Synthesis and transformation in the series of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids

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The aim of the work is to develop preparative methods for the synthesis of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids, to study the esterification reaction in this regard, to study physical and chemical properties of the obtained substances, and to predict their toxicity.

Materials and methods. Compounds were synthesized using reagents and solvents qualified as “ch.p.”. The IUPAC nomenclature as supplemented was used during the preparation. The melting temperature was determined with the capillary method according to HFC (2.2.14) on the device PTP (M). Elemental analysis was determined with the ELEMENTAR vario EL cube analyzer (manufactured in Germany) (standard – sulfonamide). IR spectra were recorded using spectrophotometer Specord M-80 (manufactured in Germany) within the range of 4000–500 cm⁻¹ (scanning was performed under the following conditions: slit program 3.0, time constant – τ = 3 s, scanning time 34 min, samples were analyzed in the form of tablets with potassium bromide). 1H NMR spectra were recorded using Varian VXR-300 spectrophotometer (manufactured in the USA), dimethyl sulfoxide-D₆ solvent, and tetramethysilane was used as an internal standard. The spectra were decoded using the computer program ADVASP 1.43. Thin layer chromatography was performed using Sorbfil plates (analytical, size 10 × 15 cm, base: polymer substrate, sorbent: silica gel STX-1A, grain: 5–17 μm, layer thickness: 110 μm combination – silicazole).

Results. The synthesis of new 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids was carried out. These products became a basis for synthesis of a number of relevant esters. Physical and chemical properties were investigated for the synthesized compounds. The structure of the obtained substances was confirmed by elemental analysis, IR-spectroscopy, 1H NMR-spectrometry, and their individuality was established by thin-layer chromatography. Computer GUSAR-online prediction of acute toxicity of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids and their esters was performed.

Conclusions. Preparative methods for the synthesis of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids have been developed, for which esterification reactions have been studied. Thus, physical and chemical properties of the received substances were investigated, and indicators of their toxicity were predicted.

Key words: 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids, esters, toxicity indicators.

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Almost every year, unknown diseases, that require specific treatment, appear; unfortunately, existing drugs are not effective for some of them. Therefore, today the creation of affordable domestically manufactured drugs with a wide range of pharmacological activity remains an urgent task of pharmacy.

Analysis of the modern literature [1–5] indicates the prospects for the search for biologically active substances among 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio) acetic acids and their esters. Analysis of the work of the scientific school of ZSMU, namely the dissertation [1] and articles [2–5], demonstrates that compounds containing 2-, 3-, 4-methoxy and 3,4,5-trimethoxyphenyl substituents are highly active antimicrobial, antifungal agents and can be the basis for the creation of promising drugs [2–4]. The pharmacological activity of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids and their esters has not been sufficiently studied. Therefore, the synthesis, study of physicochemical and biological properties of 1,2,4-triazole-3-thioacetic acids containing 2,4- and 3,4-dimethoxyphenyl substituents, as well as acid esters, from our point of view have scientific novelty and theoretical and practical significance.

Aim

The purpose of the work is to develop preparative methods for the synthesis of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids, to study the esterification reaction in this regard, to study physical and chemical properties of the obtained substances, and to predict their toxicity.

Materials and methods

In the course of practical part of the research, 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids were synthesized (compounds 2.5, 2.8, Fig. 1). This class of compounds was obtained in two ways. In the first case, the production of acids was carried out by the reaction...
between 5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-thions (compounds 2.1, 2.2, 2.3, 2.4, Fig. 1) and monochloroacetic acid in the alkaline environment. As a solvent, a mixture of water and dimethylformamide in a ratio of 1 : 1 was used.

The second method was based on the interaction of (5- (2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetonitrile (compounds 2.3, 2.4, Fig. 1) with hydrochloric acid; water was used as a solvent.

Esters of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids were synthesized in order to reduce the acidic properties and prolong the biological action.

It is known that esters of 2-(1,2,4-triazole-3-thio)acetic acids demonstrate diuretic, neuroleptic, anti-inflammatory, moderate antimicrobial and other types of biological activity [3–5]. Moreover, the strength of these compounds action is influenced by both the substituents of 1,2,4-triazole nucleus, and the structure of a complex ester group.

Acid esters have high biological activity and can also be intermediates for the synthesis of amides, hydrazides, ylide hydrazides, and bicyclic structures – derivatives of 1,2,4-triazole.

Esters of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.6, 2.7, 2.9, 2.10, Fig. 2) were obtained by a method involving the interaction of the corresponding 5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-thione (compounds 2.1, 2.2, Fig. 2) with methyl or ethyl chloroacetic acid ester in the presence of an equimolecular amount of alkali.

In order to achieve better yields of products and their higher purity, another method was used to obtain esters of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.6, 2.7, 2.9, 2.10, Fig. 2), which involves the esterification of the above mentioned acids (compounds 2.5, 2.8) with methyl and ethyl chloroacetic acid in the presence of a catalytic amount of concentrated sulfuric acid. After the reaction, the excess alcohol was evaporated, the residue was first thoroughly washed with sodium bicarbonate solution (to pH 7–8), then with water (to pH 7), the precipitate was filtered off, further washed with water and dried.

Samples of esters of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.6, 2.7, 2.9, 2.10, Fig. 2) were obtained by two methods that did not result in melting point depression.

After recrystallization, the individuation of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.5, 2.8) and their esters (compounds 2.6, 2.7, 2.9, 2.10) was confirmed by thin layer chromatography. The acetone : hexane : water was used as the mobile medium.

1H NMR-spectra of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.5, 2.8) and their esters (compounds 2.6, 2.7, 2.9, 2.10) were characterized by the presence of multiple signals of aromatic protons at 6.64–7.83 ppm, doublet signals of protons of methoxy groups at 3.65–3.90 ppm, singlet signals of the thiomethylene group have also been recorded at 3.32–3.43 ppm. In acetic acids, characteristic singlet signals of carboxyl groups were present at 2.12–2.34 ppm, and in esters of acetic acids there were signals of protons of the methyl group of the alcohol residue at 1.23–3.87 ppm.

The individuality of the synthesized compounds was proved by thin layer chromatography. The acetone : hexane : propanol 2 : 1 : 1 system was used as the mobile medium. 

Data on the percentage of elements (C, H, N, S) in the samples of the obtained compounds. The IR spectra of acids (compounds 2.5, 2.8) were additionally characterized by the absorption bands of CO-C groups in the range of 1283–1227 cm⁻¹ [6]. The IR spectra of esters (compounds 2.6, 2.7, 2.9, 2.10) were obtained by a method involving the interaction of (5- (2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetonitrile (compounds 2.3, 2.4, Fig. 1) with hydrochloric acid; water was used as a solvent.
2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acid (compounds 2.5, 2.8)

Method A. In a glass of 250 ml 1 mole of the corresponding acetonitrile (2.3, 2.4) nitrile and 65 ml of hydrochloric acid were put, dissolved and left at room temperature for 5 days. Then 200 ml of water were added, precipitated, and the whole mixture was filtered off and dried.

Method B. 1 mole of the corresponding 5-(2,4- or 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-thione (compound 2.1), dissolved in 20 ml of dimethylformamide, was put into a 250 ml round bottom flask, equipped with a reflux condenser. 1 mole of NaOH (pre-dissolved in 20 ml of water) and 1 mole of monochloroacetic acid were added. It was heated to the pH of the solution in a slightly acidic medium, after which the resulting solution was evaporated.

As a result of the reactions, the following compounds were obtained: yellow (2.5) and orange (2.8) crystalline substances insoluble in water, soluble in solutions of alkalins and alkali metal carbonates, as well as in organic solvents and solutions of mineral acids. For analysis, substances were recrystallized from a mixture of dimethylformamide-water 2:1.

2-((5-(2,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acid (2.5). Yield 82 %, m.p. = 143–145 °C. Adsorption maxima in IR-spectra \( \nu_C=1598 \text{ cm}^{-1} \). H NMR (400 MHz, DMSO-d6) \( \delta 3.38 \text{ (2H, s, S-CH}_3 \text{); 3.84–3.90} \text{ (6H, d, O-CH}_2 \text{); 4.2} \text{ (1H, s, CH); 6.67–7.83} \text{ (3H, m, C}_6 \text{H}_5 \text{); 12.34} \text{ (1H, s, COOH).Calcd for C}_{12}H_{17}N_{2}O_{5}S %: C, 48.81; H, 4.44; N, 14.23; S, 10.86. Found %: C, 48.82; H, 4.45; N, 14.21; S, 10.86.

2-((5-(3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acid (2.8). Yield 83 %, m.p. = 82–84 °C. Adsorption maxima in IR-spectra \( \nu_C=1602 \text{ cm}^{-1} \). H NMR (400 MHz, DMSO-d6) \( \delta 3.38 \text{ (2H, s, S-CH}_3 \text{); 3.65–3.83} \text{ (6H, d, O-CH}_2 \text{); 4.22} \text{ (1H, s, CH); 6.98–7.49} \text{ (3H, m, C}_6 \text{H}_5 \text{); 12.32} \text{ (1H, s, COOH).Calcd for C}_{12}H_{17}N_{2}O_{5}S %: C, 48.81; H, 4.44; N, 14.23; S, 10.86. Found %: C, 48.82; H, 4.45; N, 14.21; S, 10.86.

Esters of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.6, 2.7, 2.9, 2.10)

Method A. To a solution of 0.01 mole of sodium hydroxide in 5 ml of water, 0.01 mole of the corresponding 5-R-1,2,4-triazole-3-thione (2.1, 2.2) in 50 ml of ethanol and 0.01 mole of monochloroacetic acid methyl ester were added. The mixture was boiled for 5 hours, the solvent was evaporated, the residue was washed with distilled water, and crystallized from a mixture of ethanol-water 3:1.

Method B. A mixture of 0.01 mole of the corresponding 2-((5-(2,4- or 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acid (compound 2.5, 2.8), 30 ml of alcohol (methanol, ethanol) and 0.5 ml of concentrated sulfuric acid was boiled for 10 hours, the solvent was evaporated, the residue was neutralized with sodium bicarbonate solution, which resulted in obtaining compounds 2.6, 2.7, 2.9, 2.10. White crystalline substances were insoluble in solutions of alkalis and alkali metal carbonates, sparingly soluble in water, and soluble in organic solvents. For analysis, it was purified by recrystallization from ethanol-water 3:1.

Ethyl 2-((5-(2,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetate (2.7). Yield 87 %, m.p. = 129–131 °C. Adsorption maxima in IR-spectra \( \nu_C=1598 \text{ cm}^{-1} \). H NMR (400 MHz, DMSO-d6) \( \delta 1.25 \text{ (3H, s, CH}_3 \text{); 3.40} \text{ (2H, s, S-CH}_3 \text{); 3.87} \text{ (6H, d, O-CH}_2 \text{); 4.23} \text{ (1H, s, CH); 6.67–7.77} \text{ (3H, m, C}_6 \text{H}_5 \text{). Calcd for C}_{17}H_{23}O_7S %: C, 52.00; H, 5.30; N, 12.99; S, 9.91. Found %: C, 51.99; H, 5.28; N, 13.00; S, 9.90.

Ethyl 2-((5-(3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetate (2.9). Yield 78 %, m.p. = 120–122 °C. Adsorption maxima in IR-spectra \( \nu_C=1563 \text{ cm}^{-1} \). H NMR (400 MHz, DMSO-d6) \( \delta 1.25 \text{ (3H, s, CH}_3 \text{); 3.37} \text{ (2H, s, S-CH}_3 \text{); 3.84} \text{ (6H, d, O-CH}_2 \text{); 4.31} \text{ (1H, s, CH); 6.69–7.81} \text{ (3H, m, C}_6 \text{H}_5 \text{). Calcd for C}_{17}H_{23}O_7S %: C, 50.48; H, 4.89; N, 13.58; S, 10.36. Found %: C, 50.50; H, 4.90; N, 13.50; S, 9.90.

In order to exclude potentially toxic substances as unsurprising objects of experimental pharmacological screening at the stage preceding the synthetic part, the prediction of acute toxicity of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.5, 2.8) and their esters (compounds 2.6, 2.7, 2.9, 2.10) was performed using the GUSAR-online program. Computer prediction of acute toxicity of synthesized compounds was performed according to the structural formulas of the compounds in the online version of the program GUSAR-online [7].

Results

As predicted by GUSAR-online for tested 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids (compounds 2.5, 2.8) and their esters (compounds 2.6, 2.7, 2.9, 2.10), the average lethal dose of LD50 for the corresponding acetic acids was when administered: intraperitoneally – from 455.4 to 480.1 mg/kg, intravenously – from
315.1 to 340.5 mg/kg, orally – from 578.8 to 1235.0 mg/kg and subcutaneously – from 1043.0 to 1150.0 mg/kg. The average lethal dose of LD$_\text{50}$ for the corresponding esters of acetic acid was when administered: intraperitoneally – from 801.9 to 866.1 mg/kg, intravenously – from 246.6 to 351.0 mg/kg, orally – from 955.3 to 1457.0 mg/kg and subcutaneously – from 1121.0 to 2015.0 mg/kg.

According to the results of the prediction of the toxicity index, it should be noted that all compounds belong to low-toxic and practically non-toxic substances, which corresponds to the 4$\text{th}$ and 5$\text{th}$ toxicity class according to the classification of K. K. Sidorov and according to the OECD classification [8].

### Discussion

New 2-((5-(2,4- and 3,4-dimethoxyphenyl)-1,2,4-triazole-3-yl)thio)acetic acids were synthesized, becoming the basis for the production of a number of suitable esters. The structure of the obtained substances was confirmed by elemental analysis, the obtained results confirm the data on the percentage of elements (C, H, N, S) in the samples of the obtained compounds, IR-spectroscopy, in the IR-spectra of acids (compounds 2.5, 2.8) CH$_\text{2}$, absorption bands -COOH groups at 1760 cm$^{-1}$ [6]. Besides, the IR spectra of esters (compounds 2.6, 2.7, 2.9, 2.10) were additionally characterized by absorption bands of CO-C groups in the range of 1283–1227 cm$^{-1}$ [6] and $^1$H NMR-spectrometry. In acetic acids, characteristic singlet signals of carboxyl groups were present at 12.32–12.34 ppm, and in esters of acetic acids there were signals of protons of the methyl group of the alcohol residue at 1.23–3.87 ppm, and their individuality was established by thin layer chromatography. According to the results of computer GUSAR-online prediction of toxicity indicators, it should be noted that all compounds were low-toxic and virtually non-toxic substances, which corresponds to 4 and 5 toxicity class according to the classification of K. K. Sidorov and the OECD classification [8].

### Conclusions

1. Preparative methods have been developed and the synthesis of 2-((5-(2,4- and 3,4-dimethoxyphenyl)-3H-1,2,4-triazole-3-yl)thio)acetic acids and their esters has been carried out.

2. The structure of the obtained compounds was confirmed by elemental analysis of IR-spectroscopy, $^1$H NMR-spectra, and their individuality was justified by thin layer chromatography.

3. Computerized prediction of acute toxicity was performed for the synthesized compounds, which showed the possibility of searching for potential drugs based on them.

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### Conflicts of interest

authors have no conflict of interest to declare.