

The vicinal F–C–C–F moiety as a tool for influencing peptide conformation

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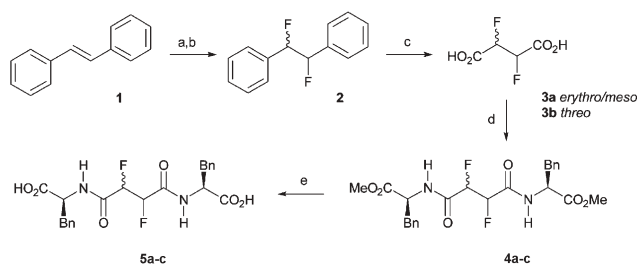
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Diastereoisomers of bis(amino acid amides) of 2,3-difluorosuccinic acid have been prepared and the *erythro*- and *threo*-isomers display very different conformations.

Strategic replacement of fluorine for hydrogen has been widely employed in the pharmaceutical industry to protect candidate bioactives against adventitious metabolism^{1,2} or in the development of mechanism-based enzyme inhibitors.³ We have been exploring another aspect of fluorine incorporation into organic molecules whereby the polarity of the C–F bond is used as a tool for influencing the conformation of organic compounds.⁴ There are attractive prospects here, not only in the bioactives arena, but also for the development of performance materials products.⁵ Recently we reported that α -fluoroamides have a preferred stereoelectronic orientation, where the C–F bond is *syn*-coplanar with the N–H bond and *anti*-coplanar to the amide carbonyl.⁶ This is a significant effect in energy terms, with a barrier to rotation of around 8.0 kcal mol⁻¹ when evaluated in small molecules. Seebach has recently shown that pairs of β -amino acid derived oligopeptides, which differ only in which enantiomer of 2-fluoro- β -alanine is present at the centre of the molecule, possess different secondary structures.⁷ The C–F bond is argued to be a significant contributor to these experimental observations. Thus diastereoisomers generated by the interconversion of a single C–F bond can differently and significantly influence the conformation of larger molecules through such stereoelectronic effects.

The lower energy *gauche*- versus *anti*-conformer for 1,2-difluoroethane has been widely studied and the fluorine *gauche*-effect is a well known phenomenon in organofluorine chemistry.⁸ The effect extends to *erythro*- and *threo*-stereoisomers of organic compounds such as 2,3-difluorobutane⁹ and 9,10-difluorostearic acids; compounds which differ in physical properties and display quite distinct behaviour, as a consequence of a single stereogenic C–F bond inversion.¹⁰ In this study the α -fluoroamide and vicinal fluorine *gauche*-preferences are explored in *erythro*- and *threo*-diastereoisomers of a series of bis(amino acid) 2,3-difluorosuccinamides. It was anticipated that the two different diastereoisomers would display different conformational preferences as a result of the relative stereochemistry of the vicinal C–F bonds.

The diastereoisomeric mixture of the *erythro/meso*- (**3a**) and *threo*-isomers (**3b**) was prepared as illustrated in Scheme 1. Treatment of *trans*-stilbene **1** with *N*-bromosuccinamide in the presence of hydrogen fluoride, followed by addition of silver fluoride, generated a 4:1 mixture of *erythro*- and



Scheme 1 Reagents and conditions: a) NBS, HF/py, Et₂O; b) AgF, 73%; c) O₃, AcOH, H₂O₂, 45%; d) PheOMe, EDCI, HOBt, DMF, 83%; e) aq. HCl–acetone, 100%.

threo-stereoisomers **2**, according to the one pot procedure reported by Olah.¹¹ The major product is the *erythro/meso*-isomer, consistent with a predominant double inversion mechanism due to aryl participation.¹² Both of the diastereoisomers of **2** could be purified by a combination of chromatography and crystallisation.

For preparative purposes **2** was used either as the 1:1 isomeric mixture or as the predominant *erythro/meso*-stereoisomer which can conveniently be crystallised to purity. Treatment with ozone¹³ followed by oxidative work up generated the corresponding difluorosuccinic acids **3** without any stereochemical loss. Yagupolskii *et al.*¹⁴ have prepared *erythro*-difluorosuccinic acid **3a** by direct treatment of L-tartaric acid with SF₄ in the presence of hydrogen fluoride, and the *threo*-stereoisomer **3b** was prepared in a similar manner by Hudlicky.¹⁵ Our attempts to use DAST in such a reaction failed and thus the synthetic route depicted in Scheme 1 offers a practical route, certainly for the small scale preparation of **3a** and **3b**.

To prepare a bis(amino acid amide), (*S*)-phenylalanine methyl ester and 2,3-difluorosuccinic acids **3** (1:1 diastereoisomeric mix) were treated with EDCI/HOBt to form the bis(amino ester amides) **4** as a diastereoisomeric mixture. Interestingly, all of the three stereoisomers of **4** (Fig. 1) could be separated by flash chromatography. This allowed the hydrolysis of the individual stereoisomers **4a–c** under acidic conditions, to generate the corresponding dicarboxylic acids **5a**, **5b**, and **5c**.

Both the *threo*- and the *erythro*-stereoisomers, **5a** and **5b** respectively, gave suitable crystals for X-ray analysis and the resultant structures are shown in Figs. 2 and 3.† The approximate relative relationships of the functional groups along C2–C3 for **5a** and **5b** are illustrated by the Newman projections. It is notable that for each structure a *gauche*-relationship is maintained between the vicinal fluorines (F–C–C–F angle 54° in **5a** and 77.1° in **5b**). Also, the α -fluoroamide functionalities hold an approximate planar arrangement in each case. This is distorted a little in one of the

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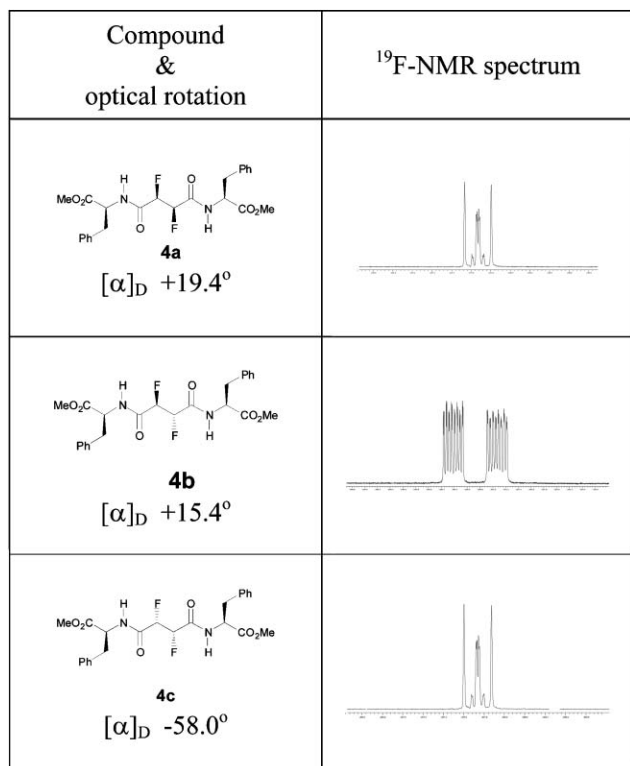


Fig. 1 Structures, optical rotation values and ^{19}F -NMR spectra of **4a–c**.

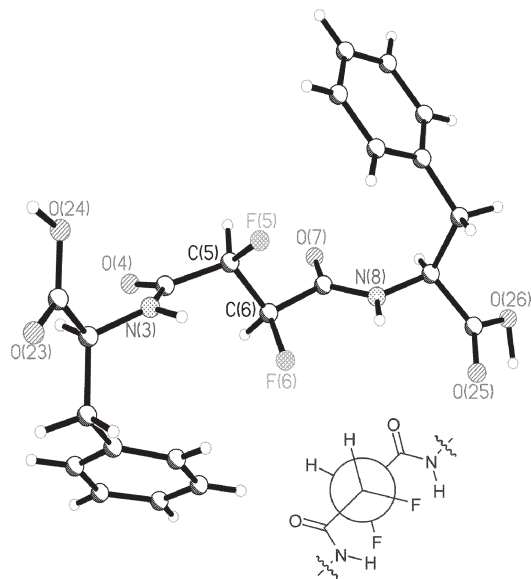


Fig. 2 X-Ray structure of **5a**, which was obtained from **4a** by direct hydrolysis. The carbon chain conformation of the 2,3-difluorosuccinamide moiety is very different from **5b** (Fig 3), but conforms to that predicted by the fluorine *gauche*-effect and the α -fluoride amide effect. Selected bond lengths (\AA) and torsion angles ($^\circ$) for **5a**: N3–C4 1.338(7), N3–H(3N) 0.9800(11), C4–O4 1.214(7), C5–F5 1.397(7), C5–C6 1.495(10), C5–H(5A) 1.0000, C7–O7 1.220(7), C7–N8 1.332(7), N(8)–H(8N) 0.9800(12); O4–C4–C5–F5 155.4(6), N3–C4–C5–F5 $-23.8(8)$, F5–C5–C6–F6 49.0(6), F6–C6–C7–O7 $-165.6(6)$, F6–C6–C7–N8 $-12.2(8)$. Flack parameter 0.23(13).

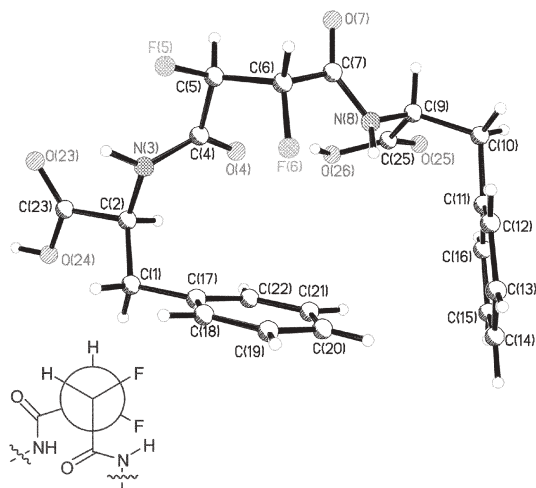


Fig. 3 X-Ray structure of **5b**, which was obtained by direct hydrolysis of **4b**. There were two molecules of **5b** in the asymmetric unit with similar conformations; only one is shown. The 2,3-difluorosuccinamide moiety is very different from **5a** (Fig 2). Selected bond lengths (\AA) and torsion angles ($^\circ$) for **5b**: N3–C4 1.349(9), N3–H(3N) 0.9800(13), C4–O4 1.236(8), C5–F5 1.406(7), C5–C6 1.538(9), C7–O7 1.229(8), C7–N8 1.345(8), N(8)–H(8N) 0.9800(11); O4–C4–C5–F5 $-176.1(6)$, N3–C4–C5–F5 4.8(8), F5–C5–C6–F6 75.2(6), F6–C6–C7–O7 $-178.0(5)$, F6–C6–C7–N8 3.4(8). Flack parameter $-1.6(14)$.

amides in **5a** [N(3)–C(4)–C(5)–F(5) dihedral angle of -23.8°]. In the crystal packing diagram the amide N(3)–H is involved in an intermolecular hydrogen bonding interaction with the carbonyl of another amide (not shown) which results in a small deviation from the ideal *anti*-planar geometry and a close N(H)···F interaction to fluorine F(5). However, in the main, the *anti*-planar α -fluoroamide is a feature of these structures.

The 2,3-difluorosuccinamide moiety in structure **5a** has the carbon chain in an *anti*-zig-zag conformation with a C–C–C torsion angle of 163° , whereas in **5b** this torsion angle is 78.3° . Thus for the different diastereoisomers the main chain of the 2,3-difluorosuccinamide moiety is contorted quite differently. In both cases the fluorines are *gauche*, whereas the amides are *anti* to each other in **5a** but *gauche* in **5b**. Thus the relative configuration of the C–F bonds is influencing the overall conformation of these bis(amides), and in a predictable manner. These solid state conformations are also supported by solution state NMR studies.

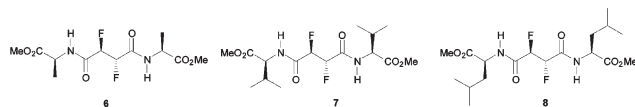
The ^{19}F -NMR spectra for each of the diastereoisomers of **5** are shown in Fig. 1 and the $^3J_{\text{HH}}$ and $^3J_{\text{HF}}$ coupling constants for **4** and **5** are shown in Table 1. The *threo*-stereoisomers give rise to an AA'XX' spin system due to the magnetic non-equivalence of the vicinal fluorines and their geminal hydrogens. The *erythro*-stereoisomers **4b** and **5b** have truly diastereotopic fluorines and

Table 1 NMR derived coupling constants (J) for compounds **4** and **5**

Compound	$^3J_{\text{HF}}$	$^3J_{\text{HH}}$	$^3J_{\text{FF}}$	$^2J_{\text{HF}}$
<i>threo</i> - 4a	30.5	1.3	12.0	46.1
<i>erythro</i> - 4b	23.0	1.3	12.0	48.0
<i>threo</i> - 4c	30.9	1.4	12.3	45.6
<i>threo</i> - 5a	31.8	1.0	12.0	44.9
<i>erythro</i> - 5b	24.1	1.3	12.3	47.4
<i>threo</i> - 5c	31.1	1.5	10.0	45.6

therefore show first order spectra. In a similar manner to the Karplus relationship for $^3J_{\text{HH}}$ coupling constants, the magnitude of the $^3J_{\text{HF}}$ coupling constants have an angular dependence, although these values are quite sensitive to the nature of the other substituents.¹⁶ On the other hand the $^3J_{\text{FF}}$ coupling constants are not reliable indicators of vicinal relationships.¹⁷

The large $^3J_{\text{HF}}$ (~30 Hz) and small $^3J_{\text{HH}}$ (1.0–1.5 Hz) coupling constant values for the *threo-a/c* isomers can be contrasted with the smaller $^3J_{\text{HF}}$ (~23 Hz) coupling constants for the difluoro-*erythro-b* isomers. In the *threo-a/c* isomers the larger $^3J_{\text{HF}}$ coupling constants indicate that both C–F bonds have a vicinal C–H bond *anti* to each other, as found in the solid state structure of **5a**, and they also conserve a fluorine *gauche*-relationship (see Newman projections, Figs 2 and 3). The smaller $^3J_{\text{HF}}$ coupling constants of the *erythro*-series are consistent with a solution conformation where the vicinal fluorines are predominantly *gauche* with respect to each other. Concurrently, the small $^3J_{\text{HH}}$ coupling constants in each series indicate a vicinal *gauche*-H–H relationship in each case (see Newman projections in Figs 2 and 3). Hence it can be concluded that for each diastereoisomeric series the vicinal fluorines prefer a *gauche*-relationship, which influences the relative orientation of the peripheral amides. We note that the *threo*-diastereoisomers (**4a** and **4c**) had very different optical rotation $[\alpha]_{\text{D}}$ values ranging from -58.0° to $+19.4^\circ$ (Fig 1).



A series of amino acids, including (*S*)-alanine, (*S*)-valine and (*S*)-leucine, were then coupled as their methyl esters to *erythro*-meso-difluorosuccinic acid **3a**, prepared from the purified meso-difluorodiphenylethane **2a** by means of ozonolysis. This generated the *erythro*-stereoisomers **6**, **7** and **8** respectively, in enantiomerically pure form. The NMR coupling constants of all these compounds resemble closely those values for the *erythro*-compounds **4b** and **5b**, and thus similar solution state conformations of the difluorosuccinamide moiety can be assumed.

In summary, the bis(*S*)amino acid ester) amides of 2,3-difluorosuccinic acid, **4a–c**, have been prepared in diastereomerically pure form. The solid state conformations of the hydrolysed bis(acids) **5a** and **5b** suggest that the α -fluoro amides adopt an approximately *anti*-planar arrangement and solution state NMR data of **4** and **5** indicate that the vicinal fluorines adopt a *gauche*-conformation within the difluorosuccinamide moiety, and that the amide groups are orientated differently for the two diastereoisomeric series. Such behaviour of the C–F bond should be useful in the design of peptide mimetics and in influencing the conformation of bioactive molecules.

Notes and references

† Crystal data for pseudopeptide **5a**: $M = 486.87$; orthorhombic, space group $P2(1)2(1)2(1)$; $a = 4.9826(19)$, $b = 22.066(10)$, $c = 24.319(9)$ Å; $V = 2673.7(18)$ Å³; $T = 173(2)$ K; $Z = 4$; $\mu = (\text{Mo-K}\alpha) 1.709 \text{ mm}^{-1}$; reflections: total = 35848, unique 4760 ($R_{\text{int}} 0.1355$); $R_1 = 0.1260$, $wR_2 = 0.3296$ for 4760 observed data [$I > 2\sigma(I)$]. CCDC 271257.

Crystal data for pseudopeptide **5b**: $M = 448.42$; monoclinic, space group $P2(1)$; $a = 11.121(2)$, $b = 6.1323(10)$, $c = 30.671(6)$ Å; $\beta = 90.179(6)^\circ$; $V = 2091.8(7)$ Å³; $T = 93(2)$ K; $Z = 4$; $\mu = (\text{Mo-K}\alpha) 0.115 \text{ mm}^{-1}$; reflections: total = 13463, unique 6970 ($R_{\text{int}} 0.0887$); $R_1 = 0.0825$, $wR_2 = 0.1525$ for 4011 observed data [$I > 2\sigma(I)$]. CCDC 271258. See <http://dx.doi.org/10.1039/b506010a> for crystallographic data in CIF or other electronic format.

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