Pergamon

Tetrahedron Vol. 51, No. 4, pp. 1187-1196, 1995
Elsevier Science Ltd
Printed in Great Britain
0040-4020/95 $9.50+0.00

0040-4020(94)01014-5

Enantioselective Synthesis of *S*-*o*-Carboranylalanine via Methylated Bislactim Ethers of 2,5-Diketopiperazines

Wilhelm Karnbrock, Hans-Jürgen Musiol and Luis Moroder*

Max-Planck Institut für Biochemie, Am Klopferspitz 18a, 82152 Martinsried, Germany

Abstract: A new synthesis of *S*-*o*-carboranylalanine by the Schöllkopf-Hartwig procedure is described. The metalated bislactim ether of cyclo-(D-Val-Gly) reacted with propargylbromide to yield with 52 % de the (5*S*)-propargyl adduct. Its boronation with 6,9-bis(acetonitrile)-decaborane proceeded in satisfactory yields despite the steric hindrance of the bislactim ether, but surprisingly thermal isomerization was found to occur to some extent at the chiral C-5 atom. Although all attempts to separate the diastereomeric 12:1 mixture failed so far, this synthetic approach represents a significant improvement because of the relatively facile access to *S*-*o*-carboranylalanine in high enantiomeric purity.

Key words: *S*-*o*-carboranylalanine, enantioselective synthesis, bislactim ether, boron neutron capture therapy.

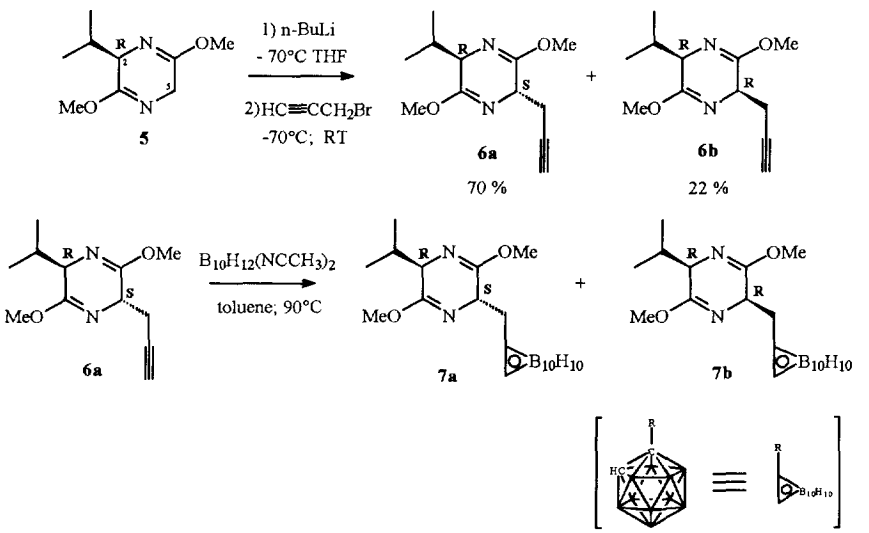
o-Carboranylalanine is a potential phenylalanine mimetic since the dimensions of the carborane cage (1,2-$C_2B_{10}H_{12}$) are only slightly larger than the space occupied by a benzene ring rotating about its C(1)-C(4) axis[1]. *o*-Carboranylalanine containing analogues of enkephalin[2-4] and angiotensin II[5] were found to retain their bioactivities despite the function of the replaced phenylalanine moieties. Vice versa, substitution of the planar phenylalanine side chain for the pseudoaromatic highly lipophilic and electronegative carborane moiety in bradykinin and substance p[3] led to almost complete loss of hormonal activity[6]. Nevertheless, this boron-rich amino acid could be an interesting tool in Boron Neutron Capture Therapy (BNCT)[7,8], particularly for selective receptor-mediated targeting of *o*-carboranylalanine-peptide hormone analogues to cancerous cells.

In view of the promising applications of this boron-rich amino acid we have focussed on possible improvements of the synthesis of enantiopure *S*-*o*-carboranylalanine. Of the two syntheses proposed by Schwyzer and coworkers[1,2] the boronation of Boc-Pra-OMe with the 6,9-bis(acetonitrile)-decaborane was found to be feasible despite the presence of an acidic proton of the carboxamide group, which probably was shielded by the bulky Boc-moiety. All our attempts to improve this multistep synthesis failed and the yields of the final product were lower than those reported[2] but in agreement with the findings of Andersson et al.[10]. Of the new asymmetric syntheses of *S*- or *R*-o-carboranylalanine reported so far, alkylation of 2-*tert*-butyl-1-*tert*-butyloxycarbonyl-3-methyl-4-imidazolidinone with propargylbromide according to the procedure of Fitzi and Seebach[11] followed by boronation[10] requires drastic acid conditions in the hydrolytic step[15]. Alkylation of N-(diphenyl-methylene)aminoacetonitrile with propargyl bromide and addition of decaborane followed by acid hydrolysis has been proposed to be an efficient new procedure[12]. However, only a racemic mixture of the amino acid is obtained by this method. Finally a multistep new enantioselective synthesis of o-carboranylalanine has been presented by Radel and Kahl[13], but without experimental details. By analogy with

the synthesis reported by Wyzlic and Soloway[12], we have prepared the N-(diphenylmethylene)-*S*-propargylglycine methyl ester **(1)** by transimination of benzophenone imine with H-Pra-OMe·HCl according to O'Donnell and Polt[14]. Subsequent boronation with decaborane in acetonitrile/benzene[9] or with 6,9-bis(acetonitrile)-decaborane in toluene was found to give mainly N-(diphenylmethyl)-*S*-propargylglycine methyl ester **(2)** and N-(diphenylmethyl)-*S*-*o*-carboranylalanine methyl ester besides traces of the desired compound. Under both reaction conditions, reduction of the C-N double bond took place in contrast to what has been reported by Wyzlic and Soloway[12]. Yet this is in agreement with previous findings that reduction of aldehydes occurs during boronation of acetylenic compounds[16]. In order to examine the effect of steric hindrance of the diphenylmethylene moiety on the addition reaction, the benzaldehyde Schiff base **3** was used, but again N-(benzyl)-*S*-*o*-carboranylalanine methyl ester **(4)** was formed. Moreover, the rate of boronation could not be enhanced by this approach, thus excluding this synthetic route as suitable for the preparation of the target compound.

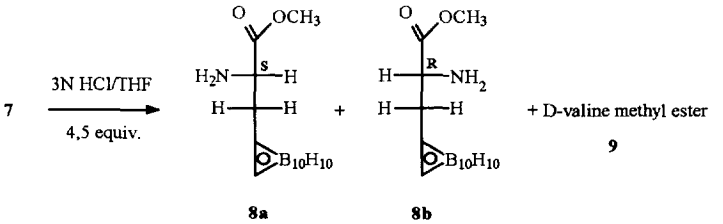
We then applied the method of Schöllkopf-Hartwig[17,18] as shown in Scheme 1. Alkylation of the metalated bislactim ether of cyclo-(D-Val-Gly) **5** took course in 52 % de. The low degree of diastereoselectivity of the alkylation step with linear alkyl halides is known[17,18] and has been attributed to an uncomplete shielding of the diastereotopic side of the planar dihydropyrazine anion by the isopropyl group. Subsequent reaction of **6a** with 6,9-bis(acetonitrile)-decaborane afforded the desired *o*-carboranyl derivative as a diastereomeric mixture of **7a** and **7b** (12:1), *i.e.* 84 % de as determined by ^{1}H NMR analysis.

Scheme 1:



The configuration at the chiral atoms C-2 and C-5 was determined after hydrolysis of 7 at the stage of the amino acid methyl esters. For the *o*-carboranylalanine methyl ester the chiral shift reagent tris[3(2,2,2-trifluoro-1-hydroxyethylidene)-d-camphorato]-europium ($Eu(TFC)_3$) confirmed partial isomerization, whereas valine methyl ester was treated with pentafluoropropionic acid anhydride, and then analysed by gas chromatography on a Chirasil-Val glas capillary column[19]. A value of 98.5 % D-Val and 1.5% L-Val was determined by this procedure.

Scheme 2:



It has been reported by Zeiß[20] that thermal isomerization can occur at chiral atom C-2. Therefore, the significant degree of isomerization at C-5 has to be attributed to a temperature dependent imine-enamine tautomerism[20,21]. This was confirmed by heating the diastereomer **6a** in toluene (at 90°C) in absence of 6,9-bis(acetonitrile)-decaborane and monitoring its partial conversion to diastereomer **6b** by hplc. The different rates of isomerization at the atoms C-2 and C-5 have to be attributed to electronic effects of the β-substituents[21,22]. Up to now all attempts to separate the 12:1 diastereomeric mixture of **7a/7b** chromatographically have failed even by using hplc.

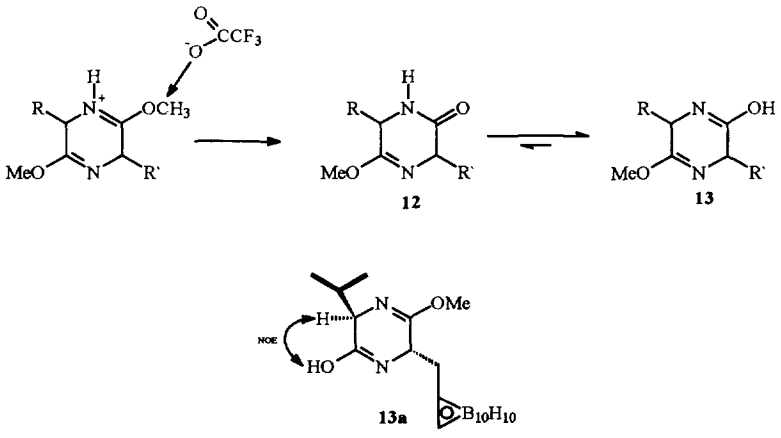
Difficulties also came up in the hydrolysis of the bislactim ether **7.** Several conditions were examined, and the results are summarized in Table 1. Use of standard conditions, *i. e.* of 0.25 M HCl led to a suspension which did not dissolve completely even upon days of incubation at room temperature and, thus, low yields of amino acid esters were obtained. Addition of MeOH or dioxane as cosolvent was found to enhance the reaction rate significantly, but the concomitant formation of undesired by-products was observed. By using 0.1 M trifluoroacetic acid in acetonitrile[23] again fast hydrolysis was observed with the formation of the desired amino acid methyl esters, which however, were contaminated by at least 4 by-products. Finally, with 4.5 equivalents of 3 M HCl in THF[24] optimal conditions could be elaborated for the hydrolysis of the bislactim ether 7.

Table 1: Hydrolysis of bislactim ether **7** under different conditions

Reaction conditions	2.2 equiv. 0.25 M HCl	2.2 equiv. 0.25 M HCl/MeOH	2.2 equiv. 0.25 M HCl/dioxane	3 equiv. 0.1 M trifluoroacetic acid/acetonitrile	4.5 equiv. 3 M HCl/THF
Yield of compound **8**	low	42	41	low	74
Annotations	suspension even after several days	main side product: compound **11** (34 %)	main side product: compound **11** (not isolated)	among at least 4 side products the main product: compound **13 a**	pratically no side products

The formation of the dipeptides H-Val-Xxx-OMe and H-Xxx-Val-OMe as well as of the diketopiperazine has been reported[23] as possible side reactions in this hydrolytic step. In fact, we were able to isolate and characterize one of these compounds, *i.e.* *o*-carboranylalanyl-D-valine methyl ester (**11**). As a possible intermediate of the diketopiperazine formation the heterocycle **12** (Scheme 3) seems to be quite reasonable because it can rearrange to compound **13** via an amide-iminol tautomerism, which was already observed in similar cases[25,26]. Upon treatment of the bislactim ether **7** with 0.1M trifluoroacetic acid a compound was isolated which was identified as **13a** by NMR spectroscopy.

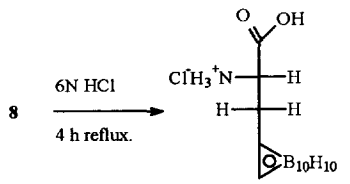
Scheme 3:



Our experience in the hydrolysis of dihydropyrazines confirm the findings of Duthaler[27] that optimal reaction conditions have to be developed for the hydrolysis of different bislactim ethers.

Acid hydrolysis of the *o*-carboranylalanine methyl ester **8** was the final step of the synthesis. Although the

Scheme 4:



10

target molecule, *i.e.* *S*-*o*-carboranylalanine **10** could not be obtained as enantiopure compound, the Schöllkopf-Hartwig procedure proved to be very efficient at least in terms of the relatively fast and facile accessibility.

Experimental Section

THF was freshly distilled from $LiAlH_4$ and toluene from NaH. (2*R*)-(-)-3,6-Dimethoxy-2-isopropyl-2,5-dihydropyrazine, silica gel 60 and butyllithium were purchased from E. Merck A.G. (Darmstadt), decaborane from Aldrich (Steinheim) and propargyl bromide from Fluka A.G. (Neu-Ulm). 6,9-Bis(acetonitrile)-decaborane was prepared according to known procedures (29). Glasware used in the boronation was decontaminated in a Br_2/MeOH bath. ^1H - and ^{13}C - NMR spectra were recorded on a Bruker AM500 spectrometer operating at 500.13 and 100.61 MHz; TMS was used as internal standard. ^{11}B - NMR spectra were recorded on a Bruker ACP200 spectrometer operating at 64.21 MHz. IR spectra were measured on the Perkin Elmer FT-IR 1760 spectrometer on NaCl plates. FAB-MS spectra were obtained on a Finnigan MAT 900 and EI-MS spectra on a Varian MAT 312, respectively. TLC was carried out on silica gel 60 plates (Merck AG) and compounds were visualized by fluorescamine, chlorine/o-tolidine and with $PdCl_2$ reagent (25 mg in 10 ml 1M HCl, 100 °C) for boron-containing compounds. Analytical hplc was performed on Nucleosil^R C_8 (4 x 25 cm, 5 µm particle size, 300 Å) columns, Marchery - Nagel (Düren), using the following buffers: A_1) 15 % CH_3CN/85 % H_3PO_4 (2 %), A_2) 15 % CH_3CN/85 % H_2O, B_1) 80 % CH_3CN/20 % H_3PO_4 (2 %), B_2) 80 % CH_3CN/20 % H_2O, flow rate : 1 mL/min

Abbreviations: Standard abbreviations as recommended by the IUPAC-IUB Commission on Biochemical Nomenclature (*J. Biol. Chem.* **1972**, *247*, 977) were used for amino acids and related derivatives; Pra, propargylglycine; Boc, tert-butyloxycarbonyl; Me, methyl; MeOH, methanol; THF, tetrahydrofuran; AcOEt, ethyl acetate; de diastereomeric excess.

N-(Diphenylmethylene)-*S*-propargylglycine methyl ester (1)

S-Propargylglycine methyl ester hydrochloride (109 mg; 0.66 mmol) was treated in 2.5 ml of CH_2Cl_2 with benzophenonimine (115 µl; 0.66 mmol) at room temperature for 24h. The solvent was removed, and the residue distributed between ether and water. The organic layer was dried with $MgSO_4$, evaporated to dryness and then the residue was chromatographed on 15 g silica gel with *n*-hexane/AcOEt, 4:1. Yield: 126 mg (65 %).

^{1}H-NMR (CD_3OD): 2.29 (dd, 1H, ^4J = 2.5 and 2.9,CH); 2.72, 2.79 (2ddd, 2H, ^2J = 16.7, ^3J = 5.3 and 8.5, ^4J = 2.5 and 2.9, CH_2); 3.71 (s, 3H, OCH_3); 4.28 (dd, 1H, ^3J = 5.3 and 8.5, α-CH); 7.25, 7.35, 7.41, 7.49, 7.57 (5m, 10H, aromatic CH); ^{13}C-NMR (CD_3OD): 23.96 (CH_2); 52.87 (OCH_3); 65.32 (α-CH); 71.93 (CH, alkyne); 81.27 (C, alkyne); 129.16, 129.33, 129.67, 130, 130.19, 131, 131.84, 133.7, 137.18, 140.62 (aromatic C); 172.46, 174.44 (C-N, imine; C, carbonyl); IR (cm^{-1}): 3294 (C-H, alkyne); 3058 (C-H, aromatic); 2952 (C-H, aliphatic); 2122 (C-C, alkyne); 1742 (CO, ester); 1624 (C-N, imine); FAB-MS: m/z = 293 (20%), 292 (84%), 73 (100%); $[M + H]^+$ = 292 calcd. for $C_{19}H_{18}NO_2$.

N-(Diphenylmethyl)-*S*-propargylglycine methyl ester (2)

Decaborane (40 mg; 0.34 mmol) was refluxed in 2 ml of benzene with 26 µl of acetonitrile for 2 h, and then N-(diphenylmethylene)-*S*-propargylglycine methyl ester (95 mg; 0.33 mmol) in 0.5 ml of benzene was added. After 15 h the reaction mixture was cooled to room temperature, filtered and evaporated. The residue was dissolved in MeOH and stirred overnight. The solvent was removed and the crude product extracted with boiling *n*-hexane. The *n*-hexane layer was treated with charcoal, evaporated, and the residue was chromatographed on 10 g silica gel 60 with $CHCl_3$/petroleum ether, 1:1. Yield: 40 mg (15 %). hplc: linear gradient from 100 % A_1 to 100 % B_1 in 25 min followed by isocratic elution with B_1 for 5 min, t_R = 12.9 min (> 95 %).

^{1}H-NMR ($CDCl_3$): 2.03 (t, 1H, ^4J = 2.6, H-C, alkyne); 2.43 (s, 1H, N-H); 2.62 (dd, 2H, ^3J = 5.7, ^4J = 2.6, CH_2); 3.41 (t, 1H, ^3J = 5.7, α-CH); 3.74 (s, 3H, OCH_3); 4.89 (s, 1H, Ph_2CH); 7.21 (dd, 2H, ^3J = 15.5, ^nJ = 7.5, aromatic CH); 7.29 (dd, 4H, ^3J = 15.5, ^nJ = 7.5); 7.38 (d, 2H, ^nJ = 7.5); 7.44 (d, 2H, ^nJ = 7.5); ^{13}C-NMR ($CDCl_3$): 24.28 (CH_2); 52.68 (OCH_3); 57,91 (Ph_2CH) 65.79 (α-CH); 71.85 (CH, alkyne); 80.12 (C, alkyne); 127.96, 128.02, 128.22, 129.23, 143.3, 144.4 (aromatic C); 174.47 (CO); IR (cm^{-1}): 3300 (C-H, alkyne; N-H); 3050 (C-H, aromatic); 2950 (C-H, aliphatic); 2120 (C-C, alkyne); 1740 (CO, ester); FAB-MS: m/z = 295 (10%), 294 (50%); $[M + H]^+$ = 294 calcd. for $C_{19}H_{20}NO_2$.

N-(Phenylmethylene)-*S*-propargylglycine methyl ester (3)

S-Propargylglycine methyl ester hydrochloride (115 mg; 0.7 mmol) was treated at 0°C in 1.5 ml of CH_2Cl_2 containing 60 mg $MgSO_4$ with benzaldehyde (71 µl; 0.7 mmol) and triethylamine (98 µl; 0.7 mmol). The reaction mixture was allowed to reach room temperature during 4 h and was stirred overnight. Next, it was filtered and the filtrate was evaporated and distributed between ether and water. The organic layer was dried with $MgSO_4$ and evaporated to dryness. Yield: 133 mg (88%).

^{1}H-NMR (CD_3OD): 2.33 (dd, 1H, ^4J = 2.6 and 2.6, HC, alkyne); 2.7, 2.89 (2ddd, 2H, ^2J = 17, ^3J = 5.1 and 8.7, ^4J = 2.6, CH_2); 3.75 (s, 3H, OCH_3); 4.18 (dd, 1H, ^3J = 5.1 and 8.7, α-CH); 7.44 (mt, 2H, HC, aromatic); 7.49 (mt, 1H, HC, aromatic); 7.79 (md, 2H, HC, aromatic); 8.41 (s, 1H, HC, imine); ^{13}C-NMR (CD_3OD): 23.9 (CH_2); 52.89 (OCH_3); 72.37, 72.69 (α-CH, CH, alkyne); 80.83 (C-alkyne); 129.79, 132.68, 136.7 (C aromatic); 167.47, 172.49 (C-N, imine, CO); IR (cm^{-1}): 3289 (C-H, alkyne); 3029 (C-H, aromatic); 2953 (C-H, aliphatic); 2122 (C-C, alkyne); 1742 (CO, ester); 1642 (C-N, imine); EI-MS: m/z = 216 (2%), 215 (14%), 214 (93%), 129 (100%); M^+ = 215 calcd. for $C_{13}H_{13}NO_2$.

N-(Benzyl)-*S*-*o*-carboranylalanine methyl ester (4)

N-(Phenylmethylene)-*S*-propargylglycine methyl ester (62 mg; 0.29 mmol) was treated in 5 ml of toluene with 6,9-bis(acetonitrile)-decaborane (62 mg; 0.31 mmol) for 3.5 h at 90 °C. The reaction mixture was cooled to room temperature, the solvent was evaporated, and the residue was dissolved in MeOH. After stirring for 12 h, the MeOH was evaporated, and the residue chromatographed on 11 g silica gel 60 with petroleum ether/AcOEt, 7:1. Yield: 15 mg (16%). hplc: linear gradient from 100 % A_1 to 100 % B_1 in 25 min followed by isocratic elution with B_1 for 5 min, t_R = 18.5 min (95 %).

^{1}H-NMR ($CDCl_3$): 2.4, 2.63 (2dd, 2H, ^2J = 15.4, ^3J = 10.1 and 3.4, CH_2); 3.44 (dd, 1H, ^3J = 10.1 and 3.4, α-CH); 3.56, 3.78 (2d, 2H, ^2J = 12.5, Ph-CH_2); 3.76 (s, 3H, OCH_3); 4.44 (s, 1H, HC, carboranyl); 7.26 (md, 2H, HC, aromatic); 7.32 (mt, 1H, HC, aromatic); 7.36 (mt, 2H, HC- aromatic); 1.4 - 2.8 (10H, BH); ^{13}C-NMR ($CDCl_3$): 40.15 ($\underline{C}H_2$); 52.71, 53.23 ($O\underline{C}H_3$, α-$\underline{C}H$); 60.28 (HC, carboranyl); 73.24 (C, carboranyl); 128.52, 129.31, 129.46, 138.71 (C, aromatic); 173.6 ($\underline{C}O$); ^{11}B-NMR ($CDCl_3$): - 1.34, - 4.13, - 9.1, - 11.96; IR (cm^{-1}): 3317 (N-H); 3062 (C-H, aromatic); 2954 (C-H, aliphatic); 2586 (B-H); 1738 (CO, ester); FAB-MS: m/z = 337.8 (2.5%), 337.3 (1.5%), 336.3 (5%), 335.3 (4%), 334.3 (2.5%); $[M + H]^+$= 337 calcd. for $C_{13}H_{26}{}^{11}B_{10}NO_2$.

(2*R*,5*S*)-3,6-Dimethoxy-2-isopropyl-5-propargyl-2,5-dihydropyrazine (6a)

Under an argon atmosphere 1.44 ml of 1.6 M butyllithium in *n*-hexane (2.3 mmol) was added dropwise to a precooled (-70° C) stirred solution of (2*R*)-(-)-3,6-dimethoxy-2-isopropyl-2,5-dihydropyrazine (**5**) (0.4 ml; 2.2 mmol) in 6 ml of THF . After additional stirring for 20 min at -70° C a precooled solution of propargyl bromide (1.73 ml; 2.2 mmol) in 2 ml of THF was added, and the reaction mixture was stirred for 5 h at -70 °C. Then the reaction mixture was allowed to reach room temperature overnight. The solvent was removed in vacuo, and the oily residue was distributed between 3 x 50 ml AcOEt and 50 ml water. The organic layers were washed with brine, dried over $MgSO_4$, and evaporated to dryness. The crude product was chromatographed on 60 g silica gel with CH_2Cl_2/AcOEt (99:1) as eluent. Yield: 0.35 g, (70 %) of the (2*R*,5*S*)-diastereomer (**6a**; R_f= 0.23, CH_2Cl_2/AcOEt (99:1)) and 0.11 g (22 %) of the (2*R*,5*R*)-diastereomer (**6b**; R_f= 0.12, CH_2Cl_2/AcOEt (99:1)).

6a: hplc: linear gradient from 100 % A_2 to 100 % B_2 in 25 min then isocratic B_2 5 min, t_R = 16.5 min (> 95 %); ^{1}H-NMR ($CDCl_3$): 0.68, 1.05 [2d, 6H, ^3J = 6.7 and 6.8, $(C\underline{H}_3)_2CH$]; 1.88 (dd, 1H, ^4J = 2.6, C-H, alkyne); 2.23 - 2.31 [m, 1H, $(CH_3)_2\underline{C}H$]; 2.67, 2.73 (2ddd, 2H, ^2J = 16.5, ^3J = 4.5 and 5, ^4J = 2.6; CH_2); 3.71, 3.72 (2 s, 6H, 3- and 6- OCH_3); 4.03 (dd, 1H, ^3J = 3.4 and ^5J = 3.4, 2-H); 4.11 (dt, 1H, ^3J = 4.5 and ^5J = 3.4, 5-H); ^{13}C-NMR ($CDCl_3$): 17.26, 19.78 [$(H_3\underline{C})_2CH$]; 25.78 ($\underline{C}H_2$); 32.37 [$(H_3C)_2\underline{C}H$]; 53.25 (3- and 6- $O\underline{C}H_3$); 55.12, 61.70 (2- and 5- $\underline{C}H$); 70.64 ($\underline{C}H$, alkyne); 81.27 ($\underline{C}$, alkyne); 162.56, 165.56 (C-3 and C-6); IR (cm^{-1}): 3300 (HC, alkyne); 2960 (HC, aliphatic); 1700 (C-N, bislactim); EI-MS: m/z = 222 (10%); 183 (68%); 141 (100%); M^+ = 222 calcd. for $C_{12}H_{18}N_2O_2$.

6b: hplc: linear gradient from 100 % A_2 to 100 % B_2 in 25 min then isocratic B_2 5 min, t_R = 17.4 min (95 % (2*R*,5*R*); 4 % (2*R*,5*S*)); ^{1}H-NMR ($CDCl_3$): 0.74, 1.08 [2d, 6H, ^3J = 6.8, $(C\underline{H}_3)_2CH$]; 1.93 (dd, 1H, ^4J = 2.6, alkyne - C-H); 2.23 - 2.31 [m, 1H, $(CH_3)_2\underline{C}H$]; 2.60, 2.77 (2ddd, 2H, ^2J = 16.6, ^3J = 4.7 and 6.8, ^4J = 2.6; CH_2); 3.7 (2 s, 6H, 3- und 6- OCH_3); 3.93 (dd, 1H, ^3J = 5.3 and ^5J = 3.7, 2-H); 4.15 (dt, 1H, ^3J = 6.8 and ^5J = 3.7, 5-H); ^{13}C-NMR ($CDCl_3$): 18.09, 20.32 [$(H_3\underline{C})_2CH$]; 25.79 ($\underline{C}H_2$); 31.55 [$(H_3C)_2\underline{C}H$]; 53.15 (3- and 6- $O\underline{C}H_3$); 55.20, 61.53 (2- and 5- $\underline{C}H$); 70.79 ($\underline{C}H$, alkyne); 81.94 ($\underline{C}$, alkyne); 162.12, 164.73 (C-3 and C-6); IR (cm^{-1}): 3299 (HC, alkyne); 2960 (HC, aliphatic); 1698 (C-N, bislactim).

Thermal isomerization of diastereomer **6a**: 29 mg of **6a** were dissolved in 1.5 ml of toluene and the solution was kept at 90°C; partial isomerization to **6b** was monitored by hplc. hplc: linear gradient from 100 % A_2 to 50 % A_2 /50 % B_2 in 30 min; toluene t_R = 20, **6a** t_R = 23.9, **6b** t_R = 25.9.

3,6-Dimethoxy-2-isopropyl-5-(1,2-dicarba-*closo*-dodecaboran(12)-1-yl)methyl-2,5-dihydropyrazine (7)

To 6,9-bis(acetonitrile)-decaborane (336 mg; 1.66 mmol) (2*R*,5*S*)-3,6-dimethoxy-2-isopropyl-5-propargyl-2,5-dihydropyrazine (336 mg; 1,51 mmol) in 17 ml of toluene was added at room temperature under nitrogen atmosphere. The reaction mixture was kept at 90°C for 4 h, then the suspension was filtered and the solvent was distilled off. The resulting oil was chromatographed on 60 g silica gel with CH_2Cl_2/petroleum ether, 3/1 (R_f= 0.54), as eluent and rechromatographed on 18 g silica gel with petroleum ether/AcOEt, 9/1 (R_f= 0.43). Upon evaporation of the solvent from the combined fractions, the resulting oily residue solidified by standing in the cold. Yield: 180 mg of **7a/7b,** 12:1 (35 %); mp: 76-77° C. hplc: linear gradient from 100 % A_2 to 100 % B_2 in 25 min followed by isocratic elution with B_2 for 5 min, t_R = 19.7 min (> 95 %).

7a: ^{1}H-NMR (CD_2Cl_2): 0.67, 1 [2d, 6H, ^3J = 6.7, $(C\underline{H}_3)_2CH$]; 2.17 - 2.24 [m, 1H, $(CH_3)_2C\underline{H}$]; 2.28, 3.01 (2dd, 2H, ^2J = 14.9, ^3J = 11.4 and 2.5, CH_2); 3.62, 3.65 (2 s, 6H, 3- and 6- OCH_3); 3.93 (dd, 1H, ^3J = 3.6 and ^5J = 3.6, 2-H); 4.07 (ddd, 1H, ^3J = 11.4 and 2.5, ^5J = 3.6, 5-H); 4.78 (s, 1H, C-H, carboranyl); 1.2 - 3 (10H, BH); ^{13}C-NMR (CD_2Cl_2): 16,77, 19.08 $[(H_3\underline{C})_2CH]$; 32.35 $[(H_3C)_2\underline{C}H]$; 41.62 ($\underline{C}H_2$); 53.04 (3- and 6- $O\underline{C}H_3$); 54.97, 60.34, 61.09 (2- and 5- $\underline{C}H$; $\underline{C}H$, carboranyl); 74.02 (C, carboranyl); 160.87, 165.06 (C-3 and C-6); ^{11}B-NMR (CD_2Cl_2): - 1.68; - 4.65; - 9.32; - 12.41.

7b: ^{1}H-NMR (CD_2Cl_2): 0.64, 1.01 [2d, 6H, ^3J = 6.7 and 7, $(C\underline{H}_3)_2CH$]; 2.18 - 3.01 (2dd, 2H, ^2J = 14.7, ^3J = 11.8 and 2.4, CH_2); 2.19 - 2.28 [m, 1H, $(CH_3)_2C\underline{H}$] 3.61, 3.64 (2 s, 6H, 3- and 6- OCH_3); 3.92 (dd, 1H, ^3J = 3.6, ^5J = 4.8, 2-H); 4.12 (ddd, 1H, ^3J = 11.8 and 2.4, ^5J = 4.8, 5-H); 4.89 (s, 1H, C-H carboranyl); 1.2 - 3 (10H, BH); ^{13}C-NMR (CD_2Cl_2): 16,99, 19.35 $[(H_3\underline{C})_2CH]$; 31.25 $[(H_3C)_2\underline{C}H]$; 42.74 ($\underline{C}H_2$); 52.89, 52.96 (3- and 6- $O\underline{C}H_3$); 55.08, 60.61, 60.69 (2- and 5- $\underline{C}H$, $\underline{C}H$, carboranyl); 73.83 (C, carboranyl); 160.48, 164.65 (C-1 and C-4);^{11}B-NMR (CD_2Cl_2): - 2.06; - 4.67; - 8.48; - 9.24; - 10.1; -12.28.

7:IR (cm^{-1}): 3060, 2950 (C-H); 2590 (B-H); 1697(C-N, bislactim); EI-MS: m/z = 342 (6%), 341 (12%), 340 (13%), 339 (8%), 338 (4%), 327 (48%), 326 (94%), 325 (100 %), 324 (64%), 323 (28%), 299 (22%), 298 (48%), 297 (51%), 296 (34%), 295 (16%); M^+ = 342 calcd. for $C_{12}H_{28}{}^{11}B_{10}N_2O_2$.

***o*-Carboranylalanine methyl ester (8)**

To a suspension of compound 7 (0.2 g, 0.6 mmol) in 0.9 ml of 3N HCl, THF was added under stirring until a clear solution was obtained. After stirring for 16 h at room temperature the reaction mixture was evaporated, and the residue distributed between water and ether. The aqueous layer was adjusted to pH 10 with ammonia (25%) and extracted two more times with ether. The combined organic layers were washed with water and brine, dried with $MgSO_4$, filtered, and evaporated. By distillation at $8 \cdot 10^{-3}$ mbar at room temperature 27 mg of the valine methyl ester (**9**) (35 % yield) was obtained. The residual material (146 mg) was chromatographed on 15 g silica gel 60 by gradient elution with petroleum ether/AcOEt, 4:1 to 1:1. Yield: 110 mg (74%) of **8a/8b**. mp: 54°C; $[a]_D^{24}$ = -2.7° (c =1, MeOH);

^{1}H-NMR ($CDCl_3$): 1.44 (s, 2H, NH_2); 1.5 - 2.9 (10H, BH); 2.26 (2dd, 2H, ^2J = 15.3, ^3J = 3.4 and 10.2, CH_2); 3.51 (dd, 1H, ^3J = 3.4 and 10.2, α-CH); 3.67 (s, 3H, OCH_3); 4.54 (s; 1H; CH, carboranyl); ^{13}C-NMR ($CDCl_3$): 41.43 ($\underline{C}H_2$); 53.39, 54.77 (α-$\underline{C}H$, $O\underline{C}H_3$); 60.42 ($\underline{C}H$, carboranyl); 73.38 ($\underline{C}$, carboranyl); 174.79 ($\underline{C}O$); ^{11}B-NMR ($CDCl_3$): - 3.09; - 6.04; - 10.76; - 11.99; - 13.67; IR (cm^{-1}): 3402, 3332 (NH_2); 3050 (CH, carboranyl); 2955 (CH, aliphatic); 2569 (BH); 1716 (CO, ester); EI-MS: m/z = 248, 247, 246, 245, 244, 243, 242, 241; M^+ = 248, 247, 246, 245, 244, 243, 242, 241 calcd. for $C_6H_{10}B_{10}NO_2$.

Side products of the hydrolysis of 7

o-Carboranylalanyl-D-valine methyl ester (11)

^{1}H-NMR ($CDCl_3$): 0.91, 0.94 [2d, 6H, ^3J = 7 and 6.7, $(C\underline{H}_3)_2CH$]; 1.63 (s, 2H, NH_2); 2.18 [m, 1H, $(CH_3)_2C\underline{H}$]; 2.35, 2.94 (2dd, 2H, ^2J = 15.5, ^3J = 8.8 and 3.3, CH_2); 3.5 (dd, 1H, ^3J = 8.8 and 3.3); 3.73 (s, 3H, OCH_3); 4.01 (s, 1H, C-H, carboranyl); 4.42 (dd, 1H, ^3J = 8.8 and 4.9, α-CH); 7.71 (d, 1H, ^3J = 8.8, amide - NH); 1.6 - 2.8 (10H, BH); coupling between 4.42 und 7.71 confirmed in a ^{1}H - ^{1}H - COSY spectrum;^{13}C-NMR ($CDCl_3$): 18.44, 19.74 $[(H_3\underline{C})_2CH]$; 31.69 $[(H_3C)_2\underline{C}H]$; 43.4 $(\underline{C}H_2)$; 52.89 $(O\underline{C}H_3)$; 55.88 (α-C, carboranylalanine); 57.94 (α-C, Val); 62.66 (CH, carboranyl); 73.83 (C, carboranyl); 172.84, 173.32 (CO of carboranylalanine and Val); assignment of signals with the help of a HETCOR programme; ^{11}B-NMR ($CDCl_3$): - 2.90; - 6.05; - 10.19; - 12.63; - 13.48; IR (cm^{-1}): 3328 (H_2N and HN, amide); 3059 (CH, carboranyl); 2966 (CH, aliphatic); 2590 (BH); 1740 (CO, ester); 1661, 1518 (amide I and II); FAB-MS: m/e= 347 (50%); 346 (90%); 345 (100%); 344 (70%); 343 (30%); $[M+H]^+$ = 347 calcd. for $C_{11}H_{28}{}^{11}B_{10}N_2O_3$.

3-Hydroxy-6-methoxy-2-isopropyl-5-(1,2-dicarba-closo-dodecaboran(12)-1-yl)methyl-2,5-dihydropyrazine (13a)

^{1}H-NMR ($CDCl_3$): 0.88, 0.98 [2d, 6H, ^3J = 6.7 and 6.8, $(C\underline{H}_3)_2CH$]; 2.18 [m, 1H, $(CH_3)_2C\underline{H}$]; 2.45, 3.17 (2dd, 2H, ^2J= 15.2, ^3J = 10.3 and 3.2, CH_2); 3.69(s, 3H, OCH_3); 3.86 (dd, 1H, ^3J = 5.7 and ^5J = 3.3, 2-H); 4.08 (dt, 1H, ^3J = 10.3 and 3.2, ^5J = 3.3, 5-H); 4.42 (s, 1H, C-H carboranyl); 6.65 (s, 1H, OH); 1.6 - 2.9 (10H, BH); ^{1}H - ^{1}H - COSY - spectrum: no crosspeak with the singulett at 6.65 ppm; NOESY spectrum: crosspeak between 6.65 and 3.86 ppm and between 4.08 and 0.88 ppm; the NMR data were consistent with the structure **13a**; ^{13}C-NMR ($CDCl_3$): 17.26, 19.09 $[(H_3\underline{C})_2CH]$; 33.25 $[(H_3C)_2\underline{C}H]$; 41.36 $(\underline{C}H_2)$; 54.24 $(O\underline{C}H_3)$; 57.66, 59.85, 60.45 (2- and 5- CH; CH, carboranyl); 74.15 (C, carboranyl); 161.5, 170.09 (C-3 and C-6);^{11}B-NMR ($CDCl_3$): - 2.91; - 5.73; - 10.67; - 13.80; IR (cm^{-1}): 3209 (OH); 3066 (CH, carboranyl); 2964 (CH, aliphatic); 2589 (BH); 1677 (C-N, bislactim); FAB-MS: m/z = 329(12%); 328(30%), 327(35%), 326(20%), 325(8%); $[M+H]^+$ = 329 calcd. for $C_{11}H_{26}{}^{11}B_{10}N_2O_2$.

***o*-Carboranylalanine hydrochloride (10)**

A suspension of *o*-carboranylalanine methyl ester (**8**) (101 mg; 0.41 mmol) in 3.3 ml of 6N HCl was refluxed for 4 h. The resulting almost clear solution was filtered, and kept at 4°C for 12 h. The resulting precipitate was collected and upon concentration a second fraction was obtained. Yield: 87 mg (78%); homogeneous on tlc (n-BuOH/H_2O/HOAc, 4:1:1; 2-Propanol/Pyridine/H_2O, 7:6:6 and $CHCl_3$/MeOH/HOAc 95:5:3 with R_f-values according to Lit.[28,1]); mp: 230°-235°C (dec); $[a]_D^{24}$ = - 6.0° (c = 0.1, H_2O); Lit.[28]: - 6.7° (c = 0.3, H_2O) and Lit.[1]: - 21.0° (c = 1, H_2O) both for *S-o*-carboranylalanine.

^{1}H-NMR (CD_3OD): 1.6 - 2.9 (10H, BH); 2.74, 3.14 (2dd, 2H, ^2J = 16.2, ^3J = 4.9 und 6.3; CH_2); 4.14 (dd, 1H, ^3J = 4.9 und 6.3; α - CH); 4.73 (s, 1H, CH - carboranyl); ^{13}C-NMR (CD_3OD): 39.01($\underline{C}H_2$); 53.27 (α - CH); 64.15 (CH - carboranyl); 73.25 (C - carboranyl); 170.07 (C - carbonyl);^{11}B-NMR (CD_3OD): - 2.36; - 5.03; - 9.31; - 11.74; - 12.49; IR (cm^{-1}): 3400 (NH_3^+); 3058 (CH, carboranyl); 2942 (CH, aliphatic); 2586 (BH); 1754 (CO, carboxylic group); FAB-MS m/e: 234 (20%); 233 (40%); 232 (45%); 231 (30%); 230 (10%); $[M+H]^+$ = 234; 233; 232; 231; 230 calcd. for $C_5H_{18}B_{10}NO_2$.

Acknowledgment

This study was supported by a research grant of Glaxo S. p. A., Via Fleming 2, I - 37100 Verona. The authors thank Dr. I. Zetl for ^{1}H- and ^{13}C-NMR, H. Zink for ^{11}B-NMR measurements, and Prof. Dr. W. Schäfer as well as I. Buhrow for the mass spectra. Thanks are also given to Dr. D. Donati, Dr. S. Rudolph-Böhner and Dr. D. Stephenson for helpful discussions.

References

1. Leukart, O.; Caviezel, M.; Eberle, A.; Escher, E.; Tun-Kyi, A.; Schwyzer, R. *Helv. Chim. Acta* **1976**, *59*, 2184-2187.
2. Fauchere, J.-L.; Leukart, O.; Eberle, A.; Schwyzer, R. *Helv. Chim. Acta* **1979** *62*, 1385-1395.
3. Leukart, O.; Escher, E.; Regoli, D.; Schwyzer, R. *Helv. Chim. Acta* **1979**, *62*, 546-552.
4. Schwyzer, R.; Do, K.Q.; Eberle, A.N.; Fauchere, J.-L. *Helv. Chim. Acta* **1981**, *64*, 2078-2083.
5. Escher, E.; Guillemette, G.; Leukart, O.; Regoli, D. *Eur. J. Pharmacol.* **1980**, *66*, 267-272.
6. Couture, R.; Drouin, J.-N.; Leukart, O.; Regoli, D. *Can. J. Physiol. Pharmacol.* **1979**, *57*, 1437-1442.
7. Hawthorne, M.F. *Angew. Chem.* **1993**, *105*, 997-1033 *Int. Ed. Engl.* **1993**, *32*, 950 - 984.
8. Pettersson, O.-A.; Olsson, P.; Lindström, P.; Sjöberg, S.; Carlsson, J.; *Melanoma Res.* **1993**, *3*, 369-376.
9. Heying, T.L. *Inorg. Chem.* **1963** , *2*, 1089 - 1092.
10. Andersson, A.; Andersson, J.; Burgman, J.-O.; Capola, J.; Carlsson, J.; Conde, H.; Crawford, J.; Graffmann, S.; Grusell, E.; Holmburg, A.; Johansson, E.; Larsson, B.S.; Liljefors, T.; Lindström, P.; Malmquist, J.; Pellettiere, L.; Pettersson, O.; Ponten, J.; Roberti, A.; Russel, K.; Reist, H.; Salford, L.; Sjöberg, S.; Sternerlöw, B.; Strömberg, P.; Westermark, B. *Progress in Neutron Capture Therapy for Cancer*; Allen, B.J. et al. Eds.; Plenum Press: New York, 1992; pp 41 -52.
11. Fitzi, R.; Seebach, D. *Tetrahedron* **1988**, *44*, 5277 - 5292.
12. Wyzlic, I.M.; Soloway A.H. *Tetrahedron Lett.* **1992**, *33*, 7489 - 7490.
13. Radel, P.A.; Kahl, S.B. *Abstr. Fifth Int. Symp. Neutron Capture Ther. Cancer*, **1992**, 5.
14. O'Donnell, M.; Polt, R.L. *J. Org. Chem.* **1982**, *47*, 2663-2666.
15. Blank, S.; Seebach, D. *Angew. Chem.* **1993**, *105*, 1780 - 1781 *Int. Ed. Engl.* **1993**, *32*, 1765 - 1766.
16. Prashar, J.K.; Lama, D.; Moore, D.E. *Tetrahedron Lett.* **1993**, *34*, 6799-6800.
17. Schöllkopf, U.; Groth, C.; Deng, C. *Angew. Chem.* **1981**, *93*, 793 - 795 *Int. Ed. Engl.* **1981**, *20*, 798 - 800.
18. Schöllkopf, U. *Pure and Appl. Chem.* **1983**, *55*, 1799-1806.
19. Frank, H.; Nicholson, G.; Bayer, E. *J. Chromatogr.* **1978**, *167*, 187 - 196.
20. Zeiss, H.-J. *Tetrahedron Lett.* **1987**, *28*,1255 - 1258.
21. Shainyan, B.; Mirskova, A. *Russ. Chem. Rev.* **1979**, *48*, 107 - 117.
22. Ahlbrecht, H.; Fischer, S.; *Tetrahedron* **1970**, *26*, 2837 - 2848.
23. Beulshausen, T.; Groth, U.; Schöllkopf, U. *Liebigs Ann. Chem.* **1991**, 1207 - 1209.
24. Groth, U.; Huhn, T.; Porsch, B.; Schnuck, C.; Schöllkopf, U. *Liebigs Ann. Chem.* **1993**, 715 - 719.
25. Kol'tsov, A.I.; Kheifets, G.M.; *Russ. Chem. Rev.***1972**, *41*, 452 - 467.
26. Wheland, *Advanced Organic Chemistry*; Wiley: New York. 3rd ed. 1960 pp 663 - 730.
27. Duthaler, R.O.; *Tetrahedron*, **1994**, *50*, 1539 - 1650.
28. Leukart, O.; Dissertation ETHZ, No. 5972, 1977.
29. Schaeffer, R. *J. Am. Chem. Soc.* **1957**, *79*, 1006 -1007.

(*Received in Germany* 14 *October* 1994; *revised* 11 *November* 1994; *accepted* 12 *November* 1994)