Chemistry of Heterocyclic Compounds, Vol. 32, No. 1, 1996

PORPHYRINS.

32.* MASS SPECTROMETRIC STUDY OF vic-DIHYDROXY- AND vic,vic-TETRAHYDROXYCHLORINS

A. S. Moskovkin and G. V. Ponomarev

A mass spectrometric investigation has been carried out on dihydroxy- and tetrahydroxychlorins, derivatives of vic-diols of etioporphyrin-I and octaethylporphyrin, the vic-diol and vic,vic-tetraol of the tetraethyl ester of coproporphyrin-I, dimers of bischlorins linked together with a methylene bridge, and also of their trimethylsilylated products. It was established that in the spectra of porphyrin diols the molecular ion peaks were either of low intensity or were completely absent. In the spectra of trimethylsilyl derivatives, the ion peaks of maximum intensity were caused by the silylation of only one of each two neighboring hydroxyl groups in the initial compounds.

In recent years more publications have appeared on the prospects of using derivatives of the chlorin series compared to porphyrins for the photodynamic therapy of cancer [2]. One of the convenient methods of obtaining chlorins is the synthesis of vic-dihydroxychlorins by the oxidation of the corresponding porphyrins with osmium tetroxide [3].

The aim of the present work was to study the mass spectrometric behavior of a series of hydroxy substituted porphyrins synthesized by us previously [4, 5].

It is known that the mass spectrometric identification of di- and polyhydroxy substituted compounds encounters some difficulty due to the inclination of their molecular ions M^+ to dehydrate [6]. Characteristics of these compounds are a low intensity for M^+ ion peaks and a high, often maximal, intensity for the $[M-H_2O]^+$ ion peaks. Study of the mass spectra of porphyrin diols and tetraols (di- and tetrahydroxychlorins) is thus of significant interest.

On the one hand a distinctive feature of the majority of porphyrins is the presence in their mass spectra of intense M^+ peaks and a small number of less intense peaks of fragment ions. On the other hand at the temperature which is needed to evaporate the sample in the ion source (200° and greater) thermal decomposition of the initial hydroxy compounds is possible in addition to the mass spectrometric dehydration of M^+ ions. To protect the hydroxyl groups they are usually converted into O-acetyl or O-trimethylsilyl derivatives [7], which enables the molecular ion of the initial compound to be determined from the characteristic shift of masses.

The mass spectrometric behavior under the action of electron impact has been studied in the present work for the dihydroxy substituted etioporphyrin I (Ia), octaethylporphyrin (IIa), the tetraethyl ester of coproporphyrin I (IIIa), the triethyl isopropyl ester (IVa), tetrahydroxycoproporphyrin I (Va), 2-hydroxy-3,3,7,7,8,8,12,13,17,18-octaethylchlorin (VI), the octaethylporphyrin ketone (VII), and its Pd and Pt complexes (VIII) and (IX), and also the chlorin dimers (X) and (XI).

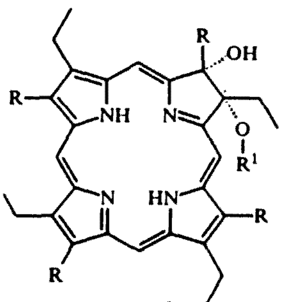
The corresponding trimethylsilyl derivatives of (Ib)-(Vb) were obtained by the reaction of chlorins (Ia)-(Va) with bis(trimethylsilyl)trifluoroacetamide [8].

The intensity of molecular ion peaks in the mass spectra of hydroxychlorins (Ia), (IIa), and (Va) did not exceed 10% and they were completely absent from the spectra of compounds (IIIa) and (IVa) (see Table 1). Dehydration was the main fragmentation process of the M^+ ions of compounds (Ia)-(Va) leading to the appearance in their spectra of $[M-H_2O]^+$ ion peaks

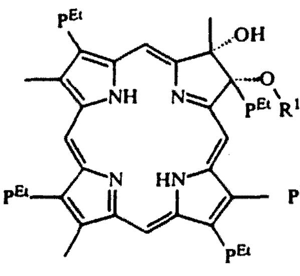
---

*For Part 31 see [1].

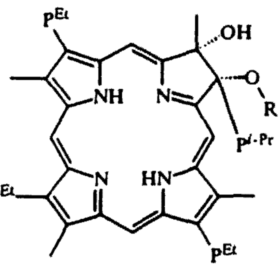
All-Russian Center of Catastrophy Medicine "Protection," Russian Ministry of Public Health and the Pharmaceutical Industry, Moscow 123182. Institute of Biological and Medicinal Chemistry of the Russian Academy of Medical Sciences, Moscow 119832. Translated from Khimiya Geterotsiklicheskikh Soedinenii, No. 1, pp. 43-48, January, 1996. Original article submitted December 13, 1995.



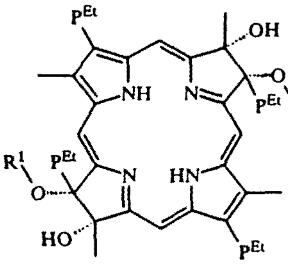
I–II a,b



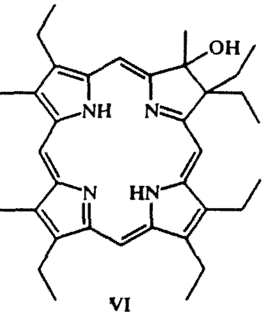
III a,b



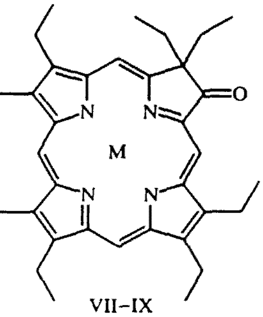
IV a,b



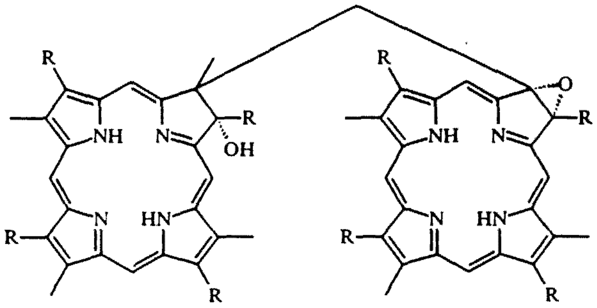
V a,b



VI



VII–IX



X, XI

Ia,b, XI, R = Me; IIa,bR = Et; XI, R = P^{Et}; Ia—Va, R^1 = H; Ib—VbR^1 = $SiMe_3$; VII, M = 2H; VIII, M = Pd; IX, M = Pt; where P^{Et} = $CH_2CH_2COOC_2H_5$; $P^{i\text{-}Pr}$ = $CH_2CH_2COOCH(CH_3)_2$

maximal in intensity (fragment Φ_1). Fission of a water molecule from the molecular ion M^+ probably occurs with the participation of a hydrogen atom from the neighboring hydroxyl group as a result of which the Φ_1 fragment has the structure of the corresponding oxirane. For compound (Va), having four hydroxyl groups disposed in pairs on opposite pyrrole rings, fission of only one water molecule also occurs with the formation of a peak for $[M-H_2O]^{+\cdot}$ probably because the oxirane Φ_1 fragment formed is sufficiently stable in spite of the presence of the remaining two vicinal hydroxyl groups. As is known [4] the tetraol (Va) may exist as two isomers in which the vicinal hydroxyl groups may be on one side (as a so-called cis isomer) or on different sides of the macrocycle (as the so-called trans isomer). However the individual cis-(Va) and trans-(Va) have completely identical mass spectra.

The further decomposition of Φ_1 fragments is linked with the removal of methyl and ethyl radicals typical of derivatives of etioporphyrin and octaethylporphyrin [9]. Stepwise decomposition of the ester groups occurred in the case of the coproporphyrin derivatives.

TABLE 1. Characteristic Ion Peaks and Their Intensities in the Mass Spectra of Compounds (Ia, b)-(Va, b)

Fragment	Value of *m/z* (intensity, %)				
	Ia	IIa	IIIa	IVa	Va
M^+	512 (6)	568 (5)	800 (–)	814 (—)	834 (10)
$[M-CH_3]^+$	—	—	—	—	—
$[M-C_2H_5]^+$	—	—	—	—	—
$[M-OC_2H_5]^+$	—	—	—	—	—
$[M-CH(CH_3)_2]^+$	—	—	—	—	—
Φ_1*	494 (100)	550 (100)	782 (100)	796 (100)	816 (100)
$[\Phi_1-H_2O]^+$	—	—	—	—	798 (27)
$[\Phi_1-HOSi(CH_3)_3]^+$	—	—	—	—	—
$[\Phi_1-CH_3]^+$	479 (9)	535 (6)	—	—	—
$[\Phi_1-C_2H_5]^+$	465 (18)	521 (7)	—	—	—
$[\Phi_1-OC_2H_5]^+$	—	—	737 (6)	751 (5)	771 (3)
$[\Phi_1-OCH(CH_3)_2]^+$	—	—	—	737 (3)	—
$[\Phi_1-CO_2-C_2H_4]^+$	—	—	710 (9)	724 (8)	—
$[\Phi_1-CH_2CO_2C_2H_5]^+$	—	—	695 (7)	709 (5)	—
$[\Phi_1-(CH_2)_2CO_2C_2H_5]^+$	—	—	691 (8)	681† (9)	715 (6)
M^{2+}	—	—	—	—	—
Φ_1^{2+}	247 (4)	275 (5)	391 (2)	398 (1)	408 (1)
Fragment	Ib	IIb	IIIb	IVb	Vb
M^+	584 (100)	640 (100)	872 (100)	886 (100)	978 (100)
$[M-CH_3]^+$	569 (4)	625 (2)	857 (5)	871 (3)	963 (3)
$[M-C_2H_5]^+$	555 (5)	611 (2)	—	—	—
$[M-OC_2H_5]^+$	—	—	827 (7)	841 (4)	933 (3)
$[M-CH(CH_3)_2]^+$	—	—	—	827 (2)	—
Φ_1*	494 (25)	550 (20)	782 (74)	796 (35)	888 (55)
$[\Phi_1-H_2O]^+$	—	—	—	—	—
$[\Phi_1-HOSi(CH_3)_3]^+$	—	—	—	—	798 (5)
$[\Phi_1-CH_3]^+$	—	—	479 (6)	535 (4)	—
$[\Phi_1-C_2H_5]^+$	465 (12)	521 (8)	—	—	—
$[\Phi_1-OC_2H_5]^+$	—	—	737 (7)	751 (2)	843 (3)
$[\Phi_1-OCH(CH_3)_2]^+$	—	—	—	737 (2)	—
$[\Phi_1-CO_2-C_2H_4]^+$	—	—	710 (8)	724 (2)	816 (1)
$[\Phi_1-CH_2CO_2C_2H_5]^+$	—	—	695 (6)	709 (2)	801 (1)
$[\Phi_1-(CH_2)_2CO_2C_2H_5]^+$	—	—	681 (8)	681† (6)	787 (5)
M^{2+}	292 (1)	320 (3)	436 (2)	443 (2)	489 (3)
Φ_1^{2+}	247 (—)	275 (2)	391 (2)	398 (1)	444 (2)

---

*The fragment Φ_1 corresponds to the $[M-H_2O]^+$ ion in the spectra of compounds (Ia)-(Va) and to the $[M-HOSi(CH_3)_3]^+$ ion in the spectra of compounds (Ib)-(Vb).

†Fragment $[\Phi_1\text{-}CH_2CH_2CO_2CH(CH_3)_2]^+$.

The mass spectra of the trimethylsilyl derivatives of (Ib)-(Vb) contain peaks maximal in intensity shifted from the M^+ ions by 72 for compounds (Ia)-(IVa) [chlorins (Ib)-(IVb)] and by 144 atomic mass units [compound (Vb)] (Table 1). The mass shift indicates that probably only one of the vicinal hydroxyl groups was subjected to trimethylsilylation due to steric hindrance.

Decomposition of the M^+ ions of compounds (Ib)-(Vb) occurs predominantly with the formation of Φ_1 fragments second in intensity in the mass spectra. The remaining peaks in the spectra of compounds (Ib)-(Vb) are caused by the fission of methyl, ethyl, and ester groups from the M^+ and Φ_1 ions.

Peaks for doubly charged Φ_1^{2+} ions were present in the spectra of all the compounds (Ia, b)-(Va, b) and doubly charged molecular ions M^{2+} were present in the spectra of compounds (Ib)-(Vb).

It is interesting that the monohydroxychlorin (VI) was significantly more stable than the vic-dihydroxychlorins. Dehydration of its molecular ion occurs to an insignificant extent and the intensity of the $[M-H_2O]^+$ ion peak in the spectrum did not exceed 2%. This is probably linked with the impossibility of fission of a water molecule with the formation of the corresponding oxirane without preliminary rearrangement of the molecular ion.

TABLE 2. Characteristic Ion Peaks and Their Intensities in the Mass Spectra of Compounds (VI)-(IX)*

Fragment	Value of m/z (intensity, %)			
	VI	VII	VIII	IX
M^+	552 (100)	550 (100)	654 (100)	742 (70)†
$[M-CH_3]^+$	537 (4)	535 (5)	639 (2)	727 (2)
$[M-H_2O]^+$	534 (2)	—	—	—
$[M-C_2H_5]^+$	523 (15)	521 (14)	625 (12)	713 (10)
$[M-C_2H_5-CH_3]^+$	508 (1)	506 (6)	610 (6)	698 (4)
$[M-C_2H_5-2CH_3]^+$	493 (3)	491 (4)	595 (8)	683 (6)
$[M-2C_2H_5-CH_3]^+$	479 (3)	477 (1)	581 (2)	669 (4)
M^{2+}	276 (9)	275 (2)	327 (11)	371 (7)

---

[*] In the spectra of compounds (VIII) and (IX) values are given for the ^{106}Pd and ^{194}Pt isotopes respectively.

[†] m/z 743 (100).

TABLE 3. Characteristic Ion Peaks and Their Intensities in the Mass Spectra of Compounds (X) and (XI)

Fragment	Value of m/z (intensity, %)		Fragment	Value of m/z (intensity, %)	
	X	XI		X	XI
M^+	988 (78)	1564 (8)	$[\Phi_3-CH_3]^+$	463 (24)	—
$[M-CH_3]^+$	975 (0,3)	—	$[\Phi_3-C_2H_5]^+$	449 (5)	—
$[M-H_2O]^+$	970 (5)	1546 (0,2)	$[\Phi_3-OC_2H_5]^+$	—	721 (6)
$[M-C_2H_5]^+$	959 (0,2)	—	$[\Phi_3-COOC_2H_5]^+$	—	693 (22)
$[M-OC_2H_5]^+$	—	1519 (3)	$[\Phi_3-CH_2COOC_2H_5]^+$	—	679 (16)
$[M-COOC_2H_5]^+$	—	1491 (0,6)	Φ_2^+ *	247 (5)	391 (0,2)
Φ_2^+ *	494 (55)	782 (11)	Φ_3^+ †	239(19)	383 (3)
Φ_3^+ †	478 (100)	766 (100)			

---

[*] Φ_2^+ is the fragment corresponding to the ion of the epoxyporphyrin monomer.

[†] φ_3^+ is the fragment corresponding to the ion of the porphyrin monomer.

The porphyrin ketones (VII)-(IX) occupy a special position. In spite of their possible formal assignment as chlorin derivatives these are some of the most stable tetrapyrrole compounds. The molecular ion peaks have maximum intensity in their mass spectra (Table 2) and the main fragmentation processes of the M^+ ions are linked with the sequential fission of methyl and ethyl groups [9].

The porphyrin ketones (VII)-(IX) occupy a special position. In spite of their possible formal assignment as chlorin derivatives these are some of the most stable tetrapyrrole compounds. The molecular ion peaks have maximum intensity in their mass spectra (Table 2) and the main fragmentation processes of the M^+ ions are linked with the sequential fission of methyl and ethyl groups [9].

A characteristic feature of chlorin diols containing a geminal methyl group together with a hydroxy group, such as the derivatives of etioporphyrin I and coproporphyrin I, is their inclination towards cationic dimerization in the presence of zinc acetate [5]. We have synthesized two dimers (X) and (XI) in the mass spectra of which fragments were observed characteristic of each of the macrocycles in addition to fairly intense peaks of the molecular ions (Table 3). As in the case of compound (VI), dehydration of the M^+ ions of the dimers (X) and (XI) occurred to an insignificant extent. The formation of fragments Φ_3 and Φ_2, having the structures of the porphyrin monomer and its oxirane analog respectively, probably occurs as a result of both mass spectrometric fragmentation of the M^+ ions and by the possible thermal decomposition of the initial dimers.

It has been established from a mass spectrometric study of the di- and tetrahydroxy substituted chlorins (Ia)-(Va) that it is necessary to obtain their trimethylsilyl derivatives (Ib)-(Vb), in which trimethylsilylation occurs only at one of the two neighboring hydroxyl groups, in order to identify these compounds reliably. On the other hand, preliminary hydroxyl group protection is not required to identify the monohydroxyoctaethylchlorin (VI). Reliable interpretation of mass spectra without previous trimethylsilylation is also possible for chlorin dimers containing an oxirane grouping in one of the macrocycles and a hydroxyl group in the other.

EXPERIMENTAL

The synthesis of dihydroxychlorins was effected according to [3], of the tetrahydroxychlorin as in [4], chlorin dimers [5], chlorin (IVa) by heating chlorin (IIIa) in isopropyl alcohol in the presence of anhydrous sodium acetate, chlorin (VI) by the reduction of porphyrin ketones (VII) with $NaBH_4$ in a mixture of chloroform and methanol, and complexes (VIII) and (IX) by heating porphyrin ketone (VII) with $PdCl_2$ in DMF for 30 min at 105°C and with $PtCl_2$ in benzonitrile for 2 h at 215-220°C respectively.

Mass spectra were obtained on a Finnigan MAT 90 instrument by direct insertion of samples into the ion source with an ionizing energy for electrons of 70 eV. Evaporator temperatures were 190-210°C for compounds (I)-(VI), and 300-330°C for porphyrinketones (VII)-(IX) and dimers (X) and (XI).

REFERENCES

1. D. V. Yashunskii, A. S. Moskovkin, and G. V. Ponomarev, Khim. Geterotsikl. Soedin., No. 7, 906 (1995).
2. R. K. Pandey, F.-Y. Shiau, M. Isaac, S. Ramaprasad, T. J. Dougherty, and K. M. Smith, Tetrahedron Lett., **33**, 7818 (1992).
3. C. K. Chang and C. C. Sotiriou, J. Org. Chem., **52**, 926 (1987).
4. G. V. Ponomarev and A. M. Shul'ga, Abstr. 15th Intern. Symposium on Macrocyclic Chemistry, Odessa (1990) p. 76.
5. G. V. Ponomarev and A. M. Shul'ga, Khim. Geterotsikl. Soedin., No. 1, 126 (1992).
6. N. S. Vul'fson, V. G. Zaikin, and A. I. Mikaya, Mass Spectrometry of Organic Compounds [in Russian], Khimiya, Moscow (1986), p. 169.
7. P. B. Terent'ev, Mass Spectrometry in Organic Chemistry [in Russian], Vysshaya Shkola, Moscow (1979), p. 171.
8. M. C. Dutt, J. Chromatog., **248**, 115 (1982).
9. D. Dolphin (ed.), The Porphyrins, Vol. 3, Academic Press, New York (1978), p. 395.