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The effect of a biocidal paint as a disinfectant and insecticide on the growth performance and blood biochemical parameters of broiler chickens

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SUMMARY

The aim of this study was to evaluate the effects of liming and a biocidal paint, used in poultry for disinfection and insect control, on the growth performance and blood biochemical parameters of broiler chickens. It was postulated that in successive trials, the active ingredients contained in the slow-release, water-based, long-lasting biocidal paint would not significantly compromise bird performance or blood biochemistry. The experiment covered three production cycles. Each production cycle was an individual trial, and thus there were three trials during the experiment. Two groups were studied in each trial, and each group consisted of one poultry house. The rearing conditions, diet, length of the rearing period, and building specifications were the same for both groups in each trial. The experiment was performed on unsexed Ross 308 chickens that were raised until 40 days of age under standard conditions. The distinguishing (experimental) factor was the method of disinfection and insect control liming (group L) was used in one poultry house and biocidal paint (group BP) in the other. Broilers from group BP (biocidal paint) were found to have better growth performance than birds from group L (liming). Application of the biocidal paint had no significant long-term negative effect on the growth performance or blood biochemical parameters of the chickens. However, changes were noted in blood cholesterol and triglyceride concentrations, depending on the disinfection insect control method. The average values of these parameters were significantly higher in group BP, but the differences were at the significance threshold. The results of the study suggest that the innovative biocidal paint can be used in poultry houses as an effective disinfectant and insecticide.

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INTRODUCTION

The global poultry industry has been growing dynamically for many years (Mottet and Tempio, 2017; Biesek et al., 2020). The European poultry market is expected to continue to grow (van Limbergen et al., 2020), but at a much slower rate than in the last decade. Market growth will be less dynamic, but the long-term outlook is still optimistic (EC, 2019). Poultry is popular among consumers on account of its high quality and lower price in comparison with other meat types.

The rapid growth of the poultry industry and increasing poultry consumption necessitate measures to maintain high broiler performance and promote bird health. This is not an easy task, and despite considerable research, disease control in poultry production continues to be a serious challenge (Battersby et al., 2017). The main sources of infection in poultry farming include the employees (Ridley et al., 2011), other livestock, the immediate environment (Ogden et al., 2014), pests (such as the lesser mealworm) (Mituniewicz and Dzik, 2020; Dzik and Mituniewicz, 2020), rodents (Cavia et al., 2019) and farm equipment (Battersby et al., 2016).

The production cycle, on broiler farms is particularly short, which promotes intensive rearing (Biesek et al., 2020; Wahyono and Utami, 2018). Flock health management is a critical consideration on all poultry farms, as it contributes to higher profits. Preventive procedures such as cleaning and disinfection are essential for maintaining high biosecurity standards on farms, improving performance, and keeping birds in good health (de Castro Burbarelli et al., 2017). Many infectious and non-infectious agents that compromise broiler performance and increase mortality have been identified (Jones et al., 2018). Considerable progress in disease prevention and control has been made over the years, but poultry diseases, in particular infectious diseases, are very difficult to eliminate from commercial poultry farms. For this reason, cleaning and disinfection should play a key role in daily farming practice (de Castro Burbarelli et al., 2015).

Numerous commercial products are available to help to meet high hygiene standards on poultry farms. These products should be safe for humans and birds and should not affect poultry performance or health. However, the extent to which disinfection and insect control directly affect broiler performance and health has not often been studied. Further research is needed to assess the effectiveness of commercial disinfectants and insecticides (Witkowska et al. 2019; Dzik and Mituniewicz, 2020; Dzik et al. 2022) and their impact on poultry (Witkowska et al. 2007). Therefore, this study was undertaken to determine the effects of liming and a biocidal paint, applied in poultry houses for disinfection and insect control on the growth performance and blood biochemical parameters of broiler chickens. It was postulated that in successive trials the active ingredients contained in the biocidal paint would not significantly influence bird performance or blood biochemistry. This is an important consideration, as the biocidal paint is a slow-release, water-based, long-lasting disinfectant and insecticide.

MATERIALS AND METHODS

As the experiment involved a routine hygiene procedure carried out on a production farm the consent of the Local Ethics Committee for Experiments on Animals was not required by Polish law, as no treatment, including medical treatment, invasive diagnostics, or procedures causing psychological or social discomfort to the participants, were used in the study. The experiment was conducted in accordance with European Union (EU) Directive 2010/63/EU.

Housing

Production trials were carried out on a large commercial poultry farm (Pomerania region, Poland). The experiment involved three production cycles. Each production cycle was an individual trial, so that there were three trials during the experiment. Two groups were studied in each trial, and each group consisted of one poultry house (Table 1). The rearing conditions, diet, length of the rearing period, and building specifications were the same for both groups in each trial. The distinguishing (experimental) factor was the method of disinfection and insect control liming (group L) was used in one poultry house and biocidal paint (group BP) in the other.

Table 1

Experimental design

Experiment	Group L	Group BP
Trial I	Poultry house with limewash	Poultry house with biocidal paint
Trial II	Poultry house with limewash	Poultry house with biocidal paint
Trial III	Poultry house with limewash	Poultry house with biocidal paint

The experiment was performed on unsexed Ross 308 chickens that were raised until 40 days of age under standard conditions, on straw bedding, in buildings with controlled temperature, humidity and ventilation. Rearing in each group began with 20 500 birds. The stocking density was ~43 kg/m^2 (the poultry producer had obtained the necessary authorization to exceed the standard maximum stocking rate as specified in the Regulation of the Minister for Agriculture and Rural Development of 15 February 2010 on the requirements and procedures for keeping species of farm animals for which standards of protection are laid down in European Union legislation). The birds had free access to drinking water and were fed complete diets *ad libitum*, in accordance with poultry nutrient requirements in successive growth stages (starter: 1-10 days, grower I: 11-20 days, grower II: 21-31 days, finisher: 32-40 days). The nutritional value of the diets is presented in Table 2 (nutritional value of the feed according to the manufacturer's label).

Table 2

Nutritional value of chickens' diets

Composition (%)	Starter	Grower I	Grower II	Finisher
Crude protein	22,95	20,95	19,74	19,20
Crude fat and oil	5,72	5,87	6,71	7,47
Crude ash	5,39	4,49	4,04	3,88
Crude fibre	3,37	3,26	2,93	2,81
Lysine	1,37	1,22	1,16	1,10
Calcium	0,80	0,61	0,51	0,46
Methionine	0,59	0,53	0,51	0,49
Total phosphorus	0,53	0,43	0,35	0,34
Sodium	0,14	0,13	0,13	0,13

Bedding was removed after each production cycle, and the houses were cleaned and disinfected for three weeks. Limewash was applied in group L, and a slow-release, water-based long-lasting biocidal paint (Kleib Sp. z o.o., Brzesc Kujawski, Poland) was applied in group BP, as comprehensive protection against pathogens and insects. In group L, before each production cycle, the house was cleaned and the walls and floor were fogged with limewash using a hydrodynamic painting unit (Faska Sp. z o.o., Fabianki, Poland) at 300 mL/m^2. In group BP, the house was cleaned and the walls and floor were painted with a biocidal paint using a hydrodynamic painting unit (Faska Sp. z o.o., Fabianki, Poland) at 400 mL/m^2, in accordance with the manufacturer's instructions, only once at the beginning of the experiment (Table 3).

Table 3

Application of hygiene method

Trial	Application	
	Limewash	Biocidal paint
I	7 days before trial I	
II	7 days before trial II	7 days before trial I
III	7 days before trial III	

The biocidal paint, which is intended for use in poultry houses, contains the following active ingredients:

- insecticide active ingredients:
  - chemical – permethrin (0,04% by weight), a synthetic pyrethroid widely used against insects (Bissinger et al., 2011; Fankhauser et al., 2015; Pham et al., 2019; PubChem, 2022). Permethrin acts as a contact killer and stomach poison and affects the nervous system of insects; it is effective against eggs, larvae, pupae and adults (Hartley and Kidd 1983); it is fast-acting and highly effective even in low doses (Zaim et al., 2000; Mosqueira et al., 2010).
  - optical – a mixture of ultramarine and violet 23 (0,004% by weight) with repellent properties (Pubchem, 2022). As numerous beetle species have blue- and/or violet-sensitive photoreceptors (Cronin and Bok 2016; Sharkey et al., 2017), the addition of a blue pigment (ultramarine) and a violet pigment (violet 23) to the paint may provide additional protection against *Alphitobius diaperinus*.
- antimicrobial active ingredients:
  - zinc pyrithione (0,12% by weight) – inhibits fungal growth through copper influx and damage to iron-sulphur proteins (Reeder et al., 2011; Monte-Serrano et al., 2015); unlike other antimicrobial ingredients such as silver and copper, zinc-based ingredients are broad-spectrum antimicrobial agents (Abd Ali et al., 2021).
  - 1,2-benzisothiazol-3(2H)-one (0,05% by weight) – a broad-spectrum antimicrobial agent (Wang et al., 2018; Di Martino, 2020).

The biocidal paint was certified by the National Hygiene Institute in Warsaw, Poland (No. HK/B/0571/01/2016) and patented at the Polish Patent Office (No. 232978).

Growth performance

In each trial, 250 broiler chickens selected randomly from group L and group BP were weighed at 1, 7, 14, 21, 28, 35 and 40 days of age to determine their body weights (BW, kg) and body weight gain (BWG). Mortality (%) was monitored daily. The feed conversion ratio (FCR), which defines feed intake in g per g BWG, was calculated for the entire experimental period (1-40 days) and for each week of rearing (Colombino et. al., 2020). Additionally, at the end of rearing, the European Broiler Index (EBI) was calculated based on the following formula (Zaboli et al., 2017):

$$EBI = \frac{BW \ \times survival\,rate \ \times 100}{PP \ \times FCR},$$

where: BW – body weight (kg), survival rate – 100 - mortality (%), PP – length of the production period (days), FCR – feed conversion ratio (g feed/g body weight gain).

Blood parameters

In each trial, blood was collected post mortem from the jugular vein of 250 broilers (selected randomly from group L and group BP). Whole blood was placed in plastic test tubes containing heparin as an anticoagulant. The samples were centrifuged at 3000×g for 10 min at room temperature, and plasma was collected for further analyses. Blood samples were analysed for levels of aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transpeptidase (GGT), urea, cholesterol and triglycerides in the Cobas 6000 analyser according to commercial keys (F. Hoffmann-La Roche Ltd., Basel, Switzerland). Laboratory analyses were performed in the certified Analytical Laboratory at the Municipal Hospital in Olsztyn, Poland (ISO 9001:2000 certificate No. 134B-2002-AQ-GDA-RvA).

Statistical analysis

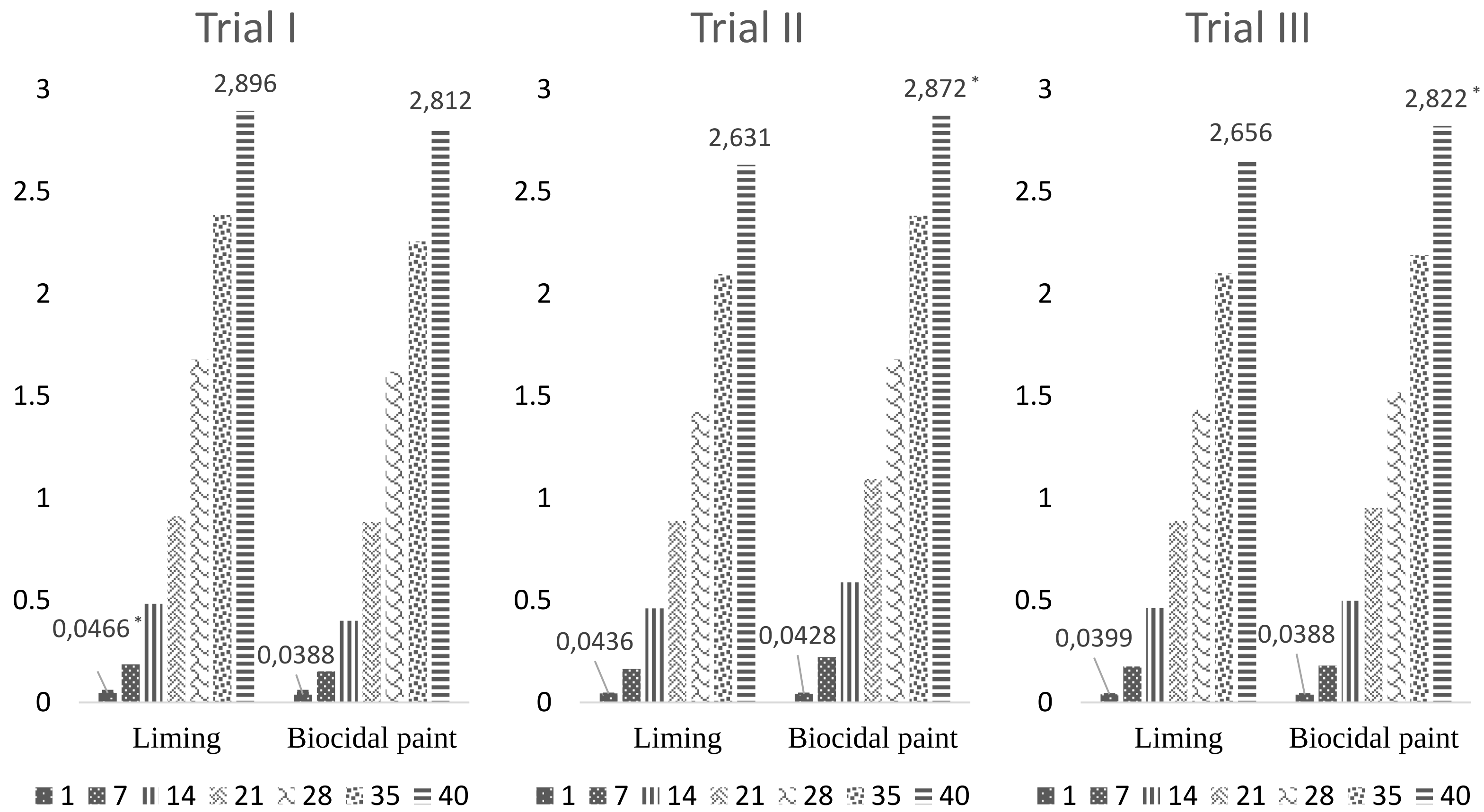
Statistical analyses were performed using Statistica v. 13.3.0 software (TIBCO Software, 2017). The normality of data distribution was verified with the Shapiro–Wilk test. The mean values of the other growth performance and blood biochemical parameters of broilers in groups BP and L were compared by Student's *t*-test. A one-way repeated measures analysis of variance (ANOVA) followed by Tukey's test was used to evaluate whether application of the biocidal paint had a significant long-term effect on the growth performance or blood biochemical parameters of birds in successive trials. In all tests, the results were regarded as significant at $P < 0{,}05$.

RESULTS AND DISCUSSION

In trial I, one-day-old broilers from groups L and BP differed significantly in initial BW, but no significant differences were found in their final BW. In trials II and III, no significant differences were noted in the initial BW of birds from groups L and BP, whereas their final BW was significantly different (Fig. 1). Application of the biocidal paint had no long-term significant effect on the BW of chickens – the average values of this parameter did not differ significantly across trials.

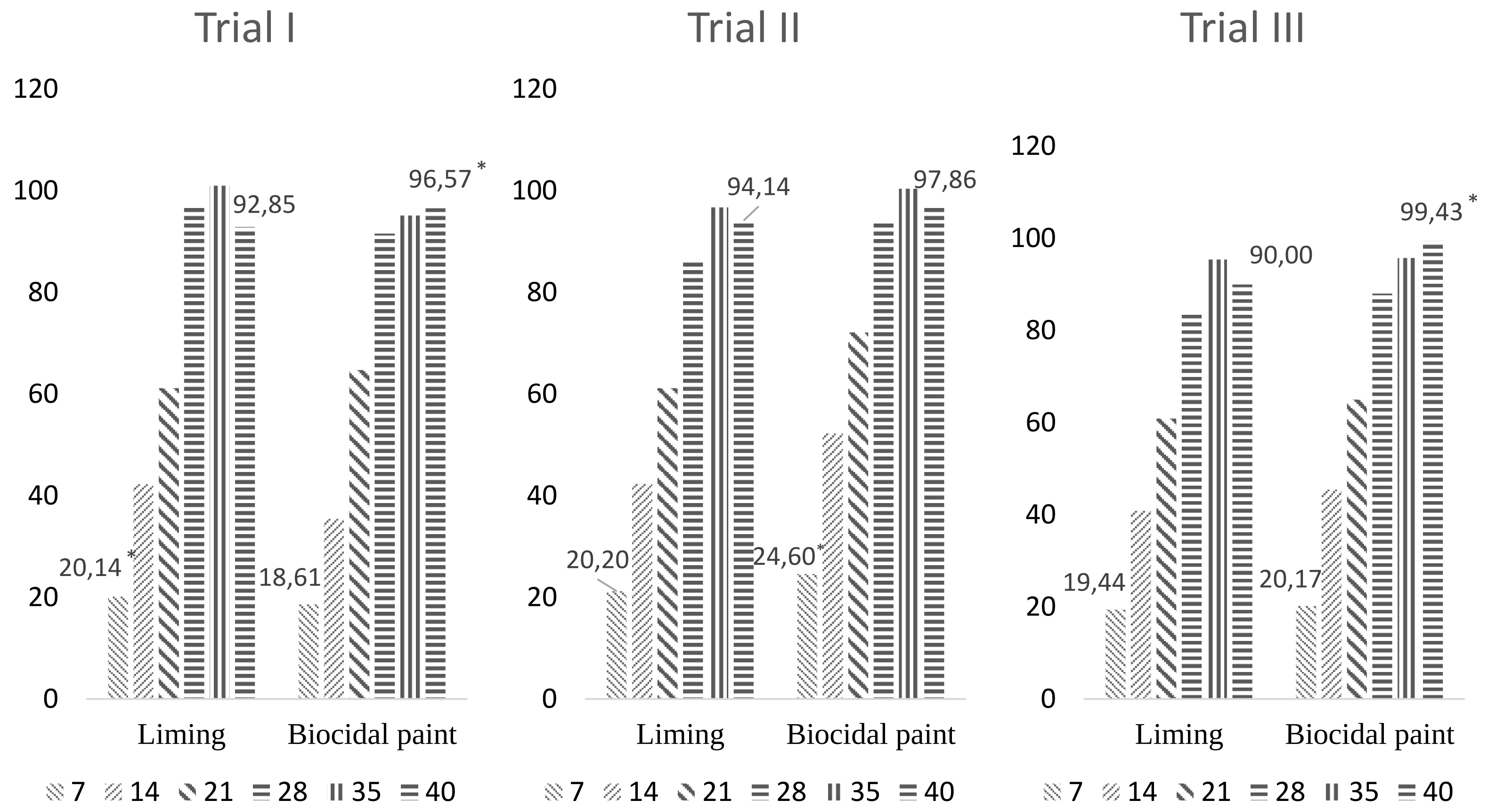
Body weight gain (BWG) was similar in both groups throughout the experiment, and the differences observed were at the significance threshold. However, significantly higher values of this parameter were noted more frequently in group BP (Fig. 2). Application of the biocidal paint had no significant long-term effect on the BWG of chickens – the average values of this parameter did not differ significantly across trials.

It should be stressed that irrespective of the disinfection and insect control method used, all 40-day-old broilers had BW of 2,631 kg to 2,896 kg. In trials II and III, the final BW of birds was significantly higher in group BP than in group L. Moreover, long-term exposure to the biocidal paint did not decrease the BW of birds throughout the experiment. The body weights of broilers noted in this study were comparable with those reported by Witkowska et al. (2007) and higher than those reported by other researchers for birds raised under similar management and housing conditions (de Castro Burbarelli et al., 2015; de Castro Burbarelli et al., 2017; Witkowska et al., 2019). The BW and BWG values obtained in the research are similar to the standard growth rate of Ross 308 chickens (Aviagen, 2019).



*significant differences ($P < 0,05$) between groups L (liming) and BP (biocidal paint). Long-term exposure to the biocidal paint had no significant effect on the average values of the parameters across trials.

Fig. 1. Average body weight of broiler chickens (kg) during rearing (days)



*significant differences ($P < 0,05$) between groups L (liming) and BP (biocidal paint). Long-term exposure to the biocidal paint had no significant effect on the average values of the parameters across trials.

Fig. 2. Average daily body weight gain (g) of broiler chickens during rearing (days)

Table 4

Average values of performance parameters of broiler chickens in trials I, II and III

	Parameter		Liming	Biocidal paint
Trial I	Initial body weight (kg)		0,0466±0,28	0,0388±0,22
	Final body weight (kg)		2,896±99	2,812±95
	Mortality (%)		5,16	1,98
	Feed conversion rate (g feed/g body weight gain)	1-7 days	0,71±0,17	0,68±0,10
		7-14 days	1,15±0,22	1,22±0,19
		14-21 days	1,35±0,12	1,31±0,08
		21-28 days	1,52±0,11	1,47±0,18
		28-35 days	1,69±0,06	1,64±0,02
		35-40 days	1,83±0,07	1,89±0,09
		1-40 days	1,75±0,13	1,74±0,15
	European Broiler Index		392,44	395,07
Trial II	Initial body weight (kg)		0,0436±0,31	0,0428±0,25
	Final body weight (kg)		$2,631^{b}$±111	$2,872^{a}$±119
	Mortality (%)		5,36	1,63
	Feed conversion rate (g feed/g body weight gain)	1-7 days	0,73±0,10	0,80±0,04
		7-14 days	1,22±0,08	1,25±0,07
		14-21 days	1,44±0,02	1,46±0,05
		21-28 days	1,62±0,07	1,60±0,10
		28-35 days	1,74±0,05	1,77±0,09
		35-40 days	1,89±0,05	1,88±0,05
		1-40 days	1,74±0,05	1,77±0,11
	European Broiler Index		349,77	407,21
Trial III	Initial body weight (kg)		0,0399±0,19	0,0388±0,20
	Final body weight (kg)		$2,656^{b}$±107	$2,822^{b}$±110
	Mortality (%)		5,43	1,36
	Feed conversion rate (g feed/g body weight gain)	1-7 days	0,99±0,05	1,01±0,02
		7-14 days	1,28±0,05	1,31±0,01
		14-21 days	1,49±0,03	1,50±0,09
		21-28 days	1,70±0,02	1,69±0,07
		28-35 days	1,88±0,04	1,80±0,05
		35-40 days	1,94±0,02	1,95±0,04
		1-40 days	1,77±0,03	1,75±0,04
	European Broiler Index		357,10	396,56

Different lowercase letters (a, b) indicate significant differences ($P < 0,05$) between groups L (liming) and BP (biocidal paint). Long-term exposure to the biocidal paint had no significant effect on the average values of the parameters across trials standard deviation (SD).

Research has shown that mortality in poultry flocks is determined by numerous factors, and that it has decreased in recent years (Witkowska et al., 2007; van Limbergen et al., 2020; Utnik-Banaś, 2017; Witkowska et al., 2019), most likely due to improved flock management strategies and the search for innovative technological solutions in poultry farming. In the present study, in trial I, bird mortality was 3,18% lower in group BP than in group L. In trials II and III, the difference between groups reached 3,73% and 4,07%, respectively. According to Szőllősi and Szűcs (2014), average mortality rates in broilers in selected EU member states range from 3,7% to 4,5%. In the current experiment, mortality in group BP never exceeded 2%, which is highly satisfactory in terms of both production profitability and bird welfare. Properly selected disinfection and insect control methods play a key role in poultry farms. Moreover, increased mortality generates economic losses.

The FCR in both groups was somewhat higher than the values reported by de Castro Burbarelli et al. (2015) and Witkowska et al. (2019) and the standard growth rate of Ross 308 chickens (Aviagen, 2019). On the other hand, higher FCR values were noted in a study investigating the effects of dietary modifications on the productive performance of broilers (Mohammed and Abbas, 2009). According to Szőllősi and Szűcs (2014), the average FCR for broilers in Poland is 1,76; and similar values were obtained for both groups in this study. The FCR was better in group BP, as manifested by higher final BW at comparable feed consumption.

Broiler production efficiency in both groups was evaluated based on the EBI. The higher the value of the index, the higher the performance and the better the health status of birds (Bhamare et al., 2016; Kryeziu et al., 2018). In the present study, the EBI ranged from 349,77 to 407,21, and it was higher in group BP in each trial.however, the EBI values were similar in both groups, most likely because final BW was higher in group L, possibly due to its significantly higher initial value in comparison to group BP. These results indicate that the biocidal paint did not compromise bird performance, as the EBI values did not decrease in successive trials. Agricultural progress has contributed to an improvement in the performance parameters analysed in this study, leading to better economic results.

Analysis of the average values of blood parameters presented in Table 5 revealed changes in blood cholesterol and triglyceride concentrations in trials II and III, depending on the disinfection and insect control method. The average values of these parameters were significantly higher in group BP in trials II and III, but the differences were at the significance threshold. No significant differences in the levels of AST, ALT, GGT or urea were found between groups. Long-term exposure to the biocidal paint had no significant effect on blood parameters, whose average values did not differ significantly across trials.

Table 5

Average values of selected blood biochemical parameters of broiler chickens in trials I, II and III

Parameter	Trial I		Trial II		Trial III	
	Liming	Biocidal paint	Liming	Biocidal paint	Liming	Biocidal paint
AST (U/L)	338,73±56,23	329,92±56,39	281,08±88,02	291,76±88,90	301,04±60,59	328,79±55,81
ALT (U/L)	2,78±0,05	2,79±0,11	2,71±0,09	2,65±0,03	2,67±0,10	2,71±0,11
GGT (U/L)	20,01±1,31	21,72±1,75	19,98±1,15	20,99±1,44	22,21±2,18	21,91±1,64
Urea (mg/dL)	1,21±0,23	1,28±0,12	1,38±0,28	1,30±0,21	1,29±0,15	1,25±0,10
Cholesterol (mg/dL)	169,21±5,34	168,77±7,21	153,47^{b}±4,98	171,58^{a}±11,21	151,56^{b}±5,01	169,26^{a}±9,58
Triglycerides (mg/dL)	128,09±2,32	126,07±2,61	117,25^{b}±2,09	125,49^{a}±2,58	115,28^{b}±1,99	128,51^{a}±2,78

AST – aspartate aminotransferase, ALT – alanine aminotransferase, GGT – gamma-glutamyl transpeptidase
Different lowercase letters (a, b) indicate significant differences (P < 0,05) between groups L (liming) and BP (biocidal paint). Long-term exposure to the biocidal paint had no significant effect on the average values standard deviation (SD) of the parameters across trials.

Changes in blood biochemical parameters can be the first symptom of disease or poisoning with toxic substances (Jurani et al., 2004; Rajman et al., 2006; Özeby and Esen, 2007; Tessari et al., 2010; Erdelyi et al., 2011). These parameters are more difficult to interpret in poultry than in other livestock species. Birds are characterized by greater individual variation than in other animals, and the ranges of physiological parameters are broader (Fritz et al., 2000; Piotrowska et al., 2011; Kowalczuk-Vasilew et al., 2017).

The liver is the main organ responsible for numerous digestive, metabolic and production processes; for this reason its susceptibility to damage can vary. Liver damage is assessed based on the serum concentrations of specific liver enzymes. These enzymes can lead to dysfunctions in various bodily systems, which can compromise bird health and performance. AST, ALT and GGT are bioindicators of liver function; increased levels of these enzymes are usually indicative of liver damage (Aikpitanyi and Egweh, 2020).

Many researchers have found that AST activity in chickens increases with age (Silva et al., 2007; Tessari et al., 2010; Kudair and Al-Hussary, 2010). High or excessive AST levels can be indicative of poisoning with toxic substances. However, AST activity is also observed in other organs (the heart, kidneys, brain and skeletal muscles), and the specificity of this enzyme as an indicator of liver damage may be limited (Tessari et al., 2010). According to Kokore et al. (2021), the AST level in poultry should not exceed 230 U/L. In the present study, AST levels exceeded this value in both groups (L and BP), ranging from 281,08 to 338,73 U/L during the entire experiment. Nevertheless, biocidal paint does not appear to exert negative effects on AST activity.

Serum ALT levels were determined in order to test for any toxic effect of the biocidal paint (Ozer et al., 2008). No significant differences in the activity of this enzyme were noted in response to the disinfection and insect control methods, and it was comparable in all birds.

The levels of GGT, which is a useful indicator of liver disease, were also determined in this study. GGT levels in poultry are usually highest at 35 days of age. This may be explained by the fact that feed intake is highest in the fifth week of production, which intensifies metabolic processes in the liver (Silva et al., 2007). In the present study, GGT levels ranged from 19,98 to 22,21 U/L. Similar values (20,40-23,15 U/L) were reported by Safaeikatouli et al. (2010), whereas GGT activity in the blood serum of broilers was lower (by about 5,5 U/L) in a study by Subapriya et al. (2007). These results confirm that GGT activity is influenced by high genetic variation in birds as well as by other factors.

Serum urea levels in birds and their health implications have been studied since the first half of the 20th century (Dyer and Roe, 1933; Howell, 1939). The reference range for birds is 3,3-7,2 mg/dL. Urea is the final product of protein metabolism in the body. High urea levels may be indicative of kidney dysfunction. In birds fed protein-rich diets, excess urea is usually excreted in the urine (Karsawa and Maeda, 1994). To exclude the effect of this factor in the current experiment, all broilers were fed identical diets with identical protein content. Serum urea levels ranged from 1,21 to 1,38 mg/dL in both groups. These values can be considered normal and indicative of adequate dietary protein utilization and the absence of toxic substances in the birds' immediate environment (Nworgu et al., 2007; Polat et al., 2011).

Blood and serum cholesterol levels are also affected by genetic factors, feed type, and medicines (Hargis, 1988). Serum triglyceride levels are mainly determined by diet, hormones and diseases (Regar et al., 2019). In the present study, the blood cholesterol concentrations of broilers ranged from 151,56 to 169,21 mg/dL in group L, and from 168,77 to 171,58 mg/dL in group BP. The respective values for triglyceride concentrations were 115,28-128,09 mg/dL and 125,49-128,51 mg/dL. According to Regar et al. (2019), normal serum cholesterol and triglyceride levels in broiler chickens are 125 to 200 mg/dL and $\leq$ 150 mg/dL, respectively. Even the highest values of these parameters determined in this study did not exceed those limits. The elevated blood cholesterol and triglyceride concentrations in chickens from group BP may have been associated with their significantly higher BW. Cholesterol comes from two sources: exogenous cholesterol is derived from feed, and endogenous cholesterol is produced by the body (Regar et al., 2019).

Based on the results of the liver enzyme analysis and serum urea levels, as well as the cholesterol and triglyceride levels, no toxic effects for chickens should be attributed to the biocidal paint, especially given that changes in the biochemical parameters of the blood are most often the first symptom of poisoning by toxic substances.

CONCLUSIONS

The biocidal paint was used only once as a disinfectant and insecticide and did not compromise the growth performance or blood biochemistry of broiler chickens. Long-term exposure to the biocidal paint also had no significant negative effect on bird performance or blood biochemical parameters. The results of this study suggest that the biocidal paint can be recommended as a long-lasting disinfection and insect control method that is less laborious to apply than liming and positively affects the performance and health status of chickens by improving sanitary conditions in the poultry house.

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