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Design of a Novel Secondary Structure Scaffolding Device: Induction of a Reverse Turn in Tetrapeptides by Incorporating a β-Amino Acid and Stereocontrolled Free Radical α-Substitution Reactions in Peptide Motifs

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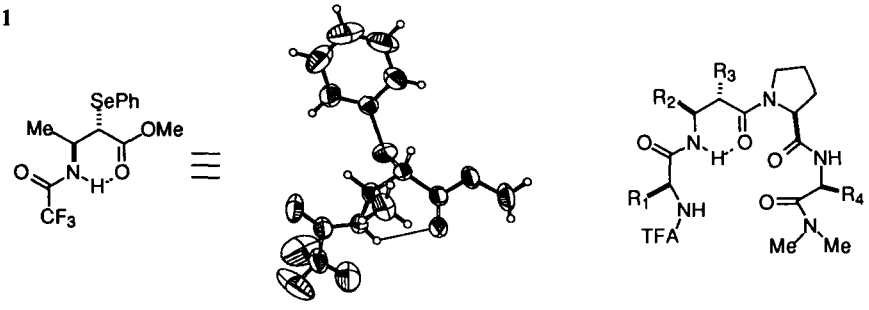
Abstract: The incorporation of α-substituted β-amino acids into a tetrapeptide motif induces a reverse turn in aprotic solvent systems as revealed by NOESY and ROESY techniques, and by CD spectra. The conformations of these compounds have been studied by molecular modeling, and further supported by performing a highly stereoselective free radical allylation reaction on an α-phenylseleno β-amino acid unit within the tetrapeptide. © 1997 Elsevier Science Ltd.

The de novo design of secondary structures of peptides is an area which has attracted considerable attention in recent years[1]. In this regard, β-turns play an important structural and conformational role in many biologically relevant proteins.[2] These motifs have been the focus of many studies in the design and synthesis of peptidomimetics and related molecules.[3] Common strategies for constructing β-turn-like secondary structures consist in incorporating a conformationally rigid peptidomimetic cyclic molecule,[2-4] a conformationally rigidified acyclic peptidomimetic molecule[3] such as an α,β-unsaturated γ-amino acid,[5] an α,α-dialkylamino acid,[6] or a D-α-amino acid.[7] As an extension of our studies on the highly stereoselective free radical allylations of α-phenylseleno β-amino acids by exploiting intramolecular H-bonding as a stereocontrolling element,[8] we report herein on the synthesis and conformational aspects of a tetrapeptide that comprises an α-substituted β-amino acid.[9]

In our previous studies, we found that α-substituted β-amino acid derivatives adopt a conformation in which the N-H and C=O(NMe$_2$ or OMe) groups are coplanarly aligned in space to form a pseudo six-membered ring *via* an intramolecular H-bond.[8] Based on solution and X-ray crystallographic data as well as on molecular modeling data, we hypothesized that the incorporation of a β-amino acid in a peptide chain might enforce a reverse turn, thus generating a β-turn-like secondary structure (Figure 1).

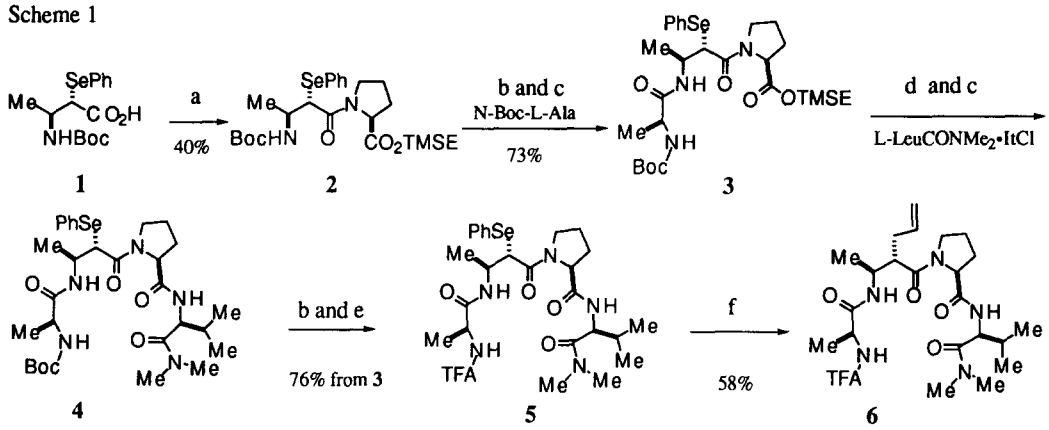
Figure 1



Proposed reverse turn structure

The synthesis of the intended tetrapeptides followed an unencumbered path as shown in Scheme 1. Thus, treatment of the dianion of Boc-β-amino-L-butyric acid methyl ester with phenylselenyl bromide gave almost exclusively the corresponding (*S*)-α-phenylselenyl derivative which was carefully hydrolyzed to the acid **1**. Peptide coupling with 2-trimethylsilylethyl L-prolinate in the presence of PyBOP or TBTU/DIPEA gave the dipeptide [(α-*R*-phenylseleno)-Boc-β-HAla]-Pro-$OCH_2CH_2SiMe_3$, **2**. Acid hydrolysis of the Boc group followed by peptide coupling with Boc-Ala-OH gave the corresponding tripeptide **3** in 73% yield. Finally, removal of the TMSE ester group and coupling of the resulting proline tripeptide with H-Val-NMe_2 led to the desired tetrapeptide **4** harboring an enantiopure α-phenylseleno β-amino acid. The corresponding "natural" tetrapeptide sequence BocAla-Ala-Pro-ValNMe_2 was synthesized in a similar way.

Scheme 1



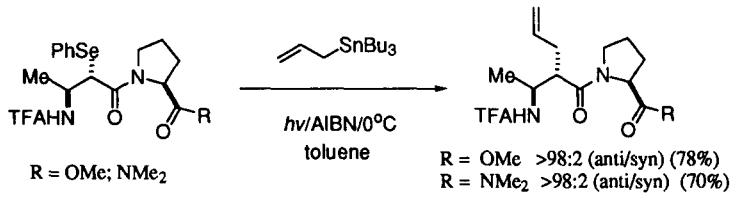
Reaction conditions: a. PyBOP/DIPEA/CH_3CN/-78 to 0°C, 2-trimethylsilylethyl prolinate; b. 4N HCl/dioxane; c. TBTU/DIPEA /CH_3CN/0°C; d. TBAF/THF; e. TFAA/TEA/CH_2Cl_2/-78°C; f. allyl tributylstannane/AIBN/*hv*/CH_2Cl_2/0°C.

In order to maximize intramolecular H-bonding as in our previous studies,[8] we converted the Boc group in the tetrapeptide **4** into the corresponding N-trifluoroacetyl analog **5**. Treatment of the latter with allyl tributylstannane in the presence of AIBN under irradiation afforded a compound **6** as a single diastereomer in which the phenylseleno group had been replaced by an allyl group with retention of configuration. That the free radical allylation had taken place with a greater than 95:5 ratio of stereoselectivity as in related β-amino acid amides[8], was ascertained by preparing the product **6** independently utilizing the preformed α-(*S*)-allyl β-amino L-butyric acid[8] as for the sequence shown in Scheme 1. Model studies in the stereocontrolled free radical allylation of dipeptides containing an α-phenylseleno β-amino acid related to **2** led to a single diastereomers (anti/syn >98:2) as shown in Scheme 2.[10, 11]

Although we could not obtain *X*-ray quality crystals to study the solid-state three-dimensional structure and conformation of the two tetrapeptides **5** and **6**, we secured convincing spectroscopic evidence regarding the relative disposition of the two peptidic chains. The solution conformation of peptides **5** and **6** were investigated in $CDCl_3$ with NOESY and ROESY techniques.[12] The qualitative yet clearly discernible long distance nOe's are indicative of the presence of a reverse turn in **5** and **6** (Figure 2). Molecular modeling

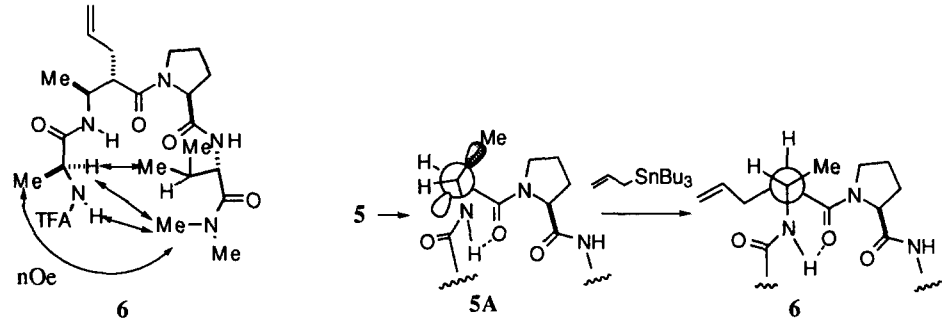
studies (Macromodel 4.5 version,[13] Amber force field) with nOe derived internuclear distances as constraints also suggest the prevalence of a β-turn-like conformation in **5** and **6**.

Scheme 2



The CD spectra of **5** and **6** measured in 1,1,2,2-tetrachloroethane showed in each case, a band at about 220 nm indicative of a reverse turn which supports the nOe data.[14,15] It is therefore possible that the β-amino acid unit in the tetrapeptide still adopts a conformation similar to that in the corresponding monomer (Figure 1). This is further supported by the unprecedented chemical conversion of **5** to **6** *via* a highly stereoselective free radical allylation reaction as in the esters and amides of α-phenylseleno-β-N-substituted amino acids.[8] The successful stereocontrolled α-C-allylation under mild free radical conditions within peptidic motifs is most likely due to approach of the allyl group from the less hindered side of a conformationally biased and H-bonded α-amino radical **5A**, as illustrated in Figure 2.

Figure 2



In summary, β-amino acids can be used as scaffolding devices for the design and assembly of secondary structures of peptides.[17] The localized 3-dimensional organization of peptidic subunits harboring such α-substituted β-amino acids may induce important conformational and functional properties that cannot be realized with their counterparts derived from α-amino acids. Further studies in these and related areas will be reported in due course.

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