Synthesis and the diuretic activity of 8-aminosubstituted of 7-(2-hydroxy-3-p-metoxyphenoxypropyl-1)-3-methylxanthine

It has been found that natural xanthines, as well as their synthetic analogs, possess the diuretic effect. Analysis of the literature proves that there is a great opportunity of applying synthetic derivatives of N-methylated xanthines as potential diuretics.

Aim. To develop preparative methods of the synthesis of 8-aminosubstituted of 7-(2-hydroxy-3-p-metoxyphenoxypropyl-1)-3-methylxanthine and study their physical, chemical and biological properties.

Results. The synthesis of a series of 8-aminosubstituted of 7-(2-hydroxy-3-p-metoxyphenoxypropyl-1)-3-methylxanthines was carried out. According to the results of the biological testing the compounds synthesized belong to the toxicity of class IV. 7-(2-Hydroxy-3-p-methoxyphenoxypropyl-1)-8-(furyl-2-methylamino)-3-methylxanthine shows the highest diuretic activity, and hence, requires a more in-depth study since it is twice more active than hydrochlorothiazide. It should be emphasized that all compounds synthesized exhibit a marked diuretic effect.

Experimental part. 8-Bromo-7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methylxanthine was obtained by heating 8-bromo-3-methylxanthine with p-methoxyphenoxymethyloxirane in butanol-1 and in the presence of N,N-dimethylbenzylamine. 8-Aminosubstituted of 7-(2-hydroxy-3-p-metoxyphenoxypropyl-1)-3-methylxanthine was obtained by boiling of bromoalcohol with the primary and secondary amines. The structure of the compounds synthesized was unambiguously confirmed by NMR-spectroscopy. The acute toxicity of the compounds obtained was studied by Kerber method. The study of the diuretic activity of the compounds was carried out using Ye. Berkhin method. Hydrochlorothiazide was used as a reference substance.

Conclusions. Simple methods for the synthesis of 8-amino-7-(2-hydroxy-3-p-metoxyphenoxypropyl-1)-3-methylxanthines have been developed. The structure of the compounds synthesized has been confirmed by the method of NMR 1H-spectroscopy. The acute toxicity and the diuretic activity of the compounds obtained have been studied.

Key words: xanthine; organic synthesis; NMR-spectroscopy; acute toxicity; diuretic effect
To find original and non-toxic diuretics is an important task of modern pharmaceutical chemistry since diuretics are widely used in the comprehensive treatment of various cardiovascular diseases [1-3]. It has been found that natural xanthines (theophylline, theobromine, caffeine), as well as their synthetic analogs (euphyllin, etofylline), possess the diuretic effect [4]. Analysis of the literature proves that there is a great opportunity of applying synthetic derivatives of N-methylated xanthises as potential diuretics [5-7].

The aim of this work is to develop preparative methods of the synthesis of 3-methylxanthine derivatives not described earlier and study their physical, chemical and biological properties.

**Results and Discussion**

As the initial compound 8-bromo-7-(2-hydroxy-3-p-methoxyphenoxypropyl)-3-methylxanthine (2) was selected since its analogs revealed a high biological effect [7-12]. As shown in Scheme, the initial bromoalcohol 2 was obtained by heating 8-bromo-3-methylxanthine with p-methoxyphenoxymethylsulfoxide. Compounds 4, 5, 11, 13 are soluble in dioxane, dimethylformamide, and diethyl ether. Meanwhile, they are insoluble in water and diethyl ether. The 8-amino derivatives of 7-(2-hydroxy-3-p-methoxyphenoxypropyl)-3-methylxanthine (3-13) obtained are white crystalline compounds that are insoluble in water and diethyl ether. Meanwhile, they are soluble in dioxane, dimethylformamide, and dimethylsulfoxide. Compounds 4, 5, 11, 13 are soluble in diluted mineral acids. The structure of 8-aminoxanthines 3-13 synthesized was proven by NMR-spectroscopy data (Tab. 1). The spectra have clear proton signals, which are substituents of the appropriate form and intensity located in the relevant part of the spectrum.

According to the results of the biological testing the compounds synthesized belong to the toxicity of 2 of the N-propyl residue its methylene and methine protons in the NMR spectrum were recorded as two multiplets at 4.45-4.15 ppm (3H) and 3.88 ppm (2H). Aromatic protons of the p-methoxyphenoxypropyl residue were registered in the form of an intense singlet at 6.78 ppm (4H) indicating their magnetic equivalence.

The presence of the bromine atom in position 8 of the xanthine molecule allowed studying the reactions of bromoalcohol 2 with various amines. It was found that when heating compound 2 with the primary and secondary amines of the aliphatic or heterocyclic series, the reaction of the bromine atom substitution by the amine residue occurred under brief boiling of synthons in the aqueous dioxane medium along with formation of the corresponding 8-amino-substituted (3-5, 8-13) (Scheme). The reaction with aromatic amines in these conditions was not performed.

The synthesis of 8-m-tolylaminoxanthine (6) and p-ethoxyphenylaminoxanthine (7) was carried out by boiling synthons in the excess of amine without solvents. The 8-amino derivatives of 7-(2-hydroxy-3-p-methoxyphenoxypropyl)-3-methylxanthine (3-13) obtained are white crystalline compounds that are insoluble in water and diethyl ether. Meanwhile, they are soluble in dioxane, dimethylformamide, and dimethylsulfoxide. Compounds 4, 5, 11, 13 are soluble in diluted mineral acids. The structure of 8-aminoxanthines 3-13 synthesized was proven by NMR-spectroscopy data (Tab. 1). The spectra have clear proton signals, which are substituents of the appropriate form and intensity located in the relevant part of the spectrum.
class IV. Their LD$_{50}$ is in the range of 290-835 mg/kg. The most toxic ones are 8-n-butylaminoxanthine 3 (290 mg/kg) and 4-benzylpiperidinoxanthine 12 (302 mg/kg). Virtually, 8-(furyl-2-methylamino)xanthine 8 (835 mg/kg) is a non-toxic compound. It should be mentioned that compound 8 shows the highest diuretic activity, and hence, requires a more in-depth study since it is twice more active than hydrochlorothiazide (a reference substance). It should be emphasized that all compounds synthesized exhibit a marked diuretic effect. Apart from 8-furylmethylamino derivative 8, such compounds as butylaminoxanthine 3 (231.8 %), N,N-diethylaminoethylaminoxanthine 5 (199.3 %), m-tolylaminoxanthine 6 (243.5 %), p-ethoxyphenylaminoxanthine 7 (21.0 %) are even more active than hydrochlorothiazide. Aminoxanthines 9 (186.2 %) and 12 (185.2 %) containing benzyl residue in their structure are practically equivalent to hydrochlorothiazide.

The above-mentioned details clearly indicate a great opportunity and feasibility of further search for non-toxic diuretics among xanthine derivatives.

Table 1

The values of the chemical shift in NMR-spectra of the compounds synthesized (2-13)

| Compound | N$^1$H (s, 1H) | CH$_{aax}$ | C$^4$NH | OH (d, 1H) | N$^1$CH$_2$CHCH$_2$O | OCH$_3$ (s, 3H) | N$^1$CH$_2$ (s, 3H) | Other signals |
|----------|---------------|---------|--------|--------|------------------|--------------|----------------|-------------|
| 1        | 2             | 3       | 4      | 5      | 6                | 7            | 8              | 9           |
| 2        | 11.25         | 2-13.25 | 6.78   | 4.15   | 3.88 (m, 2H)     | 3.30         |                 |             |
| 3        | 10.14         | 6.79    | 4.10   | 3.40   | 3.85 (m, 2H)     | 3.35         |                 |             |
| 4        | 10.20         | 6.83    | 4.48   | 4.35   | 3.50             | 3.30         |                 |             |
Experimental Part

The melting point was determined using the open capillary method with PTP-M device. Elemental analysis was performed using an Elementar Vario cube device; NMR-spectra were taken on a Bruker SF-300 spectrometer (with the operating frequency of 300 MHz, DMSO as a solvent, and TMS as an internal standard). These data corresponded to the elemental analysis calculated.

Analytical data of the compounds synthesized are given in Tab. 1, 2.

The synthesis of 8-bromo-7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methylxanthine (2). Boil the mixture of 24.5 g (0.1 Mole) of 8-bromo-3-methylxanthine [13], 19.8 g (0.11 Mole) of p-methoxyphenoxyoxirane, 1 ml of N,N-dimethylbenzylamine in 150 ml of butanol-1 for 3 h. Then filter the mixture being hot, rinse in a cold dioxane and water and crystallize from the aqueous dioxane.

The synthesis of 8-n-butylamino-7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methylxanthine (3). Boil the mixture of 4.25 g (0.01 Mole) of bro-

| Compound | M. p., °C | The empirical formula | Yield, % |
|----------|-----------|-----------------------|---------|
| 2        | 190-192   | C_{16}H_{17}BrN_{4}O_{5} | 87.1     |
| 3        | 241-242   | C_{20}H_{27}N_{5}O_{5}  | 91.1     |
| 4        | 221-222   | C_{20}H_{28}N_{6}O_{5}  | 81.0     |
| 5        | 216-217   | C_{22}H_{32}N_{6}O_{5}  | 87.0     |
| 6        | 286-287   | C_{23}H_{25}N_{5}O_{5}  | 85.1     |
| 7        | 241-242   | C_{24}H_{27}N_{5}O_{6}  | 66.4     |
| 8        | 226-227   | C_{24}H_{27}N_{5}O_{6}  | 70.3     |
| 9        | 172-173   | C_{24}H_{27}N_{5}O_{5}  | 94.6     |
| 10       | 192-193   | C_{20}H_{25}N_{5}O_{5}  | 89.1     |
| 11       | 200-201   | C_{21}H_{28}N_{6}O_{5}  | 93.2     |
| 12       | 209-210   | C_{24}H_{27}N_{5}O_{6}  | 84.6     |
| 13       | 183-184   | C_{22}H_{28}N_{6}O_{5}  | 69.6     |
moxanthine, 2, 3 ml (0.03 Mole) of n-butylamine, 30 ml of water with 30 ml of dioxane for 1 h. After that, cool the mixture, and add 100 ml of water. Filter the precipitate, rinse in water and crystallize from the aqueous dioxane.

Compounds 4, 5, 8-13 were obtained in a similar way. Compounds 4, 5, 11, 13 were purified using the reprecipitation method. Compounds 8-10, 12 were crystallized from the aqueous dioxane.

The synthesis of 7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methyl-8-m-tolylaminoxanthine (6).

Boil the solution of 4.25 g (0.01 Mole) of bromoxanthine 2 in 15 ml of m-toluidine for 1 h. Then cool it, and add 150 ml of propanol-2. Filter the precipitate, rinse in propanol-2, acetone and water and then crystallize from the aqueous DMF.

Compound 7 was obtained in a similar way.

The acute toxicity of the compounds synthesized was studied by Kerber method [8] in white mice weighing 18-24 g.

The study of the diuretic activity of the compounds was carried out using Ye. Berkhin method [9]. The compounds studied were injected intraperitoneally in the dose of 1/20 LD50 as a 3-5 % thin aqueous suspension stabilized by Tween-80 30 min prior to the water load. Hydrochlorothiazide in the dose of 25 mg/kg was used as a reference substance.

Data of the biological effects of the compounds synthesized are shown in Tab. 3.

**Conclusions**

1. Simple methods for the synthesis of 8-amino-7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methylxanthines have been developed.
2. The structure of 7,8-disubstituted derivatives of 3-methylxanthine synthesized has been confirmed by the method of NMR 1H-spectroscopy.
3. The acute toxicity and the diuretic activity of the compounds obtained have been studied. For more in-depth pharmacological studies 7-(2-hydroxy-3-p-methoxyphenoxypropyl-1)-3-methyl-(furyl-2)-methylaminoxanthine (8) has been proposed, it increases diuresis by 3 times and is twice more active than hydrochlorothiazide.

**Conflicts of Interest:** authors have no conflict of interest to declare.

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