2nd International Electronic Conference on Synthetic Organic Chemistry (ECSOC-2), http://www.mdpi.org/ecsoc/, September 1-30, 1998

[d0001]

Selective Epimerisation of a Fungal Cyclopeptolide via an 2-Amino-oxazole Intermediate - Conformational Consequences

B. Oberhauser*, B. Grohmann and H. Sperner

Novartis Forschungsinstitut GmbH
Brunner Str. 59
A-1235 Wien, Austria
Fax: ++43-1-86634 354

Abstract

The selective epimerisation of the cyclic depsiheptapeptide (cyclopeptolide HUN-7293) **1** at a single amino acid (Leu^5) was achieved in four steps. A *regio*-selective thionation at one of its six amide-bonds provided the corresponding thioamide **2**, which upon *S*-benzylation followed by an unusual silver-promoted *intra*-molecular cyclisation, was converted to the 2-aminooxazole derivative **4**. Re-opening of the oxazole ring by acid hydrolysis regenerated the cyclopeptolide, but preferentially with inverted stereochemistry at Leu^5 **5**. The influence of this inversion on the backbone conformation is discussed.

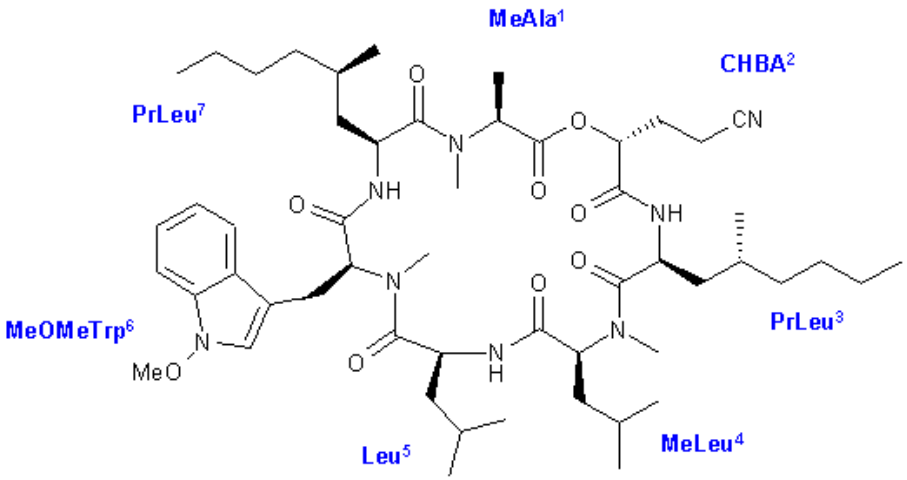


Fig. 1: Structure of the fungal metabolite HUN-7293 (**1**)

Introduction

The cyclic depsiheptapeptide HUN-7293 **1**, a fungal fermentation product, potently inhibits VCAM-1 expression on activated endothelial cells[1]. In addition to *N*-methylated amino acids MeAla and MeLeu, it contains the uncommon constituents D-4-cyano-2-hydroxybutyric acid (CHAB), *N*1'-methoxy-*N*-methyltryptophan (MeOMeTrp) and d-propylleucine (PrLeu). Its constitution (Fig. 1) and 3-dimensional structure has been determined by 2D-NMR methods and X-ray diffraction analysis (Fig. 2)[2]. The 3-dimensional structure is characterised by two methylated *cis*-amide bonds and two trans-annular hydrogen bonds resulting in a tightly packed backbone conformation. This allows a close hydrophobic contact of the sidechains of hydroxy acid^2 (CHBA^2), amino acid^6 (MeOMeTrp^6) and amino acid^5 (Leu^5) as indicated by a strong magnetic shielding effect of the aromatic ring on the Leu^5 b-protons (Leu^5 b-H: d = -0.36 ppm).

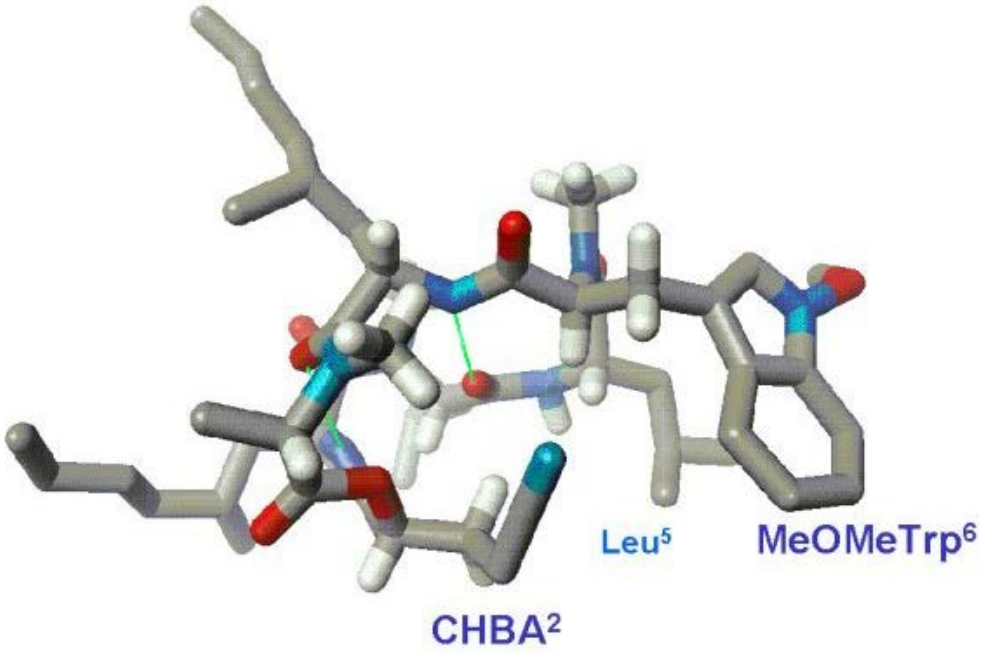
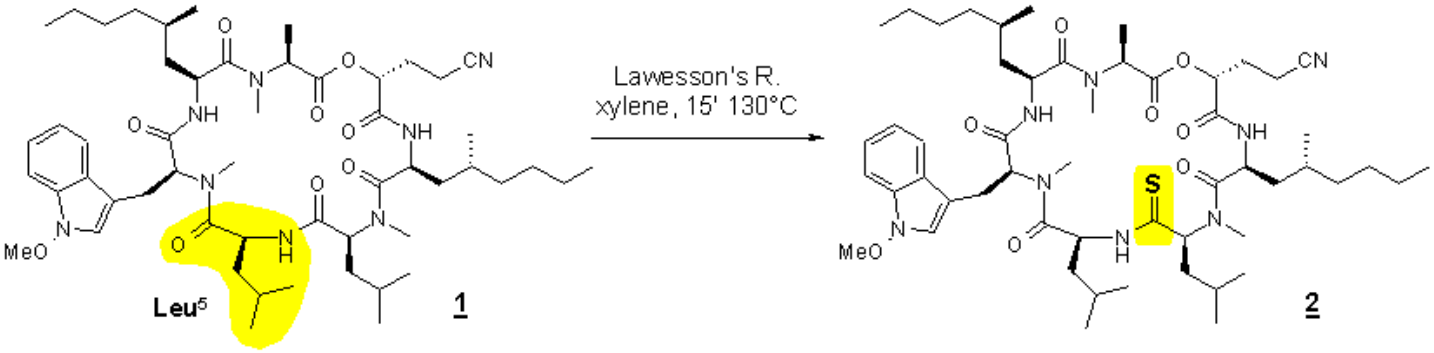


Fig. 2: X-ray crystal structure of HUN-7293, 3D: msv-file or mol-file

Results and Discussion

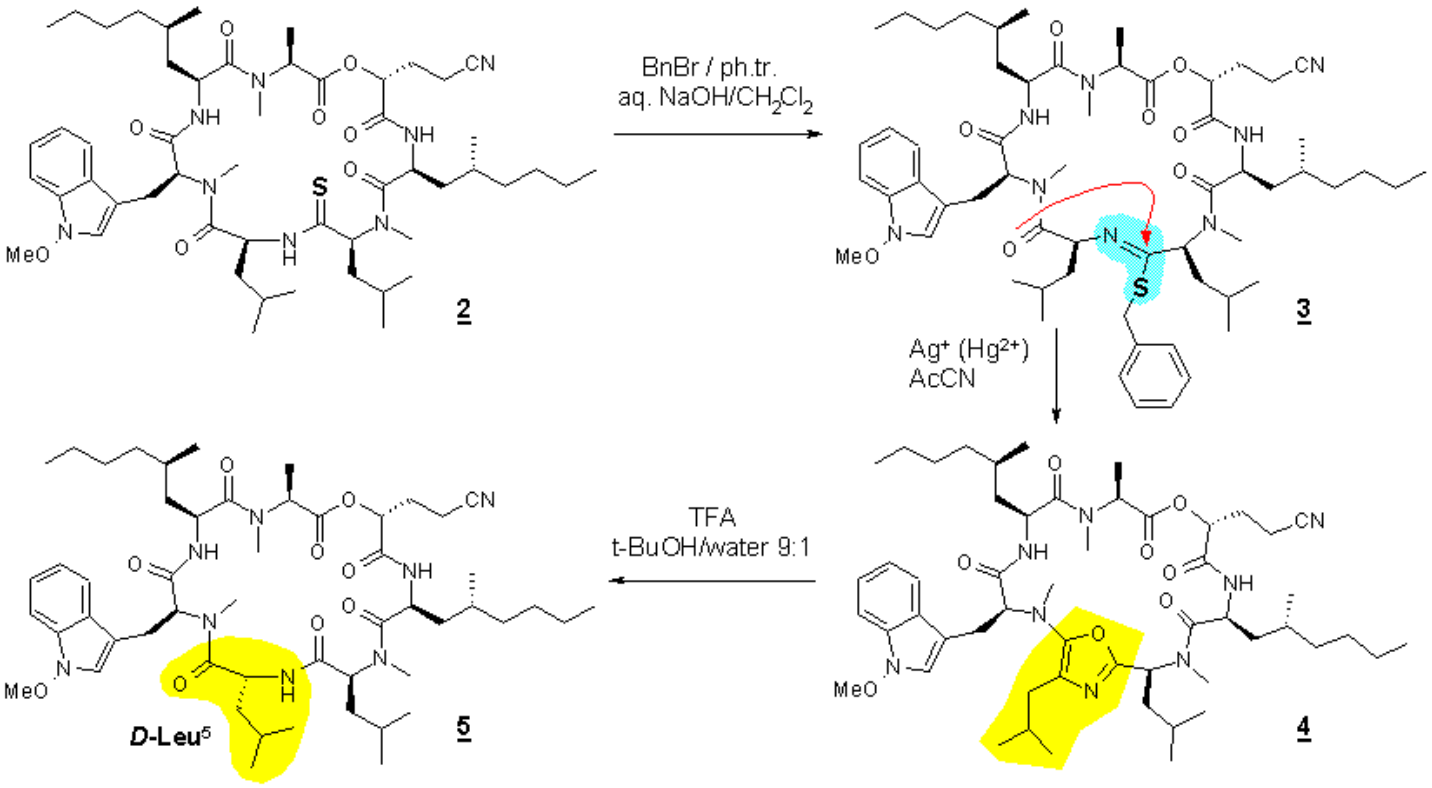
Highly *regio*-selective thionation of the most reactive amide carbonyl between MeLeu[4] and Leu[5] (the only primary amide NH-group, which is not involved in *intra*-molecular hydrogen bonding) was easily achieved with 0.5 equivalents Lawesson's reagent in xylene at 130°C (Scheme 1). As by-products, the double thioamide (additional thionation at MeOMeTrp[6]-PrLeu[7]) and a triple thioamide (thionation at all primary amides) could be isolated.



Scheme 1: Selective transformation of a single amide bond with Lawesson's reagent

Alkylation of the mono-thioamide **2** with benzyl bromide under phase-transfer conditions (aq. NaOH, dichloromethane, ultrasound) resulted in a quantitative transformation into the S-benzyl thioamidate **3**. Treatment of this thioamidate with silver- or mercury salts in acetonitrile destroyed the chirality at Leu[5] by the unusual formation of a peptide-bridged 2-aminooxazole **4**. In contrast to other oxazole-containing peptidic natural products, no sidechain atoms are involved in the ring-closure of this rather exotic type of a rigid dipeptide mimetic[5-9]. The formation of the heterocycle can be explained via the activation of the thioamidate by co-ordination of the heavy metal at the sulphur (i.e. formation of an excellent leaving group). Due to the lack of an external nucleophile, this forces the molecule to undergo an *intra*-molecular cyclisation

involving the adjacent carbonyl group of Leu (Scheme 2).



Scheme 2: 2-Aminooxazole as achiral dipeptide-mimetic

Utilising the sensitivity of 2-aminooxazoles towards acid hydrolysis, the two masked amid bonds could easily be regenerated by treatment with TFA in aqueous t-butanol. This led to the formation of the D-Leu^5 epimer **5** together with HUN-7293 itself in a 2:1 ratio. The predominance of one diastereomer probably reflects the rigid structure of the peptide-bridged oxazole, as also indicated by a single NMR-conformation. The stereochemistry of the predominant diastereomer **5** was established by 2D-NMR methods and independently proven via partial synthesis.

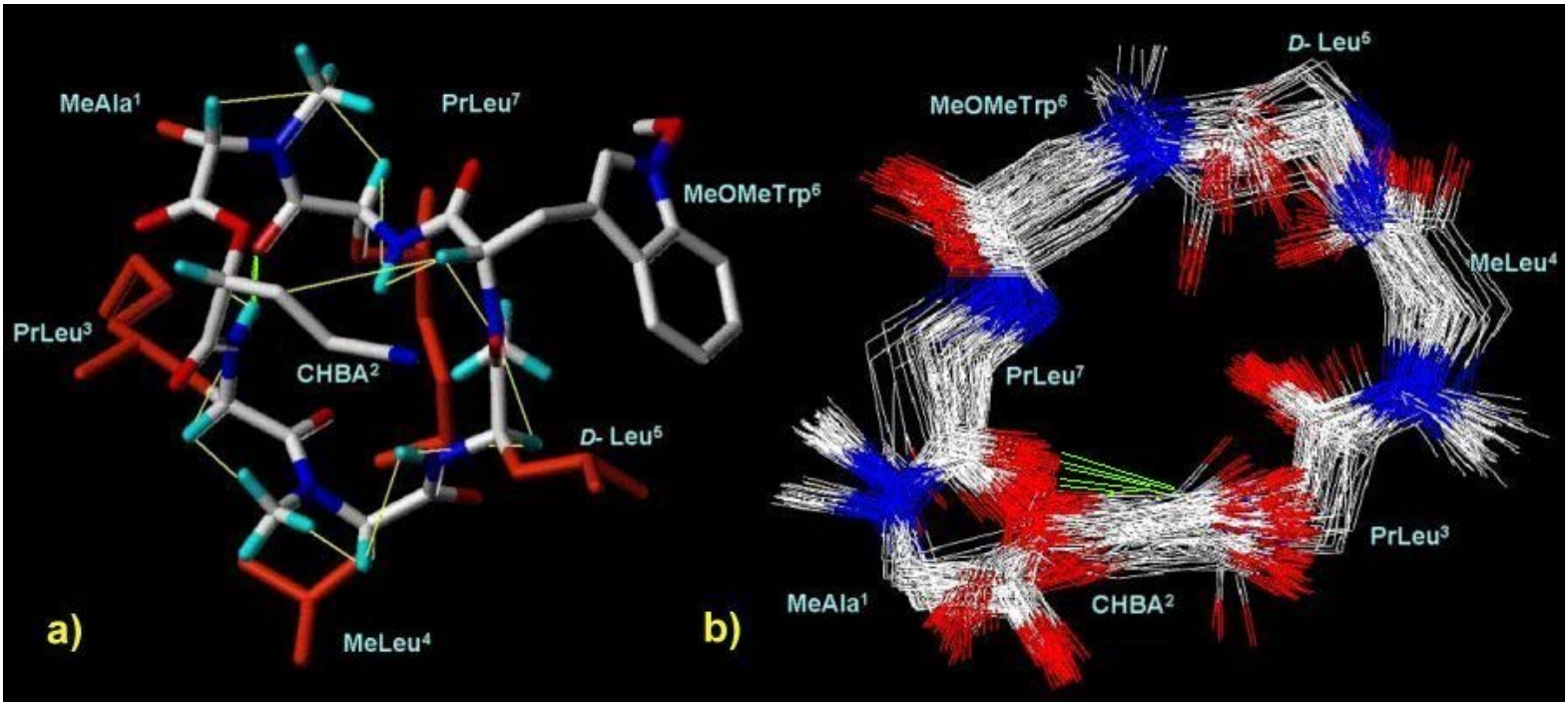


Figure 3:
a) Observed ROESY distance constraints (yellow) in an low energy conformation with a possible hydrogen bridge (green) PrLeu^3 NH - CO PrLeu^7 (3D: msv-file or mol-file)
b) Ensemble of 100 energy-minimised backbone structures compatible with the distance constraints (simulated annealing[10]); Due to the lack of distance constraints at the lactone moiety, two equally populated conformations at MeAla^1 are visible.

In contrast to HUN-7293 **1**, the 5-*epi* derivative **5** adopts a single conformation in $CDCl_3$ solution. Although dramatic changes in the conformation could be expected due to the epimerisation, 5-*epi* HUN-7293 shares some characteristic NMR-features with the parent compound:

- high field shift of the Leu^5 NH
- ROESY cross-peaks of CHBA^2-MeOMeTrp^6 indicating a "back-folding" of the CN-sidechain onto the ring (Fig. 3a)

The main difference was the absence of the extreme high-field shift of one Leu^5 b-proton. A simulated annealing analysis was performed utilising the available set of backbone ROESY distance-constrains (Fig. 3b). The resulting structure was compatible with the observed $^3J_{NH\text{-}Ha}$ coupling constants, whereas coupling constants of sidechain protons $^3J_{Ha\text{-}Hb}$ could only be explained by a mixed population of sidechain conformers.

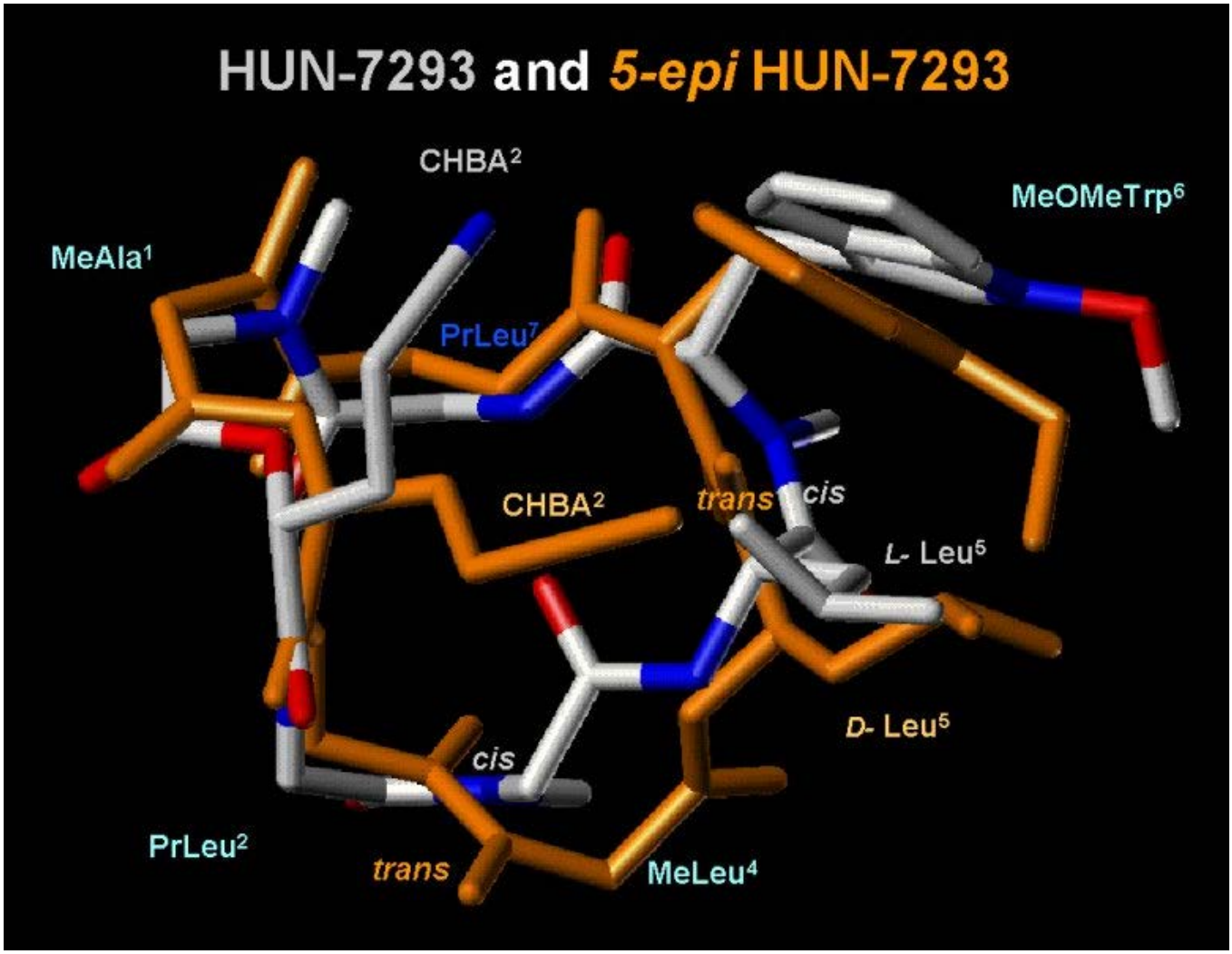


Figure 4: Overlay of the natural product HUN-7293 (X-ray structure, grey) and its Leu^5-epimer (NMR structure, orange; 3D: msv-file or mol-file)

Conclusions

Because of the known sensitivity of the cyclopeptolides 3D conformation towards even minor structural changes, we were surprised to see that the 5-*epi* HUN-7293 adopts an overall similar conformation. The different chirality of Leu^5 is obviously compensated by the flipping of both *cis*-amide bonds to the *trans*-conformation still allowing at least one trans-annular hydrogen bond and the hydrophobic interaction of the CHBA^2, MeOMeTrp^6 and Leu^5 (Fig. 4).

References

1. Foster, C.A., Dreyfuss, M., Mandak, B., Meingassner, J.G., Naegeli, H.U., Nussbaumer, A., Oberer, L., Scheel, G., Swoboda, E.-M. (1994) **Pharmacological modulation of endothelial cell-associated adhesion molecule expression: Implications for future treatment of dermatological diseases.** *J. Dermatol.* **21**, 847-854.
2. Hommel, U., Weber, H.-P., Oberer, L., Naegeli, H.U., Oberhauser, B., Foster, C.A. (1995) **The 3D-structure of a natural inhibitor of cell adhesion molecule expression.** *FEBS Letters* **379**, 69-73.
3. Seebach, D., Ko, S.Y., Kessler, H., Köck, M., Reggelin, M., Schmieder, P., Walkinshaw, M.D., Bölsterli, J.J., Bevec, D. (1991) **Thiocyclosporines: preparation, solution and crystal structure, and immunosuppressive activity.** *Helv. Chim. Acta* **74**, 1953-1990.
4. Eberle, M.K., Jutzi-Eme, A.-M., Nuninger, F. (1994) **Cyclosporin A: Regioselective ring opening and fragmentation reactions via Thioamides. A route to semisynthetic cyclosporins.** *J. Org. Chem. 59*, 7249-7258.
5. Lipshutz, B.H., Huff, B.E., McCarthy, K.E., Mukarram, S.M.J., Siahaan, T.J., Vaccaro, W.D., Webb, H., Falick, A.M..

Miller, T.A. (1990) **Oxazolophanes as masked cyclopeptide alkaloid equivalents: cyclic peptide chemistry without peptide couplings.** *J. Am. Chem. Soc.* **112**, 7032-41.
6. Lipshutz, B.H., Hungate, R.W., McCarthy, K.E. (1986) **Chiral induction in originally racemic amino acids via 5-acyl and 5-acyloxyaminooxazoles.** *Isr. J. Chem.* **27**, 49-55.
7. Lipshutz, B.H., Hungate, R.W., McCarthy, K.E. (1983) **Heterocycles in synthesis. Dipeptides via unmasking of 5-acyl- and 5-acyloxyaminooxazoles.** *Tetrahedron Lett.* **24**, 5155-8.
8. Lipshutz, B.H., Hungate, R.W., McCarthy, K.E. (1983) **Heterocycles as masked diamide/dipeptide equivalents. Formation and reactions of substituted 5-(acylamino)oxazoles as intermediates en route to the cyclopeptide alkaloids**. *J. Am. Chem. Soc.* **105**, 7703-13.
9. Engelfried, C., Ludwig, K., Jaenichen, H., Koenig, W.A. (1979**) Reactions of amino acids and peptides. II. Preparation of 2-aminooxazole derivatives from peptide amides with hexafluoroacetic anhydride.** *Liebigs Ann. Chem.* **1979,** 973-85.
10. simulated annealing: Sybyl, 100 cycles 700K 1000fs, 200K 1000fs, energy minimisation: Tripos forcefield, Gasteiger charges, no violations of distance constraints greater than 20 pm (ROESY)

Comments

During 1-30 September 1998, all comments on this poster should be sent by e-mail to ecsoc@listserv.arizona.edu with d0001 as the message subject of your e-mail. After the conference, please send all the comments and reprints requests to the author.
