ELSEVIER

www.elsevier.nl/locate/farmac

Il Farmaco 54 (1999) 846–851

IL FARMACO

Short Communication

Synthesis and antinociceptive activity of some 3-substituted benzothiazolone derivatives

Bilge Çakir [a], Aysel Ulucay [b], Deniz S. Dogruer [a], Askin Isimer [b], M. Fethi Sahin [a,*]

[a] *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Gazi University, Hipodrom, Ankara 06330, Turkey*
[b] *Center of Pharmaceutical Sciences, Gulhane Military Medical Academy, Etlik, Ankara, Turkey*

Received 4 March 1999; accepted 6 August 1999

Abstract

Thirteen 3-substituted benzothiazolone derivatives have been synthesized. Their chemical structures have been elucidated by IR and NMR spectral data and by elemental analyses. Among these compounds, 1-{3-[2(3*H*)-benzothiazolon-3-yl]propanoyl}morpholine (**5b**); 1-{3-[2(3*H*)-benzothiazolon-3-yl]propanoyl}-4-benzylpiperidine (**5c**); 1-{3-[2(3*H*)-benzothiazolon-3-yl]-propanoyl}-4-phenylpiperazine (**5d**); 3 - [3 - (4 - benzylpiperidine - 1 - yl)propyl] - 2(3*H*)-benzothiazolone (**5k**); 3 - [3 - (4 - benzylpiperazine-1-yl)propyl]-2(3*H*)-benzothiazolone (**5l**); 3-[3-(4-phenylpiperazine-1-yl)propyl]-2(3*H*)-benzothiazolone (**5m**) have been found to be significantly more active than the others.

Keywords: Antinociceptive activity; Benzothiazolone derivatives; Synthesis

1. Introduction

As it is widely accepted there are two classes of compounds which are used in clinical analgesia: (a) peripheral analgesics, which can be divided into two subclasses namely cyclooxygenase inhibitors (NSAIDs) and peripheral opioids, dipyrone can be added to this group; (b) central opioids. However, each class has disadvantages. NSAIDs frequently cause gastrointestinal disorders, while opiates lead to tolerance, physical dependency and addiction. The aim of current analgesic research is to develop new NSAIDs without such side effects [1,2].

Benzothiazolinone derivatives have also been reported as potent analgesic agents. In 1995, Ferreira and co-workers screened the antinociceptive activity of 6-benzoylbenzothiazolone and concluded that it might release an endogenous, opioid like substance from the adrenal glands exerting the antinociceptive activity [3]. The compound was claimed to have little or no anti-inflammatory activity. On the other hand, one of the benzothiazolinone derivatives, 4-[(5-chloro-2-oxo-3-benzothiazolinyl)acetyl]-1-piperazinylethanol hydrochloride (Tiaramide HCl, Fig. 1) is a well-known anti-inflammatory agent. It was also proven to have analgesic antihistaminic activity with a low incidence of mild side effects [4,5].

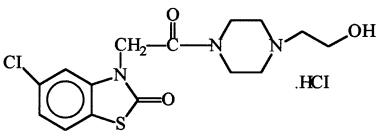


Fig. 1. Tiaramide hydrochloride.

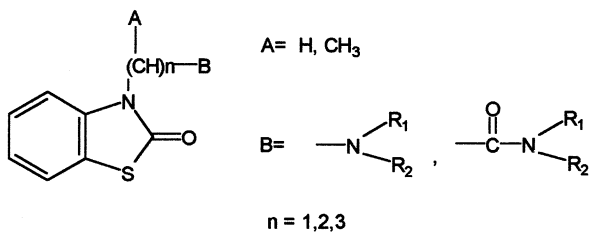
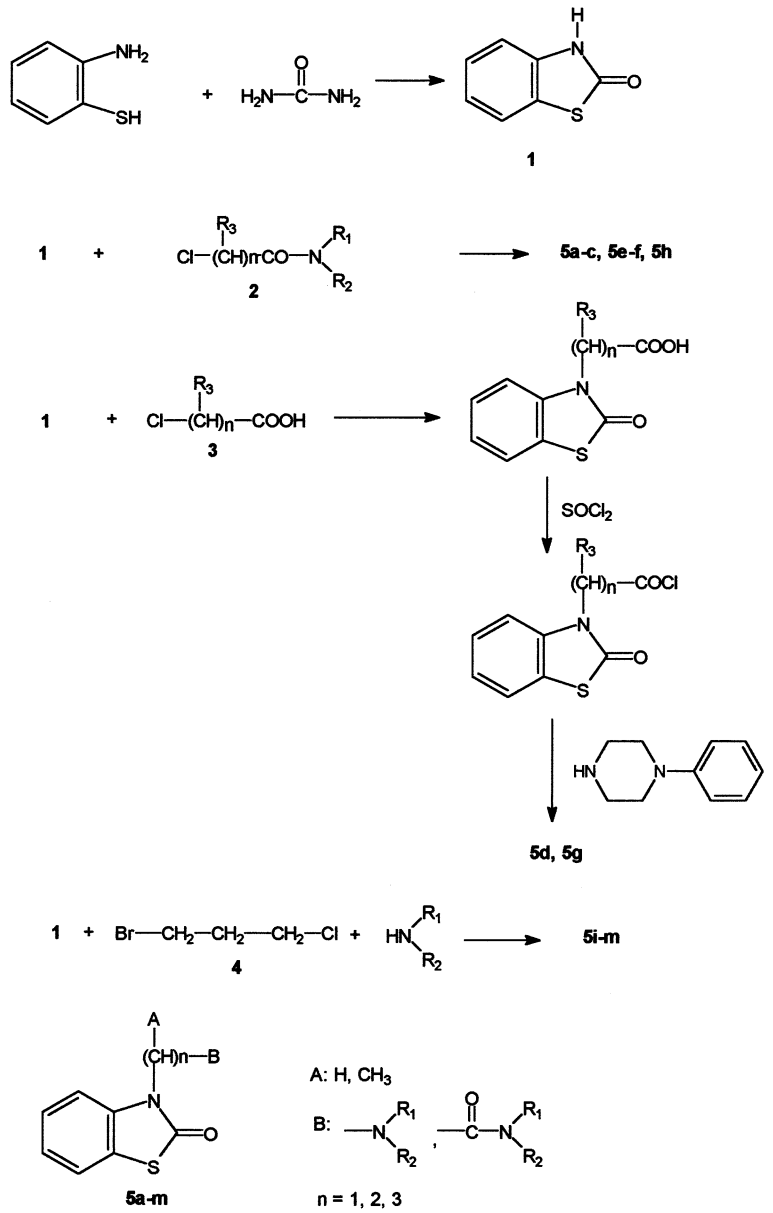


Fig. 2. The general formula of the compounds to be synthesized.

* Corresponding author. Tel.: +90-312-223 5018.

0014-827X/99/$ - see front matter
PII: S0014-827X(99)00111-1



Scheme 1. Synthesis of the title compounds.

On the other hand several benzoxazolone and oxazolopyridine derivatives have been synthesized and claimed to have significant analgesic activity, and the authors claimed that if these compounds have a three-carbon chain, between the two nitrogens on the substituents at position three, they exhibit better analgesic activity. The authors also stressed the significance of phenylpiperazino grouping at the side chain on the oxazole nitrogen of the ring of the compound they synthesized [6,7].

Therefore, in order to evaluate the importance of the feature of the side chain mentioned above in the title compounds, the synthesis and investigation of the antinociceptive activity of the compounds shown in Fig. 2 was pursued.

2. Chemistry

The syntheses of the title compounds have been realized as illustrated in Scheme 1. Synthesis of 2(3*H*)-benzothiazolone (**1**) was carried out by the reaction of *o*-aminothiophenol with urea [8]. 2(3*H*)-Benzothiazolone was reacted either with 3-chloropropanamide derivatives **2**, or 3-chloropropanoic acid (**3**) followed by thionyl chloride to give the acid chloride, which afterwards was reacted with an appropriate amine to obtain the final compounds **5a**–**5h**. 2(3*H*)-Benzothiazolone was also reacted with bromochloropropane (**4**) and an amine in the presence of potassium carbonate to form the final compounds **5i**–**5m** (Scheme 1). In the synthesis of the aminopropylbenzothiazolinone derivatives, the

symmetrical amines could have been obtained. Therefore, it was thought to reduce the corresponding amides to aminopropylbenzothiazolinones but this attempt was not successful. As a result the above mentioned synthetic method was utilized. There was a possibility to obtain symmetrical diamines instead of the expected compounds, however, in the work-up step the title compounds were obtained pure.

3. Experimental

3.1. Chemistry

All the chemicals used in this study were purchased from either Aldrich or Merck AG. 3-[2(3*H*)-Benzothiazolon-3-yl]propanoic acid and 3-[2(3*H*)-benzothiazolon-3-yl]propanoyl chloride were synthesized in our laboratory. The melting points are 142°C for the acid and 67°C for the acid chloride, which were in accordance with those cited in the literature [9]. Therefore, they were used in the next steps without further analysis.

The melting points of the compounds were determined using an Electrothermal 9000 melting point apparatus and were uncorrected.

The IR spectra of the compounds were recorded on a Perkin–Elmer 1330 IR spectrophotometer. The ^{1}H NMR spectra were recorded on a Bruker 200 FT NMR spectrometer.

3.1.1. Synthesis of 2(or 3)-chloropropanoylamines

A total of 0.03 mol of amine was dissolved in 30 ml of chloroform and 0.01 mol of 2- or 3-chloropropanoyl chloride was added dropwise while stirring. The mixture was stirred for 5 min while cooling and refluxed for a further 10 min on a water bath. The chloroform solution was extracted with 5% HCl and washed with water, dried over anhydrous sodium sulfate, and filtered. Chloroform was evaporated under diminished pressure. The crude amide was obtained.

3.1.2. Synthesis of the amides (**5a–5h**)

3.1.2.1. Method A. Metallic sodium (0.01 mol) was dissolved in 30 ml of ethanol. The ethanol was evaporated. The residue was dissolved in 20 ml of *N*,*N*-dimethylformamide and 0.01 mol of 2-oxobenzothiazoline, and 0.01 mol of the corresponding chloroamide was added. The eventual solution was refluxed and the reaction progression was monitored through TLC on silica gel using a 1:4 methanol–chloroform mixture as the solvent system. When the reaction was completed the mixture was poured into 50 ml of ice-cold water and stirred. The oil that separated was taken into 50 ml of acetone and treated with activated charcoal. The acetone was evaporated and the residue was crystallized from an acetone–water mixture.

3.1.2.2. Method B. The compound 2(or 3)-(2-oxobenzothiazolin-3-yl) propanoyl chloride (0.03 mol) was dissolved in 120 ml of heptane. Triethylamine (0.03 mol) and phenylpiperazine (0.03 mol) were added. The eventual mixture was stirred for 18 h at room temperature. The organic solvent was evaporated and the residue was treated with 50 ml of water, stirred for an extra hour at room temperature, then extracted with chloroform, dried over sodium sulfate, and filtered. Chloroform was evaporated to dryness and the residue was crystallized with ethanol–water mixture.

Compounds **5c** and **5e** were purified through column chromatography on silica gel 230–400 mesh and using a 2:3 benzene–ethyl acetate mixture as eluent.

3.1.3. Synthesis of 3-(3-aminopropyl)-2-oxobenzothiazoline hydrochlorides (**5i–5m**)

A total of 0.1 mol of 2(3*H*)-benzothiazolone, 0.03 mol of potassium carbonate, 1-bromo-3-chloropropane and an appropriate amine were placed in 30 ml of anhydrous ethanol and refluxed for 12 to 20 h depending on the amine used. Ethanol was evaporated at the end of the reaction and the residue was treated with 50 ml of water and extracted with ether. The ethereal portions were pooled and dried over sodium sulfate, concentrated on a rotary evaporator and treated with ethereal HCl. The precipitate thus formed, was filtered and dried at room temperature, crystallized out from an appropriate solvent and dried at room temperature.

3.2. Biology

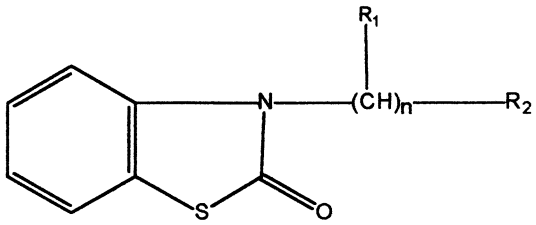
3.2.1. Animals

Albino mice of both sexes (23 ± 3.0 g), which are a local breed, were employed. The animals were housed in groups of eight, with food and tap water ad libitum, and were received in the laboratory at least 2 days before the experiments to allow them to get accustomed to the environment. The food was withdrawn on the day before the experiment, but they were allowed free access to tap water. The treatment was performed in full awareness of the test animals. Acetic acid (Merck A.G), carboxymethyl cellulose sodium salt (CMC Na) (Aldrich) and aspirin (Bayer) were used.

3.2.2. Analgesic activity test

A modified Koster's test was employed [10]. The Koster test was first used by Koster et al. in mice for a dosage of 60 mg/kg acetic acid (0.6 percentage solution) by intraperitoneal injection to produce repeated characteristic stretching movements [11]. Safak et al. used acetic acid at a dose level of 300 mg/kg (3% solution)

Table 1
Chemical structure, yield percentages, melting points and crystallization solvents of the compounds synthesized



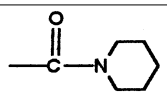
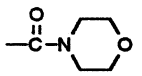
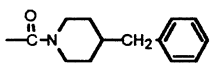
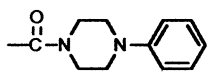
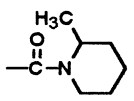
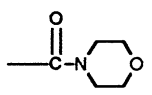
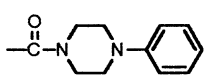
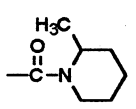
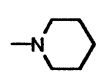
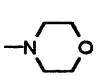
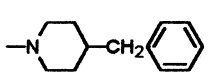
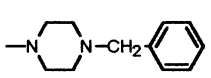
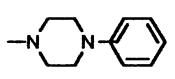
Comp.	*n*	R_1	R_2	Empirical formula	M.p. (°C)	Yield (%)	Method of synthesis	Cryst. solvent
5a	2	H		$C_{15}H_{18}N_2O_2S$	80–83	48	A	acetone–water
5b	2	H		$C_{14}H_{16}N_2O_3S$	100–103	19.2	A	acetone–water
5c	2	H		$C_{22}H_{24}N_2O_2S$	93–98	12.4	A	
5d	2	H		$C_{20}H_{21}N_3O_2S$	126–128	21.9	B	ethanol–water
5e	2	H		$C_{16}H_{20}N_2O_2S$	oil	31.5	A	
5f	1	CH_3		$C_{14}H_{16}N_2O_3S$	102–104	48.3	A	acetone–water
5g	1	CH_3		$C_{20}H_{21}N_3O_2S$	176–179	32.7	B	acetone–water
5h	1	CH_3		$C_{16}H_{20}N_2O_2S$	74–77	39.1	A	cyclohexane
5i	3	H		$C_{15}H_{20}N_2OS.HCl$	189–191	57.0		acetone
5j	3	H		$C_{14}H_{18}N_2O_2S.HCl$	191–194	48.3		acetone
5k	3	H		$C_{22}H_{26}N_2OS.HCl$	265–270	31.8		methanol–ether
5l	3	H		$C_{21}H_{25}N_3OS.HCl$	244–247	53.7		acetone
5m	3	H		$C_{20}H_{23}N_3OS.HCl$	302	25.6		ethanol–ether

Table 2
NMR spectral data of the compounds **5a**–**5m**

Comp.	Solvent	^{1}H MNR (δ ppm)
5a	$CDCl_3$	1.44–1.50 (m, 4H), 1.54–1.62 (m, 2H), 2.78 (t, 2H), 3.32 (t, 2H), 3.51 (t, 2H), 4.30 (t, 2H), 7.14 (t, 1H), 7.22 (d, 1H), 7.32 (t, 1H), 7.41 (d, 1H)
5b	DMSO-d_6	2.70 (t, 2H), 3.10–3.80 (m, 8H), 4.15 (t, 2H), 7.00–7.75 (m, 4H)
5c	DMSO-d_6	0.89–0.95 (m, 2H), 1.49 (t, 2H), 1.65–1.75 (m, 1H), 2.39–2.45 (m, 2H), 2.66–2.84 (m, 6H), 4.13–4.17 (m, 2H), 7.11–7.21 (m, 4H), 7.26 (t, 2H), 7.36 (d, 2H), 7.63 (d, 1H)
5d	DMSO-d_6	2.67 (t, 2H), 2.92–2.97 (m, 4H), 3.40 (t, 2H), 3.45 (t, 2H), 4.06 (t, 2H), 6.67 (t, 1H), 6.80 (d, 2H), 7.05–7.11 (m, 3H), 7.24–7.27 (m, 2H), 7.52 (d, 1H)
5e	DMSO-d_6	0.90–1.80 (m, 9H), 1.90–3.30 (m, 5H), 4.00–4.30 (t, 2H), 7.00–7.80 (m, 4H)
5f	DMSO-d_6	1.50 (d, 3H), 2.90–3.70 (m, 8H), 5.60 (q, 1H), 7.00–7.90 (m, 4H)
5g	DMSO-d_6	1.50 (d, 3H), 3.00–3.50 (m, 8H), 5.65 (q, 1H), 6.50–7.80 (m, 9H)
5h	DMSO-d_6	1.00–1.80 (m, 12H), 2.80 (m, 3H), 5.50 (q, 1H), 7.10–7.80 (m, 4H)
5I	DMSO-d_6	1.51–1.92 (m, 4H), 2.09–2.14 (m, 2H), 3.08 (t, 2H), 3.31–3.36 (m, 6H), 4.03 (t, 2H), 7.22 (t, 1H), 7.40 (t, 1H), 7.47 (d, 1H), 7.67 (d, 1H), 10.50 (s, 1H)
5j	$CDCl_3$	2.32–2.39 (m, 2H), 3.21 (t, 2H), 3.33–3.42 (m, 4H), 3.82–4.02 (m, 4H), 4.12 (t, 2H), 7.19 (t, 1H), 7.30 (d, 1H), 7.36 (t, 1H), 7.47 (d, 1H), 12.4 (s, 1H)
5k	$CDCl_3$	1.54–1.81 (m, 3H), 2.02–2.11 (m, 2H), 2.44–2.67 (m, 6H), 3.00–3.03 (m, 2H), 3.46–3.55 (m, 2H), 4.09 (t, 2H), 7.10 (d, 2H), 7.16–7.22 (m, 3H), 7.27 (t, 2H), 7.36 (t, 1H), 7.44 (d, 1H), 12.4 (s, 1H)
5l	DMSO-d_6	1.74–2.27 (m, 2H), 3.20–3.61 (m, 10H), 4.03 (t, 2H), 4.31 (s, 2H), 7.21 (t, 1H), 7.36–7.48 (m, 5H), 7.63–7.67 (m, 3H), 12.00 (s, 1H)
5m	DMSO-d_6	1.66–1.70 (m, 2H), 2.20 (t, 2H), 2.26 (t, 4H), 2.89 (t, 4H), 3.85 (t, 2H), 6.59 (t, 1H), 6.72 (d, 2H), 7.02 (t, 3H), 7.19–7.25 (m, 2H), 7.46 (d, 1H)

[10]. It was preferred to utilize a 300 mg/kg acetic acid dose in this study. Acetylsalicylic acid (ASA) was used as the reference drug.

Each compound was suspended in 0.5% carboxymethyl cellulose at a concentration of 10 mg/ml and given orally to mice in groups of eight at the 100 mg/kg dose. One hour after this administration, pain was induced by intraperitoneal injection of 3% solution of acetic acid at the 300 mg/kg level. The control group 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; percentage analgesic activity was calculated by using the formula:

$$\text{Precentage antinociceptive activity} = \frac{n - n'}{n} \times 100$$

n = average number of 'stretching' of the control group

n' = average number of 'stretching' of the test group

The reference was administered according to the test protocol.

4. Results and discussion

Thirteen 2(3*H*)-benzothiazolone derivatives were synthesized. Their chemical structures, melting points and percentage yields are shown in Table 1. Their chemical structures have been elucidated by elemental analysis and data obtained from IR (the IR spectra indicated the presence of two carbonyl bands at 1685–1655 and 1645 and 1625 cm^{-1} for the ring carbonyl and amide carbonyl, respectively, for compounds **5a**–**5h**) and NMR spectral analyses. Compounds **5i**–**5m** have been obtained as their hydrochloride salts. Tables 2 and 4 illustrate the ^{1}H NMR spectral data and elemental analyses, respectively.

Table 3 lists the antinociceptive activity of the compounds as the percentage inhibition of stretching movement of the animals in comparison to the control group. Eight out of 13 compounds were found to be

Table 3
Antinociceptive activity of the compounds **5a**–**5m**

Comp.	Antinociceptive activity (%)	Activity compared with aspirin
Aspirin	40.96	1.00
5a	21.46	0.52
5b	71.76 *	1.75
5c	77.52 *	1.89
5d	77.74 *	1.82
5e	55.16	1.34
5f	35.59	0.87
5g	15.99	0.39
5h	15.99	0.39
5I	60.08	1.46
5j	27.27	0.66
5k	87.89 *	2.14
5l	76.47 *	1.86
5m	97.17 *	2.37

* $P < 0.05$.

Table 4
Elemental analysis of the compounds synthesized

Comp.	Calc. elemental analysis (Found) (%)		
	C	H	N
5a	62.04 (61.69)	6.25 (5.73)	9.65 (9.56)
5b	57.52 (57.93)	5.52 (5.64)	9.59 (9.52)
5c	69.45 (70.21)	6.36 (5.67)	7.36 (7.14)
5d	65.37 (65.64)	5.76 (5.45)	11.44 (11.42)
5e	63.13 (63.18)	6.62 (6.87)	9.20 (8.81)
5f	57.52 (57.61)	5.52 (5.59)	9.59 (9.55)
5g	65.37 (65.41)	5.76 (5.76)	11.44 (11.30)
5h	63.13 (63.68)	6.62 (6.76)	9.20 (9.06)
5i	57.59 (58.17)	6.44 (6.46)	8.95 (9.03)
5j	53.41 (53.92)	6.08 (6.01)	8.90 (8.85)
5k	64.68 (64.78)	6.72 (7.47)	7.18 (6.99)
5l	57.27 (56.49)	6.18 (6.38)	9.54 (9.64)
5m	67.95 (68.37)	6.55 (5.92)	11.89 (11.72)

more active than aspirin as antinociceptive agents. Compounds **5b**–**d**, and **5k**–**m** exhibited significantly higher activity than aspirin. The difference between the activity of these compounds and that OF aspirin was found statistically significant by using the Mann–Whitney U test. These six compounds have three carbons between the two nitrogens on the side chain at position 3.

The positive effect of the side chain length on the activity is more evident when comparing activities of compounds bearing the same amido group (compounds **5b**, **5d**, **5e** versus **5f**, **5g**, **5h**). With the exception of **5b**, all the most active compounds (**5c**, **5d**, **5k**, **5l**, **5m**) bear a terminal phenyl moiety, thus, this structural feature can be considered useful though not sufficient for analgesic activity. Compound **5g**, bearing a phenyl piperazino group, was poorly active.

Finally, all other structural features being equal, amino compounds were more active then amides (**5k**, **5m** versus **5c**, **5d**).

Acknowledgements

The Turkish Scientific and Technological Council have supported this study (TUBITAK: project number SBAG/AYD 200).

References

[1] J.H. Jaffe, W.R. Martin, Opioid analgesics and antagonists, in: L.S. Goodman, A. Gilman, T.W. Rall, A.S. Nics, P. Taylor (Eds.), Pharmacological Basis of Therapeutics, Pergamon, New York, 1990, pp. 485–521.
[2] D. Clinch, A.K. Banerjee, G. Ostick, D.W. Levy, Nonsteroidal anti-inflammatory drugs and gastrointestinal adverse effects, J. R. Coll. Physicians Lond. 17 (1983) 228–230.
[3] S.H. Ferreira, B.B. Lorenzotti, M. Devissaguet, D. Lesieur, Y. Tsouderos, S14080, a peripheral analgesic acting by release of an endogenous circulating opioid-like substance, Br. J. Pharmacol. 114 (1995) 303–308.
[4] T. Takashima, Y. Kadoh, S. Kumada, Pharmacological investigations of benzothiazoline derivatives, Arzneim.-Forsch. 22 (1972) 711–715.
[5] K. Tsurumi, Y. Hiramatsu, M. Nozaki, M. Hayashi, H. Fujimura, Anti-inflammatory activity of tiaramide hydrochloride, 4-[2-(5-chloro-2-oxo-3-benzothiazolinyl)acetyl]-1-piperazineethanol hydrochloride, Arzneim.-Forsch. 22 (1972) 716–724.
[6] M.C. Viaud, P. Jamoneau, C. Flouzat, J.G.B. Espiard, B. Pfeiffer, P. Renard, D.H. Caignard, G. Adam, G.J. Guillaumet, *N*-Substituted oxazolo[5,4-*b*]pyridine-2(1*H*)-ones: a new class of non-opiate antinociceptive agents, J. Med. Chem. 38 (1995) 1278–1286.
[7] M.C. Bermann, J.P. Bonte, I. Lesieur-Demarquilly, M. Debaert, D. Lesieur, M. Leinot, J. Benoit, C. Labrid, Pharmacomodulation du benzoxazolinone par la structure aryl-piperazinique, Eur. J. Med. Chem. 17 (1982) 85–88.
[8] T.H. Fife, J.E.C. Hatchins, M.S. Wang, Higly efficient intramolecular nucleophilic reactions. The cyclization of *p*-nitrophenyl-*N*-(2-mercaptophenyl)-*N*-methylcarbamate and phenyl-*N*-(2-aminophenyl)-*N*-methylcarbamate 97 (1975) 5878–5882.
[9] W. Nimich, Benzothiazolyl carboxylic acids, J. Pract. Chem. 27 (1965) 220–224 Ref. Chem. Abstr. 62 (1965) 11797.
[10] C. Safak, H. Erdogan, E. Palaska, R. Sunal, S. Duru, Synthesis of 3-(2-pyridinylethyl)benzoxazolinone derivatives: potent analgesic and anti-inflammatory compounds inhibiting prostaglandin E_2, J. Med. Chem. 35 (1992) 1296–1299.
[11] R. Koster, M. Anderson, E.J. Debeer, Acetic acid for analgesic screening, Fed. Proc. 18 (1959) 412.