



Research article

Phytotoxicity as an indicator of stability of broiler production residues



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ABSTRACT

Beddings used for successive broiler lots act as substrate to absorb water and feed from the excreta and may be subsequently used as agricultural fertilizers. This study evaluated the physicochemical characteristics and the phytotoxicity of beddings used to raise five consecutive broiler lots in five aviaries. Samples were collected for beddings not used yet and for beddings used at each of the five broiler lots. Lettuce and cucumber seeds were considered as phytotoxicity bioindicators. As beddings were used for greater number of lots, N, Ca, K, Mg and P contents generally increased, but the C content decreased, the pH alkalized and humidity was reduced ($P < 0.05$). The germination index for lettuce and cucumber seeds was reduced with increased bedding ($P < 0.05$). Beddings used for at least five broiler lots presented decreased C:N ratio and would not be recommended for agricultural use due to its toxicity for both tested seeds.

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1. Introduction

Brazil reached a relevant position in the world broiler market, ranked as the third greatest industry and the leading exporter (Mathews and Haley, 2015). As also occurs around the world, large poultry inventories are housed in relatively small areas. Hence, if not efficiently managed, dejects of poultry farms may incur in environmental risk due to potential pollution (Kelleher et al., 2002; Sharpley et al., 2007; Bolan et al., 2010). In poultry farms, beddings are used to provide thermal comfort, avoiding direct contact of the animals with the floor (Miles et al., 2011). Beddings act as substrate for absorption of water and feed, incorporating the animals' excreta (Corrêa et al., 2012), also functioning as digester, since their physicochemical characteristics are altered as the excreta is continuously absorbed (Gao et al., 2010). However, the accumulation of organic material inside beddings may favor proliferation of microorganisms that may be pathogenic (Omeira et al., 2006; Huang et al., 2011).

With the expansion of poultry production, cellulose-rich

bedding material tends to become scarcely available. Thus, reutilizing beddings in consecutive broiler lots is a common strategy employed to reduce production costs (Dai-Prá et al., 2009). In Brazil, beddings are commonly used for 5–8 consecutive lots. Beddings used for poultry production are frequently used in agriculture as organic fertilizers (Sistani et al., 2003; Ogunwande and Osunade, 2011), to improve chemical, physical and biological characteristics of soils, benefitting nutrients recycling (Liu et al., 2009; Atencio et al., 2010; Keener et al., 2014). Nonetheless, recurring use of organic residues over time may result in negative consequences for soils destined to agriculture, if the water table is contaminated by nitrites (Jiang et al., 2011). That may occur when unstabilized beddings are placed in the soil, inhibiting the development of vegetable cultures (Fuente et al., 2011).

Many physicochemical parameters, such as temperature, pH, nutrient content and C:N ratio can be used as indicators of stabilization of organic composts (Corrêa et al., 2009; Ogunwande and Osunade, 2011). However, as those parameters do not provide enough information to ensure that residues of poultry production are environmentally safe for agricultural use (Jurado et al., 2014), evaluations of phytotoxicity are suggested as supplementary methods to determine compost stabilization (Zuconi et al., 1981; Tiquia and Tam, 1998; Ozores-Hampton et al., 1999; Delgado et al., 2010, 2012; Galende et al., 2014). Phytotoxicity trials consider

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the germination and growth of roots of distinct vegetable species as indicators of biological responses to stabilization of biosolids. The objectives of this study were to evaluate the physicochemical parameters of beddings used for consecutive lots of broilers and to determine the influence of the agricultural use of such beddings on the growth of vegetables used as bioindicators.

2. Material and methods

2.1. Experimental design

This study was conducted in five aviaries. Throughout this study, the same methods were applied in all aviaries. Each aviary was 12 m wide × 100 m long and housed 18,000 broilers per lot, from their first day of life until 42 days of age. At the beginning of the study, each aviary received a 0.15 m bedding composed of 50% rice husk (*Oryza sativa*) and 50% shavings (*Pinus elliottii*), which are low-cost and highly available materials commonly used in aviaries in Brazil.

Prior to the first animal contact (day 0), bedding samples were collected from twenty points of each aviary. Those samples were homogenized to compose a 1.0 kg sample (one per aviary), as recommended by Cook et al. (2011). Using the same method, additional samples were collected from each aviary at the end of five consecutive broiler lots, which added to the sample collected at day 0 resulted in a total of 30 broiler lots. The physicochemical parameters evaluated subsequently were obtained by the average of the samples collected in the five aviaries.

2.2. Physicochemical analyses

Bedding samples were analyzed as described by Page et al. (1982). The dry matter content was determined after incubation of samples at 105 °C for 24 h. The C content was determined by the Walkley–Black method, the N content was estimated by the Kjeldahl method and the C:N ratio was determined by calculation. The P content was determined by atomic absorption spectrophotometry and the K content was determined by flame photometry. The pH was measured by a digital pH meter.

2.3. Phytotoxicity test

Seeds of lettuce (*Lactuca sativa*) and cucumber (*Cucumis sativus*) were the bioindicators for the phytotoxicity test, following procedures adapted from those described by Zucconi et al. (1981). Ten g of each bedding sample were diluted in 100 mL distilled water and left to rest for 24 h. Then, the content was filtered in paper filters (Whatman n. 3 BioAmerica®, Miami, FL, USA) using a vacuum pump. Thereafter, 5 mL of the filtered content were seeded in triplicate in Petri dishes with 2 cm diameter, including either 20 lettuce seeds or 10 cucumber seeds. The dishes were covered with parafilm, to favor gas exchange and to reduce humidity losses. Subsequently, dishes with lettuce seeds were incubated at 20 °C for 48 h, whereas dishes with cucumber seeds were incubated at 20 °C for 72 h. For each vegetable, dishes including 5 mL distilled water were used as controls, in triplicate. Thereafter, the number of germinated seeds per plate was counted and their root length was measured using a caliper rule.

A germination index (GI) was calculated through the formula: $GI = (G^*Lm/Lc)$, as described by Zucconi et al. (1981): G was the number of germinated seeds in sample dishes divided by the number of germinated seeds in control dishes; Lm was the average root length in sample dishes (in mm); and Lc was the average root length (in mm) in control dishes. A GI lower than 100% indicates that the germination power and root length for that seed was

impaired by the contact with the tested solution, whereas values greater than 100% indicate that both traits were improved (Zucconi et al., 1981; Tiquia and Tam, 1998).

2.4. Statistical analyses

Due to lack of normality according to the Shapiro–Wilk test, physicochemical parameters were compared across the number of broiler lots through the Kruskal–Wallis analysis of variance for non-parametric data. After transformation to the logarithmic scale, the GI was compared across the number of lots through analysis of variance, with comparison of means by the Tukey test. To allow interpretation, these data were reported in their original scale. All statistical analyses were conducted using Statistix® (2008).

3. Results and discussion

The C content in unused beddings and in beddings used for only one broiler lot were similar ($P > 0.05$), but greater ($P < 0.05$) than the contents in beddings used for two or more lots (Table 1). That may have been a consequence of C volatilization as CO₂, due to the breathing activity of microorganisms present inside the bedding (Murphy et al., 2014). On the other hand, the N content increased as beddings were used for more broiler lots ($P < 0.05$). Thus, across all lots, the N excreted by the poultry was apparently greater than the N volatilized to the atmosphere (Cook et al., 2011). The C:N ratio was greatest for unused beddings and decreased as beddings were used in consecutive lots ($P < 0.05$). That might suggest that beddings used for only two lots might be adequate organic agricultural fertilizers, since their C:N ratio was inferior to 25 (Jurado et al., 2014). However, as beddings were used across more lots, the greater N content would result in increased content of NH₃, which commonly leads to phytotoxicity (Khan et al., 2014; Bittsánszky et al., 2015), as evidenced by the GI observed for lettuce and cucumber seeds, which worsened as beddings were used across more lots (Table 2). Besides being phytotoxic for the biological model tested in the present study, such beddings may also lead to detrimental effects for the environment due to potential pollutant effect of NH₃. Therefore, beddings used for broiler production would not be suitable for agricultural use without any further processing, as it is commonly done by poultry producers (Keener et al., 2014), even after being used for five lots. Unused beddings were the only ones presenting C:N ratio greater than 160:1 (Table 1) and the greatest GI ($P < 0.05$) for both tested seeds (Table 2). In the specific case of cucumber seeds, the GI equal to 109.6% observed for samples collected at day 0 (Table 2) indicate that this GI is 9.6% points greater compared to that observed in control dishes. The germination of such seeds may have been favored by some specific characteristics of the rice husk/shavings combination, since unused beddings had no contact with nutrients excreted by the poultry.

The contents of Ca, K, P and Mg also increased as bedding use increased (Table 1), although they were generally similar for beddings used on 2–4 broiler lots and achieved their greatest levels in beddings used for five lots ($P < 0.05$). Under normal conditions, all such elements are not volatilized, presenting sedimentary biogeochemical cycles, which may explain their richer content in beddings used for greater number of lots (Owen et al., 2008; Pote et al., 2012). Nevertheless, despite of their apparently greater agronomic value, the poor germination response observed for both seeds in the phytotoxicity tests (Table 2) indicates that such beddings are not recommended as organic fertilizers for agricultural use without additional treatment (Zucconi et al., 1981).

Unused beddings presented acid pH, likely due to the presence of organic acids in the shavings and rice husk. As such substances were degraded by the microbiota formed after the addition of

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