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Original paper

Thiophene isosteres: synthesis and pharmacological study of 3-(azol-1-yl)thieno isothiazole-1,1-dioxides

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Summary — Three series of new 3-(azol-1-yl)thieno isothiazole-1,1-dioxides were synthesized and tested for anti-inflammatory and related pharmacological activities as well as for acute toxicity. Acetylsalicylic acid was used as the reference standard.

Résumé — **Isostères du thiophène: synthèse et étude pharmacologique d'(azolyl-1-)-3-thiéno isothiazole-dioxydes-1,1.** *Trois séries de nouveaux (azol-yl-1)-3-thiéno isothiazole-dioxydes-1,1 ont été synthétisées et testées dans le but d'étudier leurs activités anti-inflammatoire et activités pharmacologiques voisins ainsi que la toxicité aiguë. L'acide acétylsalicylique a été utilisé comme référence dans les essais biologiques.*

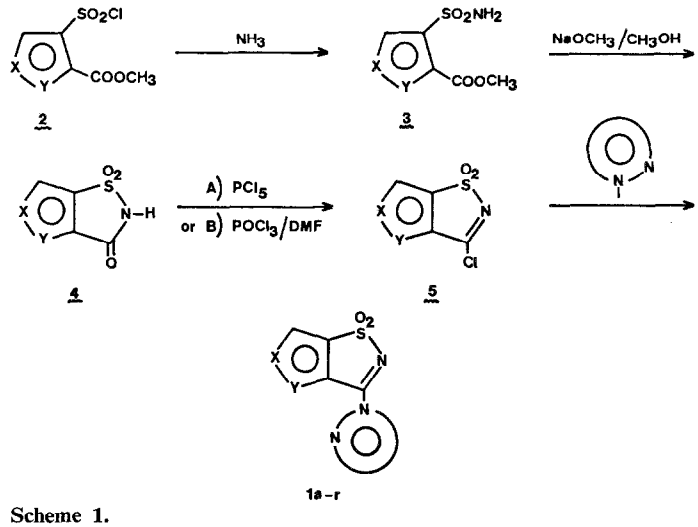
3-(azol-1-yl)thieno[3,4-*d*]isothiazole-1,1-dioxides / 3-(azol-1-yl)thieno[2,3-*d*]isothiazole-1,1-dioxides / analgesic activity / anti-pyretic activity

Introduction

During the last few years, various 3-*N*-derivatives of 1,2-benzisothiazole-1,1-dioxide (saccharine) have been investigated in relation with their activities as diuretic, hypoglycemic [1] and hypotensive [2—4] agents. These observations and our interest in the search for new compounds with promising pharmacological activities led us to synthesize three series of 3-(azol-1-yl)thieno isothiazole-1,1-dioxides **1a—r** which show a clear relationship of bioisosterism with the mentioned benzene compounds and a great similarity with the thieno-1,2-thiazine analogs of 'oxicams', mainly tenoxicam, whose structure and anti-inflammatory activity have been described [5]. Before reporting the anti-hypertensive properties of these compounds, we describe herein their synthesis and the results obtained in the evaluation of their analgesic, anti-pyretic, anti-inflammatory and anti-arthritic activities. Acetylsalicylic acid (ASA) was included in all tests for comparison purposes.

Chemistry

Compounds **1** correspond to the series of the hitherto unknown 3-(azol-1-yl)-thieno[3,4-*d*]isothiazole-1,1-dioxides **1a—f**, 3-(azol-1-yl)-thieno[2,3-*d*]isothiazole-1,1-dioxides **1g—l** and 5-phenyl-3-(azol-1-yl)-thieno[2,3-*d*]isothiazole-1,1-dioxides **1m—r**. Following the route depicted in Scheme 1,



Scheme 1.

they were synthesized from thieno-saccharines **4** by chlorination with phosphorus pentachloride (Method A) or phosphorus oxychloride-*N*,*N*-dimethylformamide (Method B), and subsequent nucleophilic displacement of the chlorine atom of the 3-chloro-thieno isothiazole-1,1-dioxides **5** by pyrazoles, indazole and benzotriazole.

Method A is a modification of the procedure originally employed in the synthesis of 1,2-benzisothiazole-1,1-dioxide derivatives [6, 7]. It consists of heating to the fusion tempera-

ture a mixture of phosphorus pentachloride, the thieno-saccharines **4** and the corresponding azoles. With this method, however, final products, such as **1a** and **1c** result, chlorinated on the azole nucleus. Better results were obtained with Method B which uses formamide chloride as the halogenating agent. This was formed by the addition of catalytic amounts of *N,N*-dimethylformamide to phosphorus oxychloride. Its liquid character allowed the rapid evaporation of the excess of reagent and the easy formation of compounds **1** in shorter reaction times and satisfactory yields.

Intermediate thieno-saccharines **4** were obtained in two steps, as outlined in Scheme 1. Thus, chlorosulfonyl derivatives **2** were treated with ammonia gas in methylene chloride solution to give the corresponding sulfonamides **3** which in turn cyclized to **4** with sodium methoxide.

4-Chlorosulfonyl-3-methoxycarbonyl-thiophene **2** (X = S, Y = CH) was synthesized following a previously published procedure [8]. 3-Chlorosulfonyl-4-methoxycarbonyl-thiophene **2** (X = CH, Y = S) and its 5-phenyl-derivative **2** (X = C—Ph, Y = S) were better prepared by a new method used in our laboratory [9], which involves a Meerwein type reaction of the diazonium salts of the corresponding 3-amino-2-methoxycarbonyl-thiophenes [10—12] with sulfur dioxide and cupric chloride in acetic acid. The 5-phenyl-derivative was obtained in 70% yield as a white crystalline solid which turned out to be somewhat unstable to sunlight; however, this did not prevent its manipulation and analysis.

The structures of the new compounds were confirmed by elemental and spectroscopic analyses. Table I lists the yields and the physical and analytical data of all the products obtained. Their main spectroscopic characteristics are summarized in Table II.

Pharmacological Results and Discussion

All new thieno isothiazole-1,1-dioxides **1a—r** were subjected to a series of tests in order to evaluate their analgesic, anti-pyretic, anti-inflammatory and anti-arthritic activities as well as their ulcerogenic effects and acute toxicity. The results obtained in these assays are given in Table III.

In the analgesic test (phenylquinone-induced writhing method), several compounds tested at dose levels of 10 and 50 mg/kg, were active. Thus, at the dose of 10 mg/kg, compounds **1c**, **1h**, **1i** and **1j** were clearly effective, particularly **1i** which showed an analgesic activity aproximately 6 times higher than that of acetylsalicylic acid (ASA). At 50 mg/kg, compounds **1c**, **1i** and **1j** showed a degree of activity comparable to that of ASA.

As regards the anti-pyretic activity (yeast-induced fever test), the same molecules were shown to possess activities similar to that of ASA. The other compounds were only slightly or not at all effective.

In the carrageenin edema test in the mouse, compound **1c** showed a good anti-inflammatory activity, but slightly lower than that of the reference drug. Similar results were found when this assay was carried out in the rat. Moreover, in adjuvant-induced arthritis (*M. butyricum* test), compounds **1c**, **1h** and **1m** were the most effective, especially the 5-phenyl-derivative **1m** which was as active as ASA.

All these compounds were shown to have interesting values of acute lethal toxicity (LD_{50} = 400 mg/kg, i.p.) and scarce ulcerogenic effects at the assayed dose (250 mg/kg, *p.o.*), at variance with aspirin which, at the same dose, produced 40% ulcerogenic manifestations.

From the above results, some broad generalizations could be drawn. The title compounds presented predominantly analgesic and anti-pyretic activities, though some of them were also as effective as aspirin in the anti-inflammatory and anti-arthritic tests. In general, there is a good correlation between their analgesic and anti-pyretic activities. Of the three series studied, the one containing the unsubstituted thieno[2,3-*d*]isothiazole-1,1-dioxide structure exhibited the best analgesic and anti-pyretic properties; however, the introduction of a phenyl group on the thiophene ring caused the activities to decrease. In each of the three series, the maximal analgesic and anti-pyretic activities corresponded to compounds bearing the 3,5-dimethylpyrazole moiety at position 3. Among the products assayed, **1c** appears to be the most promising compound, with activities similar to those of aspirin in the tests performed.

Experimental protocols

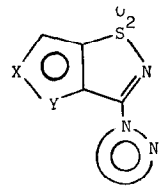
Chemistry

All melting points were taken on a Gallenkamp capillary apparatus and are uncorrected. Infrared spectra were determined with a Perkin—Elmer Model 257 instrument. ^{1}H NMR spectra were obtained with a Varian EM-390 spectrometer, using tetramethylsilane as the internal standard. Chemical shifts are expressed in δ units. Analyses indicated by symbols were within $\pm$0.4% of the theoretical values. The purity of compounds was verified by thin—layer chromatography (TLC) which was run on silica gel GF_{254} (Merck) with cyclohexane—ethyl acetate mixtures (2:1 and 1:1, v/v, respectively) as eluents.

3-Sulfamoyl-2-methoxycarbonyl thiophene **3** *(X = CH, Y = S)*
3-Chlorosulfonyl-2-methoxycarbonyl thiophene [9] (24.0 g, 0.1 mol) was dissolved in methylene chloride (200 ml), the solution was cooled to 0°C, and ammonia gas was introduced during a 1 h period. After an additional 2 h of stirring, the mixture was washed to neutrality with 10% aqueous hydrochloric acid and then with water (3×10 ml), and the organic phase was separated, dried (sodium sulfate) and concentrated. The crude product was recrystallized from water to give 19.4 g (88%), mp: 123—124°C. Anal. $C_6H_7NO_4S_2$ (C, H, N, S). IR (nujol): 3330, 3250 cm^{-1} (NH_2); 1710 cm^{-1} (C=O). ^{1}H NMR ($CDCl_3$): δ 7.65 (s, 2H, thiophene); 5.90 (s, 2H, NH_2 exchangeable); 3.95 (s, 3H, CH_3).

5-Phenyl-3-chlorosulfonyl-2-methoxycarbonyl thiophene **2** *(X = C—Ph, Y = S)*
5-Phenyl-3-amino-2-methoxycarbonyl thiophene [11] (13.9 g, 0.06 mol) was added gradually to a vigorously stirred 35% hydrochloric acid solution (30 ml). The reaction mixture was stirred at room temperature for 30 min. It was then cooled below 0°C (ice—salt bath) and diazotized with sodium nitrite (4.2 g, 0.06 mol) in water (8.4 ml). The resulting diazonium salt was stirred for one more hour at this temperature and then added to a freshly prepared solution obtained by the addition of cupric chloride (3.6 g) in water (3.6 ml) to acetic acid (32 ml) saturated with SO_2. The resulting reaction mixture was poured onto ice—water (250 ml) and the solid so formed was collected, washed with water, dried and crystallized. Yield 13.3 g (70%), mp: 159—160°C (benzene—ethanol). Anal. $C_{12}H_9ClO_4S_2$ (C, H, Cl, S). IR (nujol): 1735 cm^{-1} (C=O); 1385, 1185 cm^{-1} (SO_2). ^{1}H NMR ($CDCl_3$): δ 7.75 (s, 1H, thiophene); 7.65—7.35 (m, 5H, benzene); 4.00 (s, 3H, CH_3).

Table I. 3-(Azol-1-yl)thieno isothiazole-1,1-dioxides **1a—r.**



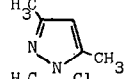
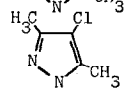
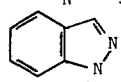
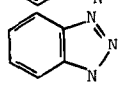
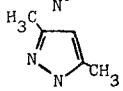
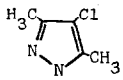
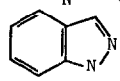
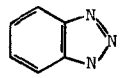
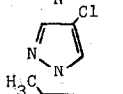
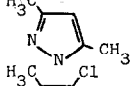
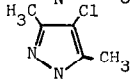
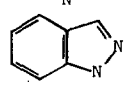
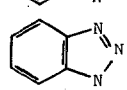
Compound	X	Y		Yield(%) (method)	M.p.(°C) solvent MeCN	Formula (m.w.)
1a	S	C-H		50(A) 60(B)	251-253	$C_8H_5N_3O_2S_2$ (239.27)
1b	S	C-H		73(A)	236-238	$C_8H_4ClN_3O_2S_2$ (273.71)
1c	S	C-H		42(A) 45(B)	228-231	$C_{10}H_9N_3O_2S_2$ (267.32)
1d	S	C-H		24(A)	268-270(d)	$C_{10}H_8ClN_3O_2S_2$ (301.76)
1e	S	C-H		73(A)	312-314	$C_{12}H_7N_3O_2S_2$ (289.32)
1f	S	C-H		35(A)	~255(d)	$C_{11}H_6N_4O_2S_2$ (290.32)
1g	C-H	S		59(A) 65(B)	242-243	$C_8H_5N_3O_2S_2$ (239.27)
1h	C-H	S		65(A)	222-224	$C_8H_4ClN_3O_2S_2$ (273.71)
1i	C-H	S		42(A) 27(B)	207-208	$C_{10}H_9N_3O_2S_2$ (267.32)
1j	C-H	S		42(A)	236-237	$C_{10}H_8ClN_3O_2S_2$ (301.76)
1k	C-H	S		36(A)	314-316	$C_{12}H_7N_3O_2S_2$ (289.32)
1l	C-H	S		44(A)	~260(d)	$C_{11}H_6N_4O_2S_2$ (290.32)
1m	C-Ph	S		42(A) 72(B)	250-252(d)	$C_{14}H_9N_3O_2S_2$ (315.36)
1n	C-Ph	S		75(A)	294-296(d)	$C_{14}H_8ClN_3O_2S_2$ (349.80)
1o	C-Ph	S		30(A) 35(B)	263-265(d)	$C_{16}H_{13}N_3O_2S_2$ (343.41)
1p	C-Ph	S		34(A)	257-259(d)	$C_{16}H_{12}ClN_3O_2S_2$ (377.86)
1q	C-Ph	S		53(A)	293-296(d)	$C_{18}H_{11}N_3O_2S_2$ (365.42)
1r	C-Ph	S		45(A)	~270(d)	$C_{17}H_{10}N_4O_2S_2$ (366.41)

Table II. IR and ^1H NMR data for compounds **1a—r.**

Compound	$\nu_{max.}^{SO_2}$ (cm^{-1})	Solvent	δ(ppm)
1a	1330, 1170	DMSO-d_6	6.80 (q, 1H, pyrazole); 8.20 (d, 1H, pyrazole, J= 1.5 Hz), 8.60 (m, 3H, 2 thiophene + 1 pyrazole).
1b	1340, 1180	DMSO-d_6	8.35 (s, 1H, pyrazole); 8.60 (q, 2H, thiophene, J= 3.0 Hz); 8.95 (s, 1H, pyrazole).
1c	1340, 1175	CF_3COOH	2.45 (s, 3H, CH_3); 2.70 (s, 3H, CH_3); 6.40 (s, 1H, pyrazole); 8.10 (d, 1H, thiophene, J= 2.4 Hz); 8.50 (d, 1H, thiophene, J= 2.4 Hz).
1d	1340, 1175	CF_3COOH	2.40 (s, 3H, CH_3); 2.70 (s, 3H, CH_3); 8.05 (d, 1H, thiophene, J= 2.6 Hz); 8.60 (d, 1H, thiophene, J= 2.6 Hz).
1e	1330, 1175	DMSO-d_6	7.45-7.85 (m, 2H, indazole); 8.05 (m, 1H, indazole); 8.55 (m, 1H, indazole); 8.65 (d, 1H, thiophene; J= 2.4 Hz); 8.75 (d, 1H, thiophene, J= 2.4 Hz); 8.85 (d, 1H, indazole, J= 0.9 Hz).
1f	1330, 1180	CF_3COOH	7.85 (m, 2H, benzotriazole); 8.20 (d, 1H, thiophene, J= 2.5 Hz); 8.35 (d, 1H, benzotriazole, J= 7.5 Hz); 8.60 (d, 1H, benzotriazole, J= 8.4 Hz); 8.80 (d, 1H, thiophene, J= 2.5 Hz).
1g	1330, 1165	CF_3COOH	6.25 (q, 1H, pyrazole); 6.95 (d, 1H, thiophene, J= 5.1 Hz); 7.50 (d, 1H, pyrazole, J= 2.7 Hz); 7.65 (d, 1H, thiophene, J= 5.1 Hz); 7.90 (d, 1H, pyrazole, J= 3 Hz).
1h	1340, 1165	CF_3COOH	7.00 (d, 1H, thiophene, J= 5.1 Hz); 7.40 (s, 1H, pyrazole); 7.55 (d, 1H, thiophene, J= 5.1 Hz); 7.85 (s, 1H, pyrazole).
1i	1335, 1170	CF_3COOH	2.35 (s, 3H, CH_3); 2.70 (s, 3H, CH_3), 6.30 (s, 1H, pyrazole); 7.45 (d, 1H, thiophene, J= 5.1 Hz); 8.00 (d, 1H, thiophene, J= 5.1 Hz).
1j	1340, 1170	CF_3COOH	1.90 (s, 3H, CH_3); 2.20(s, 3H, CH_3); 7.00 (d, 1H, thiophene, J= 5.1 Hz); 7.55 (d, 1H, thiophene, J= 5.1 Hz).
1k	1330, 1170	DMSO-d_6	7.65 (m, 2H, indazole); 7.80 (d, 1H, thiophene, J= 4.8 Hz); 8.10 (m, 1H, indazole); 8.35 (m, 1H, indazole); 8.45 (d, 1H, thiophene, J= 4.8 Hz); 8.90 (d, 1H, indazole, J= 0.75 Hz),
1l	1345, 1170	CF_3COOH	7.60 (d, 1H, thiophene, J= 5.1 Hz); 7.90 (m, 2H, benzotriazole); 8.25 (d, 1H, thiophene, J= 5.1 Hz); 8.35 (d, 1H, benzotriazole, J= 7.8 Hz); 8.65 (d, 1H, benzotriazole, J= 8.4 Hz).
1m	1340, 1165	DMSO-d_6	6.90 (q, 1H, pyrazole); 7.50 (t, 3H, benzene); 7.85 (m, 2H, benzene), 8.25(m, 2H, thiophene + pyrazole); 8.70 (d, 1H, pyrazole, J= 3.0 Hz).
1n	1340, 1170	DMSO-d_6	7.55 (t, 3H, benzene); 7.90 (m, 2H, benzene); 8.30 (s, 1H, thiophene) 8.50 (s, 1H, pyrazole), 9.10 (s, 1H, pyrazole).
1o	1330, 1165	CF_3COOH	2.40 (s, 3H, CH_3); 2.70 (s, 3H, CH_3); 6.30 (s, 1H, pyrazole); 7.45 (t, 3H, benzene); 7.65 (m, 3H, 2 benzene + 1 thiophene).
1p	1340, 1170	CF_3COOH	2.45 (s, 3H, CH_3); 2.75 (s, 3H, CH_3); 7.50 (t, 3H, benzene); 7.65 m, 3H, 2 benzene + 1 thiophene).
1q	1330, 1170	DMSO-d_6	7.40-8.10 (m, 8H, 5 benzene + 3 indazole); 8.25 (s, 1H, thiophene); 8.40 (m, 1H, indazole); 8.90 (d, 1H, indazole, J= 0.6 Hz).
1r	1350, 1180	DMSO-d_6	7.55 (m, 3H, benzene); 7.95 (m, 4H, 2 benzene + 2 benzotriazole); 8.40 (m, 3H, 1 thiophene + 2 benzotriazole).

Table III. Pharmacological activities of **1a—r.**

No.	Activity							Toxicity LD_{50} (mg/kg, i.p.)
	analgesic[a] (mg/kg)		anti-pyretic[b]	anti-inflammatory[c]		anti-arthritic[d] AD_{50} (mg/kg, *p.o.*)	ulcerogenic[e] % ulcer production	
	10	50		mouse	rat			
1a	0	0	0	17	31	250	0	400
1b	0	54	—0.3	0	—	—	10	>500
1c	50	88	—1.8	38	58	125	5	>500
1d	0	61	—0.2	14	20	>250	5	>500
1e	0	47	—0.5	20	30	>250	10	>500
1f	19	22	0	0	—	—	10	>500
1g	13	36	0	18	28	>250	0	>500
1h	35	49	—1.5	27	45	150	0	500
1i	76	90	—2.2	25	33	250	0	>500
1j	42	86	—1.7	5	—	—	5	>500
1k	0	0	0	5	—	—	0	>500
1l	20	61	—0.5	3	—	—	0	>500
1m	24	26	0	29	52	100	5	>500
1n	0	0	0	15	25	>250	5	>500
1o	0	56	—0.6	0	—	—	0	>500
1p	0	0	0	7	—	—	0	>500
1q	20	12	0	3	—	—	0	>500
1r	0	0	0	0	—	—	0	>500
ASA	13	80	—2.0	41	61	100	40	—

[a] % inhibition.
[b] Dose 200 mg/kg, *p.o.* Decrease of rectal temperature (°C) 2 h after dosing.
[c] Dose 200 mg/kg, *p.o.* % reduction of paw swelling in the carrageenin edema assay.
[d] AD_{50} are approximate values. They were obtained after administration of 4 progressively increasing doses. The ± standard errors of the mean measures of each assayed dose did not exceed 25%.
[e] Dose 250 mg/kg, *p.o.*

5-Phenyl-3-sulfamoyl-2-methoxycarbonyl thiophene **3** *(X = C—Ph, Y = S)*
This compound was obtained from **2**, following the procedure employed for **3** (X = CH, Y = S). Yield 81%, mp: 199—201°C (ethanol). Anal. $C_{12}H_{11}NO_4S_2$ (C, H, N, S). IR (nujol): 3370, 3280 cm^{-1} (NH_2); 1720 cm^{-1} (C=O). ^1H NMR (acetone-d_6): δ 7.75 (m, 3H, thiophene and benzene); 7.45 (m, 3H, benzene); 6.70 (s, 2H, NH_2 exchangeable); 3.90 (s, 3H, CH_3).

*2,3-Dihydro-3-oxo-thieno[3,4-*d*]isothiazole-1,1-dioxide* **4** *(X = S, Y = CH)*
This compound was prepared by the method described in [8].

*2,3-Dihydro-3-oxo-thieno[2,3-*d*]isothiazole-1,1-dioxide* **4** *(X = CH, Y = S)*
3-Sulfamoyl-2-methoxycarbonyl thiophene (5.9 g, 0.036 mol) in methanol (30 ml) was added to a 2 M methanolic solution of sodium methoxide (18 ml) and the mixture was refluxed for 24 h. It was cooled to room temperature and acidified with concentrated hydrochloric acid, and the precipitated product was filtered and washed to neutrality with water. By evaporation of the methanol, a second crop of the product was collected. The product was dried and recrystallized from water to yield 4.3 g (85%), mp: 209—212°C. Anal. $C_5H_3NO_3S_2$ (C, H, N, S). IR (nujol): 1715 cm^{-1} (C=O). ^1H NMR (acetone-d_6): δ 8.30 (d, J = 5 Hz, 1H, thiophene); 8.00 (s, 1H, NH exchangeable); 7.00 (d, J = 5 Hz, 1H, thiophene).

*5-Phenyl-2,3-dihydro-3-oxo-thieno[2,3-*d*]isothiazole-1,1-dioxide* **4** *(X = C—Ph, Y = S)*
This compound was prepared analogously to **4** (X = CH, Y = S) starting from 5-phenyl-3-sulfamoyl-2-methoxycarbonyl thiophene (14.8 g, 0.05 mol). Yield 90%, mp: 255—258°C (ethanol—water). Anal. $C_{11}H_7NO_3S$ (C, H, N, S). IR (KBr): 1720 cm^{-1} (C=O). ^1H NMR (acetone-d_6): δ 7.95 (s, 1H, thiophene); 7.80 (m, 2H, benzene); 7.50 (m, 3H, benzene); 7.00 (s, 1H, NH exchangeable).

3-(Azol-1-yl)thieno isothiazole-1,1-dioxides ***1a—r***

d B. A solution of the thieno-saccaharine **4** (0.1 mol), phosphorus pentachloride (0.11 mol) and the corresponding azole (0.2 mol) was heated at fusion temperature for 1—3 h. The mixture was washed with hot water and then with acetone to give the desired products which were recrystallized from acetonitrile.
Method B. A solution of the thieno-saccharine **4** (0.1 mol), phosphorus oxychloride (100 ml) and *N,N*-dimethylformamide (0.3 ml) was refluxed for 90 min. Evaporation of the excess phosphorus oxychloride gave an oil to which the corresponding azole (0.2 mol) was added. The mixture was heated for 30 min at 130—150°C. Work up as above yielded the final compounds.

Pharmacology

Acute toxicity
Toxicity was tested by administering the test compounds i.p. to male Swiss mice weighing 23—28 g each. The LD_{50} values were calculated according to the method described by Litchfield and Wilcoxon [13].

Analgesic activity
The test devised by Siegmund *et al.* [14], which measures the pain produced by the i.p. injection of a solution of phenylquinone (5 mg/kg), was performed on lots of 10 mice which had been fasted for 16 h. The products, suspended in 50% acacia resin, were administered 1 h before phenylquinone and the number of times that the animal stretched or writhed between 7 and 12 min after the application of the algogenic agent was counted. The control group was given only acacia resin, 1 ml/100 g of body weight.

Anti-pyretic activity
For this test, male Wistar rats weighing 170—210 g were used (4 lots of 6 animals each). Hyperthermia was produced by a slightly modified

version of Smith's and Adams' methods [15, 16], involving the s.c. injections of a 15% suspension of brewer's yeast (1.5 g/kg) 18 h before the products were administered. During this time, the animals were given no food. Their rectal temperature was taken immediately and 2, 4, 6 and 8 h after the *p.o.* administration of the products suspended in acacia resin. The results were obtained according to the method of Brownlee [17], as described by Berger and Bates [18].

Anti-inflammatory activity
The method of Winter *et al.* [19] was applied to lots of 10 female Wistar rats weighing 180—220 g which had been fasted for 16 h. 60 min after the products had been administered orally, inflammation was induced by the subplantar injection of a 1% suspension of carrageenin. The volumetric measurement was taken on a plethysmograph (Ugo Basile) immediately after the injection of the irritant and 2 h later.

Anti-arthritic activity
A variation of the technique reported by Winter and Nuss [20] was used. Polyarthritis was induced in rats by a 0.1 ml injection of a 0.6% suspension of *Mycobacterium butyricum* in liquid paraffin into the right foot pads. Lots of 10 male Wistar rats weighing 150—180 g were used. Treatment with the products began the same day and lasted for 20 days. Plethysmographic measurements were taken starting on day 8. The AD_{50} values were assessed according to Colot [21].

Ulcerogenic activity
The ulcerogenic activity was studied using a method similar to that of Domenjoz [22]. Lots of 10 male Wistar rats were fasted for 16 h before and during the entire experiment. The dose was divided into 2 portions which were given at 6 h intervals. 18 h after the 2nd dose, the animals were sacrificed, their stomachs removed and opened along the greater curvature. The mucosal surface was then examined under the microscope and the number of ulcerations was recorded.

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References

1 Whitehead C. W. & Traverso J. J. (1960) *J. Org. Chem.* 25, 413—416
2 Traverso J. J., Whitehead C. W., Bell J. F., Boaz H. E. & Willard P. W. (1967) *J. Med. Chem.* 10, 840—844
3 Whitehead C. W., Traverso J. J., Bell J. F. & Willard P. W. (1967) *J. Med. Chem.* 10, 844—849
4 Willard P. W., Whitehead C. W. & Traverso J. J. (1967) *J. Med. Chem.* 10, 849—852
5 Binder D., Hromatka O., Geissler F., Schmied K., Noe C. R., Burri K., Pfister R., Strub K. & Zeller P. (1987) *J. Med. Chem.* 30, 678—682
6 Meadow J. E. & Reid E. E. (1943) *J. Am. Chem. Soc.* 65, 457—459
7 Stephen E. & Stephen H. (1957) *J. Chem. Soc.* 490—492
8 Rossy P. A., Hoffmann W. & Müller N. (1980) *J. Org. Chem.* 45, 617—620
9 Corral C., Lissavetzky J. & Valdeolmillos A. M. (1985) *Org. Prep. Proc. Int.* 17, 163—165
10 Huddleston P. R. & Barker J. M. (1979) *Syn. Commun.* 9, 731—733
11 Hartmann H. & Liebscher J. (1984) *Synthesis* 275—276
12 Liebscher J., Neumann B. & Hartmann H. (1983) *J. Prakt. Chem.* 325, 915—918
13 Litchfield J. T. & Wilcoxon F. (1949) *J. Pharmacol. Exp. Ther.* 96, 99
14 Siegmund E., Cadmus T. & Lu G. (1957) *Proc. Soc. Exp. Biol. Med.* 95, 729
15 Smith P. K. & Hamburger W. E. (1935) *J. Pharmacol. Exp. Ther.* 54, 346
16 Adams S. S., Hebborn P. & Nicholson J. S. (1968) *J. Pharm. Pharmacol.* 20, 305
17 Brownlee G. (1939) *Quart. S. Pharm. Pharmacol.* 12, 45
18 Berger F. M. & Bates H. M. (1973) *Pharmacology* 9, 164
19 Winter C. A., Risley E. A. & Nuss G. W. (1962) *Proc. Soc. Exp. Biol. Med.* 111, 544
20 Winter C. A. & Nuss G. W. (1966) *Arthritis Rheum.* 9, 394
21 Colot M. (1972) *in: Notions techniques de Pharmacologie Générale* Masson, Paris
22 Domenjoz R. (1960) *Ann. N. Y. Acad. Sci.* 86, 263