NEW AMARYLLIDACEAE ALKALOIDS FROM *NARCISSUS PAPYRACEUS KER-GAWLER*

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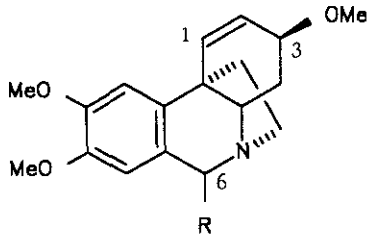
Abstract- Three new alkaloids, O-methylpapyramine (**1**), O-methylmaritidine (**2**) and 9-O-demethylhomolycorine N-oxide (**3**) have been isolated from aerial parts of *Narcissus papyraceus Ker-Gawler*, together with lycorine, papyramine, pseudolycorine, homolycorine and 9-O-demethylhomolycorine.

Narcissus papyraceus Ker-Gawler (Amaryllidaceae) is a widely distributed plant of the Mediterranean and Macaronesic Region.[1] The plant is believed to be toxic for herbivorous mammalians. The toxicity might be related to the alkaloids content which is five times higher in the aerial part than in the bulbs.[2] Earlier work on the constituents of the bulbs describes the isolation of nine alkaloids.[3] In this paper we report the isolation from aerial parts of *N. papyraceus* of three new alkaloids: O-methylpapyramine (**1**), O-methylmaritidine (**2**) and 9-O-demethylhomolycorine N-oxide (**3**), together with homolycorine (**4**) and 9-O-demethylhomolycorine (**5**), papyramine (**6**), lycorine (**7**) and pseudolycorine (**8**). The last three alkaloids were also presents in the bulbs.[3]

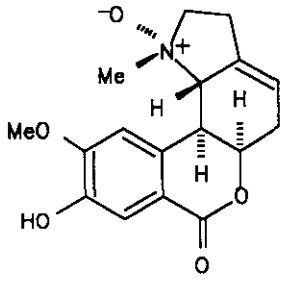
Dried aerial parts of *N. papyraceus* were extracted with methanol. Acid-base extraction afforded the crude basic material that was chromatographed using basic alumina and EtOAc-ethanol to give alkaloids **1-8**.

O-Methylpapyramine (**1**) was eluted from the column with EtOAc and was purified by column chromatography (EtOAc-hexane 4:1). Its ms was consistent with a molecular formula $C_{19}H_{25}NO_4$ (M^+ 331). A base peak at M^+-55 suggested a 5,10b-ethanophenanthridine skeleton without substituent at C-11.[4,5] The ^1H nmr spectrum revealed the presence of two olefinic and two p-oriented aromatic protons, two O-Me aromatic groups and two O-Me aliphatic groups but no signal due to a N-methyl group. The chemical shift of two methine carbon atoms in the ^{13}C nmr spectrum (δ 72.4 and 96.4 ppm) readily located both aliphatic methoxyl groups at C-3 and C-6. The ß configuration at C-3 was deduce from the $J_{2,3}$ = 4.6 Hz and by parcial synthesis from papyramine (**6**), of known configuration. The configuration at C-6 was also ß (trans to the ethane bridge) since 2D-NOESY and nOe diff.

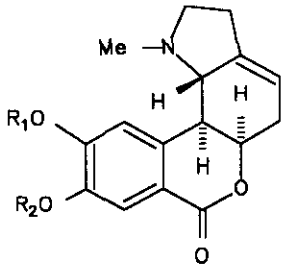
experiments exhibited nOe between H-6 and H-12 (8%). As expected, acidic hydrolysis of **1** afforded papyramine (**6**), however no **1** was formed when **6** was treated with MeOH/HCl, excluding the possibility of being an artifact. Preparation of 6-O-acetylpapyramine was required to achieve the introduction of the O-Me group at C-6, as previously observed for 6-methoxybuphanidrine.[6]



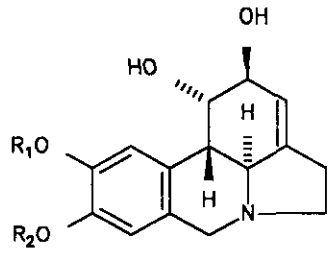
1, R = β-OMe
2, R = H
6, R = α,β-OH



3



4, $R_1 = R_2$ = Me
5, $R_1 = R_2$ = H



7, $R_1 + R_2 = CH_2$
8, R_1 = H R_2 = Me

O-Methylmaritidine (**2**) was eluted from the column with EtOAc-ethanol (9.9:0.1). Ms fragmentation suggested a 5,10b-ethanophenanthridine skeleton. Its ^{1}H nmr was positively compared with the reported data for maritidine,[7,8] although a singlet at δ 3.82 indicated a 3-methoxy substituent as the main difference. The configuration ß at C-3 was set up on the basis of the coupling constants of H-1, H-2 and H-3 ($J_{1,3}$ = 0 and $J_{2,3}$ = 4.8 Hz) and by comparison with those reported for maritidine and epimaritidine.[9] Furthermore, the structure of **2** was confirmed by its partial synthesis from papyramine. Reaction of **6** with thionyl chloride followed by lithium aluminium hydride reduction afforded O-methylmaritidine.

The third new compound in *N. papyraceus* was 9-O-demethylhomolycorine N-oxide (**3**) which was eluted with EtOAc-ethanol (10:2). A base peak at m/z 109 suggested a lycorenine-type alkaloid.[10] Characteristic of the ^1H nmr was the presence of one O-Me group and a low field N-Me group in agreement with the N-oxide functionality. The 2D-COSY spectrum located the O-Me group at C-10, as in 9-O-demethylhomolycorine (**5**). Treatment of **5** with 36% H_2O_2 gave a single N-oxide which was shown to be identical to **3**. The α configuration of the N-oxide group was deduce from the 2D-NOESY spectrum since nOe was observed between the H-11 and both N-Me and O-Me (nOe diff. experiments gave a nOe beetwen H-11/N-Me and H-11/O-Me of 2% and 4% respectively). The isolation of **3** confirms the presence of N-oxides among the Amaryllidaceae alkaloids.[11]

Papyramine (**6**) was the main alkaloid found in this plant. It exist in solution as an equilibrium mixture of epimers at C-6, which was showed to be solvent dependent. The α/ß ratio was: 3/2 in $CDCl_3$, 1/1 in DMSO and 2/1 in $CDCl_3$-TFA. ^1H and ^{13}C nmr data for each epimer are given in the experimental part.

Lycorine, homolycorine and 9-O-demethylhomolycorine were fully identified by comparison with authentic samples.

EXPERIMENTAL

Melting points were determined in a Gallenkamp apparatus and are uncorrected. Optical rotations were measured with a Pekin-Elmer Model 241 polarimeter. The uv espectra were recorded in a Hewlett-Packard 8452A spectrophotometer, ir spectra with a Beckman Model Aculab IV spectrophotometer, nmr spectra with a Bruker WP-200 SY instrument in the solvent specified with Me_4Si as internal standard, operating at 200 MHz for ^1H and 50 MHz for ^{13}C. Mass spectra were recorded on a HP GC/MS 5988A and high resolution mass spectra on a Kratos MS-50 instrument. The absorbent used for column chromatography was basic alumina 60 (Merck 1067) and silica gel 60 (Merck 7734).

Isolation of alkaloids from Narcissus papyraceus Ker-Gawler.- Aerial parts of *N. papyraceus*, collected in Campanillas (Málaga), were dried to 2.75 kg, powdered and extracted in a soxhlet apparatus with MeOH (15 l, 9 days). The extracts were concentrated and the resulting syrup was dissolved in 3% HCl (3 l) and washed with hexane. The aqueous layer was brought up to pH 8 with concentrated NH_4OH and extracted with EtOAc. This extract (40 g) was finally chromatographed on basic alumina using EtOAc step gradient (0-100%) with ethanol to give compounds **1-8** in the following order of elution:

O-methylpapyramine (**1**) (10:0), homolycorine (**6**) (10:0), O-methylmaritidine (**2**) (9.9:0.1), lycorine (**4**) (9.5:0.5), 9-O-demethylhomolycorine (**7**) (9:1), papyramine (**8**) (8.5:1.5), 9-O-demethylhomolycorine N-oxide (**3**) (8:2) and pseudolycorine (**5**) (7.5:2.5).

(-)-O-Methylpapyramine (1)- (Yield 0.3 g). Amorphous. $[\alpha]_D^{24}$ - 31 ° (c 0.1, MeOH). Uv (MeOH) λ_{max} nm (log ϵ): 206 (4.72); 230(h) (3.98); 286 (3.67). Ir ($CHCl_3$) ν_{max} (cm^{-1}): 1610, 1510, 1470, 1410, 1330, 1272, 1250, 1225, 1120. ^1H Nmr ($CDCl_3$) δ (ppm): 6.73 (s, H-10); 6.71 (s, H-7); 6.59 (d, H-1, $J_{1,2}$ = 10 Hz); 5.90 (dd, H-2, $J_{2,1}$ = 10, $J_{2,3}$ = 4.6 Hz); 4.43 (s, H-6); 3.79 (s, OMe arom.); 3.77 (s, OMe arom.); 3.75 (m, H-3); 3.52 (s, OMe); 3.47 (dd, H-4a, $J_{4a,4eq}$ = 4.37, $J_{4a,4ax}$ = 13.5 Hz); 3.26 (s, OMe); 3.28 (ddd, H-12, $J_{12,11'}$= 4.8, $J_{12,11}$ = 9.7, J_{gem} = 13.5 Hz); 2.65 (ddd, H-12', $J_{12',11}$ = 5.7, $J_{12',11'}$= 8.3, J_{gem} = 13.5 Hz); 1.85 (m, H-4eq, 2H-11); 1.52 (ddd, H-4ax, $J_{4ax,3}$ = 4.1, $J_{4ax,4a}$ = 13.5, J_{gem} = 13.5). ^{13}C Nmr ($CDCl_3$) δ (ppm): 148.2 (C-9); 147.3 (C-8); 138.1(C-10a); 132.4 (C-1); 125.5 (C-6a); 125.3 (C-2); 112.4 (C-7); 105.0 (C-10); 96.4 (C-6); 72.4 (C-3); 57.1 (C-4a); 56.8 (OMe); 56.0 (OMe); 55.8 (OMe); 55.7 (OMe); 48.4 (C-12); 43.7 (C-10b); 41.1 (C-11); 28.5 (C-4). Eims m/z (%): 331 (M^+, 50); 300 (40); 276 (100); 245 (60); 214 (20); 201 (10). Hrms: Calcd for $C_{19}H_{25}NO_4$: 331.1784. Found: 331.1794 .

(+)-O-Methylmaritidine (2)- (Yield 0.05 g). mp 240-242 °C (crystallized as HCl derivative from MeOH-EtOAc); $[\alpha]_D^{23}$ + 16 ° (c 0.1, MeOH). Uv (MeOH) λ_{max} nm (log ϵ): 236 (h) (4.23); 286 (4.30). Ir (KBr) ν_{max} cm^{-1}: 1610, 1600, 1580, 1475, 1440, 1425, 1060. ^1H Nmr ($CDCl_3$) δ (ppm): 6.81 (s, H-10), 6.57 (s, H-7), 6.49 (d, H-1, $J_{1,2}$ = 10.1 Hz), 6.08 (dd, H-2, $J_{2,1}$ = 10.1, $J_{2,3}$ =4.8 Hz), 4.80 (d, H-6, J_{gem} = 15.8 Hz), 4.25 (d, H-6', J_{gem} = 15.8 Hz), 4.08 (ddd, H-12', $J_{12',11}$ = 4.3, $J_{12',11'}$= 12.0, J_{gem} = 12.6 Hz), 3.9 (dd, H-4a, $J_{4a,4ax}$ = 13.5, $J_{4a,4eq}$ = 3.8 Hz), 3.85 (m, H-3), 3.86 (s, OMe), 3.82 (s, OMe), 3.34 (s, OMe), 3.27 (ddd, H-12, $J_{12,11'}$= 7.1, $J_{12,11}$ = 8.8, J_{gem} = 12.6 Hz), 2.95 (ddd, H-4eq, J_{gem} = 13.6, $J_{4eq,4a}$ = 3.8, $J_{4eq,3}$ = 2 Hz), 2.37 (ddd, H-11, $J_{11,12'}$= 4.3, $J_{11,12}$ = 8.8, J_{gem} = 12.5 Hz), 2.22 (ddd, H-11', $J_{11',12}$ = 7.1, $J_{11',12'}$= 12.0, J_{gem} = 12.5 Hz), 1.73 (ddd, H-4ax, J_{gem} = 13.5, $J_{4ax,4a}$ = 13.5, $J_{4ax,3}$ = 3.9 Hz). ^{13}C Nmr, ($CDCl_3$) δ (ppm): 149.0 (C-9), 148.9 (C-8), 138.0 (C-10a), 128.2 (C-1), 127.3 (C-2), 116.5 (C-6a), 110.0 (C-7), 106.2 (C-10), 70.7 (C-3), 64.8 (C-4a), 57.1 (C-6), 56.1 (3xOMe), 52.3 (C-12), 44.9 (C-10b), 40.6 (C-11), 25.6 (C-4). Eims m/z (%) : 301 (M^+, 42), 286 (24), 270 (22), 231 (100), 203 (26.6). Hrms: Calcd for $C_{18}H_{23}NO_3$: 301.1671. Found: 301.1663 .

(+)-9-0-Demethylhomolycorine N-oxide (3)- (Yield 0.3 g). mp 153-154 °C (from ethanol); $[\alpha]_D^{23}$ + 19 ° (c 0.1, MeOH). Uv (MeOH) λ_{max} nm (log ϵ): 230 (4.43), 270 (4.28), 304 (3.9); (MeOH-NaOH) : 208 (5.34), 248 (4.57), 274 (h) (4.30), 340 (3.82). Ir (KBr) ν_{max} cm^{-1} : 3180, 1700, 1580, 1480, 1430, 1320, 1300, 1250, 1220, 1200, 1050. ^1H Nmr ($CDCl_3$-CD_3OD) δ (ppm) : 7.44 (s, H-8), 7.24 (s, H-11), 5.76 (br d, H-4, J = 2.2 Hz), 4.83 (m, H-5a), 3.96 (s, OMe), 3.87 (br d, H-11c, $J_{11c,11b}$ = 9.9 Hz), 3.57 (m, 2 H-2), 3.5 (dd, H-11b, $J_{11b,11c}$ = 9.9, $J_{11b,5a}$ = 2.0 Hz), 2.94 (s, N-Me), 2.88 (m, H-3), 2.72 (m, H-3'), 2.64 (m, 2 H-5). ^{13}C Nmr ($CDCl_3$-CD_3OD) δ (ppm) : 167.1 (C-7), 153.4 (C-10), 147.9 (C-9), 135.6 (C-3a), 134.9 (C-11a), 121.0 (C-4), 117.6 (C-7a), 117.1 (C-8), 112.6 (C-11), 79.3 (C-11c), 78.8 (C-5a), 70.7 (C-2), 56.8 (OMe), 56.2 (N-Me), 37.4 (C-11b), 31.4 (C-5), 26.2 (C-3). Eims m/z (%): 317 (M^+ 10), 281 (10), 240 (10), 165 (5), 125 (20), 109 (100), 108 (20).

(-)-Lycorine (7)- (Yield 1.3 g). mp 255 °C (from methanol); $[\alpha]_D^{23}$ - 68 ° (c 0.24, EtOH); lit:[12] mp 257-258 °C; $[\alpha]_D$ - 78.5 ° (EtOH).

(-)-Pseudolycorine (8)- (Yield 0.075 g). mp 236-237 °C (from ethanol); $[\alpha]_D^{22}$ - 60 ° (c 0.04, MeOH); lit:[13] mp 237-240 °C.

(+)-Homolycorine (4)- (Yield 0.14 g). mp 166-167 °C (ethyl ether); $[\alpha]_D^{23}$ + 86 ° (c 0.1, MeOH); lit:[14] mp 175 °C (H_2O); $[\alpha]_D$ + 85 ° (EtOH).

(+)-9-0-Demethylhomolycorine (5)- (Yield 3.5 g). mp 213-214 °C (from EtOAc); $[\alpha]_D^{24}$ + 111 ° (c 0.1, MeOH); lit:[15] mp 213-214 °C.

Papyramine (6)- (Yield 4.5 g). mp 112 °C (EtOAc - ether). $[\alpha]_D^{22}$ 0 ° (c 0.25, MeOH). Uv (MeOH) λ_{max} nm (log ϵ): 206 (4.61), 238 (3.77), 282 (3.82). Ir (KBr) ν_{max} cm^{-1}: 3040, 3000, 1580, 1490, 1440, 1410, 1380, 1070. Eims m/z (%): 317 (M^+,60), 286 (20), 268 (10), 262 (100), 247 (40), 219 (10), 203 (20).

α-Epimer:

^1H Nmr ($CDCl_3$) δ (ppm): 6.99 (s, H-7), 6.76 (s, H-10), 6.62 (d, H-1, $J_{1,2}$ = 10.0 Hz), 5.97 (dd, H-2, $J_{2,1}$ = 10.0, $J_{2,3}$ = 4.9 Hz), 5.86 (s, H-6), 3.86 (s, 2 OMe), 3.85 (m, H-3), 3.7 (ddd, H-12, $J_{12,11'}$ = 4.6, $J_{12,11}$ = 8.2, J_{gem} = 13.9 Hz), 3.58 (dd, H-4a, $J_{4a,4eq}$ = 4.1, $J_{4a,4ax}$ = 13.5 Hz), 3.35 (s, OMe), 3.00 (ddd, H-12', $J_{12',11}$ = 3.9, $J_{12',11'}$ = 9.6, J_{gem} = 13.9 Hz), 2.20 (br d, H-4eq, J_{gem} = 13.5 Hz), 1.96 (m, 2H-11), 1.74 (ddd, H-4ax, $J_{4ax,3}$ = 4.1, $J_{4ax,4a}$ = 13.5, J_{gem} = 13.5 Hz). ^{13}C Nmr ($CDCl_3$) δ (ppm): 148.0 (C-9), 147.4 (C-8), 136.1 (C-10a), 132.2 (C-1), 127.0 (C-6a), 125.3 (C-2), 110.6 (C-7), 105.0 (C-10), 86.7 (C-6), 72.1 (C-3), 61.9 (C-4a), 55.8 (OMe), 55.6 (OMe), 44.4 (C-10b), 42.1 (C-12), 41.7 (C-11), 28.4 (C-4).

ß-Epimer:

^1H-Nmr ($CDCl_3$) δ ppm: 6.85 (s, H-7), 6.8 (s, H-10), 6.64 (d, H-1, $J_{1,2}$ = 10 Hz), 5.97 (dd, H-2, $J_{2,1}$ = 10, $J_{2,3}$ = 4.9 Hz), 5.14 (s, H-6), 3.85 (m, H-4a), 3.86 (s, 2 OMe), 3.82 (m, H-3), 3.36 (m, H-12), 3.31 (s, OMe), 2.82 (ddd, H-12', $J_{12',11}$ = 6.5, $J_{12',11'}$ = 8.4, J_{gem} = 13.8 Hz), 2.05 (m, H-4eq), 1.96 (m, 2H-11), 1.6 (ddd, H-4ax, $J_{4ax,3}$ = 4.1, $J_{4ax,4a}$ = 13.8, J_{gem} = 13.8 Hz). ^{13}C Nmr ($CDCl_3$) δ (ppm): 148.3 (C-9), 147.4 (C-8), 137.1 (C-10a), 131.8 (C-1), 125.6 (C-2), 112.0 (C-7), 105.0 (C-10), 88.6 (C-6), 72.1 (C-3), 56.4 (C-4a), 55.8 (OMe), 55.6 (OMe), 47.5 (C-12), 43.8 (C-10b), 40.6 (C-11), 27.8 (C-4).

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REFERENCES

1. B. Valdés, in **"Flora Vascular de Andalucia Occidental"**, Vol. **3**, ed. by B. Valdéz, S. Talavera, and E. Sanchez-Galiano, Ketres Editora, S. A., Barcelona, 1987, p. 468.
2. R. Suau, A. I. Garcia, and R. Rico, **Acta Botánica Malacitana**, 1988, **13**, 189.
3. H. Shanhai, M. Guangen, and S. Guoquiang, **Huaxue Xuebao,** 1981, **39,** 529 (**Chem. Abstr.**, 1982, **96,** 139653r)
4. P. Longevialle, D. H. Smith, A. L. Burlingame, H. M. Fales, and R. G. Highet, **Org. Mass. Spectroscopy,** 1973, **7,** 401.
5. P. Longevialle, H. M. Fales, R. G. Highet, and A. L. Burlingame, **Org. Mass. Spectroscopy,** 1973, **7,** 417.
6. M. R. Slabaugh and W. C. Wildman, **J. Org. Chem.,** 1971, **36,** 3202.
7. W. C. Wildman, **"The Alkaloids"**, Vol. **11,** ed. by R. H. F. Manske and H. L. Holmes, Academic Press, New York, Ch. 10.
8. S. Ghosal, A. Razdan, and S. Razdan, **Phytochemistry,** 1985, **24,** 635.
9. M.A. Schwartz and R.A. Holton, **J. Am. Chem. Soc.,** 1970, **92,** 1090.
10. T. Ibuka, H. Irie, S. Uyeo, K. Kotera, and Y. Nakagawa, **Tetrahedron Lett.,** 1966, 4745.
11. R. Suau, A. I. Gómez, R. Rico, M. P. Vázquez, L. Castedo, and R. Riguera, **Phytochemistry,** 1988, **27,** 3285.
12. W. C. Wildman, **"The Alkaloids"**, Vol. **6,** ed. by R. H. F. Manske, Academic Press, New York, 1960, Ch. 9.
13. J. M. Llabrés, F. Viladomat, J. Bastida, C. Codina, M. Serrano, M. Rubiralta, and M. Feliz, **Phytochemistry**, 1986, **25**, 1453.
14. T. Kametani, **" The Chemistry of the Isoquinoline Alkaloids. 1."**, Elsevier, Publ. Company, New York, 1969.
15. S. Uyeo and N. Yanaihara, **J. Chem. Soc.,** 1959, 172.

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