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Relationships between broiler final weights and microbiota of certain segments of the intestine

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Abstract. Only healthy animals can expect good production results. Gastrointestinal tract (GIT) health is of particular importance in broilers. GIT health has been protected by antibiotics as growth promoters for years. Since their use is forbidden, alternatives are required. One alternative is the use of medium chain fatty acid (MCFA) in broiler nutrition, in order to ensure the health of digestive tract, that is, prevent the activity of pathogenic bacteria, coccidias and viruses. Today, commercial MCFA supplement is used on the market in nutrition of broilers and piglets. Previous experiences of using MCFA in nutrition of broilers suggest that MCFA can be used as a substitute for antibiotics. In the duodenum of experimental broilers (a group of birds fed with added MCFA), the numbers of *Enterococcus* spp. and *E. coli* were significantly correlated with bird weight, but this was not the case in other intestinal segments (ileum or caecum).

1. Introduction

Food safety is not complete without including safety of agricultural production, which must be provided despite the growing challenges associated with it, especially the growing world population (population bomb), but also other factors (global warming, environmental pollution, agricultural land shrinkage, etc.). In livestock production, the increase in meat production has been largely based on the selection of animals. The production of beef, pork and poultry is still on the rise. The largest increase in meat production in recent decades was the increase in livestock (pig) production for pork. However, poultry meat production will continue to grow at a higher annual rate than pork, and particularly more than beef. This increase in poultry production is based on economic reasons (fast turnover of capital, fattening lasts for 42 days), increasing demand for this meat, and it being a meat that has no religious restrictions. In addition, poultry is especially appreciated by consumers due to its low fat content, especially in breast meat (about 1%). Today's broiler hybrids have been selected so that the heaviest part of the carcass is the breast. In order to fully exploit the genetic potential of today's most common broiler hybrids (Cobb, Ross, Hubbard), paragenetic factors, such as care, accommodation, health care, and especially nutrition, are very important.

The aim of this study was to examine the significance of correlations between the final weight of fattening broilers and the microbiota of individual segments of the intestine.

2. Materials and methods

A detailed description of the method of cultivation, keeping, nutrition of broilers, as well as the settings of this experiment was published in Baltić et al. [1]. For the purpose of this work, broilers were fed in all stages of fattening with added MCFA, i.e., a commercial preparation Aromabiotic®, with an average fatty acid composition of: capric acid (2.28 ± 0.05%), caprylic acid (36.85 ± 0.03%), capric acid (37.88 ± 1.60%) and lauric acid (24.50 ± 0.45%). Samples for microbiological analyses were taken from seven broilers' digestive tracts (duodenum, ileum, cecum) from each group (three groups).

The results obtained were compared by statistical analysis using Microsoft Excel 2010 and GraphPad Prism software, version 7.00 for Windows (GraphPad Software, San Diego, California USA, www.graphpad.com). Pearson's correlation was used to determine relationships between broiler final weights and microbiota of segments of the intestine (duodenum, ileum, caecum). Differences were considered significant if P<0.05.

3. Results and discussion

The results obtained for correlations between broiler final weights (g) and determined numbers of isolated bacteria (log CFU/g) from examined intestine segments are present in Fig. 1 (duodenum), Fig. 2 (ileum) and Fig. 3 (caecum).

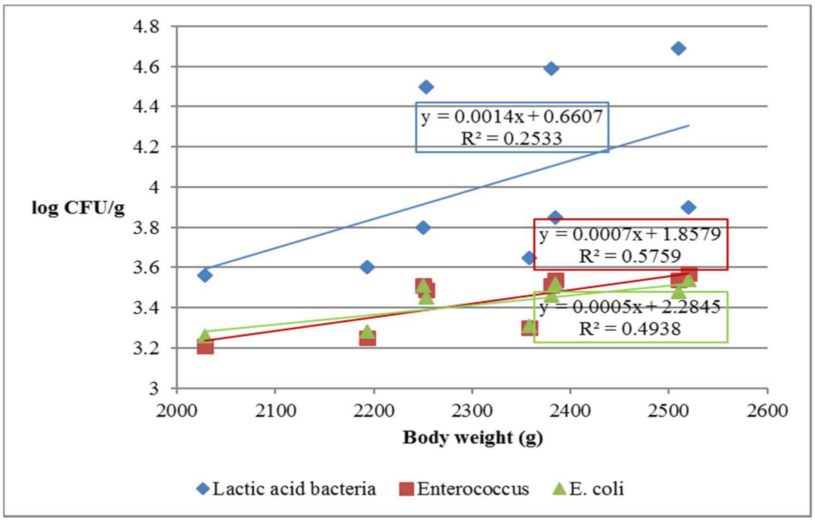

Figure 1. Correlation between broiler final weights (g) and determined number of isolated bacteria (log CFU/g) from duodenum

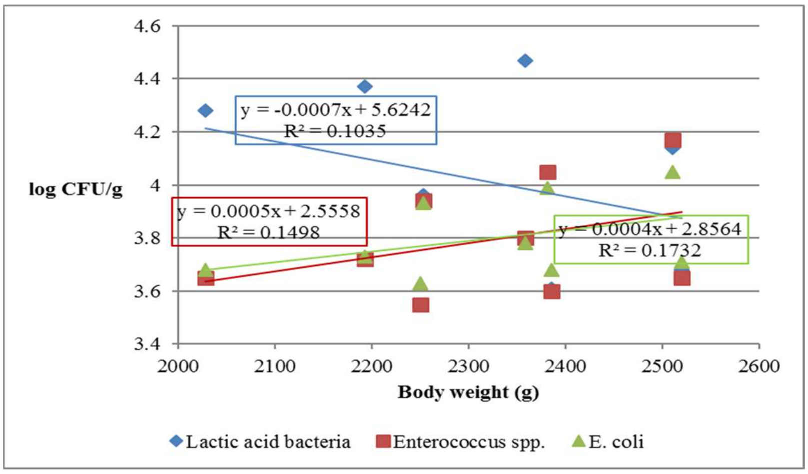

Figure 2. Correlation between broiler final weights (g) and determined number of isolated bacteria (log CFU/g) from ileum

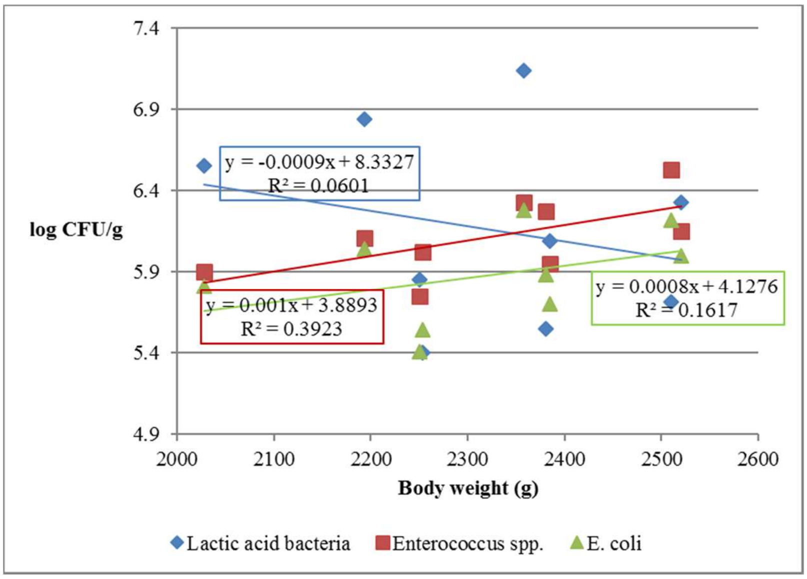

Figure 3. Correlation between broiler final weights (g) and determined number of isolated bacteria (log CFU/g) from caecum

Medium and strongly (r = 0.759, r = 0.703, respectively) significant ($P < 0.05$) relationships were found between broiler final weights and numbers of *Enterococcus spp.*, and *E. coli*, respectively, in duodenum. An insignificant medium correlation (r = 0.503) was determined between broiler final weights and the number of lactic acid bacteria in the duodenum. A negative, medium correlation between broiler final weights and number of lactic acid bacteria, *Enterococcus* spp., and *E. coli* (r = -0.322, r = 0.387, and r = 0.416, respectively), but these correlations were not significant.

The medium relationship (r = 0.626) between broiler final weights and number of *Enterococcus spp*. in caecum was not significant. Low correlation dependence (r = 0.402) between the number of *E. coli* in caecum and final broiler final weights was not significant. There was no correlation between broiler final weights and lactic acid bacteria in caecum (r = -0.245). Table 1 shows correlations between broiler final weights and microbiota of the intestinal segments studied.

Table 1. Correlation between broiler final weights and microbiota of intestinal segments

Intestine segments	Intestine microbiota	Correlation coefficient (r)	Interpretation of correlation dependence*	Significance of the difference
Duodenum	Lactic acid bacteria	0.503	Medium	ns
	Enterococcus spp.	0.760	Strong	p<0.05
	E. coli	0.703	Medium	p<0.05
Ileum	Lactic acid bacteria	- 0.322	Medium	ns
	Enterococcus spp.	0.387	Medium	ns
	E. coli	0.416	Medium	ns
Caecum	Lactic acid bacteria	- 0.245	No relationship	ns
	Enterococcus spp.	0.626	Medium	ns
	E. coli	0.402	Weak	ns

Legend: ns - not significant; *Source: Colton [2]

In feeding of poultry, it is especially important to preserve the health of gastrointestinal tract (GIT), especially of young animals/birds. Without a healthy GIT there is no healthy animal, feed is poorly used and production results are unsatisfactory [3]. The harmful agents (bacteria, parasites, toxins, etc.) in GIT are sourced from the environment in which the animals are located (air, equipment) [4], feed and water. The functions of the GIT are to protect the animal from harmful agents (infectious and non-infectious), transporting food ingredients through it, digesting them, and then absorbing nutrients and energy. The GIT is simultaneously a metabolic and immunological organ [5]. Finally, in GIT there are numerous types of bacteria, and according to some data, there are 640 different types of bacteria and over 20 different hormones [6]. The GIT eliminates harmful and non-immaterial food ingredients. Feed can greatly affect the integrity of intestinal mucosa and microbiota of GIT. Among the pathogenic bacteria in the GIT, the most commonly found are *Campylobacter*, *Salmonella*, *Clostridia*, *E. coli*, *Staphylococcus* [7,8,9,10], and among the protozoa, *Eimeria* spp., [11]. In GIT there are also useful bacteria: *Lactobacillus* spp., *Bifidobacterium* spp., *Enterococcus* spp. For a healthy GIT is important to maintain eubiosis, i.e., a balanced state of the pathogenic and useful microbiota [12,13].

Over 50 years in broiler feeding, antibiotics were used as growth promoters or for prevention of adverse actions of pathogenic bacteria in the GIT. After it became clear that bacterial resistance is an increasing public health problem, the use of antibiotics as growth promoters was prohibited in animal nutrition in the EU in 2006 and then in most other countries [14,15,16]. Since the ban on the use of antibiotics in animal nutrition, alternatives have been sought (probiotics, prebiotics, phytogens, additives, enzymes, short-chain and medium-chain fatty acids and other food supplements). Today, more attention is paid to strategies that would stimulate the strengthening of the immune system in animal nutrition [17,4,18,19,20]. For this purpose, amino acids, vitamins, microelements and linoleic acid are used [21,22,23,24,25,26,20,27,28,29,30,31]. In the Cobb Brochure Guide, it is recommended that 1% linoleic acid be used in all three phases of production [32]. MCFAs improve the production results of broilers in fattening (higher final mass, higher growth, better food conversion), and improve lean meat production (higher carcass weight, higher breast mass and higher weight of drumstick with thigh), as well as meat quality parameters (less fat in meat breasts).

MCFAs affect digestive tract microbiota, including coccidias, viruses and, thus, protect the health of broilers. These fatty acids integrate into the cell membrane or enter the cell in a non-desiccated form, especially at a low pH, thereby adversely affecting metabolism of bacterial cells [33].

The effectiveness of the bactericidal effect of MCFA depends on the amount of added MCFA, so the higher added amount the more pronounced is the effect. The reason for less antibacterial activity in lower GIT sections could be because MCFA is already very efficiently absorbed in the duodenum, and so is less available for absorption in the jejunum, meaning its effect here is less pronounced. However, if 3% MCFA is used, there can be a reduction in the number of bacteria [7]. Reduction of potential pathogens in competition with nutrients is significant because pathogens can damage the cellular epithelium of the GIT, which is manifested by reduced nutrition absorption, especially in the small intestine. On the other hand, some bacteria have a positive effect in the digestive tract because they have the ability to ferment and create, for example, butyrates which promote the function of enterocytes and the immune system [34]. It has been suggested that MCFA also have an inhibitory effect *via Lactobacillus* [12], but there are opposite opinions [35].

The GIT of broilers makes up a complex microbiota system that plays important roles in the digestibility and absorption of food, the development of the immune system and the health of animals, and the exclusion of pathogenic activity [10]. Previous studies have shown that nutrition of broilers affects the microbiota of the GIT, or its diversity.

Due to numerous conditions related to the environment in which broilers are cultivated, the microbiota of the GIT can be very different and often changes. The microbiota of the GIT can contain pathogens in large numbers, and this can badly affect the immune system and production results. The potential changes in the number of bacteria is best illustrated by the fact that in a 35-fold increase in bacterial in numbers in the microbial ileum, lactobacilli accounted for over 90% of the increase [10,5].

Coccidiosis is the most economically most important livestock disease in the world and is the most important health problem of its kind [36]. It is believed that coccidiosis losses are two billion dollars in poultry production annually (death, treatment, prevention). In India, in broiler cultivation, 95.6% of the total losses is associated with coccidiosis-related losses. There are seven species of coccidiosis in chicken: *Eimeria acervulina*, *Eimeria brunetti*, *Eimeria mitis*, *Eimeria necatrix*, *Eimeria pracox* and *Eimeria tenella*, while *Eimeria tenellais* is the most common cause of coccidiosis. Over the last forty years, several different strategies of their control have been developed. The incidence of coccidiosis is significantly influenced by the season and it is most common in the autumn (45,12%), then the summer (30,84%) and the spring (23,81%), and least prevalent in winter (20,29%) [36].

4. Conclusion

In the duodenum of the experimental broilers (a group of birds fed with added MCFA), the numbers of *Enterococcus* spp. and *E. coli* were significantly correlated with bird weight, but numbers of lactic acid bacteria were not. No significant correlations were found between the numbers of bacteria (*Enterococcus* spp., *E. coli* or lactic acid bacteria) determined in other, lower intestinal segments (ileum or caecum), where medium or weak insignificant correlations with bird weight were mostly found.

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