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The effect of a probiotic composed of *Lactobacillus* and yeasts, and of flavomycin on the performance and faecal microflora of broiler chickens

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ABSTRACT

The experiment was carried out on 99 female broilers, allocated to 3 groups of 33 birds, kept in individual cages. A wheat- and soyabean meal-based diet was formulated and divided into 3 batches: C (unsupplemented), A (supplemented with flavomycin), and P (supplemented with a probiotic composed of lactic acid bacteria, yeasts, and yucca extract). Diets were fed from day 1 to 41 of life. Final BW was on average 2.4 kg in all groups, FCR was 1.63 kg feed/kg BWG, neither BWG nor FCR nor mass of the liver, pancreas and gastrointestinal tract were significantly influenced by supplementing the diet with either additive. GI tract digesta pH was lower in groups P and A than in the control group, significantly ($P<0.05$) lower in the crop and ileal content, only. In group P the *Lactobacilli* and *E. coli* counts in excreta were higher ($P<0.05$) in 1- to 3-week old chickens than in group C, while the *Clostridium* count, lower ($P<0.05$) than in group A. In the following weeks the tendency was similar, but insignificant. It may be concluded that the studied probiotic can be considered to be a substitute for antibiotic growth promoters in broiler diets.

KEY WORDS: probiotic, antibiotic growth promoter, faecal microflora, broiler chickens

INTRODUCTION

Various alternative strategies have been proposed for reducing the problems associated with the planned withdrawal of antibiotic growth promoters (AGP), one of them is to populate the intestinal tract with bacteria (Patterson and Burkholder,

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2003) or yeasts (Line et al., 1998), which influence the environment of the gut and favour the establishment of beneficial rather than detrimental species. Modulation of the intestinal population of bacteria by feeding different probiotics is currently under active study, aimed at identifying appropriate bacterial strains and substrates along with the conditions under which they are effective (Apajalahti et al., 2004). The purpose of this study was to compare the effect of a probiotic composed of defined strains of lactic acid bacteria (LAB), yeasts, and a yucca extract, and AGP on the performance of broiler chickens and their faecal microflora.

MATERIAL AND METHODS

The probiotic preparation LABYuc-Probio® was provided by Mifarmex GmbH. It contained in 1g: 4.7×10^7 LAB (*L. casei/paracasei* ŁOCK 0920, *L. brevis* ŁOCK 0944 and *L. plantarum* ŁOCK 0945), 2×10^3 yeasts (*S. cerevisiae* ŁOCK 0140) and 50 mg yucca Schidigera extract. The microorganisms originated from the Pure Cultures Collection of Industrial Microorganisms (ŁOCK 105), Technical University of Łódź; they were resistant to gastric juice and bile and had a high fermenting ability.

Three wheat- and soyabean meal-based diets were formulated, each as starter, grower, and finisher. The starter and grower contained coccidiostat (Cycostat). Each diet was divided into 3 batches. Batch C was unsupplemented, batch A was supplemented with 14 mg/kg of flavomycin, batch P, with 1g/kg (starter and grower) or 0.5 g/kg (finisher) of the probiotic. All diets were cold pelleted.

Ninety-nine 1-day-old female broilers were divided into 3 equal groups. For the first week of life they were kept in groups, then they were placed in individual cages. Chickens were given the experimental diets *ad libitum*: starters (day 1-21), growers (day 22-35), finishers (day 36-41). Feed intake and body weight were measured in weekly intervals. At the end of the experiment, all of the birds were slaughtered, blood samples were taken and centrifuged at 8500 × g for 8 min. The liver, pancreas and crop, stomach, jejunum, ileum and caeca were weighed, digesta were collected, diluted with water (1:1, w/w) and pH was measured using a WTW pH/340 pH meter. In plasma the content of ammonia was measured on a Vitros (Johnson and Johnson) apparatus using NH_3DT slides.

Fresh excreta samples were taken from 5 chickens per group at weekly intervals. Excreta were suspended in buffered 1% peptone water (1:9 w/v), then serial decimal dilutions were made, avoiding aeration. The following bacteria species were identified: *Lactobacilli* on MRS agar medium, using a double-layer technique and anaerobic incubation at 35°C/72 h; *Clostridium* on TCS agar and anaerobic incubation at 37°C/18-24 h; *Enterobacteriaceae* on VRBD agar and aerobic incubation at 35°C/ 24 h; *Enterococcus* on Esculine Bile agar and aerobic incubation at 37°C/72 h; the most probable number (MPN) of *E. coli* on McConkey agar and aerobic incubation

at 35°C/24 h, total number of bacteria on Plate Count agar and aerobic incubation at 35°C/24 h, and total number of yeasts on YGC agar and aerobic incubation at 20°C/120 h. The specific morphology of cells was checked under a microscope (Olympus CX-41). Each determination was done in triplicate. The results are presented as colony forming units (cfu) or MPN per gram of excreta. The results were subjected to one-way analysis of variance using Statgraphics Plus ver. 5.1.

RESULTS AND DISCUSSION

The final body weight averaged 2.4 kg in all groups, FCR was 1.63 kg feed/kg BWG, mortality (3 birds) was not connected with experimental factors. The European Broiler Index equalled about 360 for all groups. BWG, FCR, liver weight (Table 1), pancreas and gastrointestinal tract weight (data not shown) were not significantly influenced by supplementing the diets with either additive. The blood ammonia concentration was within normal physiological values in all groups. There was a tendency towards a lower digesta pH in groups P and A than in the control group, but the differences were significant in crop and ileal contents only (Table 1). The decrease in digesta pH in groups A and P indicated a positive effect of both additives on the activity of microflora producing short-chain organic

Table 1. Performance, liver weight, NH_3 in blood and pH of digesta in 41-day old broilers

Dietary treatment	BWG g	FCR, g feed/g BWG	Liver % LBW	NH_3 µM/L	Digesta pH in			
					crop	jejunum	ileum	caeca
Control	2361	1.62	2.51	173	4.88[a]	5.85	6.80[a]	6.68
Antibiotic	2367	1.63	2.37	161	4.51[b]	5.62	6.20[b]	6.56
Probiotic	2351	1.63	2.51	187	4.66[ab]	5.71	6.67[a]	6.53
SEM	25	0.01	0.07	11	0.10	0.08	0.15	0.06

[ab] means in columns with no common superscripts were significantly different at P<0.05

acids. However, the influence of both applied preparations on the bacterial flora found in excreta was inconclusive, with significant differences between groups being found only in the first 3 weeks of life. In 3-week-old chickens from group P (Table 2) the number of *Lactobacillus* and *E. coli* in excreta was higher (P<0.05) than in the control group, while the number of *Clostridium,* lower (P<0.05) than in

Table 2. Faecal microflora of 3-week-old broilers, log cfu/g

Dietary treatment	Bacteria total	Yeasts total	*E. coli*	*Entero-bacteriaceae*	*Entero-coccus*	*Clostridium*	*Lactobacillus*
Control	9.96	6.05	6.98[a]	7.25	6.13	4.43[ab]	9.42[a]
Antibiotic	9.80	6.24	8.05[b]	8.06	6.86	5.73[a]	9.78[ab]
Probiotic	9.67	6.09	8.48[b]	8.25	5.92	3.67[b]	10.40[b]
SEM	0.22	0.37	0.29	0.42	0.32	0.63	0.31

[ab] means in columns with no common superscripts were significantly different at P<0.05

group A. In the following weeks the tendency was similar, but the differences were insignificant (data not shown). This may have been due to the relatively small number of tested samples (5 per group), however, the horizontal transfer of probiotic organisms to untreated birds cannot be excluded (birds on control and experimental treatments were caged adjacently). Patterson and Burkholder (2003) underlined, that stress status is important in detecting growth performance responses, and studies in which there is no response to AGP should not be considered negative to probiotic treatments. The present study was performed under optimal environmental conditions; in the environment of a broiler farm, colonization of the gastrointestinal tract by LAB bacteria may have greater influence on the health status and performance of birds. It may be concluded the studied probiotic preparation may be considered as a substitute of AGP in broiler diets.

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STRESZCZENIE

Wpływ probiotyku, złożonego z bakterii *Lactobacillus* i drożdży, oraz flawomycyny na wyniki odchowu i mikroflorę odchodów kurcząt brojlerów

Doświadczenie przeprowadzono na 99 kurkach brojlerach, podzielonych na 3 grupy po 33 ptaki, utrzymywane indywidualnie. Dieta pszenno-sojowa została podzielona na 3 części, C (nieuzupełniona), A (z dodatkiem flawomycyny) i P (z dodatkiem probiotyku, składającego się z bakterii kwasu mlekowego, drożdży i ekstraktu z juki). Diety skarmiano od 1 do 41 dnia życia. Końcowa masa ciała wyniosła we wszystkich grupach średnio 2,4 kg, przy zużyciu 1,63 kg paszy/kg BWG. Dodatek do diety probiotyku bądź antybiotyku nie wpłynął na wydajność odchowu, masę wątroby, trzustki i przewodu pokarmowego. W treści przewodu pokarmowego w grupach A i P pH było niższe niż w grupie C, istotnie ($P<0,05$) tylko w treści wola i jelita biodrowego. W grupie P liczba *Lactobacilli* i *E. coli* w odchodach była wyższa ($P<0,05$) u kurcząt w wieku 1 do 3 tygodni niż w grupie C, podczas gdy liczba *Clostridium* niższa ($P<0,05$) niż w grupie A. W następnych tygodniach tendencja była podobna, lecz nieistotna statystycznie. Wyniki wskazują, że badany probiotyk może zastąpić antybiotykowy stymulator wzrostu w mieszankach dla kurcząt brojlerów.