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Pyrimidines. XVII. 1-(Tetrahydro-2-pyranyl)cytosine, -uracil, -thymine and Related Compounds (1)

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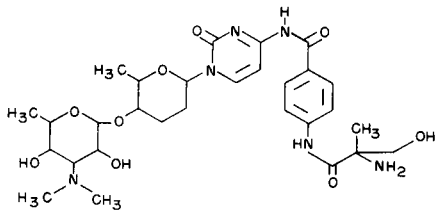
In view of the reported biological activities of antibiotics Amicetin and Blasticidin S, 1-(tetrahydro-2-pyranyl)cytosine, -uracil, -thymine and related compounds were synthesized. These compounds were prepared by the Hilbert-Johnson method from 2-chlorotetrahydropyran and corresponding 2,4-dimethoxypyrimidines.

Naturally occurring pyrimidines bearing a tetrahydro-2-pyranyl ring substituted at position-1 of pyrimidine have been reported. Amicetin (2), for instance, an antibiotic isolated from *Streptomyces vinaceus-drappus* (2b) and *Streptomyces plicatus* (2e), was found to consist of a cytimidine unit and an amosamine unit linked through a 6-methyltetrahydro-2-pyranyl ring (I) (2g). This antibiotic was reported to inhibit M protein synthesis as well as growth of streptococcal cells at the same concentration (3). Blasticidin S, another antibiotic isolated from the culture of *Streptomyces griseochromogens* which possesses both antitumor activity (4) and inhibitory activity against rice blast disease (5), was found to be a 1-(substituted dihydro-2-pyranyl)cytosine (6) (II).

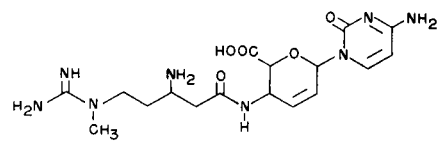
A number of pyrimidine derivatives containing a sugar moiety other than the naturally occurring ribose or deoxyribose have recently been studied. Among these, cytosine arabinoside was found to possess antiviral activity against DNA viruses such as herpes simplex vaccinia and the viral antigen of Simian virus 40 (7). In addition, activity against various malignant neoplasms was also reported (8).

Substitution of tetrahydropyran or tetrahydrofuran ring systems for ribose in a number of purine derivatives has resulted in compounds with considerable antitumor activity (9). 9-(Tetrahydro-2-furyl)-6-mercaptopurine exhibited a therapeutic index (against Ca-755) of approximately 200 as compared to 30 for 6-MP against the same tumor (9c). 9-(Tetrahydro-2-pyranyl)-6-mercaptopurine possesses a therapeutic index comparable to 6-MP riboside in Ca-755 (10). Contrary to general belief, this activity is perhaps due not to the parent purine bases resulting from *in vivo* dealkylation, but rather to the contribution of the tetrahydropyranyl or tetrahydrofuryl moiety. This is best illustrated by the report that 9-(tetrahydro-2-furyl)*adenine* demonstrated confirmed activity against Ca-755 (9c). In a parallel case, a number of 9-substituted-6-mercaptopurines were effective inhibitors of 6-MP-*resistant* H. Ep. No. 2 cells and Ca-755 cells (11).

The foregoing information clearly indicated that pyrimidines containing a tetrahydropyranyl or tetrahydrofuryl ring at the 1-position should be investigated. In view of the reported biological activities of the antibiotics Amicetin (I) and Blasticidin S (II), synthesis of 1-(tetrahydro-2-pyranyl)pyrimidines was undertaken in our laboratory.



I



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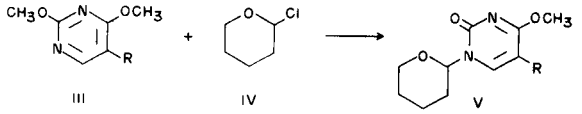

III

a. R = H
b. R = CH_3
c. R = Br
d. R = I

IV

V

a. R = H
b. R = CH_3
c. R = Br
d. R = I

VI

a. R = H, X = O
b. R = CH_3, X = O
c. R = Br, X = O
d. R = I, X = O
e. R = H, X = S

VII

a. R = H, Y = NH_2
b. R = H, Y = $NHCH_3$
c. R = CH_3, Y = NH_2
d. R = Br, Y = NH_2
e. R = I, Y = NH_2

Preparation of 1-(tetrahydro-2-pyranyl)pyrimidines by direct condensation of dihydropyran with pyrimidines, as in the case of the preparation of 9-(tetrahydro-2-pyranyl)purines (9), could not be achieved in our hands. Consequently, other methods of synthesis were sought. Hilbert and Johnson in 1930 (12) reported that 2,4-dialkoxypyrimidines can be caused to rearrange into 1,3-dialkyluracils and, furthermore, alkylation of these dialkoxypyrimidines with alkyl halide readily yielded 1-substituted uracils. This convenient synthetic method has since been used for the preparation of several pyrimidine derivatives (13). In our laboratory it was found that when *freshly prepared* 2-chlorotetrahydropyran (IV) (14) was condensed with 2,4-dimethoxypyrimidine (IIIa) (12), the desired 1-(tetrahydro-2-pyranyl)-2-oxo-4-methoxy-1,2-dihydropyrimidine (Va) was obtained in good yield. The 4-methoxy group of the resulting compound was quite labile in aqueous mineral acid and was readily converted to the 1-substituted uracil (VIa) at room temperature. The corresponding cytosine analog (VIIa) and 1-(tetrahydro-2-pyranyl)-2-oxo-4-methylamino-1,2-dihydropyrimidine (VIIb) were obtained by treating with aqueous ammonia and aqueous methylamine, respectively.

The Hilbert-Johnson method was extended to the preparation of derivatives of other naturally occuring pyrimidines and their analogs. 2,4-Dimethoxy-5-methylpyrimidine (IIIb) (15) and IV gave 1-(tetrahydro-2-pyranyl)-2-oxo-4-methoxy-5-methyl-1,2-dihydropyrimidine (Vb), isolated as a clear oil. This was allowed to stir in dilute hydrochloric acid to give the thymine analog VIb. The corresponding 4-amino derivative (VIIc) was readily prepared from Vb and ammonia under similar reaction conditions. 1-(Tetrahydro-2-pyranyl)-5-bromouracil (VIc) and 1-(tetrahydro-2-pyranyl)-5-bromocytosine (VIId) as well as the corresponding 5-iodo analogs (VId and VIIe) were prepared accordingly. 1-(Tetrahydro-2-pyranyl)-4-thiouracil (VIe) was obtained by refluxing a mixture of Va and sodium hydrosulfide in methanol. At the present no effort is being made to resolve the racemic mixtures which were undoubtedly formed during the condensation reactions between III and IV.

The ultraviolet absorption spectra of VIa and VIIa bear striking resemblances to those of uridine and cytidine, respectively. Substitution of methyl, bromo, or iodo group at position-5 of these pyrimidines caused progressive bathochromic shifts of the ultraviolet absorption maxima. Ultraviolet absorption properties of VIe and 4-thiothymidine (16) were also essentially similar.

EXPERIMENTAL (17)

1-(Tetrahydro-2-pyranyl)-2-oxo-4-methoxy-1,2-dihydropyrimidine (Va).

To 48 g. (0.40 mole) of *freshly prepared* 2-chlorotetrahydropyran (14) (IV) in 400 ml. of dichloromethane was added 58 g. (0.41 mole) of 2,4-dimethoxypyrimidine (IIIa). The solution was stirred at room temperature for 30 hours. A small amount of precipitate, which formed during that time, was removed by filtration. The filtrate was evaporated on a water bath under reduced pressure to give an oily substance. The oil was stirred with petroleum ether (b.p. 35-60°) to remove any unreacted IIIa. The solvent layer was decanted and the remaining oil dissolved in 300 ml. of anhydrous ether. The ethereal solution was evaporated to dryness *in vacuo* at room temperature. The residual solid which weighed 60 g. (71.5% yield) and melted at 64-66°, was recrystallized from small amounts of anhydrous ether to give 51 g. (60.7% yield) of analytically pure product, m.p. 68-69°; λ max (CH_3OH), 274 mμ (ε, 7,800).

Anal. Calcd. for $C_{10}H_{14}N_2O_3$: C, 57.1; H, 6.72; N, 13.3. Found: C, 56.8; H, 7.14; N, 13.4.

1-(Tetrahydro-2-pyranyl)-2-oxo-4-methoxy-5-methyl-1,2-dihydropyrimidine (Vb).

To 38 g. (0.32 mole) of *freshly prepared* IV in 350 ml. of dichloromethane was added 50 g. (0.33 mole) of 2,4-dimethoxy-5-methylpyrimidine (15) (IIIb). The solution was stirred for 30 hours at room temperature and the solvent evaporated under reduced pressure. The resulting oil was stirred with 300 ml. of petroleum ether (b.p. 35-60°) and the ether layer decanted. This process was repeated three times. The last trace of unreacted IIIb was removed by heating the oily residue at 90° under reduced pressure for 1 hour. The remaining oil was then dissolved in 200 ml. of anhydrous ether, boiled, treated with charcoal and filtered. The clear ethereal solution was then poured into 600 ml. of petroleum ether and a small amount of insoluble material was removed by filtration. The filtrate was then evaporated on a water bath under reduced pressure to yield 38 g. of clear oil (λ max (CH_3OH), 280 mμ), which was used directly for the preparation of VIb and VIIc.

1-(Tetrahydro-2-pyranyl)-2-oxo-4-methoxy-5-bromo-1,2-dihydropyrimidine (Vc).

This compound was prepared from 95 g. of 2,4-dimethoxy-5-bromopyrimidine (18) (IIIc), 50 g. of *freshly prepared* 2-chlorotetrahydropyran (IV) and 300 ml. of dichloromethane according to the procedure for the preparation of Vb. A yield of 95 g. of Vc was obtained as clear, viscous oil (λ max (CH_3OH), 280 mμ). It was used directly for the preparation of VIc and VIId.

1-(Tetrahydro-2-pyranyl)-2-oxo-4-methoxy-5-iodo-1,2-dihydropyrimidine (Vd).

This compound was similarly prepared from 50 g. of 2,4-dimethoxy-5-iodopyrimidine (13a) (IIId) and 22.5 g. of *freshly prepared* IV in 250 ml. of dichloromethane. A solid crude product was obtained which, upon two recrystallizations from ethyl acetate, gave 29 g. (46.0%) yield) of Vd, m.p. 153-156°; λ max (CH_3OH), 298 mμ (ε, 5,700).

Anal. Calcd. for $C_{10}H_{13}IN_2O_3$: C, 35.7; H, 3.90; N, 8.33. Found: C, 35.9; H, 4.10; N, 8.33.

1-(Tetrahydro-2-pyranyl)uracil (VIa).

To a solution of 10 g. of Va in 50 ml. of water was added 3 ml. of concentrated hydrochloric acid. The mixture was stirred at room temperature for 20 minutes and the precipitate that had formed was filtered, washed with water and acetone, and dried at 80° to give 9.3 g. (97.5% yield) of white crystals, m.p. 185-187°. Recrystallization from water yielded analytically pure product, m.p. 188-190°; λ max (H_2O), 259 mμ (ε, 11,000); λ max (*p*H 1), 259 mμ (ε, 12,900); λ max (*p*H 11), 230 (ε, 5,900), 259 mμ (ε, 9,700).

Anal. Calcd. for $C_9H_{12}N_2O_3 \cdot {}^1/_2H_2O$: C, 52.8; H, 6.40; N, 13.7. Found: C, 53.2; H, 6.73; N, 13.9.

1-(Tetrahydro-2-pyranyl)thymine (VIb).

A mixture of 20 g. of Vb, 150 ml. of water and 5 ml. of concentrated hydrochloric acid was stirred at room temperature for 1 hour and allowed to cool overnight in the refrigerator. The resulting precipitate was filtered and washed with iced water. Recrystallization of the crude product from 300 ml. of boiling water gave 8 g. (42.6% yield) of analytically pure product, m.p. 175-176°; λ max (H_2O), 265 mμ (ε, 8,600); λ max (*p*H 1), 265 mμ (ε, 9,500); λ max (*p*H 11), 277 (ε, 6,750), 265 mμ (ε, 7,800).

Anal. Calcd. for $C_{10}H_{14}N_2O_3$: C, 57.1; H, 6.72; N, 13.3. Found: C, 57.2; H, 6.75; N, 13.6.

1-(Tetrahydro-2-pyranyl)-5-bromouracil (VIc).

To a suspension of 13 g. of Vc in 150 ml. of water and 10 ml. of concentrated hydrochloric acid at less than 10° was added, with stirring, 50 ml. of ethanol. The resulting solution was rapidly stirred at less than 20° for 30 minutes then evaporated *in vacuo* with no external heating. The crude solid, m.p. 194-196°, was washed with ether and recrystallized from 50% aqueous methanol to give 9 g. (72.6% yield) of VIc, m.p. 202-203°; λ max (H_2O), 277 mμ (ε, 10,400);

λ max (*p*H 1), 277 mμ (ε, 11,000); λ max (*p*H 11), 232 (ε, 6,600), 274 mμ (ε, 7,400).

Anal. Calcd. for $C_9H_{11}BrN_2O_3$: C, 39.3; H, 4.02; N, 10.2. Found: C, 39.0; H, 4.08; N, 10.4.

1-(Tetrahydro-2-pyranyl)-5-iodouracil (VId).

To a solution of 10 g. of Vd in 260 ml. of methanol and 130 ml. of water at less than 20° was added, with stirring, 10 ml. of concentrated hydrochloric acid. The mixture was stirred for 2 hours at 20-25° then evaporated to dryness *in vacuo* at room temperature. The residue was washed well with cold water then recrystallized from water to give 8.2 g. (85.5% yield) of VId. The product softened at 140° and melted between 152° and 162°; λ max (H_2O), 283 mμ (ε, 8,000); λ max (*p*H 1), 288 mμ (ε, 7,100); λ max (*p*H 11), 230 (ε, 11,000), 283 mμ (ε, 5,800).

Anal. Calcd. for $C_9H_{11}IN_2O_3$: C, 33.5; H, 3.44; N, 8.70. Found: C, 33.6; H, 3.68; N, 8.64.

1-(Tetrahydro-2-pyranyl)-2-thiouracil (VIe).

A moderate stream of dry hydrogen sulfide was passed into a methanolic solution (150 ml.) of 3.5 g. of sodium methoxide and 10 g. of Va. During this time the solution was heated and refluxed for 2 hours. The reaction mixture was then evaporated to dryness *in vacuo* and the residue dissolved in 150 ml. of water. With cooling, the solution was adjusted to *p*H 1 with dilute hydrochloric acid. The resulting yellow precipitate was filtered and washed with cold water. The crude product was recrystallized three times from water to give 1.2 g. (11.9% yield) of analytically pure VIe, m.p. 180-181°; λ max (H_2O), 245 (ε, 2,900), 328 mμ (ε, 22,200); λ max (*p*H 1), 246 (ε, 3,600), 329 mμ (ε, 24,000); λ max (*p*H 11), 234 (ε, 4,600), 314 mμ (ε, 21,200).

Anal. Calcd. for $C_9H_{12}N_2O_2S$: C, 51.0; H, 5.70; N, 13.2. Found: C, 50.6; H, 5.95; N, 13.1.

1-(Tetrahydro-2-pyranyl)cytosine (VIIa).

A stream of gaseous ammonia was passed through a refluxed and stirred solution of 20 g. of Va in 250 ml. of 28% aqueous ammonia at a moderate rate for 3 hours. The resulting solution was allowed to cool and the precipitate was filtered, washed with acetone and dried at 80° to give 14 g. (75.5% of yield) of white crystals, m.p. 250-252°. Recrystallization from water afforded analytically pure product, m.p. 253-255°; λ max (H_2O), 268 mμ (ε, 9,200); λ max (*p*H 1), 276 mμ (ε, 13,650); λ max (*p*H 11), 235 (ε, 7,700), 268 mμ (ε, 9,200).

Anal. Calcd. for $C_9H_{13}N_3O_2$: C, 55.4; H, 6.71; N, 21.5. Found: C, 55.2; H, 6.63; N, 21.4.

1-(Tetrahydro-2-pyranyl)-2-oxo-4-methylamino-1,2-dihydropyrimidine (VIIb).

A solution of 10 g. of Va and 30 ml. of 40% methylamine in 150 ml. of water was stirred at room temperature for 90 minutes. The reaction mixture was then evaporated to dryness *in vacuo* and the residue was added to 200 ml. of boiling benzene. To the boiling suspension was added just enough methanol to afford a complete solution. It was then decolorized with charcoal and filtered. The volume of the filtrate was then reduced to 90-100 ml. and upon cooling, 8 g. (80.4% yield) of analytically pure VIIb was collected, m.p. 185°; λ max (H_2O), 235 (ε, 12,400), 268 mμ (ε, 15,700); λ max (*p*H 1), 279 mμ (ε, 18,200); λ max (*p*H 11), 239 (ε, 11,800), 268 mμ (ε, 15,700).

Anal. Calcd. for $C_{10}H_{15}N_3O_2$: C, 57.5; H, 7.23; N, 20.1. Found: C, 57.7; H, 7.51; N, 19.8.

1-(Tetrahydro-2-pyranyl)-5-methylcytosine (VIIc).

A stream of gaseous ammonia was passed through a refluxed and stirred solution of 15 g. of Vb in 300 ml. of 28% aqueous ammonia at a moderate rate for 6 hours. The resulting solution was evaporated under reduced pressure to a gummy residue. This was triturated with ethyl acetate, which gave a crystalline solid. It was added into 100 ml. of boiling benzene followed by the addition of a few drops of methanol. The resulting clear solution was treated with charcoal and filtered. The volume of the filtrate was reduced until the sign of cloudiness appeared. It was cooled and the precipitate which had formed was collected by filtration. After drying at 80° for 18 hours the product, which is analytically pure, weighed 13 g. (92.9% yield), m.p. 222-223°; λ max (H_2O), 275 mμ (ε, 10,000); λ max (*p*H 1), 284 mμ (ε, 16,000); λ max (*p*H 11), 235 (ε, 4,600), 275 mμ (ε, 10,450).

Anal. Calcd. for $C_{10}H_{15}N_3O_2$: C, 57.5; H, 7.23; N, 20.1. Found: C, 57.6; H, 7.45; N, 20.4.

1-(Tetrahydro-2-pyranyl)-5-bromocytosine (VIId).

A mixture of 20 g. of Vc and 220 ml. of ethanolic ammonia (saturated at 0°) was heated in a sealed steel bomb at 100° for 12 hours. The reaction mixture was evaporated to dryness and the residue recrystallized from a mixture of water and methanol to give 9.2 g. (48.5% yield) of VIId, m.p. 225-229° dec.; λ max (H_2O), 285 mμ (ε, 8,200); λ max (*p*H 1) 297 mμ (ε, 13,200); λ max (*p*H 11), 235 (ε, 8,200), 285 mμ (ε, 8,200).

Anal. Calcd. for $C_9H_{12}BrN_3O_2$: C, 39.4; H, 4.40; N, 15.3. Found: C, 39.2; H, 4.70; N, 15.1.

1-(Tetrahydro-2-pyranyl)-5-iodocytosine (VIIe).

This compound was similarly prepared from 10 g. of Vd and 120 ml. of ethanolic ammonia. Recrystallization of the crude reaction product from methanol-water gave 7.5 g. (78.5% yield) of VIIe which started to decompose at 190° and melted at 204-206°; λ max (H_2O), 291 mμ (ε, 6,400); λ max (*p*H 1), 306 mμ (ε, 9,300); λ max (*p*H 11), 227 (ε, 12,500), 291 mμ (ε, 6,500).

Anal. Calcd. for $C_9H_{12}IN_3O_2$: C, 33.7; H, 3.76; N, 13.1. Found: C, 33.9; H, 3.81; N, 13.0.

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