EI and CI Mass Fragmentation of Tryptamine, Tetrahydro-β-carboline and Some of their Derivatives

Jukka Gynther

Department of Pharmaceutical Chemistry, University of Kuopio, P.O. Box 6, SF-70211 Kuopio, Finland

Gynther, J., 1988. EI and CI Mass Fragmentation of Tryptamine, Tetrahydro-β-Carboline and Some of their Derivatives. – Acta Chem. Scand., Ser. B 42: 433–441.

EI and CI mass spectra of tryptamine and eleven of its derivatives, as well as those of tetrahydro-β-carboline (1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole, THBC) and twelve of its derivatives were recorded and interpreted. An intense ion, most probably possessing a quinolinium ion structure, is produced from tryptamine derivatives by EI. The retro-Diels-Alder reaction is a prominent EI fragmentation pathway of β-carbolines (BC's), and a bulky 1-substituent os also easily split off.

CI causes ammonia expulsion from tryptamine derivatives with a primary amino group in the side-chain; in the case of secondary amines and tryptophan derivatives, $(MH)^+$ is the base peak when methane is used as the reaction gas. $(MH)^+$ is the base peak in the CI spectra of BC derivatives, except in the case of derivatives which have a free carboxyl group, the loss of which often yields the base peak.

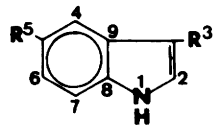
Only few previous reports have dealth with the mass spectrometric fragmentation of tryptamines and β-carbolines (9*H*-pyrido[3,4-b]indoles, BC's).[1–5] Several tryptamine and BC derivatives are found in trace amounts in human and animal tissues,[6–13] and it has been suggested that some BC's have neuromodulator and endocrinologic functions;[14–16] consequently these compounds should be of interest from a pharmacological point of view. Gas-liquid chromatography-mass spectrometry (GLC-MS) has been most effectively and widely used in analytical investigations on these compounds because of the high specificity and sensitivity of this method.

EI ionization was earlier most commonly used in GLC-MS analysis of BC's. However, as the result of intensive fragmentation, a reduced sensitivity is usually encountered when recording weak molecular ions. On the other hand, the specificity of the analysis may be unsatisfactory when only one intense fragment ion is monitored.

CI may provide some advantages over EI as far as quantitative analysis of trace amounts of compounds is concerned. When recording intense $(MH)^+$ ion peaks, increased analytical sensitivity is often obtained without loss of specificity. In this study, EI and CI mass spectra of tryptamine and eleven of its derivatives (Fig. 1), as well as THBC and twelve of its derivatives (Fig. 2), were recorded and interpreted.

Experimental

Chemicals. The following compounds were purchased from Fluka AG (Buchs, Switzerland): Tryptamine (**1**), tryptophan (**9**), 5-methoxytryptophan (**10**) and 5-hydroxyindole-3-acetic acid (**12**). From Sigma (St. Louis, USA) were: 5-Hydroxytryptamine (**4**), *N*-methyltryptamine (**5**), 5-hydroxy-*N*-methyltryptamine (**6**), melatonin (**7**), indole-3-acetaldehyde (**11**), 2-methyl-tetrahydro-β-carboline (2-methyl-1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole, **17**), 6-hydroxy-1-methyl-tetrahydro-β-carboline (6-hydroxy-1-methyl-1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole, **21**), norharmane (9*H*-pyrido[3,4-b]indole, **23**), harmol (1-methyl-9*H*-pyrido[3,4-b]indole, **24**) and harminic acid (7-methyl-1*H*-pyrrolo[2,3-c]pyridine-2,3-di-



	R^3	R^5
	β α ω	
1	$CH_2CH_2NH_2$	H
2	$CH_2CD_2NH_2$	H
3	$CD_2CD_2NH_2$	H
4	$CH_2CH_2NH_2$	OH
5	$CH_2CH_2NHCH_3$	H
6	$CH_2CH_2NHCH_3$	OH
	α β ω α'β'	
7	$CH_2CH_2NHCOCH_3$	OCH_3
8	$CD_2CD_2NHCOCH_3$	OCH_3
9	$CH_2CH(COOH)NH_2$	H
10	$CH_2CH(COOH)NH_2$	OCH_3
11	CH_2COOH	OH
12	CH_2CHO	H

Fig. 1. The molecular structures of tryptamine (**1**) and eleven of its derivatives.

carboxylic acid, **25**). All other compounds studied were synthesized in our laboratory according to Refs. 3 and 17–19.

Tryptamine-α,α-d_2 (**2**) was synthesized from 11.5 mmol of indole-3-acetamide using 28.8 mmol of lithium aluminium deuteride in 50 ml of dry tetrahydrofuran. The mixture was heated under reflux overnight and the organic phase was separated and evaporated. The oily residue was dissolved in dry dichloromethane and the product precipitated as the hydrochloride salt by addition of saturated ethanolic hydrochloric acid. The free base was obtained by dropwise addition of dilute sodium hydroxide to an aqueous solution of **2** hydrochloride; yield 3.2 mmol (28 %), m.p. 114–117 °C. IR: 1080 cm^{-1} (C–N), 3270 cm^{-1} (N–H). Isotopic purity 93.5 %.

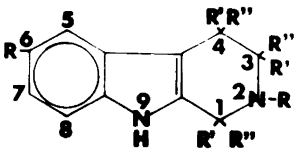
Tryptamine-α,α,β,β-d_4 (**3**) was synthesized from 17.3 mmol of indole-3-glyoxylyl amide by heating under reflux with 53.6 mmol of lithium aluminium deuteride as in the case of **2**; yield 4.4 mmol (25 %), m.p. 115–117 °C. IR: 1080 cm^{-1} (C–N), 3270 cm^{-1} (N–H). Isotopic purity 96.4 %.

Melatonin-α,α,β,β-d_4 (**8**) was prepared from 5-methoxy-tryptamine-α,α,β,β-d_4 by stirring a mixture of 10 ml of acetic anhydride and 2.6 mmol of 5-methoxytryptamine-α,α,β,β-d_4 in 25 ml of pyridine for 1 h. The mixture was extracted with dichloromethane, and the dichloromethane phase washed with dilute HCl. After evaporation of the solvent, **8** was recrystallized from benzene; yield 1.8 mmol (69 %), m.p. 115–117 °C. IR: 1080 cm^{-1} (C–N), 1200 cm^{-1} (O–C), 1650 cm^{-1} (C=O), 3280 cm^{-1} (N–H). ^{1}H NMR: δ 8.18 (N^1–H), 1.25 (C^2–H), 7.02 (C^4–H), 7.26 (C^6–H), 6.86 (C^7–H), – [C(α)–H], – [C(β)–H], 5.60 [N(ω)–H], 1.91 [C(β')+H], 3.88 (O–C–H). Isotopic purity 86.2 %.

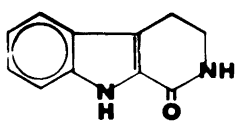
Tetrahydro-β-carboline (1,2,3,4-tetrahydro-9*H*-pyrido-[3,4-b]indole, **13**) was synthesized from 11.6 mmol of **18** by suspending it in 25 ml of water and adding 3 ml of concentrated hydrochloric acid. The mixture was then heated under reflux for 30 min. After cooling, the solution was made alkaline by dropwise addition of NaOH. The white precipitate obtained was filtered off and washed with water; yield 10.2 mmol (88 %), m.p. 200–202 °C. IR: 3290 cm^{-1} (N–H). ^{1}H NMR: δ 4.02 (C^1–H). 1.82 (N^2–H), 3.18 (C^3–H), 2.76 (C^4–H), 6.98–7.43 (C^{5-8}–H), 7.74 (N^9–H).

Tetrahydro-β-carboline-3,3,4,4-d_4 (**14**) was synthesized from 2.3 mmol of deuterated **18** in the same way as the unlabelled **13**; yield 1.6 mmol (70 %), m.p. 199–202 °C. IR: 3290 cm^{-1} (N–H). ^{1}H NMR: δ 4.01 (C^1–H), 1.72 (N^2–H), – (C^3–H), – (C^4–H), 7.01–7.65 (C^{5-8}–H), 7.85 (N^9–H). Isotopic purity 76.3 %.

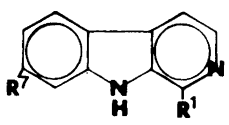
1-Methyl-tetrahydro-β-carboline (1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole, **15**) was synthesized from 25.5 mmol of **1** hydrochloride dissolved in 10 ml of water/ethanol (85:15 v/v). 0.1 ml of acetaldehyde was added and the pH of the mixture was adjusted to 3 by addition of 2 M hydrochloric acid. The mixture was heated under reflux



	$R^{1'}$	$R^{1''}$	R^2	$R^{3'}$	$R^{3''}$	$R^{4'}$	$R^{4''}$	R^6
13	H	H	H	H	H	H	H	H
14	H	H	H	D	D	D	D	H
15	CH_3	H	H	H	H	H	H	H
16	C_2H_5	H	H	H	H	H	H	H
17	H	H	CH_3	H	H	H	H	H
18	COOH	H	H	H	H	H	H	H
19	H	H	H	H	COOH	H	H	H
20	H	H	H	H	$COOCH_3$	H	H	H
21	CH_3	H	H	H	H	H	H	OH

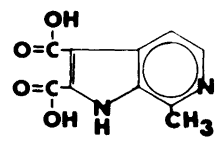


22 1-oxo-THBC



23 Norharmane $R^1=R^7=H$

24 Harmol $R^1=CH_3$, $R^7=OH$



25 Harminic acid

Fig. 2. The molecular structures of tetrahydro-β-carboline (**13**) and twelve of its derivatives.

for 30 min, and after cooling, **15** was extracted from the alkalinized solution using ethyl acetate. The solvent was evaporated and the brownish viscous liquid product was crystallized from dichloromethane/petroleum ether (2:1); yield 4.6 mmol (18 %), m.p. 174–176 °C. IR: 3290^{-1} (N–H).

1-Ethyl-tetrahydro-β-carboline (1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole, **16**) was synthesized from 25.5 mmol of **1** hydrochloride and 0.1 mmol of propionaldehyde according to the procedure given for **15**; yield 3.9 mmol (15 %), m.p. 115–117 °C. IR: 3290 cm^{-1} (N–H).

Tetrahydro-β-carboline-1-carboxylic acid (1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole-1-carboxylic acid, **18**) was synthesized from **1** hydrochloride (25.5 mmol) dissolved in 100 ml of water. 26.5 mmol of glyoxylic acid monohydrate was added and the pH of the solution was adjusted to 4 by adding 1 M KOH. The solution was stirred for 1 h, during which time a precipitate was formed. After filtration and washing with cold water the yield was 20.8 mmol (82 %), m.p. 205–208 °C. IR: 1650 cm^{-1} (C=O), 3300 cm^{-1} (N–H).

Tetrahydro-β-carboline-3-carboxylic acid (1,2,3,4-tetrahydro-9*H*-pyrido[3,4-b]indole-3-carboxylic acid, **19**) was prepared from 24.5 mmol of **1** in alkalinized water solution (pH 9).

30 % formalin (25 mmol) was added, and after stirring at room temperature for 2 h the solution was heated under reflux for 3 h **19** crystallized out from neutralized solution. After successive washings with water, methanol and dichloromethane, the yield was 11.2 mmol (46 %), m.p. 301–305 °C. IR: 1630 cm^{-1} (C=O), 3390 cm^{-1} (N–H), 3450 cm^{-1} (O–H).

Tetrahydro-β-carboline-3-methylcarboxylate (**20**) was synthesized from 4.6 mmol of **19** dissolved in methanol. Dry hydrogen chloride was bubbled through the solution until it became clear, maintaining the temperature below 40 °C. The white precipitate obtained was filtered off and washed with methanol; yield 3.5 mmol (76 %), m.p. 250–254 °C. IR: 1735 cm^{-1} (C=O), 3200 cm^{-1} (N–H).

1-Oxo-tetrahydro-β-carboline (**22**) was synthesized using 2,3-piperidinedione-3-phenylhydrazone (5.4 mmol) and 10 ml of 90 % formic acid. The solution was heated under reflux for 30 min, and after addition of water to the hot solution an oily phase separated out. This oily layer was redissolved by dropwise addition of hot ethanol. After cooling, yellow crystals were obtained which were recrystallized from dilute ethanol; yield 3.8 mmol (70 %), m.p. 201–203 °C. IR: 1655 cm^{-1} (C=O), 3230 cm^{-1} (N–H).

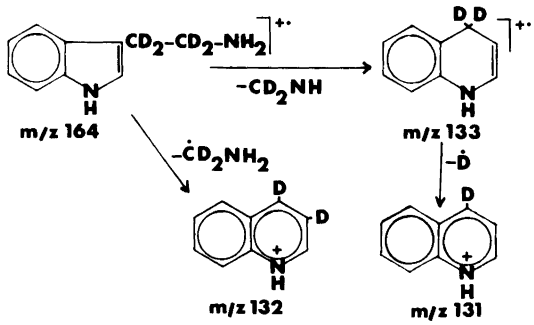
Melting points were recorded using a capillary melting point instrument and are uncorrected. IR spectra of the synthezised compounds were recorded in the solid state on a Pye Unicam SP 1050 infrared spectrophotometer using KBr disks. ^1H NMR spectra were obtained on a Bruker AM-250 MHz instrument. EI and CI mass spectra were recorded using a Jeol JMS-D 300 instrument with a JMA 2000 mass analysis system at a resolution of 1000. Methane was used as the CI reaction gas, and a solids-insertion probe was used for sample admission. The ionizing electron beam energy was 70 eV in the EI, and 200 eV in the CI technique. The ionization current was 300 μA, and the ionization chamber was maintained at 230 °C.

Results and discussion

EI mass fragmentation of tryptamine derivatives. The expulsion of $(CH_4N)^{\cdot}$ from the ethylamino side-chain of **1** yields the ion at *m/z* 130 which is the base peak, most probably with the quinolinium ion structure.[1] This ion is also formed by the successive loss of CH_3N and $H^{\cdot}$ from the molecular ion, which is confirmed by analogous fragmentation of **2** and **3**. In the case of **3**, the loss of $(CD_2NH+D)^{\cdot}$ yields an ion $(M-33)^+$ which is more intense than the corresponding ion $(M-31)^+$ in the spectra of **1** and **2**. This suggests that the quinolinium ion in which one deuterium is retained can result from a hydroquinolinium ion after the loss of one deuterium from the β-position of the parent **3** ion (Scheme 1).

Further cleavage of CHN from the quinolinium ion yields an ion $(C_8H_7)^+$ at *m/z* 103 (**1** and **2**) and an ion $(C_8H_6D)^+$ at *m/z* 105 (**3**) (Table 1). The fragmentation pattern of **4** is quite similar to that of **1**. After the cleavage of $(CH_4N)^{\cdot}$, the resulting hydroxyquinolinium ion remains unaffected by EI. In the case of *N*-methyltryptamines, the positive charge is most often retained by the nitrogen in the side-chain, yielding the ion $(C_2H_6N)^{+\cdot}$ at *m/z* 44, but the quinolinium and hydroquinolinium ions are also abundant (Table 1). The cleavage of $(C_3H_6NO)^{\cdot}$ from the acetylated ethylamino group of **7** yields the methoxyquinolinium ion at *m/z* 160. Another significant ion is formed by the loss of C_2H_5NO from the molecular ion. The methoxyquinolinium ion is further split off by the loss of CH_3 radical from the methoxy group and the subsequent loss of CO. The fragmentation pathway of **7** is confirmed by the analogous fragmentation of **8** (Table 1).

The peak-pattern of the EI mass spectra of **9** and **10** appears quite simple. The loss of $(C_2H_4NO_2)^{\cdot}$ yields the quinolinium or methoxyquinolinium ion, respectively, as the base peak



Scheme 1. Supposed EI fragmentation of tryptamine-α,α,β,β-d_4 (**3**).

Table 1. EI mass spectra of tryptamine and some of its derivatives [Relative intensity (%)].

	$[M]^{+\cdot}$	Quinolinium ion	Hydroquinolinium ion	Other ions
1	160(16)	130(100)	131(57)	117(12), 103(10)
2	162(14)	130(100)	131(78)	131(28), 103(14)
3	164(19)	132(100)	133(62)	134(7), 131(14), 105(7)
4	176(23)	146(100)	147(53)	
5	174(2)	130(52)	131(75)	132(9), 44(100)
6	190(3)	146(54)	147(68)	159(9), 44(100)
7	232(25)	160(100)	161(12)	174(13), 173(93), 145(16)
8	236(17)	162(100)	163(25)	177(31), 176(69), 147(22)
9	204(6)	130(100)	131(11)	117(7)
10	234(7)	160(100)	161(12)	145(11), 117(6)
11	191(38)	146(100)	147(11)	117(5)
12	159(26)	130(100)	131(11)	103(9), 77(13), 64(11)

(Table 1). Further fragmentation of the methoxyquinolinium ion proceeds like that of melatonin (cf. above).

A hydroxyquinolinium ion at *m/z* 146 is formed from **11** by the loss of $(CHO_2)^{\cdot}$; further loss of $(CHO)^{\cdot}$ yields an ion at *m/z* 117. In the mass spectrum of **12**, the most abundant ion at *m/z* 130 is produced by the loss of $(CHO)^{\cdot}$ from the molecular ion with the relative intensity of 25% (Table 1).

CI mass fragmentation of tryptamine derivatives. The protonated amino group of primary indoleethylamines (**1–4**) is lost as ammonia in the presence of methane reaction gas, this process yielding the base peak $(MH-NH_3)^+$. In contrast to the EI ionization, the cleavage of $(CH_4N)^{\cdot}$ is not favoured, and the intensity of the $(M)^{+\cdot}$ peak for these compounds remains quite low (Table 2).

An interesting ion, viz. $(M+12)^+$, is found in the CI spectra of all the primary amines studied. This ion clearly arises by immonium ion formation from the primary amino group, but this ion would also appear to be due to loss of ammonia from the $(M+C_2H_5)^+$ ion. The $(M+C_2H_5)^+$ ion is found in the CI spectrum of **6–8**, **11** and **12**, but

Table 2. CI mass spectra of tryptamine and some of its derivatives [Relative intensity (%)].

	$[M+29]^+$	$[M+12]^+$	$[M+1]^+$	$[M]^+$	Other peaks
1	189(−)	172(10)	161(23)	160(11)	145(11), 144(100)
2	191(−)	174(18)	163(37)	162(15)	147(46), 146(100)
3	193(−)	176(10)	165(18)	164(9)	149(11), 148(100)
4	205(−)	188(7)	177(4)	176(28)	160(100), 148(70), 147(55)
5	203(−)	186(−)	174(30)	174(4)	145(47), 132(28), 131(100), 130(15)
6	219(9)	202(−)	191(100)	190(2)	160(29), 148(13), 147(14)
7	261(33)	244(−)	233(100)	232(47)	202(14), 174(50), 173(17), 160(11)
8	165(24)	248(−)	237(100)	236(50)	206(10), 178(55), 177(27), 176(27), 162(18)
9	233(−)	216(−)	205(1)	204(1)	188(16), 159(10), 130(100)
10	263(−)	246(−)	235(4)	234(1)	218(23), 189(14), 174(12), 161(16), 160(100), 130(30)
11	220(7)	203(−)	192(100)	191(11)	148(67), 147(11), 146(27)
12	188(17)	171(−)	160(100)	159(26)	130(26)

Table 3. EI mass spectra of some beta-carbolines [Relative intensity (%)].

	$[M]^{+\cdot}$	Other peaks							
13	172(34)	171(11)	144(19)	143(100)	115(9)				
14	176(32)	175(16)	146(19)	145(100)	144(11)	117(10)			
15	186(62)	185(25)	172(14)	171(100)	157(46)	156(31)	144(12)	143(11)	
16	200(16)	172(14)	171(100)	170(8)	156(7)	144(7)			
17	186(25)	185(7)	144(15)	143(100)	142(7)				
18	216(3)	172(68)	171(100)	170(22)	169(42)	155(32)	144(26)		
		143(32)	130(17)	115(17)					
19	216(23)	172(60)	171(100)	170(19)	169(40)	155(32)	144(35)	143(77)	115(19)
20	230(54)	171(46)	169(22)	144(35)	143(100)	115(11)			
21	202(57)	201(25)	188(13)	187(100)	174(10)	173(48)	172(31)	160(9)	146(9)
22	186(77)	157(42)	130(15)	129(100)	128(16)	102(12)	98(28)		
23	168(100)	169(15)	167(9)	141(8)	114(9)				
24	198(100)	199(15)	197(22)	170(11)					
25	220(−)	214(7)	213(8)	176)	159(29)	158(74)	131(17)		
		130(33)	129(17)	104(14)	103(17)				

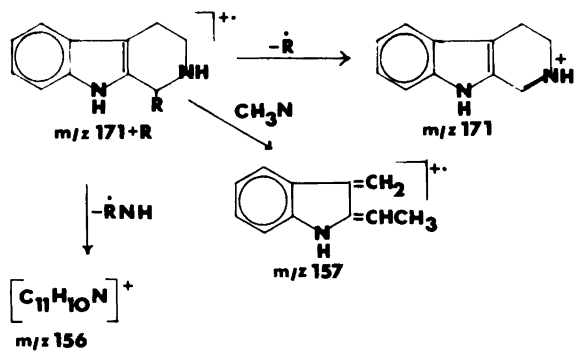
the ion $(MH-NH_3)^+$, which is typical of the primary amines, is now absent. $(MH)^+$ and $(M)^{+\cdot}$ peaks in the CI spectra of **9** and **10** are very small. The cleavage of $C_2H_5NO_2$ yields the base peaks of **9** and **10**, these ions possessing quinolinium and methoxyquinolinium ion structures, respectively. The loss of ammonia from these compounds results in ions with an abundance of about 20 % (Table 2).

In the CI spectrum of tryptamines with a secondary amino group, $(MH)^+$ is the base peak except for **5**. The secondary amino group is lost from **6, 7** and **8**, yielding ions $(MH-CH_5N)^+$, *m/z* 160, $(MH-C_2H_5NO)^+$, *m/z* 174, and $(MH-C_2H_5NO)^+$, *m/z* 178, respectively.

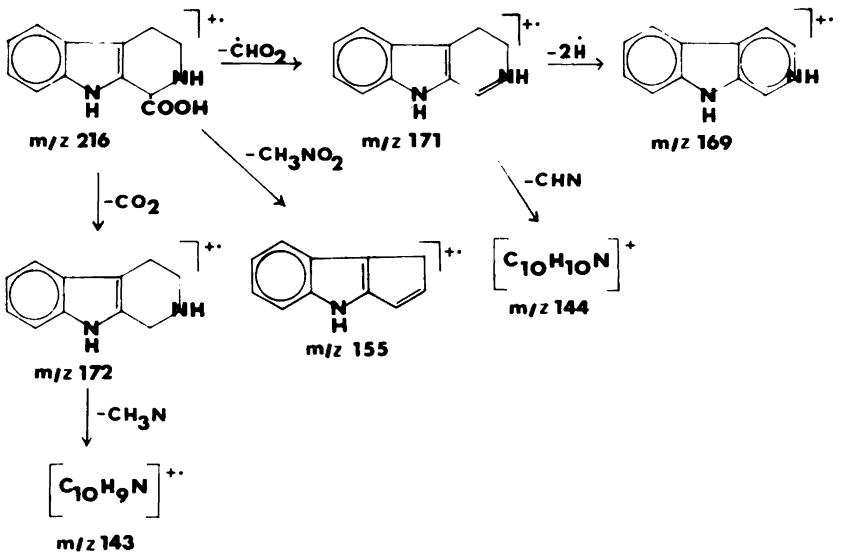
The loss of CO_2 and CH_2O_2, from the protonated **11**, produces two abundant ions at *m/z* 148 and *m/z* 146; however, $(MH)^+$ is the base peak, as in the case of **12**. This compound is further fragmented by the loss of CH_2O, the resultant ion showing the quinolinium ion structure (Table 2).

EI mass fragmentation of BC's. The retro-Diels-Alder reaction is favoured in the fragmentation of THBC's lacking bulky substituents in the 1-position.[4] The expulsion of CH_3N yields the most abundant ion, $(C_{10}H_9N)^{+\cdot}$. Analogously, an ion $(C_{10}H_7D_2N)^{+\cdot}$ is formed when CHD_2N is lost from **14**. This reaction mechanism is also operative in the case of **17** (Table 3).

Radical cleavage from the 1-position of THBC's yields the most abundant ion, $(M-R^1)^+$, this ion possessing the dihydro-β-carbolinium structure. **15** is also fragmented by the retro-Diels-Alder mechanism, yielding $(C_{11}H_{11}N)^{+\cdot}$, or by the expulsion of $(CH_4N)^{\cdot}$, which results in $(C_{11}H_{10}N)^+$ (Scheme 2). In the spectrum of **18**, the most abundant ion, $(C_{11}H_{11}N_2)^+$, most probably having the dihydro-β-carbolinium ion structure, is associated with the loss of CHO_2 radical from the molecular ion. The β-carbolinium ion results when hydrogen is lost from the dihydro-β-carbolinium ion (Scheme 3). The expulsion of CHN from the dihydro-β-carbolinium ion yields $(C_{10}H_{10}N)^+$. The loss of CO_2 from the molecular ion introduces an ion $(C_{11}H_{12}N_2)^{+\cdot}$; this ion is further fragmented by the retro-Diels-Alder



Scheme 2. Supposed EI fragmentation of 1-substituted THBC's.



Scheme 3. Supposed EI fragmentation of THBC-1-carboxylic acid (**18**).

mechanism, $(C_{10}H_9N)^{+\cdot}$ being the resultant ion. An interesting ion, $(C_{11}H_9N)^{+\cdot}$, with a relatively high abundance (32 %) is clearly the resultant ion from the expulsion of CH_3NO_2 from the molecular ion (Scheme 3).

EI mass fragmentation of **19** is comparable to that of **18**. However, the ion at *m/z* 143, as the result from the loss of CO_2 and the retro-Diels-Alder reaction, is appreciably more abundant in the case of **19**, as is the $(M)^{+\cdot}$ peak (Table 3). The retro-Diels-Alder reaction is the main fragmentation pathway of **20**. The cleavage of $(C_2H_3O)^{\cdot}$ from the molecular ion yields another abundant ion, in addition to quite intense $(M)^{+\cdot}$ (Table 3). EI fragmentation of **21** by the retro-Diels-Alder reaction yields $(C_{10}H_9NO)^{+\cdot}$, but the most abundant ion, $(C_9H_7N)^{+}$, is produced from the former by the loss of CO. The unsubstituted or 6-hydroxy-substituted benzene ring of BC's remains unaffected by EI, in the contrary to that of their 6-methoxy analogues.[5]

In the EI mass spectra of each of the THBC's analyzed, the $(M)^{+\cdot}$ ion is quite abundant except in the spectra of THBC's with a carboxyl substituent (Table 3). In the case of aromatic BC's the molecular ion is most abundant, and other quantitatively important ions are obtained through loss of H$^{\cdot}$ and CHN only (Table 3).

Compound **25**, with a molecular structure substantially different from that of the other BC's studied here, shows a peculiar EI mass spectrum. Facile expulsion of CO_2 yields $(C_9H_8N_2O_2)^{+\cdot}$ at *m/z* 176 as the base peak. The loss of H_2O from the latter ion yields $(C_9H_6N_2O)^{+\cdot}$, followed by loss of CO and CHN resulting in the ions $(C_8H_6NO)^{+\cdot}$ and $(C_8H_5NO)^{+\cdot}$, respectively, with considerable abundances (Table 3).

CI mass fragmentation of BC's. The base peak in the CI spectra of THBC's is most often $(MH)^{+}$, but the fragmentation of the tetrahydropyridine ring yields several other peaks (Table 4). In the case of **13**, the retro-Diels-Alder process introduces the very abundant ion $(MH-29)^{+}$. The same mechanism is operative for each of the THBC's studied, although the resultant peak intensities are lower (Table 4). The derivatives of THBC with an alkyl substituent in the 1-position undergo cleavage of this group, but substantially lower ion abundances are observed here than in the corresponding EI spectra. The indolyl moiety of the THBC's remains unaffected by CI.

In the CI mass fragmentation of **18**, the loss of CHO_2 radical and CO_2 yields $(C_{11}H_{12}N_2)^{+\cdot}$ and $(C_{11}H_{13}N_2)^{+\cdot}$, respectively, the latter further undergoing the retro-Diels-Alder reaction and formation of the most abundant ion $(C_{10}H_{10}N)^{+}$. The expulsion of CO_2 from **19** results in the base peak

Table 4. CI mass spectra of some beta-carbolines [Relative intensity (%)].

	$[M+H]^{+\cdot}$	Other peaks
13	173(100)	174(12), 172(14), 144(20), 143(12)
14	177(100)	178(14), 176(18), 175(10), 146(18), 145(14)
15	187(100)	188(15), 186(27), 172(11), 158(13), 144(13)
16	201(100)	202(15), 200(15), 172(13)
17	187(100)	188(13), 186(25), 185(10), 172(12), 144(22)
18	217(−)	173(25), 172(70), 171(37), 170(26), 145(13), 144(100), 143(56)
19	217(−)	174(13), 173(100), 172(31), 171(21), 144(58)
20	231(100)	232(14), 230(22), 172(11), 144(50)
21	203(100)	204(14), 202(27), 201(19), 188(13), 187(21)
22	187(100)	188(12), 186(14)
23	169(100)	170(14), 168(7)
24	199(100)	200(13), 198(9)
25	221(−)	205(13), 178(11), 177(100), 161(11), 159(7), 133(69)

formation. This ion is further split off by the retro-Diels-Alder mechanism. $(MH)^+$ is the base peak in the CI spectrum of **20**. An acetate radical is not cleaved off as readily as CO_2 or (CHO_2) from the corresponding acid; however, the retro-Diels-Alder fragmentation mechanism shows high activity, this reaction being excluded in the case of **21**.

In the CI spectra of aromatic BC's, i.e. **23** and **24**, $(MH)^+$ is the base peak and practically all other feasible peaks are present (Table 4). The $(M)^{+\cdot}$ peak in the EI spectrum of **25** is undetectably small, as is the $(MH)^+$ peak in the CI spectrum. The base peak $(MH-44)^+$ at *m/z* 177 results here from the loss of CO_2 from the protonated parent molecule. Further loss of another CO_2 yields an intensive ion $(C_8H_9N_2)^+$ at *m/z* 133. The expulsion of oxygen from the protonated molecule and from the ion $(MH-44)^+$ probably yields ions $(MH-16)^+$ at *m/z* 205 and $(MH-60)^+$ at *m/z* 161, respectively (Table 4).

In conclusion, EI fragmentation of tryptamine derivatives most often yields the quinolinium ion as the base peak. THBC's are fragmented mainly by the retro-Diels-Alder mechanism, but a bulky substituent in the 1-position results in an alternative fragmentation pathway. The degree of saturation and the nature of substituents in the tetrahydropyridine ring are the two factors showing the strongest effects on the EI fragmentation of these compounds. The possibility of ammonia expulsion during CI, when methane is used as the reaction gas, reduces the abundance of the $(MH)^+$ peak in the spectra of tryptamines. In the CI spectra of BC's, on the other hand the $(MH)^+$ peak is most often the base peak, apart from for the derivatives which have a free carboxyl group.

When methane is used as the reaction gas, CI does not provide any advantages over EI in the MS analysis of tryptamines and BC's. However, the use of other reaction gases, such as ammonia or butane, would in some respects be superior to the CI method considered in this study.

References

1. Couch, M. W. and Williams, C. M. *Anal. Biochem. 50* (1972) 612.
2. Hattox, S. E. and Murphy, R. C. *Biomed. Mass Spectrom. 2* (1975) 272.
3. Shaw, G. J., Wright, G. J. and Milne, W. A. *Biomed. Mass Spectrom. 4* (1977) 348.
4. Coutts, R. T., Locock, R. A. and Slywka, G. W. A. *Org. Mass Spectrom. 3* (1970) 879.
5. Gynther, J., Peura, P. and Salmi, S. *Acta Chem. Scand., Ser. B 39* (1985) 849.
6. Kari, I., Peura, P. and Airaksinen, M. M. *Biomed. Mass Spectrom. 7* (1980) 549.
7. Kari, I. *FEBS. Lett. 127* (1981) 277.
8. Peura, P., Kari, I. and Airaksinen, M. M. *Biomed. Mass Spectrom. 7* (1981) 559.
9. Leino, M., Kari, I., Airaksinen, M. M. and Gynther, J. *Exp. Eye Res. 36* (1983) 135.
10. Beck, O., Bosin, T. R., Lundman, A. and Borg, S. *Biochem. Pharmacol. 31* (1982) 2517.
11. Beck, O. and Bosin, T. R. *Biomed. Mass Spectrom. 6* (1979) 19.

12. Beck, O. and Flodberg, G. *Biomed. Mass Spectrom. 11* (1984) 155.
13. Artigas, F., Sunol, C., Tusell, J. M., Martinez, E. and Gelpi, E. *Biomed. Mass Spectrom. 11* (1984) 142.
14. Barchas, J. D., Akil, H., Elliott, G. R., Holman, R. B. and Watson, S. J. *Science 200* (1978) 964.
15. Airaksinen, M. M., Ho, B. T., An, R. and Taylor, D. *Arzneim.-Forsch. 14* (1978) 42.
16. Meyer, J. S. and Buckholtz, N. S. *Res. Commun. Chem. Pathol. Pharmacol. 14* (1976) 205.
17. Peura, P., Gynther, J. and Ylinen, M. *Acta Pharm. Fenn. 91* (1982) 55.
18. Ho, B. T. and Walker, K. E. *Org. Synth. 51* (1971) 136.
19. Lippke, K. P., Schunack, W. G., Wenning, W. and Muller, W. E. *J. Med. Chem. 26* (1983) 499.

Received December 18, 1987.