# UDC 636.5:614.4 DOI: 10.15587/2519-8025.2022.266239

# EFFECTIVENESS OF ECOLOGICALLY SAFE DISINFECTANTS AGAINST *PSEUDOMONAS AERUGINOSA* AND THE POULTRY MAIN BACTERIOSIS PATHOGENS

# Andriy Berezovskiy, Tetiana Fotina, Yevheniia Vashchyk, Dmytro Berezhnyi, Dmytro Morozenko

**The aim:** study of the effectiveness of environmentally safe disinfectants against P. aeruginosa and pathogens of the main bacteriosis of poultry at test facilities.

*Materials and methods.* To study the antimicrobial action of the investigated disinfectants against a mixture of epizootic cultures of E. coli, P. aeruginosa, S. aureus, S. typhimurium, isolated from poultry, bacteriological studies were carried out on test objects: galvanized iron, wooden bars (painted and unpainted), red brick, cutouts from plaster, size  $10 \times 10$  cm.

**Results.** The working solutions of the new disinfectant "Dezsan" were studied in comparison with the control agents: "Virocid" and "Bi-dez" at a concentration of 0.01; 0.1; 0.25; 0.5; 1; 1.5 % in relation to suspension cultures of E. coli, P. aeruginosa, S. aureus, S. typhimurium. In this case, it was established, that the "Dezsan" agent showed an antimicrobial effect on rough test objects after exposure for 3 hours at a concentration of 0.1 %, and at a concentration of 0.25 % - for 1 hour. On smooth surfaces, the agent neutralized bacterial cultures at a concentration of 0.1 % after exposure for 1 hour. The preparation "Bi-dez" at a concentration of 0.25 % was effective on smooth surfaces after exposure for 1 hour, on rough surfaces (brick, plaster) - at a concentration of 0.5 % after exposure for 3 hours or more. The working solution of 1 % concentration neutralized bacterial cultures on all types of surfaces after exposure for 1 hour or more. "Virocid" agent after exposure for 1 hour neutralized bacterial cultures on smooth surfaces in concentrations of 0.25 % and higher; on rough surfaces, the growth of cultures was not detected when using a 0.5 % solution.

**Conclusions.** Environmentally safe disinfectants "Dezsan" and "Shumerske sryblo" compared to the control ones ("Bi Dez" and "Virotsid") show an active antimicrobial effect at a concentration of 0.25 % and 3 %, respectively, against the suspension of epizootic cultures of E. coli, P. aeruginosa, S. aureus and S. typhimurium on different types of production surfaces, which justifies the feasibility of their use based on the principle of rotation of disinfectants for the prevention of bacterial pseudomonosis of poultry

Keywords: poultry, pseudomonosis, E. coli, P. aeruginosa, S. aureus, S. typhimurium, effectiveness, prevention

#### How to cite:

Berezovskiy, A., Fotina, T., Vashchyk, Y., Berezhnyi, D., Morozenko, D. (2022). Effectiveness of ecologically safe disinfectants against *Pseudomonas aeruginosa* and the poultry main bacteriosis pathogens. ScienceRise: Biological Science, 3 (32), 00-00. doi: http://doi.org/10.15587/2519-8025.2022.266239

© The Author(s) 2022 This is an open access article under the Creative Commons CC BY license hydrate

## 1. Introduction

Poultry farming, as a branch of animal husbandry, is progressive and constantly improving. This especially applies to the prevention of bacterial diseases [1, 2]. It differs from other branches of animal husbandry in its high reproduction rate and prematurity, which makes it the main source of providing the population with proteins of animal origin [3].

The use of modern technologies allows in broiler production to reduce the period of poultry fattening to 35 days, to ensure an average daily gain of more than 50 grams for feed conversion of 1.75 kg, and to obtain more than 230 kg of broiler meat in slaughter weight per laying hen of the parent flock; in egg production, more than 340 eggs can be obtained for an average annual laying hen with feed conversion of 1.17 kg [4]. However, in

Veterinary research

conditions of intensification of production, the effect of factors, contributing to the disruption of normal microflora in farm poultry, is noted. Non-observance of veterinary and sanitary norms and zoohygiene requirements causes a violation of the balance between normal and conditionally pathogenic microflora of the gastrointestinal tract, which, against the background of constant stress and a decrease in the natural resistance of the body of birds, leads to an increase in the pathogenic and virulent properties of microorganisms and causes the development of an infectious disease [5].

**The aim of the research** study of the effectiveness of environmentally safe modern disinfectants against *P. aeruginosa* and pathogens of the main bacteriosis of poultry at test facilities.

To study the antimicrobial action of the investigated disinfectants against a mixture of epizootic cultures of E. coli, P. aeruginosa, S. aureus, S. typhimurium, isolated from pat material from poultry, bacteriological studies were carried out on test objects in accordance with the methodological recommendations "Use of the latest means and methods of rehabilitation of poultry facilities and control of their effectiveness", 2007. The test objects were: galvanized iron, wooden bars (painted and unpainted), red brick and plaster cutouts, measuring 10×10 cm. Before applying the cultures, the test objects were subjected to heat treatment for the purpose of sanitation. The research was conducted in a bacteriological box. Using a sterile pipette, a mixture of 1 billion suspended microbial cells of bacterial cultures of E. coli, P. aeruginosa, S. aureus, S. typhimurium in an isotonic solution was applied to test objects in enameled cuvettes. An hour later, using a sprayer, aerosols of disinfectant solutions were sprayed at the rate of 10 cm<sup>3</sup> per 10 cm<sup>2</sup> for exposure from 1 hour up to 1 day Working solutions of disinfectants "Dezsan", "Virocid" and "Bi-dez" at a concentration of 0.01 were studied; 0.1; 0.25; 0.5; 1; 1.5 %. Control test objects were irrigated with sterile distilled water.

The composition of the "Bi-dez" product:  $100 \text{ cm}^3$  of the drug contains active substances (g): polyhexamethyleneguanidine hydrochloride – 6.5; dodecyldipropylene triamine – 6.5. Excipients: glutamic acid, cocoamidopropyl betaine, demineralized water (NVF BROVAFARMA LLC, Ukraine).

The composition of "Dezsan":  $100 \text{ cm}^3$  of the drug contains active substances: alkyldimethylbenzylammonium chloride – 4.8 %, octyldecyldimethylammonium chloride – 1.44 %, didecyldimethylammonium chloride – 2.16 %, glutaraldehyde – 10 % (Brovafarma Ltd., Ukraine).

"Virocid" contains a composition of two quaternary ammonium compounds: alkyldimethylbenzylammonium chloride – 17.06 %, didecyldimethylammonium chloride – 7.8 %), glutaraldehyde – 10.7 %, isopropanol – 14.6 %, turpentine derivative – 2.0 % ("CID LINES NV/SA", Belgium).

To determine the quality of disinfection with sterile cotton swabs that were previously immersed in tubes with MPB, washings were taken from the surfaces of the test objects and again placed in tubes with MPB, which were incubated in a thermostat at a temperature of + 38 °C, followed by recording the growth of the culture after 12, 24 and 48 hours. The presence of the development of signs of culture growth (turbidity, change in the color of MPB, the formation of a film on the surface and sediment at the bottom of the test tube) indicated the absence of antimicrobial action of the studied disinfectant.

#### 3. Research results

According to the results of our monitoring bacteriological studies of pathological material and washings from the production surfaces of poultry premises, on average, in all farms of different technological direction, escherichia prevailed – 37.58 % and Pseudomonas aeruginosa – 22.98 %, coccus microflora was detected in 20.23 % of cases. The number of *Proteus, Campylobacter, Enterobacter, Citrobacter, Klebsiella, Yersinia, and Clostridium* cultures was 19.21 %. Thus, the frequency

of isolation of *P. aeruginosa* and *E. coli* was 3 times higher than the cases of coccal microflora isolation and 3.15 times higher than the frequency of other causative agents of poultry bacteriosis.

Our results and the data of other researchers regarding the isolation of bacterial flora from poultry farms differed somewhat, which can be explained by climatic features, different methods and focus of research. A higher percentage of P. aeruginosa isolation confirms the effectiveness of our proposed medium, which was used to isolate the pathogen from pat material and facilities of poultry farms. Thus, according to the results of studies by Stegnii B.T., Gliebova K.V., Petrenchuk E.P. et al. [6], the percentage of productive poultry, affected by salmonella, is 7.7 % of the total number of poultry examined. The share of pathogenic E. coli cultures accounts for 14.7 % of the number of isolated pathogens. The frequency of isolation of Enterobacter, Citrobacter, and Proteus cultures is 3.9 %, 15.6 %, and 8.2 %, respectively. The percentage of poultry, infected with representatives of the Staphylococcus, Pseudomonas, Neisseria and Ornithobacterium families, is not significant [7].

Disinfection measures, aimed at destroying pathogens in the environment and preventing their penetration into the bird's body, are an integral part of the effective fight against bacterial infections. Effectively carried out disinfection measures in premises for growing poultry, hatcheries allow to prevent the spread of pathogens and the occurrence of epizootic outbreaks. Long-term use of disinfectants of the same chemical group causes the development of microflora resistance to antimicrobial and disinfectant drugs. There is a need to constantly search for new effective antimicrobial substances. The environmental aspect is extremely important when choosing disinfectants. Effective, but aggressive substances (formalin, caustic soda, chlorine-containing disinfectants, etc.) are recognized by the world as ecologically dangerous, have an irritating and carcinogenic effect, so they are abandoned in many countries [8, 9].

Epizootic well-being in the economy directly depends on timely and regular disinfection with effective disinfectants. The competitiveness of modern poultry enterprises is determined by many factors, including the quality of disinfection measures. The main purpose of disinfection is not only to improve the health of the livestock industry, but also to prevent infectious diseases in healthy farms. Therefore, preventive disinfection is gaining more and more popularity as a combination of disinfection measures, carried out in the absence of infectious diseases, its purpose is to prevent the occurrence and spread of infections [10–12].

The working solutions of the new disinfectant "Dezsan" were studied in comparison with the control agents: "Virocid" and "Bi-dez" at a concentration of 0.01; 0.1; 0.25; 0.5; 1; 1.5 % in relation to suspension cultures of *E. coli*, *P. aeruginosa*, *S. aureus*, *S. typhimurium*. In this case, it was established, that the "Dezsan" agent showed an antimicrobial effect on rough test objects after exposure for 3 hours at a concentration of 0.1 %, and at a concentration of 0.25 % – after 1 hour. On smooth surfaces, the agent neutralized bacterial cultures at a concentration of 0.1 % after exposure for 1 hour. The results of the study are presented in Table 1.

Table
-------

	Exposure, hours	Growth of bacterial cultures on test objects / concentration of disinfection solution, %																	
Test objects		"Dezsan"						"Bides"						"Virocide"					
		0.01	0.1	0.25	0.5	1	1.5	0.01	0.1	0.25	0.5	1	1.5	0.01	0.1	0.25	0.5	1	1.5
Galvanized iron	1	+		—	—		1	+	+	-	I	I	1	+	+	_	-	Ι	_
	3	+	I	-	_	Ι	١	+	I	_	I		١	+	Ι	_		I	-
	24	+	I	-	_	Ι	١	+	I	_	I		١	+	Ι	_		I	-
Painted wood	1	+	I	-	_	Ι	١	+	+	_	I		١	+	+	_		I	-
	3	+	I	-	_	Ι	١	+	I	_	I		١	+	Ι	_		I	-
	24	+	I	-	_	Ι	١	+	I	_	I		١	+	Ι	_		I	-
Unpainted wood	1	+	I	-	_	Ι	١	+	+	+	I		١	+	+	_		I	-
	3	+	I	-	_	Ι	١	+	+	_	I		١	+	+	_		I	-
	24	+	I	-	_	Ι	١	+	I	_	I		١	+	Ι	_		I	-
Brick	1	+	+	—	—	١	-	+	+	+	+	-	-	+	+	+	-	Ι	—
	3	+	١		_	١	١	+	+	+	١		١	+	+	+	Ι	Ι	-
	24	+	I	-	-	١	١	+	+	-	I	I	١	+	١	-	1	I	-
Plaster	1	+	+	-	-	١	١	+	+	+	+	I	١	+	+	+	1		-
	3	+		-	-	Ι	_	+	+	+	I	Ι	_	+	+	+	-	-	-
	24	+	١	_	_	١	_	+	+	-	١	_	_	+	Ι	-	Ι		-

Antimicrobial effect of modern disinfectants on suspended cultures of *E. coli*, *P. aeruginosa*, *S. aureus*, *S. typhimurium* on various test objects

*Note:* "+" – *presence of growth,* "–" – *absence of growth* 

The preparation "Bi-dez" at a concentration of 0.25 % was effective on smooth surfaces after exposure for 1 hour, on rough surfaces (brick, plaster) – at a concentration of 0.5 % after exposure for 3 hours or more. A working solution of 1 % concentration neutralized bacterial cultures on all types of surfaces after exposure for 1 hour or more. The "Virocid" agent after exposure for 1 hour neutralized bacterial cultures on smooth surfaces in concentrations of 0.25 % and higher; on rough surfaces the growth of cultures was not detected when using a 0.5 % solution.

Thus, the studied disinfectant "Dezsan" and the control "Bi-dez" and "Virocid" on all types of surfaces in 3 hours of exposure showed an antimicrobial effect against the suspension of epizootic cultures of *E. coli*, *P. aeruginosa*, *S. aureus* and *S. typhimurium*, isolated from the pat material from the bird, in a concentration of 0.1 %, 0.5 % and 0.5 %, and after exposure for 1 hour – in 0.25 %, 1 % and 0.5 %, respectively.

In comparison with modern studies of other authors, it should be noted, that standard disinfection schemes in poultry farming do not give results in terms of complete destruction of pathogens. This is caused by various factors, among which the leading role belongs to antibiotic resistance, in particular, Salmonella [13]. Peracetic acid, hydrogen peroxide, and formaldehyde proved to be the best disinfectants for poultry rooms, but their effectiveness was not ideal [14]. According to foreign authors, the selection and testing of disinfection is an important problem [15], which determines the relevance and necessity of conducting our research.

**Research limitations.** The research was conducted exclusively on such test objects as galvanized iron, painted and unpainted wood, brick and plaster. Therefore, the effectiveness of the tested disinfectants on other materials cannot be guaranteed.

**Prospects for further research.** A promising direction of research is the study of other concentrations of disinfectants for further use in poultry farming for disease prevention.

# 4. Conclusions

Environmentally safe disinfectants "Dezsan" and "Shumerske sryblo" compared to the control ones ("Bi Dez" and "Virotsid") show an active antimicrobial effect at a concentration of 0.25 % and 3 %, respectively, against the suspension of epizootic cultures of *E. coli, P. aeruginosa, S. aureus* and *S. typhimurium* on different types of production surfaces, which justifies the feasibility of their use based on the principle of rotation of disinfectants for the prevention of bacterial pseudomonosis of poultry.

## **Conflict of interests**

The authors declare that they have no conflict of interest in relation to this study, including financial, personal, authorship, or any other, that could affect the study and its results, presented in this article.

## Financing

The study was performed without financial support.

# Acknowledgments

The authors express their gratitude to the limited liability company "Brovafarma" (Ukraine) for their support in conducting this research and providing disinfectants for testing.

#### References

1. Jeni, R. E., Dittoe, D. K., Olson, E. G., Lourenco, J., Corcionivoschi, N., Ricke, S. C.et al. (2021). Probiotics and potential applications for alternative poultry production systems. Poultry Science, 100 (7), 101156. doi: https://doi.org/10.1016/j.psj.2021.101156

2. Redweik, G. A. J., Jochum, J., Mellata, M. (2020). Live Bacterial Prophylactics in Modern Poultry. Frontiers in Veterinary Science, 7. doi: https://doi.org/10.3389/fvets.2020.592312

3. Chen, S., Yong, Y., Ju, X. (2021). Effect of heat stress on growth and production performance of livestock and poultry: Mechanism to prevention. Journal of Thermal Biology, 99, 103019. doi: https://doi.org/10.1016/j.jtherbio.2021.103019

4. Bovkun, G.F. (2004). Rol mikroflory pri zabolevanii organov pishchevareniia u tcypliat. Veterinariia, 4, 14-16.

5. Al-Khalaifah, H. S. (2018). Benefits of probiotics and/or prebiotics for antibiotic-reduced poultry. Poultry Science, 97 (11), 3807–3815. https://doi.org/10.3382/ps/pey160

6. Stehnii, B. T., Hliebova, K. V., Petrenchuk, E. P., Bobrovytska, I. A., Maiboroda, O. V. (2014). Epizootolohichnyi monitorynh bakterialnykh khvorob ptytsi v Ukraini. Veterynarna medytsyna, 98, 99–102.

7. Stehnii, B. T., Hliebova, K. V., Petrenchuk, E. P., Zaremba, I. A., Maiboroda, O. V. (2013). Analiz epizootychnoho monitorynhu bakterialnykh zakhvoriuvan silskohospodarskoi, dykoi ta dekoratyvnoi ptytsi na terytorii Skhodu Ukrainy Veterynanrna medytsyna, 97, 232–233.

8. Prokudina, N. O. (2016). Suchasni dezinfektanti: pliusi ta minusi. Suchasne ptakhivnitctvo, 4 (161), 19-22.

9. Zon, G. A., Vashchik, E. V., Moroz, O. S. (2010). Rezultati poshuku suchasnikh dezinfektciinikh rechovin, aktivnikh do zbudnika psevdomonozu ptitci. Aktualnye problemy sovremennogo ptitcevodstva. Alushta, 89–95.

10. Mandyhra, Yu. M. (2017). Sanitarna otsinka zastosuvannia u tvarynnytstvi dezinfikuiuchykh zasobiv na osnovi poliheksametylenhuanidynu. Kharkiv.

11. Prokudina, N. O. (2016). Suchasni dezinfektanty: pliusy ta minusy. Suchasne ptakhivnytstvo, 4 (161), 19-22.

12. Mandyhra, M. S., Lysytsia, A. V., Zhyhaliuk, S. V., Dmytriiev, I. M., Velychko, Yu. M., Andrushchuk, I. L. et al. (2012). Analiz zasobiv dlia veterynarnoi dezinfektsii. Veterynarna medytsyna, 96, 163–165.

13. Newton, K., Gosling, B., Rabie, A., Davies, R. (2020). Field investigations of multidrug-resistant Salmonella Infantis epidemic strain incursions into broiler flocks in England and Wales. Avian Pathology, 49 (6), 631–641. doi: https://doi.org/10.1080/03079457.2020.1809634

14. Maertens, H., De Reu, K., Van Weyenberg, S., Van Coillie, E., Meyer, E., Van Meirhaeghe, H. et al. (2018). Evaluation of the hygienogram scores and related data obtained after cleaning and disinfection of poultry houses in Flanders during the period 2007 to 2014. Poultry Science, 97 (2), 620–627. https://doi.org/10.3382/ps/pex327

15. Wales, A. D., Gosling, R. J., Bare, H. L., Davies, R. H. (2021). Disinfectant testing for veterinary and agricultural applications: A review. Zoonoses and Public Health, 68 (5), 361–375. Portico. https://doi.org/10.1111/zph.12830

> Received date 04.08.2022 Accepted date 22.09.2022 Published date 30.09.2022

Andriy Berezovskiy, Doctor of Veterinary Sciences, Professor, Department of Veterinary Expertise, Microbiology, Zoohygiene and Safety and Quality of Livestock Products, Sumy National Agrarian University, Gerasima Kodratieva str., 160, Sumy, Ukraine, 40000

**Tetiana Fotina,** Doctor of Veterinary Sciences, Professor, Head of Department, Department of Veterinary Expertise, Microbiology, Zoohygiene and Safety and Quality of Livestock Products, Sumy National Agrarian University, Gerasima Kodratieva str., 160, Sumy, Ukraine, 40000

Yevheniia Vashchyk\*, Doctor of Veterinary Sciences, Associate Professor, Department of Veterinary Medicine and Pharmacy, National University of Pharmacy Pushkinska str., 53, Kharkiv, Ukraine, 61002

**Dmytro Berezhnyi,** PhD, Associate Professor, Department of Veterinary Medicine and Pharmacy, National University of Pharmacy, Pushkinska str., 53, Kharkiv, Ukraine, 61002

**Dmytro Morozenko,** Doctor of Veterinary Sciences, Senior Researcher, Head of Department, Department of Veterinary Medicine and Pharmacy, National University of Pharmacy, Pushkinska str., 53, Kharkiv, Ukraine, 61002

\*Corresponding author: Yevheniia Vashchyk, e-mail: yevgeniavashik@gmail.com