Synthesis and Antiinflammatory Activity of 7-Methanesulfonylamino-6-phenoxychromones. Antiarthritic Effect of the 3-Formylamino Compound (T-614) in Chronic Inflammatory Disease Models

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A group of derivatives of 7-methanesulfonylamino-6-phenoxychromone (1) at the pyrone and phenoxy rings was synthesized starting with 4-chloro-3-nitroanisole and evaluated against acute and chronic inflammations in oral administration in animals. Significant potency in the rat models of carrageenin-induced edema (CPE) and adjuvant-induced arthritis (AA) was realized with 2′-fluoro and 2′,4′-difluoro derivatives (9a and 9d), and 3-formylamino derivative (19a) and its 2′-fluoro and 2′,4′-difluoro compounds (22a and 22d), displaying AA therapeutic effect of ED_{40}=2.5—7.1 mg/kg/d for 7 d and AA prophylactic effect of 53—70% inhibition at the dosage of 3 mg/kg/d for 22 d. To identify a candidate for further pharmacological study, the five compounds were subjected to evaluation of their gastro-ulcerogenic liability, leading to selection of the fluorine-free compound 19a which did not cause acute ulceration at 300 mg/kg in oral administration in rats. Compound 19a (ED_{40}=3.6 mg/kg in established AA) possessed good therapeutic efficacy against type II collagen-induced arthritis in DBA/1J mice with doses of 30 and 100 mg/kg, suggesting the development of 19a (designated T-614) as a prospective disease-modifying antirheumatic agent. In addition, a preparative-scale synthetic route to T-614 has been established.

Key words antiinflammatory activity; methanesulfoanilide; chromone; structure–activity relationship; antiarthritic activity; T-614

The majority of currently marketed nonsteroidal antiinflammatory drugs (NSAIDs) are chemically hydroxylic acids as represented by indomethacin and ibuprofen (carboxylic) and piroxicam (enolic). These NSAIDs are believed to exhibit their antiinflammatory activity by inhibiting the enzyme cyclooxygenase (COX) that catalyzes the biosynthesis of prostaglandins and thromboxane from arachidonic acid, the mechanism correlated with unwanted side-effects such as gastrointestinal and renal toxicity.[1] It was in the mid-1970s that R-805, or nimesulide [*N*-(4-nitro-2-phenoxyphenyl)-methanesulfonamide] was reported by Swingle *et al*.[2] as the first member of the methanesulfoanilide class of NSAID (N–H acid of pK_a 6—7), which was characterized by low gastrointestinal toxicity in conventional animal models. Since then, structural modifications of nimesulide aimed at enhancing its antiinflammatory activity and reducing the toxicity have been carried out by other groups to yield some advanced agents such as flosulide (CGP-28238),[3] NS-398 (2-cyclohexyloxy analogue of nimesulide),[4] and FK-3311[5] (Chart 1). These new sulfoanilides which proved more potent and yet less ulcerogenic than nimesulide were subsequently demonstrated to display selective inhibition of the inducible cyclooxygenase (COX-2) upregulated at inflammatory sites without affecting the constitutive COX-1 which is associated with homeostatic prostanoid synthesis. Unexpectedly, however, subsequent testing with NS-398 and flosulide has revealed that the classical NSAID-like ulcerogenic liability is still an issue and these two are not being developed further.[6]

In our study aimed at developing an advanced sulfoanilide antiinflammatory agent, we designed the methanesulfoanilides having chromone and 2,3-dihydrochromone rings, particularly compound **1** or **2** (Chart 1) as a lead structure in which the amide nitrogen is located at the *para* position with respect to the carbonyl group as seen in the structures of flosulide and FK-3311. Conventional assays for antiinflammatory activity and ulcerogenicity liability in animal models proved that the chromone compound **1** deserves to receive structural modifications (Table 1). Accordingly, derivatives of **1** at the phenoxy ring as well as at the pyrone ring were synthesized and their structure–activity relationship studied in terms of inhibition of rat carrageenin-induced paw edema and rat adjuvant-induced arthritis, in addition to ulcerogenic liability. The best pharmacological profile has been obtained with *N*-(3-formylamino-4-oxo-6-phenoxy-4*H*-7-chromenyl)-methanesulfonamide, which was designated as T-614.[7] This paper describes the synthesis of a series of derivatives of **1** and the details of their structure–activity relationship leading to our assignment of T-614 for further investigation as an antiarthritis agent. In addition, a preparative-scale synthetic route to T-614 has been investigated.

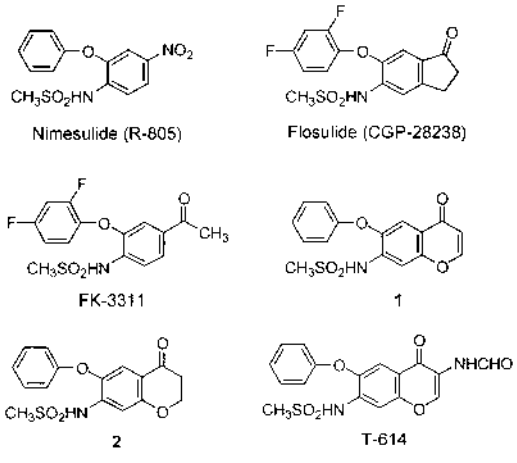

Chart 1

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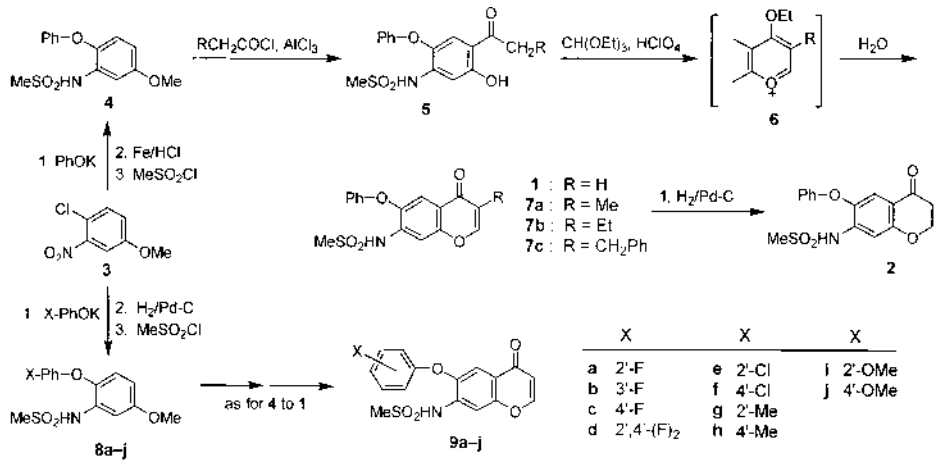

Chart 2

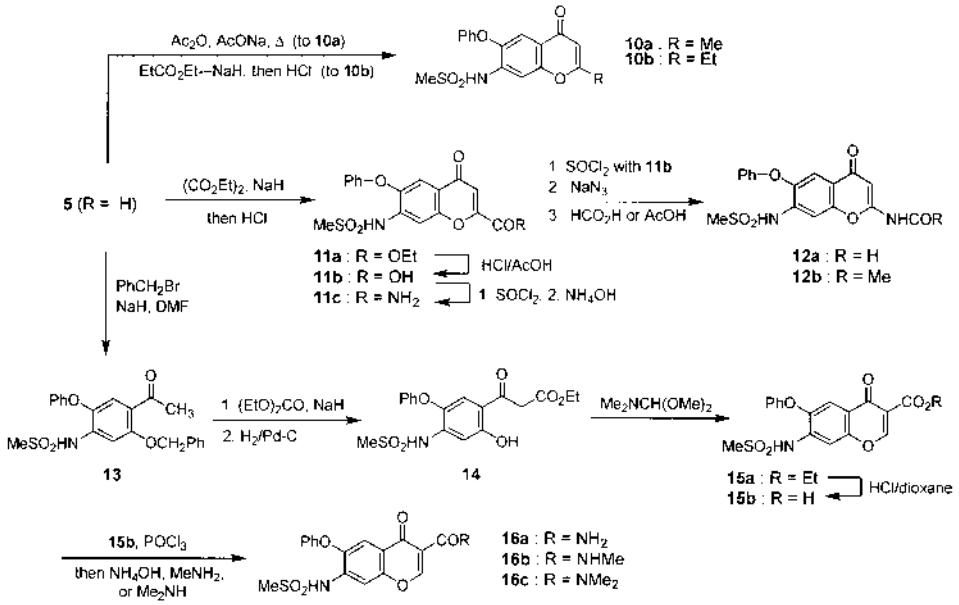

Chart 3

Chemistry

N-(4-Oxo-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (**1**) and its 2,3-dihydro compound (**2**) were synthesized starting with commercially available 4-chloro-3-nitroanisole (**3**) (Chart 2). Reaction of **3** with phenol in the presence of *tert*-BuOK in hot *N*,*N*-dimethylformamide (DMF) afforded 3-nitro-4-phenoxyanisole,[8] which was converted to the sulfonamide **4** by reduction of the nitro group with iron powder followed by *N*-sulfonylation with methanesulfonyl chloride in pyridine (an overall yield of 50%). Friedel–Crafts acylation of **4** with acetyl chloride in dichloromethane using 2 equivalents of $AlCl_3$ afforded 2′-hydroxyacetophenone compound (**5**, R=H) in 84% yield. This product was then subjected to the pyrone-ring annulation with triethyl orthoformate in the presence of perchloric acid to yield a benzopyrylium salt **6**,[9] which on treatment with hot water provided **1** in 85% yield. Catalytic hydrogenation of **1** in the presence of Pd–C in acetic acid afforded **2**. 3-Alkyl derivatives of **1** (**7a—c**) were obtained from **4** by using appropriate acyl chlorides (RCH_2COCl) in the Friedel–Crafts acylation step. The chromones **9a—j** having substituted phenoxy group at the 6-position were similarly prepared *via* intermediacy of **8a—j** obtained by the displacement reaction of **3** with desired phenols before manipulation of the nitro group.

A group of derivatives of **1** at the pyrone-ring other than **7a—c** were prepared from 2-hydroxy-4-methanesulfonylamino-5-phenoxyacetophenone (**5**, R=H) as illustrated in Chart 3. The 2-methyl derivative **10a** was obtained by heating **5** (R=H) with acetic anhydride in the presence of sodium acetate (Kostanecki–Robinson reaction),[10] whereas 2-ethyl compound (**10b**) was most conveniently prepared by a Claisen ester condensation with ethyl propionate.[11] The 2-carboxylic ester **11a** was obtained by sodium hydride-mediated condensation of **5** (R=H) with diethyl oxalate.[12] The ester was hydrolyzed under acid conditions to give carboxylic acid **11b**, which was converted to the carboxamide **11c** *via* a chloride and to 2-formylamino and 2-acetoamino chromones (**12a**, **b**) using the Curtius reaction. For the preparation of 3-carboxylic ester **15a** and carboxamides **16a—c**,

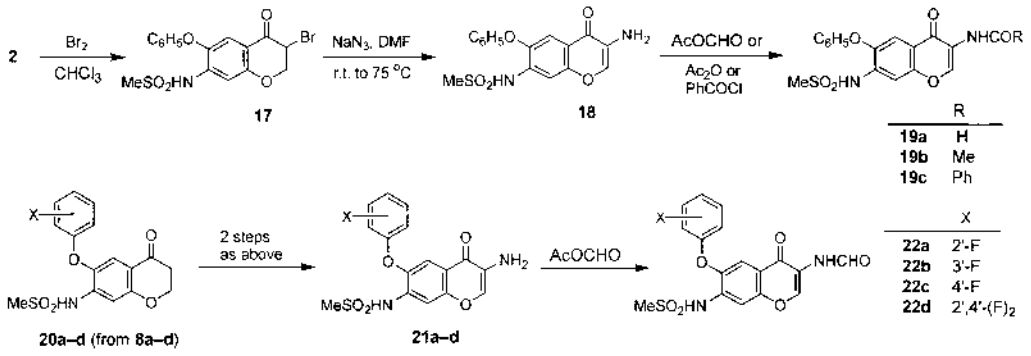

Chart 4

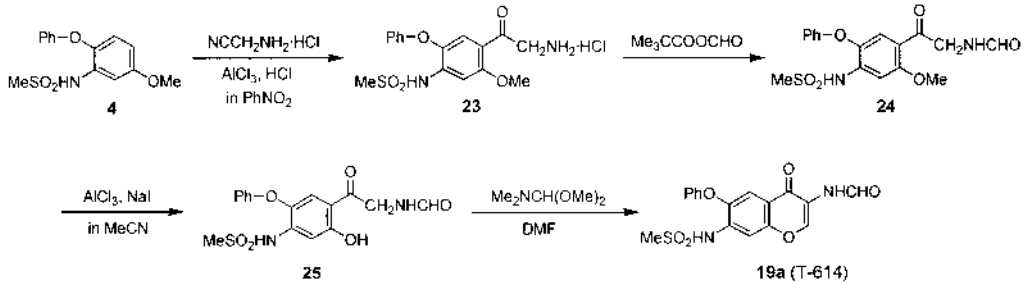

Chart 5

O-benzylated compound **13** was used. The 2-hydroxybenzoylacetate derivative **14** derived from **13** in two steps, which involve NaH-mediated ethoxycarbonylation with diethyl carbonate and subsequent catalytic hydrogenolysis, was subjected to pyrone-ring annulation with $Me_2NCH(OMe)_2$[13] to afford **15a**. It was then converted to **16a**—**c** using a standard 3-step reaction: alkaline ester-hydrolysis to **15b**, chlorination with thionyl chloride, and amidation reaction with appropriate amines.

Chart 4 shows the synthetic route of 3-acylamino compounds (**19a**—**c**, **22a**—**d**) which utilizes a novel amination method originally reported by Szabo and Nemeth.[14a] Typically, treatment of the 2,3-dihydrochromone **2** with bromine in chloroform afforded α-bromo ketone **17** in 97% yield, which was allowed to react with sodium azide in DMF at 70—75 °C for 1 h to give a 3-amino-chromone **18** in 80% yield. *N*-Acylation leading to **19a—c** was carried out by conventional acylation procedures. The 3-amino precursors **21a**—**d** for the syntheses of **22a**—**d** (fluorine-substituted phenoxy analogues of **19a**) were obtained from **8a**—**d** following the same procedure as employed in the preparation of **18**.

After the compound **19a** (T-614) was selected as a candidate for further evaluation of its pharmacological properties, we investigated an alternative, efficient synthetic route in terms of number of steps and overall yield. A four-step synthesis established is illustrated in Chart 5. Thus, the readily available starting material **4** was allowed to react with aminoacetonitrile hydrochloride in nitrobenzene in the presence of 1 equivalent of $AlCl_3$ and excess hydrogen chloride[15] at 25—30 °C to give an aminoacetophenone **23** in 90% yield. This material was treated with formic trimethylacetic anhydride at room temperature affording 3-formylamino compound **24** (91% yield), which was then subjected to *O*-demethylation reaction with aluminum chloride (2 eq) plus sodium iodide (1 eq) in acetonitrile solvent.[16] Reaction of the resulting product **25** with *N*,*N*-dimethylformamide dimethylacetal in DMF provided **19a** in 67% overall yield from **4**.

Table 1. Antiinflammatory Activity and Gastric Toxicity of Compounds **1** and **2**, and Reference Drugs in Oral Administration in Rats

Compound	Carrageenin-induced paw edema[a] ED_{30} (mg/kg)	Adjuvant-induced arthritis[b] ED_{40} (mg/kg/d)	Gastric toxicity[c] UD_{50} (mg/kg)
1	4.2	19	>300
2	>100	>30	>300
Indomethacin	2.0	0.80	2.2
Nimesulide	2.5	1.6	170
Flosulide	1.4	0.25	>300
FK-3311	4.4	5.7	NT[d]

a) Male Donryu rats (overnight fast) received subplantar injection of carrageenin (1 mg) to the left hindpaw 1 h after oral administration of a test compound, then 3 h later the edema volume of each rat was measured. *b*) Arthritis in Lewis rats was induced by intradermal injection of heat-killed *M. tuberculosis* (0.6 mg) in mineral oil at the tail base. A test compound was orally administered once daily from day 18 through 24 after the adjuvant injection. Volumes of both hindpaws were measured on day 25. *c*) Male Wistar rats (24-h fast) received oral administration of a test compound. After another 5-h fast without water supply, the stomachs were examined for the presence of mucosal lesions to obtain the UD_{50} values. *d*) Not tested.

Pharmacological Results and Discussion

The chromone **1** and its 2,3-dihydro compound **2** were first evaluated for their antiinflammatory activity in the rat carrageenin-induced paw edema (CPE) and in the rat established adjuvant-induced arthritis (AA). The result obtained in oral administration (Table 1) shows that compound **1** is much more potent than **2** in both the acute and chronic inflammation models (ED_{30}=4.2 mg/kg in CPE; ED_{40}=19 mg/kg/d in AA). When compared the antiinflammatory efficacy of **1**

Table 2. Antiinflammatory Activity and Gastric Toxicity of Chromone Derivatives in Oral Administration in Rats

Compd.	Carrageenin-induced paw edema[a]		Adjuvant-induced arthritis (AA)				Gastric toxicity[d]	
			Established AA test[b]		Non-established AA test[c]			
	% inhibition of paw volume at 10 mg/kg	ED_{30} (mg/kg)	% reduction of paw swelling at 10 mg/kg/d	ED_{40} (mg/kg/d)	% prevention of paw swelling at 0.3 mg/kg/d	at 3 mg/kg/d	UD_{50} (mg/kg)	Lesion index at 300 mg/kg (incidence)
7a	33		9					
7b	25							
7c	10							
9a	46	3.4	39	3.1	7	56	>300	2.9 (1/7)
9b	32	12	25	18				
9c	35	4.8	34	8.3	4	23		
9d	39	3.4	36	2.5	17	70	>300	2.9 (1/7)
9e	34	6.0	37	4.7	9	34		
9f	25	65	17	54				
9g	29		17					
9h	29		15					
9i	16		1					
9j	28		13					
10a	33	5.0	30	6.2	6	27		
10b	14		−3					
11a	22		−7					
11b	18		6					
11c	19		8					
12a	9		−3					
12b	12		−9					
15a	15		6					
15b	21		27					
16a	32	8.3	36	12				
16b	27		7					
16c	16		−5					
19a	40	3.6	33	3.6	44	57	>300	0.7 (0/8)
19b	17	80	9					
19c	14		8					
22a	34	7.2	38	3.2	14	59	>300	6.1 (3/8)
22b	29	4.5	25	10				
22c	39	4.6	25	25				
22d	51	5.3	39	7.1	6	53	>300	2.1 (1/7)
Nimesulide		2.5		1.6	32	59	170	17.6 (6/7)
Flosulide		1.4		0.25	50	68	>300	8.1 (3/7)

a) See footnote *a* of Table 1. *b*) See footnote *b* of Table 1. The reduction data were obtained on day 22. *c*) The prophylactic effects were evaluated by dosing test compounds on days 0 through 21 after adjuvant injection. *d*) See footnote c of Table 1. Lesion index means the sum of (1) average number of lesions, (2) average severity of lesions, and (3) % ulcer-incidence/10.

with those of some known arylsulfonamide agents shown in Table 1, the activity of **1** in preventing CPE is comparable to that of FK-3311, but 2- to 3-fold weaker than nimesulide and flosulide. With regard to therapeutic efficacy on the AA inflammation, the chromone **1** proved to be significantly less active than indomethacin as well as the sulfoanilide reference drugs. We then conducted a comparative study of gastric ulcerogenicity, which is an important adverse effect associated with classical NSAIDs in general. Compound **1** showed no indication of gastric ulceration up to 300 mg/kg in single oral administration in rats, and the UD_{50} value (the dose that causes gastric mucosal lesion in 50% of animals) was evaluated to be greater than 300 mg/kg. This value was of the same level as observed with flosulide but much greater than indomethacin (2.2 mg/kg) and nimesulide (170 mg/kg).

On the basis of the above comparison study, chromone **1** was selected as a lead for performing chemical modification which involves introduction of a variety of substituents at the pyrone ring as well as at the phenoxy group with the aim of enhancing the marginal activity, particularly against the established AA. First, the effect of introducing alkyl group to the pyrone ring was studied. Among the 2- and 3-alkyl derivatives, it was only 2-methyl compound (**10a**) that showed acceptable potency in the AA assay (ED_{40}=6.2 mg/kg/d in once daily oral administration for 7-consecutive days) and also in CPE inhibition (ED_{30}=5.0 mg/kg) as shown in Table 2.

The effect of placing a substituent on the phenoxy ring at C(6) proved quite sensitive to polarity and position of the substituent. Thus, noticeable activity-enhancement against established AA was realized with those compounds having a halogen substituent at the *ortho* position with respect to the phenoxy oxygen as observed with **9a** (2′-fluoro, ED_{40}=3.1 mg/kg), **9d** (2′,4′-difluoro, ED_{40}=2.5 mg/kg), and **9e** (2′-chloro, ED_{40}=4.7 mg/kg). The 4′-fluorophenoxy compound **9c** was about 2.5-fold less potent than **9a** (2′-fluorophenoxy) but roughly equipotent to **10a** in CPE inhibition. Furthermore, the 3′-fluorophenoxy and 4′-chlorophenoxy compounds (**9b**, **9f**) were considerably less active showing ED_{40} values of 18 and 54 mg/kg in the AA assay, respectively, although they were more potent than the methyl and methoxy

derivatives **9g**—**j**.

We next turned to the evaluation of compounds bearing an amide functionality, which is attached at the pyrone ring in the constitutionally isomeric form of (chromon-2 or 3-yl)carboxamide or (2 or 3-acylamino)chromone. Of the carboxamide compounds (**11c**, **16a**—**c**), the primary 3-carboxamide **16a** was most potent but unacceptable in terms of its ED_{40} (AA)=12 mg/kg and ED_{30} (CPE)=8.3 mg/kg. Among the acylamino compounds (**12a**, **b** and **19a**—**c**), on the other hand, we found the 3-formylaminochromone **19a** displaying remarkable potency, ED_{40} (AA)=3.6 mg/kg and ED_{30} (CPE)=3.6 mg/kg, the values being comparable to those obtained with **9a** (2′-fluoro) and **9d** (2′,4′-difluoro).

Encouraged by finding the potential 3-formylamino compound **19a**, we synthesized its mono and difluoro derivatives at the phenoxy ring (**22a**—**d**). In accordance with the structure–activity relationship previously obtained with the corresponding 3(H) samples (**9a**—**d**), compounds **22a** (2′-fluoro/3-formylamino) and **22d** (2′,4′-difluoro/3-formylamino) showed good AA-therapeutic activity but remained at the levels roughly comparable to the fluorine-free molecule **19a**.

We then conducted defining prophylactic anti-AA potency of the eight compounds (**9a**, **c**—**e**; **10a**; **19a**; **22a**, **d**) which were selected for their good CPE-inhibition (ED_{30} values of 3.4—7.2 mg/kg) as well as noticeable therapeutic activity against the established AA (ED_{40} values of 2.5—8.3 mg/kg). The assessment with Lewis rats was carried out by once daily oral administration of test compounds (0.3 and 3.0 mg/kg) for 22-consecutive days beginning on the day of the adjuvant injection. The prophylactic activities (Table 2), which are roughly equipotent to nimesulide and flosulide, were obtained with **9a**, **9d**, **19a**, **22a** and **22d** (suppression ranging from 53 to 70% at 3.0 mg/kg).

At this stage of the selection of the five compounds (Chart 6), they were evaluated for their potential gastric toxicity in fasted rats in terms of the UD_{50} value and the lesion index (Σ average number of lesions+average severity of lesions+% incidence×0.1),[17] which were determined 5 h after oral administration of each compound. The data shown in Table 2 indicate that all of the five compounds display low ulcerogenic liability with the acute UD_{50} values of >300 mg/kg, significantly less toxic than nimesulide (UD_{50}=170 mg/kg), and that the fluorine-free compound **19a** has the best safety profile as seen by its smallest lesion index (0.7 at 300 mg/kg), which was smaller than that of flosulide (8.1).

On the basis of its overall pharmacological profile described above, compound **19a** was selected for further investigations focusing on its antiarthritic potential. Accordingly, we tested the effect of **19a** on collagen-induced arthritis (CIA) in DBA/1J mice using a progression-preventive dosing regimen, in which once daily oral administration of the test compound was initiated on day 21 shortly after the secondary immunization with a mixture of bovine type II collagen and Freund's complete adjuvant (FCA). Compound **19a** effectively reduced progression of paw-swelling, suppression of 38% at 30 mg/kg and 57% at 100 mg/kg after 2 weeks (Table 3). Evidence for the beneficial effect against joint degeneration was obtained by an X-ray radiographic analysis of the joints, which was carried out after termination of the paw-swelling evaluation. The inhibition % data derived from severity scores (Table 3) indicate that compound **19a** (89% at 30 mg/kg) is significantly more potent than nimesulide and flosulide. To understand the remarkable anti-CIA potency of **19a**, we measured serum levels of interleukin-6 (IL-6) in dosing **19a** and nimesulide, since enhanced production of the cytokine in CIA mice has been reported.[18] Concentrations of IL-6 were determined by an enzyme-linked immunosorbent assay (ELISA) using blood samples from animals receiving administration of a test compound from day 0 through day 34 (30 mg/kg/d). In the mice treated with **19a**, there was observed an 88% reduction of IL-6 level, whereas nimesulide gave much smaller reduction of 26%.

In conclusion, chemical modification of compound **1**, which involves introduction of a variety of substituents at the phenoxy and pyrone groups, has provided us with compound **19a** (designated T-614) which demonstrates potent antiarthritic activity in chronic inflammatory disease models. It is a member of the methanesulfoanilide agents as represented

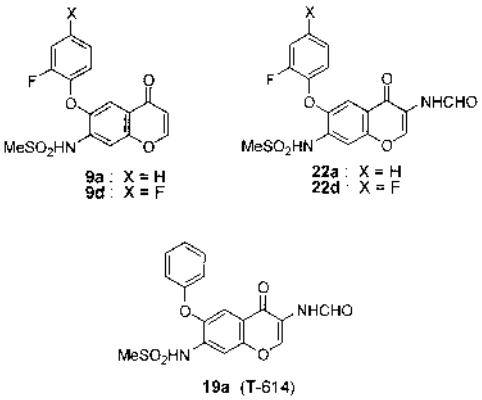

Chart 6

Table 3. Effect on type II Collagen-induced Arthritis in Male DBA/1J Mice

Compound	Dose (mg/kg/d)	Paw-swelling[a]		Joint-degeneration[b]	
		Arthritis score[c]	% inhibition[d]	Severity score[c]	% inhibition[d]
Control	—	6.9±0.7	—	15.5±3.1	—
19a (T-614)	30	4.3±0.6	38	1.6±0.6	89*
	100	3.0±1.2	57*	1.9±1.8	88*
Nimesulide	30	6.0±1.2	13	14.3±3.6	8
Flosulide	30	5.4±1.5	22	12.6±3.9	19

a) Arthritis in mice was induced by injection of a mixture of bovine type II collagen and FCA on days 0 and 21. Test compounds were orally administered once daily starting on day 21 and over the period of 2 weeks. *b*) The paws were removed on day 35 for counting the number of degenerated joints by an X-ray radiographic analysis as described in the experimental section. *c*) The scoring system is described in the experimental section, and the data are expressed as mean±S.E. (n=7). *d*) *, $p<0.05$ as compared with the control by non-parametric Dunnett test.

Table 4. Characterization Data of *N*-(5-Methoxy-2-phenoxyphenyl)methanesulfonamides

Compd.	Formula	mp, °C (Crystallization solvent)	*Anal*. Calcd (Found)		
			C	H	N
8a	$C_{14}H_{14}FNO_4S$	109—110.5 (iso-PrOH)	54.01 (53.98)	4.53 (4.55)	4.50 (4.58)
8b	$C_{14}H_{14}FNO_4S$	58—59.5 (iso-Pr_2O)	54.01 (53.97)	4.53 (4.47)	4.50 (4.60)
8c	$C_{14}H_{14}FNO_4S$	83—84 (iso-Pr_2O)	54.01 (53.96)	4.53 (4.73)	4.50 (4.60)
8d	$C_{14}H_{13}F_2NO_4S$	76—77 (iso-PrOH)	51.06 (51.05)	3.98 (3.98)	4.25 (4.25)
8e	$C_{14}H_{14}ClNO_4S$	108—109 (iso-PrOH)	51.30 (51.04)	4.31 (4.53)	4.27 (4.30)
8f	$C_{14}H_{14}ClNO_4S$	104—106 (iso-Pr_2O)	51.30 (51.18)	4.31 (4.48)	4.27 (4.18)
8g	$C_{15}H_{17}NO_4S$	114—115.5 (iso-PrOH)	58.62 (58.58)	5.58 (5.60)	4.56 (4.47)
8h	$C_{15}H_{17}NO_4S$	115—116 (iso-PrOH)	58.62 (58.57)	5.58 (5.60)	4.56 (4.72)
8i	$C_{15}H_{17}NO_5S$	70—71.5 (iso-Pr_2O)	55.72 (55.68)	5.30 (5.35)	4.33 (4.41)
8j	$C_{15}H_{17}NO_5S$	104—105 (iso-PrOH)	55.72 (55.76)	5.30 (5.32)	4.33 (4.35)

Table 5. Characterization Data of *N*-(4-Oxo-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide Derivatives

Compd.	Formula	mp, °C (Crystallization solvent)	*Anal*. Calcd (Found)		
			C	H	N
7a	$C_{17}H_{15}NO_5S$	164—165 (iso-PrOH)	59.12 (59.00)	4.38 (4.33)	4.06 (3.99)
7b	$C_{18}H_{17}NO_5S$	120—121 (EtOH)	60.16 (59.92)	4.77 (4.96)	3.90 (3.86)
7c	$C_{23}H_{19}NO_5S$	182—183 (EtOH)	65.55 (65.23)	4.54 (4.49)	3.32 (3.20)
9a	$C_{16}H_{12}FNO_5S$	174.5—175.5 (EtOH)	55.01 (54.92)	3.46 (3.51)	4.02 (3.96)
9b	$C_{16}H_{12}FNO_5S$	212—213 (MeCN)	55.01 (55.03)	3.46 (3.44)	4.01 (4.11)
9c	$C_{16}H_{12}FNO_5S$	211—213 (EtOH)	55.01 (55.03)	3.46 (3.45)	4.01 (4.11)
9d	$C_{16}H_{11}F_2NO_5S$	182—183 (AcOEt)	52.32 (52.39)	3.02 (2.96)	3.81 (3.85)
9e	$C_{16}H_{12}ClNO_5S$	151—152 (EtOH)	52.54 (52.11)	3.31 (3.40)	3.83 (3.98)
9f	$C_{16}H_{12}ClNO_5S$	185—186 (MeCN)	52.54 (52.68)	3.31 (3.21)	3.83 (3.66)
9g	$C_{17}H_{15}NO_5S$	140—141 (EtOH–iso-Pr_2O)	59.12 (59.13)	4.38 (4.38)	4.06 (4.10)
9h	$C_{17}H_{15}NO_5S$	151—152 (EtOH–H_2O)	59.12 (59.11)	4.38 (4.46)	4.06 (3.95)
9i	$C_{17}H_{15}NO_6S$	122.5—123.5 (EtOH–iso-Pr_2O)	56.50 (56.53)	4.18 (4.18)	3.88 (3.90)
9j	$C_{17}H_{15}NO_6S$	122—124 (EtOH)	56.50 (56.45)	4.18 (4.24)	3.88 (3.90)
19a	$C_{17}H_{14}N_2O_6S$	236—238 (MeCN)	54.54 (54.35)	3.77 (3.68)	7.48 (7.52)
19b	$C_{18}H_{16}N_2O_6S$	254—256 (MeCN)	55.66 (55.81)	4.15 (4.09)	7.21 (7.22)
19c	$C_{23}H_{18}N_2O_6S$	243—245 (MeCN)	61.33 (61.20)	4.03 (3.84)	6.22 (6.36)
22a	$C_{17}H_{13}FN_2O_6S$	256—257 (MeCN)	52.04 (51.88)	3.34 (3.18)	7.14 (7.15)
22b	$C_{17}H_{13}FN_2O_6S$	232—233 (MeCN)	52.04 (52.26)	3.34 (3.55)	7.14 (7.44)
22c	$C_{17}H_{13}FN_2O_6S$	237—238 (MeCN)	52.04 (52.19)	3.34 (3.35)	7.14 (7.16)
22d	$C_{17}H_{12}F_2N_2O_6S$	240—241 (MeCN)	49.76 (49.67)	2.95 (3.03)	6.83 (6.99)

by nimesulide and flosulide (Chart 1) but has structural features of a chromone core-framework and two chemically different amide groups, formamide and methanesulfonamide functionalities at the remote 3- and 7-positions, respectively. Important pharmacological profiles of T-614 reported in this paper are (1) its low gastro-ulcerogenic liability in oral administration[19,20] and (2) its notable activity against progressive joint destruction in mice CIA models, the potency being significantly greater than nimesulide and flosulide. Clinical evaluations of T-614 as a slow-acting (disease-modifying) antirheumatic agent[21] are in progress.

Experimental

Chemistry Melting points were determined using a Büchi 535 melting point apparatus and are uncorrected. Combustion elemental analysis (C, H, N) was carried out at the analytical department of Toyama Chemical Company, Ltd., using a Yanako MT-3 instrument. ^{1}H-NMR spectra were recorded on a JEOL FX 60 or LA 400 spectrometer with TMS as an internal standard. Infrared spectra were obtained with a Hitachi 260-30 spectrometer. Column chromatography was performed using Merck Silica gel 60 (70—230 mesh).

***N*-(5-Methoxy-2-phenoxyphenyl)methanesulfonamide (4) and Its Substituted 2-Phenoxy Analogues (8a—j)** Potassium *tert*-butoxide (13.5 g, 0.12 mol) was added to a stirred solution of phenol (11.3 g, 0.12 mol) and 4-chloro-3-nitroanisole (**3**) (18.8 g, 0.10 mol) in dry DMF (100 ml), and the mixture was heated at 110 °C for 4 h before pouring into ice-water. The whole was extracted with AcOEt, and the organic phase was sequentially washed with 2 N HCl and water, dried on $MgSO_4$, and concentrated. The residue was purified by silica gel chromatography (hexane–toluene=3 : 1) to give 3-nitro-4-phenoxyanisole (20.6 g, 84%), mp 37.5—38.5 °C after recrystallization from iso-Pr_2O–hexane. *Anal*. Calcd for $C_{13}H_{11}NO_4$: C, 63.67; H, 4.52; N, 5.71. Found: C, 63.81; H, 4.50; N, 5.51. The nitro compound obtained here (20.0 g, 79 mmol) was dissolved in a mixture of 50% EtOH (200 ml) and 4 N HCl (5 ml), and to the solution was added iron powder (13.7 g) portionwise over 20 min at 65—70 °C. After continued stirring at the same temperature for 30 min, the reaction mixture was filtered and the filtrate was diluted with water to precipitate the reduction product. It was purified by recrystallization from toluene to afford 3-amino-4-phenoxyanisole (12.6 g, 72%), mp 112—112.5 °C. *Anal*. Calcd for $C_{13}H_{13}NO_2$: C, 72.54; H, 6.09; N, 6.51. Found: C, 72.59; H, 5.99; N, 6.45.

The amino compound (10.0 g, 46 mmol) was dissolved in dry pyridine (50 ml), and the solution was cooled to 0—5 °C and treated with $MeSO_2Cl$ (5.6 g, 49 mmol). After being stirred at room temperature for 1 h, the reaction mixture was poured into water, and the whole was extracted with AcOEt. The organic phase was successively washed with 2 N HCl and water, dried on $MgSO_4$, and concentrated under reduced pressure. The residue was crystallized from EtOH to give **4** (11.1 g, 82%), mp 109.5—111 °C. *Anal.* Calcd for $C_{14}H_{15}NO_4S$: C, 57.32; H, 5.15; N, 4.78. Found: C, 57.21; H, 5.04; N, 4.79.

The analogues **8a**—**j** having halo, methyl and methoxy substituents at the 2-phenoxy group were also prepared from **3** by subjecting them to the same sequence of reactions, except that reduction of the nitro group was performed by Pd–C catalyzed hydrogenation. Their characterization data are

given in Table 4.

***N*-(4-Oxo-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (1) (General Procedure for the Synthesis of 7a—c and 9a—j)** Aluminum chloride (26.5 g, 0.20 mol) was added to a stirred and cooled (0—5 °C) solution of **4** (29.3 g, 0.10 mol) and acetyl chloride (7.7 g, 98 mmol) in dry CH_2Cl_2 (300 ml). After being stirred at 15—20 °C for 1 h, the mixture was poured onto ice-water and phases were separated. The organic layer was washed with water, dried on $MgSO_4$, and concentrated. The solid residue was crystallized from 2-propanol to give **5** (R=H) (27.0 g, 84%), mp 153—155 °C, IR (KBr) 3240, 1625 cm^{-1}. *Anal*. Calcd for $C_{15}H_{15}NO_5S$: C, 56.07; H, 4.71; N, 4.36. Found: C, 56.07; H, 4.63; N, 4.46.

A stirred suspension of **5** (R=H) (5.0 g, 15.6 mmol) in triethyl orthoformate (50 ml) was treated with 70% $HClO_4$ (4.5 g) at room temperature. After 1 h, crystalline precipitate of a benzopyrylium salt (**6**) (R=H) was filtered and washed with ether. This material was treated with boiling water (50 ml) for 5 min to give **1** (4.4 g, 85%) after recrystallization from MeCN, mp 217—218 °C, IR (KBr) 3240, 1625 cm^{-1}. *Anal*. Calcd for $C_{16}H_{13}NO_5S$: C, 58.00; H, 3.95; N, 4.23. Found: C, 57.94; H, 3.88; N, 4.05.

Compounds **7a**—**c** (3-alkyl derivatives of **1**) were prepared from **4** by the same 2-step procedure in which appropriate α-substituted acetyl chlorides were used in the Friedel–Crafts acylation step. Compounds **9a**—**j** were prepared from **8a**—**j** utilizing the same 2-step procedure as described for the transformation of **4** to **1**. The characterization data for **7a**—**c** and **9a**—**j** are given in Table 5.

***N*-(4-Oxo-6-phenoxy-2,3-dihydro-4*H*-7-chromenyl)methanesulfonamide (2) and Its Phenoxy-Substituted Compounds (20a—d)** A solution of **1** (10.0 g, 30 mmol) in AcOH (200 ml) was stirred at 40—45 °C under an atmospheric pressure of H_2 in the presence of 5% Pd–C and for 1 h. The hydrogenation product **2** was obtained by filtration of the catalyst and evaporation of the solvent, mp 143—144 °C after recrystallization from MeOH. *Anal*. Calcd for $C_{16}H_{15}NO_5S$: C, 57.65; H, 4.54; N; 4.20. Found: C, 57.60; H, 4.43; N, 4.21. Using the same catalytic hydrogenation procedure, the following fluoro derivatives **20a**—**d** were prepared from **8a**—**d**.

N-[4-Oxo-6-(2-fluorophenoxy)-2,3-dihydro-4*H*-7-chromenyl]methanesulfonamide (**20a**): mp 131—132 °C from EtOH (70% yield). *Anal*. Calcd for $C_{16}H_{14}FNO_5S$: C, 54.70; H, 4.02; N, 3.99. Found: C, 54.69; H, 4.02; N, 3.93.

N-[4-Oxo-6-(3-fluorophenoxy)-2,3-dihydro-4*H*-7-chromenyl]methanesulfonamide (**20b**): mp 146—147 °C from EtOH (68% yield). *Anal*. Calcd for $C_{16}H_{14}FNO_5S$: C, 54.70; H, 4.02; N, 3.99. Found: C, 54.88; H, 4.04; N, 4.14.

N-[4-Oxo-6-(4-fluorophenoxy)-2,3-dihydro-4*H*-7-chromenyl]methanesulfonamide (**20c**): mp 167—168 °C from EtOH (74% yield). *Anal*. Calcd for $C_{16}H_{14}FNO_5S$: C, 54.70; H, 4.02; N, 3.99. Found: C, 54.91; H, 4.08; N, 3.86.

N-[4-Oxo-6-(2,4-difluorophenoxy)-2,3-dihydro-4*H*-7-chromenyl]methanesulfonamide (**20d**): mp 163.5—165 °C from EtOH (86% yield). *Anal*. Calcd for $C_{16}H_{13}F_2NO_5S$: C, 52.03; H, 3.55; N, 3.79. Found: C, 51.94; H, 3.51; N, 3.91.

***N*-(4-Oxo-2-methyl-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (10a)** A mixture of **5** (R=H) (3.21 g), Ac_2O (5.45 g), and AcONa (4.1 g) was stirred and heated at 130—140 °C for 1.5 h. The mixture was then cooled to room temperature and AcOEt (200 ml) and water (100 ml) were added. The organic layer was separated, washed with water and brine, dried on $MgSO_4$, and concentrated. The solid residue was crystallized from AcOEt to give **10a** (0.86 g, 25%), mp 186.5—187 °C. *Anal*. Calcd for $C_{17}H_{15}NO_5S$: C, 59.12; H, 4.38; N, 4.06. Found: C, 59.27; H, 4.41; N, 4.11.

***N*-(4-Oxo-2-ethyl-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (10b)** Sodium hydride (60% dispersion in mineral oil, 0.93 g) was added to a stirred suspension of **5** (R=H) (1.5 g, 4.67 mmol) and ethyl propanoate (25 ml), and the mixture was heated under reflux for 2 h before pouring into ice-water. The whole was extracted with AcOEt after adjusting its pH at 5 with 4 N HCl. The organic phase was concentrated, and the residue was dissolved in a mixture of 12 N HCl (2.0 ml) and acetic acid (18 ml) before heating at 70—80 °C for 30 min. The mixture was then cooled to room temperature and extracted with AcOEt. The extract was washed with water, dried, and concentrated. The residual solid was crystallized from 2-propanol to give **10b** (1.3 g, 78%), mp 187.5—188.5 °C. *Anal*. Calcd for $C_{18}H_{17}NO_5S$: C, 60.16; C, 4.77; N, 3.90. Found: C, 60.20; H, 4.70; N, 3.90.

7-[(Methylsulfonyl)amino]-4-oxo-6-phenoxy-4*H*-2-chromenecarboxylic Acid (11b) Sodium hydride (60% dispersion in mineral oil, 3.1 g) was added to a stirred suspension of **5** (R=H) (5.0 g, 15.6 mmol) and diethyl oxalate (4.9 g, 33.5 mmol) in dry EtOH (85 ml), and the mixture was heated under reflux for 1.5 h before pouring into ice-water. The whole was extracted with AcOEt after acidification to pH 2 with 4 N HCl followed by filtration of the crystalline precipitate. This material was dissolved in a mixture of 12 N HCl (1.0 ml) and AcOH (50 ml), and the solution was heated at 80 °C for 10 min before cooling and extraction with AcOEt. The organic phase was washed with water, dried, and concentrated. The residue was crystallized from a mixture of 2-propanol and AcOEt to afford **11a** (ethyl ester of **11b**) (3.5 g, 56%), mp 155—156 °C. *Anal*. Calcd for $C_{19}H_{17}NO_7S$: C, 56.57; H, 4.25; N, 3.47. Found: C, 56.44; H, 4.21; N, 3.51.

A suspension of **11a** (3.5 g, 8.7 mmol) in AcOH (30 ml) and 12 N HCl (20 ml) was heated under reflux for 1 h. The reaction mixture was diluted with water to precipitate the product, which was crystallized from AcOH to give **11b** (3.0 g, 92%), mp 256—258 °C. *Anal*. Calcd for $C_{17}H_{13}NO_7S$: C, 54.40; H, 3.49; N, 3.73. Found: C, 54.44; H, 3.71; N, 3.85.

7-[(Methylsulfonyl)amino]-4-oxo-6-phenoxy-4*H*-2-chromenecarboxamide (11c) Thionyl chloride (3.8 g) and DMF (0.1 ml) was added to a stirred suspension of **11b** (3.0 g, 8.0 mmol) in CH_2Cl_2 (30 ml), and the mixture was heated under reflux for 1.5 h before concentration under reduced pressure. The residual chloride (3.1 g), IR (neat) 1760 cm^{-1}, was poured into an excess amount of ice-cooled 25% NH_4OH solution, and the whole was stirred at room temperature for 30 min. The whole was acidified to pH 2 with 4 N HCl, and extracted with AcOEt. The organic extract was washed with water, dried, and concentrated. The residue was crystallized from MeOH to give **11c** (0.60 g, 20%), mp >280 °C. *Anal*. Calcd for $C_{17}H_{14}N_2O_6S$: C, 54.54; H, 3.77; N, 7.48. Found: C, 54.66; H, 3.68; N, 7.35.

2-Formylamino and 2-Acetoamino Derivatives of 1 (12a and 12b) A solution of the chloride of **11b** (3.1 g, obtained in the above section) in tetrahydrofuran (THF) (80 ml) was added over 10 min to a stirred solution of sodium azide (1.26 g, 19 mmol) in water (10 ml) at 5—10 °C. After being stirred at the same temperature for 1.5 h, the reaction mixture was filtered to obtain the deposited azide product, which was dried over P_2O_5 under reduced pressure, 1.45 g, (46%), IR (KBr) 2115 cm^{-1}. A portion of the azide (1.0 g, 2.5 mmol) in MeCN (10 ml) containing formic acid (0.17 g, 3.7 mmol) was heated under reflux for 1.5 h. The mixture was cooled and filtered to remove insoluble material, and the filtrate was concentrated under reduced pressure. The residue was subjected to silica gel chromatography (toluene–AcOEt=1 : 1), followed by crystallization of the homogeneous product from MeCN to give **12a** (0.27 g, 29%), mp 214—216 °C. *Anal*. Calcd for $C_{17}H_{14}N_2O_6S$: C, 54.54; H, 3.77; N, 7.48. Found: C, 54.70; H, 3.78; N, 7.55.

The *N*-acetyl compound **12b** was obtained by decomposing the azide intermediate in the presence of AcOH (1.5 eq), mp 236—238 °C after recrystallization from EtOH (31% yield). *Anal*. Calcd for $C_{18}H_{16}N_2O_6S$: C, 55.66; H, 4.15; N, 7.21. Found: C, 55.81; H, 3.88; N, 6.95.

Ethyl 3-{2-Hydroxy-4-[(methylsulfonyl)amino]-5-phenoxyphenyl}-3-oxopropanoate (14) *O*-Benzylation of **5** (R=H) with benzyl bromide in the presence of NaH in DMF was carried out according to a standard procedure to give **13** in 50% yield, mp 132—134 °C from toluene. *Anal*. Calcd for $C_{22}H_{21}NO_5S$: C, 64.22; H, 5.14; N, 3.40. Found: C, 63.84; H, 5.16; N, 3.67.

Sodium hydride (60% dispersion in mineral oil, 1.6 g) was added to a stirred solution of **13** (4.11 g, 9.96 mmol) in diethyl carbonate (20 ml) and DMF (20 ml). After being heated at 90—100 °C for 30 min, the reaction mixture was cooled and poured into ice-water. The aqueous phase was separated and washed with ether, then extracted with AcOEt at pH 5 adjusted with 4 N HCl. The organic extract was washed with water, dried on $MgSO_4$, and concentrated. The residue was purified by silica gel chromatography (toluene–AcOEt=3 : 1), followed by crystallization from iso-Pr_2O to give *O*-benzyl ether of **14** (3.2 g, 66%), mp 85—90 °C. *Anal*. Calcd for $C_{25}H_{25}NO_7S$: C, 62.10; H, 5.21; N, 2.90. Found: C, 62.38; H, 5.36; N, 2.69. This material was subjected to 5% Pd–C catalyzed hydrogenolysis in EtOH to afford **14** in 90% yield, mp 111.5—112.5 °C after recrystallization from a mixture of 2-propanol and AcOEt. *Anal*. Calcd for $C_{18}H_{19}NO_7S$: C, 54.95; H, 4.87; N, 3.56. Found: C, 55.12; H,4.85; N, 3.16.

Ethyl 7-[(Methylsulfonyl)amino]-4-oxo-6-phenoxy-4*H*-3-chromenecarboxylate (15a) A solution of **14** (3.9 g, 10 mmol) in DMF (40 ml) was treated with $Me_2NCH(OMe)_2$ (2.6 g, 22 mmol) at room temperature for 1 h before pouring into water. The whole was extracted with AcOEt after acidification with 4 N HCl. The organic phase was washed with water, dried, and concentrated under reduced pressure. The residue was crystallized from EtOH to give **15a** (3.6 g, 89%), mp 167—168 °C. *Anal*. Calcd for $C_{19}H_{17}NO_7S$: C, 56.57; H, 4.25; N, 3.47. Found: C, 56.54; H, 4.49; N, 3.20. A mixture of the ester **15a** (2.0 g, 5 mmol), dioxane (40 ml) and 6 N HCl (20 ml) was heated under reflux for 30 min before dilution with water. Crystalline precipitate was filtered and recrystallized from AcOH to give **15b** (1.7 g, 91%), mp >280 °C. *Anal*. Calcd for $C_{17}H_{13}NO_7S$: C, 54.40; H, 3.49; N, 3.73. Found: C, 54.34; H, 3.35; N, 3.67.

Amides (16a—c) of 15b Phosphorus oxychloride (4.6 g, 30 mmol) was added dropwise to a stirred and cooled (between −5 and −10 °C) suspen-

sion of **15b** (3.8 g, 10 mmol) in DMF (75 ml). After continued stirring at the same temperature for 3 h, the mixture was added dropwise to 25% NH_4OH at 10—20 °C. After 30 min, the whole was acidified to pH 4 with 4 N HCl to precipitate crude **16a**, which was recrystallized from AcOH, mp 251—253 °C (2.8 g, 75%). *Anal*. Calcd for $C_{17}H_{14}N_2O_6S$: C, 54.54; H, 3.77; N, 7.48. Found: C, 54.32; H, 3.77; N, 7.70. Compounds **16b** and **16c** were obtained using the same amidation procedure.

16b, mp 255—256 °C after recrystallization from EtOH. *Anal*. Calcd for $C_{18}H_{16}N_2O_6S$: C, 55.66; H, 4.15; N, 7.21. Found: C, 55.86; H, 4.37; N, 7.25.

16c, mp 213—215 °C after recrystallization from AcOEt. *Anal*. Calcd for $C_{19}H_{18}N_2O_6S$: C, 56.71; H, 4.51; N, 6.96. Found: C, 56.73; H, 4.51; N, 6.88.

***N*-(3-Amino-4-oxo-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (18) and Fluorinated 6-Phenoxy Analogues (21a—d)** Bromine (16.3 g, 0.10 mol) was added over 30 min to a stirred solution of **2** (33.3 g, 0.10 mol) in $CHCl_3$ (300 ml) which was maintained at 25—30 °C. After continued stirring at the same temperature for 30 min, the reaction mixture was treated with water (100 ml). The organic phase was successively washed with 5% $Na_2S_2O_3$, water and brine, then dried on $MgSO_4$ and concentrated. The residual 3-bromo compound **17** (40.1 g, 97%) showed mp 144—145.5 °C and IR (KBr) 1680 cm^{-1} after recrystallization from toluene. The crude bromide (40.1 g) was dissolved in DMF (280 ml), and NaN_3 (13.9 g, 0.21 mol) was added before heating at 70—75 °C under stirring for 1 h. The reaction mixture was cooled and poured into a mixture of AcOEt (1500 ml) and H_2O (300 ml), then the whole was acidified (pH <1) with HCl. The aqueous layer was separated and, after washing with AcOEt, was made pH 4 with 10% NaOH and extracted with AcOEt. The AcOEt solution was washed with water, dried on $MgSO_4$, and concentrated. The residual solid was recrystallized from EtOH to give **18** (27.7 g, 80%), mp 162—163 °C. ^{1}H-NMR (60 MHz in DMSO-d_6) δ: 3.19 (3H, s), 5.50—7.00 (2H, br), 7.04—7.49 (5H, m), 7.35 (1H, s), 7.62 (1H, s), 7.94 (1H, s). *Anal*. Calcd for $C_{16}H_{14}N_2O_5S$: C, 55.48; H, 4.07; N, 8.09. Found: C, 55.46; H, 4.00; N, 7.91.

The following 3-amino chromones were prepared from **8a**—**d** using the same 3-step sequence of reactions: catalytic hydrogenation; bromination, and azidation (the yields refer to those based on the 2,3-dihydro intermediates **20a**—**d**).

N-[3-Amino-4-oxo-6-(2-fluorophenoxy)-4*H*-7-chromenyl]methanesulfonamide (**21a**): mp 173—174 °C after recrystallization from iso-Pr_2O–AcOEt (75% yield). *Anal*. Calcd for $C_{16}H_{13}FN_2O_5S$: C, 52.75; H, 3.60; N, 7.69. Found: C, 52.95; H, 3.71; N, 7.85.

N-[3-Amino-4-oxo-6-(3-fluorophenoxy)-4*H*-7-chromenyl]methanesulfonamide (**21b**): mp 207—208 °C after recrystallization from EtOH (68% yield). *Anal*. Calcd for $C_{16}H_{13}FN_2O_5S$: C, 52.75; H, 3.60; N, 7.69. Found: C, 52.79; H, 3.56; N, 7.55.

N-[3-Amino-4-oxo-6-(4-fluorophenoxy)-4*H*-7-chromenyl]methanesulfonamide (**21c**): mp 204—206 °C after recrystallization from EtOH (72% yield). *Anal*. Calcd for $C_{16}H_{13}FN_2O_5S$: C, 52.75; H, 3.60; N, 7.69. Found: C, 52.96; H, 3.55; N, 7.91.

N-[3-Amino-4-oxo-6-(2,4-difluorophenoxy)-4*H*-7-chromenyl]methanesulfonamide (**21d**): mp 202—202.5 °C after recrystallization from EtOH (55% yield). *Anal*. Calcd for $C_{16}H_{12}F_2N_2O_5S$: C, 50.26; H, 3.16; N, 7.33. Found: C, 50.13; H, 3.06; N, 7.42.

***N*-(3-Formylamino-4-oxo-6-phenoxy-4*H*-7-chromenyl)methanesulfonamide (19a=T-614)** Acetic formic anhydride, prepared *in situ* by heating a mixture of HCO_2H (27.6 g, 0.60 mol) and Ac_2O (30.6 g, 0.30 mol) at 40—45 °C for 1.5 h, was added to a solution of **18** (34.6 g, 0.10 mol) in CH_2Cl_2 (400 ml), and the solution was stirred at room temperature for 1 h before addition of iso-Pr_2O. The precipitate was collected by filtration and recrystallized from MeCN to give **19a** (27.3 g, 73%), mp 236—238 °C. ^{1}H-NMR (400 MHz in DMSO-d_6) δ: 3.22 (3H, s, CH_3SO_2), 7.15 (2H, dd, J=8.5, 1.0 Hz, H-2′), 7.25 (1H, tt, J=7.5, 1.0 Hz, H-4′), 7.32 (1H, s, H-8), 7.47 (2H, dd, J=8.7, 7.5 Hz, H-3′), 7.70 (1H, s, H-5), 8.32 (1H, d, J=1.5 Hz, CHO), 9.27 (1H, s, H-2), 9.79 (1H, br s, NHCHO), 10.05 (1H, s, NHSO$_2$Me). ^{13}C-NMR (100 MHz in DMSO-d_6) δ: 40.59 (CH_3SO_2), 108.85 (C-5), 111.00 (C-8), 117.61 (C-4a), 119.48 (C-2′), 123.09 (C-3), 124.53 (C-4′), 130.12 (C-3′), 135.27 (C-7), 145.52 (C-2), 145.66 (C-6), 151.38 (C-8a), 155.47 (C-1′), 160.34 (CHO), 169.69 (C-4).

Compounds **22a**—**d** were prepared from the corresponding phenoxy precursors (**21a**—**d**) according to the same *N*-formylation procedure, and *N*-acetyl and *N*-benzoyl derivatives of **18** (**19b**, **c**) were obtained using Ac_2O and C_6H_5COCl, respectively. The characterization data of **19a**—**c** and **22a**—**d** are given in Table 5.

A Preparative-scale Synthesis of 19a (T-614) Aluminum chloride (109 g, 0.82 mol) and then $H_2NCH_2CN \cdot HCl$ (37.9 g, 0.41 mol) were added portionwise to a stirred nitrobenzene (300 ml) at room temperature. After being stirred at 40 °C for 1 h, the mixture was cooled to 10 °C and treated with **4** (100 g, 0.34 mol), followed by saturation of the whole with dry HCl over the period of 10 h at the temperature of 25—30 °C. The reaction mixture was poured into ice-cooled 4 N HCl (500 ml), and the precipitated solid product was filtered, washed successively with AcOEt and 2-propanol, and dried to give the semi-hydrate of *N*-[4-(2-aminoacetyl)-5-methoxy-2-phenoxyphenyl]methanesulfonamide hydrochloride (**23**) (122 g, 90%), which was used for the next step without further purification. An analytical sample was obtained by recrystallization from aqueous acetone, mp 167—169.5 °C (decomp.), IR (KBr) 1675 cm^{-1}. *Anal*. Calcd for $C_{16}H_{19}ClN_2O_5S \cdot 0.5H_2O$: C48.54; H, 5.09; N, 7.09. Found: C, 48.48; H, 5.13; N, 6.95.

Sodium formate (41.2 g, 0.61 mol) was added to a solution of trimethylacetyl chloride (36.6 g, 0.30 mol) in acetone (300 ml), and the mixture was vigorously stirred at room temperature for 5 h before addition of **23** (100 g, 0.25 mol) and continued stirring for 3 h. Water (900 ml) was added to the reaction mixture, and the crystalline precipitate was filtered, washed with water and 2-propanol, and dried to give *N*-[4-(2-formylaminoacetyl)-5-methoxy-2-phenoxyphenyl]methanesulfonamide (**24**) (87 g, 91%), which was used for the next step without purification. An analytical sample was obtained by recrystallization from MeCN, mp 153—154 °C, IR (KBr) 1679, 1655 cm^{-1}. *Anal*. Calcd for $C_{17}H_{18}N_2O_6S$: C, 53.96; H, 4.79; N, 7.40. Found: C, 54.07; H, 4.81; N, 7.48.

Sodium iodide (43.6 g, 0.29 mol) and **24** (100 g, 0.26 mol) were added to a solution of $AlCl_3$ (70.5 g, 0.53 mol) in MeCN (300 ml) which was prepared at a temperature below 20 °C by cooling with ice-water. After being stirred at 20 °C for 3 h, the reaction mixture was poured into 1% aqueous Na_2SO_3 (900 ml), and the pale yellow precipitate was filtered, washed with water and EtOH, and air-dried to obtain *N*-[4-(2-formylaminoacetyl)-5-hydroxy-2-phenoxyphenyl]methanesulfonamide (**25**) (91.5 g, 95%), mp 173—174.5 °C after recrystallization from MeCN, IR (KBr) 1664, 1639 cm^{-1}. *Anal*. Calcd for $C_{16}H_{16}N_2O_6S$: C, 52.74; H, 4.43; N, 7.69. Found: C, 52.74; H, 4.32; N, 7.46.

N,*N*-Dimethylformamide dimethylacetal (40.9 g, 0.34 mol) was added to a stirred solution of **25** (50.0 g, 0.14 mol) in DMF (150 ml) at 10 °C, and the mixture was stirred at 15 °C for 8 h before addition of CH_2Cl_2 (250 ml) and water (500 ml). The insoluble crystalline material was filtered, washed successively with CH_2Cl_2 (150 ml), water (150 ml) and EtOH (150 ml), then dried to give **19a** (T-614) (44.4 g, 87%), mp 236—238 °C after recrystallization from MeCN, which was identical with a sample prepared by *N*-formylation of **18**.

Pharmacological Methods The carrageenin-induced rat paw edema assay was carried out using procedures described by Winter *et al*. and Otterness and Moore.[22] Assessment of therapeutic and prophylactic effects on rat adjuvant arthritis was carried out according to the method of Pearson.[23a] Collagen arthritis was induced in male DBA/1J mice by a modification of the method described by Wooley.[24] Gastro-ulcerogenic study in rats was performed according to the experimental procedures detailed by Ono and co-workers,[25] and the lesion index (L. I.) was calculated by the method originally described by Robert *et al*.[17a] All test samples were prepared as fine suspensions by homogenizing the finely powdered samples in 0.5% carboxymethylcellulose sodium (CMC-Na, Tokyo Kasei) and were administered orally *via* a gavage needle.

Inhibition of Carrageenin-Induced Paw Edema in Rats Male Donryu rats (100—130 g), fasted overnight with free access to water, were given orally *via* a gavage needle either the vehicle (0.5% aqueous CMC-Na) or a test compound (10 mg/kg) suspended in the same solvent (1.0 ml/100 g body weight). One hour later, a 1% solution of Seakem-402 carrageenin (Marine Colloids, Springfield, NJ) in sterile saline (0.1 ml) was injected into the plantar surface of the left hindpaw. Three hours after the irritant injection, the paw volume was measured using a plethysmometer. The increase in paw volume was compared with that in the vehicle control group to calculate inhibition %. The data shown are those obtained on the basis of the average of 6 or 7 rats. The ED_{30} values were determined by a linear regression analysis using the data with three to five doses.

Inhibition of Adjuvant-Induced Arthritis in Rats Adjuvant arthritis was induced in male Lewis rats (7-week-old, 150—170 g) by intradermal injection of a suspension of heat-killed *Mycobacterium tuberculosis* (Difco Laboratories) in mineral oil (0.6 mg/0.1 ml) at the base of the tail. Eighteen days after the adjuvant injection, animals showing typical inflammation in the hindpaws were grouped (n=5 or 6) and received once daily oral administration of a test compound (10 mg/kg) suspended in 0.5% aqueous CMC-Na (1.0 ml/100 g body weight) over a period of 4 d (for % reduction) or 7 d (for ED_{40} values). Twenty-four hours after the last dosing, the volumes of both hindpaws were measured using a plethysmometer to obtain inhibition

% in comparison to vehicle-treated controls. For the assessment of prophylactic effects, once daily oral administration of a test compound (0.3 or 3.0 mg/kg) to rats (5 or 6 animals per group) started shortly after the adjuvant injection and continued over the period of 22 d.

Gastric Ulcerogenicity in Rats Male Wistar rats of 180—200 g (7 or 8 rats per group) were fasted for 24 h with free access to water before receiving oral administration of a test compound or solvent vehicle (CMC-Na). After continued fast for 5 h without water supply, the rats were sacrificed by carbon dioxide inhalation. The stomachs were removed and fixed with 1% formalin for 30 min before opening along the greater curvature and rinsing with tap water. The mucosa was examined for the presence of lesions using a stereoscopic microscope. Severity of lesions was graded according to the following score scales (0 to 4): 0=normal; 1=petechiae; 2=a lesion smaller than 1 mm in length; 3=a lesion between 1 and 3 mm in length; 4=a lesion greater than 3 mm in length. A total severity score for each rat was calculated by multiplying the number of lesions by their severity rating and summing the products. The UD_{50} value, the dose that caused gastric lesions of the scores ≥ 2 in 50% of the animals, was calculated by the method of Litchfield and Wilcoxon.[26] The L. I., an overall assessment of damage, was calculated by the method of Robert and others,[17] which is the sum of (1) average number of lesions, (2) average severity of lesions, and (3) % incidence divided by 10.

Collagen-Induced Arthritis in Mice Male DBA/1J mice (8-week-old, $n=7$ per group; Charles River, Japan) were primed by an intradermal injection at several sites of the hip with 0.2 ml of an emulsion consisting of equal volumes of Freund's complete adjuvant (FCA) (Nacalai Tesque, Kyoto) and bovine type II collagen of 2 mg/ml 0.1 M acetic acid (Funakoshi, Tokyo). After 3 weeks, the mice received a booster of the same amount of the FCA/collagen emulsion (0.2 ml) as given in the primary immunization, followed by once daily oral dosing for 2 weeks. The arthritis scores of 0 to 3 were determined on day 35 according to the following criteria: 0=noninvolved; 1=swelling of one or two toes, or slight swelling of the ankle; 2=swelling of one or two toes accompanied by slight swelling of the ankle, or moderate swelling of the ankle; 3=extensive swelling of the paws. The maximum possible score for an arthritic mouse was 12 (3 points for each paw). For joint-degeneration study, the paws were cut off after bleeding at *ca*. 8 mm above the wrist and ankle joints and subjected to radiographic analysis using an X-ray apparatus (SOFTEX-cmbw, Softex, Tokyo): exposure of the paw samples placed on X-ray films (Fuji FR, 12×16.5 cm) with 36 kV and 10 mA for 8 s at the distance of 64 cm. The films were inspected for the 42 joints per mouse, which are connected with the following bones: forepaw bones of carpus, five metacarpals, and four proximal phalanges (2nd to 5th); hindpaw bones of calcaneus, tarsal, five metacarpals, and four proximal phalanges (2nd to 5th). The counting of affected joints (maximum possible score of 42 per mouse) was performed by two persons not aware of the experimental procedures.

Serum IL-6 assay was performed with the mice receiving a sample-dosing for 34 consecutive days starting on day 0. Blood samples were collected on day 35 *via* interior vena cava under ether-anesthesia and kept at −30 °C in a freezer before use. Concentration of IL-6 was determined using a mouse ELISA kit (BioSource International) and according to the instructions provided by the regent supplier.

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References and Notes

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