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New polyfunctionalized 2-hydrazinoanthraquinone derivatives as potential antimicrobial agents

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Aim. Synthesis and study of new polyfunctionalized 2-hydrazinoanthraquinone derivatives as potential antimicrobial agents. **Methods.** Organic synthesis, NMR and LC-MS spectroscopy, agar diffusion and broth microdilution methods. **Results.** A series of anthraquinonehydrazone derivatives are synthesized using the reaction of 2-(morpholinodiazenyl)anthracene-9,10-dione with methylene active compounds in the acetic acid medium. The screening of antimicrobial activity identified the compounds with significant effects against the tested microorganisms with MIC value <186.9 μM. Compounds 5 and 11 with MIC <93.5 μM are effective against yeast fungi whereas compound 5 with MIC <186.9 μM is effective against *P.putida*, which is multidrug resistant to antibiotics. **Conclusions.** The obtained hydrazino-anthraquinone derivatives constitute an interesting background for the design of new synthetic agents with antimicrobial activity.

Keywords: anthraquinones, methylene active compounds, 4-thiazolidinones, antimicrobial activity

Introduction

Anthraquinone derivatives are one of the key quinone molecular platforms, characterized by a strong synthetic and pharmacological potential due to their high reactivity and molecular

affinity to various bio targets. Thus, the planar structure of the aromatic anthracene-9,10-dione core provides its ability for DNA intercalation [1], which was the basis of the construction of several anticancer drugs, in particular daunorubicin, adriamycin and mitoxantrone. Additionally, 9,10-anthraquinone derivatives have been identified as potent antimicrobial [2], antioxidant [3], laxative [4], and anti-inflammatory [5] agents. Anthraquinones are also the building blocks of many dyes, electrolytes in flow batteries and digester additives in papermaking [6]. Significant pharmacological interest in anthraquinone derivatives is also based on the possibility of combination with other pharmacophore fragments, thus providing various biologically active compounds. Examples of pharmacophores are thiazole/thiazolidinone derivatives and their structure analogs, which display anticancer [7, 8], antimicrobial [9, 10], antiviral [11], anti-inflammatory [12, 13], and antitrypanosomal activities [14].

In our previous works, we reported the synthesis of 1-hydrazinoanthraquinones starting from 1-(morpholinodiazenyl)anthracene-9,10-dione and various methylene active compounds (MACs), e.g. thiazolidinones and their analogs, Meldrum's acid, acetylacetone, β-dinitriles, *etc.* [15]. Some of the mentioned hybrid molecules displayed significant anticancer and moderate antimicrobial activities [16].

Thus, the aim of our work was the design and synthesis of isomeric polyfunctionalized 2-hydrazinoanthraquinones containing thiazolidine, hydantoin, and other MACs, and evaluation of their antimicrobial activity (Figure 1).

Materials and Methods

Chemistry

All materials were purchased from Sigma-Aldrich and used without purification. The uncorrected melting points were determined in open capillary tubes using the Cole-Parmer

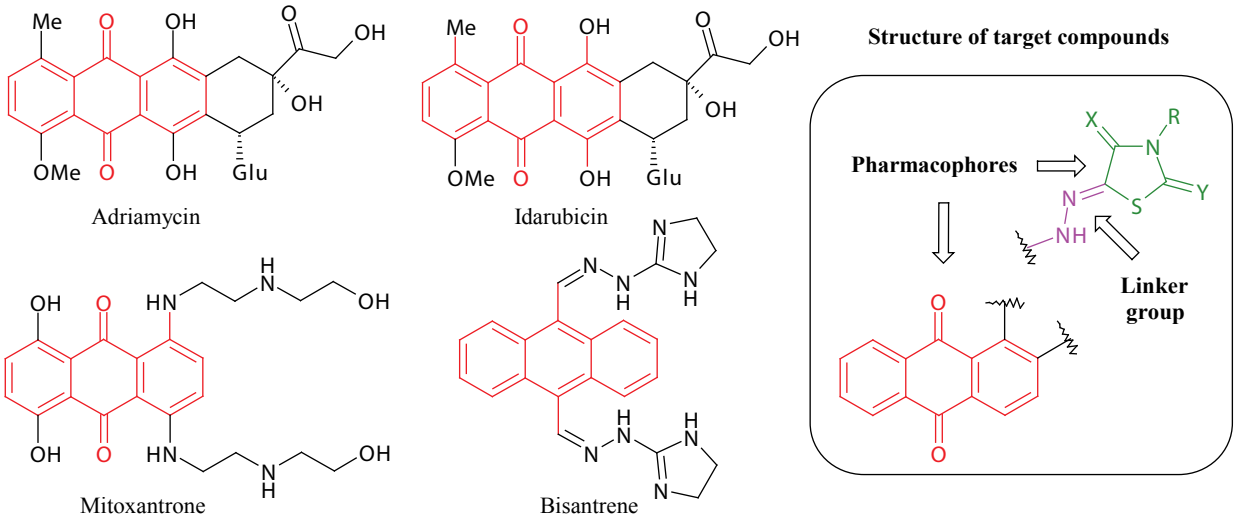


Fig. 1. Background for targeted compound synthesis.

IA9200 Melting Point Apparatus. Thermo Scientific's FlashSmart Elemental Analyzer was used to conduct the elemental evaluations. The Varian Gemini (^{1}H NMR at 400 MHz) instrument was used to record the ^{1}H NMR spectra in DMSO-d_6. The chemical shifts (δ) are given in the ppm units relative to TMS as reference (0.00). The Finnigan MAT INCOS-50 instrument was used to record the LC-MS spectra. The starting 2-(morpholinodiazenyl) anthracene-9,10-dione was prepared according to the reported method [16].

General procedure for synthesis of polyfunctionalized 2-hydrazinoanthraquinones 1–11

A mixture of 2-(morpholinodiazenyl)anthracene-9,10-dione (10 mmol), appropriate methylene active compound (10 mmol), and sodium acetate (10 mmol, for the synthesis of compounds **1, 5** and **6**) in acetic acid (20 mL) was heated under reflux for 30 min, then cooled, and the obtained solid was filtered, washed by water, absolute ethanol and ethoxyethane, dried and crystallized from a mixture dimethylformamide : ethanol (1:2).

5-(2-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)thiazolidine-2,4-dione **(1)**. Yield: (72 %); m.p.: 154–156 ºC. ^{1}H NMR: δ 7.86–7.90 (m, 3H, arom.), 8.07–8.14 (m, 4H, arom.), 8.35 (s, 1H, NH), 12.00 (s, 1H, NH). Anal. calcd. for ($C_{17}H_9N_3O_4S$): C, 58.12; H, 2.58; N, 11.96. Anal. found: C, 58.28; H, 2.40; N, 11.83. LC-MS: *m/z* 352 $[M+H]^+$.

2-(2-(4-Oxo-2-thioxothiazolidin-5-ylidene) hydrazineyl)anthracene-9,10-dione **(2).** Yield: (68 %); m.p.: 152–154 ºC. ^{1}H NMR: δ 7.60 (m, 1H, arom.), 7.86 (m, 2H, arom.), 8.03 (s, 1H, arom.), 8.12 (m, 2H, arom.), 8.23 (m, 1H, arom.), 10.98 (s, 1H, NH), 12.93 (s, 1H, NH). Anal. calcd. for ($C_{17}H_9N_3O_3S_2$): C, 55.58; H, 2.47; N, 11.44. Anal. found: C, 55.70; H, 2.38; N, 11.32. LC-MS: *m/z* 368 $[M+H]^+$.

3-(5-(2-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)-4-oxo-2-thioxothiazolidin-3-yl)propanoic acid **(3)**. Yield: (78 %); m.p.: 154–156 ºC. ^{1}H NMR: δ 2.48 (m, 2H, CH_2), 3.29 (m, 2H, CH_2), 7.89–7.91 (m, 2H, arom.), 7.90–7.93 (m, 2H, arom.), 8.08 (d, 1H, J = 8.6 Hz, arom.), 8.14 (s, 1H, arom.), 8.20 (d, 1H, J =8.2 Hz, arom.), 9.31 (s, 1H, NH). Anal. calcd. for ($C_{20}H_{13}N_3O_5S_2$): C, 54.66; H, 2.98; N, 9.56. Anal. found: C, 54.78; H, 3.11; N, 9.40. LC-MS: *m/z* 440 $[M+H]^+$.

2-(2-(4-Amino-2-oxothiazol-5(2H)-ylidene) hydrazineyl)anthracene-9,10-dione **(4).** Yield: (83 %); m.p.: >240 ºC. ^{1}H NMR: δ 7.61 (d, 1H, J = 8.4 Hz, arom.), 7.87 (m, 2H, arom.) 7.93 (s, 1H, arom.), 8.12–8.14 (m, 3H, arom.), 9.29 (s, 2H, NH_2), 10.81 (s, 1H, NH). Anal. calcd. for ($C_{17}H_{10}N_4O_3S$): C, 58.28; H, 2.88; N, 15.99. Anal. found: C, 55.11; H, 3.02; N, 16.14. LC-MS: *m/z* 351 $[M+H]^+$.

5-(2-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)imidazolidine-2,4-dione **(5).** Yield: (69 %); m.p.: >240 ºC. ^{1}H NMR: δ 7.85–7.90 (m, 3H, arom.), 7.91 (d, 1H, J = 8.9 Hz, arom.), 8.10–8.20 (m, 3H, arom.), 8.32 (s, 1H, NH), 8.81 (s, 1H, NH), 10.54 (s, 1H, NH). Anal. calcd. for ($C_{17}H_{10}N_4O_4$): C, 61.08; H, 3.02; N, 16.76. Anal. found: C, 61.21; H, 2.82; N, 16.54. LC-MS: *m/z* 335 $[M+H]^+$.

5-(2-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)-2,2-dimethyl-1,3-dioxane-4,6-dione **(6)**. Yield: (82 %); m.p.: >240 ºC. ^{1}H NMR: δ 2.39 (s, 6H, 2*Me), 7.80–7.95 (m, 5H, arom), 8.23 (d, 2H, J = 8.2 Hz, arom.), 10.18 (s, 1H, NH). Anal. calcd. for ($C_{20}H_{14}N_2O_6$):

C, 63.49; H, 3.73; N, 7.40. Anal. found: C, 63.61; H, 3.52; N, 7.54. LC-MS: *m/z* 379 $[M+H]^+$.

5-(2-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)-2-thioxodihydropyrimidine-4,6(1H,5H)-dione **(7).** Yield: (84 %); mp >240 ºC. ^{1}H NMR: δ 7.93–7.95 (m, 2H, arom.), 8.08 (dd, 1H, *J* = 1.8, 8.5 Hz, arom.), 8.21–8.23 (m, 2H, arom.), 8.28 (d, 1H, *J* = 8.5 Hz, arom.), 8.37 (s, 1H, arom.), 12.55 (s, 1H, NH), 12.70 (s, 1H, NH), 14.13 (br.s, 1H, NH). Anal. calcd. for ($C_{18}H_{10}N_4O_4S$): C, 57.14; H, 2.66; N, 14.81. Anal. found: C, 57.31; H, 2.52; N, 15.00. LC-MS: *m/z* 379 $[M+H]^+$.

2-Cyano-2-(2-(9,10-dioxo-9,10-dihydroanthracen-2-yl)hydrazineylidene)acetic acid (**8**). Yield: (77 %); m.p.: >240 ºC. ^{1}H NMR: δ 7.89–7.91 (m, 2H, arom.), 8.03 (s, 1H, arom.), 8.13 (d, 1H, *J* = 8.6 Hz, arom.), 8.17–8.20 (m, 2H, arom.), 8.22 (d, 1H, *J* = 8.6 Hz, arom.), 12.20 (s, 1H, NH), 14.00 (s, 1H, COOH). Anal. calcd. for ($C_{17}H_9N_3O_4$): C, 63.95; H, 2.84; N, 13.16. Anal. found: C, 63.81; H, 2.99; N, 13.30. LC-MS: *m/z* 320 $[M+H]^+$.

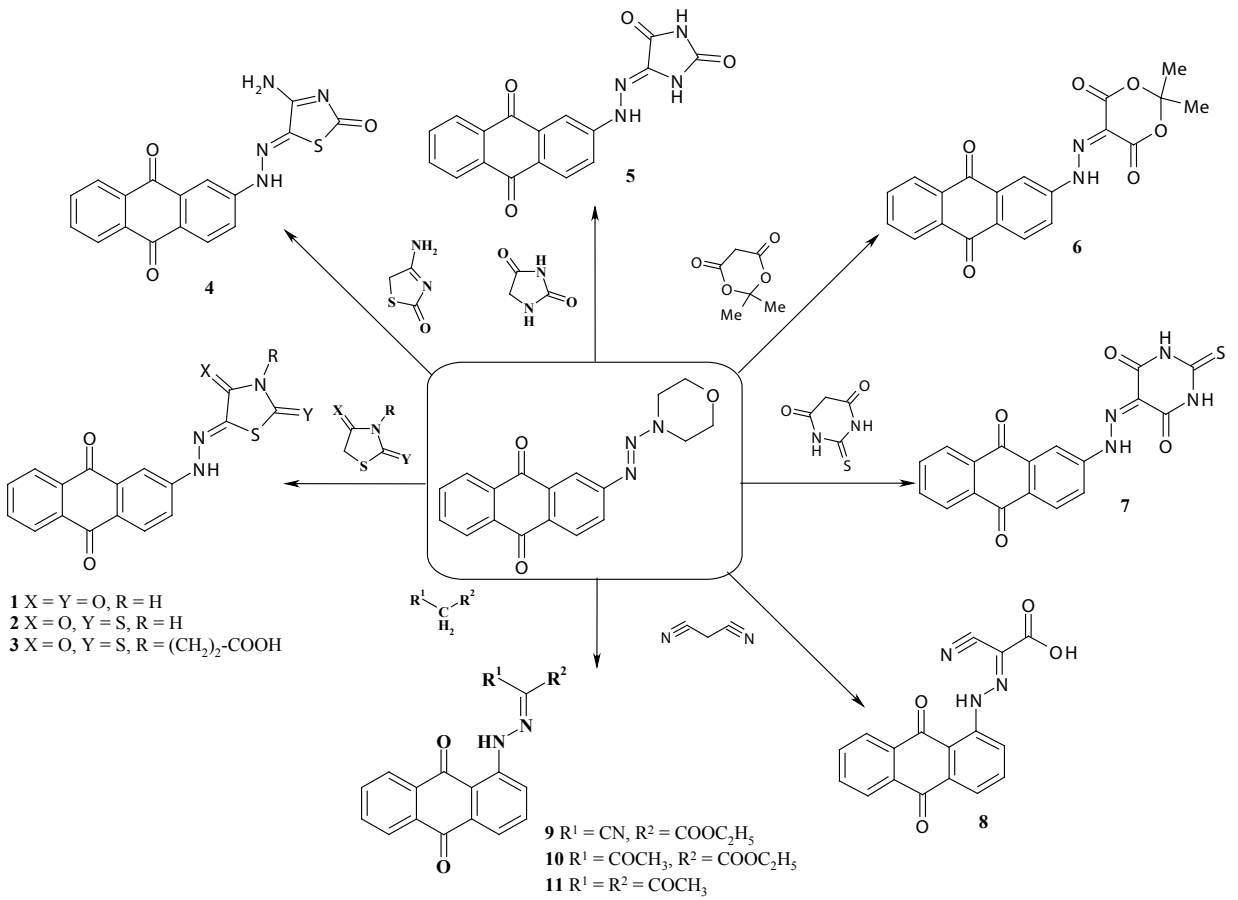
Ethyl 2-cyano-2-(2-(9,10-dioxo-9,10-dihydroanthracen-1-yl)hydrazineylidene)acetate (**9**). Yield: (70 %); m.p.: 158–160 ºC. ^{1}H NMR: 1.33 (t, 3H, *J* = 7.1 Hz, Me,), 4.33 (q, 2H, *J* = 7.3 Hz, CH_2), 7.90–7.91 (m, 3H, arom.), 8.18–8.20 (m, 3H, arom.), 8.34 (s, 1H, arom.), 9.46 (s, 1H, NH). Anal. calcd. for ($C_{19}H_{13}N_3O_4$): C, 65.70; H, 3.77; N, 12.10. Anal. found: C, 65.87; H, 3.98; N, 11.99. LC-MS: *m/z* 348 $[M+H]^+$.

Ethyl 2-(2-(9,10-dioxo-9,10-dihydroanthracen-1-yl)hydrazineylidene)-3-oxobutanoate (**10**). Yield: (72 %); m.p.: 160–162 ºC. ^{1}H NMR: 1.31 (t, 3H, *J* = 7.0 Hz, Me), 2.41 (s, 3H, Me), 4.37 (q, 2H, *J* = 7.0 Hz, CH_2), 7.71 (d, 1H, *J* = 8.1 Hz, arom.), 7.85–7.86 (m, 2H, arom.), 7.93 (s, 1H, arom.), 8.07–8.10 (m, 3H, arom.), 11.63 (s, 1H, NH). Anal. calcd. for ($C_{20}H_{16}N_2O_5$): C, 65.93; H, 4.43; N, 7.69. Anal. found: C, 66.07; H, 4.68; N, 7.89. LC-MS: *m/z* 365 $[M+H]^+$.

1-(2-(2,4-Dioxopentan-3-ylidene)hydrazineyl)anthracene-9,10-dione **(11)**. Yield: (68 %); m.p.: 150–152 ºC. ^{1}H NMR: δ 3.91 (s, 6H, 2*Me), 7.82 (d, 1H, *J* = 8.4 Hz, arom.), 7.89–7.94 (m, 2H, arom.), 8.08 (s, 1H, arom.), 8.17–8.23 (m, 4H, NH, arom.). Anal. calcd. for ($C_{19}H_{14}N_2O_4$): C, 68.26; H, 4.22; N, 8.38. Anal. found: C, 68.04; H, 4.11; N, 8.58. LC-MS: *m/z* 335 $[M+H]^+$.

Antimicrobial activity

Using the agar diffusion and serial dilution resazurin-based microdilution tests (RBMA), the synthesised compounds were evaluated *in vitro* for their antibacterial and antifungal properties [17]. For this, a 5.5 mm-diameter agar well was filled with 100 µL (1 mg/mL) of the tested compound. Using a calipers, the diameter of the growth retardation was determined with a 0.1 mm error. As a control, the discs containing dimethyl sulfoxide (DMSO), vancomycin, ciprofloxacin, and clotrimazole were employed. Due to limited solubility of the test chemicals in diluted DMSO, pure DMSO was used as a solvent. Additionally, Petri dishes were incubated at 37 °C for 24 hours for bacteria and at 25 °C for 24–48 hours for fungi using Mueller-Hinton agar and Saburo agar (for fungi). The RBMA method required the addition of 15 mL of 0.02 % resazurin to each well of a 96-well plate along with 50 µl of nutritive medium (Mueller-



Scheme 1. Synthesis of polyfunctionalized 2-hydrazinoanthraquinones **1–11**.

Hinton broth or glucose broth), 50 μl of the suspension of the microbe (McFarland 2.0), and 100 μl of the tested compounds. The MALDI TOF method (Bruker, Bremen, Germany) and 16S rRNA gene sequences were utilised to identify the reference and clinical microbial and fungal strains (Table 1). All clinical isolates had various patterns of antibiotic resistance and were either extensively drug-resistant (EDR) or multidrug-resistant (MDR). A patient with healthcare-associated infections (HAIs) from local hospital had clinical strains isolated from them [18].

Results

Chemistry

As a continuation of our studies on polyfunctionalized anthraquinonehydrazones [15, 16, 19, 20] we performed the reaction of isomeric 2-(morpholinodiazenyl)anthracene-9,10-dione with methylene active compounds to give the corresponding hydrazine derivatives **1–11** (Scheme 1). The reaction is carried out in acetic acid medium. In the case of synthesis of compounds **1**, **5** and **6** using methylene active compounds with weak CH-acidity, name-

ly thiazolidine-2,4-dione, 4-amino-2-oxo-2*H*-thiazole and imidazolidine-2,4-dione, anhydrous sodium acetate was added as a catalyst to enhance deprotonation of an active methylene group. Noteworthy, the interaction of morpholinetriazene antraquinone with malononitrile under the similar reaction conditions is accompanied with hydrolysis of the nitrile group to the carboxyl group providing derivative **8**.

The structure and purity of synthesized compounds **1–11** were confirmed by the ^{1}H NMR and LC-MS spectral data. Thus, in the ^{1}H NMR spectra of the synthesized com-

Table 1. In vitro **antimicrobial activity of synthesized compounds (zone of growth inhibition at conc. 1 mg/mL after 24–48 h)**

Compound	Zone of growth inhibition (mm ± SE)*									
	Gram-negative bacteria					Gram-positive bacteria			Fungi	
	Reference strains		Clinical strains			Reference strains	Clinical strains		Reference strains	Clinical strains
	Pseudomonas aeruginosa (ATCC 27853 (F-51))	*Raoultella terrigena* (ATCC 33257)	*Pseudomonas putida* N 182	*Escherichia coli* N 168	*Proteus mirabilis* N 113	*Staphylococcus aureus* (ATCC 25923 (F-49))	*Staphylococcus aureus* N 23	*Enterococcus faecalis* N 26	*Candida. albicans* (ATCC 885–653)	*Candida albicans* N 139
1	00	00	00	00	00	00	6.9± 0.3	00	00	00
2	00	00	00	00	00	00	00	00	00	7.1±0.3
3	00	00	00	12.2± 0.2	00	00	00	00	00	11.2± 0.3
4	00	00	12.1± 0.2	00	00	00	00	00	00	00
5	00	00	12.2± 0.2	00	00	00	00	00	00	11.2± 0.3
6	00	00	00	00	00	00	6.4± 0.3	00	00	00
7	00	00	00	9.9± 0.3	00	00	00	10.2 ± 0.4	00	00
8	00	00	00	00	00	00	00	00	00	00
9	00	00	00	00	00	00	00	00	00	00
10	00	00	00	00	00	00	00	00	00	00
11	00	00	00	00	00	00	00	00	00	11.4±0.2
DMSO	00	00	00	00	00	00	00	00	00	00
Vancomycin	-	-	-	-	-	32.0 ± 0.5	11.4 ± 0.3	9.0 ± 0.2	-	-
Ciprofloxacin	35.0 ± 0.3	30.0 ± 0.5	10.0 ± 0.2	14.0 ± 0.4	16.0 ± 0.3	35.0 ± 0.5	9.0 ± 0.2	10.0 ± 0.2	-	-
Clotrimazole	-	-	-	-	-	-	-	-	18.0 ± 0.5	11.0 ± 0.3

* Results shown with significantly difference from the control of the solvent (DMSO); 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*, 30–40 mm for *E.coli*); Clotrimazole 10 μg (inhibition zone 12–17 mm for *Candida spp*; d of well 5.5 mm

pounds, the signals of NH proton of the arylhydrazo group were assigned at 8.20–12.55 ppm. Characteristic aromatic protons of the anthraquinone fragment of the synthesized compounds are manifested by the signals between 7.60–8.37 ppm. The formation of compound **8** as a product of spontaneous hydrolysis of the nitrile group to carboxyl was confirmed by the ^{1}H NMR and LC-MS spectral data. In the ^{1}H NMR spectra of compound **8** the signal of carboxyl group appears as broad singlet at d 14.00 ppm. The molecular ion peak observed at *m/z* value of 319.0 $[M + H]^+$ in the positive ionization mode in the mass spectrum confirmed the formation of the target 2-cyano-2-(2-(9,10-dioxo-9,10-dihydroanthracen-2-yl) hydrazineylidene)acetic acid **8**.

Antimicrobial activity

According to the screening of antimicrobial activity of polyfunctionalized 2-hydrazinoanthraquinones (Table 1), the highest activity was shown by compounds **3, 4, 5, 7, 11**. Thus, the best activity among mentioned compounds was shown by compound **5** against extensively drug-resistant *P.putida* N182 and *C.albicans* N139. Compound **3** displayed the activity against multi-drug-resistant *E.coli* N168 and *C.albicans* N139, similar to compound **7,** which showed the activity against *E.coli* N168 and *E.faecalis* N26. Compound **4** possessed the activity against *P.putida* N182. Compound **11** showed the selective activity against *C.albicans* N139. The tested compounds did not possess the significant antimicrobial activity against Gram-positive microorganisms, except for compound **7,** which was active against *E.faecalis* N26.

Five hit-compounds (Table 2) were tested for the minimum inhibitory concentration (MIC). The best activity showed compounds **5** and **11** against *C.albicans* N139 (93.5 μM), and compound **5** against *P.putida* N182 (186.9 μM), which corresponded to the clotrimazole and ciprofloxacin result.

In general, the results of screening antimicrobial activity of 2-hydrazinoanthraquinones revealed an absence of correlation of this type of biological activity with the structure of the CH-acid fragment of the studied compounds. In particular, the anthraquinone-hydrazone conjugates based on 4-thiazolidinone and its structural analogs showed no appreciable antimicrobial activity, except for compound **5**, which contains an imidazolidine fragment. On the other hand, a number of polyfunctional anthraquinone hydrazones based on some heterocyclic compounds, in particular, Meldrum's

Table 2. **MIC value (μM) of compounds 3–5, 7 and 11 against bacterial and yeast species**

Strain/Compound	μM						
	3	**4**	**5**	**7**	**11**	Ciprofloxacin	Clotrimazole
Pseudomonas putida N 182	00	00	<186.9	-	-	<188.6	-
Escherichia coli N 168	00	-	-	00	-	<93.5	-
Enterococcus faecalis N 26	-	-	-	00	-	<93.5	-
Candida albicans N 139	00	-	<93.5	-	<93.5	-	< 90.6

00 — results with no significant difference from the control of the solvent (DMSO); - not tested

Table 3. **Physicochemical and pharmacokinetic properties of the hit-compounds 3–5, 7 and 11.**

	3	**4**	**5**	**7**	**11**
Physicochemical properties					
Molecular weight	439.46	350.35	334.29	378.36	334.33
Num. heavy atoms	30	25	25	27	25
Num. arom. heavy atoms	12	12	12	12	12
Num. rotatable bonds	5	2	2	2	4
Num. H-bond acceptors	6	5	5	5	5
Num. H-bond donors	2	2	3	3	1
Molar Refractivity	118.83	98.14	94.46	106.67	92.37
TPSA Å²	173.53	139.28	116.73	148.82	92.67
Consensus log Po/w	2.35	1.94	1.04	1.24	2.54
Lipinski Rule	Yes	Yes	Yes	Yes	Yes
Pharmacokinetics					
GI absorption	Low	High	High	Low	High
BBB permeant	No	No	No	No	No
P-gp substrate	No	No	No	No	No
Log Kp (SP) (cm/s) (skin permeation)	–6.42	–6.31	–6.78	–6.71	–5.32
Bioavailability Score	0.11	0.55	0.55	0.55	0.55

acid and thiobarbituric acid (compounds **6** and **7**), and some alicyclic methylene active compounds (compound **11**), showed a moderate level of antimicrobial activity against some reference and clinical strains of microorganisms.

Molecular and pharmacokinetic properties

The molecular and pharmacokinetic properties of hit-compounds **3–5**, **7** and **11** were calculated using the SwisAdme online server of the Swiss Institute of Bioinformatics [21]. According to these data, all compounds are suited to drug-likeness parameters, and may exhibit high gastrointestinal absorption (except **3** and **7**), although do not cross the blood-brain barrier. Since they were not shown to be P-glycoprotein substrates in the tests, the polyfunctionalized 2-hydrazinoanthraquinones could not be connected to the drug's excretion.

The compounds indicated above may have minimal skin penetration across the cell membrane due to their negative skin permeability. All the predictive data allow considering the hit-compounds **3–5**, **7** and **11** as the prospective drug-like candidates for further in-depth studies (Table 3).

Discussion

Similarly to other quinones, anthraquinone-based derivatives manifest their pharmacological potential by implementing two mechanisms of biological activity [22, 23]. The first

is based on the redox properties of anthraquinones with further subsequent reduction and re-oxidation under physiological conditions, including the generation of reactive oxygen species (ROS), in particular, superoxide anion (O_2^-), hydroxyl radicals (•OH) and hydrogen peroxide (H_2O_2). The second mechanism, by which anthraquinones interact with biotargets is based on high electrophilicity, which allows them to form covalent bonds with nucleophilic agents, especially with thiol groups of glutathione, proteins, nucleophilic amino acid groups [24]. As a result, this functionality made it possible to identify various biologically active compounds among anthraquinones, including antimicrobial agents [15, 25].

In general, antimicrobial resistance (AMR) is a major issue in healthcare, and there is a direct correlation between the spread of AMR globally and the rise in the infectious disease mortality, particularly from the infections linked to healthcare. WHO forecasts that, by 2050, the AMR pathogens would surpass cancer as the leading cause of death globally, exceeding the total effect of all other causes of death. The WHO recommends the discovery of novel synthetic compounds with antimicrobial activity as one of the most effective short-term strategies for controlling AMR bacteria [26, 27].

One of other interesting groups of compounds with a polypharmacological profile, the methylene active compounds (4-thiazolidinone-based derivatives, hydantoin, *etc.*) have many reaction centers and are reasonably open to chemical modification/combination with other biophores. As a result, this class of chemical substances attracts keen attention in the search for the "drug-like molecules" with antimicrobial activity, effective against the diverse mechanisms of AMR, including HAIs infections] [28].

EDR *P.putida* and especially the strains resistant to carbapenems and fluoroquinolones are particularly dangerous for the health care system [29,30]. In our study, the tested compound 5 showed significant antimicrobial activity against *P.putida* with MIC like ciprofloxacin, which allows it to be considered as a potential drug-like molecule. *P.putida* is a rare pathogen that causes various infections [31], and our strain was isolated from a patient with HAI pneumonia, which is interesting for further research. *Proteus mirabilis* was an object of our experiment, because it is a common cause of community-acquired infections [32], was isolated as HAI urinary tract infection pathogen, and was multi-drug resistant.

Candida albicans is the most common fungal pathogen. Recently, the drug resistance of *C. albicans* is increasingly severe. The therapeutic potential of antifungal 4-thiazolidinone-based derivatives is a promising strategy to develop novel antifungal agents. In our study, the tested compounds **5** and **11** showed the significant antimicrobial activity against *C. albicans* with MIC like clotrimazole, which allows it to be considered as a potential drug candidate molecule.

Compound **5**, which showed both a selective effect on *P.putida* and an antifungal effect, provokes interest but the mechanism of its action requires further research. Compound **5** also demonstrated antimicrobial activity against planktonic forms *P.putida* and fungi, which may indicate a bacteriostatic mechanism of action, in particular on protein synthesis [33], but this too needs further study. The high

activity of derivative **5** is probably caused by the presence of an imidazolidine fragment in the structure, which is the basis of several highly active antimicrobial agents [34, 35]. Accordingly, the combination with another pharmacophore fragment, namely the anthraquinone core, make it possible to obtain the hit compound for further in-depth study.

Drug-likeness descriptors (Lipinski and Veber rules) for synthesized compounds were calculated with SwissADME. Thus, Lipinski's rule states that an orally active drug/compound should pass the following criteria: molecular weight (MW) < 500 Da, number of H-bond donors (HBDs) < 5, LogP < 5, number of H-bond acceptors (HBAs) < 10. Veber *et al.* identified two other relevant descriptors: number of rotatable bonds (NBR) < 10 and polar surface area (PSA) < 140 $Å^2$ [36]. SwissADME computational analysis of the tested polyfunctionalized 2-hydrazinoanthraquinones indicated no violations of these rules, suggesting that they would display well-behaved gastrointestinal absorption or permeation.

Conclusions

New polyfunctionalized 2-hydrazinoanthraquinones **1–11** have been synthesized with high yields via azo-coupling reaction using 2-(morpholinodiazenyl)anthracene-9,10-dione and methylene active compounds.

The preliminary results of antimicrobial activity allowed us to identify the active compound **5**, which has shown the best activity against extensively drug-resistant *P.putida* N182 and *C.albicans* N139. It revealed a potential for in-depth study of the mentioned compound for the construction of novel antimicrobial agents.

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Нові поліфункціональні похідні 2-гідразиноантрахінону як потенційні антимікробні агенти

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Мета. Синтез і дослідження нових поліфункціональних 2-гідразиноантрахінонів як потенційних антимікробних агентів. **Методи.** Органічний синтез, спектроскопія ЯМР та хромато-мас-спектрометрія, метод дифузії в агар та мікрометод серійних розведень. **Результати.** Взаємодією 2-(морфолінодіазеніл)антрацен-9,10-діону з метиленовими активними сполуками в середовищі оцтової кислоти синтезовано ряд похідних антрахінон-гідразону. Скринінг антимікробної активності виявив сполуки зі значним впливом на досліджувані мікроорганізми зі значенням МІК <186.9 мкМ. Сполуки 5 і 11 з МІК <93.5 мкМ ефективні проти дріжджових грибів, а сполука 5 з МІК <186.9 мкМ ефективна проти полірезистентної до антибіотиків *P.putida*. **Висновки.** Синтезовані похідні гідразиноантрахінону є цікавою основою для створення нових синтетичних агентів з антимікробною активністю.

Ключові слова: антрахінони, метилен активні сполуки, 4-тіазолідинони, антимікробна активність.

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