Phytochemistry, 1973, Vol. 12, pp 2051 to 2054 Pergamon Press Printed in England

THREE NEW ALKALOIDS FROM *BUXUS PAPILLOSA*

MAURICE SHAMMA and VASSIL ST GEORGIEV
Department of Chemistry, The Pennsylvania State University, University Park, PA 16802, U S A

and

GHULAM A MIANA and F SULTANA KHAN
Institute of Chemistry, University of Islamabad, Islamabad, Pakistan

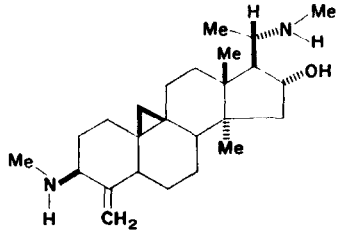
(*Received* 24 *January* 1973 *Accepted* 9 *March* 1973)

Key Word Index—*Buxus papillosa*, Buxaceae, steroidal alkaloids

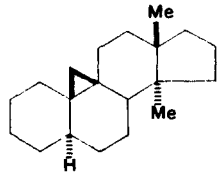
Abstract—Three new steroidal alkaloids have been obtained from *Buxus papillosa* Schneider These are (−)-cyclobuxupaline-*C* (IV), (+)-cyclopapilosine-*D* (VII) and (+)-buxamine-*C* (IX) A known alkaloid also present is desoxy-16-buxidienine (X)

INTRODUCTION

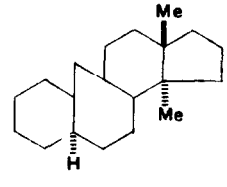
PLANTS of the genus *Buxus* (Buxaceae) are a rich source of steroidal alkaloids [1] Since Brown and Kupchan established the structure of cyclobuxine-*D* (I),[2] isolated from *B sempervirens* L, several *Buxus* species have been characterized It is noteworthy that except for irehine,[3] which is a 3β-hydroxy-20α-dimethylamino-Δ^5-pregnene, *Buxus* alkaloids can be divided into two groups, namely derivatives of cyclo-9β,19-pregnane-5α (II), and derivatives of *abeo*-9(10 → 19)-pregnane-5α (III) [4]



(I)



(II)



(III)

[1] (a) ČERNY, V and ŠORM, F (1967) *The Alkaloids* (MANSKE, R H F, ed), Vol 9, p 375, Academic Press, New York, (b) BROWN, JR, K S (1970) in *Chemistry of the Alkaloids* (PELLETIER, S W, ed), p 631, Van Nostrand-Reinhold, New York, (c) GOUTAREL, R (1971) in *Specialist Periodical Reports The Alkaloids*, Vol 1, p 407, The Chemical Society (London), London, (1972) *ibid* Vol 2, p 266

[2] BROWN, JR, K S and KUPCHAN, S M (1962) *J Am Chem Soc* **84**, 4590, 4592

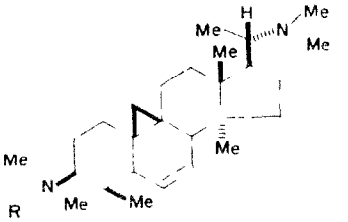
[3] VOTICKÝ, Z and TOMKO, J (1965) *Coll Czech Chem Commun* **30**, 348

[4] (a) HERLEM-GAULIER, D, KHOUNG-HUU-LAINÉ, F, STANISLAS, E and GOUTAREL, R (1965) *Bull Soc Chim Fr* 657, (b) NAKANO, T and TERAO, S (1965) *J Chem Soc* 4512.

As part of a study of the medicinal plants of Pakistan, we had occasion to look at the alkaloids of *B papillosa* Schneider [5] In the course of these investigations several alkaloids were isolated,[6 7] and herein we report the structures of three new alkaloids

RESULTS

The first alkaloid, (—)-cyclobuxupaline-*C* (IV), $C_{27}H_{46}N_2$, showed IR bands at 3340 cm^{-1} (N–H) and 1645 cm^{-1} (C=C) The NMR spectrum of the alkaloid contained signals for four *C*-methyl singlets at δ0 70 (3H), 0 74 (3H) and 0 95 (6H), a secondary C_{21}-methyl doublet centered at δ0 83 (*J* 6 Hz), and a dimethylamino and an *N*-methyl singlet at δ2 20 (6H) and 2 38 (3H), respectively A broad absorption was also present at δ5 30 (2H) attributable to the vinylic hydrogens of a disubstituted double bond [4a 8 9]



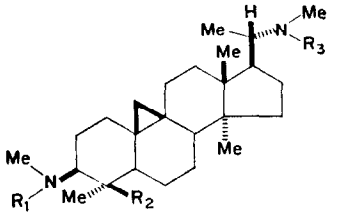
(IV) R = H
(V) R = Ac

The MS of cyclobuxupaline-*C* (IV) exhibited a parent peak *m/e* 398 (5) for the formula $C_{27}H_{46}N_2$, and a base peak *m/e* 72 corresponding to the fragment Me $-CH=N^+(Me)_2$ thus indicating that the dimethylamino group is located at C_{20} [10]

The alkaloid could be readily acetylated to *N*-acetylcyclobuxupaline-*C* (V), $C_{29}H_{48}N_2O$ More significant was the hydrogenation of IV with Adams catalyst which gave rise to crystalline (+)-dihydrocyclobuxupaline-*C* (VI), $C_{27}H_{48}N_2$, which showed no signals for vinylic protons in the NMR spectrum A salient feature of the spectrum was an *AB* system centred at δ0 25 and 0 50 (J_{gem} = 5 Hz) due to the cyclopropyl methylene The change in the sign of the specific rotation upon hydrogenation is consonant with the presence of a C-6(7) double bond in a *Buxus* alkaloid [4a,8 9] Comparison (TLC, IR and NMR spectra) of (+)-dihydrocyclobuxupaline-*C* with the known alkaloid (+)-cycloprotobuxine-*C* (VI)[11-13] showed the two materials to be identical, so that structure IV can be assigned to cyclobuxupaline-*C*

[5] STEWART, R R (1958) *Pak J Forestry* **8**, 62
[6] IKRAM, M , MIANA, G A and MAHMUD, F (1968) *Pak J Sci Ind Res* **11**, 253, (1969) *Chem Abstr* **71**, 779b
[7] IKRAM, M , MIANA, G A , SULTANA, F and MAHMUD, F (1968) *Pak J Sci Ind Res* **11**, 488, (1969) *Chem Abstr* **71**, 88432s
[8] KHUONG-HUU-LAINE, F , MAGDALEINE, M J , BISSET, N G and GOUTAREL, R (1966) *Bull Soc Chim Fr* 758
[9] NAKANO, T and TERAO, S (1964) *Tetrahedron Letters* 1035, 1045
[10] DOLEJŠ, L , HANUŠ, V , VOTICKY, Z and TOMKO, J (1965) *Coll Czech Chem Commun* **30**, 2869
[11] CALAME, J P and ARIGONI, D (1964) *Chimia* **18**, 185
[12] KHUONG-HUU, F , PARIS R , RAZAFINDRAMBAO, R , CAVE, A and GOUTAREL, R (1971) *Compt Rend* **273C**, 558
[13] We wish to thank Professor R GOUTAREL, CNRS, Gif-sur-Yvette, for a generous gift of cycloprotobuxine-*C* and desoxy-16-buxidienine

The second new alkaloid, (+)-cyclopapilosine-*D* (VII), $C_{26}H_{46}N_2O$, showed IR absorption bands for O–H and N–H at 3380 (broad) and 3300 (sharp) cm^{-1}, respectively In the NMR spectrum, there were signals for three tertiary *C*-methyl groups as singlets at δ0 98 (3H) and 1·15 (6H), one secondary C_{21}-methyl group at δ1 08 (3H, *d*, *J* 5 Hz), two *N*-methyl groups at δ2 41 and 2 50, an *AB* system due to the methylene protons of a hydroxymethyl function at δ4 12 and 4 72 (2H, *dd*, J_{gem} 10 Hz), and another *AB* system corresponding to the cyclopropyl methylene protons at δ0 30 and 0 72 (2H, *dd*, J_{gem} 4 Hz)



(VI) $R_1 = H$, $R_2 = R_3 = Me$
(VII) $R_1 = R_3 = H$, $R_2 = CH_2OH$
(VIII) $R_1 = R_3 = Ac$, $R_2 = CH_2OAc$

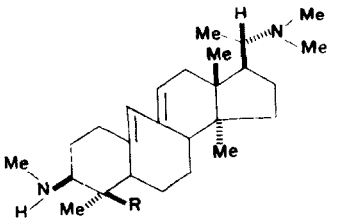
The MS of cyclopapilosine-*D* (VII) showed a molecular ion *m/e* 402 (5), and an *m/e* 58 (100) ion corresponding to the fragment Me–CH=N^+H–Me Acetylation furnished *N,N,O*-triacetylcyclopapilosine-*D* (VIII), $C_{32}H_{52}N_2O_4$, whose IR spectrum contained strong bands at 1630 (amide C=O) and 1730 cm^{-1} (ester C=O), and no N–H absorption The NMR spectrum of VIII showed singlets for three acetyl groups at δ1 97, 2 02 and 2 06 The *AB* system that had originally been at δ4 12 and 4 72 in cyclopapilosine-*D* was now shifted downfield to δ4 58 and 5 00, thus indicating that the hydroxymethyl group in the alkaloid is situated at C_4 [4 9]

Two secondary nitrogen functions are present, one at C_3 and the other at C_{20}, and the cyclopropane methylene must bridge C_9 and C_{10} as found in cyclobuxupaline (IV), and in several other alkaloids of the Buxaceae It follows that cyclopapilosine-*D* must be VII

The third new alkaloid is (+)-buxamine-*C* (IX), $C_{27}H_{46}N_2$, whose UV spectrum, λ_{max}^{EtOH} 228sh, 235, 243 and 252 nm (log ε 3 95, 4 12, 4 17 and 3 96) is characteristic of an *abeo*-9(10 → 19)-diene system [14–17] This spectrum is, in fact, close to that of the well characterized alkaloid desoxy-16-buxidienine (X),[12] λ_{max}^{EtOH} 238, 246 and 254 nm, which we have also found in *B papillosa* [13] Furthermore, the IR spectrum of buxamine-*C* (IX) showed IR bands at 1640, 1605 and 975 cm^{-1}, attributable to a heteroannular diene [18 19]

The NMR spectrum of buxamine-*C* (IX) included signals for four tertiary *C*-methyl groups as singlets at δ0 70, 0 73, 0 75 and 1 02, one secondary C_{21}-methyl group at δ0 85 (3H, *d*, *J* 6 Hz), one dimethylamino group as a singlet at δ2 20, one *N*-methyl group as a singlet at δ2 47, and finally two vinylic protons as two peaks—one at δ5 97 (1H, broad *s*), and the other at δ5 52 (1H, *m*)—a splitting pattern characteristic of a heteroannular diene system [18]

[14] KUPCHAN, S M and ASBUN, W L (1964) *Tetrahedron Letters* 3145
[15] PUCKETT, R T, SIM, G A, ABUSHANAB, E and KUPCHAN, S M (1966) *Tetrahedron Letters* 3815
[16] STAUFFACHER, D (1964) *Helv Chim Acta* **47**, 968
[17] CALAME, J P (1965) Doctoral Dissertation, E T H, Zurich
[18] KUPCHAN, S M, KENNEDY, R M, SCHLEIGH, W R and OHTA, G (1967) *Tetrahedron* **23**, 4574



(IX) R = Me
(X) R = CH_2OH

The positions of the *N*-methyl and *N*-dimethyl groups can be derived from the MS data The base peak *m/e* 72 belongs to the Me–CH=N^+(Me)$_2$ fragment, and an *m/e* 44 peak for Me–N^+H=Me is also observed [19] Buxamine-*C* must, therefore, be represented by IX, a structure closely related to that of the accompanying alkaloid desoxy-16-buxidienine (X) [20]

EXPERIMENTAL[21]

NMR spectra were run at 60 Mc in $CDCl_3$ solution using TMS as internal standard MS were obtained at low and high resolution on an MS-9 instrument M ps are uncorrected All TLC were on Merck F-254

(−)-*Cyclobuxupaline*-C (IV) has m p 111–113° (acetone) and $[\alpha]_D^{25}$ −37° ($CHCl_3$, *c* 1 0) High resolution mass measurement M^+ Calcd for $C_{27}H_{46}N_2$ *m/e* 398 3660 Found *m/e* 398 3653

(+)-*Cyclopapilosine*-D (VII) has m p 233–235° (acetone) and $[\alpha]_D^{25}$ +54° ($CHCl_3$, *c* 0 86) High resolution mass measurement M^+ Calcd for $C_{26}H_{46}N_2O$ *m/e* 402 3610 Found *m/e* 402 3594

(+)-*Buxamine*-C (IX) has m p 153–155° (acetone) and $[\alpha]_D^{25}$ +24° ($CHCl_3$, *c* 1 12) High resolution mass measurement M^+ Calcd for $C_{27}H_{46}N_2$ *m/e* 398 3690 Found *m/e* 398 3689

(+)-*Desoxy*-16-*buxidienine* (X) has m p 183–185° (acetone) and $[\alpha]_D^{25}$ +33° ($CHCl_3$, *c* 1 00) High resolution mass measurement M^+ Calcd for $C_{27}H_{46}N_2O$ *m/e* 414 3609 Found *m/e* 414 3609

N-*Acetylcyclobuxupaline*-C (V) 6 mg cyclobuxupaline-*C*, 0 3 ml Ac_2O and 0 5 ml pyridine were left at room temp for 60 hr *N*-acetylcyclobuxupaline-*C* (7 mg) crystallized from acetone, m p 199–201°, IR (KBr) 1635 cm^{-1} (amide C=O) High resolution mass measurement M^+ Calcd for $C_{29}H_{48}N_2O$ *m/e* 440 3765 Found *m/e* 440 3765

Dihydrocyclobuxupaline-C (VI) Cyclobuxupaline-*C* (25 mg) was stirred for 5 hr at room temp with 15 mg Adams catalyst in 10 ml HOAc The reaction mixture was filtered and neutralized with aq$NaHCO_3$ The product was extracted into CH_2Cl_2 and recovered as crystals (28 mg) m p 129–130°, $[\alpha]_D^{25}$ + 26·7° ($CHCl_3$, *c* 0 98) High resolution mass measurement M^+ Calcd for $C_{27}H_{48}N_2$ *m/e* 400 3815 Found *m/e* 400 3840

N,N,O-*Triacetylcyclopapilosine*-D (VIII) Acetylation of cyclopapilosine-*D* gave 12 mg of VIII, m p 238–240° (CH_2Cl_2) High resolution mass measurement M^+ Calcd for $C_{32}H_{52}N_2O_4$ *m/e* 528 3925 Found *m/e* 528 3899

Acknowledgments —The authors are grateful to the National Institutes of Health for grant CA-11450, and to Professor Robert Goutarel for alkaloid samples

[19] TOMKO, J , BAUEROVA, O , VOTICKY, Z , GOUTAREL, R and LONGEVIALLE, P (1966) *Tetrahedron Letters* 915

[20] The stereochemistry and absolute configuration of the *Buxus* alkaloids have been conclusively determined by Kupchan and Nakano (a) BROWN, JR , K S and KUPCHAN, S M (1964) *J Am Chem Soc* **86**, 4424, 4430, (b) Ref 4b and 15

[21] The isolation procedure for the alkaloids which was carried out by G A M and F S K will be reported in a separate paper