THE SYNTHESIS OF PYRAZOLES. A SIMPLE PREPARATIVE SYNTHESIS OF C-NUCLEOSIDIC ANTIBIOTICS FORMYCIN AND FORMYCIN B

Ladislav KALVODA

Institute of Organic Chemistry and Biochemistry, Czechoslovak Academy of Sciences, 166 10 Prague 6

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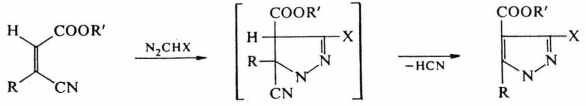
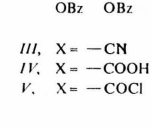
It was found that the esters of 3-cyano-2-propenoic acids (which are easily accessible by acylation of hydrogen cyanide in the presence of an excess of alkoxycarbonylmethylenetriphenylphosphorane) afford pyrazoles on cycloaddition of diazo compounds and simultaneous elimination of hydrogen cyanide. The diazo compound adds with its nitrogen to the α position and with its carbon atom to the β position with respect to the nitrile group. The cycloaddition was used as a key reaction step in the simple synthesis of C-nucleosidic antibiotics formycin (*I*) and formycin B (*II*).

Recently a simple synthesis of the esters of 3-cyano-2-alkenoic acids was elaborated in our laboratory, consisting in the action of acylation reagents on alkoxycarbonylmethylenetriphenylphosphoranes in the presence of hydrogen cyanide[1]. These cyano esters were found valuable as intermediates in the synthesis of 2-substituted meleinimides[1], which was used successfully in the synthesis of the C-nucleosidic antibiotic showdomycin[2]. In this paper we should like to point out the possibility of utilizing them also in the synthesis of 3(5)-substituted pyrazoles, namely in the synthesis of C-nucleosidic antibiotics formycin (*I*) and formycin B (*II*). In the past the syntheses of formycin[3] B and oxoformycin[4] were described, which also start from nitrile *III*, but which are substantially more complex.

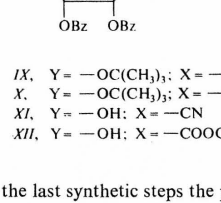
During the study of the reactivity of the double bond in the esters of 3-cyano--2-alkenoic acids we found that they afford on cycloaddition of diazo compounds an unstable adduct that easily eliminates hydrogen cyanide under formation of corresponding pyrazoles. This cycloaddition takes place in such a way that the diazo-compound is bound by its carbon atom to the position β and by its nitrogen atom in the position α to the nitrile group.



This reaction course presented the possibility of utilizing this cycloaddition as the key reaction step in the synthesis of C-nucleosidic antibiotics formycin and formycin B. During their synthesis the easy accessibility of 2,3,5-tri-O-benzoyl-β-D-ribofuranosyl cyanide[5] was made use of, which was converted by acid hydrolysis fo 2,4,6-tri-O-benzoyl-2,5-anhydro-D-allonic acid[2] (*IV*). The latter was converted to chloride *V*, which was not isolated, and reacted with tert-butoxycarbonylmethylene-triphenylphosphorane (*VI*) in the presence of hydrogen cyanide to afford ester-nitrile *VII* (the configuration Z for the ester-nitrile *VII* was assigned on the basis of its easy cyclization to imide *VIII*). In subsequent steps diazoacetonitrile was added to ester-nitrile *VII*, or also ethyl diazoacetate, under formation of substituted pyrazoles *IX* or *X*, respectively. The tert-butyl group was split off from pyrazoles *IX* and *X* under acid catalysis and acids *XI* and *XII* were obtained which were submitted to Curtius degradation in a modification developed by Ninomiya and coworkers[6] for urethans *XIII* and *XIV*.

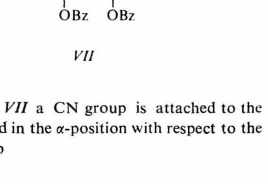


III, X = —CN
IV, X = —COOH
V, X = —COCl



IX, Y = —OC(CH$_3$)$_3$; X = —CN
X, Y = —OC(CH$_3$)$_3$; X = —COOC$_2$H$_5$
XI, Y = —OH; X = —CN
XII, Y = —OH; X = —COOC$_2$H$_5$

VI

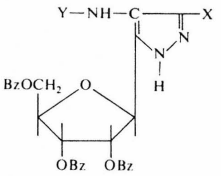


VII

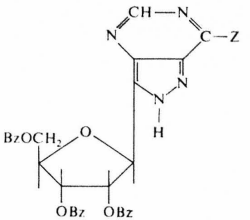
In formula *VII* a CN group is attached to the double bond in the α-position with respect to the sugar group

In the last synthetic steps the protecting trichloroethyl group was cleaved reductively from the obtained urethans, and in amino derivatives *XV* and *XVI* the pyrimidine cycle was closed by heating with formamidine acetate[7] under formation of benzoates *XVII* and *XVIII*. Using alkali catalyzed methanolysis of the protecting benzoyl groups in the benzoates *XVII* and *XVIII* formycin (*I*) and formycin B (*II*) were

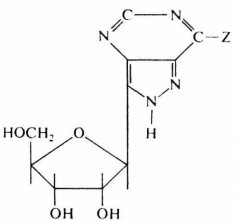
obtained, the properties of which are identical with those of substances obtained by fermentation.



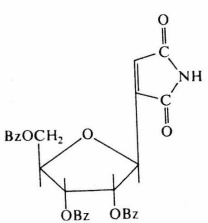
XIII, X = $-COOC_2H_5$; Y = $-COOCH_2CCl_3$
XIV, X = $-CN$; Y = $-COOCH_2CCl_3$
XV, X = $-COOC_2H_5$; Y = $-H$
XVI, X = $-CN$; Y = $-H$



XVII, Z = $-OH$
XVIII, Z = $-NH_2$



I, X = $-NH_2$
II, X = $-OH$



VIII

XIX

EXPERIMENTAL

The melting points were determined on a Kofler block. The analytical samples were dried at 70°C/0·1 Torr for 5 h, unless stated otherwise. Thin-layer chromatography was carried out on silica gel Merck GF_{254} (type 60); the substances were inspected under ultraviolet light or detected by carbonization after spraying with 10% sulfuric acid in methanol. The solutions were dried over anhydrous magnesium sulfate and concentrated at 40°C bath temperature.

(Tert-Butoxycarbonylmethyl)triphenylphosphonium Chloride (*XIX*)

Chloroacetic acid (94·5 g; 1·0 mol) and sulfuric acid (2·0 ml) were added to a solution of isobutylene (300 ml) in benzene (1·0 l) and the solution allowed to stand at room temperature for 48 h. It was then washed with water (300 ml) and dilute ammonia until alkaline. The benzene extract was stabilized with N,N-dimethylaniline (2 ml), dried and concentrated to about 700 ml. Triphenylphosphine (131 g; 0·5 mol) was added to the concentrate and the mixture boiled for 8 h. After cooling the precipitated product *XIX* was filtered off under suction and washed with ether. Yield 200 g (98%), m.p. 166—167°C. For $C_{24}H_{26}ClO_2P$ (412·9) calculated: 69·81% C, 6·35% H, 7·60% P, 8·58% Cl; found: 70·12% C, 6·52% H, 7·34% P, 8·62% Cl.

(Tert-Butoxycarbonylmethylene)triphenylphosphorane (*VI*)

Chloroform (400 ml) and ice (200 g) were added to a stirred solution of chloride *XIX* (200 g; 0·48 mol) in water (400 ml), followed by a 10% aqueous sodium hydroxide solution until the reaction of the mixture was permanently alkaline. The chloroform layer was separated, dried and concentrated. Ether was added to the residue and the solution was allowed to crystallize. The separated product *VI* was suction-dried, and washed with ether. Yield 145 g (77%), m.p. 149 to 150°C. For $C_{24}H_{25}O_2P$ (376·4) calculated: 76·57% C, 6·60% H, 8·22% P; found: 76·15% C, 6·78% H, 8·08% P.

Tert-Butyl-(*Z*)-3-cyano-3-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)propenoate (*VII*)

A suspension of nitrile *III* (4·7 g; 10 mmol) in acetic acid (20 ml and conc. hydrochloric acid (5·0 ml) was heated at 100°C under stirring for 15 min. After cooling water was added (50 ml) followed by ethyl acetate (50 ml). The organic layer was separated, washed with water, dried, the solvent evaporated and the residue co-distilled with toluene (2 × 20 ml). Thionyl chloride (7·0 ml), ether (7·0 ml) and dimethylformamide (1 drop) were added to the residue and the mixture was refluxed for 45 min, then concentrated and the residue co-distilled with toluene (3 × 20 ml). The residue was dissolved in benzene (50 ml), hydrogen cyanide (2·0 ml) was added and the mixture cooled to 0°C. Phosphorane *VI* (8·4 g; 22 mmol) was added under stirring and the mixture stirred for another hour at room temperature, then washed with water and the organic layer was separated, dried and concentrated. Ethanol was added to the residue which induced the crystallization of a product which was filtered off under suction and washed with ethanol. Yield 3·55 g (60%), m.p. 149—150·5°C, $[\alpha]_D^{20}$ —60·2° (*c* 0·5 in $CHCl_3$). For $C_{34}H_{31}NO_9$ (597·6) calculated: 68·33% C, 5·23% H, 2·34% N; found: 68·74% C, 5·43% H, 2·76% N.

2-(2,3,5-Tri-O-benzoyl-β-D-ribofuranosyl)maleinimide (*VIII*)

Ester-nitrile *VII* (0·6 g; 1 mmol) was added to a mixture of acetic acid (2·5 ml), acetic anhydride (2·5 ml) and sulfuric acid (0·5 ml) and the mixture was heated on a boiling water bath for 15 min.

After cooling it was diluted with ethyl acetate (30 ml) and water (30 ml), the organic layer was washed with a saturated aqueous solution of sodium hydrogen carbonate, dried and evaporated. The product, *VIII*, was isolated from the reaction mixture by chromatography on silica gel in benzene–ethyl acetate 9 : 1 (TLC R_F *VIII* = 0·32; R_F *VII* = 0·70 in the same system). Yield 420 mg (78%), $[\alpha]_D^{25}$ −28·5° (*c* 0·5 in $CHCl_3$). IR spectrum ($CHCl_3$): *ν* (NH) 3440 w; *ν* (CO) 1782 w, sh, 1734 vs, 1714 m, sh. For $C_{30}H_{23}NO_9$ (541·5) calculated: 66·54% C, 4·28% H, 2·59% N; found: 66·58% C, 4·42% H, 2·58% N.

4-Tert-Butoxycarbonyl-5-cyano-3-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)pyrazole (*IX*)

Water (10 ml) and sodium nitrite (6·9 g; 0·1 mol) were added to a suspension of aminoacetonitrile hydrochloride (10·0 g; 0·11 mol)[8] in ether (100 ml) and the mixture was stirred at room temperature for 2 h. The ethereal layer was separated, dried over calcium chloride and evaporated to about 25 ml volume (it is most important to keep the volume at this value, because in one case a serious explosion took place in our laboratory when the mixture was concentrated *in vacuo* at 40°C). Dichloromethane (25 ml) and ester-nitrile *VII* (6·0 g; 10 mmol) were then added to the obtained ethereal solution of diazoacetonitrile and the mixture was allowed to stand in darkness at room temperature for 4 weeks. It was then concentrated and the residue co-distilled with xylene (2 × × 50 ml), the residue was dissolved in toluene (10 ml), triethylamine (3·0 ml) was added and the mixture allowed to stand at room temperature for one hour, then concentrated and the residue co-distilled with toluene (2 × 50 ml). For analysis the product was freed from a small amount of impurities by chromatography in benzene–ethyl acetate 9 : 1 (TLC: R_F *IX* = 0·30 in the same system). Yield 5·5 g (86%), $[\alpha]_D^{25}$ −4·5° (*c* 0·5 in $CHCl_3$). For $C_{35}H_{31}N_3O_9$ (637·7) calculated: 65·93% C, 4·90% H, 6·59% N; found: 65·75% C, 4·89% H, 6·41% N.

4-Tert-Butylcarbonyl-5-ethoxycarbonyl-3-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)pyrazole (*X*)

Ester-nitrile *VII* (6·0 g; 10 mmol) was added to a solution of ethyl diazoacetate (4·2 g; 30 mmol) in toluene (25 ml) and the mixture heated at 60—70°C for 20 h, then concentrated and the residue co-distilled with xylene (2 × 50 ml). The residue was dissolved in toluene (10 ml), triethylamine (3 ml) was added to it and the mixture was allowed to stand at room temperature for 1 h. It was then concentrated and the residue co-distilled with toluene (2 × 50 ml). For analysis the product was purified by chromatography on silica gel in benzene–ethyl acetate 9 : 1 (TLC: R_F *X* = 0·30 in the same system). Yield 5·0 g (73%), $[\alpha]_D^{25}$ +6·4° (*c* 0·5 in $CHCl_3$). For $C_{37}H_{36}O_{11}N_2$ (684·7) calculated: 64·90% C, 5·30% H, 4·09% N; found: 64·67% C, 5·53% H, 3·97% N.

5-Ethoxycarbonyl-4-(2,2,2-trichloroethoxycarbamoyl)-3-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)pyrazole (*XIII*)

Pyrazole *X* (prepared by condensation of 6·0 g, 10 mmol of esternitrile *VII* with ethyl diazoacetate) was dissolved in formic acid (30 ml; 98%) and the solution was heated at 60—70°C for 30 min. After concentration the residue was co-distilled with a mixture of dioxane and toluene 1 : 1 (2 × 50 ml). Triethylamine (1·12 g; 11 mmol), trichloroethanol (1·64 g; 11 mmol), diphenylphosphorylazide[6] (3·03 g; 11 mmol), and tolene (30 ml) were added to the residue and the mixture was heated at 100°C for 5 h. Benzene was then added (50 ml) followed by a saturated aqueous solution of sodium hydrogen carbonate (50 ml). The organic layer was separated, dried and evaporated. The residue was dissolved in benzene (50 ml) and filtered through a column of silica gel (3 × 16 cm) which was subsequently washed with a mixture of ethyl acetate and benzene (1 : 9; 500 ml) and the combined filtrates were evaporated, the residue dissolved in ether and the solution

allowed to crystallize. The separated product was filtered off under suction and washed with ether. Yield 4·50 g (58%, calculated per *VII*), m.p. 174–175°C. $[\alpha]_D^{25}$ +37·1° (*c* 0·5 in $CHCl_3$). For $C_{35}H_{30}Cl_3N_3O_{11}$ (775·0) calculated: 54·24% C, 3·90% H, 5·42% N, 13·72% Cl; found: 54·71% C, 3·91% H, 5·72% N, 13·17% Cl.

5-Cyano-4-(2,2,2-trichloroethoxycarbamoyl)-3-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)pyrazole (*XIV*)

Pyrazole *IX* (prepared on condensation of 6·0 g of ester-nitrile *VII* with diazoacetonitrile) was converted to *XIV* in the same manner as pyrazole *X* to urethan *XIII*. After filtration through a silica gel column 2·6 g (36%, calculated per *VII*) were obtained of a syrup which would not crystallize. For analysis the product was purified by chromatography on silica gel in benzene–ethyl acetate 9 : 1 mixture (TLC: R_F *XIV* = 0·40 in the same mixture); $[\alpha]_D^{25}$ −4·5° (*c* 0·5 in $CHCl_3$). For $C_{35}H_{31}N_3O_9$ (637·7) calculated: 65·93% C, 4·90% H, 6·59% N; found: 65·75% C, 4·89% H, 6·41% N.

2′,3′,5′-Tri-O-benzoylformycin B (*XVII*)

Zinc dust (3·0 g) and ammonium chloride (1·0 g) were added to a suspension of carbamoyl derivative *XIII* (500 mg; 0·65 mmol) in methanol (20 ml) and the mixture was refluxed for 45 min under stirring. The unreacted zinc was filtered off and the filtrate evaporated. Benzene (20 ml), water (20 ml) and conc. ammonia (5 ml) were added to the residue and the organic layer was separated and concentrated. The residue was dissolved in methylcellosolve (3 ml) and formamidine acetate[9] (200 mg; 2 mmol) was added to the solution which was then heated at 100°C for 5 h. Water (20 ml) was then added, followed by ethyl acetate (20 ml) and the organic layer was separated, dried and evaporated. The residue was dissolved in benzene and the solution allowed to stand for crystalization. The separated product was filtered off under suction and washed with benzene. Yield 250 mg (67%), m.p. 185–186°C (recrystallization), m.p. 233–237°C. $[\alpha]_D^{25}$ −84·1° (*c* 0·5 in acetone). For $C_{31}H_{24}N_4O_8$ (580·6) calculated: 64·14% C, 4·17% H, 9·65% N; found: 64·36H C, 4·57% H, 9·59% N.

2′,3′,5′-Tri-O-benzoylformycin (*XVIII*)

Ammonium chloride (4·0 g) and zinc dust (10 g) were added to a solution of carbamoyl derivative *XIV* (2·6 g; 4·1 mmol) in methanol (80 ml) and the mixture was refluxed for 45 min. Zinc was filtered off and the filtrate evaporated. The residue was triturated with benzene (80 ml), water was added (80 ml) followed by conc. ammonia (20 ml), and the benzene layer was separated, dried and evaporated) The residue was mixed with methylcellosolve (10 ml), formamidine acetate (1·15 g; 12·3 mmol) and the mixture was heated at 100°C for 5 h. After cooling water (80 ml) was added to the mixture, then ethyl acetate (80 ml) and the organic layer was separated, dried and concentrated. The residue was dissolved in benzene and allowed to stand for crystallization. The separated product was filtered off under suction and washed with a benzene–ether mixture (1 : 1); yield 720 mg (29·5%), m.p. 237–238°C. $[\alpha]_D^{25}$ −79·5 (*c* 0·5 in acetone). For $C_{31}H_{25}N_5O_7$ (579·6) calculated: 64·24% C, 4·35% H, 12·08% N; found: 64·08% C, 4·27% H, 12·03% N.

Formycin (*I*)

A solution of sodium methoxide in methanol (1·0 ml of a 1M solution) was added to a suspension of tribenzoate *XVIII* (500 mg; 0·86 mmol) in methanol (20 ml) and the mixture was allowed to

stand at room temperature for 20 h. Acetic acid (1·0 ml) was then added to it and the mixture concentrated. Ether (20 ml) and water (20 ml) were added to the residue and the aqueous layer was separated and filtered through a column (2 × 2·5 cm) of Dowex 50 X 8 in H^+ form. The column was first washed with water and formycin was then eluted with 5% aqueous ammonia. The eluate was concentrated to a thin syrup which was allowed to crystallize. Yield 207 mg (90%) of a product of m.p. 153–155°C. $[\alpha]_D^{25}$ −39·3° (*c* 0·5 in 0·1M-HCl). Literature[10] gives m.p. 149–150°C, $[\alpha]_D$ −40·0° (*c* 1 in 0·1M-HCl). For analysis the sample was dried at room temperature and 0·1 Torr for 1 h. For $C_{10}H_{13}N_5O_4.H_2O$ (285·3) calculated: 42·11% C, 5·30% H, 24·55% N; found: 41·88% C, 5·43% H, 25·00% N.

Formycin B (*II*)

Tribenzoate *XVII* (500 mg; 0·86 mmol) was suspended in absolute methanol (20 ml), sodium methoxide in methanol (1·0 ml of a 1M solution) was added to it and the mixture was allowed to stand at room temperature for 20 h. Acetic acid (1·0 ml) was then added and the mixture evaporated. The residue was treated with ether (20 ml) and water (20 ml) and the aqueous layer was separated and filtered through a column (2 × 2·5 cm) of Dowex 50 in pyridine cycle. The filtrate was evaporated, the residue codistilled with water (2 × 20 ml), and the residue crystallized from a small amount of water. Yield 220 mg (95%); m.p. 243–245°C. Literature[3] gives m.p. 245–249°C. $[\alpha]_D^{25}$ −52·0° (*c* 0·5 in water); lit.[11] gives $[\alpha]_D^{20}$ 51·5° (*c* 1 in water). For analysis the sample was dried at 130°C and 0·1 Torr for 1 h. For $C_{10}H_{12}N_4O_5$ (268·2) calculated: 44·78% C, 4·51% H, 20·89% N; found: 44·41% C, 4·82% H, 20·68% N.

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