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Activation of the Carboxylic Acids by Anhydride Formation with *N*-Acyl-*N*-Alkyl Carbamic Acid[1]

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The mixed anhydrides with *N*-acyl carbamic acid (**IV**) were prepared by the reaction of carboxylic acids (**III**) with *N*-chlorocarbonyl lactams (**IIa**) or sec. amides (**IIb**). Decarboxylation of **IV** yielded *N*-acyl lactams (**VI**), while symmetrical anhydrides (**VII**) were obtained by the side reaction of **IV** with carboxylic acids (**III**). Aminolysis of **IV** yielded carboxylic acid amides (**IX**) in high yield and purity. **IVb** was used in the preparation of aminosubstituted beta-lactam antibiotics (**XII**), as a new way of activating *N*-protected phenylglycine (**IIIg**), during acylation of 6-APA or 7-ADCA (**XI**).

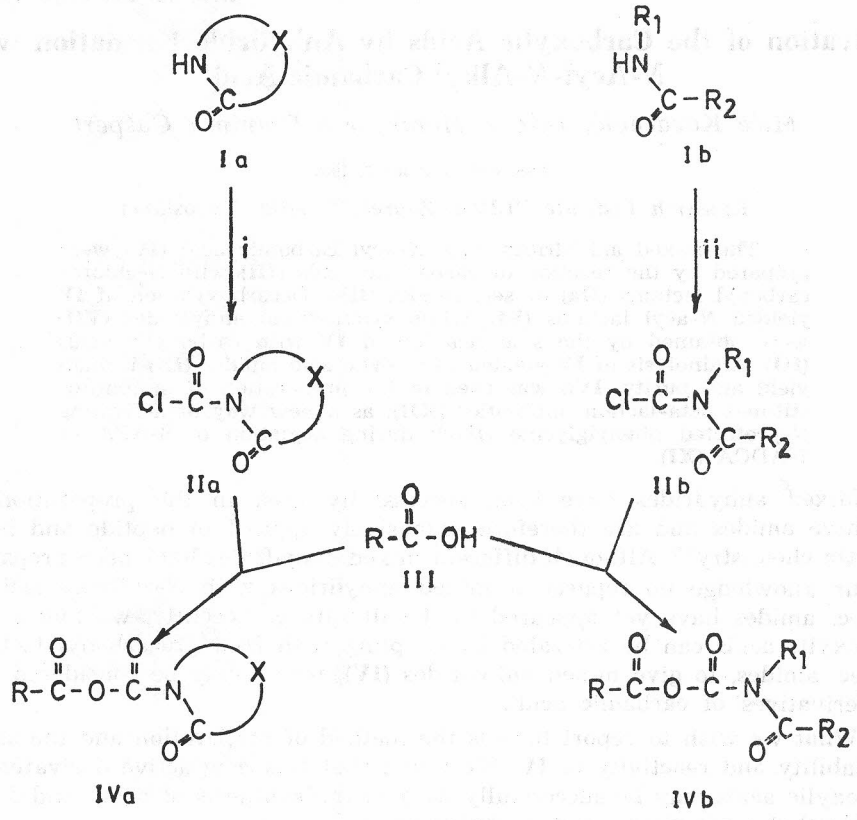
Mixed anhydrides have been successfully used in the preparation of sensitive amides and are therefore extensively applied in peptide and beta-lactam chemistry[2,3]. Although different mixed anhydrides have been prepared, to our knowledge no reports of mixed anhydrides with *N*-carboxy lactams or sec. amides have yet appeared in the literature. Recently, we found that carboxylic acids can be activated by coupling with *N*-chlorocarbonyl lactams or sec. amides, to give mixed anhydrides (**IV**), which may be considered also as derivatives of carbamic acid[4].

What we wish to report here is the method of preparation and the study of stability and reactivity of **IV**. We found that this new active derivative of carboxylic acids may be successfully used in the synthesis of acid amides and semisynthetic penicillins and cephalosporins.

The necessary *N*-chlorocarbonyl intermediates **II** can be prepared by the reaction of carbonyl dichloride with metal or silyl derivatives of lactams or sec. amides[5,6]. However, the procedure used in this paper is a slight modification of the described one, since it is more convenient to use triethylamine than to prepare metal or silyl derivatives[7]. All the new prepared *N*-chlorocarbonyl compounds show characteristic absorption bands in IR spectrum at 1790—1810 cm^{-1}, and are quite stable compounds, isolated from the reaction mixture in high yield.

The mixed anhydrides **IV** were prepared by the method similar to those used for the preparation of carbonic acid monoesters R—CO—O—CO—OR_1 (**V**)[8]. The carboxylic acid (**III**) is dissolved in an inert organic solvent in the presence of ter. base, cooled to 0 °C, and *N*-chlorocarbonyl intermediate **II** is added by stirring. After a few minutes amine hydrochloride starts to separate

SCHEME I



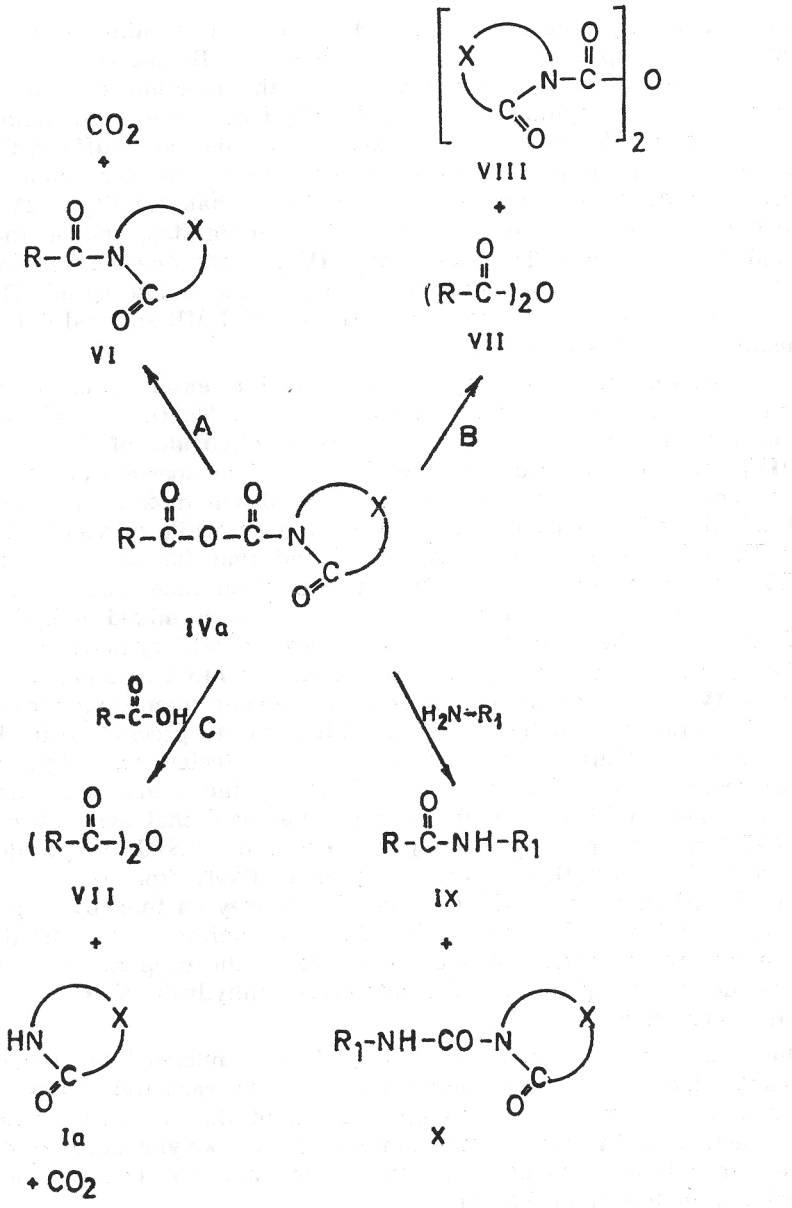
R = Ph-CH$_2$, R_1 = Ph , R_2 = CH$_3$

X = $(CH_2)_3$, $CH_2-CH-S-C(CH_3)_2-CH$ (N-$COCH_3$)

i = $COCl_2$ / TEA

ii = Cl-Si$(CH_3)_3$ / TEA, $COCl_2$

SCHEME II



R = $Ph-CH_2$, Ph-CH-NH-CO-O-CH_2-Ph, Ph-CH-NH-CO-O-CH_2-CCl_3

R_1= $Ph-CH_2$, Ph-CH-$COOCH_3$

X = $(CH_2)_3$, CH_2-CH-S-C$(CH_3)_2$-CH
N
$COCH_3$

and the mixed anhydride is formed during the next 30 minutes. This is substantiated by the appearance of the new bands in IR spectrum at 1815 and 1745 cm^{-1}, detected in a sample taken from the reaction solution. In most reactions with nucleophiles the mixed anhydrides are used immediately, without isolation. However, some mixed anhydrides are sufficiently stable at room temperature to be isolated by evaporation of a mother liquor, obtained after filtration of amine hydrochloride. It is found that stability of **IV** depends on the reaction conditions in the anhydride forming step, and on the nature of R and X substituents. The most stable **IV** was the one derived from phenylacetic acid and *N*-carboxybutyrolactam isolated as a viscous oil. The anhydride structure of **IV** was deduced from IR and ^1H NMR spectral data, as well by chemical reactions outlined in scheme II.

While studing the reactivity of **IV**, special attention was given to the reaction conditions which favour the side reactions. There is an obvious similarity in the side reactions between the mixed anhydrides of *N*-acyl carbamic acids (**IV**) and the anhydrides with carbonic acid monoesters (**V**). One of the common side reactions has been the loss of carbon dioxide. In the case of mixed anhydride **V**, decarboxylation gives an ester, while **IV** yields the *N*-acyl lactam **VI** (scheme II, pathway A). We found that formation of semicyclic imid **VI** occured in all cases in which the reaction time was extended over 30 minutes. Another side reaction, common to both mixed anhydrides (**IV** and **V**) has been the formation of the carboxylic acid symetrical anhydride **VII**. We observed that formation of **VII** was the favoured reaction when mixed anhydride **IV** was more soluble in reaction solvent than symmetrical anhydride **VII**, particularly in the case when **VII** starts to precipitate in the cours of the reaction. Thus in the reaction of *N*-protected phenylglycine with *N*-chlorocarbonyl butyrolactam during 2 hours, the symmetrical anhydride **VII** was formed in 50% yield. It has been suggested that symmetrical anhydride (**VII**) was formed by disproportionation of mixed anhydride **V**[9]. It seems to us that formation of **VII** results more likely from the side reaction of **IV** with carboxylic acid (**III**) (scheme II, pathway C) than by disproportionation of **V** (scheme II, pathway B). This assumption is supported by the evidence that we recovered 90% of lactam **Ia** in the preparation of **VII**, and failed to detect the presence of summetrical anhydride **VIII** or any coresponding derivatives.

The reactivity of mixed anhydride **IV** with nucleophiles revealed the significant selectivity of nucleophilic attack on the carbonyl group of carboxylic acid, with a minimum »wrong« opening of the anhydride bond. Thus, in the reaction of **IV** with simple amines, the carboxylic acid amides were obtained in 80—85% yields, the only side products being *N*-acyl urea (**X**), isolated in less than 5% yield.

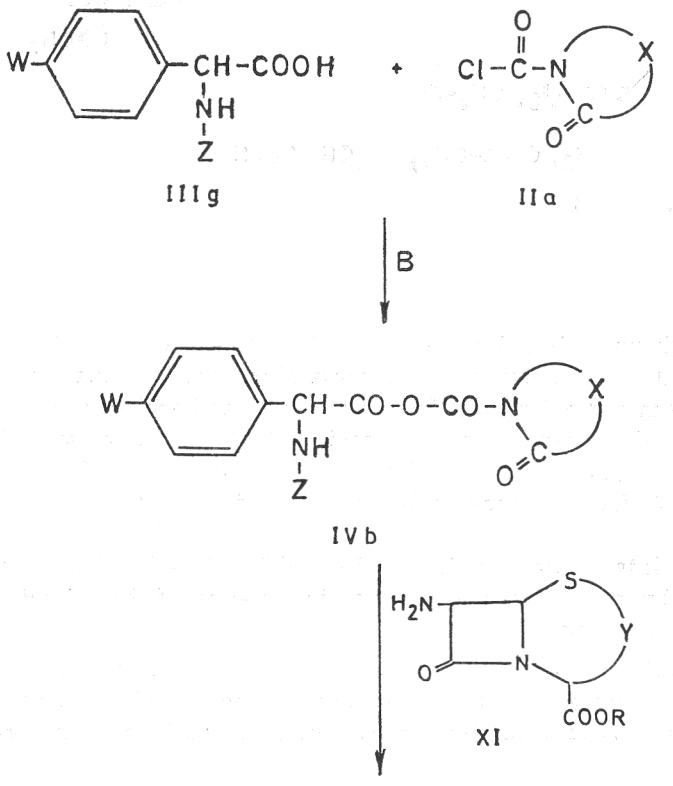
It seemed to us that **IV** might also prove to be a suitable way for the activation of aminoacids. Therefore, we prepared **IVb** with D-α-phenylglycine (**IIIg**; W=H, OH), which is incorporated in several clinically important beta-lactam antibiotics (ampicillin, amoxycillin, cephalexine and cephaloglycine)[4,10b].

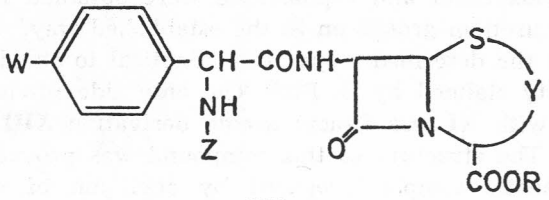
The preparation of *N,O*-protected semisynthetic penicillins and cephalosporins (**XII**) illustrates the use of **IV** as an activation form for *N*-protected phenylglycine with benzyl or trichloroethyl urethan as the amino protective

group. Ampicillin, amoxycillin and cephalexine were obtained from **XII** by removal of ester and urethan groups on in the established way.[3] All the final products, prepared in the described ways, were identical to standard samples with biological activity claimed by B. Ph.[17] The only side product observed in aminolysis of **IV** with **XI** was *N*-acyl ureido derivative **XIII**, isolated in less than 10% yield. The structure of this compound was proved by comparison with the authentic sample, prepared by acylation of 6-APA with *N*-chlorocarbonyl butyrolactam. It is noteworthy that no more racemisation of **IIIg** occured by activation v i a **IV**, than by other methods, since the final products obtained in this way had the same values of optical rotations as reported in the literature.

In the preparation of aminosubstituted penicillins an enamine group is used instead of urethan, since removal of this group is easier and has already occured during the isolation of the product. The procedure for the production of aminopenicillins is illustrated by the preparation of ampicillin and amoxycillin trihydrate. This procedure includes a treatment of Dane's salt of

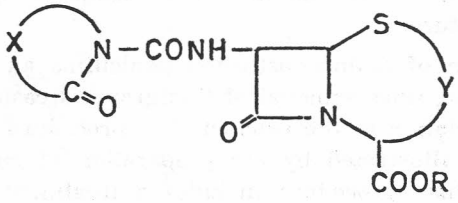
SCHEME III





XII

+



XIII

Z = Ph-CH_2-O-CO, Cl_3C-CH_2-O-CO, CH_3O-CO-CH=C-CH_3, H

X = $(CH_2)_3$, $(CH_2)_4$, $(CH_2)_5$, $(CH_2)_6$, CH_2-CH-S-$C(CH_3)_2$-CH (N—$COCH_3$)

Y = $C(CH_3)_2$, CH_2-C(=O)-CH_3

R = Ph-CH_2, Cl_3C-CH_2, Ph_2CH, Na, H

W = H, OH

D-α-phenylglycine (**IIIg**; Z=CH_3O—CO—CH=C(—CH_3)—) with *N*-chlorocarbonyl butyrolactam in cold dry acetone, containing catalytic quantity of pyridine, which gives mixed anhydride (**IVb**) in situ, and which is then treated with sodium salt of 6-aminopenicillanic acid (6-APA) in aq. acetone. Under these reaction conditions protection of the phenolic group in **IIIg** (W=OH) is not neccessary. A free aminopenicillin is obtained withous isolation of unstable *N*-protected intermediate (**XII**; Z=CH_3O—CO—CH=C(—CH_3)—) which is readily hydrolysed by cold diluted acid, prior to isolation of **XII** (Z=H, R=H) from the reaction solution.

The results on the application of mixed anhydride **IV** in the preparation of carboxylic acid amides are promising for the use of **IV** in the preparation of other carboxylic acid derivatives. Further work in this direction is in progress.

EXPERIMENTAL

Melting points are uncorrected. The IR spectra were recorded on a Perkin-Elmer Infracord Model 257 G. The UV spectra were recorded with SP8-100 Unicam spectrometer. The ^{1}H NMR spectra were done with an EM-360 Varian spectrometer unless otherwise stated, with TMS as the internal standard. Chemical shifts are given in ppm (δ).

TLC was conducted on original plates (Merck, Kieselgel HF_{254}) followed by detection with iodine vapours and water or with UV absorption in solvent systems as stated:

(A) benzene-acetone (9 : 1)
(B) methylene chloride- ether (4 : 1)

Optical rotations were measured on Carl Zeiss polarimeter POLAMAT.

General Method for the Preparation of N-*Chlorocarbonyl Lactams* (**IIa**)

A solution of lactam (0.1 mol), triethylamine (10.1 g, 0.1 mol) and toluene (80 ml) was cooled to 0 °C, whereupon a 20% solution (w/w) of $COCl_2$ in toluene (52 ml) was added over a period of 30 minutes. The reaction mixture was stirred for 1 hour and precipitated triethylamine hydrochloride was filtered off. The mother liquor was extracted with water (2 × 60 ml), dried ($MgSO_4$) and evaporated under reduced pressure.

N-Chlorocarbonyl Butyrolactam /**IIa**; $X = -(CH_2)_3-$/

Yield: 11.8 g (80%); colourless oil; b. p. 112—114 °C/ 0.13 Pa.

Anal. $C_5H_6NO_2Cl$ (147.56) calc'd: C 40.65; H 4.10; N 9.49; Cl 24.03%
found: C 40.55; H 4.43; N 9.71; Cl 25.20%

IR spectrum (film): 1810(vs), 1764(m), 1720(s), 1350(m), 1265(s), 1227(m), 1170(s), 1095(m) and 885(w) cm^{-1}.

^{1}H NMR spectrum ($CDCl_3$): 1.78—2.90/m, $(CH_2)_2CO$/, 3.65—4.35 /m, CH_2N/.

N-*Chlorocarbonyl Valerolactam* /**IIa**; $X = -(CH_2)_4-$/

Yield: 13.3 g (82.7%); colourless oil; b. p. 98—105°C/0.13 Pa (decomp.)
IR spectrum (CH_2Cl_2): 2960(m), 2880(w), 1800(s), 1720(vs), 1650(m), 1385(w), 1332(w), 1295(w), 1245(w), 1200(m), 1175(m), 1123(s), 985(w) and 950 (w) cm^{-1}.

N-*Chlorocarbonyl Caprolactam* /**IIa**; $X = -(CH_2)_5-$/

Yield: 12.8 g (73%); viscous oil; b. p. 88—91 °C /0.13 Pa (decomp.) Lit.[11] b. p. 97 °C /13 Pa.

IR spectrum (CH_2Cl_2): 2940(s), 1790(vs), 1715(vs), 1445(m), 1335(m), 1200(s), 1150(s), 1075(s), 970(s) and 900(m) cm^{-1}.

N-*Chlorocarbonyl Enantholactam* /**IIa**; $X = -(CH_2)_6-$/

Yield: 16.8 g (87.4%); viscous oil; b. p. 103—105 °C/ 0.13 Pa (decomp.)
IR spectrum (film): 2920(m), 1790(s), 1715(vs), 1440(m), 1360(m), 1330(m), 1225(m), 1190(s), 1115(m), 1082(m), 995(m) and 730(m) cm^{-1}.

N^3-*Chlorocarbonyl*-N^8-*acetyl-7,7-dimethyl-6-thia-3,8-diazabicyclo*/*3.2.1.*/*octan-2-one* /**IIa**; $X = -CH_2-CH(N(COOCH_3))-S-C(CH_3)_2-CH-$/

The crude product was suspended in ether (50 ml), stirred at 5 °C for 10 minutes, crystals filtered and dried. Yield: 23.5 g (85%); m. p. 105—7 °C.
IR spectrum (CH_2Cl_2): 1800(s), 1730(s), 1660(vs), 1400(s), 1290(s), 1200(vs), 1160(s) and 1110(m) cm^{-1}.

^{1}H NMR spectrum ($CDCl_3$): 1.46, 1.68 and 1.53, 1.68/4s, 2 $C(CH_3)_2$/ 2.22, 2.30/2s, $COCH_3$/, 3.7—4.3/m, CH_2/, 4.43, 5.08/ 2s, C_1—H/, 5.7, 6.2 /2m, C_5—H/.

N-Chlorocarbonyl Acetanilide /**IIb**; $R_1=C_6H_5$, $R_2=CH_3$/

To a solution of acetanilide (13.5 g; 0.1 mol), triethylamine (11.1 g; 0.11 mol) and methylene chloride (100 ml) trimethylchlorsilane (12 g; 0.11 mol) was added and the reaction mixture was mixed for 1 hour at a temperature of 40 °C. Precipitated triethylamine hydrochloride was filtered off, mother liquor cooled to 0 °C and a 20% solution (*w/w*) of $COCl_2$ in toluene (52 ml) was added over a period of 15 minutes. The reaction mixture was stirred for 1 hour at 0 °C, cold water (50 ml) added and after 1 minute the organic layer was separated, dried ($MgSO_4$) filtered and evaporated under reduced pressure. The crude product was suspended in *n*-hexane (30 ml), stirred for 5 minutes, filtered off and dried. Yield: 13.7 g (69.4%); m. p. 55—57 °C.

IR spectrum (KBr): 1765(s), 1730(s,b), 1610(w), 1490(w), 1490(w), 1425(m), 1370(m), 1195(s,b), 1010(m), 825(m), 760(s), and 700(m) cm^{-1}.

General Method for the Preparation of Mixed Anhydrides (**IV**)

To a solution of acid (4 mmol), pyridine (0.32 g; 4 mmol) in methylene chloride (10 ml), solution of *N*-chlorocarbonyl lactam or sec. amide (4 mmol) in methylene chloride (10 ml) was added over a period of 10 minutes at 0 °C. The reaction mixture was stirred for 15 minutes, cold water (20 ml) added and after 1 minute the organic layer separated, dried ($MgSO_4$), filtered and evaporated under reduced pressure.

Phenylacetic N-carboxy butyrolactam anhydride /**IVa**; $R = C_6H_5CH_2$, $X = (CH_2)_3$/

Yield: 0.83 g (73%); viscous oil; IR spectrum (film): 2920(m), 1820(s), 1750(s), 1362(w), 1328(w), 1035(vs) and 725(m) cm^{-1}.

^{1}H NMR spectrum ($CDCl_3$): 1.80—2.25 (m, CH_2), 2.35—2.75 (m, CH_2—CO), 3.55 (s, CH_2—Ph), 3.71—4.20 (m, CH_2—N), 7.20 (s, C_6H_5).

Phenylacetic N-acetyl, N-phenyl carbamic anhydride

/**IVb** $R = C_6H_5CH_2$, $R_1 = C_6H_5$, $R_2 = CH_3$/

Yield: 0.85 g (72.9%) viscous oil; IR spectrum (CH_2Cl_2): 3015(w), 1822(vs), 1760(vs,b) 1600(m), 1500(m), 1200(vs) and 1035(vs) cm^{-1}.

Phenylacetic N^3*-carboxy-*N^8*-acetyl-7,7-dimethyl-6-thia-3,8-diazabicyclo-/3.2.1./octan-2-one anhydride* (**IVa**; $R = C_6H_5CH_2$,

$X = —CH_2—CH(—S—C(CH_3)_2—CH—)—N(—COCH_3)—$

Yield: 0.76 g (51%); viscous oil; IR spectrum (CH_2Cl_2): 3015(w), 2920(w), 1825(s), 1755(m,sh), 1715(s) and 1035(vs) cm^{-1}.

Rearrangement of Mixed Anhydrides (**IV**) *into* N*-Acyl Lactams (Semicyclic Imides)* /**VI**/

N*-Phenylacetyl butyrolactam* /**VI**; $R = C_6H_5CH_2$ $X = —(CH_2)_3—$/

A solution of mixed anhydride **IVa** /$R=C_6H_5CH_2$, $X=—(CH_2)_3—$/ (0.5 g; 2 mmol), triethylamine (0.01 g; 0.1 mmol) and methylene chloride (10 ml) was stirred for 10 hours at 25 °C. The solvent was evaporated and ether (5 ml) added and stirred for 5 minutes, filtered off and the mother liquor was evaporated to oily residue.

The crude product was chromatographed over silica gel using a benzene-acetone gradient. Compound with R_F 0.65 (system A) was separated.

Yield: 0.19 g (47%); m. p. 50—1 °C.

Anal. $C_{12}H_{13}NO_2$ (203.1) calc'd: C 70.90; H 6.45; N 6.89%
found: C 70.50; H 6.10; N 6.67%

IR spectrum (CH_2Cl_2): 2980(w), 1740(vs), 1695(s), 1356(s), 1230(w), 1190(m) and 1070(w) cm^{-1}.

^1H NMR spectrum ($CDCl_3$): 1.75—2.22 (m, CH_2), 2.36—2.75 (m, CH_2CO), 3.60—3.90 (m, CH_2N), 4.17 (s, $CH_2C_6H_5$) and 7.20 (s, C_6H_5).

N^3Phenylacetyl-N^8-acetyl-7,7-dimethyl-6-thia-3,8-diazabicyclo/3.2.1/octan-2-one /**VI**; $R = C_6H_5CH_2$, $X = -CH_2-CH(N(COCH_3))-S-C(CH_3)_2-CH-$/

A solution of mixed anhydride **IVa** ($R = C_6H_5CH_2$, $X = -CH_2-CH(N(COCH_3))-S-C(CH_3)_2-CH-$)

(0.38 g, 1 mmol) in toluene (10 ml) was stirred for 8 hours at 100 °C. The reaction mixture was cooled to 25 °C, extracted with water (10 ml), dried ($MgSO_4$), filtered and evaporated. The residue was stirred with ether for 5 minutes at 0 °C and crystals were filtered off. Yield: 0.12 g (35%); m. p. 154—6 °C. /Lit. [16]m. p. 154—6 °C./

N-(N'-2',2,2'-Trichloroethyloxycarbonyl-D-α-phenylglycyl)-butyrolactam

/**VI**; $R = C_6H_5-CH(NH-CO-O-CH_2CCl_3)-$, $X = -(CH_2)_3-$/

a) To a solution of *N*-2,2,2-trichloroethyloxycarbonyl-D-α-phenylglycine (0.65 g, 2 mmol), pyridine (0.16 g, 2 mmol) and benzene (15 ml) solution of *N*-chlorocarbonylbutyrolactam (0.29 g, 2 mmol) in benzene (5 ml) was added dropwise during 10 minutes at 15 °C and the reaction mixture stirred for 2 hours. Crystals were filtered off and the mother liquor was extracted with water, dried ($MgSO_4$), filtered and evaporated under reduced pressure. The residue was crystallised from ether and *n*-hexane.

Yield: 0.16 g (20.6%); m. p. 138—140 °C; $[\alpha]_D^{23} + 20^0$ (c 0.5; CH_2Cl_2); R_f 0.73 (system B).

Anal. $C_{15}H_{15}N_2O_4Cl_3$ (393.86) calc'd: C 45.70; H 3.84; N 7.11; Cl 26.96%
found: C 45.65; H 3.50; N 7.05; Cl 26.20%

IR spectrum (KBr): 3340(s), 1745(vs), 1680(s), 1535(s), 1365(vs), 1247(s), 1100(m), 1040(m), 820(m), 705(m) and 715(m) cm^{-1}.

^1H NMR spectrum ($CDCl_3$): 1.70—2.28 (m, CH_2), 2.31—2.70 (m, CH_2—CO), 3.43—4.00 (m, CH_2—N), 4.61 (s, CH_2—O), 6.03 (d, J = 7Hz, NH) 6.48 (d, J = 7Hz, CH), and 7.27 (s, C_6H_5).

b) To a solution of 2,2,2-trichloroethyloxycarbonyl-D-α-phenylglycine (0.65 g, 2 mmol), *N*-chlorocarbonylbutyrolactam (0.29 g, 2 mmol) in benzene (15 ml) at 80 °C pyridine (0.16 g, 2 mmol) was added and the reaction mixture was stirred for 2 hours. Crystals were filtered off and the mother liquor was worked up as described above.

Yield: 0.22 g (27.9%); m. p. 139—140 °C.

N-(N'-Benzyloxycarbonyl-D-α-phenylglycyl)-butyrolactam

/**VI**; $R = C_6H_5—\underset{|}{C}H—NH—CO—O—CH_2C_6H_5$, $X = —(CH_2)_3—$/

To a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (0.57 g, 2 mmol), *N*-chlorocarbonyl butyrolactam (0,29 g, 2 mmol) in benzene (15 ml) at 80 °C, pyridine (0.16 g, 2 mmol) was added and the reaction mixture was stirred for 2 hours. Crystals were filtered off and the mother liquor was extracted with water (2 × 10 ml), dried ($MgSO_4$), filtered and evaporated under reduced pressure. The residue was chromatographed over silica gel using methylene chloride-ether gradient. A compound with R_f 0.56 (system B) was separated.

Yield: 0.25 g (35%); m. p. 97—8 °C; $[\alpha]_D^{23}$ — 28° (c 0.25, CH_2Cl_2).

Anal. $C_{20}H_{20}N_2O_4$ (352.38) calc'd: C 68.11; H 5.72; N 7.90%
found: C 68.35; H 5.73; N 7.78%

IR spectrum (KBr): 3355(m), 2980(w), 1740(s), 1718(vs), 1685(vs), 1525(s), 1455(w), 1365(s), 1240(vs), 1195(w), 1080(w), 1040(m), 756(m) and 700(m) cm^{-1}.
^1H NMR spectrum ($CDCl_3$): 1.55—2.17 (m, CH_2), 2.12—2.67 (m, CH_2—CO), 3.27—3.93 (m, CH_2—N), 4.92 (s, CH_2—O), 5.81 (d, J = 8Hz, NH), 6.44 (d, J = 8Hz, CH) and 6.75—7.45 (m, 2 C_6H_5).

Formation of Symmetrical Anhydrides (**VII**):
Bis (N-2,2,2-trichloroethyloxycarbonyl-D-α-phenylglycine) anhydride (**VII**);

$R = C_6H_5—\underset{|}{C}H—NH—CO—O—CH_2CCl_3$

a) To a solution of N-2,2,2-trichloroethyloxycarbonyl-D-α-phenylglycine (0.65 g, 2 mmol), pyridine (0.16 g, 2 mmol) and methylene chloride (10 ml), solution of *N*-chlorocarbonyl butyrolactam (0.29 g, 2 mmol) in methylene chloride (5 ml) was added dropwise during 5 minutes at 0 °C. The reaction mixture was stirred for 2 hours, crystals were filtered off and washed with cold methylene chloride (1 ml).

Yield: 0.2 g (31%); m. p. 183—5 °C.

UV spectrum (dioxan) λ_{max}: 252 nm (log ε 3.19), 258 nm (log ε 2.98) 264 nm (log ε 2.86) (shoulder)

IR spectrum (KBr): 3350(s), 1825(s), 1715(vs), 1515(s,b), 1380(w), 1350(w), 1320(w), 1295(w), 1235(m), 1080(vs), 1045(vs), 940(m) 820(w), and 720(s) cm^{-1}.
^1H NMR spectrum (DMSO-d_6; Jeol 90 Mc): 4.91 (s, CH_2O), 5.32 (d, J = 10Hz, CH), 7.28—7.46 (m, C_6H_5) and 8.75 (d, J = 10Hz, NH).

b) According to the procedure described above, benzene was used as a solvent, and reaction mixture was stirred at 15 °C for 2 hours. Crystals were filtered off, stirred with cold water for 5 minutes at 5 °C, filtered off and dried.

Yield: 0.35 g (55%); m. p. and spectral data as above.

N-Benzylphenylacetamide (**IX**; $R = R_1 = C_6H_5CH_2$)

To a solution of mixed anhydride **IVa** ($R = C_6H_5CH_2$—; $X = —(CH_2)_3—$) (0.6 g, 2.4 mmol) and methylene chloride (15 ml), solution of benzylamine (0.27 g, 2.5 mmol) in methylene chloride (5 ml) was added at 0 °C and the reaction mixture stirred for 2 hours. After addition of water (15 ml) the mixture was stirred for 5 minutes and the organic layer separated, dried ($MgSO_4$), and after filtration, the solvent was evaporated under reduced pressure. The residual solid was stirred with ether and crystals were filtered and dried.

Yield 0.44 g (82.2%); m. p. 118—121 °C. Lit. [12]: m. p. 122 °C. R_f: 0.43 (system A)

The mother liquor was evaporated under reduced pressure and the residue chromatographed over silica gel using benzene-acetone gradient and the compound with R_f 0.56 (system A) was separated.

Yield: 0.017 g (3.2%) of compound **X** /$R_1 = C_6H_5CH_2$; $X = —(CH_2)_3—$); m. p. 64—6 °C.

N-(N'-Benzylcarboxamide)-butyrolactam (**X**; $R_1 = C_6H_5CH_2$, $X = -(CH_2)_3-$)

To a solution of *N*-chlorocarbonyl butyrolactam (0.74 g, 5 mmol) and toluene (20 ml) at 0 °C, benzylamine (1.1 g, 10 mmol) was added and the reaction mixture stirred for 1 hour. Benzylamine hydrochloride was filtered off; mother liquor was mixed with water (15 ml) and dil. hydrochloric acid (1 : 1) added till pH 3. The organic layer was separated, washed with water (15 ml), dried ($MgSO_4$) filtered off and evaporated. The residue was stirred with ether and crystals filtered off.

Yield: 0.68 g (62%); m. p. 65—6 °C; R_f 0.55 (system A)

Anal. $C_{12}H_{14}N_2O_2$ (218.25) calc'd: C 66.03; H 6.47; N 12.84%
found: C 65.73; H 6.17; N 12.57%

IR spectrum (KBr): 3315(m), 1695(s,b), 1530(m), 1480(w), 1380(m), 1263(m), 835(w), 753(m), 695(m), and 655(m) cm^{-1}.
^1H NMR spectrum ($CDCl_3$): 1.75—2.30 (m, CH_2), 2.40—2.8 (m, CH_2—CO), 3.63—4.12 (m, CH_2—N), 4.40—4.70 (m, $CH_2C_6H_5$), 7.60 (s, C_6H_5) and 9.10 (b, CO—NH).

N-Benzyloxycarbonyl-D-α-phenylglycine benzylamide (**IX**;

$R = C_6H_5\underset{|}{C}H—NH—CO—O—CH_2—C_6H_5$, $R_1 = C_6H_5—CH_2$)

To a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (1.14 g, 4 mmol), pyridine (0.32 g, 4 mmol) and methylene chloride (10 ml), a solution of *N*-chlorocarbonyl butyrolactam (0.62 g, 4.2 mmol) and methylene chloride (5 ml) was added at 0 °C and the reaction mixture stirred for 20 minutes. A solution of benzylamine (0.43 g, 4 mmol) in methylene chloride (5 ml) was added dropwise over period of 15 minutes and the reaction mixture stirred for 1 hour at 0 °C. Water (15 ml) was added, the mixture was stirred for 5 minutes, the organic layer separated and dried ($MgSO_4$). After evaporation the solid residue was recrystallised from ethyl acetate.

Yield: 1.29 g (85%); m. p. 186—190 °C; $[\alpha]_D^{23} = -105°$ (c 0.5, CH_2Cl_2); R_f 0.78 (system B).

Anal. $C_{23}H_{22}N_2O_3$ (374.42) calc'd: C 73.77; H 5.92; N 7.48%
found: C 73.30; H 5.60; N 7.10%

IR spectrum (KBr): 3310(s), 1688(s), 1650(vs), 1520(s), 1254(s), 750(m) and 695(s) cm^{-1}.
^1H NMR spectrum (DMSO-d_6): 4.28 (d, J = 6Hz, CH_2—N), 5.06 (s, CH_2O), 5.34 (d, J = 8Hz, CH—N), 6.90—7.70 (m, 3 C_6H_5), 7.97 (d, J = 8Hz, NH—CO—O), and 8.75 (m, NH—CO).

N-Benzyloxycarbonyl-D-α-phenylglycyl-L-phenylalanine methyl ester

(**IX**; $R = C_6H_5\underset{|}{C}H—NH—CO—O—CH_2C_6H_5$, $R_1 = C_6H_5\underset{|}{C}H—COOCH_3$).

To a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (0.57 g, 2 mmol), pyridine (0.16 g, 2 mmol) and methylene chloride (10 ml), a solution of *N*-chlorocarbonyl butyrolactam (0.30 g, 2 mmol) in methylene chloride (5 ml) was added dropwise during 5 minutes at 0 °C. The reaction mixture was stirred for 20 minutes, a solution of L-phenylalanine methylester (0.36 g, 2 mmol) in methylene chloride (5 ml) was added and the solution stirred for 1 hour. Water was added, the mixture stirred for 5 minutes, the organic layer separated, washed with water (10 ml), dried ($MgSO_4$) and filtered off. Evaporation of solvent gave solid residue which was recrystallized from isopropanol.

Yiled: 0.69 g (77%); m. p. 166—8 °C; $[\alpha]_D^{23} = -22.7°$ (c 0.44, CH_2Cl_2); R_f 0.75 (system B).

Anal. $C_{26}H_{26}N_2O_5$ (446.5) calc'd: C 69.93; H 5.87; N 6.27%
found: C 70.25; H 6.05; N 6.52%

IR spectrum (KBr): 3320(s), 1735(m), 1685(m), 1645(s), 1520(s), 1440(w), 1380(w), 1377(w), 1345(w), 1255(m), 1230(m), 1045(w), and 700(s) cm^{-1}.

^{1}H NMR spectrum ($CDCl_3$): 2.95 (d, J = 5Hz, $CH_2C_6H_5$), 3.73 (s, CH_3O), 5.06 (s, O—CH_2—C_6H_5), 5.28 (d, J = 7Hz, CH), 6.33 (d, J = 7Hz, NH) 6.44—7.50 (m, NH, CH, $3C_6H_5$).

*6-/N-(Benzyloxycarbonyl)-D-α-phenylglycylamido/-penicillanic acid benzyl ester (**XII**; Z = $C_6H_5CH_2$—O—CO—, R = $C_6H_5CH_2$, Y = $C(CH_3)_2$, W = H/*

To a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (0.57 g, 2 mmol), pyridine (0.16 g, 2 mmol) and methylene chloride (10 ml), a solution of *N*-chlorocarbonyl butyrolactam (0.29 g, 2 mmol) in methylene chloride (5 ml) was added at 0 °C and the reaction mixture stirred for 15 minutes. A solution of 6-aminopenicillanic acid benzylester (0.58 g, 1.9 mmol) in methylene chloride (10 ml) was added and the reaction mixture was stirred for 1 hour at 0 °C. Cold water (15 ml) was added, the mixture stirred for 5 minutes, organic layer separated, dried ($MgSO_4$), filtered and evaporated. The crude residue was chromatographed over silica gel using a methylene-chloride-ether gradient. Two compounds were isolated.

Compound XII/Z = $C_6H_5CH_2$—O—CO, R = $C_6H_5CH_2$, Y = $C(CH_3)_2$, W = H); with R_f 0.75 (system B).

Yield: 0.87 g (75.5%); foam; (amorphous product[13]); $[\alpha]_D^{20}$ + 90° (c 0.5, CH_2Cl_2).

Anal. $C_{31}H_{31}N_3O_6S$ (573.65) calc'd: C 64.90; H 5.45; N 7.33; S 5.59%
found: C 65.22; H 5.64; N 7.52; S 5.12%

IR spectrum ($CHCl_3$): 3390(m), 3010(m), 1783(vs), 1725(s), 1690(s), 1490(s), 1295(m), 1260(m), 1195(m), 1150(m), 1040(m) and 960 (m) cm^{-1}.
^{1}H NMR spectrum ($CDCl_3$): 1.30 and 1.40 (2s, $2CH_3$), 4.30 (s, C_3—H), 4.90 and 5.03 (2s, $2OCH_2$—C_6H_5), 5.00—5.65 (m, CH, C_5—H, C_6—H), 6.07 (d, J = 7Hz, NH), 6.77 (d, J = 8.5Hz, NH) and 6.90—7.50 (m, $2C_6H_5$).
Compound **XIII**/R = C_6H_5—CH_2, X = —$(CH_2)_3$—, Y = $C(CH_3)_2$/ with R_f 0.66 (system B) yield: 0.075 g (9.5%); m. p. 115—118 °C; $[\alpha]_D^{23}$ + 100° (c 0.5, CH_2Cl_2).
IR and ^{1}H NMR spectral data identical to spectra of sample prepared by procedure described below.

*6-(Butyrolactam-N-amido)-penicillanic acid benzylester (**XIII**; R = $C_6H_5CH_2$—, X = —$(CH_2)_3$—, Y = $C(CH_3)_2$/*

To a solution of 6-aminopenicillanic acid benzylester (0.6 g, 2 mmol), triethylamine (0.3 g, 3 mmol) and benzene (20 ml), solution of *N*-chlorocarbonyl butyrolactam (0.29 g, 2 mmol) in benzene (10 ml) was added at 0 °C and the reaction mixture stirred for 2 hours. Crystals of triethylamine hydrochloride were filtered off, the mother liquor was washed with water (10 ml) and stirred with water (10 ml) by addition of dil. HCl (1 : 1) till pH 2.5. The organic layer was separated, washed with water (10 ml), dried ($MgSO_4$), filtered and evaporated. The solid residue was chromatographed over silica gel using a methylene chloride -ether gradient. Compound with R_f 0.66 (system B) was separated.

Yield: 0.57 g (68.3%); m. p. 115—8 °C; $[\alpha]_D^{23}$ + 100° (c 0.5, CH_2Cl_2).

Anal. $C_{20}H_{23}N_3O_5S$ (417.47) calc'd: C 57.54; H 5.55; N 10.07; S 7.68%
found: C 57.32; H 5.33; N 10.10; S 7.20%

IR spectrum (KBr): 3240(m), 3200(m), 2965(m), 1775(vs), 1745(s), 1710(vs), 1520(vs), 1382 (m), 1300(m), 1250(m), 1200(m), 1180(m), 970(m), 745(m), and 690(m) cm^{-1}.
^{1}H NMR spectra ($CDCl_3$): 1.44 and 1.67 (2s, $2CH_3$), 1.77—2.33 (m, CH_2), 2.41—2.80 (m, CH_2—CO), 3.70—4.00 (m, CH_2—N), 4.55 (s, C_3—H), 5.22 (s, CH_2O), 5.50—5.87 (m, C_5—H, C_6—H), 7.47 (s, C_6H_5) and 9.33 (d, J = 8Hz, NH).

*6-(D-α-aminophenylacetamido)-penicillanic acid trihydrate (ampicilline trihydrate) /**XII**; R = Z = W = H, Y = $C(CH_3)_2$/*

To a solution of *N*-chlorocarbonyl butyrolactam (6.45 g, 44 mmol), pyridine (0.05 g) and acetone at —10 °C, sodium salt of *N*-(1-methoxycarbonylpropen-2-yl)-D-α-phenylglycine (11.6 g, 43 mmol) was added and the reaction mixture stirred for 5 minutes. The resulting mixture, containing the mixed anhydride, was added

with vigorous stirring into a solution of sodium salt of 6-aminopenicillanic acid (8.2 g, 38 mmol) in water (30 ml) and acetone (30 ml). The reaction mixture was stirred for 1 hour at —10 °C, dil. HCl (1 : 1) added dropwise to pH 2 and stirring was continued for 15 minutes by adding more acid if necessary. The mixture was extracted with methylenechloride (150 ml), the aqueous layer was concentrated at low temperature and reduced pressure to remove acetone. Dil. NaOH (20%) was added dropwise till pH 5 at 10 °C. A suspension was stirred for 2 hours at 10 °C, crystals were filtered and washed with acetone.

Yield: 9.55 g (62%) ampicillin trihydrate. $[\alpha]_D^{23}$ + 291° (c 0.5, H_2O) water: 12.9% Microb. assay: 97.5%.
IR spectrum identical to standard.

6-(D-α-amino-p-hydroxyphenylacetamido)-penicillanic acid trihydrate (Amoxycilline trihydrate) /**XII**; $R = Z = H$, $W = OH$, $Y = C(CH_3)_2$/

To a solution of *N*-chlorocarbonyl butyrolactam (6.45 g, 44 mmol), *N*-methylmorpholine (0.04 g) and acetone (65 ml) at —20 °C potassium salt of *N*-(1-methoxycarbonylpropen-2-yl)-D-α-amino-p-hydroxyphenylglycine (12.3 g, 40 mmol) was added at —20 °C and the reaction mixture stirred for 10 minutes. The resulting mixture, containing the mixed anhydride was added with vigorous stirring to a solution of triethylamine salt of 6-aminopenicillanic acid, prepared by dissolving 6-aminopenicillanic acid (8.2 g, 38 mmol) in water (30 ml) and acetone (30 ml) with addition of triethylamine (3.8 g, 38 mmol). The reaction mixture was stirred for 1 hour at —20 °C, dil. HCl (1 : 1) was added dropwise at 0 °C to pH 2. Methylene chloride (80 ml) was added, the mixture stirred for 2 minutes, the aqueous layer separated and was again extracted with methylene chloride (80 ml). After filtration conc. NH_4OH was added dropwise to pH 5 and the suspension was stirred for 2 hours at 5 °C. Crystals were filtered, washed with acetone and dried.

Yield: 11.2 g (70.5%) amoxicilline trihydrate. $[\alpha]_D^{23}$ + 293° (c 0.2, H_2O) water: 13.2%. Microb. assay: 95.2%
IR spectrum identical to standard.

7-/N-(Benzyloxycarbonyl)-D-α-phenylglycylamido/-3-methyl-3-cephem-4-carboxylic Acid Diphenylmethyl Ester /**XII**; $R = CH(C_6H_5)_2$,

$Z = C_6H_5CH_2—O—CO—$, $Y = —CH_2—\overset{\|}{C}—CH_3$, $W = H$/

a) To a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (0.57 g, 2 mmol), pyridine (0.16 g, mmol) and methylene chloride (10 ml), a solution of *N*-chlorocarbonylbutyrolactame (0.29 g, 2 mmol) in methylene chloride (5 ml) was added at —10 °C and the reaction mixture stirred for 15 minutes. A solution of 7-amino-3-methyl--3-cephem-4-carboxylic acid diphenylmethyl ester (0.70 g, 1.85 mmol) methylene chloride (10 ml) was added and the mixture stirred for 2 hours at 0 °C. Cold water (10 ml) was added, the mixture stirred for 5 minutes. The organic layer was separated, washed with 5% HCl (5 ml), water (10 ml), 1N $NaHCO_3$ (5 ml), water (10 ml) and dried. After filtration solution was evaporated and solid residue crystallized from benzene.

Yield: 0.94 g (78.4%); m. p. 203—5 °C; $[\alpha]_D^{23}$ — 10° (c 2, CH_2Cl_2); R_f 0.77 (sistem B).

Anal. $C_{37}H_{33}N_3O_6S$ (647.72) calc'd: C 68.61; H 5.14; N 6.49; S 4.95%
found: C 68.30; H 5.20; N 6.49; S 4.30%

IR spectrum (KBr): 3310(m), 1775(s), 1710(m), 1685(m), 1658(vs) 1520(s), 1375(w), 1355(w), 1297(w), 1235(w), 1218(m), 1150(w), 935(w) and 690(m) cm^{-1}.
^1H NMR spectrum (DMSO-d_6): 2.00 (s, CH_3), 3.30—3.50 (m, CH_2—S), 5.00 (d, J = 4 Hz, C_6—H), 5.08 (s, $CH_2C_6H_5$), 5.50 (d, J = 8Hz, CH—C_6H_5), 5.50—5.85 (m, C_7—H), 6.87 (s, CH—O), 7.20—7.70 (m, $4C_6H_5$), 8.05 (d, J = 8Hz, NH—CO), and 9.30 (d, J = 8 Hz, NH—CO—O).

b) To a solution of N^3-chlorocarbonyl-N^8-acetyl-7,7-dimethyl-6-thia-3,8-diazabicyclo/3.2.1./octan-2-one (0.55 g, 2 mmol) in methylene chloride (10 ml), a solution of *N*-benzyloxycarbonyl-D-α-phenylglycine (0.57 g, 2 mmol) pyridine (0.18 g, 2.3 mmol)

in methylene chloride (5 ml) was added and the reaction mixture stirred for 15 minutes. A solution of 7-amino-3-methyl-3-cephem-4-carboxylic acid diphenylmethyl ester (0.60 g, 1.6 mmol) in methylene chloride (5 ml) was added and the reaction mixture stirred for 1 hour at 0 °C. Cold water was added (10 ml) and worked up as under a/. A solid residue was chromatographed over silica gel using a methylene-chloride-ether gradient. Compound with R_f 0.77 (system B) was separated.

Yield: 0.87 g (84%); m. p. 203—5 °C.

7-/N-(2',2',2'-Trichloroethyloxycarbonyl)-D-α-phenylglycylamido/-3-methyl--3-cephem-4-carboxylic acid 2',2',2'-trichloroethyl ester /**XII**;

$R = Cl_3CCH_2$—, $Z = Cl_3CCH_2$—O—CO—, $Y = CH_2$—$\overset{\|}{C}$—CH_3, $W = H$/

a) According to previous procedure for **XII** (under a/) using N-2',2',2'-trichloroethyloxycarbonyl-D-α-phenylglycine and 2',2',2'-trichloroethyl ester of 7-ADCA instead of diphenylmethyl ester the crude product obtained was chromatographed over silica gel using methylene chloride -ether gradient. A compound with R_f 0.78 (system B) was separated.

Yield: 0.86 g (78.2%); m. p. 104—6 °C; IR and ^{1}H NMR spectral data are indentical to the data reported in the literature[14].

b) The procedure was repeated using *N*-chlorocarbonylvalerolactam instead of *N*-chlorocarbonyl butyrolactam. A crude product was purified by crystallization from carbon tetrachloride and ether.

Yield: 0.83 g (75%); m. p. and spectra identical.

7-/N-(2',2',2'-Trichlorethyloxycarbonyl)-D-α-phenylglycilamido)-3-methyl--3-cephem-4-carboxylic acid diphenylmethyl ester

/**XII**; $R = CH(C_6H_5)_2$, $Z = Cl_3CCH_2$—O—CO—, $Y = CH_2$—$\overset{\|}{C}$—CH_3, $W = H$/

a) According to the previous procedure using diphenylmethylester instead of trichlorethyl ester, a crude product was obtained, which was purified by crystallization from isopropanol.

Yield: 0.87 g (84%); m. p. 177—183°; $[\alpha]_D^{23} = -10^0$ (c 0.5, CH_2Cl_2); R_f 0.76 (system B). /Lit.[15] $[\alpha]_D^{23} = -8.6$ (c 1, dioxane)./

IR spectrum (KBr): 3320(s), 3035(w), 2950(w), 1785(vs), 1715(vs), 1670(vs), 1530(m), 1500(m), 1453(w), 1375(m), 1225(s), 723(m), and 700(m) cm^{-1}.

^{1}H NMR spectrum ($CDCl_3$): 1.96 (s, CH_3), 3.03 (m, CH_2—S), 4.58 (s, CH_2—O), 4.75 (d, J = 5Hz, C_6—H), 5.37 (d, J = 7Hz, NH), 5.66 (2d, J= 5 and 9Hz, C_7—H), 6.45 (d, J = 7Hz, CH), 7.90 (s, CHO), and 6.86—7.50 (m, $3C_6H_5$ and NH).

b) The procedure was repeated by using *N*-chlorocarbonyl caprolactam instead of *N*-chlorocarbonyl butyrolactam.

Yield: 0.88 g (68.7%) of the product with the same physical and spectral data as those reported under a/.

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REFERENCES

1. Taken in part from *Ph. D. Thesis* of M. K., University of Zagreb, 1981.
2. E. Schröder and K. Lübke, *The Peptides,* Vol. **I**, Academic Press, New York, 1965., p. 85.
3. J. H. C. Nayler, *Advances in Penicillin Research, in: Adv. Drug Res.* **7** (1973) pp. 1—106; G. V. Kaiser and S. Kukolja, *Modifications of the beta-Lactam System,* in: E. H. Flynn (Ed.), *Cephalosporins and Penicillins,* Academic Press, New York (1972) pp. 74—94.

4. M. Kovačević, J. J. Herak, and B. Gašpert, *Yugoslav Patent Appl.* **P-1408/80** (1980); *Brit. Patent Appl.* **2 078 724** (1982).
5. H. B. König, W. Schröck, K. G. Metzger, and H. Diesselkotter, *Ger. Offenlengsschrift* **20 25 414** (1971).
6. V. F. Mironov, V. D. Seludiakov, and V. P. Koziukov, *Žur. Obšćei Him.* **39** (1969) 220; *Dokl. Akad. Nauk SSSR,* **192** (1970) 1056.
7. M. Kovačević, J. J. Herak, and B. Gašpert, *Yugoslav Patent Appl.* **P-1410/80** (1980); *Ger. Offenlegungsschrift* **31 20 415** (1981).
8. P. Stelzer, *Asymmetrische Anhydride,* in E. Müler (Ed.), *Houben-Weyl Methoden der Organischen Chemie,* IV Auf., G. Thieme Verlag, Stuttgart, 1974, Band **XV/2**, pp. 169—205.
9. N. F. Albertson, *Synthesis of Peptides with Mixed Anhydrides,* in: A. C. Cope (Ed.), *Organic Reactions,* Vol. **12**, p. 157; J. Wiley & Sons, New York, 1962.
10. M. Kovačević, J. J. Herak, and B. Gašpert, a) *Yugoslav Patent Appl.* **P-2928/80** (1980); *Ger. Offenlegunsschrift* **31 25 486** (1982); b) *Yugoslav Patent Appl.* **P-1898/81** (1981); *Swiss Patent Appl.* **4658/82** (1982).
11. G. Falkenstein and H. Doerfel, *Makrom. Chem.* **127** (1969) 34.
12. *Beilsteins Handbuch der Organischen Chemie,* IV Auf., J. Springer Verlag, Berlin, 1935, Band **12** (I) p. 458.
13. *Novo Terapeutisk Lab., Ger. Offenlegunsscrhift* **23 64 759** (1974).
14. R. R. Chauvette, P. A. Pennington, C. W. Ryan, R. D. Cooper, F. L. José, I. G. Wright, E. M. VanHeyningen, and G. W. Huffman, *J. Org. Chem.* **36** (1971) 1259.
15. R. Bywood, G. Gallagher, and D. Walker, *Ger. Offenlegungsschrift* **23 11 597** (1973).
16. J. J. Herak, M. Kovačević, I. Lukić, and B. Gašpert, *Croat. Chem. Acta* **52** (1979) 311.
17. *British Pharmacopoeia,* Department of Health and Social Security — London (1980) 31, 35, 86.

SAŽETAK

Aktiviranje karboksilnih kiselina s pomoću mješovitog anhidrida s *N*-acil-*N*-alkil karbaminskom kiselinom

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Priređeni su mješoviti anhidridi **IV** reakcijom karboksilnih kiselina **III** s *N*-klorokarbonil laktamima (**IIa**) i sekudarnim amidima (**IIb**). Dekarboksilacijom **IV** daju polucikličke imide **VI**, dok sporednom reakcijom nastaju simetrični anhidridi **VII**. Aminolizom su iz **IV** dobiveni amidi karboksilnih kiselina u visokom iskorištenju i čistoći. Stoga je **IVb** iskorišten za pripravu aminosupstituiranih beta-laktamskih antibiotika (**XII**), što je nova metoda aktiviranja *N*-zaštićenog fenilglicina (**IIIg**), tijekom aciliranja 6-APK odnosno 7-ADCK (**XI**).