D)]	4.9	10:90

[a] Determined by HPLC on a Daicel Chiralpak AD-H column.

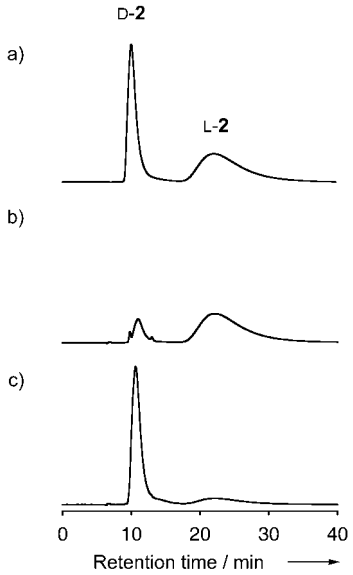


Figure 3. HPLC traces obtained on a chiral stationary phase of a) *rac*-**2** (reference) and b), c) **2** extracted from inclusion complexes with L-**1** and D-**1**, respectively (column: Daicel Chiralpak AD-H, eluent: hexane/2-propanol/Et_2NH = 75:25:0.1).

guests **4** and shorter-chain **5** as nonhelical reference compounds (Scheme 1). Although the number of atoms along the main chain of **4** is identical to that of **2**, the molecular length of **5** is more likely to be similar to that of **2** adopting a helical conformation. Spectroscopic titration experiments showed that the association constants of these nonhelical guests with L-**1** are one order of magnitude smaller than those observed for helical **2**, and more importantly, their enantiomers were not discriminated stereochemically (Table 1, entries 3 and 4).[7] From these observations we can conclude that a helix–helix (host–guest) interaction is responsible for the stereochemical guest selection in the complexation between **1** and **2**. Moreover, substituents on the helical chains play a role in the helix–helix interaction upon bundling.[2–4,13] For example, smaller association constants are found for the complexation of L-**1** with the enantiomers of helical guest **3**, which does not contain benzylidene units, than those with **2** (Table 1, entry 2). In particular, the L enantiomer of **3** is preferentially selected by L-**1**, but the observed $K_{assoc}(L\supset L)/K_{assoc}(L\supset D)$ ratio of 3.3:1 is clearly smaller than that of L-**1**⊃**2**.

In conclusion, we have demonstrated one-pot optical resolution of helical peptidic guests in solution by using a cyclodimeric zinc porphyrin host bearing single-handed oligopeptide units. In conjunction with control experiments on nonhelical chiral guests, the results clearly show that the host and guest molecules stereochemically recognize their helical structures, rather than the point chiralities (Leu) in their chains, upon bundling of the host molecule in the confined cavity. We believe that exploration of asymmetric transformations through artificial peptide bundling is worthy of further investigation.

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