ION TRAP TANDEM MASS SPECTROMETRY OF *C*- AND *N*-METHYL, BENZYL, AND PRENYL SUBSTITUTED 2-OXOPYRROLIDINOINDOLINES

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This paper is dedicated to Professor Pedro Joseph-Nathan in recognition of his 50 years of outstanding scientific trajectory.

ABSTRACT

The electron impact induced fragmentations of *C*- and *N*-methyl, benzyl, and prenyl substituted 2-oxopyrrolidinoindolines were studied using an ion trap mass spectrometer (IT-MS). Correlations of characteristic fragment ions of the 2-oxopyrrolidinoindoline skeleton with specific modifications of the substituents around it were supported by stepwise fragmentation MS/MS analysis and accurate mass measurements. The MS^3 spectra evidenced the neutral loss of methyl- or benzylisocyanate from the 2-oxopyrrolidine ring, which would result in a rearranged stable quinolinium ion. *www.relaquim.com*

Keywords: 2-oxopyrrolidinoindolines, mass spectra, EI-ion trap, MS/MS analysis, quinolinium ion, accurate mass.

RESUMEN

Se estudiaron las fragmentaciones inducidas por impacto electrónico de 2-oxopirrolidinoindolinas sustituidas en *C* y *N* por grupos metilo, bencilo, y prenilo usando un espectrómetro de trampa de iones (EM-TI). Las correlaciones de iones-fragmento característicos del esqueleto de 2-oxopirrolidinoindolinas con modificaciones específicas de los sustituyentes alrededor del mismo se sustentaron por análisis de fragmentación gradual EM/EM y mediciones de masa exacta. Los espectros EM^3 evidenciaron la pérdida neutra de metil- o bencilisocianato proveniente del anillo de 2-oxopirrolidina que podría resultar en un ión reordenado quinolinio estable. *www.relaquim.com*

Palabras clave: 2-oxopirrolidinoindolinas, espectros de masa, trampa de iones-IE, análisis EM/EM, ión quinolinio, masa exacta.

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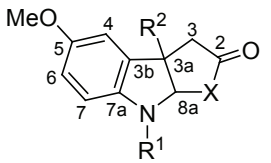
INTRODUCTION

Pyrrolidinoindolines are the basic nuclei of a number of alkaloids that have been isolated from a widespread series of natural sources, including amphibians, plants, and marine organisms (Anthoni *et al.*, 1990; Aygün and Pindur, 2003; Spande *et al.*, 1988; Tokuyama and Daly, 1983). These alkaloids exhibit an impressive array of promising biological properties which includes anticholinesterase activitiy (Rivera-Becerril *et al.*, 2008; Thal *et al.*, 1996; Yu *et al.*, 2010). Owing to their medicinal relevance and structural complexity, pyrrolidinoindoline alkaloids have served as a fertile area for the development of chemical strategies for their synthesis (Crich and Banerjee, 2007; Kim and Movassaghi, 2009; Morales-Ríos and Suárez-Castillo, 2008; Morales-Ríos *et al.*, 2001; Steven and Overman, 2007). A structural survey of this alkaloid family reveals a central *cis*-fused pyrrolidinoindoline core that in all cases incorporates a quaternary center at the C(3a) site. In addition, N(1), C(3a), and N(8) positions have been shown to incorporate broad variation in substituents.

Mass spectrometry (MS) has proven to be a successful approach for structural elucidation of furoindolines (Clayton and Reed, 1963; Morales-Ríos *et al.*, 2011) and pyrrolidinoindolines (Fales *et al.*, 1970; Rubino and Zecca, 1991; Spande *et al.*, 1988; Spiteller and Spiteller-Friedmann, 1963). In the current study, we were interested in establishing and validating MS fragmentation patterns of a series of 2-oxopyrrolidinoindolines **1a**-**1f** diversely substituted at N(1), C(3a), and N(8) by methyl, benzyl, and prenyl groups (Fig. 1) in order to provide correlations of characteristic fragment ions of the 2-oxopyrrolidinoindoline skeleton with specific modifications of the substituents around it.



1a: $R^1 = R^2$ = Bn, X = NMe

1b: $R^1 = R^2$ =  , X = NMe

1c: R^1 = Bn, R^2 =  , X = NMe

1d: $R^1 = R^2$ = Bn, X = NBn

1e: $R^1 = R^2$ =  , X = NBn

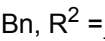
1f: R^1 = Bn, R^2 =  , X = NBn

Figure 1. Structures of target molecules **1a**-**1f**.

MATERIALS AND METHODS

General

Melting point was measured on a Fisher-Johns apparatus and is uncorrected. NMR experiments were performed using Varian Mercury spectrometers working at 300 and 75.4 MHz for ^{1}H and ^{13}C, respectively. Chemical shifts are reported in ppm downfield from tetramethylsilane. IR spectrum was measured with a Perkin-Elmer 16 FPC FT infrared spectrophotometer. All solvents and reagents were purchased in the reagent grade quality and were used without further purification. Solvents for chromatography were purified by distillation. Column chromatography was performed on Silica Gel 60 (230-400 mesh) from Aldrich. 2-Oxopyrrolidinoindolines **1a**-**1c** are known and were synthesized as described (Morales-Ríos *et al.* 2012) from the corresponding 2-oxofuroindolines **3a**-**3c** by treatment with methylamine.

EI-MS Analysis

The electron impact mass spectrometry (EI-MS) analyses were performed using an ion trap Varian Saturn 2000 spectrometer coupled with a Varian 3800 gas chromato-

graph. The MS conditions were as follows: transfer line heater, 280 °C; ion source temperature, 220 °C; electron impact ionization (EI) mode; ionization energy, 70 eV; electron multiplier voltage (EMV), 1950 V. The typical mass spectrum was recorded by averaging 1200 scans from *m/z* 20 to 650 at a scan rate of 1 s/scan. For multistage sequencing MS^2/MS^3, the compounds were introduced by direct insertion probe and the precursor ions were selected within an isolation width of 2 u. Exact mass measurements for the ions of interest were recorded on a Jeol JMS-GCMate II instrument or on an Agilent LCTOF spectrometer at the UCR Mass Spectrometry Facility, University of California, Riverside, with an error less than ±3 ppm for all ions discussed.

General lactamization procedure

To a solution of the appropriate 2-oxofuroindoline **3a**, **3b** or **3c** (0.65 mmol) in MeOH (20 mL) was added $BnNH_2$ (2.3 equiv, 0.17 mL). The reaction mixture was kept at room temperature for 5-120 h. After this, the solvent was removed under reduced pressure and the residue was suspended in EtOAc (30 mL). The suspension was washed successively with a 5% aq HCl solution (2 x 10 mL) and brine (2 x 10 mL). The organic layer was dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The crude product was purified by flash chromatography (7:3 hexane/EtOAc).

1,3a,8-Tribenzyl-5-methoxy-2-oxo-2,3,3a,8a-tetrahydro-8*H*-pyrrolo[2,3-*b*]indole (1d). Following the general procedure, a mixture of **3a** (250 mg) and $BnNH_2$ in MeOH was refluxed for 96 h to give **1d** (219 mg, 71%) as colorless crystals, mp 118-119 °C. TLC: R_f 0.42 (7:3 hexane/EtOAc); IR ($CHCl_3$) υ_{max} 3010, 2934, 1678, 1602, 1496 cm^{-1}. ^1H NMR ($CDCl_3$) δ 6.69 (1H, dd, *J* = 8.6, 2.6 Hz, H6), 6.58 (1H, d, *J* = 2.5 Hz, H4), 6.39 (1H, d, *J* = 8.5 Hz, H7), 4.80 (1H, s, H8a), 3.73 (3H, s, OMe), 2.93 and 2.82 (2H, AB, *J* = 17.2 Hz, H3), *N*1-Bn: 7.28-7.13 (overlapped, $2H_m$, H_p), 6.85 (2H, m, $2H_o$), 4.92 and 3.93 (2H, AB, *J* = 17.5 Hz, CH_2), *C*3a-Bn: 7.28-7.13 (overlapped, $2H_m$, H_p), 6.65 (2H, m, $2H_o$), 2.80 and 2.66 (2H, AB, *J* = 13.5 Hz, CH_2), *N*8-Bn: 7.28-7.13 (overlapped, $2H_m$, H_p), 6.96 (2H, m, $2H_o$), 4.04 and 3.75 (2H, AB, *J* = 15.7 Hz, CH_2); ^{13}C NMR ($CDCl_3$) δ 172.4 (C2), 154.1 (C5), 143.9 (C7a), 136.0 (C3b), 114.2 (C6), 111.0 (C7), 110.2 (C4), 84.9 (C8a), 55.9 (OMe), 51.3 (C3a), 41.9 (C3), *N*1-Bn 136.5 (C_i), 128.3 ($2C_m$), 127.6 ($2C_o$), 127.3 (C_p), 43.5 (CH_2), *C*3a-Bn: 136.2 (C_i), 130.1 ($2C_o$), 128.6 ($2C_m$), 126.9 (C_p), 44.7 (CH_2), *N*8-Bn: 138.7 (C_i), 128.5 ($2C_m$), 127.5 ($2C_o$), 127.4 (C_p), 54.7 (CH_2); EIMS *m/z* (%) $M^{+\bullet}$ 474 (100), 383 (42), 292 (6), 250 (13), 91 (23).

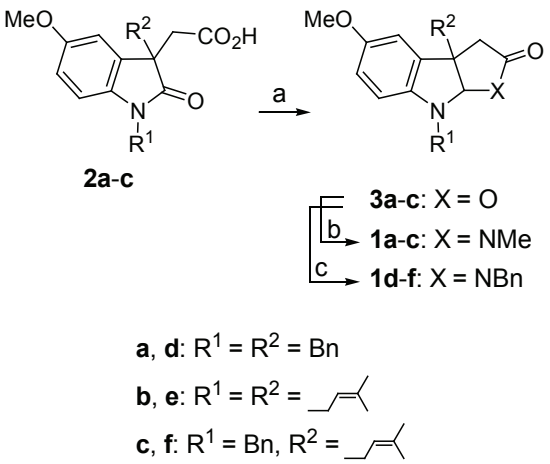
1-Benzyl-5-methoxy-3a,8-bis(3-methyl-2-buten-1-yl)-2-oxo-2,3,3a,8a-tetrahydro-8*H*-pyrrolo[2,3-*b*]indole (1e). Following the general procedure, a mixture of **3b** (223 mg) and $BnNH_2$ in MeOH was refluxed for 5 h to give **1e** (197 mg, 70%) as pale yellow oil. TLC: R_f 0.20 (7:3 hexane/EtOAc); IR ($CHCl_3$) υ_{max} 3006, 2972, 1676, 1598, 1494 cm^{-1};^1H NMR ($CDCl_3$) δ 6.69 (1H, dd, *J* = 8.4, 2.5 Hz, H6), 6.67 (1H, d, *J* = 2.5 Hz, H4), 6.50 (1H, d, *J* = 8.5 Hz, H7), 4.62 (1H, s, H8a), 3.75 (3H, s, OMe), 2.81 and 2.72 (2H, AB, *J* = 17.3 Hz, H3), *N*1-Bn: 7.32 (2H, m, $2H_m$), 7.28 (1H, m, H_p), 7.21 (2H, m, $2H_o$), 5.05 and 4.13 (2H, AB, *J* = 15.7 Hz, CH_2), *C*3a-Pre: 5.00 (1H, partially overlapped, ABX, tm, *J* = 7.4 Hz, CH=), 2.39 and 2.24 (2H, ABX, *J* = 14.6, 8.0, 7.4 Hz, CH_2), 1.70 (3H, s, Me), 1.48 (3H, s, Me); *N*8-Pre: 5.10 (1H, partially overlapped, ABX, tm, *J* = 7.0 Hz, CH=), 3.74 and 3.60 (2H, ABX, *J* = 15.8, 7.7, 7.0 Hz, CH_2), 1.59 (3H, s, Me), 1.74 (3H, s, Me); ^{13}C NMR ($CDCl_3$) δ 173.0 (C2), 154.1 (C5), 143.5 (C7a), 137.7 (C3b), 113.6 (C6), 111.1 (C7), 109.8 (C4), 85.5 (C8a), 55.8 (OMe), 50.2 (C3a), 41.5 (C3), *N*1-Bn: 136.3 (C_i), 128.5 ($2C_m$), 127.3 ($2C_o$), 127.2 (C_p), 43.4 (CH_2), *C*3a-Pre: 135.9 (C=), 118.4 (CH=), 37.3 (CH_2), 25.9 (Me), 17.9 (Me), *N*8-Pre: 135.1 (C=), 120.8 (CH=), 48.8 (CH_2), 25.6 (Me), 17.6 (Me); EIMS *m/z*

(%) $M^{+\bullet}$ 430 (100), 362 (17), 293 (67), 160 (22), 91 (10).

1,8-Dibenzyl-5-methoxy-3a-(3-methyl-2-buten-1-yl)-2-oxo-2,3,3a,8a-tetrahydro-8*H*-pyrrolo[2,3-*b*]indole (1f). Following the general procedure, a mixture of **3c** (236 mg) and $BnNH_2$ in MeOH was refluxed for 120 h to give **1f** (244 mg, 83%) as pale yellow oil. TLC: R_f 0.48 (7:3 hexane/EtOAc); IR ($CHCl_3$) υ_{max} 3016, 2934, 1678, 1598 cm^{-1}; ^1H NMR ($CDCl_3$) δ 6.67 (1H, overlapped, H4), 6.64 (1H, partially overlapped, dd, J = 8.4, 2.6 Hz, H6), 6.38 (1H, d, J = 8.0 Hz, H7), 4.70 (1H, s, H8a), 3.74 (3H, s, OMe), 2.78 (2H, s, H3), *N*1-Bn: 7.32-7.22 (overlapped, $2H_m$, H_p), 7.03 (2H, m, $2H_o$), 5.04 and 3.93 (2H, AB, J = 15.6 Hz, CH_2), *C*3a-Pre: 4.91 (1H, ABX, tm, J = 7.3 Hz, CH=), 2.29 and 2.13 (2H, ABX, J = 14.5, 8.1, 7.3 Hz, CH_2), 1.66 (3H, s, Me), 1.39 (3H, s, Me), *N*8-Bn: 7.32-7.22 (overlapped, $2H_m$, H_p), 7.12 (2H, m, $2H_o$), 4.33 and 4.18 (2H, AB, J = 16.2 Hz, CH_2); ^{13}C NMR ($CDCl_3$) δ 173.2 (C2), 154.1 (C5), 143.9 (C7a), 137.0 (C3b), 113.5 (C6), 110.3 (C7), 110.1 (C4), 86.4 (C8a), 55.9 (OMe), 50.1 (C3a), 41.8 (C3), *N*1-Bn: 136.2 (C_i), 128.6 ($2C_m$), 127.6 ($2C_o$), 127.4 (C_p), 43.8 (CH_2), *C*3a-Pre: 136.0 (C=), 118.3 (CH=), 37.4 (CH_2), 25.9 (Me), 18.0 (Me), *N*8-Bn: 138.7 (C_i), 128.5 ($2C_m$), 127.3 (C_p), 127.2 (C_o), 55.3 (CH_2); EIMS m/z (%) $M^{+\bullet}$ 452 (100), 383 (41), 292 (3), 250 (7), 91 (2).

RESULTS AND DISCUSSION

The preparation of 2-oxopyrrolidinoindolines **1a**-**1f** was achieved in two steps from the sodium salt of 2-(1,3-dialkyl-2-oxo-3-indolyl)acetic acids **2a**-**2c**. The reductive cyclization of **2a**-**2c** with $LiBHEt_3$ gave the corresponding 2-oxofuroindolines **3a**-**3c**. Stirring MeOH solutions of **3a**-**c** with methylamine at room temperature for 2-24 h or with benzylamine in boiling methanol for 5-120 h gave the expected 2-oxopyrrolidinoindolines **1a**-**1f** in a combined yield of 40-65% for the two-step process (Scheme 1).



Scheme 1. Preparation of 2-oxopyrrolidinoindolines **1a**-**1f**. a) NaH/THF, rt, 10 min, then $LiBHEt_3$/THF, rt, 6 h; (b) 40% aqueous $MeNH_2$/MeOH, rt, 2-24 h; (c) $BnNH_2$/MeOH, reflux, 5-120 h.

The electron impact mass spectra (EI-MS) of compounds **1a**-**1f** (Table 1) were complemented by tandem mass spectrometry (MS/MS). This technique involves the isolation of a specific ion, and the subsequent fragmentation thereof. Figure 2 illustrates MS^1 and MS^2 spectra obtained from **1a**-**1f**. In single IT-MS mode (Fig. 2, left side), the base peaks corresponding to $M^{+\bullet}$ were observed along with several fragment ions. These spectra are very alike, exhibiting common peaks that correspond to losses of 91 Da for **1a** and **1d**, and of 69 Da for **1b**, **1c**, **1e** and **1f**. The accurate mass measurement and the stepwise fragmentation MS^2 analysis of **1a**-**1f** (Fig. 2, right side) revealed that such losses involved the cleavage of the benzyl or prenyl moieties (Table 1). The EI-MS of compounds **1a** or **1d** acquired in MS^3 mode revealed that the fragment ions were very similar from those of **1c** or **1f**, respectively, evidencing that the base peaks in the MS^2 mode (Fig. 2, right side) were formed from loss of the angular C(3a)-substituent moiety. Whereas, compounds **1b** and **1e**, both characterized by the presence of two prenyl groups at positions C(3a) and N(8), exhibit significant peaks in the MS^2 mode at m/z 285 and

217 for **1b** and at *m/z* 362 and 293 for **1e** attributed to the loss of one or two of the prenyl groups from the molecular ions (Fig. 2, right side). The elemental compositions of these ions were confirmed using accurate mass measurements, which afforded relative errors within the range -2.7 to 1.4 ppm (Table 2).

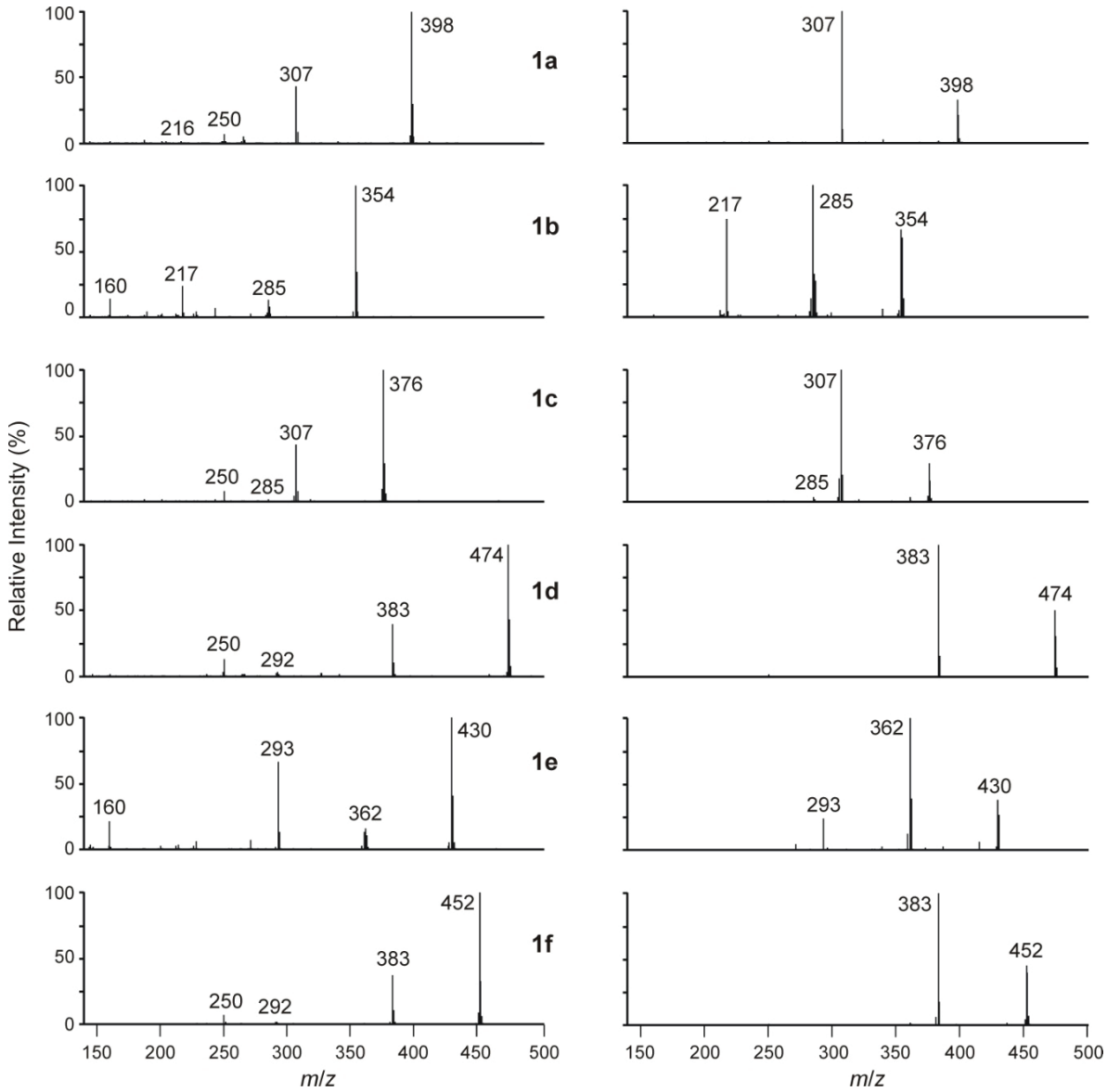


Figure 2. MS^1 (left side) and MS^2 (right side) spectra of **1a**-**1f** obtained by direct insertion probe at 300 °C.

Table 1. Characteristic *m*/*z* values (relative intensity, %) in EI-MS spectra[a] of **1a**-**1f**

Ion assignment	**1a**	**1b**	**1c**	**1d**	**1e**	**1f**
$M^{+\bullet}$	398 (100)	354 (100)	376 (100)	474 (100)	430 (100)	452 (100)
$[M-Bn]^+$	307 (43)	-	285 (1)	383 (42)	-	-
$[M-Pre]^+$	-	285 (14)	307 (44)	-	362 (17)	383 (41)
$[M-Bn-Bn]^{+\bullet}$	216 (2)	-	-	292 (6)	-	-
$[M-Pre-Pre + H]^+$	-	217 (24)	-	-	293 (67)	-
$[M-Pre-Bn]^{+\bullet}$	-	-	-	-	-	292 (3)
$[M-Bn-C_3H_7N]^+$	250 (7)	-	-	-	-	-
$[M-Pre-C_3H_7N]^+$	-	-	250 (8)	-	-	-
$[M-Bn-C_9H_{11}N]^+$	-	-	-	250 (13)	-	-
$[M-Pre-C_9H_{11}N]^+$	-	-	-	-	-	250 (7)
$[M-Pre-Pre-C_3H_7N + H]^+$	-	160 (15)	-	-	-	-
$[M-Pre-Pre-C_9H_{11}N + H]^+$	-	-	-	-	160 (22)	-

[a] By direct insertion probe at 300 °C.

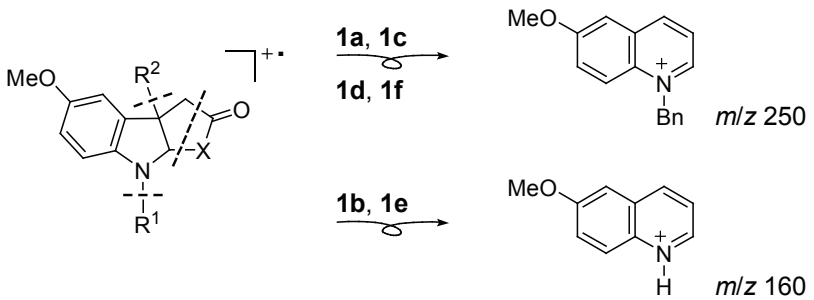
Table 2. High-resolution mass measurements for major fragment ions of **1a**-**1f**

Ion	Measured mass (*m*/*z*)	Calculated mass (*m*/*z*)	Elemental composition	Error (ppm)
1a				
$M^{+\bullet}$	398.1996	398.1994	$C_{26}H_{26}N_2O_2$	+0.4
$[M-Bn]^+$	307.1451	307.1447	$C_{19}H_{19}N_2O_2$	+1.4
$[M-Bn-Bn]^{+\bullet}$	216.0897	216.0899	$C_{12}H_{12}N_2O_2$	-0.8
$[M-Bn-C_3H_7N]^+$	250.1233	250.1232	$C_{17}H_{16}NO$	+0.4
1b				
$M^{+\bullet}$	354.2309	354.2307	$C_{22}H_{30}N_2O_2$	+0.5
$[M-Pre]^+$	285.1600	285.1603	$C_{17}H_{21}N_2O_2$	-1.1
$[M-Pre-Pre + H]^+$	217.0976	217.0977	$C_{12}H_{13}N_2O_2$	-0.5
$[M-Pre-Pre-C_3H_7N + H]^+$	160.0765	160.0762	$C_{10}H_{10}NO$	+1.6
1c				
$M^{+\bullet}$	376.2153	376.2151	$C_{24}H_{28}N_2O_2$	+0.6
$[M-Bn]^+$	285.1604	285.1603	$C_{17}H_{21}N_2O_2$	+0.3
$[M-Pre]^+$	307.1440	307.1447	$C_{19}H_{19}N_2O_2$	-2.1
$[M-Pre-C_3H_7N]^+$	250.1231	250.1232	$C_{17}H_{16}NO$	-0.4
1d				
$M^{+\bullet}$	474.2300	474.2307	$C_{32}H_{30}N_2O_2$	-1.5
$[M-Bn]^+$	383.1757	383.1760	$C_{25}H_{23}N_2O_2$	-0.7

Table 2. *continued*

Ion	Measured mass (*m/z*)	Calculated mass (*m/z*)	Elemental composition	Error (ppm)
$[M-Bn-Bn]^{+\bullet}$	292.1211	292.1212	$C_{18}H_{16}N_2O_2$	-0.3
$[M-Bn-C_9H_{11}N]^+$	250.1229	250.1232	$C_{17}H_{16}NO$	-1.2
1e				
$M^{+\bullet}$	430.2622	430.2620	$C_{28}H_{34}N_2O_2$	+0.4
$[M-Pre]^+$	362.1990	362.1994	$C_{23}H_{26}N_2O_2$	-1.2
$[M-Pre-Pre+H]^+$	293.1282	293.1290	$C_{18}H_{17}N_2O_2$	-2.7
$[M-Pre-Pre-C_9H_{11}N+H]^+$	160.0764	160.0762	$C_{10}H_{10}NO$	+1.0
1f				
$M^{+\bullet}$	452.2461	452.2464	$C_{30}H_{32}N_2O_2$	-0.6
$[M-Pre]^+$	383.1754	383.1760	$C_{25}H_{23}N_2O_2$	-1.5
$[M-Pre-Bn]^{+\bullet}$	292.1208	292.1212	$C_{18}H_{16}N_2O_2$	-1.3
$[M-Pre-C_9H_{11}N]^+$	250.1231	250.1232	$C_{17}H_{16}NO$	-0.4

Continuing with the fragmentation process of **1a**-**1f**, the cleavage of the lactam ring give rise to *m/z* 160 ion in the MS^3 spectra of **1b** and **1e** and to *m/z* 250 ion in the MS^3 spectra of **1a**, **1c**, **1d**, and **1f** (Scheme 2). These ions could derived from neutral loss of methyl- or benzylisocyanate (MeN=C=O or BnN=C=O), which are concurrent with the elimination of the prenyl group at N8 in the case of **1b** and **1e**. The *m/z* 160 and 250 are probably formed with simultaneous rearrangement to the stable ring-expanded quinolinium ions, whose plausible structures are shown in Scheme 2. The accurate mass measurements (Table 2) confirmed the composition of the fragments of interest, i.e. *m/z* 250.1233 for **1a** (calcd for $C_{17}H_{16}NO^+$ 250.1232) and *m/z* 160.0765 for **1b** (calcd for $C_{10}H_{10}NO$ + H^+ 160.0762).



Compound	MS^2	MS^3
1a	398 → 307	307 → 250, 216
1b	354 → 285, 217	285 → 160
1c	376 → 307, 285	307 → 250
1d	474 → 383	383 → 292, 250
1e	430 → 362, 293	362 → 160
1f	452 → 383	383 → 292, 250

Scheme 2. Fragmentation patterns and quinolinium structures detected in the MS^3 ionization process for **1a**-**1f**.

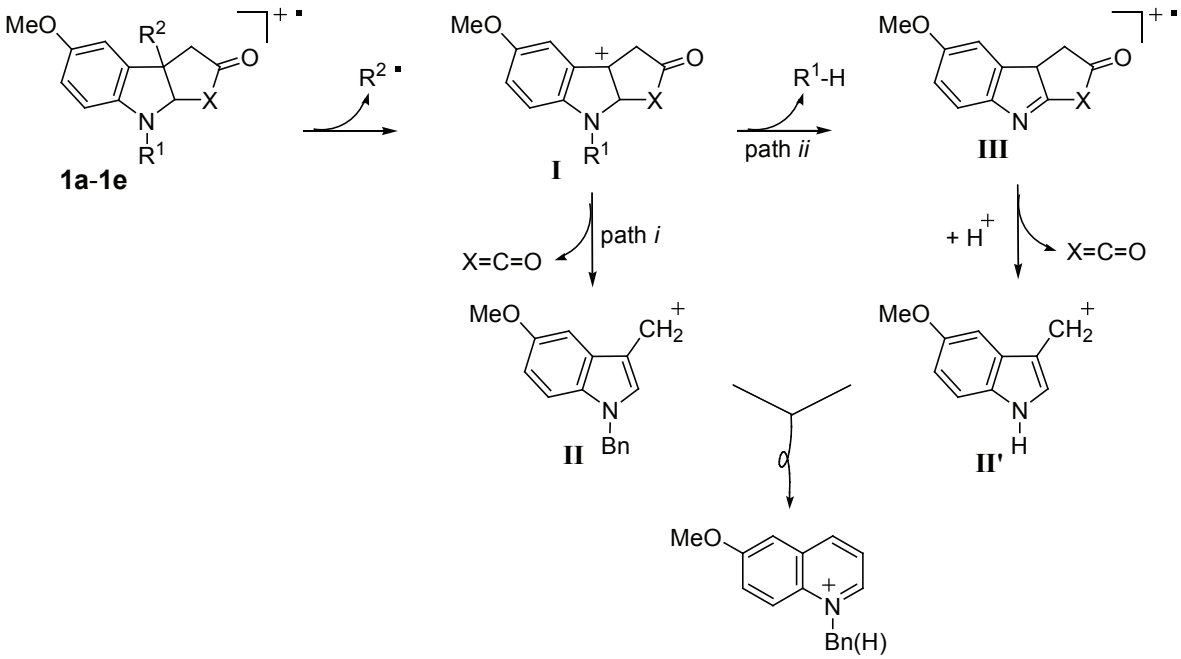


Figure 3. Fragment ion structures proposed for **1a**-**1e**. Path *i*: The main fragmentation pattern of **1a**, **1c**, **1d** and **1f.** Path *ii*: The fragmentation pattern of **1b** and **1e**.

Although the dominant fragmentation pathway for **1a**, **1c**, **1d** and **1f** includes the consecutive losses of $R^{2\bullet}$ and X=C=O from $M^{+\bullet}$ to give ion **II** (Fig. 3, path *i*), an alternative fragmentation route of ion **I** proceeds via elimination of $Bn^{\bullet}$ to give ion **III** (path *ii*) nevertheless in very low relative intensity (≤ 3%, Table 1). In contrast, elimination of $Pre^{\bullet}$ from ion **I** (path *ii*) is the only one fragmentation pathway for **1b** and **1e** giving $[M-Pre-Pre + H]^+$ ion **III** in 24% and 67%, respectively (Table 1).

CONCLUSIONS

The present study provided insight into the fragmentation patterns of diversely substituted 2-oxopyrrolidinoindolines **1a**-**1f**. The multiple-stage capability of the IT-MS together with accurate mass measurements on high-resolution instruments were invaluable for establishing fragmentation pathways, and greatly aided in proposing fragment ion structures.

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REFERENCES

Anthoni, U., Nielsen, P.H., Pereira, M., Christophersen, C. (1990) Bryozoan secondary metabolites: a chemotaxonomical challenge. *Biochemistry & Molecular Biology* **96B**: 431-437.

Aygün, A., Pindur, U. (2003) Chemistry and biology of new marine alkaloids from the indole and annelated indole series. *Current Medicinal Chemistry* **10**: 1113-1127.

Clayton, E., Reed, R.I. (1963) The mass spectra of physostigmine and some related compounds. *Tetrahedron* **19**: 1345-1357.

Crich, D., Banerjee, A. (2007) Chemistry of the hexahydropyrrolo[2,3-b]indoles: configuration, conformation, reactivity, and applications in synthesis. *Accounts of Chemical Research* **40**: 151-161.

Fales, H.M., Lloyd, H.A., Milne, G.W.A. (1970) Chemical ionization mass spectrometry of complex molecules. II. Alkaloids. *Journal of the American Chemical Society* **92**: 1590-1597.

Kim, J., Movassaghi, M. (2009) Biogenetically inspired syntheses of alkaloid natural products. *Chemical Society Reviews* **38**: 3035-3050.

Morales-Ríos, M.S., López-Camacho, P.Y., Jacobo-Cabral, C.O., Pérez-Rojas, N.A., Trujillo-Serrato, J.J., Burgueño-Tapia, E., Suárez-Castillo, O.R., Joseph-Nathan P. (2011) Unimolecular rearrangements of ketene-*O,O*-acetals and fragmentations occurring in the gas phase. *Journal of Mass Spectrom*etry **46**: 489-495.

Morales-Ríos, M.S., Rivera-Becerril, E., López Camacho, P.Y., Pérez-Rojas, N.A., Suárez-Castillo, O. R. (2012) Preparation of *O*-methyl substituted 2-oxofuro- and 2-oxopyrrolidinoindolines by reductive lactonization of oxindol-3-ylacetic acids. *Natural Product Communications* **7**: 1445-1451.

Morales-Ríos, M.S., Suárez-Castillo O.R. (2008) Synthesis of marine indole alkaloids from *Flustra foliacea. Natural Product Communications* **3**: 629-642.

Morales-Ríos, M.S., Suárez-Castillo, O.R., Trujillo-Serrato, J.J., Joseph-Nathan P. (2001) Total syntheses of five indole alkaloids from the marine bryozoan *Flustra foliacea. Journal of Organic Chemistry* **66**: 1186-1192.

Rivera-Becerril, E., Joseph-Nathan, P., Pérez-Alvarez, V.M., Morales-Ríos, M.S. (2008) Synthesis and biological evaluation of (-)- and (+)-debromoflustramine B and its analogues as selective butyrylcholinesterase inhibitors. *Journal of Medicinal Chemistry* **51**: 5271-5284

Rubino, F.M., Zecca, L. (1991) Retrosynthetic fragmentation in the fast atom bombardment mass spectra of eserine and some related compounds. *Organic Mass Spectrometry* **26**: 961-966.

Spande, T. F., Edwards, M.W., Pannell, L.K., Daly, J.W. (1988) Pseudophrynamine A: an unusual prenyl pyrrolo[2,3-*b*]indole ester from an australian frog, *Pseudophryne coriacea* (Myobatrachidae). *Journal of Organic Chemistry* **53**: 1222-1226.

Spiteller, G., Spiteller-Friedmann, M. (1963) Key fragments in alkaloid mass spectra. *Tetrahedron Letters* 147-152.

Steven, A., Overman, L.E. (2007) Total synthesis of complex cyclotryptamine alkaloids: stereocontrolled construction of quaternary carbon stereocenters. *Angewandte Chemie International Edition* **46**: 5488-5508.

Thal, L.J., Schwartz, G., Sano, M., Weiner, M., Knopman, D., Harrell, L., Bodenheimer, S., Rosser, M., Philpot, M., Schor, J., Goldberg, A. (1996) A multicenter double-blind study of controlled-release physostigmine for the treatment of symptoms secondary to Alzheimer's disease. *Neurology* **47***: 1389-1395.*

Tokuyama, T., Daly, J.W. (1983) Steroidal alkaloids (batrachotoxins and 4β-hydroxybatrachotoxins), "indole alkaloids" (calycanthine and chimonanthine) and a piperidinyldipyridin. *Tetrahedron* **39**: 41-47.

Yu, Q.-s., Holloway, H.W., Luo, W., Lahiri, D.K., Brossi, A., Greig, N.H. (2010) Long-acting anticholinesterases for myasthenia gravis: synthesis and activities of quaternary phenylcarbamates of neostigmine, pyridostigmine and physostigmine. *Bioorganic & Medicinal Chemistry* **18**: 4687-4693.