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SYNTHESIS OF NEW S-(ω-FLUORALKYL) 4-SUBSTITUTED BENZENESULFONOTHIOATES AND THEIR ANTIMICROBIAL ACTIVITY

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New S-(ω-fluoralkyl) 4-amino- and 4-acetamidobenzenesulfonothioates were synthesized by the reaction of sodium salts of the substituted thiosulfonic acids and corresponding ω-fluoralkylbromides. The study of their antimicrobial activity against thirteen reference and clinical bacterial and fungal strains using agar diffusion method was conducted. All compounds showed significant antimicrobial activity, two of them showed higher activity against Candida albicans than known antifungal compound of sulfonothioate class, S-ethyl ester of 4-aminobenzenethiosulfonic acid.

Keywords: sulfonothioates, fluoralkylbromides, antibacterial activity, antifungal activity, clinical strains.

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Introduction

S-alkyl sulfonothioates are structural analogs of phytoncids (defence molecules) found in plants of the genus *Allium*, and some others, for example cabbage [1]. These compounds are S-esters of sulfinothioic acid, the most common among them is allicin (S-allyl prop-2-ene-1-sulfinothioate) which biological activity is well-known. In a dose-dependent manner, allicin inhibit or kill both bacteria and fungi, including resistant strains like MRS [2]. It acts against numerous Gram positive and Gram-negative bacteria such as *Escherichia coli*, *Salmonella enterica*, *Shigella* (*Shigella boydii*, *Shigella flexneri*, *Shigella sonnei*), *Enterococcus faecalis*, *Staphylococcus aureus*, *Streptococcus* (*Streptococcus faecalis*, *Streptococcus mutans*, *Streptococcus pyogenes*), *Klebsiella aerogenes*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, as well as against a number of fungi: *Candida albicans* and *Aspergillus niger*. In addition, it shows antiviral activity both *in vitro* and *in vivo* [3]. However, allicin is quite unstable due to the presence of tetravalent sulfur and readily decomposes even at room temperature [3]. In contrast, S-esters of sulfonothioic acids are much more stable. Their biological activity, especially antifungal, has also been widely studied [4]. The most well-known compound from this class is S-ethyl 4-aminobenzenesulfonothioate (4d); for example its considerable antifungal effect on *Candida tropicalis* was described and some aspects of its action in subfungicidal concentration was studied [5]. In addition, for thiosulfonic compounds other types of activity have been found, for example antiaggregation [6] and many others.

The biological activity of fluorine-containing S-esters of sulfonothioic acids has not been studied much. However, a number of such compounds have been obtained since esters of thiosulfonic acids are effective fluoroalkyllating and fluoroalkylthiolating reagents in organic synthesis. For example, radical fluoroalkyllation of aldehydes with $Ph-SO_2-S-R_f$ was described, where R_f is $-CF_3$, $-C_2F_5$, $-CF_2H$ or $-CH_2F$ group [7]. Compounds of such type are prepared by electrophilic fluoroalkylthiolation of $R-SO_2Na$ or of $R-SO_2Cl$ with *in situ* generation of $R-SO_2^{(-)}$ [8]. From this list, only compounds with $-CH_2F$ group can be prepared under harsh conditions with the nucleophilic substitution reaction of $R-SO_2SNa$ and CH_2FCl [9].

The compounds of $R-SO_2-S-(CH_2)_n-CH_2F$ type are much more accessible synthetically as they

can be prepared with the nucleophilic substitution reaction of available and chemically active 1-bromo-ω-fluoroalkanes and sodium or potassium salts of substituted sulfonothioic S-acids. The change of alkyl substituents in compounds with known antimicrobial action to their close isosteres, ω-fluorinated alkyls, may be interesting in terms of improvement of biological activity.

Antimicrobial resistance (AMR) is a global issue that leads to increased mortality due to infectious diseases and places a more significant financial burden on the healthcare system. One short-term method to combat AMR is to search for new antimicrobial compounds among synthetic derivatives [10,11]. Therefore, the aim of our work was the synthesis of new S-fluoroalkyl 4-substituted benzenesulfonothioates and the investigation of their antimicrobial activity against a number of bacterial and fungal strains.

Experimental

General methods

Melting points were determined in open capillary tubes in a «Mettler Toledo ID 50» apparatus and were uncorrected. ^{1}H NMR spectra were recorded using a «Varian Mercury» (Varian Inc., Palo Alto, CA) 400 MHz spectrometer with DMSO-d_6 as solvent and solvent signal as an internal standard; the coupling constants are given in Hz. The elemental analysis was performed by means of a Euro Vector EA-3000 (Eurovector SPA, Redavalle, Italy) microanalyzer. Elemental analyses were within ±0.4% inaccuracy with respect to the theoretical values. The progress of the reaction was monitored by TLC on Silufol UV-254 plates. For preparative chromatography silica gel with 60 E pore size and 40–63 μm fraction was used. Chemical ionization mass spectrometry was performed on Agilent 1100 Series (LC/MSD Trap) Spectrometer using the gradient elution: A) H_2O+0.1% HCOOH; and B) CH_3CN+0.1% HCOOH. All starting materials and solvents were obtained from commercially available sources and used without additional purification.

General procedure for the synthesis of S-(w-fluoroalkyl) 4-aminobenzenesulfonothioates (4a–c) and S-(w-fluoroalkyl) 4-acetamidobenzenesulfonothioates (5a–c)

To the suspension of 2 mmol of sodium 4-aminobenzenesulfonothioate (1) or sodium 4-acetamidobenzenesulfonothioate (2) in 3 ml of freshly distilled DMF, 3 mmol (1.5 eq) of appropriate 1-bromo-ω-fluoroalcane (3a–c) was added in single portion. The mixture was stirred for 2 hours at room temperature and then 0.5 ml of diethanolamine was added to remove the excess of alkylating agent. After additional stirring for 5 hours at room temperature, the mixture was saturated with 50 ml of water and extracted 3×10 ml of DCM; combined organic extracts were washed with 5×30 ml of water, filtered through Na_2SO_4 and thin silica gel layer (washed with DCM) and the solvent was evaporated in water jet pump vacuum. The residue was dissolved in 2 ml of $CHCl_3$ and chromatographed on 30 cm^3 silica gel column using $CHCl_3$ to $CHCl_3$/EtOAc 1:1 for *(4a–c)* and $CHCl_3$ to $CHCl_3$/EtOAc 3:1 for *(5a–c)* gradient with TLC monitoring: in $CHCl_3$/EtOAc 3:1 system amino derivatives (4a–c) and acetylamino derivatives (5a–c) have R_f of about 0.3 and 0.5, respectively. Hexane/EtOAc systems can be used for isocratic eluation, but if gradient eluation is used, a $CHCl_3$ to $CHCl_3$/EtOAc system should be used because the obtained compounds have very little solubility in hexanes. The products were obtained as almost colorless oils with different viscosity, in some cases *(4a, 5a, 5b, 5c)* they crystallized at −30^{0}C for several weeks forming radial crystal growths.

S-(2-fluoroethyl) 4-aminobenzenesulfonothioate (4a)

Yield 69%, colorless crystals, m.p.=48–51^{0}C. ^{1}H NMR (400 MHz, DMSO-d_6) δ, ppm: 7.54 (d, *J*=8.7 Hz, 2H, 2CH (Ar)), 6.65 (d, *J*=8.8 Hz, 2H, 2CH (Ar)), 6.41 (s, 2H, NH_2), 4.55 (t, *J*=5.9 Hz, 1H, $-CH_2F$), 4.43 (t, *J*=5.9 Hz, 1H, $-CH_2F$), 3.30 (t, *J*=5.9 Hz, 1H, $-CH_2-$), 3.24 (t, *J*=5.9 Hz, 1H, $-CH_2-$). Calcd. for $C_8H_{10}FNO_2S_2$ (%): C 40.84; H 4.28; F 8.07; N 5.95; S 27.25. Found: C 40.80; H 4.31; F 8.04; N 6.01; S 27.31. LC–MS m/z: $[M+H]^+$: 236.0.

S-(2-fluoroethyl) 4-acetamidobenzenesulfonothioate (5a)

Yield 72%, colorless crystals, m.p.=75–77^{0}C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.48 (s, 1H, NH), 7.89 (d, *J*=8.9 Hz, 2H, 2CH (Ar)), 7.84 (d, *J*=9.0 Hz, 2H, 2CH (Ar)), 4.58 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 4.46 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 3.38 (t, *J*=5.7 Hz, 1H, $-CH_2-$), 3.31 (t, *J*=5.7 Hz, 1H, $-CH_2-$), 2.11 (s, 3H, Ac). Calcd. for $C_{10}H_{12}FNO_3S_2$ (%): C 43.31; H 4.36; F 6.85; N 5.05; S 23.12. Found: C 43.22; H 4.41; F 6.92; N 4.99; S 23.08. LC–MS m/z: $[M+H]^+$: 278.0.

S-(3-fluoropropyl) 4-aminobenzenesulfonothioate (4b)

Yield 71%, almost colorless oil. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.52 (d, *J*=8.7 Hz, 2H, 2CH (Ar)), 6.65 (d, *J*=8.8 Hz, 2H, 2CH (Ar)), 6.37 (s, 2H, NH_2), 4.48 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 4.36 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 3.00 (t, *J*=7.3 Hz, 2H, $-CH_2-$ (1')), 2.00–1.81 (m, 2H, $-CH_2-$ (2')). Calcd. for $C_9H_{12}FNO_2S_2$ (%): C 43.36; H 4.85; F 7.62; N 5.62; S 25.72. Found: C 43.42; H 4.91;

F 7.58; N 5.54; S 25.66. LC–MS m/z: $[M+H]^+$: 250.0.

S-(3-fluoropropyl) 4-acetamidobenzenesulfonothioate (5b)

Yield 73%, colorless crystals, m.p.=87–90°C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.47 (s, 1H, NH), 7.87 (d, *J*=9.1 Hz, 2H, 2CH (Ar)), 7.84 (d, *J*=10.6 Hz, 2H, 2CH (Ar)), 4.48 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 4.36 (t, *J*=5.7 Hz, 1H, $-CH_2F$), 3.07 (t, *J*=7.3 Hz, 2H, $-CH_2-$ (1')), 2.10 (s, 3H, Ac), 1.99–1.84 (m, 2H, $-CH_2-$ (2')). Calcd. for $C_{11}H_{14}FNO_3S_2$ (%): C 45.35; H 4.84; F 6.52; N 4.81; S 22.01. Found: C 45.40; H 4.79; F 6.48; N 4.77; S 21.93. LC–MS m/z: $[M+H]^+$: 292.0.

S-(4-fluorobutyl) 4-aminobenzenesulfonothioate (4c)

Yield 68%, almost colorless low viscosity oil. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.52 (d, *J*=8.8 Hz, 2H, 2CH (Ar)), 6.64 (d, *J*=8.8 Hz, 2H, 2CH (Ar)), 6.35 (s, 2H, NH_2), 4.42 (t, *J*=5.6 Hz, 1H, $-CH_2F$), 4.30 (t, *J*=5.6 Hz, 1H, $-CH_2F$), 2.96 (t, *J*=6.9 Hz, 2H, $-CH_2-$ (1')), 1.70–1.53 (m, 4H, $2CH_2$). Calcd. for $C_{10}H_{14}FNO_2S_2$ (%): C 45.61; H 5.36; F 7.21; N 5.32; S 24.35. Found: C 45.57; H 5.42; F 7.17; N 5.40; S 24.41. LC–MS m/z: $[M+H]^+$: 264.0.

S-(4-fluorobutyl) 4-acetamidobenzenesulfonothioate (5c)

Yield 73%, colorless crystals, m.p.=87–89°C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.46 (s, 1H, NH), 7.87 (d, *J*=9.1 Hz, 2H, 2CH (Ar)), 7.83 (d, *J*=9.2 Hz, 2H, 2CH (Ar)), 4.42 (t, *J*=5.5 Hz, 1H, $-CH_2F$), 4.30 (t, *J*=5.4 Hz, 1H, $-CH_2F$), 3.04 (t, *J*=6.8 Hz, 2H, $-CH_2-$ (1')), 2.10 (s, 3H, Ac), 1.69–1.54 (m, 4H, $2CH_2$). Calcd. for $C_{12}H_{16}FNO_3S_2$ (%): C 47.20; H 5.28; F 6.22; N 4.59; S 21.00. Found: C 47.13; H 5.33; F 6.18; N 4.63; S 20.93. LC– MS m/z: $[M+H]^+$: 306.1.

Antimicrobial activity

The studied compounds were tested *in vitro* for their antibacterial and antifungal activities using agar diffusion [12–14]. For this purpose, 100 μL (1 mg/mL) of the tested compound was placed in an agar well with a diameter of 5.5 mm. The diameter of the growth retardation was measured using a micrometer with an error of 0.1 mm. Dimethyl sulfoxide (DMSO) (Sigma-Aldrich, St. Louis, MO, USA), vancomycin (discs) (Thermo Fisher Scientific Inc., Waltham, MA, USA), ciprofloxacin (discs) (Thermo Fisher Scientific Inc., Waltham, MA, USA), and clotrimazole (discs) (Mast Group Ltd., Liverpool, UK) were used as controls. Pure DMSO was used as a solvent due to the poor solubility of the test compound in dilute DMSO. In addition, Mueller-Hinton agar (Sigma-Aldrich, St. Louis, MO, USA) and Saburo agar (for fungi) (Sigma-Aldrich, St. Louis, MO, USA) were used, and Petri dishes were incubated at 37°C for 24 h for bacteria and at 25°C for 24–48 h for fungi. Reference antibacterial and antifungal compounds were used: Vancomycin 30 μg (inhibition zone 17–21 mm for S. aureus); Ciprofloxacin 5 μg (inhibition zone 25–33 mm for *P. aeruginosa*, 22–30 mm for *S. aureus*, and 30–40 mm for *E.coli*); Clotrimazole 10 μg (inhibition zone 12–17 mm for *Candida spp*); diameter of well 5.5 mm.

For minimal inhibitory concentration (MIC) determination, resazurin-based microdilution assay was used. A mixture comprising nutrient broth (either Mueller-Hinton or glucose broth), a suspension of microbes, and the compounds under investigation was prepared in a 96-well plate. Specifically, each well was filled with 50 μl of nutrient broth, 50 μl of microbial suspension standardized to a McFarland 1.0 concentration, and 100 μl of the compounds being tested. Additionally, 15 μl of 0.02% resazurin solution was added to every well to facilitate the assessment.

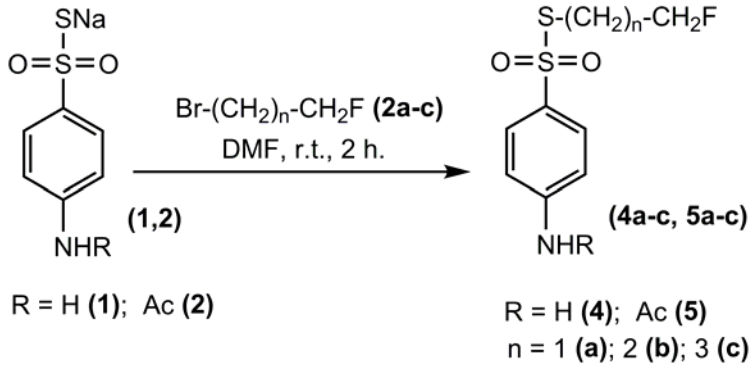
Thirteen reference and clinical microbial and fungal strains were used, previously identified by the MALDI TOF system (Bruker, Bremen, Germany) and 16S rRNA gene sequences. All clinical strains were multi-drug-resistant or extensively drug-resistant with different antibiotic resistance patterns. Clinical strains were isolated from a patient with healthcare-associated infections (respiratory tract, blood, urine) from Lviv regional hospitals. All testing was repeated triplicate. All biofilm-forming strains had the pox gene, which is responsible for the formation of biofilms.

Results and discussion

Synthesis

Reaction of sodium or potassium S-salts of substituted sulfonothioic acids with different alkylating agents is the main method of synthesis of sulfonothioic acids S-esters. The reaction can be conducted in various polar solvents such as acetone or acetonitrile or even water [15], but the usage of DMF is convenient due to the fast and clean course of the reaction in mild conditions. The alkylation of (1, 2) was carried out in dry, freshly distilled DMF at room temperature for 2 hours with 1.5 excess of alkylating agent (Scheme 1).

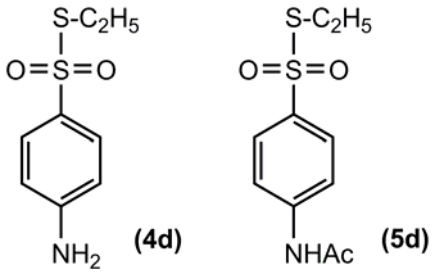
Under these conditions, products of N-alkylation were not found in the reaction mixture. Excess of the alkylating agents was removed by the addition of diethanolamine with the formation of water-soluble compounds. This was done in anticipation of problems with the crystallization of products and the difficulty of removing the relatively high-boiling alkylating agents from the obtained oils. After the aqueous workup



Scheme 1. Synthesis of the new S-(ω-fluoralkyl) 4-substituted benzenesulfonothioates

followed with the filtration of the DCM extract through thin silica gel layer and removing the solvent *in vacuo*, the products were sufficiently clean by LC-MS (93–95%), but for biological studies were additionally purified by column chromatography on silica gel. After preparative chromatography obtained products had LC-MS purity >98%. Due to rather low melting point and very slow crystallization, even crystalline products (4a, 5a–c) cannot be effectively purified by recrystallization.

Also with the same method, two known compounds with high antimicrobial activity (S-ethyl 4-aminobenzenesulfonothioate (4d) and S-ethyl 4-acetamidobenzenesulfonothioate (5d) (Figure) were synthesized as comparative compounds for the biological studies. These compounds are in fact «defluorinated» analogues of compounds (4a) and (5a), respectively.



S-(alkyl) 4-substituted benzenesulfonothioates with known antimicrobial activity

Structure of the products was confirmed by ^{1}H NMR spectroscopy and mass spectrometry. In ^{1}H-NMR spectra of amino derivatives (4a–c) protons of *p*-substituted aromatic ring are observed as a typical broad AB system at 7.5–6.6 ppm with SSCC ≈8.8 Hz. However, in case of N-acetyl derivatives (5a–c), they are observed as two very close doublets at 7.8 ppm (Δδ≈0.04 ppm), indicating almost the same electron effects of N-acetyl and $-SO_2S-$Alk groups on aromatic ring. For amino derivatives (4a–c), protons of $-NH_2$ group are observed as a singlet at 6.4 ppm, while for N-acetyl derivatives (5a–c), amide proton is observed at 10.4 ppm. Presence of terminal fluorine atom in all compounds is confirmed by the observation of H–F coupling constants (≈5.7 Hz) in $-CH_2F$ groups which protons are observed as two triplets at 4.58–4.30 ppm. MS spectra of obtained compounds show expected molecular masses and isotopic distribution.

Antimicrobial activity

The study of the antimicrobial activity of six new compounds was carried out using the agar diffusion method using reference and clinical strains of fungi, Gram-positive and Gram-negative bacteria. For Gram-negative bacteria (Table 1), it demonstrated that all compounds were highly active against clinical strain *Pseudomonas aeruginosa* N197 (up to 30.9 mm inhibition zone for 4c), moderately active against reference strain *Raoultella ornithinolytica* DSM 7464 and almost inactive against clinical strain *Pseudomonas luteola* N198 and reference strain *Pseudomonas aeruginosa* ATCC 10145. Only one compound (4a) showed moderate activity against *Klebsiella varicola* 3958 clinical strain almost at the same level as its known «defluorinated» analog (4d). In addition, all compounds were active against wild non-pathogenic strains *Lactobacillus paracasei* L10 and L11.

For Gram-positive bacteria (Table 2), all studied compounds were highly active against *Staphylococcus aureus* biofilm-forming Nb2 (>30 mm) and moderately active against other studied strains: *Staphylococcus aureus subsp. aureus* ATCC 25923 biofilm-forming, *Staphylococcus aureus* biofilm-forming Nb5 and *Staphylococcus epidermidis* ATCC 12228 non-biofilm forming.

The study of the antifungal activity (Table 3) demonstrated that all compounds are highly active against both reference and clinical *Candida albicans* strains as well as against *Aspergillus niger*

Table 1

Zones of growth inhibition of Gram-negative bacteria

Substance	Zone of growth inhibition (mm±SE)						
	Reference strains		Clinical strains			Wild non-pathogenic strains	
	Pseudomonas aeruginosa ATCC 10145	*Raoultella ornithinolytica DSM 7464*	*Pseudomonas aeruginosa* N197	*Pseudomonas luteola* N198	*Klebsiella varicola 3958*	*Lactobacillus paracasei* L10	*Lactobacillus paracasei* L11
4a	9.0	13.2	22.6	13.0	10.0±0.2	19.7	24.4
5a	10.0	12.0	18.9	13.0	00.0	15.2	18.9
4b	9.0	8.0	22.4	13.0	00.0	22.1	25.4
5b	10.0	9.0	19.0±0.2	17.0	00.0	19.6	20.9
4c	10.0	7.0	30.9±0.2	14.0	00.0	24.3	25.5
5c	10.0	9.0	25.0	13.0	00.0	20.5	19.3
4d	9.0	13.3	31.5±0.2	19.7	11.0±0.2	26.7	32.0
5d	9.0	11.0	20.6±0.2	13.0	00.0	21.4	24.9
DMSO	9.0	00.0	00.0	13.0	00.0	9.5	9.5
C1*	–	–	–	–	–	–	–
C2*	>35.0±0.3	–	<20.0±0.2	<20.0±0.2	<14.0±0.4	>35.0±0.3	>35.0±0.3

Note: * – C1 stands for Vancomycin, and C2 stands for Ciprofloxacin.

Table 2

Zones of growth inhibition of Gram-positive bacteria

Substance	Zone of growth inhibition (mm±SE)			
	Reference strains			
	Staphylococcus aureus subsp. aureus ATCC 25923 biofilm-forming	*Staphylococcus epidermidis* ATCC 12228 non-biofilm forming	*Staphylococcus aureus* biofilm-forming Nb2	*Staphylococcus aureus* biofilm-forming Nb5
4a	12.0±0.2	12.0	>30.0±0.2	15.0
5a	19.3±0.2	17.8	>30.0±0.2	10.0
4b	19.3±0.2	15.4	>30.0±0.2	17.9
5b	13.1±0.2	12.0	>30.0±0.2	12.9±0.2
4c	18.1±0.2	22.8	>30.0±0.2	16.4±0.2
5c	16.3±0.2	11.3	>30.0±0.2	14.1
4d	26.7±0.2	24.5	>30.0±0.2	27.4±0.2
5d	12.0±0.2	13.1	>30.0±0.2	14.1±0.2
DMSO	00.0	00.0	00.0	00.0
C1*	32.0±0.5	>30.0±0.5	<11.5±0.3	–
C2*	35.0±0.5	>30.0±0.5	<10.0±0.2	–

Note: * – C1 stands for Vancomycin, and C2 stands for Ciprofloxacin.

(26.5–36.4 mm). These compounds directly inhibited the growth of *Aspergillus* rather than delaying the maturation of spores.

For the most sensitive strains, minimal inhibitory concentration (MIC) values (mM) of synthesized compounds were determined (Table 4).

All compounds showed MIC less than 1 mM against *Candida albicans* (ATCC 885-653), two compounds (5a and 4c) showed slightly higher activity (0,22 and 0,23 mM respectively) than known compound 5d (0,24 mM). Synthesized compounds were less active (0.47–1.6 mM) against *Pseudomonas aeruginosa* N197 than known 4d (0.28 mM); in general, amino derivatives (4a–d) were more active than N-acylated derivatives (5a–d). Only compound 5a showed high activity (0.45 mM) against *Staphylococcus aureus* biofilm-forming Nb2, other compounds showed MIC in the range of 2.0–3.7 mM.

It can be observed that synthesized compounds

Table 3

Zones of growth inhibition of Gram-positive bacteria

Substance	Zone of growth inhibition (mm±SE)		
	Fungi		
	Reference strains		Clinical strains
	Candida. albicans (ATCC 885-653)	*Aspergillus niger*	*Candida albicans* N 85
4a	31.0±0.2	28.0±0.2	27.2±0.2
5a	29.6±0.2	28.0±0.2	27.3±0.2
4b	30.0	32.0	24.5
5b	30.2±0.3	36.4±0.5	27.9±0.2
4c	28.5±0.3	36.4±0.5	29.1±0.2
5c	32.2	36.4±0.5	25.0
4d	30.7±0.3	36.4±0.5	28.7±0.2
5d	32.7±0.3	36.4±0.5	26.7±0.2
DMSO	10.5	00.0	9.5
C3*	–	00.0	<6.0±0.2

Note: * – C3 stands for Clotrimazole.

Table 4

MIC value of compounds against bacterial and fungi species

Strain	MIC, mM							
	4a	5a	4b	5b	4c	5c	4d	5d
Staphylococcus aureus biofilm-forming Nb2	2.12	0.45	2.00	3.40	3.70	3.20	0.58	0.48
Pseudomonas aeruginosa N197	0.53	0.90	0.50	1.70	0.48	1.60	0.28	0.48
Lactobacillus paracasei L10	0.13	0.90	0.25	0.85	0.47	1.70	0.035	0.24
Candida albicans (ATCC 885-653)	0.27	0.22	0.98	0.85	0.23	0.81	0.28	0.24

are less active (MIC>0.13 mM) against *Lactobacillus* which are part of the normal non-pathogenic microbiota than known compound 4d with MIC 0.035 mM, and N-acylated derivatives (5a–d) are generally less active than amino derivatives (4a–d).

Conclusions

Six new S-(ω-fluoralkyl) 4-amino- (4a–c) and 4-acetamidobenzenesulfonothioates (5a–c) were synthesized by the reaction of sodium salts of the corresponding thiosulfonic acids and ω-fluoralkylbromides: 1-bromo-2-fluoroethane, 1-bromo-3-fluoropropane and 1-bromo-4-fluorobutane. The study of their antimicrobial activity against thirteen reference and clinical strains of fungi, Gram-positive and Gram-negative bacteria was carried out using agar diffusion method.

The study of the antimicrobial activity using the agar diffusion method showed that all synthesized compounds were highly active against *Pseudomonas aeruginosa* clinical strain and *Staphylococcus aureus* biofilm-forming Nb2 strain, moderately active against other studied *Staphylococcus aureus* strains and *Raoultella ornithinolytica,* and almost inactive against *Pseudomonas luteola* and *Pseudomonas aeruginosa* ATCC 10145 reference strain. Only one synthesized compound (4a) showed moderate activity against *Klebsiella varicola* (10.0 mm). All compounds were highly active against both reference and clinical *Candida albicans* strains as well as against *Aspergillus niger*.

For the most sensitive strains, minimal inhibitory concentrations with resazurin-based microdilution assay was determined. All compounds showed significant antimicrobial activity against *Pseudomonas aeruginosa* N197 (0.48–1.70 mM), *Staphylococcus aureus* biofilm-forming Nb2 (0.45–3.70 mM) and *Candida albicans* (0.22–0.98 mM); two of them (5a and 4c) showed slightly higher activity against *Candida albicans* than known antifungal compound of thiosulfonate class: S-ethyl ester of 4-aminobenzenethiosulfonic acid (4d) (0.24 mM).

It can be concluded that introduction of terminal fluorine atom does not significantly change the profile of antimicrobial activity of S-(alkyl) 4-substituted benzenesulfonothioates. The synthesized ω-fluorinated compounds (4a–c, 5a–c) as well as reference compounds 4d and 5d non-selectively inhibited the growth of both Gram-positive and Gram-negative

microbiota, as well as fungi, they directly inhibited the growth of *Aspergillus* rather than delaying the maturation of spores.

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СИНТЕЗ НОВИХ S-(ω-ФТОРАЛКІЛ) 4-ЗАМІЩЕНИХ БЕНЗОЛСУЛЬФОНОТІОАТІВ ТА ЇХ АНТИМІКРОБНА АКТИВНІСТЬ

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Нові S-(ω-фторалкіл) 4-аміно- та 4-ацетамідобензолсульфонотіоати були синтезовані реакцією натрієвих солей заміщених тіосульфокислот з відповідними ω-фторалкілбромідами. Здійснено дослідження їх антимікробної активності проти тринадцяти еталонних і клінічних штамів бактерій і грибів методом дифузії в агарі. Усі сполуки виявили значну антимікробну активність, дві з них показали вищу активність щодо *Candida albicans*, ніж відома протигрибкова сполука класу тіосульфонатів – S-етиловий ефір 4-амінобензолтіосульфонової кислоти.

Ключові слова: сульфонотіоати, фторалкілброміди, антибактеріальна активність, протигрибкова активність, клінічні штами.

SYNTHESIS OF NEW S-(ω-FLUORALKYL) 4-SUBSTITUTED BENZENESULFONOTHIOATES AND THEIR ANTIMICROBIAL ACTIVITY

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New S-(ω-fluoralkyl) 4-amino- and 4-acetamidobenzenesulfonothioates were synthesized by the reaction of sodium salts of the substituted thiosulfonic acids and corresponding ω-fluoralkylbromides. The study of their antimicrobial activity against thirteen reference and clinical bacterial and fungal strains using agar diffusion method was conducted. All compounds showed significant antimicrobial activity, two of them showed higher activity against Candida albicans than known antifungal compound of sulfonothioate class, S-ethyl ester of 4-aminobenzenethiosulfonic acid.

Keywords: sulfonothioates; fluoralkylbromides; antibacterial activity; antifungal activity; clinical strains.

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