

## Article

# Dust and Bacterial Air Contamination in a Broiler House in Summer and Winter

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**Abstract:** This study aimed to investigate dust and bacterial air contamination in a broiler house during different seasons. The study was carried out in commercial housing conditions during five weeks of the rearing cycle in summer and winter. The total dust concentration ranged from 1.90 to 4.50 mg/m<sup>3</sup> in summer and from 2.80 to 5.10 mg/m<sup>3</sup> in winter. The total bacterial count ranged from 2.85 × 10<sup>4</sup> to 1.03 × 10<sup>5</sup> CFU/m<sup>3</sup> in summer and from 2.12 × 10<sup>4</sup> to 2.28 × 10<sup>5</sup> CFU/m<sup>3</sup> in winter. The study results showed the dust concentration to be increased in winter as compared to summer, yielding a significant correlation ( $r = 0.602$ ,  $p < 0.05$ ) with a significantly higher airborne bacterial count in winter ( $p < 0.001$ ). Furthermore, dust concentration showed significant correlations ( $p < 0.05$ ) with air temperature ( $r = -0.418$ ), relative humidity ( $r = 0.673$ ), and broiler activity ( $r = 0.709$ ), while bacterial count yielded significant correlations ( $p < 0.05$ ) with air temperature ( $r = -0.756$ ), relative humidity ( $r = 0.831$ ), and airflow rate ( $r = 0.511$ ). The results obtained in the study can prove useful in the field. Seasonal variability in dust and bacterial air contamination should be considered in the development of guidelines or standards of air quality in broiler housing and evaluation of the effectiveness of remedial strategies.

**Keywords:** poultry; air quality; seasonal variability; airborne dust; airborne bacteria



**Citation:** Ravić, I.; Ostović, M.; Ekert Kabalin, A.; Kovačić, M.; Matković, K.; Gottstein, Ž.; Horvatek Tomić, D. Dust and Bacterial Air Contamination in a Broiler House in Summer and Winter. *Agriculture* **2024**, *14*, 778. <https://doi.org/10.3390/agriculture14050778>

Academic Editors: Anna Koziorowska and Bartosz Piechowicz

Received: 22 April 2024

Revised: 14 May 2024

Accepted: 16 May 2024

Published: 18 May 2024



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## 1. Introduction

The air in intensive livestock production houses is always contaminated with some level of pollutants such as dust and microorganisms, with high concentrations of these pollutants posing a threat to animal and human health and welfare, production efficiency, and the external environment [1–3].

Dust in livestock houses is up to 90% of organic origin [4]. The sources of dust are animals and their excrements, litter, feed, and microbial cells or fragments [5,6]. According to aerodynamic particle diameter, dust is classified as the total or inhalable dust (<100 μm), thoracic dust (<10 μm), and respirable dust (<4 or 5 μm) [7,8]. The smaller the particles, the deeper they penetrate in the respiratory system and are health riskier [9]. In livestock houses, the respirable fraction generally accounts for 5% to 10% of inhalable particles [8]. The impact of dust particles on health can be described as mechanical, chemical, infectious, immunosuppressive, allergic, and toxic [10].

Saprophytes are the most common microorganisms detected in livestock house air; however, pathogenic microorganisms may also occur [11,12]. About 90% of the microorganisms identified in livestock house air were gram-positive bacteria [13,14], with staphylococci, streptococci, and enterococci being the most common [11]. The high proportion

of gram-positive bacteria in livestock house air can be attributed to their resistance to environmental conditions owing to their tough cell wall [14]. Unlike these, gram-negative bacteria account for a small proportion of airborne bacteria ranging from 0.02% to 5.2% [15].

Comparative research on air quality in livestock houses has indicated that dust concentration is highest in the air of poultry houses. So, Radon et al. [16] report on the median total airborne dust concentration in poultry houses of  $7.01 \text{ mg/m}^3$ , while Takai et al. [17] report on the mean concentration of inhalable and respirable dust of  $3.60 \text{ mg/m}^3$  and  $0.45 \text{ mg/m}^3$ , respectively. In the study by Kim and Ko [18], the mean concentration of total and respirable airborne dust in poultry houses was  $4.39 \text{ mg/m}^3$  and  $2.33 \text{ mg/m}^3$ , respectively. Skóra et al. [12] report on the mean concentration of total airborne dust in poultry houses of  $1.44 \text{ mg/m}^3$  and Bakutis et al. [19] on  $11.4 \text{ mg/m}^3$ . Ellen et al. [20] found the airborne dust concentration in poultry houses to range from  $0.02 \text{ mg/m}^3$  to  $81.33 \text{ mg/m}^3$  of inhalable dust and from  $0.01 \text{ mg/m}^3$  to  $6.5 \text{ mg/m}^3$  of respirable dust.

Poultry houses are livestock houses with the highest level of airborne microorganisms as well [14,16,21]. Duan et al. [22] found that the total bacterial count in the air of poultry houses ranged from  $3.80 \times 10^5$  to  $2.57 \times 10^6 \text{ CFU/m}^3$ , whereas according to Bródka et al. [23], it ranged from  $4.74 \times 10^4$  to  $1.89 \times 10^8 \text{ CFU/m}^3$ . In the study performed by Bakutis et al. [19], the total bacterial count in the air of poultry houses did not exceed  $10^5 \text{ CFU/m}^3$ . These findings are consistent with the report by Matković et al. [24] that the bacterial count in the air of poultry houses usually ranges from  $10^4$  to  $10^8 \text{ CFU/m}^3$ .

Besides animal species, animal category and housing systems have also been found to affect the concentration of air contaminants in livestock houses. So, the concentrations of airborne dust and bacteria were higher in broiler houses as compared with laying hen houses [14,18,25], as well as in broiler houses with deep litter systems as compared with other floor systems [26,27]. Other factors that can contribute to poultry house air contamination with dust and bacteria include poultry age, weight and activity, stocking density, lighting programs, type of bedding and feeding, type of ventilation and ventilation rate, farm management, microclimate conditions, and seasons, as well as the method and time of sampling [6,11,20,28,29].

Regarding season and microclimate conditions, previous studies have demonstrated no clear consensus on their impact on airborne dust and bacterial concentrations in broiler houses. For instance, Saleh et al. [30] and Wójcik et al. [31] report that the total bacterial count in broiler house air is higher in winter than in summer, whereas opposite results were recorded by Lawniczek-Walczyk et al. [32] and Chen et al. [33]. In the study by Lawniczek-Walczyk et al. [32], bacterial count in broiler house air showed a significant positive correlation with relative humidity but no significant correlation with air temperature, whereas Banhazi et al. [34] recorded a significant positive correlation between bacterial count and air temperature but no significant correlation with relative humidity. Kim and Ko [18] and Saleh et al. [30] determined the lower total dust concentration in broiler houses in summer than in winter attributing it to a higher ventilation rate in summer. In contrast, Vučemilo et al. [35] recorded no significant correlation between airflow rate and total dust concentration in a broiler house.

This study contributes to the assessment of seasonal airborne dust and bacterial contamination in intensive broiler production housing.

## 2. Materials and Methods

### 2.1. Animals, Housing, and Farm Management

The study was carried out in a commercial broiler house during five weeks of rearing cycle in summer (July–August 2016) and winter (December–January 2016/2017). The farm is located in northwest Croatia with a moderate continental climate. The poultry house is of a closed type, length 104 m, width 12 m, and height 4 m, with a controlled microclimate. During both rearing cycles, there were 18,000 Ross hybrid broilers in the house. From the beginning of the rearing period, broilers were evenly distributed all over the house and kept on a litter made of pine sawdust (2-cm lower layer) and chopped wheat straw (8-cm

upper layer), stocking density up to 33 kg/m<sup>2</sup>. The litter was not turned over nor was new litter added during the two rearing cycles. The house is heated by fuel oil heaters, while ventilation is forced via negative pressure. Longitudinal walls have 49 air inlets, a width of 0.54 m, and a height of 0.26 m, with covers made of solid plastic, which are automatically opened and closed. Five exhaust fans are located along the mid-ceiling. Four sidewall fans are only operating at high temperatures but were out of function during the study period. The ventilation system operated at a minimum rate in winter, reaching a maximum rate in summer. Ventilation rates were set according to the manufacturer's instructions (Big Dutchman International GmbH, Vechta, Germany). Lighting is artificial with a total of 32 neon light fixtures of 36-W. The lighting regimen was compliant with recommendations for Ross broilers [36]. Chickens were fed a complete feeding mixture (Biodar, Varaždin, Croatia), with both feed and water offered ad libitum from 390 round pan feeders and 1116 nipple drinkers with cups. The poultry house was cleaned and disinfected with high-pressure cleaners and rested for two weeks after each rearing cycle. Floor was disinfected with caustic soda (PCC Rokita SA, Brzeg Dolny, Poland) and the rest of the house, as well as an equipment, with Ecocid<sup>®</sup> S (Krka d.d., Novo Mesto, Slovenia). After the litter material was spread, the house was fumigated with a Formaster G tablet (Formaster di Emanuela Magnani & C.s.a.s., Piacenza, Italy).

## 2.2. Data Collection

The concentration of airborne dust and bacteria was measured once a week during each rearing cycle. A SAS 100<sup>TM</sup> instrument (PBI International, Milan, Italy) was used to determine bacterial count, with an airflow rate of 10 L/10 s. The sampling head was sterilized prior to each sampling and disinfected between measurements with 70% isopropyl alcohol (VWR International PBI, Milan, Italy). The air samples were collected into Petri dishes with Tryptic Soy Agar (Biolife, Milan, Italy) for enumeration of total aerobic mesophilic bacteria, as reported elsewhere [37–41]. After incubation at 37 °C for 24 h, the grown colonies (CFU/m<sup>3</sup>) were counted using a digital colony counter (J.P. Selecta, Barcelona, Spain), with results corrected as per the table and formula attached to the instrument. Dust in the house air (mg/m<sup>3</sup>) was sampled by use of the SKC pump (SKC Ltd., Blandford Forum, UK), with airflow through the filter (Whatman International Ltd., Maidstone, UK) at a rate of 4 L/min over 4 h. Total dust concentration in the poultry house air was determined by weighing the filter before and after air sampling. The filters were weighed in the laboratory at an air temperature of 22 °C (±2 °C) and relative humidity of 45% (±5%). Measurements were performed in the morning at nine sites (bacteria) and at three sites (dust) in the poultry biozone, with the dust sampling pump being relocated every 80 min. Measurements of air temperature, relative humidity, and airflow rate in the house were also performed at weekly intervals, as reported by Horvatek Tomić et al. [42]. Broiler activity was also assessed by direct observation of broilers employing the method reported by Weeks et al. [43] and Shields et al. [44]. Once a week, the behavior of 120 broilers was observed for one hour at five-minute intervals at different sites within the house and the results were expressed as the rate (%) of active broilers.

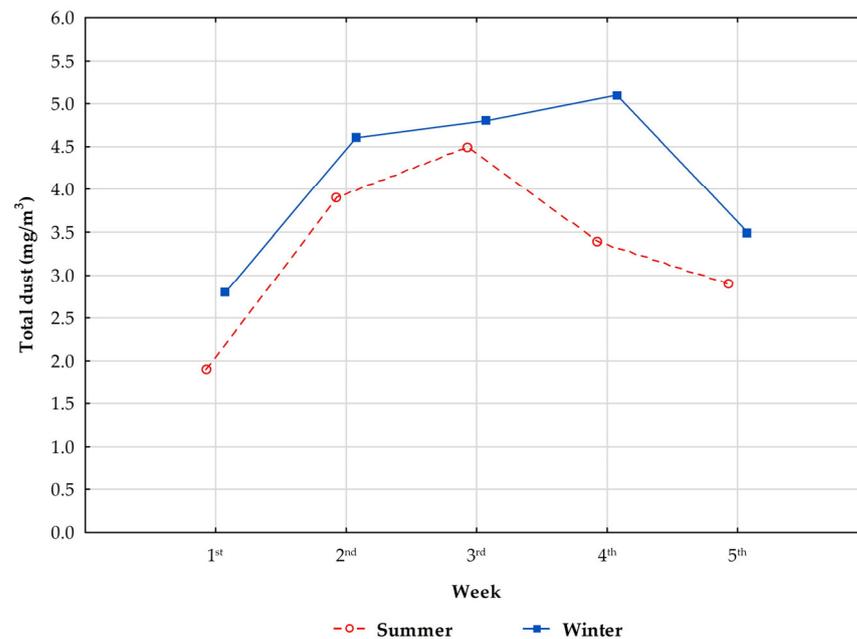
## 2.3. Statistical Analysis

All analyses were conducted by Statistica v. 14.1.0.8 reference software (Cloud Software Group, Inc., Palo Alto, CA, USA, 2023, Data Science Workbench, <http://tibco.com>, accessed on 11 December 2023). Depending on the normality of data distribution as assessed by the Kolmogorov–Smirnov test, the statistical significance of differences in the average values of dust concentration, bacterial count, and rate of active broilers, as well as bacterial count according to particular rearing weeks between the seasons, was calculated by use of a Student's *t*-test and Mann-Whitney U-test. Differences in bacterial count among particular rearing weeks within a season were analyzed by one-way ANOVA and Tukey HSD test for post hoc analysis or Friedman ANOVA. Correlations among investigated parameters were

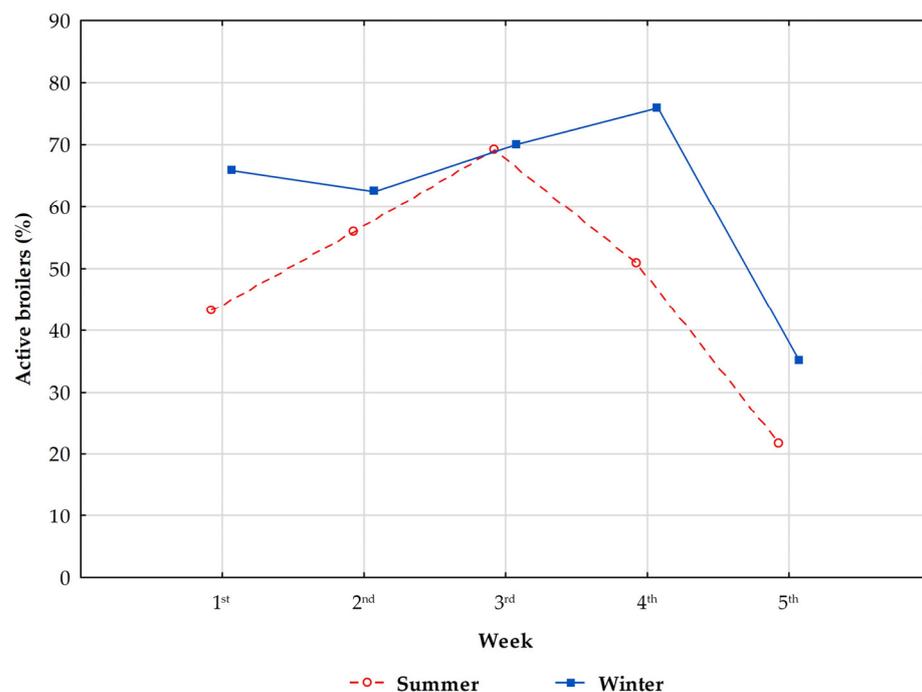
determined by Spearman rank order correlations. In all tests,  $p < 0.05$  was considered as a statistically significant value.

### 3. Results and Discussion

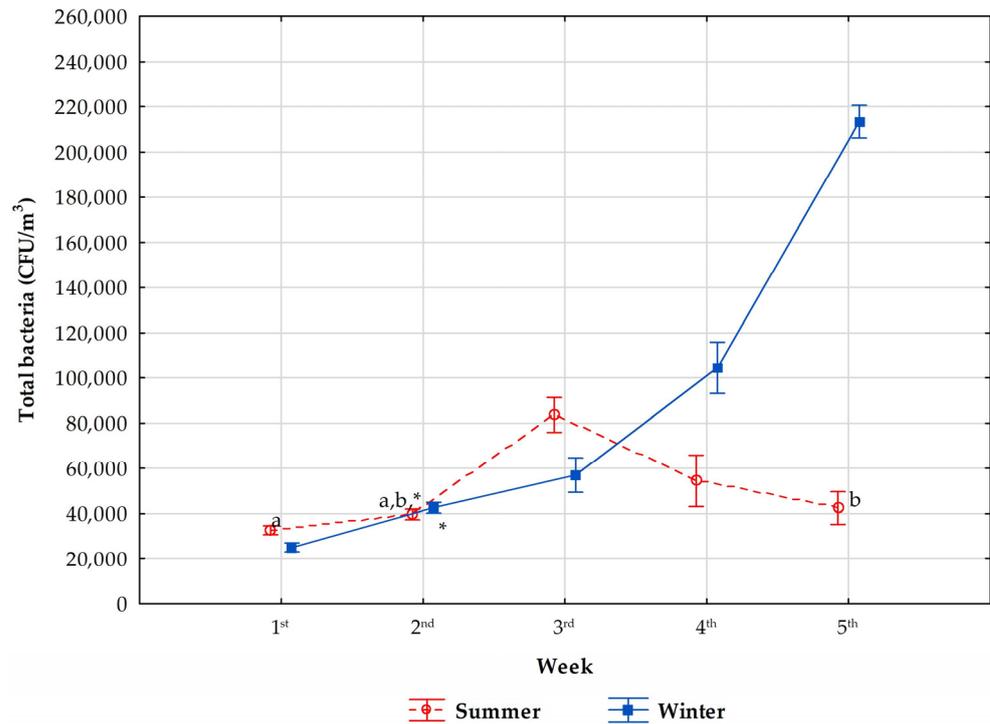
The total airborne dust concentration, rate of active broilers, and total airborne bacterial count according to rearing weeks in summer and winter are illustrated in Figures 1–3, whereas the average values of these parameters throughout the summer and winter rearing cycles are presented in Table 1. Correlations among investigated parameters are shown in Table 2.



**Figure 1.** Total dust concentration in broiler house air per rearing week in summer and winter.



**Figure 2.** The rate of active broilers (of 120 broilers observed weekly) per rearing week in summer and winter.



**Figure 3.** Mean ( $\pm 95\%$  confidence interval) total bacterial count in broiler house air per rearing week in summer and winter. <sup>a,b</sup> Same letters mark weekly values of the same season that were not significantly different, while others differed significantly ( $p < 0.01$ ). \* Asterisk marks weekly values in different seasons that were not significant, others differed significantly ( $p < 0.001$ ).

**Table 1.** Total dust concentration and total bacterial count in broiler house air and rate of active broilers during five weeks of rearing cycle in summer and winter.

| Parameter   | Summer  | Winter  |
|---|---|---|
| Total dust (mg/m <sup>3</sup> )<br>Mean $\pm$ SD (Min–Max)                            | 3.32 $\pm$ 0.99<br>(1.90–4.50)                                    | 4.16 $\pm$ 0.97<br>(2.80–5.10)                                    |
| Total bacteria (CFU/m <sup>3</sup> )<br>Median (Min–Max)                              | 4.13 $\times 10^4$ *<br>(2.85 $\times 10^4$ –1.03 $\times 10^5$ ) | 5.70 $\times 10^4$ *<br>(2.12 $\times 10^4$ –2.28 $\times 10^5$ ) |
| Active broilers (% of 600 broilers<br>observed per season)<br>Mean $\pm$ SD (Min–Max) | 48.17 $\pm$ 17.55<br>(21.67–69.17)                                | 61.83 $\pm$ 15.81<br>(35.00–75.83)                                |

\*  $p < 0.001$ .

**Table 2.** Correlations among microclimate parameters, total dust, and total bacteria in broiler house air and rate of active broilers.

| Parameter                            | Air Temperature (°C) | Relative Humidity (%) | Airflow Rate (m/s) | Total Dust (mg/m <sup>3</sup> ) | Total Bacteria (CFU/m <sup>3</sup> ) | Active Broilers (%) |
|--------------------------------------|----------------------|-----------------------|--------------------|---------------------------------|--------------------------------------|---------------------|
| Air Temperature (°C)                 | 1.000                | −0.443 *              | −0.356 *           | −0.418 *                        | −0.756 *                             | 0.093               |
| Relative Humidity (%)                |                      | 1.000                 | 0.405 *            | 0.673 *                         | 0.831 *                              | 0.397 *             |
| Airflow Rate (m/s)                   |                      |                       | 1.000              | −0.043                          | 0.511 *                              | −0.187              |
| Total Dust (mg/m <sup>3</sup> )      |                      |                       |                    | 1.000                           | 0.602 *                              | 0.709 *             |
| Total Bacteria (CFU/m <sup>3</sup> ) |                      |                       |                    |                                 | 1.000                                | 0.198               |
| Active Broilers (%)                  |                      |                       |                    |                                 |                                      | 1.000               |

\*  $p < 0.05$ .

The total airborne dust concentration was consistent with the results previously reported in broiler houses [18,34,35,45], although some studies recorded much higher dust concentrations in the air of such houses [37]. In both seasons, dust concentration was lowest at the beginning of the rearing cycle, showing a rise thereafter and eventually decreasing at the end of the rearing cycle (Figure 1). These findings could be ascribed to higher broiler weight and lower activity at the end of the rearing cycle, as also discussed by Vučemilo et al. [35,45]; however, these authors did not quantify broiler activity. Our study indicated a comparable pattern of dust concentration and broiler activity during the rearing cycle in both seasons (Figures 1 and 2), where the mean rate of active broilers did not differ significantly ( $p > 0.05$ ) between the seasons (Table 1). There was a significant positive correlation ( $r = 0.709$ ;  $p < 0.05$ ) between the rate of active broilers and dust concentration (Table 2). Calvet et al. [7] report on a strong positive correlation between broiler activity and air concentration of dust particles less than 10  $\mu\text{m}$  in diameter.

Although the mean dust concentration was higher in winter than in summer (4.16  $\text{mg}/\text{m}^3$  and 3.32  $\text{mg}/\text{m}^3$ , respectively), there was no significant between-season difference ( $p > 0.05$ ) (Table 1). The same was indicated by the results reported by Lawniczek-Walczyk et al. [32]. Kim and Ko [18] recorded a significantly higher mean total dust concentration in broiler house air in winter than in summer. In the study by Saleh et al. [30], the total dust concentration in broiler houses was also higher in winter as compared to summer, reaching even double values at the end of the rearing cycle in winter. In the studies by Kim and Ko [18] and Saleh et al. [30], the lower dust concentration measured in summer was ascribed to a higher house ventilation rate in summer, as also discussed by Takai et al. [17]. In our study, there was no significant correlation between airflow rate and dust concentration ( $r = -0.043$ ;  $p > 0.05$ ) (Table 2), as also reported by Vučemilo et al. [35]. Banhazi et al. [34] reported a significant negative correlation between airflow rate and respirable dust concentration in the air of broiler houses but failed to record a significant correlation with inhalable particle concentration, indicating that these particles may not be easily eliminated by increasing airflow rate.

High temperature triggers a cascade of events that can influence airborne dust concentration, such as increased ventilation rate [11]. Although in our study there was no significant correlation between airflow rate and dust concentration, a significant negative correlation was recorded for air temperature with airflow rate ( $r = -0.356$ ;  $p < 0.05$ ) and dust concentration ( $r = -0.418$ ;  $p < 0.05$ ) (Table 2). In contrast, other authors report a significant positive correlation between air temperature and total dust concentration in the air of broiler houses [34,35], whereas no significant correlation was found between air temperature and smaller dust particles [32,34]. Zhao et al. [11] report on the correlation of airborne dust concentration and air temperature to change from positive to negative at extremely high temperatures, possibly because animal activity is reduced at high temperatures, thus decreasing the airborne dust concentration. In our study, there was no significant correlation between air temperature and the rate of active broilers ( $r = 0.093$ ;  $p > 0.05$ ) (Table 2).

Vučemilo et al. [35] report that high air humidity can precipitate sedimentation of dust particles, whereas low air humidity results in high airborne dust concentration. These authors, however, recorded a significant positive correlation between relative humidity and total dust concentration in the air of a broiler house, as also confirmed by our study ( $r = 0.673$ ;  $p < 0.05$ ) (Table 2). This finding may be related to the significant positive correlation among the rate of active broilers, dust concentration, and relative humidity, where higher broiler activity yielded higher dust concentration, as well as higher air humidity ( $r = 0.397$ ;  $p < 0.05$ ) (Table 2). Banhazi et al. [34] recorded a significant negative correlation between relative humidity and respirable dust concentration in broiler houses, however, without any effect of air humidity on inhalable dust concentration. In the study by Lawniczek-Walczyk et al. [32], relative humidity in broiler houses had no impact on airborne dust particles less than 10  $\mu\text{m}$  in diameter.

The values of air temperature, relative humidity, and airflow rate in the broiler house were consistent with those reported elsewhere. The mean temperature and airflow rate did not differ significantly between the seasons, whereas the mean relative humidity was significantly higher in winter as compared with summer [42]. There was a significant negative correlation between air temperature and relative humidity ( $r = -0.443$ ;  $p < 0.05$ ), while airflow rate showed a significant negative correlation with air temperature and a significant positive correlation with relative humidity ( $r = 0.405$ ;  $p < 0.05$ ) (Table 2). On the one hand, relative air humidity was expected to decrease with increased airflow rate, whereas, on the other hand, it was quite understandable that airflow rate increased due to broiler growth and increased humidity in the house.

Dust can act as a carrier of microorganisms [4,8,11], as also demonstrated by this study. Dust concentration yielded a significant positive correlation with bacterial count in the air ( $r = 0.602$ ;  $p < 0.05$ ) (Table 2). The total bacterial count in the broiler house air was consistent with the results reported by Vučemilo et al. [45] and Matković et al. [46], whereas higher values have been reported by Saleh et al. [30], Wójcik et al. [31], Lawniczek-Walczyk et al. [32], Oppliger et al. [37], and Haas et al. [47]. In our study, the median bacterial count was significantly higher ( $p < 0.001$ ) in winter ( $5.70 \times 10^4$  CFU/m<sup>3</sup>) than in summer ( $4.13 \times 10^4$  CFU/m<sup>3</sup>) (Table 1), which is consistent with the results reported by Saleh et al. [30] and Wójcik et al. [31] but opposite to the results reported by Lawniczek-Walczyk et al. [32] and Chen et al. [33]. According to Chen et al. [33], the higher bacterial count in broiler house air in summer should be attributed to high temperature and humidity that favor the growth of microorganisms.

The bacterial burden of air in broiler houses has been shown to rise with broiler age and stage of the production cycle [32,37,45,47,48]; in addition to animal growth, it is caused by increased concentration of feces, litter, and feed residues [6]. Saleh et al. [30] and Wójcik et al. [31] report on comparable findings. These authors attributed lower bacterial count determined in the broiler house air in the last rearing week both in summer and winter to reduced broiler activity (although it was not quantified) and higher indoor airflow rate, respectively. On the other hand, Banhazi et al. [34] did not record any effect of the airflow rate on the bacterial count in broiler house air. In this study, bacterial count in the broiler house air in summer increased by the middle of the rearing cycle to decrease thereafter, whereas, in winter, it showed a continuous rise throughout the rearing cycle. A comparison of the seasonal patterns showed that bacterial count was significantly higher ( $p < 0.001$ ) at the beginning and in the middle of the rearing cycle in summer, whereas, in winter, it was significantly higher ( $p < 0.001$ ) at the end of the rearing cycle due to a continuous increase in their number (Figure 3). There was a significant positive correlation between airflow rate and bacterial count ( $r = 0.511$ ;  $p < 0.05$ ) (Table 2), suggesting that a higher indoor airflow rate is associated with higher bacterial circulation. There was no significant correlation between the rate of active broilers and airborne bacterial count ( $r = 0.198$ ;  $p > 0.05$ ) (Table 2).

Air temperature yielded a significant negative correlation ( $r = -0.756$ ;  $p < 0.05$ ) and relative humidity yielded a significant positive correlation ( $r = 0.831$ ;  $p < 0.05$ ) with airborne bacterial count (Table 2). The latter correlation could explain their significantly higher median count during the rearing cycle in winter as compared to summer. Lawniczek-Walczyk et al. [32] also report on a significant positive correlation between bacterial count in broiler house air and relative humidity but no significant correlation with air temperature, whereas Banhazi et al. [34] describe opposite results.

As the effects of the factors influencing dust and bacterial concentration in livestock housing air are interrelated, they require integrated research [11]. Improved knowledge of these contaminants and factors influencing their concentrations is important for the assessment of their impact, proposing appropriate mitigation strategies and setting the standards of air quality in livestock housing, including poultry houses [21,25,49].

#### 4. Conclusions

Study results indicated that there were seasonal differences in broiler house air contamination with dust and bacteria, suggesting it to be greater in winter. Airborne dust and bacterial concentrations were found to be affected by the same factors including air temperature and relative humidity but also by additional factors such as airflow rate (bacteria) and broiler activity (dust), the latter factor being hardly quantified in previous research. The findings obtained in our study can prove useful in the field. Seasonal variability in concentrations of air contaminants should be taken into account when developing the guidelines or standards of air quality in broiler housing and assessing the effectiveness of remedial strategies.

**Author Contributions:** Conceptualization, I.R., M.O. and D.H.T.; Methodology, I.R., M.O. and D.H.T.; Validation, I.R., M.O. and D.H.T.; Formal Analysis, A.E.K.; Investigation, I.R., M.O., D.H.T. and M.K.; Resources, I.R., M.O., D.H.T. and M.K.; Data Curation, A.E.K.; Writing—Original Draft Preparation, I.R., M.O. and D.H.T.; Writing—Review and Editing, I.R., M.O., A.E.K., M.K., K.M., Ž.G. and D.H.T.; Visualization, A.E.K., K.M. and Ž.G.; Supervision, M.O. and D.H.T.; Project Administration, M.O. and D.H.T.; Funding Acquisition, I.R. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research received no external funding.

**Institutional Review Board Statement:** The study protocol was approved by the Ethics Committee in Veterinary Medicine, Faculty of Veterinary Medicine, University of Zagreb, Zagreb, Croatia (class: 640-01/16-17/43; record no.: 251-61-01/139-16-2; 21 April 2016).

**Data Availability Statement:** The original contributions presented in the study are included in the article, further inquiries can be addressed to the corresponding author.

**Conflicts of Interest:** Author Matija Kovačić was employed by the company Kovačić Family Farm. The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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