

## IDENTIFICATION OF *LACTOBACILLUS* SPP. FROM BROILER LITTER IN BRAZIL

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### SHORT COMMUNICATION

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#### ABSTRACT

*Lactobacillus* spp. were identified in 100 broiler litter samples collected from different poultry-rearing regions in Brazil. Ten different *Lactobacillus* species were identified: *L. plantarum*, *L. casei* subsp. *pseudoplantarum*, *L. delbrueckii* subsp. *delbrueckii*, *L. reuteri*, *L. murinus*, *L. agilis*, *L. delbrueckii* subsp. *lactis*, *L. salivarius* subsp. *salicinus*, *L. viridescens* and *L. amylophilus*.

**Key words:** *Lactobacillus* spp., broiler litter, chicken.

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*Lactobacillus* species are a bacterial group with morphologic, metabolic and physiologic common characteristics. They are Gram-positive, non-motile, non spore-forming, catalase negative, cytochrome absent, facultative anaerobes, aerotolerant, nutritionally demanding, acid-tolerant, with strictly fermentative metabolism, and lactic acid is the main product of carbohydrates fermentation (6). They are found in nutritionally rich habitats, such as milk, meat, vegetables, grains and in enteric normal microbiota from different animal species (2).

Studies on the intestinal bacteria and pathogen control by intestinal microbiota gained great interest in avian production in the last years (8). Avian enteric microbiota is formed according to the anatomic and physiologic characteristics conditions of the gastrointestinal tract. The microbial population on broiler litter is extremely diversified due to the continuous deposition of birds' excreta, secretions and desquamations. In this microenvironment, strict aerobes or anaerobes, and facultative anaerobes are the outstanding genera (7).

*Lactobacillus* colonizes avian enteric tract since the first day of life. These bacteria can colonize the small intestine of chicks, being ileum the segment with greater *Lactobacillus* diversity (5).

Broiler litter can be defined as vegetal material (wood, shaves, rice husk, coffee straw, sugar cane and other byproducts) that

contains excreta (feces and urine), skin debris, feathers and feed residues (1). A "new" litter is the one that has not yet received production of birds, while the so-called "re-used" litter is that which has already received production of bird flocks once or more times.

The aim of this survey was to identify *Lactobacillus* spp. in broiler litter samples from different poultry-rearing regions in Brazil.

One hundred samples of broiler litters from nine poultry-rearing areas in Brazil were used (Table 1). Each poultry-rearing farm sent 300 to 500 g of broiler litter collected in different points of the broiler houses. The samples were fractionated in 20g packages and kept at 4°C in a sterile recipient.

Isolation of *Lactobacillus* species was carried out as follows: 5g of each sample broiler litter were added to 50 ml of MRS broth (Difco, Detroit, ML, USA) and kept at 37°C for 24 hours. Afterwards, the sample was grown on MRS agar and maintained at 37°C for 24 to 72 hours. Identification of bacteria was done as described by Klander and Weiss (6). The bacteria were lyophilized and kept at -20°C.

Out of the one hundred samples studied, twenty-three (23%) *Lactobacillus* species were isolated: *L. plantarum* (39.1%), *L. casei* subsp. *pseudoplantarum* (17.4%), *L. delbrueckii* subsp. *delbrueckii* (8.7%), *L. reuteri* (8.7%), *L. agilis* (4.3%), *L. murinus*

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**Table 1.** Results of *Lactobacillus* identification according to broiler litter samples in different poultry-rearing regions.

Origin of litters	Number of broiler litter samples n= 100	<i>Lactobacillus</i> identification n=23	Kind of broiler litter	Indicator of litter utilization
São Paulo	20	<i>L. plantarum</i> (2)*, <i>L. murinus</i> (1)	Wood shaves	New
Minas Gerais	10	<i>L. plantarum</i> (1), <i>L. casei</i> subsp. <i>pseudoplantarum</i> (4)	Wood shaves	New
Santa Catarina	16	<i>L. plantarum</i> (2), <i>L. amilophilus</i> (1)	Wood shaves	New
Rio Grande do Sul	22	<i>L. plantarum</i> (3), <i>L. delbrueckii</i> subsp. <i>lactis</i> (1), <i>L. salivarius</i> subsp. <i>salicinus</i> (1), <i>L. delbrueckii</i> subsp. <i>delbrueckii</i> (2), <i>L. reuteri</i> (2)	Wood shaves	Re-used 4x
Paraná	13	<i>L. plantarum</i> (1)	Rice husk	New
Mato Grosso	5	No isolation of <i>Lactobacillus</i>	Wood shaves	New
Mato Grosso do Sul	5	<i>L. agilis</i> (1)	Rice husk	Re-used 5x
Pernambuco	4	<i>L. viridiensis</i> (1)	Sugar cane byproducts	New
Paraíba	5	No isolation of <i>Lactobacillus</i>	Rice husk	New

\* Indicates the number of *Lactobacillus* isolates according to the sampled litters collected from each state.

(4.3%), *L. delbrueckii* subsp. *lactis* (4.3%), *L. salivarius* subsp. *salicinus* (4.3%), *L. viridescens* (4.3%) and *L. amylophilus* (4.3%).

Several factors may interfere on *Lactobacillus* viability in the litter, such as litter pH, bird age, environmental temperature, state of re-use and others (3). In this study, the isolation of *Lactobacillus* species was not affected by the litter original kind, nor by the times the litter was reused (Table I).

The microbial population in broiler litter is highly diversified. Regarding the bacterial population on the enteric tract, *Enterobacteriaceae*, *Lactobacillus*, *Micrococcus*, *Pseudomonas*, *Bacillus*, *Sarcinae*, *Clostridium* and anaerobic bacteria are the main bacterial groups (3).

In this study, *L. agilis*, *L. murinus* and *L. viridiensis* were isolated from broiler litters. These species neither are commonly found in avian environments, nor in commercial birds' enteric tract, as some other *Lactobacillus* species, which are often isolated from avian gut. Therefore, their importance for the enteric microbiota balance, as well as their role in the development of intensively raised birds are being speculated(4).

These results showed the diversity of *Lactobacillus* species isolated in broiler litter samples obtained from different poultry-rearing areas in Brazil. Other studies should be carried out in order to better understand the importance of these species upon broiler intestinal microbiota regulation, reduction of ammonium in the shed, and their interference on the elimination of pathogens, such as *Salmonella* and *Campylobacter* sp.

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#### RESUMO

##### Identificação de *Lactobacillus* spp de cama de frango no Brasil

Foram identificadas cepas de *Lactobacillus* spp. de 100 amostras de camas de frango coletadas de diferentes regiões de produção avícola do Brasil. Foram isoladas dez espécies diferentes de *Lactobacillus*: *L. plantarum*, *L. casei* subsp. *pseudoplantarum*, *L. delbrueckii* subsp. *delbrueckii*, *L. reuteri*, *L. murinus*, *L. agilis*, *L. delbrueckii* subsp. *lactis*, *L. salivarius* subsp. *salicinus*, *L. viridescens*, *L. amylophilus*.

**Palavras-chave:** *Lactobacillus* spp., cama de frango, frango.

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