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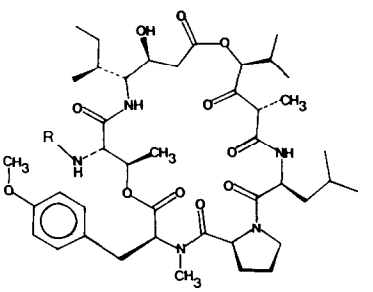
TOTAL SYNTHESIS OF THE DIDEMNINS - 1. SYNTHESIS OF THE PEPTOLIDE RING[1]

U.Schmidt*, M.Kroner and H.Griesser
Institut für Organische Chemie, Biochemie und Isotopenforschung der Universität Stuttgart,
Pfaffenwaldring 55, D-7000 Stuttgart 80, Bundesrepublik Deutschland

Abstract - The 23-membered peptolide ring of the didemnins is formed in a two phase cyclization of a linear ω-amino-pentafluorophenyl ester in 70% yield without high dilution. (2RS,4S)-2,5-dimethyl-4-hydroxy-3-oxohexanoic acid (Hip) and (3S,4R,5S)-isostatine are synthesized by acylations of trimethylsilyl malonates.

The cyclic depsipeptides didemnin A-C were isolated from the tunicate Tridemnum solidum by K.L.Rinehart et al. in 1981[2]. The biological properties are dependent on the structure of the side chain; only didemnin B exhibits strong cancerostatic and immunosuppressive activities and has been entered into clinical trials. The originally reported suggestion for the structure has been revised several times: 1) The configuration of N-methylleucine in the side chain has been identified as R[3]; 2) M.M.Joullie et al. established the 2S,4S-configuration for the hydroxyisovalerylpropionic acid (Hip)[4]; 3) By means of NMR investigations, B.Castro et al. demonstrated that (3S,4R,5S)-isostatine and not statine is a ring building block[5]. A reliable elucidation of the structure and configuration was recently achieved by X-ray crystallography[6] and total synthesis[7] (ring closure at the nitrogen atom of isostatine in 18% yield). The synthesis of didemnin A analog containing statine as a building block has been reported by T.Shioiri et al.[8] (ring closure at the nitrogen atom of N,O-dimethyltyrosine).



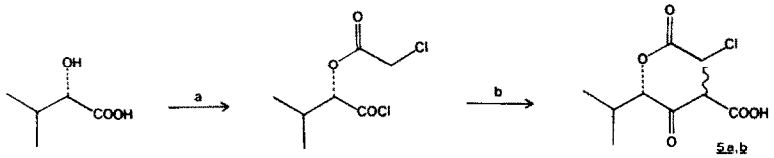
1 didemnin A, R = H-R-MeLeu
2 didemnin B, R = H-Lac-Pro-R-MeLeu
3 didemnin C, R = H-Lac-R-MeLeu
4a R = Z

After having completed the synthesis of the didemnin analog containing statine we learned of the corrected didemnin structure[9]. We now report on the construction of the didemnin ring with the Z-protecting group. Substitution of the latter with amino acids or lactylamino acids makes possible the formation of all naturally occurring didemnins and also the structural variations in the side chain that determine the biological properties. Our methods for the construction of the linear substrate and for the ring closure (at the nitrogen atom of proline in 70% yield) are completely different from the methods used by Shioiri and Rinehart. A simple method for the synthesis of (2RS,4S)-2,5-dimethyl-4-hydroxy-3-oxohexanoic acid ((2RS,4S)-Hip) is described. The preparation of (3S,4R,5S)-4-amino-3-hydroxy-5-methylheptanoic acid ((3S,4R,5S)-isostatine) is reported for the first time. - For the construction of difficulty accessible peptide bonds the coupling reagent 1-phenyl-2-tetrazoline-5-thione/t-butyl isocyanide[10] and the thiol esters of 4,6-dimethyl-2-thiopyridone-3-carbonitrile[11] were used successfully. - Because the

amino group of the isostatine derivative and the methylamino group of tyrosine have proved not to be easily attacked in our experiments we do not consider these positions to be especially suitable for the ring closure.

Preparation of (2RS,4S)-2,5-Dimethyl-4-hydroxy-3-oxohexanoic Acid ((2RS,4S)-Hip) and (3S,4R,5S)-4-Amino-3-hydroxy-5-methylheptanoic Acid ((3S,4R,5S)-Isostatine)

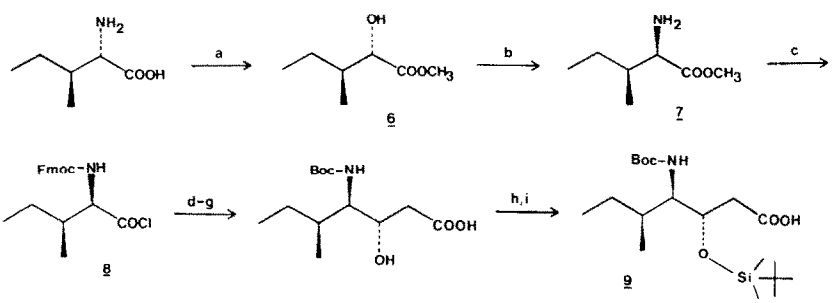
The skeletons of (2RS,4S)-Hip and isostatine were built up with the help of a β-keto ester synthesis developed previously by us using trimethylsilyl malonate[12]. (S)-2-(Chloroacetoxy)-isovaleryl chloride (from (S)-valine) reacted smoothly with the lithium enolate of bis-(trimethylsilyl)methylmalonate. Subsequent hydrolysis gave a mixture of the two diastereomeric β-keto acids (Scheme 1).



Scheme 1

a) Cl-CH_2-COCl/pyridine/CH_2Cl_2/20°C/15 h//$SOCl_2$/50°C/3 h/79%; b) MeC(Li)(CO_2SiMe_3)$_2$/THF/-60° → 20°C/15 h/1-N-$KHSO_4$/84%.

The expensive (2R,3S)-alloisoleucine was obtained from commercially available (2S,3S)-isoleucine via the corresponding α-hydroxycarboxylic acid. Fmoc-(2R,3S)-alloisoleucine chloride was then allowed to react with the lithium enolate of methyl trimethylsilyl malonate to furnish the β-keto ester, reduction of which resulted in a diastereomeric mixture [3S,4R,5S/3R,4R,5S (77:23)]. The 3S,4R,5S-diastereomer could be purified by recrystallization. After acidic hydrolysis the amino group was deprotected in aqueous alkaline solution and the corresponding Boc derivative was prepared (Scheme 2).

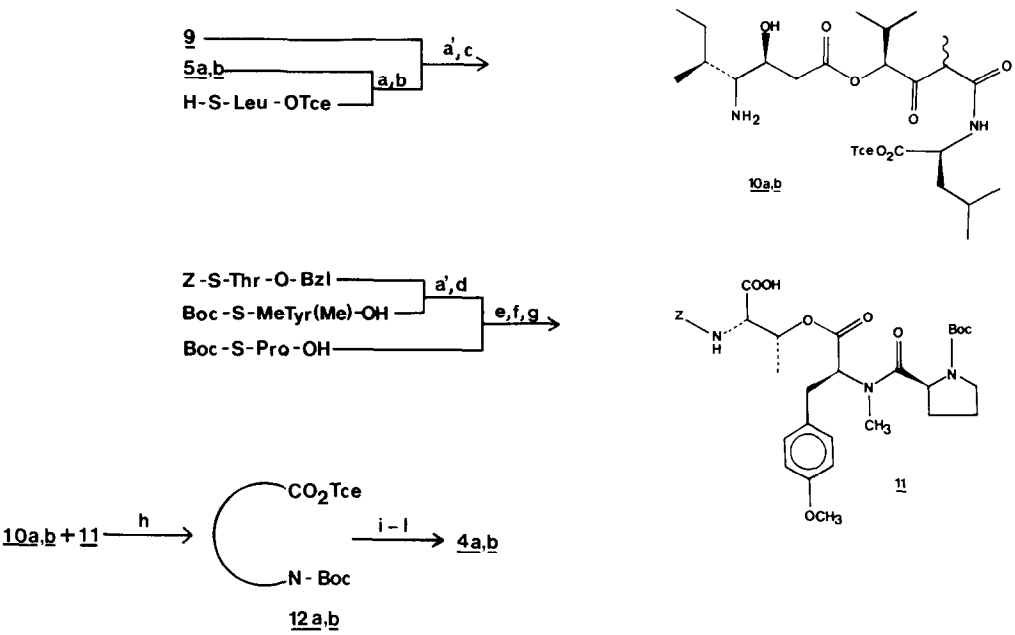


Scheme 2

a) i-C_5H_9ONO/CH_3COOH//CH_3OH/HCl; b) diethyl azodicarboxylate/PPh_3/HN_3/toluene/0°C/2 h//H_2/Pd/HCl/60%; c) FmocCl/pyridine/CH_2Cl_2/0°C/1 h/87%//HCl/H_2O/dioxane/100°C/2 d/90%//$SOCl_2$/CH_2Cl_2/40°C/2 h/91%; d) MeO_2C-CH^-Li-CO_2SiMe_3/THF/-70° → 20°C/1 h/H_2O/quant.; e) $NaBH_3CN$/CH_3COOH/THF/20°C/20 h/90%//separation of diastereomers by recrystallization from ethyl acetate/petroleum ether; f) HCl/H_2O/dioxane/100°C/5 h/93%; g) 1-N-NaOH/40°C/2 h/Boc_2O/dioxane/20°C/24 h/96%; h) TBDMS-Cl/imidazole/DMF/20°C/15 h/quant.; i) 1-N-NaOH/ dioxane/20°C/0.1 h/quant.

Construction of the Linear Peptolide and Ring Closure

The linear peptolide was built up using known methods. For the acylation of N,O-dimethyltyrosine, the activation method using phenyltetrazolinethione/isocyanide developed earlier by us[10] proved to be useful. - The following activation methods were checked for the difficult acylation of the isostatine derivative (10+11; yields in brackets): pentafluorophenyl ester (33%); mixed anhydride from pivaloyl chloride (21%; mainly wrong opening); $(C_6H_5O)_2P(O)N_3$ (45%); thiol ester of 4,6-dimethyl-2-thiopyridone-3-carbonitrile[11] (55%). Therefore the last proved to be the reagent of choice. All intermediates containing Hip were diastereoisomeric mixtures. Some of them have separated f.e. 12. - The ring closure was achieved using the pentafluorophenyl ester in a two phase system ($H_2O/CHCl_3$)[13]. By this way a nearly pure product was isolated. For spectroscopic investigations it was purified easily using MPLC (ethyl acetate/petroleum ether (6/4); 70% yield of chromatographically purified cyclopeptolides). The reaction product consists of the two diastereoisomers with respect to the configuration in the 2-position of the Hip unit (50/20 probably S/R). The isolated R-diastereoisomer 4b[14] rearranges slowly, in solution and by acid or base catalysis, to the more stable isomer as does the (2-epi-Hip)-didemnin A (Scheme 3). - All new compounds gave satisfactory ^{1}H-NMR and MS data[15].



Scheme 3

a) DCC/CH_2Cl_2/0°C/2 h/90%; a') DCC/DMAP/CH_2Cl_2/-20°C/15 h/90%; b) $(CH_2)_5N$-CS-NH_2/dioxane/NEt_3/50°C/3.5 h/85%; c) HF/CH_3CN/20°C/15 h/quant.;d) HCl-dioxane/20°C/1 h/quant.; e) phenyltetrazolinethione/t-butylisonitrile/CH_2Cl_2/20°C/15 h/75%; f) H_2/Pd/1 eq. HCl/ethanol/20°C/2 h/100%; g) Z-Cl/$NaHCO_3$/H_2O/1 h/70%; h) 4,6-dimethyl-2-thiopyridone-3-carbonitrile/Ph_3P/CH_2Cl_2/-20°C/1 h/11/20°C/15 h/55%; i) Zn/CH_3COOH/H_2O/20°C/15 h/quant.; j) DCC/C_6F_5OH/-20°C/15 h; k) $Me_3SiOSO_2CF_3$/CH_2Cl_2/20°C/3 h; l) $NaHCO_3$/H_2O/$CHCl_3$/20°C/30 min; j,k,l: 69%.

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Notes and References

1 Amino Acids and Peptides-65. Cyclopeptides-12. Part 64: U.Schmidt, W.Siegel, K.Mundinger, Tetrahedron Lett., in press; Part 11: U.Schmidt, D.Weller, ibid. 27, 3495.

2 K.L.Rinehart,Jr. J.B.Gloer, J.C.Cook,Jr., J.Am.Chem.Soc. 103, 1857 (1981).

3 K.L.Rinehart,Jr., Anal.Chem.Symp.Ser. 24, 119 (1985).

4 W.R.Ewing, K.L.Bhat, M.M.Joullie, Tetrahedron 42, 5863 (1986).

5 B.Castro, 10th American Peptide Symposium, St.Louis, MO (1987); Paper P-292. We thank Prof. Castro for personal information (October 1987).

6 D.v.d.Helm, 14th Meeting Intern. Congress of Crystallography, Perth, Australia (1987); Paper C-52/03.2-8.

7 K.L.Rinehart,Jr., V.Kishore, S.Nagarajan, R.J.Lake, J.B.Gloer, F.A.Bozich, K.-M.Li, R.E.Maleczka,Jr., W.L.Todsen, M.H.G.Munro, D.W.Sullins, R.Sakai, J.Am.Chem.Soc. 109, 6846 (1987).

8 T.Shioiri, Japan Symposium on Peptide Chemistry, Kobe, Japan (1987); P 111.

9 We thank Prof.Rinehart for personal information (September 27, 1987).

10 U.Schmidt, M.Dietsche, Angew.Chem. 94, 145 (1982); Angew.Chem.Int.Ed.Engl. 21, 143 (1982).

11 U.Schmidt, K.Schefenacker, Liebigs Ann.Chem. 1985, 1254; U.Schmidt, B.Potzolli, ibid. 1987, 935.

12 U.Schmidt, M.Schwochau, Tetrahedron Lett. 10, 875 (1967); idem, Monatsh.Chem. 98, 1492 (1967).

13 U.Schmidt, A.Lieberknecht, H.Griesser, J.Talbiersky, J.Org.Chem. 47, 3261 (1982); U.Schmidt, R.Utz, A.Lieberknecht, H.Griesser, B.Potzolli, J.Bahr, K.Wagner, P.Fischer, Synthesis 1987, 236.

14 The S-diastereoisomer 4a moves faster on normal phase MPLC (ethyl acetate/petroleum ether (1/1)).

15 4a: mp 226-227°C, FAB-MS $(MH)^+$ = 950 (glycerol), $[\alpha]_D^{20}$ = -166.5 (c = 0.76,$CHCl_3$); ^1H-NMR-data (250 MHz, $CDCl_3$) δ = 7.57 (d,J=8.9Hz,1H), 7.35-7.23 (6H), 7.09 (d, J= 8.5Hz,2H), 6.85 (d,J=8.5Hz,2H), 5.50 (d,J=10Hz,1H), 5.22 (d,J=3.4Hz,1H), 5.07 (d,d, J=12.5Hz,J'=22Hz,2H), 5.04 (m,1H), 4.72 (m,2H), 4.59 (m,1H), 4.11 (d,t,J=4.3Hz, J'= 9.6Hz,1H), 4.01 (d,d,J=9.5Hz,J'=19.5Hz, 1H), 3.92 (q,J=7Hz,1H), 3.80 (s,3H), 3.72 (m, 1H), 3.63-3.56 (m,2H), 3.38 (d,d,J=4.2Hz, J'=14.2Hz,1H), 3.18 (d,d,J=10.6Hz,J'=14.2Hz, 1H), 3.13 (d,J=17.2Hz,1H), 2.97 (m,1H), 2.55 (s,3H), 2.53 (d,d,J=10.5Hz, J'=17.2Hz,1H), 2.34 (m,1H), 2.25-1.10 (10H), 1.27 (d,J=7.0Hz,3H), 1.24 (d,J=7.8Hz,3H), 1.00-0.80 (18H). 4b: 7.36-7.25 (6H), 7.07 (d,J=8.5Hz,2H), 6.83 (d,J=8.5Hz,2H), 6.49 (d,J=8.3Hz,1H), 5.59 (d,J=6.0Hz,1H), 5.33 (d,J=5.1Hz,1H), 5.14 (m,1H), 5.03 (q,J=12.2Hz,2H), 4.72 (m,1H), 4.60 (d,d,J=4.1Hz,J'=7.9Hz,1H), 4.39 (s,br,1H), 4.19 (q,J=7.1Hz,1H), 4.18-4.08 (m,2H), 3.78 (s,3H), 3.76 (d,d,J=1.9Hz,J'=4.1Hz,1H), 3.64-3.49 (m,2H), 3.35 (d,d,J=4.2Hz,J'= 14.2Hz,1H), 3.16 (d,d,J=10.6Hz,J'=14.2Hz,1H), 2.94 (d,d,J=4.9Hz,J'=7.4Hz,1H), 2.88 (s,br,1H), 2.50 (m,1H), 2.63 (s,3H), 2.37 (m,1H), 2.28-1.10 (10H), 1.27 (d,J=4.1Hz, 3H), 1.18 (d,J=6.4Hz,3H), 1.00-0.78 (18H).

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