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ALKALOIDAL CONSTITUENTS OF *FUMARIA INDICA*

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Key Word Index—*Fumaria indica*; Fumariaceae; isoquinoline; paprafumine; paprarine; papraline; *seco*phthalideisoquinoline.

Abstract—Three new alkaloids, paprafumine, paprarine and papraline, were isolated from the aerial parts of *Fumaria indica*. Their structures were determined by spectral studies. Three more alkaloids isolated for the first time from this species were identified as cryptopine, raddeanine and oxocoptisine.

INTRODUCTION

We have previously reported the isolation of five new alkaloids from the aerial parts of *Fumaria indica* [1–4]. During our continuing phytochemical investigations, three new alkaloids, paprafumine (**1**), paprarine (**2**) and papraline (**3**), have been isolated, along with three other known alkaloids identified for the first time from this species

RESULTS AND DISCUSSION

Fumaria indica is bitter, slightly acidic and astringent. The plant extract is regarded as a laxative, diuretic and alterative, and is said to be beneficial in dyspepsia and scrofulous skin infection. Seeds of the plant are also used as a mild analgesic.

Ethanolic extracts of the aerial parts of *F. indica* were evaporated to a gum which was then partitioned between chloroform and water at pH ~1.5 and pH ~9. Compounds **1** and **3** were isolated from the chloroform extracts obtained by extraction at pH ~9 while compounds **2**, **4**, **5** and **6** were isolated by extraction at pH ~1.5.

Paprafumine (**1**), $C_{22}H_{23}NO_8$, exhibited a ^{1}H NMR spectrum which closely resembled that of narceimicine [5], except that it showed an additional three-proton singlet at δ3.10, due to the methyl protons of the ester group substituted at C-8. The presence of this methoxy carbon was confirmed by a signal at δ57.6 in the ^{13}C NMR spectrum. The UV absorption maxima at 331 and 209 nm were characteristic of *seco*phthalide isoquinoline alkaloids [6], while the IR bands at 4235 and 3645 cm^{-1} were due to hydrogen-bonded OH (phenolic) stretching vibrations. Another IR absorption appeared at 1708 cm^{-1} due to the carbonyl group. The presence of C=C was indicated by an intense absorption at 1482 cm^{-1}. The mass spectrum of **1** showed no $[M]^+$, but the $[M-18]^+$ appeared at *m/z* 411.1326 corresponding to the formula, $C_{22}H_{21}NO_7$ (calc. 411.1318), due to loss of water. The $[M]^+$ was determined by positive-ion FAB-mass spectrometry [7]. The peak at *m/z* 368.1163 resulted from the loss of a carbomethoxy group, that at *m/z* 236.0330 by cleavage of the C-14a/C-14 bond. The remaining half of the molecule also appeared at *m/z* 192.1042. A fragment at *m/z* 158.0942 was due to the loss of the *N*-dimethyl-bearing-side-chain, which is a characteristic feature of *seco*phthalideisoquinoline alkaloids [8].

The ^{1}H NMR spectrum ($CDCl_3$–CD_3OD, 49:1, 400 MHz) of **1** showed two downfield sharp singlets at δ7.71 and 6.70 due to the C-1 and C-4 aromatic protons. The protons of two methylenedioxy groups resonated as singlets at δ5.97 and 6.35 indicating the absence of any chiral centre. Two two-proton multiplets at δ2.35 and 2.98 were due to the C-5 and C-6 methylenes. Two downfield doublets at δ6.88 and 7.25 (J = 7.9 Hz) were assigned to the C-11 and C-12 aromatic protons, respectively. A three-proton singlet at δ3.10 was assigned to the ester methoxy protons. The somewhat upfield chemical shift of the methoxy signal was attributed to the shielding effect of the aromatic ring of the isoquinoline moiety.

The ^{13}C NMR spectrum (Table 1) of **1** showed all 22 carbon resonances. Multiplicity assignments were made using DEPT pulse sequences [9]. The ^{13}C NMR data are comparable with that of bicucullinine [10]. These observations led to structure **1** for this new *seco*phthalideisoquinoline base.

The second new alkaloid, paprarine (**2**), showed strong UV absorption at 293 and 214 nm, characteristic of a *seco*phthalideisoquinoline skeleton [6, 11]. The IR spectrum exhibited a weak absorption at 3285 cm^{-1}, which indicated the presence of a hydroxy function. The lactone function was inferred from the presence of an absorption

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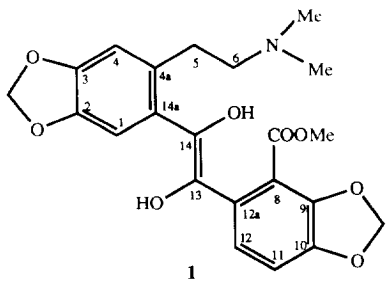

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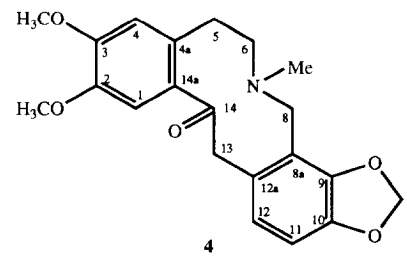

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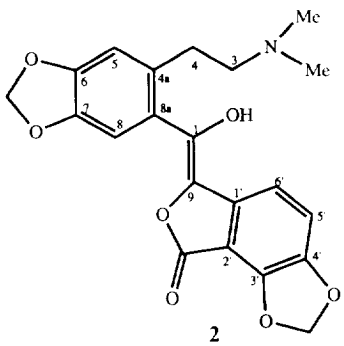

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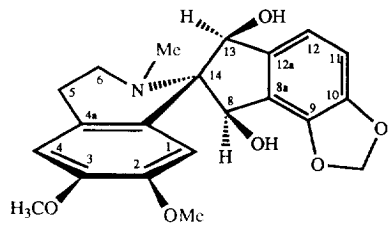

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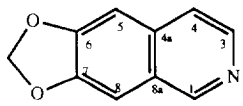

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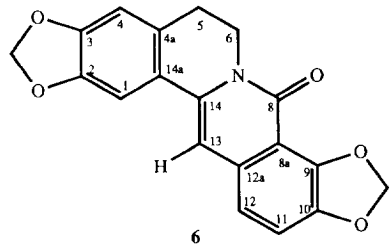

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band at 1714 cm $^{-1}$. Other IR absorptions were at 1592 (C=C) and 1125 (C–O–C) cm $^{-1}$. The mass spectrum of **2** displayed the $[M^+]$ at *m/z* 397.1138, corresponding to the molecular formula $C_{21}H_{19}NO_7$, indicating twelve degrees of unsaturation [11]. The loss of a methyl radical $[CH_3]$ yielded an ion at *m/z* 382.0927 ($C_{20}H_{16}NO_7$, calc., 382.0911). The peak at *m/z* 336.0629 ($C_{19}H_{12}O_6$) was due to the loss of $[H_2O + C_2H_5N]$, that at *m/z* 322 due to the loss of $[CH_2{=}N^+Me_2 + H_2O]$, while the base peak at *m/z* 58 was due to $[CH_2{=}N^+Me_2]$.

The ^{1}H NMR spectrum of **2** ($CDCl_3$–CD_3OD, 49:1) was particularly informative, showing two singlets at δ6.64 and 6.63, assigned to the C-8 and C-5 aromatic protons. The C-3 and C-4 methylene protons of **2** appeared as multiplets at δ3.12 and 2.88, respectively. Two doublets at δ6.79 and 6.96 (J = 7.9 Hz) were due to the C-5′ and C-6′ protons, respectively, while the two methylenedioxy protons appeared as singlets at δ5.89 and 6.04. The ^{13}C NMR spectrum was very weak due to paucity of sample and some carbons, such as C-9, C-1′, C-4′ and C=O, could not be detected. The observed ^{13}C NMR signals were closely comparable to those of the reported *N*-methyl hydrastine [4] and adlumidiceine enol lactone [13]. On the basis of the above-mentioned spectroscopic evidence, structure **2** is assigned to this new isoquinoline base.

Papraline (**3**), $C_{10}H_7NO_2$ ($[M]^+$ *m/z* 171.0512) was obtained from the chloroform extracts obtained on extraction of the aqueous solutions at pH 9. It was identified as the simple isoquinoline alkaloid, papraline, by comparison of its ^{1}H NMR, ^{13}C NMR and mass spectroscopic data with those reported for nigellimine *N*-oxide [14]. The UV spectrum was characteristic of an isoquinoline chromophore, whereas the IR spectrum indicated the presence of a conjugated C=N (2738 cm $^{-1}$) group and an aromatic moiety (1518–1470 cm $^{-1}$). The ^{1}H NMR spectrum showed signals for five aromatic protons. The singlet at δ9.16 was due to the C-1 proton, while two doublets integrating for one proton each at δ8.20 ($J_{3,4}$ = 5.7 Hz) and 7.91 ($J_{4,3}$ = 5.7 Hz) were due to the C-3 and C-4 protons, respectively. The downfield chemical shifts for these protons (C-1 and C-3) are due to the deshielding effect of the iminic nitrogen. The other two singlets at δ7.31 and 7.45 were due to C-5 and C-8 aromatic protons. The methylenedioxy protons appeared at δ6.31 as a two proton singlet.

The ^{13}C NMR spectrum (Table 1) of **3** showed eight carbon signals instead of ten, with two overlapping signals for C-1/C-3 and C-5/C-8 carbons, which appeared at δ123.2 and 103.3, respectively. The aromatic C-4 appeared at δ104.7. A DEPT experiment also showed only one methylenedioxy carbon at δ103.5. The C-4a and

Table 1. ^{13}C NMR spectral data of **1**, **2**, and **3**

C	1	2	3
1	108.3	196.3	23.2
2	145.9	—	—
3	150.7	62.6	123.2
4	110.8	35.2	103.3
4a	134.7	125.8	135.1
5	28.0	106.6	104.7
6	56.2	151.2	142.3
7	—	51.1	146.0
8	125.0	106.1	103.3
8a	—	135.6	136.3
9	151.6	137.6	—
10	146.0	117.2	—
11	124.8	114.7	—
12	112.0	152.4	—
12a	127.2	—	—
13	189.5	119.6	—
14	192.5	—	—
14a	132.7	—	—
NMe	—	—	—
$N(Me)_2$	41.7	55.0	—
OMe	57.6	—	—
$O-CH_2-O$	101.9	101.2	103.5
$O-CH_2-O$	102.0	103.6	—
C=O	167.5	168.5	—

C-8a quaternary carbons appeared at δ136.1 and 136.3, while the oxygenated C-6 and C-7 quaternary carbons resonated at δ142.3 and 146.0.

The known bases, cryptopine (**4**) [15], raddeanine (**5**) [16] and oxocoptisine (**6**) [17], were also isolated and identified by comparison of their spectral data (UV, IR, ^1H NMR, mass) with those reported in the literature. These alkaloids have not been previously reported from this species.

EXPERIMENTAL

^1H NMR recorded at 300, 400 and 500 MHz and ^{13}C NMR at 100 MHz on a 400 MHz instrument. TLC was performed on silica gel pre-coated plates (GF-254, 0.2 mm).

Mature, whole plants of *F. indica* were collected (20 kg dry wt) in March 1988, from wheat fields in the suburbs of Karor, 145 km from Multan city (Punjab, Pakistan) and identified by the plant taxonomist at the Department of Botany, University of Karachi. EtOH extracts of the plant were concd, acidified with 5% HCl and filtered. The soln (pH 1.5) was extracted with $CHCl_3$, then dried (Na_2SO_4) and evapd under red. pres. The resulting gummy material (26 g) 'Fr. A' was loaded onto a silica gel (70–230 mesh ASTM) column and eluted with $CHCl_3$–MeOH mixts of increasing polarity. The resulting frs on repurification, yielded paprarine (**2**), cryptopine (**4**), raddeanine (**5**) and oxycoptisine (**6**).

The aq. acidic extract was basified with NH_4OH (pH ~ 9) and extracted with $CHCl_3$. The $CHCl_3$ extracts were evapd under red. pres. The resulting dried material (38 g) 'Fr. B' was loaded on to a silica gel column and eluted with $CHCl_3$–MeOH mixts of increasing polarity. The frs thus obtained were further chromatographed to afford **1** and **3**.

Paprafumine (**1**). The fr. obtained by CC of 'Fr. B' eluted with $CHCl_3$–MeOH (21:4) when subjected to repeated TLC on silica gel using $CHCl_3$–MeOH–NH_4OH (82:17:1) afforded pure paprafumine (**1**) as an amorphous solid, (28 mg) (1.4×10^{-4}% yield). UV λ_{max}^{MeOH} nm (log ε): 331 (4.05), 209 (4.48); λ_{min} nm (log ε): 280 (3.87). IR ν_{max}^{KBr} cm^{-1}: 4235, 3645 (OH), 2898 (C–H), 1708 (C=O), 1482 (C=C). ^1H NMR ($CDCl_3$–CD_3OD, 49:1, 400 MHz): δ7.71 (1H, *s*, H-1), 7.25 (1H, *d*, $J_{12,11}$ = 7.9 Hz, H-12), 6.88 (1H, *d*, $J_{11,12}$ = 8.0 Hz, H-11), 6.70 (1H, *s*, H-4), 6.35 (2H, *s*, $O-CH_2-O$), 5.97 (2H, *s*, $O-CH_2-O$), 3.10 (3H, *s*, ester-OCH_3), 2.98 (2H, *m*, H-6), 2.60 (6H, *s*, $N(CH_3)_2$), 2.35 (2H, *m*, H-5). FAB-MS: +ve *m/z* 430 $[M+H]^+$. EIMS *m/z* (rel. int.): 411 $[M-18]^+$ (38), 368 (22), 236 (26), 219 (18), 192 (78), 167 (42), 158 (12), 149 (100), 58 (64). ^{13}C NMR: see Table 1.

Paprarine (**2**). A fr. obtained on CC of 'Fr. A' eluted with $CHCl_3$–MeOH (83:17) was subjected to prep. TLC on silica gel with $CHCl_3$–MeOH (4:1) to afford a new *seco*phthalideisquinoline enol lactone, paprarine (**2**), as an amorphous solid, (22 mg) (11×10^{-5}% yield). $[\alpha]_D^{20} = 0°$ (MeOH). UV λ_{max}^{MeOH} nm (log ε): 293 (0.78), 214 (1.56); λ_{min} nm (log ε): 275 (0.41). IR ν_{max}^{KBr} cm^{-1}: 3285 (OH), 1714 (C=O), 1592 (C=C) and 1125 (C–O–C). ^1H NMR ($CDCl_3$–CD_3OD, 49:1, 400 MHz): δ6.96 (1H, *d*, $J_{3',2'}$ = 7.9 Hz, H-3'), 6.79 (1H, *d*, $J_{2',3'}$ = 7.9 Hz, H-2'), 6.64 (1H, *s*, H-8), 6.63 (1H, *s*, H-5), 6.04 (2H, *s*, $O-CH_2-O$), 5.89, (2H, *s*, $O-CH_2-O$), 3.12 (2H, *m*, H-3), 2.88 (2H, *m*, H-4), 2.81 (6H, *s*, NMe_2). MS *m/z* (rel. int.): 397 (16), 382 (6), 336 (20), 204 (28), 193 (8), 190 (12), 149 (32), 58 (100). ^{13}C NMR: see Table 1.

Papraline (**3**). The fr. obtained by CC of 'Fr. B' eluted with $CHCl_3$–MeOH (19:1) was purified by prep. TLC on silica gel using petrol (40–60°)–Me_2CO (41:9). This afforded an amorphous solid (23 mg, yield 11.5 $+10^{-5}$% yield). UV λ_{max}^{MeOH} nm (log ε): 325 (2.43), 312 (2.37), 233 (3.47), 195 (2.85): λ_{min} nm (log ε): 316 (2.33), 300 (2.22), 200 (2.43). IR ν^{CHCl_3} cm^{-1}: 2915, 2840 (C–H), 2738, (C=N), 1518–1470 (C=C). ^1H NMR ($CDCl_3$, 500 MHz): δ9.16 (1H, *s*, H-1), 8.20 (1H, *d*, *J* = 5.7 Hz, *s*, H-3), 7.91 (1H, *d*, *J* = 5.7 Hz, *s*, H-4), 7.45 (1H, *s*, H-8), 7.31 (1H, *s*, H-5), 6.31 (2H, *s*, $O-CH_2-O$). MS *m/z* (rel. int.): 173 (100), 143 (6), 115 (32), 88 (28), 62 (38).

Cryptopine (**4**). The column loaded with 'Fr. A' on elution with $CHCl_3$–MeOH (97:3) afforded two major alkaloids, cryptopine (**4**) along with protopine. Further purification was performed by TLC using petrol (40–60°)–Me_2CO (7:3). The crystalline material obtained (21 mg, 10.5×10^{-5} % yield) had mp 156–158°. UV λ_{max}^{MeOH} nm (log ε): 286 (2.80), 206 (3.75), 193 (3.18). λ_{min} nm (log ε): 260 (2.58), 197 (3.77). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 2898 (C–H), 1718 (C=O), 1585 (C=C). ^1H NMR ($CDCl_3$, 400 MHz): δ7.02 (1H, *s*, H-1), 6.71 (1H, *s*, H-4), 6.71 (2H, *m*, H-11, H-12), 5.95 (2H, *s*, $O-CH_2-O$), 3.92 (3H, *s*, OMe), 3.92 (3H, *s*, OCMe), 3.75 (2H, *m*, H-13), 3.09 (2H, *m*, H-8), 2.83 (2H, *m*, H-6), 2.21 (3H, *s*, N-Me), 2.11 (2H, *m*, H-5). MS *m/z* (rel. int.): 369 $[M]^+$ (6), 283 (4), 267 (5), 221 (2), 190 (9), 179 (18), 148 (100).

(±)-*Raddeanine* (**5**). The fr. obtained by CC of 'Fr. A' eluted with $CHCl_3$–MeOH (98.5:1.5) afforded three alkaloids. This fr. was further purified by alumina TLC using petrol (40–60°)–Me_2CO (9:1). The faster moving alkaloid was raddeanine (**5**) obtained as an amorphous solid (8 mg 4×10^{-5} % yield). $[\alpha]_D^{25} = 0°$. UV λ_{max}^{MeOH} nm (log ε): 352 (3.38), 294 (3.69), 261 (3.98), 234 (4.36), 202 (4.60); λ_{min} nm (log ε): 315 (3.28), 279 (3.53), 255 (3.96), 219 (4.24). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3684 (OH), 1595 (C=C). ^1H NMR ($CDCl_3$, 300 MHz): δ 7.54 (1H, *d*, $J_{12,11} = 8.1$ Hz, H-12), 7.06 (1H, *d*, $J_{11,12} = 8.1$ Hz, H-11), 6.72 (1H, *s*, H-1), 6.65 (1H, *s*, H-4), 6.15 (2H, *s*, O–CH_2–O), 6.12 (1H, *s*, H-8), 5.86 (1H, *dd*, $J = 4.4$ and 1.2 Hz, H-13), 3.78 (3H, *s*, OMe), 3.73 (3H, *s*, OMe), 3.52 (2H, *m*, H-6), 2.95 (2H, *m*, H-5), 2.73 (3H, *s*, N-Me). MS *m/z* (rel. int.): 385 (22), 370 (16), 352 (6), 338 (7), 325 (10), 324 (5), 206 (10), 149 (42), 71 (64), 57 (100).

8-*Oxocoptisine* (**6**). The fr. obtained by CC of 'Fr. A' eluted with $CHCl_3$–MeOH (23:2) afforded two minor alkaloids. These were purified by repeated TLC in $CHCl_3$–MeOH–NH_4OH (95:4.5:0.5), which afforded an amorphous solid (8 mg, 4×10^{-5} % yield). $[\alpha]_D^{25} = 0°$ (MeOH). UV λ_{max}^{MeOH} nm (log ε): 296 (2.08), 203 (3.91), 194 (3.46); λ_{min} nm (log ε): 273 (3.04). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 2878 (C–H), 1725 (C=O), 1618 (C=C). ^1H NMR ($CDCl_3$, 400 MHz): δ 7.22 (1H, *s*, H-1), 7.16 (1H, *d*, $J_{12,11} = 8.3$ Hz, H-12), 7.03 (1H, *d*, $J_{11,12} = 8.3$ Hz, H-11), 6.73 (1H, *s*, H-4), 6.69 (1H, *s*, H-13), 6.21 (2H, *s*, O–CH_2–O), 5.99 (2H, *s*, O–CH_2–O), 4.28 (2H, *t*, $J_{6,5} = 4.8$ Hz, H-6), 2.89 (2H, *t*, $J_{5,6} = 4.8$ Hz, H-5). MS *m/z* (rel. int.): 335 $[M]^+$ (82), 320 (98), 292 (22), 163 (35), 160 (38), 85 (60), 69 (100).

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