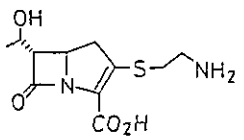
SYNTHESIS OF OPTICALLY ACTIVE 3-MERCAPTOPYRROLIDINE DERIVATIVES. SYNTHETIC INTERMEDIATES OF CARBAPENEM RS-533 AND ITS ISOMER

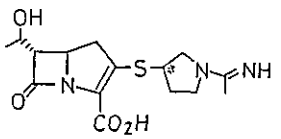
Tomoyuki Shibata, Kimio Iino, and Yukio Sugimura*
Chemical Research Laboratories, Sankyo Co., Ltd.
1-2-58 Hiromachi, Shinagawa-ku, Tokyo, 140, Japan

Abstract- The optically active 3-mercaptopyrrolidine derivatives(3a, 3b, 4a and 4b), which are synthetic intermediates of the carbapenem RS-533 (2a) and its isomer (2b), were synthesized by convenient and practical methods.

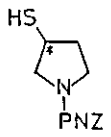
Since the discovery of thienamycin (1)[1], with its high antibacterial activity and broad spectrum, strong interest has been focused on the carbapenem antibiotics. We, also, have been greatly interested in carbapenem synthesis and have already reported the synthesis of RS-533 (2a)[2], which is more potent in activity and biologically more stable than thienamycin. For the synthesis of 2a or its isomer 2b, optically active mercaptane 3 or 4 are necessary. Only two reports are known about the synthesis of optically active 3-hydroxypyrrolidine.[3, 4] We herein report a number of convenient methods for the synthesis of 3 and 4.



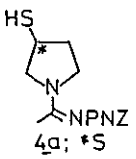
1; THIENAMYCIN



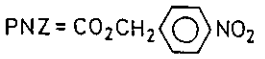
2a: *S RS-533
2b: *R



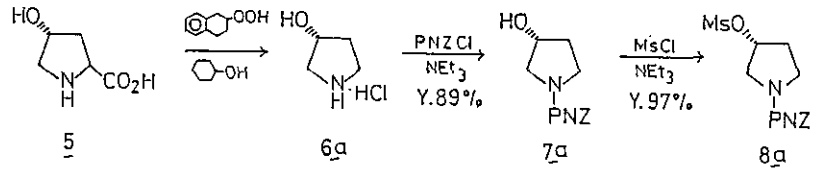
3a: *S
3b: *R

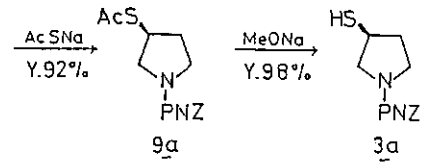


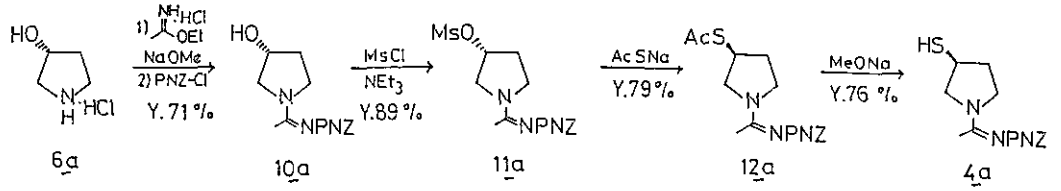
4a: *S
4b: *R



Mercaptane 3a and 4a, which are necessary for the synthesis of RS-533 (2a), have an S-configuration at the C-3 of pyrrolidine and can be synthesized by the conversion of the OH group of L-hydroxyproline (5), which is a natural amino acid, to an SH group via inversion of the stereochemistry. When 5 was heated with a catalytic amount of tetralinehydroperoxide in cyclohexanol, decarboxylation occurred and 6a was obtained.[3] Protection of NH group by the p-nitrobenzyloxycarbonyl group using p-nitrobenzyloxycarbonyl chloride and triethylamine in methylene chloride gave 7a in 89% yield. The hydroxy group of 7a was mesylated with methanesulfonyl chloride and triethylamine in methylene chloride to give 8a in 97% yield. Treatment of 8a with sodium thiolacetate in dimethylformamide gave 9a with inversion of the configuration in 92% yield.[5] Hydrolysis of 9a by sodium methoxide gave 3a in 98% yield. In contrast, treatment of 6a with ethoxyacetimidate hydrochloride in methanol followed by p-nitrobenzyloxycarbonylation gave 10a in 71% yield. Following similar methods, 4a was synthesized from 10a.

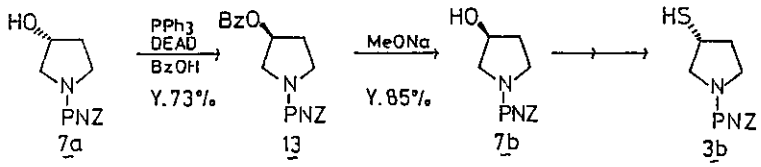


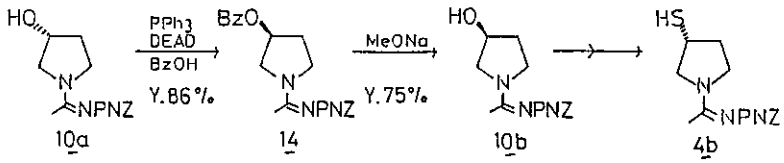




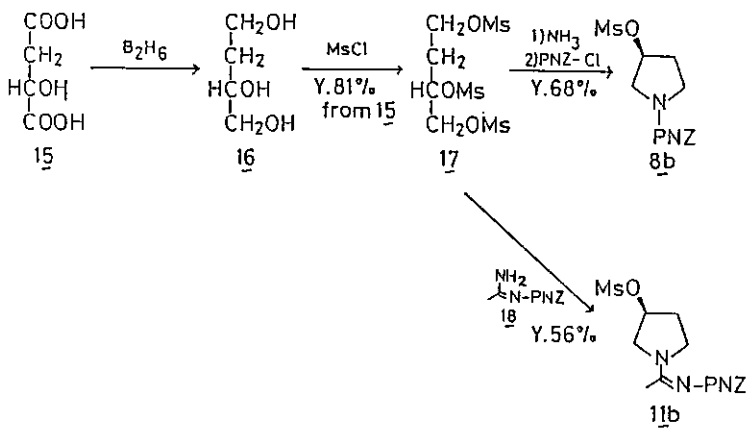
But mercaptane 3b and 4b, which are necessary for the synthesis of 2b, have an R-configuration at the C-3 of pyrrolidine. So the inversions of the OH group of 7a and 10a using Mitsunobu reaction[6] (7a → 13 → 7b, and 10a → 14 → 10b) are necessary. And following conversion to an SH group by the methods mentioned

above gave 3b and 4b, respectively. Therefore we investigated more convenient methods for the synthesis of 3b or 4b.

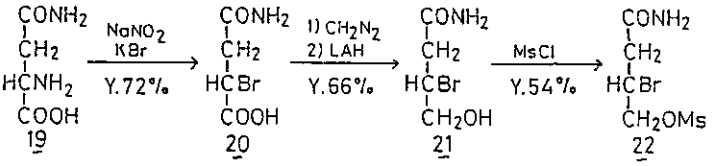


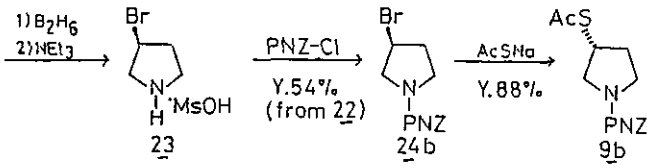


First, as the starting material, L-malic acid (15) was used. Reaction of 15 with diborane gave triol 16[7], quantitatively. Without purification, treatment of 16 with 4.5 equiv. of methanesulfonyl chloride and triethylamine gave trimesylate 17 in 81% yield from 15. Treatment of 17 with ammonia in ethanol in the presence of triethylamine at room temperature for 3-4 days and subsequent treatment with p-nitrobenzyloxycarbonyl chloride gave 8b in 68% yield. When amidine 18 was used, instead of ammonia, 11b was obtained in 56% yield.[8] The 8b and 11b thus obtained showed the same $(\alpha)_D$ values as those of 8b and 11b obtained from L-hydroxyproline.

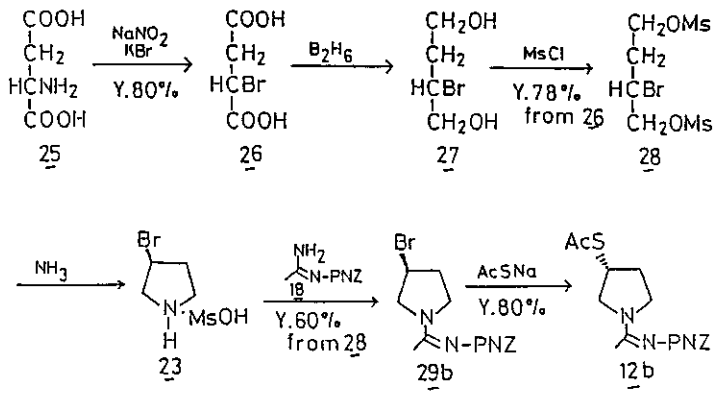


Next, L-asparagine (19) was used as the starting material. The amino group of 19 was converted to the bromo group with sodium nitrite and potassium bromide to give 20 with retention of the stereochemistry in 72% yield.[9] Esterification of 20 with diazomethane and subsequent reduction with lithium aluminum hydride gave alcohol 21 in 66% yield. Treatment of 21 with methanesulfonyl chloride and triethylamine gave mesylate 22 in 54% yield. Reduction of 22 with diborane and subsequent treatment with triethylamine in ethanol at room temperature for 1 week gave the ring closure compound 23, which was treated with p-nitrobenzyloxycarbonyl chloride to give 24b in 54% yield from 22. Treatment of 24b with sodium thiolacetate in dimethylformamide gave thiolacetate 9b with inversion of the stereochemistry in 88% yield. The 9b thus obtained showed the same $[\alpha]_D$ value as that of 9b obtained from L-hydroxyproline.

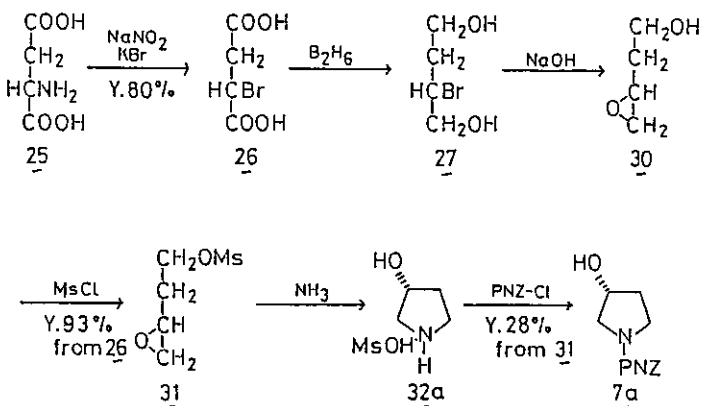




In the third method L-asparatic acid (25) was used as the starting material. The amino group of 25 was converted to the bromo group to give bromide 26 with retention of the stereochemistry in 80% yield.[10] Reduction of 26 with diborane and subsequent mesylation gave dimesylate 28 in 78% yield. Treatment of 28 with ammonia in ethanol in the presence of triethylamine at room temperature for 1 week gave 23. Without purification, 23 was reacted with 18 to give 29b in 60% yield from 28. Treatment of 29b with sodium thiolacetate in dimethylformamide gave 12b with the inversion of the stereochemistry in 80% yield. The 12b thus obtained showed the same $[\alpha]_D$ value as that of 12b obtained from L-hydroxyproline.



If D-asparatic acid is used as the starting material, 4a is obtained by the methods mentioned above. In addition, 3a was also obtained from L-asparatic acid by the following methods. Reduction of 26 with diborane and subsequent treatment with sodium hydroxide gave epoxide 30 in almost quantitative yield. Without purification, 30 was mesylated to give 31 in 93% yield. Treatment of 31 with ammonia in methanol in the presence of alumina at room temperature for 1 day gave 32a, which was reacted with p-nitrobenzyloxycarbonyl chloride to give 7a in 28% yield from 31. The 7a thus obtained showed the same $[\alpha]_D$ value as that of 7a obtained from L-hydroxyproline.



Thus, both 3 and 4 could be synthesized from the cheap starting materials, l-malic acid, L-asparagine and L-asparatic acid in short steps.

EXPERIMENTAL

IR spectra were recorded on a Jasco A-102 spectrometer. Nuclear magnetic resonance spectra were recorded on a Varian XL-100A or a Varian EM-360L spectrometer. Chemical shifts are reported in parts per million (δ) using tetramethylsilane as an internal standard. Optical rotations were recorded on a Perkin-Elmer 241 spectropolarimeter.

3-(R)-HYDROXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (7a) from 6a.
To a suspension of 3-(R)-hydroxypyrrolidine hydrochloride (1.31g) in methylene chloride (20ml), triethylamine (1.4ml) was added under ice water bath cooling. After stirring for 10 min, p-nitrobenzyloxycarbonyl chloride (2.3g) and triethylamine (1.4ml) were added successively at 0°C. After additional stirring for 30 min at the same temperature, the mixture was poured into water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated to give crystals which were recrystallized from benzene-ethyl acetate to give 7a (2.5g) as pale yellow crystals, mp 99-100°C. Yield 89%. NMR($CDCl_3$) 1.8-2.2(2H, m), 3.17(1H, s), 3.4-4.8(4H, m), 4.35-4.65(1H, m), 5.20(2H, s), 7.52,8.17(4H, A_2B_2, J=8.5Hz). IR($CHCl_3$) 3400, 1690, 1610, 1520cm^{-1}. $[\alpha]_D$ -18.5° (c=2.8, $CHCl_3$). Anal. Calcd for $C_{12}H_{14}N_2O_5$: C, 54.13; H, 5.30; N, 10.52. Found: C, 54.06; H, 5.32; N, 10.77.

3-(R)-METHANESULFONYLOXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (8a) from 7a.
To a solution of 7a (3.80g) in methylene chloride (80ml), triethylamine (2.4ml) and methanesulfonyl chloride (1.3ml) were added dropwise simultaneously at 0°C. After being stirred for 30 min the reaction mixture was poured into ice water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated under reduced pressure to give 8a. Recrystallization from benzene gave colorless crystals (4.78g), mp 101-102°C. Yield 97%. NMR($CDCl_3$) 2.0-2.6(2H, m), 3.10(3H, s), 3.3-4.0(4H, m), 5.1-5.5(1H, m), 5.25(2H, s), 7.58,8.20(4H,A_2B_2, J=8.5Hz). IR(KBr) 1715, 1605, 1520cm^{-1}. $[\alpha]_D$ -25.8° (c=0.9, $CHCl_3$). Anal. Calcd for $C_{13}H_{16}N_2O_7S$: C, 45.35; H, 4.68; N, 8.14; S, 9.31. Found: C, 45.39; H, 4.69; N, 8.17; S, 9.45.

3-(S)-ACETYLTHIO-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (9a) from 8a.
To a suspension of NaH (467mg) in dimethylformamide (50ml), thiolacetic acid (1.3ml) was added dropwise at 0°C under nitrogen atmosphere. To the mixture 8a (4.78g) was added and the mixture was heated to 65°C and stirred for 3 h at the same temperature. After cooling, the mixture was poured into ice water and extracted with ethyl acetate. The extract was washed with water, dried over $MgSO_4$ and evaporated under reduced pressure to give an oil which was purified by silica gel column chromatography eluting with hexane-ethyl acetate (2:1) to give 9a (4.14g), mp 69-70°C. Yield 92%. NMR($CDCl_3$) 1.8-2.2(2H, m), 2.34(3H, s), 3.2-4.2(4H, m), 5.22(2H, s), 7.53,8.22(4H, A_2B_2, J=9Hz). IR(KBr) 1715, 1685, 1605, 1520cm^{-1}. $[\alpha]_D$ -25.3° (c=0.9, $CHCl_3$). Anal. Calcd for $C_{14}H_{16}N_2O_5S$: C, 51.84; H, 4.97; N, 8.64; S, 9.89. Found: C, 51.80; H, 4.90; N, 8.68; S, 10.12.

3-(S)-MERCAPTO-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (3a).
To a solution of 9a (4.14g) in methanol (40ml), sodium methoxide (28% methanol solution, 2.7ml) was added at 0°C, and the mixture was stirred for 1 h at room temperature. After quenching the reaction with acetic acid (1ml), the reaction mixture was concentrated to half volume by evaporation and the concentrated mixture was poured into brine and extracted with ethyl acetate. The extract was washed with brine, dried over $MgSO_4$ and evaporated to give 3a (3.48g), mp 58-59°C. Yield 98%. NMR($CDCl_3$) 1.81(1H, d, J=6Hz), 1.7-2.7(2H, m), 3.2-4.1(5H, m), 5.25(2H, s), 7.55,8.17(4H, A_2B_2, J=8.5Hz). IR(KBr) 1715, 1605, 1515cm^{-1}. $[\alpha]_D$ +12.2° (c=1, $CHCl_3$). Anal. Calcd for $C_{12}H_{14}N_2O_4S$: C, 51.05; H, 5.00; N, 9.92; S, 11.36. Found: C, 51.02; H, 4.88; N, 9.89; S, 11.59.

3-(R)-HYDROXY-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (10a) from 6a.
To a suspension of 6a (613mg) in methanol (10ml), sodium methoxide (28% methanol solution, 1.05ml) was added. Under ice water cooling, ethoxyacetimidate hydrochloride (1.65g) was added and the mixture was stirred for 1 h, after which sodium methoxide (28% methanol solution, 2.73ml) was added. After stirring for an additional 30 min under ice water cooling, the solvent was removed from the mixture under reduced pressure. Benzene was added to the mixture and evaporated under reduced pressure. The residue was dissolved in tetrahydrofurane (20ml) and sodium hydride (55% oil dispersion, 218mg) was added. To the mixture, a solution of p-nitrobenzyloxycarbonyl chloride (1.5g) in methylene chloride (20ml) was

added dropwise. After stirring for 1 h, the mixture was extracted with methylene chloride, washed with water, dried over $MgSO_4$ and evaporated to give a residue which was loaded to a silica gel column. The fraction eluted with ethyl acetate gave 10a (1.09g), mp 116-117°C. Yield 71%. NMR(DMSO-d_6) 2.24(3H, s), 2.7-3.5 (5H, m), 3.5-4.2(2H, m), 4.7-5.1(1H, m), 5.30(2H, s), 7.60,8.20(4H, A_2B_2, J=9Hz). IR(KBr) 1675, 1610, 1580, 1520cm^{-1}. $[\alpha]_D$ -13.2° (c=0.3, $CHCl_3$). Anal. Calcd for $C_{14}H_{17}N_3O_5$: C, 54.72; H, 5.58; N, 13.67. Found: C, 54.71; H, 5.58; N, 13.62.

3-(R)-METHANESULFONYLOXY-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (11a) from 10a.

To a solution of 10a (3.22g) in methylene chloride (50ml), were added methanesulfonyl chloride (0.93ml) and triethylamine (1.67ml) successively under ice water cooling. After being stirred for 30 min the mixture was poured into ice water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated under reduced pressure to give an oil which was loaded to a silica gel column. The fraction eluted with ethyl acetate gave 11a (3.60g) as colorless crystals, mp 79-81°C. Yield 89%. NMR($CDCl_3$) 2.31(3H, s), 2.0-2.6(2H, m), 3.05(3H, s), 3.4-4.0(4H, m), 5.17(2H, s), 5.0-5.5(1H, m), 7.55,8.15(4H, A_2B_2, J=9Hz). IR($CHCl_3$) 1680, 1610, 1560, 1525cm^{-1}. $[\alpha]_D$ -29.8° (c=1, $CHCl_3$). Anal. Calcd for $C_{15}H_{19}N_3O_7S$: C, 46.75; H, 4.97; N, 10.90; S, 8.32. Found: C, 46.87; H, 5.00; N, 10.75; S, 8.29.

3-(S)-ACETYLTHIO-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (12a) from 11a.

To a suspension of sodium hydride (55% oil dispersion, 0.735g) in dimethylformamide (30ml) was added thiolacetic acid (1.25ml) at 0°C under a nitrogen atmosphere. After being stirred for 10 min at 0°C, 11a (4.0g) was added and the mixture was stirred at 65°C for 3 h. After cooling, the reaction mixture was poured into water and extracted with ethyl acetate. The extract was washed with water, dried over $MgSO_4$ and evaporated under reduced pressure to give an oil which was purified by silica gel column chromatography. The fraction eluted with benzene-ethyl acetate (2:1) was collected to give 12a (3.0g) as an oil. Yield 79%. NMR($CDCl_3$) 1.8-2.2(2H, m), 2.30(3H, s), 2.35(3H, s), 3.2-4.2(5H, m), 5.16(2H, s), 7.50,8.10(4H, A_2B_2, J=9Hz). IR($CHCl_3$) 1690, 1610, 1560, 1525cm^{-1}.

$[\alpha]_D$ -28.3° (c=1, $CHCl_3$). Anal. Calcd for $C_{16}H_{19}N_3O_5S$: C, 52.59; H, 5.24; N, 11.50; S, 8.77. Found: C, 52.35; H, 5.25, N, 11.22; S, 8.50.

3-(S)-MERCAPTO-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (4a).
To a solution of 12a (3.0g) in methanol (100ml), was added sodium methoxide (28% methanol solution, 1.64ml) at -10°C, and the mixture was stirred for 30 min at 0°C. The reaction was quenched by the addition of acetic acid (0.5ml) and the reaction mixture was concentrated to half volume by evaporation under reduced pressure. The concentrated mixture was poured into brine and extracted with ethyl acetate. The extract was washed with brine, dried over $MgSO_4$ and evaporated to give an oil which was purified by silica gel column chromatography. The fraction eluted with benzene-ethyl acetate (2:1) gave 4a (2.0g), mp 78-81°C. Yield 76%. NMR($CDCl_3$) 1.7-2.7(3H, m), 2.33(3H, s), 3.2-4.1(5H, m), 5.22(2H, s), 7.54,8.17(4H, A_2B_2, J=8.5Hz). IR(KBr) 1670, 1610, 1560, 1520cm^{-1}. $[\alpha]_D$ +24.8° (c=1, $CHCl_3$). Anal. Calcd for $C_{14}H_{17}N_3O_4S$: C, 52.00; H, 5.30; N, 12.99; S, 9.92. Found: C, 51.95; H, 5.20; N, 12.88; S, 10.08.

3-(S)-BENZOYLOXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (13).
Under nitrogen atmosphere, to a solution of 7a (50g), triphenylphosphine (98.5g) and benzoic acid (45.8g) in anhydrous tetrahydrofuran (800ml) was added a solution of diethyl azodicarboxylate (59ml) in anhydrous tetrahydrofuran (500ml). After being stirred for 2 h at room temperature, the mixture was evaporated under reduced pressure to give an oil. To this oil, benzene and hexane were added to give crystals which were removed by filtration. The filtrate was stood overnight to give crystals, which were collected by filtration. The collected crystals were washed with hexane-benzene to give 13 (24g). The combined filtrate and washings were concentrated and the residue obtained was loaded to a silica gel column and eluted with benzene-ethyl acetate (5:1) to give 13 (27g), mp 116-118°C. Yield 73%. NMR($CDCl_3$) 2.0-2.5(2H, m), 3.5-3.9(4H, m), 5.21(2H, s), 5.4-5.7(1H, m), 7.2-7.7(5H, m), 7.8-8.3(4H, m). IR(KBr) 1715, 1605, 1520cm^{-1}. $[\alpha]_D$ +51.8° (c=1, $CHCl_3$).

3-(S)-HYDROXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (7b) from 13.
Under nitrogen atmosphere, sodium methoxide (28% methanol solution, 13.8ml) in methanol (25ml) was added dropwise to a solution of 13 (24.5g) in anhydrous

tetrahydrofuran (400ml) and anhydrous methanol (200ml) at -5°C. After being stirred for 1 h at the same temperature, acetic acid (4ml) was added and the mixture was evaporated under reduced pressure to half volume and subsequently diluted with ethyl acetate, washed with brine, dried over $MgSO_4$ and evaporated to give crystals. The crystals were washed with ether to give 7b (15.2g), mp 99-100°C. Yield 86%. $[\alpha]_D$ +19.5° (c=0.4, $CHCl_3$).

3-(S)-BENZOYLOXY-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (14).
Under nitrogen atmosphere, to a solution of 10a (49g), triphenylphosphine (83.4g) and benzoic acid (38.8g) in anhydrous tetrahydrofuran (1200ml) was added a solution of diethyl azodicarboxylate (50ml) in anhydrous tetrahydrofuran (450ml). After being stirred for 2 h at room temperature, the mixture was evaporated under reduced pressure to give an oil. To this oil, ether (200ml) was added to give crystals, which were collected by filtration and washed with ether to give 14 (56.1g), mp 129-131°C. Yield 86%. NMR($CDCl_3$) 1.9-2.6(2H,m), 2.32 and 2.38(in total 3H, s x 2), 3.5-4.1(4H, m), 5.20(2H, s), 5.4-5.8(1H, m), 7.2-7.7(3H, m), 7.8-8.3(2H, m), 7.54,8.14(4H, A_2B_2, J=9Hz). IR(KBr) 1710, 1680, 1605, 1560, 1515cm^{-1}. $[\alpha]_D$ +63.9° (c=1, $CHCl_3$).

3-(S)-HYDROXY-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (10b) from 14.
Under nitrogen atmosphere, sodium methoxide (28% methanol solution, 12.6ml) in methanol (400ml) was added dropwise to a solution of 14 (55g) in anhydrous tetrahydrofuran (900ml) and anhydrous methanol (400ml) at 0°C. After being stirred for 2 h under ice water cooling, acetic acid (3.7ml) was added and the mixture was evaporated under reduced pressure to half volume and subsequently diluted with ethyl acetate, washed with brine and dried over $MgSO_4$. This extract was concentrated to a volume of 100ml to give crystals. The crystals were collected by filtration, washed with ethyl acetate and ether to give 10b (31g), mp 116-117°C. Yield 75%. $[\alpha]_D$ +13.5°(c=1, $CHCl_3$).

(2S)-1,2,4-TRIMETHANESULFONYLOXYBUTANE (17).
Under nitrogen atmosphere, boron trifluoride etherate (50ml) was added to a suspension of sodium borohydride (11.4g) in tetrahydrofuran (100ml) and the mixture was sttired for 5 min at room temperature. To the mixture, a solution of l-malic acid (10.0g) in tetrahydrofuran (100ml) was added dropwise and the

resulting solution was stirred for 3 h at room temperature. Methanol (200ml) was added dropwise to the mixture and the resulting solution was refluxed for 1 h. After removal of the solvent, the resulting residue was extracted with tetrahydrofuran (150ml). The extract was concentrated under reduced pressure to give a crude triol 16, which could be used in the next reaction without purification. To the solution of crude triol in tetrahydrofuran (300ml), triethylamine (59.7ml) and methanesulfonyl chloride (33.5ml) were added dropwise and the resulting solution was stirred for 2 h at 0°C. The mixture was concentrated under reduced pressure and the resulting residue was extracted with ethyl acetate. The extract was washed with water, dried over $MgSO_4$ and evaporated to give crude trimesylate as white crystals, which were recrystallized from ethanol-isopropylether to afford pure 17 (21.3g), mp 79.5-80°C. Yield 84%. NMR($CDCl_3$) 1.9-2.4(2H, m), 3.05(3H, s), 3.08(3H, s), 3.13(3H, s), 4.0-4.7(4H, m), 4.7-5.2(1H, m). IR(KBr) 1350, 1325, 1175, 1165cm^{-1}. $[\alpha]_D$ -24.4° (c=0.5, $CHCl_3$). Anal. Calcd for $C_7H_{16}O_9S_3$: C, 24.70; H, 4.74; S, 28.26. Found: C, 24.83; H, 4.74; S, 28.53.

3-(S)-METHANSULFONYLOXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (8b) from 17.
To a suspension of 17 (340mg) in ethanol (8ml) triethylamine (0.14ml) was added and ammonia gas was bubbled for 1 h at room temperature. After being stirred for 3 days at room temperature the mixture was evaporated under reduced pressure. The resulting residue was suspended in methylene chloride (8ml), and triethylamine (0.14ml) was added. After stirring for 10 min, p-nitrobenzyloxycarbonyl chloride (230mg) and triethylamine (0.14ml) were added successively at 0°C. After being stirred at the temperature, the mixture was poured into water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated to give crystals which were recrystallized from benzene to give pure 8b (230mg), mp 101-102°C. Yield 67%. $[\alpha]_D$ +25.6° (c=1, $CHCl_3$).

3-(S)-METHANESULFONYLOXY-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (11b) from 17.
To a suspension of 17 (500ml) in ethanol (10ml) triethylamine (0.21ml) was added and ammonia gas was bubbled through the resulting mixture for 1 h at room temperature. After being stirred for 3 days at room temperature, the mixture was evaporated under reduced pressure. The resulting residue was suspended in

ethanol (5ml) and methylene chloride (10ml), and 18 (350mg) was added to this suspension. The mixture was stirred for 6 h and left to stand overnight at room temperature. After dilution with methylene chloride, the mixture was washed with water, dried over $MgSO_4$ and evaporated to give a partially crystallized oil. After removing the crystals by filtration and washing with ethyl acetate, the combined filtrate and washing were evaporated to give an oil which was charged to a silica gel column. The fraction eluted with ethyl acetate-benzene (2:1) gave 11b (318mg). Yield 56%. $[\alpha]_D$ +30.8° (c=2.2, $CHCl_3$).

3-(S)-BROMO-4-HYDROXYBUTANAMIDE (21).

To a suspension of 20 (7g) in methanol (100ml) was added a solution of diazomethane in ether until the reaction mixture became yellow. The solvent was evaporated under reduced pressure and tetrahydrofuran (70ml) was added to the residue. To the resulting solution, lithium aluminum hydride (1.4g) was added in small portions. After being stirred for 3 h, the reaction was quenched with diluted hydrochloric acid. Two layers were separated and the aqueous layer was extracted with ethyl acetate. The tetrahydrofuran layer and ethyl acetate layer were combined, washed with brine, dried over $MgSO_4$ and evaporated to give crude 21 (4.3g) as an oil which could be used in the next reaction without purification. Yield 66%. NMR(CD_3OD) 2.61(1H, dd, J=16, 9Hz), 2.98(1H, dd, J=16, 6Hz), 3.6-3.8(2H, m), 4.1-4.5(1H, m). IR(neat) 3350, 3200, 1660, 1610cm^{-1}.

3-(S)-BROMO-4-METHANESULFONYLOXYBUTANAMIDE (22).

To a solution of 21 (570mg) in tetrahydrofuran (10ml), were added methanesulfonyl chloride (0.49ml) and triethylamine (0.88ml) at 0°C and the mixture was stirred for 1 h at room temperature. The reaction mixture was poured into water and extracted with ethyl acetate. The extract was washed with brine, dried over $MgSO_4$ and evaporated to give an oil which was loaded to a silica gel column. The fraction eluted with ethyl acetate-hexane (10:1) gave 22 (443mg) as an oil. Yield 54%. NMR(CD_3OD) 2.5-3.0(2H, m), 3.12(3H, s), 4.4-4.6(2H, m), 4.6-4.8(1H, m).

3-(S)-BROMO-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (24b).

To a solution of 22 (123mg) in tetrahydrofuran (1.5ml), borane-tetrahydrofuran complex (1M tetrahydrofuran solution, 1.4ml) was added and refluxed for 3 h. The

solvent was then evaporated under reduced pressure. To the resulting residue, methanol (2ml) was added and the resulting mixture was refluxed for 30 min. The solvent was removed by evaporation and then ethanol (3ml) and triethylamine (0.14ml) were added. After being stirred for 1 week at room temperature, triethylamine (0.07ml) and p-nitrobenzyloxycarbonyl chloride (108mg) were added. The mixture was stirred for 10 min at room temperature, diluted with ethyl acetate, washed with water and brine, dried over $MgSO_4$ and evaporated to give a residue which was loaded to a silica gel column. The fraction eluted with hexane-ethyl acetate (1:1) gave 24b (88.2mg) as colorless crystals, mp 77.5-79°C. Yield 54%. NMR($CDCl_3$) 2.1-2.6(2H, m), 3.4-4.0(4H, m), 4.3-4.7(1H, m), 5.21(2H, s), 7.46,8.15(4H, A_2B_2, J=9Hz). IR($CHCl_3$) 1700, 1610, 1525cm^{-1}. $[\alpha]_D$ -32° (c=1.5, $CHCl_3$). Anal. Calcd for $C_{12}H_{13}N_2O_4Br$: C, 43.47; H, 3.98; N, 8.51; Br, 24.48. Found: C, 43.49; H, 3.91; N, 8.65; Br, 24.37.

3-(R)-ACETYLTHIO-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (9b) from 24b.
To a suspension of sodium hydride (55% oil dispersion, 18mg) in dimethylformamide (0.5ml), thiolacetic acid (0.05ml) was added at 0°C under nitrogen atmosphere. After being stirred for 10 min at the same temperature, a solution of 24 (68.2mg) in dimethylformamide (0.5ml) was added to the reaction mixture and refluxed for 3 h. After cooling, the reaction mixture was diluted with methylene chloride and washed with water and brine, dried over $MgSO_4$ and evaporated. The residue was loaded to a silica gel column. The fraction eluted with hexane-ethyl acetate (1:1) gave 9b (59.4mg). Yield 88%. $[\alpha]_D$ +25.4° (c=1, $CHCl_3$).

2-(S)-BROMO-1,4-DIMETHANESULFONYLOXYBUTANE (28).
Under nitrogen atmosphere, borane-tetrahydrofuran complex (1M tetrahydrofuran solution, 75ml) was added dropwise to a solution of 26 (4.9g) in tetrahydrofuran (50ml) at 0°C. The reaction mixture was then stirred for 5 h at room temperature. After the addition of methanol (10ml), the reaction mixture was refluxed for 1 h and evaporated to give diol (27). According to its NMR spectrum the resulting 27 was almost pure and could be used in the next reaction without purification. The compound 27 thus obtained was dissolved in methylene chloride (100ml) and stirred with methanesulfonyl chloride (5.8ml) and triethylamine (10.5ml) at room temperature for 1.5 h. The mixture was poured into water and extracted with methylene chloride. The extract was washed with water, dried over

$MgSO_4$ and evaporated to give an oil which was purified by silica gel column. The fraction eluted with benzene-ethyl acetate (1:1) gave dimesylate 28 (6.28g). Yield 78%.

27: NMR($CDCl_3$) 1.9-2.3(2H, m), 3.6-4.0(4H, m), 4.0-4.5(1H, m).

28: NMR($CDCl_3$) 2.0-2.5(2h, m), 3.03(3H, s), 3.07(3H, s), 4.0-4.6(4H, m).

3-(S)-BROMO-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (29b).

To a suspension of 28 (590mg) in ammonia saturated ethanol (30ml), was added triethylamine (0.23ml) and the mixture was stirred for 5 days at room temperature. The resulting solution was then evaporated under reduced pressure. The residue was dissolved in ethanol (3ml) and methylene chloride (6ml), and stirred with 18 (430mg) for 5 h at room temperature. The mixture was diluted with methylene chloride, washed with water, dried over $MgSO_4$ and evaporated under reduced pressure. The residue thus obtained was purified by silica gel column chromatography. The fraction eluted with ethyl acetate-hexane (4:1) was collected to give 29b (399mg) as an oil. Yield 60%. NMR($CDCl_3$) 2.0-2.6(2H, m), 2.26 and 2.33(in total 3H, each s), 3.4-4.3(4H, m), 4.3-4.8(1H, m), 5.23(2H, s), 7.60,8.20(4H, A_2B_2, J=8.5Hz). IR($CHCl_3$) 1670, 1610, 1560, 1525cm^{-1}.

3-(R)-ACETYLTHIO-1-(N-p-NITROBENZYLOXYCARBONYLACETIMIDOYL)PYRROLIDINE (12b) from 29b.

Under nitrogen atmosphere, thiolacetic acid (0.1ml) was added to a suspension of sodium hydride (55% oil dispersion, 35mg) in dimethylformamide (10ml) at 0°C. A solution of 29b in dimethylformamide (2ml) was added to the mixture, and stirred for 3 h at 55°C. The resulting mixture was diluted with ethyl acetate, washed with water, dried over $MgSO_4$ and evaporated to give an oil. This oil was purified by silica gel column chromatography. The fraction eluted with benzene-ethyl acetate (2:1) was collected to give 12b (212mg) as an oil. Yield 80%. $[\alpha]_D$ +28.5° (c=2.4, $CHCl_3$).

(2R)-4-METHANESULFONYLOXY-1,2-EPOXYBUTANE (31).

Under nitrogen atmosphere, borane-tetrahydrofuran complex (1M tetrahydrofuran solution, 15ml) was added dropwise to a solution of 26 (0.98g) in tetrahydrofuran (10ml) at 0°C. The mixture was then stirred for 5 h at room temperature. After the addition of aqueous NaOH (0.8g NaOH in H_2O 2ml), the mixture was refluxed for

3 h. After cooling, anhydrous K_2CO_3 was added to the mixture to absorb the water. The organic layer was separated by decantation and evaporated under reduced pressure. The resulting oil was dissolved in methylene chloride (20ml) and treated with methanesulfonyl chloride (1ml) and triethylamine (2ml) for 30 min at room temperature. The mixture was poured into water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated to give an oil which was purified by silica gel column chromatography. The fraction eluted with benzene-ethyl acetate (1:1) gave 31 (0.77g) as an oil. Yield 93%. NMR($CDCl_3$) 2.49(1H, dd, J=4, 2Hz), 2.78(1H, t, J=4Hz), 2.95(1H, m), 3.07(3H, s), 4.29(2H, t, J=6Hz). $[\alpha]_D$ +27.4° (c=0.1, $CHCl_3$).

3-(R)-HYDROXY-1-p-NITROBENZYLOXYCARBONYLPYRROLIDINE (7a) from 31.
Alumina B (Super I) was added to a solution of 31 (356mg) in methanol (70ml). Ammonia was bubbled through the mixture for 5 h, after which it was stirred overnight at room temperature. After removal of alumina by filtration, the filtrate was concentrated under reduced pressure. The residue was dissolved in methylene chloride (20ml) and treated with p-nitrobenzyloxycarbonyl chloride (500mg) and triethylamine (0.33ml) at 0°C for 30 min. The resulting solution was poured into ice water and extracted with methylene chloride. The extract was washed with water, dried over $MgSO_4$ and evaporated under reduced pressure. The residue was purified by silica gel column chromatography, eluted with benzene-ethyl acetate, and recrystallized from benzene-ethyl acetate to give 7a (160mg), mp 99-100°C. Yield 28%. $[\alpha]_D$ -18.5° (c=1, $CHCl_3$).

REFERENCES

1. H.Kropp, J.S.Kahan, F.M.Kahan, J.Sandelof, G. Darland and J.Birnbaum, Abstract of 16th Intersci.Conf.Antimicrob.Agent and Chemother., P. 228, Chicago, 1976.
2. T.Miyadera, Y.Sugimura, T.Hashimoto, T.Tanaka, K.Iino, T.Shibata and S.Sugawara, J.Antibiotics, 36, 1034(1983).
3. T.Suyama and S.Kanao, Yakugakuzasshi, 85, 531(1965).
4. M.M.Bowers Nemia, J.Lee and M.M.Joullie, Synth.Commun., 13, 1117(1983).
5. D.Danneels, M.Anteunis, L.V.Acker and D.Tavernier, Tetrahedron, 31, 327(1975).
6. O.Mitsunobu, Synthesis, 1981, 1.
7. S.Hanessian, A.Ugolini, D.Dube and A.Glamyan, Can.J.Chem., 62, 2146(1984).
8. This new amidine formation reaction was developed by M. Hashimoto et al. M.

Hashimoto, Y.Okada, H.Yoshihara, T.Takebayashi, Y.Eda snd Y.Osanai (Sankyo Co., Ltd.), Jpn. Kokai Tokkyo Koho, JP 59,212460(1984).

9. S.Kallenberg, Chem.Ber., 50, 90(1917).

10. P.Walden, Chem.Ber., 28, 2766(1895).

Received, 18th December, 1985