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Synthesis of Hydroquinone Monosulfates Having Carboxyalkyl and Hydroxyalkyl Side Chains

KAYOKO OKAMOTO,* EIJI MIZUTA, KAZUHIDE KAMIYA, and ISUKE IMADA

Central Research Division, Takeda Chemical Industries, Ltd., Jusohonmachi, Yodogawa-ku, Osaka 532, Japan

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Sulfate conjugates of ubiquinone metabolites (Ia, Ib) and related compounds including 6-(10-hydroxydecyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinone (idebenone, Id-10) were synthesized. Chemical shifts in the proton nuclear magnetic resonance (^{1}H-NMR) spectra of the 1- and 4-sulfates were assigned by investigating differences in the low-field shifts between model compounds and the corresponding sulfates, and by regio-specific synthesis of the 4-sulfates.

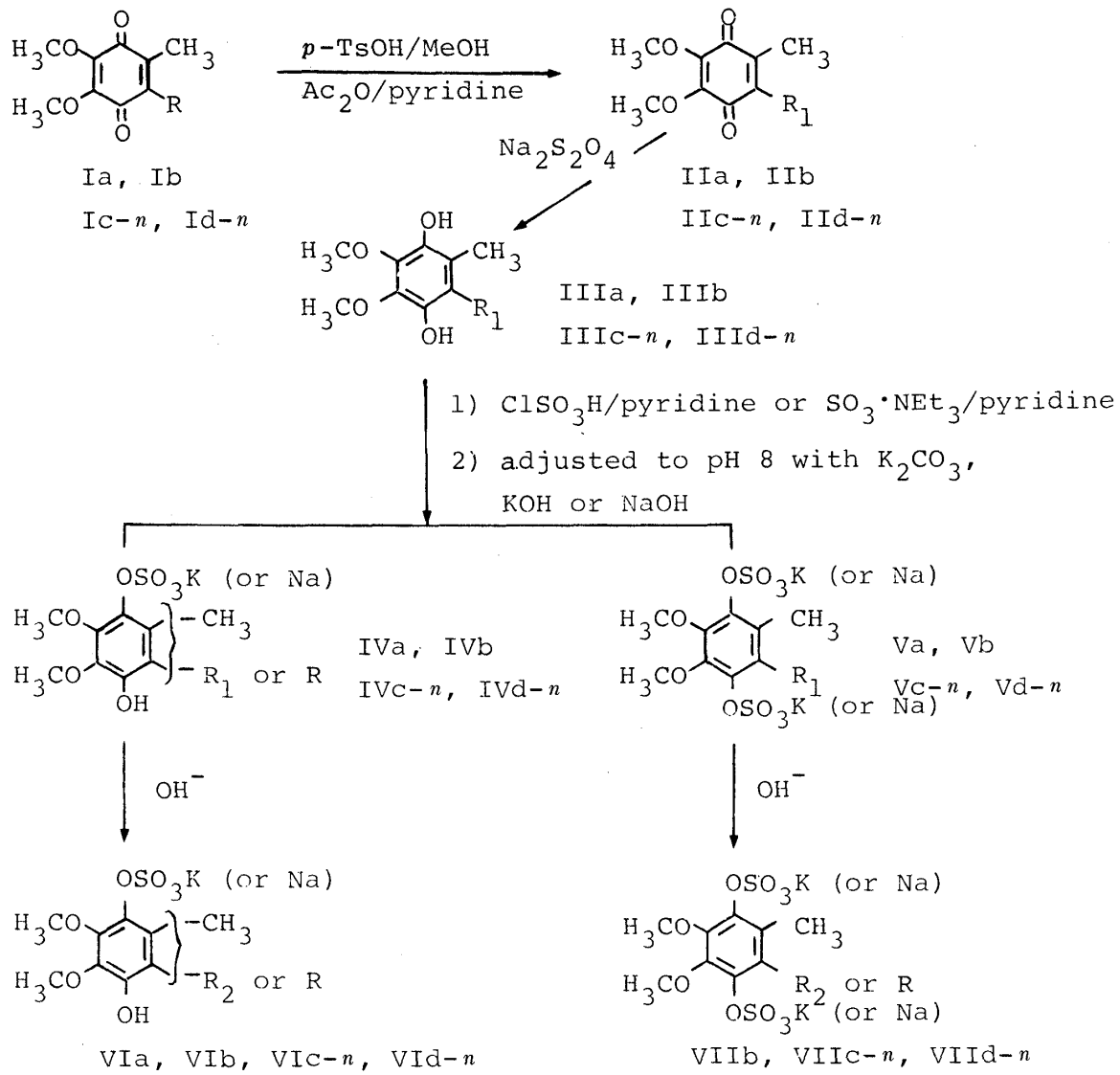
Keywords——idebenone; sulfate conjugate; hydroquinone 1-sulfate; hydroquinone 4-sulfate; SIMS

In a previous paper,[1] we reported that in several animal species, ubiquinone-7 is converted to 6-(5-carboxy-3-methyl-2-pentenyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinone (Ia) and 6-(3-carboxybutyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinone (Ib), and excreted in the urine as a sulfate conjugate. Since these quinonyl acids showed some interesting biological effects,[2] we also synthesized the related compounds 6-(ω-carboxyalkyl)- and 6-(ω-hydroxyalkyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinones (Ic-*n*, Id-*n*) and investigated their structure–activity relationships.[2b-e]

Sulfate conjugates of steroid,[3] calciferol[4] and carotenoid[5] have been found in animal sources and may have significant physiological roles. In this report, we describe the synthesis of some sulfate conjugates, especially monosulfates of the metabolites (Ia, Ib) and related quinones.

Monosulfates were synthesized by a modification of the method described for the synthesis of metabolites of ubiquinone-7[6] (Chart 1). The carboxylic acid (Ia, Ib or Ic-*n*) was converted to the methyl ester (IIa, IIb or IIc-*n*), reduced with sodium hydrosulfite to the hydroquinone (IIIa, IIIb or IIIc-*n*), and then converted to the sulfate (IVa, IVb, IVc-*n*, Va, Vb or Vc-*n*) by treatment with chlorosulfonic acid in pyridine or sulfur trioxide triethylamine complex ($SO_3 \cdot NEt_3$)[7] in the presence of sodium hydrosulfite. The reaction mixture was separated by column chromatography into two fractions, one containing monosulfates (IVa, IVb or IVc-*n*) and the other containing small amounts of disulfate (Va, Vb or Vc-*n*). The product was hydrolyzed with alkali hydroxide (KOH, NaOH) to obtain the monosulfates [VIa, VIb or VIc-*n* ($n=4$, 10)] and disulfate (VIIb or VIIc-10). These were obtained as sulfate or sulfate/carboxylate salts (K or Na), and the monosulfates were mixtures of 1- and 4-sulfates. One isomer, the disodium salt of 6-(3-carboxypropyl)-2,3-dimethoxy-5-methylhydroquinone 4-sulfate (VIc-4A),[8] was obtained by the hydrolysis of the isomer (IVc-4A) isolated by diethylaminoethyl (DEAE)-cellulose column chromatography of 2,3-dimethoxy-6-(3-methoxycarbonylpropyl)-5-methylhydroquinone monosulfate (IVc-4). Next, the sulfates of 6-(ω-hydroxyalkyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinone (Id-*n*) were obtained as follows. 6-(10-Hydroxydecyl)-2,3-dimethoxy-5-methyl-1,4-benzoquinone (idebenone, Id-10) was acetylated to the acetate (IId-10). Then IId-10 was reduced with sodium

hydrosulfite to the hydroquinone and converted to the sulfates as described above. The reaction mixture was separated by column chromatography into two fractions, monosulfates (IVd-10) and disulfate (Vd-10). Alkaline hydrolysis of these compounds gave VId-10 and VIId-10, respectively. Hydroquinone 4-sulfate (VId-10A) was obtained by recrystallization from water as colorless needles, mp 178—181 °C. In a similar manner, the monosulfates VId-4 and VId-11 were obtained from Id-4 and Id-11, respectively.



Compound	R	R_1	R_2
Ia—VIa	$-CH_2CH=C(CH_3)CH_2CH_2COOH$	$-CH_2CH=C(CH_3)CH_2CH_2COOCH_3$	$-CH_2CH=C(CH_3)CH_2CH_2COOK$
Ib—VIIb	$-CH_2CH_2CH(CH_3)COOH$	$-CH_2CH_2CH(CH_3)COOCH_3$	$-CH_2CH_2CH(CH_3)COOK$
Ic-*n*—VIIc-*n*	$-(CH_2)_{n-1}COOH$	$-(CH_2)_{n-1}COOCH_3$	$-(CH_2)_{n-1}COOK$ (or Na)
Id-*n*—VIId-*n*	$-(CH_2)_nOH$	$-(CH_2)_nOCOCH_3$	—

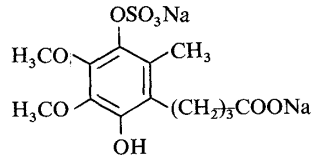
Chart 1

The structure of VId-10A was determined on the basis of the chemical shift of the ring methyl (2.22 ppm) in the proton nuclear magnetic resonance (^{1}H-NMR) spectrum (Table I), absorptions at 280 and 273 nm (phenol) in the ultraviolet (UV) spectrum, absorptions at 1280, 1230 and 1050 cm^{-1} (SO_2) in the infrared (IR) spectrum, a positive color reaction (phenolic hydroxyl) with the ferric chloride–potassium ferricyanide reagent,[9] and elemental analysis. The monosulfates were equimolar mixtures of 1- and 4-sulfates, because two methyl and two methylene signals were observed at the same intensity in their ^{1}H-NMR spectra (Table I). The ^{1}H-NMR signal due to the ring methyl was shifted to low field by *o*- or *m*-hydroxy sulfation, probably due to the electron-attracting character of the sulfonyl group. To confirm the assignment of these signals, we compared the methyl signals of *o*-cresol (VIII), *m*-cresol (X) and their sulfates (IX, XI) (Table II). Sulfation of the hydroxyl group caused the methyl signals of *o*-cresol and *m*-cresol to shift to lower field by 0.13 and 0.07 ppm, respectively. The low-field shift of the ring methyl and methylene signals was larger with *o*-hydroxy than *m*-hydroxy sulfation, and the signals of the ring methyl and methylene of the monosulfates were assigned as shown in Table I. A similar result has been reported for the ring proton in the sulfation of estrogen catechol.[10] This conclusion was confirmed by the regio-specific synthesis of one isomer. That is, VIc-4A and VIc-10A were obtained by the Elbs persulfate oxidation[11] of 4-(2-hydroxy-3,4-dimethoxy-6-methylphenyl)butyric acid (XII-4) and 10-(2-hydroxy-3,4-dimethoxy-6-methylphenyl)decanoic acid (XII-10),[2c] respectively (Chart 2). Their chemical shifts agreed well with the above results.

The results of mass spectrometry supported our conclusions on the structures of these compounds. Fast atom bombardment mass spectrometry (FABMS) of estrogen glucuro-

TABLE I. ^{1}H-NMR Chemical Shifts of Ring-CH_3 and Ring-CH_2 in Hydroquinone, Hydroquinone Monosulfates and Hydroquinone Disulfates

Compound No. (salt form)	Chemical shift value (ppm) in D_2O			
	Ring-CH_3		Ring-CH_2	
	1-Sulfate	4-Sulfate	1-Sulfate	4-Sulfate
VIa (K)	1.95	2.04	3.27	
VIb (K)	2.11	2.22	2.32—2.74	
Hydroquinone of Ic-4	2.12		2.66	
VIc-4 (K)	2.17	2.24	2.72	2.64
VIc-4A (K)		2.24		2.65
VIc-4 (Na)	2.17	2.25	2.71	2.63
VIc-4A (Na)[a]		2.25		2.64
VIc-10 (K)	2.10	2.21	2.72	2.54
VIc-10A (K)		2.23		2.56
VId-10 (K)	2.10	2.22	2.73	2.53
VId-10A (K)[b]		2.22		2.54
VIIb (K)	2.17		2.65	
VIIc-10 (K)	2.29		2.78	
VIId-10 (Na)	2.20		2.62	

a) 

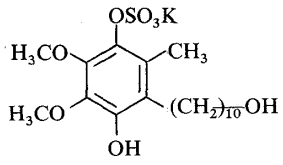
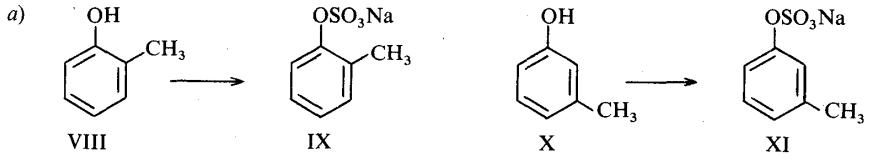
b) 

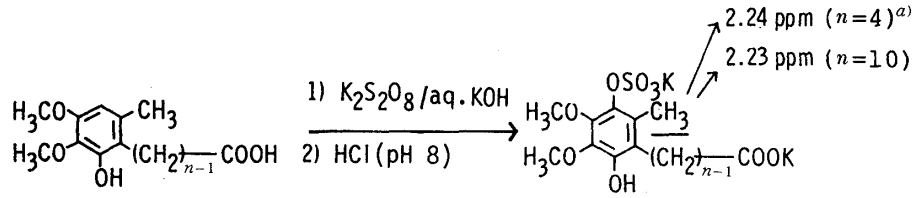
TABLE II. ^{1}H-NMR Chemical Shifts of CH_3 of *o*-Cresol, *m*-Cresol and Their Sulfates in D_2O

Compound[a)]	ppm of CH_3	Low-field shift on sulfation
o-Cresol (VIII)	2.21	—
o-Cresol sulfate (IX)	2.34	0.13
m-Cresol (X)	2.40	—
m-Cresol sulfate (XI)	2.47	0.07

a)



VIII IX X XI



XII-*n* (*n*=4, 10) VIc-*n*A (*n*=4, 10)

Chart 2. Alternate Synthetic Route for Hydroquinone Monosulfate

a) Refer to Table I.

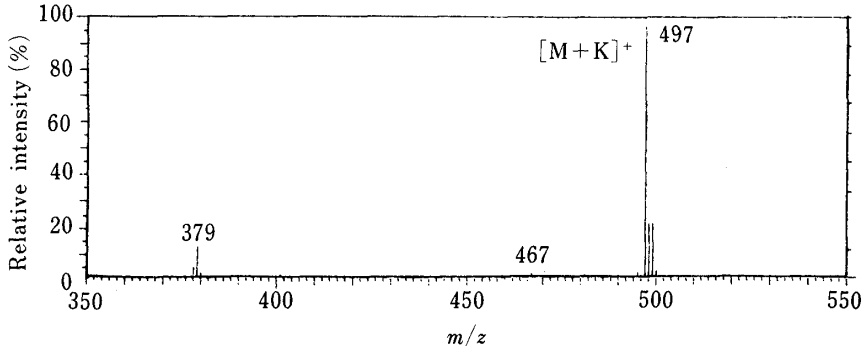


Fig. 1. Mass Spectrum of VId-10A Obtained by Secondary Ion Mass Spectrometry (SIMS)

TABLE III. Mass Spectra of Hydroquinone Monosulfates and Disulfate Obtained by Secondary Ion Mass Spectrometry (SIMS)

Compound[a)]	Molecular formula	Molecular weight	Ions: *m/z* (%)[b)]	
			$[M+K]^+$	$[M+K-118]^+$
VId-4 (K)	$C_{13}H_{19}KO_8S$	374	413 (100)	295 (3)
VId-10A (K)	$C_{19}H_{31}KO_8S$	458	497 (100)	379 (14)
VId-11 (K)	$C_{20}H_{33}KO_8S$	472	511 (100)	393 (14)
VIId-10 (K)	$C_{19}H_{30}K_2O_{11}S_2$	576	615 (100)	497 (59)

a) K in parentheses after the compound number shows the form of the salt. *b)* Relative intensity.

nide and sulfate has recently been reported.[12] The sulfates described above were analyzed by secondary ion mass spectrometry (SIMS) in xenon gas. As can been seen in Fig. 1, the spectrum of the potassium salt of VId-10A was clear and simple, and showed $m/z=497$ ($[M+K]^+$) ion as a dominant ion in the molecular ion region. In general, the $[M+K]^+$ ion and the $[M+K-118]^+$ ion were dominant in the SIMS spectra of 6-(ω-hydroxyalkyl)-2,3-dimethoxy-5-methylhydroquinone sulfates (Table III). On the other hand, the $[M+H]^+$ and $[M+K(\text{or Na})]^+$ ions were dominant in the SIMS spectra of 6-(ω-carboxyalkyl)-2,3-dimethoxy-5-methylhydroquinone sulfates with no $[M+K(\text{or Na})-118]^+$ ion being observed (Table IV).

Table IV. Mass Spectra of Hydroquinone Monosulfates Obtained by SIMS

Compound[a]	Molecular formula	Molecular weight	Ions: m/z (%)[b]		
			$[M+H]^+$	$[M+K \text{ (or Na)}]^+$	$[M-H+2K \text{ (or 2Na)}]^+$
VIa (K)	$C_{16}H_{20}K_2O_9S$	466	467 (72)	505 (100)	543 (24)
VIb (K)	$C_{14}H_{18}K_2O_9S$	440	441 (87)	479 (100)	517 (18)
VIc-4A (Na)	$C_{13}H_{16}Na_2O_9S$	394	395 (100)	417 (88)	439 (29)
VIc-4 (K)	$C_{13}H_{16}K_2O_9S$	426	427 (100)	465 (44)	—

a) K and Na in parentheses after the compound number show the form of the salt. *b*) Relative intensity.

Table V. Effects of Quinones (Ic-4, Id-10), Hydroquinone (Id-10 Hydroquinone) and Hydroquinone Monosulfates (VIc-4A, VId-10A) on the Release of Hydrolases from the Lysosomal Fraction and on the Activity of Cyclic AMP Phosphodiesterase

Compound	Effect[a] on release of lysosomal hydrolases			Effect[b] on phosphodiesterase		
	Concentration (M)	% hydrolases released		Concentration (M)	% inhibition	
		β-Glucuronidase	Acid phosphatase		Expt. I	Expt. II
None (control)	—	100	100	—	0	0
Id-10	2×10^{-5}	76	64	2.5×10^{-4}	47	49
	2×10^{-4}	76	64	5×10^{-4}	61	
				1×10^{-3}	64	
Ic-4	2×10^{-5}	119	142	5×10^{-4}	33	
				1×10^{-3}	39	
Id-10 hydroquinone	2×10^{-6}	105	110			
	2×10^{-5}	78	80			
VId-10A	2×10^{-8}	102	89	5×10^{-4}		70
	2×10^{-7}	131	146	1×10^{-3}		89
	2×10^{-6}	128	148			
	2×10^{-5}	132	166			
VIc-4A	2×10^{-6}	104	105	5×10^{-4}		5
	2×10^{-5}	129	160	1×10^{-3}		6
Theophylline				1×10^{-4}		13

a) The effect on membrane stability was assayed by measuring the hydrolase released from the lysosomal fraction of rat liver during incubation at 37 °C for 90 min as already described,[2a] and the values are given as % of the control (control: 100%). None of the test compounds inhibited the lysosomal hydrolases. *b*) Phosphodiesterase activity was determined by measuring inorganic phosphate after incubation for 30 min at 37 °C, as described by Butcher and Sutherland.[15] The incubation mixture (1 ml) consisted of 0.5 M Tris–HCl, pH 7.5 (0.2 ml), 20 mM $MgSO_4$ (0.1 ml), 20 mM cyclic AMP (0.1 ml), 0.1% 5′-nucleotidase (0.1 ml), 0.05% bovine heart phosphodiesterase (0.2 ml), a solution of the test compound in dimethyl sulfoxide (5—10 μl) and water. The percent inhibition with respect to the control is shown (control: 0%).

Sulfate conjugates of steroid,[3] calciferol[4] and carotenoid[5] occur as intermediate metabolites in animal sources, and have been considered to be one of the active forms. Further, Id-10, after being administered clinically and experimentally, appeared in the plasma mainly in the form of its hydroquinone monosulfate (VId-10).[13] This result suggested that the main conjugate metabolites of ubiquinone-7 may be VIa and VIb.

Some quinone compounds (Ia, Ib, Ic-*n*, and Id-*n*) have been reported to affect the stability of the lysosomal membrane[2c] and the tissue level of cyclic nucleotides.[2c,14] To investigate the structure–activity relationships, especially the relation to sulfate conjugation in metabolism, the effects of the monosulfates (VIc-4A, VId-10A) on the stability of rat liver lysosomes and bovine heart phosphodiesterase were compared with those of the corresponding quinone compounds (Table V). As a result of sulfation, Id-10 lost its membrane-stabilizing activity. VIc-4A did not show a stabilizing activity, like Ic-4. These results support our earlier finding[2c] that a decrease in the hydrophobicity of the compounds is accompanied with a decrease in membrane-stabilizing activity. The inhibition of phosphodiesterase activity[15] by

TABLE VI. Physicochemical Data for Potassium Salts of IVa, IVb, Va, Vb, VIa, VIb, VIc-*n* ($n=4$, 10), VIIb and VIIc-10

Compound No. (salt form)	IR ν_{max}^{KBr} cm^{-1}	UV $\lambda_{max}^{H_2O}$ nm (ε)	^{1}H-NMR (D_2O) δ
IVa (K)	1730 (ester) 1250, 1050 (SO_2)		1.62 (3H, s, $=CCH_3$), 1.94 and 2.04 (3H, s, CH_3 on the ring), 2.10—2.49 (4H, m, CH_2COO, $=CCH_2$), 3.05—3.29 (2H, m, CH_2 on the ring), 3.35 (3H, s, $COOCH_3$), 3.70 (3H, s, OCH_3), 3.79 (3H, s, OCH_3)
IVb (K)	1730 (ester) 1250, 1040 (SO_2)		1.04 (3H, d, $J=6$ Hz, CH_3), 1.34—1.75 (2H, m, CH_2), 1.95 and 2.05 (3H, s, CH_3 on the ring), 2.29—2.60 (3H, m, CH_2 on the ring), 3.05 (3H, s, $COOCH_3$), 3.67 (3H, s, OCH_3), 3.73 (3H, s, OCH_3)
Va (K)	1730 (ester) 1250, 1050 (SO_2)		1.67 (3H, s, $=CCH_3$), 2.10 (3H, s, CH_3 on the ring), 2.17—2.57 (4H, m, CH_2COO, $=CCH_2$), 3.24 (2H, m, CH_2 on the ring), 3.37 (3H, s, $COOCH_3$), 3.79 (6H, s, OCH_3)
Vb (K)	1730 (ester) 1250, 1040 (SO_2)		1.07 (3H, d, $J=6$ Hz, CH_3), 1.50—1.77 (2H, m, CH_2), 2.14 (3H, s, CH_3), 2.42—2.82 (3H, m, CH_2 on the ring, CH), 3.55 (3H, s, $COOCH_3$), 3.79 (6H, s, OCH_3)
VIa (K)	1560 (COOK) 1260, 1050 (SO_2)	273, 280	1.70 (3H, s, $=CCH_3$), 1.95 and 2.04 (3H, s, CH_3 on the ring), 2.19 (4H, br, CH_2COO, $=CCH_2$), 3.27 (2H, m, CH_2 on the ring), 3.67 (3H, s, OCH_3), 3.77 (3H, s, OCH_3)
VIb (K)	1560 (COOK) 1250, 1040 (SO_2)	274, 281	1.07 (3H, d, $J=6$ Hz, CH_3), 1.40—1.72 (2H, m, CH_2), 2.02 and 2.14 (3H, s, CH_3 on the ring), 2.25—2.65 (3H, m, CH_2 on the ring, CH), 3.69 (3H, s, OCH_3), 3.79 (3H, s, OCH_3)
VIc-4 (K)	1560 (COOK) 1250, 1040 (SO_2)	275, 280	1.69 (2H, q, CH_2), 2.17 and 2.24 (3H, s, CH_3 on the ring), 2.26 (2H, t, CH_2COO), 2.64 and 2.72 (2H, t, CH_2 on the ring), 3.86 (3H, t, OCH_3), 3.92 (3H, t, OCH_3)
VIc-10 (K)	1720 (COOH) 1250, 1050 (SO_2)	273.5, 279	1.21 (14H, br, CH_2), 2.10 and 2.21 (3H, s, CH_3 on the ring), 2.26 (2H, t, CH_2COO), 2.54 and 2.72 (2H, m, CH_2 on the ring), 3.86 (3H, s, OCH_3), 3.91 (3H, s, OCH_3)
VIIb (K)	1560 (COOK) 1250, 1040 (SO_2)		1.04 (3H, d, $J=6$ Hz, CH_3), 1.60 (2H, m, CH_2), 2.17 (3H, s, CH_3 on the ring), 2.65 (3H, m, CH_2 on the ring, CH), 3.80 (6H, s, OCH_3)
VIIc-10 (K)	1720 (COOH) 1250, 1050 (SO_2)	278	1.29 (14H, br, CH_2), 2.29 (3H, s, CH_3 on the ring), 2.34 (2H, t, CH_2COO), 2.78 (2H, t, CH_2 on the ring), 3.94 (6H, s, OCH_3)

Id-10 was almost the same as that by the sulfate conjugate, though Ic-4 lost its inhibitory activity on sulfation. This may indicate that there is no relation between the mechanisms of these effects.

Experimental

Melting points were measured with a Yanagimoto micro melting point apparatus, and are uncorrected. UV spectra were recorded in EtOH or H_2O with a Hitachi EPS-3T spectrophotometer. IR spectra were obtained with Hitachi EPI-S2 and -215 spectrophotometers, and SIMS in xenon gas with a Hitachi M-80A. ^{1}H-NMR spectra were run on Varian HA-100, T-60 and JEOL JNM-GX 400 spectrophotometers in D_2O with tetramethylsilane (TMS) as an external standard. Chemical shifts are given as δ values (ppm): s, singlet: d, doublet; t, triplet; q, quartet; br, broad; m, multiplet. Phosphodiesterase from beef heart in glycerol (Boehringer Mannheim), 3′,5′-cyclic adenosine monophosphate (cyclic AMP) (Boehringer Mannheim), and snake venom (Sigma) were purchased from the indicated sources. The lysosomal fractions of rat liver were prepared from 6- to 8-week-old female Sprague-Dawley rats as already described.[2a] The effects on membrane stability of the rat liver lysosomal fraction and on beef heart phosphodiesterase activity were determined by methods similar to those described previously.[2c] Protein concentrations were determined by the Lowry method.[16]

General Procedure for the Synthesis of 6-(ω-Carboxyalkyl)-2,3-dimethoxy-5-methylhydroquinone Monosulfate and Disulfate, Potassium Salts [VIa, VIb, VIIb, VIc-*n* and VIIc-*n* (*n*=4, 10)——Quinonyl carboxylic acid (Ia, Ib or Ic-*n*) was esterified to give IIa, IIb or IIc-*n* in MeOH in the presence of a small amount of *p*-TsOH as an acid catalyst. The resulting ester (IIa, IIb or IIc-*n*) was dissolved in ethyl ether and the ethereal solution was vigorously shaken with 10% $Na_2S_2O_4$. The solvent was evaporated off *in vacuo* and the resulting hydroquinone was dissolved in pyridine. Chlorosulfonic acid (1—1.5 eq mol) was then added to the solution under N_2 at -10 to 0 °C. After being stirred at 25 °C for 3—48 h, the mixture was adjusted to pH 8—9 with 1 N K_2CO_3 or KOH, and extracted with ethyl ether to remove quinone and the hydroquinone compounds (IIa, IIb, IIIa, IIIb, IIc-*n* or IIIc-*n*). The aqueous layer was concentrated *in vacuo* and the residue was dissolved in hot EtOH. The insoluble inorganic salts were removed by filtration and the filtrate was subjected to column chromatography on silica gel (Merck, 200—400 mesh). The first fraction eluted with MeOH–$CHCl_3$ (2 : 5, v/v) was concentrated *in vacuo* to obtain the potassium salts of IVa or IVb. The fraction eluted with MeOH was concentrated *in vacuo* to obtain the potassium salts of Va or Vb. These monosulfates or disulfates were dissolved in 1 N KOH and the solution was warmed to 60 °C for 2 h. The reaction mixture was evaporated *in vacuo* after being adjusted to pH 8, and the residue was dissolved in hot EtOH. The insoluble materials were removed by filtration, and the filtrate was concentrated *in vacuo* to obtain a residue, which was subjected to column chromatography on Sephadex LH-20 or G-15 and developed with H_2O, giving VIa, VIb or VIIb. When Ic-*n* was converted to its hydroquinone sulfates, the product of alkaline hydrolysis was obtained without isolation of the intermediate (IVc-*n*). Potassium salts of VIa, VIb and VIc-*n* were obtained as powders from MeOH–ethyl ether. The physicochemical data are shown in Table VI.

Table VII. Physicochemical Data for Sodium Salts of IVc-4, IVc-4A, VIc-4 and VIc-4A

Compound No. (salt form)	IR ν_{max}^{KBr} cm^{-1}	UV $\lambda_{max}^{H_2O}$ nm (ε)	^{1}H-NMR (D_2O) δ
IVc-4 (Na)	1730 (ester) 1250, 1050 (SO_2)	276, 282.5	1.66—1.92 (2H, m, CH_2), 2.14 and 2.23 (3H, s, CH_3 on the ring), 2.42 (2H, t, J=7 Hz, CH_2COO), 2.64 and 2.80 (2H, t, J=7 Hz, CH_2 on the ring), 3.66 (3H, s, $COOCH_3$), 3.86 (3H, s, OCH_3), 3.93 (3H, s, OCH_3)
IVc-4A (Na)	1730 (ester) 1250, 1050 (SO_2)	275 (1334), 282.5 (1532)	1.65—1.93 (2H, m, CH_2), 2.23 (3H, s, CH_3 on the ring), 2.42 (2H, t, J=7 Hz, CH_2COO), 2.66 (2H, J=7 Hz, CH_2 on the ring), 3.65 (3H, s, $COOCH_3$), 3.86 (3H, s, OCH_3), 3.92 (3H, s, OCH_3)
VIc-4 (Na)	1570 (COONa) 1260, 1050 (SO_2)		1.63—1.86 (2H, m, CH_2), 2.17 and 2.25 (3H, s, CH_3 on the ring), 2.26 (2H, t, CH_2COO), 2.63 and 2.71 (2H, t, CH_2 on the ring), 3.86 (3H, s, OCH_3), 3.92 (3H, s, OCH_3)
VIc-4A (Na)	1570 (COONa) 1260, 1050 (SO_2)	275, 280.5	1.73 (2H, q, J=7.6 Hz, CH_2), 2.25 (3H, s, CH_3 on the ring), 2.26 (2H, t, J=7.6 Hz, CH_2COO), 2.64 (2H, t, J=7.6 Hz, CH_2 on the ring), 3.86 (3H, s, OCH_3), 3.92 (3H, s, OCH_3)

6-(3-Carboxypropyl)-2,3-dimethoxy-5-methylhydroquinone Monosulfate, Disodium Salt (VIc-4)——A solution of IIIc-4 in pyridine was added to a suspension on $SO_3 \cdot NEt_3$ complex[7] in pyridine under cooling at -30 °C. After being adjusted to pH 8 with 1 N NaOH, the reaction mixture was treated in the usual manner and the residue was purified by DEAE-cellulose column chromatography. The first fraction gave sodium salts of IVc-4 and the second fraction, a white powder. The sodium salts of IVc-4 were recrystallized from a small amount of H_2O to yield the sodium salt of IVc-4A as colorless crystals, mp 82—85 °C. *Anal.* Calcd for $C_{14}H_{19}NaO_9S \cdot 3H_2O$: C, 38.18; H, 5.72; S, 7.27. Found: C, 37.98; H, 5.85; S, 7.08. A solution of the sodium salts (53 mg) of IVc-4 in 1 N NaOH (1 ml) was stirred at 60 °C for 30 min. After being adjusted to pH 8 with dil. HCl, the mixture was evaporated *in vacuo* and the resulting residue was dissolved in hot EtOH. The insoluble materials were removed by filtration and the filtrate was evaporated *in vacuo*. The resulting powder was washed with $CHCl_3$, giving the sodium salts of VIc-4 as a white powder. Yield, 40 mg. Similarly, the sodium salt of VIc-4A was obtained as a white powder from the sodium salt of IVc-4A. Physicochemical data of the sodium salts of IVc-4, IVc-4A, VIc-4 and VIc-4A are shown in Table VII.

General Procedure for the Synthesis of 6-(ω-Hydroxyalkyl)-2,3-dimethoxy-5-methylhydroquinone Monosulfate and Disulfate, Sodium Salt and Potassium Salt [VId-*n* (*n*=4, 10 and 11), and VIId-10]——The quinone compound Id-*n* was acetylated in pyridine with acetic anhydride. IIId-*n* was prepared in a manner similar to that described for IIIa, IIIb or IIIc-*n* from the corresponding quinones (IId-*n*). Chlorosulfonic acid was added to a solution of IIId-*n* in

Table VIII. Physicochemical Data for Potassium and Sodium Salts of IVd-10, Vd-10, VId-*n* (*n*=4, 10, 11) and VIId-10

Compound No. (salt form)	IR ν_{max}^{KBr} cm^{-1}	UV $\lambda_{max}^{H_2O}$ nm (ε)	^{1}H-NMR (D_2O) δ
IVd-10 (K)	1730 (ester) 1250, 1050 (SO_2)		1.18 (16H, br, CH_2), 1.90 (3H, s, $OCOCH_3$), 1.98 and 2.11 (3H, s, CH_3 on the ring), 2.46 (2H, m, CH_2 on the ring), 3.70 (3H, s, OCH_3), 3.80 (3H, s, OCH_3)
IVd-10 (Na)	1730 (ester) 1250, 1050 (SO_2)		1.23 (16H, br, CH_2), 1.99 (3H, s, $OCOCH_3$), 2.08 and 2.21 (3H, s, CH_3 on the ring), 2.44—2.80 (2H, m, CH_2 on the ring), 3.80 (3H, s, OCH_3), 3.88 (3H, s, OCH_3), 3.98 (2H, t, CH_2O)
Vd-10 (Na)	1740 (ester) 1250, 1050 (SO_2)		1.17 (16H, br, CH_2), 1.99 (3H, s, $OCOCH_3$), 2.19 (3H, s, CH_3 on the ring), 2.71 (2H, t, CH_2 on the ring), 3.86 (6H, s, OCH_3), 3.99 (2H, t, J=6 Hz, CH_2O)
VId-4 (K)	1280, 1240, 1050 (SO_2)		1.38—1.66 (4H, m, CH_2), 2.08 and 2.21 (3H, s, CH_3 on the ring), 2.63 and 2.71 (2H, t, CH_2 on the ring), 3.53 (2H, t, CH_2O), 3.80 (3H, s, OCH_3), 3.90 (3H, s, OCH_3)
VId-10 (K)	1280, 1230, 1050 (SO_2)	275, 280	1.20 (16H, br, CH_2), 2.10 and 2.22 (3H, s, CH_3 on the ring), 2.53 and 2.73 (2H, m, CH_2 on the ring), 3.51 (2H, t, J=6 Hz, CH_2O), 3.81 (3H, s, OCH_3), 3.90 (3H, s, OCH_3)
VId-10A (K)	1280, 1230, 1050 (SO_2)	273.5 (1090), 280 (1100)	1.21 (16H, br, CH_2), 2.22 (3H, s, CH_3 on the ring), 2.54 (2H, m, CH_2 on the ring), 3.52 (2H, t, J=6 Hz, CH_2O), 3.81 (3H, s, OCH_3), 3.89 (3H, s, OCH_3)
VId-10 (Na)	1260, 1050 (SO_2)	275, 280	1.21 (16H, br, CH_2), 2.10 and 2.21 (3H, s, CH_3 on the ring), 2.53 (2H, m, CH_2 on the ring), 3.53 (2H, t, J=6 Hz, CH_2O), 3.80 (3H, s, OCH_3), 3.89 (3H, s, OCH_3)
VId-11 (K)	1280, 1240, 1050 (SO_2)	274, 280	1.28 (18H, br, CH_2), 2.11 and 2.22 (3H, s, CH_3 on the ring), 2.61 (2H, m, CH_2 on the ring), 3.53 (2H, t, CH_2O), 3.81 (3H, s, OCH_3), 3.90 (3H, s, OCH_3)
VId-11A (K)	1280, 1240, 1050 (SO_2)	274, 280	1.22 (18H, br, CH_2), 2.22 (3H, s, CH_3 on the ring), 2.55 (2H, m, CH_2 on the ring), 3.52 (2H, t, CH_2O), 3.80 (3H, s, OCH_3), 3.88 (3H, s, OCH_3)
VIId-10 (Na)	1250, 1050 (SO_2)	278	1.20 (16H, br, CH_2), 2.20 (3H, s, CH_3 on the ring), 2.62 (2H, m, CH_2 on the ring), 3.51 (2H, t, CH_2O), 3.82 (6H, s, OCH_3)

Table IX. Physicochemical Data for Potassium Salts of VIc-4A and VIc-10A

Compound No. (salt form)	IR ν_{max}^{KBr} cm^{-1}	^{1}H-NMR (D_2O) δ
VIc-4A (K)	1560 (COOK), 1250, 1050 (SO_2)	1.70 (2H, q, J=7.5 Hz, CH_2), 2.24 (3H, s, CH_3 on the ring), 2.26 (2H, t, CH_2COO), 2.65 (2H, t, J=7.5 Hz, CH_2 on the ring), 3.86 (3H, s, OCH_3), 3.92 (3H, s, OCH_3)
VIc-10A (K)	1570 (COOK), 1250, 1050 (SO_2)	1.29 (14H, br, CH_2), 2.19 (2H, t, CH_2COO), 2.23 (3H, s, CH_3 on the ring), 2.56 (2H, t, CH_2 on the ring), 3.85 (3H, s, OCH_3), 3.93 (3H, s, OCH_3)

pyridine [five-fold excess (v/w) over IIId-*n*] in the presence of a small amount of $Na_2S_2O_4$ under ice-cooling. Sodium or potassium salts of IVd-*n* (n=4, 10 and 11) and Vd-10 were obtained as described for IVa, IVb and IVc-*n*. Sodium salt of Vd-10: wax. *Anal.* Calcd for $C_{21}H_{32}Na_2O_{12}S_2 \cdot 4H_2O$: C, 38.73; H, 6.15; S, 9.84. Found: C, 38.53; H, 5.72; S, 9.31. These salts were hydrolyzed with NaOH or KOH to give alkali metal salts of VId-*n* (n=4, 10 and 11) and VIId-10. The potassium salts of VId-10A and VId-11A were obtained as colorless crystals from the potassium salts of VId-10 and VId-11, respectively, by recrystallization from H_2O. Potassium salt of VId-10A: mp 178—181 °C. *Anal.* Calcd for $C_{19}H_{31}KO_8S$: C, 49.76; H, 6.81; S, 6.99. Found: C, 49.74; H, 6.72; S, 6.93. Potassium salt of VId-11A: mp 181—182 °C. *Anal.* Calcd for $C_{20}H_{33}KO_8S \cdot 1/2H_2O$: C, 49.87; H, 7.11; S, 6.65. Found: C, 49.99; H, 7.00; S, 6.43. Physicochemical data of the alkali metal salts of IVd-10, Vd-10, VId-*n* (n=4, 10 and 11) and VIId-10 are listed in Table VIII.

Synthesis of 6-(3-Carboxypropyl)-2,3-dimethoxy-5-methylhydroquinone 4-Sulfate, Dipotassium Salt (VIc-4A) and 6-(9-Carboxynonyl)-2,3-dimethoxy-5-methylhydroquinone 4-Sulfate, Dipotassium Salt (VIc-10A) by the Elbs Persulfation[9]——One mol of 4-(2-hydroxy-3,4-dimethoxy-6-methylphenyl)butyric acid (XII-4) or 10-(2-hydroxy-3,4-dimethoxy-6-methylphenyl)decanoic acid (XII-10) was dissolved in 10% KOH (5 mol). A 5% aqueous solution of potassium persulfate (1 mol) was slowly added to the above solution under ice-cooling. The mixture was stirred continuously for 2 h and then left standing overnight. After being acidified to pH 5 with dil. HCl, the reaction mixture was extracted with ethyl acetate and the unreacted starting material (XII-4 or XII-10) was recovered (*ca.* 37%). After being adjusted to pH 8 with 10% KOH, the mixture was concentrated *in vacuo*. The residue was dissolved in MeOH and the insoluble materials were removed by filtration. For purification, the methanolic solution was filtered in batches on silica gel (200—400 mesh) and the filtrate was treated three times with charcoal, then evaporated. The residue was purified on a column packed with Sephadex G-15 to obtain the potassium salt of VIc-4A or VIc-10A. The physicochemical data are shown in Table IX.

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