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Synthesis of a Partial Sequence of Proinsulin Using the A-Chain of Natural Insulin. IV.[1] Synthesis of a Peptide Corresponding to Positions 31—81 of Bovine Proinsulin

Hiroko AIBA and Yasutsugu SHIMONISHI*

Institute for Protein Research, Osaka University, Yamada-kami, Suita, Osaka 565

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For the synthesis of bovine proinsulin, the *S*-sulfonate of the Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain, which corresponds to positions 41—81 of bovine proinsulin, was coupled with Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–Pro–Gln–Val–Gly–N_3. The *S*-sulfonate of the Arg(Tos)–Arg(Tos)–Glu–Val–Glu–Gly–Pro–Gln–Val–Gly–Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain, correspoding to positions 31—81 of bovine proinsulin, was obtained in a pure form by removal of the protecting groups from the coupled product and chromatography on QAE-Sephadex A-25.

In preceding papers[1–3] we reported the synthesis of the *S*-sulfonates of the Arg–A-chain,[2] Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain[3] and Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain,[1] which correspond to positions 60—81, 53—81, and 41—81 respectively, of bovine proinsulin,[4] using the *S*-sulfonate of the A-chain of natural bovine insulin as a starting material. In the conventional procedure, the low solubility of larger polypeptides protected with hydrophobic groups causes serious problems not only in their reactivity in solution but also in purification. Attempts have been made to find protecting groups that increase the solubility of larger protected polypeptides. The Bunte-salt[2,5] was found to be suitable as both a protecting group for cysteine residues in the A-chain of insulin, as well as for increasing the solubility of the A-chain and its elongated derivatives in buffers and organic solvents such as DMF and DMSO[6] in the presence of water. It was found feasible to carry out the elongation reaction of the A-chain in homogeneous solution and purification of the product by ion-exchange chromatography. In the present work, the procedure was applied to the synthesis of the *S*-sulfonate of the sequence corresponding to positions 31—81 of bovine proinsulin (Fig. 1). The synthesis was carried out as a step in the synthesis of the prohormone.

The synthesis was performed by the reaction of the *S*-sulfonate of sequence 41—81 of bovine proinsulin[1] with a protected peptide azide with the sequence, Arg–Arg–Glu–Val–Glu–Gly–Pro–Gln–Val–Gly, which corresponds to positions 31—40 of the prohormone. Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–Pro–Gln–Val–Gly–OBzl[6] (IX) was synthesized (Fig. 2), the protected decapeptide being constructed from two appropriately protected smaller fragments (V) and (VIII). The synthesis of (V), Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–OH, was started by condensing Z–Val–ONSu[7] with H–Glu(OBu^t)–Gly–OH, obtained by catalytic hydrogenation of its corresponding Z derivative[3] and used directly without isolation. The tripeptide (I) Z–Val–Glu(OBu^t)–Gly–OEt, thus synthesized, was catalytically hydrogenated. The resulting tripeptide ester was not isolated, but coupled directly with Z–Glu(OBu^t)–ONp[8] to give Z–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–OEt (II). The Z group of the protected tetrapeptide (II) was then removed by catalytic hydrogenation, and the resulting tetrapeptide ester was acylated with Z–Arg(Tos)–OH[9] by means of DCC to give Z–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–OEt (III). The protected pentapeptide (III) was once again catalytically hydrogenated and allowed to react with Boc–Arg(Tos)–ONSu[3] to give the protected hexapeptide (IV), Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–OEt. IV was saponified under the usual conditions to give the corresponding acid (V). Synthesis of the latter fragment (VIII), Boc–Pro–Gln–Val–Gly–OBzl, was started by coupling Boc–Val–OH[10] with H–Gly–OBzl by DCC. The dipeptide derivative (VI) thus synthesized was treated with TFA. The resulting dipeptide ester was not isolated, but directly elongated by two further single-reactions using Boc–Gln–ONp[11] and Boc–Pro–ONSu[7] for acylation. The protected tetrapeptide (VIII), Boc–Pro–Gln–Val–Gly–OBzl, thus obtained was treated with TFA. The resulting peptide ester was not isolated, but directly coupled with the

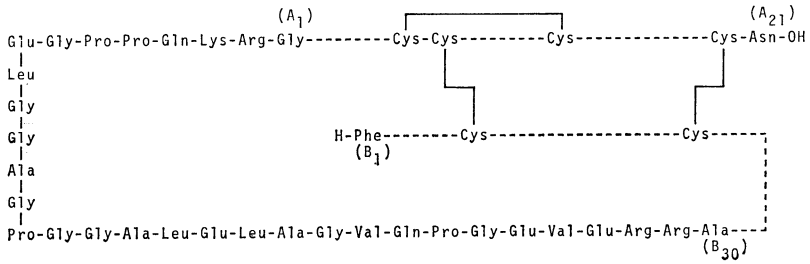

Fig. 1. Structural model of bovine proinsulin.

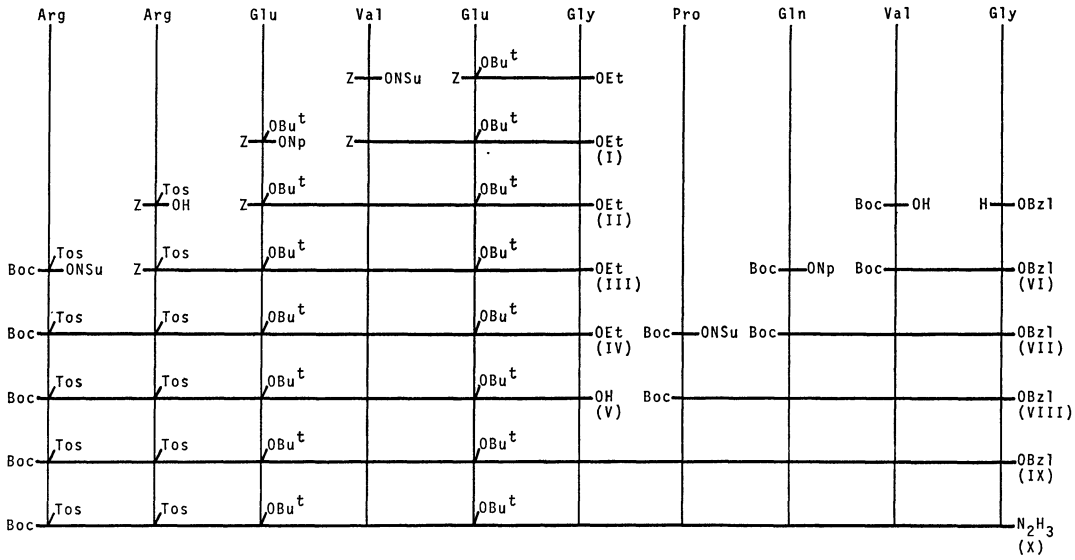

Fig. 2. Scheme for synthesis of protected decapeptide (sequence corresponding to positions BC_1 to C_8 in bovine proinsulin).

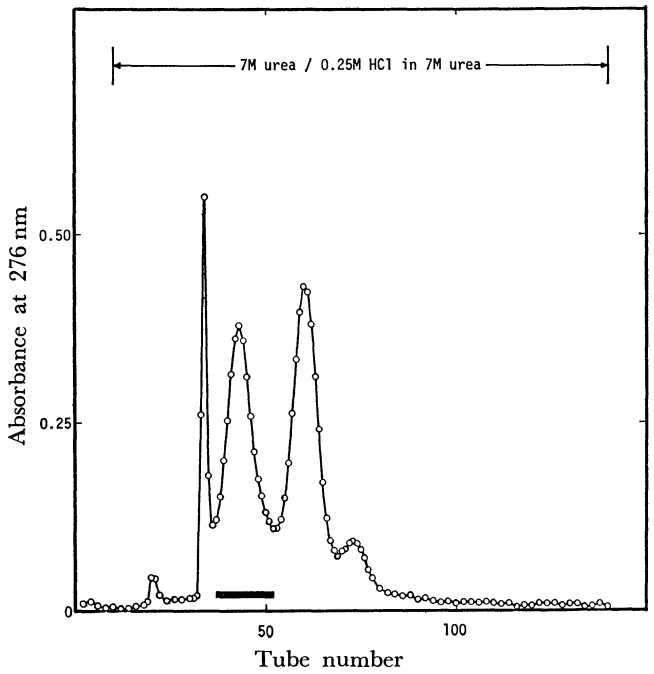

Fig. 3. Chromatography of the *S*-sulfonate of the Arg(Tos)–Arg(Tos)–Glu–Val–Glu–Gly–Pro–Gln–Val–Gly–Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain on QAE-Sephadex A-25 under the conditions described in the text.

hexapeptide acid V by DCC in the presence of HOBt[12] to give the protected decapeptide ester (IX), Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–Pro–Gln–Val–Gly–OBzl, which was then converted to its corresponding hydrazide (X) by the usual method.

Boc–Arg(Tos)–Arg(Tos)–Glu(OBu^t)–Val–Glu(OBu^t)–Gly–Pro–Gln–Val–Gly–N_3, prepared from its corresponding hydrazide (X) by Rudinger's method[13] without being isolated, was coupled with the *S*-sulfonate of the Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-cahin.[1] Azide couplings in the synthesis of peptides corresponding to positions 53—81[3] and 41—81[1] of bovine proinsulin mainly gave the target material, leaving only a little unchanged starting material. On the other hand, on azide coupling starting material remained considerably unreacted, even though the coupling reaction was performed under similar conditions to those described in the preceding papers.[1,3] However, the target product ,the *S*-sulfonate of a peptide corresponding to positions 31—81 of bovine proinsulin, Arg(Tos)–Arg(Tos)–Glu–Val–Glu–Gly–Pro–Gln–Val–Gly–Ala–Leu–Glu–Leu–Ala–Gly–Gly–Pro–Gly–Ala–Gly–Gly–Leu–Glu–Gly–Pro–Pro–Gln–Lys[Z(2-Cl)]–Arg–A-chain, could be easily separated from the starting material and the side-reaction product by ion-exchange chromatography on QAE-Sephadex A-25 after treatment of the reaction product with TFA (Fig. 3). The amino acid ratio of the acid

Table 1. Amino acid ratios of peptides synthesized[a]

Amino acid	Protected [31—40]	*S*-Sulfonate of [41—81][b]	*S*-Sulfonate of [31—81]
Lys		0.92(1)	1.00(1)
Arg	1.87(2)	1.00(1)	2.76(3)
Asp		1.96(2)	2.04(2)
Ser		1.94(2)	1.99(2)
Glu	3.14(3)	7.62(7)	11.27(10)
Pro	1.02(1)	2.88(3)	4.40(4)
Gly	2.00(2)	7.10(7)	9.15(9)
Ala		4.00(4)	4.00(4)
Cys		nd[c]	1.39(2)
Val	2.09(2)	1.67(2)	3.52(4)
Ile		0.55(1)	0.60(1)
Leu		5.00(5)	5.23(5)
Tyr		1.72(2)	1.98(2)

a) Molar ratios of individual amino acids are shown relative to that of glycine or alanine. Numbers in parentheses are theoretical values. b) Cited from Ref. 1. c) Cystine was not determined.

hydrolysate of the purified *S*-sulfonate is given in Table 1. Edman-degradation[14] of the purified material released the phenylthiohydantoin derivative of ω-tosylarginine as the N-terminal amino acid residue, which was identified on polyamide thin-layer by comparison with an authentic sample.

Thus a peptide with 51 amino acid residues, which shares a little over 62% of the 81 amino acid residues of bovine proinsulin, could be prepared in a homogeneous reaction system and purified easily by chromatography on an ion-exchanger.

Experimental

The general experimental and analytical methods are the same as reported.[3] UV-Absorption spectra were recorded on a Hitachi-spectrophotometer, type-124 and ^{1}H-NMR spectra on a JEOL PFT-100 pulse Fourier transform NMR spectrometer locked in deuterium and equipped with an FT-1a pulse control system. Chemical shifts were measured with TMS as an internal reference.

5-[3-[3-(p-Tolylsulfonyl)guanidino]propyl]-3-phenyl-2-thiohydantoin Hydrochloride (PTH-derivative of H-Arg(Tos)-OH). The title compound was prepared from H-Arg(Tos)-OH,[9] as reported[14]; mp 139—141 °C; $[\alpha]_D^{20}$ −9.7° (*c* 0.6, EtOH); UV (EtOH): λ_{max} 269.5 nm ($\varepsilon=15.1\times10^3$), λ_{min} 252 (8.6×10^3); ^{1}H-NMR (D_2O in DMSO-d_6): CH_2-CH_2(1, 2) δ 1.47—1.75 (4H, m), CH_3(aromatic) 2.32 (3H, s), CH_2(3) 3.11 (2H, t), CH(ring) 4.41 (1H, t), H(aromatic) 7.23—7.68 (9H, m); (DMSO-d_6): NH(ring) 10.57 (1H, s) ppm. Found: C, 49.47; H, 4.80; N, 14.50; S, 13.57; Cl, 7.05%. Calcd for $C_{20}H_{23}O_3N_5S_2\cdot HCl$: C, 49.83; H, 5.02; N, 14.53; S, 13.30; Cl, 7.36%.

Z-Val-Glu(OBu^t)-Gly-OEt (I). Z-Glu(OBu^t)-Gly-OEt[3] (16.5 g, 39.1 mmol) was dissolved in EtOH (250 ml) and hydrogenated over 5% palladium-charcoal catalyst. The catalyst was filtered off and the filtrate was concentrated to an oil under reduced pressure. The oil was dissolved with Z-Val-ONSu[7] (13.0 g, 37.4 mmol) in $CHCl_3$ (150 ml), stirred at room temperature for 2 d and then concentrated to a syrup. The syrup was dissolved in AcOEt and washed successively with 0.1 M HCl, 5% aqueous $NaHCO_3$ and water, dried and then concentrated to a crystalline solid, which was collected with AcOEt and hexane; wt 17.5 g. The crude material was recrystallized from EtOH; wt 16.2 g (83.1%), mp 141—143 °C, $[\alpha]_D^{27}$ −31.6° (*c* 2.2, EtOH).

Found: C, 59.89; H, 7.75; N, 8.09%. Calcd for $C_{26}H_{39}O_8N_3$: C, 59.87; H, 7.54; N, 8.06%.

Z-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-OEt (II). Compound I (13.0 g, 24.9 mmol) was suspended in EtOH (250 ml), and hydrogenated over 5% palladium-charcoal at 40 °C. The catalyst was filtered off and the filtrate was concentrated to an oil. The oil was dissolved with Z-Glu(OBu^t)-ONp[8] (12.0 g, 26.2 mmol) in $CHCl_3$ (150 ml) and stirred at room temperature for a day, The solution was washed successively with 0.1M HCl, 5% aqueous $NaHCO_3$ and water, dried and then concentrated to a syrup. The syrup was triturated with hexane; wt 14.3 g. The crude material was recrystallized from AcOEt and hexane; wt 14.1 g (80.1%), mp 149—150 °C, $[\alpha]_D^{27}$ −8.8° (*c* 2.0, DMF).

Found: C, 59.33; H, 7.95; N, 7.83%. Calcd for $C_{35}H_{54}O_{11}N_4$: C, 59.47; H, 7.70; N, 7.93%.

Z-Arg(Tos)-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-OEt (III). Compound II (2.80 g, 3.96 mmol) was dissolved in hot MeOH (150 ml), and hydrogenated over 5% palladium-charcoal catalyst at room temperature. The catalyst was filtered off and the filtrate was concentrated to a solid. The solid was dissolved in THF (100 ml) with Z-Arg(Tos)-OH, which was prepared from its cyclohexylammonium salt[9] (2.50 g, 4.45 mmol), cooled to −5 °C— −10 °C and mixed with HOBt (0.8 g, 5.93 mmol) and DCC (0.9 g, 4.37 mmol). The mixture was stirred at the same temperature for 1 h and at room temperature for 1 d. The precipitate formed was filtered off and the filtrate was concentrated to a syrup. The syrup was dissolved in AcOEt, washed successively with 0.1M HCl, 5% aqueous $NaHCO_3$ and water, dried and concentrated to a crystalline residue; wt 3.60 g. The crude product was recrystallized from EtOH and hexane; wt 3.48 g (86.4%), mp 179.5—181.5 °C, $[\alpha]_D^{21}$ −7.5° (*c* 2.0, DMF).

Found: C, 56.51; H, 7.13; N, 10.95; S, 3.44%. Calcd for $C_{48}H_{72}O_{14}N_8S$: C, 56.67; H, 7.14; N, 11.02; S, 3.15%.

Boc-Arg(Tos)-Arg(Tos)-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-OEt (IV). Compound III (5.10 g, 5.01 mmol) was dissolved in hot MeOH (200 ml) and hydrogenated over 5% palladium-charcoal catalsyt at 35 °C. The catalyst was filtered off and the filtrate was concentrated to a solid. The solid was dissolved with Boc-Arg(Tos)-ONSu[3] (3.90 g, 7.41 mmol) in DMF (50 ml), stirred at room temperature for 1 d and then concentrated to a residue. The residue was dissolved in $CHCl_3$ and washed successively with 0.1M HCl, 5% aqueous $NaHCO_3$ and water, dried, and concentrated to a residue. The residue was collected from $CHCl_3$ and ether; wt 6.06 g. The crude product was reprecipitated from $CHCl_3$ and ether; wt 4.69 g (70.4%), mp 141—142 °C, $[\alpha]_D^{21}$ −8.1° (*c* 2.0, DMF). Amino acid ratio in the acid hydrolysate: Glu, 2.13 (2); Gly, 1.00 (1); Val, 1.04 (1); Arg, 1.95 (2).

Found: C, 52.30; H, 7.15; N, 12.80; S, 4.91%. Calcd for $C_{58}H_{92}O_{17}N_{12}S_2\cdot 2H_2O$: C, 52.39; H, 7.28; N, 12.64; S, 4.82%.

Boc-Arg(Tos)-Arg(Tos)-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-OH (V). Compound IV (1.30 g, 0.977 mmol) was dissolved in MeOH (30 ml) and 1 M NaOH (3 ml) was added to the solution. The solution was stirred at room temperature for 2 h and then neutralized with 1 M HCl. MeOH was evaporated from the solution and the residue was mixed with 0.1 M HCl. The precipitate formed was filtered and reprecipitated from MeOH and ether; wt 1.20 g (94.5%), mp 136—139 °C, $[\alpha]_D^{21}$ −7.5° (*c* 2.0, DMF).

Found: C, 51.46; H, 6.81; N, 12.86; S, 5.04%. Calcd for $C_{56}H_{88}O_{17}N_{12}S_2\cdot 2H_2O$: C, 51.67; H, 7.12; N, 12.92; S, 4.93%.

Boc-Val-Gly-OBzl (IV). Boc-Val-OH prepared from its dicyclohexylammonium salt[10] (20.0 g, 50.1 mmol) was added to a solution of H-Gly-OBzl·TosOH (20.2 g, 59.9 mmol) and TEA (8.4 ml) in THF (200 ml). The solution was cooled below 0 °C, mixed with a solution of DCC (11.3 g, 54.9 mmol) in THF (50 ml) and stirred at the same temperature for 1 h and at room temperature for 1 d. It was then concentrated to a syrupy residue, dissolved in AcOEt and washed successively with 0.1 M HCl, 5% aqueous $NaHCO_3$ and water. The washed solution was dried and then concentrated to a syrup, which was crystallized from hexane; wt 16.0 g. The crude material was recrystallized from EtOH and hexane; wt 10.5 g (57.7%), mp 74.5—76.5 °C, $[\alpha]_D^{20}$ −8.3° (*c* 2.0, DMF).

Found: C, 62.78; H, 7.97; N, 7.75%. Calcd for $C_{19}H_{28}O_5N_2$: C, 62.62; H, 7.74; N, 7.69%.

Boc-Gln-Val-Gly-OBzl (VII). Compound VI (8.40 g, 23.1 mmol) was dissolved in 3.65 M HCl in dioxane (63 ml) and stirred at room temperature. After 1 h the solution was mixed with 3.65 M HCl in dioxane (10 ml) and stirred at room temperature for 30 min. The solution then was

concentrated to a syrup, which was solidified in ether. The solid was dissolved with TEA (3.2 ml) and Boc-Gln-ONp[11] (8.40 g, 22.9 mmol) in DMF (200 ml), stirred at room temperature for 2 d and then concentrated to dryness. The residue was suspended in $CHCl_3$, washed successively with 0.1 M HCl, 5% aqueous $NaHCO_3$ and water, filtered, and then dried; wt 10.0 g. The crude material was crystallized from a mixture of EtOH and MeOH; wt 9.50 g (84.1%), mp 203.5—204.5 °C, $[\alpha]_D^{20}$ −17.3° (*c* 2.0 DMF).

Found: C, 58.68; H, 7.35; N, 11.48%. Calcd for $C_{24}H_{36}O_7N_4$: C, 58.52; H, 7.37; N, 11.38%.

Boc-Pro-Gln-Val-Gly-OBzl (VIII). Compound VII (9.40 g, 19.1 mmol) was dissolved in 3.65 M HCl in dioxane (52 ml), stirred at room temperature for 60 min and then concentrated to a syrup. The syrup was triturated in ether, dissolved with TEA (2.7 ml) and Boc-Pro-ONSu[7] (7.10 g, 22.8 mmol) in DMF (150 ml), stirred at room temperature for 2 d and then concentrated to a solid. The solid was suspended in $CHCl_3$, washed successively with 0.1 M HCl, 5% aqueous $NaHCO_3$ and water, filtered, and dried over P_2O_5; wt 9.71 g. The crude material was reprecipitated from DMF, EtOH, and water; wt 8.66 g (76.7%), mp 210 °C (sintered) and 216—217 °C (decomp), $[\alpha]_D^{20}$ −38.2° (*c* 2.0, DMF). Amino acid ratio in the acid hydrolysate: Glu, 1.12 (1); Pro, 0.99 (1); Gly, 1.00 (1); Val, 1.04 (1).

Found: C, 59.02; H, 7.33; N, 11.89%. Calcd for $C_{29}H_{43}O_8N_5$; C, 59.07; H, 7.35; N, 11.88%.

Boc-Arg(Tos)-Arg(Tos)-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-Pro-Gln-Val-Gly-OBzl (IX). Compound VIII (1.95 g, 3.31 mmol) was stirred in TFA (7 ml) at room temperature for 60 min. The solution was concentrated to a syrupy residue, which was triturated in ether. The material was dissolved wtih compound V (3.88 g, 2.98 mmol), *N*-methylmorpholine (0.43 ml) and HOBt (0.6 g) in DMF (50 ml). The solution was cooled below 0 °C, mixed with a solution of DCC (0.68 g, 3.30 mmol) in DMF (20 ml) and stirred at the same temperature for 1 h and at room temperature for 1 d. The mixture was stirred with HOBt (0.2 g), *N*-methylmorpholine (0.25 ml) and DCC (0.3 g) at room temperature for 2 d. The precipitate formed was filtered off and the filtrate was concentrated to dryness. The residue was collected with DMF and water and reprecipitated from MeOH and AcOEt; wt 4.54 g (86.0%), mp 201.5—203.5 °C (melt and dec), $[\alpha]_D^{20}$ −30.8° (*c* 1.0, DMF).

Found: C, 54.31; H, 7.02; N, 13.07; S, 3.74%. Calcd for $C_{80}H_{121}O_{22}N_{17}S_2 \cdot 2H_2O$: C, 54.19; H, 7.10; N, 13.43; S, 3.62%.

Boc-Arg(Tos)-Arg(Tos)-Glu(OBu^t)-Val-Glu(OBu^t)-Gly-Pro-Gln-Val-Gly-N_2H_3 (X). Compound IX (1.42 g, 0.802 mmol) was dissolved in DMF (10 ml) and mixed with 100% hydrazine hydrate (0.5 g). The solution was stirred at room temperature for 4 h and concentrated to a syrup, which was triturated in ether; wt 1.35 g (100%), mp 207.5—209.5 °C (dec).

Found: C, 52.07; H, 7.34; N, 16.07; S, 4.01%. Calcd for $C_{73}H_{117}O_{21}N_{19}S_2 \cdot H_2O$: C, 52.22; H, 7.14; N, 15.85; S, 3.82%.

S-Sulfonate of the Arg(Tos)-Arg(Tos)-Glu-Val-Glu-Gly-Pro-Gln-Val-Gly-Ala-Leu-Glu-Leu-Ala-Gly-Gly-Pro-Gly-Ala-Gly-Gly-Leu-Glu-Gly-Pro-Pro-Gln-Lys[Z(2-Cl)]-Arg-A-Chain. Compound X (0.83 g, 0.5 mmol) was dissolved in *N*-methylpyrrolidone (8 ml) and mixed with 3.0 M HCl in dioxane (1 ml) and then isopentyl nitrite (70 mg) in *N*-methylpyrrolidone (1 ml) at −20 °C. The solution was stirred at −20 °C——25 °C for 75 min, mixed with *N*-methylmorpholine (0.4 ml) and the *S*-sulfonate of the Ala-Leu-Glu-Leu-Ala-Gly-Gly-Pro-Gly-Ala-Gly-Gly-Leu-Glu-Gly-Pro-Pro-Gln-Lys[Z(2-Cl)]-Arg-A-chain[1] (236 mg, *ca.* 50 μmol), and stirred at 2—3 °C for a week in a refrigerator.

The solution then was mixed with 0.1M NH_4HCO_3 (20 ml) and subjected to gel-filtration on Sephadex G-25 (3×60 cm) in 0.1M NH_4HCO_3. The eluate with absorption at 280 nm was collected and lyophilized. The lyophilized material was dissolved in TFA (10 ml) in an ice-water bath, stirred at room temperature for an hour and concentrated to a syrup. The syrupy material was dissolved in 0.1 M NH_4HCO_3 (50 ml) with a small amount of ammonia water, lyophilized and redissolved with Na_2SO_3 (0.4 g), $Na_2S_4O_6$ (0.4 g) and EDTA·2Na (0.01 g) in 8 M urea (10 ml) at pH 8.0. The solution was stirred at room temperature for 2 d and then charged on a column (3×60 cm) of Sephadex G-25 in 0.1 M NH_4HCO_3. The eluate with absorption at 280 nm was collected and lyophilized. The lyophilized material was charged on a column (9×140 mm) of QAE-Sephadex A-25 (acetate cycle), equilibrated with 7 M urea, and material was eluted with a linear gradient of 7 M urea (250 ml) to 0.25 M HCl in 7 M urea (250 ml) (Fig. 3). The fraction (shown by a bar), with an absorption at 276 nm, was desalted on a column (3×60 cm) of Sephadex G-25 using 0.1 M NH_4HCO_3 and lyophilized; 87.6 mg.

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6) The abbreviations used are as follows: DMF, *N*,*N*-dimtheylformamide: DMSO, dimethyl sulfoxide; DCC, dicyclohexylcarbodiimide; HOBt, 1-hydroxybenzotriazole; TFA, trifluoroacetic acid; THF, tetrahydrofuran; TEA, triethylamine; Other abbreviations used are those recommended by the IUPAC-IUB: *J. Biol. Chem.*, **247**, 977 (1972).
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