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THE EFFECTIVENESS OF SURFACTANTS AS COMPOUNDS FOR CREATING DISINFECTANTS WITH A WIDE SPECTRUM OF ACTION

Abstract

Along with the use of antibiotics, the use of disinfectants is crucial in the fighting against multi-resistant strains of bacteria that are dangerous not only for animals but also for humans. A new complex disinfectant could be used as a prevention method. Therefore, the aim of the work was to evaluate the effectiveness surfactants (anionic, cationic and non-ionic) as compounds for the creating of disinfectants with a wide spectrum of action. Compositions with guanidine-containing oligomer (GCO) inhibited S. aureus and C. albicans most effectively. At concentrations of 1000 and 100 ppm, 100 % of cells were inactivated. The biocidal effect against representatives of Gram-negative bacteria was weaker, which should be considered during creation of antimicrobial agents active against pathogenic strains of E. coli. With a rational combination of effective components and their synergism, it is possible to significantly reduce the concentration of the working solution compared to already used commercial disinfectants. For example, the concentration of glutaraldehyde in composition containing 3 % GCO, 1 % Triton X-100, 1 % Trilon B and 1 % glutaraldehyde was in 9 times lower than of Lysoformin 3000. Also, most surfactants used were not characterized by mutagenic activity, which is one of the main criteria for disinfectants usage since this reduces the risks of developing bacterial resistance to antibiotics and other biocides. Thus, the development and implementation of new disinfectants, which could help to fight against multi-resistant strains of bacteria, is an indispensable part of comprehensive programs in controlling and prevention of common diseases in animal husbandry and medicine, particularly, colibacillosis.

Keywords: anionic, cationic and non-ionic surfactants, bactericidal and fungicidal activity, disinfectants, pathogenic *E. coli.*

Introduction

Microorganisms with multiple drug resistance (MDR) are currently considered as a significant threat to the health care system, agriculture, veterinary medicine, and economy in general. According to WHO forecasts, infections caused by multiresistant strains of bacteria may lead to an increase in the number of unpreventable deaths in future [1,2]. It is assumed that over the next 30 years, the world economy may suffer in lack of more than 100 trillion dollars due to the spread of MDR strains of microorganisms. This primarily concerns low- and middle-income countries [1-3].

Despite the variety of vaccination programs, antibiotics are still being the most used strategy for prevention and treatment in the agricultural industry. For example, due to the growing selective pressure and irrational antibiotic use against pathogenic *E. coli*, the emergence of the antibiotic multiresistant phenomenon has limited

treatment options and increased public health concern. The potential transfer of MDR genetic determinants directly by contact and indirectly into the food chain, water, and manure, among others became possible [2,7]. Also, regarding the fact that MDR strains of microorganisms are becoming more and more common, it is very important to investigate the cause and origin of such cultures, as well as their possible habitats, to assess the potential threat originating from MDR microorganisms. In this context, disinfectants and antiseptics are important factors in the development of such microorganisms, since they are widely used not only in medical practice, industrial sector, but also in private households [4]. Moreover, the SARS-CoV-2 pandemic led to a significant increase in the use of disinfectants and antibiotics because it was one of the main widely distributed ways to fight against the disease [5,6].

In turn, this may pose a potential risk to human health, as it is shown that the exposure of disinfectants could be linked with antibiotic's resistance. For example, in some Gram-negative bacteria, resistance to benzalkonium chloride and chlorhexidine is linked with resistance to ampicillin, cefotaxime, sulfamethoxazole and ceftazidime, sulfamethoxazole, imipenem, cefotaxime, tetracycline, respectively [3,7].

One of the possible solutions to this problem is the creation of multi-component disinfectant, the individual substances of which, on the one hand, allow expanding the number of targets of the disinfectant, and on the other hand, are characterized by a synergistic effect. In addition, it is desirable that these agents prevent the initial attachment of bacteria to various abiotic surfaces and the subsequent formation of biofilm as a result totally killed the bacteria [8,9]. Also, such multi-component disinfectants should be included into the complex program of control and prophylaxis of widely spread veterinary diseases such as colibacillosis, salmonellosis, etc. Therefore, the aim of the work was to evaluate the effectiveness surfactants as compounds for the creation of disinfectants with a wide spectrum of action.

Material and methods

Various classes of surfactants were used to create compositions with wide spectrum activity. Explored surfactants were synthesized at the Institute of Macromolecular Chemistry of the National Academy of Sciences of Ukraine (guanidine-containing oligomer (GCO), surfactant 2 and surfactant 3) [10]. Also, we chose characterized compounds such as Trilon B (Netherlands); Triton X-100 (Merk, Germany); glutaraldehyde, (Sigma-Aldrich, USA), OP-10). Three versions of the compositions were tested, which differed in the composition of the components and their ratio. They were designated as: composition 1 (1 % GCO and 1 % surfactant 2), composition 2 (1 % GCO, 1 % surfactant 2 and 1 % Trilon B) and composition 3 (3 % GCO, 1 % Triton X-100, 1 % Trilon B and 1 % glutaraldehyde). As a comparison control, we used Lysoformin 3000 (9.5 % glutaraldehyde, 9.6 % didecyldimethylammonium chloride and 7.5 % of glyoxal) for disinfection of all types of surfaces and medical equipment.

Disinfectant activity was measured against Gram-positive, Gram-negative bacteria and yeasts such as *Staphylococcus aureus* CCM 209, *Escherichia coli* BE and *Candida albicans* UCM Y-690, respectively. The optical density of microbes' suspension was 1.5 Mc Farland units [10]. The biocidal activity of individual components and compositions were determined by the Gould suspension method [11,12]. After counting the CFU on the agar medium, we calculated the arithmetic mean values of the number of cells/ml and established the number of viable cells by the formula:

$$C = \lg \frac{N_t}{N_{tk}},$$

where N_t — the number of bacteria that survived after contact with experimental surfactants; N_{tk} — the number of bacteria that grew in the control over the same time period.

Based on the obtained data, it was determined the decimal logarithm and effectiveness of the experimental surfactants due to the scale in Table 1. It was taken into account that compound was considered as effective only when the number of viable cells decreased more than by -4.0 lg CFU/ml [12].

Table 1

The scale for evaluating the bactericidal activity of the test substances

Value of C	Number of cells that died, %
From -2.0 to -2.9	99.000
From -3.0 to -3.9	99.900
From -4.0 to -4.9	99.990
From -5.0 to -5.9	99.999
From -6.0 and more	> 99.999

The effectiveness of surfactants and research compositions against test cultures was determined at concentrations range of 10000–0.1 ppm (where, 1 part/million (ppm) = 0.9988590004 mg/L [13], 1 ppm = 0.0001 %) directly at the moment of contact with the cells of microorganisms (0 min) and after exposure for 15 and 30 minutes, respectively. After the exposure time, the samples were sown to Nutrient agar medium (NA, Himedia, India) using the Gould technique [11]. As a control, we used standardized suspensions of test-culture cells, which were not exposed to the test compounds or compositions.

All variants of experiments were carried out in three or more independent repetitions (n). The obtained results were used to calculate arithmetic mean values and root mean square errors. The data were checked for normality of distribution, after which appropriate criteria were applied for statistical processing [14].

The mutagenic activity of experimental surfactants was determined in the Ames test [15-17]. It was used *Salmonella typhimurium* TA98 and TA100 strains, which had been characterized by different mutations in the histidine operon: *hisD3052* reading frame shift and *hisG46* base pair replacement, respectively. These strains were grown on Nutrient broth (NB, Himedia, India) at 37 °C for making standardized suspension with optical density 0.7–0.8 units at wavelengths λ = 540 nm. Then suspension was mixed in semi-liquid agar with experimental surfactants in the concentration range of 0.1; 1.0; 10.0; 100.0 and 1000.0 μg/plate. The obtained mixture was applied to the surface of the lower layer of agar medium at Petri dishes. Distilled water served as negative control. The solution of $K_2Cr_2O_7$ was used as a model mutagen and positive control, because the number of His^+-revertant colonies of *S. typhimurium* TA98 increased in 22.6 times, and *S. typhimurium* TA100 in 8.7 times. This indicated the sensitivity of the test strains to the action of mutagenic compounds.

After cultivation at 37 ºC for 48 hours, the number of His^+ revertant colonies was determined. The mutagenicity ratio (MR) was calculated as the number of His^+-revertants in the treated sample to the number of spontaneous revertants. If the value of MR is higher than 2.0, then the sample was considered as mutagenic; MR below 1.7 — no mutagenic activity, MR — 1.7–2.0 characterized a compound as a potential mutagen [18]. The presence of a mutagenic effect was recognized by two conditions. First condition was the presence of a statistically significant difference between the number of colonies of His^+-revertants and the negative control according to the Mann-Whitney test ($p < 0.05$) [14]. Also, the number of CFU His^+-revertants in the experiment had to exceed the spontaneous mutation background of *S. typhimurium* TA98 and *S. typhimurium* TA100 at least in 2.0 and 1.8 times, respectively.

Results and discussion

In our previous research [10] and the data of other authors [19], it was found that the guanidine-containing oligomer has antimicrobial activity against various representatives of pro- and eukaryotic microorganisms. Taking into account such feature of GCO as spectrum of action, stability during storage, lack of odor and relative simplicity of synthesis, we assume that this oligomer could be promising for the creation of broad-spectrum antimicrobial agents possibly active against even multiresistant pathogenic *E. coli*. In addition, we proposed to combine it with anionic (Trilon B, surfactant 2, surfactant 3) and non-ionic (Triton X-100 and OP-10) surfactant, as well as glutaraldehyde in order to strengthen the general antimicrobial activity and improve the cleaning properties [20-22].

We determined the disinfectant activity of individual compounds against *S. aureus* CCM 209, *E. coli* BE and *C. albicans* UCM Y-690 before creating the compositions. The guanidine-containing oligomer proved to be effective against staphylococcal cells (Fig. 1). At a concentration of 1000 ppm, regardless of the duration of contact (0–30 min), a complete suppression of the viability of *S. aureus* CCM 209 cells were observed. When the concentration decreased to 100 ppm at the time of adding the culture to the surfactant solution (0 min), the viability of the cells decreased to -4.4 lg CFU/ml, and after exposure for 15–30 minutes, cells of staphylococci under the influence of GCO totally lost their viability, which confirmed the effectiveness of the compound. When the concentration of GCO was reduced to 10 ppm and the exposure time was 30 min, the number of dead cells was 99.99 %, and with contact from 0 to 15 min, the number of dead cells did not exceed 99.0 %. Therefore, the last concentration was ineffective.

It was also found that the *E. coli* BE strain was more resistant to the influence of GCO. The number of viable cells at the highest concentration and longest exposure even did not reach -3.0 lg CFU/ml (Fig. 2), and thus, the biocidal effect was insufficient. This fact indicated a crucial role of *E. coli* as a marker of disinfectant activity. Therefore, regarding high prevalence and negative impact of pathogenic *E. coli* in poultry and swine farming it is necessary to provide more careful disinfection of facilities.

Instead, the guanidine-containing oligomer showed significant inhibitory activity against cells of the *C. albicans* UCM Y-690. The growth of these yeast at 1–1000 ppm was not observed at all after 0–30 min exposure. However, at the concentration of 0.1 ppm, fungicidal activity was insufficient for effective disinfection (Fig. 3).

After the comparison of guanidine-containing oligomer and Lysoformin 3000 effect to *S. aureus* CCM 209 cells, it was found that they had similar

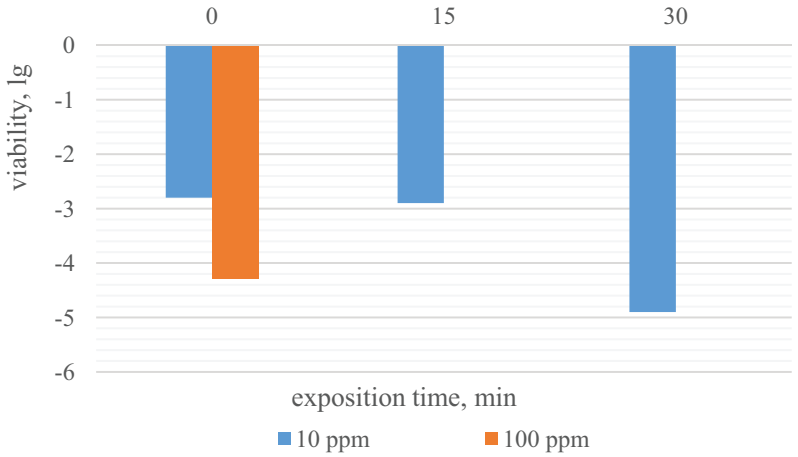

Fig. 1. The influence guanidine-containing oligomer to viability of *S. aureus* CCM 209 cells

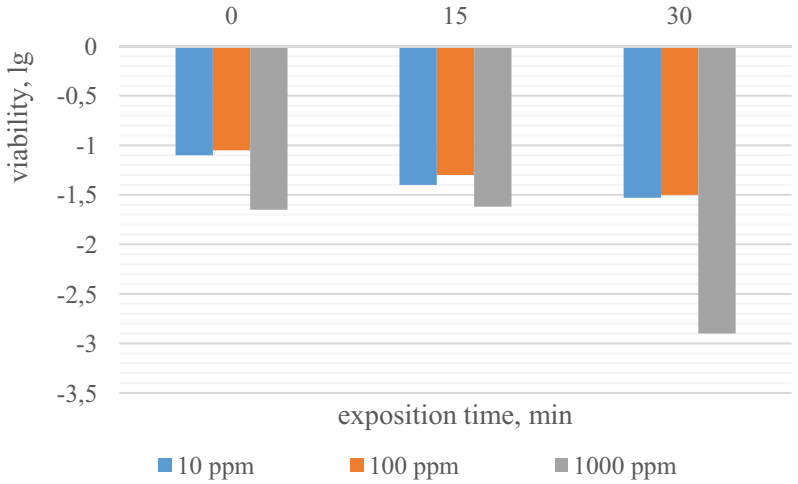

Fig. 2. The influence guanidine-containing oligomer to viability of *E. coli* BE cells

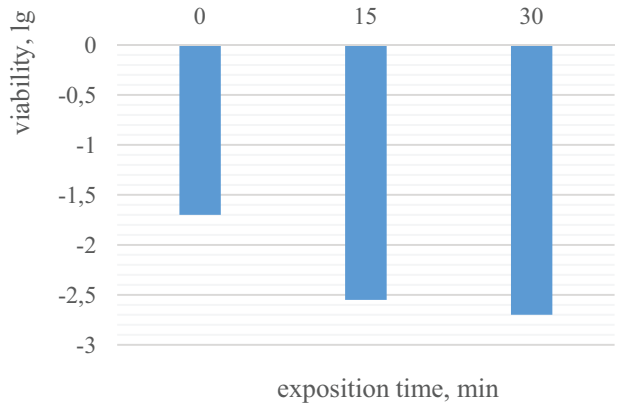

Fig. 3. The influence guanidine-containing oligomer to viability of *C. albicans* UCM Y-690

activity, as both inhibited the viability of staphylococcal cells to 100 % at a concentration of 100 ppm and exposure for 30 minutes. In the case of Gram-negative bacteria, the number of dead cells of the *E. coli* BE at a concentration of GCO of 1000 ppm and an exposure of 30 min was 99.0 %, while Lysoformin 3000 inhibited the bacterial growth to 100 %. The effect of the guanidine-containing oligomer to *C. albicans* cells was the most effective and inhabited their growth to 100 % in 10 ppm solution. However, Lysoformin 3000 at the mentioned concentration was not effective at all.

Therefore, it was necessary to improve the disinfecting activity of the guanidine-containing oligomer against Gram-negative bacteria by combining it with other surfactants, which also have antimicrobial activity. For that reason it was created composition 1, which additionally to GCO contained surfactant 2.

It was shown that after 0 min exposure of composition 1 at a concentration 1000 ppm, the number of viable cells of staphylococci decreased to -4.0 lg CFU/ml. These data prove the effectiveness of the created composition (GCO and surfactant 2) against *S. aureus* CCM 209 cells. The growth of *C. albicans* UCM Y-690 was also absent under the action of this composition, but at the concentration 100 ppm. However, we were unable to achieve a significant effect on the viability of *E. coli* BE with this combination of studied surfactants. At the concentration of 1000 ppm, the number of *E. coli* cells that died because of exposure to composition 1 were near 1.0 lg CFU/ml (Fig. 4).

Also, to enhance the disinfecting activity against Gram-negative bacteria, we created composition 2, to which, in addition to GCO and surfactant 2, was added Trilon B. It was established that under the action of composition 2, with a final concentration of 1000 ppm the growth of *S. aureus* CCM 209 was completely inhibited. Thus, in the case of Gram-positive bacteria, we again observed an increase of the disinfecting activity compared to GCO. Composition 2 and 1 also characterized by similar effectivity against *C. albicans* UCM Y-690 strain. Moreover, the growth of yeast was totally inhibited at the concentration of 100 ppm.

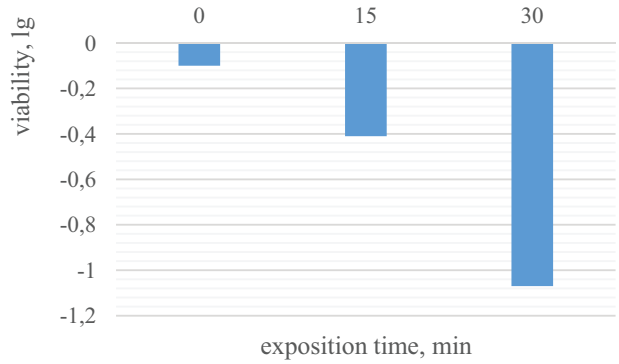

Fig. 4. The influence composition 1 to viability of *E. coli* BE cells

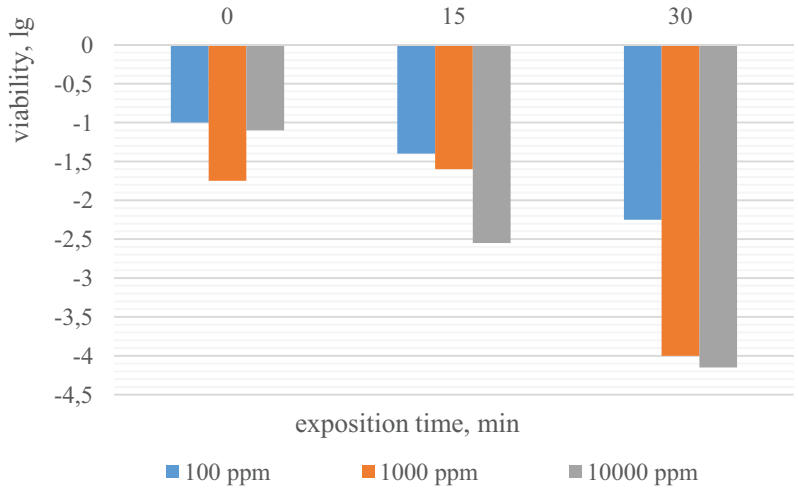

Fig. 5. The influence of composition 2 to viability of *E. coli* BE cells

Considering the disinfectant activity of composition 2 against *E. coli* BE, it was established that the required level of cell destruction efficiency (-4.0 lg CFU/ml) was achieved only under 30 min exposure and concentrations not less than 1000 ppm (Fig. 5). Lower concentrations and shorter time intervals usage were ineffective. It should be noted that additional washing abilities of disinfecting composition could improve its effectiveness against colibacillosis in veterinary because of enhancing cleaning possibility.

The combination of three components in composition 2 improved its disinfecting activity, compared to composition 1, which at 1000 ppm and similar exposure conditions inhibited less than 99 % of *E. coli* BE cells. So, it was determined that Trilon B is able to increase the effect of GCO and surfactant 2 against Gram-negative bacteria. However, the activity of both compositions was significantly lower than Lysoformin 3000, that inhibited the growth of all tested strains after 30 minutes exposure at 100 ppm.

On the basis of obtained results and literature data, which prove the feasibility of combining different classes of surfactants, as well as regarding the wide spectrum of glutaraldehyde activity [3,6,20-25], we tested multicomponent composition based on GCO, Trilon B, Triton X-100 and glutaraldehyde. It was shown that at 1000 ppm it totally inhibited the growth staphylococci and yeast-like fungi cells. However, at 100 ppm, the disinfection efficiency against *S. aureus* CCM 209 slightly decreased, but the fungicidal activity was preserved, because the growth of *C. albicans* UCM Y-690 was inhibited to 100 %. When the concentration was reduced to 10 ppm, composition 3 lost its ability to inhibit the growth of yeast-like fungi.

The Gram-negative bacteria were more resistant to composition 3 comparing yeasts and Gram-positive microbes. Nevertheless, it was observed that it is quite possible to achieve significant disinfection efficiency of this composition by selecting the duration of exposure to bacterial cells and optimal concentrations. The usage of composition 3 at 1000 ppm concentration reduced the number of viable *E. coli* BE cells to -4.0 lg and -5.2 lg CFU/ml after 15 and 30 minutes of exposure, respectively. When the concentration of composition 3 was increased to 10000 ppm the number of viable *E. coli* cells were decreased to -4.4 lg CFU/ml. Also, it was detected 100 % activity after 30 minutes of exposure (Fig. 6).

The obtained disinfection efficiency against *E. coli* BE fully responds to requirements of disinfectants, since the number of viable cells decreased by -4.0 lg CFU/ml or more. The 10000 ppm concentration of composition 3 and time of exposure near 15–30 minutes considered to be effective. Thus, during cleaning, preparation process of facilities and biosecurity measure in poultry and swine farming disinfectants based on surfactants and aldehydes ought to be a part of complex prophylaxis program against antibiotic resistance pathogenic *E. coli* strains and colibacillosis.

Based on obtained data it was determined that composition 3 characterized by highest rate of disinfection activity against Gram-positive, Gram-negative bacteria and yeasts among all tested compounds. For example, after 30 min exposure with concentration of 1000 ppm its disinfection rate was in 3.5 times higher than activity of GCO. It was

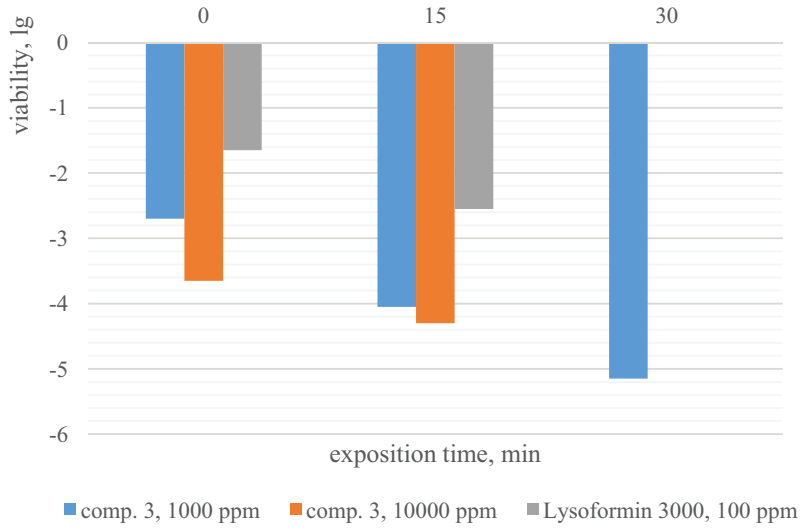

Fig. 6. The influence of composition 3 and Lysoformin 3000 to viability of *E. coli* BE cells

also determined that, under similar conditions, this composition completely inhibited the growth of Gram-positive *S. aureus* CCM 209 cells, and at a concentration of 100 ppm, it neutralized *C. albicans* cells by 100 %.

One of important features that should be considered during creation of new antimicrobial agents is their mutagenicity rate, which indicates the possibility of using this or that surfactant in general [18]. After determining the disinfectant activity of the compositions, we have been investigating the mutagenic potential of individual components by the Ames test. Compounds with high mutagenicity are not desirable to use for disinfection since mutations are the main source of variability and acquired resistance of microorganisms to biocides [26,27].

It was found that GCO in the concentration range of 0.1–100 μg did not have a mutagenic effect relatively to both investigated strains of *S. typhimurium* TA98 and TA100. The number of His^+-revertant colonies ranged from 34±8 to 48±2 for *S. typhimurium* TA98 and 99±18 to 143±25 for *S. typhimurium* TA100. The ratio of the number of revertant colonies in the experiment to the control did not exceed 1.2 and 1.4, respectively (Table 2). This is also confirmed by the absence of a statistically significant difference compared to the spontaneous mutation rate.

After studying the mutagenic effect of glutaraldehyde and surfactant 2, it was shown that the last mention compound had a slight mutagenic potential regarding *S. typhimurium* TA98 test strain. The number of CFU His^+-revertant for this strain varied depending on the concentration and statically differed compared to the spontaneous mutation background. Confirmation of surfactant 2 mutagenic effect was the ratio of the number of His^+-revertant colonies to the spontaneous mutations rate (Table 2). However, this compound did not show mutagenic activity against *S. typhimurium* TA100 strain, because of the ratio of the number of CFU in the experiment to the spontaneous mutations ranged from 0.9 to 1.7. Thus surfactant 2 could have a low mutagenic potential to *S. typhimurium* TA98 and did not influence to *S. typhimurium* TA100 at all.

Determination of the mutagenic activity of glutaraldehyde was carried out in the range of concentrations (10–0.1 μg), which did not inhibit the growth of the test strains of salmonella (Table 2). The number of His^+-revertants to *S. typhimurium* TA98 under the influence of glutaraldehyde ranged from 23±6 to 32±5 CFU, and *S. typhimurium* TA100 – 142±15 to 251±4. The absence of mutagenic effect of glutaraldehyde to *S. typhimurium* TA98 and TA100 also was proven by the ratio of CFU in the experiment to the spontaneous mutation rate, which ranged from 0.7 to 1.0 and 0.9 to 1.6, respectively (Table 2). Under the influence of $K_2Cr_2O_7$ (positive control), the number of CFU His^+-revertant colonies of *S. typhimurium* TA98 and TA100 was in 58.7 and 12.5 times higher, respectively.

Therefore, we found that the guanidine-containing oligomer and glutaraldehyde did not cause frameshift or base-pair substitution mutations of *S. typhimurium* TA98 and TA100.

It should be noted that surfactant 2 was also possible to use in antimicrobial compositions, since it had bed characterized by high antimicrobial activity and at the same time low mutagenicity. If it is necessary to replace or introduce additional components, nonionic surfactant OP-10 could be chosen, as it possesses a high biocidal activity [10]. Also, OP-10 at concentrations from 0.1 to 1000.0 μg did not show mutagenic potential against *S. typhimurium* TA98 and TA100 strains. Instead, surfactant 3 was characterized by high mutagenic activity against both mentioned strains, which made its further use impossible. The study of the mutagenic potential of Trilon B and Triton X-100 was not provided because according to literature data that feature had not been detected [28,29].

Conclusion

Compositions with guanidine-containing oligomer was characterized by high biocidal activity against Gram-positive bacteria and representatives of the genus *Candida*. At the same time, there is a need to increase its efficiency against Gram-negative microbes such as pathogenic *E. coli*, because colibacillosis is the most widespread disease among poultry and swine farming, and the appearance of multiresistant strains harmful for humans is sporadically recorded.

It was determined that the spectrum of action and the disinfectant ability could be significantly improved by introducing cationic, anionic, nonionic surfactants and glutaraldehyde in various combinations. With a rational combination of effective components and their synergism, it is possible to significantly reduce the concentration of the working solution compared to already used commercial disinfectants. For example, the concentration of glutaraldehyde in composition 3 (3 % GCO, 1 % Triton X-100, 1 % Trilon B and 1 % glutaraldehyde) was in 9 times lower than of Lysoformin 3000.

Table 2

Mutagenic activity of composition components

Test-cultures	Compound	Dosage (μg/plate)	Number of His^+-revertants	CFU surfactants to SM ratio
S. typhimurium TA98	GCO	100.0	46–50	1.2
		10.0	38–44	1.1
		1.0	36–50	1.1
		0.1	26–42	0.9
	Surfactant 2	100	505–623	18.2
		10	188–198	6.2
		1	34–36	1.1
		0.1	37–27	1.0
	Glutaric aldehyde	1000	gi	–
		100	gi	–
		10	37–27	1
		1	29–17	0.7
		0.1	31–21	0.8
	control	200	1793–1849	58.7
	SM	–	40–22	–
S. typhimurium TA100	GCO	100.0	95–147	1.2
		10.0	99–119	1.1
		1.0	81–117	1.0
		0.1	118–168	1.4
	Surfactant 2	100	169–151	1.0
		10	276–254	1.7
		1	251–231	1.5
		0.1	167–129	0.9
	Glutaric aldehyde	1000	gi	–
		100	157–166	1.0
		10	127–157	0.9
		1	243–207	1.4
		0.1	247–255	1.6
	control	200	1923–2013	12.5
	SM	–	125–189	–

Remark. SM – spontaneous mutation; positive control – $K_2Cr_2O_7$; gi – growth inhibition.

Also, guanidine-containing oligomer, surfactant 2 and glutaraldehyde were not characterized by mutagenic activity, which is one of the main criteria for disinfectants usage since this reduces the risks of developing bacterial resistance to antibiotics and other biocides.

Implementing various combinations of surfactants that are introduced into newly created disinfectant, it is possible to enhance not only the disinfecting activity (composition 3), but also prevent the formation of cross-resistance in different groups of microorganisms. That is why Triton X-100 can be replaced by a non-ionic surfactant of industrial production – OP-10, that is characterized by high biocidal activity and does not show mutagenic effects. This would help to improve washing features and increase overall antimicrobial activity, which is especially important for livestock farming with a low level of management and biosecurity.

Thus, the development and implementation of new disinfectants, which could help to fight against multiresistant strains of bacteria, is an indispensable part of comprehensive programs in controlling and prevention of common diseases in animal husbandry and medicine, particularly, colibacillosis.

References

1. Collignon P. Fact sheets on sustainable development goals: health targets: Antimicrobial resistance. Emerging Infect. Dis. 2017;6:434-36. doi:10.3201/eid0604.000428
2. Morrison L, Zembower TR. Antimicrobial resistance – global report on surveillance. Gastrointest. Endosc. Clin. N. Am. 2020;30:619-35. doi:10.1016/j.giec.2020.06.004
3. Basiry D, Entezari Heravi N, Uluseker C, Kaster KM, Kommedal R, Pala-Ozkok I. The effect of disinfectants and antiseptics on co- and cross-selection of resistance to antibiotics in aquatic environments and wastewater treatment plants. Front Microbiol. 2022;13:13:1050558. doi:10.3389/fmicb.2022.1050558
4. Russell AD. Bacterial adaptation and resistance to antiseptics, disinfectants and preservatives is not a new phenomenon. J. Hosp. Infect. 2004;57;97-104. doi:10.1016/j.jhin.2004.01.004
5. Usman M, Farooq M, Hanna K. Environmental side effects of the injudicious use of antimicrobials in the era of COVID-19. Sci. Total Environ. 2020;745:141053. doi:10.1016/j.scitotenv.2020.141053
6. Ghafoor D, Khan Z, Khan A, Ualiyeva D, Zaman N. Excessive use of disinfectants against COVID-19 posing a potential threat to living beings. Current Research in Toxicology. 2021;2:159-68. doi:10.1016/j.crtox.2021.02.008
7. Kampf G. Biocidal agents used for disinfection can enhance antibiotic resistance in Gram-negative species. Antibiotics. 2018;7(4):110. doi:10.3390/antibiotics7040110
8. Wei T, Yu Q, Chen H. Responsive and Synergistic Antibacterial Coatings: Fighting against Bacteria in a Smart and Effective Way. Adv. Healthcare Materials 2019;8(3):e1801381. doi:10.1002/adhm.201801381
9. Pemmada R, Shrivastava A, Dash M, Cui K, Kumar P, Ramakrishna S, Zhou Y, Thomas V, Nanda HS. Science-based strategies of antibacterial coatings with bactericidal properties for biomedical and healthcare settings. Current Opinion in Biomedical Engineering. 2023;25:100442. doi:10.1016/j.cobme.2022.100442
10. Furtat I, Nechypurenko O, Vakuliuk P, Vortman M, Shevchenko V. Antimicrobial activity of traditional and newly synthetized surface-active agents as a basis for the creation of new disinfectants. NaUKMA Research Papers. Biology and Ecology. 2022;5:25-32. doi:10.18523/2617-4529.2022.5.25-32
11. Gold J. Quantity and quality in the diagnosis of urinary tract infections. British Journal of Urology. 1965;37(1):7-12. doi:10.1111/j.1464-410X.1965.tb09567.x
12. Furtat I, Lupatsii M, Murlanova T, Vakuliuk P, Gaidai A, Biliayeva O, Sobczuk H, Golub A. Nanocomposites with ornidazole-antibacterial and antiadhesive agents against Gram-positive and Gram-negative bacteria. Applied Nanoscience. 2020;10:3193-3203. doi:10.1007/s13204-020-01260-x
13. Convert Part/million (ppm) to Milligram/liter. [Internet]. Available from: https://www.unitconverters.net/concentration-solution
14. Glantz SA. Primer of Biostatistics. 7th ed. The McGraw-Hill Companies; 2012. 327 p.
15. Maron DM, Ames BN. Revised methods for the Salmonella mutagenicity test. Mutation Research. 1983;113(3-4):173-215. doi:10.1016/0165-1161(83)90010-9
16. Sundar R, Jain MR, Valani D. Chapter Ten – Mutagenicity Testing: Regulatory Guidelines and Current Needs. In: Kumar A, et al., ed. Mutagenicity: Assays and Applications. Academic Press; 2018. p. 191-228. doi:10.1016/B978-0-12-809252-1.00010-9
17. Vijay U, Gupta S, Mathur P, Suravajhala P, Bhatnagar P. Microbial Mutagenicity Assay: Ames Test. Bio Protoc. 2018;8(6):e2763. doi:10.21769/BioProtoc.2763
18. Obłąk E, Piecuch A, Guz-Regner K, Dworniczek E. Antibacterial activity of gemini quaternary ammonium salts. FEMS Microbiology Letters. 2014;350(2):190-8. doi:10.1111/1574-6968.12331
19. Vortman MYa, Pysmenna YuB, Rudenko AV, Tretyak VV, Lemeshko VN, Shevchenko VV. Fungicidical and bactericidical activity of alkyl-substituting polyetherguanidines. Biologichni Studii. 2020;14(3):65-78. doi:10.30970/sbi.1403.630
20. Russell AD. Glutaraldehyde: current status and uses. Infect Control Hosp Epidemiol. 1994;15(11):724-33. doi:10.1086/646845
21. Sehmi SK, Allan E, MacRobert AJ, Parkin I. The bactericidal activity of glutaraldehyde-impregnated polyurethane. Microbiology open. 2016;5(5):891-97. doi:10.1002/mbo3.378
22. Lin W, Guan X, Cao J, Niu B, Chen Q. Bactericidal mechanism of glutaraldehyde-didecyldimethylammonium bromide as a disinfectant against Escherichia coli. Journal of Applied Microbiology. 2017;122(3):676-85. doi:10.1111/jam.13384
23. Zhou C, Wang Y. Structure–activity relationship of cationic surfactants as antimicrobial agent. Current Opinion in Colloid & Interface Science. 2020;45:28-43. doi:10.1016/j.cocis.2019.11.009
24. Rutala WA, Weber DJ. Disinfectants used for environmental disinfection and new room decontamination technology. Am. J. Infect. Control. 2013;41(5):S36-S41. doi:10.1016/j.ajic.2012.11.006
25. Sayed SRM, Ezzat AO, Yassin MT, Abdelbacki AMM. Synthesis, Characterization and Application of Novel Cationic Surfactants as Antibacterial Agents. Separations. 2023;10(2):97. doi:10.3390/separations10020097
26. Monarca S, Richardso SD, Feretti D, Grottolo M, Thruston JrAD, Zani C, Navazio G, Ragazzo P, Zerbini I, Alberti A. Mutagenicity and disinfection by-products in surface drinking water disinfected with peracetic acid. Environmental Toxicology and Chemistry. 2002;21(2):309-318. doi:10.1002/etc.562021 0212
27. Thomas DN, Wills JW, Tracey H, Baldwin SJ, Burman M, Williams AN, Harte DSG, Buckley RA, Lynch AM. Ames test study designs for nitrosamine mutagenicity testing: qualitative and quantitative analysis of key assay parameters. Mutagenesis. 2024;39(2):78-95. doi:10.1093/mutage/gead033
28. Narasimhamurthy MK. Assessment of *in vivo* mutagenic potency of ethylenediaminetetraacetic acid in albino mice. Food and Chemical Toxicology. 1991;29(12):845-49. doi:10.1016/0278-6915(91)90112-K
29. Ballal NV, Rao BN, Mala K, Bhat KS, Rao BSS. Assessment of genotoxic effect of maleic acid and EDTA: a comparative *in vitro* experimental study. Clin Oral Invest. 2013;17:1319-27. doi:10.1007/s00784-012-0818-x

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ЕФЕКТИВНІСТЬ ПОВЕРХНЕВО-АКТИВНИХ РЕЧОВИН ЯК КОМПОНЕНТІВ ДЛЯ СТВОРЕННЯ ДЕЗІНФЕКЦІЙНИХ ЗАСОБІВ ШИРОКОГО СПЕКТРА ДІЇ

Мета. Поряд із використанням антибіотиків застосування дезінфектантів має вирішальне значення в боротьбі з мультирезистентними штамами бактерій, небезпечними не лише для тварин, а й для людини. З огляду на це метою роботи було оцінити ефективність ПАР як сполук для створення дезінфекційних засобів широкого спектра дії. **Методи.** Дезінфікувальну дію досліджених катіонних, аніонних та нейоногенних ПАР щодо представників грампозитивних і грамнегативних бактерій, а також грибів вивчали щодо штамів *Staphylococcus aureus* CCM 209, *Escherichia coli* BE та *Candida albicans* УКМ Y-690. Біоцидну активність визначали суспензійним методом Gould шляхом підрахунку клітин, що вижили після обробки дослідженими ПАР та композиціями, створеними на їхній основі, за концентрацій у діапазоні 10000–0,1 ppm. Мутагенну активність сполук вивчали в тесті Еймса щодо тест-штамів *Salmonella typhimurium* TA98 і TA100. **Результати.** Композиції з гуанідиновмісним олігомером найефективніше пригнічували *S. aureus* та *C. albicans*. За концентрації 1000 і 100 ppm спостерігалася 100 % загибель клітин цих штамів. Біоцидний ефект щодо представників грамнегативних бактерій був дещо слабшим, що потрібно враховувати під час створення антимікробних засобів, зокрема активних щодо патогенних штамів *E. coli*. Спектр дії та дезінфікувальна здатність досліджених у роботі композицій залежали від складу компонентів, які використовували для їхнього створення. Також показано, що більшості ПАР, вивчених у роботі, які використовували для створення антимікробних композицій, не була притаманна мутагенна активність. **Висновки.** На прикладі досліджених композицій різного хімічного складу, створених на основі гуанідиновмісного олігомеру, показано перспективність його застосування як основного компонента дезінфікувальних засобів. Отже, розроблення та впровадження нових дезінфектантів, які могли б допомогти в боротьбі з мультирезистентними штамами бактерій, є невідокремною частиною комплексних програм контролю та профілактики поширених захворювань у тваринництві та медицині, зокрема колібактеріозу.

Ключові слова: аніонні, катіонні та нейоногенні поверхнево-активні речовини, бактерицидна та фунгіцидна дія, дезінфікувальні засоби, патогенні *E. coli*.

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