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Synthesis of *N*-substituted 4,6-dioxo-imidazo[3,4-c] thiazoles and their analgesic activity in mice

Bernadette Hugon, Catherine Rubat, Pascal Coudert, Fernand Leal, Joseph Fialip and Jacques Couquelet

Abstract

A new series of *N*-substituted dioxo-imidazo[3,4-c]thiazoles have been prepared and evaluated for their analgesic activity. The structures of these new derivatives were confirmed by IR, ^{1}H NMR and ^{13}C NMR spectra, and by elemental analysis. When administered intraperitoneally to mice all derivatives were devoid of any toxic effect, even at the high dose of 800 mg kg^{-1}. In the phenylbenzoquinone-induced abdominal constriction test in mice, eight of the nine synthesized compounds exhibited significant antinociceptive properties with ED50 values (50% effective dose) ranging from 46.7 to 104.7 mg kg^{-1} intraperitoneally. Further investigation demonstrated that analgesic activity of the most effective derivatives **5e** and **5f** partly involved opioidergic and/or noradrenergic pathways.

Introduction

Despite an ever growing body of knowledge about endogenous nociceptive and antinociceptive systems, clinical treatment of pain today is dominated by two main groups of analgesics: the opioids such as morphine and codeine and the non-steroidal anti-inflammatory drugs including aspirin and ibuprofen. Given the reluctance to use opiates because of their liability towards physical dependence, tolerance, respiratory depression and constipation, and the limitations in efficacy of the peripheral analgesics associated to classical drawbacks i.e. gastrointestinal lesions (Clinch et al 1983), the quest is to develop new potent analgesic agents with the efficacy of morphine without the undesired and use-limiting side effects.

In the literature, a class of compounds represented by arylpiperazine derivatives (Valeri et al 1991; Pick et al 1992) seemed to bring an element of hope concerning these problems. The introduction of (4-aryl-piperazin-1-yl)alkyl moieties on various heterocyclic nuclei such as benzoxazolinones (Erdogan et al 1991; Palaska et al 1993; Gökhan et al 1996), oxazolopyridine (Flouzat et al 1993; Viaud et al 1995), pyridazine (Moreau et al 1996; Rohet et al 1997) and pyrazolotriazine (Mavel et al 1993) led to favourable antinociceptive compounds. On this basis we decided to synthesize new *N*-substituted dioxo-imidazo[3,4-c]thiazoles, which share structural similarities with analgesic thiazolotriazine derivatives of the arylpiperazine class (Issartel et al 1998). We have described their analgesic activity in the phenylbenzoquinone-induced abdominal constriction test in mice and have studied their structure–activity relationships. The most active derivatives were evaluated for their possible interactions with opioidergic and noradrenergic systems.

Groupe de Recherches en Pharmacochimie, Laboratoire de Chimie Thérapeutique, Faculté de Pharmacie, Université d'Auvergne, 63001 Clermont-Ferrand Cedex, France

Bernadette Hugon, Pascal Coudert, Fernand Leal, Jacques Couquelet

Laboratoire de Pharmacologie, Faculté de Pharmacie, Université d'Auvergne, 63001 Clermont-Ferrand Cedex, France

Catherine Rubat, Joseph Fialip

Correspondence: P. Coudert, Laboratoire de Chimie Thérapeutique, Faculté de Pharmacie, 28, place H. Dunant, 63001 Clermont-Ferrand Cedex, France.

Materials and Methods

Chemistry

Melting points were determined on a Reichert apparatus (Isi, Paris, France) and were uncorrected. The infrared (IR) spectra were recorded on a Beckman 4240 spectrophotometer (Beckman Instruments, Gagny, France) in KBr pellets. The proton and carbon nuclear magnetic resonance (^{1}H NMR and ^{13}C NMR) spectra were run on a Brüker AC-400 (400 MHz) spectrometer (Wissembourg, France) with tetramethylsilane as the internal standard. Compound purity was determined by TLC on precoated plates (silica gel $60F_{254}$ Merck, Darmstadt, Germany) and spots were visualized with UV light or iodine. Elemental analyses were performed at the Service Central d'Analyses (Vernaison, France). Starting materials were purchased from Acros (Noisy-le-Grand, France).

Methyl 3-(2-*chloroethylcarbamoyl*)-4-*carboxylate* (**3**)
The starting ester, methyl thiazolidine 4-carboxylate, (**2**) was prepared by the procedure described by Margoum et al (1984). 2-Chloroethyl isocyanate (1.06 g, 10 mmol) was added to a solution of **2** (1.47 g, 10 mmol) in 15 mL anhydrous cyclohexane. The mixture was stirred at room temperature for 2 h and the solvent was evaporated to $\frac{2}{3}$. After cooling the resulting precipitate was filtered off and **3** (2.25 g, 89 %) was used without further purification: mp 62°C; IR υ cm^{-1} (KBr): 1748, 1721 (2 C═O). Due to its instability, derivative **3** could not be purified and no correct ^{1}H NMR spectrum could be obtained.

5-(2-*chloroethyl*)-4,6-*dioxo*-6*a*,7-*dihydro*(2*H*)-*imidazo*[3,4-*c*]*thiazole* (**4**)
Compound **3** (2.53 g, 10 mmol) was dissolved in 20 mL diethyl ether and 2 mL HCl was added. The mixture was stirred at room temperature for 2 h and then evaporated to dryness. The resulting residue was treated with water (20 mL) and the solution was extracted with chloroform (2×20 mL). The organic phase was dried over $MgSO_4$ and evaporated to furnish the desired product **4** as an oil (2.16 g, 98 %): $n_D^{20} = 1.555$; IR υ cm^{-1} (NaCl) 1780, 1720 (2 C═O); ^{1}H NMR ($CDCl_3$) δ 3.05 (m, 1H, SCH), 3.30 (m, 1H, SCH), 3.65 (m, 2H, NCH_2), 3.80 (m, 2H, CH_2Cl), 4.15 (d, 1H, SCHN), 4.35 (dd, 1H, CHCO), 5.00 (d, 1H, SCHN); ^{13}C NMR ($CDCl_3$) δ 32.5 (SCH_2); 40.2 (NCH_2); 40.5 (SCH_2N); 48.8 (CH_2Cl); 64.5 (*C*HCO); 158.7 (NCON); 171.4 (CO).

5-[2-(4-*arylpiperazin*-1-*yl*)*ethyl*]4,6-*dioxo*-6*a*,7-*dihydro*-(2*H*)-*imidazo*[3,4-*c*]*thiazole hydrochlorides* (**5a**–**i**)
To a solution of **4** (3.31 g, 15 mmol) in 60 mL acetonitrile was added the appropriate *N*-arylpiperazine (15 mmol) and sodium carbonate (1.59 g, 30 mmol). The solution was heated at reflux for three days and the solvent was e'vaporated. The desired products **5** were obtained as oils after chromatography on a silica gel column (35–70 μm; eluent, ethyl acetate/hexane 60:40). The *N*-substituted dioxo-imidazothiazoles **5** were dissolved in dry diethyl ether. The salts were prepared by bubbling gaseous hydrochloric acid into cooled solutions of the bases. The resulting precipitates were washed with diethyl ether, collected by filtration and dried.

Behavioural studies

Swiss male mice (18–22 g; Dépré, Saint-Doulchard, France) were used in all experiments. The animals were kept in groups of 10 in a temperature-controlled room with a 12-h light–dark cycle. Food and water were freely available during the experiment. The allocation of animals to the different groups was randomized and the experiments were carried out under blind conditions. The IASP Committee for Research and Ethical Issues Guidelines (Zimmermann 1983) were followed. All compounds were administered intraperitoneally in saline (0.9 % NaCl).

Acute toxicity studies
The compounds were administered intraperitoneally at graded doses (100, 200, 400, 600 and 800 mg kg^{-1}). Animals were kept under observation for eight days to detect any sign of toxicity.

Phenylbenzoquinone-induced abdominal constriction test
A 0.02 % solution (ethanol:water, 5:95) of phenylbenzoquinone (Eastman Kodak, Rochester, NY) maintained at 37°C was intraperitoneally injected to mice 30 min after the intraperitoneal administration of drugs. The number of abdominal constrictions of each animal was counted between the fifth and the fifteenth minute after the injection of the irritant (Siegmund et al 1957; Linee 1972).

Antagonism of drug antinociception by naloxone
The protocol used for the evaluation of the effect of naloxone (Narcon, Du Pnt de Nemours, Paris, France) on drug-induced analgesia was similar to that described for the phenylbenzoquinone test. Naloxone (1 mg kg^{-1}, s.c.) was injected 5 min before intraperitoneal ad-

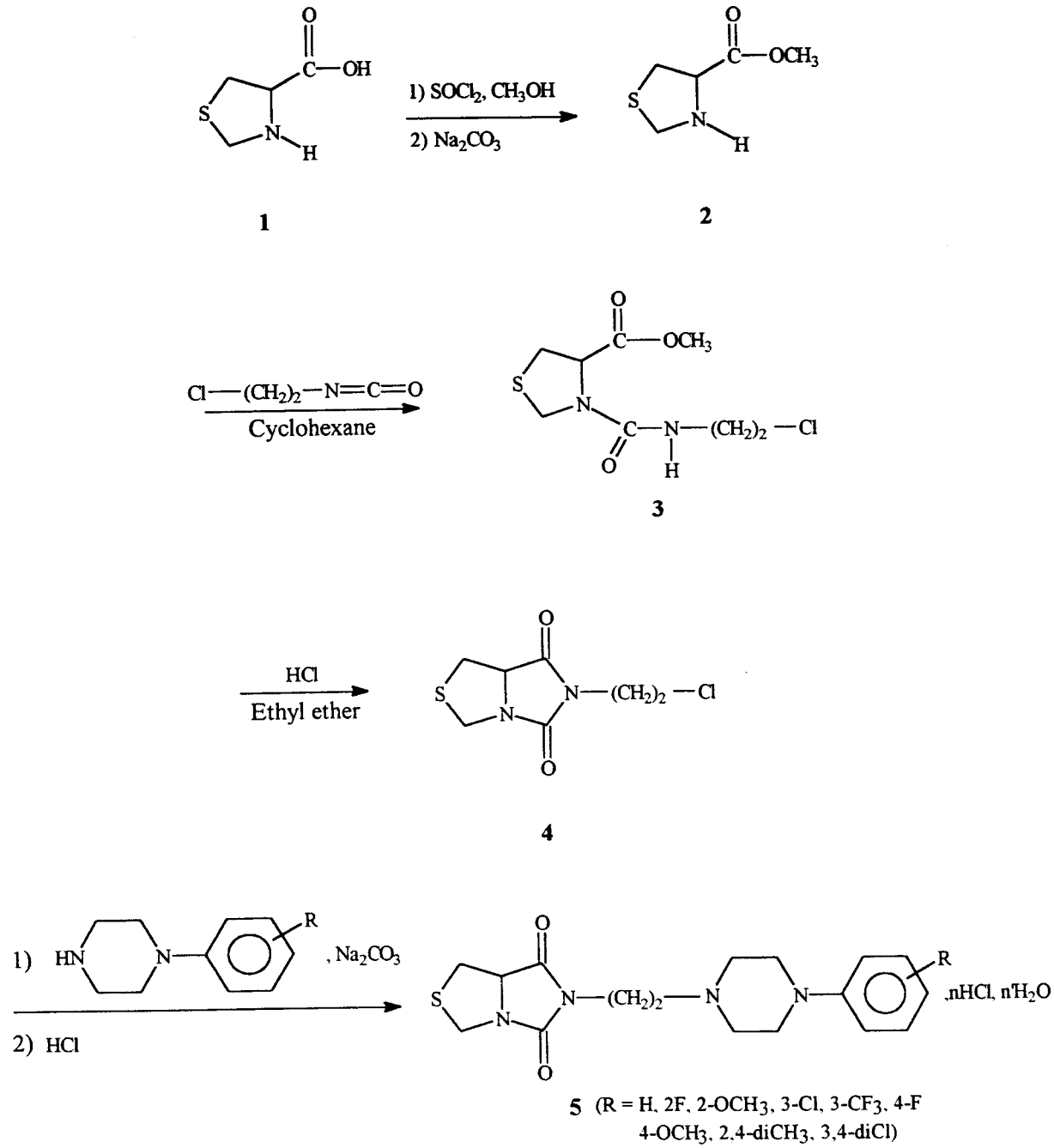


Figure 1 Synthesis of the *N*-substituted dioxo-imidazothiazoles **5**.

ministration of the phenylbenzoquinone solution (Nevinson et al 1991).

Antagonism of drug antinociception by yohimbine

Thirty minutes after simultaneous administration of drugs (i.p.) and yohimbine (1 mg kg^{-1}, i.p.; Sigma, Montluçon, France), phenylbenzoquinone-solution was given intraperitoneally. Five minutes after injection of the irritant, mice were observed for abdominal constriction for a 10-min period (Luttinger et al 1985).

Effects on maximal electroshock seizures

Test drugs were administered to the mice 30 min before the animals were subjected to maximal electroshock

through corneal electrodes. Protection against seizures was defined as the abolition of the hind limb tonic extensor component of seizures (Krall et al 1978).

Data analysis

Statistical analysis of the results was performed using the method of Schwartz (1984). The ED50 (50 % effective dose) values were determined by the method of Carmines et al (1980). The significance of the pharmacological data expressed as mean ± s.e. were analysed by the Student's *t*-test. Data from the maximal electroshock seizure test were analysed by means of the chi-square test with Yate's correction.

Results and Discussion

Preparation of the *N*-substituted dioxo-imidazothiazoles **5** was achieved from L-thiaproline (**1**) according to Figure 1. Ester **2** was prepared by a procedure described by Margoum et al (1984). The reaction of 2-chloroethyl isocyanate with **2** furnished the ureide **3**. Cyclization of the imidazole ring occurred in acidic medium via a mechanism of addition–elimination and led to derivative **4**. Target compounds **5** in basic form resulted from the nucleophilic substitution of the chlorine atom by arylpiperazinyl moieties. Imidazothiazole salts **5a**–**i** were obtained by treatment of the corresponding bases with hydrogen chloride in dry ethyl ether and were isolated as racemates. The structures of **5a**–**i** were supported by elemental analyses (Table 1) and spectral data (Tables 2 and 3).

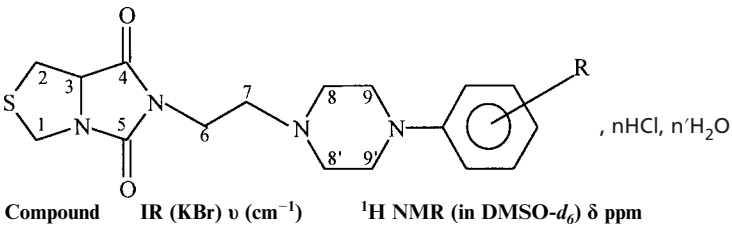
Intraperitoneal acute toxicity was investigated in mice. Test compounds **5** did not display any significant behavioural and neurotoxic effects even at the high dose of 800 mg kg^{-1}. Imidazothiazoles **5** were evaluated for analgesic activity in the phenylbenzoquinone-induced abdominal constriction test in mice and compared with paracetamol, aspirin, morphine, noramidopyrine, tramadol and trazodone. With ED50 values (50 % effective dose) ranging from 46.7 to 104.7 mg kg^{-1} intraperitoneally (Table 4), the derivatives **5**, with the exception of **5g,** were several times more potent than the currently used drug paracetamol (ED50 = 231.3 mg kg^{-1}, i.p.). On the whole, they were also equipotent to noramidopyrine (ED50 = 68.5 mg kg^{-1} i.p.). The effect of substituents on the aromatic ring on the analgesic activity was examined. It appeared that presence of an electron-donating substituent in the para position of the phenyl nucleus (**5g**) abolished analgesic properties while electron withdrawing ones led to active compounds (**5f**, **5i**). Similarly, electron-withdrawing substituents in the meta position (**5d**, **5e**) also produced significant antinociceptive effects. Substitution in the ortho position (**5b**,

Table 1 Physical data for compounds **5a**–**i**.

Compound	R	Yield (%)	mp (°C)	Formula	Elemental analysis[a]					
					C	H	N	S	Cl	F
5a	H	44	150	$C_{17}H_{22}N_4O_2S \cdot 2HCl \cdot H_2O$	46.68	5.95	12.81	7.32	16.25	
					46.82	5.90	12.82	7.46	16.16	
5b	2-F	46	122	$C_{17}H_{21}N_4FO_2S \cdot 2HCl \cdot H_2O$	44.83	5.49	12.31	7.03	15.60	4.18
					44.71	5.34	12.28	7.18	15.66	4.06
5c	2-OCH_3	45	148	$C_{18}H_{24}N_4O_3S \cdot 2HCl \cdot 1.5H_2O$	45.38	6.09	11.76	6.72	14.92	
					45.48	5.97	11.82	6.78	14.79	
5d	3-Cl	44	124	$C_{17}H_{21}N_4ClO_2S \cdot HCl \cdot H_2O$	46.90	5.52	12.87	7.36	16.32	
					47.00	5.48	12.97	7.31	16.27	
5e	3-CF_3	29	154	$C_{18}H_{21}N_4F_3O_2S \cdot 2HCl \cdot H_2O$	42.77	4.95	11.09	6.34	14.06	11.29
					42.87	4.87	11.16	6.52	13.99	11.14
5f	4-F	31	145	$C_{17}H_{21}N_4FO_2S \cdot 2HCl \cdot H_2O$	44.83	5.49	12.31	7.03	15.60	4.18
					44.97	5.53	12.29	6.93	15.56	4.07
5g	4-OCH_3	19	102	$C_{18}H_{24}N_4O_3S \cdot 2HCl \cdot 2H_2O$	44.54	6.19	11.55	6.60	14.64	
					44.52	6.06	11.53	6.53	14.60	
5h	2,4-diCH_3	47	126	$C_{19}H_{26}N_4O_2S \cdot 2HCl \cdot 3H_2O$	45.51	6.79	11.18	6.39	14.17	
					45.45	6.92	11.29	6.22	14.17	
5i	3,4-diCl	48	132	$C_{17}H_{20}N_4Cl_2O_2S \cdot 1.5HCl \cdot H_2O$	41.82	4.82	11.48	6.56	25.47	
					41.85	4.88	11.39	6.58	25.64	

[a]Upper values: calculated; lower ones: found.

Table 2 IR and ^{1}H NMR spectral data for compounds **5a–i**



, nHCl, n′H_2O

Compound	IR (KBr) υ (cm^{-1})		^{1}H NMR (in DMSO-d_6) δ ppm
	NH^+	C=O	
5a	2370	1770–1710	3.30 (m, 2H, H_2), 3.40 (m, 2H, H_7), 3.25–3.85 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 6.90–7.30 (m, 5H, H arom), 8.60 (br s, 2H, H_2O), 11.80 (br s, 2H, 2HCl)
5b	2420	1780–1710	3.30 (m, 2H, H_2), 3.45 (m, 2H, H_7), 3.30–3.70 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.65 (t, 1H, H_3), 4.85 (d, 1H, H_1), 5.30 (br s, 2H, H_2O), 7.20 (m, 4H, H arom), 11.80 (br s, 2H, 2HCl)
5c	2410	1780–1710	3.30 (m, 2H, H_2), 3.45 (m, 2H, H_7), 3.30–3.75 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 3,85 (s, 3H, OCH_3), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 6.10 (br s, 3H, 1.5 H_2O), 6.95–7.05 (m, 4H, H arom), 11.75 (br s, 2H, 2HCl)
5d	2360	1780–1710	3.25 (m, 2H, H_2), 3.40 (m, 2H, H_7), 3.20–3.85 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 6.90–7.30 (m, 4H, H arom), 8.60 (br s, 2H, H_2O), 11.80 (br s, 2H,1H,HCl)
5e	2340	1780–1710	3.25 (m, 2H, H_2), 3.45 (m, 2H, H_7), 3.35–3.90 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 4.00 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 7.15–7.50 (m, 4H, H arom), 8.30 (br s,2H, H_2O), 11.90 (br s, 2H, 2HCl)
5f	2340	1780–1710	3.30 (m, 2H, H_2), 3.40 (m, 2H, H_7), 3.20–3.75 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 7.15 (m, 4H, H arom), 7.35 (br s, 2H, H_2O), 11.80 (br s, 2H, 2HCl)
5g	2380	1780–1710	3.30 (m, 2H, H_2), 3.45 (m, 2H, H_7), 3.30–3.90 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.75 (s, 3H, OCH_3), 3.90 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.65 (t, 1H, H_3), 4.85 (d, 1H, H_1), 6.00 (br s, 4H, 2H_2O), 7.00–7.20 (m, 4H, H arom), 11.80 (br s, 2H, 2HCl)
5h	2530	1780–1710	2.20 (s, 6H, 2CH_3), 3.25 (m, 2H, H_2), 3.45 (m, 2H, H_7), 3.30–3.70 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.85 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.65 (t, 1H, H_3), 4.85 (d, 1H, H_1), 6.15 (br s, 6H, 3H_2O), 7.00 (m, 3H, H arom), 11.70 (br s, 2H, 2HCl)
5i	2340	1780–1710	3.20 (m, 2H, H_2), 3.40 (m, 2H, H_7), 3.40–3.85 (m, 8H, $H_8H_{8'}H_9H_{9'}$), 3.90 (m, 2H, H_6), 4.25 (d, 1H, H_1), 4.60 (t, 1H, H_3), 4.85 (d, 1H, H_1), 5.80 (br s, 2H, H_2O), 7.10–7.50 (m, 3H, H arom), 11.80 (br s, 1.5H, 1.5HCl)

5c) did not offer particular interest in comparison with the parent compound **5a**, except for the disubstituted derivative **5h** where it counterbalanced the baneful influence of a para electron-donating group as it was observed in the f **5g**.

Due to their superior analgesic activity, the pharmacology of **5e** and **5f** was investigated further. The diminished antinociceptive effect of **5e** by a low dose of naloxone provided good evidence that this effect was partly mediated by opioid μ-receptors (Table 5). The analgesic activity of **5f** was not significantly affected by naloxone.

Considering that activation of α_2-adrenoceptors in the central nervous system produces analgesia in a number of animal models (Luttinger et al 1985; Suh et al 1995) we decided to explore possible involvement of the α_2-adrenergic pathway in the analgesic effects of imidazothiazoles **5e** and **5f**. As we observed with the α-sympathomimetic agent clonidine, the activity of **5e** and **5f** was significantly attenuated by pretreatment with the α_2-adrenergic antagonist yohimbine (Table 6), although this antagonism was less marked than with the reference drug. It suggested that antinociceptive response was only partly mediated by an α_2-adrenergic mechanism.

Considering that the arylpiperazinyl moiety seems to contribute to the emergence of anticonvulsant activity (Malawska et al 1997; Malawska & Antkiewicz-Michaluk 1999) and that compounds **5** possess an

Table 3 ^{13}C NMR spectral data for compounds **5**.

Compound	δ ppm (in DMSO-d_6)
5a	30.8 (C_2), 33.2 (C_7), 45.3 ($C_9C_{9'}$), 48.0 (C_1), 50.1 (C_8), 50.6 ($C_{8'}$), 52.3 (C_6), 64.9 (C_3), 116.5, 120.5, 129.4 (C arom), 149.1 (C ipso), 158.3 (C_5), 172.1 (C_4)
5b	30.8 (C_2), 33.2 (C_7), 46.6 ($C_9C_{9'}$), 48.0 (C_1), 50.5 (C_8), 51.0 ($C_{8'}$), 52.4 (C_6), 64.8 (C_3), 116.3, 119.6, 125.0 (C arom), 138.3 (C ipso), 156.1, 153.6 (CF), 158.3 (C_5), 172.0 (C_4)
5c	30.8 (C_2), 33.2 (C_7), 46.7 ($C_9C_{9'}$), 48.0 (C_1), 50.6 (C_8), 51.1 ($C_{8'}$), 52.5 (C_6), 55.5 (OCH_3), 64.9 (C_3), 112.1, 118.6, 120.9, 124.1 (C arom), 138.5 (COMe), 151.9 (C ipso), 158.3 (C_5), 172.1 (C_4)
5d	30.8 (C_2), 33.1 (C_7), 44.5 ($C_9C_{9'}$), 48.0 (C_1), 49.9 (C_8), 50.4 ($C_{8'}$), 52.3 (C_6), 64.9 (C_3), 114.1, 115.3, 119.2, 130.7 (C arom), 134.0 (C Cl), 150.7 (C ipso), 158.3 (C_5), 172.0 (C_4)
5e	30.8 (C_2), 33.2 (C_7), 44.5 ($C_9C_{9'}$) 48.1 (C_1), 49.9 (C_8), 50.4 ($C_{8'}$), 52.3 (C_6), 64.9 (C_3), 111.7, 115.8, 119.3, 130.2 (C arom), 125.7 (CF_3), 129.9 (CCF_3), 149.9 (C ipso), 158.3 (C_5),172.1 (C_4)
5f	30.8 (C_2), 33.2 (C_7), 45.9 ($C_9C_{9'}$), 48.0 (C_1), 50.1 (C_8), 50.7 ($C_{8'}$), 52.3 (C_6), 64.9 (C_3), 115.7, 118.1 (C arom), 146.1 (C ipso), 158.0, 155.6 (CF), 158.3 (C_5), 172.1 (C_4)
5g	30.8 (C_2), 33.3 (C_7), 47.5 ($C_9C_{9'}$), 48.0 (C_1), 49.7 (C_8), 50.3 ($C_{8'}$), 52.3 (C_6), 55.4 (OCH_3), 64.9 (C_3), 114.7, 119.3 (C arom), 141.1 (COMe), 155.4 (C ipso), 158.3 (C_5), 172.1 (C_4)
5h	17.4 (CH_3), 20.4 (CH_3), 30.9 (C_2), 33.3 (C_7), 48.0 ($C_9C_{9'}$), 48.1 (C_1), 51.1 (C_8), 51.6 ($C_{8'}$), 52.5 (C_6), 64.9 (C_3), 118.9, 127.1, 131.7 (C arom), 132.9 (CMe), 147.1 (C ipso), 158.4 (C_5), 172.1 (C_4)
5i	30.8 (C_2), 33.2 (C_7), 44.4 ($C_9C_{9'}$), 48.0 (C_1), 49.7 (C_8), 50.3 ($C_{8'}$), 52.3 (C_6), 64.9 (C_3), 115.8, 117.0, 130.7 (C arom), 131.9 (C Cl), 149.2 (C ipso), 158.3 (C_5), 172.1 (C_4)

Table 4 Results from the phenylbenzoquinone test (analgesic activity).

Compound	ED50 (mg kg^{-1}, i.p.)[b]
5a	76.8 (62.7–94.0)
5b	104.7 (60.9–179.9)
5c	79.6 (60.0–105.7)
5d	72.9 (62.7–84.8)
5e	46.7 (20.6–105.9)
5f	53.7 (27.4–105.3)
5g	Inactive at 100 mg kg^{-1} intraperitoneally
5h	75.7 (52.6–108.9)
5i	77.6 (51.3–117.4)
Paracetamol	231.3 (147.3–363.2)
Aspirin	3.0 (1.5–6.1)
Morphine[a]	0.6 (0.3–1.1)
Noramidopyrine	68.5 (22.8–205.3)
Tramadol	8.6 (4.3–17.3)
Trazodone	10.2 (7.1–14.6)

[a]Subcutaneous route. [b]95% confidence intervals in parentheses.

Table 5 Effect of naloxone (1 mg kg^{-1}, s.c.) on compound **5e**- and **5f**-induced analgesia in the phenylbenzoquinone test.

Compound (i.p. route)	% Analgesia	
	Drug plus saline	Drug plus naloxone
5e (100 mg kg^{-1})	75.1 ± 2.1*	43.2 ± 13.8†
5f (100 mg kg^{-1})	91.8 ± 2.8*	90.6 ± 4.9 (NS)
Morphine (1.5 mg kg^{-1})[a]	77.4 ± 5.2*	15.8 ± 9.5†
Tramadol (15 mg kg^{-1})	85.8 ± 6.1*	45.8 ± 14.1†

[a]Subcutaneous route. *$P < 0.05$ compared with saline. †$P < 0.05$ compared with drug plus saline. NS, not significant.

Table 6 Effect of yohimbine (1 mg kg^{-1}, i.p.) on compound **5e**- and **5f**-induced analgesia in the phenylbenzoquinone test.

Compound (i.p. route)	% Analgesia	
	Drug plus saline	Drug plus yohimbine
5e (100 mg kg^{-1})	75.1 ± 2.1*	51.4 ± 5.3†
5f (100 mg kg^{-1})	91.8 ± 2.8*	74.4 ± 4.8†
Clonidine (0.05 mg kg^{-1})[a]	99.4 ± 1.1*	27.5 ± 16.1†
Tramadol (15 mg kg^{-1})	85.8 ± 6.1*	55.3 ± 12.9†

[a]Subcutaneous route. *$P < 0.05$ compared with saline. †$P < 0.05$ compared with drug plus saline.

hydantoin ring such as phenytoin, we evaluated **5a**–**i** in the maximal electroshock seizure test (Krall et al 1978). However, none of the imidazothiazoles **5** were able to protect mice at the dose of 100 mg kg^{-1} intraperitoneally in this test.

In conclusion, the results suggested that in this series of imidazothiazole derivatives **5e** and **5f** had a superior analgesic profile enhanced by their lack of toxicity at the high dose of 800 mg kg^{-1} intraperitoneally. The mechanism underlying their antinociceptive properties remains unknown to some degree.

In this preliminary pharmacological study, no attempts were made to explore serotonergic pathways.

However, taking into account the presence of arylpiperazinyl moieties, the serotonergic activity of which is well established (Fuller et al 1978; Kahn & Wetzler 1991), in structures **5e** and **5f**, further experiments are needed to ascertain the mechanisms involved in the analgesic effects of **5e** and **5f**.

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