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Short Communication

Synthesis and antinociceptive activity of 6-substituted-3-pyridazinone derivatives

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Abstract

A series of 3-pyridazinones carrying morpholino, arylpiperidino and arylpiperazino moiety in the position 6 **IIa–g** were synthesized and evaluated for antinociceptive activity. In the modified Koster test in mice 4-(4-fluorophenyl) piperazine, **IIf**, was found the most active compound. All the compounds except **IId** were more active than aspirin in the antinociceptive activity test.

Keywords: 6-Substituted-3-pyridazinone derivatives; Antinociceptive activity; Koster test

1. Introduction

Pain is a clinical status that human beings have been coping with for centuries. Non-steroidal analgesic drugs (NSAIDs) on the market have several serious side effects such as gastro-intestinal irritation and kidney toxicity, so copious research projects have been focussing on the development of new and moderately less toxic NSAIDs. Many heterocyclic compounds have been investigated for analgesic activity.

Arylpyridazinones have been the subject of intensive synthetic investigations, because they possess a wide spectrum of pharmacological potencies. Various 3-(2H)-pyridazinone derivatives have been described and their cardiotonic [1,2] antisecretory and antiulcer [3] as well as analgesic and anti-inflammatory [4,5] activities have been investigated. Some 3-(2H)-pyridazinone derivatives endowed with analgesic properties have been reported recently [6]. 4-Ethoxy-2-methyl-5-morpholino-3(2H)pyridazinone (emorfazone, **1**) and its derivatives have emerged as being of particular interest. Derivatives of *N*-substituted-4, 6-diaryl-3-pyridazinones (**2**) have been screened for their analgesic anti-inflammatory and antipyretic activities [7]. A number of compounds showed a good dose-dependent activity in the phenylquinone-induced writhing test in mice orally. In this series introduction of an arylpiperazinomethyl moiety at position 2 of the pyridazinone ring resulted in increased analgesic activity.

A series of 4-phenyl-6-aryl-2- [3-arylpiperazin-1-yl) propyl] pyridazin-3-ones (**3**) related to trazodone (**4**) have been synthesized and evaluated for analgesic activity. In the phenylquinone-induced writhing test, most compounds have been found several times more potent than acetaminophen and noramidopyridine [8].

Additionally, 5-arylidene-pyridazin-3-one (**5**) derivatives carrying an arylpiperazinoalkyl moiety in the position 2 were synthesized and evaluated for analgesic activity [9]. In the phenylquinone induced writhing test, these pyridazinones exhibited potent analgesic activity.

Recently peripherally acting analgesic 3-arylpiperazinyl-5-pyridazines (**6**) have been synthesized [10]. They were also several times more potent than acetaminophen and noramidopyrine. Pyridazines with a fluorine atom at phenyl ring of benzyl moiety were equipotent of the reference drug trazodone (**4**) (Fig. 1).

As part of the continuing study for new analgesics in our laboratory, we initiated a research program based on the synthesis 3-(2H)-pyridazinone derivatives bearing morpholinyl, arylpiperazinyl, arylpiperidinyl sub-

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stituents at position 6, and some of them were found to have analgesic activity.

2. Chemistry

The title compounds 6-substituted-3-(2H)-pyridazinone derivatives were synthesized according to Scheme 1. Compounds **Ia**–**g** have been reported in literature [11,12]. Compounds **IIa**–**g** are synthesized for the first time in this study.

Synthesis of derivatives **I** was carried out by the reaction of arylpiperazine, arylpiperidine, and morpholine with 3,6-dichloropyridazine. Hydrolysis of **I** with glacial acetic acid gave new 6-substituted-3-(2H)-pyridazinone derivatives **II** (Scheme 1). Compounds **II** are listed in Table 1 and their spectral data and elemental analyses (Table 2) elucidated their structures.

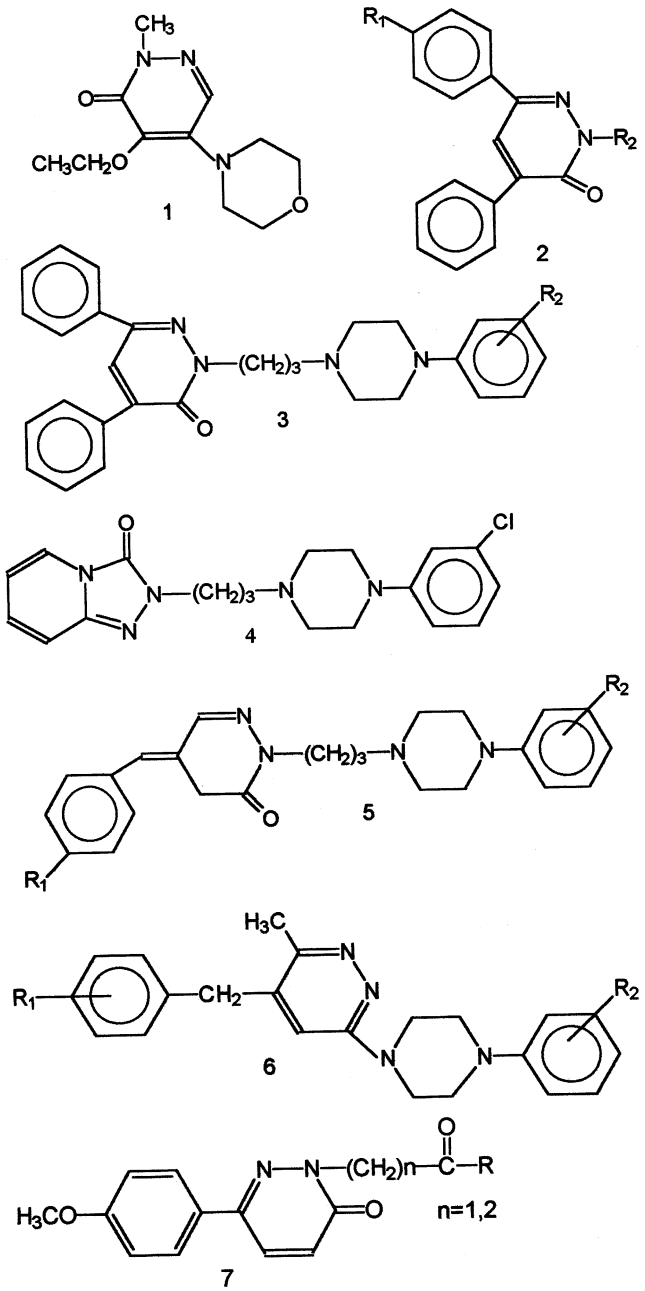
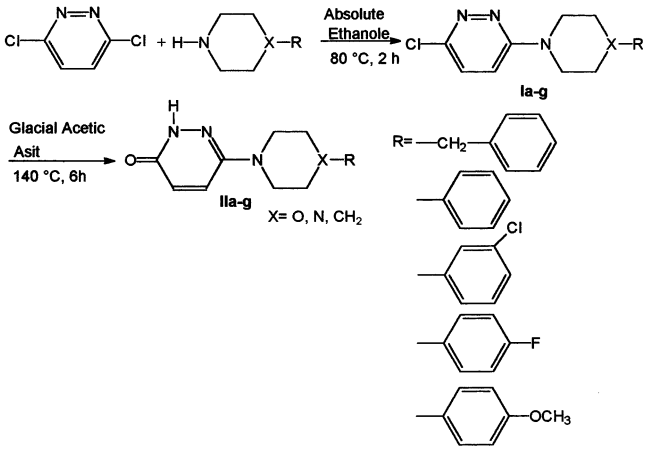


Fig. 1. Analgesic pyridazinone derivatives.



Scheme 1. Synthesis of compounds **II**.

3. Pharmacology

Preliminary pharmacological screening of title compounds for analgesic activity was done with modified Koster test in mice using aspirin as the reference [13]. Analgesic activity of the synthesized compounds is given in Table 3.

4. Experimental

4.1. Chemistry

4.1.1. Materials

The fine chemicals and all the solvents used in this study were purchased locally from Merck A.G., Aldrich.

6-Substituted-3-chloropyridazines were synthesized in our laboratory according to data in the literature [11,12].

4.1.2. Instrumental analysis

Melting points of the compounds were determined on Electrothermal 9200 melting points apparatus and the values given are uncorrected.

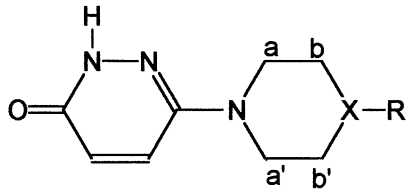
The IR spectra were recorded in KBr on a Perkin Elmer 1330 Spectrophotometer at approximately 2% concentration. The NMR spectra were recorded on Bruker 200 FT-NMR spectrometer in DMSO-d_6 (Merck). Me_4Si is used as an internal standard.

Elemental analysis: Leco 932 C, H, N elemental analyser, Analytical Laboratory of TUBITAK, Ankara, Turkey.

4.1.3. Method

*4.1.3.1. Synthesis of 6-substituted-3-(2H)-pyridazinone derivatives (**IIa–g**).* A solution of 0.05 mol of 6-substituted-3-chloropyridazine in 30 ml glacial acetic acid was refluxed for 6 h. The acetic acid was removed under reduced pressure, the residue dissolved in water and extracted with $CHCl_3$. The organic phase was dried

Table 1
Physical constant of 6-substituted-3-(2H)-pyridazinone derivative (**II**)



Compound no.	X	R	M.p (°C)	Yield (%)	Formula	Anal. (%) Calc. (found)
IIa	O		181–183	67	$C_8H_{11}N_3O_2$	C: 53.03 (52.78) H: 6.12 (6.095) N: 23.19 (22.85)
IIb	CH_2	benzyl	187–188	63	$C_{16}H_{19}N_3O$	C: 71.35 (71.37) H: 7.11 (6.76) N: 15.60 (15.25)
IIc	N	benzyl	113–115	68	$C_{15}H_{18}N_4O$	C: 66.65 (66.49) H: 6.71 (6.45) N: 20.73 (20.28)
IId	N	phenyl	202–206	57	$C_{14}H_{16}N_4O$	C: 65.61 (65.54) H: 6.29 (6.16) N: 21.86 (21.74)
IIe	N	3-chlorophenyl	208–210	64	$C_{14}H_{15}ClN_4O$	C: 57.83 (57.40) H: 5.20 (4.88) N: 19.27 (19.34)
IIf	N	4-fluorophenyl	147–148	60	$C_{14}H_{15}FN_4O$	C: 61.30 (61.18) H: 5.51 (5.55) N: 20.43 (20.19)
IIg	N	3-methoxyphenyl	287–290	59	$C_{15}H_{18}N_4O_2$	C: 62.92 (62.57) H: 6.34 (6.79) N: 19.57 (19.43)

Table 2
Spectral analyses of 6-substituted-3-(2H)-pyridazinone derivatives (**2**) (see structure formula in Table 1)

Comp. No.	R	IR (KBr) cm^{-1}		^1H NMR (DMSO-d_6) (ppm)
		C=O	C=N	
IIa		1690	1580–1490	3.63 (t, 4H, morpholine b+b′), 3.83 (m, 4H, morpholine a+a′), 7.48 (d, 1H, CH=), 7.63 (d, 1H CH=), 12.40 (s, 1H, pyridazinone NH).
IIb	benzyl	1660	1580–1490	1.23 (m, 1H, piperidine H_4), 1.67 (m, 4H, piperidine b+b′), 2.57 (m, 4H, piperidine a+a′), 3.77 (d, 2H, CH_2), 6.72–7.35 (m, 7H, C_6H_5+CH=CH), 12.00 (s, 1H, pyridazinone NH).
IIc	benzyl	1650	1580–1490	2.45 (m, 4H, piperazine b+b′), 3.20 (m, 4H, piperazine a+a′), 3.55 (s, 2H, CH_2), 6.73–7.52 (m, 7H, C_6H_5+CHCH), 12.05 (s, 1H, pyridazinone NH).
IId	phenyl	1660	1590–1495	3.20 (m, 4H, piperazine b+b′), 3.30 (m, 4H, piperazine a+a′), 6.50–7.70 (m, 7H, C_6H_5+CH=CH), 12.00 (s, 1H, pyridazinone NH).
IIe	3-chlorophenyl	1660	1595–1490	3.25 (m, 4H, piperazine b+b′), 3.35 (m, 4H, piperazine a+a′), 6.82–7.61 (m, 6H, C_6H_4+CH=CH), 12.05 (s, 1H, pyridazinone NH).
IIf	4-fluorophenyl	1690	1590–1505	3.15 (m, 4H, piperazine b+b′), 3.35 (m, 4H, piperazine a+a′), 6.72–7.50 (m, 6H, C_6H_4+CH=CH), 12.10 (s, 1H, pyridazinone NH).
IIg	3-methoxyphenyl	1680	1590–1500	3.28 (m, 4H, piperazine b+b′), 3.34 3.35 (m, 4H, piperazine a+a′), 6.80–7.63 (m, 6H, C_6H_4+CH=CH), 12.00 (s, 1H, pyridazinone NH)

Table 3
Antinociceptive activity of 6-substituted-3-pyridazinone derivatives **IIa–g**

Compound	% Antinociceptive activity
Aspirin	40.96
IIa	63.05
IIb	53.00
IIc	67.86
IId	14.25
IIe	71.08
IIf	80.72 *
IIg	46.98
Control	0

* $P = 0.05$.

over Na_2SO_4 and evaporated under reduced pressure. The residue was purified by recrystallization from ethanol.

4.2. Pharmacology

4.2.1. Materials

Locally bred Swiss albino mice of both sexes (255.0 g) were employed. The animals were housed in groups of eight with food and tap water ad libitum and were received to the laboratory at least 2 days before the day of experiment to allow them to get accustomed to the environment. Food was withdrawn one day before the experiment, but they were allowed free access to tap water. The experiments have been performed in full awareness of the test animals. Acetic acid (Merck AG), carboxymethyl cellulose sodium salt (CMC Na) (Aldrich), Aspirin (Bayer), Gauge callipers (Peacock, Ozaki Co., Tokyo) were used.

4.2.2. Method

4.2.2.1. Antinociceptive activity test. Modified Koster's test was employed 14. Koster test has been first used by Koster et al. in mice as 60 mg/kg acetic acid (0.6% solution) given by i.p. injection to produce repeated characteristic stretching movements [13]. This method has been modified later by Safak et al. [14] using acetic acid at the dose level of 300 mg/kg (3% solution). Acetylsalicylic acid (ASA) was used as reference [15].

Each compound was suspended in 0.5% carboxymethyl cellulose to form a solution at the concentration 10 mg/ml and given orally to mice in groups of eight at a dose of 100 mg/kg. One hour after this administration; pain was induced by intraperitoneally injection of 3% solution of acetic acid at 300 mg/kg. The control group animals received carboxymethyl cellulose 1 h prior to injection of acetic acid. Animals were placed in private cages 5 min after acetic acid injection and the number of 'stretching' per animal was recorded during the following 10 min period; percent analgesic activity was calculated by using the formula:

$$\text{Percent anti-nociceptive activity} = \frac{n - n'}{n} \times 100$$

where n is the average number of 'stretching' of control group and n' is the average number of 'stretching' of test group.

According to this calculation maximum antinociceptive activity could be 100% whereas it equals 0% in control. The reference drug was administered according to the test protocol.

5. Result and discussion

If one evaluates the antinociceptive activities of the compounds, it can easily be seen that all the compounds are more active than aspirin except **IId**. Compound **IIf** exhibited the highest antinociceptive activity among 6-substituted-3-(2H)-pyridazinone derivatives synthesized. This compound, **IIf**, bears 4-(4-fluorophenyl)piperazine moiety at the position 6 of the ring. There are many studies in the literature about the antinociceptive activity of the pyridazinone derivatives and in one of them, it was pointed out that the pyridazinone derivatives carrying fluorophenyl, fluorophenylpiperazine and trifluoromethyl-phenylpiperazine moieties might show highest antinociceptive activity, among the compounds, 4-phenyl-6-aryl-2-[3-arylpiperazin-1-yl)propyl]pyridazin-3-ones (**4**), synthesized in the study mentioned, the compound which bears 4-fluorophenyl substituent at the position 6 was the most active one [8]. In the same fashion, among the derivatives of 4,6-diaryl-3-pyridazinones (**2**), the compounds carrying fluor atom at the position R_1 and 3-threefluoromethylphenyl-piperazinomethyl moiety at position R_2 exhibited maximum antinociceptive activity [7]. In addition to these findings, derivatives of 3-[6-(4-methoxyphenyl)-3(2H)-pyridazino-2-yl]-acetamide and 3 - [6 - (4 - methoxyphenyl) - 3(2H) - pyridazino - 2 - yl]-propanamide (**7**) were synthesized and it was reported that fluorophenylpiperazine and trifluoromethylphenylpiperazine moiety on the amide part of these compound have positive influences on their antinociceptive activity [16]. These results demonstrated that there could be a positive contribution of these moieties to antinociceptive activity of such compounds. Further studies are in progress on this subject.

Acknowledgements

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