

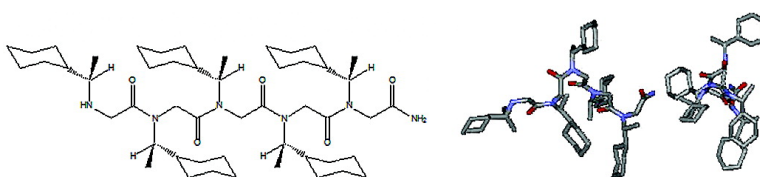
Article

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*J. Am. Chem. Soc.*, **2003**, 125 (44), 13525-13530 • DOI: 10.1021/ja037540r • Publication Date (Web): 11 October 2003

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## Structural and Spectroscopic Studies of Peptoid Oligomers with $\alpha$ -Chiral Aliphatic Side Chains

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**Abstract:** Substantial progress has been made in the synthesis and characterization of various oligomeric molecules capable of autonomous folding to well-defined, repetitive secondary structures. It is now possible to investigate sequence–structure relationships and the driving forces for folding in these systems. Here, we present detailed analysis by X-ray crystallography, NMR, and circular dichroism (CD) of the helical structures formed by N-substituted glycine (or “peptoid”) oligomers with  $\alpha$ -chiral, aliphatic side chains. The X-ray crystal structure of a *N*-(1-cyclohexylethyl)glycine pentamer, the first reported for any peptoid, shows a helix with *cis*-amide bonds,  $\sim 3$  residues per turn, and a pitch of  $\sim 6.7$  Å. The backbone dihedral angles of this pentamer are similar to those of a polyproline type I peptide helix, in agreement with prior modeling predictions. This crystal structure likely represents the major solution conformers, since the CD spectra of analogous peptoid hexamers, dodecamers, and pentadecamers, composed entirely of either (*S*)-*N*-(1-cyclohexylethyl)glycine or (*S*)-*N*-(*sec*-butyl)glycine monomers, also have features similar to those of the polyproline type I helix. Furthermore, this crystal structure is similar to a solution NMR structure previously described for a peptoid pentamer comprised of chiral, aromatic side chains, which suggests that peptoids containing either aromatic or aliphatic  $\alpha$ -chiral side chains adopt fundamentally similar helical structures in solution, despite distinct CD spectra. The elucidation of detailed structural information for peptoid helices with  $\alpha$ -chiral aliphatic side chains will facilitate the mimicry of biomolecules, such as transmembrane protein domains, in a distinctly stable form.

### Introduction

Spectroscopic and computational studies of  $\alpha$ -peptide oligomers have shown that the spatial arrangement and chemistry of the individual amino acid side chains play a significant role in directing the formation of various classes of helical and sheetlike secondary structures. For example,  $\alpha$ - and  $3_{10}$ -helices are two common secondary structures found in proteins. Such helices, which have *trans*-amide bonds, are stabilized by both intrachain hydrogen bonding and by steric constraints imposed on the backbone by the side chains.<sup>1–3</sup> For instance, experimental and theoretical investigations have shown that alanine,<sup>4–6</sup> leucine,<sup>7</sup> and glutamic acid<sup>8</sup> residues are strong  $\alpha$ -helix formers, while

$\alpha$ -aminoisobutyric (Aib)-based peptides can form either  $\alpha$ - or  $3_{10}$ -helices, or a combination thereof, depending upon chain length and sequence.<sup>9,10</sup> Thus, the chemical identity and, particularly, the steric bulk and flexibility of side chains directly influences the tendency of the peptide backbone to adopt a particular repeating conformation.<sup>11</sup> Proline-rich sequences, with their uniquely constrained amino acids, adopt completely different types of helices that may have either *cis*- or *trans*-amide bonds, termed polyproline type I or type II, respectively, depending on solvent environment and the time since dissolution.<sup>12</sup> An understanding of how a certain amino acid monomer predisposes the formation of a particular secondary structure in

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$\alpha$ -peptides has enabled the de novo design of bioactive molecules.<sup>3,13–16</sup>

Researchers have also studied the propensity of synthetic, nonnatural oligomers of various classes to fold into biomimetic secondary structures.<sup>17–20</sup> Many of these foldamer classes offer increased in vivo stability and potential therapeutic advantages in comparison to  $\alpha$ -peptides.<sup>20</sup> A deeper understanding of sequence–structure relationships in these oligomer systems promises to enable the design of structured, bioactive species. Examples of nonnatural foldameric oligomers include oligo-N-substituted glycines (peptoids),<sup>21,22</sup> oligoureas,<sup>23,24</sup> oligopyrrolinones,<sup>25</sup>  $\beta$ -peptides,<sup>18,26–35</sup>  $\gamma$ -peptides,<sup>36,37</sup> and oligo(phenylene ethynyls).<sup>38</sup>

Sequence–structure relationships in oligo-N-substituted glycines, in particular, are the focus of this study. Peptoids have been shown to be protease resistant,<sup>39</sup> and biocompatible<sup>40</sup> and are easy to produce in long-chain form with relatively complex sequences.<sup>41</sup> Additionally, peptoids have been developed for use in many biological applications.<sup>17,20,21,40</sup> Superficially, peptoids have a primary structure that is similar to that of  $\alpha$ -peptides. Whereas the side chains of  $\alpha$ -peptides are pendant groups of

the main-chain  $\alpha$ -carbon, the side chains of peptoids are instead linked to the polymer backbone through the amide nitrogen. Because of this structural difference, peptoids lack amide protons, which obviates the formation of the intrachain hydrogen bonds that partially stabilize folded structure in  $\alpha$ -peptides. And unlike natural peptides, the peptoid backbone is inherently achiral. However, it has been shown that sufficient steric direction to form stable peptoid helices of a particular handedness can be provided instead by  $\alpha$ -chiral side chains.<sup>42–45</sup> The helical structures adopted by peptoids with  $\alpha$ -chiral, aromatic side chains, in particular (*S*)- and (*R*)-*N*-(1-phenylethyl) side chains, have been studied in detail.<sup>42–47</sup> Still, folded structure in entirely aliphatic peptoids has not been investigated previously. Since helical peptoids with  $\alpha$ -chiral, aliphatic side chains are closer mimics of typical trans-membrane or membrane-disruptive polypeptides (e.g., sequences rich in leucine, isoleucine, valine, etc.), they are arguably of greater biological relevance than sequences with predominantly aromatic side chains.

In this work, we have sought to better understand the preferred secondary structure of peptoids containing entirely  $\alpha$ -chiral, aliphatic side chains. We present the first X-ray crystal structure of a (*R*)-*N*-(1-cyclohexylethyl)glycine pentamer. We have also characterized the solution structures of related, longer homooligomers of (*S*)-*N*-(1-cyclohexylethyl)glycine and (*S*)-*N*-(1-sec-butyl)glycine by circular dichroism (CD) and 2D-NMR. These results, taken together with the crystal structure, provide useful insight into the manner in which aliphatic side-chain chemistry and chain length affect peptoid secondary structure.

## Materials and Methods

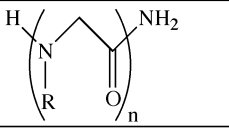
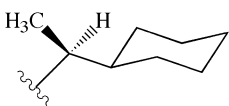
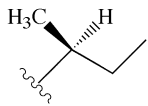
**Peptoid Synthesis, Purification, and Characterization.** Peptoid oligomers were synthesized on solid phase (Rink amide resin, Novabiochem, San Diego, CA) by a previously described submonomer methodology<sup>22</sup> and were subsequently analyzed, purified, and reanalyzed by HPLC, also as previously described.<sup>22,43,46</sup> The (*R*)-*N*-(1-cyclohexylethyl)glycine (*Nrch*), (*S*)-*N*-(1-cyclohexylethyl)glycine (*Nsch*), and (*S*)-*N*-(2-butyl)glycine (*Nssb*) monomers (Table 1) were derived from the amines (*R*)-1-cyclohexylethylamine, (*S*)-1-cyclohexylethylamine, and (*S*)-2-butylamine, respectively. All amines were obtained from Aldrich Chemical Co. (Milwaukee, WI) at >99% purity and used without additional purification. Electrospray mass spectrometry was performed at Northwestern University's Analytical Chemistry Facility (Micromass Quattro II mass spectrometer). All CD spectra were acquired in acetonitrile solution, with sample preparation and data acquisition methods as described elsewhere.<sup>46</sup>

**NMR Data Acquisition.** Peptoid samples were dissolved in acetonitrile-*d*<sub>3</sub> at concentrations of 1–2 mg/mL. NMR spectra were acquired on a Varian Inova 600 at 25 °C. Natural abundance 2D heteronuclear single quantum coherence spectroscopy (HSQC) experiments were collected with the following parameters: The spectral width was 6000 Hz in the <sup>1</sup>H dimension and 12 067 Hz in the <sup>13</sup>C dimension. The

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**Table 1.** N-Substituted Glycine Side Chains

		<i>N</i> -substituted glycine oligomer, or <i>peptoid</i>
R = Side chain	Designator	
	<i>Nsch</i> = ( <i>S</i> )- <i>N</i> -(1-cyclohexylethyl)glycine	
	<i>Nssb</i> = ( <i>S</i> )- <i>N</i> -(2-butyl)glycine	

**Table 2.** Peptoid Homooligomer Structures, Mass Confirmation, and Crude Yield<sup>a</sup>

peptoid oligomer	monomer sequence (amino-to-carboxy)	molar mass calcd:found	crude yield (%) <sup>b</sup>
1	( <i>Nrch</i> ) <sub>5</sub>	823.0:823.2	~90
2	( <i>Nsch</i> ) <sub>5</sub>	823.0:823.2	~90
3a	( <i>Nsch</i> ) <sub>6</sub>	1020.5:1020.9	~89
3b	( <i>Nsch</i> ) <sub>12</sub>	2024.1:2024.4	~58
3c	( <i>Nsch</i> ) <sub>15</sub>	2525.9:2524.1	~40
4a	( <i>Nssb</i> ) <sub>6</sub>	696.0:696.3	~72
4b	( <i>Nssb</i> ) <sub>12</sub>	1374.9:1374.5	~40
4c	( <i>Nssb</i> ) <sub>15</sub>	1714.3:1715.4	~70 <sup>c</sup>

<sup>a</sup> All compounds were purified to >97% homogeneity before analysis by CD. <sup>b</sup> As estimated by analytical reversed-phase HPLC of crude product. <sup>c</sup> Amine coupling times were doubled for this peptoid.

numbers of t1 increments, transients, and t2 complex data points were 512, 32, and 512, respectively. Data were processed with FELIX97 (Molecular Simulations). HSQC spectra were processed as phase-sensitive in both dimensions and were zero-filled to give a final data matrix of 1024 × 1024 points.

**X-ray Crystallography.** Data were collected on an (*Nrch*)<sub>5</sub> crystal with dimensions of 0.18 × 0.20 × 0.43 mm<sup>3</sup> and measured with a SMART CCD area detector with graphite monochromated Mo K $\alpha$  radiation. Data reduction was performed using SAINT, XPREP, and SADABS. The structure was solved by SHELXS and refined by SHELXTL. The crystal was grown by slow evaporation from methanol solution at room temperature. See Supporting Information for details.

## Results and Discussion

**Synthesis and Purification of Peptoid Oligomers.** We have created a series of peptoid oligomers of varying lengths and side-chain chemistries, incorporating the monomers shown in Table 1. Homooligomers of each of these monomers, comprised of 5, 6, 12, and 15 residues, were synthesized in good crude yield, ranging from 40 to 90% as estimated by analytical reversed-phase HPLC (Table 2). All compounds were purified to >97% homogeneity by preparative reversed-phase HPLC (as verified by analytical HPLC) before crystallization and/or analysis by CD and 2D-NMR. The molar masses of purified samples were confirmed by electrospray mass spectrometry (Table 2).

**X-ray Crystallographic Study of (*Nrch*)<sub>5</sub>.** Crystals of a peptoid pentamer (**1**), composed of  $\alpha$ -chiral aliphatic *Nrch*

monomers, were grown via slow evaporation from methanol solution. Figure 1 shows the crystal structure of **1**, the first solved for any peptoid oligomer. In its crystalline form, **1** adopts a left-handed helical conformation with repeating *cis*-amide bonds. The periodicity of this helix is approximately 3 residues per turn, with a pitch of ~6.7 Å as measured between corresponding main-chain atoms at residues 2 and 5. The carbonyl groups of residues 2–5 are aligned with the helix axis. An intermolecular hydrogen bond is present between the carbonyl oxygen of residue 2 and the terminal amide NH<sub>2</sub> group of an adjacent unit cell (not shown).

It is important to note here that the handedness of a peptoid helix is controlled in a simple and predictable fashion by the choice of side-chain enantiomer. For instance, enantiomeric peptoids containing  $\alpha$ -chiral aromatic side chains give rise to mirror-image CD that reflects identical helical conformations of opposing handedness.<sup>42,43</sup> Therefore, peptoid homooligomers incorporating either *Nrch* or *Nsch* monomers are analogous in structure, although of opposing handedness.

Accordingly, the backbone  $\phi$ – $\psi$  angles of **1** are similar to those observed for a polyproline type I peptide helix, yet are opposite in sign, reflecting opposing *Nrch* peptoid chirality. The backbone and side-chain  $\chi_1$  torsion angles of **1** (Table 3) are consistent with values that were predicted by a previous modeling study for a peptoid octamer with  $\alpha$ -chiral aromatic (*S*)-*N*-(1-phenylethyl)glycine (*Nspe*) monomers.<sup>44,45</sup> The sole exception is the  $\psi$  angle of residue 4, which is 141.7° and slightly outside the range predicted ( $\psi > 150^\circ$  or  $\psi < -150^\circ$ ). Importantly, this structure is also similar (though opposite in handedness) to the predominant solution conformation experimentally observed by 2D-NMR for a para-substituted *Nspe* pentamer in methanol solution.<sup>44,45</sup>

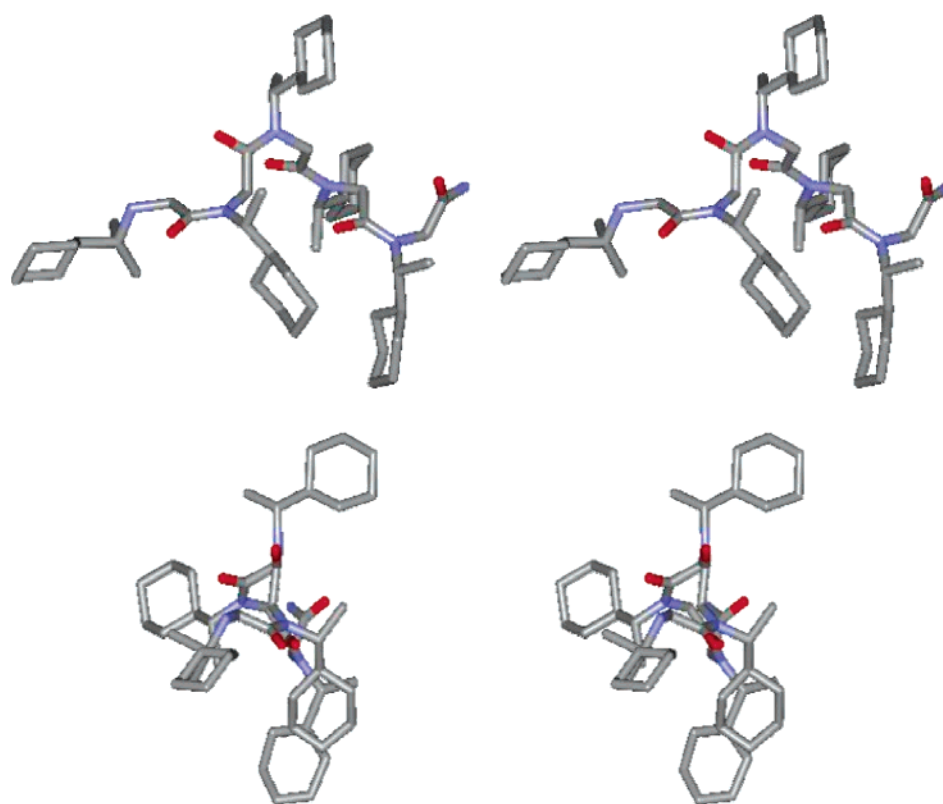
**CD Spectra of Peptoids with  $\alpha$ -Chiral Aliphatic Side Chains.** As observed for short-chained peptides, the CD spectrum of *Nrch*<sub>5</sub> (**1**) is relatively weak and is the mirror image of that observed for the analogous peptoid of opposing chirality, *Nsch*<sub>5</sub> (**2**) (see Supporting Information). Yet, even at the pentamer length, crystallographic studies strongly suggest that **1** has a strong conformational ordering and that its lowest free-energy conformation is a polyproline type I-like helix with *cis*-amide bonds. Consequently, the weak CD signals observed most likely reflect partial helical ordering in the solution state. This would be consistent with prior observations of nascent secondary structure in proline oligomers as short as 3–5 residues.<sup>48</sup>

CD spectra of an *Nsch* 6-mer, 12-mer, and 15-mer (**3a**, **3b**, and **3c**, respectively) are displayed in Figure 2a. Although the crystal structure presented in Figure 1 is an oligomer with all (*R*) side chains, we chose to investigate these longer homooligomers of (*S*)-*N*-(1-cyclohexylethyl)glycine to more closely mimic the stereochemistry of natural peptides. Whereas the *Nsch*<sub>6</sub> CD spectrum is weak, those of *Nsch*<sub>12</sub> and *Nsch*<sub>15</sub> show a distinct maximum at 210 nm and two shallow minima at 200 and 225 nm, respectively. Spectra intensify as chain length increases, in a manner consistent with prior CD studies of aromatic side chain-containing peptoids.<sup>42,46</sup> Intriguingly, the spectral features displayed most intensely by *Nsch*<sub>15</sub> are characteristics that are typically associated with a polyproline type I helix.<sup>49</sup>

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**Figure 1.** Stereo diagrams of the X-ray crystal structure of *Nrch* pentamer **1**. The structure is seen perpendicular (top) and parallel (bottom) to the long axis of the helix. Hydrogen atoms have been omitted for clarity. Note the alignment of the carbonyl groups parallel to the helix axis, consistent with the CD spectra observed for peptoid oligomers of this class.

**Table 3.** Observed Torsion Angles of (*Nrch*)<sub>5</sub> As Determined by X-ray Crystallography<sup>a</sup>

residue	$\omega$	$\phi$	$\psi$	$\chi_1$
1			-173.9	
2	-0.1	86.3	151.7	113.9
3	-3.4	67.6	-177.4	116.3
4	3.0	82.9	141.7	128.4
5	0.1	64.1	-162.5	116.9

<sup>a</sup> Values are the inverse of those for peptoids containing side chains of opposing chirality (*Nsch*).

The CD spectrum of *Nsch*<sub>6</sub> (**3a**) has comparable shape and intensity to that of the helical *Nrch*<sub>5</sub> (**1**). As previously discussed, their opposite chirality is responsible for their mirror-image spectra, which reflect opposite helical handedness. Therefore, these results suggest that the *N*-(1-cyclohexylethyl)glycine pentamer also nascently adopts a polyproline type I helix in solution, consistent with the crystal structure presented above.

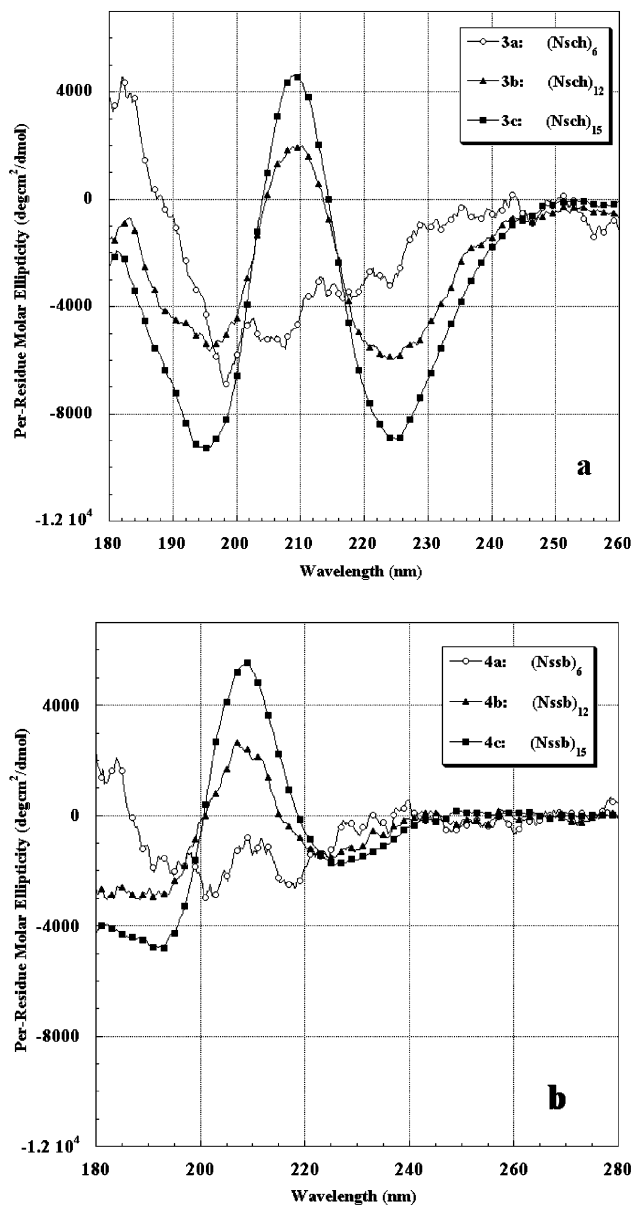
CD spectra similar to those of the *Nsch* homooligomers (**3a**, **3b**, and **3c**) are observed for *Nssb* homooligomers (**4a**, **4b**, and **4c**) at each corresponding chain length. However, the CD bands at 200 and 225 nm are somewhat reduced in intensity among the *Nssb* peptoids relative to the *Nsch* oligomers (Figure 2b). This reduced intensity is attributed to the greater extent of backbone flexibility in peptoids substituted with the less bulky *sec*-butyl side chains. However, despite the size difference between side chains (cyclic vs branched aliphatic), the shapes of the CD spectra and the consistent increase in spectral intensity with increasing chain length are qualitatively similar for these two classes of aliphatic side chain-containing peptoid oligomers.

As with the *Nsch* oligomers, we find that as chain length is extended, a more intense and well-defined CD spectrum is obtained, indicating a more ordered and stable helical fold. This observation is consistent with prior length-dependent CD studies of *m*-phenylene ethynylene oligomers,<sup>50</sup> phenylacetylene oligomers,<sup>51</sup>  $\beta$ -peptides,<sup>52,53</sup> and polyprolines.<sup>54</sup>

Temperature dependence studies have shown the thermal stability of polyproline helices,<sup>55,56</sup> and our CD experiments similarly demonstrate that the helical structure of peptoids containing  $\alpha$ -chiral aliphatic side chains is also quite stable to increased temperature. The CD spectra of pentadecamers **3c** and **4c** show only minor reductions in intensity as temperature increases from 15 to 75 °C (see Supporting Information). This thermal stability is consistent with a structure predominantly stabilized by steric repulsions rather than by intrachain hydrogen bonds.

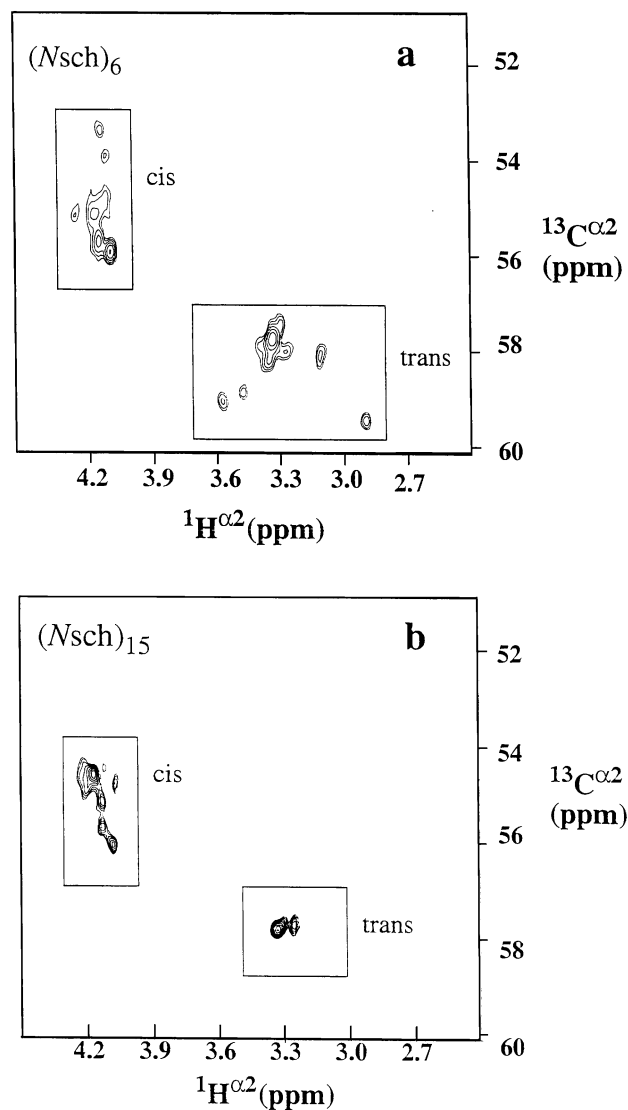
**2D-NMR Studies.** We conducted 2D-NMR experiments, specifically <sup>1</sup>H–<sup>13</sup>C HSQC analyses, to gain further insight into how conformation and structure evolve as oligomer length increases. Analysis of <sup>1</sup>H–<sup>13</sup>C HSQC spectra reveals differences

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**Figure 2.** A comparison of the CD spectra of the (a) *Nsch* 6-mer, 12-mer, and 15-mer (**3a**, **3b**, and **3c**, respectively) and (b) *Nssb* 6-mer, 12-mer, and 15-mer (**4a**, **4b**, and **4c**, respectively). Peptoid concentration was  $\sim 60 \mu\text{M}$  in acetonitrile; spectra were acquired at room temperature.

in the chemical shift distribution of the spectral regions that correspond to characteristic carbon–proton cross-peaks for short (hexamer) and long (pentadecamer) *Nsch* oligomers. Following a study in which a variety of peptoid nonamers based on *Nspe* monomers<sup>46</sup> were labeled with  $^{13}\text{C}$  groups at different positions (not shown, manuscript in preparation), these cross-peaks were unambiguously assigned to the tertiary substituted carbon methyne groups of  $\alpha$ -chiral side chains of peptoids. Analysis of the  $^1\text{H}$ – $^{13}\text{C}$  HSQC spectra of *Nsch* hexamer **3a** reveals that the number of peaks for proton–carbon correlation far exceeds that expected for a single helical conformation (Figure 3a). The multiplicity of solution conformations adopted by hexamer **3a** most likely arises from cis–trans isomerization about the amide bonds.<sup>44</sup> The chemical shift values of the peaks corresponding to the tertiary substituted carbon methyne clearly indicate the presence of both cis and trans conformations. For this hexamer,



**Figure 3.**  $^1\text{H}$ – $^{13}\text{C}$  HSQC spectra of peptoids with  $\alpha$ -chiral substituents. (a) *Nsch* **3a** and (b) *Nsch* **3c**. Panel displays the resonance cross-peaks for the tertiary substituted methyne protons. Peptoid concentration was  $\sim 2 \text{ mM}$  in acetonitrile- $d_3$ , and spectra were acquired at  $25^\circ\text{C}$ .

the cis:trans ratio, as determined by volume integration of the appropriate peaks, was  $\sim 1.0:1.0$ . This is consistent with the relatively weak CD spectrum we observed for this oligomer.

Figure 3b shows  $^1\text{H}$ – $^{13}\text{C}$  HSQC spectral regions corresponding to the tertiary substituted carbon methyne groups in the side chains of *Nsch*<sub>15</sub>. The HSQC spectra of the 15-mer show that the chemical distribution of peaks corresponding to the methyne protons is narrow (Figure 3b). The high degree of degeneracy of these peaks indicates that each residue in this longer peptoid resides in a similar chemical environment. Although extensive degeneracy of 2D-NMR peaks also may imply that the molecule is unstructured, the evolution and intensification of a polyproline type I CD spectrum among *Nsch* peptoids that occurs between 6 and 15 monomers in length suggests that the degeneracy here is the hallmark of increased order. In light of the X-ray crystal structure of *Nrch*<sub>5</sub> (Figure 1), which, together with CD data, suggests that even short peptoids may adopt nascently helical structure in solution, these results provide strong evidence that the highly repetitive conformation adopted by *Nsch*<sub>15</sub> in solution

is also helical. Volume integration of the peaks corresponding to the spectra of  $Nsch_{15}$  15-mer **3c** indicates the existence of two major families of dynamically interconverting conformers, but in this case they are populated in a ratio of  $\sim 2.6:1$  (*cis*-amide-dominated:*trans*-amide-dominated), consistent with the assertion that these oligomers gradually adopt a single, more well-ordered folded conformation as chain length is increased.

The HSQC spectra of the related  $Nssb_{15}$  oligomer **4c** are similar to those observed for  $Nsch_{15}$ , insofar as they are fundamentally degenerate and show many fewer cross-peaks than would account for each proton-carbon correlation (see Supporting Information). Again, 2D-NMR shows a ratio of dynamically interconverting conformers in a ratio of  $\sim 2.6:1$  (*cis*-amide-dominated:*trans*-amide-dominated). Also, the CD data from Figure 2b show a spectral evolution toward polyproline type I character that suggests that this peptoid adopts a highly repetitive, helical conformation similar to that of  $Nsch_{15}$ . Moreover, the CD spectra of  $(Nssb)_n$  oligomers closely resemble those of  $(Nsch)_n$  oligomers. We therefore conclude that peptoid oligomers with  $\alpha$ -chiral, *sec*-butyl side chains also adopt similar helical conformations.

It is intriguing that CD spectra resembling that of a polyproline type I helix are observed for peptoids based on  $Nsch$  and  $Nssb$  monomers, while peptoids with  $Nspe$  monomers give an  $\alpha$ -helix-like CD spectrum.<sup>42,46,47,57</sup> However, the results presented here indicate that they form helices of essentially the same structure. Prior evidence of the steric and electronic factors stabilizing peptoid helices<sup>42-45</sup> strongly indicates that the reasons for this difference in CD derive from effects of side-chain structure. The  $Nspe$  monomer most effectively constrains the peptoid backbone into the polyproline type I-like conformation because it can exhibit not only steric repulsions between bulky side chains, but also electronic repulsions between aromatic  $\pi$  and carbonyl lone-pair electrons.<sup>44,45</sup> In contrast, the  $Nsch$  monomer allows for greater conformational heterogeneity due to its lack of aromaticity, while  $Nssb$  is both more flexible and substantially less bulky than either  $Nsch$  or  $Nspe$ . For example, while 2D-NMR shows a 3.2:1 ratio of *cis*-amide-dominated:*trans*-amide-dominated conformers in  $Nspe_{15}$  (data not shown<sup>58</sup>), the ratio is only  $\sim 2.6:1$  in both  $Nsch_{15}$  and  $Nssb_{15}$ . Hence, we believe that only  $Nspe$ -based helices have a sufficiently high degree of backbone conformational stability to yield an exciton split  $\pi\pi^*$  transition in the CD spectrum, as observed for peptide  $\alpha$ -helices. An alternative explanation would be that signals generated by aromatic  $Nspe$  side chains may be strong enough to provide an anomalous signature in the far-UV CD that is not observed among  $Nsch$ - or  $Nssb$ -containing peptoids. Theoretical and experimental studies have demonstrated that exciton and coupled oscillator interactions between aromatic side chains can make substantial contributions to the far-UV CD profile.<sup>59</sup> Additional contributions may also arise from coupling interac-

tions between  $Nspe$  side-chain and backbone groups. Evaluation of the relative importance of these influences is the subject of ongoing investigations.

## Conclusions

Following an analysis of both crystallographic and spectroscopic data, we have shown that peptoid oligomers with  $\alpha$ -chiral aliphatic side chains adopt helices similar to both the polyproline type I helix and the helical structure adopted by peptoids with  $\alpha$ -chiral aromatic side chains. In particular, we have presented the first crystal structure solved for any peptoid oligomer, finding that an  $Nrch$  pentamer adopts a left-handed helix with *cis*-amide bonds. CD and 2D-NMR results obtained in acetonitrile solution suggest that peptoids of this class fold into a helical structure for which the degree of conformational homogeneity increases with increasing chain length. At long chain lengths, these  $\alpha$ -chiral aliphatic oligomers predominantly adopt a repetitive helical structure with *cis*-amide bonds.

We also present the new finding that peptoids with  $\alpha$ -chiral aliphatic side chains exhibit polyproline type I-like CD spectra in acetonitrile solution, consistent with the X-ray crystal structure of  $Nrch_5$ . This is in contrast to peptoids with  $\alpha$ -chiral aromatic side chains, which despite their polyproline type I-like structure exhibit CD spectra that qualitatively resemble that of a peptide  $\alpha$ -helix. The discovery that peptoids with aliphatic side chains can also form biomimetic helical structures in a manner similar to the aromatic-containing peptoids previously described has important implications for biomimicry. The capacity to mimic predominantly aliphatic peptide  $\alpha$ -helices is essential for designing accurate nonnatural analogues of transmembrane proteins and other small, active protein domains. These findings may also be of interest to those studying the folding propensities of polyprolines, since in many ways this class of peptoids is analogous to these natural molecules.

**Acknowledgment.** We are grateful to Ishwar Radhakrishnan for helpful discussions and to Fred Hollander and his X-ray lab at U.C. Berkeley. This material is based upon work supported by the National Science Foundation under Grant No. BES-9870386. We acknowledge the use of the Keck Biophysics Facility at Northwestern University ([www.biochem.nwu.edu/Keck/Keckmain.html](http://www.biochem.nwu.edu/Keck/Keckmain.html)) for CD measurements. C.W.W. and J.A.P. were supported by an NIH Biophysics Training Grant (Grant No. 5 T32 GM08382-10). T.J.S. was supported by the Illinois Minority Grant Incentive Program and a grant from the NIH (R01 DE 13030).

**Supporting Information Available:** CD spectra, 2D-NMR spectra, and crystal parameters (CIF data) of peptoid oligomers (PDF). This material is available free of charge via the Internet at <http://pubs.acs.org>.

JA037540R

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