

INNOVATIVE METHODS OF DISINFECTING LIVESTOCK FACILITIES

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Summary. Parasitic diseases in animals are widespread throughout the world and cause significant economic losses to the livestock industry. The most effective and economically justified measure for preventing these diseases among susceptible livestock is to implement high-quality veterinary and sanitary measures. To achieve this, it is essential to use effective disinfectants that have been proven to work in both laboratory and production environments. The work was carried out at the Laboratory of Veterinary Sanitation, Parasitology and Bee Diseases Study in the National Scientific Center 'Institute of Experimental and Clinical Veterinary Medicine' (Kharkiv, Ukraine). The effectiveness of the disinfectants was determined in accordance with existing regulatory documents. Based on these results, a method for disinfecting livestock facilities was developed. This method involves using a preparation containing peroxyacetic acid, hydrogen peroxide, acetic acid, stabilizing additives, and water. The exposure time ranges from 6 to 48 h, and the consumption rate is 500 ml/m². We propose a disinfection method involving a disinfectant containing potassium monopersulfate, sodium dichloroisocyanurate, sodium hexametaphosphate, sulfamic acid, malic acid, sodium alkylbenzyl sulfonate, sodium sulfate, and water. The exposure time is 3 h, and the consumption rate is 300 ml/m². Another method involves a preparation containing a mixture of quaternary ammonium compounds, glutaraldehyde, isopropyl alcohol, nonionic surfactants, and deionized water. This method requires an exposure time of 3–24 h and a consumption rate of 500 ml/m². The disinfectant, containing didecyltrimethylammonium chloride, glutaraldehyde, benzalkonium chloride, surfactants, orthophosphoric acid, and water, has been proven effective at a 72-hour exposure rate of 500 ml/m² for soil disinfection. The proposed disinfection methods have been proven to meet biosafety and bioprotection requirements, and are easy to use, environmentally friendly, highly effective, and cost-effective. The results presented in this article significantly supplement existing sanitary and hygienic protocols in animal husbandry. Further research should focus on developing a comprehensive, scientifically based, integrated system for protecting farm animals

Keywords: disinfectant, concentration, exposure, soil, disinfection, effectiveness, helminth eggs

Introduction. In light of climate change and changes in livestock farming practices, addressing the issue of parasitic diseases in animals is becoming particularly relevant (Amoah et al., 2018; Bogach et al., 2020). Despite the success in controlling human and animal diseases, parasitic diseases remain a leading health concern (Kwong et al., 2021; Tadege et al., 2022; Elghryani et al., 2023). A variety of antiparasitic agents are used to treat parasitized animals. The effectiveness of these agents depends on the type of parasite, the animal's overall health, and the range of antiparasitic activity (Rolbiecki and Izdebska, 2024; Zhang et al., 2024). An infected animal is a constant source of pathogens and poses a threat to healthy livestock. Thus, a significant level of sanitary contamination with exogenous helminth forms (from 21.7% to 45.6%) and soil contamination (from 20% to 36.6%) was found during the examination of livestock facilities, including a pig farm, sheep farm, dairy farm, and dog training center (Paliy et al., 2018a). According to other reports, 41.3% of soil samples are contaminated with helminth eggs. The parasites detected were *Ascaris* spp. (39.0%), *Trichuris* spp. (26.0%), *Ancylostoma/Strongyloides* (22.0%), *Toxocara* spp. (4.0%), *Taenia* spp. (3.0%), and unidentified eggs (6.0%) (Paller and Babia-Abion, 2019). Gurmassa et al. (2023) report an even higher level of helminth contamination in the environment. According to their data, helminth eggs were present in 57.1% of soil samples and 18.6% of fecal

samples. The most common types of helminths were *Ascaris lumbricoides* and *Trichuris trichiura* hookworms. In Kenya, 26.8% of households have soil contaminated with one or more types of helminth eggs, with *A. lumbricoides* being the most common (19.4%) (Steinbaum et al., 2016). In Chile, at least one parasite egg was found in 24.21% of the studied samples (Castro-Seriche, Fernández and Landaeta-Aqueveque, 2020). Additionally, 52% of 29 different source samples were found to have a prevalence of potentially infectious helminth eggs (Grego et al., 2018). The widespread distribution of helminth parasites and their potential danger to livestock necessitate antiparasitic measures, such as disinfecting the environment (Paliy, 2018). Various chemical compounds are used for this purpose, differing in composition, physicochemical and toxicological characteristics, and spectrum of antimicrobial activity (Ponomarenko et al., 2021; Paliy et al., 2024b). At the same time, it has been reported that the treatment of *A. suum* eggs with many commercially available disinfectants does not affect embryogenesis. While some disinfectants can delay or stop the development of invasive *A. suum* eggs, they are unlikely to completely kill them (Oh et al., 2016).

Therefore, it is crucial to identify effective disinfectants that align with modern livestock farming requirements, offering high efficacy and cost-effectiveness.

The study aimed to develop methods for disinfecting livestock facilities contaminated with exogenous helminth forms using innovative disinfectants.

Materials and methods. Experimental studies were conducted at the Laboratory of Veterinary Sanitation, Parasitology and Bee Diseases Study in the National Scientific Center 'Institute of Experimental and Clinical Veterinary Medicine' (Kharkiv, Ukraine).

Disinfectants with different contents of active and auxiliary substances were used to evaluate the effectiveness of disinfection and develop disinfection methods.

Disinfectant No. 1 contains peracetic acid, hydrogen peroxide, acetic acid, stabilizing additives, and water.

Disinfectant No. 2 contains potassium monopersulfate, sodium dichloroisocyanurate, sodium hexametaphosphate, sulfamic acid, malic acid, sodium alkylbenzyl sulfonate, and sodium sulfate in water.

Disinfectant No. 3 contains quaternary ammonium compounds (QACs), glutaraldehyde, isopropyl alcohol, nonionic surfactants, and deionized water.

Disinfectant No. 4 contains didecylmethylammonium chloride, glutaraldehyde, benzalkonium chloride, surfactants, orthophosphoric acid, and water.

The effectiveness of the disinfectants was determined under current regulatory documents (Paliy et al., 2020b).

Results and discussions. The planned research evaluated the effectiveness of innovative disinfectants from various chemical groups for use in animal husbandry. Based on the results, scientifically proven methods of applying disinfectants were developed, along with appropriate usage methods.

First, a disinfectant composition containing optimal proportions of peracetic acid, hydrogen peroxide, acetic acid, stabilizing additives, and water was selected for the disinfection of *Ascaris suum* eggs.

Before disinfection, livestock facilities are cleaned of manure, feed residue, and debris. Technological equipment is cleaned of technical and organic contamination. The floor, ceiling, and walls are washed with pressurized water. After the water is removed from the premises, disinfection is carried out using a product with different compositions, exposure times, and consumption rates of 500 ml/m² (Table 1).

Composition 1: peroxyacetic acid — 0.2%, hydrogen peroxide — 0.3%, acetic acid — 0.48%, stabilizing additives — 0.1%, water — 98.92%.

Composition 2: peroxyacetic acid — 0.3%, hydrogen peroxide — 0.45%, acetic acid — 0.72%, stabilizing additives — 0.15%, water — 98.38%.

Composition 3: peroxyacetic acid — 0.4%, hydrogen peroxide — 0.6%, acetic acid — 0.96%, stabilizing additives — 0.2%, water — 97.84%.

Composition 4: peroxyacetic acid — 0.5%, hydrogen peroxide — 0.75%, acetic acid — 1.2%, stabilizing additives — 0.25%, water — 97.3%.

As shown in Table 1, disinfectants 1 and 2 are ineffective against exogenous helminth contamination in livestock facilities. Meanwhile, disinfectants 3 and 4

completely disinfect treated surfaces contaminated with animal helminth eggs when exposed for 48 h and applied at a rate of 500 ml/m². No corrosion was observed when the treated surfaces were examined after contact with the disinfectant.

Table 1 — Disinfection activity of disinfectant No. 1

Compo- sition	Terms for determining the viability of <i>Ascaris suum</i> eggs, days														
	3			6			14			21			28		
	Exposure, h														
	6	24	48	6	24	48	6	24	48	6	24	48	6	24	48
1	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
3	+	+	+	+	+	+	+	+	-	+	+	-	+	+	-
4	+	+	+	+	+	+	+	+	-	+	+	-	+	+	

Notes: '–' — egg death; '+' — egg development.

Based on these results, a Ukrainian patent for a utility model No. 144297 was obtained (Paliy et al., 2020a).

Such disinfectants are widely used in various fields (De Rezende, De Lima and Santos, 2023). The effectiveness of hydrogen peroxide-based disinfectants can reportedly be increased by adding silver and copper nanoparticles (Orta De Velásquez et al., 2008). The disinfecting effect of hydrogen peroxide can be enhanced through a synergistic effect with ozone (Ibáñez-Cervantes et al., 2024).

The next task involves selecting a disinfectant composition that contains potassium monopersulfate, sodium dichloroisocyanurate, sodium hexametaphosphate, sulfamic acid, malic acid, sodium alkylbenzyl sulfonate, sodium sulfate, and water. The task also involves developing a method for disinfecting veterinary control facilities. This method includes treating the facilities with a disinfectant by spraying it for 3 h at a consumption rate of 300 ml/m².

After pre-disinfection, mechanical cleaning, and water removal, disinfection is carried out with the preparation (Table 2).

Composition 1: potassium monopersulfate — 0.25%, sodium dichloroisocyanurate — 0.0125%, sodium hexametaphosphate — 0.125%, sulfamic acid — 0.0375%, malic acid — 0.0375%, sodium alkylbenzyl sulfonate — 0.025%, sodium sulfate — 0.0125%, water — 99.5%.

Composition 2: potassium monopersulfate — 0.5%, sodium dichloroisocyanurate — 0.025%, sodium hexametaphosphate — 0.25%, sulfamic acid — 0.075%, malic acid — 0.075%, sodium alkylbenzyl sulfonate — 0.05%, sodium sulfate — 0.025%, water — 99.0%.

Composition 3: potassium monopersulfate — 0.75%, sodium dichloroisocyanurate — 0.0375%, sodium hexametaphosphate — 0.375%, sulfamic acid — 0.1125%, malic acid — 0.1125%, sodium alkylbenzyl sulfonate — 0.075%, sodium sulfate — 0.0375%, water — 98.5%.

Table 2 — Disinfection activity of disinfectant No. 2

Compo- sition	Terms for determining the viability of <i>Ascaris suum</i> eggs, days														
	3			6			14			21			28		
1	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Notes: ‘-’ — egg death; ‘+’ — egg development.

As shown in Table 2, the disinfectant preparation of composition 1 is ineffective against veterinary control objects contaminated with exogenous helminth forms. Disinfectants with compositions 2 and 3 were proven to completely disinfect treated surfaces contaminated with animal helminth eggs when exposed for 3 h at a consumption rate of 300 ml/m². Upon inspection of the treated surfaces after contact with the disinfectants, no corrosion or discoloration was observed.

Based on these results, a Ukrainian patent for a utility model No. 156093 was obtained (Paliy et al., 2024a).

Our results expand the range of applications for disinfectants containing the active ingredient monopersulfate for disinfestation purposes. The effectiveness of this group of disinfectants against bacteria and viruses has previously been established (Sonthipet, Ruenphet and Takehara, 2018). These preparations are widely used in human medicine (Umemura et al., 2022).

Ren et al. (2024) reported that peracetic acid, hydrogen peroxide, and potassium monopersulfate are suitable for low-temperature environmental disinfection.

To develop a method for disinfecting surfaces contaminated with *Ascaris suum* eggs, the effectiveness of various compositions of active ingredients was investigated. These compositions contained a mixture of quaternary ammonium compounds (QACs), glutaraldehyde, isopropyl alcohol, nonionic surfactants (SASs), and deionized water. The exposure time ranged from 3 to 24 h, and the consumption rate was 500 ml/m².

After pre-disinfection, mechanical cleaning, and removal of water, the preparation is applied (Table 3).

Composition 1: mixture of QACs — 0.5%, glutaraldehyde — 0.22%, isopropyl alcohol — 0.16%, nonionic surfactants (SAS) — 0.1%, deionized water — 99.02%.

Composition 2: mixture of QACs — 0.75%, glutaraldehyde — 0.33%, isopropyl alcohol — 0.24%, nonionic surfactants (SAS) — 0.15%, deionized water — 98.53%.

Composition 3: mixture of QACs — 1.0%, glutaraldehyde — 0.44%, isopropyl alcohol — 0.32%, nonionic surfactants (SAS) — 0.2%, deionized water — 98.04%.

As shown in Table 3, treatment of the *Ascaris suum* test culture with preparations 1 and 2 did not affect the development of *Ascaris suum* eggs. Conversely, it was determined that preparation No. 3 delayed the development of the test culture eggs and ultimately led to

their death; thus, the agent exhibited disinfestation properties.

Table 3 — Disinfection activity of disinfectant No. 3

Compo- sition	Terms for determining the viability of <i>Ascaris suum</i> eggs, days														
	3			6			14			21			28		
	Exposure, h														
	3	6	24	3	6	24	3	6	24	3	6	24	3	6	24
1	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Notes: ‘-’ — egg death; ‘+’ — egg development.

Based on these results, a Ukrainian patent for a utility model No. 130430 was obtained (Paliy et al., 2018b).

The following utility model is based on developing a method to disinfect soil contaminated with *Toxocara canis* eggs. This method uses a disinfectant containing didecyldimethylammonium chloride, glutaraldehyde, benzalkonium chloride, surfactants (SAS), orthophosphoric acid, and water. The exposure time is 72 h, and the consumption rate is 500 ml/m².

After pre-disinfection mechanical cleaning and removal of water from the premises, disinfection is carried out using the following preparations (Table 4):

Composition 1: didecyldimethylammonium chloride — 0.0675%, glutaraldehyde — 0.45%, benzalkonium chloride — 0.24%, surfactants (SAS), orthophosphoric acid — 2.2425%, water — 97%.

Composition 2: didecyldimethylammonium chloride — 0.090%, glutaraldehyde — 0.60%, benzalkonium chloride — 0.32%, surfactants (SAS), orthophosphoric acid — 2.990%, water — 96.0%.

Composition 3: didecyldimethylammonium chloride — 0.1125%, glutaraldehyde — 0.75%, benzalkonium chloride — 0.40%, surfactants (SAS), orthophosphoric acid — 3.7375%, water — 95%.

Table 4 — Disinfection activity of disinfectant No. 4

Compo- sition	Terms for determining the viability of <i>Toxocara canis</i> eggs, days				
	3	6	14	21	28
	Exposure, h / consumption rate, ml/m ²				
	72 / 500	72 / 500	72 / 500	72 / 500	72 / 500
1	+	+	+	+	+
2	-	-	-	-	-
3	-	-	-	-	-

Notes: ‘-’ — egg death; ‘+’ — egg development.

As shown in Table 4, treating soil contaminated with *T. canis* eggs with compound 1 did not affect the development of the eggs. Conversely, it was found that applying compositions 2 and 3 for 72 h at a rate of 500 ml/m² delayed the development of *T. canis* eggs in the test culture and resulted in their death. In other

words, the compositions exhibited disinfection properties.

Based on these results, a Ukrainian patent for a utility model No. 137488 was obtained (Paliy et al., 2019).

Khorolskyi (2022) also recommends using glutaraldehyde-containing disinfectants to control and prevent rabbit pasalurosis, as they effectively disinfect environmental objects and animal housing facilities.

The results of the studies show that the proposed disinfection methods meet modern agro-industrial production requirements, are effective and environmentally safe, and are economical and easy to use.

It is well-known that helminthiasis spreads among susceptible animals on livestock farms and complexes when veterinary and sanitary standards are not followed. This requires the scientifically-based use of highly effective deworming and disinfection agents (Paliy et al., 2018a; Labana et al., 2024).

Many antimicrobial agents are available for disinfection in animal husbandry, presenting practitioners with the task of selecting the most effective and cost-efficient option (Paliy et al., 2020b; Tyski, Bocian and Laudy, 2024). However, not all disinfectants meet modern requirements for effective disinfection in animal husbandry. Of the disinfectants tested, solutions containing 3.0% cresol, 0.2% sodium hypochlorite, and 0.02% sodium hypochlorite delayed, but did not prevent, the embryonation of purified *A. suum* eggs after three weeks of incubation. After six weeks, undeveloped eggs

completed embryonation regardless of exposure time, except when 10.0% povidone-iodine was used. Nevertheless, a 10.0% povidone-iodine solution inactivates most eggs after 5 minutes, although it never completely kills them, even after 60 minutes of exposure (Oh et al., 2016). Therefore, prior evaluation of a disinfectant is required, along with the development of regulations for its use.

Based on the results of our research, we have established effective application methods for four modern disinfectants to destroy exogenous helminth forms.

Using the proposed disinfection methods will improve the effectiveness of health and preventive measures against parasitic diseases in susceptible animals in agricultural enterprises.

Conclusions. Regimens for using aldehyde- and acid-containing disinfectants to decontaminate livestock facilities contaminated with exogenous helminths have been developed. These regimens can be used on livestock farms and complexes.

These methods of disinfection in animal husbandry using these agents meet environmental protection requirements and ensure the production of safe, high-quality animal products. They are also easy to use, highly effective, and cost-effective.

Results obtained from the practical implementation of these methods will allow us to propose new, effective disinfection protocols.

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