Studies on Anti-inflammatory Agents. VI.¹⁾ Synthesis and Pharmacological Properties of 2,3-Diarylthiophenes

Kiyoshi TSUJI,*^a Katsuya NAKAMURA,^a Takashi OGINO,^a Nobukiyo KONISHI,^a Takashi TOJO,^a Takehiro OCHI,^b Nobuo SEKI,^b and Masaaki MATSUO^a

Medicinal Chemistry Research Laboratories^a and Medicinal Biology Research Laboratories,^b Fujisawa Pharmaceutical Co., Ltd., 1–6, Kashima 2-chome, Yodogawa-ku, Osaka 532–8514, Japan.

Received August 11, 1997; accepted October 14, 1997

A series of novel 5-substituted-2,3-diarylthiophenes has been synthesized and found to be active in the rat adjuvant arthritis (AA) model and/or in the yeast-induced hyperalgesia (Randall–Selitto) assay. Among the compounds synthesized herein, 2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-(trifluoromethyl)thiophene (**6a**) exhibited the most potent activities on AA, collagen-induced arthritis (CIA) and the delayed-type hypersensitivity response to type II collagen. 5-Bromo-2-[4-(methylamino)phenyl]-3-[4-(methylsulfinyl)phenyl]thiophene (**38**) is also a potent inhibitor of AA, CIA, hyperalgesia and *in vitro* tumor necrosis factor- α production.

Key words anti-inflammatory agent; 2,3-diarylthiophene; antirheumatic activity; analgesic activity; structure–activity relationship

Rheumatoid arthritis (RA) is a chronic inflammatory disease characterized by painful and swollen joints with possible progression to irreversible joint damage. It is believed to have an underlying autoimmune pathology. The drug treatment initially involves the administration of nonsteroidal anti-inflammatory drugs (NSAIDs) such as indomethacin and diclofenac. These drugs relieve pain and stiffness, but have no significant effect on immunological responses. Except for the mildest cases, disease-modifying antirheumatic drugs (DMARDs) such as gold salts and D-penicillamine are used to suppress immunological processes and/or joint destruction. New and potentially disease-modifying therapies for RA that have recently come under investigation include tenidap, leflunomide, anti-tumor necrosis factor(TNF)- α antibody, cyclosporin and FK506.²⁾

We previously reported a novel compound **1** (FR115068), which was discovered through chemical modification of the anti-inflammatory agent FK3311, and which possesses a broad spectrum of both anti-inflammatory and immunoregulatory activities.³⁾ Aiming at developing new compounds exerting potent and broad-spectrum activities like **1**, we conducted chemical modification of the structurally distinct thiophene-type agent **2**.⁴⁾ Recently, a number of thiophene derivatives, which include selective cyclooxygenase(COX)-2 inhibitors such as 2,3-diarylthiophene **3** and 3,4-diarylthiophene **4**, have been reported.⁵⁾ These reports encouraged us to publish our studies on **2**-related 2,3-diarylthiophenes. In this paper, we describe the syntheses and pharmacological activities of the thiophene derivatives represented by the general structure of **5**.

Chemistry

The syntheses of 5-(perfluoroalkyl, carbamoyl and aminomethyl)thiophenes (**6**, **13**, **15**) are summarized in Chart 1. Perfluoroalkylation of **2** with sodium perfluoroalkane carboxylates and CuI yielded **6**.^{6,7)} The deoxybenzoin **7** was treated with Vilsmeier reagent, followed by cyclization with thioglycolic acid, to give **9** and **10**.⁸⁾ Decarboxylation of **10** in the presence of Cu

powder also afforded **9**. Oxidation of **10** with H₂O₂ in AcOH gave **11**, which was activated with PCl₅ or 1,1'-carbonyldiimidazole (Im₂CO) and then treated with the appropriate amines, affording the carboxamides **13**. 5-(Dialkylamino)methyl derivatives **15c–e** were prepared by reduction of **12** and subsequent oxidation *via* **14c–e**. The amino and methylamino groups of **14a, b** were protected, followed by oxidation with *m*-chloroperbenzoic acid (mCPBA) and deprotection with HBr to give **15a, b**.

The syntheses of 5-(acetyl, carboxymethyl and sulfa-moyl)thiophenes (**17**, **19**, **20**) and 3-[4-(methylsulfonyl-amino)phenyl]thiophene **22** are shown in Chart 2. Compound **9** was oxidized and acetylated with acetyl chloride and TiCl₄ catalyst to afford **17**. The carboxymethyl de-

Fig. 1

* To whom correspondence should be addressed.

Chart 1

Chart 2

Chart 3

rivative **19** was obtained by oxidative rearrangement of **17** by the method of McKillop *et al.*⁹⁾ and subsequent hydrolysis of the ester **18**. Chlorosulfonylation and amidation of **3** gave the sulfamoyl derivatives **20**. Similarly, methylsulfonylation of the amino derivative **21** yielded

22.¹⁰⁾ The nitrile **23** was prepared by dehydration of the amide **13a** (Chart 3).¹¹⁾ 5-(Substituted amino)thiophenes (**26**, **27**) were synthesized *via* the amino derivative **25**, which was obtained from **3** by nitration and reduction with Fe.

5-Bromo-2-[4-(methylamino)phenyl]thiophene **38** was obtained as shown in Chart 4. The deoxybenzoin **30** was

Chart 4

prepared by the method of Bender and Hill.¹²⁾ Reaction of **30** with Vilsmeier reagent and ethyl thioglycolate, hydrolysis, decarboxylation, and subsequent bromination with bromine afforded the 5-bromothiophene derivative **34**. The methylamino derivative **38** was prepared from **34** by reduction, protection with a formyl group, methylation, careful oxidation with mCPBA, and deprotection *via* **35** and **36**. The formylamino intermediate **35** was oxidized to give **37**.

Results and Discussion

The compounds synthesized in this study were first tested for anti-inflammatory and analgesic activities through oral administration to rats. The chronic anti-inflammatory activity was assessed in terms of inhibition of adjuvant arthritis (AA). The analgesic activity against inflammation-related pain was evaluated as relative potency in the yeast-induced hyperalgesia (Randall–Selitto) assay. The test results are summarized in Tables 1–3.

As a first step in the chemical modification of **2**, we investigated various 5-substituted thiophene derivatives to find alternative substituents to the bromine of **2**. Table 1 shows the results for perfluoro, amino and carboxy alkyl derivatives (**6**, **15**, **19**). Perfluoro compounds **6a**, **b** exhibited very potent inhibitory activities toward AA (98% and 88% inhibition at 10 mg/kg, respectively). Amino and carboxy compounds (**15b**–**e**, **19**) also showed potent anti-inflammatory activities. Among these active compounds, **6a** was selected as a lead compound because of its exceptionally strong anti-inflammatory activity ($\text{ED}_{50} = 0.030 \text{ mg/kg}$). The (methylsulfonyl)phenyl moiety at the 3-position of the thiophene ring of **6a** was replaced with a (methylsulfonylamino)phenyl group, in the expectation that broader-spectrum activities, such as those of **1**, would be obtained. The resultant compound **22** also significantly suppressed AA. Moreover, perfluoro (**6**, **22**), amino (**15a**, **c**, **d**) and carboxy (**19**) alkyl derivatives exhibited moderate analgesic activities.

Various carbamoyl and sulfamoyl derivatives (**13**, **20**) were investigated as shown in Table 2. *N*-Methyl and *N*-unsubstituted derivatives (**13b**, **20a**, **b**) exhibited the most potent anti-inflammatory activity. Bulky substituents

Table 1. Investigation of 5-(Substituted alkyl) Derivatives

Compd.	R	Adjuvant arthritis % inhibition ^{a)} (10 mg/kg <i>p.o.</i>)	Randall–Selitto relative potency ^{b)} (32 mg/kg <i>p.o.</i>)
6a	CF_3	98 ^{c)}	1.20 ^{d)}
6b	CF_2CF_3	88 ^{c)}	1.13
15a^{e)}	CH_2NH_2	43 ^{d)}	1.30 ^{c)}
15b^{e)}	CH_2NHMe	86 ^{c)}	1.03
15c^{f)}	CH_2NMe_2	79 ^{c)}	1.19 ^{d)}
15d^{f)}	CH_2NMeEt	73 ^{c)}	1.33 ^{c)}
15e	CH_2NEt_2	66 ^{c,g)}	1.06
19	CH_2COOH	78 ^{c)}	1.27 ^{c)}
22		73 ^{c,h)}	1.17

a) Uninjected paw. b) Ratio of the pain threshold in the treated vs. control animals. c) $p < 0.01$. d) $p < 0.05$, significant difference from control (Student's *t*-test). e) HBr salt. f) HCl salt. g) 3.2 mg/kg. h) 7 mg/kg.

(e.g., **13c**, **f**–**h**, **20c**) gave less active compounds, but introduction of bulky carbamoyl groups resulted in fairly potent analgesic activities (**13c**, **g**, **h**). Among the substituted amino derivatives (**26**, **27**) in Table 2, only carbamates **27a**, **b** showed potent anti-inflammatory activities. On the other hand, the sterically small and electron-withdrawing cyano (**23**) and nitro (**24**) derivatives very strongly inhibited AA.

In the previous paper,¹⁾ we reported on the 1-[4-(methylamino)phenyl]pyrazole derivative **39** (Table 3), which possessed not only potent anti-inflammatory ($\text{ED}_{50} = 0.30 \text{ mg/kg}$) and analgesic ($\text{ED}_{30} = 11 \text{ mg/kg}$) activities, but also hydrophilic character. We tried the introduction of the methylamino moiety of **39** in the present study, as depicted in Table 3. Compound **38** and its synthetic intermediate **37** significantly inhibited both AA and pain in the Randall–Selitto assay; in particular, the activities of **38** were superior to those of **39**.

Based on the above evaluation and additional dose–

Table 2. Investigation of 5-Carbamoyl, Sulfamoyl, Amino and Miscellaneous Derivatives

Compd.	R	Adjuvant arthritis % inhibition ^{a)} (10 mg/kg <i>p.o.</i>)	Randall–Selitto relative potency ^{b)} (32 mg/kg <i>p.o.</i>)
13b	CONHMe	74 ^{c)}	1.31 ^{c)}
13c	CONMe ₂	31	1.40 ^{c)}
13f	CONHEt	49 ^{d)}	1.18
13g	CON NMe	57 ^{d)}	1.32 ^{c)}
13g	CON NMe	57 ^{d)}	1.32 ^{c)}
13h	CON(OH)Me	22	1.41 ^{c)}
20a	SO ₂ NH ₂	84 ^{c)}	1.28 ^{d)}
20b	SO ₂ NHMe	89 ^{c)}	1.01
20c	SO ₂ NMe ₂	60 ^{c)}	1.09
26a	NHMe	16 ^{e)}	NT
26b	NMe ₂	57 ^{d,e)}	NT
27a	NHCO ₂ Me	81 ^{c)}	1.15 ^{d)}
27b	NHCO ₂ Et	79 ^{c)}	1.09
27c	NHSO ₂ Me	53 ^{c)}	1.17
17	COMe	47 ^{d)}	1.41 ^{c)}
23	CN	96 ^{c,e)}	0.98 ^{f)}
24	NO ₂	87 ^{c,e)}	1.08

a–d) Refer to Table 1. *e)* 3.2 mg/kg. *f)* 10 mg/kg. NT: Not tested.

Table 3. Investigation of 2-[4-(Substituted amino)phenyl] Derivatives

Compd.	X	Adjuvant arthritis % inhibition ^{a)} (3.2 mg/kg <i>p.o.</i>)	Randall–Selitto relative potency ^{b)} (10 mg/kg <i>p.o.</i>)
37	CHONH	76 ^{c)}	1.26 ^{d)}
38	MeNH	93 ^{c)}	1.35 ^{c)}
39^{e)}		74 ^{c)}	1.30 ^{c)}

a–d) Refer to Table 1. *e)* Ref. 1.

response studies, **6a** and **38** were chosen for further investigation. Compounds **6a** and **38** reduced chronic inflammation in the AA model with ED₅₀ values of 0.030 and 0.32 mg/kg, respectively (Table 4). They also exhibited analgesic activity in the Randall–Selitto assay with ED₃₀ values of 58 and 5.4 mg/kg, respectively. These data indicated that both **6a** and **38** had potency equivalent to that of standard NSAIDs such as indomethacin, but were devoid of gastrointestinal toxicity. The data in Table 4, furthermore, indicate that **6a** and **2** are selective COX-2 inhibitors. On the other hand, the COX-inhibitory activity of **38** was marginal.

To evaluate further the antirheumatic activities of **6a** and **38**, we examined the effect on the type II collagen-induced arthritis (CIA) model in mice in comparison

Table 4. Anti-inflammatory, Analgesic and COX-Inhibitory Activities of **6a**, **38** and Reference Compounds

	6a	38	2^{a)}	Indomethacin
Adjuvant arthritis ^{b)}				
ED ₅₀ (mg/kg <i>p.o.</i>) ^{c)}	0.030	0.32	0.085	0.15
UD ₅₀ (mg/kg <i>p.o.</i>) ^{d)}	> 3.2	> 3.2	> 3.2	0.069
Safety Index ^{e)}	> 106	> 10	> 38	0.46
Randall–Selitto ^{b)}				
ED ₃₀ (mg/kg <i>p.o.</i>)	58	5.4	1.3	3.4
Cyclooxygenase ^{f)}				
COX-1 (IC ₅₀ , μM)	5.6	96	11	0.23
COX-2 (IC ₅₀ , μM)	0.072	> 10	0.020	0.61
Selectivity ^{g)}	77		550	0.38

a) Synthesized according to ref. 8. *b)* In rats. *c)* Uninjected paw. *d)* The median dose for production of GI lesions. *e)* UD₅₀/ED₅₀. *f)* *In vitro* (human). *g)* Selectivity = IC₅₀ (COX-1)/IC₅₀ (COX-2).

Table 5. Effects of **6a**, **38** and Reference Compounds on Type II Collagen-induced Arthritis and DTH Response in Mice

Compd.	Dose (mg/kg <i>p.o.</i>)	CIA (% inhibition)		DTH response % inhibition
		Arthritic index	Anti C II Ab ^{a)}	
6a	0.1	56 ^{b)}	– 3	NT
	1	76 ^{b)}	9	68 ^{b)}
	10	89 ^{b)}	32 ^{c)}	79 ^{b)}
38	1	12	21	1
	3.2	47 ^{c)}	21	NT
	10	61 ^{b)}	26	0
2^{d)}	1	44	2	39 ^{b)}
	3.2	79 ^{b)}	22	38 ^{b)}
	10	65 ^{c)}	17	37 ^{b)}
1^{e)}	10	41 ^{b)}	51 ^{b)}	NT
	100	43 ^{b)}	61 ^{b)}	NT

a) Anti-type II collagen antibody. *b)* *p* < 0.01. *c)* *p* < 0.05 vs. control (Student's *t*-test). *d)* Ref. 8. *e)* Ref. 3. NT: Not tested.

with **1** and **2**. As shown in Table 5, the thiophenes (**6a**, **38**, **2**) suppressed the severity of arthritis measured in terms of the arthritic index more potently than **1**; 56% inhibition was observed even at a dose as low as 0.1 mg/kg of **6a**. Since the induction of arthritis in CIA is closely associated with a high level of both humoral and cellular immunity to type II collagen, the effects of the compounds on these immune responses to collagen were tested. Although **1** reduced the level of anti-collagen antibody in association with the inhibition of the arthritic index, neither **6a**, **38** nor **2** was effective in reducing the plasma level of anti-collagen antibody. This suggested that the suppression of arthritis by these compounds was not due to the reduction of anti-collagen antibody. We next investigated their effects on the cellular immunity to type II collagen monitored in terms of the delayed-type hypersensitivity (DTH) response. Compound **6a** strongly inhibited the DTH response to type II collagen. On the other hand, **38** and **2** showed no or moderate activities.

TNF-α is one of the cytokines critically involved in cellular immunity. Interestingly, **38** as well as **2** strongly inhibited TNF-α production by human peripheral blood mononuclear cells (PBMC) stimulated with lipopolysaccharide (LPS), as shown in Table 6, while indomethacin was inactive (up to 10 μg/ml). Taken together, the results

Table 6. Effects of **38** and **2** on the Production of TNF- α from Human PBMC

Compd.	% Inhibition ^{a)}		
	0.1 μ g/ml	1 μ g/ml	10 μ g/ml
38	40	86	100
2	13	81	101

a) Average values of duplicate experiments.

in Tables 5 and 6 indicate that **6a** and **38** suppressed the arthritic induction in CIA through the inhibition of cellular immunity to collagen.

In conclusion, the chemical modification of **2** has led to the novel compounds **6a** and **38**, which possess a broad spectrum of both anti-inflammatory and immunoregulatory activities, as shown in the AA, Randall-Selitto, COX, CIA, DTH response and TNF- α production assays. These activities suggest that thiophene derivatives such as **6a** or **38** have potential not only to reduce arthritic inflammation and pain, but also to ameliorate immunological abnormalities of RA in the clinic.

Experimental

Melting points were measured on a Mitamura capillary melting-point apparatus and are uncorrected. IR spectra were recorded on a Shimadzu IR-408 spectrophotometer. ¹H-NMR spectra were taken with a Varian EM-390 or a Bruker AC-200 instrument using tetramethylsilane as an internal standard. Electron impact MS were obtained with a Hitachi M80 mass spectrometer. Organic extracts were dried over anhydrous MgSO₄. Column chromatography was performed using Kieselgel 60 (70–230 mesh, E. Merck).

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-(trifluoromethyl)thiophene (6a) A mixture of **2** (7.2 g, 17.5 mmol),⁸⁾ CF₃COONa (9.4 g, 67.4 mmol) and CuI (6.5 g, 33.7 mmol) in *N,N*-dimethylacetamide (109 ml) was refluxed for 5 h. CH₂Cl₂ (200 ml) and 1.5N HCl (200 ml) were added. The insoluble material was removed by filtration and the filtrate was separated. The organic layer was washed with H₂O, dried and evaporated. The residue was chromatographed (toluene–EtOAc, 50:1) over silica gel to yield **3** (3.2 g, 55%) and a crude mixture of **6a** and **3** (3.0 g). Concentrated HNO₃ (2 ml) was added dropwise to 3.0 g of the crude mixture in Ac₂O (50 ml), and the whole was stirred at 0 °C for 2 h. NaHCO₃ (1.0 g) was added and the whole was evaporated. The residue was chromatographed (toluene–EtOAc, 50:1) over silica gel and the product was recrystallized from EtOH to give **6a** (1.04 g, 15%) as pale brown crystals, mp 145–146 °C. IR (Nujol): 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.20 (3H, s), 7.1–8.0 (9H, m). MS *m/z*: 400 (M⁺). Anal. Calcd for C₁₈H₁₂F₄O₂S₂: C, 53.99; H, 3.02. Found: C, 54.47; H, 3.21. Compound **24** (0.6 g, 9%), a by-product of the reaction, was obtained after the second column chromatography.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-(pentafluoroethyl)thiophene (6b) This compound was prepared similarly to **6a**, mp 183–185 °C (toluene–EtOAc). IR (Nujol): 1600, 1550, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.25 (3H, s), 7.1–8.0 (9H, m). MS *m/z*: 450 (M⁺). Anal. Calcd for C₁₉H₁₂F₆O₂S₂: C, 50.67; H, 2.69. Found: C, 51.56; H, 2.71.

3-Chloro-3-(4-fluorophenyl)-2-[4-(methylthio)phenyl]propenal (8) A mixture of POCl₃ (20.6 ml, 221 mmol) and *N,N*-dimethylformamide (DMF) (22.8 ml, 295 mmol) in ClCH₂CH₂Cl (74 ml) was stirred at room temperature for 30 min, then a solution of **7** (38.3 g, 148 mmol)¹³⁾ in ClCH₂CH₂Cl (139 ml) was added. The whole was refluxed for 10 h, cooled, washed twice with H₂O, dried, and evaporated. The residue was washed with toluene to give **8** (28.8 g, 64%) as a pale brown solid.¹⁴⁾ IR (Nujol): 1680, 1600, 1500 cm⁻¹. ¹H-NMR (CDCl₃) δ : 2.53 (3H, s), 6.9–7.8 (8H, m), 9.68 (1H, s).

2-(4-Fluorophenyl)-3-[4-(methylthio)phenyl]thiophene (9) and 2-(4-Fluorophenyl)-3-[4-(methylthio)phenyl]thiophene-5-carboxylic Acid (10) A mixture of **8** (16 g, 52.2 mmol), thioglycolic acid (3.81 ml, 54.7 mmol) and Et₃N (16 ml, 115 mmol) in pyridine (80 ml) was stirred at 70 °C for

1 h and then refluxed for 3 h. The solvent was evaporated, and the residue was dissolved in a mixture of EtOAc and H₂O. The organic layer was separated, washed with dilute HCl, dried and evaporated. The oily residue (20 g) was chromatographed¹⁵⁾ over silica gel to afford **9** (5.8 g, 37%) and **10** (10.8 g, 60%).¹⁴⁾ **9**: mp 81–83 °C, off-white solid. IR (Nujol): 1600, 1505 cm⁻¹. ¹H-NMR (CDCl₃) δ : 2.50 (3H, s), 6.8–7.5 (10H, m). MS *m/z*: 300 (M⁺). **10**: Pale brown solid. IR (Nujol): 2500, 1680, 1600, 1540, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 2.50 (3H, s), 7.1–7.5 (8H, m), 7.80 (1H, s).

A mixture of **10** (17.7 g, 51.3 mmol) and Cu powder (3.6 g, 56.7 mmol) in quinoline (31 ml) was refluxed for 5 h. EtOAc was added and the whole was filtered. The filtrate was washed successively with H₂O, dilute HCl and H₂O, dried and evaporated. The residue was recrystallized from EtOH to give **9** (13.3 g, 86%) as pale brown crystals. The IR, ¹H-NMR, MS and melting point were identical with those of **9** synthesized above.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxylic Acid (11) A mixture of **10** (10.8 g, 31.4 mmol) and H₂O₂ (30%; 9.3 ml, 81.6 mmol) in AcOH (108 ml) was stirred at 70 °C for 3 h. The mixture was cooled to 5 °C, and the precipitates were collected and recrystallized from EtOH to afford **11** (8.1 g, 68%) as pale yellow crystals, mp 264–265 °C. IR (Nujol): 1680, 1600, 1545, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.28 (3H, s), 7.1–8.1 (9H, m). MS *m/z*: 376 (M⁺). Anal. Calcd for C₁₈H₁₃FO₄S₂·1/2H₂O: C, 56.09; H, 3.66. Found: C, 56.12; H, 3.64.

***N*-Methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxamide (13b)** A mixture of **11** (7.1 g, 18.8 mmol) and PCl₅ (4.1 g, 19.8 mmol) in toluene (100 ml) and tetrahydrofuran (THF) (25 ml) was stirred at room temperature for 1 h, then concentrated to dryness to give the acid chloride (8.1 g) as a pale yellow solid. IR (film): 1750, 1600, 1540, 1510 cm⁻¹.

The above acid chloride (1.4 g) was added to a stirred mixture of 25% MeNH₂ (2 ml), THF (15 ml) and H₂O (5 ml) at 5 °C. The resultant mixture was stirred at room temperature for 2 h. EtOAc and H₂O were added, and the organic layer was separated, washed with H₂O, dried and evaporated. The residue was chromatographed (CHCl₃–EtOAc, 1:1) over silica gel and the product was crystallized from Et₂O to give **13b** (1.0 g, 81%) as colorless crystals, mp 169–170 °C. IR (Nujol): 3450, 1650, 1600, 1550, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 2.83 (3H, d, *J* = 4 Hz), 3.26 (3H, s), 7.2–8.1 (9H, m), 8.4–8.8 (1H, m). MS *m/z*: 389 (M⁺), 359. Anal. Calcd for C₁₉H₁₆FNO₃S₂: C, 58.59; H, 4.14; N, 3.60. Found: C, 58.34; H, 4.40; N, 3.54.

The following compounds were prepared from **11** in the same manner as described for **13b**.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxamide (13a): mp 233–234 °C (EtOH–EtOAc), off-white crystals. IR (Nujol): 3480, 3200, 1675, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.28 (3H, s), 7.2–8.1 (11H, m). MS *m/z*: 375 (M⁺), 357. Anal. Calcd for C₁₈H₁₄FNO₃S₂: C, 57.58; H, 3.76; N, 3.73. Found: C, 57.54; H, 4.07; N, 3.54.

***N*-Hydroxy-*N*-methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxamide (13h)**: mp 95–98 °C (iso-Pr₂O), pale brown powder. IR (Nujol): 1600, 1540, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.24 (3H, s), 3.32 (3H, s), 7.2–8.0 (9H, m), 10.78 (1H, s). MS *m/z*: 405 (M⁺). Anal. Calcd for C₁₉H₁₆FNO₄S₂·1/2H₂O: C, 55.06; H, 3.89; N, 3.37. Found: C, 55.18; H, 4.12; N, 2.93.

***N,N*-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxamide (13c)** A mixture of **11** (1.0 g, 2.66 mmol) and 1,1'-carbonyldiimidazole (0.45 g, 2.8 mmol) in THF (15 ml) was refluxed for 2 h. Me₂NH·HCl (0.3 g, 3.68 mmol) and K₂CO₃ (0.39 g, 2.8 mmol) were added, and the resultant mixture was stirred at room temperature for 2 h and then refluxed for 2 h. EtOAc was added, and the mixture was washed successively with H₂O, aqueous NaHCO₃, H₂O and dilute HCl, dried and evaporated. The residue was recrystallized from EtOH to give **13c** (0.76 g, 71%) as pale yellow crystals, mp 172–173 °C. IR (Nujol): 1605, 1545, 1490 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ : 3.21 (3H, s), 3.27 (6H, s), 7.0–8.1 (9H, m). MS *m/z*: 403 (M⁺), 359. Anal. Calcd for C₂₀H₁₈FNO₃S₂: C, 59.53; H, 4.50; N, 3.47. Found: C, 59.38; H, 4.53; N, 3.36.

The following compounds were obtained from **10** or **11** in the same manner as described for **13c**.

***N,N*-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylthio)phenyl]thiophene-5-carboxamide (12c)**: Oil.¹⁴⁾ IR (film): 2920, 1620, 1600, 1540 cm⁻¹. ¹H-NMR (CDCl₃) δ : 2.51 (3H, s), 3.28 (6H, s), 6.8–7.5 (9H, m). MS *m/z*: 371 (M⁺), 327.

N-Ethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carboxamide (**13f**): mp 149–151 °C (EtOH), colorless needles. IR (Nujol): 3400, 1640, 1550, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 1.14 (3H, t, *J* = 7 Hz), 3.25 (3H, s), 3.2–3.4 (2H, m), 7.1–7.6 (6H, m), 7.90 (2H, d, *J* = 8 Hz), 7.96 (1H, s), 8.60 (1H, t, *J* = 6 Hz). MS *m/z*: 403 (M⁺). Anal. Calcd for C₂₀H₁₈FNO₃S₂: C, 59.53; H, 4.50; N, 3.47. Found: C, 59.71; H, 4.50; N, 3.50.

1-{2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenoyl}-4-methylpiperazine (**13g**): mp 68–72 °C (CHCl₃), off-white powder. IR (Nujol): 1610, 1545, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.12 (3H, s), 2.37 (4H, brs), 3.23 (3H, s), 3.72 (4H, brs), 7.1–8.0 (9H, m). MS *m/z*: 458 (M⁺). Anal. Calcd for C₂₃H₂₃FNO₃S₂·1/2H₂O: C, 59.08; H, 4.96; N, 5.99. Found: C, 59.34; H, 5.08; N, 5.35.

N,N-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylthio)phenyl]-5-thenylamine Hydrochloride (**14c**) A solution of **12c** (1.95 g, 5.26 mmol) in benzene (15 ml) was added dropwise to an ice-cooled suspension of LiAlH₄ (0.27 g, 7 mmol) in Et₂O (10 ml). The mixture was refluxed for 9 h and cooled. Then 4 N NaOH (10 ml) was added dropwise, and EtOAc was added to the mixture. The insoluble material was removed by filtration and the filtrate was washed with H₂O, dried and evaporated. The residue was chromatographed (toluene–EtOAc, 2:1) over silica gel to afford **14c** (1.5 g, 78%) as a yellow oil.¹⁴ IR (film): 2770, 1600, 1510 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.35 (6H, s), 2.50 (3H, s), 3.66 (2H, s), 6.8–7.5 (9H, m). MS *m/z*: 357 (M⁺), 313.

N,N-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenylamine Hydrochloride (**15c**) A mixture of **14c** (1.4 g, 4 mmol) and mCPBA (2.6 g, 12 mmol) in CH₂Cl₂ (25 ml) was stirred overnight. The insoluble material was removed by filtration, and the filtrate was washed with aqueous NaHCO₃, dried and evaporated. The residue was dissolved in 30% HCl in MeOH (1 ml) and the solvent was evaporated. The residue was pulverized with Et₂O to give **15c** (1.4 g, 81%) as a pale brown powder, mp 192–194 °C. IR (Nujol): 3400, 2550, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.23 (3H, s), 3.55 (6H, s), 5.25 (2H, s), 7.2–8.1 (9H, m). MS *m/z*: 389 (M⁺), 345. Anal. Calcd for C₂₀H₂₀FNO₂S₂·HCl·H₂O: C, 54.10; H, 5.22; N, 3.15. Found: C, 53.64; H, 4.74; N, 3.09.

The following compounds were prepared from **14d** or **14e**¹⁰ in the same manner as described for **15c**.

N-Ethyl-*N*-methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenylamine Hydrochloride (**15d**): mp 95–100 °C (dec.) (EtOH), off-white solid. IR (Nujol): 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 1.40 (3H, t, *J* = 8 Hz), 3.20 (3H, s), 3.37 (3H, s), 3.77 (2H, q, *J* = 8 Hz), 5.15 (2H, s), 7.1–8.0 (9H, m). MS *m/z*: 403 (M⁺), 345. Anal. Calcd for C₂₁H₂₂FNO₂S₂·HCl·2H₂O: C, 53.05; H, 4.63; N, 2.94. Found: C, 52.87; H, 4.83; N, 2.57.

N,N-Diethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenylamine (**15e**): mp 98–102 °C (hexane–EtOAc), pale yellow powder. IR (Nujol): 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 1.20 (6H, t, *J* = 7 Hz), 3.0–3.3 (4H, m), 3.23 (3H, s), 4.52 (2H, s), 7.1–7.9 (9H, m). MS *m/z*: 417 (M⁺), 345. Anal. Calcd for C₂₂H₂₄FNO₂S₂·5/2H₂O: C, 57.14; H, 5.20; N, 3.03. Found: C, 57.52; H, 5.68; N, 2.80.

N-(Benzyloxycarbonyl)-2-(4-fluorophenyl)-3-[4-(methylthio)phenyl]-5-thenylamine (**16a**) Benzyl chloroformate (1.0 ml, 7.0 mmol) was added dropwise to an ice-cooled solution of **14a** (2.1 g, 6.36 mmol)¹⁰ and Et₃N (1.27 ml, 9.12 mmol) in CH₂Cl₂ (20 ml). The mixture was stirred at room temperature for 3 h, washed successively with H₂O and aqueous NaHCO₃, dried and evaporated. The residue was chromatographed (CHCl₃) over silica gel to afford **16a** (1.3 g, 48%) as a yellow oil.¹⁴ ¹H-NMR (DMSO-*d*₆) δ: 2.45 (3H, s), 4.39 (2H, d, *J* = 6 Hz), 5.14 (2H, s), 6.9–7.5 (14H, m), 8.00 (1H, t, *J* = 6 Hz). MS *m/z*: 463 (M⁺).

N-(Benzyloxycarbonyl)-*N*-methyl-2-(4-fluorophenyl)-3-[4-(methylthio)phenyl]-5-thenylamine (**16b**): This compound was obtained from **14b**¹⁰ in the same manner as described for **16a**: Yellow oil.¹⁴ ¹H-NMR (DMSO-*d*₆) δ: 2.45 (3H, s), 2.93 (3H, s), 4.60 (2H, s), 5.14 (2H, s), 7.1–7.4 (14H, m). MS *m/z*: 477 (M⁺).

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenylamine Hydrobromide (**15a**) **16a** (1.3 g, 2.81 mmol) was oxidized with mCPBA (1.3 g, 6.04 mmol) at room temperature overnight, and chromatographed (toluene–EtOAc, 2:1) over silica gel. The resultant sulfone (oil; 0.78 g, 1.58 mmol) was dissolved in 30% HBr in AcOH (10 ml). The solution was stirred at room temperature for 1 h and evaporated. The residue was washed with iso-Pr₂O to give **15a** (0.7 g, 53%) as a white powder, mp > 200 °C. IR (Nujol): 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.24 (3H, s), 4.32 (2H, s), 7.1–7.5 (7H, m), 7.80 (2H, d, *J* = 8 Hz), 8.37 (2H, brs). MS *m/z*: 361 (M⁺). Anal. Calcd for C₁₈H₁₆FNO₂S₂·

HBr·2/3H₂O: C, 47.58; H, 4.07; N, 3.08. Found: C, 48.02; H, 4.03; N, 2.59.

N-Methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thenylamine Hydrobromide (**15b**): This compound was obtained from **16b** in the same manner as described for **15a**: mp 100–110 °C (dec.) (Et₂O). IR (Nujol): 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.64 (3H, s), 3.24 (3H, s), 4.44 (2H, s), 7.2–7.6 (7H, m), 7.90 (2H, d, *J* = 8 Hz), 9.00 (2H, brs). MS *m/z*: 375 (M⁺). Anal. Calcd for C₁₉H₁₈FNO₂S₂·HBr·H₂O: C, 48.30; H, 3.84; N, 2.53. Found: C, 48.09; H, 4.13; N, 2.53.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene (**3**): This compound was obtained from **9** in the same manner as described for **15c**: mp 155–156 °C (EtOH), pale brown solid. IR (Nujol): 1595, 1505 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.23 (3H, s), 7.0–8.0 (10H, m). MS *m/z*: 332 (M⁺). Anal. Calcd for C₁₇H₁₃FO₂S₂: C, 61.42; H, 3.94. Found: C, 61.46; H, 3.91.

5-Acetyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene (**17**) TiCl₄ (2.7 ml, 24.2 mmol) was added dropwise to a stirred solution of **3** (5.0 g, 15.1 mmol) and AcCl (2.2 ml, 30.2 mmol) in benzene (50 ml) at 5–10 °C. The mixture was stirred at room temperature for 4 h, poured into ice-H₂O, and extracted with EtOAc. The extract was washed with aqueous NaHCO₃, dried and evaporated. The residue was recrystallized from EtOH–EtOAc to give **17** (3.7 g, 66%) as pale brown crystals, mp 169–172 °C. IR (Nujol): 1670, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.58 (3H, s), 3.21 (3H, s), 7.0–8.0 (8H, m), 8.11 (1H, s). MS *m/z*: 374 (M⁺), 359. Anal. Calcd for C₁₉H₁₅FO₃S₂: C, 60.94; H, 4.04. Found: C, 60.46; H, 4.12.

Methyl 2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-acetate (**18**) A mixture of **17** (1.0 g, 2.67 mmol), Ti(NO₃)₃·3H₂O (1.4 g, 3.21 mmol) and 70% HClO₄ (3 ml) in MeOH (15 ml) and dioxane (7 ml) was stirred at room temperature for 7 h. The insoluble material was removed by filtration, and CHCl₃ and H₂O were added to the filtrate. The organic layer was separated, dried and evaporated to give **18** (1.2 g) as an oil.¹⁴ IR (film): 1740, 1670, 1600, 1510 cm⁻¹. ¹H-NMR (CDCl₃) δ: 3.10 (3H, s), 3.81 (3H, s), 3.91 (2H, s), 6.9–8.0 (9H, m). MS *m/z*: 404 (M⁺), 345.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-acetic Acid (**19**) A mixture of **18** (2 g, 4.95 mmol) and 4 N NaOH (3.7 ml, 14.8 mmol) in THF (20 ml) was stirred at room temperature for 1 h. H₂O and toluene were added, and the aqueous layer was separated, acidified with HCl and extracted with EtOAc. The extract was dried and evaporated. The residue was recrystallized from EtOH–H₂O to give **19** (1.6 g, 81%) as pale orange crystals, mp 171–173 °C. IR (Nujol): 1700, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.24 (3H, s), 3.93 (2H, s), 7.1–8.0 (9H, m). MS *m/z*: 390 (M⁺), 345. Anal. Calcd for C₁₉H₁₅FO₄S₂: C, 58.45; H, 3.87. Found: C, 58.18; H, 3.84.

N-Methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-sulfonamide (**20b**) **3** (1.1 g, 3.31 mmol) was added portionwise to ClSO₃H (1 ml, 15 mmol) and the resultant mixture was stirred at room temperature for 1 h. The mixture was added to an ice-cooled mixture of 25% MeNH₂ (20 ml) and THF (20 ml), and the whole was stirred at 0 °C for 1 h and then extracted with EtOAc. The extract was washed with H₂O, dried and evaporated. The residue was chromatographed (CHCl₃–MeOH, 20:1) over silica gel and the product was recrystallized from EtOH to give **20b** (1.1 g, 77%) as colorless crystals, mp 199–201 °C. IR (Nujol): 3300, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.63 (3H, s), 3.25 (3H, s), 7.2–8.0 (9H, m). MS *m/z*: 425 (M⁺). Anal. Calcd for C₁₈H₁₆FNO₄S₃: C, 50.81; H, 3.79; N, 3.29. Found: C, 50.59; H, 3.78; N, 3.24.

The following compounds were prepared in the same manner as described for **20b**.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-sulfonamide (**20a**): mp 195–197 °C (Et₂O), pale brown crystals. IR (Nujol): 3340, 3250, 1600 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.30 (3H, s), 7.2–8.1 (11H, m). MS *m/z*: 411 (M⁺). Anal. Calcd for C₁₇H₁₄FNO₄S₃: C, 49.62; H, 3.43; N, 3.40. Found: C, 50.36; H, 3.53; N, 3.06.

N,N-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-sulfonamide (**20c**): mp 189–191 °C (EtOH). IR (Nujol): 1600, 1535, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.79 (6H, s), 3.25 (3H, s), 7.2–8.0 (9H, m). MS *m/z*: 439 (M⁺). Anal. Calcd for C₁₉H₁₈FNO₄S₃·2/3H₂O: C, 50.54; H, 4.32; N, 3.10. Found: C, 50.25; H, 4.06; N, 3.03.

4'-[2-(4-Fluorophenyl)-5-(trifluoromethyl)-3-thienyl]methanesulfonamide (**22**) A mixture of **21** (0.49 g, 1.45 mmol)¹⁰ and MeSO₂Cl (0.135 ml, 1.75 mmol) in pyridine (5 ml) was stirred overnight at room temperature. The solvent was evaporated, and the residue was dissolved in THF (5 ml)

and treated with 4N NaOH (1.5 ml) for 30 min. EtOAc and dilute HCl were added, and the organic layer was separated, washed with brine, dried and evaporated. The residue was chromatographed (toluene–EtOAc, 10:1) over silica gel and the product was washed with EtOH to give **22** (0.52 g, 86%) as pale brown crystals, mp 150–152 °C. IR (Nujol): 3320, 1600, 1560, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.02 (3H, s), 7.1–7.5 (8H, m), 7.85 (1H, s), 9.90 (1H, s). MS *m/z*: 415 (M⁺), 336. *Anal.* Calcd for C₁₈H₁₃F₄NO₂S₂: C, 52.04; H, 3.15; N, 3.37. Found: C, 51.99; H, 3.22; N, 3.20.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]thiophene-5-carbonitrile (23) A mixture of **13a** (4.8 g, 12.8 mmol) and MeSO₂Cl (8.8 g, 76.8 mmol) in pyridine (25 ml) was stirred at 50 °C for 4 h. The mixture was evaporated and the residue was triturated in dilute HCl. The precipitates were collected and recrystallized from EtOH to afford **23** (3.6 g, 79%) as crystals, mp 139–140 °C. IR (Nujol): 2220, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.24 (3H, s), 7.1–7.6 (6H, m), 7.91 (2H, d, *J* = 8 Hz), 8.24 (1H, s). MS *m/z*: 357 (M⁺). *Anal.* Calcd for C₁₈H₁₂FNO₂S₂: C, 60.56; H, 3.39; N, 3.92. Found: C, 60.68; H, 3.44; N, 3.85.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-nitrothiophene (24) HNO₃ (d = 1.42; 2.6 ml, 41.7 mmol) was added dropwise to a stirred solution of **3** (6.0 g, 18.1 mmol) in Ac₂O (98 ml) at 0 °C. The mixture was stirred at 0 °C for 2 h, treated with NaHCO₃ (1 g), and evaporated. A solution of the residue in EtOAc was washed with aqueous NaHCO₃, dried and evaporated. The residue was chromatographed (toluene–EtOAc, 20:1) over silica gel and the product was recrystallized from EtOH to give **24** (4.4 g, 48%) as yellow crystals, mp 155–156 °C. IR (Nujol): 3100, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.28 (3H, s), 7.1–8.1 (8H, m), 8.42 (1H, s). MS *m/z*: 377 (M⁺). *Anal.* Calcd for C₁₇H₁₂FNO₄S₂: C, 54.10; H, 3.21; N, 3.71. Found: C, 54.09; H, 3.57; N, 3.63.

2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thiophenamine (25) A mixture of **24** (3.6 g, 9.55 mmol), Fe powder (3.6 g) and NH₄Cl (0.36 g) in EtOH (58 ml) and H₂O (22 ml) was stirred under reflux for 1 h. The insoluble material was removed by filtration and washed with DMF (40 ml). The filtrate was evaporated and the residue was triturated with H₂O. The precipitates were collected and recrystallized from EtOH to give **25** (2.7 g, 82%) as a pale brown powder, mp 207–209 °C. IR (Nujol): 3450, 3350, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.25 (3H, s), 5.8 (2H, s), 6.8–8.0 (9H, m). MS *m/z*: 347 (M⁺). *Anal.* Calcd for C₁₇H₁₄FNO₂S₂·1/6H₂O: C, 58.27; H, 4.12; N, 4.00. Found: C, 57.97; H, 4.27; N, 3.97.

***N*-Methyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thiophenamine (26a) and *N,N*-Dimethyl-2-(4-fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thiophenamine (26b)** MeI (3 g, 21.3 mmol) was added to an ice-cooled mixture of **25** (1.85 g, 5.33 mmol) and K₂CO₃ (2.5 g, 16 mmol) in DMF (20 ml). The whole was stirred at room temperature for 5 h and poured into ice-H₂O. The precipitates were collected and chromatographed (toluene–EtOAc, 4:1) over silica gel, and the desired, isolated products were independently recrystallized from EtOH to give **26a** (0.9 g, 58%) and **26b** (0.5 g, 31%). **26a**: mp 183–185 °C. IR (Nujol): 1600, 1515 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.93 (3H, s), 3.06 (3H, s), 6.05 (1H, s), 6.8–7.9 (8H, m). MS *m/z*: 361 (M⁺). *Anal.* Calcd for C₁₈H₁₆FNO₂S₂·5/9H₂O: C, 58.20; H, 4.64; N, 3.77. Found: C, 57.87; H, 4.20; N, 3.31. **26b**: mp 130–135 °C (dec.). IR (Nujol): 1600, 1555, 1520 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.97 (6H, s), 3.06 (3H, s), 5.94 (1H, s), 6.9–7.9 (8H, m). MS *m/z*: 375 (M⁺). *Anal.* Calcd for C₁₉H₁₈FNO₂S₂: C, 60.80; H, 4.80; N, 3.73. Found: C, 60.84; H, 4.59; N, 3.57.

Methyl *N*-[2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thienyl]-carbamate (27a) ClCO₂Me (0.23 ml, 3.02 mmol) in MeCN (1 ml) was added dropwise to a stirred solution of **25** (1.1 g, 3.02 mmol) and pyridine (0.24 ml, 3.02 mmol) in MeCN (8 ml) and THF (10 ml) at –20 °C. The mixture was stirred at 5 °C for 1 h, and EtOAc and H₂O were added. The organic layer was dried and evaporated. The residue was chromatographed (toluene–EtOAc, 10:1) over silica gel and the product was recrystallized from EtOH to give **27a** (0.84 g, 69%) as pale red crystals, mp 103–108 °C (dec.). IR (Nujol): 3330, 1720, 1600, 1580, 1540 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.28 (3H, s), 3.80 (3H, s), 6.77 (1H, s), 7.2–8.1 (8H, m), 10.94 (1H, s). MS *m/z*: 405 (M⁺), 373. *Anal.* Calcd for C₁₉H₁₆FNO₄S₂: C, 56.28; H, 3.98; N, 3.45. Found: C, 55.73; H, 3.82; N, 3.29.

Ethyl *N*-[2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thienyl]carbamate (**27b**): This compound was prepared in the same manner as described for **27a**: mp 70–75 °C (hexane–EtOAc), pale brown

powder. IR (Nujol): 3330, 1720, 1580, 1530 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 1.27 (3H, t, *J* = 7 Hz), 3.22 (3H, s), 4.19 (2H, q, *J* = 7 Hz), 6.67 (1H, s), 7.1–7.9 (8H, m), 10.91 (1H, s). MS *m/z*: 419 (M⁺), 373. *Anal.* Calcd for C₂₀H₁₈FNO₄S₂: C, 57.27; H, 4.32; N, 3.36. Found: C, 57.55; H, 4.65; N, 3.06.

***N*-[2-(4-Fluorophenyl)-3-[4-(methylsulfonyl)phenyl]-5-thienyl]-methanesulfonamide (27c)** This compound was prepared in the same manner as described for **22**: mp 87–90 °C (toluene–EtOAc), pale brown powder. IR (Nujol): 3200, 1600, 1510 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 3.17 (3H, s), 3.26 (3H, s), 6.98 (1H, s), 7.1–8.0 (8H, m), 10.4 (1H, br s). MS *m/z*: 425 (M⁺), 346. *Anal.* Calcd for C₁₈H₁₆FNO₄S₃: C, 50.81; H, 3.79; N, 3.29. Found: C, 50.55; H, 3.95; N, 3.21.

3-[4-(Methylthio)phenyl]-2-(4-nitrophenyl)acrylic Acid (29) A mixture of **28** (2.0 g, 13.2 mmol), 4-nitrophenylacetic acid (2.4 g, 13.2 mmol) and NaOMe (0.8 g, 14.8 mmol) in Ac₂O (10 ml) was refluxed for 24 h. CHCl₃ and aqueous NaHCO₃ were added, and the aqueous layer was separated, acidified with HCl and extracted with EtOAc. The extract was washed with H₂O, dried and evaporated, and the residue was washed with EtOH to afford **29** (1.6 g, 42%), mp 217–219 °C (dec.).¹⁴ IR (Nujol): 1690, 1670, 1610, 1590, 1515 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.42 (3H, s), 6.98 (2H, d, *J* = 8 Hz), 7.10 (2H, d, *J* = 8 Hz), 7.42 (2H, d, *J* = 8 Hz), 7.83 (1H, s), 8.25 (2H, d, *J* = 8 Hz), 12.8 (1H, s). MS *m/z*: 315 (M⁺).

2-[4-(Methylthio)phenyl]-1-(4-nitrophenyl)ethanone (30) A mixture of **29** (1.6 g, 5.08 mmol) and PCl₅ (1.1 g, 5.59 mmol) in CH₂Cl₂ (20 ml) was stirred at room temperature for 1 h. The solvent was evaporated to give the acryloyl chloride as a solid.

A solution of the above chloride in Me₂CO was added dropwise to an ice-cooled mixture of NaN₃ (0.5 g, 7.62 mmol), H₂O (10 ml) and Me₂CO (10 ml); the pH of the reaction mixture was kept at 7.5 by addition of aqueous Na₂CO₃. The whole was stirred at room temperature for 2 h and extracted with EtOAc. The extract was washed with brine, dried and evaporated to afford the acryloyl azide as a solid.

A mixture of the above azide (2.0 g), AcOH (20 ml) and H₂O (10 ml) was refluxed for 3 h and poured into ice-H₂O. The precipitates were collected and washed with H₂O to give **30** (1.1 g, 75%), mp 105–107 °C.¹⁴ IR (Nujol): 1695, 1600, 1520 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.47 (3H, s), 4.28 (2H, s), 7.1–7.3 (4H, m), 8.13 (2H, d, *J* = 8 Hz), 8.27 (2H, d, *J* = 8 Hz). MS *m/z*: 287 (M⁺).

Ethyl 3-[4-(Methylthio)phenyl]-2-(4-nitrophenyl)thiophene-5-carboxylate (**31**): This compound was prepared from **30** in the same manner as described for **10**: Oil.¹⁴ IR (Nujol): 1710, 1600, 1520 cm⁻¹. ¹H-NMR (CDCl₃) δ: 1.30 (3H, t, *J* = 7 Hz), 2.48 (3H, s), 4.20 (2H, q, *J* = 7 Hz), 7.1–7.3 (4H, m), 7.47 (2H, d, *J* = 8 Hz), 7.82 (1H, s), 8.18 (2H, d, *J* = 8 Hz). MS *m/z*: 399 (M⁺).

3-[4-(Methylthio)phenyl]-2-(4-nitrophenyl)thiophene-5-carboxylic Acid (**32**): This compound was prepared from **31** in the same manner as described for **19**: mp 190–195 °C (dec.) (H₂O).¹⁴ IR (Nujol): 1690, 1595, 1515 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.47 (3H, s), 6.6–8.3 (9H, m). MS *m/z*: 371 (M⁺).

3-[4-(Methylthio)phenyl]-2-(4-nitrophenyl)thiophene (**33**): This compound was prepared from **32** in the same manner as described for **9**: mp 135–140 °C (EtOAc), pale brown solid.¹⁴ ¹H-NMR (DMSO-*d*₆) δ: 2.47 (3H, s), 6.8–8.2 (10H, m).

5-Bromo-3-[4-(methylthio)phenyl]-2-(4-nitrophenyl)thiophene (34) Following the procedure reported previously,⁸ **33** was treated with Br₂ to give **34** (90% yield) as a pale brown powder, mp 230 °C (dec.) (EtOAc).¹⁴ IR (Nujol): 1600, 1515 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.50 (3H, s), 7.1–8.2 (9H, m). MS *m/z*: 407, 405.

5-Bromo-2-[4-(formylamino)phenyl]-3-[4-(methylthio)phenyl]thiophene (35) A mixture of Ac₂O (3.3 g, 32.3 mmol) and HCO₂H (1.5 g, 32.6 mmol) was stirred at 50 °C for 30 min. 2-(4-Aminophenyl)-5-bromo-3-[4-(methylthio)phenyl]thiophene (4.0 g, 10.6 mmol), which was obtained from **34** in the same manner as described for **25**,¹⁰ was added to the mixture and the whole was stirred at room temperature for 2 h, then poured into ice-H₂O. The precipitates were collected and washed with H₂O to give **35** (3.8 g, 88%), mp 150–160 °C (dec.).¹⁴ IR (Nujol): 1690, 1600 cm⁻¹. ¹H-NMR (DMSO-*d*₆) δ: 2.48 (3H, s), 6.8–7.7 (9H, m), 8.29 (1H, s), 9.40 (1H, s). MS *m/z*: 405, 403.

5-Bromo-2-[4-(*N*-methylformylamino)phenyl]-3-[4-(methylthio)phenyl]thiophene (36) NaH (60%; 0.45 g, 11.3 mmol) was added to an ice-cooled solution of **35** (3.8 g, 9.41 mmol) in DMF (50 ml). The mixture was stirred at 5 °C for 30 min, and then MeI (3 g, 26.8 mmol) was added. The whole was stirred for 2 h and evaporated. Ice-H₂O was

added to the residue, and the precipitates were collected and washed with H₂O to give **36** (3.8 g, 97%), mp 150–155 °C (dec.).¹⁴⁾ IR (Nujol): 1685, 1600 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.48 (6H, s), 6.9–7.6 (9H, m), 8.52 (1H, s). MS *m/z*: 419, 417.

5-Bromo-2-[4-(formylamino)phenyl]-3-[4-(methylsulfinyl)phenyl]thiophene (37) A mixture of **35** (1.0 g, 2.48 mmol) and mCPBA (0.5 g, 2.32 mmol) in CH₂Cl₂ (80 ml) was stirred at 0 °C for 1 h. The mixture was washed successively with aqueous NaHCO₃ and H₂O, dried and evaporated. The residue was chromatographed (CH₂Cl₂–Me₂CO, 5:1) over silica gel to give **37** (1.0 g, 96%) as a pale yellow powder, mp 130–133 °C (dec.). IR (Nujol): 3250, 1690, 1600, 1515 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.76 (3H, s), 6.9–8.1 (10H, m), 8.36 (1H, s). MS *m/z*: 421, 419. Anal. Calcd for C₁₈H₁₄BrNO₂S₂: C, 51.43; H, 3.33; N, 3.33. Found: C, 51.56; H, 3.70; N, 3.15.

5-Bromo-2-[4-(methylamino)phenyl]-3-[4-(methylsulfinyl)phenyl]thiophene (38) A mixture of 5-bromo-2-[4-(*N*-methylformylamino)phenyl]-3-[4-(methylsulfinyl)phenyl]thiophene (2.0 g, 4.61 mmol), which was obtained from **36** in the same manner as described for **37**,¹⁰⁾ and concentrated HCl (10 ml) in MeOH (100 ml) and THF (20 ml) was stirred at room temperature for 2 h. The mixture was evaporated and the residue was triturated in aqueous NaHCO₃ to give a powder. The crude powder was chromatographed (CHCl₃–MeOH, 25:1) over silica gel to afford **38** (0.5 g, 29%), mp 160–164 °C (dec.). IR (Nujol): 3350, 1610, 1520 cm⁻¹. ¹H-NMR (CDCl₃) δ: 2.74 (3H, s), 2.76 (3H, s), 6.4–7.7 (9H, m). MS *m/z*: 407, 405. Anal. Calcd for C₁₈H₁₆BrNOS₂·H₂O: C, 50.94; H, 4.28; N, 3.30. Found: C, 50.66; H, 3.82; N, 3.14.

Biological Methods. AA, CIA, Randall-Selitto and COX Assays These experiments were carried out according to the procedures described in previous reports.^{3,11)}

Effects on the DTH Response to Bovine Type II Collagen in Mice Seven male DBA/1 mice were used per group. The mice were sensitized at the tail base with 125 μg of type II collagen emulsified in complete Freund's adjuvant containing *Mycobacterium tuberculosis* strain H37Rv (Wako Pure Chemical Industries Ltd., Osaka, Japan). Two weeks later, a 0.04 ml challenge dose of 2.5 mg/ml type II collagen in phosphate-buffered saline (PBS) was injected into the plantar region of the right hind foot, and 0.04 ml of PBS into the left hind foot to act as a control. Twenty-four hours after challenge, the volumes of the two hind feet were measured with a volume meter (Muromachi MK-550). The drug was administered orally on consecutive days (except holidays), starting from the sensitization. Data were expressed as % inhibition compared with the vehicle control for each study.

Effects on the Production of TNF-α (in Vitro) Human PBMC were isolated from heparinized venous blood from 2 healthy adult male volunteers. PBMC (1.0 × 10⁶ cells in 1.0 ml culture medium/well) were stimulated with LPS (Sigma, St. Louis, MO) 10 μg/ml for 24 h at 37 °C in the presence or absence of a drug. The PBMC culture supernatants were assayed for TNF-α levels by enzyme-linked immunosorbent assay (ELISA). The effects of the drug on TNF-α production were expressed as % inhibition compared with values obtained in the absence of the drug.

Acknowledgments The authors are grateful to Drs. H. Senoh, T. Fujii, and their colleagues in the pharmacological division for biological assays, and Drs. K. Sakane and G. W. Spears in the Medicinal Chemistry Research Laboratories for useful suggestions during the preparation of this paper. Thanks are also due to the staff members of the Analytical Research Laboratories for elemental analyses and spectral measurements.

References and Notes

- 1) Part V: Tsuji K., Konishi N., Spears G.W., Ogino T., Nakamura K., Tojo T., Ochi T., Shimojo F., Senoh H., Matsuo M., *Chem. Pharm. Bull.*, **45**, 1475–1481 (1997).
- 2) a) Madhok R., *Lancet*, **346**, 481–485 (1995); b) Parnham M. J., *Exp. Opin. Invest. Drugs*, **4**, 777–779 (1995); c) Elliott M. J., Maini R. N., Feldmann M., Long-Fox A., Charles P., Katsikis P., Brennan F. M., Walker J., Bijl H., Ghayeb J., Woody J. N., *Arthritis Rheum.*, **36**, 1681–1690 (1993); d) Maini R. N., Elliott M. J., Brennan F. M., Feldmann M., *Clin. Exp. Immunol.*, **101**, 207–212 (1995); e) Kuo E. A., Hambleton P. T., Kay D. P., Evans P. L., Matharu S. S., Little E., McDowall N., Jones C. B., Hedgecock C. J. R., Yea C. M., Chan A. W. E., Hairsine P. W., Ager I. R., Tully W. R., Williamson R. A., Westwood R., *J. Med. Chem.*, **39**, 4608–4621 (1996); f) Niwayama S., Turk B. E., Liu J. O., *ibid.*, **39**, 3044–3045 (1996); g) Baba A., Kawamura N., Makino H., Ohta Y., Taketomi S., Sohda T., *ibid.*, **39**, 5176–5182 (1996); h) Various antirheumatic agents were reported at the American College of Rheumatology 60th National Scientific Meeting (Orlando, FL, 1996), see *Arthritis Rheum.*, **39**, No.9 (Supplement) (1996).
- 3) Nakamura K., Tsuji K., Konishi N., Okumura H., Matsuo M., *Chem. Pharm. Bull.*, **41**, 894–906 (1993).
- 4) a) Gans K. R., Balles E. A., Galbraith W., Haber S. B., Kerr J. S., Newton R. C., Schmidt W. K., Smith C., Hewes W. E., Ackerman N. R., *Pharmacologist*, **29**, 138 (1987); b) Gans K. R., Galbraith W., Roman R. J., Haber S. B., Kerr J. S., Schmidt W. K., Smith C., Hewes W. E., Ackerman N. R., *J. Pharmacol. Exp. Ther.*, **254**, 180–187 (1990).
- 5) a) Leblanc Y., Gauthier J. Y., Ethier D., Guay J., Mancini J., Riendeau D., Tagari P., Vickers P., Wong E., Prasit P., *Bioorg. Med. Chem. Lett.*, **5**, 2123–2128 (1995); b) Bertenshaw S. R., Talley J. J., Rogier D. J., Graneto M. J., Rogers R. S., Kramer S. W., Penning T. D., Koboldt C. M., Veenhuizen A. W., Zhang Y., Perkins W. E., *ibid.*, **5**, 2919–2922 (1995); c) Gauthier J. Y., Leblanc Y., Black W. C., Chan C. C., Cromlish W. A., Gordon R., Kennedey B. P., Lau C. K., Léger S., Wang Z., Ethier D., Guay J., Mancini J., Riendeau D., Tagari P., Vickers P., Wong E., Xu L., Prasit P., *ibid.*, **6**, 87–92 (1996); d) Pinto D. J. P., Copeland R. A., Covington M. B., Pitts W. J., Batt D. G., Orwat M. J., Lam G. N., Joshi A., Chan Y. C., Wang S., Trzaskos J. M., Magolda R. L., Kornhauser D. M., *ibid.*, **6**, 2907–2912 (1996).
- 6) Plummer E. L., U.S. Patent 4617316 (1986) [*Chem. Abstr.*, **106**, 101899n (1987)].
- 7) a) Matsui K., Tobita E., Ando M., Kondo K., *Chem. Lett.*, **1981**, 1719–1720; b) Carr G. E., Chambers R. D., Holmes T. F., Parker D. G., *J. Chem. Soc., Perkin Trans. 1*, **1988**, 921–926.
- 8) Haber S. B., U.S. Patent 4820827 (1989) [*Chem. Abstr.*, **100**, 22571x (1984)].
- 9) McKillop A., Swann B. P., Taylor E. C., *J. Am. Chem. Soc.*, **93**, 4919–4920 (1971).
- 10) Matsuo M., Tsuji K., Konishi N., Nakamura K., PCT Int. Appl. WO 91 19708 (1991) [*Chem. Abstr.*, **116**, 194141d (1992)].
- 11) Tsuji K., Nakamura K., Konishi N., Tojo T., Ochi T., Senoh H., Matsuo M., *Chem. Pharm. Bull.*, **45**, 987–995 (1997).
- 12) Bender P. E., Hill D. T., U.S. Patent 4153706 (1979) [*Chem. Abstr.*, **91**, 74622e (1979)].
- 13) Hinkley D. F., Conn J. B., U.S. Patent 3647858 (1972) [*Chem. Abstr.*, **76**, 153425m (1972)].
- 14) This product was used for the next step without further purification.
- 15) Elution was started with hexane-toluene (10:1), then continued with toluene, CHCl₃, and finally CHCl₃–MeOH (5:1).