NEW SYNTHESIS OF RIBONUCLEOSIDE CARBOCYCLIC ANALOGUES* **

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Reaction of (±)-1,2-O-isopropylidene-3*t*-benzoyloxymethyl-5*t*-(2,2,2-trichloroethoxycarbonyl)-aminocyclopentane-1*r*,2*c*-diol (*III*) with zinc in methanol afforded the amino derivative *IV*, the condensation of which with 5-amino-4,6-dichloropyrimidine (*V*) yielded the acyclic intermediate *VI*. Reaction of compound *VI* with triethyl orthoformate in the presence of hydrogen chloride, ammonolysis, debenzoylation, and acidic hydrolysis furnished (±)-aristeromycin (*I*). The racemate *I* was resolved into the optical antipodes *Ia* and *Ib* by chromatography on a column of cellulose. Reaction of the amine *IV* with N-(ethoxycarbonyl)amide of 3-ethoxy-2-ethoxycarbonylacrylic acid and methanolysis furnished the 2′,3′-O-isopropylidene-5-methoxycarbonyl derivative *XIV* from which the carbauridine (±)-1-(2*t*,3*t*-dihydroxy-4*c*-hydroxymethylcyclopent-1*r*-yl)uracil (*II*) was prepared by alkaline hydrolysis, decarboxylation, and removal of the protecting group by the action of acetic acid.

In an earlier paper[1], we have described a synthetic approach to the synthesis of a (±)-2*t*,3*t*-dihydroxy-4*c*-hydroxymethylcyclopentyl-1*r*-amine derivative as the intermediate in preparation of racemic carbocyclic analogues of ribonucleosides; in these isosters, the original ribofuranose ring is replaced by the corresponding substituted cyclopentane derivative. In the present paper, we wish to report the last step in the preparation of the racemic aristeromycin, namely, (±)-9-(2*t*,3*t*-dihydroxy-4*c*-hydroxymethylcyclopent-1*r*-yl)adenine (*I*), and the carbocyclic analogue of uridine, namely, (±)-1-(2*t*,3*t*-dihydroxy-4*c*-hydroxymethylcyclopent-1*r*-yl)uracil (*II*), from the above mentioned intermediate.

The two syntheses have in common formation of the purine or pyrimidine ring from the substituted cyclopentylamine *IV*. Compound *IV* was prepared *in situ* by reaction of (±)-1,2-O-isopropylidene-3*t*-benzoyloxymethyl-5*t*-(2,2,2-trichloroethoxycarbonylamino)cyclopentane-1*r*,2*c*-diol (*III*) in methanol with zinc in the presence of ammonium chloride[1].

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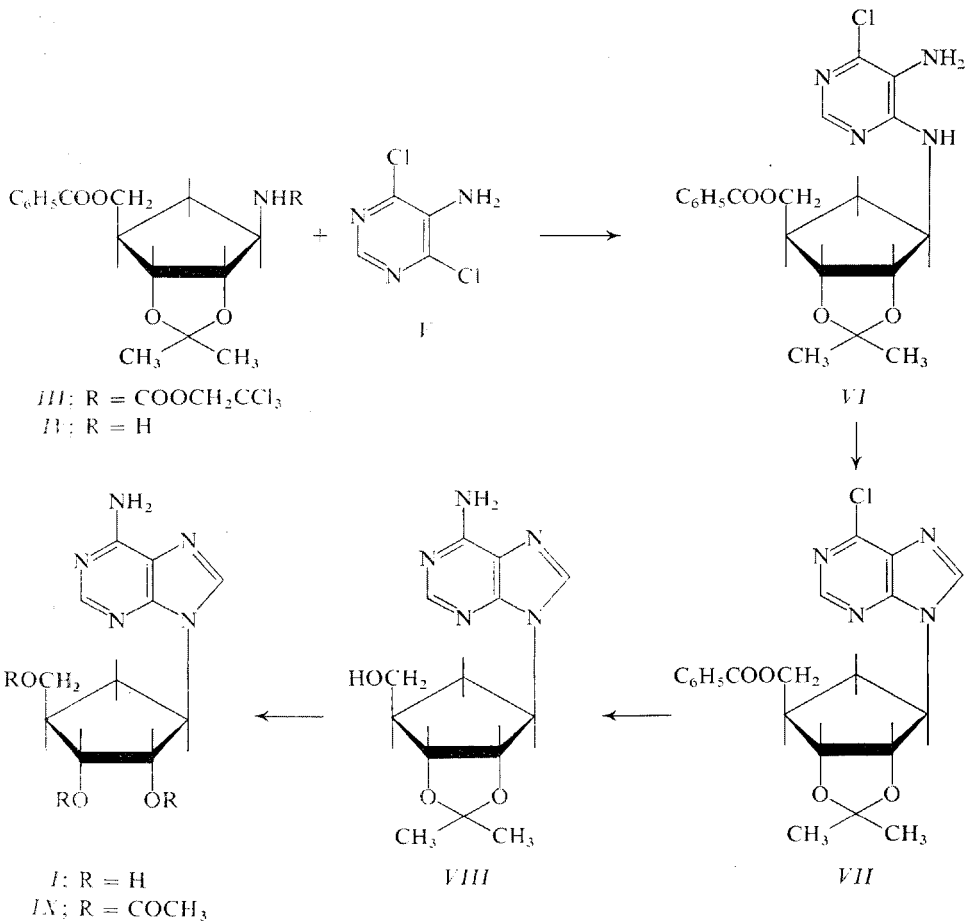
For purposes of the preparation of (±)-aristeromycin (*I*), the amino derivative *IV* was heated with 5-amino-4,6-dichloropyrimidine (*V*) in 1-butanol in the presence of triethylamine. The thus-obtained acyclic intermediate *VI* was converted in the next step by the action of triethyl orthoformate in the presence of hydrochloric acid (*cf.*[2]) into the 6-chloropurine derivative *VII*. The cyclisation was quantitative and the chromatographically homogeneous product was directly transformed to an adenine derivative by ammonolysis in methanolic solution. This treatment results not only in substitution at position 6 of the heterocyclic ring but also in ammonolysis of the benzoyl group at position 5′ of the cyclopentane ring with the formation of (±)--aristeromycin 2′,3′-isopropylidene derivative *VIII*. This compound was isolated in pure state and its structure was established by elemental analysis, mass spectrum,



SCHEME 1

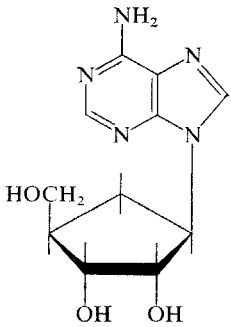
and ^{1}H-NMR spectrum which simultaneously confirmed the isomeric homogeneity of the product. Furthermore, the difference between chemical shifts of signals belonging to the doublet of the isopropylidene group ($\Delta\delta$ = 0·21) suggests (by analogy to ribofuranose derivatives[3]) that the present structure is similar to that of β-anomers since in the sugar series the $\Delta\delta$ value is equal to 0·18–0·23 p.p.m. with β-anomers and to 0–0·10 p.p.m. with α-anomers. This finding along with the established structure of the starting urethan *III* and the amine *IV* (*via* the acetyl derivative, *cf.*[1]) may be considered as a sufficient evidence for the absence of any change in configuration of the carbon atom at position 1 of the cyclopentane ring during the synthesis of the adenine ring from compound *IV*.

The cleavage of the 2′,3′-O-isopropylidene derivative* *VIII* in acetic acid is much slower than that of the analogous 2′,3′-O-isopropylideneadenosine and affords pure (±)-aristeromycin (*I*). The mass spectrum of *I* exhibits a molecular peak *m/e* = = 305 as well as the appropriate group of base peaks, BH^+ and BH_2^+ *m/e* = 134, 135, and 136, and does not differ from the mass spectrum of an authentic specimen. The ^{1}H-NMR spectrum displays H_2 and H_8 signals of the adenine ring and a group of three unresolved multiplets of proton signals due to the cyclopentane ring (*cf.*[2]). The UV absorption spectrum of compound *I* at pH 2 (λ_{max} 258 nm) is also in accordance with that of an authentic material. Compound *I* is entirely homogeneous on electrophoresis in a borate buffer solution and its mobility under these conditions simultaneously confirms the presence of a *cis*-diol system capable of the formation of a borate complex in alkaline media. The preparative yield of (±)-aristeromycin (*I*) from the cyclopentylamine derivative *III* is satisfactory and comparable with those of an alternative route reported elsewhere[2].

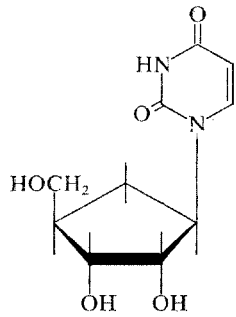
The behaviour of compound *I* on thin-layer chromatography (silica gel) and paper chromatography in several solvent systems suggests a complete homogeneity. On the other hand, a sharp separation of compound *I* into two spots was observed on paper chromatography in the solvent system 2-propanol–conc. aqueous ammonia–water (7 : 1 : 2). As shown after elution of these spots with 0·01M-HCl, the UV spectra were identical and the two substances were present in a strictly equimolar ratio. The two substances, compound *Ia* (lower R_F value) and compound *Ib* (higher R_F value) do not differ on rechromatography in other solvent systems or on electrophoresis. The above separation is also limited to cellulose since on silica gel in the same solvent system compound *I* behaves as homogeneous. When the above prepared compound *I* was subjected to preparative chromatography on a column of cellulose powder in the solvent system stated, the products *Ia* and *Ib* were isolated

* To emphasize the analogy to isosteric ribonucleosides, a notation is used analogous to that of nucleosides; positions 1′, 2′, and so on thus correspond to positions on the cyclopentane ring. Position 5′ designates the hydroxymethyl group and the methylene group on the ring is designated 6′.

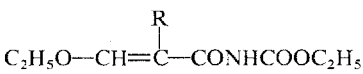
in pure crystalline state; their mass and ^{1}H-NMR spectra are identical and correspond to those of compound prior to the separation. Furthermore, conversion of the two substances *Ia* and *Ib* to the 2′,3′-O-isopropylidene derivatives *VIIIa* and *VIIIb* by the reaction with 2,2-dimethoxypropane yielded products of identical ^{1}H-NMR and mass spectral data and R_F values on paper chromatography and thin-layer chromatography on silica gel.



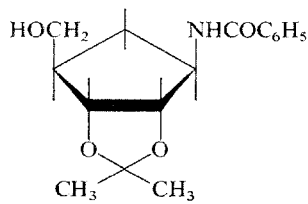
I



II



X; R = H
XII; R = $COOC_2H_5$



XI

The products *Ia* and *Ib* exhibit optical activity. Their $[\alpha]_D^{20}$ values in 1M-HCl are opposite and their CD spectra both in water and 0·01M-HCl are enantiomeric (Fig. 1). The CD spectra of compounds *VIIIa,b* obtained from separated components *Ia,b* are also enantiomeric. It may be thus assumed on the basis of all these findings that (±)-aristeromycin (*I*) was successfully resolved into enantiomers by chromatography on cellulose in the solvent system stated. The designations (+) and (−) refer to $[\alpha]_D$ in 0·1M-HCl; the (+)-series is derived from compound *Ia*. In the literature, the CD spectra of the naturally occurring aristeromycin have not been reported; the absolute configuration corresponding to that of adenosine was assigned on the basis of X-ray analysis[4]. In our hands, a sample of the naturally occurring aristeromycin did not exhibit under similar conditions any Cotton effects in the whole wavelength region. On the other hand, the specimen obtained by another synthetic procedure[2] may be resolved into compounds *Ia* and *Ib* under the above conditions.

The CD spectra of compounds *Ia* and *Ib* are not identical with those of D- and L-adenosine (Fig. 1*b*) but correspond to CD spectra of 8-bromoadenosine derivatives with *anti* conformation. Consequently, the earlier assignment of the absolute configuration to compounds *Ia,b* appears problematic. On the basis of comparison with spectra of 8-bromoadenosine, compound *Ib* (of the higher mobility) can be preliminarily assigned the absolute configuration corresponding to D-ribonucleosides while that of L-ribonucleosides belongs to compound *Ia* of a lower mobility.

The carbocyclic analogue *II* of uridine has not been so far reported. Attention has been paid[2,5–8] exclusively to the preparation of purine derivatives which are more readily accessible *via* the ring formation. The attempted preparation of the series *II* compounds by classical closures of the pyrimidine ring, *e.g.*, by the reaction of compound *IV* with the N-(ethoxycarbonyl)amide of 3-ethoxyacrylic acid[9] (*X*) under conditions recommended for preparation of uracil derivatives, did not meet with success. Two compounds were isolated from the reaction mixture, namely, the amide of 3-ethoxyacrylic acid and a substance which was assigned the structure *XI* on the basis of mass spectrum (m/e = 292, M-15 = $C_{15}H_{18}NO_4$) and ^{1}H-NMR spectrum. Compound *XI* might be formed from the amino derivative *IV* under the reaction conditions used by an intra- or intermolecular (O → N)-transbenzoylation. The occurrence of this side reaction can obviously be ascribed to the insufficient reactivity of the 3-alkoxy derivative *X* towards the amine *IV*.

In an earlier paper[10] of this Series, we have reported on the preparation of 5-ethoxycarbonyluridine by the reaction of 2,3-O-isopropylidene-β-D-ribofuranosylamine with N-(ethoxycarbonyl)amide of 3-ethoxy-2-ethoxycarbonylacrylic acid (*XII*);

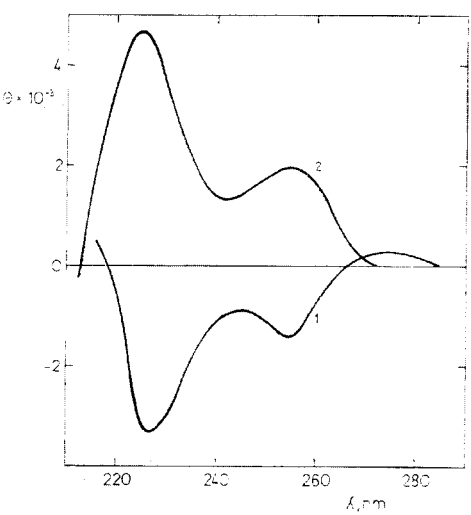


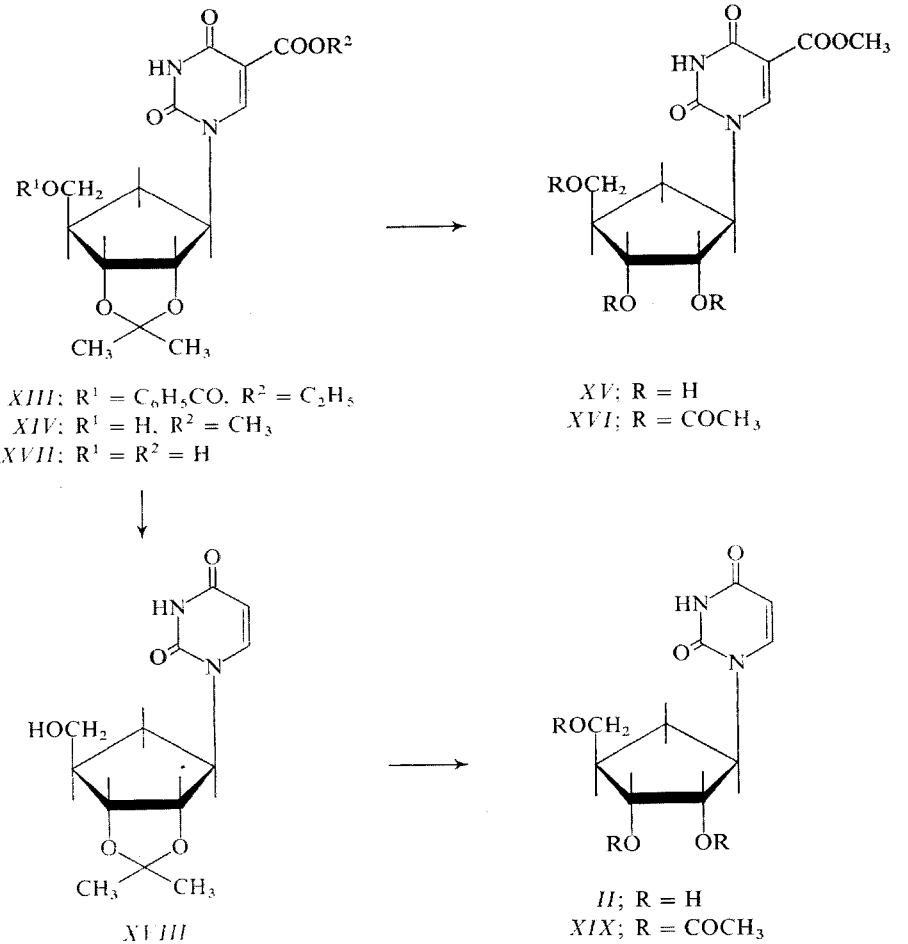
FIG. 1
Circular Dichroism Spectra (in 0·01M-HCl)
1 Compound *Ib*, *2* compound *Ia*.

the amide *XII* was prepared for the first time and description of its preparation was included. The condensation is extraordinarily easy since the derivative *XII* is highly reactive. The reaction of compounds *IV* and *XII* was effected by reflux in methanol in the presence of triethylamine[10]. Under these conditions a single product was obtained and characterised by the structure *XIII* which may be inferred from ^{1}H-NMR spectrum, analysis, and mass spectrum containing the molecular peak *m/e* 458 and a system of regressive peaks, inter alia the 5-ethoxycarbonyluracil peak *m/e* 184 and the benzoic acid peak.

Removal of the benzoyl group from position 5′ of compound *XIII* is extraordinarily difficult. It was accomplished by a prolonged reflux in 0·2M methanolic sodium methoxide. Compound *XIV* was isolated as the single product and its structure confirmed by ^{1}H-NMR spectrum and mass spectrum. Methanolysis of the benzoyl ester in compound *XIII* was simultaneously accompanied by transesterification of the ethoxycarbonyl group at position 5 of the uracil ring with the formation of the corresponding methyl ester. The ^{1}H-NMR spectrum of compound *XIV* exhibits the presence of a doublet of the isopropylidene group, signals of NH and H_6 protons of the uracil ring as well as a singlet of the methyl group bound as ester; the cyclopentane portion of the molecule affords again three multiplets with the corresponding number of protons, the attempted analysis of which did not meet with success. Compound *XIV* was therefore subjected to acidic hydrolysis with the formation of the free carbocyclic nucleoside *XV* which was directly without isolation acetylated. The ^{1}H-NMR spectrum of the resulting chromatographically homogeneous triacetyl derivative *XVI* was again incapable of analysis in the cyclopentane region. An unambiguous assignment of the anomeric structure of compound *XIV* from the $J_{1',2'}$ coupling constant was therefore not possible. Analogously to the aristeromycin derivative *VIII* it may be however inferred from the difference ($\Delta\delta$ = = 0·19 p.p.m.) of chemical shifts in the doublet of proton signals belonging to the isopropylidene group of compound *XIV* that compounds *XIII* to *XV* are "β-anomers"*, *i.e.*, the 1*r*,4*c*-isomers.

By the alkaline hydrolysis of compound *XIV*, the salt of the 5-carboxy derivative *XVII* is readily obtained. Aqueous solution of this salt was passed through a pyridinium ion exchange resin to afford the pyridinium salt containing effluent which was evaporated and dried *in vacuo* to remove pyridine. The thus-obtained free acid *XVII* was then subjected to thermal decarboxylation; in contrast to the nucleoside series[11], the present case requires the temperature of boiling quinoline. The decarboxylation is rather sensitive to the reaction time since an excessive reaction time results in a considerable decomposition of products. The reaction was preferably checked by paper electrophoresis in an alkaline buffer solution; this method makes possible determina-

* The designation "β-anomer" for the 2*t*,3*t*-dihydroxy-4*c*-hydroxymethylcyclopent-1*r*-yl derivative of the heterocyclic base is used in analogy to nucleosides.



SCHEME 2

tion of the resulting neutral derivative *XVIII* in the presence of the starting acidic compound *XVI*. The reaction mixture was processed by chromatography on silica gel to afford the chromatographically homogeneous 2′,3′-O-isopropylidene derivative of the carbocyclic uridine analogue *XVIII*. The structure of compound *XVIII* was unequivocally established by mass spectrum and ^{1}H-NMR spectrum which exhibits H_5 and H_6 protons of the uracil nucleus, NH group, and three non-analysable multiplets of the proton signals belonging to the cyclopentane residue. The $\Delta\delta$ value (0·18 p.p.m.) demonstrates the isomeric purity and the 1*r*,4*c*-configuration of compound *XVIII* (*vide supra*). Acidic hydrolysis of compound *XVIII* yielded as the single product the chromatographically and electrophoretically pure nucleoside

analogue *II*. The structure of compound *II* may be inferred from the above intermediates (particularly compound *XVIII*) and the mass spectrum (*m/e* 242 (M^+), *m/e* 113 BH uracil). Similarly to compound *XV*, we did not succeed in resolving by peracetylation the multiplets of the cyclopentane ring protons. Unfortunately, this effect is characteristic of the present type of compounds even in various solvents[1]. The IR spectra of acetates *XVI* and *XIX* in tetrachloromethane exhibit bands of ν(C=O) 1755 cm^{-1} (acetate), ν(C=O) 1695 and 1714 cm^{-1} (uracil), and ν(C=N) 1635 cm^{-1} with compound *XIX* and 1623 cm^{-1} with compound *XV* (heterocyclic base). An additional ν(C=O) band at 1730 cm^{-1} in the spectrum of compound *XVI* corresponds to the $COOCH_3$ group on the uracil ring.

In contrast to (±)-aristeromycin (*I*), the carbocyclic uridine analogue *II* cannot be resolved into optical antipodes by chromatography on cellulose. The resolution in the solvent system mentioned above also failed in the case of the 5-ethoxycarbonyl derivative *XV* and the corresponding more polar 5-carboxy derivative (obtained by alkaline hydrolysis). The resolution thus appears to be limited to the adenine derivatives but nevertheless is abolished by hydrophobisation of the molecule such as acetylation or conversion to the isopropylidene derivative as well as by introduction of polar substituents such as phosphorylation of compound[1,2] *I*. Compounds of the uracil series lack the not quite obvious influence of the adenine ring system which contributes to the separation of the enantiomers of compound *I* along with the hydrophilic character of groupings on the cyclopentane part of the molecule.

The present syntheses of (+)- and (−)-aristeromycin (*Ia* and *Ib*) and compound *II* exemplify the applicability of a novel type of cyclopentylamine derivatives in the preparation of the carba analogues of ribonucleosides. The functional groups such as the benzoyl group and the isopropylidene group are stable enough in the required transformations but may be readily removed from the cyclopentane ring if need be. Thus, *e.g.*, the 2,3-O-isopropylidene derivatives *VIII* and *XVIII* may also be used in other reactions such as selective 5'-phosphorylation and the like[1,2]. The present key analogues of ribonucleosides of both the pyrimidine and purine series may be transformed into additional analogues such cytidine or inosine by methods closely analogous to those of the nucleoside chemistry.

EXPERIMENTAL

Melting points were taken on a heated microscope stage (Kofler block) and were not corrected. Unless stated otherwise, the solutions were taken down on a rotatory evaporator at 35°C/15 Torr and analytical samples were dried over phosphorus pentoxide at 0·1 Torr. Paper chromatography was performed by the descending technique on paper Whatman No 1 (preparative runs on paper Whatman No 3 MM) in the solvent systems S_1, 2-propanol–conc. aqueous ammonia–water (7 : 1 : 2); S_2, 2-propanol–conc. aqueous ammonia–0·1M triethylammonium borate (7 : 1 : 2); and S_3, 1-butanol–acetic acid–water (5 : 2 : 3). Paper electrophoresis was carried out at 20 V/cm for 1 h (for the technique see ref.[13]) in the buffer solutions E_1, 0·1M triethylam-

TABLE I

Chromatography and Electrophoresis

Compound	R_F				E2[a]	E3[b]	Compound	R_F		
	S1	S2	S3	S7				S4	S5	S6
Adenosine	0·57	0·52	0·45	—	0·90	0·55				
Uridine	0·50	0·40	0·40	—	1·00	0	*V*	0·35	—	—
							VI	0·16	—	—
							VII	0·45	—	—
I	0·54	0·50	0·45	0·26	0·70	0·55	*IX*	—	—	0·46
Ia	0·37	0·50	0·45	0·26	0·70	0·55	*XI*	—	0·20	—
Ib	0·54	0·50	0·45	0·26	0·70	0·55	*XII*	0·35	—	—
II	0·52	0·50	0·42	—	1·00	—0·10	*XIII*	—	—	0·80
VIII	0·72	—	—	0·30	—	—	*XIV*	—	—	0·40
XV	0·54	—	—	—	—	—	*XVI*	—	0·30	—
XVII	0·46	—	—	—	—	—	*XVIII*	—	—	0·35
							XIX	—	—	0·42

[a] Referred to uridine; [b] referred to adenine.

monium hydrogen carbonate (pH 7·5); E_2, 0·1M triethylammonium borate (pH 7·5); and E_3, 1M acetic acid. Thin-layer chromatography was performed on ready-for-use Silufol UV_{235} (Kavalier Glassworks, Votice, Czechoslovakia) silica gel sheets in the solvent systems S_4, chloroform; S_5, chloroform–ethanol (95 : 5); S_6, chloroform–ethanol (90 : 10); and S_7, chloroform–ethanol (80 : 20). Chromatography on 16 × 40 × 0·3 cm preparative layers of fluorescent silica gel was performed with the use of the material produced by Service Laboratories of this Institute. Column chromatography was carried out on the Pitra silica gel (particle size, 30—60 micron; 100 g or 250 g runs); 30 ml fractions were taken and checked by thin-layer chromatography in the above solvent systems. The preparative chromatography on cellulose was effected on a 100 × 3·2 cm column (Sephadex, Uppsala, Sweden) of powdered cellulose (Macherey & Nagel MN 300) equilibrated with the solvent system S_1 to the loss of UV absorption; elution rate, 15 ml per hour; the fractions were taken in 1 h intervals and checked by paper chromatography in the solvent system S_1. The UV absorption of the eluate was continuously checked by the Uvicord apparatus (LKB, Uppsala, Sweden). The UV absorption spectra were taken on a Specord UV apparatus (Carl Zeiss, Jena) in 0·01M-HCl. The CD spectra were recorded on a Jouan Dichrograph apparatus. The ^{1}H-NMR spectra were measured on a Varian 100 apparatus in deuteriochloroform or hexadeuteriodimethyl sulfoxide (hexamethyldisiloxane as internal standard); chemical shifts are expressed in the δ scale (p.p.m.) and the coupling constants in Hz.

(±)-1,2-O-Isopropylidene-3*t*-benzoyloxymethyl-5*t*-(5-amino-6-chloropyrimidin-4-yl)aminocyclopentane-1*r*,2*c*-diol (*VI*)

A mixture of compound[1] *III* (11·25 g; 25 mmol), powdered zinc (17·5 g), ammonium chloride (17·5 g), and methanol (200 ml) was refluxed with stirring for 1 h, filtered while hot, and the material on the filter washed portionwise with hot methanol (total 200 ml). The filtrate and washings

were combined and evaporated under diminished pressure. The residue was extracted with two 200 ml portions of boiling chloroform, the extract filtered, and the filtrate evaporated. The residual crude amino derivative *IV* was treated with 5-amino-4,6-dichloropyrimidine[14] (5·0 g; 30·5 mmol), 1-butanol (70 m), and triethylamine (10 ml). The whole mixture was stirred at 120°C for 14 h under reflux condenser protected against atmospheric moisture and evaporated under diminished pressure. The residue was taken up into 150 ml of chloroform, the solution washed with two 50 ml portions of water, dried over anhydrous magnesium sulfate, filtered, and the filtrate evaporated under diminished pressure. The residue was crystallised from ethanol to afford 6·7 g (64%) of compound *VI*, m.p. 172–174°C. For $C_{20}H_{23}ClN_4O_4$ (418·9) calculated: 57·34% C, 5·53% H, 8·46% Cl, 13·37% N; found: 57·65% C, 5·30% H, 8·51% Cl, 13·20% N.

(±)-Aristeromycin (*I*)

A mixture of compound *VI* (6·7 g; 16 mmol), triethyl orthoformate (100 ml), and conc. hydrochloric acid (2·5 ml) was stirred until the solid dissolved (for 2 h) and the solution kept at room temperature overnight. As shown by chromatography in the solvent system S_5, the reaction was quantitative. The solution was then neutralised with triethylamine, evaporated at 40°C/0·1 Torr, the residue taken up into chloroform (200 ml), the solution washed with water (50 ml), dried over anhydrous magnesium sulfate, filtered, and evaporated. The residue was heated at 100°C for 8 h in an autoclave with 30% methanolic ammonia (75 ml), the mixture evaporated under diminished pressure, and the residue refluxed for 1 h in methanolic sodium methoxide (100 ml of 0·2M solution). After neutralisation with dry Dowex 50 X 8 (H^+ cycle) ion exchange resin, the mixture was filtered and the material on the filter washed with methanol. The filtrate and washings were combined and evaporated under diminished pressure to dryness. Acetic acid (100 ml of 80% aqueous solution) was then added to the residue, the whole refluxed for 2 h, evaporated, the residue coevaporated with three 50 ml portions of water, and finally crystallised from water. Yield, 3·3 g (78%) of compound *I*, m.p. 255–256°C. For $C_{11}H_{15}N_5O_3$ (265·3) calculated: 49·79% C, 5·69% H, 26·41% N; found: 49·70% C, 5·70% H, 26·28% N. Mass spectrum: m/e 265 (M^+), 134, 135, 136 (B, BH, BH_2), 248 ($M-NH_3$), 234 ($M-CH_2OH$), 136 ($B-CH=CH_2$). UV spectrum (pH 2): λ_{max} 259 nm (ε_{max} 13 700). ^{1}H-NMR spectrum (hexadeuteriodimethyl sulfoxide): 1·70–2·50 (m, 3 H) $H_{4'}$ + 2 H_6; 3·62 (m, 2 H) 2 $H_{5'}$; 4·15 (m, 2 H) $H_{2'}$ + $H_{3'}$; 5·07 (m, 1 H) $H_{1'}$; 8·40 (s, 1 H) H_2; 8·53 (s, 1 H) H_8.

2′,3′-O-Isopropylidene-(±)-aristeromycin (*VIII*)

A mixture of compound *I* (0·5 g; 1·9 mmol), 2,2-dimethoxypropane (10 ml), dimethylformamide (10 ml) and 6M hydrogen chloride in dimethylformamide (0·5 ml) is stirred at room temperature overnight, neutralised with triethylamine, evaporated at 40°C/0·1 Torr, and the residue crystallised from 50% aqueous ethanol. Yield, 0·4 g (69%) of compound *VIII* which does not melt up to 260°C. For $C_{14}H_{19}N_5O_3$ (305·4) calculated: 55·07% C, 6·27% H, 22·94% N; found: 55·85% C, 6·26% H, 23·26% N. Mass spectrum: *m/e* 305 (M^+), M-15, *m/e* 134, 135, 136 (B, BH, BH_2). ^{1}H-NMR spectrum (hexadeuteriodimethyl sulfoxide and CF_3COOD): 1·25 + 1·46 (2 × s, 2 × 3 H) $(CH_3)_2C$; 2·17 (m, 3 H) $H_{4'}$ + 2 $H_{5'}$; 3·70 (m, 2 H) 6′-CH_2; 4·75 (m, 3 H) $H_{1'}$ + + $H_{2'}$ + $H_{3'}$; 8·34 (s, 1 H) H_2; 8·44 (s, 1 H) H_8.

2′,3′,5′-Tri-O-acetyl-(±)-aristeromycin (*IX*)

A mixture of compound *I* (0·5 g; 1·9 mmol), acetic anhydride (10 ml), and boron trifluoride etherate (0·5 ml) was stirred at room temperature for 2 h and evaporated at 40°C/0·1 Torr.

The residue was coevaporated under the same conditions with toluene (20 ml) and then dissolved in chloroform (50 ml). The solution was washed with water (20 ml), dried over anhydrous magnesium sulfate, filtered, and the material on the filter washed with chloroform. The filtrate and washings were evaporated and the residue chromatographed on a layer of silica gel in the solvent system S_6. The band of the product was eluted with methanol, the eluate evaporated, and the residue crystallised from ethanol. Yield, 0·4 g (53·5%) of compound *IX*, m.p. 168–169°C. For $C_{17}H_{21}N_5O_6$ (391·4) calculated: 52·16% C, 5·40% H, 17·89% N; found: 51·64% C, 5·49% H, 18·00% N. By the action of methanolic sodium methoxide (0·05M; 30°C; 2 h), compound *IX* is split into compound I consisting of equal amounts of *Ia* and *Ib* (chromatography in the solvent system S_1). ^{1}H-NMR spectrum (deuteriochloroform): 1·73 + 2·03 + 2·10 (3× s, 3× 3 H) $OCOCH_3$; 1·90–2·50 (m, 2 H) 2 $H_{6'}$; 2·68 (m, 1 H), $H_{4'}$; 4·23 (2 dd, 2 H, $J_{4',5'} = 7·0$. $J_{4',5''} = 6·0$, $J_{gem} = 11·0$) 2 $H_{5'}$; 5·20–5·60 (m, 3 H) $H_{1'} + H_{2'} + H_{3'}$; 7·85 (brs, 2 H) NH_2; 8·04 (s, 1 H) H_2; 8·20 (s, 1 H) H_8.

Resolution. A suspension of compound *I* (0·5 g; 1·9 mmol) in water (2 ml) was brought into solution by the addition of minimum conc. hydrochloric acid, the whole applied to a column of powdered cellulose in the solvent system S_1, and the UV-active fractions checked by paper chromatography in S_1. The appropriate fractions were pooled and evaporated under diminished pressure; the other fractions were also combined and rechromatographed under the same conditions. Residues of the separate fractions of compounds *Ia* and *Ib* were crystallised from 80% aqueous ethanol. Yield (from total 1·5 g of compound *I*), 0·8 g of compound *Ia*, m.p. 120–121°C (decomp.). Mass spectrum: *m/e* 265 (mol. peak), 248 ($C_{11}H_{14}N_5O_2$), and 234 ($C_{10}H_{12}N_5O_2$). UV spectrum (pH 2): λ_{max} 260 nm (ε_{max} 13700). ^{1}H-NMR spectrum: identical with that of the racemate *I*. Optical rotation: $[\alpha]_D^{20}$ −18·6° (*c* 0·5, 1M-HCl). The yield of the other antipode *Ib* was 0·6 g; m.p. 142–144°C (decomposition); $[\alpha]_D^{20}$ +19·1° (*c* 0·5, 1M-HCl). The mass, UV, and ^{1}H-NMR spectra of compound *Ib* were identical with those of compound *Ia*. CD spectra (pH 2) of *Ia*: 255 nm (+1910), 242 min (+1310), 225 (+4660), 218 (0); of *Ib*: 275 nm (310), 266 (0), 255 (−1400), 244 min (−890), 226·5 (−3300), 218 (0); of the natural aristeromycin: not measurable.

2′,3′-O-Isopropylidene-(+)- and -(−)-aristeromycin (*VIIIa* and *VIIIb*)

Compound *Ia* or *Ib* (0·5 g; 1·9 mmol) was converted to the isopropylidene derivative analogously to the racemate *I*. Thus, compound *Ia* yielded 100 mg of the chromatographically homogeneous derivative *VIIIa*, m.p. 217–218°C. The UV and mass spectra were identical with those of the racemate *VIII*, ^{1}H-NMR spectrum (hexadeuteriodimethyl sulfoxide) was also identical with that of compound *VIII*; $\Delta\delta$ CH_3 (isopropylidene) 0·21.

Compound *Ib* yielded 120 mg of the antipode *VIIIb*, m.p. 223–225°C. The UV, mass, and ^{1}H-NMR spectrum were identical with those of the racemate *VIII*; $\Delta\delta$ CH_3 (isopropylidene) 0·21.

Condensation of Compound *IV* with the N-(Ethoxycarbonyl)amide of 3-Ethoxyacrylic Acid (*X*)

A residue of compound *IV* (prepared from 25 mmol of compound *III* by the procedure described in the case of compound *V*) was refluxed in methanol (100 ml) with compound[9] *X* (5·6 g; 30 mmol) for 1 h. Triethylamine (30 ml) was then added and the reflux continued for 6 h. The mixture was evaporated under diminished pressure, the residue refluxed with methanolic sodium methoxide (100 ml of 0·2M solution), the mixture neutralised with dry Dowex 50 X 8 (H^+) ion exchange resin, filtered, and the material on the filter washed with methanol. The filtrate

and washings were combined and evaporated under diminished pressure. The residue was chromatographed on a column of silica gel (200 g) in chloroform (9 : 1 chloroform–ethanol as eluant). Yield, 2·22 g (30·5%) of compound *XI*, m.p. 245–247°C (ethanol–light petroleum); R_F value 0·48 (in S_6). For $C_{16}H_{21}NO_4$ (291·3) calculated: 65·96% C, 7·26% H, 4·80% N; found: 66·20% C, 7·60% H, 4·83% N. Mass spectrum: *m/e* 292 (M^+), 276 ($C_{15}H_{18}NO_4$), 169 ($C_9H_{15}NO_2$). ^{1}H-NMR spectrum (deuteriochloroform): 1·32 + 1·52 (2× s, 2× 3 H) $(CH_3)_2C$; 2·0–2·35 (m, 2 H); 3·15–3·50 (m, 3 H); 4·38 (br d, 2 H); 4·70 (m, 2 H); 8·45 (br, 1 H) NH; 7·3–7·65 (3 H) + + 7·90–8·05 (2 H) arom. protons.

An additional product was obtained from the reaction mixture by recrystallisation from ethanol–light petroleum. Yield, 0·6 g (5·2 mmol) of the amide of 3-ethoxyacrylic acid, m.p. 130–132°C; R_F value 0·33 (in S_6). Mass spectrum: *m/e* 115 (M^+), 100 (M-15), 86, 71. For $C_5H_9NO_2$ (115·1) calculated: 52·17% C, 7·88% H, 12·17% N; found: 51·91% C, 8·04% H, 12·22% N.

1-(±)-(2,3-O-Isopropylidene-4*c*-hydroxymethyl-2*t*,3*t*-dihydroxycyclopent-1*r*-yl)-5-methoxycarbonyluracil (*XIV*)

A mixture of the crude residue *IV* (prepared from 25 mmol of compound *III* as described above), compound[10] *XII* (7·8 g; 30 mmol), and ethanol (100 ml) was refluxed for 3 h. Triethylamine (20 ml) was then added and the reflux continued for 8 h. The mixture was evaporated under diminished pressure and the residue refluxed in methanolic sodium methoxide (100 ml of 0·2M solution) for 2 h. The mixture was neutralised with dry Dowex 50 X 8 (H^+) ion exchange resin, filtered, and the material on the filter washed with methanol (200 ml). The filtrate and washings were combined and taken down under diminished pressure. Crystallisation of the residue from ethanol yielded 3·40 g (40%) of compound *XIV*, m.p. 233–234°C. For $C_{15}H_{20}N_2O_7$ (340·3) calculated: 52·94% C, 5·92% H, 8·23% N; found: 52·57% C, 5·94% H, 7·99% N. Mass spectrum: *m/e* (M^+), M-15, M-32. ^{1}H-NMR spectrum (deuteriochloroform): 1·33 + 1·52 (2× s, 2× 3 H) $(CH_3)_2C$; 1·75–2·20 (m, 3 H); 3·70–3·90 (m, 2 H); 4·60–4·90 (m, 3 H); 3·60 (br, 1 H) OH; 3·84 (s, 3 H) $COOCH_3$; 8·42 (s, 1 H) H_6; 11·50 (br, 1 H) NH.

1-(±)-(2,3-O-Isopropylidene-4*c*-hydroxymethyl-2*t*,3*t*-dihydroxycyclopent-1*r*-yl)uracil (*XVIII*)

A solution of compound *XIV* (6·8 g; 20 mmol) in 2M-NaOH (80 ml) was refluxed for 2 h, cooled down, neutralised with Dowex 50 X 8 (H^+) ion exchange resin, filtered, and the material on the filter washed with water. The filtrate and washings were combined and evaporated under diminished pressure. The residue was taken up into 40% aqueous pyridine (50 ml) and the solution applied to a column of pyridinium Dowex 50 X 8 ion exchange resin (150 ml); the column was eluted with 20% aqueous pyridine (1 000 ml). The eluate was evaporated under diminished pressure, the residue coevaporated with three 50 ml portions of pyridine, and dried over phosphorus pentoxide overnight. A mixture of the residue and quinoline (40 ml) was refluxed for 12 h, evaporated (80°C/0·1 Torr), the residue taken up into 50% aqueous methanol (100 ml), neutralised with Dowex 50 X 8 (H^+) ion exchange resin (50 ml), stirred for 10 min, the mixture filtered, and the material on the filter washed with methanol (300 ml). The filtrate and washings were combined and evaporated under diminished pressure. The residue was chromatographed on a column of silica gel (100 g) in chloroform; 95 : 5 chloroform–ethanol was used as eluant. The product-containing fractions were pooled, evaporated, and the residue crystallised from ethanol–light petroleum. Yield, 1·10 g (23·5%) of compound *XVIII*, m.p. 250–251°C. For $C_{13}H_{18}N_2O_5$ (282·3) calculated: 55·30% C, 6·42% H, 9·92% N; found: 55·09% C, 6·47% H, 9·83% N. ^{1}H-NMR

spectrum (deuteriochloroform): 1·20 + 1·38 (2× s, 2× 3 H) $(CH_3)_2C$; 1·60—2·10 (m, 3 H) (2 $H_{5'}$ + $H_{4'}$); 3·40—3·80 (m, 3 H) ($2H_{6'}$ + 5'-OH); 4·40—4·65 (m, 3 H) ($H_{1'}$ + $H_{2'}$ + $H_{3'}$); 5·48 br d, 1 H; $J_{5,NH} \leqq 1·0$, $J_{5,6} = 8·0$) H_5; 7·35 (d, 1 H; $J_{6,5} = 8·0$) H_6; 11·05 (br, 1 H) NH.

1-(±)-(2,3-O-Isopropylidene-4*c*-benzoyloxymethyl-2*t*,3*t*-dihydroxycyclopent-1*r*-yl)-5-ethoxycarbonyluracil (*XIII*)

A mixture of the amino derivative *IV* (prepared from 6 mmol of compound *III*), compound[10] *XII* (1·7 g; 6·5 mmol), and methanol (25 ml) was refluxed for 1 h, treated with triethylamine (4 ml), refluxed 2·5 h, and evaporated under diminished pressure. The residue was chromatographed on two layers of silica gel in the solvent system S_5. The *XIII*-containing bands (R_F value 0·43) were eluted with methanol (500 ml), the eluate evaporated under diminished pressure, and the residue crystallised from ethanol. Yield, 1·20 g (43·7%) of compound *XIII*, m.p. 245°C. For $C_{23}H_{26}N_2O_8$ (458·5) calculated: 60·24% C, 5·71% H, 6·11% N; found: 59·67% C, 5·91% H, 6·53% N. Mass spectrum: *m/e* 458 (M^+), 443 (M-15), 429 (M-29), 184 (BH^+), 167 (BH—NH_3) On heating under reflux condenser in 0·2M methanolic sodium methoxide for 1 h, compound *XIII* is quantitatively converted into compound *XIV*.

1-(±)-(4*c*-Hydroxymethyl-2*t*,3*t*-dihydroxycyclopent-1*r*-yl)uracil (*II*)

A solution of compound *XVIII* (1·4 g; 5 mmol) in 80% aqueous acetic acid (50 ml) was refluxed for 2 h, evaporated under diminished pressure, the residue coevaporated with three 20 ml portions of water and three 20 ml portions of ethanol, and finally crystallised from a mixture of ethanol and ether. Yield, 0·97 g (80%) of compound *II*, m.p. 130—132°C (decomp.). For $C_{10}H_{14}N_2O_5$ (242·2) calculated: 49·60% C, 5·82% H, 11·57% N; found: 49·26% C, 5·43% H, 11·30% N. Mass spectrum: *m/e* 242 (M^+), 224 (M-18), 113 (BH^+). UV spectrum (pH 2): λ_{max} 262 nm (ε_{max} 10200).

1-(±)-(4*c*-Acetoxymethyl-2*t*,3*t*-diacetoxycyclopent-1*r*-yl)uracil (*XIX*)

A mixture of compound *II* (0·40 g; 1·65 mmol), pyridine (10 ml), and acetic anhydride (10 ml) was kept at room temperature overnight and evaporated at 40°C/0·1 Torr. The residue was coevaporated with toluene under the same conditions and finally chromatographed on one layer of silica gel in the solvent system S_6. The *XIX*-containing band (R_F value 0·42) was eluted with methanol, the eluate evaporated under diminished pressure, and the residue crystallised from chloroform–light petroleum. Yield, 0·40 g (66%), m.p. 143—145°C. For $C_{16}H_{20}N_2O_8$ (368·3) calculated: 52·17% C, 5·47% H, 7·60% N; found: 51·98% C, 5·55% H, 7·53% N. ^{1}H-NMR spectrum (deuteriochloroform): 1·75—2·75 (m, 3 H); 1·95—2·09 (3× s, 3× 3 H) $OCOCH_3$; 4·05—4·35 (m, 2 H); 5·20—5·55 (m, 3 H); 5·67 (d, 1 H; $J_{5,6} = 8·0$, $J_{5,NH} \leqq 1·0$) H_5; 7·44 (d, 1 H, $J_{6,5} = 8·0$) 10·90 (br, 1 H) NH. IR spectrum (tetrachloromethane): ν(C=N) 1635 cm^{-1}, ν(C=O) 1714 cm^{-1} (s, sh), 1695 cm^{-1} (vs), 1755 cm^{-1} (s), 1744 cm^{-1} (s, sh); ν(NH) 3405 cm^{-1}.

1-(±)-(4*c*-Acetoxymethyl-2*t*,3*t*-diacetoxycyclopent-1*r*-yl)-5-methoxycarbonyluracil (*XVI*)

A solution of compound *XIII* (0·5 g; 1·47 mmol) in 80% aqueous acetic acid was refluxed for 1 h, evaporated under diminished pressure, the residue coevaporated with three 20 ml portions of pyridine, and the final residue stirred with a pyridine–acetic anhydride mixture (10 ml each) at room temperature overnight. The mixture was processed analogously to compound *XIX*.

Crystallisation from ethyl acetate–light petroleum yielded 0·45 g (72%) of compound *XVI*, m.p. 163–164°C; R_F value 0·30 (in S_5). For $C_{18}H_{22}N_2O_{10}$ (426·4) calculated: 50·69% C, 5·20% H, 6·57% N; found: 50·64% C, 5·40% H, 6·18% N. ^{1}H-NMR spectrum (deuteriochloroform): 1·70–2·60 (m, 3 H), 1·91 + 2·02 + 2·18 (3× s, 3× 3 H) $OCOCH_3$; 3·81 (s, 3 H) 5-$COOCH_3$; 4·0–4·35 (m, 2 H); 5·20–5·60 (m, 3 H); 8·46 (s, 1 H) H_6; 11·54 (br, 1 H) NH. IR spectrum (tetrachloromethane): ν(C=N) 1623 cm^{-1}, ν(C=O) 1695 cm^{-1} (s), 1714 cm^{-1} (s, sh), 1720 cm^{-1} (vs), 1754 cm^{-1} (s), ν(NH) 3398 cm^{-1}.

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