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The Influence of the Mineral–Microbial Deodorizing Preparation on Ammonia Emission and Growth Performance in Turkey Production

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Abstract: In our previous in vitro research and also in laying hen production, attempts were made to minimise ammonia emissions in poultry houses with the use of Deodoric® biopreparation. The objective of the present research was to evaluate the influence of the Deodoric® on ammonia (NH_3) emission and turkey growth performance in a semi-industrial production system. Significant differences in NH_3 emission (p-value < 0.001), body weight (p-value < 0.001) and relative humidity (p-value < 0.001) were observed between the control group (C) and the experimental group (E) where Deodoric®was applied. In group C, an increase in ammonia concentration in air could have contributed to a decrease in the body weight of turkeys, but the above correlation was not observed in group E. In the control group, a relatively strong correlation between NH_3 emission and temperature (p-value = 0.0009; r = 0.74) and moderate correlations between NH_3 emission vs. relative humidity (p-value = 0.01; r = 0.59), air speed (p-value = 0.015; r = 0.60) and cooling (p-value = 0.005; r = 0.66) were noted. Studied correlations were not observed in group E. The preparation did not affect microbial levels in manure or body samples. Throughout the experiment, significant differences in the number of mesophilic bacteria (for the model: F = 46.14, p-value = 0.09; for mesophilic microorganisms: F = 3.29, p-value = 0.045) and *Campylobacter* spp. (for the model: F = 24.96, p-value = 0.008; for *Campylobacter* spp.: F = 0.25, p-value = 0.64) were not observed between group C and group E. The administration of Deodoric® to manure decreased NH_3 concentration in the air and increased weight gains in the experimental group of turkeys relative to group C. Preparation may be applied in poultry farms to improve poultry farming conditions.

Keywords: ammonia; animal welfare; deodorizing additive; farm hygiene; poultry

1. Introduction

Agricultural production, including farming and animal breeding, can be a source of high emission of odorous gases in air. Both animal-related and odorous gases are closely related to manure production. Manure is responsible for greenhouse gases and odorous gases [1]. Odorous substances include aldehydes, ketones, sulphur compounds and nitrogen compounds, including ammonia. According to research, ammonia (NH_3) emission is most significantly correlated with the emission of odorous gases [2,3]. Livestock manures contain N in both organic (proteins, amino polysaccharides, and nucleic acids) and inorganic forms. The conversion of organic nitrogen to NH_3 is mediated by a host of enzymes produced by heterotrophic microbes [4].

Ammonia has been implicated in air, soil, and water degradation [5]. Its control is currently an important problem due to environmental protection and cost-effectiveness [6]. The emission and composition of odorous substances are determined by many factors, including biofilm, sanitary conditions and nutrition (protein intake) [3]. Currently, numerous provisions referring to the problem of ammonia as an important air pollutant have been introduced in Europe [7–11].

Global ammonia emissions in 1990 were estimated at 54 Tg N yr^{-1}, and the most important emitters were China, India and Europe [12]. In 2008, it was estimated that the global NH_3 emission from agricultural systems is 27–38 Tg N yr^{-1} [13]. Since 1990 in Europe, the emissions have been reduced over time in countries; however, currently, we are observing stagnation [14]. The agricultural sector remains the major source of NH_3 emissions; despite emissions falling by 26% since 1990, agriculture contributed 96% of total emissions in 1990, and 94% in 2011 [14]. The majority of NH_3 in the atmosphere arises from livestock manure [15]. Animal excreta are responsible for around 80% of ammonia emissions in agriculture [16]. Most of the emissions from livestock production come from animal houses and storage systems (31–55%); smaller contributions come from the spreading of animal manure (23–38%) and grazing animals (17–37%) [13,15]. Livestock farms (cattle and pigs) are regarded as the main sources of ammonia in the agricultural sector [17–19]. Livestock farming is responsible for around 75% of global NH_3 emissions that occur at all stages of manure production and animal breeding in Europe [3,20]. In example, poultry rearing produces approximately 6% of the total NH_3 emissions in the air in Germany [21]. Van der Hoek [16] calculated that one turkey emits 0.92 kg NH_3 $animal^{-1}$ yr^{-1}. Other studies revealed that the average daily mean emission rate of ammonia by one laying hen was 0.95 g [22]. According to the European Economic and Social Committee, global ammonia emissions will continue to increase due to population growth and, consequently, increased animal production. [23] In 2016, the European Union introduced the National Emission Ceilings Directive which requires that the EU countries must submit an annual NH_3 emission. This will ensure better monitoring of the current problem. The majority of the reduction in NH_3 emissions is due to the combination of reduced livestock numbers across Europe (especially cattle) and the lower use of nitrogenous fertilisers. Despite the above, this issue has not yet been properly resolved in poultry breeding, which is now considered as the future of the European agricultural sector, as evidenced by the continuous increase in the number of poultry flocks.

Previous studies revealed that ammonia increases susceptibility to disease and inhibits poultry growth [24–26]. According to many studies, odorous substances, including ammonia, pose a threat to the environment, humans and animals, and therefore future studies should concentrate on decreasing odorous gas emissions [27–32]. Measures aiming to decrease and control ammonia concentration should play an important role in the protection of public health and should also be controlled to protect the health of farm personnel [33,34]. Rylander and Carvalheiro [35] reported that farm employees are at increased risk of respiratory infections, chronic bronchitis and toxic pneumonia. The negative effects of NH_3 on cells cultures were observed by Nowak et al. [31,32]. In the cited study, odorous gases, including ammonia, exerted an adverse influence on chicken LMH cells [31,32]. Moreover, ammonia combined with other factors, such as dust, may negatively affect the human and animal respiratory system [27–30]. Moreover, NH_3 reacts with acidic compounds in atmosphere and forms PM2.5 particles that cause lung diseases [15,36]. In the European Union, for each animal species kept indoors, the NH_3 threshold is 20 ppm [37]. The American Conference of Governmental Industrial Hygienists set the human threshold exposure limit to ammonia concentration in air of 35 ppm at 15 min [38]. Exposure to ammonia concentration of 300 ppm in air can be hazardous to human health and may be life threatening [39]. In studies of other poultry species, high ammonia emission compromised body weight in chickens [39–43]. For this reason, high ammonia concentration may cause financial losses in poultry production [44]. Exposure to ammonia can prolong the rearing period due to a higher feed conversion ratio. Therefore, innovative solutions in environmental and public health protection are required to promote the development of modern poultry farming that guarantees high levels of security for farm employees, the environment and the communities residing in the

vicinity of poultry farms. Currently, there is an increase in awareness of the effects of atmospheric ammonia pollution, especially in relation to human health and animal production [45].

New solutions should be developed to address the problem of ammonia emissions in poultry houses. Whyte stated that gas and dust emissions in animal housing can be controlled only partially [46]. Currently, ammonia contamination in animal farms is regulated mainly by biofilters [47], which are expensive and not always reliable. Chemical substances for decreasing ammonia concentration have been proposed, including phosphoric acid (H_3PO_4), ferrous sulphate heptahydrate ($FeSO_4 \cdot 7H_2O$) and alum [$Al_2(SO_4)_3 \cdot 18H_2O$] [48–51], but they may lead to the excessive accumulation of potentially toxic substances in the environment. Recently, Anderson et al. [52] developed new litter amendment made from alum mud, bauxite, and sulfuric acid which may be used as an effective alternative litter amendment for reducing NH_3 emissions from poultry litter. According to Santoso et al. [53], the supplementation of dried *Bacillus subtilis* cultures in poultry diet significantly decreased NH_3 emission in poultry houses. Attempts have also been made to control ammonia emissions with the use of innovative products, such as biochar or ammonia-oxidizing bacteria [54,55].

A microbiological-mineral preparation for deodorization, Deodoric®, was formulated to solve the problem of high NH_3 concentration in the air in poultry farms. In our previous studies, attempts were made to minimize odour emissions at poultry farms using Deodoric®. Deodorizing preparation was prepared according to the procedure described by Borowski et al. [56]. Biopreparation consists of spray-dried microcapsules with six active bacterial strains. The deodorizing biopreparation may inhibit the growth of opportunistic pathogens in poultry litter [57]. The product also decreases the emission of odorous gases, including indole, pyridine, hydrogen sulphide hydrocarbons, aldehydes and phenols. The tested biopreparation was microbiologically stable throughout the previous experiments [58]. Under laboratory conditions, odorous compounds in exhaust air, especially ammonia, was reduced by more than 90% after 2 days.

The experiment presented in this manuscript was carried out as part of a three-year project (a mineral-microbial preparation for the removal of odorous compounds from poultry production premises) that was conducted in several stages.

This article is a continuation of studies carried out in laboratory and model conditions. Samples of poultry litter (2–5 kg) were evaluated under laboratory conditions (including the methodology of application, doses, investigation of odorous gases inhibition, the influence of the sorbent, determination of the Deodoric's composition and microbiological research). Five animals per experimental and control group were studied under model conditions in three repetitions (this stage of research involved the evaluation of odorous gases decrease, doses and application methods, the influence of the sorbent, an assessment of microclimate conditions and microbiological analyses).

The current article is a continuation of the research conducted in the industrial scale in poultry, carried out on laying hens. However, research on a different production group and a different species of poultry is needed due to different technologies in animal production. [59]

The objective of this research was to investigate the influence of the mineral-microbial preparation on NH_3 concentration, emission and final body weight of turkeys in a semi-industrial production system.

2. Experiments

2.1. Broiler Turkeys and Production Premisses

On 20 December 2016, 10-week-old female Big-6 broiler turkeys (commercial turkey breed—Hybrid Converter; producer—Hybrid Turkey, Olsztyn, Poland) were transported from an industrial farm to the Department of Avian Diseases, Faculty of Veterinary Medicine of the University of Warmia and Mazury in Olsztyn. This unit has a specialized pavilion for proper animal keeping. The animals were handled in accordance with the guidelines for Care and Use of Laboratory Animals of the Faculty of Veterinary Medicine at the University Warmia and Mazury in Olsztyn and the National Research Council [60]. The experiment was performed in two separated turkey houses with an area of 20 m^2

each, and it lasted 88 days. On 22 March 2017, turkeys were euthanized in the specialized abattoir (the Department of Commodity Science and Animal Improvement, Faculty of Animal Bioengineering of the University of Warmia and Mazury in Olsztyn).

Turkeys were divided into the experimental group (E) and control group (C), which consisted of 50 birds each. Turkeys were fed specialized diets ad libitum from automatic feeders with free access to water. Birds received a complete feed (composition: crude protein 19.20%, oils and crude fats 4.20%, crude fiber 3.50%, raw ash 5.70%, lysine 1.17%, methionine 0.44%, calcium 1.00%, phosphorus 0.57%, sodium 0.16%). Microclimate conditions were operated by automatic climate controller Stienen MFC-3VAD (Stienen Bedrijfselektronica b.v., Nederweert, Netherland). This device has minimum ventilation functionality. The fans are switched on and off according to a time-proportional minimum (duty cycle) as and when ventilation requirements decrease. These controllers also have a voltage free heating contact. The respective device parameters were set to: temperature = 21 °C; heating = −2 ΔT °C; minimum fan speed = 10%; maximum fan speed = 80%; bandwidth ventilation = 4 °C; ventilation mode: automatic to given parameters; and were set as identical in both turkey houses. Additionally, ventilation system equipped with HEPA filters and enabling maintaining a pressure cascade in corridors, bird boxes and sanitary locks, which excludes the possibility of cross-contamination of experimental rooms. The volume flow rate was controlled by device MFC-VAD, in the range 175–220 m^3 h^{-1} (calculation based on ventilator volume flow and minimum/maximum fan speed), what corresponds to around 3 times the amount of air exchange per 1 hour in the room. Both rooms had the same ventilation rate.

The birds were kept on shredded dry wheat straw litter (depth—20 cm). The initial straw layer was spread on a clean floor. Dry wheat straw humidity was estimated at about 9%. Weekly, each turkey house was supplied with the similar amount of fresh wheat straw.

2.2. Deodoric® Biopreparation

Every week, the deodorizing biopreparation was applied manually on the top of the litter in the experimental poultry house in the amount of 3.6 kg based on the following calculations: 5 turkeys per m^2 produce around 1.3 kg of excreta daily (adult individuals) and 9.0 kg of excreta weekly per m^2. Five grams of dried material per 500 g of excreta (effective dose determined in a laboratory experiment), i.e., 90 g of dried material per week/m^2, 90 g of sorbent + 90 g of dried material = 180 g of Deodoric®/week/m^2; 20 m^2 × 180 g = 3600 g = 3.6 kg. The biopreparation was composed of two parts. The first part was dried material containing a mixture of the following microorganisms: *Lactobacillus plantarum* (ŁOCK 0996), *Leuconostoc mesenteroides* (ŁOCK 0964) *Bacillus megaterium* (ŁOCK 0963) *Bacillus subtilis* (ŁOCK 0962) and *Pseudomonas fluorescens* (ŁOCK 0961), which was spray dried with trehalase (5% *w*/*v*) and maltodextrin (Maltodextrin N 15% *w*/*v*; DE = 7–13, HORTIMEX Sp. z o. o.). The second part was mineral sorbent composed of perlite (15%) and bentonite (85%) (1:1 *v*/*v*).

2.3. Measurements

Once a week, the birds were individually weighed from 24 December 2016 until the end of the experiment (22 March 2017). In groups C and E, ammonia emission was measured in the morning (8.00 a.m.) and afternoon (3.00 p.m.) from the first day of Deodoric® application until the end of the experiment. Selected microclimate parameters were monitored in both poultry houses: relative humidity and temperature were measured with the ST-8820 Multi-Function Environment Meter (CEM, Shenzhen, China). This has semiconductor sensors in the probe for humidity and ambient temperature measurements. Air speed and cooling were determined with Hill's dry kata-thermometer (Technical and Laboratory Glass Manufacturing Plant GOMAR, Warsaw, Poland) based of the protocols described by Hill et al. and Mochida [42,43]; NH_3 concentration in air was measured with the Dräger X-am® 5000 gas detector (Drägerwerk AG and Co. KGaA, Lübeck, Germany), whose operation is based on electrochemical sensors. The devices had been validated by the manufacturers before the experiment.

Measurements were performed 2 weeks before the application of Deodoric® in order to show possible differences that could disturb the result and from the first day of Deodoric® application until the end of the experiment. Measurements were always made at the height of the torso of the animals, in the same place indoors. The methodology of conducted microclimate parameters measurements was based on cited publications [61–65] and industry standard BN86/880-03. The ventilation amplitude varied slightly (175–220 m^3 h^{-1}), and therefore the concentration of ammonia during the experiment also did not change significantly. A 3-fold measurement was carried out twice a day at constant times. This approach meets the conditions of quasi-continuous measurements. NH_3 emission was calculated using the formula E = C V; where C stated for NH_3 concentration, and to the V was volume flow rate (assuming an average volume flow rate of 197.5 m^3 h^{-1}). Litter humidity measurements were carried out at 1, 7 day and 14 day after putting birds in turkey houses, by drying to constant dry mass using a Mac110NH moisture analyzer (Radwag, Radom, Poland). The litter had an initial humidity of approximately 63%. The final humidity of litter in group C was 57% and in group E was 77%.

2.4. Bacteriological Identification Methods

Swabs from the feet, sternum, beak, trachea and air sack were collected for microbiological examinations upon slaughter. Twenty samples from each body area were transported to the bacteriological laboratory at the Department of Microbiology and Clinical Immunology, Faculty of Veterinary Medicine. A detailed description of the methodology may be found in a previous publication [59].

2.5. Quantitative Microbiological Analyses

Samples of litter for microbiological examination were collected from 5 different locations in every experimental pen. The collected litter was pooled to obtain representative laboratory samples. Litter for microbiological analyses was collected 5 times at 20-day intervals (1–2 January, 2—22 January, 3—11 February, 4—3 March and 5—21 March) Turkey manure samples of 10 g each were placed in an Erlenmeyer flask containing 90 cm^3 of 0.85% NaCl and shaken on a shaker for 15 min. A detailed description of the methodology may be found in a previous publication [59].

2.6. Statistical Analysis

The analysis of variance (ANOVA) was preceded by Bartlett's test for homogeneity of variance. Statistically significant differences in ammonia, body weight, microclimate parameters, counts of mesophilic microorganisms and Campylobacter spp. concentration between group C and group E were determined by repeated measures ANOVA. An r-Pearson correlation coefficient was created to evaluate the correlations between microclimate parameters turkeys age, and NH_3 emission. Daily counts of mesophilic microorganisms and *Campylobacter* spp. differences isolated from litter were analyzed using Student's t-test for independent samples. Standard deviation (*SD*), 95% confidence interval (*CI 95%*), mode (*MO*), median (*ME*) and variance (*V*) were also defined. Differences were considered significant at p-value < 0.05. Data were processed statistically in the Statistica 13.1 program with a medical application.

3. Results

Before the application of Deodoric®, differences in the microclimate conditions between group E and group C, including relative humidity (p-value = 0.71), temperature (p-value = 0.55), air speed (p-value = 0.54) and cooling (p-value = 0.53), were not observed.

The final mean slaughter weight of turkeys was 12.41 kg in group C and 13.16 kg in group E. The average percentage of NH_3 reduction over the entire rearing period reached 43.63% (*SD* = 14.57). The measured NH_3 emission and selected microclimate parameters throughout the entire experiment are presented in Table 1.

Table 1. Ammonia concentration, emission and microclimate parameters determined during the experiment in the control group (group C) and the experimental group (group E).

Measured Parameters n = 56	Control Group						Experimental Group					
	$\bar{x}$	*SD*	*CI 95%*	*ME*	*MO*	*V*	$\bar{x}$	*SD*	*CI 95%*	*ME*	*MO*	*V*
NH_3 concentration (ppm)	25.68	11.65	7.96–14.01	24.00	m	135.71	14.43	5.14	3.96–6.83	17.00	18.00	25.14
NH_3 emission (g/h)	3.58	1.70	1.26–2.58	3.36	m	135.71	2.02	0.68	0.50–1.04	2.38	2.52	25.14
Temperature (°C)	20.88	1.17	0.87–1.82	21.20	21.40	1.39	21.28	0.98	0.72–1.52	21.40	m	0.96
Humidity (%)	55.79	6.12	4.52–9.48	55.40	m	37.50	52.80	4.43	3.27–6.86	52.60	m	19.62
Air speed (m/s)	0.052	0.031	0.02–0.05	0.05	0.06	0.001	0.05	0.048	0.02–0.05	0.065	0.05	0.0002
Cooling (W/m^2)	1.85	0.18	0.13–0.28	1.87	1.98	0.032	1.98	0.17	0.12–0.26	1.93	1.84	0.027

Legend: n—number of measurments; *x*—mean; *SD*—Standard deviation; *CI 95*—95% confidence interval; *ME*—median; *MO*—mode; *V*—variance; m—multiple.

Significant differences in NH_3 concentration in air (for the model: $F = 297.58$, p-value < 0.000001; for NH_3 emission: $F = 12.97$, p-value = 0.0013), body weight (for the model: $F = 2073.95$, p-value < 0.000001; for body weight: $F = 154.39$, p-value= 0.042) and relative humidity (for the model: $F = 127.11$, p-value < 0.000001; for relative humidity: $F = 23.55$, p-value = 0.003) were demonstrated between group C and group E. Significant differences in temperature (for the model: $F = 54.77$, p-value = 0.00015; for temperature: $F = 11.21$, p-value = 0.11), air speed (for the model: $F = 14.29$, p-value = 0.0061; for air speed: $F = 8.35$, p-value = 0.098) and cooling (for the model: $F = 2073.95$, p-value = 0.0022; for cooling: $F = 5.63$, p-value = 0.44) were not observed between the experimental and control group. The litter humidity in group C on 1 day was 63% (*SD* = 18), after 7 days 79% (*SD* = 14), and after 14 days 77% (SD = 16). The litter humidity in group E on 1 day was 64% (*SD* = 18), after 7 days 79% (SD = 14), after 14 days (7 days after biopreparation application) was 57% (SD = 16). Values varied due to different spaces in the turkey houses, i.e., the humidity was very high near water troughs, and lower at a distance.

In group C, a relatively strong correlation between NH_3 concentration and temperature and moderate correlations between NH_3 concentration vs. relative humidity, air speed and cooling were noted. No significant differences were noted between NH_3 concentration and age. In group E, significant linear relationships between NH_3 concentration vs. temperature, relative humidity, air, cooling and age were not found. Detailed correlations data are shown in Table 2.

Table 2. Pearson's correlation coefficient between NH_3 concentration in air and microclimate parameters in turkey houses (*r*).

Correlations Between Environmental Parameters	Control Group		Experimental Group	
	p-value	*r* Coefficient	*p*-value	*r* Coefficient
NH_3/temperature	0.0009	0.74	0.19	0.44
NH_3/humidity	0.01	0.59	0.1	0.61
NH_3/air speed	0.015	0.60	0.1	0.53
NH_3/cooling	0.005	0.66	0.09	0.42
NH_3/age	0.11	0.75	0.35	−0.31

r—Pearson's correlation coefficient; n = 56; significance level set at p-value < 0.05.

A statistical analysis of the number of changes in the mesophilic microorganisms isolated from litter did not reveal significant differences between groups on different collection dates (for the model: $F = 46.14$, p-value = 0.09; for mesophilic microorganisms: $F = 3.29$, p-value = 0.045). The number of mesophilic bacteria increased significantly in both groups on the last sampling date (Figure 1).

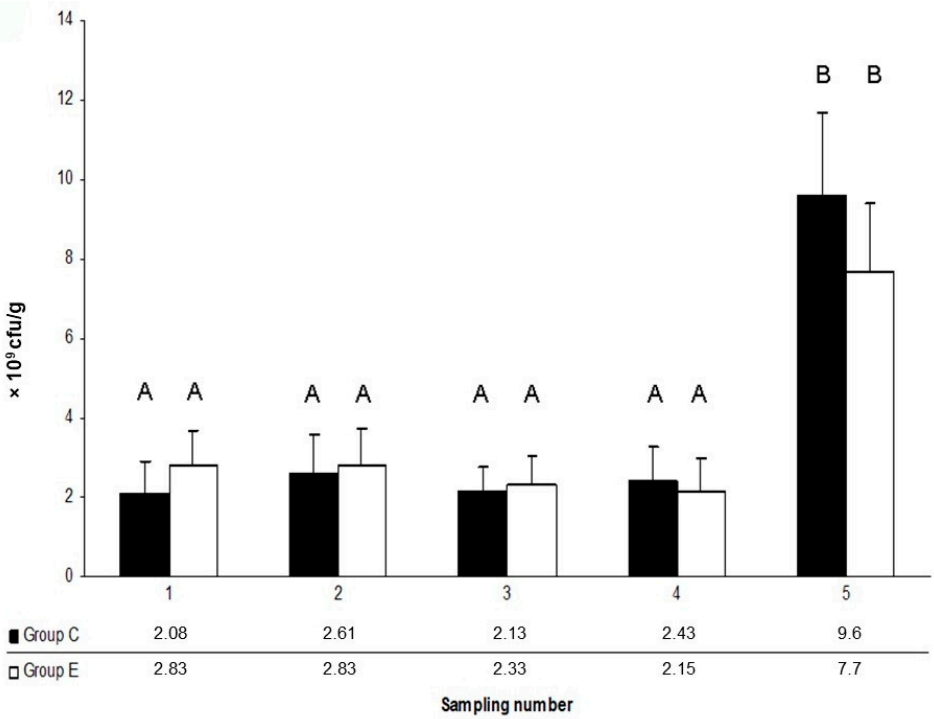

Figure 1. Changes in mesophilic microbial number during the experiment (p-value ≤ 0.05). Bars with different letters represent samples collected at 20-day intervals (number of measurments n = 5; 1—2 January, 2—22 January, 3—11 February, 4—3 March and 5—21 March). Group C—control group, group E—experimental group. Letters A and B above the bars are significantly different at p-value ≤ 0.05.

However, significant changes in *Campylobacter* spp. number were not found for the entire model (for the model: $F = 24.96$, p-value = 0.008; for *Campylobacter* spp.: $F = 0.25$, p-value = 0.64). The results of the k-nearest neighbours' algorithm in microbiological tests indicate that the deviations in mesophilic bacteria number on day 78 and in *Campylobacter* spp. number on day 20 should be regarded as a statistical anomaly (Figure 2).

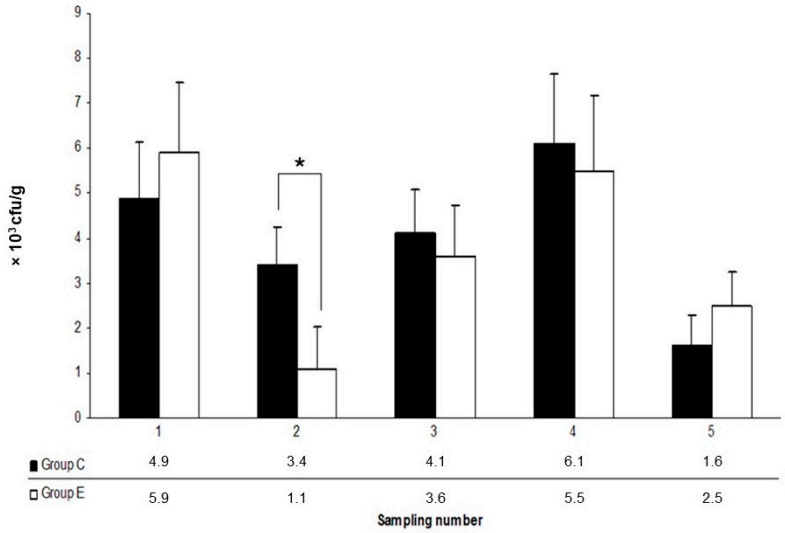

Figure 2. Differences in *Campylobacter* spp. number during the experiment (*—p-value ≤ 0.05). Samples were collected at 20-day intervals (number of measurements n = 5; 1—2 January, 2—22 January, 3—11 February, 4—3 March and 5—21 March).

A qualitative analysis of microbiological swabs collected from selected areas of the body and the respiratory tract did not reveal changes in the composition of commensal microbiota relative to the control group. In group E and group C, *Escherichia coli* and *Proteus vulgaris* were identified in air sacs; *E. coli*, *P. mirabilis* and *Enterococcus* spp. were detected in the trachea; *E. coli* and *P. mirabilis* were identified in beaks; *E. coli*, *P. mirabilis* and *Campylobacter jejuni* were detected on feet; *E. coli* and *P. mirabilis* were detected on the sternum.

4. Discussion

Several commercialized bioactive preparations for decreasing ammonia concentration in air in poultry houses have been developed to date. Deodoric® during animal rearing can be directly applied to the litter, as demonstrated by previous studies performed under laboratory conditions and by the tests conducted in poultry houses in a pilot plant (present study). Previous studies analysing the Deodoric composition were carried out to evaluate the mechanisms of action of the mineral and microbial components of the preparation [66]. Ammonia emission is an important determinant of housing conditions, and it may have a greater impact on animal welfare than stocking density [67]. For this reason, in this study, attempts were made to improve animal welfare by decreasing ammonia concentration in turkey houses. The results of published studies investigating the impact of the sorbent without microorganisms demonstrate that the sorbent mainly promoted manure drying, but also decreased odorant concentration by approximately 30–70%, depending on the compound. The microorganisms present in the biopreparation exerted an antagonistic effect on manure microbiota and decreased odorant emission by 20–40%, depending on the compound. In the present study, the average difference in NH_3 concentration between the C group and the E group was 43.63%. Significant differences between group C and group E in NH_3 concentration in air and relative humidity were noted. The NH_3 concentrations in group C were high compared to literature values [19], which could be due to the period of research (winter), which resulted in an increased concentration of ammonia in the air in turkey houses. Our study suggests that the Deodoric® preparation significantly influenced relative humidity which is mainly responsible for NH_3 concentration in air. Previous studies on laying hens and current studies on turkey broilers, have similar conclusions including reduction in odorants emission, improvement of zootechnical conditions, drying of litter) [59]. This confirms the effectiveness of the preparation in other species and various technological groups. A similar mechanism of action is also observable.

In previous studies, the analysed preparation also improved microclimate parameters in livestock facilities. The adverse effects of ammonia on poultry performance have been demonstrated in the literature [26,39,68]. Deaton et al. [69] reported that exposure to 200 ppm of NH_3 over 17 days significantly inhibited poultry performance. It may suggest that the tested preparation could minimise ammonia's adverse impact on animal health and welfare.

High ammonia concentration may also increase the prevalence of respiratory diseases, including Newcastle disease and mycoplasmosis [24,26,70–72]. In previous studies, Deodoric® inhibited potentially pathogenic microorganisms. Interestingly, the tested biopreparation also modified the microbiota of broiler turkeys. Considerable fluctuations in *Campylobacter* spp. counts were noted throughout the entire experiment in both groups. A significant reduction in *Campylobacter* spp. counts was observed in group E only on day 20. However, no such observations were made in manure samples analysed on different dates. The noted decrease could be attributed to uneven distribution of Deodoric® in the sampling area. This observation is validated by the results of our previous in vitro study [57], which suggests that the dose of the preparation should be increased to inhibit bacterial growth. The obtained microbiological results may indicate that the preparation stabilizes the environment by preventing the rapid development of the studied microorganisms. As a result, the animals' innate and acquired immune responses are not triggered. It is also worth of noticing that the tested biopreparation did not affect the counts of mesophilic bacteria in turkey microbiota. The above was confirmed by microbiological tests of samples from different parts of

the body. Throughout the experiment, total mesophilic counts were comparable in both groups. However, on the last sampling date, the bacterial population increased significantly in both groups relative to previous analytical dates. The above could be attributed to an increase in body weights and, consequently, greater litter compaction in the experimental pens. High stocking could deplete the buffer capacity of the environment and contribute to the rapid proliferation of the examined bacteria. A qualitative analysis of respiratory swabs and samples collected from studied body areas did not reveal changes in local microbiota. The same microorganisms were identified in the remaining areas of the body in both groups. The lack of differences between research groups indicate that the biopreparation did not interfere with microbial stability, thus maintaining the activity of local immunological system (skin-associated lymphoid tissues, SALT; mucosa-associated lymphatic tissue, MALT) at the physiological level. The applied preparation had a stabilizing effect on the microbiological environment of turkeys without affecting the body's microbiota. Deodoric® may also limit the losses associated with unnecessary activation of the immune system.

The feeding of birds may also affect NH_3 concentration in the air. Nahm [73] suggested that reductions in environmental nitrogen and NH_3 pollution caused by poultry farms can be achieved through improved diet formulation based on available nutrients in the ingredients, reducing crude protein levels and adding synthetic amino acids or with enzyme supplementation. Silaban et al. [74], found that reducing dietary crude protein to 15% in laying hens lowered NH3. The mentioned research indicates that the feed used by us during the experiment could not have a significant influence on the ammonia reduction in group C and E.

Previous research demonstrated that in poultry houses, selected microclimate parameters directly affect ammonia concentration [75–79]. Nimmermark and Gustafsson [75] demonstrated that the control of relative humidity and temperature may inhibit NH_3 emissions and its concentration in the air. Elliott and Collins [76] stated that NH_3 emission is mainly influenced by temperature, relative humidity and litter pH. According to Ni [77], NH_3 concentration in the air is strongly corelated with the temperature of the manure or air and the air velocity on the manure surface. Several other studies have also confirmed dependencies between NH_3 volatilization vs. pH level of manure, relative humidity, ventilation rate and the temperature [78,79]. The results of these studies reveal a similar linear relationship between NH_3 concentration vs. relative humidity and temperature in group C and a lack of statistically significant correlations between measured microclimate parameters and the NH_3 concentration in air in group E. These data indicate that the Deodoric® minimised the relationships between ammonia emission and microclimate conditions. Moreover, the lack of any statistically significant correlations between NH_3 concentration in the air and cooling/air speed indicates that room ventilation in group E did not interfere with our results. No linear relationships in group E may be indicated by tested biopreparation characteristics, including a decrease in pH, microbial competition for biological compounds present in litter and manure drying. The tested preparation due to its properties may also have an impact on dry matter content in turkey litter. It should be mentioned that even small variations in the dry matter content of litter can have an impact on reducing ammonia emission, which can be achieved by maintaining the poultry litter dry matter percentage below 60–70% [80,81]. Generally, a higher dry matter content of litter slows down the volatilization of ammonia [80]. In example, Kroodsma et al. [82] showed an inverse relationship between the amount of manure ammonia loss and the dry matter content.

Due to the fact that increased population and industrial activities have caused negative environmental impacts worldwide, the effectiveness of various strategies to reduce greenhouse gases emissions deserves special attention [83]. In previous studies, we performed a detailed analysis of volatile odor compounds in laboratory conditions [58]; however, we have not conducted studies on the Deodoric® influence on greenhouse gas emissions. In studies in animal rooms (laying hens and broilers) after using the Deodoric® preparation, no effect on CO_2 reduction was observed. We cannot say whether Deodoric® will positively or negatively affect the emission of these gases, and therefore this aspect requires further research.

5. Conclusions

The presented results give a tentative indication that this product has an impact on ammonia release in turkey houses. The studied product may decrease the adverse health consequences of ammonia emissions, which may improve the safety of farm personnel. Further research should focus on the effect of Deodoric® doses on the immune and respiratory systems of birds. Toxicological tests should also be conducted to determine the safety of high doses of the preparation. However, the results of this study indicate that Deodoric® can be safely used in the described doses in turkey farms to increase profits and to improve poultry health, animal welfare and growth performance.

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