



## Production effectivity of feed mixtures with the probiotic *Enterococcus faecium* to the quality of layers' eggs

E. HORNIAKOVÁ, L. BUŠTA

Slovak Agricultural University, Nitra, Slovakia

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### ABSTRACT

In a trial with hens of laying type Isa Brown we tested the influence of the probiotic preparation IMB 52 by measuring the weight of yolk, the weight of white and the content of minerals in yolk and white.

The experiment was performed in a four group laying test with 3 repeatings, with the total number of 432 hens aged 24 weeks. The chickens were divided to one control group and three trial groups. They were enclosed in three platen cage technology. For the feeding we used a feed mixture of maize, soybean meal, rape seed oil with an addition of minerals and vitamins. The trial groups were fed with the addition of the probiotic preparation IMB 52 (Biomim GmBH Company, Austria) based on *Enterococcus faecium* in different concentrations. The feed for group A contained 2.5.10<sup>11</sup>, the second for group B contained 5.0.10<sup>11</sup> and third for group C contained 1.10<sup>12</sup> cfu.t<sup>-1</sup>. Observed values in trial groups were always compared to control group.

The lowest value of egg weight was registered in the control group (62.77 g). In all trial groups higher values were observed, in the C group 63.03 g, in the B group 63.45 g and in the A group 63.65 g. Differences between control group compared to A and B group were statistically significant ( $P < 0.05$ ). The yolk weight, as well as the egg weight, were lowest in the control group (19.08 g), but highest in group B (16.16 g).

The content of Ca, P, Mg, Fe and Zn in the original dry matter did not confirm a definite tendency of change in the dependence of the trial interference. The content of Ca and P in yolk was higher in the groups with the probiotic preparation, the content of Mg and Zn in yolk was higher in the control group. Statistically significant decreased values were noticed only between the control group and the trial B group concerning the content of Mg and Zn ( $P < 0.05$ ). The content of K in white was significantly decreased in all trial groups compared to the control group ( $P < 0.05$ ).

**Keywords:** laying hens, probiotics (*Enterococcus faecium*), mineral content, egg weight

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### INTRODUCTION

According to the intention of specialists and customers, the usage of minimal chemical preparations and processes of agricultural production are used to implement new progressive and safer biological methods. Phasing out harmful chemotherapeutic and antibiotic preparations in breeding studies is evident, thus moving on to the substitution by probiotics, thus moving on to new biological methods of stimulating growth and utility of poultry. The main common characteristics of probiotics is their biological influence to effect the organism, in which they are stimulating physiological and biological functions and thereby induce an increasing productive potential upon the animals.

Probiotics are specifically selected for host organisms for which according to actual scientific information they have no detrimental effect on the health of the animal and the consumer.

Breeders of poultry since the beginning of mass production have been trying to minimize costs by decreasing feed intake, increasing feed consumption, decreasing mortality, improving health, thus reducing the costs for treatments.

Problems began to be recognized when higher sensitivity of animals to different affections and influence of environment was observed, particularly the breeding of animals to higher production was affected, which is a consequence of the high requirements of animals on the environment. Science in poultry production is thus driven

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**Correspondence:** E-mail: Erika.Horniakova@uniag.sk

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to find new ways, which can help to solve remaining problems. One possible solution is the application of growth stimulators based on probiotics.

Probiotics actually have extensive application abilities. To the host they have very beneficial biological effects, which are not connected to any lateral risks for the biological environment. These facts lead to an enhanced application of probiotics.

Fuller [4] in 1992 defines probiotics as biopreparations, which contain living cells or metabolites of stabilized autochthonous microorganisms, which optimize the occupancy and composition of the enteric microflora of humans and animals with stimulating effects on digestive processes and the immunity of the organism. Schrezenmeier and de Vrese [12] further exemplify the favorable influence of cultured microorganisms on digestion.

Different suggestions for the term probiotic were used in general for any product containing living microorganisms, see Michalík [10]. The term probiotic is used for the identification of products, which contain living microorganisms, favorably influencing the enteric microflora as well as stabilize the health of the host and have positive effects on the gastro-intestinal, the aspiratory and the urogenital system, according to Ferenčík and Ebringer [3]. The most frequently used strain in farm animal nutrition are the bacteria *Enterococcus faecium* M-74, which have a good reproduction ability, are adaptable to environmental changes, and show tolerance to the variation of the pH and the temperature. *Enterococcus faecium* is generally defined as non-pathogenic, non-hemolytic and non-proteolytic, with a favorable multiplicative ability.

Probiotic lactobacilli are the most commonly species used as silage inoculants. Among enterococci that belong to the lactic acid bacteria strain (LAB), bacteria have been found with probiotic character. It is believed that the inoculated populations of LAB genotypes become dominant in silage, thereby increasing the lactic acid concentration and decrease pH values, gas, and protein decomposition, see Inglis et al. [8]. Probiotic LAB are also fed to livestock to improve intestinal microbial balance, including elimination or reduction of undesirable microorganisms, see Zhao et al. [14]. Viable microbial preparations, basically including lactic acid bacteria, have been proposed as a supplement in animal fodder, Cavazzoni and Adami [1].

Research of probiotics influence to the utility and health of poultry is considered by many authors. A multitude of experiments were aimed to the application of probiotics for broiler chicks. In these trials it was expressly confirmed, that positive effects of probiotics on the health of animals were present, see Corrier et. al [2], for feed utilisation see Horniaková [6] and for quality of production see Horniaková and Krivánek [7] and Haščík et. al [5]. Further Yoruk et. al [13], Panda et. al [11] and Kurtoglu et. al [9] - in trials with layers of type ISA –

Brown - confirmed positive effects of probiotics, which contain bacterial cultures (*Lactobacillus*, *Bifidobacterium*, *Streptococcus* and *Enterococcus*) on egg production, better feed consumption and the weight of eggs. These differences however were not statistically significant.

In this paper we wanted to verify the influence of the application of the probiotic preparation IMB 52 (*Enterococcus faecium*) on egg weight, egg yolk and egg white, and further on the weight and content of minerals in egg yolk and egg white.

## MATERIALS AND METHODS

Biomim IMB 52 is a preparation based on microorganisms of the strain *Enterococcus faecium* DSM 3530, with a content of at least  $2.5 \times 10^9$  living germs/g. The product is produced by the BIOMIN Company which is located in Austria. The producer is interested on the extension of the application of the preparation to other breeds of animals. This trial was the groundwork for a revision of efficiency of this preparation in farm layers.

The problematic was studied with a group trial with layers of laying type ISA - Brown, which on the peak of the laying amounts to show a production of 92 -93%, a weight at age 20 weeks of 1400 g, an egg weight in 24. week of age being 57.0g, an egg weight in 48. week of age of 63.9 g and feed intake from 24. week of age amounting to 118 g/day.

The experiment was carried out in the experimental basis ÚKSÚP at the Biological - testing station in Vígľaš as a 4 - group laying test in 3 repetitions. The trial was realized with chicks of the hybrid ISA – Brown type, from a brooder firm in Nitra, with a total a number of chicks totaling 432 animals of age 24 weeks. Growing chicks were divided to one control group and 3 trial groups and were stocked with three platen cage technology. Every group had 108 layers, and in one cage were two layers. Each cage had a dimension of floor surface 0.18 m<sup>2</sup>, the width was 0.4 m, depth 0.45 m and height 0.4 m. Microclimatic conditions and light modes were regulated automatically with strategies established for conditions for these laying hybrids. The trial was finished after a laying cycle of 8 months.

The layers were fed with the feed mixture HYD - 10, which was offered in dry granular status. The content and nutritional values in the feed mixture are shown in table 1, 2 and 3.

The trial groups (A, B, C) were fed with the addition of the probiotic preparation IMB 52 (Biomim GmbH Company, Austria) based on *Enterococcus faecium* with different concentrations. The group A was offered  $2.5 \cdot 10^{11}$ , the second group B was given  $5.0 \cdot 10^{11}$  and third C group  $1 \cdot 10^{12}$  cfu.t-1 that represents 0.1, 0.2 and 0.4 percents of total amount.

In trial the following parameters were observed: egg weight, weight of egg white, egg yolk weight and

content of minerals in egg white and yolk. Parameters determined in the trial groups were compared with parameters determined in the control group. Statistical

data evaluation was provided by the program Microsoft Excel. For the statistical relevance of the difference of mean values a two sided t-test was used.

**Table 1: Feed mixture formulas**

Components of complet feed mixture	Units	HYD -10/K	HYD -10/A	HYD - 10/B	HYD - 10/C
Corn	%	66.5	66.5	66.5	66.5
Soy bean meal	%	21.3	21.3	21.3	21.3
CaCO <sub>3</sub>	%	9.0	9.0	9.0	9.0
Monocalciumphosphate	%	0.8	0.8	0.8	0.8
NaCl	%	0.4	0.4	0.4	0.4
Rape seed oil	%	1.0	1.0	1.0	1.0
VMZ SLOV - vit -	%	1.0	1.0	1.0	1.0
Together	-	100.0	100.0	100.0	100.0
Biomim IMB 52	%	-	0.1	0.2	0.4

The Biomim preparation is produced by the Biomim Comp., Austria

**Table 2: Content of vitamin – mineral premix SLOV - VIT**

Dry matter	g/kg	995
L - lysine	g/kg	220
DL - methionin	g/kg	180
Threonin	g/kg	20
Vitamin A	m.j./kg	1000
Vitamin D3	g/kg	25
Vitamin E (á- tokoferol)	mg/kg	7400
Vitamin B2	mg/kg	660
Vitamin B12	mg/kg	3200
Mn	mg/kg	6000
Cu	mg/kg	2000
Zn	mg/kg	6000
Fe	mg/kg	10000

**Table 3: Chemical analysis of the complete feed mixture from 24- 43 weeks of laying hens (average values)**

Parameters	Units	HYD - 10/K	HYD -10/A	HYD -10/B	HYD - 10/C
Dry matter	g/kg	902.0	899.0	901.0	900.0
Crude protein	g/kg	157.0	156.0	157.0	154.0
Fat	g/kg	44.0	44.0	46.0	45.0
Fibre	g/kg	23.0	25.0	24.0	23.0
Nitrogen free extract	g/kg	554.0	552.0	551.0	552.0
Ash	g/kg	124.0	122.0	123.0	126.0
Ca	g/kg	38.0	38.0	38.0	39.0
P	g/kg	5.0	5.1	5.0	5.0
Na	g/kg	2.4	2.5	2.3	2.2
Sugar as saccharose	g/kg	29.0	29.0	30.0	31.0
Amyloid	g/kg	448.0	444.0	437.0	444.0
Vitamin A	m.j./kg	7647.0	8723.0	7286.0	7793.0
Metabolizable energy	MJ/kg	11.80	11.71	11.69	11.73

## RESULTS AND DISCUSSION

The lowest value of egg weight (table 4) was in the control group (62.77 g). In all trial groups higher values of egg weights were observed in the following sequence: C group (63.03 g), B group (63.45 g) and A group (63.65 g). The determined tendency was statistically significant in the A and B trial group ( $P < 0.05$ ). This tendency is confirmed by Kurtoglu et.al [9] who carried out a trial with probiotics, observed higher values of egg weight, but not statistically significant.

Yolk weights were higher in all trial groups, when compared with the control group. The highest value was in the B trial group (16.96 g), then in A group (16.87 g) followed by the C group (16.83 g). This tendency was statistically significant in the B trial group ( $P < 0.05$ ). The percentage of yolk content in the eggs was in the control group 26.62 %, in the A group 26.50 %, in the B group 26.73 % and in the C trial group 26.70 %. When comparing the weight of the egg white in the control group and in trial groups, a tendency of higher weights

of the egg white in all trial groups was detected. The higher values, when compared with the control group (39.67 g), was in the A group (40.56 g), then in the B group (40.13 g) and finally the C group (39.82 g). The percentage content of egg white in the egg was 63.2 % in the control group, 63.72 % in the A group, 63.25 % in the B group and 63.18 % in the C group. The observed tendency of the weight of egg white increase was statistically significant in the A trial group. This tendency was confirmed by Yoruk et.al [13] and Panda et. al [11], who in trials with layers of laying type ISA Brown have observed increased values of produced egg masses in trial groups, that were fed with feed mixtures augmented with probiotic preparations. Their observed tendencies were not statistically significant.

The mineral content of the trial eggs was very varied (table 5, 6). The most important change of mineral content in egg yolk was the decreased content of Mg in the B group, K in B and C group and Zn in the B group. When comparing the control group and trial groups, the content of most minerals in trial groups was lower than in the control

**Table 4: Egg weight, weight of egg yolk and weight of egg white**

Group	Parameter								
	Egg weight (g)			Yolk weight (g)			White weight (g)		
	$\bar{x}$	sx	v %	$\bar{x}$	sx	v %	$\bar{x}$	sx	v %
K	62,77	1,22	1,94	16,71	0,37	2,23	39,67	1,17	2,96
A	63,65 <sup>b</sup>	0,94	1,48	16,87	0,35	2,07	40,56 <sup>b</sup>	0,83	2,05
B	63,45 <sup>b</sup>	1,04	1,64	16,96 <sup>b</sup>	0,44	2,62	40,13	0,77	1,92
C	63,03	1,11	1,76	16,83	0,36	2,13	39,82	0,77	1,92

x - average; sx - standard deviation; v - variance coefficient; b - differences between values are significant on the level  $\alpha = 0.05$

**Table 5: Content of minerals in egg yolk**

Group	Minerals - average values						
	Ca	P	Mg	K	Fe	Cu	Zn
K	652,26	4911,26	132,67	1491,46	87,57	4,34	56,20
A	651,52	5459,98	121,30	1467,78	79,87	3,99	53,20
B	644,76	4872,91	117,11 <sup>b</sup>	1418,43 <sup>b</sup>	85,24	4,11	51,87 <sup>b</sup>
C	643,78	4759,67	127,17	1421,57 <sup>b</sup>	83,22	4,96	54,78

b - differences between values are significant on the level  $\alpha = 0.05$

**Table 6: Content of minerals in egg white**

Group	Minerals - average values							
	Ca	P	Mg	Na	K	Fe	Cu	Zn
K	11,01	181,63	95,07	3608,65	888,74	6,02	0,36	1,71
A	11,74	179,44	92,00	3765,91	630,06 <sup>b</sup>	6,11	0,31	1,69
B	12,82	183,17	88,69	3484,70	660,07 <sup>b</sup>	5,20	0,30	2,25
C	10,86	209,85	84,83	3470,24	651,26	4,69	0,33	2,09

b - differences between values are significant on the level  $\alpha = 0.05$

group. The content of minerals in egg white was also greatly fluctuating in comparison with the control group when compared with the trial groups. We have observed the same tendency concerning mineral contents in egg yolk. Statistical significant changes were the decreased content of K in the A and B trial groups. In consequence of the absence of literature sources and experiments, that evaluate changes in the mineral content in eggs in trials using probiotic preparations, we were not able to discuss observed values and changes with other authors.

## CONCLUSIONS

From results of the experiment it can be concluded, that layers of laying type ISA – Brown, fed with the probiotic supplement IMB 52 (*Enterococcus faecium* 3530), there was a tendency of higher values of egg weight, egg yolk weight and weight of egg white in the trial groups. The most important changes of the values were in the A and B group, where we determined statistically significant increases of the egg weight ( $P < 0.05$ ). Also the weights of egg yolk and egg white were increased, but statistically significance was only observed in the B group for egg yolk and in the A group for egg white ( $P < 0.05$ ). The content of minerals was varied, but most important are the decreased values of Mg, K and Zn in the B and C trial groups, when evaluating the content of minerals in egg yolk. The same tendency was observed for K in the trial groups A and B, where the content of K was lower than in the control group and this tendency was statistically significant ( $P < 0.05$ ). It is necessary to study this problem in further experimental works, since there is an absence of published sources for changes of mineral content in egg yolk and egg white. From these trial results, the probiotic preparation based on *Enterococcus faecium* has positive effects on egg weight, the weight of egg white and egg yolk weight.

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**Authors' address:** Horniaková Erika, Slovak Agricultural University Nitra, Trieda A. Hlinku 2, 949 76 Nitra, Slovakia.