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Thiol Compounds. VI.[1] Mass Spectra of (2*R*,4*R*)-2-(2-Hydroxyphenyl)-3-(3-mercaptopropionyl)-4-thiazolidinecarboxylic Acid and Related Compounds

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The electron impact mass spectra of (2*R*,4*R*)-2-(2-hydroxyphenyl)-3-(3-mercaptopropionyl)-4-thiazolidinecarboxylic acid (**1**), SA 446, and the related compounds **2**—**7** are discussed. The fragmentation patterns were classified into three types of characteristic ring fragmentation (1,3-, 2,5- and 3,5-cleavages) with or without substituent cleavage. The fragment ions were established by the shifting method with the labelled derivatives **4′** and **4″** and the elemental compositions were confirmed by high-resolution mass spectrometry.

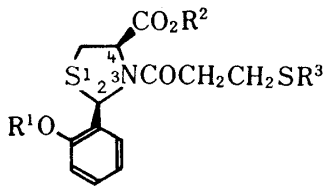
Keywords——(2*R*,4*R*)-2-(2-hydroxyphenyl)-3-(3-mercaptopropionyl)-4-thiazolidinecarboxylic acid; thiazolidine; thiol; electron impact mass spectrometry; shifting method

We have studied the syntheses and structure-activity relationships of 2-aryl-3-mercaptoacyl-4-thiazolidinecarboxylic acids having inhibitory activity against angiotensin I-converting enzyme.[1c] (2*R*,4*R*)-2-(2-Hydroxyphenyl)-3-(3-mercaptopropionyl)-4-thiazolidinecarboxylic acid (**1**), SA 446, was found to be the most potent among them.

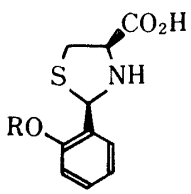
During development of the synthetic procedures for thiazolidines, mass spectrometry was of use for structure elucidation. Further, the mass spectra of the thiazolidines and the metabolites showed characteristic fragmentations. 3-Acyl-2-aryl-4-thiazolidinecarboxylic acids have not previously been studied with respect to mass spectral behavior.

In the present study, the electron impact mass spectra of **1** and **2**—**4** as methyl derivatives of **1** in Chart 1 are reported as part of a preliminary study of the metabolites, because **1** possesses three hydrophilic functional groups (SH, OH and CO_2H) in the molecule and the thiol group(s) of *N*-(2-mercapto-2-methylpropanoyl)-L-cysteine (SA 96)[2] is biologically methylated.[3]

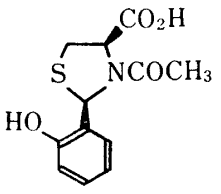
The absolute configurations of **2**, **3** and **4** were established to be (2*R*,4*R*) by ^{1}H-nuclear magnetic resonance (^{1}H-NMR) spectroscopy in the same manner as for **1**.[1b]



1: $R^1=R^2=R^3=H$
2: $R^1=R^2=H$, $R^3=CH_3$
3: $R^2=H$, $R^1=R^3=CH_3$
4: $R^1=R^2=R^3=CH_3$
4′: $R^1=CH_3$, $R^2=R^3=CD_3$
4″: $R^1=R^3=CH_3$, $R^2=CD_3$



5: R=H
6: $R=CH_3$



7

Chart 1

In addition, the mass spectra of **5**—**7** are discussed. Their fragment ions were established by the shifting method with the labelled derivatives **4'** and **4''** and by high-resolution mass spectrometry.

Results and Discussion

All of the thiazolidines **1**—**7** gave molecular ions in the electron impact mass spectra. These ions are consistent with the structural assignment in Table I. However, the relative abundances of the molecular ions were low except in the case of **5**.[4] They were established as molecular ions (MH^+) by chemical ionization mass spectrometry.

The mass spectra of typical compounds are shown in Fig. 1.

Table I. Mass Spectral Data for Thiazolidines

Compd.	$[M]^{+\cdot}$	$m/z^{a,b}$ (% relative intensity)											
		a	**b**	**c**	**d**	**e**	**f**	**g**	**h**	**i**	**j**	**k**	**l**
1	313	120	—	137	241	153	132	224	—	208	180	61	—
	(4)	(25)		(60)	(10)	(12)	(42)	(22)		(26)	(8)	(100)	
2	327	120	—	137	255	153	132	224	283[a]	208	180[b]	61	75
	(2)	(20)		(50)	(5)	(30)	(29)	(22)	(6)	(12)	(42)	(100)	(54)
3	341[a]	134	236	151	269[b]	—	132	238[b]	—	222	194	61	75
	(7)	(54)	(88)	(50)	(38)		(18)	(98)		(72)	(24)	(100)	(55)
4	355[a]	134	236	151	269	—	146	252[b]	—	222	194	61	75
	(3)	(44)	(50)	(38)	(14)		(16)	(100)		(45)	(29)	(69)	(44)
5	225[a]	120	—	137	—	153[b]	132	—	—	—	—	—	—
	(48)	(62)		(46)		(30)	(100)						
6	239	134	—	151	—	167	132	—	—	—	—	—	—
	(28)	(30)		(6)		(6)	(26)						
7	267[a]	120	—	137	195[b]	153	132	224	—	208	180	—	—
	(27)	(35)		(100)	(68)	(54)	(52)	(26)		(8)	(18)		

a) and *b*) indicate precursor and daughter ions, respectively, in metastable transition.
Remaining peaks higher than 20 % are as follows:
1: 51(22), 55 (75), 57(34), 69(28), 74(26), 76(30), 77(28), 89(22),106(30), 138(25), 176(30), 178(22).
2: 51(23), 55(25), 65(22), 77(30), 107 (36), 122(44), 138(24).
3: 55(38), 59(22), 77(24), 91(48), 101(55), 103 (24), 107(28), 119(38), 121(24), 152(28), 220(43).
4: 55(22), 59(22), 91(26).
5: 51(60), 52(25), 59(27), 63(20), 65(39), 77(70), 91(42), 102 (22), 105(28), 107(40), 118(40), 119(25), 121(40), 133(64), 134(34), 146 (52), 147(52), 148(20), 179(30), 192(20).
6: 51(30), 65(23), 77(35), 91 (100), 107(20), 118(22), 119(60), 121(22), 147(23), 161(21).
7: 51(30), 59(34), 61(31), 65(28), 77(37), 78(22), 86(24), 88(24), 91(20), 121(32), 122(28), 133(35), 138(30), 178(26).

The fragmentation patterns in the electron impact mass spectra were classified into ring fragmentations and substituents cleavage.

We identified three types of characteristic ring fragmentation pathways (1,3-, 2,5- and 3,5-cleavages) in all of the thiazolidines (**1**—**7**). However, 1,4-cleavage as reported for 2-phenyl-4-thiazolidinecarboxylic acid[5] was observed only in **5**. Cleavages of the substituents were also observed with or without ring fragmentation. The elemental compositions of the fragment ions were established by high-resolution mass spectrometry (Table II).

Ions a and b in Chart 2 (1,3-Cleavage)

The ion **a** in **1**—**7** appeared at m/z 120 or 134, the difference of 14 *u* depending on the nature of R^1, as shown in Chart 2. The relative abundances of the peak at m/z 134 in the spectra of **3** and **4** were higher than those of the peak at m/z 120 in **1** and **2**.

Vestling *et al.*[5] observed ions in which the sulfur-containing fragment retains the charge in 2-phenyl-4-thiazolidinecarboxylic acid. However, we merely observed a minor peak at m/z 104 in **5**, and ion **a** retaining a positive charge at the nitrogen atom was observed as a

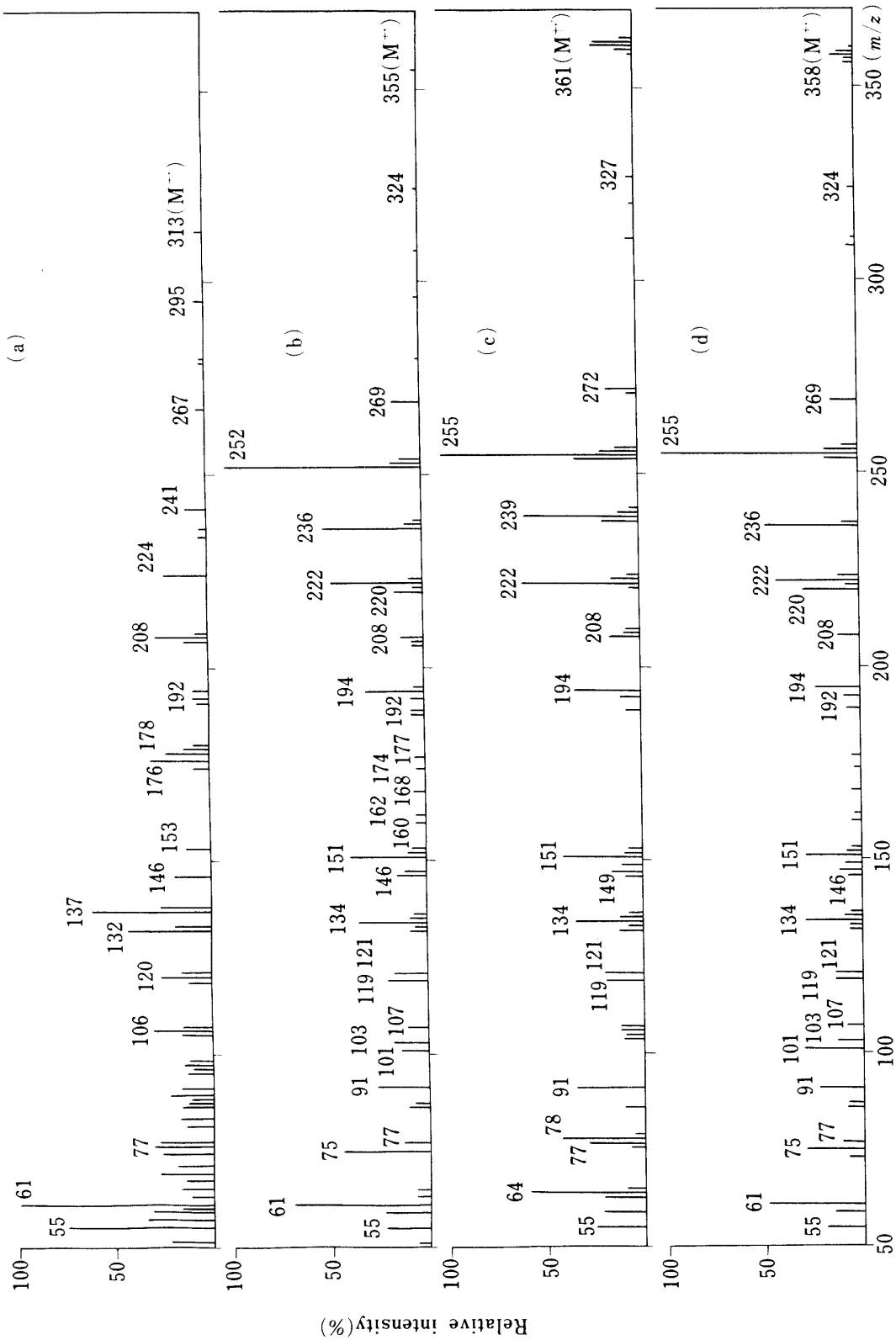


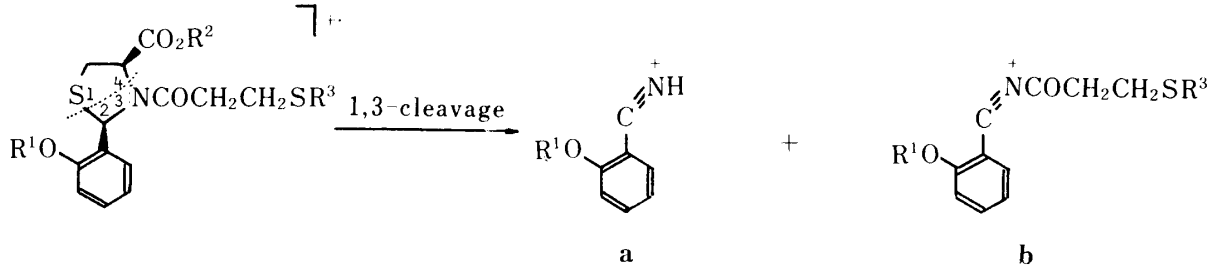
Fig. 1. Mass Spectra of Thiazolidines
(a), **1**; (b), **4**; (c), **4′**; (d), **4″**.

strong peak. The difference of behavior may be due to the presence of the *N*-acyl group in our compounds.

Ion **b** is shown in Chart 4.

Table II. High-Resolution Mass Spectral Data for Thiazolidines

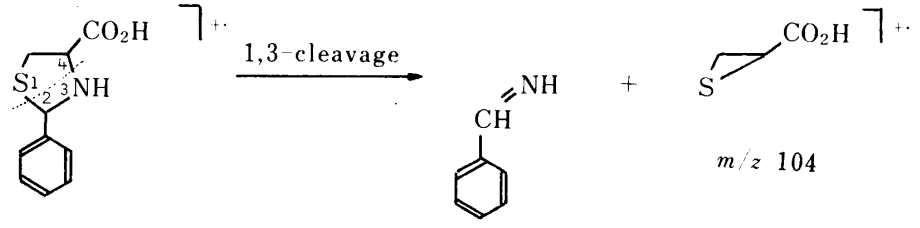
Ion	Compd.	Elemental composition	Calcd	Observed	Error (*mu*)
a	**3**	C_8H_8NO	134.0605	134.0594	−1.1
	4	C_8H_8NO	134.0605	134.0618	1.3
	5	C_7H_6NO	120.0449	120.0430	−1.9
b	**3**	$C_{12}H_{14}NO_2S$	236.0745	236.0714	−3.1
	4	$C_{12}H_{14}NO_2S$	236.0745	236.0724	−2.1
c	**3**	C_8H_7OS	151.0217	151.0228	1.1
d	**3**	$C_{12}H_{15}NO_2S_2$	269.0544	269.0579	3.5
	4	$C_{12}H_{15}NO_2S_2$	269.0544	269.0536	−0.8
e	**5**	C_7H_7NOS	153.0248	153.0256	0.8
g	**3**	$C_{11}H_{12}NO_3S$	238.0537	238.0502	−3.5
	4	$C_{12}H_{14}NO_3S$	252.0694	252.0660	−3.4
i	**3**	$C_{11}H_{12}NO_2S$	222.0588	222.0596	0.8
	4	$C_{11}H_{12}NO_2S$	222.0588	222.0609	2.1
j	**2**	$C_9H_{10}NOS$	180.0483	180.0515	3.2
	3	$C_{10}H_{12}NOS$	194.0639	194.0662	2.3
	4	$C_{10}H_{12}NOS$	194.0639	194.0647	0.8
k	**2**	C_2H_5S	61.0111	61.0119	0.8
	3	C_2H_5S	61.0111	61.0112	0.1
	4	C_2H_5S	61.0111	61.0121	1.0
l	**2**	C_3H_7S	75.0268	75.0258	−1.0
	3	C_3H_7S	75.0268	75.0272	0.4
	4	C_3H_7S	75.0268	75.0284	1.6



a **b**

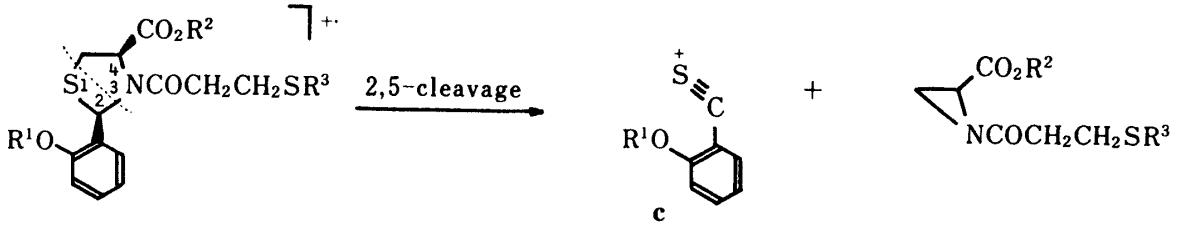
1,2 : R^1=H *m/z* 120
3,4 : R^1=CH_3 *m/z* 134

3,4 : R^1=R^3=CH_3 *m/z* 236



m/z 104

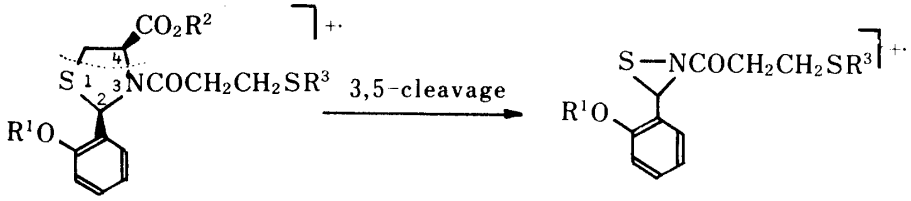
Chart 2



c

1,2 : R^1=H *m/z* 137
3,4 : R^1=CH_3 *m/z* 151

Chat 3



d

1 : R^1=R^3=H *m/z* 241
2 : R^1=H, R^3=CH_3 *m/z* 255
3,4 : R^1=R^3=CH_3 *m/z* 269

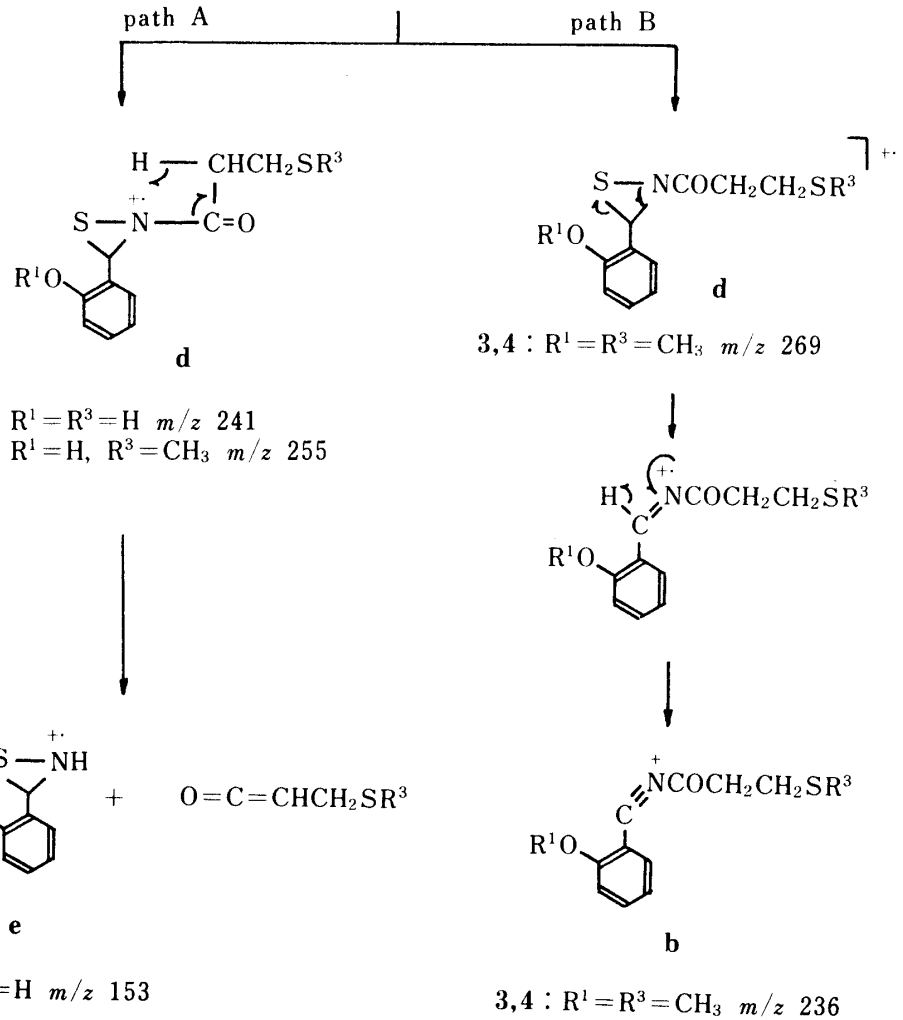


Chart 4

Ion c in Chart 3 (2,5-Cleavage)

Ions at *m/z* 137 and 151 were seen with significant intensities in **1** and **2**, and **3** and **4**, respectively. In these cases, positive charges were retained on the sulfur-containing fragment, and the ion (*m/z* 137) itself appeared as a base peak in **7**. The fragment ion peak at *m/z* 151 in **3** could be established by high-resolution mass spectrometry.

Ions d, e and b in Chart 4 (3,5-Cleavage)

The thiaziridine-type ion **d** arose first by 3,5-cleavage. The ions derived from $[M]^{+\cdot}$ at *m/z* 241 in **1**, *m/z* 255 in **2** and *m/z* 269 in **4** were generally weak, but the peak at *m/z* 269 in **3** was observed with significant intensity through the metastable supported transition observed in **7**. The elemental composition of the ion was established by high-resolution mass

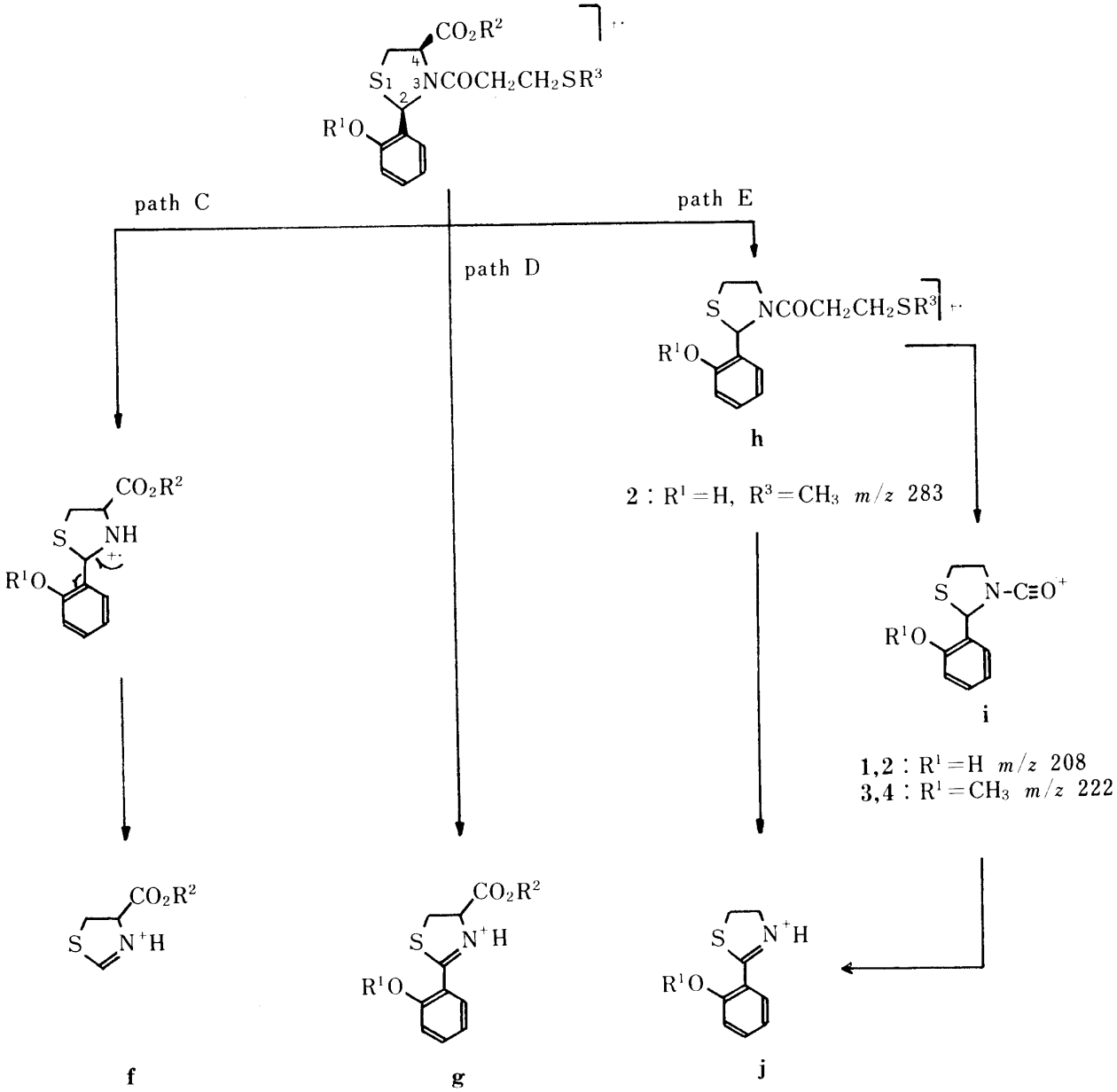


Chart 5

spectrometry (Table II). In addition, the deuterium-labelled derivative **4'** showed a reasonable 3 *u* shift (*m/z* 269→*m/z* 272), whereas no shift was seen in the case of **4''** (Fig. 1).

The further fragmentation of ion **d** occurs by two pathways (paths A and B). Path A is the elimination of a ketene moiety from ion **d** to form ion **e** at *m/z* 153, whereas path B involves the elimination of a sulfur atom and successive loss of H· from ion **d** to form ion **b** mentioned above at *m/z* 236. Ion **b** was also established by the observation of a 3 *u* shift in **4'** and no shift in **4''**.

Ions f, g, h, i, and j in Chart 5 (Cleavage of the Substituents)

Other fragmentation processes of thiazolidines are shown in Chart 5.

Eliminations of the 2-aryl group by path C were observed. The relative abundance of ion **f** formed from **1** was 42% and the fragment ion at *m/z* 132 in **5** was the base peak.[6] These results are in contrast to the limited elimination of the C_2-phenyl group in 2-phenylthiazolidines.[5]

Path D leads to ion **g**, which originates from $[M]^{+\cdot}$ with metastable supported elimination of the mercaptoacyl group in **3** and **4**. The resulting ion peaks were among the most intense.

In path E, the elimination of carboxylic acid moiety from $[M]^{+\cdot}$ gave the unstable ion **h** observed in only **2**,[7] while ions **i** and **j** gave intense peaks. Because the metastable ion for the **h**→**j** process was observed, the ion **h** was concluded to be a precursor of these ions.

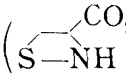
Ions k and l

Two characteristic ions **k** and **l** derived from the mercaptoacyl moiety appeared with significant intensity in *S*-methyl derivatives (**2**—**4**).[8]

McLafferty-type rearrangement ions observed in 2-(methylthio)propionic acid derivatives[9] were not seen in the cases of **2**—**4**.

$CH_3\overset{+}{S}=CH_2$ **k** $\qquad$ $CH_3\overset{+}{S}=CHCH_3$ **l**

Other Ions

The phenyl ion was observed at *m/z* 77 in all of the thiazolidines. In compound **6**, a tropilium ion (at *m/z* 91) appeared as the base peak, and the counterpart ion  at *m/z* 119 was also seen.

Experimental

Melting points were determined in open capillary tubes with a Yamato melting point apparatus and are uncorrected. Specific rotations were measured with a JASCO DIP-4 polarimeter. Infrared (IR) spectra were recorded on a JASCO A-302 spectrometer. ^{1}H-NMR spectra were measured with a JEOL PMX-60 spectrometer using tetramethylsilane as an internal standard. Mass spectra were measured with a Hitachi M-52 spectrometer equipped with a direct insertion system under the following conditions: ionization voltage, 70 eV; ion accelerating voltage, 3.0 kV; ion source and direct probe temperature, 80—100°C. The high-resolution mass spectra were taken on a Hitachi RMU-7L double-focusing mass spectrometer. Operating conditions: source temperature, 100°C; ionization voltage, 70 eV; accelerating voltage, 3.2 kV.

Compounds **5** and **6** were synthesized by reaction of L-cysteine with aryl aldehydes (*e.g.*, salicylaldehyde and *o*-methoxybenzaldehyde).[1b] In the IR spectra, a characteristic zwitterion band was observed at *ca.* 1620 cm^{-1}. The ^{1}H-NMR spectra revealed two ABX systems due to C_5–H_2 and C_4–H. The signals at δ 2.97 (dd, J=10 and 9 Hz), 3.22 (dd, J=10 and 6 Hz) and 3.83 (dd, J=9 and 6 Hz) can be assigned to A̲BX (C_5–H), AB̲X (C_5–H) and ABX̲ (C_4–H) in one configuration of **5**, whereas the signals at δ 3.02 (dd, J=10 and 6 Hz), 3.33 (dd, J=10 and 6 Hz) and 4.22 (t, J=6 Hz) can be assigned to that in the other configuration of **5**. The corresponding methine proton at C_2 appeared at δ 5.65 and 5.85 (each singlet). Thus, **5** and **6** were mixtures of two diastereoisomers.

Compounds **2** and **3** were synthesized by reaction of **5** (9.6 g, 0.043 mol) or **6** (4.8 g, 0.02 mol) with 3-(methylthio)propionyl chloride (5.9 g, 0.043 mol or 2.8 g, 0.02 mol) in triethylamine (15.3 ml, 0.11 mol or

Table III. Physicochemical Properties of Thiazolidines

Compd.	mp (°C) (Recrystn. solvent)	$[\alpha]_D$ deg. (c, solv., °C)	Formula	Analysis (%) Calcd (Found) C	H	N	IR $\nu_{C=O}^{KBr}$ cm^{-1} COO	CON
2	102.5—105.5 (EtOH–H_2O)	+159.7 (1.2, MeOH, 26)	$C_{14}H_{17}NO_4S_2 \cdot 1/2H_2O$	49.98 (49.95)	5.39 (5.26)	4.16 (4.14)	1748	1632
3	129—130 (EtOAc)	+166.4 (1.0, MeOH, 28)	$C_{15}H_{19}NO_4S_2$	52.77 (52.79)	5.61 (5.49)	4.10 (4.12)	1758	1610
4	Oil						1749 (film)	1658
5	174—175 (dec.) (DMF–H_2O)	−187.0 (1.0, DMSO, 28)	$C_{10}H_{11}NO_3S$	53.32 (53.23)	4.92 (4.97)	6.22 (6.26)	1615	—
6	137—139 (dec.) (DMF–H_2O)	−214.6 (1.0, DMSO, 28)	$C_{11}H_{13}NO_3S$	55.21 (55.09)	5.48 (5.43)	5.85 (5.88)	1625	—
7	171—173 (dec.) (EtOAc–MeOH)	+212.2 (0.5, MeOH, 25)	$C_{12}H_{13}NO_4S$	53.92 (54.10)	4.90 (5.03)	5.24 (5.05)	1737	1603

Table IV. ^{1}H-NMR Data for Thiazolidines

Compd.	Solvent	Chemical shifts δ
2	Acetone-d_6	1.90 (3H, s, CH_3), 2.30—2.93 (4H, m, CH_2CH_2), 3.10—3.70 (2H, m, C_5-H), 4.90 (1H, t, J=8 Hz, C_4-H), 6.52 (1H, s, C_2-H), 6.60—8.17 (4H, m, aromatic H), 8.90 (2H,br s, OH and CO_2H)
3	$CDCl_3$	1.88 and 2.00 (3H, each s, SCH_3), 2.30—2.90 (4H, m, CH_2CH_2), 3.22 (2H, d, J=8 Hz, C_5-H), 3.80 (3H, s, OCH_3), 4.90 (1H, t, J=8 Hz, C_4-H), 6.33 (1H, s, C_2-H), 6.70—8.00 (4H, m, aromatic H), 11.27 (1H, s, CO_2H)
4	$CDCl_3$	1.90 (3H, s, SCH_3), 2.21—2.88 (4H, m, CH_2CH_2), 3.05 (1H, dd, J=14 and 9 Hz, A$\underline{B}$X, C_5-H), 3.30 (1H, dd, J=14 and 7 Hz, A$\underline{B}$X, C_5-H), 3.82 (3H, s, OCH_3 or CO_2CH_3), 3.89 (3H, s, CO_2CH_3 or OCH_3), 4.85 (1H, dd, J=9 and 7 Hz, AB$\underline{X}$, C_4-H), 6.34 (1H, s, C_2-H), 6.76—8.18 (4H, m, aromatic H)
5	DMSO-d_6	2.97 (dd, J=10 and 9 Hz, A$\underline{B}$X, C_5-H), 3.02 (dd, J=10 and 6 Hz, A$\underline{B}$'B'X', C_5-H), 3.22 (dd, J=10 and 6 Hz, A$\underline{B}$X, C_5-H), 3.33 (dd, J=10 and 6 Hz, A'$\underline{B}$'X', C_5-H), 3.83 (dd, J=9 and 6 Hz, AB$\underline{X}$, C_4-H), 4.22 (t, J=6 Hz, A'B'$\underline{X}$', C_4H), 5.65 and 5.85 (1H, each s, C_2-H), 6.33—7.43 (4H, m, aromatic H), 7.43—9.00 (3H, br, OH, NH and CO_2H)
6	DMSO-d_6	2.94 (dd, J=10 and 9 Hz, A$\underline{B}$X, C_5-H), 2.95 (dd, J=10 and 6 Hz, A$\underline{B}$'B'X', C_5-H), 3.21 (dd, J=10 and 6 Hz, A$\underline{B}$X, C_5-H), 3.33 (dd, J=10 and 6 Hz, A'$\underline{B}$'X', C_5-H), 3.80 (dd, J=9 and 6 AB$\underline{X}$, C_4-H), 3.82 (3H, s, CH_3), 4.12 (t, J=6 Hz, A'B'$\underline{X}$', C_4-H), 5.67 and 5.85 (1H, each s, C_2-H), 6.72—7.58 (4H, m, aromatic H), 7.58—9.00 (2H, br, NH and CO_2H)
7	$CDCl_3$	1.87 and 2.13 (3H, each s, $COCH_3$), 3.22 (1H, dd, J=11 and 8.5 Hz, A$\underline{B}$X, C_5-H), 3.33 (1H, dd, J=11 and 6.5 Hz, A$\underline{B}$X, C_5-H), 4.82 (1H, dd, J=8.5 and 6.5 Hz, AB$\underline{X}$, C_4-H), 6.40 (1H, s, C_2-H), 6.58—8.10 (4H, m, aromatic H)

7.0 ml, 0.05 mol)-acetone (100 ml or 50 ml) as descrived in a previous paper.[1c] Compound **2** was converted to a dicyclohexylamine (DCHA) salt for purification, and then the free acid was obtained by treatment with potassium bisulfate. Compounds **4**, **4'** and **4''** were synthesized by reaction of **3** and the corresponding thiol with diazomethane or deuterio-diazomethane. In the ^{1}H-NMR spectra of **2**, **3** and **4**, the methine proton at C_2 appeared as a singlet in each case and the coupling constant ($J_{AX}+J_{BX}$) of the methine proton at C_4 was 16 Hz. The configurations of **2**, **3** and **4** were similar to those of a series of (2*R*,4*R*)-2-aryl-3-(3-mercaptopropionyl)-4-thiazolidinecarboxylic acids.[1b] Thus, the structures of **2**, **3** and **4** were established.

Compound **7** was reported in a previous paper.[1b]

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References and Notes

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