OCH_3), 6.52 (1H, s, 3-H), 6.79 (1H, s, 8-H), 8.02 (1H, s, 11-H). The NMR data was well consistent with that reported in the literature.[4b]

(±)-10-Hydroxy-1,2,9-trimethoxyaporphine (XIVa)——XIIIa (72 mg) was treated with 2% $PdCl_2$ (1 ml) and active carbon (50 mg) in CH_3OH (15 ml) according to the procedure given for XIIa to afford colorless crystals (XIVa) (50 mg, 88%), mp 180—182° (decomp.). An analytical sample had mp 184.5—185.5° (ether–acetone) (lit.[9] mp 140—145°). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3530 (OH). NMR δ: 2.55 (3H, s, NCH_3), 3.64, 3.82, 3.86 (each 3H, $3\times OCH_3$), 6.58 (1H, s, 3-H), 6.77 (1H, s, 8-H), 8.03 (1H, s, 11-H). *Anal.* Calcd. for $C_{20}H_{23}O_4N$: C, 70.36; H, 6.79; N, 4.10. Found: C, 70.05; H, 6.78; N, 4.13. The NMR data was consistent with that reported in the literature.[9]

(±)-N-Methyllaurotetanine (XIVb)——XIIIb (540 mg) was reduced with 2% $PdCl_2$ (5 ml) and active carbon (250 mg) in CH_3OH (100 ml) according to the procedure given for XIIa to afford colorless crystals (XIVb) (quantitative), mp 138—142°, which were recrystallized from ether–acetone to give colorless prisms (278 mg, 65%), mp 142—144°. Further recrystallization from acetone yielded colorless prisms, mp 144—145° (lit.[5] oil or powder). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3530 (OH). NMR δ: 2.37 (3H, s, NCH_3), 3.62 (3H, s, OCH_3), 3.83 (6H, s, $2\times OCH_3$), 6.57 (1H, s, 3-H), 6.78 (1H, s, 8-H), 8.06 (1H, s, 11-H). *Anal.* Calcd. for $C_{20}H_{23}O_4N$: C, 70.36; H, 6.79; N, 4.10. Found: C, 70.05; H, 6.84; N, 4.00. Oxalate; colorless prisms, mp 201—203° (decomp.) (acetone–CH_3OH) (lit. mp 212° (decomp.),[5a] 199—202° (decomp.)[5b]). The NMR data was consistent with that reported in the literature.[5b]

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Synthesis of Stereoisomeric Alanine Containing Peptide Derivatives[1]

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As analogues of D-cycloserine, the part structure of penicillin and D–D carboxypeptidase substrate of peptidoglycan of bacterial cell wall, D-alanine derivatives and their stereoisomers and D-alanyl-D-alanine derivatives and their stereoisomers (R_1-Ala-R_2, R_1-Ala-Ala-R_2: R_1=Z, H; R_2=$NHNH_2$, $NHCH_3$, $NHCH_2CH_2OH$) were synthesized. All compounds obtained did not show any antibacterial activity against *Staphylococcus aureus*, *Sarcina lutea*, *Pseudomonas aeruginosa* and *Escherichia coli*.

Some antibiotics inhibit the growth of bacteria due to interfering the synthesis of peptidoglycan of the cell walls.[3] It is well known that the structure of D-cycloserine is similar to that of D-alanine[4] and the highly reactive CO–N bond in the β-lactam ring of penicillin is the analogue of the CO–N bond in the peptide, D-alanyl-D-alanine.[5,6]

Our studies were directed to the synthesis of D-alanyl-D-alanine analogues which are analogues not only of D-cycloserine and penicillin but also of the substrates of D-D carboxypepti-

1) Abbreviations used are those recommended by IUPAC-IUB Commission on Biochemical Nomenclature: *Biochemistry*, **5**, 3485 (1966); *ibid.*, **6**, 362 (1967); *ibid.*, **11**, 1726 (1972). Z=benzyloxycarbonyl.
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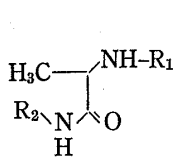
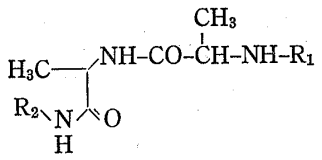


1a : R_1=Z, R_2=NH_2
1b : R_1=H, R_2=NH_2
1c : R_1=Z, R_2=CH_3
1d : R_1=H, R_2=CH_3
1e : R_1=Z, R_2=CH_2CH_2OH
1f : R_1=H, R_2=CH_2CH_2OH



2a : R_1=Z, R_2=NH_2
2b : R_1=H, R_2=NH_2
2c : R_1=Z, R_2=CH_3
2d : R_1=H, R_2=CH_3
2e : R_1=Z, R_2=CH_2CH_2OH
2f : R_1=H, R_2=CH_2CH_2OH

Chart 1

dase-transpeptidase in bacterial cell wall. The present report describes the synthesis of stereoisomeric alanine derivatives (**1a, b, c, d, e, f**) and (**2a, b, c, d, e, f**) shown in Chart 1 and alanylalanine (**2i**) to study their antibacterial activity.

N^{α}-Benzyloxycarbonylalanine hydrazide (**1a**)[7] was synthesized by the hydrazinolysis of N^{α}-benzyloxycarbonylalanine methyl ester which was made from N^{α}-benzyloxycarbonylalanine and diazomethane. N^{α}-Benzyloxycarbonylalanine methylamide (**1c**) and N^{α}-benzyloxycarbonylalanine ethanolamide (**1e**) were synthesized by *p*-nitrophenyl ester method[8] from N^{α}-benzyloxycarbonylalanine *p*-nitrophenyl ester and methylamine and monoethanolamine respectively. Debenzyloxycarbonylation was performed by hydrogenolysis over Palladium catalyst to form alanine hydrazide (**1b**), alanine methylamide (**1d**) and alanine ethanolamide (**1f**). N^{α}-Benzyloxycarbonylalanylalanine hydrazide (**2a**)[9] was prepared by hydrazinolysis of the corresponding methyl ester (**2g**)[10–12] from which alanylalanine (**2i**)[13] was prepared through N^{α}-benzyloxycarbonylalanylalanine (**2h**).[14] Elongation of (**1d**) and (**1f**) was performed by *p*-nitrophenyl ester method to give N^{α}-benzyloxycarbonylalanylalanine methylamide (**2c**) and N^{α}-benzyloxycarbonylalanylalanine ethanolamide (**2e**) respectively. Debenzyloxycarbonylation was performed by the same method as described above to give alanylalanine hydrazide (**2b**), alanylalanine methylamide (**2d**) and alanylalanine ethanolamide (**2f**).

All compounds obtained above did not show any antibacterial activity against gram positive organisms, *Staphylococcus aureus* and *Sarcina lutea* and gram negative organisms, *Escherichia coli* and *Pseudomonas aeruginosa* in the concentration of 100 μg/ml.

Synthesis of other alanine containing peptide derivatives is under way in our laboratory.

Experimental

All melting points were taken by the capillary method and are uncorrected. Optical rotations were determined in an automatic polarimeter Model DIP-180 (Japan Spectroscopic Co., Ltd.). Thin-layer chromatography was performed on silica gel (Kieselgel G. Merck). *Rf* values refer to the following solvent systems: Rf_1 *n*-BuOH–AcOH–H_2O (4:1:5), Rf_2 *n*-BuOH–pyridine–AcOH–H_2O (4:1:1:2), Rf_3 *n*-BuOH–AcOH–17% ammonia (11:6:3).

N^{α}-Benzyloxycarbonylalanine Methylamide (1c)——To the solution of methylamine (prepared from 2.72 g of methylamine hydrochloride and 40 ml of 1 N NaOH) in dimethylformamide (DMF) (50 ml) was added 6.88 g of N^{α}-benzyloxycarbonylalanine *p*-nitrophenyl ester and this reaction mixture was stirred over night at room temperature. After evaporation of the solvent, the residue was extracted with AcOEt, which was

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washed with 5% Na_2CO_3, 1 N HCl and water, dried over Na_2SO_4 and evaporated. To the residue, ether was added to form crystalline, which was recrystallized from AcOEt and ether; yield L, 1.3 g (28%); D, 2.8 g (59%), mp L, 127—128°; D, 126—127°, $[\alpha]_D^{23}$ L, −13.0° (c=1.0, MeOH); D, +11.9° (c=1.0 MeOH). *Anal.* Calcd. for $C_{12}H_{16}O_3N_2$: C, 61.0; H, 6.83; N, 11.9. Found: L, C, 61.0; H, 6.74; N, 11.9; D, C, 61.3; H, 6.88; N, 12.0.

N$^{\alpha}$-Benzyloxycarbonylalanine Ethanolamide (1e)——To 2.44 g of ethanolamine in DMF (20 ml) was added 6.88 g of N$^{\alpha}$-benzyloxycarbonylalanine p-nitrophenyl ester and the reaction mixture was stirred over night at room temperature. After removal of the solvent, the residue was extracted with AcOEt, which was washed with 1 N HCl, 5% Na_2CO_3 and water, dried over Na_2SO_4 and evaporated to form white crystalline. This was recrystallized from AcOEt and ether; yield L, 1.8 g (34%); D, 1.6 g (30%), mp L, 115—116°; D, 113—115°, $[\alpha]_D^{23}$ L, −11.8° (c=1.0, MeOH); D, +11.3° (c=1.0, MeOH). *Anal.* Calcd. for $C_{13}H_{18}O_4N_2$: C, 58.6; H, 6.81; N, 10.5. Found: L, C, 58.8; H, 6.94; N, 10.6; D, C, 59.1, H, 7.07; N, 10.7.

N$^{\alpha}$-Benzyloxycarbonylalanylalanine Methylamide (2c)——Alanine methylamide (**1d**) prepared from 0.41 g of (**1c**) in 15 ml of DMF was stirred with 0.83 g of N$^{\alpha}$-benzyloxycarbonylalanine p-nitrophenyl ester over night at room temperature. After removal of DMF, to the residue was added AcOEt to form crystalline, which was recrystallized from MeOH and AcOEt; yield L-L, 0.5 g (83%); L-D, 0.5 g (83%); D-L, 0.5 g (83%); D-D, 0.5 g (83%), mp L-L, 201—202°; L-D, 207—208°; D-L, 208—209°; D-D, 198—199°, $[\alpha]_D^{23}$ L-L, −35.7° (c=1.0, MeOH); L-D, +9.4° (c=1.0, MeOH); D-L, −10.4° (c=1.0, MeOH); D-D, +33.5° (c=1.0, MeOH). *Anal.* Calcd. for $C_{15}H_{21}O_4N_3$: C, 58.6; H, 6.89; N, 13.7. Found: L-L, C, 58.4; H, 6.81; N, 13.6, L-D, C, 58.7; H, 6.96; N, 13.6, D-L, C, 58.6; H, 6.80; N, 13.7, D-D, C, 58.9; H, 7.02; N, 13.4.

N$^{\alpha}$-Benzyloxycarbonylalanylalanine Ethanolamide (2e)——Alanine ethanolamide (**1f**) obtained from 0.53 g of (**1e**) in 15 ml of DMF was stirred with N$^{\alpha}$-benzyloxycarbonylalanine p-nitrophenyl ester (0.83 g) at room temperature over night. After removal of DMF, to the residue AcOEt was added to give solid material, which was collected by filtration and recrystallized from MeOH and AcOEt; yield L-L, 0.60 g (89%); L-D, 0.60 g (89%); D-L, 0.60 g (89%); D-D, 0.60 g (89%), mp L-L, 183—185°; L-D, 158—160°; D-L, 158—160°; D-D, 184—185°, $[\alpha]_D^{23}$ L-L, −33.5° (c=1.0, MeOH), L-D; +10.4° (c=1.0, MeOH); D-L, −9.3° (c=1.0, MeOH); D-D, +34.5°, (c=1.0, MeOH). *Anal.* Calcd. for $C_{16}H_{23}O_5N_3$: C, 57.0; H, 6.87; N, 12.5. Found: L-L, C, 57.4; H, 7.10; N, 12.5, L-D, C, 57.5; H, 7.10; N, 12.5, D-L, C, 57.5; H, 7.04; N, 12.4, D-D, C, 57.2; H, 7.09; N, 12.5.

TABLE I. Physical Properties and *Rf* Values of Thin-Layer Chromatography

No.	Compound	mp (°C)	$[\alpha]_D^{23}$	Yield (%)	Rf_1	Rf_2	Rf_3
1-b_1	H-AlaNHNH$_2$·2HCl	—	+16.76 (H_2O)	82	0.20	0.45	0.39
1-b_2	-D-	—	−17.67 (H_2O)	75	0.20	0.45	0.40
1-d_1	H-AlaNHCH$_3$·HCl	—	+16.25 (H_2O)	89	0.35	0.63	0.50
1-d_2	-D-	—	−17.54 (H_2O)	87	0.35	0.63	0.50
1-f_1	H-AlaNHCH$_2$CH$_2$OH	86—87	+ 6.79 (MeOH)	88	0.27	0.57	0.55
1-f_2	-D-	87—88	− 8.00 (MeOH)	84	0.27	0.57	0.55
2-b_1	H-Ala·AlaNHNH$_2$	140—142	−42.17 (MeOH)	80	0.05	0.45	0.36
2-b_2	-L- -D-	126—127	+52.17 (MeOH)	70	0.05	0.49	0.42
2-b_3	-D- -L-	124—125	−52.60 (MeOH)	74	0.05	0.50	0.42
2-b_4	-D- -D-	139—140	+41.79 (MeOH)	80	0.05	0.45	0.36
2-d_1	H-Ala·AlaNHCH$_3$	116—117	−30.81 (MeOH)	54	0.15	0.60	0.51
2-d_2	-L- -D-	138—139	+43.63 (MeOH)	76	0.15	0.65	0.51
2-d_3	-D- -L-	136—137	−42.61 (MeOH)	76	0.15	0.65	0.51
2-d_4	-D- -D-	115—117	+29.88 (MeOH)	61	0.15	0.60	0.51
2-f_1	H-Ala·AlaNHCH$_2$CH$_2$OH	139—140	−27.33 (MeOH)	83	0.17	0.57	0.51
2-f_2	-L- -D-	157—158	+34.81 (MeOH)	83	0.17	0.61	0.45
2-f_3	-D- -L-	156—157	−34.05 (MeOH)	83	0.17	0.61	0.45
2-f_4	-D- -D-	140—141	+26.22 (MeOH)	83	0.17	0.57	0.51

General Procedure for Debenzyloxycarbonylation of N-Protected Derivatives——N$^{\alpha}$-Benzyloxycarbonyl derivatives in MeOH were hydrogenated over Palladium catalyst. After removal of Palladium, the solvent was evaporated to dryness followed by the addition of ether or lyophilization of their 1 N HCl solution to give desired compounds.

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