Carbohydrate Research, 70 (1979) 239–246

ELECTRON-IMPACT MASS SPECTROMETRY OF METHYL *O*-METHYL-GLUCOPYRANOSIDURONAMIDES*†

VINCENT MIHÁLOV, VLADIMÍR KOVÁČIK, AND PAVOL KOVÁČ
Institute of Chemistry, Slovak Academy of Sciences, 809 33 Bratislava (Czechoslovakia)
(Received April 26th, 1978; accepted for publication in revised form, July 3rd, 1978)

ABSTRACT

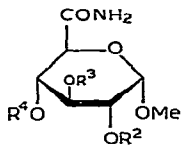
Mass-spectral fragmentation of the complete series of methyl *O*-methyl-α-D-glucopyranosiduronamides has been studied. Based on 70- and 12-eV spectra, deuteration (OD and ND_2) experiments, and metastable-transition and exact-mass measurements, new cleavage-reactions resulting from the simultaneous presence of hydroxyl and amido groups in the molecule have been found. The mass spectra provide information useful in the determination of number and locations of methyl groups in methyl *O*-methylhexopyranosiduronamides.

INTRODUCTION

Systematic study of uronic acid derivatives by mass spectrometry has shown that the fragmentation[2] of methyl (methyl 2,3,4-tri-*O*-methyl-α-D-glucopyranosid)-uronate does not differ qualitatively from that[3–6] of permethylated methylhexopyranosides. On the other hand, quantitative differences, attributed to the presence of the electron-withdrawing methoxycarbonyl group, were evident. The presence of hydroxyl groups in methyl (methyl *O*-methylhexopyranosid)uronates drastically alters the mode of fragmentation[7,8] of these compounds, giving rise to new fragmentation-series not observed for the fully methylated compounds. Fragmentation[9] of methyl 2,3,4-tri-*O*-methyl-α-D-glucopyranosiduronamide has also been found to proceed through a different, overall fragmentation-scheme. In the series of glycuronamides **1–8**, both hydroxyl and amido groups are present. Evaluation of the effect of interactions between these two types of functional group upon the mass-spectral fragmentation of the pyranoid ring, and clarification of the fragmentation of this series of derivatives was the theoretical aim of the present work. Glycuronamides are readily obtainable compounds often used in the identification of partially methylated uronic acid derivatives[10]. To find means whereby mass spectrometry could be used in the identification of this class of substances was the analytical purpose of the work described herein.

---

*Dedicated to Professor Roy L. Whistler.
†Part XIV of the series: Mass Spectrometry of Uronic Acid Derivatives: for Part XIII, see ref. 1.



1 – 8

	R^2	R^3	R^4
1	H	H	H
2	Me	H	H
3	H	Me	H
4	H	H	Me
5	Me	Me	H
6	Me	H	Me
7	H	Me	Me
8	Me	Me	Me

RESULTS AND DISCUSSION

The mass spectra of **1–8** obtained at 12 eV, simpler to some extent than those measured at 70 eV, are shown in Table I. Only a few of the compounds in this study produced weak molecular ions (appearing in the spectra at *m/e* 207, 221, and 235, according to the number of methyl groups present). Of the decompositions common in the fragmentation of permethylated hexopyranosides[3–6] and hexuronic acids[2], only fissions following Series A, F, H, and J (Table II) occur. Serial eliminations of methanol, observed[9] in the fragmentation of methyl 2,3,4-tri-*O*-methyl-α-D-glucopyranosiduronamide, compete with eliminations of water. The $[M - ROH]^{+\cdot}$ ions thus produced decompose by further elimination of water or methanol to afford the ions $[M - 2ROH]^{+\cdot}$. The previously observed[9] elimination of methyl formate from the $[M - ROH]^{+\cdot}$ ions also occurs. These characteristic features were confirmed by analyzing the spectra of *O*- and *N*-deuterated analogues and by measurement of metastable transitions. The absence of ion peaks of the pyranoid E_1 series confirms that the C-5–$CONH_2$ bond is more resistant to homolytic cleavage than either the C-5–CO_2OMe or the C-5–CH_2OMe bond.

O- And *N*-deuteration of the 2,4-di-*O*-methyl derivative **6** showed that the hydrogen atom of the amido group also participates in the elimination of water from the molecular ions (Scheme 1).

As may be seen for methyl *O*-methylhexopyranosides[11] and the corresponding methyl glycuronates[7] bearing a free hydroxyl group at C-2 or C-4, cations having the elemental compositions $C_7H_{13}O_4$, $C_6H_{11}O_4$, and $C_5H_9O_4$ (*m/e* 161, 147, and 133, shifting to *m/e* 161, 148, and 135, respectively after deuteration) are formed from the amides **1–5** and **7** (Table II). Metastable-transition measurements proved that, in cases where these ions are formed from the HO-4 compounds **1–3** and **5**, their decomposition is accompanied by liberation of carbon monoxide to give ions appearing at *m/e* 133, 119, and 105, respectively (Scheme 2). The ease of liberation of carbon monoxide from the ions of *m/e* 161, 147, and 133 formed from **1–3** and **5** supports the previous assumption[11] that these ions possess a 5-membered ring structure of the E_1 type (Scheme 2). On the other hand, fragmentation of compounds **4** and **7** is not accompanied by the elimination of carbon monoxide and, hence, the peaks at *m/e* 147 and 161 represent ions, formed after a transfer of a hydrogen atom

TABLE I

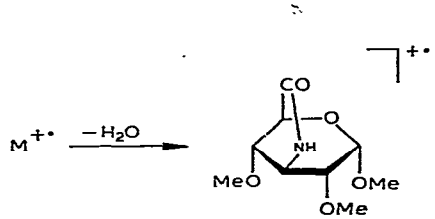
MASS-SPECTRAL DATA[a] FOR METHYL *O*-METHYL-α-D-GLUCOPYRANOSIDURONAMIDES **1–8**

m/e	% Σ_{45} × 100							
	—	2[b]	3	4	2,3	2,4	3,4	2,3,4
235					35	16	31	
222			11					
221			29					
218								25
217						48	25	116
207	36							
205					30		37	
204					64	48	48	
203		51			123	136	75	
202							35	
191			26	27		32	33	
190		56	50	119				
189	12	102	38	41				
188				68				
186						160	37	150
185							62	66
176	58			61				
175	40							
174							50	
173							37	
172		127	23	41	227		149	
171		130	14	146	94			
161		45			617		109	
160	54			87		64	56	
159			65	132		56	188	
158	153		131			48		
157	102		83			155		258
149	43		25		324		62	
148			47	251				
147		66	688	54				
146		38						
145	36	38	41	114				
144		40			58	37	67	
143		112		96	74	53	251	
141							37	
133	1529				1006			
131	95				32			
130	51	45	133	96	58		839	
129	95	102	83	105	94	50	79	
128				36				
127		45	23	70	42	176	113	
126		38		178	32	42	161	342
125		35						
119		357	1376					
118			98	320				
117	139	1351	1645	274	1461	53	377	
116	183	91	568	93			62	

TABLE I *(continued)*

m/e	% Σ_{45} × 100							
	—	2[b]	3	4	2,3	2,4	3,4	2,3,4
115	161	56	179	174			138	
114		96		68	48	56	67	
113			44	595			100	
112	124	122	41	119	103	48	75	
105	549							
104	806	408	101					
103	513	196	65		56			
102		102	65	2060	123	2364	1259	300
101	58		822	137	942	3832	671	6265
100	80		59	146	422		62	
99				73	584	64	46	
98				82		58	62	
97				36			33	
96				132		64	33	
95		53						
89			239	70	292	152	251	
88	113	1581	329	128	747	482	293	1253
87	1063	2091	688	1328	211	91	251	
86	190		269	54				
85	168	122	269	251	110	235	608	
83		53						
75	2126	1071	658	870	1624	268	1931	1002
74	315	612	359	824	51	509	503	
73	296	224	26		105	321		217
72				201		117	251	
71	296	86	113		48		52	
69			44		29		37	
68			26				29	
61	95	40	32	45		26	37	
60	139	76	23	50	32	37	35	
59	95	40	55	41		26	33	
58		35	44	43			37	
57			41					
45	172	51	179	50	84	58	54	

[a]12 eV. [b]The numbers refer to the positions of the methyl groups.



m/e 235 (238) M −H_2O , *m/e* 217 (218)

Scheme 1

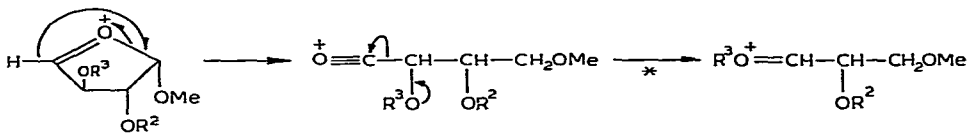
TABLE II

FEATURES CHARACTERISTIC OF THE FRAGMENTATION OF METHYL *O*-METHYL-α-D-GLUCOPYRANOSIDURONAMIDES **1–8**

Ions	m/e	% Σ_{45}[a]							
		—	*2*[b]	*3*	*4*	*2,3*	*2,4*	*3,4*	*2,3,4*
A_1	218								.
	204					..	.	.	
	190		..	.	...				
	176	..							
A_2	186						...		...
	172		...	.	.	...		...	
	158	...		...					
M — ROH	217						.	.	...
	203		..			...	...	..	
	189	.	...	.	.				
	175	.							
M — 2ROH	185							..	..
	171		...	.	...	..			
	157	...		..					
M — ROH — HCO_2Me	157						...	.	...
	143		...		..	..	.	...	
	129	..	...	..	...				
$C_7H_{13}O_4$	161		.			xx		x	
$C_6H_{11}O_4$	147		..	x	.				
$C_5H_9O_4$	133	xx							
$C_5H_{11}NO_4$	149	.		.		...		..	
$C_6H_{13}O_3$	133					xx			
$C_5H_{11}O_3$	119		...	xx					
$C_4H_9O_3$	105	xx							
$C_5H_9O_3$	117	...	xx	xx	...	xx	.	...	
$C_4H_7O_3$	103	x	...	..					
$C_3H_6NO_3$	104	x	...	...					
$C_4H_8NO_2$	102		...	..	xxx	...	xxx	xx	...
F_1	101	..		x	...	x	xxx	x	xxx
	87	xx	xxx	x	xx	...	..	...	
H_1	88		xx	...	...	x	...	...	xx
	74	...	x	...	x	..	...	...	
	60	...	..	.	.				
$J_1 + C_2H_5NO_2$	75	xxx	xx	x	x	xx	...	xx	x

[a]Peak intensities: ., < 0.5%; .., 0.5–1.0%; ..., 1–5%; x, 5–10%; xx, 10–20%; xxx, > 20%. [b]The numbers refer to the position of the methyl groups.

from the hydroxyl group on C-2, having acyclic and not furanoid structures. The ions $[C_5H_{11}NO_4]^{\ddagger}$, appearing at *m/e* 149 (151), are analogous to the $[C_6H_{12}O_5]^{\ddagger}$ ions formed[7] from methyl(methyl O-methylhexopyranosid)uronates bearing a free hydroxyl group at C-4. The formation of $C_5H_9O_3]^+$ and $[C_4H_7O_3]^+$ ions appearing at *m/e* 117 and 103, characteristic of the fragmentation of methyl (methyl *O*-methylhexopyranosid)uronates[7], also occurs for the amides. Exact-mass measurements of



R^2	R^3				
Me	Me	*m/e*	161 (161)	$C_6H_{13}O_3$ *m/e*	133 (133)
H	Me	*m/e*	147 (148)	$C_5H_{11}O_3$ *m/e*	119 (120)
H	H	*m/e*	133 (135)	$C_4H_7O_3$ *m/e*	105 (107)

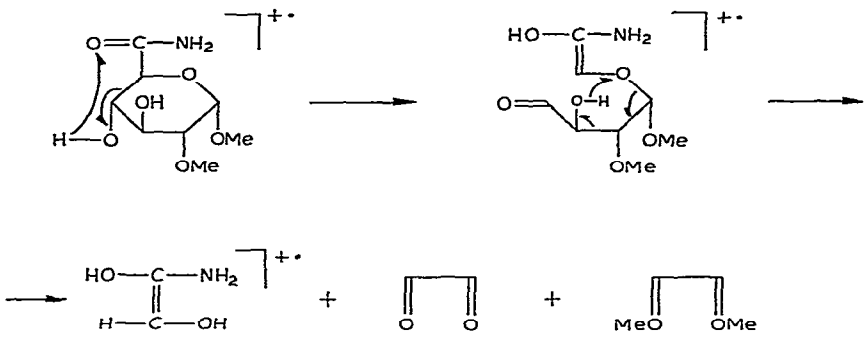
Scheme 2

the ions having *m/e* 117 formed from the 2,3-dimethyl ether **5** showed that this peak signaling their formation is a doublet consisting of $[C_5H_9O_3]^+$ and $[C_4H_7NO_3]^{+\cdot}$ ions in the ratio of 3:2. This conclusion was confirmed by deuteration, which caused a partial shift of the peak at *m/e* 117 to *m/e* 119. The ions $[C_4H_7NO_3]^{+\cdot}$ are analogous to the ions $[C_5H_8O_4]^{+\cdot}$ having *m/e* 132 formed in the fragmentation of methyl (methyl 2,3-di-*O*-methyl-α-D-glucopyranosid)uronate[7].

$C_4H_8NO_2$ *m/e* 102 (105) $C_3H_6NO_3$ *m/e* 104 (108)

Electron-impact on compounds **4**, **6**, and **7**, which bear a methoxyl group at C-4, results in the formation of an intense $[C_4H_8NO_2]^+$ ion appearing at *m/e* 102 (*m/e* 105 after deuteration). These ions are formed by transfer of a hydrogen atom from HO-2 or HO-3 to the carbonyl group, followed by conjugated transfer of electrons along the 6-membered ring. The ions $[C_3H_6NO_3]^+$ (*m/e* 104) are formed from the unsubstituted glycoside **1**. The structure shown was deduced on the basis of the observed elemental composition and, after deuteration, the observed shift by four units to higher *m/e* values.

Exact-mass measurements showed that the peak at *m/e* 75 in the spectra of **1** and **2**–**4** is a doublet. Whereas the $[C_3H_7O_2]^+$ particles constitute J_1 ions, the formation of the ions $[C_2H_5NO_2]^+$ has no analogy in any carbohydrate derivatives studied thus far. The *m/e* value at which J_1 ions appear remains unchanged on attempted deuteration, whereas peaks attributed to the ions $[C_2H_5NO_2]^{+\cdot}$ are shifted to *m/e* 79. The relative contributions of the two types of ion are shown in Table III. The formation of the $[C_2H_5NO_2]^{+\cdot}$ ions may be rationalized by a double rearrangement of hydrogen atoms of the hydroxyl groups present in the molecule. The hydrogen atom subsequently migrates to the carbonyl and ring-oxygen atoms. Consequently, the bond is cleaved to give, finally, the conjugated $[C_2H_5NO_2]^{+\cdot}$ ions



$C_2H_5NO_2$ *m/e* 75 (79)

Scheme 3

TABLE III

THE RELATIVE CONTRIBUTIONS OF THE $[C_2H_5NO_2]^+$ AND $[C_3H_7O_2]^+$ IONS TO THE INTENSITY OF THE PEAK AT *m/e* 75

Compound	*Percent*	
	$C_2H_5NO_2$	$C_3H_7O_2$
1	71	29
2	89	11
3	38	62
4	43	57

as shown in Scheme 3 for their formation from the 2-*O*-methyl derivative **2**. The fact that the peak at *m/e* 75 comprises two ion-species also explains the high intensity of these peaks in the spectra of the derivatives **1**, **2**, and **4**, not containing a methoxyl group at C-3.

The mass spectra of **1–8** (Table I) show characteristic differences (Table II) that may be used for unambiguous determination of both number and positions of methyl groups in methyl *O*-methylhexopyranosiduronamides.

EXPERIMENTAL

Compounds **1–8** were prepared[10] by ammonolysis of the corresponding methyl (methyl *O*-methyl-α-D-glucopyranosid)uronates. For deuteration, the compounds were dissolved in 10:1 CH_3OD–D_2O, and the solvents were removed directly in the mass spectrometer. The degree of deuteration achieved was 53–85%. Mass spectra were obtained at 70 and 12 eV and an emission of 100 μA by using a JMS 100 D instrument. The temperature at the site of evaporation (200–245°) was measured

with an MS-DPT-01 direct-probe, temperature-control unit, and that in the ionizing chamber was 180°. Exact-mass measurements were performed at a resolution of 10,000. Metastable transitions were measured with an MS-MT-01 metastable-ion detector.

ACKNOWLEDGMENT

The authors are grateful to Mr. J. Mrva for skilful technical assistance.

REFERENCES

1 V. MIHÁLOV, V. KOVÁČIK, AND P. KOVÁČ, *Chem. Zvesti*, in press.
2 V. KOVÁČIK AND P. KOVÁČ, *Org. Mass Spectrom.*, 9 (1974) 172–180.
3 N. K. KOCHETKOV, N. S. WULFSON, O. S. CHIZHOV, AND B. M. ZOLOTAREV, *Tetrahedron*, 19 (1963) 2209–2224.
4 N. K. KOCHETKOV AND O. S. CHIZHOV, *Tetrahedron*, 21 (1965) 2029–2047.
5 K. HEYNS AND D. MÜLLER, *Tetrahedron*, 21 (1965) 55–68.
6 K. HEYNS AND H. SCHARMANN, *Tetrahedron*, 21 (1965) 507–513.
7 V. KOVÁČIK AND P. KOVÁČ, *Org. Mass Spectrom.*, 10 (1975) 376–383.
8 V. KOVÁČIK, V. MIHÁLOV, AND P. KOVÁČ, *Chem. Zvesti*, 32 (1978) 258–266.
9 V. KOVÁČIK AND P. KOVÁČ, *Carbohydr. Res.*, 38 (1974) 25–34.
10 P. KOVÁČ, *Carbohydr. Res.*, 31 (1973) 323–330.
11 K. HEYNS, K. R. SPERLING, AND H. F. GRÜTZMACHER, *Carbohydr. Res.*, 9 (1969) 79–97.