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Articles

Design, Synthesis, and Pharmacology of 3-Substituted Sodium Azulene-1-sulfonates and Related Compounds: Non-Prostanoid Thromboxane A_2 Receptor Antagonists

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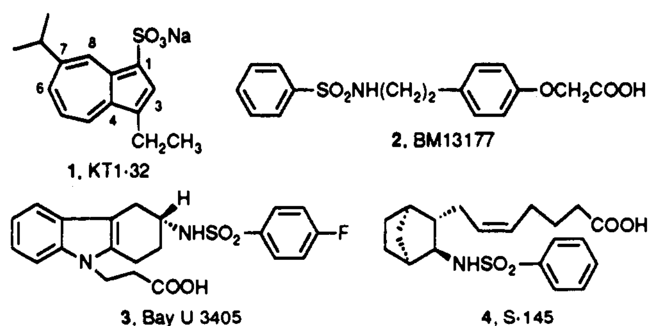
A series of novel azulene-1 carboxylic acid derivatives **28–30**, azulene-1 sulfonic acid sodium salts **41a–c**, and related compounds were synthesized. These compounds were tested for TXA_2 receptor antagonistic activity. The inhibitory concentrations (IC_{50}) of these compounds for vascular contraction (TXA_2 τ receptor) and platelet aggregation (TXA_2 α receptor) induced by (15*S*)-15-hydroxy-11α,9α-(epoxymethano)prosta-5(*Z*),13(*E*)-dienoic acid (U-46619) were obtained. Azulene-1-sulfonic acid sodium salts **41a–c** were over 3 times more potent than azulene-1-carboxylic acids **28–30**. The most potent compound, **41b**, was 4 orders of magnitude more potent than a TXA_2 antagonist, BM13,177, in inhibiting vascular contraction (τ receptor) and had an IC_{50} of 9.0×10^{-10} M. Compound **41b** was also found to be a τ receptor selective antagonist (IC_{50} of contraction/IC_{50} of aggregation = 378) and to have no TXA_2 synthetase inhibitory activity at concentrations up to 10^{-4} M and no partial agonistic activity at concentrations up to 10^{-5} M in rabbit aorta (τ receptor) and up to 10^{-4} M in rabbit platelet-rich plasma (α receptor). In a radioligand binding assay using rabbit gel-filtered platelets, compound **41b** had a high-affinity binding for the TXA_2 receptor. In an in vivo study, compound **41b** inhibited U-46619-induced sudden death in mice at a dose of 0.3 mg/kg and its duration of action was over 8 h when administered orally at 3 mg/kg.

Introduction

TXA_2 has been implicated in the etiology and pathology of many disorders such as coronary vasospasm,[1] myocardial ischemia,[2] asthma,[3] and peptic ulcer.[4,5] TXA_2 receptor antagonists may be more useful than TXA_2 sythetase inhibitors, because these compounds antagonize the effects of TXA_2 and PGH_2 and do not lead to accumulation of endoperoxide intermediates.[6] TXA_2 receptor antagonists also do not inhibit the synthesis of vasodilating prostaglandins like prostacyclin.[7] This imples that the synthetase inhibitors inhibit prostacyclin synthesis, which is incorrect. On the other hand, TXA_2 synthetase inhibitors are less effective in diseases in which TXA_2 may be produced.[6]

During the course of our screening for antiulceratives, we reported an azulene derivative, 3-ethyl-7-isopropylazulene-1-sulfonic acid sodium salt (KT-32, **1**), as an antiulcerative agent.[8] Recently, we have found that compound **1** has a weak TXA_2 antagonistic activity.[9]



Therefore, a derivative of compound **1** may prove to be a novel non-prostanoid TXA_2 antagonist.

In a previous paper,[10] we reported the synthesis and pharmacological activity of 3-substituted sodium azulene-1-sulfonate **41b**. This compound was shown to be a selective thromboxane A_2 (TXA_2) τ-receptor[11] antagonist without partial agonistic activity.

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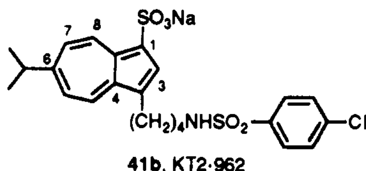
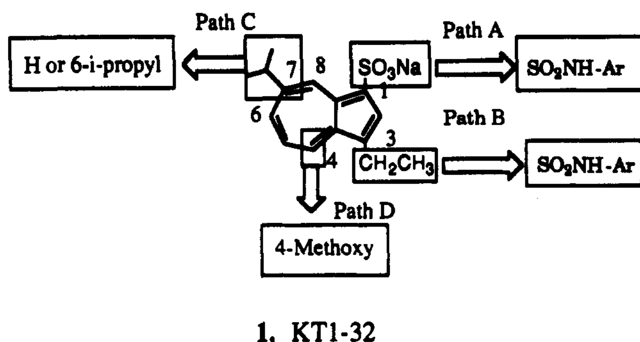


In this paper, we describe the details of design, synthesis, and pharmacology of 3-substituted sodium azulene-1-sulfonate compounds which possess TXA_2 receptor antagonistic activities.

Chemistry

Modification of 1-Sodium Sulfonate (Path A) and the 3-Ethyl Group (Path B). TXA_2 receptor antagonists like BM13,177 (2),[12] S-145,[13] and Bay U 3405 (3)[14] have a sulfonamide group in the molecule and it seems that this group is necessary for the biological activity. Therefore, we introduced an arylsulfonamide group at the 1-position of compound 1 (Path A).



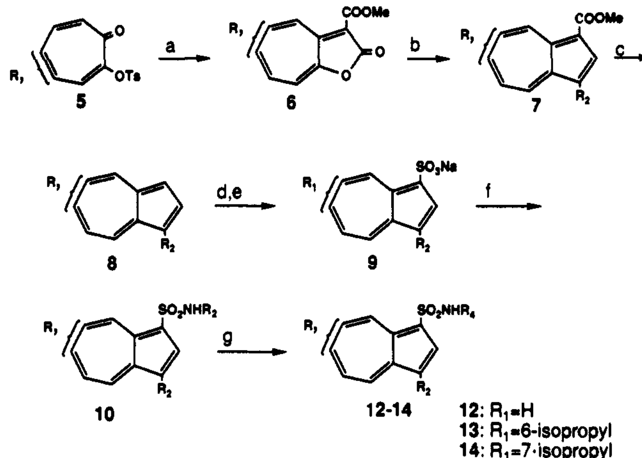
1, KT1-32

Modification of the 3-ethyl group with arylsulfonamide was accomplished by fixing the 1-position of azulene with a carboxylic acid, which was a precursor of azulene-1-sulfonic acid sodium salt as shown in Path B. In addition, the length of alkyl chain in $(CH_2)_nNHSO_2$-aryl at the 3-position of azulene was varied from $n = 2$ to 8 to optimize the biological activity. Furthermore, replacement of the carboxylic acid at the 1-position of azulene with CH_2COOH or CH=CHCOOH was performed to investigate whether these groups have the same activities as sodium sulfonate.

Effects of the 7-Isopropyl (Path C) and 4-Methoxy Group (Path D). To explore the effect of the 7-isopropyl group of compound 1 on TXA_2 receptor antagonistic activity, the activity of 7-isopropyl-substituted azulenes was compared with those of 6-isopropyl-substituted and unsubstituted azulenes (Path C). In the azulene chemistry, modification of the azulene ring was limited to introduction of an isopropyl group at the 6- or 7-position and a methoxy group at the 4-position (Path D).[15] In order to compare the potencies of 7-unsubstituted, 7-isopropyl-, 6-isopropyl-, and 4-methoxyazulene analogues, the substituent group at the 1-position of azulene was fixed with phenoxyacetic acid bearing a sulfonamide.

The synthesis of target compounds was started from the known tosyltropolone 5 and Scheme I outlines the general synthesis of 6,7-unsubstituted, 6-isopropyl, or 7-isopropyl-3-alkylazulene-1-sulfonamides 12–14. We have recently reported the preparation of 7-unsubstituted and 7-isopropyl-3-alkylazulene-1-sulfonic acid sodium salt[8] and 6-isopropyl-3-alkylazulene-1-sulfonic acid sodium salt.[16] Tosyltropolone 5 was reacted with dimethyl malonate in sodium methoxide to give 6- or 7-isopropyl-3-(methoxycarbonyl)-2*H*-cyclohepta[*b*]furan-2-one (6). Condensation of 6 with an in situ generated morpholino enamine of the appropriate aldehyde gave 1-(methoxycarbonyl)azulene 7.[17] Decarboxylation of 7 with 100% phosphoric acid at 95 °C yielded compound 8. Sulfonation of 8 with pyridine–sulfur trioxide complex in benzene, followed by treatment with sodium hydroxide, yielded the corresponding sulfonic acid sodium salt 9. Chlorination of 9 with thionyl chloride and subsequent amide formation with corresponding amine 11 in the usual manner yielded compound 10. Hydrolysis of 10 in 1 N KOH/EtOH gave 1-sulfonamides 12–14 (Table I).

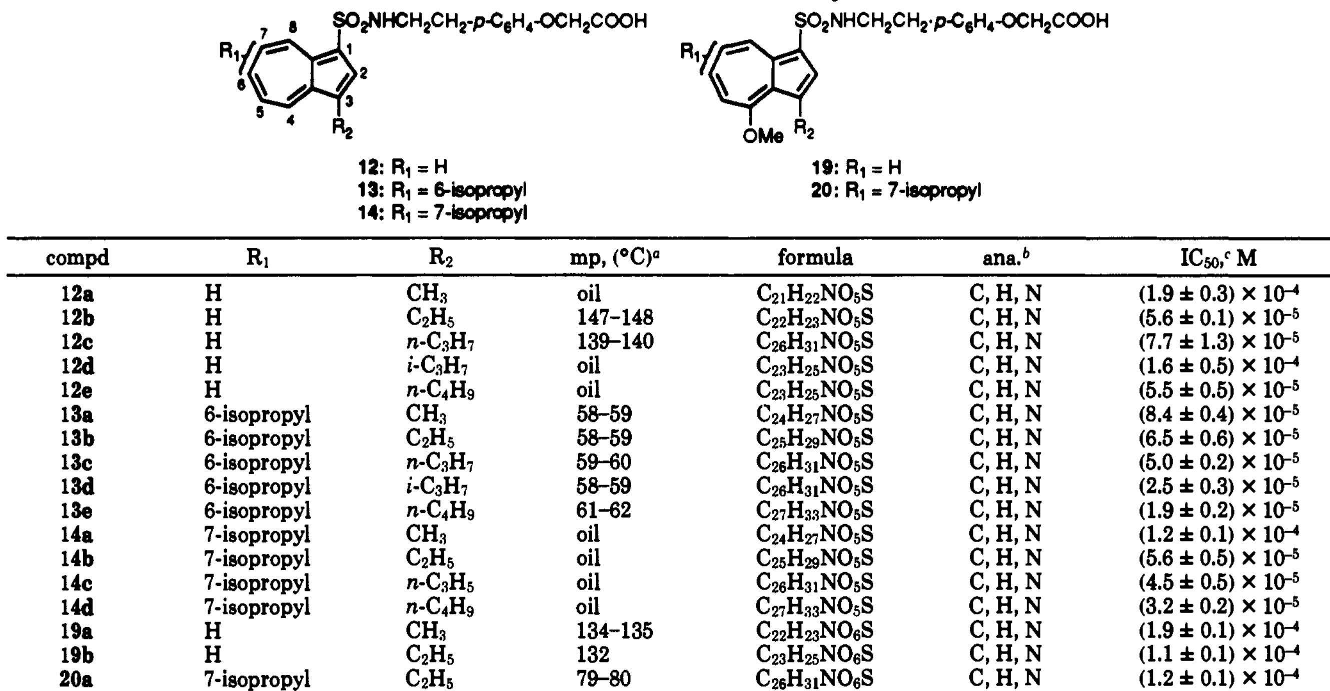
Scheme I[a]



[a] Reagents: (a) dimethyl malonate, MeONa/MeOH, 0 °C; (b) R_2CH_2CHO, morpholine/EtOH, reflux; (c) 100% phosphoric acid, 95 °C; (d) SO_3-Py complex, benzene, reflux; (e) MeONa/MeOH; (f) $SOCl_2$/THF, 0 °C; R_3NH_2/HCl (11), Et_3N, THF/DMF, 0 °C; (g) 1 N KOH/EtOH.

The 4-methoxyazulene derivatives were also prepared according to the method previously reported[8] (Scheme II). The reaction of 5 with diethyl malonate and sodium ethoxide, followed by methylation with diazomethane, yielded the corresponding 4-methoxy-3-(ethoxycarbonyl)-2*H*-cyclohepta[*b*]furan-2-one 15. Under more basic conditions using sodium ethoxide instead of sodium methoxide in this reaction, 4-hydroxy-3-(ethoxycarbonyl)-2*H*-cyclohepta[*b*]furan-2-one 15 was obtained as a major product, whereas 3-(methoxycarbonyl)-2*H*-cyclohepta[*b*]furan-2-one 6 was obtained by using sodium methoxide.[18] Condensation of 15 with morpholino enamine as shown in Scheme II yielded 4-methoxy-1-(ethoxycarbonyl)azulene 16. Compounds 19 and 20 were prepared from 16 in 6 steps similar to those used for the conversion of 7 to 12 and 14 (Table I). The introduction of the sulfonylamino group on the side chain at the 3-position is shown in Scheme III. 6- or 7-isopropyl-3-(methoxycarbonyl)-2*H*-cyclohepta[*b*]furan-2-one 6 reacted with $BnO(CH_2)_nCH_2CHO$ 21 and morpholine to give compound 22, and subsequent debenzylation with $AlCl_3$ in anisole yielded compound 23. Mitsunobu reaction[19] of 23 with 24, followed by treatment with hydrazine hydrate, resulted in the corresponding amino esters 26. Condensation of compound 26 with benzenesulfonyl chloride and hydrolysis with 10% NaOH–EtOH yielded compounds 28–30 (Table II). Alternatively, compound 26 was obtained by the reaction of compound 6 with $CH(O)CH_2(CH_2)_n$-*N*-phthalimide 24 followed by treatment with hydrazine hydrate.

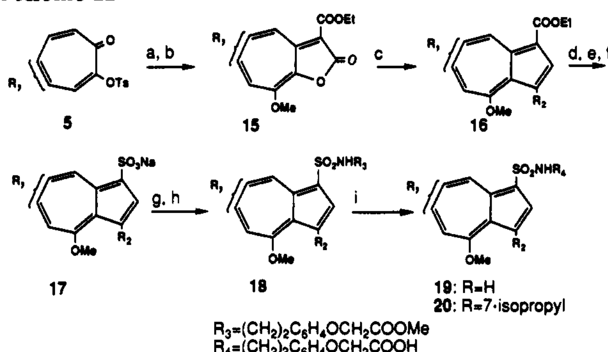
Table I. Unsubstituted and Substituted Azulene-1-sulfonamide 12–14 and 4-Methoxyazulene-1-sulfonamide 19–20



compd	R_1	R_2	mp, (°C)[a]	formula	ana.[b]	IC_{50},[c] M
12a	H	CH_3	oil	$C_{21}H_{22}NO_5S$	C, H, N	$(1.9 \pm 0.3) \times 10^{-4}$
12b	H	C_2H_5	147–148	$C_{22}H_{23}NO_5S$	C, H, N	$(5.6 \pm 0.1) \times 10^{-5}$
12c	H	n-C_3H_7	139–140	$C_{26}H_{31}NO_5S$	C, H, N	$(7.7 \pm 1.3) \times 10^{-5}$
12d	H	i-C_3H_7	oil	$C_{23}H_{25}NO_5S$	C, H, N	$(1.6 \pm 0.5) \times 10^{-4}$
12e	H	n-C_4H_9	oil	$C_{23}H_{25}NO_5S$	C, H, N	$(5.5 \pm 0.5) \times 10^{-5}$
13a	6-isopropyl	CH_3	58–59	$C_{24}H_{27}NO_5S$	C, H, N	$(8.4 \pm 0.4) \times 10^{-5}$
13b	6-isopropyl	C_2H_5	58–59	$C_{25}H_{29}NO_5S$	C, H, N	$(6.5 \pm 0.6) \times 10^{-5}$
13c	6-isopropyl	n-C_3H_7	59–60	$C_{26}H_{31}NO_5S$	C, H, N	$(5.0 \pm 0.2) \times 10^{-5}$
13d	6-isopropyl	i-C_3H_7	58–59	$C_{26}H_{31}NO_5S$	C, H, N	$(2.5 \pm 0.3) \times 10^{-5}$
13e	6-isopropyl	n-C_4H_9	61–62	$C_{27}H_{33}NO_5S$	C, H, N	$(1.9 \pm 0.2) \times 10^{-5}$
14a	7-isopropyl	CH_3	oil	$C_{24}H_{27}NO_5S$	C, H, N	$(1.2 \pm 0.1) \times 10^{-4}$
14b	7-isopropyl	C_2H_5	oil	$C_{25}H_{29}NO_5S$	C, H, N	$(5.6 \pm 0.5) \times 10^{-5}$
14c	7-isopropyl	n-C_3H_5	oil	$C_{26}H_{31}NO_5S$	C, H, N	$(4.5 \pm 0.5) \times 10^{-5}$
14d	7-isopropyl	n-C_4H_9	oil	$C_{27}H_{33}NO_5S$	C, H, N	$(3.2 \pm 0.2) \times 10^{-5}$
19a	H	CH_3	134–135	$C_{22}H_{23}NO_6S$	C, H, N	$(1.9 \pm 0.1) \times 10^{-4}$
19b	H	C_2H_5	132	$C_{23}H_{25}NO_6S$	C, H, N	$(1.1 \pm 0.1) \times 10^{-4}$
20a	7-isopropyl	C_2H_5	79–80	$C_{26}H_{31}NO_6S$	C, H, N	$(1.2 \pm 0.1) \times 10^{-4}$

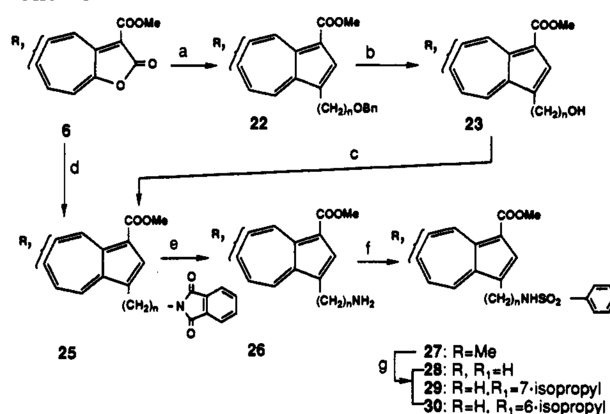
[a] Melting points are uncorrected [b] C, H, N analyses were within ±0.4% of the calculated values. [c] Inhibition of U-46619 (3×10^{-8} M) induced contraction of rat aorta IC_{50} values represent the mean ±SEM and were calculated by regression analysis from the three dose groups of four different preparations.

Scheme II[a]



[a] Reagents: (a) diethyl malonate, EtONa/EtOH, 0 °C; (b) CH_2N_2/THF; (c) R_2CH_2CHO, morpholine/EtOH, reflux; (d) 100% phosphoric acid, 95 °C; (e) SO_3–Py complex, benzene, reflux; (f) MeONa/MeOH; (g) $SOCl_2$/THF, 0 °C; (h) $R_3NH_2{\cdot}HCl$ (**11**), Et_3N, THF/DMF, 0 °C; (i) 1 N KOH/EtOH.

Scheme III[a]



[a] Reagents: (a) $BnO(CH_2)_nCH_2CHO$ (**21**), morpholine/EtOH, reflux; (b) $AlCl_3$/anisole, 0 °C; (c) phthalimide, Ph_3P, EtOOCN=NCOOEt/THF, 0 °C; (d) $CH(O)CH_2(CH_2)_nNpht$ (**24**), morpholine/EtOH; (e) $NH_2NH_2{\cdot}H_2O$/EtOH; (f) $ClSO_2Ph$, $NaHCO_3$/acetone–H_2O; (g) 10% NaOH/MeOH.

The homologation of the carboxylic acid at the 1-position is shown in Scheme IV. Demethoxycarbonylation of **27** with 100% phosphoric acid yielded **31**, which upon homologation using Anderson's stepwise construction[20] yielded azulene-1-acetic acid **34**. Compound **31** was formylated by Vielsmeir–Haack reaction,[21] subjected to Horner–Emmons reaction[22] with triethyl phosphonoacetate, followed by hydrolysis with 10% NaOH, and yielded α,β-unsaturated acid **37** (Table III). The compounds with a phenyl group or an olefin were also prepared **38–40**.

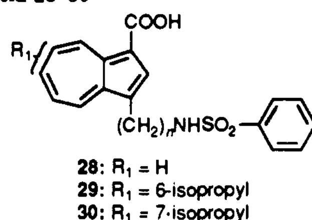
Compound **38** was prepared as shown in Scheme V. The key aldehyde **47** was prepared in a four-step sequence from **43**, prepared from reduction of **42**. **43** was subjected to Wittig reaction to give olefin **44**, followed by benzylation to give **45**. Then, **45** was subjected to hydrogenation and oxidation to give **47**. **47** was condensed with **6** in a same manner outlined in Scheme I to give target compound **38**.

Preparation of *cis*- and *trans*-olefin compounds **39** and **40** is summarized in Scheme VI. The aldehyde intermediates **50** and **53** were obtained after PCC oxidation of reported *cis*-alcohol **49**[23] and *trans*-alcohol **52**.[24] **50** and **53** were reacted with **6** in an analogous manner in Scheme I to give **39** and **40**.

Results and Discussion

The compounds described herein were initially tested for their thromboxane receptor antagonistic activities. They were tested in isolated rat thoracic aorta precontracted by the TXA_2 mimetic (15*S*)-hydroxy-11α,9α-(epoxymethano)prosta-5(*Z*),13(*E*)-dienoic acid, U-46619[25] (3×10^{-9} M). The concentrations that caused 50% relaxation are shown in Tables I–III. The modification of the 1-sulfonic acid group of KT1-32 to a 1-sulfonamide (**12–14**) did not affect TXA_2 receptor antagonistic activity and there was no difference in the activity among the unsubstituted compounds **12a–e**, the 6-substituted derivatives **13a–e**, and the 7-substituted analogues **14a–d** (Table I). At the same time, there appears to be no

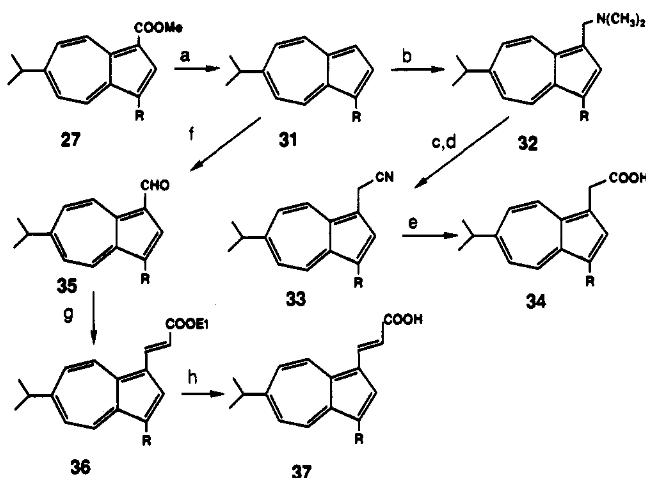
Table II. 3-(Sulfonylamino)azulene-1-carboxylic Acid 28–30



compd	R_1	*n*	mp, (°C)[a]	formula	anal.[b]	IC_{50},[c] M
28a	H	2	193–195	$C_{19}H_{17}NO_4S$	C, H, N, S	$(2.1 \pm 0.3) \times 10^{-5}$
28b	H	3	187–188	$C_{20}H_{19}NO_4S$	C, H, N, S	$(4.8 \pm 0.7) \times 10^{-6}$
28c	H	4	168–169	$C_{21}H_{21}NO_4S$	C, H, N, S	$(5.7 \pm 0.6) \times 10^{-7}$
28d	H	5	170–171	$C_{22}H_{23}NO_4S$	C, H, N, S	$(8.4 \pm 3.1) \times 10^{-7}$
28e	H	6	168–169	$C_{23}H_{25}NO_4S$	C, H, N, S	$(1.4 \pm 0.1) \times 10^{-7}$
28f	H	7	143–144	$C_{24}H_{27}NO_4S$	C, H, N, S	$(1.8 \pm 0.1) \times 10^{-8}$
28g	H	8	182–184	$C_{25}H_{29}NO_4S$	C, H, N, S	$(3.8 \pm 0.4) \times 10^{-8}$
29a	6-isopropyl	2	165–166	$C_{22}H_{23}NO_4S$	C, H, N, S	$(6.0 \pm 0.1) \times 10^{-5}$
29b	6-isopropyl	3	120–122	$C_{23}H_{25}NO_4S$	C, H, N, S	$(4.8 \pm 0.1) \times 10^{-6}$
29c	6-isopropyl	4	167–168	$C_{24}H_{27}NO_4S$	C, H, N, S	$(7.0 \pm 0.6) \times 10^{-8}$
29d	6-isopropyl	5	75–76	$C_{25}H_{29}NO_4S$	C, H, N, S	$(1.2 \pm 0.2) \times 10^{-7}$
29e	6-isopropyl	6	117–118	$C_{26}H_{31}NO_4S$	C, H, N, S	$(7.0 \pm 0.5) \times 10^{-8}$
29f	6-isopropyl	7	119–120	$C_{27}H_{33}NO_4S$	C, H, N, S	$(1.2 \pm 0.2) \times 10^{-7}$
30a	7-isopropyl	2	166–167	$C_{22}H_{23}NO_4S$	C, H, N, S	$(1.5 \pm 0.1) \times 10^{-5}$
30b	7-isopropyl	3	78–80	$C_{23}H_{25}NO_4S$	C, H, N, S	$(7.4 \pm 0.1) \times 10^{-6}$
30c	7-isopropyl	4	142–143	$C_{24}H_{27}NO_4S$	C, H, N, S	$(3.1 \pm 0.2) \times 10^{-6}$
30d	7-isopropyl	5	146–147	$C_{25}H_{29}NO_4S$	C, H, N, S	$(2.6 \pm 0.1) \times 10^{-6}$
30e	7-isopropyl	6	85–88	$C_{26}H_{31}NO_4S$	C, H, N, S	$(9.2 \pm 0.8) \times 10^{-7}$
30f	7-isopropyl	7	124–125	$C_{27}H_{33}NO_4S$	C, H, N, S	$(9.3 \pm 1.2) \times 10^{-7}$

[a] Melting points are uncorrected [b] C, H, N analyses were within ±0.4% of the calculated values. [c] Inhibition of U-46619 (3×10^{-6} M) induced contraction of rat aorta. IC_{50} values represent the mean ±SEM and were calculated by regression analysis from the three dose groups of four different preparations.

Scheme IV[a]

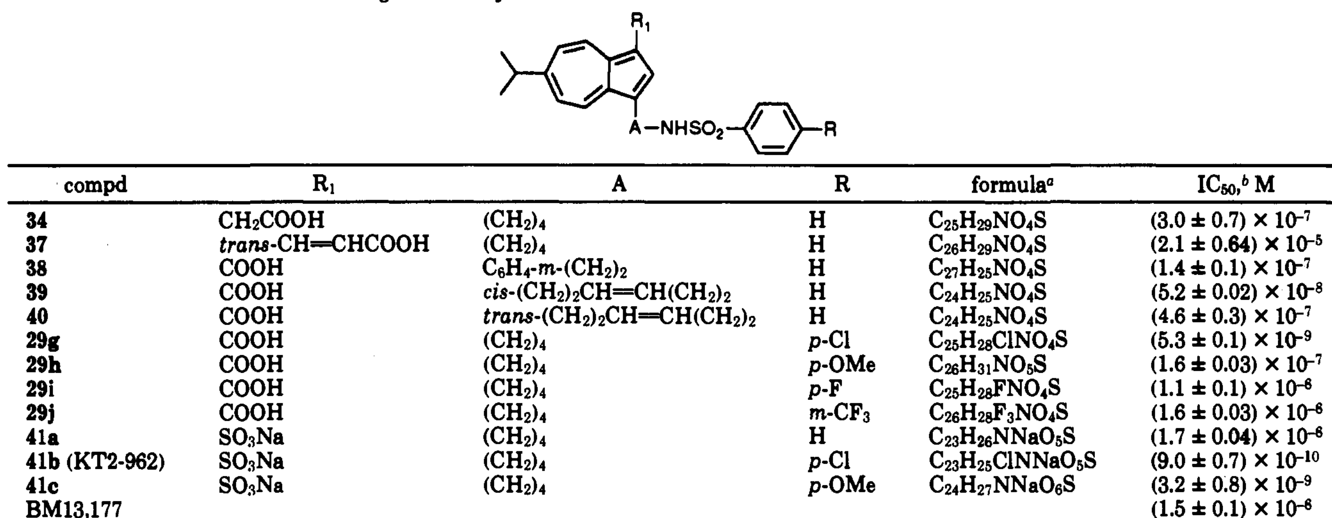


[a] Reagents: (a) 100% phosphoric acid, 120 °C; (b) $(Me)_2NCH_2N(Me)_2$, $(HCHO)_n$, AcOH; (c) MeI; (d) KCN; (e) 0.6 N KOH; (f) $POCl_3$, DMF; (g) $(EtO)_2P(O)CH_2COOEt$, NaH; (h) 10% NaOH; (i) SO_3-Py complex; (j) MeONa/MeOH.

correlation between TXA_2 receptor antagonistic activity and the increase in alkyl chain length at the 3-position of these compounds. The introduction of a 4-methoxy group to the compounds with 1-sulfonamides (**19**) caused a loss in TXA_2 receptor antagonistic activity (IC_{50}) of about 1 order of magnitude from 10^{-5} to 10^{-4} M.

As shown in Table II, the introduction of an (sulfonylamino)alkyl group at the 3-position of compound **1** gave a stepwise increase in activity as the number of methylenes was increased. For example, with compounds **28a–g**, the maximum activity was obtained when the alkyl chain was heptyl ($n = 7$, $IC_{50} = 1.8 \times 10^{-8}$ M). In the 6-isopropyl-substituted compounds **29a–f**, the maximum activity was obtained when the alkyl group was butyl ($n = 4$, **29c**, $IC_{50} = 7.0 \times 10^{-8}$ M). Further homologation of the alkyl chain to heptyl ($n = 7$) did not increase the activity. The activities of 7-isopropyl-substituted compounds **30a–f** were weaker than those of unsubstituted compounds **28a–g** and 6-isopropyl-substituted compounds **29a–f**, and a clear activity relationship with the lengthening of the alkyl chain was not demonstrated. Among the series of unsubstituted, 6-substituted, and 7-substituted compounds, 6-substituted compounds were selected for further optimization. The activities of 6-substituted compounds **29a–f** correlated to alkyl-chain length better than those of unsubstituted or 7-isopropyl-substituted compounds. Among the 6-substituted compounds, the greatest activity was found in compound **29c** as compared to the shorter and the longer chain compounds **29b** and **29d** (Table II). This suggests that there is an optimal chain length of carbon atoms for the maximum biological activity. Compound **29c** was selected for further modification. As shown in Table III, unsaturation of the $(CH_2)_4$ side chain at the 3-position, to yield vinyl- or phenyl-containing compounds **38–40** failed to increase the potency, though cis-olefinic compound **39** retained the same potency as **29c**. Addition of the electron-withdrawing and -donating substituents to the phenyl group of **29c** (compounds **29g** and **29h**) also increased the potencies about 70 and 4 times compared to the potency of compound **29c**. Replacement of the carboxylic acid at the 1-position of azulene (**29c** and **29g**) with sulfonic acid sodium salt (**41a**, $IC_{50} = 1.7 \times 10^{-8}$ M, and **41b**, $IC_{50} = 9 \times 10^{-10}$ M) increased the potency over 3 times compared to that of **29c**. Replacement of the carboxylic group with sodium sulfonate seemed to be useful for providing TXA_2 antagonists. To our knowledge, this is the first example of the use of the sodium sulfonate group to prepare TXA_2 receptor antagonists. Lee et al.[26] reported that a neutral ion pair drug could permeate through hydrophobic membranes due to its lipophilicity, and **41a,b** might be well-

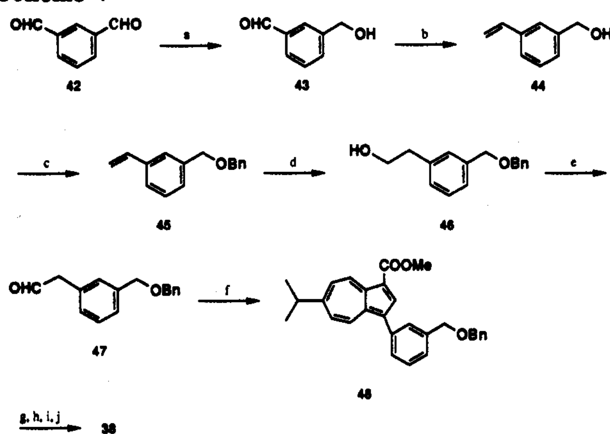
Table III. Structure and Pharmacological Activity in Vitro



compd	R_1	A	R	formula[a]	IC_{50},[b] M
34	CH_2COOH	$(CH_2)_4$	H	$C_{25}H_{29}NO_4S$	$(3.0 \pm 0.7) \times 10^{-7}$
37	*trans*-CH=CHCOOH	$(CH_2)_4$	H	$C_{26}H_{29}NO_4S$	$(2.1 \pm 0.64) \times 10^{-5}$
38	COOH	C_6H_4-*m*-$(CH_2)_2$	H	$C_{27}H_{25}NO_4S$	$(1.4 \pm 0.1) \times 10^{-7}$
39	COOH	*cis*-$(CH_2)_2CH{=}CH(CH_2)_2$	H	$C_{24}H_{25}NO_4S$	$(5.2 \pm 0.02) \times 10^{-8}$
40	COOH	*trans*-$(CH_2)_2CH{=}CH(CH_2)_2$	H	$C_{24}H_{25}NO_4S$	$(4.6 \pm 0.3) \times 10^{-7}$
29g	COOH	$(CH_2)_4$	*p*-Cl	$C_{25}H_{28}ClNO_4S$	$(5.3 \pm 0.1) \times 10^{-9}$
29h	COOH	$(CH_2)_4$	*p*-OMe	$C_{26}H_{31}NO_5S$	$(1.6 \pm 0.03) \times 10^{-7}$
29i	COOH	$(CH_2)_4$	*p*-F	$C_{25}H_{28}FNO_4S$	$(1.1 \pm 0.1) \times 10^{-6}$
29j	COOH	$(CH_2)_4$	*m*-CF_3	$C_{26}H_{28}F_3NO_4S$	$(1.6 \pm 0.03) \times 10^{-6}$
41a	SO_3Na	$(CH_2)_4$	H	$C_{23}H_{26}NNaO_5S$	$(1.7 \pm 0.04) \times 10^{-6}$
41b (KT2-962)	SO_3Na	$(CH_2)_4$	*p*-Cl	$C_{23}H_{25}ClNNaO_5S$	$(9.0 \pm 0.7) \times 10^{-10}$
41c	SO_3Na	$(CH_2)_4$	*p*-OMe	$C_{24}H_{27}NNaO_6S$	$(3.2 \pm 0.8) \times 10^{-9}$
BM13,177					$(1.5 \pm 0.1) \times 10^{-6}$

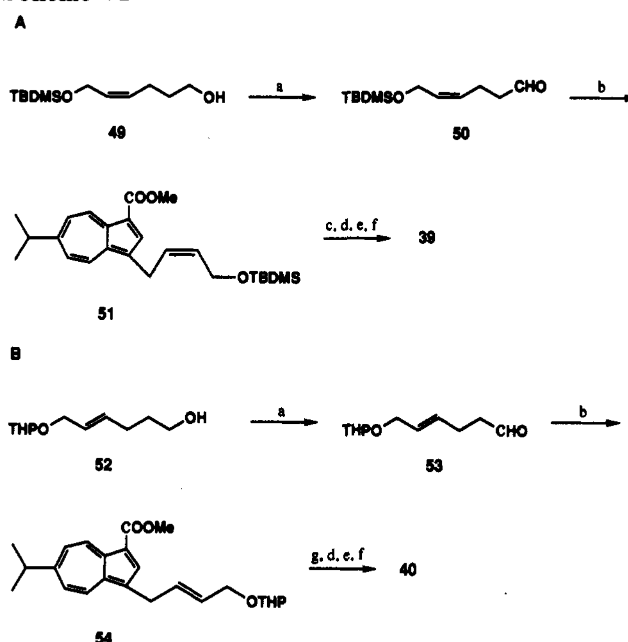
[a] C, H, N analyses were with ±0.4% of the calculated values. [b] Inhibition of U-46619 (3×10^{-8} M) induced contraction of rat aorta. IC_{50} values represent the mean ± SEM and were calculated by regression analysis from the three dose groups of four different preparations.

Scheme V[a]



[a] Reagents: (a) $NaBH_4$, THF, -45 °C, 67%; (b) methyltriphenylphosphonium bromide, *t*-BuOK, THF, rt, 91%; (c) benzyl bromide, NaH, DMF, rt, 94%; (d) (i) 9-BBN, THF, rt; (ii) 6 N NaOH, 31% H_2O_2, rt, 82%; (e) SO_3-Py, NEt_3, DMSO, rt, 91%; (f) **6**, morpholine, EtOH, reflux, 39%; (g) $AlCl_3$, anisole, 0 °C, 87%; (h) PPh_3, CBr_4, CH_2Cl_2, 0 °C, 96%; (i) benzenesulfonamide, NaH, THF-HMPA, reflux, 34%; (j) 10% NaOH, MeOH, reflux, 45%.

Scheme VI[a]



[a] Reagents: (a) PCC, AcONa, CH_2Cl_2, 0 °C; (b) **6**, morpholine, EtOH, reflux; (c) *n*-Bu_4NF, THF, rt; (d) PPh_3, CBr_4, CH_2Cl_2, 0 °C; (e) benzenesulfonamide, NaH, THF-HMPA, reflux; (f) 10% NaOH, MeOH, reflux; (g) AcOH, H_2O, THF, 45 °C.

absorbed from the gastrointestinal tract if this concept of ion formation is correct.

Extension of the carbon chain of the azulene nucleus 1-position (compounds **34** and **37**) decreased the activities. Compound **41b** was the most potent inhibitor of U-46619-induced contraction in rat aorta ($IC_{50} = 9.0 \times 10^{-10}$ M), the pA_2[27] value of **41b** being 9.78 (Schild analysis:[28] slope = 1.04, $n = 4$), and also had no inhibitory effect up to 10^{-5} M on contractile responses induced by norepinephrine, Ca^{2+}, histamine, or serotonin. On the basis of these findings, **41b** is a specific TXA_2 receptor antagonist. Needleman et al. first reported that platelet and vascular TXA_2 receptors might be different,[29] and mediation by the α receptor for aggregation and τ receptor for vasocontraction has been postulated.[11] The selectivities of these compounds for τ (rabbit aorta) versus α [rabbit platelet-rich plasma (PRP)] receptor are shown in Table IV. In this comparison, **29g**, **29h**, **41b**, and **41c** are >667, >233, 378, and 660 times more potent against the τ receptor than the α receptor and are more selective for the τ receptor (higher indices) than BM13,177 in an assay. These findings are in agreement with the report that non-prostanoid TXA_2 antagonists have different structural requirements for α versus τ receptors.[30] A critical part in the development of TXA_2 receptor antagonists is whether these agents possess a partial agonistic activity,[31] and efforts to diminish the partial agonistic activity of TXA_2 receptor antagonists have been described.[32] As shown in Table V, section A, these compounds were evaluated as to whether they possess any partial agonistic activity in rabbit aorta (τ receptor) and in rabbit PRP (α receptor). These compounds had neither partial τ receptor agonistic activities at concentrations up to 10^{-5} M in rabbit aorta nor partial α receptor agonistic activities at concentrations up to 10^{-4} M in rabbit PRP, although other prostanoid TXA_2 receptor antagonists tend to be partial agonists.[33-35]

For further characterization of **41b,c**, the TXA_2 synthetase inhibitor activities of these compounds were examined. **41b** and **41c** had no effect on TXA_2 formation in human platelets when evaluated at concentrations up

Table IV. Inhibition of U-46619-Induced Rabbit Aorta Contraction and Rabbit Platelet Aggregation

compd	R_1	R	IC_{50},[a] M cont[b]	IC_{50},[a] M aggrn[c]	selectivity index:[d] vascular
29c	COH	H	$(1.6 \pm 0.1) \times 10^{-6}$	$>10^{-4}$	>63
29g	COOH	Cl	$(1.5 \pm 0.1) \times 10^{-7}$	$>10^{-4}$	>667
29h	COOH	OMe	$(4.3 \pm 0.5) \times 10^{-7}$	$>10^{-4}$	>233
41a	SO_3Na	H	$(1.1 \pm 0.1) \times 10^{-6}$	$>10^{-4}$	>91
41b (KT2-962)	SO_3Na	Cl	$(2.3 \pm 0.5) \times 10^{-8}$	$(8.7 \pm 0.5) \times 10^{-6}$	378
41c	SO_3Na	OMe	$(9.7 \pm 1.3) \times 10^{-8}$	$(6.4 \pm 0.7) \times 10^{-5}$	660
BM13,177			$(1.3 \pm 0.2) \times 10^{-6}$	$(7.1 \pm 0.4) \times 10^{-6}$	5

[a] IC_{50} values represent the mean ± SEM and were calculated by regression analysis from the three dose groups of four different preparations. [b] Contraction of rabbit aorta was induced by 3×10^{-8} M of U-46619. [c] Aggregation of rabbit platelet-rich plasma was induced by 2.5×10^{-6} M U-46619. [d] The selectivity index for the τ receptor was the difference between the IC_{50} values in the aggregation and contraction assays.

Table V

A. Evaluation of Partial Agonistic Activities

compd	rabbit aorta[a]	rabbit PRP[b]
29h	0	0
41a	0	0
41b (KT2-962)	0	0
41c	0	0

B. Effects of **41b** and **41c** on TXB_2 Formation of Human Platelets

compd	IC_{50},[c] M
41b (KT2-962)	$>10^{-4}$
41c	$>10^{-4}$

C. Displacement of the Specific Binding of $[^3H]$SQ29548[d] to Rabbit Gel-Filtered Platelets

compd	IC_{50},[e] M
41b (KT2-962)	1.4×10^{-8}

[a] Relative values (the value of 40 mM KCl is taken as 100). The concentration of each compound is 10^{-5} M. [b] Relative values (the value of 0.1 μM U-46619 is taken as 100). The concentration of each compound is 10^{-4} M. [c] The concentration which caused 50% inhibition of TXB_2 formation. [d] SQ29,548 is a TXA_2/PGH_2 receptor antagonist; see ref 36. [e] IC_{50} values were calculated as per ref 36 with slight modifications.

to 10^{-4} M (Table V, section B). In a radioligand binding assay with rabbit gel-filtered platelets,[36] compound **41b** inhibited the binding of SQ 29,548[37] with a IC_{50} value of 0.014 μM and possessed high affinity for the TXA_2 receptor (Table V, section C).

In an in vivo experiment of U-46619-induced sudden death in mice, a minimum orally effective dose of **41b** was 0.3 mg/kg and the duration of the effect was over 8 h at a dose of 3 mg/kg, whereas the effective duration of sulotroban was 4 h at a dose of 30 mg/kg (Figure 1). Since U-46619-induced mortality in mice may be due to a combination of vasoconstriction and thrombosis,[38] these results suggest that **41b** prevents TXA_2-induced vasoconstriction and thrombosis.

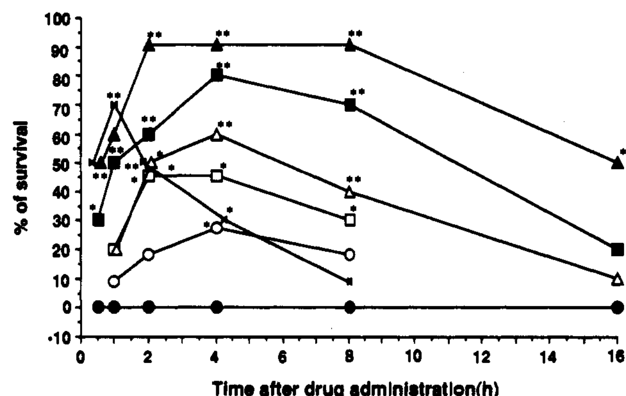


Figure 1. Protective effects of **41b** (KT2-962) and BM13,177 against U46619-induced sudden death in mice; ●, control; ○, 0.3 mg/kg; □, 1 mg/kg; △, 3 mg/kg; ■, 10 mg/kg; ▲, 30 mg/kg; ×, 30 mg/kg BM13,177. Each drug orally administered prior to injection of U46619 (800 μg/kg). At the indicated time after administration of drug, U46619 was injected intravenously and the incidence of death within 5 min was determined. Each point represents the survival percent in a group of 10–20 mice. $^*P < 0.05$ and $^{**}P < 0.01$, compared with control group.

Conclusion

In summary, we have shown the modification of 7-isopropyl-3-ethylazulene-1-sulfonic acid (compound **1**) and the effects of a 7-isopropyl group and the introduction of a sulfonamide group at the 3-position of compound **1**. 6-Isopropyl-substituted series having a sulfonamide group at the 3-position of the azulene ring showed more potent TXA_2 receptor antagonistic activities. After the optimization of the 6-substituted series, compound **41b** was found to be the most potent of this series and also found to be a τ receptor selective TXA_2 antagonist with no partial agonistic and TXA_2 synthetase activities. In an in vivo study, compound **41b** inhibited U-46619-induced sudden death and had a long duration of action. Due to its in vivo activity, compound **41b** has been selected for clinical evaluation.

Experimental Section

Analysis. Melting points were recorded on a Yamato MP-21 apparatus and are uncorrected. ^{1}H-NMR spectra were recorded on a Hitachi R-90H (90-MHz) spectrometer. Chemical shifts were expressed in parts per million relative to internal tetramethylsilane (δ = 0) or chloroform (δ = 7.26). IR spectra were obtained on a Hitachi 270-30 spectrophotometer. Mass spectra were recorded with a Hitachi M-80B mass spectrometer. Thin-layer chromatography (TLC) was carried out on 0.25-mm precoated silica gel plates from E. Merck (60F-254) with UV light and/or 7% phosphomolybdic acid in ethanol–heat as visualizing agent. Column chromatography was conducted by using silica gel (Fuji Devison BW-200, 150–325 mesh).

Solvents. Ether and THF were distilled over sodium–benzophenone ketyl under an Ar atmosphere. CH_2Cl_2, toluene, triethylamine, pyridine, and DMSO were distilled over CaH_2 under an Ar atmosphere.

N-[2-[4-(carboxymethoxy)phenyl]ethyl]-3-methylazulene-1-sulfonamide (10a). A solution of 1.50 g (6.14 mmol) of 3-methylazulene-1-sulfonic acid sodium salt[39] in 20.0 mL of DMF at 0 °C was treated with 1.18 mL (18.4 mmol) of $SOCl_2$. The mixture was stirred for 30 min at 0 °C and then treated with a solution of 1.59 g (6.14 mmol) of ethyl 4-(2-aminoethyl)-phenoxyacetate hydrochloride in 20 mL of DMF and 20.7 mL (100.0 mmol) of Et_3N and stirred for 1 h at 0 °C. The mixture was poured into ice–water, extracted with ethyl acetate, washed with 10% aqueous HCl, saturated aqueous $NaHCO_3$, water, and brine, and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude sulfonamide, which was chromatographed on silica gel. Elution with EtOAc–n-hexane (1:1) gave 1.97 g (75.0% yield) of **10a** as a violet oil: IR (neat) 3274, 2968, 2920, 1746, 1443, 1419, 1386, 1299, 1197, 1146, 1125 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ = 1.31 (t, 3 H), 2.43 (s, 3 H), 2.75 (t, 2 H), 3.15 (q, 2 H), 4.27 (q, 2 H), 4.48 (bt, 1 H), 4.52 (s 2 H), 7.27 (q, 4 H), 7.38 (t, 2 H), 7.74 (d, 1 H), 7.98 (s, 1 H), 8.37 (d, 1 H), 9.02 (d, 1 H); MS m/z 427 (M$^+$).

N-[2-[4-(Carboxymethoxy)phenyl]ethyl]-3-methylazulene-1-sulfonamide (12a). To a solution of 1.90 g (4.44 mmol) of ester in 20 mL of MeOH was added 15.0 mL of 1 N aqueous KOH at 0 °C, and the mixture was stirred for 1 h. The mixture was adjusted to pH 7.0 with 10% aqueous HCl; the solvent was removed under reduced pressure. The pH was adjusted to pH 2–3 with 10% aqueous HCl, and the solution was extracted with EtOAc. The EtOAc extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude acid, which was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (10:1) gave 1.73 g (97.4% yield) of **12a** as violet crystals: mp 38–39 °C, IR (KBr) 3016, 2926, 1734, 1518, 1299, 1215, 1146 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ = 2.50 (s, 3 H), 2.54 (t, 2 H), 3.06 (t, 2 H), 4.55 (s, 2 H), 6.55–6.94 (m, 4 H), 7.44 (q, 2 H), 7.80 (d, 1 H), 7.97 (s, 1 H), 8.46 (d, 1 H), 9.02 (d, 1 H); MS m/z 399 (M$^+$).

Methyl 6-Isopropyl-3-(4-phthalimidobutyl)azulene-1-carboxylate (25c). To a stirred, ice-cold solution of 0.50 g (1.66 mmol) of methyl 6-isopropyl-3-(4-hydroxybutyl)azulene-1-carboxylate were added 0.54 g (3.67 mmol) of phthalimide and 0.963 g (3.67 mmol) of triphenylphosphine in 5.0 mL of THF, followed by 0.64 g (3.67 mmol) of diethyl azodicarboxylate. The mixture was allowed to warm to room temperature and stirred for 12 h. Removal of the solvent under reduced pressure followed by purification by chromatography on a silica gel column with ether–n-hexane (1:2) gave 0.68 g (96.8% yield) of **25c** as a violet oil: IR (neat) 3004, 2950, 1767, 1704, 1578, 1446, 1398, 1368, 1218, 1050 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ = 1.37 (d, 6 H), 1.66–1.91 (m, 4 H), 2.90–3.24 (t + m, 3 H), 3.74 (t, 2 H), 3.82 (s, 3 H), 7.20–7.90 (m, 6 H), 8.09 (s, 1 H), 8.32 (d, 1 H), 9.45 (d, 1 H); MS m/z 429 (M$^+$).

Methyl 3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-carboxylate (27c). To a solution of 0.68 g (1.59 mmol) of phthalimide in 5.0 mL of EtOH was added 0.20 g (3.98 mmol) of hydrazine hydrate, and mixture was refluxed for 2 h. The reaction mixture was cooled to room temperature, diluted with $CHCl_3$, and filtered. The filtrate was concentrated under reduced pressure to give an oily product which was dissolved in 10 mL of acetone. To this stirring solution at 0 °C were added a solution of 0.16 g (1.91 mmol) of $NaHCO_3$ in 5 mL of water and then dropwise a solution of 0.25 mL (1.91 mmol) of benzenesulfonyl chloride in 5.0 mL of acetone. The mixture was stirred at 0 °C for 30 min and the organic was removed under reduced pressure. The remaining aqueous solution was extracted with ethyl acetate. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude sulfonamide, which was chromatographed on silica gel. Elution with ethyl acetate–n-hexane (1:4) gave 0.79 g (98.9% yield) of **27c** as violet crystals: mp 79–80 °C; IR (KBr) 3320, 3010, 1677, 1449, 1422, 1326, 1215, 1158 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ = 1.35 (d, 6 H), 1.20–1.93 (m, 4 H), 2.80–3.27 (t + m, 5 H), 3.90 (s, 3 H), 4.69 (bt, 1 H), 7.17–7.89 (m, 7 H), 8.00 (s, 1 H), 8.22 (d, 1 H), 9.43 (d, 1 H); MS m/z 440 (M$^+$).

3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-carboxylic Acid (29c). To a solution of 1.10 g (2.50 mmol) of **27c** in 22.0 mL of MeOH was added 11.0 mL of 10% aqueous NaOH, and the mixture was refluxed for 4 h. The mixture was cooled to room temperature, with removal of solvent under reduced pressure. The aqueous layer was washed with $CHCl_3$, and then the solution was adjusted to pH 2–3 with 10% aqueous HCl and extracted with ethyl acetate. The ethyl acetate was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude oil, which was chromatographed on silica gel. Elution with ethyl acetate–n-hexane (2:1) gave 0.89 g (83.6% yield) of **29c** as violet crystals: mp 167–168 °C; IR (KBr) 3274, 2950, 1653, 1581, 1461, 1326, 1248, 1155 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ = 1.35 (d, 6 H), 1.20–1.80 (m, 4 H), 2.62–3.40 (t + m, 5 H), 3.30 (bt, 1 H), 7.31–7.84 (m, 7 H), 7.98 (s, 1 H), 8.37 (d, 1 H), 9.36 (d, 1 H), 12.01 (bs, 1 H); MS m/z 381 (M$^+$ – 44).

1-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene (31). A mixture of 1.53 g (3.46 mmol) of **27** (R = $(CH_2)_4$-$NHSO_2Ph$, R_1 = 6-isopropyl) and 7.5 mL of anhydrous phosphoric acid was heated and stirred at 120 °C for 20 min. The reaction mixture was then poured into ice–water and extracted with ethyl acetate. The ethyl acetate extract was washed with water and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude product, which was chromatographed on silica gel. Elution with ethyl acetate–n-hexane (1:3) gave 1.23 g (93.1% yield) of **31** as violet crystals: mp 75–76 °C; IR (KBr) 3236, 2920, 1575, 1446, 1326, 1152, 1062 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ = 1.33 (d, 6 H), 1.20–1.95 (m, 4 H), 2.76–3.20 (t + m, 5 H), 4.35 (bt, 1 H), 6.98 (d, 2 H), 7.11–7.90 (s + m, 7 H), 8.00 (d, 1 H), 8.17 (d, 1 H); MS m/z 381 (M$^+$).

3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-sulfonic Acid Sodium Salt (41b). A mixture of 0.50 g (1.31 mmol) of **31** and 0.42 g (2.62 mmol) of SO_3–pyridine complex in 10 mL of benzene was refluxed for 3 h. The solvent was removed under reduced pressure, and the residue was dissolved in 10 mL of MeOH. Next, 0.25 mL of 10% sodium methoxide was added at room temperature, and the mixture was stirred for 24 h. The solvent was removed under reduced pressure, and the residue was dissolved in water. The mixture was extracted with THF, washed with brine, and dried over Na_2SO_4. Filtration and evaporation of the solvent under reduced pressure provided the crude sodium salt, which was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (10:1) gave 0.46 g (72.6% yield) of **41b** as violet crystals: mp 207–208 °C; IR (KBr) 3268, 2944, 1578, 1446, 1401, 1323, 1155, 1092, 1038 cm^{-1}; ^{1}H NMR (CD_3OD) δ = 1.35 (d, 6 H), 1.20–1.79 (m, 4 H), 2.75–3.04 (t + m, 5 H), 7.20–7.31 (d, 2 H), 7.40–7.90 (s + m, 6 H), 8.30 (d, 1 H), 9.00 (d, 1 H).

1-(Cyanomethyl)-3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene (33). A mixture of 0.054 g (0.53 mmol) of N,N,N′,N′-tetramethyldiaminomethane, 0.010 g (0.34 mmol) of paraformaldehyde, and 1.0 mL of acetic acid was heated to give a clear solution. This solution was cooled to room temperature and added dropwise with stirring to solution of 0.184 g (0.481 mmol) of **31** in 10 mL of CH_2Cl_2 at 0 °C. This mixture was allowed to stir for 1 h at 0 °C, placed in a refrigerator for 15 h, and then diluted with 20 mL of CH_2Cl_2, washed with water, and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude amine. This amine was dissolved in 10 mL of EtOH. To the stirring solution at room temperature were added 5.0 mL of methyl iodide and the mixture stirred for 2 h. Removal of solvent under reduced pressure gave the crude quaternary ammonium iodide. The quaternary ammonium iodide in 10 mL of EtOH was added to 0.094 g (1.44 mmol) of KCN. The mixture was heated under reflux for 1 h, diluted with 50 mL of water, and extracted with ether. The ether extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude cyanide, which was chromatographed on silica gel. Elution with ethyl acetate–n-hexane (1:2) gave 0.130 g (64.2% yield) of **33** as a violet oil: IR (neat) 3274, 2944, 2250, 1578, 1446, 1323, 1155, 1089 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ = 1.42 (d, 6 H), 1.11–1.87 (m, 4 H), 2.70–3.20 (t + m, 5 H), 4.02 (s, 1 H), 4.91 (bt, 1 H), 7.02 (d, 2 H), 7.27–7.90 (d + m, 6 H), 8.00 (d, 1 H), 8.11 (d, 1 H); MS m/z 420 (M$^+$).

3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-acetic Acid (34). To a solution of 0.11 g (0.26 mmol) of **33** in 10 mL of EtOH was added 10 mL of 0.6 M aqueous KOH, and

the mixture was refluxed for 12 h. The mixture was cooled to room temperature and the solvent removed under reduced pressure. The aqueous layer was washed with $CHCl_3$, adjusted to pH 2-3 with 10% aqueous HCl, and extracted with ethyl acetate. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude acid, which was chromatographed on silica gel. Elution with ethyl acetate gave 0.084 g (73.5% yield) of **34** as violet crystals: mp 51–52 °C; IR (KBr) 3268, 2944, 1704, 1578, 1446, 1323 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.31 (d, 6 H), 1.15–1.83 (m, 4 H), 2.77–3.15 (t + m, 5 H), 4.01 (s, 1 H), 4.30–4.60 (m, 2 H), 6.86–7.07 (dd, 2 H), 7.28–7.96 (s + m, 6 H), 7.97 (d, 1 H), 8.17 (d, 1 H); MS m/z 439 (M^+).

1-Formyl-3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene (35). DMF (4.0 mL) was treated with 0.18 mL (1.93 mmol) of $POCl_3$ at 0 °C under an Ar atmosphere. The resulting solution was stirred for 30 min and then treated with a solution of 0.490 g (1.28 mmol) of **31** in 1.8 mL of DMF and stirred for 30 min. The reaction mixture was poured into cold, 10% aqueous NaOH and extracted with ethyl acetate. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude aldehyde, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:2) gave 0.48 g (91.4% yield) of **35** as a violet oil: IR (neat) 3268, 2950, 1629, 1578, 1446, 1368, 1326, 1156, 1092 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.36 (d, 6 H), 1.50–1.90 (m, 4 H), 2.73–3.30 (t + m, 5 H), 5.03 (bt, 1 H), 7.30–7.60 (m, 5 H), 7.76–8.00 (s + m, 3 H), 8.30 (d, 1 H), 9.35 (d, 1 H), 10.33 (s, 1 H); MS m/z 409 (M^+).

(*E*)-3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-acrylic Acid Ethyl Ester (36). To a stirred suspension of 0.15 g (3.15 mmol) of a 60% NaH dispersion in 1.5 mL of THF cooled to 0 °C under an Ar atmosphere was added dropwise a solution of 0.79 g (3.51 mmol) of ethyl phosphonoacetate in 7.8 mL of THF. After the solution was stirred for 20 min, a solution of 0.48 g (1.17 mmol) of **35** in 4.8 mL of THF was added and the mixture was stirred for 2 h at room temperature. The reaction was then quenched with saturated aqueous NH_4Cl. The mixture was extracted with ethyl acetate, and the organic layer was washed with brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude ester, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:3) gave 0.54 g (96.1% yield) of **36** as a green oil: IR (neat) 3268, 2950, 1683, 1605, 1578, 1368, 1449, 1326, 1293 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.33 (d, 6 H), 1.34 (t, 3 H), 1.40–1.85 (m, 4 H), 2.70–3.13 (t + m, 5 H), 4.27 (q, 2 H), 5.05 (bt, 1 H), 6.36 (d, 2 H), 7.00–7.60 (m, 5 H), 7.60–7.90 (m, 3 H), 8.00 (s, 1 H), 8.14 (d, 1 H), 8.39 (d, 1 H); MS m/z 479 (M^+).

(*E*)-3-[4-[(Phenylsulfonyl)amino]butyl]-6-isopropylazulene-1-acrylic Acid (37). To a solution of 0.51 g (1.06 mmol) of **36** in 10 mL of MeOH was added 5.0 mL of 10% aqueous NaOH, and the mixture was stirred for 14 h at room temperature. After removal of the organic solvent under reduced pressure, the aqueous layer was washed with ethyl acetate. The solution was adjusted to pH 2-3 with 10% aqueous HCl and extracted with ethyl acetate. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude acid, which was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (10:1) gave 0.450 g (94.0% yield) of **37** as green crystals: mp 58–60 °C; IR (KBr) 3262, 2944, 1575, 1563, 1269, 1155 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.33 (d, 6 H), 1.45–1.90 (m, 4 H), 2.75–3.20 (t + m, 5 H), 4.98 (bs, 1 H), 6.37 (d, 2 H), 6.95–7.60 (m, 5 H), 7.65–7.95 (m, 3 H), 8.20 (d, 1 H), 8.20 (d, 1 H), 8.43 (d, 1 H), MS m/z 407 ($M^+ - 44$).

Methyl 3-[3-[(Benzyloxy)methyl]phenyl]-6-isopropylazulene-1-carboxylate. A mixture of 2.9 g (11.96 mmol) of [3-[(benzyloxy)methyl]phenyl]acetoaldehyde, 1.1 mL (11.96 mmol) of morpholine, and 1.47 g (5.98 mmol) of **6** in 30 mL of EtOH was heated under reflux for 20 h. After the solvent was removed under reduced pressure, the residue was dissolved in ethyl acetate. The solution was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided crude azulene, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:4) gave 1.18 g (46.5% yield) of azulene **7** (R_2 = $(PhCH_2)_2O$) as a violet oil: IR (neat) 2950, 2848, 1689, 1578, 1446, 1419, 1227, 1200 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.36 (d, 6 H), 2.90–3.30 (m, 1 H), 3.95 (s, 3 H), 4.61 (s, 2 H), 7.15–7.65 (m, 11 H), 8.35 (s, 1 H), 8.56 (d, 1 H), 9.60 (d, 1 H); MS m/z 424 (M^+).

Methyl 3-[3-(Hydroxymethyl)phenyl]-6-isopropylazulene-1-carboxylate. To a solution of 1.18 g (2.78 mmol) of the azulene prepared above in 30 mL of anisol was added 0.74 g (5.56 mmol) of $AlCl_3$ at 0 °C and the mixture was stirred for 15 min. The mixture was poured into ice–water and extracted with ethyl acetate. The ethyl acetate extract was washed with water, saturated aqueous $NaHCO_3$, and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude alcohol, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:1) gave 0.81 g (87.1% yield) of alcohol as a violet oil: IR (neat) 3400, 2944, 2860, 1689, 1449, 1419, 1227, 1200 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.36 (d, 6 H), 2.90–3.30 (m, 1 H), 3.93 (s, 3 H), 4.76 (s, 2 H), 7.20–7.65 (m, 6 H), 8.33 (s, 1 H), 8.55 (d, 1 H), 9.57 (d, 1 H); MS m/z 334 (M^+).

Methyl 3-[3-(Bromomethyl)phenyl]-6-isopropylazulene-1-carboxylate. To a solution of 0.80 g (2.39 mmol) of the above alcohol in 10.0 mL of CH_2Cl_2 were added 0.75 g (2.87 mmol) of triphenylphosphine and 0.95 g (2.87 mmol) of carbon tetrabromide at −25 °C under an Ar atmosphere. The reaction mixtre was stirred for 1 h at room temperature. The reaction was quenched with saturated aqueous $NaHCO_3$ and extracted with $CHCl_3$. The $CHCl_3$ extract was washed with brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude bromide, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:4) gave 0.91 g (95.8% yield) of bromide as a violet oil: IR (neat) 2950, 1689, 1578, 1449, 1419, 1233, 1197 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.36 (d, 6 H), 2.90–3.30 (m, 1 H), 3.94 (s, 3 H), 4.57 (s, 2 H), 7.20–7.63 (m, 6 H), 8.33 (s, 1 H), 8.52 (d, 1 H), 9.60 (d, 1 H); MS m/z 398 (M^+).

Methyl 3-[3-[(Phenylsulfonyl)amino]phenyl]-6-isopropylazulene-1-carboxylate. To a stirred suspension of 0.15 g (3.40 mmol) of 60% sodium hydride dispersion in 5.0 mL of THF cooled to 0 °C under an Ar atmosphere was added dropwise a solution of 0.54 g (3.40 mmol) of benzenesulfonamide in 5.0 mL of THF. After the solution was stirred for 20 min, a solution of 0.89 g (2.26 mmol) of bromide in 5.0 mL of HMPA was added and the mixture was stirred for 1 h at room temperature. The reaction was then quenched with saturated aqueous NH_4Cl and extracted with EtOAc. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude sulfonamide, which was chromatographed on silica gel. Elution with ethyl acetate–*n*-hexane (1:3) gave 0.36 g (33.6% yield) of sulfonamide as a violet oil: IR (neat) 2968, 1692, 1578, 1449, 1422, 1374, 1239, 1200 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.32 (d, 6 H), 2.90–3.30 (m, 1 H), 3.93 (s, 3 H), 4.49 (s, 2 H), 7.05–7.60 (m, 9 H), 7.75–8.00 (m, 6 H), 8.11 (s, 1 H), 8.30 (d, 1 H), 9.52 (d, 1 H); MS m/z 473 (M^+).

3-[3-[[(Phenylsulfonyl)amino]methyl]phenyl]-6-isopropylazulene-1-carboxylic Acid (38). To a solution of 0.26 g (0.55 mmol) of the ester prepared above in 10 mL of MeOH was added 5.0 mL of 10% aqueous NaOH and the mixture was refluxed for 10 h. After removal of the organic solvent under reduced pressure, the aqueous layer was washed with $CHCl_3$ The solution was adjusted to pH 2-3 with 10% aqueous HCl and extracted with ethyl acetate. The ethyl acetate extract was washed with water and brine and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure provided the crude acid, which was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (50:1) gave 0.035 g (13.8% yield) of **38** as violet crystals: mp 95–97 °C; IR (KBr) 3010, 2950, 1650, 1578, 1485, 1236, 1155 cm^{-1}; ^1H NMR ($CDCl_3$) δ = 1.36 (d, 6 H), 2.90–3.30 (m, 1 H), 4.25 (d, 2 H), 7.00–7.65 (m, 9 H), 7.70–8.00 (m, 2 H), 9.30 (s, 1 H), 9.48 (d, 1 H), 9.60 (d, 1 H); MS m/z 459 (M^+).

Biological Methods. Relaxing Effects on U-46619-Induced Contraction of Rat Aorta. The thoracic aortas from male rats (Wistar) were excised, cleaned, and cut into ring strips (2-mm width and 4-mm length). The tissues were placed in 10-mL organ baths containing a Krebs bicarbonate solution [(mM) NaCl (120.3), KCl (4.8), $CaCl_2$ (1.2), KH_2PO_4 (1.2), $MgSO_4$ (1.3), $NaHCO_3$ (24.2), dextrose (10.0)] that was kept at 37 °C while

being bubbled with 95% O_2 and 5% CO_2. A resting tension of 1.0 g was applied, and after equilibration for 1 h, the isomeric tention was recorded on a polygraph (Nihon Kohden) through a force-displacement transducer (Nihon Kohden). After the increase in isomeric tension due to addition of 3×10^{-8} M U-46619 became stable, cumulative concentrations of each compound were added to the bath. The concentration which caused a 50% relaxation of a U-46619-induced maximal change in tension was obtained by regression analysis of the concentration-relaxation curve.

Inhibitory Effects of U-46619-Induced Platelet Aggragation in Rabbit. Male rabbits (New Zealand) weighing 2.0–2.5 kg were anesthetized with ether; blood was withdrawn from the carotid artery through a cannulation tube with a syringe containing trisodium citrate (3.18%, $^1/_{10}$ volume). Platelet-rich plasma (PRP) was then prepared by centrifugation at 300*g* for 15 min. The supernatant was decanted, and the remaining pellet was centrifuged at 1000*g* for 20 min to produce platelet-poor plasma (PPP), which was used as a zero calibration. Platelet aggregation was measured with an aggregometer (Chrono-Log) by the method of Born.[40] PRP (500 μL) placed in a cuvette was warmed at 37 °C for 2 min, and then a solution of the compound or vehicle was added. Exactly 2 min later, 2.5×10^{-6} M U-46619 was added to PRP. The change in light transmission was recorded, with the light transmissions for PRP and PPP being taken as 0% and 100%, respectively, and the maximum light transmission after addition of U-46619 as the maximum aggregation. The concentration which caused 50% inhibition of a U-46619-induced maximum aggregation was obtained by regression analysis of the concentration–inhibition curve.

Binding Assay with Rabbit Gel-Filtered Platelets. The experiments were done according to the method described by Hanasaki et al.[41] with slight modifications. Blood from male New Zealand rabbits was collected into a syringe containing a 0.15 volume of acid citrate dextrose [(mM) trisodium citrate (85.0), citric acid (70.0), glucose (110.0)]. PRP was obtained by centrifugation at 300*g* for 15 min. The PRP was mixed with PGI_2 (0.5 μg/mL) and apyrase (25 μg/mL) and layered on 40% bovine serum albumin. Platelets were recentrifuged at 1200*g* for 15 min and resuspended in 1.0 mL of resuspension buffer [(mM) NaCl (137), KCl (2.7), $MgCl_2$ (1.0), Na_2HPO_4 (3.8), HEPES (3.8), glucose (5.6), 0.035% bovine serum albumin, pH 7.35]. Platelets were separated from plasma proteins by gel filtration through a column of Sepharose 2B and suspended in the resuspension buffer to a final concentration of 3×10^8 cells/mL. Aliquots of the platelet suspension (0.46 mL) were incubated with 5 nM [^{3}H]SQ29548 (0.075 μCi) plus 10^{-5} M BM13,505 or various concentrations of compound **41b** (KT2-962) for 30 min at 25 °C. Specific binding is defined as the difference between the binding in the presence and absence of 10^{-5} M BM13505. After the incubation, ice-cold saline (3.0 mL) was added to each tube and the reaction mixture was immediately filtered through Whatman GF/C glass-filter disks, which were then washed four times with ice-cold saline. All filtration procedures were completed within 20 s. The radioactivity was measured in a liquid scintillation counter (Aloka, LSC-700).

Effects on Thromboxane B_2 Formation. Human blood was obtained from the antecubital vein of healthy volunteers. PRP was prepared by centrifugation at 200*g* for 20 min. The PRP was further centrifuged at 2000*g* for 10 min, and the resulting pellet was washed twice with 25 mM phosphate buffer. The washed pellets were resuspended in 50 mM phosphate buffer (pH 7.4) containing 1 mM EDTA, sonicated, and stored at -80 °C until use. Sonicated platelets were preincubated with test compounds in the presence of 1 mM GSH at 37 °C for 5 min, and then the mixture was incubated with [1-^{14}C]arachidonic acid (0.1 μCi) at 37 °C for 5 min. Each reaction was terminated by the addition of 2.5 mL of cold ether/methanol(30:4, v/v) and the mixture was acidified with 200 mM citric acid. ^{14}C-Labeled eicosanoids were extracted and separated by thin-layer chromatography. The areas corresponding to each eicosanoid were removed, and the radioactivity was counted. Thromboxane formation was calculated from the amount of thromboxane B_2 generated from [^{14}C]arachidonic acid. The IC_{50} values were obtained by regression analysis of the concentration–inhibition curve.

Protective Effects against U-46619-Induced Sudden Death in Mice. This investigation was carried out according to the modified method of Myers et al.[38] using conscious male ddY mice. KT2-962 or BM13,177 (sulotroban) was orally administered in 0.5% methylcellulose solution at doses from 0.3 to 30 mg/kg. Control animals were given only 0.5% methylcellulose solution. At the indicated time after administration of each drug or vehicle, U-46619 (800 μg/kg) was injected intravenously. The effect of each drug was evaluated by measuring the incidence of death within 5 min of injection. The point of death was determined by monitoring respiration. Results are expressed as percent survival. Each group was comprised of 10–20 mice.

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References

(1) Ellis, E. F.; Oelz, O.; Roberts, L. J. Coronary arterial smooth muscle contraction by a substance released from platelets: Evidence that it is thromboxane A_2. *Science* **1976**, *193*, 1135–1137.

(2) Schror, K.; Smith, E. F., III; Bickerton, M.; Smith, J. B.; Nicolaou, K. C.; Magolda, R.; Lefer, A. M. Preservation of ischemic myocardium by pinane thromboxane A_2. *Am. J. Physiol.* **1980**, *238*, H87–H92.

(3) Coleman, R. A.; Sheldrich, R. L. G. Prostanoid-induced contraction of human bronchial smooth muscle is mediated by TP-receptors. *Br. J. Pharmacol.* **1989**, *96*, 688–692.

(4) Explugues, J. V.; Whittle, B. J. R. Close-arterial administration of the thromboxane mimetic U-466129 induces damage to the rat gastric mucosa. *Prostaglandins* **1988**, *96*, 688–692.

(5) Kitagawa, H.; Kurahashi, K.; Fujiwara, M. Gastric Mucosal Erosion Due to a Mucosal Ischemia Produced by Thromboxane A_2-Like Substance in Rats under Water-Immersion Stress. *J. Pharmacol. Exp. Ther.* **1986**, *237*, 300–304.

(6) Lefer, A. M. Comparison of the actions of thromboxane receptor antagonists in biological systems. *Drugs Today* **1985**, *21*, 283–291.

(7) Patscheke, H. Thromboxane A_2/Prostaglandin H_2 Receptor Antagonists. A New Therapeutic Principle. *Stroke* **1990**, *21* (Suppl IV), IV-139–IV-142.

(8) Yanagisawa, T.; Wakabayashi, S.; Tomiyama, T.; Yasunami, M.; Takase, K. Synthesis and Antiulcer Activities of Sodium Azulene Sulfonates. *Chem. Pharm. Bull.* **1988**, *36*, 641–647.

(9) Mochizuki, S.; Wakabayashi, S.; Tomiyama, A.; Sakata, N.; Shibata, S. Thromboxane A_2 Antagonistic Action of a New Anti-Ulcer Agent, Azuletil Sodium (KT1-32). *J. Gastroenterol.* **1989**, *24* (Suppl. 162), 194–194.

(10) Tomiyama, T.; Wakabayashi, S.; Kosakai, K.; Yokota, M. Azulene Derivatives: New Non-Prostanoid Thromboxane A_2 Receptor Antagonists. *J. Med. Chem.* **1990**, *33*, 2323–2326.

(11) Mais, D. E.; Saussy, D. L.; Chaikhouni, Jr. A.; Kochel, P. J.; Knapp, D. R.; Hamanaka, N.; Halushka, P. V. Pharmacologic Characterization of Human and Canine Thromboxane A_2/Prostaglandin H_2 receptors in Platelet and Blood vessels: Evidence for Different Receptors. *J. Pharmacol. Exp. Ther.* **1985**, *233*, 418–424.

(12) BM13,177, 4-[2-(benzenesulfonamido)ethyl]phenoxyacetic acid (Sulotroban), is a TXA_2 antagonist. It was synthesized in our laboratories. Bush, L. R.; Smith, S. G. Antagonism of U46619-induced aggregation of human and canine platelets by four TXA_2 receptor antagonists. *Thromb. Res.* **1988**, *44*, 377–389.

(13) Narisada, M.; Ohtani, M.; Watanabe, E.; Uchida, K.; Arita, H.; Doteuchi, M.; Hanasaki, K.; Kakushi, H.; Otani, K.; Hara, S. Synthesis and in vitro Activity of various Derivatives of a Novel Thromboxane Receptor Antagonist, (±)-(5Z)-7-[3-endo-[(phenylsulfonyl)amino]bicyclo[2,2,1]hept-2-exo-yl]heptenoic Acid. *J. Med. Chem.* **1988**, *31*, 1847–1854.

(14) Rosentreter, U.; Boshagen, H.; Seuter, F.; Perzborn, E.; Friedler, V. B. Synthesis and Absolute Configuration of the New Thromboxane Antagonist (3R)-3-(4-Fluorophenylsulfonamide)-1,2,3,4-tetrahydro-9-carbazolepropanoic Acid and Comparison with its Enantiomer. *Arzneim-Forsch./Drug Res.* **1989**, *39*, 1519–1521.

(15) Takase, K.; Yasunami, M. The Synthesis of Azulene Derivatives by the Reaction of 2H-cyclohepta[b]furan-2-ones with Enamines. *J. Syn. Org. Chem. Jpn.* **1981**, *39*, 1172–1182.

(16) Yanagisawa, T.; Kosakai, K.; Tomiyama, T.; Yasunami, M.; Takase, K. Studies on Anti-Ulcer Agents. II. Synthesis and Anti-Ulcer Activities of 6-Isopropylazulene-1-sodium sulfonate Derivatives. *Chem. Pharm. Bull.* **1990**, *12*, 3355–3358.

(17) Yang, P. M.; Yasunami, M.; Takase, K. The Formation of Azulene Derivatives by the Reaction of 2H-cyclohepta[b]furan-2-ones with Enamines. *Tetrahedron Lett.* **1971**, *45*, 4275–4278.

(18) Nozoe, T.; Takase, K.; Kato, M.; Nogi, T. Reaction of 2-Arylsulfonyloxytropones and Active Methylene Compounds. Formation of 8-Hydroxy-2H-cyclohepta[b]furan-2-one and 2-Amino-8H-cyclohepta[b]furan-8-one Derivatives. *Tetrahedron* 1971, *27*, 6023–6035.

(19) Mitsunobu, O. The use of Diethyl Azodicarboxylate and Triphenylphosphine in Synthesis and Transformation of Natural Products. *Synthesis* 1981, 1–28.

(20) Anderson, A. G.; Anderson, R. G.; Fujita, T. S. Displacement reactions on 1-azulylmethyl trimethylammonium iodide. *J. Org. Chem.* 1962, *27*, 4535–4539.

(21) Hafner, K.; Bernhand, C. Azulenes. IV. Azulene Aldehydes and ketones. *Justus Liebigs Ann. Chem.* 1959, *625*, 108–123.

(22) Wadworth, W. S.; Emmons, W. D. The Utility of Phosphonate Carbanions in Olefin Synthesis. *J. Am. Chem. Soc.* 1961, *83*, 1733–1738.

(23) Collonges, F.; Descotes, G. Synthesis tautomeric equilibrium, and pyrolysis of cis-6-hydroxy-4-hexenal. *Tetrahedron Lett.* 1973, 1117–1118.

(24) Corey, E. J.; Kirst, H. A. New method for the synthesis of macrolides. *J. Am. Chem. Soc.* 1972, *94*, 667–668.

(25) U-46619, (15*S*)-hydroxy-11α,9α-(epoxymethano)prosta-5(*Z*),13(*E*)-dienoic acid, is a stable PGH_2 analogue and TXA_2/PGH_2 receptor agonist. It was purchased from Cayman Chemical Co. (Ann Arbor, MI). Malmsten, C. Some biological effects of prostaglandin endoperoxide analogs. *Life Sci.* 1976, *18*, 169–176.

(26) Lee, S. J.; Kurihara-Bergstoron, T.; Kim, S. W. Ion-paired drug diffusion through polymer membranes. *Int. J. Pharm.* 1987, *45*, 59–73.

(27) pA_2 is the negative logarithm of the dose of antagonist that would require a doubling of the agonist dose to compensate for its action.

(28) Arunlakshana, O.; Schild, H. O. Some quantitative uses of drug antagonists. *Br. J. Pharmacol. Chemother.* 1959, *14*, 48–58.

(29) Needlemann, P.; Minkes, M.; Raz, A. Thromboxanes: Selective biosynthesis and distinct biological properties. *Science* 1976, *193*, 163–165.

(30) Harrold, M. W.; Grajzl, B.; Shin, Y.; Romstedt, K. J.; Feller, D. R.; Miller, D. D. Synthesis and Thromboxane A_2 Antagonist Activity of N-Benzyltrimetoquinol Analogues. *J. Med. Chem.* 1988, *31*, 1506–1512.

(31) Hall, S.; Han, W-C.; Harris, D. N.; Hedberg, A.; Ogletree, M. L. 9,11-Epoxy-9-homo-14-thiaprost-5-enoic acid Derivatives: Potent Thromboxane A_2 Antagonists. *J. Med. Chem.* 1989, *32*, 974–984.

(32) (a) Seno, K.; Hagashita, S. Thromboxane A_2 Receptor Antagonists. III. Synthesis and Pharmacological Activity of 6,6-Dimethylbicyclo-[3,1,1]heptane Derivatives with a Substituted Sulfonylamino Group at C-2. *Chem. Pharm. Bull.* 1989, *37*, 1524–1533. (b) Kamata, S.; Haga, N.; Tsuri, T.; Uchida, K.; Kushi, H.; Arita, H.; Hanasaki, K. Synthesis of thromboxane receptor antagonists with bicyclo[3,1,0]-hexane ring systems. *J. Med. Chem.* 1990, *33*, 229–239.

(33) Hanasaki, K.; Arita, H. Characterization of a new compound, S-145, as a specific TXA_2 receptor antagonist in platelets. *Thromb. Res.* 1988, *50*, 365–376.

(34) Uski, T. K. Further characterization of the contraction-mediating prostanoid receptors in feline cerebral arteries. Effects of the thromboxane-receptor antagonist AH23848. *Acta Physiol. Scand.* 1988, *133*, 519–524.

(35) Mckenniff, M.; Rodger, I. W.; Norman, P.; Gardiner, P. J. Characterization of receptors mediating the contractile effects of prostanoids in guinea-pig and human air ways. *Eur. J. Pharmacol.* 1988, *153*, 149–159.

(36) Hedberg, A.; Hall, S. E.; Ogletree, M. L.; Harris, D. N.; Liu, E. C-K. Characterization of [5,6-^{3}H]SQ29,548 as a high affinity radioligand, binding to thromboxane A_2/prostaglandin H_2-receptors in human platelets. *J. Pharmacol. Exp. Ther.* 1988, *245*, 786–792.

(37) [^{3}H]SQ29,548, tritium-labeled (1*S*-[1a,2b(5*Z*),3b,4a])-7-[3-[[2-[(phenylamino)carbonyl]hydrazino]methyl]-7-oxabicyclo[2.2.1]-hept-2-yl]-5-heptenoic acid, is a stable PGH_2 analogue and TXA_2/PGH_2 receptor antagonist. It was purchased from New England Nuclear.

(38) Myers, A.; Penhos, J.; Ramey, E.; Ramwell, P. Thromboxane Agonism and Antagonism in a Mouse Sudden Death Model. *J. Pharmacol. Exp. Ther.* 1983, *224*, 369–372.

(39) Yanagisawa, K.; Wakabayashi, S.; Tomiyama, T.; Yasunami, M.; Takase, K. Synthesis and Anti-ulcer Activities of Sodium Alkylazulene Sulfonates. *Chem. Pharm. Bull.* 1988, *36*, 641–647.

(40) Born, G. V. R. Aggregation of Blood Platelets by Adenosine Diphosphate and Its Reversal. *Nature (London)*, 1962, *194*, 927–929.

(41) Hanasaki, K.; Nakano, T.; Arita, H. Two phasic generation of thromboxane A_2 by the action of collagen on rat platelets. *Thromb. Res.* 1987, *46*, 425–436.