Synthesis of Streptolidine Lactam, a Guanidine-containing Amino-acid Lactam Moiety of Streptothricin Antibiotic Group

Mitsuhiro KINOSHITA and Yoshiharu SUZUKI

Department of Applied Chemistry, Faculty of Engineering, Keio University, Hiyoshi, Kohoku-ku, Yokohama 223

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Streptolidine lactam (**3**) present in streptothricin antibiotics was stereospecifically synthesized. 3,4-Anhydro-1,2 : 5,6-di-*O*-isopropylidene-D-iditol (**4**) was converted into the 3,4-bis(benzyloxycarbonylamino)-3,4-dideoxy-D-mannitol derivative **7** *via* the 3,4-diazido-3,4-dideoxy-D-mannitol derivative **6**. De-*O*-protection of **7** followed by two-stage oxidation with periodate-bromine afforded 2,3-bis(benzyloxycarbonylamino)-2,3-dideoxy-D-*arabono*-1,4-lactone (**8**), which was transformed into the azide lactone **9**. Selective hydrogenolysis of **9** with Raney Ni gave the *N*-protected amino sugar lactam **10**. *O*-Tetrahydropyranylation of **10** followed by hydrogenolysis afforded the *O*-protected amino sugar lactam **12**. Treatment of **12** with cyanogen bromide in water gave the *O*-tetrahydropyranylated streptolidine lactam **13**, which on mild acid hydrolysis afforded **3** (hydrochloride). The amino sugar lactam **14** obtained by hydrogenolysis of **9** with palladium black was also treated with cyanogen bromide to yield **3** (hydrobromide). Acid hydrolysis of **3** gave streptolidine (**2**)(dihydrochloride) identical with that derived from the antibiotics.

The ingenious studies on construction of the streptolin-streptothricin group of *streptomyces* antibiotics by Van Tamelen *et al.*[1] revealed the presence of a lactam ring in the streptolidine unit **A** of the intact antibiotic **1**.[2] Borders *et al.*[3] verified the lactam structure by comparison of the PMR spectra of antibiotic LL-AC-541 belonging to the streptothricin family and of streptolidine(**2**), a common degradation product of various streptothricin type antibiotics.

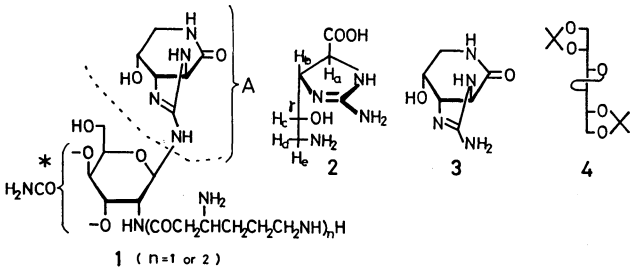


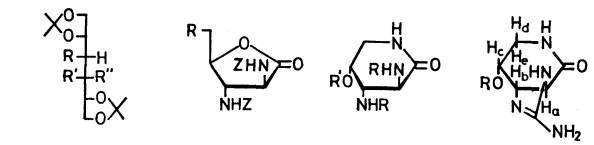
Chart 1.

During the course of synthetic studies on streptothricin antibiotics, we found it necessary to synthesize the streptolidine lactam **3** which corresponds to the cyclic guanidine-containing lactam moiety **A**, for use in a streptothricin antibiotic synthesis and for a biological test in connection with the role of **A** in the biological activity of the antibiotics.[1,5] This paper presents the first synthesis of **3** and its *O*-protected derivative **13**. Streptolidine(**2**) has been synthesized independently by Kusumoto *et al.*,[6] and Goto and Ohgi[7] *via* the amino sugar lactam **14** from D-ribose and D-xylose.[8] In the synthesis of streptolidine lactams, we also adopted the condensation reaction of amino sugar lactam with cyanogen bromide[6,7] for the cyclic guanidine ring formation. Consideration was given to the following points: (i) protection of the free hydroxyl group of the amino sugar lactam **14** stabilizes the lactam ring during the condensation reaction and (ii) the use of equivalent amounts of the pure amino sugar lactam and cyanogen bromide for the reaction minimizes any side reactions which might be caused by excess cyanogen bromide. The *O*-protected amino sugar lactam **12** and free lactam **14** were synthesized in moderate overall yields through the new stereospecific route from the 3,4-anhydro-D-iditol derivative **4** prepared from D-mannitol.[9,10]

* The indicated pattern for the carbamate group substitution has been shown to be most likely vaild for streptolin-streptothricin group (**1**) by Borders *et al.*[4]

Results and Discussion

Starting material, 3,4-anhydro-1,2 : 5,6-di-*O*-isopropylidene-D-iditol(**4**),[9] prepared *via trans*-3,4-didehydro-3,4-dideoxy-1,2 : 5,6-di-*O*-isopropylidene-D-*threo*-hexitol[10] from D-mannitol, was subjected to azidolysis with sodium azide in the presence of ammonium chloride in aqueous methyl cellosolve and the resulting azido hydroxy compound, without purification, was *O*-mesylated in the usual way, affording 3-azido-3-deoxy-1,2:5,6-di-*O*-isopropylidene-4-*O*-mesyl-D-talitol(**5**) in 61% overall yield from **4**. The mesylate **5** was allowed to react with sodium azide in DMSO to give 3,4-diazido-3,4-dideoxy-1,2 : 5,6-di-*O*-isopropylidene-D-mannitol (**6**) in 47% yield. Hydrogenolysis of **6** over palladium black in methanol followed by *N*-benzyloxycarbonylation with benzyl chloroformate in pyridine at −20 °C afforded 3,4-bis(benzyloxycarbonylamino)-3,4-dideoxy-1,2 : 5,6-di-*O*-isopropylidene-D-mannitol (**7**) in 77% yield. De-*O*-isopropylidenation of **7** with warm aqueous acetic acid followed by periodate oxidation with 1.2 equivalent of sodium periodate in aqueous acetone and subsequent oxidation with bromine in aqueous dioxane at room temperature gave 2,3-bis(benzyloxycarbonyl-



5 R=N_3, R'=MsO, R''=H
6 R=R''=N_3, R'=H
7 R=R''=ZHN, R'=H
8 R=HO
9 R=N_3
10 R=Z, R'=H
11 R=Z, R'=THP
12 R=H, R'=THP
14 R=R'=H
13 R=THP
3 R=H

Z=$PhCH_2OCO$
THP=

Chart 2.

amino)-2,3-dideoxy-D-*arabono*-1,4-lactone (**8**) in 59% yield. *O*-Mesylation of **8** in the usual way followed by treatment with sodium azide in DMSO at 100 °C afforded 5-azido-2,3-bis(benzyloxycarbonylamino)-2,3,5-trideoxy-D-*arabono*-1,4-lactone (**9**) in 61% yield.

Selective hydrogenolysis of the azido group in **9** over Raney Ni in methanol led directly[11] to 5-amino-2,3-bis-(benzyloxycarbonylamino)-2,3,5-trideoxy-D-*arabono*- 1,5-lactam (**10**) in 63% yield after recrystallization. The 4-*O*-(tetrahydro-2-pyranyl) derivative **11** (an epimeric mixture about the 2-position of the pyran) was obtained by treatment of **10** with dihydropyran and a catalytic amount of *p*-toluenesulfonic acid in DMF at 37—40 °C in 80% yield after recrystallization. Hydrogenolysis of **11** over 10% palladium on charcoal in methanol at 50 psi affroded, after silica gel column chromatography, an almost pure sample of the *O*-protected sugar lactam **12**, whose IR spectrum($CHCl_3$) showed a δ-lactam band at 1660 cm^{-1}, in 77% yield. Treatment of **12** in water with 0.95 equivalent of cyanogen bromide gave the *O*-protected cyclic guanidine lactam **13** as a crystalline hydrobromide in 51% yield after recrystallization. The ring proton coupling constants in the PMR spectrum of the hydrobromide of **13** were very similar to those of the spectral data[3] reported for the lactam structure of the streptolidine moiety in antibiotic LL-AC541 (Table). Treatment of the hydrobromide with silver carbonate followed by mild hydrolysis with 0.5 M hydrochloric acid afforded the hydrochloride of streptolidine lactam(**3**) as needles in 87% yield after recrystallization. The PMR spectrum of **3** also shows peaks consistent with the lactam structure.

On the other hand, hydrogenolysis of **9** over palladium black in methanol gave, after silica gel column chromatography, an almost pure sample of 2,3,5-triamino-2,3,5-trideoxy-D-*arabono*-1,5-lactam(**14**)[6,7] in 71% yield. *N*-benzyloxycarbonylation of **14** with *N*-(benzyloxycarbonyloxy)succinimide[14] in aqueous DMF yielded **10** in 90% yield. Treatment of **14** in water with one equivalent of cyanogen bromide for 2 h at room temperature afforded the crystalline hydrobromide of **3** in 62% yield after recrystallization. The CD curve of **3** shows the positive Cotton effect at 220 nm. In the plain negative ORD curve of **3**, however, no first positive extremum expected for the positive Cotton effect in the CD spectrum is observed. The ORD curves of streptothricin and its family show a first extremum at 227 nm in positive field through negative field.[12] This indicates that the ORD curve of **3** is more strongly affected by back ground rotation[13] than those of the antibiotics.

Hydrolysis of **3** with 3 M hydrochloric acid overnight at room temperature gave the crystalline dihydrochloride of **2** (85% yield) identical in IR, PMR (Table), and optical rotation with those derived from streptothricin antibiotics. The stereochemistry of synthetic **3** was thus completely confirmed.

In preliminary microbial tests, the synthetic streptolidine lactams **13** and **3** showed no inhibition against test microorganisms sensitive to the streptothricin antibiotic group at the concentration of 100 μg/ml.

TABLE. COMPARISON OF 90 MHz PMR DATA (D_2O)[a] OF SYNTHETIC STREPTOLIDINE LACTAM (**13** and **3**) AND STREPTOLIDINE (**2**) WITH THE REPORTED PMR DATA (100 MHz, D_2O)[a,b] OF ANTIBIOTIC LL-AC541 AND NATURAL STREPTOLIDINE

(δ, Hz)	**13**	**3**	LL-AC541	**2**	
				Synthetic	Natural
H_a	5.15	5.03	5.16	5.06	5.07
H_b	4.55	4.47	4.59	4.73	4.74
H_c	≈5.20	5.14	≈5.20	4.57	4.59
H_d	4.22	4.28	4.32	3.77	3.76
H_e	3.99	3.85	3.89	3.53	3.54
J_{ab}	14.2	14.0	14.8	4.7	4.7
J_{bc}	2.5	2.8	2.7	3.6	3.6
J_{cd}	4.5	5.5	5.5	3.4	3.4
J_{ce}	1.8	1.5	1.2	9.8	9.8
J_{de}	15.0	14.8	15.0	13.2	13.2

a) All chemical shifts are based on external reference of TMS. b) See Ref. 3.

Experimental

Melting points were detemined on a micro hot-stage and are uncorrected. IR spectra were taken on a Hitachi 225 spectrophotometer, PMR spectra on Varian A-60D and EM-390 spectrometers using TMS as internal and external standard. Specific rotations were determined with a Zeiss Photoelectric Polarimeter. CD and ORD spectra were taken on a JASCO J-20 spectropolarimeter. TLC was performed on Wakogel B-5 and colum chromatography on Wakogel C-200. Paper chromatography was conducted on Toyoroshi No. 525 with 1-butanol-acetic acid-water (3 : 1 : 1). Paper electrophoresis was carried out with a Savant IV-5000A in formic acid-acetic acid-water (1 : 3 : 36). Unless otherwise stated, hydrogenolysis was conducted at room temperature over catalyst under bubbling with hydrogen. In general, concentration was carried out under reduced pressure below 40 °C.

1) 3-Azido-3-deoxy-1,2 : 5,6-di-O-isopropylidene-4-O-mesyl-D-talitol (5). A solution of 3,4-anhydro-1,2 : 5,6-di-*O*-isopropylidene-D-iditol (**4**)[9] (36.0 mg) in 80% aqueous methyl cellosolve (1 ml) was heated with sodium azide (78.8 mg) and ammonium chloride (32.6 mg) at 120 °C for 7 h. The reaction mixture was evaporated after filtration and the residue was extracted with ethyl acetate. The dried extract was evaporated to afford a pale brown syrup. The syrup (40 mg) was mesylated with mesyl chloride (0.012 ml) in dry pyridine (0.8 ml) at room temperature for 1 h. Work-up in the usual way gave a brown syrup, which was chromatographed on silica gel with benzene-acetone (20 : 1) to afford **5** (32.6 mg, 60%) as a colorless syrup: $[\alpha]_D^{17}$ −26° (*c* 0.78, $CHCl_3$); $\nu_{max}^{CCl_4}$ 2105 (N_3), 1370 and 1180 cm^{-1} (sulfonate).

Found: C, 42.94; H, 6.25; N, 11.22; S, 8.53%. Calcd for $C_{13}H_{23}N_3O_7S$: C, 42.73; H, 6.34; N, 11.50; S, 8.78%.

2) 3,4-Diazido-3,4-dideoxy-1,2:5,6-di-O-isopropylidene-D-mannitol (6). Sodium azide (365 mg) was added to a solution of **5** (379.4 mg) in dry DMSO (7.6 ml) and the mixture was heated at 120 °C for 3.5 h. The reaction mixture was poured into cold water and extracted with chloroform. The extracts were washed with saturated NaCl solution, dried, and evaporated. The residual brown syrup was chromatographed on silica gel with benzene-acetone (40 : 1) to afford **6** (153.3 mg, 47%) as a colorless syrup: $[\alpha]_D^{28}$ +15° (*c* 0.67, $CHCl_3$), $\nu_{max}^{CCl_4}$ 2100 cm^{-1} (N_3); δ ($CDCl_3$), 1.40 and

1.46 [each s, 6H, $(CH_3)_2C$], and 3.60—4.39 (m, 8H).

Found: C, 46.49; H, 6.55; N, 26.54%. Calcd for $C_{12}H_{20}N_6O_4$: C, 46.14; H, 6.45; N, 26.91%.

*3) 3,4-Bis(benzyloxycarbonylamino)-3,4-dideoxy-1,2 : 5,6-di-O-isopropylidene-*D*-mannitol (7).* A sample of **6** (220 mg) was hydrogenolysis in methanol (6 ml) for 3 h over palladium black. The resulting colorless syrup (200 mg) was dissolved in dry pyridine (4 ml) and cooled to −20 °C, and benzyl chloroformate (0.375 ml) was added to this solution under stirring. After being stirred at −20 °C for 1.5 h, the reaction mixture was diluted with water and extracted with chloroform. The extracts were washed with saturated NaCl solution, dried and evaporated. The residue was subjected to silica gel column chromatography with benzene-acetone (20 : 1) to give **7** (288.6 mg, 77%) as a colorless syrup: $[\alpha]_D^{28}$ +13° (*c* 3.74, $CHCl_3$); δ ($CDCl_3$), 1.32 and 1.42 [each s, 6H, $(CH_3)_2C$], 3.75—4.45 (m, 8H), 5.12 (s, 4H, $PhCH_2$), 5.59—5.86 (br, 2H, NH), and 7.34 (s, 10H, Ph).

Found: C, 63.46; H, 6.88; N, 5.19%. Calcd for $C_{28}H_{36}N_2O_8$: C, 63.62; H, 6.87; N, 5.30%.

*4) 2,3-Bis(benzyloxycarbonylamino)-2,3-dideoxy-*D*-arabono-1,4-lactone (8).* A solution of **7** (82.0 mg) in acetic acid (1.6 ml) was diluted with water (0.8 ml) and warmed at 60 °C for 1 h. The solution was evaporated and the residue was co-evaporated with ether in order to remove acetic acid. The residual solid (70 mg) was dissolved in a mixture of acetone (2.1 ml) and water (0.28 ml), and solid sodium metaperiodate (39.9 mg) was added to the solution in small portions with stirring under ice-cooling. After 1 h, acetone (2 ml) was added to the mixture. The precipitate was filtered off and evaporated to give a colorless solid. Bromine (0.032 ml) was added dropwise to the ice-cooled solution of the solid in dioxane (2.5 ml) and water (0.77 ml), and the mixture was stirred for 1 h. After being kept at room temperature for 3 h, a saturated $Na_2S_2O_3$ solution (4.5 ml) was added to the reaction mixture under ice-cooling. The resulting mixture was extracted with ethyl acetate and extracts were washed with saturated NaCl solution and saturated $NaHCO_3$ solution, dried, and evaporated. The residual solid was dissolved in a small amount of ethyl acetate and filtered in order to remove sulfur. Evaporation of the filtrate afforded a pale yellow solid, which was chromatographed on silica gel with chloroform-methanol (10 : 1) to give a colorless solid. Recrystallization from ethyl acetate and petroleum ether (bp 30—60 °C) afforded needles of **8** (38.6 mg, 59%): mp 149.0—150.5 °C (dried for 24 h at 60 °C in 1 Torr over CaH_2); $[\alpha]_D^{26}$ −51° (*c* 0.89, CH_3OH): ν_{max}^{KBr} 1800 (γ-lactone), 1680 (amide I), and 1530 cm^{-1} (amide II).

Found: C, 59.82; H, 5.44; N, 6.70%. Calcd for $C_{21}H_{22}N_2O_7 \cdot 1/2H_2O$: C, 59.570 H, 5.48; N, 6.62%.

*5) 5-Azido-2,3-bis(benzyloxycarbonylamino)-2,3,5-trideoxy-*D*-arabono-1,4-lactone (9).* A sample of **8** (34.0 mg) was mesylated with mesyl chloride (0.0254 ml) in pyridine (1 ml) at room temperature for 1 h. Work-up in the usual way followed by silica gel column chromatography with benzene-acetone (6 : 1) afforded an almost pure sample of the mesylate (36.1 mg, 89%); δ ($CDCl_3$), 3.85 (s, 3H, CH_3SO_3), 4.02—4.62 (5H, H-2,3,4,5-CH_2), 4.91 (s, 4H, $PhCH_2$), 5.31—5.90 (br, 2H, NH), and 7.15 (s, 10H, Ph). The mesylate (11.5 mg) was treated with sodium azide (6.1 mg) in dry DMSO (0.3 ml) at 100 °C for 0.5 h. Work-up in the suual way followed by silica gel column chromatography with benzene-acetone (10 : 1) gave colorless crystals of **9** (6.8 mg, 68%): mp 84.0—85.0 °C (dried for 24 h at 60 °C in 1 Torr over CaH_2); $[\alpha]_D^{26}$ +20° (*c* 1.72, CH_3OH), $\nu_{max}^{CHCl_3}$ 2100(N_3), 1790(γ-lactone), 1710 (amide I), and 1500 cm^{-1} (amide II).

Found: C, 56.36; H, 4.84; N, 15.63%. Calcd for $C_{21}H_{21}N_5O_6 \cdot 1/2\ H_2O$: C, 56.24; H, 4.95; N, 15.62%.

*6) 5-Amino-2,3-bis(benzyloxycarbonylamino)-2,3,5-trideoxy-*D*-arabono-1,5-lactam (10).* (a) A sample of **9** (37.5 mg) was hydrogenolyzed in methanol with Raney Ni R-100 (Nikko Scientific and Chemical Industries Ltd.) for 4 h. Recrystallization of the crystalline product from methanol gave **10** (22.0 mg, 63%) as fine needles: mp 208—209 °C $[\alpha]_D^{25}$ −122° (*c* 0.84, DMF); ν_{max}^{KBr} 3370 (OH), 3310 (NH), 1680 (amide I), 1650 (δ-lactam), and 1530 cm^{-1} (amide II).

Found: C, 60.74; H, 5.56; N, 10.34%. Calcd for $C_{21}H_{23}N_3O_6$: C, 61.01; H, 5.61; N, 10.16%.

(b) A sample of **9** (231.5 mg) was hydrogenolyzed in methanol (8.5 ml) over palladium black for 5 h. During this period a fresh catalyst was added every hour. Evaporation of the filtered solution afforded the crude triamino sugar lactam **14** as a pale yellow syrup. A solution of *N*-(benzyloxycarbonyloxy)succinimide (283 mg) in DMF (1 ml) was added to a solution of the syrup in DMF (3 ml) and water (0.5 ml). After being kept at room temperature for 1 h, the reaction mixture was neutralized with triethylamine and evaporated. The residue (pale brown semisolid) was chromatographed on silica gel with chloroform-methanol (10 : 1) to give **10** (127.3 mg, 60% overall yield from **9**): mp 203—207 °C; $[\alpha]_D^{20}$ −127° (*c* 0.79, DMF). This sample was identical with that obtained in (a) on mixture melting point.

*7) 5-Amino-2,3-bis(benzyloxycarbonylamino)-2,3,5-trideoxy-4-O-(tetrahydro-2-pyranyl)-*D*-arabono-1,5-lactam (11).* A mixture of **10** (107.2 mg), dihydropyran (0.54 ml), *p*-toluenesulfonic acid (4.8 mg), and dry DMF (1.75 ml) was warmed at 37—40 °C for 2 h. The reaction mixture was neutralized with triethylamine and evaporated. The residual pale brown syrup was chromatographed on silica gel with chloroform-methanol (25 : 1) to afford **11** (124.6 mg, 97%) as a colorless solid. Crystallization took place on slow evaporation of the methanol solution, giving colorless needles of **11** (80% overall yield): mp 163—166 °C; $[\alpha]_D^{20}$ −59° (*c* 1.28, $CHCl_3$); δ ($CDCl_3$), 1.12—1.90 (m, 6H, 3′,4′,5′-CH_2), 3.10—4.76 (m, 8H, 5,6′-CH_2, H-2′, 2, 3, 4), 5.05 (s, 4H, $PhCH_2$), 5.50—6.33 (br, 3H, NH), and 7.30 (s, 10H, Ph).

Found: C, 62.65; H, 6.26; N, 8.53%. Calcd for $C_{26}H_{31}N_3O_7$: C, 62.76; H, 6.28; N, 8.45%.

*8) 2,3,5-Triamino-4-O-(tetrahydro-2-pyranol)-2,3,5-trideoxy-*D*-arabono-1,5-lactam (12).* A sample of **11** (150.8 mg) was hydrogenolyzed in methanol (3 ml) at 50 psi for 4 h using 10% Pd/C catalyst (total 180 mg). The catalyst was divided into four 45 mg-portions, one portion being added every hour during the reaction period. Filtration followed by removal of solvent afforded a pale yellow-green syrup. Chromatography over silica gel (0.7 g) with chloroform-methanol-17% aqueous ammonia (20 : 20 : 1) gave almost pure **12** (53.2 mg, 77%) as a colorless syrup: $\nu_{max}^{CHCl_3}$ 3580—3140 (NH_2), 3400 (NH), and 1660 cm^{-1} (δ-lactam).

9) O-(Tetrahydro-2-pyranyl)streptolidine Lactam (13) Hydrobromide. Cyanogen bromide (23.5 mg, 0.95 equiv) was added to a solution of **12** (53.2 mg) in water (0.5 ml) and the mixture was allowed to stand at room temperature for 5 h. Evaporation of the reaction mixture afforded a pale yellow solid, which was crystallized on slow evaporation of the methanol solution to give colorless needles of the hydrobromide of **13** (40.1 mg, 51%): mp 295 °C (dec); $[\alpha]_D^{20}$ −128° (*c* 0.975, H_2O); ν_{max}^{KBr} 3100 (NH), 1670 (lactam and guanidium I), and 1590 cm^{-1} (vw, guanidium II).

Found: C, 39.44; H, 5.76; N, 16.60%. Calcd for $C_{11}H_{18}N_4O_3 \cdot HBr$: C, 39.41; H, 5.71; N, 16.72%.

10) Streptolidine Lactam (3) Hydrochloride. A solution of **13** hydrobromide (37.5 mg) in water (0.9 ml) was treated with Ag_2CO_3 (15.4 mg, 0.5 equiv) for 10 min. Additional

Ag_2CO_3 (7 mg) was added to the mixture and the precipitates were filtered and washed with water (1 ml). The combined filtrates (1.9 ml) were acidified with 2M HCl (0.6 ml), and kept at room temperature for 0.5 h. The reaction mixture was neutralized (pH 7) with Amberlite CG-4B and evaporated after filtration to afford colorless crystals. Recrystallization from methanol–acetone (1 : 1) gave a pure sample of the hydrochloride of **3** (20.0 mg, 87%) as needles: mp 146—147.5 °C; $[\alpha]_D$ −124°, $[\alpha]_{436}$ −250°, and $[\alpha]_{365}$ −386° (*c* 0.99, H_2O at 16 °C); $[\theta]_{220}$ +3794 (*c* 0.11, H_2O at 18 °C); ν_{max}^{KBr} 1680 (lactam and guanidium I), and 1600 cm^{-1} (sh, guanidium II).

Found: C, 34.72; H, 5.58; N, 26.76%. Calcd for $C_6H_{10}N_4O_2 \cdot HCl$: C, 34.78; H, 5.37; N, 27.12%.

11) Preparation of Streptolidine Lactam (3) Hydrobromide from Triamino Sugar Lactam (14). A crude sample of **14** (Exp. 6b) was chromatographed through a short column of silica gel with chloroform–methanol–17% aqueous ammonia (6 : 6 : 1) to afford a practically pure sample of **14** in 72% yield. Cyanogen bromide (30.0 mg, 1 equiv) was added to a solution of the sample (39.7 mg) of **14** in water (0.4 ml), and the mixture was kept at room temperature for 2 h and then neutralized (pH 7) with Amberlite CG-4B, and evaporated. The pale yellow crystalline residue (70 mg) was recrystallized twice from methanol–acetone to give **3** hydrobromide (42 mg, 60%): mp 147—148 °C; $[\alpha]_D$ −102°, $[\alpha]_{436}$ −210°, and $[\alpha]_{365}$ −329° (*c* 0.638, H_2O at 19 °C); ν_{max}^{KBr} 1680 (lactam and guanidium I) and 1600 cm^{-1} (guanidium II). The PMR spectrum (D_2O) of the sample was identical with that of **3** hydrochloride.

Found: C, 29.02; H, 4.66; N, 22.00%. Calcd for $C_6H_{10}N_4O_2 \cdot HBr$: C, 28.70; H, 4.42; N, 22.31%.

12) Streptolidine (2) Dihydrochloride. A solution of **3** hydrochloride (10.5 mg) in 3 M HCl (0.2 ml) was allowed to stand at room temperature for 22 h. The solution was evaporated and the crystalline residue was washed with a mixture of methanol–acetone (2 : 1) to afford **2** dihydrochloride (11.0 mg, 83%). Recrystallization from water–ethanol gave an analytical sample: mp 175—180 °C (dec) [lit,[15] mp 173—190 °C (dec)]; $[\alpha]_D$ +52°, $[\alpha]_{436}$ +126°, $[\alpha]_{365}$ +195° (*c* 0.988, H_2O at 16 °C) [lit,[15] $[\alpha]_D^{22}$ +56.8° (*c* 2.35, H_2O)]; ν_{max}^{KBr} 1725 (COOH), 1680 (guanidium I), 1580 cm^{-1} (guanidium II). R_f-values on paper chromatopraghy and mobility of paper electrophresis of the synthetic product and the natural one were identical.

Found: C, 27.70; H, 5.21; N, 21.41%. Calcd for $C_6H_{12}N_4O_3 \cdot 2HCl$: C, 27.60; H, 5.41; N, 21.46%.

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