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Controlling the sign and magnitude of screw-sense preference from the C-terminus of an achiral helical foldamer†

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The global screw-sense preference of an achiral helical oligomer may be controlled by a single chiral monomer located at one terminus. Remarkably, maximal control is induced in oligomers of the achiral quaternary amino acid Aib by a single C-terminal alaninamide residue, probably because the Ala side chain, though small, is compatible with a 3_{10} helical conformation. The presence or absence of a C-terminal hydrogen bond donor determines the screw sense of the entire oligomer.

The adoption of well-defined conformations is a characteristic feature of many classes of biomolecules, and understanding the encoding of conformational features within the primary structure of proteins is an important challenge.[1] Foldamers are synthetic oligomers and polymers that likewise adopt well-defined conformations,[2] and their utility depends on using simple structural features (dipole orientation, hydrogen bonding ability, stereochemical configuration) to induce global conformational, and hence ultimately functional, consequences.[3] We[4] and others[5,6] have shown that helical oligomers containing the achiral quaternary amino acid Aib (α-aminoisobutyric acid)[7] adopt preferentially a left- or a right-handed[4a] global screw sense as a result of the local stereochemical influence of a chiral N-terminal amino acid residue,[4a–j,5a] a chiral diol bound to an N-terminal boronate binding site,[4k] or a chiral carboxylic acid ion-paired with an N-terminal amino group.[5b] These N-terminal controllers of global conformation function by providing appropriately orientated hydrogen-bond acceptors that organise the N-terminal NH groups of the overall 3_{10} helical structure[7] of the oligomer into a left- or a right-handed β-turn.[8]

Little is known about the propensity of achiral peptide helices to have their screw sense induced from the C terminus. Chiral C-terminal residues induce some degree of screw-sense preference in short achiral helices,[5c,6] but a comparison[9] between an N- and a C-terminal controller suggested that C-terminal control was subordinate to N-terminal control.

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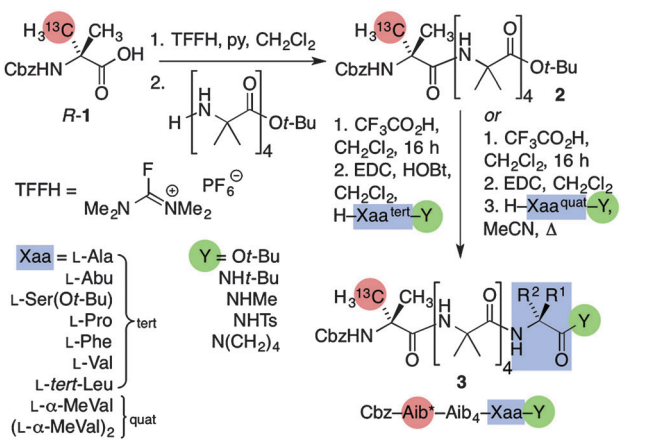
We now report a quantitative analysis of the role played by a chiral C-terminal residue in determining the global screw-sense preference of a series of otherwise achiral Aib-based oligomers **3**. The compounds in question were synthesised by ligating a range of amino acid derivatives H–Xaa–Y (esters Y = OR or amides Y = NHR or NR_2) to the C terminus of an Aib pentamer **2**, as shown in Scheme 1. Derivatives of tertiary amino acids (H–Xaa^{tert}–Y) coupled cleanly using the coupling agent EDC in the presence of HOBt. Derivatives of quaternary amino acids (H–Xaa^{quat}–Y) failed to couple under these conditions, but nonetheless cleanly opened the azlactone derivative of **2** (generated using EDC) on reflux in acetonitrile.

In order to quantify the screw-sense preference induced by the C-terminal residues, the N-terminal Aib residue of the pentamer **2** was isotopically labelled in an enantioselective manner with ^{13}C. The required protected amino acid *R*-**1** was synthesised from L-Ala by a method[10] that enriches the ^{13}C abundance in the pro-*R* methyl group to *ca.* 80% and the pro-*S* methyl group to *ca.* 20%.

The ^{13}C NMR spectrum of each oligomer **3** was acquired at +23 °C in CD_3OD, a solvent in which the NMR spectra of Aib oligomers show no concentration-dependent effects.[11] The chemical shift separation $\Delta\delta_{fast}$ between the minor and major ^{13}C NMR



Scheme 1 Synthesis of the ^{13}C labelled oligomers **3**.

Table 1 Conformational preferences in Aib oligomers **3** carrying C-terminal controllers Xaa-Y

Entry	Compound	Residue Xaa	R^1	R^2	Y	$\Delta\delta_{fast}$[a]/ppb	$\lvert\Delta\delta_{slow}\rvert$[b]/ppb	h.e.[c]/%	h.r.[d]
1	**3**-Ala-NH*t*-Bu	Ala	Me	H	NH*t*-Bu	+1807	2415	+75	88 : 12
2	**3**-AlaNHMe	Ala	Me	H	NHMe	+1883	—	+78	89 : 11
3	**3**-Ala-N$(CH_2)_4$	Ala	Me	H		−800	—	−33	33 : 67
4	**3**-Abu-NH*t*-Bu	Abu[e]	Et	H	NH*t*-Bu	+1857	—	+77	88 : 12
5	**3**-Ser(O*t*-Bu)[f]-NH*t*-Bu	Ser(O-*t*-Bu)[f]	CH_2O*t*-Bu	H	NH*t*-Bu	+668	—	+28	64 : 36
6	**3**-Pro-NH*t*-Bu	Pro	–$(CH_2)_3$CH–	H	NH*t*-Bu	+376	—	+16	58 : 42
7	**3**-Phe-NH*t*-Bu	Phe	Bn	H	NH*t*-Bu	+1676	2410	+70	85 : 15
8	**3**-Phe-NHTs	Phe	Bn	H	NHTs	+1057	—	+36	68 : 32
9	**3**-Val-NH*t*-Bu	Val	i-Pr	H	NH*t*-Bu	+1726	2420	+71	86 : 14
10	**3**-Tle-NH*t*-Bu	*tert*-Leu	*t*-Bu	H	NH*t*-Bu	+1575	—	+65	83 : 17
11	**3**-αMv-NH*t*-Bu	α-MeVal	i-Pr	Me	NH*t*-Bu	+1710	—	+70	85 : 15
12	**3**-αMv_2-NH*t*-Bu	α-$MeVal_2$	i-Pr	Me	NH*t*-Bu	+1943	—	+80	90 : 10
13	**3**-Ala-O*t*-Bu	Ala	Me	H	O*t*-Bu	−1336	—	−55	22 : 78
14	**3**-Phe-O*t*-Bu	Phe	Bn	H	O*t*-Bu	−816	2427	−34	33 : 67
15	**3**-Val-O*t*-Bu	Val	i-Pr	H	O*t*-Bu	−1112	2441	−46	27 : 73
16	**3**-Tle-O*t*-Bu	*tert*-Leu	*t*-Bu	H	O*t*-Bu	−838	—	−35	32 : 68
17	**3**-αMv-O*t*-Bu	α-MeVal	i-Pr	Me	O*t*-Bu	−1037	—	−43	28 : 72

[a] Chemical shift separation between minor and major labelled peaks $[\delta_{fast}^{minor} - \delta_{fast}^{major}]$ in the ^{13}C NMR spectrum in CD_3OD at +23 °C. [b] Modulus of the chemical shift separation between labelled peaks in the ^{13}C NMR spectrum in CD_3OD at −40 °C. Where no value for $\Delta\delta_{slow}$ was measured, an average value of $\Delta\delta_{slow}$ = 2420 was employed. [c] Helical excess = $\Delta\delta_{fast}/|\Delta\delta_{slow}|$ interpreted as $\{[P] - [M]\}/\{[P] + [M]\}$. Positive values indicate right-handed screw sense predominates. [d] Helical ratio = $[P] : [M]$. [e] Abu = (*S*)-(+)-2-aminobutyric acid = L-(+)-butyrine. [f] Serine side-chain protected as its *t*-butyl ether.

signals of each oligomer is reported in Table 1. For a number of oligomers, the ^{13}C NMR spectrum was also acquired at −70 °C in CD_3OD. Variable temperature NMR over the range −70 to +40 °C (Fig. 1b) showed that spectra at the upper and lower limits of this range provided suitable values for chemical shift separation at slow and fast exchange $\Delta\delta_{slow}$ and $\Delta\delta_{fast}$,[4c] and the values obtained at −70 °C for $\Delta\delta_{slow}$ are also reported in Table 1.

The measured values of $\Delta\delta_{slow}$ are constant within 1% across the compounds studied, consistent with the assumption[4c] that the spatial separation between the ^{13}C NMR reporter and the chiral C-terminal residue ensures that the anisochronicity of the ^{13}C labels at low temperature results entirely from their local interaction with the slowly inverting helix. Thus, at fast exchange, the value of $\Delta\delta_{fast}$ is dependent only on the equilibrium ratio of the *M* and *P* helices,[4c,e] and the value $\Delta\delta_{fast}/|\Delta\delta_{slow}|$ may be interpreted as helical excess (h.e.), as reported in Table 1.

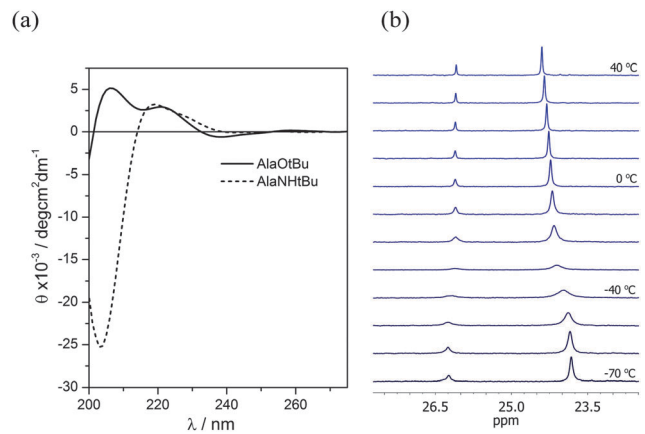


Fig. 1 (a) Circular dichroism spectra of **3**-Ala-NH*t*-Bu and **3**-Ala-O*t*-Bu at +20 °C in MeOH, measured at 7×10^{-4} mol dm^{-3}. The signs of the bands at 205 nm[4e,13] indicate a *P* (right-handed) screw-sense preference in **3**-Ala-NH*t*-Bu and an *M* (left-handed) screw-sense preference in **3**-Ala-O*t*-Bu; (b) Variable temperature ^{13}C NMR spectra of **3**-Ala-NH*t*-Bu at 10 °C intervals from −70 °C (bottom) to +40 °C (top). Coalescence occurs between −50 and −20 °C.

The sign of the helical excess was deduced from the location of the major signal arising from the ^{13}C-labelled Aib residue. The pro-*R* methyl group of an Aib residue resonates upfield of the pro-*S* methyl group in a right handed (*P*) 3_{10} helix and downfield in a left handed (*M*) 3_{10} helix.[4d,12] Positive values of $\Delta\delta_{fast}$ thus correspond to *P* helicity, a deduction confirmed by circular dichroism (Fig. 1a): the negative diagnostic band at 205 nm[4e,13] for **3**-Ala-NH*t*-Bu confirms *P* helicity, and the positive band at 205 nm for **3**-Ala-O*t*-Bu confirms *M* helicity.

The first conclusion that can be drawn from the results in Table 1 is that C-terminal secondary amides of L-amino acids, whether tertiary (entries 1 and 2, 4–10) or quaternary (entries 11 and 12), induce *P* helicity, while C-terminal esters (entries 13–17) and a tertiary amide (entry 3) induce *M* helicity. We deduce that the divergent behaviour of these two groups of compounds is due to the ability of the secondary amides to use their NH group to form an additional C-terminal hydrogen bond, promoting continuation of a 3_{10} helical structure, as illustrated in Fig. 2a. It is well established that L residues located within a 3_{10} helix promote right-handed screw-sense preference,[4a,14] and Fig. 2c illustrates the mechanism by which a bulky R^1 substituent exerts this effect by lying perpendicular to the plane of the adjacent amide group. The weak preference induced by ProNH*t*-Bu (entry 6) is likely to be due to the bend induced by a Pro residue,[15] preventing or weakening this C-terminal hydrogen bond. By contrast, the inability of C-terminal esters to continue a hydrogen-bonded network gives rise to the well-known 'Schellman motif',[16] in which dipole repulsion leads to a local helical inversion. Such a motif is evident in the X-ray crystal structure of **3**-Ala-O*t*-Bu (Fig. 3). A single L residue adopting this motif has been

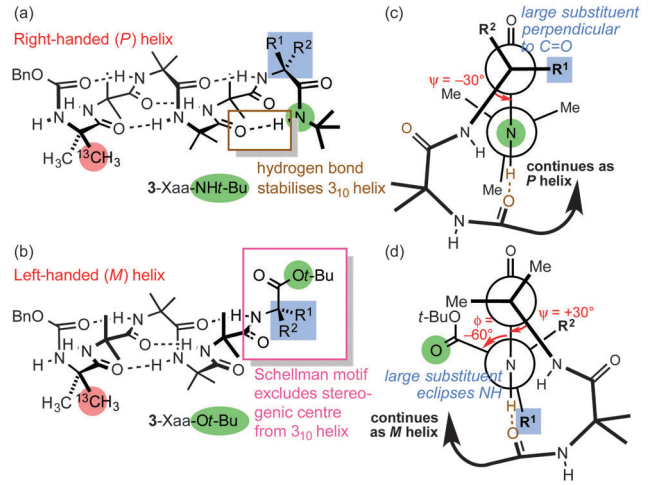


Fig. 2 Conformations of Aib oligomers **3** bearing (a) a C-terminal secondary amide function and (b) C-terminal ester (or tertiary amide) function, with (c) and (d) showing Newman projections of their C-termini viewed from N-terminal direction to illustrate the origin of the conformational preference.

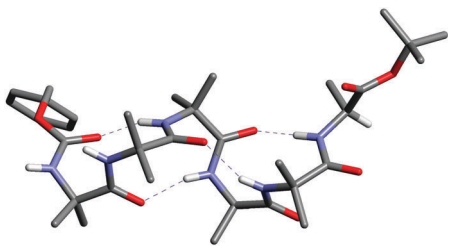


Fig. 3 X-ray crystal structure[18] of **3**-Ala-O*t*-Bu, showing a C-terminal Schellman motif and left-handed (*M*) helicity.

noted before to induce a left-handed screw sense in an otherwise achiral oligomer,[5c] and Fig. 2d illustrates the origin of the effect. The tertiary amide **3**-Ala-N$(CH_2)_4$ also exhibits *M* screw-sense, presumably also the result of a corresponding '*tert*-amide Schellman' motif.[17]

The conformational constraint imposed by their additional hydrogen bond means that the secondary amides generally control screw-sense preference to a greater degree than the esters, irrespective of the size of the amide N-substituent (compare entries 1 and 2). Within each series there are however some surprising features. Acidification of the C-terminal NH group by the tosyl group in **3**-Phe-NHTs (entry 8) fails to increase conformational preference over the *tert*-butyl amide (entry 7), and in both the amide and the ester series the greatest degree of screw-sense preference (75% h.e. for **3**-Ala-NH*t*-Bu) induced by a single chiral residue results from Ala (entries 1, 2 and 13), rather than the more bulky chiral amino acids. Only the α-methylvaline dimer of **3**-αMv_2-NH*t*-Bu (entry 12; a motif which induces comparably high screw-sense control when located at the N terminus[4d,e]) exerts more powerful control (80% h.e.) than AlaNH*t*-Bu.

The more powerful control exerted by residues Ala or Abu (entries 1–3) with smaller side chains is consistent with screw-sense control being greatest in a conformationally uniform helix that can adopt through its whole length a 3_{10} helical structure,[7] since Ala and Abu are readily accommodated by the steric demands of the 3_{10} helical environment.[19] Although the steric differentiation at the stereogenic centre of Val, Phe and *tert*-Leu (entries 7–10) is greater, these residues have a lower propensity for helix formation,[20] and presumably favour alternative conformations with lower screw-sense preferences. The lower screw-sense control induced by Ser(O*t*-Bu)NH*t*-Bu (entry 5) suggests that more remote steric bulk is likewise not well tolerated by a 3_{10} helical structure. α-MeVal, being quaternary, is compatible with a 3_{10} helix,[4d,21] the greater selectivity observed with Ala *vs.* α-MeVal being simply due to the steric differentiation between Me *vs.* H and i-Pr *vs.* Me.

In conclusion, C-terminal L amino acid residues induce a preferred right-handed screw sense as their amide derivatives and left-handed screw sense as their ester derivatives in a series of helical Aib oligomers. Screw-sense control is maximised by residues that can participate in a 3_{10} helical structure, namely L-Ala and the dimer of L-α-MeVal.

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