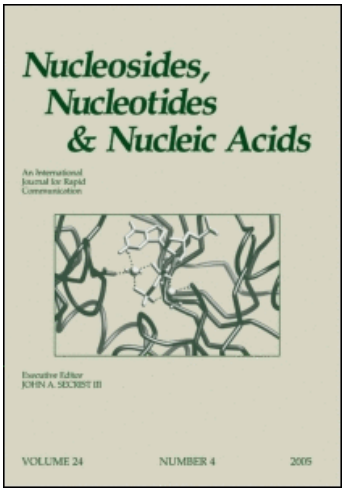

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SYNTHESIS AND BIOLOGICAL ACTIVITY OF CARBOCYCLIC CLITOCINE

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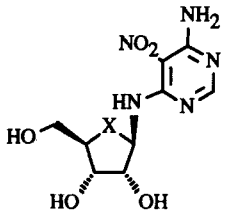
Abstract: The preparation and anti-viral activity of carbocyclic clitocine and some related compounds are described.

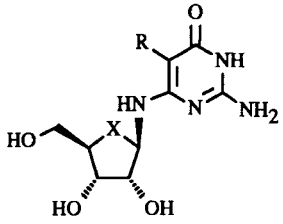
Clitocine [6-amino-5-nitro-4-(β-D-ribofuranosylamino)pyrimidine] (1) was isolated from the mushroom ***Clitocybe inversa*** by Kubo *et al.*,[1] and shown to exhibit potent insecticidal activity against the pink bollworm ***Pectinophora gossypiella***. Chemical synthesis of (1)[2,3] and further biological evaluation[2] have been reported. Clitocine exhibits potent inhibitory effects on several leukaemia cell lines (ED_{50}= 3 x 10^{-8}M) and is also a substrate and an inhibitor of adenosine kinase.

Evidently, clitocine (1) bears a close structural similarity to adenosine. Preparation of the related guanosine-type analogue (2) has also been recently reported.[4]

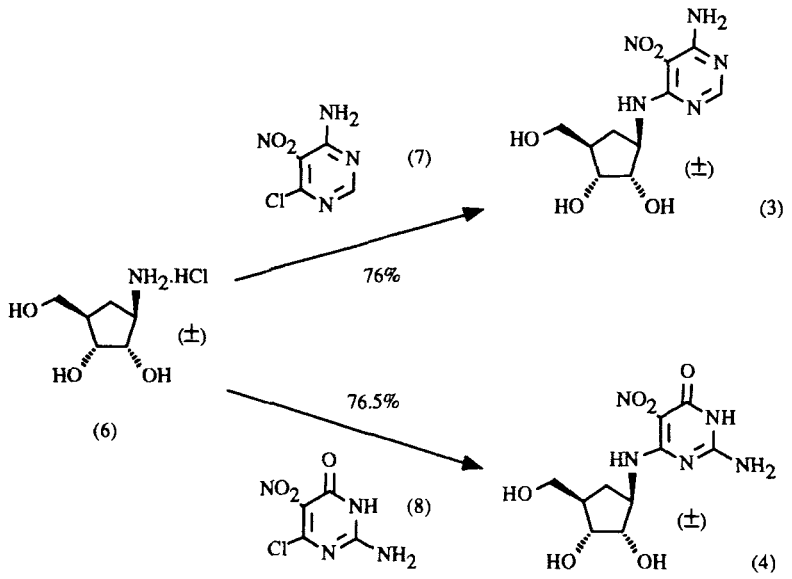
As part of our continuing interest in anti-viral carbocyclic nucleosides, the analogues (3) and (4) and the related compound (5) represented appropriate targets.



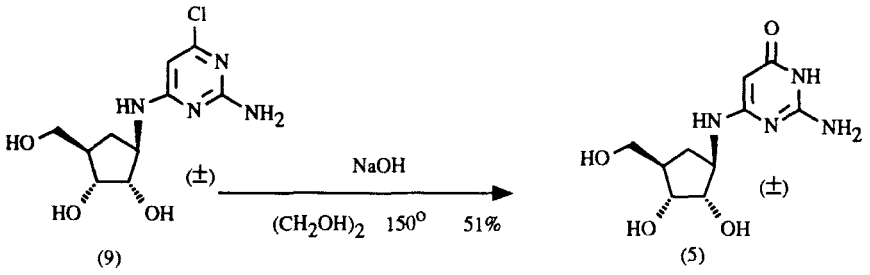
X = O (1)
X = CH_2 (3)



R = NO_2, X = O (2)
R = NO_2, X = CH_2 (4)
R = H, X = CH_2 (5)



Scheme 1.



Scheme 2.

Preparation of (±)-carbocyclic clitocine (3) has been recently reported.[5] We prepared (±)-(3) in good yield from the readily available aminotriol (6)[6] and 4-chloro-5-nitro-6-aminopyrimidine (7)[7] in triethylamine and ethanol at reflux. The guanosine-type carbocycle (4) was similarly prepared from (6) and 2-amino-6-chloro-5-nitro-4(3H)pyrimidinone (8).[8] Scheme 1.

^{1}H NMR studies of (3) and (4) have confirmed that a strong hydrogen bond exists between the NO_2 group and the 4-NH [δ(d_6DMSO) 9.7, d J=8 Hz].

Compound (5) was prepared from the known chloropyrimidine (9)[9] by hydrolysis under forcing conditions. Scheme 2.

Table 1. *in vitro* Biological Activity

Inhibition of virus replication (IC_{50})					
Compd	Extent of phosphorylation by Adenosine Kinase (A=1)	ELISA assay (μM)	Plaque reduction assay [toxicity] (μM)		Toxicity to L1210 cells (μM) (mouse DBA/2)
		influenza A (Singapore/1/57)	influenza A (Singapore/1/57)	influenza B (Victoria/102/85)	
3	1.48	70	5.2 [>50 0]	105.2 [>500]	1.75
4	0.09	99	165 [>500]	>500 [>500]	>50
5	0.11	>1mM	-	-	>50

Carbocyclic clitocine (3) is readily phosphorylated by adenosine kinase and shows potent and selective activity against influenza A (Singapore/1/57) *in vitro*. When tested *in vivo* against influenza A (Singapore) in mice (i.p.),significant activity was seen at 100mg/kg.

The compounds were routinely tested against Herpes, Retro and Myxoviruses. Significant activity was only found against influenza. Table 1.

Experimental

(1β,2α,3α,4β)-4-[[4-amino-5-nitropyrimidin-6-yl]amino]-2,3-dihydroxycyclopentane methanol (3).

A mixture of (6) (2.94g, 18.3mM), (7) (3.33g, 18.3mM) and triethylamine (12.96g, 0.128M) in ethanol (72 ml) was heated to reflux for one hour. The precipitate was collected by filtration and dried (50° *in vacuo 24h.)* to afford the **title compound** (3.96g, 76%) as a colourless solid. m.p. 219-221°. UV: λ_{max} (EtOH) 339.4 nm, $E^{1\%}_{1cm}312$. IR: ν_{max} (nujol) 3432 (sh), 3270, 1661, 1516 cm^{-1}. ^1H NMR: (d_6-DMSO) δ 9.21 (1H, d, J 8.0 Hz, CH*NH)*, 8.58 (2H, bd, NH_2), 7.99 (1H, s, pyrimidine-CH), 4.80 (1H, d, J 5.0 Hz, OH), 4.75 (1H, t, J 5 Hz, 5′ OH), 4.48 (1H, d, J 5.0 Hz, OH), 4.45 (1H, m, *CH*NH), 3.75 (2H, m, 2′ and 3′ *CH*OH), 3.39 (2H, m, CH_2OH), 2.20 (1H, m, 6′a-CH), 1.93 (1H, m, 4′-CH), 1.2 (m, 6′b-CH, partially masked by EtOH) ppm. Signals at 8.58, 4.80, 4.75 and 4.48 disappeared on addition of D_2O. Found: C, 41.58; H, 5.69; N, 22.56; $C_{10}H_{15}N_5O_5$ 0.4 EtOH 0.4 H_2O requires C, 41.72; H, 5.90; N, 22.53.

(1β,2α,3α,4β)-2-amino-6-[[2,3-dihydroxy-4-(hydroxymethyl)cyclopentyl]-amino]-5-nitropyrimidin-4-one (4)

A mixture of (6) (1.0g, 5.24 mM), (8) (0.963g, 5.24 mM) and triethylamine (3.71g, 3.67 M) in ethanol (70 ml) was heated to reflux for 1.33 hours. The resulting

precipitate was collected by filtration and washed with ethanol to afford the **title compound** (4) (1.21g, 76.5%) as colourless crystals. m.p. 183-185° (dec). IR: ν_{max} (nujol) 3466 (sh), 3298, 3225, 1722, 1640 cm $^{-1}$. ^{1}H-NMR (d_6-DMSO) δ 10.8 (1H, bs, CO*NH*), 9.62 (1H, d, J 8.0 Hz CH*NH*), 7.9 (1H, bs) and 6.7 (1H, bs, NH_2), 4.81 (1H, d, J 5.0 Hz, OH), 4.70 (1H, t, J 5.0 Hz, 5′ OH), 4.49 (1H, bs, OH), 4.38 (1H, m, *CH*NH), 3.78 (2H, m, 2′ and 3′ *CH*OH), 3.42 (2H, m, CH_2OH), 2.25 (1H, m, 6′a-CH), 1.92 (1H, m, 4′-CH), 1.18 (1H, m, 6′b-CH) ppm. Signals at 10.8, 7.9, 6.7, 4.81, 4.70 and 4.49 disappeared on addition of D_2O. Found: C, 39.51; H, 5.23; N, 22.95; $C_{10}H_{15}N_5O_6$ requires C, 39.84; H, 5.02; N, 23.25.

(1β,2α,3α,4β)-2-amino-6-[[2,3-dihydroxy-4-(hydroxymethyl)cyclopentyl]-amino]pyrimidin-4-one (5)

A mixture of compound (9) (1.0g, 3.64 mM), sodium hydroxide pellets (1.0g, 25.6 mM) and ethylene glycol (10 ml) was heated at 150° for 20 min. The solvent was removed *in vacuo* (< 1 mbar). The residue was dissolved in water, acidified with 2N hydrochloric acid and the solvent evaporated. The residue was triturated with ethanol and salt separated. The filtrate was evaporated to afford an oil that crystallized slowly from aqueous ethanol to afford the **title compound** (476 mg, 51%) as a colourless crystalline solid. m.p. 150-152 °. UV: λ_{max} (H_2O) 268.8 nm. ($E^{1\%}_{1cm}$594.9) IR: ν_{max} (nujol) 3450-2747 (several sh), 1703, 1632, 1531 cm^{-1}. ^{1}H NMR (d_6-DMSO) 8.20 (1H, bs, NH), 7.70 (2H, bs, NH_2), 5.27 (1H, bs, CH*NH*), 4.50-3.80 (6H, br, 3 x OH, H_2O and pyrimidinone-CH), 3.65 (2H, m, 2′ and 3′ *CH*OH), 3.37 (2H, m, CH_2OH), 2.12 (1H, m, 6′a-CH), 1.90 (1H, m, 4′-CH), 1.08 (1H, m, 6′b-CH) ppm. ^{13}C NMR (d_6-DMSO) 170-155 (4 br, pyrimidinone-C) 78.0, 77.0, 74.0, 64.0, 47.0, 31.5 ppm. Mass spectrum (thermospray) 257 $[MH]^+$, 239 $[MH\text{-}H_2O]^+$, 127. Found: C, 38.35; H, 6.09; N, 17.88. $C_{10}H_{16}N_4O_4$. HCl. H_2O requires C, 38.65; H, 6.16; N, 18.03.

REFERENCES

1. I. Kubo, M. Kim, W.F. Wood, H. Naoki. *Tetrahedron Letters* 1986, *27*, 4277
2. R.J. Moss, C.R. Petrie, R.B. Meyer Jr., L.D. Nord, R.C. Willis, R.A. Smith, S.B. Larson, G.D. Kini, R.K. Robins. *J. Med. Chem.* 1988, *31*, 786
3. T. Kamikawa, S. Fujie, Y. Yamagiwa, M. Kim, H. Kawaguchi. *J. Chem. Soc. Chem. Commun.* 1988, 195
4. J.C. Espie, M.F. Lhomme, C. Morat, J. Lhomme. *Tetrahedron Letters* 1990, *31*, 1423
5. C.F. Palmer, K.P. Parry, S.M. Roberts. *Tetrahedron Letters* 1990, *31*, 279
6. R.C. Cermak, R. Vince. *Tetrahedron Letters* 1981, *22*, 2331
7. S.M. Greenburg, L.O. Ross, R.K. Robins. *J. Org. Chem.* 1959, *24*, 1314
8. D.T. Browne, J. Eisinger, N.J. Leonard. *J. Amer. Chem. Soc.* 1968, *90*,7302
9. Y.F. Shealey, J.D. Clayton. *J. Pharm. Sci.* 1973, *62*, 1432