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An Acyl Migration with Oxalic Acid

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In the past few years, the structures of a number of new antibiotics have been reported in the literature; all have had the common feature of a lactone that is formed from the carboxyl function of an amino acid and the hydroxyl group of a hydroxy-amino acid.

For the synthesis of these peptide lactone antibiotics, it is necessary to synthesize a chain that will include the desired ester bond. As a preliminary investigation for this purpose, we have first prepared an ester chain, namely, *O*-(*t*-butyloxycarbonyl-L-phenylglycyl)-*N*-benzyloxycarbonyl-L-threonine (I), which has already been reported by Ondetti and Thomas.[1)]

The coupling of *t*-butyloxycarbonyl-L-phenylglycine and *N*-benzyloxycarbonyl-L-threonine was carried out by the method reported by Ondetti and Thomas; then the reaction mixture was separated by Sephadex LH-20, using chloroform as the elution solvent. By this separation method, we easily obtained the desired ester chain as an oily substance, which was then crystallized as dicyclohexylammonium salt (II) by treatment with dicyclohexylamine in an acetone - hexane solution.

The values of the physical constant and the spectroscopic data of this dicyclohexylammonium salt (II) were identical with those of the dicyclohexylammonium salt of I reported in the literature.[1)] From this finding, it is obvious that the substance containing the ester bond obtained here was the compound I. For the purification of this salt, II, it was dissolved in acetone while being heated and then allowed to stand for several hours at room temperature; this gave crystals which melted at 154—155°C, but its IR spectrum showed that this product was not the ester-bond compound, but a compound resulting from the *O,N* acyl migration. This migration has also been seen when II was dissolved in chloroform or benzene without heating and allowed to stand at room temperature. We have assumed that this *O,N* acyl migration product was *t*-butyloxycarbonyl-L-phenylglycyl-*N*-benzyloxycarbonyl-L-threonine (III), which was obtained in this case as its dicyclohexylammonium salt (IV); this structure was confirmed by synthesis, as is shown in Fig. 3.

The salt IV was treated with equal moles of oxalic acid in acetone, and then the deposited ammonium salt of oxalic acid was filtered and the acetone was removed; this left an oily substance which was crystallized as its salt in a 96% yield by the addition of dicyclohexylamine. Since the

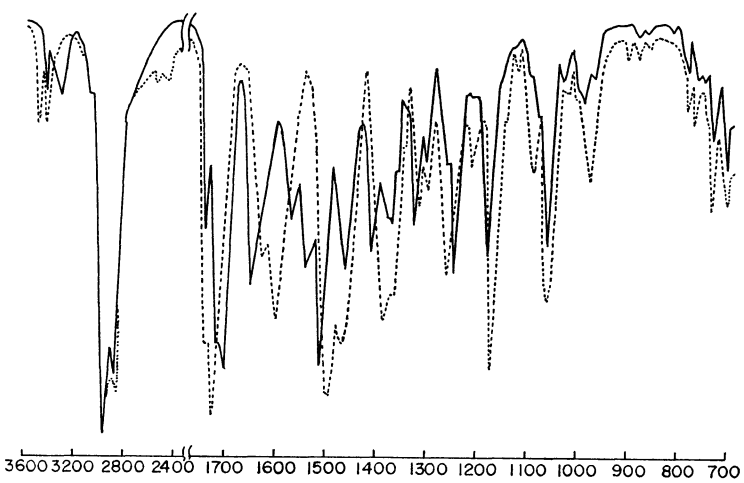

Fig. 1. Infrared spectra.
—— (II) ------ (IV)

1) M. A. Ondetti and P. L. Thomas, *J. Amer. Chem. Soc.*, **87**, 4373 (1965).

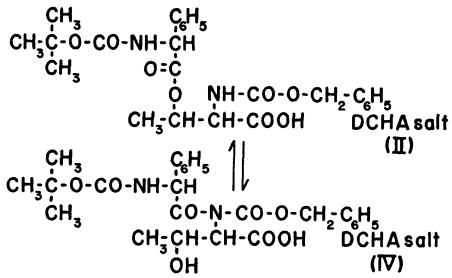

Fig. 2

IR spectrum of this salt was identical with that of the salt II, it is clear that the *N,O* acyl migration had occurred upon the treatment of the oxalic acid. Though the melting point and the specific rotation value of the salt obtained by the *N,O* acyl migration are higher than the values of the salt II, it is possible, in view of the value of specific rotation, that the purity of the former compound is slightly higher.

In the present case it was also found that the *N,O* acyl migration with oxalic acid did not affect the *N*-protecting *t*-butyloxycarbonyl- and benzyloxycarbonyl- groups and that no racemization occurred.

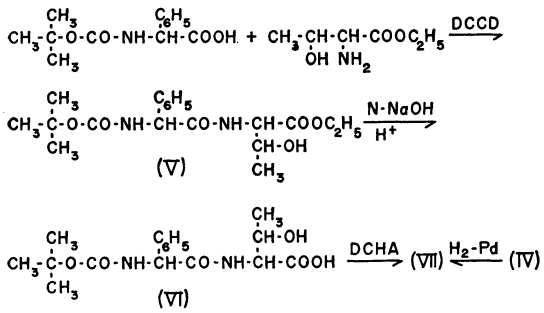

Fig. 3

t-Butyloxycarbonyl-L-phenylglycyl-L-theronine (VI) was prepared in the following way. *t*-Butyloxycarbonyl-L-phenylglycyl-L-threonine ethyl ester (V), obtained by the coupling of *t*-butyloxycarbonyl-L-phenylglycine and L-threonine ethyl ester in the presence of dicyclohexylcarbodiimide in acetonitrile, was hydrolyzed by 1N sodium hydroxide at room temperature to give VI. The dicyclohexylammonium salt of VI was found to be identical with the debenzyloxycarbonylation product of IV in melting point, IR spectra, and specific rotation value.

Experimental

***O*-(*t*-Butyloxycarbonyl-L-phenylglycyl)-*N*-benzyloxycarbonyl-L-threonine Dicyclohexylammonium Salt (II).** *t*-Butyloxycarbonyl-L-phenylglycine (920 mg, 3.5 mmol) and *N*-benzyloxycarbonyl-L-threonine (1.75 g, 3.5 mmol) and *N*-benzyloxycarbonyl-L-threonine (1.75 g, 7 mmol) were treated by the method reported in the literature.[1] The crude reaction product thus obtained showed two spots (R_f 0.79 and 0.41) on thin-layer chromatography. This mixture was separated by Sephadex LH-20, using chloroform as the elution solvent. The compound, with an R_f value of 0.41, which was shown to be the desired product, was isolated from acetone - hexane in the form of a crystalline dicyclohexylammonium salt. Yield, 640 mg (38%); mp 146.5—149°C; $[\alpha]_D^{21}$ +34.0° (*c* 0.838, DMF) (lit. mp 145—147°C; $[\alpha]_D^{28}$ +35.1°).

Found: C, 66.48; H, 8.08; N, 6.38%. Calcd for $C_{37}H_{53}O_8N_3$: C, 66.57; H, 7.95; N, 6.30%.

***t*-Butyloxycarbonyl-L-phenylglycyl-*N*-benzyloxycarbonyl-L-threonine Dicyclohexylammonium Salt (IV).** The salt II (76.8 mg, 0.114 mmol) was dissolved in hot acetone (1 m*l*) and then allowed to stand for several hours at room temperature. By the addition of hexane to this solution, crystals of IV were precipitated. Yield, 69.5 mg (90%); mp 154—155°C; $[\alpha]_D^{25}$ +41.1° (*c* 0.953, DMF).

Found: C, 66.22; H, 8.26; N, 6.51%. Calcd for $C_{37}H_{53}O_8N_3$: C, 66.57; H, 7.95; N, 6.30%.

Migration of IV to II with Oxalic Acid. To a solution of IV (188 mg, 0.282 mmol) in hot acetone (1 m*l*), we added oxalic acid-$2H_2O$ (35.5 mg, 0.282 mmol); the solution was then refluxed for 5 min and subsequently left standing for 2 hr at room temperature. A dicyclohexylammonium salt of oxalic acid thus deposited was filtered off, the mother liquor was removed *in vacuo*, and the residue was dissolved in ethyl acetate. After the filtration of an insoluble oxalic acid, the solvent was removed and the residual oil was again dissolved in 0.5 m*l* of acetone. To this solution dicyclohexylamine (0.05 m*l*) in hexane was then added. After a short time, crystalline salts were obtained. Yield, 181 mg (96.2%); mp 154—155°C; $[\alpha]_D^{25}$ +40.1° (*c* 0.983, DMF). The IR spectrum of this salt was identical with that of II, and a mixed-melting-point determination with IV was depressed to 151—152°C.

***t*-Butyloxycarbonyl-L-phenylglycyl-L-threonine Ethyl Ester (V).** *t*-Butyloxycarbonyl-L-phenylglycine (200 mg, 0.8 mmol) and L-threonine ethyl ester (117 mg, 0.8 mmol) were dissolved in acetonitrile (4 m*l*) and cooled at −3°C. A solution of dicyclohexylcarbodiimide (165 mg, 0.8 mmol) in acetonitrile (2 m*l*) was added, with stirring and cooling below −1°C; stirring was continued for 3 hr at −1°C and then for 7 hr at room temperature. The precipitate of dicyclohexylurea was filtered, and the filtrate was concentrated to dryness *in vacuo*. The oily product was dissolved in ethyl acetate; then the solution was washed with 3% hydrochloric acid, 3% sodium bicarbonate, and then with water, and dried over anhydrous sodium sulfate. The concentration of the solution *in vacuo* left a crystalline product which was recrystallized twice from ethyl acetate - petroleum ether. Yield, 170 mg (68%); mp 107—108.5°C; $[\alpha]_D^{20}$ +48.0° (*c* 1.03, abs. ethanol).

Found: C, 59.87; H, 7.45; N, 7.41%. Calcd for $C_{19}H_{28}O_6N_2$: C, 59.98; H, 7.42; N, 7.36%.

***t*-Butyloxycarbonyl-L-phenylglycyl-L-threonine (VI).** Into a solution of V (150 mg, 0.4 mmol) in ethanol (3 m*l*), 1N aqueous sodium hydroxide (0.4 m*l*, 0.4 mmol) was added with stirring. After the solution had stood at room temperature for 1.5 hr, 2 m*l* of water as

added to the reaction mixture and ethanol was removed *in vacuo*. The aqueous residue was treated with ethyl acetate, and the aqueous layer was adjusted to pH 3 to give an oily product. This product was extracted with ethyl acetate, and the solution was washed with water and dried over anhydrous sodium sulfate. The concentration of the solution *in vacuo* left a crystalline product (112 mg, 80%), which was subsequently recrystallized from ethyl acetate - petroleum ether. Yield, 98 mg (70%); mp 150—151°C; $[\alpha]_D^{26}$ +60.9° (*c* 1.04, abs. ethanol).

Found: C, 57.61; H, 6.77; N, 7.99%. Calcd for $C_{17}H_{24}O_6N_2$: C, 57.94; H, 6.87; N, 7.95%.

The dicyclohexylammonium salt (VII) of this compound (VI) was prepared in the usual way; mp 145—146°C; $[\alpha]_D^{25}$ +45.3° (*c* 1.03, DMF). No depression of the melting point was observed on admixture with the debenzyloxycarbonylation product of IV.