Regulatory Mechanisms in Biosystems

ISSN 2519-8521 (Print)
ISSN 2520-2588 (Online)
Regul. Mech. Biosyst.

Study of the antimicrobial activity of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid

V. I. Kushnir*, V. V. Stybel*, I. M. Kushnir*, L. V. Fizer**, V. I. Lubenets**, I. Y. Mazur*, B. V. Gutyj***, I. S. Semen*, U. Z. Berbeka*

**State Scientific-Research Control Institute of Veterinary Medicinal Products and Feed Additives, Lviv, Ukraine*
***Lviv Polytechnic National University, Lviv, Ukraine*
****Stepan Gzhytskyi National University of Veterinary Medicine and Biotechnologies, Lviv, Ukraine*

Article info

Received 04.05.2025
Received in revised form 03.06.2025
Accepted 27.06.2025

State Scientific-Research Control Institute of Veterinary Medicinal Products and Feed Additives, Donetska st., 11, Lviv, 79019, Ukraine. Tel.:+38-098-966-30-20. E-mail: wolodjak@gmail.com

Lviv Polytechnic National University, Bandera st., 12, Lviv, 79013, Ukraine. E-mail: liubov.v.fizer@lpnu.ua

Stepan Gzhytskyj Lviv National University of Veterinary Medicine and Biotechnologies, Pekarska st., 50, Lviv, 79000, Ukraine. Tel.:+38-068-136-20-54. E-mail: bvh@ukr.net

Kushnir, V. I., Stybel, V. V., Kushnir, I. M., Fizer, L. V., Lubenets, V. I., Mazur, I. Y., Gutyj, B. V., Semen, I. S., Berbeka, U. Z. (2025). Study of the antimicrobial activity of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid. Regulatory Mechanisms in Biosystems, 16(3), e25102. doi:10.15421/0225102

S-alkyl esters of thiosulfonic acids (R-S(O)2S-R') are structural analogs of natural sulfur-containing substances and have a broad spectrum of biological activity. The results of studying the sensitivity of microorganisms to ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid obtained using new "green" methods, particularly environmentally safe reactions under the influence of ultrasound, are presented. It was established that the studied substance's antimicrobial activity changed with a change in the medium's pH. In particular, with a decrease in the pH of the medium from 7.0 to 5.2 units, the minimum inhibitory concentration of the studied substance also decreased. Thus, the minimum inhibitory concentrations of ethyl ester of thiosulfonic acid against *Staphylococcus aureus* ATCC 6538 and *Enterococcus faecalis* IMB B-7497 decreased from 62.5 to 31.2 μg/cm^3, and *Alcaligenes faecalis* GISK 242484 – LI 415 from 62.5 to – 15.6 μg/cm^3. It is also necessary to note the decrease in the minimum inhibitory concentration of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid against isolates of *Enterobacter cloacae* and *Pseudomonas aeruginosa*, respectively, from 250 and 500 to 62.5 and 125 μg/cm^3. At a pH of 8.8 units, the minimum inhibitory concentration of the tested substance increased, compared to pH of 7.0 and 5.2 units. In particular, it was found that the minimum inhibitory concentration against *S. aureus* ATCC 6538 was 250 μg/cm^3, while at pH values of the medium of 5.2 and 8.8 units, respectively, it was 31.2 and 15.6 μg/cm^3. In addition, the minimum inhibitory concentration against *Salmonella typhimurium* 144, *E. faecalis* IMB B-7497, *A. faecalis* GISK 242484 – LI 415, and *Serratia marcescens* 1 was 250 μg/cm^3, while at pH values of the medium of 5.2 and 8.0 units, the MICs were lower. The sensitivity of gram-positive microorganisms to ethyl ester of thiosulfonic acid, in particular, *S. aureus* ATCC 6538 and *E. faecalis* IMB B-7497, with a decrease in the pH of the medium to the weakly acidic side, increased from 5.4% to 7.1%, and in the weakly alkaline, on the contrary, decreased by 3.8%. The sensitivity of gram-negative microorganisms, in particular, *Escherichia coli* ATCC 25922, *S. typhimurium* 144, *P. aeruginosa*, *E. cloacae*, *A. faecalis* GISK 242484 – LI 415, *S. marcescens* 1, with a decrease in the pH of the medium to the weakly acidic side, increased from 2.3% to 12.2%, and in the weakly alkaline medium decreased from 1.5% to 18.2%.

Keywords: minimum inhibitory concentrations; sensitivity of microorganisms; gram-positive; gram-negative microorganisms.

Introduction

Antimicrobial resistance (AMR) is a global problem of modern medicine and one of the serious challenges for humanity, caused by the acquisition of antibiotic resistance by some bacteria. Today, AMR poses a threat not only to human health but also to the economy and security of countries as a whole (Khaytovych & Polyakova, 2023). The reason for this is the excessive and inappropriate use of antibiotics in both human and veterinary medicine. As a result, infectious diseases have become difficult to treat. This increases the incidence of diseases caused by antibiotic-resistant microorganisms (Breijyeh et al., 2020). It is important to note that antibiotics are also used in animal husbandry to stimulate animal growth. When such food products are consumed, resistant bacteria will likely be transmitted to humans (Salmanov et al., 2022).

The WHO has prepared a Global Action Plan on Antimicrobial Resistance to implement therapeutic and preventive measures using effective and safe medicines. To this end, strategic objectives have been identified, among which an important place belongs to increasing investment in developing new drugs or structural modification of existing antibiotics (Breijyeh et al., 2020).

In addition, one of the essential tasks of solving the problem of antimicrobial resistance is the search for antimicrobial agents that are alternatives to antibiotics. In this aspect, sulfur-containing organic compounds, particularly S-esters of thiosulfonic acids, deserve special attention. These compounds are structural analogs of natural sulfur-containing substances, such as allicin, which have a wide range of biological activities, in particular, antimicrobial activity (Halenova et al., 2015; Lubenets et al., 2017; Martyrosian et al., 2019; Martini & Passos, 2023). In addition, they exhibit antiparasitic activity (Dmitryjuk et al., 2020) and also have antithrombotic (Bolibrukh et al., 2015) and antitumor activity (Guillamón et al., 2023). The positive effect of allicin has also been proven in experiments on rabbits when fed this substance. In particular, an increase in the body weight of the animals was found, which is associated with the presence of sulfur-containing organic compounds in their composition (Isko & Sychov, 2022).

When studying the effect of structural analogs of allicin, in particular, S-allyl-4-aminobenzenethiosulfonate and S-allyl-4-acetylaminobenzenethiosulfonate, their positive impact on the antioxidant defense system of the body of laboratory animals was established (Liubas et al., 2022). There are also data on the treatment of obesity and type 2 diabetes with drugs containing allicin, which promotes the transformation of white adipose tissue into beige, which is essential in the treatment of this pathology.

Today, one of the essential tasks of organic chemistry is the synthesis of new biologically active compounds and studying the relationship between their structure, reactivity, and biological properties. Given this, the synthesis of new sulfur-containing compounds is aimed, first of all, at establishing their antimicrobial activity. Thus, out of twenty-four synthesized sulfur-containing compounds, only two exhibited pronounced antimicrobial activity against gram-positive and gram-negative microorganisms and antifungal activity: S-phenyl 4-

(acetylamino)benzenesulfonothioate and S-phenyl 4-(3-chloropropanol) aminobenzenesulfonothioate (Lubenets et al., 2017).

Natural and synthetic thiosulfonates are the subject of research by scientists of the Department of Technology of Biologically Active Compounds, Pharmacy, and Biotechnology of the Institute of Chemistry and Chemical Technologies of the National University "Lviv Polytechnic." The research is conducted under the leadership of Professor Lubenets V. I. The scientific achievements of the Department of TBSFB include more than 900 compounds of the thiosulfonate structure. New synthesis methods and the search for prospects for the use of these biologically active compounds for applied use in various industries are constantly being developed (Vasylyuk et al., 2018; Dmitryjuk et al., 2020; Martirosyan et al., 2021; Martirosyan et al., 2022; Zaczynska et al., 2023). Therefore, a new method was developed for obtaining and synthesizing ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid as a promising low-toxic compound with antimicrobial activity. The research aimed to determine the minimum inhibitory concentrations (MIC) of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid at different pH values of the medium.

Materials and methods

Methods for obtaining the sodium salt of 4-acetylaminobenzenethiosulfonic acid and the ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid, as well as their characteristics, were described in the works of Lubenets et al. (2013, 2017, 2024).

The determination of minimum inhibitory concentrations was carried out using serial dilutions in a liquid nutrient medium. For this purpose, microorganisms were grown on meat-peptone agar and incubated in a thermostat at 35 ºC for 24 hours. After the end of the cultivation period, the culture suspension was prepared. A suspension of test cultures, equivalent to 0.5 units according to the McFarland standard, was prepared in an isotonic solution of sodium chloride. As test cultures, museum strains of the following microorganisms were used: *Staphylococcus aureus* ATCC 6538, *Escherichia coli* ATCC 25922, *Salmonella typhimurium* 144, *Enterococcus faecalis* IMB B-7497, *Alcaligenes faecalis* GISK 242484- LI 415, *Serratia marcescens* 1, and clinical strains: *Pseudomonas aeruginosa*, *Enterobacter cloacae*.

Serial dilutions were conducted in tryptone-soy broth (TSB). For this, 2 cm^3 of the nutrient broth was poured into each test tube; one test tube served as a negative control.

A working solution of the test substance with a volume of 2 cm^3 was added to the first test tube containing 2 cm^3 of TSB using a pipette with a sterile tip. This was thoroughly mixed and 2 cm^3 of the broth solution transferred to the next test tube with 2 cm^3 of TSB with a new sterile tip. This procedure was repeated as many times as necessary, and 2 cm^3 of broth was removed from the last test tube. After that, a suspension of test cultures in the amount of 0.2 cm^3 was added to each test tube with a different concentration of the test substance and to the test tube with a negative control. The rack with the test tubes was shaken and placed in a thermostat for 24 hours at 36 ± 1 ºC. After the end of the cultivation period, the presence or absence of growth of microorganisms in the test tubes was noted. The lowest concentration of the drug, at which the multiplication of microorganisms did not occur and the content of the test tube remained transparent, was the lowest inhibitory concentration.

The determination of the sensitivity of microorganisms to the test substance was carried out using the agar diffusion method on the Mueller-Hinton medium. For this, test strains of microorganisms were grown on meat-peptone agar, incubated in a thermostat at a temperature of 36 ± 1 ºC for 24 h, and a culture suspension was prepared at 0.5 units according to the McFarland standard. The nutrient medium was melted and poured into petri dishes in two layers. Uninoculated Mueller-Hinton medium was used for the lower layer and inoculated with test microorganisms for the upper layer.

After the medium solidified, holes ("wells") were made in the thickness using a sterile drill (with a diameter of 6 mm) at a distance of 28 mm from the center of the dish, into which the test agent was introduced. The petri dishes were incubated in a thermostat for 24 h at 36 ± 1 ºC. The results of the studies were taken into account by the diameter of the zones of inhibition of the growth of microorganisms around the "wells".

Results

An essential element in determining the effectiveness and safety of the synthesized compound S-ester of 4-acetylaminobenzenethiosulfonic acid is conducting microbiological and toxicological studies. The antimicrobial efficacy of the studied compound was studied in experiments to determine its minimum inhibitory concentration against test strains of microorganisms.

The minimum inhibitory concentrations of the studied agent were studied in TSB with pH indicators of the medium at pH 5.2. 7.0 and 8.8 units. The results of studies on the influence of the pH of the medium, 7.0 units on the activity of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid are given in Table 1.

Table 1
Minimum inhibitory concentrations of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid at different pH levels of the medium

Test cultures	pH of the medium / concentration, μg/cm^3		
	5.2	7.0	8.8
S. aureus ATCC 6538	31.2	62.5	250
E. coli ATCC 25922	62.5	125.0	125
S. typhimurium 144	62.5	125.0	250
P. aeruginosa	125.0	500.0	1000
E. cloacae	62.5	250.0	250
E. faecalis IMB B-7497	31.2	62.5	250
A. faecalis GISK 242484-LI 415	15.6	62.5	250
S. marcescens 1	31.2	125.0	250

When determining the minimum inhibitory concentrations of the test substance at a pH of 7.0, antimicrobial activity was established at a concentration of 62.5 μg/cm^3 against two gram-positive microorganisms: *S. aureus* ATCC 6538 and *E. faecalis* IMB B-7497, and against the gram-negative strain *A. faecalis* GISK 242484-LI 415. It should be noted that the lowest sensitivity to the test substance sulfonic acid was shown by two strains: *E. cloacae* and *P. aeruginosa*. The minimum inhibitory concentration was 250 and 500 μg/cm^3, and *E. coli* ATCC 25922, *S. typhimurium* 144, and *S. marcescens* were sensitive to the test agent at 125 μg/cm^3.

When the pH of the medium changed to 5.2 units (Table 1), the MIC of the tested substance also changed, compared to the pH of the medium of 7.0 units. Thus, the minimum inhibitory concentration against *S. aureus* ATCC 6538 and *E. faecalis* IMB B-7497 from 62.5 at a pH of 7.0 units decreased to 31.2 μg/cm^3, and against *A. faecalis* from a concentration of 62.5 decreased to 15.6 μg/cm^3.

It is also necessary to note the decrease in the MIC of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid at a pH of the medium of 5.2 units against *E. cloacae* and *P. aeruginosa*, which decreased, respectively, from 250 and 500 to – 62.5 and 125 μg/cm^3. The same trend was observed when studying all other test strains of microorganisms.

At a pH of 8.8 units (Table 1), the MIC of the studied substance increased compared to the pH of 7.0 and 5.2 units. It was found that at a pH of 8.8 units, the MIC of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid against *S. aureus* ATCC 6538, *S. typhimurium* 144, *E. faecalis* IMB B-7497, *A. faecalis* GISK 242484-LI 415, and *S. marcescens* 1 was 250 μg/cm^3. While at pH of 7.0 and 5.2 units, it was significantly lower and amounted to at pH of 5.2 units, respectively – 31.2, 62.5, 31.2, 15.6, and 31.2 μg/cm^3, and at pH of 7.0 units, respectively – 62.5, 125, 62.5, 62.5, and 125 μg/cm^3.

The minimum inhibitory concentrations of the studied substance against *P. aeruginosa* were recorded at a pH of 8.8 units. The minimum inhibitory concentration was the highest compared to pH of 5.2 and 7.0 units and amounted to 1000 μg/cm^3.

Subsequently, the diffusion method in an agar nutrient medium using "wells" determined the sensitivity of test strains of microorganisms to the studied compound at different pH values (5.2, 7.0, and 8.8 units). The concentration of the test substance was 4 mg/cm^3, the

diameter of the "wells" was 6 mm, and the solvent of the test substance served as a control.

Table 2
Sensitivity of test cultures to the test agent

Microorganisms	Control, pH 5.2/7.0/8.8	pH of the medium/zone of inhibition of the growth of microorganisms, mm		
		5.2	7.0	8.8
S. aureus ATCC 6538	0/0/0	19.6 ± 0.3^b	18.6 ± 0.3^a	18.6 ± 0.3^a
E. coli ATCC 25922	0/0/0	19.3 ± 0.3^c	18.3 ± 0.3^b	17.7 ± 0.3^a
S. typhimurium 144	0/0/0	17.7 ± 0.3^b	17.3 ± 0.3^b	16.3 ± 0.3^a
P. aeruginosa	0/0/0	9.6 ± 0.3^b	9.3 ± 0.3^b	7.6 ± 0.3^a
E. cloacae	0/0/0	16.6 ± 0.3^b	15.6 ± 0.3^a	15.3 ± 0.3^a
E. faecalis IMB B-7497	0/0/0	19.6 ± 0.3^c	18.3 ± 0.3^b	17.6 ± 0.3^a
A. faecalis GISK 242484-LI 415	0/0/0	18.3 ± 0.3^c	16.3 ± 0.3^b	5.6 ± 0.3^a
S. marcescens 1	0/0/0	20.3 ± 0.3^c	19.6 ± 0.3^b	19.3 ± 0.3^a

Note: different letters in a row indicate that data samplings are significantly ($P < 0.05$) different one from another according to the Tukey test with the Bonferroni correction.

As a result of the conducted studies, it was established (Table 1) that the test strains of microorganisms were sensitive to the studied substance at all pH values of the medium. At the same time, different activity of ethyl ester of thiosulfonic acid to microorganisms was revealed, which depended on the pH value of the medium. In particular, in a weakly acidic medium, the sensitivity of microorganisms was higher compared to the medium's neutral and weakly alkaline pH values. In addition, it was found that in a weakly alkaline medium, the activity of the studied substance was the lowest. If we analyze the sensitivity of gram-negative microflora to the studied agent, it was established that the most resistant microorganism was the *P. aeruginosa* strain, the zones of growth inhibition of microorganisms around the "wells" at pH values of 5.2, 7.0, and at a pH of 8.8 units were, respectively, – 9.6 ± 0.3, 9.3 ± 0.3 and 7.6 ± 0.3 mm ($P < 0.05$). At the same time, the activity of the studied substance increased with a decrease in the pH of the medium compared to neutral pH by 3.2%, and with an increase to slightly alkaline pH, it decreased by 18.2%.

The most sensitive gram-negative microorganism was the *S. marcescens* strain, which belongs to the Yersiniaceae family; the zones of inhibition of growth of microorganisms at a pH of 5.2, 7.0, and 8.8 units were, respectively, 20.3 ± 0.3, 19.6 ± 0.3, and 19.3 ± 0.3 mm.

It should be noted that the studies used a clinical isolate of *E. cloacae*, which was resistant to most antimicrobial drugs. This strain was less sensitive to the action of ethyl ester of thiosulfonic acid compared to other gram-negative microorganisms; the zones of inhibition of growth of microorganisms at a pH of 5.2, 7.0, and 8.8 units were, respectively, – 16.6 ± 0.3, 15.6 ± 0.3, and 15.3 ± 0.3 mm.

The sensitivity of gram-positive microorganisms *S. aureus* ATCC 6538 and *E. faecalis* IMB B-7497 to the tested agent was the same at a pH of 5.2 units; the inhibition zones of growth of microorganisms were 19.6 ± 0.3 mm. At a pH of 7.0 units, the zones of inhibition of growth were, respectively, 18.6 ± 0.3 and 18.3 ± 0.3, and at a pH of 8.8 units, 18.6 ± 0.3 and 17.6 ± 0.3 mm.

Thus, the sensitivity of gram-positive microorganisms to ethyl ester of thiosulfonic acid, in particular, *S. aureus* ATCC 6538 and *E. faecalis* IMB B-7497, with a change in the pH of the medium to the weakly acidic side, increased from 5.4% to 7.1%, and in the weakly alkaline side decreased by 3.8%. The sensitivity of gram-negative microorganisms, in particular, *E. coli* ATCC 25922, *S. typhimurium* 144, *P. aeruginosa*, *E. cloacae*, *A. faecalis* GISK 242484-LI 415, *S. marcescens* 1, with a change in the pH of the medium to the weakly acidic side, increased from 2.3% to 12.2%, and in the weakly alkaline side decreased by 1.5% to 18.2%.

Discussion

Today, the work of many scientists is aimed at the synthesis of valuable compounds with the elimination of certain environmental restrictions and the use of new "green" methods, in particular environmentally safe reactions under the influence of ultrasound (Pham et al., 2015; Heydari et al., 2022).

In this regard, a number of substances have been synthesized at the Department of Technology of Biologically Active Substances, Pharmacy and Biotechnology of Lviv Polytechnic National University, but compounds 2 and 3, obtained according to Figure 1, deserve special attention.

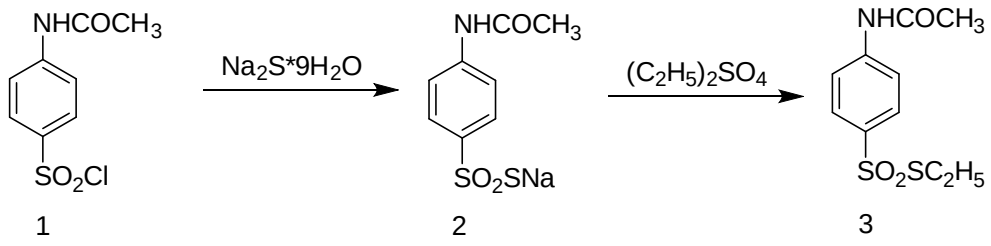

Fig. 1. Synthesis of ethyl S- ester of 4-acetylaminobenzenethiosulfonic acid as a starting compound for the synthesis of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid (*3*), 4-acetylaminobenzenesulfonyl chloride (*1*) was used, which was converted into the corresponding sodium salt of 4-acetylaminobenzenethiosulfonic acid (*2*) using a redox reaction with sodium sulfide: to obtain ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid (*3*), the sodium salt of 4-acetylaminobenzenethiosulfonic acid (*2*) was alkylated with diethyl sulfate using various solvents and under "solvent-free" conditions

It is worth noting that a new method for obtaining thiosulfonate three was developed, which corresponds to the basic principles of "green chemistry". In particular, the alkylation reaction was carried out under the influence of ultrasound without solvents and in the presence of trace amounts of water. In addition, compared to the previous ones, the alkylation method we have proposed leads to an increase in the yield of the target product by up to 90% and reduces the reaction time. S-esters of thiosulfonic acids can exhibit antioxidant activity, protecting cells from damage caused by oxidative stress. This is due to their ability to modify tyrosine kinase substrates, which affects the interaction of these substrates with both tyrosine kinase and other proteins. Such compounds can affect the regulation of the cell cycle, inhibiting cell proliferation and reducing the reproductive capacity of cells. S-alkyl esters, in particular, can initiate apoptosis, promoting programmed cell death – a critically important process for eliminating damaged cells.

Thiosulfinates, such as allicin, are effective platelet aggregation inhibitors (Briggs et al., 2000) and are crucial in preventing cardiovas-

cular diseases, including ischemic heart and brain damage. Platelet aggregation is a complex biochemical process that requires activation of the GPIIb/IIIa receptor by thromboxane A2, which provides fibrinogen binding (Yip et al., 2005). Classical antiplatelet agents, such as acetylsalicylic acid (aspirin), block the synthesis of thromboxane, thereby inhibiting the activation of this receptor. In practical studies, it was found that the S-methyl ester with an aromatic benzenesulfonyl moiety demonstrates the ability to reduce platelet aggregation. It was found that even minor changes in the structure of compounds of the S-ester class of 4-aminobenzenethiosulfonic acid can lead to the loss of antiplatelet activity. For example, acetylation of the amino group in such compounds reduces their inhibitory effect: the methyl S-ester of 4-acetylaminobenzenethiosulfonic acid had two times lower activity than the analog that did not contain the acetyl group. These results confirm the high sensitivity of the platelet aggregation process, particularly collagen-induced, to the action of the studied compounds.

Allicin can influence signaling pathways associated with the immune response, which opens up potential prospects for its therapeutic application. If allicin activates immune cells, it can enhance the body's defense against pathogens, as well as modulate immune responses, which is promising in the context of the treatment of allergies and autoimmune diseases. To confirm the immunomodulatory effect of allicin, a study was conducted on a mouse model of Morbus Bechterew – a degenerative rheumatoid lesion of the vertebrae. One of the first effects detected was the ability of allicin to inhibit the migration of neutrophil granulocytes into epithelial tissues – a process that plays a key role in the development of inflammation (Gu et al., 2013). It has also been found that allicin affects T lymphocytes by inhibiting their chemotaxis induced by the chemokine SDF1α. This effect is associated with a disruption of the actin cytoskeleton.

Additionally, allicin has been shown to inhibit neutrophil transendothelial migration (Sela et al., 2004), further supporting its potential as an anti-inflammatory agent. Allicin also affects cytoskeletal structures in various cell systems. In particular, in experiments on mouse fibroblasts (NIH-3T3 line), it was found that even at low concentrations (2 μM), allicin causes rapid depolymerization of microtubules. At the same time, the structure of the actin cytoskeleton remains stable (Prager-Khoutorsky et al., 2007). One of the central regulators of lymphocyte activation is the p21ras protein, which initiates the inactivation of RAS-GTPase by increasing its enzymatic activity. Interestingly, this protein is a direct target of allicin: as a result of thioallylcylation, p21ras is activated (Patya et al., 2004), which, in turn, can lead to increased phosphorylation of the ERK1/2 kinase, which is involved in numerous signaling cascades critical for lymphocyte activation.

Allicin can inhibit phosphatase activity, which is accompanied by increased phosphorylation of ERK1/2, a key element of the signaling cascade that transforms external signals into an intracellular response. In addition, allicin inhibits the production of reactive nitrogen species (RNS) in macrophages activated by lipopolysaccharide (LPS). Thus, allicin can directly affect inflammatory processes due to its antimicrobial properties and indirectly through the modulation of signaling in immune cells (Haase et al., 2012).

In scientific research, considerable attention has been paid to studying the biological activity of thiosulfonates, particularly their antimicrobial properties and mechanisms of action (Lubenets et al., 2017). The high efficiency, broad spectrum of antimicrobial activity, stability, and low toxicity of thiosulfoesters make them promising candidates for developing new drugs (Baranovych et al., 2001). This is especially relevant against the increasing prevalence of pathogens resistant to existing antimicrobial drugs (El Bcheraoui et al., 2018). Infectious diseases previously effectively treated are increasingly resistant to standard therapy (Boucher et al., 2009). In the context of increasing antibiotic resistance, only a few new therapeutic approaches have reached the stage of clinical trials in recent years (Halenova et al., 2017).

Conclusions

It was found that the sensitivity of microorganisms to the action of the synthesized ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid changed with the change in the pH of the medium. In particular, at a pH of 5.2 units, it increases, and at a pH of 8.0 units, it decreases, compared to a pH of 7.0. The sensitivity of gram-negative microorganisms at a pH of 5.2 units increases from 2.3% to 12.2%, and at a pH of 8.0 units, it decreases from 1.5% to 18.2%. The sensitivity of gram-positive microorganisms at a pH of 5.2 increases from 5.4% to 7.1%, and a pH of 8.0 decreases by 3.8%, compared to a pH of 7.0 units. Prospects for further research: determination of the fungicidal effect of ethyl S-ester of 4-acetylaminobenzenethiosulfonic acid.

References

Baranovych, D. B., Komarovska, O. Z., Lubenets, V. I., & Novikov, V. P. (2001). Syntez i biolohichna aktyvnist S-alkilbenzoltiosulfonativ [Synthesis and biological activity of S-alkylbenzenethiosulfonates]. Fiziolohichno Aktyvni Rechovyny, 2(30), 33–36 (in Ukrainian).

Bolibrukh, K., Polovkovych, S., Khoumeri, O., Halenova, T., Nikolaeva, I., Savchuk, O., Terme, T., Vanelle, P., Lubenets, V., & Novikov, V. (2015). Synthesis and anti-platelet activity of thiosulfonate derivatives containing a quinone moiety. Scientia Pharmaceutica, 83(2), 221–231.

Boucher, H. W., Talbot, G. H., Bradley, J. S., Edwards, J. E., Gilbert, D., Rice, L. B., Scheld, M., Spellberg, B., & Bartlett, J. (2009). Bad bugs, no drugs: No ESKAPE! An update from the Infectious Diseases Society of America. Clinical Infectious Diseases, 48(1), 1–12.

Breijyeh, Z., Jubeh, B., & Karaman, R. (2020). Resistance of Gram-negative bacteria to current antibacterial agents and approaches to resolve it. Molecules, 25(6), 1340.

Briggs, W. H., Xiao, H., Parkin, K. L., Shen, C., & Goldman, I. L. (2000). Differential inhibition of human platelet aggregation by selected *Allium thiosulfinates*. Journal of Agricultural and Food Chemistry, 48(11), 5731–5735.

Dmitryjuk, M., Szczotko, M., Kubiak, K., Trojanowicz, R., Parashchyn, Z., Khomitska, H., & Lubenets, V. (2020). S-methyl-(2-methoxycarbonyl-amino-benzimidazole-5) thiosulfonate as a potential antiparasitic agent – its action on the development of *Ascaris suum* eggs *in vitro*. Pharmaceuticals, 13(11), 332.

El Bcheraoui, C., Mokdad, A. H., Dwyer-Lindgren, L., Bertozzi-Villa, A., Stubbs, R. W., Morozoff, C., Shirude, S., Naghavi, M., & Murray, C. J. L. (2018). Trends and patterns of differences in infectious disease mortality among US counties, 1980–2014. Journal of the American Medical Association, 319(12), 1248–1260.

Gu, X., Wu, H., & Fu, P. (2013). Allicin attenuates inflammation and suppresses HLA-B27 protein expression in ankylosing spondylitis mice. BioMed Research International, 2013, 171573.

Guillamón, E., Mut-Salud, N., Rodríguez-Sojo, M. J., Ruiz-Malagón, A. J., Cuberos-Escobar, A., Martínez-Férez, A., Rodríguez-Nogales, A., Gálvez, J., & Baños, A. (2023). *In vitro* antitumor and anti-inflammatory activities of allium-derived compounds propyl propane thiosulfonate (PTSO) and propyl propane thiosulfinate (PTS). Nutrients, 15(6), 1363.

Haase, H., Hieke, N., Plum, L. M., Gruhlke, M. C., Slusarenko, A. J., & Rink, L. (2012). Impact of allicin on macrophage activity. Food Chemistry, 134(1), 141–148.

Halenova, T. I., Nikolaeva, I. V., Nakonechna, A. V., Bolibrukh, K. B., Monka, N. Y., Lubenets, V. I., & Ostapchenko, L. I. (2015). The search of compounds with antiaggregation activity among S-esters of thiosulfonic acids. The Ukrainian Biochemical Journal, 87(5), 83–92.

Halenova, T. I., Nikolaeva, I. V., Stasevych, M. V., Zvarych, V. I., Lunin, V. V., Novikov, V. P., & Savchuk, O. M. (2017). Platelet aggregation under the influence of some dithiocarbamate derivatives of 9,10-anthracenedione. Research Journal of Pharmaceutical, Biological and Chemical Sciences, 8, 1626–1632.

Heydari, S., Habibi, D., & Faraji, A. A. (2022). Green and efficient solventand catalyst-free ultrasonic dibenzylation procedure. Chemistry and Chemical Technology, 16(1), 126–132.

Isko, O. Y., & Sychov, M. Y. (2022). Vplyv riznykh rivniv chasnyka (*Allium sativum*) na pokaznyky zaboyu molodnyaka kroliv [The influence of different levels of garlic (*Allium sativum*) on slaughter indicators of young rabbits]. Tavriiskyi Naukovyi Visnyk Silskohospodarskoyi Nauky, 128, 251–256 (in Ukrainian).

Khaytovych, M. V., & Polyakova, D. S. (2023). Antymikrobna rezystentnist' ta orhanizatsiyni pytannya administruvannya antymikrobnykh preparativ v Ukrayini [Antimicrobial resistance and organizational issues of antimicrobial drug administration in Ukraine]. Ukrainskyi Medychnyi Chasopys, 153(1), 5–9 (in Ukrainian).

Liubas, N., Iskra, R., Stadnytska, N., Monka N., Havryliak, V., & Lubenets, V. (2022). Antioxidant activity of thiosulfonate compounds in experiments

in vitro and *in vivo*. Biointerface Research in Applied Chemistry, 12(3), 3106–3116.

Lubenets, V., Karpenko, O., Ponomarenko, M., Zahoriy, G., Krychkovska, A., & Novikov, V. (2013). Development of new antimicrobial compositions of thiosulfonate structure. Chemistry and Chemical Technology, 7(2), 119–124.

Lubenets, V., Vasylyuk, S., Monka, N., Bolibrukh, K., Komarovska-Porokhnyavets, O., Baranovych, D., Musyanovych, R., Zaczynska, E., Czarny, A., Nawrot, U., & Novikov, V. (2017). Synthesis and antimicrobial properties of 4-acylaminobenzenethiosulfoacid S-esters. Saudi Pharmaceutical Journal, 25(2), 266–274.

Martini, H., & Passos, J. F. (2023). Cellular senescence: all roads lead to mitochondria. The FEBS Journal, 290(5), 1186–1202.

Martirosyan, I., Pakholiuk, E., Lubenets, V., Komarovska-Porokhnyavets, O., Monka, N., Nakonechna, A., Peredriy, O., & Lutskova, V. (2021). Innovative approach to the creation of textile materials with antimicrobial properties. Science and Innovation, 17(3), 56–66.

Martirosyan, I., Pakholiuk, O., Golodyuk, G., Lutskova, V., & Lubenets, V. (2022). Investigation of antimicrobial properties of textile materials after washing. Fibres and Textiles, 29(1), 28–35.

Martyrosian, I. A., Pakholiuk, O. V., Semak, B. D., Komarovska-Porokhniavets, O. Z., Lubenets, V. I., & Pambuk, S. A. (2019). New technologies of effective protection of textiles against microbiological damage. Nanosistemy, Nanomateriali, Nanotehnologii, 17(4), 621–636.

Patya, M., Zahalka, M. A., Vanichkin, A., Rabinkov, A., Miron, T., Mirelman, D., Wilchek, M., Lander, H. M., & Novogrodsky, A. (2004). Allicin stimulates lymphocytes and elicits an antitumor effect: A possible role of p21ras. International Immunology, 16(2), 275–281.

Pham, H. T., Nguyen, N. T., Duus, F., & Luu, T. X. T. (2015). Ultrasound-accelerated synthesis of asymmetrical thiosulfonate S-esters by base-promoted reaction of sulfonyl chlorides with thiols. Phosphorus, Sulfur, and Silicon and the Related Elements, 190(11), 1934–1941.

Prager-Khoutorsky, M., Goncharov, I., Rabinkov, A., Mirelman, D., Geiger, B., & Bershadsky, A. D. (2007). Allicin inhibits cell polarization, migration and division via its direct effect on microtubules. Cell Motility and the Cytoskeleton, 64(5), 321–337.

Salmanov, A. H., Shchehlov, D. V., Artomenko, V. V., Mamonova, M. Y., & Ushkalov, V. O. (2022). Strymuvannia antymikrobnoyi rezystentnosti na pidkhodakh "Yedyne zdorovia" [Suppression of antimicrobial resistance in the "One health" approach]. AhrarMediaHrup, Kyiv (in Ukrainian).

Sela, U., Ganor, S., Hecht, I., Brill, A., Miron, T., Rabinkov, A., Wilchek, M., Mirelman, D., Lider, O., & Hershkoviz, R. (2004). Allicin inhibits SDF-1-alphainduced T cell interactions with fibronectin and endothelial cells by downregulating cytoskeleton rearrangement, Pyk-2 phosphorylation and VLA-4 expression. Immunology, 111(4), 391–399.

Vasylyuk, S. V., Shyyan, H. B., Monka, N. Y., Shyyan, A. V., & Lubenets, V. I. (2018). Syntez tiosulfoesteriv iz sym-tryazynovym frahmentom [Synthesis of thiosulfoesters with a sym-tryazine fragment]. Chemistry, Technology and Application of Substances, 2(1), 98–104 (in Ukrainian).

Yip, J., Shen, Y., Berndt, M. C., & Andrews, R. K. (2005). Primary platelet adhesion receptors. IUBMB Life, 57(2), 103–108.

Zaczynska, E., Czarny, A., Karpenko, O., Vasylyuk, S., Monka, N., Stadnytska, N., Fizer, L., Komarovska-Porokhnyavets, O., Jaranowski, M., Lubenets, V., & Zimecki, M. (2023). Obtaining and determining antiviral and antibacterial activity of S-esters of 4-r-aminobenzenethiosulfonic acid. Chemistry and Chemical Technology, 17(2), 315–324.