Synthesis and Thromboxane A_2/Prostaglandin H_2 Receptor Antagonistic Activity of Phenol Derivatives

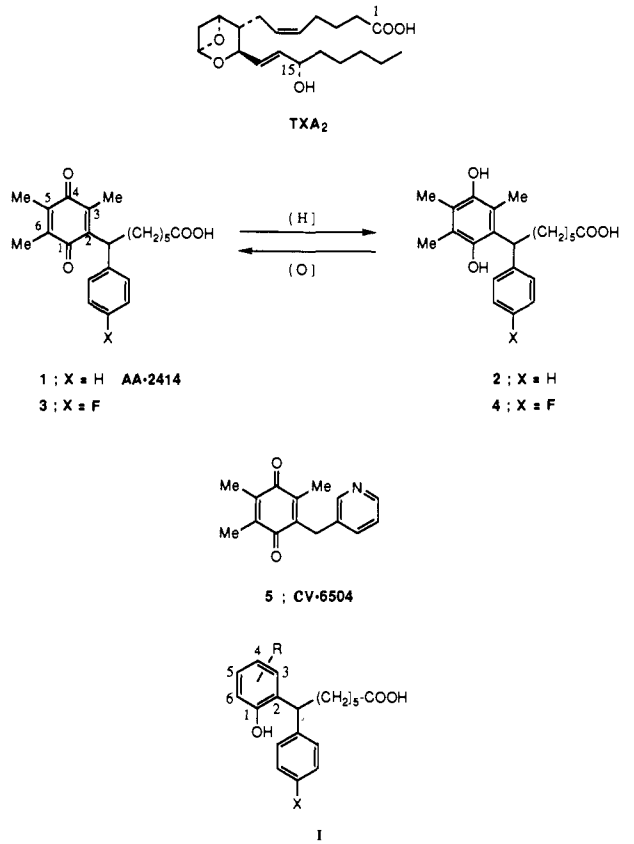
Shoji Fukumoto,† Mitsuru Shiraishi,*,† Zen-ichi Terashita,‡ Yasuko Ashida,‡ and Yoshiyuki Inada‡

Research and Development Division, Takeda Chemical Industries, Ltd., 17-85 Jusohonmachi 2-chome, Yodogawa-ku, Osaka 532, Japan. *Received November 20, 1991*

Consideration of possible structural similarities between thromboxane A_2 and the hydroquinone form of (*R*)-(+)-7-(3,5,6-trimethyl-1,4-benzoquinon-2-yl)-7-phenylheptanoic acid (*R*-(+)-AA-2414) led to the development of a new series of thromboxane A_2/prostaglandin H_2 (TXA_2/PGH_2) receptor antagonists, namely 7-(4-fluorophenyl)-7-(2-hydroxyphenyl)heptanoic acids (I). These compounds were found to be potent TXA_2/PGH_2 receptor antagonists. Compounds having either a carbonyl or a hydroxymethyl group at the para-position of the phenolic hydroxy group exhibited most potent activities in this series. Compounds **14**, **15**, **18**, and **26** inhibited the specific binding of [^{3}H]U-46619 to guinea pig platelet membranes (IC_{50} = 4.4, 80, 32, and 13 nM, respectively), and also inhibited U-46619-induced human platelet aggregation (IC_{50} = 310, 69, 79, and 78 nM, respectively). Comparison of the UV spectra of the compounds with a carbonyl group at the para-position of phenolic hydroxy group revealed that the activity tended to increase in accordance with a decrease in the torsional angle between the carbonyl group and the phenol ring. These results suggested that the spacial location of the carbonyl and hydroxymethyl oxygen are important for significant increase in activity and that the carbonyl and hydroxymethyl oxygen at the para-position of the phenolic hydroxy group might interact with one of the TXA_2/PGH_2 receptor sites.

Thromboxane A_2[1] (TXA_2) is an unstable metabolite of arachidonic acid. It is a potent inducer of platelet aggregation[2] and vascular and pulmonary smooth muscle contraction.[3,4] Consequently, TXA_2 may be involved in a variety of cardiovascular and respiratory diseases,[5] and a number of thromboxane A_2/prostaglandin H_2 (TXA_2/PGH_2) receptor antaonists[6-8] have been developed for the treatment of these diseases. In our continuing synthetic and pharmacological investigations of quinone derivatives,[9-11] we discovered a potent and specific nonprostanoid TXA_2/PGH_2 receptor antagonist, (±)-7-(3,5,6-trimethyl-1,4-benzoquinon-2-yl)-7-phenylheptanoic acid (**1**, AA-2414),[10] which is currently under clinical trial (Chart I). Generally, quinones and hydroquinones are interconvertible by redox reaction within the living body. For example, 2,3,5-trimethyl-6-(3-pyridylmethyl)-1,4-benzoquinone (**5**, CV-6504)[11] and **1**, both of which have quinone moieties, are easily reduced to their hydroquinones in the presence of leukocytes.[12] Furthermore, TXA_2/PGH_2 receptor antagonistic activity of hydroquinones **2** and **4** was greater than that of their quinones. For example, **4** was 7 times more potent than **3** in inhibition of the specific binding of [^{3}H]U-46619 to guinea pig membrane (IC_{50} = 0.2 μM and 1.4 μM, respectively). These facts suggest that the hydroquinone forms **2** and **4** may play a more important role in the development of TXA_2/PGH_2 antagonistic action. In addition, structural comparison between the *R* isomer of **2** and TXA_2 using a computer-assisted graphics method supported the idea that the hydroxy group at the 1-position of the hydroquinone ring of **2** may be more essential than that at the 4-position at inducing antagonistic activity (Chart II). With this knowledge, phenol derivatives (I, Chart I) possessing a hydroxy group at the 1-position[13] were designed in expectation of obtaining higher affinities for the TXA_2/PGH_2 receptor. Herein, we report the synthesis, structure–activity relationships, and biological evaluation of a novel series of 7-(2-hydroxyphenyl)-7-(4-fluorophenyl)heptanoic acids and related compounds.

Chart I



Chemistry

The general synthetic pathways for preparation of compounds listed in Table I are shown in Schemes I–VI.

Phenol derivatives **6–8**, **11**, **12**, **19**, **32**, **34**, **35**, **45**, **46** were synthesized by the acid-catalyzed Friedel–Crafts type al-

* Author to whom correspondence should be addressed.
† Chemistry Research Laboratories.
‡ Biology Research Laboratories.

(1) Hamberg, M.; Svensson, J.; Samuelsson, B. Thromboxane: A New Group of Biologically Active Compounds Derived from Prostaglandin Endoperoxides. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 2994–2998.
(2) Svensson, J.; Hamberg, M.; Samuelsson, B. On the Formation and Effects of Thromboxane A_2 in Human Platelets. *Acta. Physiol. Scand.* **1976**, *98*, 285–294.
(3) Hamberg, M.; Hedqvist, P.; Strandberg, K.; Svensson, J.; Samuelsson, B. Prostaglandin Endoperoxides IV. Effects on Smooth Muscle. *Life Sci.* **1975**, *16*, 451–462.
(4) Svensson, J.; Strandberg, K.; Tuvemo, T.; Hamberg, M. Thromboxane A_2: Effects on Airway and Vascular Smooth Muscle. *Prostaglandins* **1977**, *14*, 425–436.

0022-2623/92/1835-2202$03.00/0

Scheme I

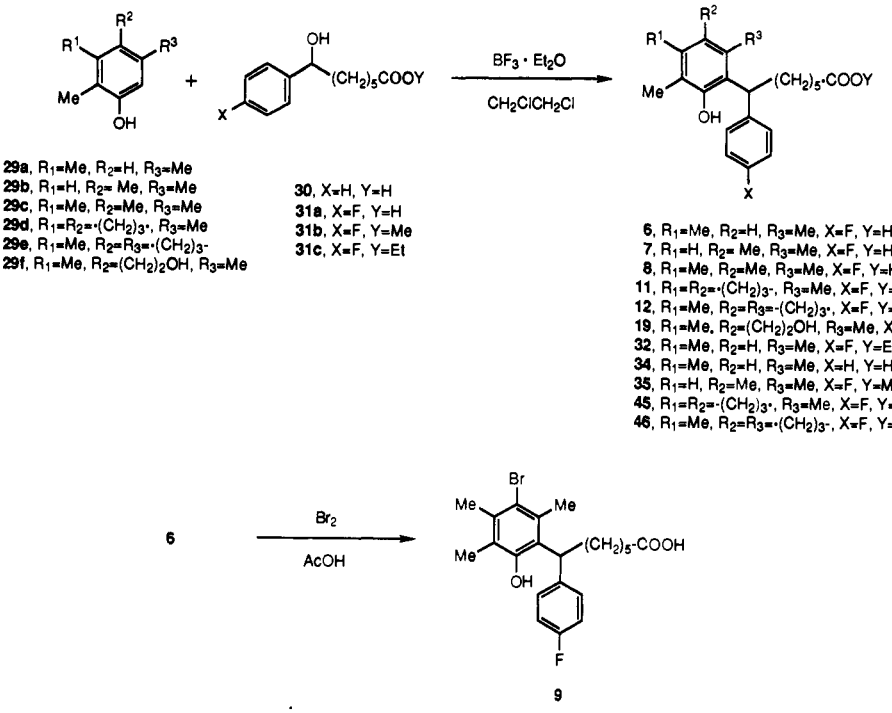
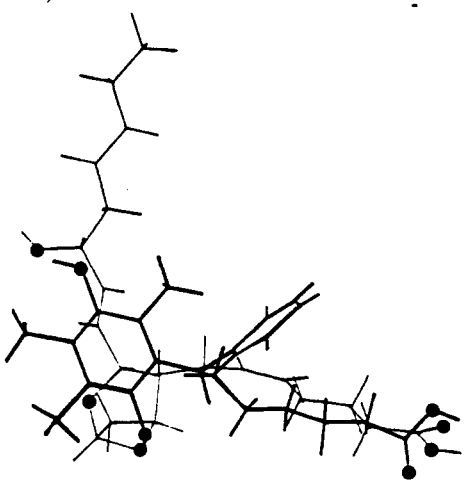


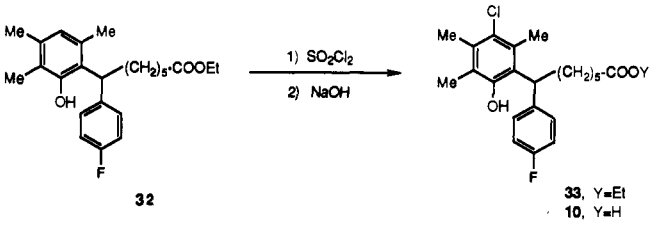
Chart II. Superposition of TXA_2 (lightface) and the *R* Isomer of **2** (boldface)[a]



[a] This chart was obtained by superposition of one of the stable conformation of TXA_2 and that of *R* isomer of **2**. Closed circles represent oxygen atoms.

kylation of various phenols[14] (**29**) with α-substituted benzyl alcohols **30** and **31** (Scheme I). Modification of the phenol moieties of the resulting compounds were subsequently carried out (Scheme I–VI).

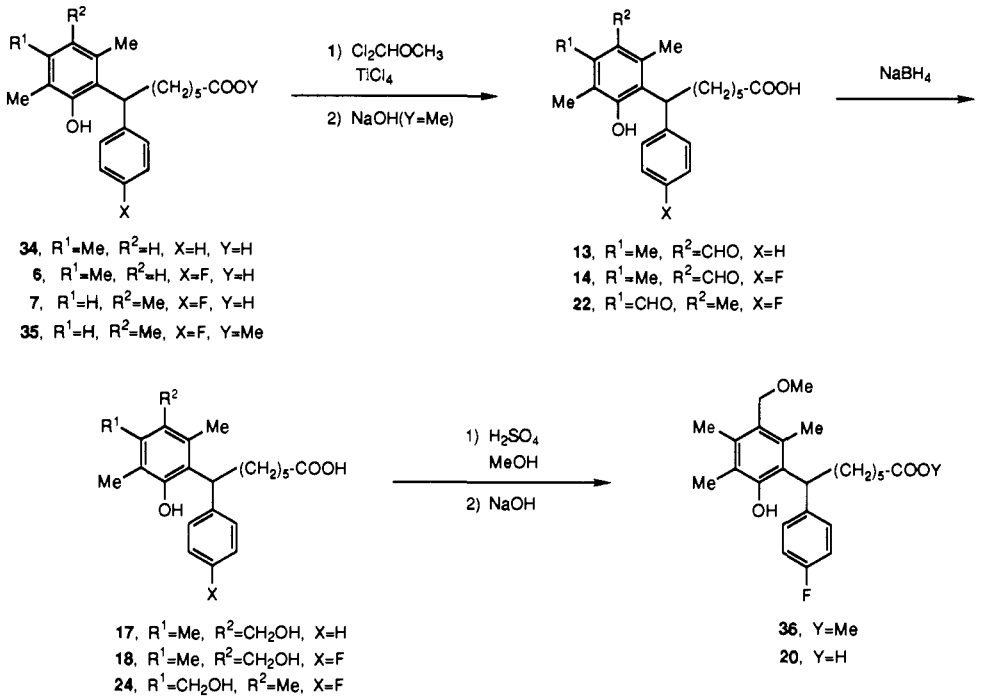
Scheme II



Bromination of **6** in acetic acid gave 4-bromophenol **9** (Scheme I). Chlorination of ethyl ester **32** with SO_2Cl_2, followed by hydrolysis, gave 4-chlorophenol **10** in 73% yield (Scheme II). Formylation of phenols **34** and **6** with Cl_2CHOCH_3–$TiCl_4$[15] in dichloromethane at a temperature of −10 to −12 °C gave 4-formylphenols **13** and **14** in good yields. An attempt to formylate free acid **7** was carried out under the same conditions, but the desired compound **22** could not be isolated. Therefore, aldehyde **22** was obtained by formylation of methyl ester **35** followed by hy-

(5) Fitzgerald, G. A.; Branch, R. A.; Jackson, E. K.; Jackson Roberts, L.; Knapp, H. R.; Oates, J. A. Clinical Implications of Prostaglandin and Thromboxane A_2 Formation. *N. Engl. J. Med.* **1988**, *319*, 689–698 and 761–767.
(6) Cross, P. E. Antithrombotic Agents. In *Annual Report on Medicinal Chemistry*; Hess, H-J., Eds.; Academic Press: New York, 1982; Vol. 17, Chapter 9, 79–88.
(7) Cross, P. E.; Dickinson, R. P. Thromboxane Synthetase Inhibitors and Antagonists. In *Annual Report on Medicinal Chemistry*; Bailey, D. M., Eds.; Academic Press: New York, 1987; Vol. 22, Chapter 10, 95–105.
(8) Hall, Steven E. Thromboxane A_2 Receptor Antagonists. *Med. Res. Rev.* **1991**, *11*, 503–579.
(9) Terao, S.; Shiraishi, M.; Kato, K.; Ohkawa, S.; Ashida, Y.; Maki, Y. Quinones. Part 2. General Synthetic Routes to Quinone Derivatives with Modified Polyprenyl Side Chains and the Inhibitory Effects of These Quinones on the Generation of the Slow Reacting Substance of Anaphylaxis(SRS-A). *J. Chem. Soc. Perkin Trans. 1* **1982**, 2909–2920.
(10) Shiraishi, M.; Kato, K.; Terao, S.; Ashida, Y.; Terashita, Z., Kito, G. Quinones. 4. Novel Eicosanoid Antagonists: Synthesis and Pharmacological Evaluation. *J. Med. Chem.* **1989**, *32*, 2214–2221.
(11) Ohkawa, S.; Terao, S.; Terashita, Z.; Shibouta, Y.; Nishikawa, K. Dual Inhibitors of Thromboxane A_2 Synthase and 5-Lipoxygenase with Scavenging Activity of Active Oxygen Species. Synthesis of a Novel Series of (3-Pyridylmethyl)benzoquinone Derivatives. *J. Med. Chem.* **1991**, *34*, 267–276.
(12) Ohkawa, S.; Terao, T.; Murakami, M.; Matsumoto, T.; Goto, G. Reduction of 2,3,5-Trimethyl-6-(3-pyridylmethyl)-1,4-benzoquinone by PB-3c Cells and Biological Activity of Its Hydroquinone. *Chem. Pharm. Bull.* **1991**, *39*, 917–921.
(13) The same numbering is used for the phenol derivatives and the quinone derivatives through this paper to prevent the number varying with each compound.
(14) 5-Hydroxy-4,7-dimethylindan was prepared from 6-methoxy-4,7-dimethyl-1-indanol; see Experimental Section. 5-Hydroxy-6,7-dimethylindan was synthesized in the same method.
(15) Reiche, A.; Gross, H.; Höft, E. *Chem. Ber.* **1960**, *93*, 88–94.

Scheme III



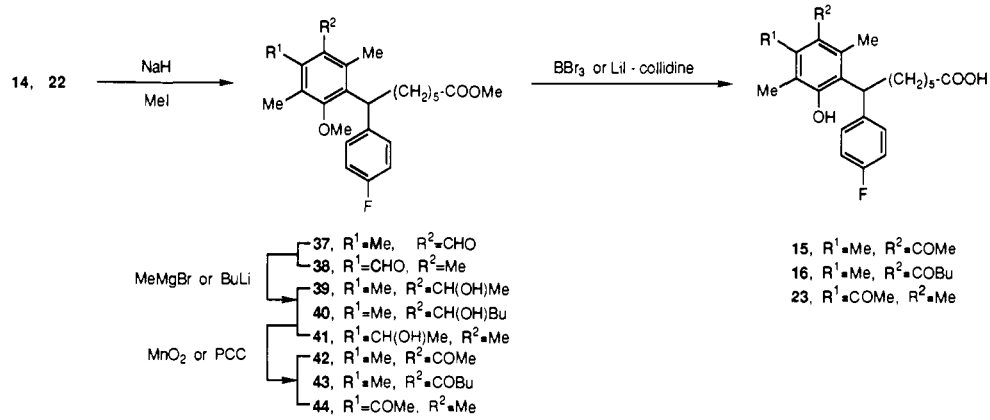
34, R^1=Me, R^2=H, X=H, Y=H
6, R^1=Me, R^2=H, X=F, Y=H
7, R^1=H, R^2=Me, X=F, Y=H
35, R^1=H, R^2=Me, X=F, Y=Me

13, R^1=Me, R^2=CHO, X=H
14, R^1=Me, R^2=CHO, X=F
22, R^1=CHO, R^2=Me, X=F

17, R^1=Me, R^2=CH_2OH, X=H
18, R^1=Me, R^2=CH_2OH, X=F
24, R^1=CH_2OH, R^2=Me, X=F

36, Y=Me
20, Y=H

Scheme IV



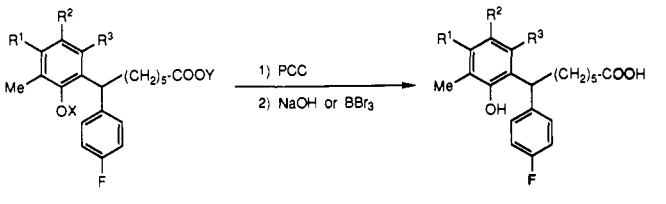
37, R^1=Me, R^2=CHO
38, R^1=CHO, R^2=Me
39, R^1=Me, R^2=CH(OH)Me
40, R^1=Me, R^2=CH(OH)Bu
41, R^1=CH(OH)Me, R^2=Me
42, R^1=Me, R^2=COMe
43, R^1=Me, R^2=COBu
44, R^1=COMe, R^2=Me

15, R^1=Me, R^2=COMe
16, R^1=Me, R^2=COBu
23, R^1=COMe, R^2=Me

drolysis. Formylated phenols **13**, **14**, and **22** were easily reduced to the 4- and 5-(hydroxymethyl)phenols **17**, **18**, and **24** with $NaBH_4$ in THF. Treatment of **18** with a catalytic amount of sulfuric acid in methanol provided 4-(methoxymethyl)phenol **36**, which was subsequently hydrolyzed to **20** (Scheme III). The protected formyl compounds **37** and **38** were allowed to react with MeMgBr or *n*-BuLi to yield diastereoisomeric pairs of the secondary alcohols **39**–**41** in good yields. Little tertiary alcohol byproduct was obtained by the competitive reaction between the ester group of **37** and MeMgBr, and even with *n*-BuLi the yield of the tertiary alcohol was only 7%. Oxidation of **39**–**41** with activated MnO_2 or pyridinium chlorochromate (PCC) gave ketones **42**–**44** in good yields. These were finally demethylated and hydrolyzed (BBr_3[16] or LiI–collidine[17]) to phenols **15**, **16**, and **23** (Scheme IV).

Indanones **25** and **26** and tetralone **27** were obtained by oxidation of indans **45** and **46** and tetraline **47** with PCC[18] in benzene, followed by deprotection (Scheme V). Carboxylic acid **21** was prepared from formylated phenol **48**

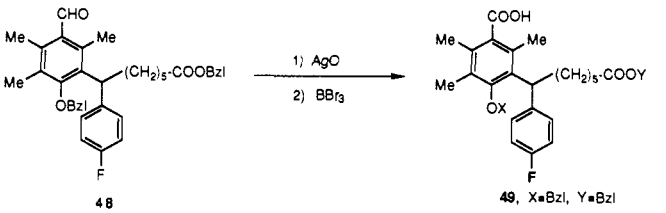
Scheme V



45, R^1=R^2=-$(CH_2)_3$-, R^3=Me, X=H, Y=Me
46, R^1=Me, R^2=R^3=-$(CH_2)_3$-, X=H, Y=Me
47, R^1=Me, R^2=R^3=-$(CH_2)_4$-, X=Me, Y=Me

25, R^1=R^2=-$(CH_2)CO$-, R^3=Me
26, R^1=Me, R^2=R^3=-$CO(CH_2)_2$-
27, R^1=Me, R^2=R^3=-$CO(CH_2)_3$-

Scheme VI



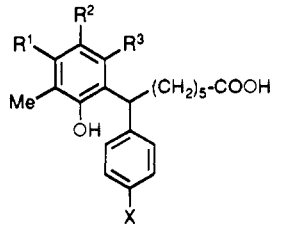
48

49, X=Bzl, Y=Bzl
21, X=H, Y=H

Bzl = CH_2Ph

(16) McOmie, J. F. W.; West, D. E. 3,3′-Dihydroxybiphenyl. *Organic Syntheses*; Wiley: New York, 1973; Collect. Vol. V, p 412.
(17) Harrison, I. T. Cleavage of Alkyl Aryl Ethers with Lithium Iodide. *Chem. Commun.* **1969**, 616.

Table I. Physical Properties and TXA_2 Receptor Binding Activity of Phenols



no.	R^1	R^2	R^3	X	synth method	% yield[a]	mp, °C	recrystn solvent[b]	formula (fw)[c]	IC_{50}, 10^{-8} M (n)[d]
6	Me	H	Me	F	A	89	128–129	A	$C_{22}H_{27}FO_3$ (358.44)	45 (3)
7	H	Me	Me	F	A	78	142–143	A	$C_{22}H_{27}FO_3$ (358.44)	24 (3)
8	Me	Me	Me	F	A	51	161–163	B	$C_{23}H_{29}FO_3$ (372.46)[e]	13 (3)
9	Me	Br	Me	F	B	83	138–140	B	$C_{22}H_{26}BrFO_3$ (437.34)[f]	20 (3)
10	Me	Cl	Me	F	C	73	165–166	A	$C_{22}H_{26}ClFO_3$ (392.88)	84 (3)
11	$-(CH_2)_3-$		Me	F	A	77	164–165	A	$C_{24}H_{29}FO_3$ (384.47)	4.9 (3)
12	Me	$-(CH_2)_3-$		F	A	80	166–167	A	$C_{24}H_{29}FO_3$ (384.47)	16 (3)
13	Me	CHO	Me	H	D	87	185–186	C	$C_{23}H_{28}O_4$ (368.45)[g]	2.0 (2)
14	Me	CHO	Me	F	D	97	202–204	A	$C_{23}H_{27}FO_4$ (386.45)	0.44 (3)
15	Me	Ac	Me	F	E	70	164–165	D	$C_{24}H_{29}FO_4$ (400.47)	8.0 (2)
16	Me	COBu	Me	F	E	53	98–99	E	$C_{27}H_{35}FO_4$ (442.55)	19 (2)
17	Me	CH_2OH	Me	H	D	75	160–161	D	$C_{23}H_{30}O_4$ (370.47)	82 (2)
18	Me	CH_2OH	Me	F	D	70	184–185	D	$C_{23}H_{29}FO_4$ (388.46)	3.2 (2)
19	Me	$(CH_2)_2OH$	Me	F	A	30	144–145	D	$C_{24}H_{31}FO_4$ (402.49)	25 (2)
20	Me	CH_2OMe	Me	F	D	35	141–142	A	$C_{24}H_{31}FO_4$ (402.49)	34 (2)
21	Me	COOH	Me	F	G	63	159–160	D	$C_{23}H_{27}FO_5$ (402.45)[h]	320 (2)
22	CHO	Me	Me	F	D	47	167–168	D	$C_{23}H_{27}FO_4$ (386.45)	3.0 (2)
23	Ac	Me	Me	F	E	77	129–130	D	$C_{24}H_{29}FO_4$ (400.47)	25 (2)
24	CH_2OH	Me	Me	F	D	64	154–155	D	$C_{23}H_{29}FO_4$ (388.46)	65 (2)
25	$-(CH_2)_2CO-$		Me	F	F	25	245–246	C	$C_{24}H_{27}FO_4$ (398.46)	7.9 (2)
26	Me	$-CO(CH_2)_2-$		F	F	27	225–226	D	$C_{24}H_{27}FO_4$ (398.46)[i]	1.3 (2)
27	Me	$-CO(CH_2)_3-$		F	F	38	160–161	D	$C_{25}H_{29}FO_4$ (412.48)	7.6 (2)
3										140 (3)
4										20 (3)
28	(BM-13505)									6.2 (5)

[a] No attempt was made to optimize yields. Numbers represent the yield for the last step. [b] A = EtOAc/hexane; B = benzene/hexane; C = acetonitrile/THF; D = acetonitrile; E = isopropyl ether/hexane. [c] Analytical results are within ±0.4% of theoretical values unless otherwise noted. [d] The IC_{50} is the concentration of compound required to reduce by 50% the specific binding of 4 nM [^{3}H]U-46619 to guinea pig platelet membrane receptors. The n values (in parentheses) are the number of experiments in which a dose–response curve was determined from two to six replicates per dose level. [e] C: calcd, 74.17; found, 73.64. [f] C: calcd, 60.42; found, 61.15. [g] C: calcd, 74.97; found, 74.48. [h] C: calcd, 68.64; found, 67.84. [i] C: calcd, 72.34; found, 71.43.

by oxidation with AgO followed by deprotection of **49** with BBr_3 (Scheme VI).

Pharmacological Results and Discussion

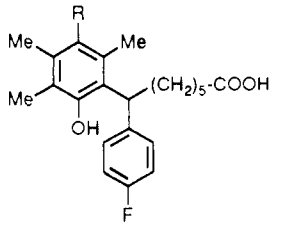
TXA_2/PGH_2 receptor antagonistic activities of the phenol derivatives described above were measured by their ability to inhibit specific binding of [^{3}H]U-46619[19,20] to guinea pig platelet membrane. The results are displayed in Table I as IC_{50} values (i.e., the concentration needed to inhibit the specific binding of [^{3}H]U-46619 by 50%). The activities of **3**, **4**, and nonprostanoid TXA_2/PGH_2 receptor antagonist BM-13505[21] (**28**) are also presented in Table I.

Interestingly, compound **6**, in which the hydroxy group at the 4-position of hydroquinone **4** was removed, maintained activity. Replacement of the C-4 hydroxy group of hydroquinone **4** by Me, Br, and Cl also resulted in retention of activity. The highly lipophilic compound **11** exhibited potent activity (IC_{50} = 49 nM), whereas its structural isomer **12** was less than one-third as active as **11**.

Introduction of a carbonyl group or hydroxymethyl group at the 4- or 5-position of the phenol ring potentiated TXA_2/PGH_2 receptor antagonistic activities. Compound **14**, having a formyl group at the 4-position, exhibited the most potent activity, and compounds **11**, **13**, **18**, **22**, and **26** also exhibited significant activities. Structure–activity relationships for the series of compounds with a carbonyl group (COR) at the 4-position are summarized below. The activity decreases in accordance with increasing size of the substituent R; i.e., IC_{50} value of **14** (R = H), **15** (R = CH_3), and **16** (R = Bu) were 4.4, 80, and 190 nM, respectively. There was a difference between the activity of the two indanone derivatives **25** and **26**. Compound **26** was about 6 times more potent than **25**. This order was opposite to that obtained in the corresponding indans **11** and **12**. The optimum number (n) of methylene groups between the phenol ring and the hydroxy group was 1 [**18** (n = 1) > **4** (n = 0) ≥ **19** (n = 2)]. Compound **18**, which has a hydroxymethyl group, was about 10 times more potent than compound **20**, which has a methoxymethyl group. Both compounds having carbonyl at the 4-position of phenol ring were more potent than the corresponding 5-substituted compounds (**14** > **22**, **15** > **23**, **18** > **24**).

(18) Rathore, R.; Saxena, N.; Chandrasekaran, S. A Convenient Method of Benzylic Oxidation with Pyridinium Chlorochromate. *Synth. Commun.* **1986**, *16*, 1493–1498.
(19) Bundy, G. L. The Synthesis of Prostaglandin Endoperoxide Analogs. *Tetrahedron Lett.* **1975**, 1957–1960.
(20) Coleman, R. A.; Humphrey, P. P. A.; Kennydy, I.; Levy, G. P.; Lumley, P. Comparison of the Actions of U-46619, a Prostaglandin H_2 Analogue, with these of Prostaglandin H_2 and Thromboxane A_2 on Some Isolated Smooth Muscle Preparations. *Br. J. Pharmacol.* **1981**, *73*, 773–778.
(21) Staiger, C.; Uebis, V.; Kaufman, B.; Brindley, F.; Neugebauer, G. Dose-range-finding Study on BM-13505, a New Thromboxane A_2 Receptor Antagonist, in Healthy Volunteers. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1986**, *332*, 385.

Table II. λ_{max} and ϵ Values in UV Spectra[a] and TXA_2 Receptor Binding Activity of Phenols



no.	R	λ_{max}, nm	ϵ	IC_{50},[b] nM
14	CHO	278	11750	4.4
15	$COCH_3$	267	3200	80
		273	2960	
16	$CO(CH_2)_3CH_3$	266	2920	190
		273	2790	
26[c]		272	13190	13

[a] Spectra were recorded in THF. [b] Inhibition of specific binding of [^{3}H]U-46619 to guinea pig platelet membrane. See Table I. [c] Structure is given in Scheme V.

These findings indicate that TXA_2/PGH_2 receptor antagonistic activity of phenol derivatives is influenced by the position of substitution of a carbonyl or hydroxymethyl group on the phenol ring. Especially, the findings about indanones **25** and **26** as well as the importance of the number of methylene groups in the compound with hydroxy groups suggest that the spacial position of the carbonyl or hydroxyl oxygen is important for significant enhancement of activity.

The importance of the spacial position of the oxygen has been demonstrated by additional experiments. The UV spectra of compounds having a carbonyl group at the 4-position of the phenol ring (**14–16, 26**) were measured. The λ_{max} and ϵ values of he $\pi \rightarrow \pi^*$ transition of these compounds are presented in Table II along with their antagonistic activity. Their λ_{max} were little affected by the carbonyl group (R), whereas their ϵ values decreased in accordance with increasing size of R, accompanied by a decrease in activity. Compounds **14** and **26** exhibited large ϵ values and potent activities, whereas **15** and **16** exhibited small ϵ values and relatively weak activities. This might be attributable to the deviation of the carbonyl group from the phenol ring's plane due to steric hindrance between the carbonyl group and the 3- and 5-position methyl groups.[22-24] The carbonyl group of **14** is on the phenol ring's plane, and the carbonyl groups of **15** and **16** are out of the phenol ring's plane. Indanone **26**, in which the carbonyl group may be fixed in the same plane as the phenol ring, exhibited a larger ϵ value and more potent activity than that of the acetyl derivative **15**, equal in size to **26**. Consequently, it is important that the carbonyl oxygen exists on the phenol ring's plane for potent TXA_2/PGH_2 receptor antagonistic activity. From these results we imagine the interaction site which interacts with the carbonyl or hydroxymethyl oxygen of phenol derivatives within the TXA_2/PGH_2 receptor.

(22) Braude, E. A.; Sondheimer, F.; Forves, W. F. Steric Effects in the Electronic Spectra of Organic Compounds. *Nature* **1954**, *173*, 117–119.
(23) Braude, E. A.; Sondheimer, F. Studies in Light Absorption. Part XI. Substituted Benzaldehydes, Acetophenones and Related Compounds. The Effects of Steric Conformation on the Electronic Spectra of Conjugated Systems. *J. Chem. Soc.* **1955**, 3754–3766.
(24) Schubert, W. M.; Sweeney, W. A.; Latovrette, H. K. Spectroscopic and Other Properties of Large Ring Mono- and Dimeric Benzocyclanones Prepared by a High-dilution Friedel–Crafts Reaction. *J. Am. Chem. Soc.* **1954**, *76*, 5462–5466.

Table III. Biological Activities of Phenols

no.[c]	IC_{50}, 10^{-8} M: rabbit aorta[a] (n = 6–15)	IC_{50}, 10^{-8} M: human platelet[b] (n = 5)
11	23 (7.2–65.5)	31 (12–80)
13	19 (8.4–51.0)	140[d] (30–620)
14	1.0 (0.42–2.9)	31[d] (15–66)
15	27 (9.2–57.4)	6.9 (4.4–10)
18	3.2 (3.0–3.5)	7.9 (6.1–10)
26	2.7 (2.1–3.3)	7.8 (5.6–11)
28 (BM-13505)	12 (9.9–14.0)	64 (47–87)

[a] The IC_{50} is the concentration of compound required to reduced U-46619 (3×10^{-8} M)-induced contraction of the rabbit aorta by 50%. 95% confidence limits determined by probit analysis are in parentheses. [b] The IC_{50} is the concentration of compound required to inhibit U-46619-induced aggregation of human platelet by 50%. 95% confidence limits determined by probit analysis are in parentheses. [c] Structures are given in Table I. [d] These lower activities might be attributable to the lability of the formyl group due to use of intact cells in the assay.

Compounds **14** and **17** with a fluoro atom at the 4-position of phenyl group showed potent activity, in comparison with the corresponding unsubstituted compounds **13** and **18**. This tendency agreed with that obtained in the previous quinone series. Compound **21** having a 4-carboxyl group, showed poor activity (IC_{50} = 3.2 μM). It might be attributable to the acidity and polarity of a carboxyl group.

Selected compounds with carbonyl or hydroxymethyl groups at the 4-position of phenol ring were further evaluated for their inhibitory effects on U-46619-induced contraction of rabbit aorta in vitro and U-46619-induced human platelet aggregation in vitro. These compounds exhibited potent TXA_2/PGH_2 receptor antagonistic activities in both assays. The results are summarized in Table III. Compound **14** exhibited the most potent activity in the rabbit aorta assay as well as in the guinea pig membrane, but was less active in the human platelet assay. Compounds **18** and **26** also showed significant activities in both in vitro tests. Compound **15** was moderately active in the rabbit aorta and showed the most potent activity (IC_{50} = 69 nM) in the human platelet assay.

In addition, the inhibitory effects on U-46619-induced bronchoconstriction in guinea pigs and rats (in vivo, po) were examined and the results are summarized as percent inhibition at the various doses tested (Table IV). Compounds **14**, **18**, and **26** exhibited very potent activities in both animals in vivo, whereas compounds **13** and **25** were less active.

In conclusion, from the consideration of structures of TXA_2 and hydroquinone form of **1**, we have found the novel phenol derivatives which are more potent than parent quinone compounds in their TXA_2/PGH_2 receptor antagonistic activity. Especially, the phenol derivatives having carbonyl or hydroxymethyl groups at 4-position of the phenol ring, such as the compounds **14**, **15**, **18**, and **26**, are orally active, nonprostanoid TXA_2/PGH_2 receptor antagonists which possess potent TXA_2/PGH_2 receptor antagonistic activities in the various species and tissues tested. We also concluded that the carbonyl or hydroxymethyl oxygen might interact with one of the TXA_2/PGH_2 receptor sites. Further biological and pharmacological evaluation of these compounds are currently in progress.

Experimental Section

Melting points were obtained with a Yanaco micro melting apparatus and are uncorrected. ^{1}H NMR spectra not specified were recorded on a Varian EM-390 spectrometer at 90 MHz in $CDCl_3$ with tetramethylsilane as an internal standard. Where elemental analyses are given, results obtained were within ±0.4% of the theoretical values. Solutions in organic solvents were dried

Table IV. Inhibitory Effect in U-46619-Induced Bronchoconstriction in Vivo

no.[b]	% inhibition[a] guinea pig 5[c]	guinea pig 1.25[c]	guinea pig 0.31[c]	rat 5[c]	rat 1.25[c]
11	76[d]** (9)[e]	37 (9)	–[f]	48** (7)	
13	30 (6)	–	–	–	–
14	84** (10)	67** (10)	33 (10)	68** (6)	54** (7)
15	76** (6)	22 (6)	–	42** (8)	–
18	–	–	57** (7)	65** (7)	40** (8)
25	42** (7)	–	–	–	–
26	95** (7)	59** (7)	–3 (5)	78** (8)	50** (7)
28 (BM-13505)	84** (7)	36* (7)	–	73** (8)	–

[a] Percent inhibition on U-46619-induced bronchoconstriction in guinea pig and rat 1 h after oral administration. [b] Structures are given in Table I. [c] Doses of drug in mg/kg. [d] Significance of differences (Dunnett's test): $*P < 0.05$, $**P < 0.01$ (vs control). [e] The number in parentheses shows the number of animals tested. [f] Not tested.

over anhydrous $MgSO_4$. Column chromatography was carried out on silica gel (Wakogel C-300, particle size 45–75 μm) by the flash chromatography technique. Yields were not maximized. All TLC was run with Merck silica gel 60 (F_{254}) plates.

7-(4-Fluorophenyl)-7-(2-hydroxy-3,4,6-trimethylphenyl)heptanoic Acid (6). To a solution of 2,3,5-trimethylphenol (**29a**, 4.6 g, 33.7 mmol) and 7-(4-fluorophenyl)-7-hydroxyheptanoic acid (**31**, 8.1 g, 33.7 mmol) in 1,2-dichloroethane (95 mL) was added dropwise boron trifluoride–ethyl ether (1.4 g, 10.1 mmol) at 60 °C. The reaction mixture was stirred for 6 h at the same temperature. After cooling, the mixture was diluted with $CHCl_3$ and washed successively with saturated aqueous $NaHCO_3$, water, dilute HCl, water, and brine, dried, and evaporated. The residue was recrystallized from isopropyl ether (IPE)–hexane to give **6** (10.8 g, 89%): TLC (EtOAc–hexane, 1:10) R_f = 0.2; ^{1}H NMR δ 1.00–1.90 (m, 6 H), 1.92–2.48 (m, 4 H), 2.01 (s, 3 H), 2.19 (s, 3 H), 2.31 (s, 3 H), 4.34 (t, J = 8.4 Hz, 1 H), 6.62 (s, 1 H), 6.83–7.43 (m, 4 H), 5.3–9.9 (br, 2 H).

7-(5-Bromo-2-hydroxy-3,4,6-trimethylphenyl)-7-(4-fluorophenyl)heptanoic Acid (9). To a solution of 7-(4-fluorophenyl)-7-(2-Hydroxy-3,4,6-trimethylphenyl)heptanoic acid (6, 1.32 g, 3.68 mmol) in acetic acid (10 mL) was added dropwise a solution of bromine (0.19 mL, 3.7 mmol) in acetic acid (2 mL) with ice cooling. The solution was stirred at room temperature for 30 min. The mixture was poured into water and extracted with EtOAc. The organic layer was washed with water, dried, and evaporated. The residue was recrystallized from EtOAc–hexane to give **9** (1.04 g, 65%): TLC (EtOAc–hexane, 1:2) R_f = 0.15; ^{1}H NMR δ 1.13–1.82 (m, 6 H), 1.82–2.55 (m, 4 H), 2.12 (s, 3 H), 2.39 (s, 3 H), 2.47 (s, 3 H), 4.52 (t, J = 7.1 Hz, 1 H), 6.80–7.40 (m, 6 H).

Ethyl 7-(5-Chloro-2-hydroxy-3,4,6-trimethylphenyl)-7-(4-fluorophenyl)heptanoate (33). Ethyl 7-(4-fluorophenyl)-7-(2-hydroxy-3,4,6-trimethylphenyl)heptanoate (**32**, 1.0 g, 2.59 mmol) was added to sulfuryl chloride (0.38 g, 2.85 mmol). The reaction mixture was stirred at room temperature for 1 h and poured into ice–water. The product was extracted with EtOAc. The extract was washed in turn with saturated aqueous $NaHCO_3$, water, and brine, dried, and concentrated to yield **33** (1.0 g, 92%) as an oil: TLC (EtOAc–hexane, 1:5) R_f = 0.5; ^{1}H NMR δ 1.00–1.80 (m, 6 H), 1.23 (t, J = 7.5 Hz, 3 H), 1.82–2.50 (m, 4 H, 2.09 (s, 3 H), 2.32 (s, 3 H), 2.40 (s, 3 H), 4.09 (q, J = 7.5 Hz, 2 H), 4.48 (t, J = 7.8 Hz, 1 H), 4.51 (s, 1 H), 6.84–7.40 (m, 4 H).

7-(5-Chloro-2-hydroxy-3,4,6-trimethylphenyl)-7-(4-fluorophenyl)heptanoic Acid (10). To the solution of the above **33** (1.0 g, 2.38 mmol) in MeOH (5 mL) was added 1 N sodium hydroxide (3.9 mL) and the mixture was stirred at room temperature for 15 h. The solvent was removed under reduced pressure. The residue was acidified with 1 N HCl and extracted with EtOAc. The organic layer was washed with water, dried, and evaporated. The residue was recrystallized from EtOAc–hexane to afford **10** (0.74 g, 79%): TLC (EtOAc–hexane, 1:2) R_f = 0.15); ^{1}H NMR δ 1.00–1.78 (m, 6 H), 1.80–2.45 (m, 4 H), 2.21 (s, 3 H), 2.29 (s, 3 H), 2.34 (s, 3 H), 4.56 (t, J = 7.2 Hz, 1 H), 6.60–7.70 (m, 6 H).

7-(4-Fluorophenyl)-7-(3-formyl-6-hydroxy-2,4,5-trimethylphenyl)heptanoic Acid (14). To a solution of **6** (7.5 g, 20.9 mmol) and dichloromethyl methyl ether (7.2 g, 62.7 mmol) in CH_2Cl_2 (80 mL) was added to a solution of $TiCl_4$ (11.9 g, 62.7 mmol) in CH_2Cl_2 (25 mL) dropwise in the temperature range of –10 to –12 °C. The mixture was stirred for 20 min and poured into ice–water. The mixture was extracted with $CHCl_3$, and the organic layer was washed with water, dried, and evaporated. The residue was recrystallized from THF–acetonitrile to afford **14** (7.8 g, 97%): TLC (EtOAc–hexane, 2:1) R_f = 0.6; ^{1}H NMR (DMSO-d_6) δ 0.80–1.67 (m, 6 H), 1.90–2.56 (m, 4 H), 2.09 (s, 3 H), 2.31 (s, 3 H), 2.34 (s, 3 H), 3.40 (br s, 1 H), 4.59 (t, J = 7.2 Hz, 1 H), 6.90–7.35 (m, 4 H), 8.79 (s, 1 H), 10.46 (s, 1 H).

7-(4-Fluorophenyl)-7-[2-hydroxy-5-(hydroxymethyl)-3,4,6-trimethylphenyl]heptanoic Acid (18). To a solution of **14** (1.2 g, 3.11 mmol) in THF (30 mL) was added $NaBH_4$ (0.059 g, 1.6 mmol) at 0 °C. The mixture was stirred at room temperature for 2 h and quenched with acetone. The solvent was removed under reduced pressure, and water was added to the residue. The mixture was acidified with 1 N HCl and extracted with EtOAc. The extract was washed with water, dried, and evaporated. The residue was recrystallized from THF–acetonitrile to provide **18** (0.85 g, 70%): TLC (EtOAc–hexane, 2:1) R_f = 0.4; ^{1}H NMR δ 1.00–1.75 (m, 6 H), 1.85–2.60 (m, 4 H), 2.07 (s, 3 H), 2.21 (s, 3 H), 2.24 (s, 3 H), 2.60–3.70 (br s, 1 H), 4.38–4.73 (m, 3 H), 6.75–7.40 (m, 6 H).

Methyl 7-(4-Fluorophenyl)-7-[2-hydroxy-5-(methoxymethyl)-3,4,6-trimethylphenyl]heptanoate (36). To a solution of **18** (0.67 g, 1.72 mmol) in MeOH (20 mL) was added concentrated H_2SO_4 (0.09 mL) at 0 °C. The mixture was stirred at room temperature for 30 min and neutralized with aqueous $NaHCO_3$. MeOH was removed under reduced pressure. The residue was extracted with EtOAc, and the organic layer was washed with water, dried, and evaporated. The residue was chromatographed on silica gel with EtOAc–hexane (1:18) as eluent to afford **36** (0.5 g, 70%) as an oil: TLC (IPE) R_f = 0.6; ^{1}H NMR δ 1.03–1.79 (m, 6 H), 1.85–2.48 (m, 4 H), 2.06 (s, 3 H), 2.29 (s, 3 H), 2.35 (s, 3 H), 3.42 (s, 3 H), 3.63 (s, 3 H), 4.30–4.65 (m, 1 H), 4.45 (s, 1 H), 4.56 (s, 1 H), 6.83–7.40 (m, 4 H).

Methyl 7-(4-Fluorophenyl)-7-(3-formyl-6-methoxy-2,4,5-trimethylphenyl)heptanoate (37). A solution of **14** (5.4 g, 14.0 mmol) in DMF (40 mL) was added dropwise to a stirred suspension of 60% NaH (1.2 g, 29.4 mmol, washed three times with hexane) in DMF (26 mL) at 0 °C. The mixture was stirred at room temperature for 30 min, MeI (4.18 g, 29.4 mmol) was added dropwise at 0 °C, and the mixture then stirred for 1 h at room temperature. Water was slowly added and the mixture was extracted with EtOAc. The organic layer was washed with water, dried, and evaporated to give **37** (5.7 g, 99%) as an oil: TLC (EtOAc–hexane, 1:4) R_f = 0.5; ^{1}H NMR δ 0.96–1.83 (m, 6 H), 1.84–2.70 (m, 4 H), 2.29 (s, 3 H), 2.30 (s, 3 H), 2.40 (s, 3 H), 3.30 (s, 3 H), 3.63 (s, 3 H), 4.60 (t, J = 7.5 Hz, 1 H), 6.80–7.30 (m, 4 H), 10.61 (s, 1 H).

Methyl 7-(4-Fluorophenyl)-7-[3-(1-hydroxyethyl)-6-methoxy-2,4,5-trimethylphenyl]heptanoate (39). To a solution of **37** (1.0 g, 2.41 mmol) in anhydrous THF (20 mL) was added dropwise a solution of MeMgBr (14.4 mmol) in THF (5 mL) at –78 °C. The mixture was quenched with aqueous $KHSO_4$ and extracted with EtOAc. The organic layer was washed with water, dried, and evaporated. The residue was chromatographed on silica gel with EtOAc–hexane (1:5) as eluent to yield **39** (0.73 g, 71%) as an oil: TLC (EtOAc–hexane, 1:4) R_f = 0.3; ^{1}H NMR δ 0.95–2.50 (m, 10 H), 1.53 (s, 3 H), 1.59 (s, 3 H), 2.19 (s, 6 H), 2.36 and 2.38

(3 H), 3.31 (s, 3 H), 3.63 (s, 3 H), 4.52 (t, J = 7.2 Hz, 1 H), 5.42 (q, J = 6.9 Hz, 1 H), 6.79–7.33 (m, 4 H).

Methyl 7-(3-Acetyl-6-methoxy-2,4,5-trimethylphenyl)-7-(4-fluorophenyl)heptanoate (42). To a solution of **39** (1.4 g, 3.25 mmol) in benzene (14 mL) was added activated MnO_2 (14.0 g) and the mixture was vigorously stirred for 1.5 h. The catalyst was filtered off and was washed with ethanol. The combined liquid was concentrated to obtain **42** (1.3 g, 93%) as an oil: TLC (EtOAc–hexane, 1:2) R_f = 0.6; ^{1}H NMR δ 0.90–1.83 (m, 6 H), 1.85–2.70 (m, 4 H), 2.01 (s, 3 H), 2.10 (s, 3 H), 2.16 (s, 3 H), 2.62 (s, 3 H), 3.28 (s, 3 H), 3.63 (s, 3 H), 4.51 (t, J = 7.2 Hz, 1 H), 6.77–7.35 (m, 4 H).

7-(3-Acetyl-6-hydroxy-2,4,5-trimethylphenyl)-7-(4-fluorophenyl)heptanoic Acid (15). A solution of **42** (1.6 g, 3.73 mmol) in CH_2Cl_2 (15 mL) was added dropwise to a solution of BBr_3 (1.4 mL) in CH_2Cl_2 (15 mL) at −78 °C. The solution was allowed to warm slowly to room temperature and stirred for 5 h. Ice–water was added to the mixture at 0 °C. The organic layer was washed with water, dried, and evaporated. The residue was recrystallized from EtOAc–hexane to afford 15 (0.87 g, 58%): TLC (EtOAc–hexane, 2:1) R_f = 0.5; ^{1}H NMR δ 1.06–1.77 (m, 6 H), 1.90–2.45 (m, 4 H), 2.03 (s, 3 H), 2.10 (s, 3 H), 2.20 (s, 3 H), 2.45 (s, 3 H), 4.37 (t, J = 8.1, 1 H), 4.5–10.0 (br s, 1 H), 6.81–7.38 (m, 4 H).

Methyl 7-(4-Fluorophenyl)-7-[3-(1-hydroxypentyl)-6-methoxy-2,4,5-trimethylphenyl]heptanoate (40). To a solution of **37** (3.2 g, 7.72 mmol) in THF (70 mL) was added n-butyllithium (1.6 M hexane solution, 6.4 mL, 10.2 mmol) dropwise at −78 °C. The reaction mixture was stirred for 1 h at the same temperature. The mixture was quenched with aqueous $KHSO_4$ and extracted, with EtOAc. The organic layer was washed with water, dried, and evaporated. The residue was chromatographed on silica gel with EtOAc–hexane (1:20) as eluent to afford **40** (2.0 g, 53%) as an oil: TLC (EtOAc–hexane, 1:4) R_f = 0.4; ^{1}H NMR δ 0.86 (t, J = 6.1 Hz, 3 H), 1.00–2.65 (m, 17 H), 2.20 (s, 6 H), 2.38 (s, 3 H), 3.30 (s, 3 H), 3.63 (s, 3 H), 4.60 (t, J = 7.5 Hz, 1 H), 5.21 (dd, J = 5.7, 8.7 Hz, 1 H), 6.78–7.37 (m, 4 H).

7-(4-Fluorophenyl)-7-(2-hydroxy-3,4,6-trimethyl-5-valerylphenyl)heptanoic Acid (16). A mixture of methyl 7-(4-fluorophenyl)-7-(2-methoxy-3,4,5-trimethyl-5-valerylphenyl)-heptanoate (43, 1.2 g, 2.49 mmol), collidine (1.8 g, 14.9 mmol), and LiI (1.0 g, 7.5 mmol) was heated to reflux for 20 h. After being cooled, the mixture was acidified by addition of 1 N HCl and extracted with EtOAc. The extract was washed with water, dried, and evaporated. The residue was chromatographed on silica gel with EtOAc–hexane (1:20) as eluted to afford 16 (0.70 g, 63%): TLC (EtOAc–hexane, 1:4) R_f = 0.6; ^{1}H NMR δ 0.93 (t, J = 6.6 Hz, 3 H), 1.10–1.88 (m, 10 H), 1.95–2.55 (m, 4 H), 2.02 (s, 3 H), 2.06 (s, 3 H), 2.16 (s, 3 H), 2.70 (t, J = 6.9 Hz, 2 H), 4.41 (t, J = 7.7 Hz, 1 H), 5.5–9.5 (br, 1 H), 6.84–7.45 (m, 4 H).

7-(4-Fluorophenyl)-7-(5-hydroxy-6,7-dimethyl-1-oxoindan-4-yl)heptanoic Acid (26). To a solution of methyl 7-(6,7-dimethyl-5-hydroxyindan-4-yl)-7-(4-fluorophenyl)heptanoate (46, 1.4 g, 3.51 mmol) in benzene (40 mL) was added a mixture of PCC (3.8 g, 17.5 mmol) and Celite (7 g), and the mixture was stirred for 2 h. The reaction mixture was filtered, and the filtrate was washed with water, dried, and evaporated. THF (10 mL) and 1 N NaOH (10 mL) were added to the residue, and the mixture was stirred at room temperature for 13 h. The solvent was removed under reduced pressure and the residue was acidified with dilute HCl and extracted with EtOAc. The organic layer was washed with water, dried, and evaporated. The residue was recrystallized from acetonitrile to obtain 26 (0.37 g, 26%): TLC (EtOAc–hexane, 1:1) R_f = 0.1; ^{1}H NMR (DMSO-d_6) δ 1.00–1.60 (m, 6 H), 1.96–2.39 (m, 4 H), 2.09 (s, 3 H), 2.41–2.62 (m, 2 H), 2.50 (s, 3 H), 2.62–2.84 (m, 1 H), 2.84–3.05 (m, 1 H), 4.34 (t, J = 7.2 Hz, 1 H), 6.98–7.17 (m, 2 H), 7.26–7.40 (m, 2 H), 9.02 (s, 1 H), 11.93 (br s, 1 H).

Benzyl 7-[2-(Benzyloxy)-5-carboxy-3,4,6-trimethylphenyl]-7-(4-fluorophenyl)heptanoate (49). A suspension of benzyl 7-[2-(benzyloxy)-5-formyl-3,4,6-trimethylphenyl]-7-(4-fluorophenyl)heptanoate (48, 2.3 g, 4.06 mmol), AgO(II) (4.5 g), dioxane (20 mL), and water (2 mL) was stirred for 12 days at 50 °C. Insoluble materials were filtered off and washed with EtOAc. The combined organic layer was concentrated. EtOAc was added to the residue and the mixture was washed with water, dried, and evaporated. The residue was purified by column chromatography on silica gel with EtOAc–hexane (1:10) as eluent to afford **49** (1.5 g, 63%) as an oil: TLC (EtOAc–hexane, 2:1) R_f = 0.5; ^{1}H NMR δ 0.95–1.78 (m, 6 H), 1.83–2.43 (m, 4 H), 2.14 (s, 3 H), 2.23 (s, 3 H), 2.31 (s, 3 H), 4.42 (dd, J = 12.0, 27.6 Hz, 2 H), 4.65 (t, J = 8.3 Hz, 1 H), 5.09 (s, 2 H), 5.5–6.5 (1 H), 6.77–7.53 (m, 4 H), 7.36 (m, 10 H).

6-Methoxy-4,7-dimethyl-1-indanol (51). To a solution of 6-methoxy-4,7-dimethyl-1-indanone[25] (**50**, 7.0 g, 36.8 mmol) in MeOH (100 mL) and THF (50 mL) was added $NaBH_4$ (1.2 g, 31.7 mmol) at 0 °C and then the mixture was stirred at room temperature for 1 h. The mixture was quenched with acetone (10 mL) and evaporated under reduced pressure. To the residue was added EtOAc and the EtOAc was washed with water, dried, and concentrated to afford **51** (7.0 g, 99%): mp 193–195 °C; TLC (EtOAc–hexane, 1:10) R_f = 0.4; ^{1}H NMR δ 1.55 (s, 1 H), 1.86–3.30 (m, 4 H), 2.23 (s, 6 H), 3.78 (s, 3 H), 5.29 (dd, J = 2.7, 6.3 Hz, 1 H), 6.60 (s, 1 H).

5-Methoxy-4,7-dimethylindan (52). A solution of 6-methoxy-4,7-dimethyl-1-indanol (**51**, 4.0 g, 20.8 mmol) in AcOH (100 mL) was hydrogenated on palladium black (0.6 g) at room temperature for 3 h. The catalyst was removed by filtration and the filtrate was evaporated. The residue was dissolved in EtOAc and washed successively with saturated aqueous $NaHCO_3$ and water, dried, and evaporated to afford **52** (3.5 g, 96%): mp 94–95 °C; TLC (EtOAc–hexane, 1:10) R_f = 0.9; ^{1}H NMR δ 1.71–2.36 (m, 2 H), 2.07 (s, 3 H), 2.19 (s, 3 H), 2.65–2.96 (m, 4 H), 3.73 (s, 3 H), 6.45 (s, 1 H).

5-Hydroxy-4,7-dimethylindan (29d). To a solution of 4,7-dimethyl-5-methoxyindan (**52**, 3.5 g, 19.9 mmol) in CH_2Cl_2 (30 mL) was added a solution of BBr_3 (5.2 g, 20.9 mmol) in CH_2Cl_2 (2 mL) at −78 °C. The solution was allowed to warm slowly to room temperature and stirred at the same temperature for 1 h. The mixture was ice-cooled and ice–water was added. The organic layer was washed with saturated $NaHCO_3$ solution and water and dried. After evaporation, the remaining residue was recrystallized from hexane to afford **29d** (3.2 g, 99%): mp 112–113 °C; TLC (EtOAc–hexane, 1:10) R_f = 0.3; ^{1}H NMR δ 1.80–2.40 (m, 2 H), 2.13 (s, 3 H), 2.16 (s, 3 H), 2.58–2.94 (m, 4 H), 4.50 (br s, 1 H), 6.52 (s, 1 H).

Biological Methods. In Vitro Experiments. [^{3}H]U-46619 Binding to Guinea Pig Platelet Membrane. The experiments were done according to the methods described by Kattelman et al.[26] with slight modifications. A syringe containing 0.315% citrate anticoagulant and 1 mM aspirines final concentrations was used to collect the blood by cardiac puncture from conscious guinea pigs. The PRP (platelet-rich plasma) fraction was collected by centrifuging the blood at 3000 rpm for 5 s at room temperature. The obtained PRP was centrifuged at 4800 rpm for 10 min at 4 °C. Platelet membranes were prepared according to the following procedure: In order to remove the residual plasma protein, the platelet pellet was washed once with 30 mL of buffer containing 25 mM Tris-HCl/5 mM $MgCl_2$ (pH 7.4) and recentrifuged to pellet the platelets. The platelets were then resuspended in 20 mL of the same buffer, and the cells were disrupted by sonication. The sonication was performed on ice using a Kontes Sonicator (Vinjeland, NJ). The platelets were sonicated for a total 90 s with a 15 s burst followed by a 15 s intermission. The sonicated mixture was centrifuged at 100000g for 1 h and the pellet was suspended in the same buffer. The protein concentration was adjusted to 1 mg/mL. The resuspended membrane fraction (0.1 mL) was incubated with 4 nM of [^{3}H]U-46619 and drugs at 25 °C for 30 min. The reaction mixture was filtered through a glass-fiber membrane (GF/C, Whatman). The membranes were quickly washed twice with 5 mL of cold buffer. The radioactivity on the glass filter was measured using a liquid-scintillation counter [Aloka, LSC-900, scintillator containing toluene (12 L), bis-MSB (12 g), DPO (180 g), and nonion (5.16 L)]. Nonspecific binding

(25) Jaysukhlal, R. M.; Meers, S. K.; Suneel, Y. D. Synthesis of Some Indan-1-ones. *J. Chem. Soc. Perkin Trans. 1* **1977**, 2089–2092.

(26) Kattelman, E. J.; Venton, D. L.; Le Breton, G. C. Characterization of U46619 Binding in Unactivated, Intact Human Platelets and Determination of Binding Site Affinities of Four TXA_2/PGH_2 Receptor Antagonists (13-APA, BM 13177, ONO 3708, SQ 29548). *Thromb. Res.* **1986**, *41*, 471.

of [^{3}H]U-46619 to the platelet was estimated in the presence of 10^{-5} M unlabeled U-46619.

Rabbit Aorta Contraction. New Zealand white rabbits (male, 2–3 kg) were killed and their thoracic aorta were placed in Krebs–Henseleit solution of the following composition (mM): NaCl, 118; KCl, 4.75; $CaCl_2$, 2.54; KH_2PO_4, 1.19; $MgSO_4$, 1.19; $NaHCO_3$, 12.5; and glucose, 10.0. Adhering fat and connective tissue were removed and spiral strips of the aorta were prepared. Each strip (2–3 mm in width, 3 cm in length) was mounted in an organ bath containing 20 mL of the Krebs–Henseleit solution, bubbled with 95% O_2–5% CO_2 gas at 37 °C. A resting tension of 2 g was isometrically recorded on an ink writing polygraph (San-ei, RECTI-HORIZ-8k) via a force-displacement transducer (Nihon Kohden, Model SB-1T). The contractile response of the strip to U-46619 (3×10^{-8} M) was examined in the absence or presence of drugs (final concentrations of drugs were from 10^{-5} to 10^{-9} M in 0.01% DMSO). The aortic strip was treated with the agents for 30 min before the addition of U-46619.

U-46619-Induced Human Platelet Aggregation. Platelet aggregation study was done as described before.[27] Blood was collected in 3.8% sodium citrate (1 mL for 9 mL of blood) by cardiac puncture from healthy male volunteers who reported to be free of medication for at least 10 days prior to blood collection. Platelet-rich plasma (PRP) and platelet-poor plasma (PPP) were obtained from the blood by centrifugation at 1000*g* for 5 s and at 1000*g* for 10 min at room temperature, respectively. The platelet density of PRP was adjusted to 400 000 platelets/μL with PPP. Platelet aggregation was measured with a photometer (Hematracer 6, Niko Bioscience) according to the method described by Born.[28] The PRP (250 μL) was preincubated at 37 °C for 2 min and then incubated for 2 min with phenol derivatives, or vehicle (25 μL), followed by stimulation with U-46619 (25 μL). The concentration of U-46619 for aggregation was used to obtain the submaximal aggregation (U-46619: 1–3 μM).

U-46619-Induced Bronchoconstriction in Guinea Pig. Male Hartly guinea pigs were used for experiments. The guinea pig, anesthetized with urethane (1.5 g/kg, ip), was fixed in a dorsal position, subjected to tracheotomy, and connected to a respirator through a cannula. A side branch of the tracheal cannula was connected to a respirator (Harvard apparatus rodent respirator Type 680) at the rate of 70 strokes/min and a constant volume of 3–5 mL.

Inflation pressure was kept constant at 10 cm of H_2O. After treatment with gallamine triethiodide (1 mg/kg, iv), U-46619 (10 μg/kg) was given through a carotid cannula and the airway resistance was measured by the overflow technique of the Konzett–Rössler method.[29] Drugs suspended in a 5% gum arabic solution were given orally 1 h before the treatment with U-46619.

U-46619-Induced Bronchoconstriction in Rat. The bronchoconstriction in male Sprage–Dawley rats induced by U-46619 (30 μg/kg, iv) was examined by the Konzett–Rössler method as described above. Drugs suspended in a 5% gum arabic solution were given orally 1 h before the treatment with U-46619.

Acknowledgment. We wish to thank Dr. S. Terao, Dr. A. Imada, Dr. K. Nishikawa, Dr. T. Aono, and Dr. S. Ohakawa for their encouragement and helpful discussions throughout this work, M. Takamoto for the computer-assisted analysis, and M. Kawamura for the human platelet assay. Thanks are also due to Dr. D. Murphy for reviewing the manuscript.

Registry No. (±)-**6**, 140901-18-4; (±)-**7**, 140874-75-5; (±)-**8**, 140874-76-6; (±)-**9**, 140874-77-7; (±)-**10**, 140874-78-8; (±)-**11**, 140874-79-9; (±)-**12**, 140901-19-5; (±)-**13**, 140874-80-2; (±)-**14**, 140874-81-3; (±)-**15**, 140874-82-4; (±)-**16**, 140874-83-5; (±)-**17**, 140874-84-6; (±)-**18**, 140874-85-7; (±)-**19**, 140874-86-8; (±)-**20**, 140874-87-9; (±)-**21**, 140874-88-0; (±)-**22**, 140874-89-1; (±)-**23**, 140874-90-4; (±)-**24**, 140874-91-5; (±)-**25**, 140874-92-6; (±)-**26**, 140874-93-7; (±)-**27**, 140901-20-8; **29a**, 697-82-5; **29b**, 496-78-6; **29c**, 488-70-0; **29d**, 28567-20-6; **29e**, 28567-19-3; **29f**, 140875-11-2; (±)-**30**, 122114-99-2; (±)-**31a**, 122115-05-3; (±)-**31b**, 140875-12-3; (±)-**31c**, 140875-13-4; (±)-**32**, 140874-94-8; (±)-**33**, 140874-95-9; (±)-**34**, 140874-96-0; (±)-**35**, 140874-97-1; (±)-**36**, 140701-21-9; (±)-**37**, 140874-98-2; (±)-**38**, 140874-99-3; **39**, 121099-68-1; **40**, 121127-02-4; **41**, 140875-00-9; (±)-**42**, 140875-01-0; (±)-**43**, 140875-02-1; (±)-**44**, 140875-03-2; (±)-**45**, 140875-04-3; (±)-**46**, 140875-05-4; (±)-**47**, 140875-06-5; (±)-**48**, 140875-07-6; (±)-**49**, 140875-08-7; **50**, 57122-11-9; (±)-**51**, 140875-09-8; **52**, 140875-10-1; MeMgBr, 75-16-1; BuLi, 109-72-8.

(27) Terashita, Z.; Tsushima, S.; Yoshioka, Y.; Nomura, H.; Inada, Y.; Nishikawa, K. CV-3988—A specific antagonist of platelet activating factor (PAF). *Life Sci.* **1983**, *41*, 1975.

(28) Born, G. V. R. Aggregation of blood platelets by adenosine diphosphate and its reversal. *Nature* **1962**, *194*, 927–929.

(29) Konzett, H.; Rössler, R. *Naunyn-Schmiedebergs Arch. Exp. Pathol. Pharmakol.* **1940**, *195*, 71–74.