Synthesis of the natural peptide product Fusahexin

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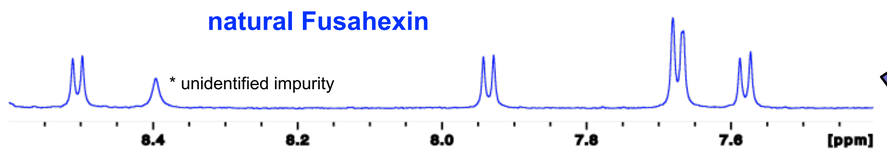
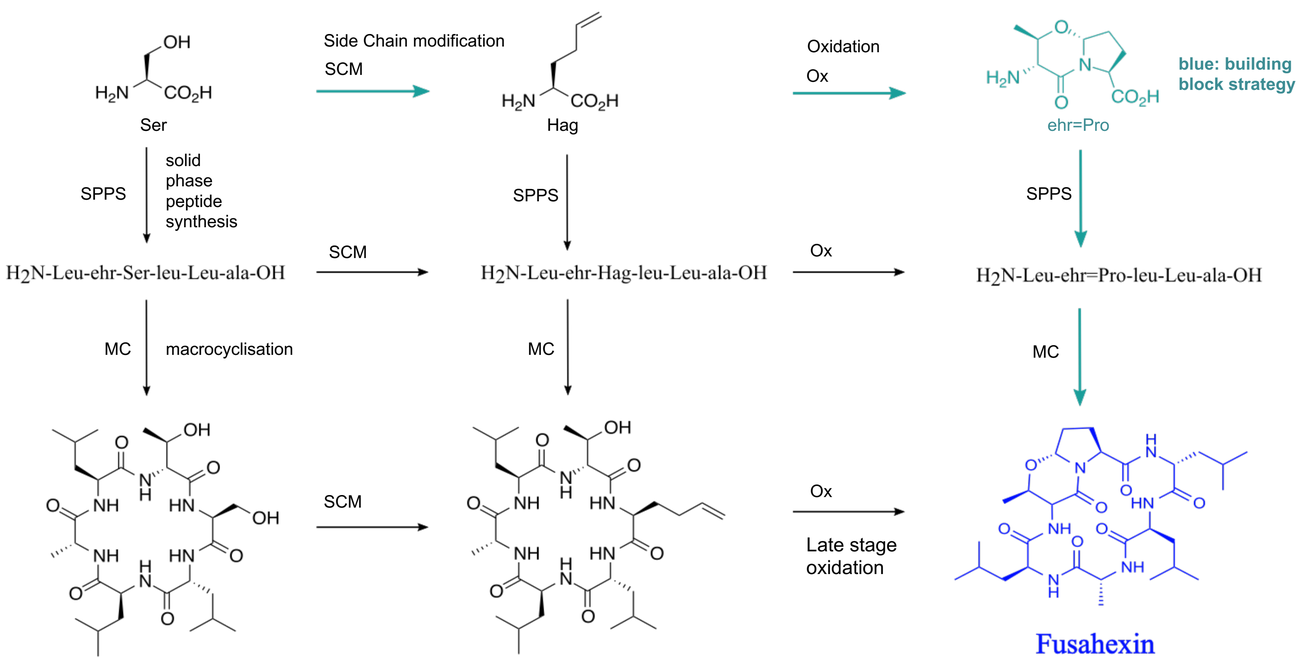
Introduction

The cyclic hexapeptide Fusahexin was isolated and characterized by WESTPHAL *et al*.[1] It contains a 6,5-bicyclic dipeptide which is similar to β-turn mimics proposed by BALDWIN and CLARIDGE[2] but in a different configuration because the D-*allo*-Thr (**ehr**) was not investigated as a β-turn mimic before. Substituents on the 6-membered ring can restrict the valerolactam puckering and identify efficient β-turn mimics.[3] The aim of this project is the synthesis of Fusahexin and the characterization of the conformational restriction exerted by ehr=Pro.

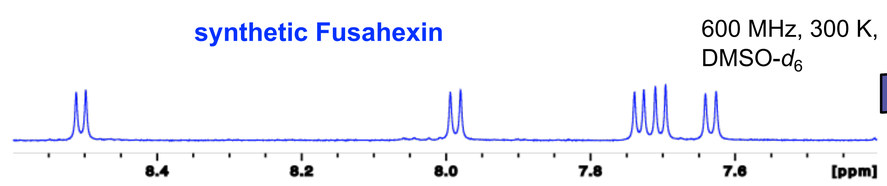
Expansion of the literature known spectra of the isolated Fusahexin from WESTPHAL, 600 MHz, 308 K, DMSO-d_6



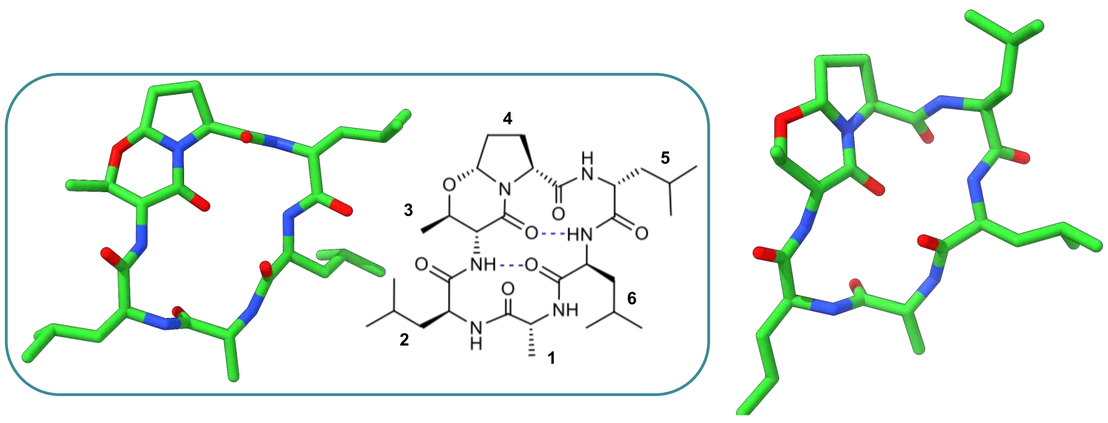
All possible strategies to the natural peptide product Fusahexin



Homoallylglycine (**Hag**) was synthesized according to the literature[5] and after cleavage of the Boc protecting group,was used in a dipeptide coupling with Fmoc-ehr-OH. The resulting dipeptide was used in a LEMIEUX-JOHNSON oxidation to yield the storable building block Fmoc-ehr=Pro-OH which was used in SPPS for the linear precursor peptide of Fusahexin.The tricyclic peptide natural product can be isolated and characterized after head to tail macrocyclisation. The synthetic Fusahexin was characterized with NMR spectroscopy and compared with the literature known data of the isolated one. Both spectra show nearly the same chemical shifts and coupling constants.



Modeling



Left: 3D model of Fusahexin *cyclo*[-Leu-ehr=Pro-leu-Leu-ala]

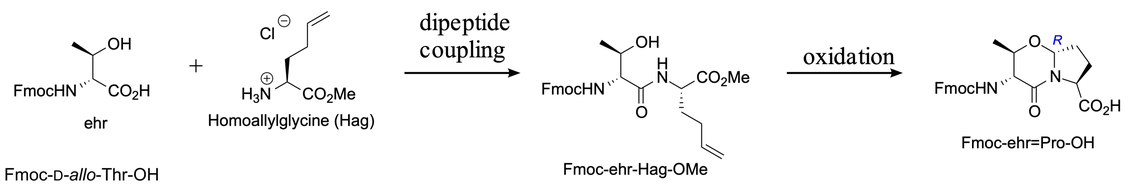
Middle: Hydrogen bonding pattern of Fusahexin

Right: 3D model of *cyclo*[-Leu-ehr=Pro-Leu-Leu-ala]

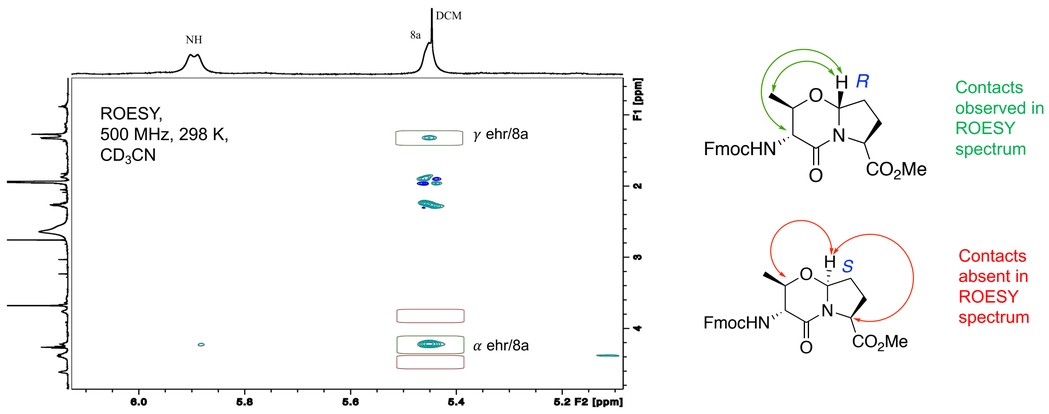
The alternating sequence of D- and L- configurated amino acids and the bicyclic ring system ehr=Pro determine the conformation of Fusahexin. NOEs, ^3J coupling constants and temperature coefficients identify the 3D structure in a computer model (left). The leu^{5}NH→Leu^{6}NH NOE is characteristic for a βII´-turn with ehr=Pro in the i to i+1 position (middle).

We inverted the configuration of amino acid 5 and characterized the cyclic hexapeptide *cyclo*[-Leu-ehr=Pro-Leu-Leu-ala] by NMR and modeling (above right). ehr=Pro remains in the long side although the β-turn flips to βI-conformation. Further peptides confirm the β-turn-inducing properties of ehr=Pro.

Building block synthesis



The building block Fmoc-ehr=Pro-OH was synthesized in 9 steps, starting from the natural amino acid L-serine. During the whole synthesis the N-terminus was protected with the Fmoc protection group which is necessary for the SPPS. The bridge head forms stereoselectively in *R* configuration independently of the peptide sequence. The stereoselective ring closure was proven by 2D-NMR like ROESY, where the bridge head proton shows a contact to the α-proton and to the γ-protons of ehr.



The independence of the bridge head configuration from the peptide sequence was further proven by the synthesis of *cyclo*[ehr-Hag-Phe-Gly-Gly-Gly] and the late-stage oxidation to *cyclo*[ehr=Pro-Phe-Gly-Gly-Gly], where the bridge head was again *R* configurated.[6]

Conclusion and Outlook

The building block Fmoc-ehr=Pro-OH can be synthesized in a 9 step synthesis with the LEMIEUX-JOHNSON oxidation as the key step. With this dipeptide the synthesis of the peptide natural product fusahexin was possible and 10 mg were isolated as a white solid. Because of the similiarity to the dipeptide which BALDWIN & CLARIDGE proposed as a β-turn mimic the Fmoc-ehr=Pro-OH building block is a possible β-turn mimic. In further studies the building bock is going to be used in more different peptide sequences to see if it is introducing a β-turn. Bioactivity studies are in progress.

References

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