Building units for N-backbone cyclic peptides. 1. Synthesis of protected N-(ω-aminoalkyl) amino acids and their incorporation into dipeptide units

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Building Units for N-Backbone Cyclic Peptides. 1. Synthesis of Protected N-(ω-Aminoalkylene)amino Acids and Their Incorporation into Dipeptide Units

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A variety of new amino acids which contain an ω-aminoalkylene group on the N^{α}-amino nitrogen were synthesized by alkylation of alkylenediamines with α-halogeno acids. The reaction proceeds with inversion of configuration; thus, optically pure products were obtained when optically active α-halogeno acids were used. The N-(ω-aminoalkylene)amino acids were protected by orthogonal protecting groups to allow their incorporation into dipeptides by the "solution" techniques and into peptides by the solid-phase peptide synthesis (SPPS) methodology. A series of dipeptide analogs of Phe-Gly, Leu-Gly, Trp-Gly, Phe-Leu, and Phe-Ala in which the nitrogen of the peptide bond is alkylated by ω-aminoalkylene chains with various lengths were prepared. These new protected N-(ω-aminoalkylene)amino acids and their derived dipeptide units may be used as building blocks for conformationally constrained N-backbone cyclic peptides.

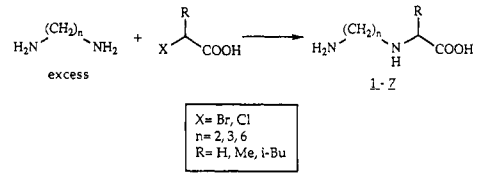
Introduction

Conformational restriction of peptides through cyclization is used for the following purposes: biologically active peptides are cyclized to achieve metabolic stability, to increase potency, to confer or improve receptor selectivity, and to control bioavailability. The possibility of controlling these important pharmacological characteristics through cyclization of linear peptides prompted the use of medium- and long-range cyclization to convert natural bioactive peptides into potential peptidomimetic drugs.[1] Cyclization also brings about structural constraints that enhance conformational homogeneity and facilitate conformational analysis.[1] Consequently, stepwise structural rigidification through cyclization may give an insight into the biologically active conformation of linear peptides, providing that their biological activity and selectivity are maintained.

The common modes of long-range (or global) cyclization are the same modes found in naturally occurring cyclic peptides. These include: (a) side chain to side chain cyclization, (b) end to end cyclization, and (c) side chain to end group cyclization. The main limitation of these classical modes of cyclization is that they require substitution of amino acid side chains by Glu/Lys or Cys in order to achieve cyclization. Such substitutions are permissible in the silent region of peptides but may lead to inactive peptides when applied to their active region.[2-8]

Recently,[9] we have suggested a new general method for medium- and long-range cyclization (cyclization between residue *i* and residue $i + n$ (with $n > 1$) of a peptide chain) in addition to the known modes described above. According to this method, called "backbone cyclization", cyclization involves the joining of N^{α} and/or the C^{α} atoms in the peptidic backbone. The concept of N-backbone cyclization is shown in Figure 1.

Scheme I



Thus, for example, to achieve N-backbone cyclization, hydrogens of the peptide bond are replaced by ω-functionalized alkylene chains that can then be either connected to side chains or ends or interconnected to form the desired cyclic peptide.

Some conformationally constrained peptides which have features similar to those shown in Figure 1 were previously described for short-range cyclizations (namely cyclization between residue *i* and residue $i + 1$ of a peptide chain). Thus, for example, cyclic units containing five-, six-, and seven-membered lactam rings were used to impose conformational restriction on biologically active peptides (ref 10 and references cited therein). These and other modes of short-range cyclization were not used previously for medium- and long-range cyclizations.

Here, we report the synthesis of new N-(ω-aminoalkylene)amino acids and their protection with orthogonal protecting groups. These protected N-(ω-aminoalkylene)amino acids can be incorporated into peptides and subsequently cyclized by the solid-phase methodology. In addition, we describe the use of the protected N-(ω-aminoalkylene)amino acids to prepare protected dipeptides. These protected dipeptides may be incorporated into peptides and cyclized by the classical methodology of peptide synthesis. One of the orthogonally protected N-(ω-aminoalkylene)amino acids was recently used by us for the synthesis of a series of N-backbone to amino end cyclic analogs of substance P by the SPPS methodology.[9,11]

Results and Discussion

1. Synthesis of N-(ω-Aminoalkylene)amino Acids and Their Orthogonal Protection. Various procedures

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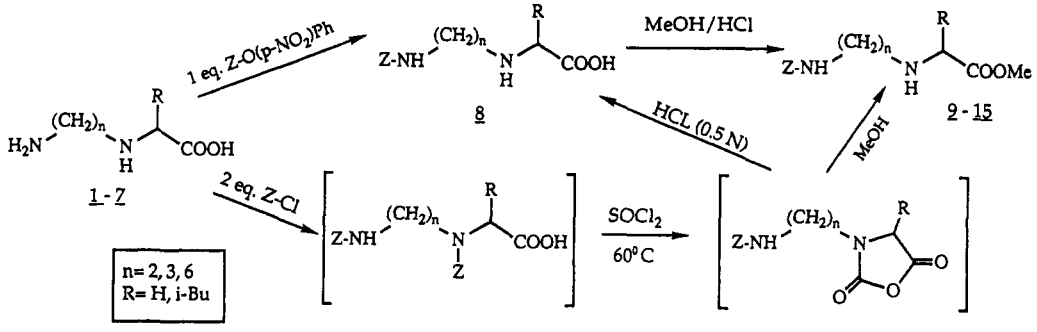
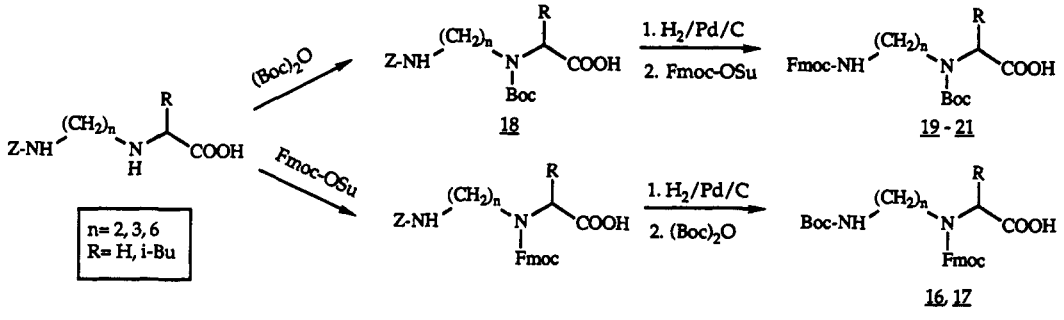
(11) Byk, G.; Halle, D.; Chorev, M.; Selinger, Z.; Gilon, C. Submitted.

0022-3263/92/1957-5687$03.00/0

Scheme II



Scheme III



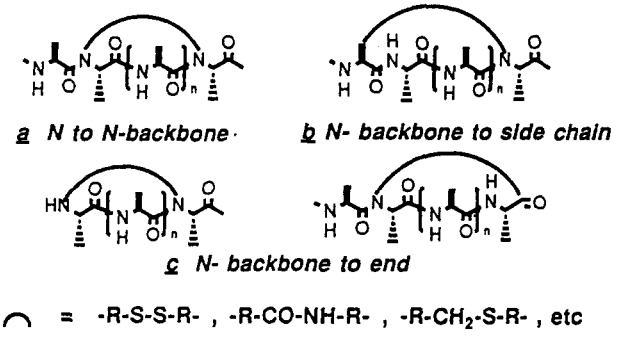


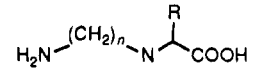
Figure 1. Concept of N-backbone cyclization.

are available for the synthesis of N-alkylamino acids (e.g., ref 12); our attempts to prepare *N*-(ω-aminoalkylene)amino acids using these procedures failed. It seems that introduction of a new function on the N-alkyl chain led to undesirable products.

Our approach, based on a modified nucleophilic substitution of alkylene diamines,[13] is outlined in Scheme I: alkylation of the appropriate alkylene diamine with unsubstituted α-halogeno carboxylic acids produced the appropriated *N*-(ω-aminoalkylene)amino acids in good yield (see Scheme I and Table I, compounds 1–3). In the case of substituted α-halogeno carboxylic acids the yields were lowered (~40%) due to the undesired formation of α,β-dehydro carboxylic acids (see Table I, compounds 4–7; see also section 1c below).

Both N,N and N,N′ dialkylations were not observed due to the use of a large excess of the alkylenediamine. When optically active α-halogeno carboxylic acids were used as alkylating agents, an S_N2 nucleophilic substitution underwent with consequent inversion of configuration (e.g., compounds 6 and 7 Table I). The optical purity of these

Table I. Data for Compounds 1–7



no.	*n*	R	method	% yield	mp, °C	anal.	$[\alpha]^{19}_D$
1	2	H	A	72	153	C, H, N	
2	3	H	A	69[a]	153	C, H, N, Cl	
3	6	H	A	53[a]	202	C, H, N, Cl	
4	2	Me	A	41[a]	198	C, H, N	D,L
5	6	Me	A	47[a]	204	C, H, N	D,L
6	2	i-Bu	A	36	211	C, H, N	−24, 4[b]
7	2	i-Bu	A	37	209	C, H, N	26[c]

[a] As dihydrochloride. [b] D enantiomer, *c* 0.3, 6 N HCl. [c] L enantiomer, *c* 0.32, 6 N HCl.

compounds was checked on a chiral HPLC column (see Experimental Section).

In order to make the *N*-(ω-aminoalkylene)amino acids useful for peptide synthesis both amino groups have to be protected by orthogonal protecting groups. The conventional methods used for the selective protection of diamino acids (e.g., cupric salts complexes, etc.) gave poor results.

This problem was solved by three different approaches:

(a) The use of selective acylating agents such as benzyl or *tert*-butyl *p*-nitrophenyl carbonate (Z-O(*p*-NO_2)Ph and Boc-*O*-(*p*-NO_2)Ph), taking advantage of the steric hindrance in the vicinity of the N^{α} that render the secondary amino function less accessible[13] (see Scheme II). The *N*-(ω-protected-aminoalkylene)amino acids obtained by this procedure were transformed into their corresponding methyl ester which were used as the amino component in the synthesis of dipeptides (see section 2 below). An improved overall yield of the *N*-(ω-protected-aminoalkylene)amino acids is obtained when the protection reaction is performed on the crude *N*-(ω-aminoalkylene)-amino acids without isolation of the latter.

(b) Synthesis of *N*-(Z-ω-aminoalkylene)amino acids or esters by selective deprotection of Z-*N*-(Z-ω-aminoalkylene)amino acids. The selective deprotection reaction was achieved by first cleaving the N^{α}-Z group through the formation of the corresponding N^{α}-carboxy anhydride[14]

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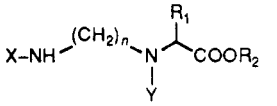
Table II. Data for Compounds 8–21



no.	n	R_1	R_2	X	Y	method	% yield	k'^a	anal.	$[\alpha]^{19}_D{}^b$
8	2	H	H	Z	H	B	37[c]		C, H, N, Cl	
9	2	H	Me	Z	H	B, C	48	1.02	C, H, N	
10	3	H	Me	Z	H	B, C	34	1.08	C, H, N	
11	6	H	Me	Z	H	B, C	23	1.15	C, H, N	
12	2	Me	Me	Z	H	B, C	19	1.15	C, H, N	D,L
13	6	Me	Me	Z	H	B, C	28	1.23	C, H, N	D,L
14	2	*i*-Bu	Me	Z	H	B, C	31[d]	1.26	C, H, N	7.27[e]
15	2	*i*-Bu	Me	Z	H	B, C	29	1.3	C, H, N	−7.1[f]
16	3	H	H	Boc	Fmoc	D	33	1.12	C, H, N	
17	6	H	H	Boc	Fmoc	D	26	1.26	C, H, N	
18	3	*i*-Bu	H	Z	Boc	E	43		C, H, N	−5.8
19	2	H	H	Fmoc	Boc	E	42	1.08	C, H, N	
20	2	H	H	Fmoc	Boc	B	85	1.08	C, H, N	
21	3	*i*-Bu	H	Fmoc	Boc	E	94	1.3	C, H, N	−7.3

[a] RP-18 column, 70% MeOH. [b] *c* 1, MeOH. [c] As hydrochloride. [d] Includes $^1/_3$ H_2O. [e] D enantiomer. [f] L enantiomer.

Table III. ^{1}H NMR Data on Compounds 9–21

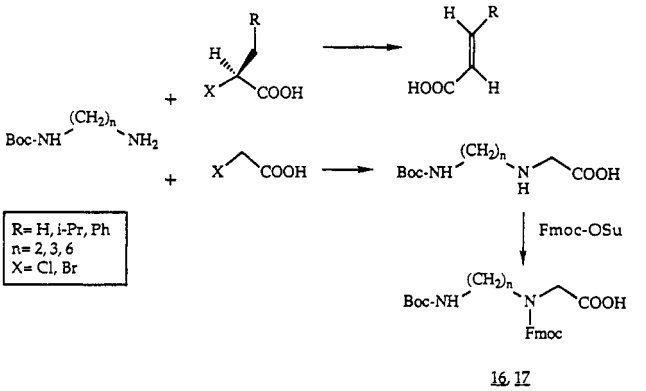
compd	^{1}H NMR
9	7.4 (5 H, Ar), 5.6 (1 H, amide), 5.08 (2 H, Bz), 3.7 (3 H, s, O-CH_3), 3.4 (2 H, s, NCH_2CO), 3.25 (2 H, m, CH_2), 2.75 (2 H, t, CH_2), 1.8 (1 H, s, amine)
10	7.35 (5 H, Ar), 5.75 (1 H, amide), 5.08 (2 H, s, Bz), 3.07 (3 H, s, OCH_3), 3.35 (2 H, s, NCH_2CO), 3.25 (2 H, m, NCH_2), 2.6 (2 H, t, NCH_2), 1.8 (1 H, s, amine)
11	7.35 (5 H, Ar), 5.35 (1 H, amide), 5.08 (2 H, s, Bz), 3.7 (3 H, s, OCH_3), 3.35 (2 H, s, NCH_2CO), 3.15 (2 H, m, NCH_2), 2.5 (2 H, m, NCH_2), 1.85 (1 H, s, amine), 1.45 (4 H, m, $NCH_2CH_2CH_2CH_2CH_2CH_2N$), 1.3 (4 H, m, CH_2CH_2)
12	7.35 (5 H, Ar), 5.4 (1 H, amide), 5.1 (2 H, Bz), 3.7 (3 H, s, OCH_3), 3.35 (2 H, m, NCH_2), 3.22 (1 H, m, αH), 2.6–2.8 (2 H, m, NCH_2), 1.85 (1 H, s, amine), 1.28 (3 H, d, CH_3)
13	7.35 (5 H, Ar), 5.3 (1 H, amide), 5.1 (2 H, s, Bz), 3.7 (3 H, s, OCH_3), 3.35 (1 H, m, αH), 3.15 (2 H, q, NCH_2), 2.5 (2 H, m, NCH_2), 1.85 (1 H, s, amine), 1.5 (4 H, m, $NCH_2CH_2CH_2CH_2CH_2CH_2N$), 1.28 (7 H, m, $CH_3CH_2CH_2$)
14, 15	7.35 (5 H, Ar), 5.5 (1 H, amide), 5.1 (2 H, s, Bz), 3.7 (3 H, s, OCH_3), 3.35 (2 H, m, NCH_2), 3.20 (H, m, αH), 2.7 (2 H, m, NCH_2), 1.9 (1 H, s, amine), 1.49 (2 H, m, CH_2), 1.45 (1 H, m, $CH_2CH(CH_3)_2$), 0.9 (6 H, d, $CH_2CH(CH_3)_2$)
16	7.7 (2 H, Ar), 7.6 (2 H, Ar), 7.2–7.4 (4 H, Ar), 4.4–4.7 (2 H, m, Bz), 4.2 (1 H, m, fluorenyl-CH), 3.8 (2 H, m, CH_2), 2.7–3.5 (4 H, m, $NCH_2CH_2CH_2N$), 1.45 (11 H, s, CH_2 + Boc)
17	7.7 (2 H, Ar), 7.6 (2 H, Ar), 7.2–7.4 (4 H, Ar), 3–4.6 (9 H, fluorenyl-CH_2, CH_2CO, 2 × CH_2N, Bz), 1–1.5 (13 H, s, CH_2CH_2 + Boc)
18	7.45 (5 H, Ar), 6.0 (1 H, NH), 5.2 (2 H, m, Bz), 3.3–3.9 (5 H, αH, $CH_2CH_2CH_2$), 1.6–2.0 (5 H, $CHCH_2$, $CH_2CH_2Ch_2$), 1.43 (9 H, Boc)
19, 20	7.7 (2 H, Ar), 7.6 (2 H, Ar), 7.2–7.4 (4 H, Ar), 4.4–4.7 (2 H, m, Bz), 4.2 (1 H, m, fluorenyl-CH), 3.8 (2 H, m, NCH_2), 2.7–3.5 (4 H, m, CH_2CH_2), 1.45 (9 H, Boc)
21	7.8 (2 H, Ar), 7.6 (2 H, Ar), 7.2–7.4 (4 H, Ar), 4.1–4.4 (4 H, CHCO, CH_2, 3–3.4 (4 H, 2 × CH_2N), 1.5–2 (4 H, 2 × CH_2)), 1.45 (9 H, Boc), 0.9 (6 H, d, $CH_2CH(CH_3)_2$)

and subsequent acid hydrolysis to give the *N*-(Z-ω-aminoalkylene)amino acids (Scheme II compound 8). Methanolysis of the N^α-carboxy anhydride gave the appropriate methyl esters (Scheme II, compounds 9–15). The *N*-(Z-ω-aminoalkylene)amino acids were converted by a three-step protocol (methods H, J, K) to the orthogonally diprotected *N*-(ω-aminoalkylene)amino acids suitable for SPPS (see Scheme III, compounds 16, 17, 19–21).

(c) The use of monoprotected alkylenediamines as starting materials for the alkylation of α-halogeno carboxylic acids. This approach gave satisfactory results only in the case of α-chloroacetic acid. In cases where the α-halogeno carboxylic acids contained β-hydrogens the α,β-dehydro carboxylic acids were the major products (see Scheme IV).

2. Synthesis of Protected Dipeptides Containing an *N*-(ω-Aminoalkylene) Group on the Amide Bond. To demonstrate the use of *N*-(ω-aminoalkylene)amino acids in peptide synthesis we have studied the acylation of various *N*-(Z-ω-aminoalkylene)amino acid methyl esters by various Boc-protected amino acids. Usually, the rate of acylation of *N*-(alkyl)amino acids incorporated in a growing peptide chain is very slow and necessitates a strong coupling reagent and an excess of the acylating amino acid.

Scheme IV



16, 17

In SPPS repeated couplings are used to ensure completion.[15] Moreover, it was reported that pseudopeptides containing a ψ(CH_2NH) bond in the backbone undergo peptide condensation through the N^α primary amine even when the secondary amine is unprotected.[15,16]

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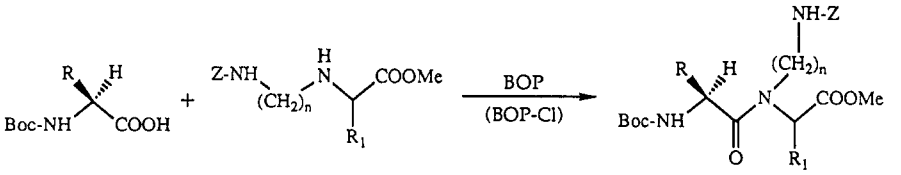
(15) Hocart, S. J.; Nekola, M. V.; Coy, D. H., *J. Med. Chem.* **1987**, *30*, 739.

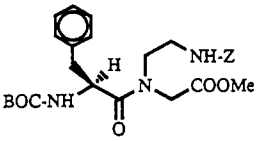
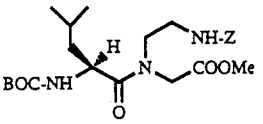
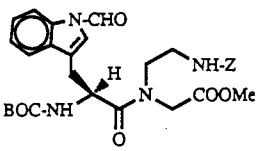
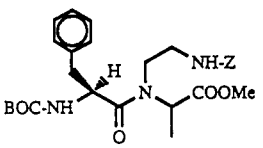
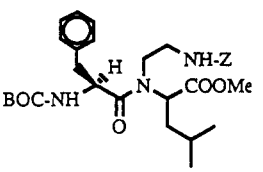
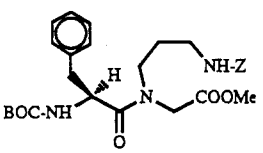
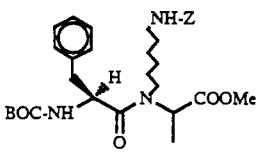
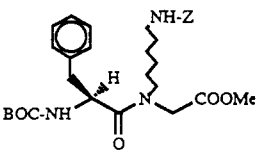
(16) Felix, A. M.; Heimer, E. P.; Wang, C.-T.; Lambros, T. J.; Fournier, A.; Mowles, T. F.; Maines, S.; Campbell, R. M.; Wegrzynski, B. B.; Toome, V.; Fry, D.; Madison, V. S., *Int. J. Peptide Protein Res.* **1988**, *32*, 441.

Scheme V



R= Bzl, i-Bu, (N-For)-3-indolyl-methyl
n= 2, 3, 6
R_1= H, Me, i-Bu

Table IV. Data for Compounds 22–29

no.	structure	method	% yield	[MH]+ calcd	[MH]+ found	k'[a]	anal.	$[\alpha]^{19}_D$[b]
22		F	87	514	514	1.35	C, H, N	-3.43
23		F	98	480	480	1.48	C, H, N	-17.56
24		F	98	581	581	1.6	C, H, N	-8.73
25		G	98	528	528	2.16 (c)	C, H, N	4.43
26		F	95	570	570	2.1 (c)	C, H, N	3.4
27		F	97	528	528	1.63	C, H, N	0.0
28		G	99	584	584	2.4 (c)	C, H, N	-4.52
29		G	96	570	570	1.82	C, H, N	-4.58

[a] RP-18 column, 80% MeOH. [b] c 1, MeOH. [c] RP-18 column, 75% MeOH.

We have found that various Boc amino acids undergo smooth coupling with the secondary amino group of *N*-(Z-ω-aminoalkylene)amino acid methyl esters to give the dipeptides **22–29** as outlined in Scheme V.

Generally strong activation methods are needed for coupling when the amino component is N-alkylated.[15] The

BOP-Cl method has proved to be superior to other coupling methods.[17] In our case, using BOP-Cl as coupling agents gave analytically pure peptides in high yields (about 95%). In some cases BOP reagent[18] was used as coupling agent with similar results and even faster coupling time than the BOP-Cl method (see Table IV).

Conclusions

In this paper we describe the preparation of *N*-(ω-aminoalkylene)amino acids, their orthogonal protection, and coupling with protected amino acids to give dipeptides. These protected *N*-(ω-aminoalkylene)amino acids and their dipeptides are useful building blocks for the synthesis of conformationally constrained peptides by the backbone cyclization method.

Experimental Section

Materials and Methods. Alkylenediamines were purchased from Merck Schuchatdt and were used without further purification. α-Chloro carboxylic acids were synthesized from the corresponding amino acids.[19] α-Bromo carboxylic acids were prepared according to modified procedure[19] (5 N HBr was used instead of 5 N HCl). Benzyl *p*-nitrophenyl carbonate and BOP-Cl were purchased from Aldrich. BOP-Cl was purified according to the known procedure.[17] BOP reagent was purchased from Richelieu Canada. Thionyl chloride was refluxed and distilled over flax oil. All solvents were analytically pure and used without further purification. HPLC was performed on a Merck Hitachi 655A equipped with a LC-5000 gradient pump and UV-vis detector with tunable wave length set at 220 nm. The flow was fixed at 1 mL/min, and the eluants were water (+0.05% TFA), MeOH, and MeCN. The columns were Lichrosphere RP-18 or RP-8 15-cm × 4.2-mm i.d. from Merck. Optical purity was checked on ChiraSpher column from Merck (5 μm, 25-cm × 4-mm i.d.). The flow was fixed at 1 mL/min, and the eluant was a mixture of *n*-hexane–dioxane–2-propanol (50/44/5). The detector was set to 254 nm. Melting points were measured on a Thomas Hoover capillary machine, and optical activity was measured on a Perkin Elmer-141 polarimeter in a 10-cm length cell with a sodium lamp at 25 °C. Microanalysis was carried out at the microanalytical department of The Hebrew University, Jerusalem. ^{1}H NMR spectra were recorded on a Bruker WP-200 pulsed FT spectrometer. Samples were dissolved in $CDCl_3$. Chemical shifts are in ppm relative to TMS internal standard. FAB-MS was determined by Prof. D. M. Desiderio, College of Medicine Department of Neurology, University of Tennessee, Memphis, TN.

Method A. Preparation of *N*-(ω-Aminoalkylene)amino Acids. The appropriate alkylenediamine (15.8 mol) was rapidly stirred at 4 °C (if the alkylenediamine is solid it was dissolved in 500 mL CH_2Cl_2) while the α-halogeno carboxylic acid (1.6 mol) was added portionwise, ensuring that each addition had solubilized. The reaction was then stirred at 25 °C for 48 h and evaporated in vacuo (60 °C).

To the resulting paste was added a solution of DMSO/ether/ethanol (3:1:1, 500 mL) and the mixture left overnight in the freezer. The precipitated zwitterions were collected by filtration on sintered glass and washed with ethanol and ether. In some cases the products were obtained as the dihydrochloride salts rather than the zwitterions (structures and chemical data see Table I). The optical purity of compounds **6** and **7** was checked on their fully protected derivatives **14** and **15** (see method C and Table II).

Method B. Selective Protection of *N*-(ω-Aminoalkylene)amino Acids. A solution of benzyl *p*-nitrophenyl carbonate (0.605 mol) in dioxane (1.3 L) was added dropwise to a stirred solution of the *N*-(ω-aminoalkylene)amino acid (0.4 mol) in 50% aqueous dioxane (2.6 L). The mixture was maintained at pH = 11 (with 2 N NaOH in an automatic titrator). After being stirred for 24 h at room temperature the mixture was evaporated to dryness, dissolved in H_2O (1.2 L), and filtered. The filtrate was extracted with EtOAc (2 × 1 L), and the aqueous layer was cooled in a water–ice bath and acidified to pH = 5.5 with 6 N HCl. After extraction with ether (2 × 1 L), the aqueous layer was acidified (pH = 1 with concentrated HCl), evaporated to dryness, and reevaporated from *i*-POH. In one case (starting material **1** Table I) crystallization from *i*-PrOH gave the acid **8** (Table II). When this procedure was applied to materials **2–7** the products were oils or were obtained in low yield. In these cases the crude monoprotected *N*-(Z-ω-aminoalkylene)amino acids were esterified according to method C below. This procedure increased considerably the overall yields of the diprotected *N*-(ω-aminoalkylene)amino acids. Alternatively, the crude monoprotected *N*-(ω-aminoalkylene)amino acids could be protected on the N^{α} with Boc (method H bellow) and then the N^{ω}-Z cleaved (method K bellow) and the N^{ω} protected with Fmoc (method J) (for structures and chemical data see Table II).

Method C. Esterification of Monoprotected *N*-(ω-Aminoalkylene)amino Acids. Crude *N*-(Z-ω-aminoalkylene)-amino acids (40 mmol) were suspended and stirred in anhydrous MeOH (600 mL), and dry HCl (H_2SO_4 trap) was bubbled for 1 h. The stirring was continued for 1 h at room temperature, and the MeOH was evaporated in vacuo.

The crude product was dissolved in water (500 mL) and washed with EtOAc (2 × 500 mL). The pH of the water was raised to 8 (saturated $NaHCO_3$) and extracted with EtOAc (3 × 300 mL). The organic phase was dried over $MgSO_4$ and evaporated to dryness in vacuo (for structures and chemical data see Tables II and III). The optical purity of the two enantiomers **14** and **15** was checked on a ChiraSpher column. Each compound gave only one peak at different *k'* whereas a mixture gave two peaks with the same *k*'s corresponding to those of the pure compounds.

Method D. Preparation of *N*-(ω-Boc-aminoalkylene)Gly.

1. Preparation of Boc-alkylenediamine. The appropriate alkylenediamine (1 mol) was dissolved in $CHCl_3$ (1 L). The stirred solution was cooled in an ice bath, and $(Boc)_2O$ (0.1 mol in 0.5 L of $CHCl_3$) was added dropwise. The solution was stirred for an additional 24 h at room temperature and the solvent evaporated to dryness in vacuo. The resulting oil was dissolved in ether (0.5 L) and washed with brine (6 × 200 mL). The etheral layer was dried on $MgSO_4$ and evaporated to dryness in vacuo. The resulting oil was dried on P_2O_5 in vacuo.

2. The appropriate monoBoc-alkylenediamine (1 mol) was rapidly stirred at 0 °C while α-chloroacetic acid (0.1 mol) was added portionwise, ensuring that each addition had solubilized. The mixture was left overnight at room temperature, and then ether was added (50 mL) and the precipitate collected by filtration, washed with ether (3 × 50 mL), and dried over P_2O_5. The solid dissolved in water (pH = 1.0) and lyophilized. The solid was dissolved in water (80 mL) and the N^{α} protected with Fmoc (method J) to give products **16** and **17** (see Tables II and III).

Method E. Selective Deprotection of Z-*N*-(Z-ω-aminoalkylene)amino Acids. *N*-(ω-Aminoalkylene)amino acids were reacted with 2 equiv of Z-Cl according to method L. The di-Z products (30 mmol) were dissolved in neat $SOCl_2$ (50 mL) and warmed to 60 °C for 0.5 h. The solution was evaporated in vacuo, and HCl (2 N, 100 mL) was added to the resulting paste. The mixture was stirred during 3 h and washed with ether (3 × 100 mL). The pH of the solution was adjusted to 9 and the N^{α} amino group protected with Boc in situ (method H). The N^{ω} Z-protecting group was removed by catalytic hydrogenation (method K) and reprotected with Fmoc (method J) (see Tables II and III). Alternately, MeOH was added to the paste instead of HCl to give, after evaporation in vacuo, esters **9–15** (Scheme II and Table II).

Method F. Coupling with BOP-Cl. BOP-Cl (1.1 mmol) was added to a stirred solution of the diprotected N-(ω-aminoalkylene)amino acids (1 mmol) followed by 1.2 mmol of DIEA in 10 mL of MeCN at −15 °C. The solution was stirred for 20 min at −15 °C, and the amino acid ester salt (1 mmol) was added in 210 mL of MeCN with 1.1 mmol of DIEA. The stirring was continued overnight at 0 °C. The solvent was evaporated in vacuo and the crude product dissolved in EtOAc and washed with saturated solutions of $KHSO_4$ (2 × 300 mL), $NaHCO_3$ (2 × 300 mL), and brine (2 × 300 mL). The organic phase was dried over $MgSO_4$ and evaporated in vacuo to dryness. (see Table IV).

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Method G. Coupling with BOP. To a stirred solution of the amino acid ester component (1 mmol) in CH_2Cl_2 (10 mL) were added BOP reagent (1.1 mmol), and diprotected N-(ω-aminoalkylene)amino acids (1.1 mmol), DIEA (3 mmol) at room temperature. After 15 min the pH was checked for basicity (in cases where the pH was lower than pH = 9, more DIEA was added) and the reaction mixture left for 1 h at room temperature. The solvent was evaporated under vacuum and the crude product dissolved in EtOAc (100 mL) and washed as described in method F above (see Table IV).

Method H. Preparation of Boc-amino Acids.[20] Amino acid (0.1 mol) was dissolved in NaOH (1 N, 200 mL) and dioxane (200 mL) added. The mixture was stirred in an ice bath, and a solution of $(Boc)_2O$ (0.14 mol) in dioxane (200 mL) was added dropwise while the pH was maintained at 9. The mixture was left stirring at room temperature overnight. The dioxane was evaporated in vacuo and the water solution washed with ether (3 × 150 mL), cooled, and acidified with saturated $KHSO_4$ solution to pH 3. The precipitate was collected by filtration, washed with cold water, and dried on P_2O_5 in vacuo to constant weight. If upon acidification an oil was formed, it was extracted with EtOAc (3 × 150 mL) which was washed with saturated NaCl, dried over $MgSO_4$, and evaporated to dryness. After being dried over P_2O_5, the residue was crystallized from EtOAc/petroleum ether.

Method I. Preparation of Fmoc-amino Acids.[21] A solution of Fmoc-OSu (0.024 mol) in MeCN (25 mL) was added at once to a stirred aqueous solution of amino acid (0.025 mol) adjusted to pH 9 with TEA. The pH was maintained at 8.5–9 with TEA. After 15 min the pH stabilized and the reaction mixture was left another 15 min. The MeCN was evaporated in vacuo, the pH adjusted to 3 with saturated $KHSO_4$, and the precipitate collected by filtration, washed with cold water, and dried over P_2O_5 to constant weight. If upon acidification an oil formed it was treated as in method H.

Method K. Removal of the Z Protecting Group.[22] To a solution of Z-amino acid (1 g) dissolved in MeOH (5 mL) were added Pd/C (10%, 1 g) and ammonium formate (1 g) with stirring. The advance of the reaction was followed by HPLC. After completion (~2 h), the catalyst was removed by filtration and the filtrate evaporated to dryness in vacuo. The residue was dissolved in water which was lyophilized.

Method I. Preparation of Z-amino Acids.[23] Z-Amino acids were prepared according to method H, but Z-Cl was used instead of $(Boc)_2O$.

Abbreviations used are in accordance to the recommendations of IUPAC-IUB Commission on Biochemical Nomenclature in: *Eur. J. Biochem.* **1984**, *138*, 9. *J. Biol. Chem.* **1989**, *264*, 663. Other abbreviations are as follows: Bzl, benzyl; Z, benzyloxycarbonyl; Boc, (*tert*-butyloxy)carbonyl; Fmoc, (fluorenylmethoxy)carbonyl; OSu, *O*-succinimide ester; BOP-Cl, bis(2-oxo-3-oxazolidinyl)phosphinic chloride; BOP, benzotriazolyl-*N*-oxytrisdimethylaminophosphonium hexafluorophosphate; DMSO, dimethyl sulfoxide; DIEA, diisopropylethylamine; TFA, trifluoroacetic acid; TEA, triethylamine.

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Registry No. 1, 24123-14-6; 2, 90495-95-7; 3, 143192-21-6; 4, 143192-22-7; 5, 143192-23-8; 6, 143192-24-9; 7, 143192-25-0; 8, 90495-98-0; 9, 128421-96-5; 10, 143192-26-1; 11, 143192-27-2; 12, 143192-28-3; 13, 143192-29-4; 14, 143192-30-7; 15, 128421-93-2; 16, 143192-31-8; 17, 143192-32-9; 18, 143192-33-0; 19, 143192-34-1; 21, 143192-35-2; 22, 143192-36-3; 23, 143192-37-4; 24, 143192-38-5; 25, 143192-39-6; 26, 143192-40-9; 27, 143192-41-0; 28, 143192-42-1; 29, 143192-43-2; $ClCH_2COOH$, 79-11-8; $H_2NCH_2CH_2NH_2$, 107-15-3; $H_2N(CH_2)_3NH_2$, 109-76-2; $H_2N(CH_2)_6NH_2$, 124-09-4; $BocNH(CH_2)_2NH_2$, 75178-96-0; $BocNH(CH_2)_6NH_2$, 51857-17-1; Boc-Leu-OH, 13139-15-6; Boc-Phe-OH, 13734-34-4; Boc-Tip(CHO)-OH, 47355-10-2.

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Stereocontrolled Synthesis of C_2-Symmetric and Pseudo-C_2-Symmetric Diamino Alcohols and Diols for Use in HIV Protease Inhibitors

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The stereocontrolled syntheses of dibenzyldiamino alcohol 1 and dibenzyldiamino diols 2–4, core units of potent C_2-symmetric and pseudo-C_2-symmetric inhibitors of HIV protease, are described, starting from phenylalanine. Stereoselective epoxidation of trans olefin 7, produced by S_N2' displacement of an allylic mesylate, followed by regiospecific epoxide opening with lithium azide provided the azido alcohol 8 as the major product. Azide reduction and deprotection led to diamine 1. Protected diamino diols 15–17 were prepared expeditiously by intermolecular titanium- or vanadium-mediated pinacol coupling of protected phenylalaninal. Methods for the stereospecific interconversion of the major (3*R*,4*R*,5*R*,6*S*) isomer to the desired (3*S*,4*R*,5*S*,6*S*) isomer via intramolecular hydroxyl inversion are described.

The human immunodeficiency virus type 1 encodes an aspartic proteinase (HIV protease) which is responsible for proteolytic processing of the gag and gag-pol gene products. These proteolytic events are required for the production of mature, infectious progeny virions; thus, HIV protease has received considerable attention as a potential target for the chemotherapy of AIDS. The availability of detailed structural information on the retroviral proteases has inspired the design of inhibitors which exploit the unique structural aspects of these enzymes. Specifically, the recognition that HIV protease exists in its active form as a C_2-symmetric homodimer has prompted interest in

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