

APPLICATION OF PROBIOTIC PREPARATION IMB 52 IN LAYING HENS' NUTRITION

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ABSTRACT

The goal of this experiment was additional verification of the probiotic preparation *Enterococcus lactiferm*, the influence of this IMB 52 feed mixture on feed consumption, nutrient consumption, production and weight of eggs in laying hens. The trial included laying hens of the ISA - Brown type, which were divided into a control group (group K) and 3 experimental groups (A, B, C), each containing 108 hens. Laying hens were fed by the feed mixture HYD - 10 upto first 43 weeks. The feed mixtures were different in concentration according to Enterococcus faecium in the probiotic preparation IMB 52. Group A was fed with $2.5.10^{11}$, the second group B with $5.0.10^{11}$ and third group C with 1.10^{12} cfu.t⁻¹. The probiotic preparation IMB 52 was produced by BIOMIN Company in Austria. The highest feed intake per hen per day was in the K group (124.07 g) as compared with experimental groups which consumed 97.90 %, 96,37 % and 97.27 % of feed, respectively. A similar tendency was found in the intake of dry matter, crude protein and energy, normalized for one egg, and the same for per kg egg mass produced. Lowest values were registered in group B. Egg weight was the highest, 63.65 g in group A, which was also statistically significant (P<0.05) in comparison with control group K. Groups B and C registered higher values of egg weight (63.45 and 63.03) compared with the control group. Laying intensity varied from 91.93 % to 92.83 % with the lowest value found in group A and the highest value in group C. These differences between groups (less than 1 %) were not statistically significant though (P>0.05). The production of egg mass was from 8.13 - 8.04 kg per head with the lowest value in group B and the highest in group C.

Key words: laying hens, probiotics (IMB 52 preparation), laying intensity, egg weight

INTRODUCTION

Poultry meat provides a significant portion of human nutrition and belongs to a significant source of biologically full-valued food-stuff. Meat of scraper poultry has high biological value and is eupeptic. Consumption of poultry meat, mainly chicken and turkey as basic representatives of scraper poultry show increasing tendency. Along with poultry meat also eggs are rated as very worthy from nutritional view as nutritious and biologically rich food.

Optimal poultry nutrition and optimal technology for feeding are important factors, which facilitates maximal exploitation of the genetic potential of poultry. By introducing the hybrid combinations of layers into mass production forms the risk of balance violation between microbial biocoenoses of the cage environment and striking power of animal organism can be augmented. In fact, the used hybrid layers of type Shaver Starcross (white shell hybrid), or Isa Brown (brown used hybrid) respond very sensitively to these imbalances. So, preventive measures were also taken to decrease the influence of these negative factors. Among the effective substances to fight the expansion of *Salmonella* infections probiotic preparations were used. Probiotics progressively replace antibiotics, because the organisms rapidly produce resistent bacterial strains. Experimental results with probiotics indicate a decrease in illness of the animals, along with better nutrient utilization and good influence to living environment.

Enterococcus faecium belongs to genus *Streptococcus* and family *Streptococcaceae*. Genus *Streptococcus* comprises of about 40 strains, out of which except pathogenic streptococci the strains belong to enterococci, pertained mostly by Lancefields classificator to group D, which is a part of natural microflora of homoeothermic animals (Voříšek, 1989).

According to Bergeys classification genus *Streptococcus belongs* to the section *Firmicutes*, class *Firmibacteria* and the 17th group (Mundt, 1986).

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Following their uncommon properties "*Strepto-coccus faecium*" was renamed to "*Enterococcus faecium*" (Schleifer and Kilpper-Bälz, 1984). *Enterococcus faecium* populate the digestive tract of humans and most other animals and also of poultry (Mundt, 1986). *Enterococcus faecium* are rounded cells, which occur in pairs or short chains. The main product of Enterococcus faecium metabolism is lactic acid. Also bacteriocines with antisalmonelossic effect are produced (Schiraldi et al., 1982).

Microbial population of the gastrointestinal tract is of prime importance for the accurate process of digestion and keeping good health. Research in probiotics for utility and health status of poultry has been considered by a lot of researchers.

Most works were aimed to use probiotics for broiler chickens. In these experiments the positive effect of probiotics to health status of animals (Corrier et al., 1994), nutrients utilization (Horniaková, 2005), quality of production (Horniaková and Krivánek, 2002; Haščík et al., 2005) were confirmed. Also as in experiments with broiler chickens and in experiments with layers ISA-Brown, it was found Yoruk et al. (2004) that addition of probiotics which contains bacterial cultures (Lactobacillus, Bifidobacterium, Streptococcus a Enterococcus), which were used in feed mixtures in the second period of laying, egg production was positively affected and this linearly increased with increasing doses of probiotics. Simultaneously lower mortality and better feed consumption was registered. Differences between egg weights in these experiments were statistically not significant.

In this paper we wanted to verify the influence of the probiotic preparation IMB 52 (*Enterococcus faecium*) in alternative feed mixtures for hens ISA-Brown laying type to feed and energy consumption, laying intensity and the amount of produced egg mass. The results were evaluated in the first laying period which is 140 days and in the period where hens reach the maximum of their potential abilities.

MATERIAL AND METHODS

Biomin IMB 52 is a preparation based on microorganisms of the strain *Enterococcus faecium* DSM 3530, with a content of at least 2.5×10^9 living germs/g for the calves up to 6 months. The producer is interested in 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 hens of laying type ISA - Brown, which on the peak of the laying amounts show a production of 92 -93%, a weight of 1400 g at age of 20 weeks, an egg weight of 57.0 g at the age of 24. weeks, an egg weight of 63.9 g at the age of in 48. weeks and feed intake amounting to 118 g/day at 24. weeks of age.

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 replications. The trial was realized with chicks of the hybrid ISA - Brown type, from a brooder firm in Nitra, with a total number of 432 chicks of 24 weeks age. Hens were divided to one control group and 3 trial groups and were stocked with three platen cage technology. Each group had 108 layers, and in one cage were two layers. Each cage had a floor surface dimension of 0.18 m², 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 and divided to 2 periods. First period of evaluation rangedbetween 24th to 43rd weeks of laying, that means a total of 140 days and the second period from 44th week to end of trial or 112 days. In this paper are evaluated the results from first laying period.

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

Components of complete feed mixture	HYD -10/1	HYD -10/2	HYD - 10/3	HYD - 10/4	
Corn	66.5	66.5	66.5	66.5	
Soybean meal	21.3	21.3	21.3	21.3	
CaCO ₃	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	
Rapeseed oil	1.0	1.0	1.0	1.0	
VMZ SLOV - vit - BR3	1.0	1.0	1.0	1.0	
Together ⁹	100.0	100.0	100.0	100.0	
Biomin IMB 52	-	0.1	0.2	0.4	

 Table 1:
 Composition of feed mixtures (% of total amount)

Biomin preparation is produced by the Biomin Company, Austria

Table 2:Content of vitamin - mineral prenSLOV - VIT - BR3							
Parameter	Unit	Amount					
Dry matter	g.kg ⁻¹	995					
Lysine	g.kg ⁻¹ g/kg	220					
Methionine	g.kg ⁻¹ g/kg	180					
Threonine	g.kg ⁻¹ g/kg	20					
Vitamin A	m.j.kg ⁻¹	1000					
Vitamin D3	g.kg ⁻¹	25					
Vitamin E (α -tocopherol)	mg.kg ⁻¹	7400					
Vitamin B2	mg.kg ⁻¹	660					
Vitamin B12	mg.kg ⁻¹	3200					
Mn	mg.kg ⁻¹	6000					
Cu	mg.kg ⁻¹	2000					
Zn	mg.kg ⁻¹	6000					
Fe	mg.kg ⁻¹	10000					

The trial groups (A, B, C) were fed with the
addition of the probiotic preparation IMB 52 based on
Enterococcus faecium with different concentrations.
The group A was offered $2.5.10^{11}$, the second group B
was given $5.0.10^{11}$ and third C group 1.10^{12} cfu.t ⁻¹ that
represents 0.1, 0.2 and 0.4 percents of the total amount,
respectively.

The method for mixing of probiotic preparation consisted of 3 steps. First 200 g of patch was prepared (120 g of wheat + 80 g of additives) and then mixed to

1000 g (800 g of wheat and 200 g a small patch) then
100 kg together with other components were established.
Total quantity of feed mixture was granulated. Probiotic
preparation was protected against heating.

In the trial the following parameters were observed: feed consumption - for the whole group which was monitored in months' intervals and in days for animal weighting. From total weight of feed and leftovers per group we have calculated feed consumption per head and day, consumption for one egg and consumption for one kg of egg mass. By calculating of the average consumption of feed and nutrients in feed mixtures we have computed also the intake of dry matter, crude protein and energy per day, per egg and per kg of egg mass. Further evaluated parameters were: laying intensity, egg weight and produced egg mass. There parameters were determined individualy (laying intensity and egg weight) in the trial groups and were compared with parameters determined in the control group. Statistical evaluation was carried out by the programme Microsoft Excel. For the statistical relevance of the difference of mean values a two sided t-test was used.

RESULTS AND DISCUSSION

In evaluating the average feed consumption for one layer per day (table 4), feed consumption in control group was 124.1 g and in the experimental groups the values observed were 119.6 g for group B, 120.7 g for group C and 121.5 g for group A. Average feed consumption per egg observed followed the same tendency and sequence as the other parameters. Values registered for

Parameter	Units	HYD - 10/1	HYD -10/2	HYD -10/3	HYD - 10/4	
Dry matter	g.kg ⁻¹	902.0	899.0	901.0	900.0	
Crude protein	g.kg ⁻¹	157.0	156.0	157.0	154.0	
Fat	g.kg ⁻¹	44.0	44.0	46.0	45.0	
Fibre	g.kg ⁻¹	23.0	25.0	24.0	23.0	
Nitrogen free extract	g.kg ⁻¹	554.0	552.0	551.0	552.0	
Ash	g.kg ⁻¹	124.0	122.0	123.0	126.0	
Ca	g.kg ⁻¹	38.0	38.0	38.0	39.0	
Р	g.kg ⁻¹	5.0	5.1	5.0	5.0	
Na	g.kg ⁻¹	2.4	2.5	2.3	2.2	
Sugar as sucrose	g.kg ⁻¹	29.0	29.0	30.0	31.0	
Starch	g.kg ⁻¹	448.0	444.0	437.0	444.0	
Vitamin A	mj.kg ⁻¹	7647.0	8 723.0	7 286.0	7 793.0	
MEN	MJ.kg ⁻¹	11.80	11.71	11.69	11.73	

Table 3: Nutrient content (g.kg⁻¹) of complete feed mixtures fed to laying hens from 24-43rd weeks (average values)

average feed consumption for 1 kg of egg was highest in the control group (2.156 kg), while in the experimental groups, values were lower and in the sequence: C group (2.068 kg), B group (2.073) and A group (2.102). This data about decreasing feed consumption were confirmed by Kurtoglu et al. (2004), Yoruk et al. (2004), Horniaková (2005) and others in their experiments with application of probiotic preparations as additives to feed mixtures. These results were statistically not significant.

Likewise in ISA – Brown layers, also in layers of Leghorn Panda et al. (2003) reported similar results. Their experimental findings showed that the application of probiotic additives significantly affected laying intensity and qualitative markers of shell weight. Feed intake for hen per day was influenced also by the values of the assumed nutritives. Dry matter consumption for per hen per day was lower in control group (111.9 g), in comparison to the experimental groups with the following values: 107.7 g for group B, 108.6 g for group C and 109.2 g for group A. Consumption of crude protein was also lower in all the experimental groups in comparison to the control group K.

Consumption of metabolizable energy per hen per day was lower in all the experimental groups in comparison with group K (1.464 MJ). All other observed parameters were lower in the experimental groups. Intake and consumption of feed and thereafter intake and consumption of nutritives per egg or per kg of egg mass

Demonstern		ŀ	K		А		В		С	
Parameter		value	index	value	index	value	index	value	index	
Average consumption	of feed									
for one hen per day	g	124.07	100.00	121.47	97.90	119.57	96.37	120.68	97.27	
for one egg	g	133.80	100.00	132.15	98.77	129.78	97.00	130.00	97.16	
for 1 kg of eggs	kg	2.156	100.00	2.102	97.50	2.073	96.15	2.068	95.92	
Average consumption	at one hen per	day								
Dry matter	g	111.91	100.00	109.20	97.58	107.73	96.26	108.61	97.05	
Crude protein	g	19.48	100.00	18.95	97.28	18.77	96.36	18.58	95.38	
MEN (10)	MJ	1.464	100.00	1.422	97.13	1.398	95.49	1.416	96.72	
Average consumption	of nutritives f	or 1 egg								
Dry matter	g	120.69	100.00	118.80	98.43	116.93	96.88	117.00	96.94	
Crude protein	g	21.01	100.00	20.62	98.14	20.38	97.00	20.02	95.29	
MEN	MJ	1.579	100.00	1.547	97.97	1.517	96.07	1.525	96.58	
Average consumption of nutritives for 1 kg of eggs										
Dry matter	kg	1.945	100.00	1.890	97.17	1.868	96.04	1.861	95.68	
Crude protein	g	338.49	100.00	327.91	96.87	325.46	96.15	318.47	94.09	
MEN	MJ	25.44	100.00	24.61	96.74	24.23	95.24	24.26	95.36	

Table 4:	Consumption of feed, dry matter, crude protein and en	ergy
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Table 5: Egg weight, intensity of egg production and amount of produced egg mass

					Parameter				
Group	Egg weight (g)			Laying intensity (%)			Amount of produced egg mass (kg)		
	x	S _X	v %	$\overline{\mathbf{x}}$	S _X	v %	x	S _X	v %
K	61.97	1.22	1.98	92.78	9.65	10.40	8.12	1.21	1.49
А	62.89b	1.32	2.09	91.93	10.85	11.80	8.09	1.33	1.65
В	62.34	1.16	1.87	92.14	9.4	9.92	8.04	1.06	1.32
С	62.52	1.10	1.77	92.83	9.1	9.80	8.13	1.18	1.45

b - differences between values are significant at the level a = 0,05

were best manifested in the experimental groups B and C. These are the groups with higher concentrations of probiotics.

During the period of the experiment from 24th to 43rd week of life span of hens, that is 140 days of laying, average production of eggs per hen was 129.8 eggs in control group, which corresponds to the relative value of 92.73 %. Higher laying intensity was found only in the experimental group C (92,83 %), and in experimental groups A and B we observed a decrease in laying intensity to 91.93 % and 92.14 % respectively. These differences were not statistically significant (P>0.05).

Major differences in probiotics usage was evaluated in animals giving lower amount of animal products. Nickolovova and Penkov (2004) resported that the average production of duck eggs (93.91 opposite to 67.88 per head) and laying intensity (45.92 % versus 34.63 %) was higher in groups which were fed with mixtures containing probiotic additives. This tendency was maintained in the whole laying period.

The average amount of produced egg mass (table 5) per layer was compared with the control group (8.12 kg). In the experimental groups a decrease in production was observed (in A and B group), which does not correspond to the data from Yoruk et al. (2004) and Panda et al. (2003), who dreported statistically significant increase of produced egg mass in ISA-Brown and Leghorn laying hens during the whole laying period. May be the positive effect is more efficient in the second laying period, when the utilisaton of nutrients is worst.

The lowest value of egg weight (table 5) was noted in the control group (61.97 g). In all the trial groups higher values of egg weights were observed in the following sequence: group A (62.89 g), group C (62.52 g) and group B (62.34 g). The determined tendency was statistically significant in the trial group B (P<0.05). This tendency was confirmed by Kurtoglu et.al (2004) who carried out a trial with probiotics, observed higher values of egg weight, but not statistically significant.

CONCLUSION

From results of experiments with the hens of laying type ISA - Brown (brown shell), which were fed by a mixture with additives of the probiotic preparation IMB 52, there was a tendency of decreasing feed consumption and nutritives for per hen, per egg and per kg of produced egg mass. Better results were noticed in experimental groups with highest concentration of active substances (IMB 52). The lowest intake was noticed in the groups B and C.

While evaluating laying intensity and egg weight we have noticed for our observed period (first half of laying period) highest laying intensity (92.3 %)

in the experimental group C and highest value of egg weight in the experimental group A. Values of laying intensity varied from 91.93 % to 92.83 % without being statistically significance (P>0.05). Values of egg weight in the experimental groups A and B were statistically significant in comparison to the control group (P<0.05).

In the amount of produced egg mass significant changes were not noticed. Lower values (8.09 and 8.04 kg, respectively) were recorded in groups A and B, while group C and control group K registred higher values, 8.13 and 8.12 kg, respectively.

From the breeding point of view it is very important which of the parameters will be preferred by breeders during the considered period, noticing that the concentration of probiotic preparation IMB 52 affects different individual parameters. Financial expenditure will be lower using probiotic preparations in regards to better health status of layers and lower costs for medicines.

REFERENCES

- CORRIER, D.E HOLLISTER, A. G NIBSET, D. J SCANLAN, C. M - BEIER, R. C - DELOACH, J. R. 1994. Compatitive exclusion of Salmonella enteriditis in Leghorn chicks. In: *Avian Diseases*, vol. 38, 1994, no. 2, p. 297 - 303.
- HAŠČÍK, P. ČUBOŇ, J. HORNIAKOVÁ, E. KRIVÁNEK, L. - KULÍŠEK, V. 2005. Vzťah medzi aplikáciou probiotického preparátu a množstvom abdominálneho tuku u výkrmových kurčiat. In: *Poľnohospodárstvo*, roč. 51, 2005, č. 11, s. 574 - 579.
- HORNIAKOVÁ, E. 2005. The influence of Enterococcus faecium M-74 bacteria on bone mineralization in chickens. In: *Proceedings* (15th European Symposium on Poultry nutrition), Hungary, p. 195 - 197.
- HORNIAKOVÁ, E. KRIVÁNEK, L. 2002. Produkčná účinnosť kŕmnych zmesí s probiotikom Lactiferm L5 pre brojlerové kurčatá. In: *Poľnohospodárstvo*, roč. 48, 2002, č. 8, s. 407 - 412.
- KURTOGLU, V. KURTOGLU, F. SEKER, E. COSKUN, B. - BALEVI,T. - POLAT, E. S. 2004. Effect of probiotic supplementation on laying hen diets on yield performance and serum and egg yolk cholesterol. In: *Food Additives & Contaminants*, vol. 21, 2004, no. 9, p. 817 – 823.
- MUNDT, J. O. 1986. Enterococci. In: SNEATH, P. H. A.: Bergey's Manual of Systemic Bacteriology, vol. 2, 1986, p. 1063 – 1065.
- NICKOLOVOVA, M. PENKOV, D. 2004. Experimental influence of Laktina probiotic on egg laying characteristics, fertility and viability in muscovy duck. In: *Journal of Central European Agriculture*, vol. 5, 2004, no. 4, p. 353 358.
- PANDA, A. K. REDDY, M. R. RAMA,R.A. O. S. V. - PRAHARAH, N. K. 2003. Production performance, serum/yolk cholesterol and immune competence of white leghorn layers as influenced by dietary supplementation with probiotic. In: *Trop Anim Health Prod.*, vol. 35, 2003, no. 1, p. 85 - 94.

- SCHIRALDI, O. BARI, C. TROTTA, F. 1982. Le traitement par Streptococcus faecium des porteurs asymptomatiques de Salmonelles. In: *Méd. et. Hyg.*, vol. 40, 1982, p. 455 -456.
- SCHLEIFER, K. H. KLIPPER-BÄLZ, R. 1984 Transfer of Streptococcus faecalis and Streptococcus faecium to the Genus Enterococcus nom. re. as Enterococcus faecalis comb. nov. and Enterococcus faecium comb. nov. In: J.

Syst. Bacteriol, vol. 34, 1984, p. 31 - 34.

- VOŘÍŠEK, K. 1989 *Probiotika a gastrointestinální mikroflóra.* Praha : VŠZ, Agronomická fakulta, 109 s.
- YORUK, M. A GUL, M. HAYIRLI, A. MACIT, M. 2004. The effects of supplementation of humate and probiotic on egg production and quality parameters during the late laying period in hens. In: *Poultry Science*, vol. 83, 2004, p. 84-88.

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