SYNTHESIS OF N-SUBSTITUTED DERIVATIVES OF 1-(4-METHOXYPHENYL)-1,5-DIHYDRO-4*H*-PYRAZOLO[3,4-D]PYRIMIDIN-4-ONE AS POTENTIAL ANTICONVULSANTS

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ABSTRACT

The high psychotropic activity of pyrimidine derivatives attracts attention and leads to the creation of new pyrimidine drugs which affect the central nervous system. As psychotropic agents, special attention deserve azolopyrimidine derivatives, including pyrazolopyrimidines.

Thus, among the pyrazolopyrimidine derivatives, compounds with antiepileptic, anticonvulsant, sedative, anxiolytic activity, ligands of benzodiazepine site of GABA receptors have been found. In addition, the ligands of 5HT-6 receptors were identified that are promising for the treatment of central nervous system, muscle relaxants.

The purpose of this research was a synthesis of alkylated derivatives of 1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidine-4-ones.

Keywords: *synthesis; pyrimidine; pyrazole; acetamides, activity prediction*

The high psychotropic activity of pyrimidine derivatives attracts attention and leads to the creation of new pyrimidine drugs which affect the central nervous system. As psychotropic agents, a special attention deserve azolopyrimidine derivatives, including pyrazolopyrimidines. Thus, the compounds with antiepileptic, anticonvulsant, sedative, anxiolytic activity (1,2), ligands of benzodiazepine site of GABA receptors (3) were found among the pyrazolopyrimidine derivatives.

This research aimed at synthesizing the N-substituted derivatives of 1-(4-methoxyphenyl)-1,5-dihydro-4***H***-pyrazolo[3,4-d]pyrimidin-4-one.

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Received: November 23, 2015
Accepted: February 5, 2016

The choice of substituent in the first position of pyrazolopyrimidine system was conditioned by the proved influence of the 4-metoxyphenyl radical in affecting the anticonvulsant activity of substances (4,5,6).

To establish the prospects of synthesis and an optimization of further pharmacological screening, we performed a prediction of the biological activity that was planned for the synthesis of compounds using a PASS computer program. N-Substituted derivatives of 1-(4-methoxyphenyl)-1,5-dihydro-4***H***-pyrazolo[3,4-d]pyrimidine-4-one were selected for a synthesis. Marked psychotropic activities such as antiepileptic, anxiolytic, antidepressant, antineurotic, and convulsant activities (Ra≥0.50) were predicted for these compounds.

The initial compound – 1-(4-methoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d]pyrimidine-4-one **4** was obtained within two stages. The first stage included the synthesis of intermediate – ethyl 5-amino-1-(4-methoxyphenyl)-1*H*-pyrazole-4-carboxylate **3** by an interaction with the

4-methoxyphenylhydrazine hydrochloride **1** and ethyletoxymethylencyanocetate **2** in isopropyl alcohol medium in the presence of triethylamine. During the second stage of the research, the resulting carboxylate **3** was heated in an excess of formamide for two days. As a result of this reaction, a target 1-(4-methoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d]pyrimidine-4-one **4** was obtained (Fig. 1).

Synthesis of N-substituted derivatives of 1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidin-4-one was carried out by an interaction of pyrazolo[3,4-d]pyrimidine-4-one **4** with N-arylsubstituted α-chloroacetamides, 2-chloro-1-(4-arylpiperazine-1-yl)-ethanones and 2-chloro-N-(4-chlorobenzyl)acetamide (Fig. 2). The reaction was performed maintaining the mixture of reagents for two hours at 70°C in dimethylformamide medium in the presence of $NaHCO_3$.

The presence of several reaction centers in the molecule of pyrazolopyrimidine **4** made it possible to achieve several directions of the reaction: substitution can occur at the nitrogen atom in position 5 (route a) as well as at the oxygen atom in position 4 (route b) (Fig. 2).

Such reactivity of pyrazolopyrimidine was predetermined by its tendency to tautomeric transformations. We also took into account the difficulty of regioselectively introducing the aforementioned radicals in the structure of 1-(4-methoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d]pyrimidin-4-one **4** and determining the position of substituents due to the

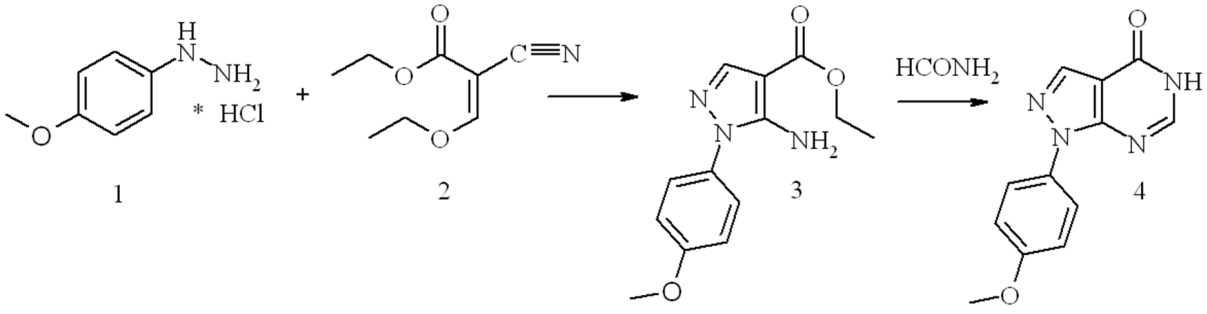

Figure 1. Synthesis of 1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidine-4-one **4**

Table 1. Data of 1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidine-4-one derivatives obtained 4, 5.1-12

Compound	Yield, %	M.p., °C	Calculated N, %	Found N, %	Mol. formula	[MH⁺]
4	89	180-182	23.13	23.22	$C_{12}H_{10}N_4O_2$	–
5.1	85	265-287	18.66	18.78	$C_{20}H_{17}N_5O_3$	376
5.2	88	280-282	17.27	17.43	$C_{21}H_{19}N_5O_4$	–
5.3	84	243-245	16.08	16.23	$C_{22}H_{21}N_5O_5$	–
5.4	85	270-272	17.09	17.24	$C_{20}H_{16}ClN_5O_3$	410
5.5	88	268-270	17.27	17.39	$C_{21}H_{19}N_5O_4$	–
5.6	86	285-287	17.98	18.02	$C_{21}H_{19}N_5O_3$	390
5.7	84	205-207	17.98	18.04	$C_{21}H_{19}N_5O_3$	–
5.8	81	250-252	16.52	16.66	$C_{21}H_{18}ClN_5O_3$	–
5.9	82	265-257	16.16	16.31	$C_{22}H_{19}N_5O_5$	–
5.10	80	255-257	16.70	16.79	$C_{21}H_{17}N_5O_5$	–
5.11	84	230-232	18.91	19.02	$C_{24}H_{24}N_6O_3$	444
5.12	84	245-247	17.71	17.98	$C_{25}H_{26}N_6O_4$	–

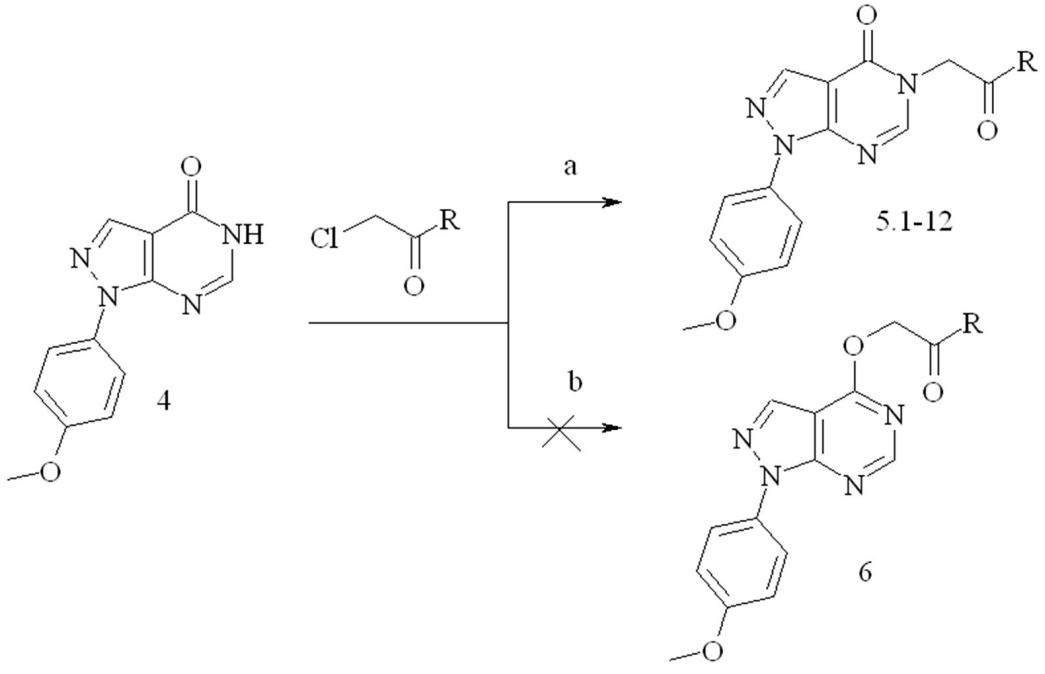

Figure 2. Synthesis of N-substituted derivatives of 1-(4-methoxyphenyl)-1,5-dihydro--pyrazolo[3,4-d]pyrimidin-4-one **5**

probability of formation of a mixture of N- and O-substituted products.

Due to chromato-mass spectrometry data the synthesized products were individual substances (Table 1) which were obtained with the satisfactory yields. After the crystallization from isopropyl alcohol, the synthesized compounds became white or light yellow crystalline substances with well-defined melting points.

Comparing the ^{1}H NMR spectra of the synthesized compounds **5** with the spectra of initial 1-(4-methoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d]pyrimidine-4-one **4**, the absence of imine signal proton of pyrimidine cycle was noted but this proton disappeared in both of the proposed structures. Furthermore, a singlet of NH-group of amide residue at δ 12.46 ppm (5.1-5.8) and additional aryl protons caused the multiplicity and intensity which corresponded to the nature and location of the substituents. The spectra of compounds 5.10-12 were characterized by two multiplets at 3.10-3.73 ppm corresponding to protons of piperazine cycle. The spectrum of compounds 5.8 was characterized by a doublet at 4.30 ppm corresponding to protons of the methylene group in a benzyl radical.

The position of the proton signals of methylene groups in the alkyl radical at 4.62-4.92 ppm showed the reaction in the direction A (Fig. 2). Since the formation of the type **6** compounds, the signals shifted to 0.2-0.4 ppm. For a more reliable verification of N-substituted derivatives formation, we used the NOESY spectroscopy data following the example of a compound 5.1. NOESY spectrum which was characterized by cross-peak CH_2 of the protons of acetamide residue and CH proton at position 6 of pyrazolo[3,4-d]pyrimidine system. This clearly indicated the formation of an N-substituted derivative and, therefore, confirmed the formation of the compounds of type **5**.

Experimental Part

All of the solvents and reagents were obtained from commercial sources. The melting points (°C) were measured with a Kofler melting point apparatus and were not corrected. 1H NMR spectra were recorded on a Varian Mercury (200 MHz) spectrometer in DMSO-d6 using TMS as an internal standard (chemical shifts are in ppm). NOESY spectra were recorded on a Varian Gemini (300 MHz) spectrometer in DMSO-d6 using TMS as an internal standard (chemical shifts are in ppm). LC/MS was recorded

Table 2. ^{1}H NMR spectra of the synthesized compounds 4, 5.1-12

Compound	CONH 1H, s	CH-3 1H, s	CH-5 1H, s	Ar-H	CH_2CO, 2H, s	OCH_3 1H, s	The signals of protons of other functional groups
4	-	8.30	8.12	7.95, 2H, d 7.12, 2H, d	-	3.75	12.43, 1H, s, NH
5.1	10.45	8.32	8.25	7.90, 2H, d 7.55, 2H, d 7.27, 2H, t 7.15-7.01, 3H, m	4.85	3.75	-
5.2	10.31	8.41	8.31	7.88, 2H, d 7.43, 2H, d 7.17, 2H, d 6.95, 2H, d	4.80	3.80 3.67	-
5.3	10.02	8.42	8.30	7.88, 2H, d 7.71, 1H, d 7.10, 2H, d 6.61-6.40, 2H, m	4.92	3.92 3.80 3.72	-
5.4	10.54	8.44	8.30	7.90, 2H, d 7.61, 2H, d 7.40, 2H, d 7.09, 2H, d	4.87	3.80	-
5.5	9.80	8.45	8.30	8.00-7.80, 3H, m 7.28-7.00, 4H, m 6.96-6.80, 1H, m	4.94	3.82 3.70	-
5.6	10.32	8.41	8.30	7.89, 2H, d 7.47, 2H, d 7.10-6.98, 4H, m	4.89	3.80	2.22, 3H, s, CH_3
5.7	-	8.41	8.31	7.62-7.32, 5H, m 7.09, 2H, m	4.62	3.75	2.42, 3H, s, CH_3
5.8	9.85	8.42	8.30	7.85, 2H, m 7.42, 2H, m 7.30, 2H, m 7.05, 2H, m	4.75	3.75	4.30, 2H, d CH_2Bn
5.9	10.22	8.43	8.30	7.88, 2H, d 7.22-6.90, 4H, m 6.78, 1H, d	4.81	3.75	4.22, 4H, s, $2CH_2$
5.10	10.32	8.45	8.31	7.88, 2H, d 7.30-7.09, 3H, m 7.00-6.81, 2H, m	4.85	3.75	5.95, 2H, s, CH_2

5.11	-	8.40	8.31	7.85, 2H, d 7.230-7.20, 2H, m 7.10, 1H, d 6.99, 2H, d 6.80, 1H, t	4.92	3.80	3.73-3.58, 4H, m, $2CH_2$; 3.26-3.10, 4H, m, $2CH_2$;
5.12	-	8.42	8.31	6.82, 2H, d 7.00, 2H, d 7.75, 2H, d 8.01, 2H, d	4.89		3.73-3.58, 4H, m, $2CH_2$; 3.26-3.00, 4H, m, $2CH_2$;

with PE SCIEX API 150EX chromatograph equipped with a mass spectrometer.

1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidine-4-one 4

The mixture of 0.1 mol (17.46g) of 4-methoxyphenylhydrazine hydrochloride 1 and 0.1 mol (15.73 ml) of ethylethoxymetylencyanocetate 2 was heated in isopropyl alcohol with triethylamine (1.1 mol, 15.28 ml) at 60°C for four hours. The reaction mixture was cooled at room temperature; the isopropyl alcohol was evaporated under the vacuum, and the residue was diluted with 200 ml of water. Then the formed precipitate of ethyl 5-amino-1-(4-methoxyphenyl)-1*H*-pyrazole-4-carboxylate 3 was filtered, rinsed with water, and dried. 0.1 mol of the obtained carboxylate 3 was heated in 200 ml of formamide at 120˚C for 48 hours. The reaction mixture was cooled at room temperature, the precipitate was isolated, diluted with isopropyl alcohol, filtered, and dried at 60-70˚C for 12 hours.

General procedure of the synthesis of N-substituted derivatives of 1-(4-methoxyphenyl)-1,5-dihydro-4H-pyrazolo[3,4-d]pyrimidin-4-one 5.

0.0015 mol (0.12 g) of sodium bicarbonate and 0.001 mol of appropriate alkyl halide were added to a solution of 0.001 mol (0.24 g) of 1-(4-methoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[-3,4-d] pyrimidine-4-one **4** in 10 ml of dimethylformamide . Then this was heated for 5 hours at 70˚C. The reaction mixture was cooled at room temperature, the precipitate was isolated, diluted by isopropyl alcohol, filtered, and dried. It became crystallized from isopropanol.

CONCLUSIONS

Using the PASS program, a synthesis of potential anticonvulsants of derivatives of pyrazolo[3,4-d]pyrimidine was planned. The synthesis of 1-(4-metoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d]pyrimidine-4-one and their alkylation was carried out. It was found that the reaction of 1-(4-metoxyphenyl)-1,5-dihydro-4*H*-pyrazolo[3,4-d] pyrimidine-4-one with α-chloroacetamides, 2-chloro-1-(4-arylpiperazine-1-yl)-ethanones and 2-chloro-N-(4-chlorobenzyl)acetamide in DMFA conditions - $NaHCO_3$ was selectively performed at position 5 of the pyrazolopyrimidine cycle to form N-substituted derivatives.

REFERENCES

1. Gavrin L. K., A. Lee B. A., Provencher W. W. Et al. Synthesis of pyrazolo[1,5-α]pyrimidinone regioisomers // J. Org. Chem. – 2007. – Vol. 72. – pp. 1043–1046.
2. Dalinger I. L., Vatsadse I. A., Shevelev S. A. Liquid-phase synthesis of combinatorial libraries based on trifluoromethyl-substituted pyrazolo[1,5-α]pyrimidine scaffold // J. Comb. Chem. –2005. – Vol. 7. – pp. 236–245.
3. Lager E., Andersson J., Nilsson M. High-affinity ligands at the benzodiazepine site of brain GABA receptors. Synthesis, pharmacology, and pharmacophore modeling // J. Med. Chem. – 2006. – Vol. 49. – pp. 2526–2533.
4. Tasso S. M., Bruno-Blanch L.E., Moon S.C., Estiu G.L. Pharmacophore searching and QSAR analysis in the design of anticonvulsant drugs // J. Mol. Struct. – 2000. – 504 (1-3). – P. 229-240.
5. Malawska B. New anticonvulsant agents // Current topics in medicinal chemistry. – 2005. – № 5. – pp. 69-85.
6. Poroikov V.V. Med. Chem. Res., 2010, Vol. 19 (S1). – p. 30.