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# SYNTHESIS AND PHARMACOLOGICAL PROPERTIES OF NEW DERIVATIVES OF 4-ALKOXY-6-METHYL-1H-PYRROLO[3,4-c]PYRIDINE-1,3(2H)-DIONES

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**Abstract:** Synthesis of 2-(2-hydroxy-3-amino)propyl derivatives of 4-alkoxy-6-methyl-1H-pyrrolo[3,4-c]pyridine-1,3(2H)-diones (**24**-**35**) is described. The chlorides used in the above synthesis exist mainly in the cyclic forms (**18**, **20**-**23**). Only chloride with benzhydryl substituent at the nitrogen atom of piperazine has the chain structure (**19**). Among the studied imides the most active analgesics in the "writhing" syndrome test proved to be compounds **30** and **31** (with $LD_{50}$ > 2000 mg/kg) containing 4-benzylpiperidino group. Furthermore, all imides suppressed significantly spontaneous locomotor activity of mice.

**Keywords:** pyrrolo[3,4-c]pyridine derivatives, 1-chloro-3-aminopropan-2-ols and their derivatives, azetidinium salts, synthesis, analgesic and sedative activities

In the previous papers (1-3) we described the chemical and pharmacological properties of the series of 4-alkoxy-6-methyl-1H-pyrrolo[3,4-c]pyridine-1,3(2H)-diones (3,4-pyridinedicarboximide) derivatives. All compounds were more active as analgesic agents than acetylsalicylic acid (ASA) in the "writhing syndrome" test ($ED_{50}$ = 0.4 –16.4 mg/kg, for aspirin $ED_{50}$ = 39.15 mg/kg). In the "hot plate" test some of them were devoid of the analgesic properties at a dose of 200 mg/kg, similarly as ASA used at the same dose. In the most cases the tested compounds were not toxic ($LD_{50}$ > 2000 mg/kg).

From among the imide derivatives synthesized hitherto compound **1** (Fig. 1) displayed the strongest antinociceptive action (1). Introduction of trifluoromethyl or methoxy groups into the phenyl substituent at the N-4 atom of piperazine in the imide **1** (compounds **3** and **5**) weakened the analgesic properties both in the "writhing syndrome" and "hot plate" tests.

Similar relationship took place in the case of compounds **2** (ethoxy homologue of **1**) and **4** in the first test. In the "hot plate" test imide **4** was more active than the parent substance **2** (1). In continuing research on this series of compounds we reported recently (3) that introduction of halogens (Cl, F) into the phenyl substituent in **1** and **2** (imides **6-13** and **14**, **15** with simultaneous elimination of OH group in the propyl chain) generated a decrease of the analgesic action. At the same time an increase of the sedative activity was observed. Worth of notice is a fact that these structural changes did not influence the toxicity of the above-mentioned compounds. All had $LD_{50}$ > 2000 mg/kg.

In order to obtain new information concerning structure activity relationship in this group of compounds we carried out further modification of the structure of 1H-pyrrolo[3,4-c]pyridine-1,3(2H)-diones **1** and **2**. This modification consisted in the replacement of: 1) phenyl substituent at N-4 of piperazine by benzyl and benzhydryl groups, 2) N-substituted piperazine by other cyclic amines (piperidine, pyrrolidine and morpholine). We wanted to know, if these structural changes would influence the toxicity, analgesic and sedative activities of the tested compounds.

With this aim we carried out the synthesis of the appropriate 3,4-pyridinedicarboximides (**24-35**) presented in Figure 2. In the case of compounds **28** and **29** the benzyl substituent was additionally introduced into the position 4 of the piperidine ring

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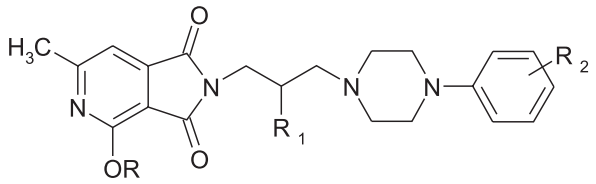

Figure 1. R, $R_1$, $R_2$ are as follows: **1** $CH_3$, OH, H; **2** $C_2H_5$, OH, H; **3** $CH_3$, OH, *m*-$CF_3$; **4** $C_2H_5$, OH, *m*-$CF_3$; **5** $CH_3$, OH, *o*-$OCH_3$; **6**, **7** $CH_3$, OH, *m*(*p*)-Cl; **8**, **9** $C_2H_5$, OH, *m*(*p*)-Cl; **10**, **11** $CH_3$, OH, *o*(*p*)-F; **12**, **13** $C_2H_5$, OH, *o*(*p*)-F; **14** $CH_3$, H, *m*-Cl; **15** $C_2H_5$, H, *m*-Cl.

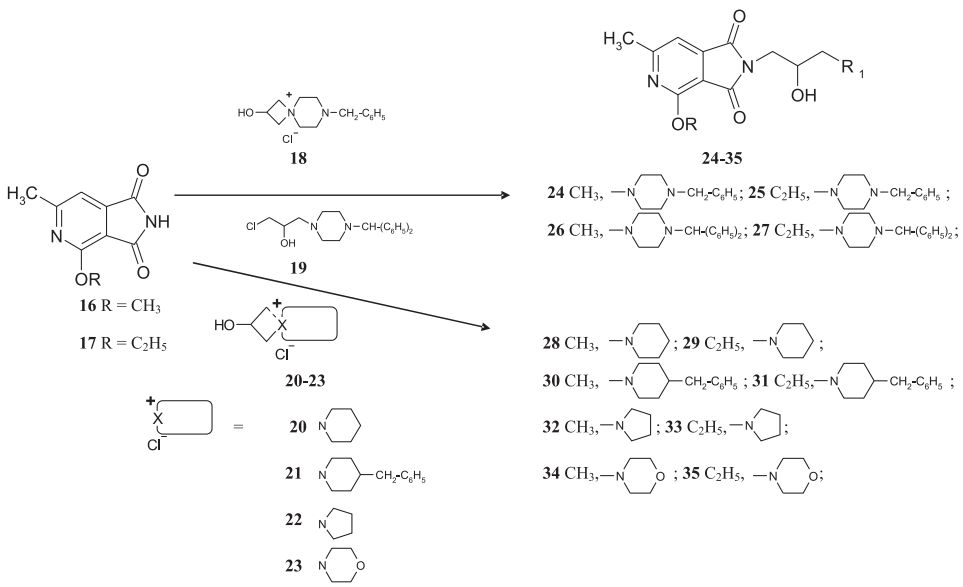

Figure 2. Synthesis of the investigated compounds

(imides **30**, **31**). We expected that the compounds obtained would exhibit the analgesic and sedative properties.

**Chemistry**

The starting materials for the synthesis of the above mentioned compounds were 4-methoxy- and 4-ethoxy-6-methyl-1H-pyrrolo[3,4-c]pyridine-1,3(2H)-diones (2-methoxy- and 2-ethoxy-6-methyl-3,4-pyridinedicarboximides (**16**, **17**) (4). They were condensed with the appropriate chain (**19**) or cyclic (**18**, **20**-**23**) chlorides in anhydrous ethanol in the presence of potassium ethoxide giving compounds **24-35** (Fig. 2). Their structures were confirmed by elemental and spectral (IR, $^1$H NMR) analyses.

The above mentioned chlorides were synthesized in the reaction of epichlorohydrin with appropriate cyclic amines in ethanol according to the literature (1, 5-9). They can exist in chain or cyclic forms. Compounds containing piperidine, pyrrolidine and morpholine moieties are known as 3-hydroxyazetidinium salts **20** (6, 7), **22** (8) and **23** (8, 9). The similar structure we ascribed to the newly synthesized chloride **21** with 4-benzylpiperidine rest, based on the results of our previous research (confirmed by X-ray analysis) as well as elemental and spectral analyses (1). Compound **21** was soluble in water and gave positive reaction for chloride ions with silver nitrate. Using the same criterion we stated that the chloride containing N-benzylpiperazine rest also formed the cyclic structure **18,** whereas its water insoluble analogue with N-benzhydryl substituent exists in the chain form **19**. In the literature (5) there is a mention concerning 1-chloro-3-[4-benzyl(and benzhydryl)-1-piperazinyl]propan-2-ols, which without isolation (after reaction of epichlorohydrin with suitable piperazines) were used as inter-

Table 1. Properties of the investigated compounds.

| Compound | Formula (molecular weight) | M.p. (°C) solvent | Yield % | IR absorptions in KBr ($cm^{-1}$) | | |
|---|---|---|---|---|---|---|
| | | | | CO | OH | Monosubstituted benzene |
| **24** | $C_{23}H_{28}N_4O_4$ (424.50) | 128-130 ethanol | 80 | 1715, 1770 | 3120 | 695, 755 |
| **25** | $C_{24}H_{30}N_4O_4$ (438.53) | 118-120 ethanol | 85 | 1715, 1770 | 3090 | 695, 755 |
| **26** | $C_{29}H_{32}N_4O_4$ (500.60) | 102-104 ethanol | 40 | 1715, 1770 | 3080 | 705, 750 |
| **27** | $C_{30}H_{34}N_4O_4$ (514.63) | 127-129 ethanol | 40 | 1720, 1775 | 3090 | 700, 750 |
| **28** | $C_{17}H_{23}N_3O_4$ (333.39) | 127-129 ethanol | 66 | 1705, 1775 | 3090 | - |
| **29** | $C_{18}H_{25}N_3O_4$ (347.42) | 115-117 ethanol | 70 | 1715, 1770 | 3080 | - |
| **30** | $C_{24}H_{29}N_3O_4$ (423.52) | 157-159 ethanol | 84 | 1720, 1780 | 3100 | 705, 760 |
| **31** | $C_{25}H_{31}N_3O_4$ (437.54) | 130-131 ethanol | 76 | 1715, 1775 | 3100 | 700, 760 |
| **32** | $C_{16}H_{21}N_3O_4$ (319.36) | 129-131 ethanol | 65 | 1730, 1780 | 3090 | - |
| **33** | $C_{17}H_{23}N_3O_4$ (333.39) | 117-119 cyclohexane | 62 | 1710, 1780 | 3100 | - |
| **34** | $C_{16}H_{21}N_3O_5$ (335.36) | 138-140 ethanol | 75 | 1700, 1775 | 3090 | - |
| **35** | $C_{17}H_{23}N_3O_5$ (349.39) | 136-137 ethanol | 76 | 1705, 1765 | 3090 | - |

mediates in the synthesis of 6-substituted purine derivatives.

## EXPERIMENTAL

### Chemistry

All the results of C, H, N determinations (carried out using a Carlo Erba Elemental Analyzer model NA-1500) were within ± 0.4% of the theoretical values. All uncorrected melting points were determined on a Boetius apparatus. The IR spectra, in KBr pellets, were measured with a Specord M 80 (C. Zeiss, Jena). $^1$H NMR spectra were determined in $CDCl_3$ solutions on a Tesla 587 A spectrometer (80 MHz), when not indicated otherwise, using TMS as an internal standard.

General method for synthesis of chlorides **18**, **19** and **21**.

Epichlorohydrin (0.02 mol) was added to a vigorously stirred solution of the appropriate cyclic amine (N-benzylpiperazine, N-benzhydrylpiperazine, 4-benzylpiperidine) (0.02 mol) in 50 mL of anhydrous ethanol. The mixture was stirred at room temperature for 5 h. The separated solid substance **19** was collected on a filter and washed first with ethanol, then with distilled water.

In the case of compounds **18** and **21** a few milliliters of diethyl ether were added to the reaction mixture and then it was left overnight. The resulting solid substance was collected on a filter and washed with a small amount of ethanol.

Chlorides **18** and **21** were water-soluble and gave positive reaction for chloride ions with silver nitrate. Compound **19** was insoluble in water.

Chloride of 2-hydroxy-7-benzyl-7-aza-4-azoniaspiro[3,5]nonane – **18**

$C_{14}H_{21}ClN_2O$, $M_w$ = 268.79 (C, H, N determinations), m.p. 133-134°C (yield 71%). $^1$H NMR in DMSO-$d_6$ (300 MHz, δ ppm): 2.48-2.63 (m-4H, -($CH_2$)$_2$-N-$CH_2$-Ph); 3.40-3.66 (m-6H, -$CH_2$-Ph + -$N^+$($CH_2$)$_2$); 4.12-4.53 (m-4H, 2 x $CH_2$); 4.58-4.69 (m-1H, C**H**); 6.64-6.66 (d-1H, O**H**); 7.25-7.37 (m-5H, arom. H).

2-Hydroxy-3-(4-benzhydryl-1-piperazinyl)propyl chloride – **19**

$C_{20}H_{25}ClN_2O$, $M_w$ = 344.89 (C, H, N determinations), m.p. 129-131°C (yield 88%).

Table 2. Acute lethal toxicity, influence on the pain reaction in the „hot-plate" and the „writhing syndrome" tests and on the spontaneous locomotor activity of the investigated compounds in mice.

| Compounds | Acute lethal toxicity | Pain reaction | | | | Spontaneous locomotor activity | |
|---|---|---|---|---|---|---|---|
| | $LD_{50}$ mg/kg (*i.p.*)[1] | "hot plate" test | | "writhing syndrome" test | | | |
| | | $ED_{50}$ (mg/kg) | $LD_{50}/ED_{50}$ | $ED_{50}$ (mg/kg) | $LD_{50}/ED_{50}$ | $ED_{50}$ (mg/kg) | $LD_{50}/ED_{50}$ |
| **24** | > 2000 | inactive | | 66.7 (51.5 – 86.4) | > 29.9 | 68.5 (51.1 – 92.0) | > 29.2 |
| **25** | 1214.6 (1094.2 – 1334.2) | 71.2 (54.7 – 92.6) | 17.05 | 51.3 (30.6 – 86.1) | 23.7 | 65.8 (46.9-92.3) | 18.46 |
| **26** | > 2000 | 162.3 (135.2 – 194.7) | > 12.32 | 107.7 (66.9 – 173.4) | 18.6 | 69.1 (51.8 – 92.1) | > 28.9 |
| **27** | > 2000 | inactive | | 36.8 (23.8 – 56.9) | > 54.3 | 64.6 (48.6 – 85.9) | > 30.9 |
| **28** | 1764.9 (1713.5 – 1870.8) | inactive | | 69.3 (50.7 – 94.7) | 25.5 | 70.2 (52.9 – 93.1) | 25.1 |
| **29** | 885.4 (851.3 – 920.8) | 187.0 (143.8 – 243) | 4.73 | 43.9 (28.0 – 68.8) | 20.2 | 43.9 (26.3 – 73.3) | 20.2 |
| **30** | > 2000 | 124.6 (89 – 174.4) | > 16.05 | 10.3 (5.95 – 17.81) | > 194.2 | 12.03 (7.51 – 19.2) | > 166.25 |
| **31** | > 2000 | 144.0 (110.7 – 187.2) | > 13.9 | 12.9 (9.21 – 18.06) | > 155.0 | 86.2 (50.7 – 146.5) | > 23.20 |
| **32** | 1624.4 (1541.1 – 1712.0) | 92.5 (66.1 – 129.5) | 17.56 | 67.3 (51.1 – 88.7) | 24.1 | 70.0 (52.4 – 93.5) | 23.2 |
| **33** | 1694.1 (1644.7 – 1744.9) | inactive | | 40.6 (25.8 – 63.8) | 41.7 | 46.7 (25.4-86.0) | 36.3 |
| **34** | 599.0 (516.4 – 694.8) | 172.1 (143.4 – 206.5) | 3.48 | 59.0 (46.7 – 74.7) | 10.1 | 70.9 (53.4 – 94.2) | 8.4 |
| **35** | 1240.0 (953 – 1612) | inactive | | 62.0 (44.3 – 86.8) | 20.0 | 85.0 (70.8 – 102.0) | 14.6 |
| Acetylsalicylic acid | 167.0 [10] | inactive at a dose of 200 mg/kg | | 39.15 (29.1 – 48.4) | 4.26 | - | - |
| Morphine | 140.0 [11] | 3.5 (3.0 – 4.1) | 40 | 2.44 (1.18 – 5.02) | 57.4 | - | - |

[1] The data are the median lethal doses with 5 % confidence limits in parentheses (n = 6)

$^1$H NMR (δ ppm): 2.02-2.86 (m-10H, 8H of piperazine + C**H**$_2$γ); 3.43-3.65 (distorted d-3H, C**H**$_2$α + OH); 3.72-4.02 (m-1H, C**H**α); 4.22 (s-1H, C**H**-(Ph)$_2$); 6.98-7.60 (m-10H, arom. H).

Chloride of 2-hydroxy-7-benzyl-4-azoniaspiro[3,5] nonane – **21**

$C_{15}H_{22}ClNO$, $M_w$ = 267.80 (C, H, N determinations), m.p. 167-168°C (yield 84%).

$^1$H NMR in DMSO-d$_6$ (300 MHz, δ ppm): 1.37-1.78 (m-5H, Pip-4-**H** + Pip-3-C**H**$_2$ + Pip-5-C**H**$_2$); 2.48-2.57 (d-2H, C**H**$_2$-Ph); 3.18-3.41 (m-2H, *Pip-2*-**H**$_{ax}$ + *Pip-6*-**H**$_{ax}$); 3.56-3.80 (2 x d-1H, Pip-2-**H**$_{eq}$ + Pip-6-**H**$_{eq}$); 4.07-4.51 (m-2H, 2 x C**H**$_2$); 4.58-4.64 (m-1H, C**H**); 6.64-6.67 (d-1H, O**H**); 7.15-7.32 (m-5H, arom. H).

General method for synthesis of 2-(2-hydroxy-3-amino)propyl derivatives of 4-methoxy and 4-ethoxy-6-methyl-1H-pyrrolo[3,4-c]pyridine-1,3 (2H)-diones (**24-35**)

A 0.01 mol of potassium was dissolved in 120 mL of anhydrous ethanol and to this solution 0.01 mol of 4-methoxy- or 4-ethoxy-6-methyl-1H-pyrrolo[3,4-c]pyridine-1,3(2H)-dione (**16**, **17**) was added. The reaction mixture was refluxed for 15 min in case of imide **16** and to the obtained suspension (imide **16**) or clear solution (imide **17**) 0.012 mol of the appropriate chloride **18**-**23** was added. The mixture was refluxed until the alkaline reaction disappeared. After filtration, the clear solution was left to crystallize (compound **30**). Then the separated product was collected on a filter and purified by crystallization from ethanol. In other cases the filtrate was evaporated to a small volume and the residue was left to crystallize. The further procedure was the same as for imide **30**.

The properties of compounds **24-35** are presented in Table 1 but the assignments of their $^1$H NMR spectra are shown below:

$^1$H NMR of compound **24** (δ ppm): 2.20-2.94 (m-13 H, 8 H of piperazine + C**H**$_2$γ + C**H**$_3$); 3.45 (s-2H, C**H**$_2$-Ph); 3.64-3.83 (distorted d-3H, C**H**$_2$α + OH); 3.85-4.25 (m-4H, OC**H**$_3$ + C**H**β); 7.13-7.49 (m-6H, arom. H).

$^1$H NMR of compound **25** (δ ppm): 1.28-1.59 (t-3H, -CH$_2$-C**H**$_3$); 2.23-2.82 (m-13H, 8H of piperazine + C**H**$_2$γ + C**H**$_3$); 3.46 (s-2H, C**H**$_2$-Ph); 3.61-3.78 (distorted d-3H, C**H**$_2$α + O**H**); 3.82-4.11 (m-1H, C**H**β); 4.43-4.78 (q-2H, -C**H**$_2$-CH$_3$); 7.10-7.46 (m-6H, arom. H).

$^1$H NMR of compound **26** (δ ppm): 2.15-3.09 (m-13H, 8H of piperazine + C**H**$_2$γ + C**H**$_3$); 3.54-3.76 (distorted d-3H, C**H**$_2$α + OH); 3.83-4.21 (m-5H, OC**H**$_3$ + C**H**β + C**H**-Ph); 7.05-7.51 (m-11H, arom. H).

$^1$H NMR of compound **27** (δ ppm): 1.30-1.58 (t-3H, -CH$_2$-C**H**$_3$); 2.23-2.94 (m-13H, 8H of piperazine + C**H**$_2$γ + C**H**$_3$); 3.66-3.83 (distorted d-3H, C**H**$_2$α + O**H**); 3.89-4.06 (m-1H, C**H**β); 4.17 (s-1H, C**H**-Ph); 4.42-4.75 (q-2H, -C**H**$_2$-CH$_3$); 7.08-7.50 (m-11H, arom. H).

$^1$H NMR of compound **28** (δ ppm): 1.33-1.72 (m-6H, 6H of piperidine); 2.24-2.52 (m-6H, 4H of piperidine + C**H**$_2$γ); 2.62 (s-3H, C**H**$_3$); 3.62-4.03 (m-4H, C**H**$_2$α + C**H**β + O**H**); 4.12 (s-3H, OC**H**$_3$); 7.18 (s-1H, arom. H).

$^1$H NMR of compound **29** (δ ppm): 1.28-1.68 (m-9H, 6H of piperidine + -CH$_2$-C**H**$_3$); 2.20-2.68 (m-9H, 4H of piperidine + C**H**$_2$γ + C**H**$_3$); 3.59-4.11 (m-4H, C**H**$_2$α + C**H**β + O**H**); 4.42-4.79 (q-2H, -C**H**$_2$-CH$_3$); 7.15 (s-1H, arom. H).

$^1$H NMR of compound **30** (300 MHz, δ ppm): 1.19-1.63 (m-5H, Pip-4-**H** + Pip-3-C**H**$_2$ + Pip-5-C**H**$_2$); 1.85-2.23 (2 × t-1H, Pip-2-**H**$_{ax}$ + Pip-6-**H**$_{ax}$); 2.37 (distorted t-2H, C**H**$_2$γ); 2.48-2.51 (d-2H, C**H**$_2$-Ph); 2.61 (s-3H, C**H**$_3$); 2.78-2.94 (2 x d-1H, Pip-2-**H**$_{eq}$ + Pip-6-**H**$_{eq}$); 3.62-3.79 (m-3H, C**H**$_2$α + O**H**); 3.97-4.03 (m-1H, C**H**β); 4.12 (s-3H, OC**H**$_3$); 7.10-7.26 (m-6H, arom. H).

$^1$H NMR of compound **31** (300 MHz, δ ppm): 1.19-1.62 (m-8H, Pip-4-**H** + Pip-3-C**H**$_2$ + Pip-5-C**H**$_2$ + -CH$_2$-C**H**$_3$); 1.86-2.22 (2 × t-1H, Pip-2-**H**$_{ax}$ + Pip-6-**H**$_{ax}$); 2.34 (distorted t-2H, C**H**$_2$γ); 2.49-2.51 (d-2H, C**H**$_2$-Ph); 2.60 (s-3H, C**H**$_3$); 2.74-2.90 (2 × d-1H, Pip-2-**H**$_{eq}$ + Pip-6-**H**$_{eq}$); 3.61-3.79 (m-3H, C**H**$_2$α + O**H**); 3.94-4.01 (m-1H, C**H**β); 4.56-4.63 (q-2H, -C**H**$_2$-CH$_3$); 6.91-7.29 (m-6H, arom. H).

$^1$H NMR of compound **32** (δ ppm): 1.55-1.95 (m-4H, 4H of pyrrolidine); 2.22-2.83 (m-9H, 4H of pyrrolidine + C**H**$_2$γ + C**H**$_3$); 3.20 (broad s-1H, O**H**); 3.62-4.26 (m-6H, C**H**$_2$α + C**H**β + OC**H**$_3$); 7.15 (s-1H, arom. H).

$^1$H NMR of compound **33** (δ ppm): 1.27-1.93 (m-7H, -CH$_2$-C**H**$_3$ + 4H of pyrrolidine); 2.23-2.88 (m-9H, 4H of pyrrolidine + C**H**$_2$γ + C**H**$_3$); 3.23-4.20 (m-4H, C**H**$_2$α + C**H**β + O**H**); 4.39-4.76 (q-2H, -C**H**$_2$-CH$_3$); 7.15 (s-1H, arom. H).

$^1$H NMR of compound **34** (δ ppm): 2.24-2.79 (m-9H, 4H of morpholine + C**H**$_2$γ + C**H**$_3$); 3.19 (broad s-1H, O**H**); 3.54-3.82 (m-6H, 4H of morpholine + C**H**$_2$α); 3.85-4.22 (m-4H, C**H**β + OC**H**$_3$); 7.19 (s-1H, arom. H).

$^1$H NMR of compound **35** (δ ppm): 1.32-1.60 (t-3H, -CH$_2$-C**H**$_3$); 2.27-2.72 (m-9H, 4H of morpholine + C**H**$_2$γ + C**H**$_3$); 3.14 (broad s-1H, O**H**); 3.55-3.81 (m-6H, 4H of morpholine + C**H**$_2$α); 3.85-4.14 (m-1H, C**H**β); 4.42-4.75 (q-2H, -C**H**$_2$-CH$_3$); 7.15 (s-1H, arom. H).

**Pharmacology**

Pharmacological procedures were carried out in the same way as described in (3).

## RESULTS AND DISCUSSION

Acute toxicity

*C*ompounds **24**, **26**, **27**, **30** and **31** after intraperitoneal administration were not toxic ($LD_{50}$ > 2000 mg/kg). The most toxic compound was **34** ($LD_{50}$ = 599 mg/kg). All examined substances were less toxic than aspirin (ASA) and morphine after intraperitoneal administration ($LD_{50}$ for ASA = 167 mg/kg (10), for morphine = 140 mg/kg (11), respectively, for mice (Table 2)). Toxic doses of all tested compounds caused sedation and decrease of the locomotor activity.

Analgesic activity:

"Hot plate" test in mice

In this test seven compounds were active at a dose of 200 mg/kg and up to a dose of 50 mg/kg. The most active compound was **25** ($ED_{50}$ = 71.2 mg/kg) but this effect was twenty fold weaker than that of morphine (Table 2).

"Writhing syndrome" test in mice

All compounds tested showed analgesic activity in this test. The most potent effects were produced by compounds **30** and **31** ($ED_{50}$ = 10.3 mg/kg) (**30**) and ($ED_{50}$ = 12.9 mg/kg) (**31**). These compounds were stronger analgesic agents than ASA but their antinociceptive effects were weaker than those of morphine. The $ED_{50}$ values and therapeutic indexes for the compounds investigated are presented in Table 2.

Locomotor activity.

All compounds tested significantly suppressed the spontaneous locomotor activity of mice during a 30 min observation period. The most potent effect was produced by compound **30** ($ED_{50}$ = 12.03 mg/kg). The $ED_{50}$ values and therapeutic indexes for the compounds investigated are presented in Table 2.

From the data presented above it can be seen that the acute toxicity of the studied imides was depend on the structure of cyclic amine in the alkyl chain and sometimes on the kind of the substituent in the position 2 of the pyridine ring. This statement confirmed the data presented below:

In the case of piperazinyl derivatives containing benzhydryl substituent at the nitrogen atom two homologues **26** and **27** were not toxic. The lack of the toxicity was also observed in the case of methoxy derivative **24** with benzyl group at N-4 of the piperazine ring. The replacement of the methoxy group in **24** by ethoxy one (imide **25**) caused an increase of the toxic effects. Compounds **28**, **29** and **32**-**35,** in which N-substituted piperazine was replaced by the other cyclic amines, were characterized by higher or lower toxicity. The most toxic substance was imide **34** containing morpholino and methoxy groups. Its ethoxy homologue **35** displayed considerably lower toxicity. The opposite effect was observed in the case of the homologous compounds **28** and **29** possessing piperidine ring in the alkyl linker. Pyrrolidine derivatives **32** and **33** showed similar toxicity despite of the difference in the structure of alkoxy group. The introduction of benzyl substituent in the position 4 of the piperidine ring in compounds **28** and **29** (imides **30** and **31**) eliminated the toxic effects.

Furthermore, the last modification afforded the strongest analgesics (**30**, **31**) among the tested compounds in the "writhing syndrome" test. Debenzylation of **30** and **31** (imides **28** and **29**) caused considerable decrease of the analgesic activity. A replacement of piperidino group in **28** and **29** by pyrrolidinyl one (imides **32** and **33**) did not influence essentially antinociceptive properties in the "writhing syndrome" test. In the case of morpholine derivatives **34** and **35** only imide **34** with methoxy group was more active as analgesic agent than piperidine and pyrrolidine analogues **28** and **32**. The opposite effect was observed in the case of 2-ethoxy homologue **35** in relation to the imides **29** and **33**. Compounds with N-benzyl- and N-benzhydrylpiperazinyl moieties (**24-27**) were characterized also by the weak analgesic properties in the "writhing syndrome" test. In this group the most active substance was imide **27**, displaying considerable stronger analgesic effects than methoxy homologue **26**. Worth of notice is a fact that in this group of compounds ethoxy derivatives **25**, **27**, **29**, and **33** were more active as analgesic agents than methoxy ones **24**, **26**, **28** and **32** in this test.

In the "hot plate" test, the strongest antinociceptive effects were produced by imide **25**, substituted at N-4 of piperazine by benzyl group. Its methoxy homologue **24** was inactive in this test. Replacement of benzyl in **24** by benzhydryl group (imide **26**) caused an appearance of the weak analgesic activity. Introduction of ethoxy group instead of methoxy one in **26** (imide **27**) led to disappearance of the analgesic action. In the case of imides having other cyclic amines, the strongest antinociceptive effects were generated by compound **32** with pyrrolidinyl and methoxy groups. The ethoxy homologue of **32** (imide **33**) was inactive in this test. Similar relationship took place in the case of morpholine derivatives **34** and **35**. Compound **34** acted

considerably weaker than its analogue **32**. Between two imides **28** and **29** possessing piperidino group only ethoxy homologue **29** was characterized by a weak analgesic activity. Introduction of benzyl group in position 4 of piperidine ring in **28** and **29** (imides **30** and **31**) caused the appearance (**30**) or increase (**31**) of the antinociceptive action.

From the data presented in Table 2 it follows that imides **25**, **26**, **29-32** and **34** acted stronger than aspirin in the "hot plate" test.

Furthermore, all the tested compounds suppressed the spontaneous locomotor activity of mice. The most active substance in this test was imide **30** containing 4-benzylpiperidino and methoxy groups. An exchange of methoxy substituent by ethoxy one in **30** (imide **31**) caused a marked decrease of action in this test. Imide **31** and morpholine derivative **35** were the weakest inhibitors of the spontaneous locomotor activity of mice. Debenzylation of **30** and **31** (compounds **28**, **29**) weakened (**28**) or increased (**29**) the inhibited properties. The imides with piperidino group (**28** and **29**) acted in this test similarly to their pyrrolidinyl analogues **32** and **33**. The same situation was observed in the group of piperazinyl derivatives (**24-27**).

Previously synthesized imides **1**, **2** (1) possessed values of $ED_{50}$ = 0.4 mg/kg (**1**) and 1.4 mg/kg (**2**) in the "writhing syndrome" test . These data indicate that in this test all the compounds studied were considerably weaker analgesic agents than the above mentioned imides **1** and **2**.

The same was observed in the "hot plate" test in which 5 compounds were inactive. The others were much weaker analgesics than the parent compounds **1** and **2** ($ED_{50}$ for **1** = 11.9 mg/kg, for **2** = 17.6 mg/kg).

Compound **2** suppressed the spontaneous locomotor activity of mice ($ED_{50}$ = 107.9 mg/kg) while imide **1** was inactive in this test. All the newly synthesized substances were more active in this test than **2**.

Imides **24**, **26**, **27**, **30** and **31** had $LD_{50}$ >2000 mg/kg similarly to the "lead" compounds **1** and **2**. The other studied imides were more toxic than the parent substances **1** and **2**.

From these results it was concluded that compounds **30** and **31** displayed potent analgesic activity in the "writhing syndrome" test in mice and were non toxic. The therapeutic indexes of the analgesic action of these substances were larger than those of other anti-writhing active imides tested.

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