Influence of a probiotic of *Bacillus toyoi* strain on performance of growing turkey poult

Einfluss eines Probiotikas – Präparates von *Bacillus toyoi* auf die Wachstumsleistung von Puten

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**Introduction**

Due to the ban on the utilization of growth-promoting antibiotics in poultry feeding in EU countries (ANON, 2003), alternative feed additives with similar effects were searched for (ANADON et al., 2006; BONNEAU and LAARVELD, 1999). Probiotics, prebiotics, symbiotics, organic acids and other natural substances have been taken into consideration (BOMBA et al., 2002; HOLZAPFEL and SCHILLINGER, 2002; TUCKER, 2002).

Probiotics added in adequate quantity to water or feed have beneficial influence on animal health (STANTON et al., 1997). Their mode of action is based on the inhibition of development of intestinal tract pathogenic bacteria, mainly by decreasing the pH of intestinal digesta. They also neutralize enterotoxins created by pathogenic *E. coli* strains, and produce the antibiotic-like substances against pathogenic microbiota: nizine, reuterin, acidophilin, acidolin and lactosin (JADAMUS et al., 2002; JIN et al., 1997). Probiotic additive to the diet may also modify some haematological and immunological parameters of blood (ÇETIN et al., 2005).

A lot of results of the investigations carried out on poultry have proved the beneficial influence of probiotics on performance and health of these animals (HOMMA and SHINOHARA, 2004; NAVA et al., 2005). These effects on performance depend on bacteria strains and their concentration in the probiotic preparation. The *Bacillus toyoi* seems to be an interesting strain of bacteria. Its positive effect was affirmed in other species of animals (JADAMUS et al., 2002; STAMATI et al., 2006). The investigations carried out by JEROCH et al. (2004) showed also beneficial influence of this probiotic on feed utilization in growing heavy turkey.

The aim of the study was to determine the influence of probiotic preparation containing different levels of *Bacillus cereus* var. *toyoi* strain (Toyocerin®) on body weight gains, feed conversion ratio, some blood indices and also on health of growing turkeys.

**Materials and Methods**

Three hundred and sixty male middle-heavy BUT 9 turkey poult were used in the experiment. They were randomly allocated to 3 groups, 120 birds each. Besides, on the first day of the trial, 12 extra birds were assigned to each group. It was because of the possibility of sexing mistakes and occurrence of possible mortality at the first few days of rearing. The birds of particular groups were randomly placed in four 12 square meter pens treated as replications, 33 birds each. There was a tuba feeder and a hanging bell drinker connected with a water reservoir in each pen. Turkeys were kept in the same poultry house, according to the norms enclosed in the instructions of rearing which were worked out in the hatchery distributing the poult. During the weighing on 42nd day of the birds’ life the females, as well as the weakest males, were eliminated. Finally only 30 birds in each pen were left for subsequent investigations.

Group I (control) of turkeys was fed with full-feed mixture (loose form) without antibiotics or other growth promoters. It contained Monensin (playing a coccidiostatic role in birds digestive tract) added to the mixture by Elancoban® preparation. Group II and III animals received the same mixture like control one but with Toyocerin® (Rubinum S.A., Barcelona, Spain) additive. Toyocerin® is a probiotic preparation, containing live cultures of *Bacillus cereus* var. *toyoi*. It was supplemented to mixtures of group II and III, providing $0.2 \times 10^9$ or $1 \times 10^9$ B. *toyoi* CFU/kg feed, respectively. This supplement was added and mixed right after production of mixtures appropriate to rearing period.

Birds were fed *ad libitum* with the above mentioned balanced full-feed mixtures, according to nutrient recommendations (NRC, 1994). Their compositions are shown in Table 1. Declared content of vitamins, microelements and coccidiostatic in the mixtures in particular rearing periods of turkeys is presented in Table 2.

Birds were weighed at the beginning of the study (1 day-old poult) and at the end of each period of trial. These terms were determined by the introduction of subsequent feeding mixtures according to the recommendation of their producer in 3rd, 6th, 9th, 12th week and also at the end of trial (18 weeks). Body weights, weekly feed and water intake in each pen, as well as mortality was recorded. Weekly body weight gains and also feed intake and feed conversion ratio per 1 kg body gain in particular periods

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1 CFU – colony forming units
were estimated. Survival rate of turkeys and also the European Efficiency Factor according to the following formula:

$$\text{EEF} = \frac{\text{survival} \times \text{live weight (kg)} / \text{age (days)} \times \text{feed conversion} \times 100}{100}$$

were calculated as well (ANDRYS et al., 2003; LEMME et al., 2006).

The samples of feed mixtures for laboratory analysis were collected two times in each feeding period. Feeds were analysed for basic nutrients (crude protein, crude fiber, ether extract and crude ash) contents (AOAC, 2000). Metabolizable energy was calculated by the European Tables formula (1989). The commercial mash feed containing TOYOCERIN was analyzed for the active ingredient, viable *Bacillus cereus toyoi* in the laboratory (Bayrisches Landesamt für Gesundheit und Lebensmittelsicherheit, D-85764 Oberschleissheim, Germany).

On 42nd, 84th and 126th day of experiment blood samples (10 ml) for analytical tests were taken from brachial vein of 24 turkeys (8 birds from each group). Then, the samples were collected in tubes containing heparin as anticoagulant. In the full blood haemoglobin content (Hb),...
Table 3. Content of nutrients and nutritive value of basal mixtures for turkeys
\( \text{Nährstoffgehalte der Grundmischungen} \)

<table>
<thead>
<tr>
<th>Item</th>
<th>Prestarter</th>
<th>Starter</th>
<th>Grower 1</th>
<th>Grower 2</th>
<th>Grower 3</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0-3</td>
<td>4-6</td>
<td>7-9</td>
<td>10-12</td>
<td>13-18</td>
</tr>
<tr>
<td>ME, kcal</td>
<td>2839</td>
<td>2905</td>
<td>2980</td>
<td>3054</td>
<td>3142</td>
</tr>
<tr>
<td>Dry matter, g</td>
<td>892.1</td>
<td>894.7</td>
<td>893.7</td>
<td>893.5</td>
<td>892.8</td>
</tr>
<tr>
<td>Crude protein, g</td>
<td>267.2</td>
<td>250.1</td>
<td>227.7</td>
<td>211.8</td>
<td>189.3</td>
</tr>
<tr>
<td>Crude fiber, g</td>
<td>39.4</td>
<td>40.2</td>
<td>40.9</td>
<td>41.3</td>
<td>45.2</td>
</tr>
<tr>
<td>Ether extract, g</td>
<td>28.6</td>
<td>36.9</td>
<td>51.2</td>
<td>58.8</td>
<td>64.3</td>
</tr>
<tr>
<td>Crude ash, g</td>
<td>71.4</td>
<td>64.4</td>
<td>59.9</td>
<td>55.0</td>
<td>50.2</td>
</tr>
<tr>
<td>Calcium, g</td>
<td>13.9</td>
<td>12.9</td>
<td>11.4</td>
<td>11.0</td>
<td>10.2</td>
</tr>
<tr>
<td>Total phosphorus, g</td>
<td>10.2</td>
<td>9.4</td>
<td>8.6</td>
<td>8.2</td>
<td>7.4</td>
</tr>
</tbody>
</table>

Results and discussion

The analysis of basic nutrients, calcium, phosphorus and metabolizable energy in mixtures given in particular rearing periods (Table 3) corresponded with the recommendations (NRC, 1994).

The average concentration of Bacillus cereus in feed of group II during the whole trial oscillated around 0.18 ± 0.02 × 10^7 CFU kg⁻¹ diet, while in group III it amounted to 0.96 ± 0.03 × 10^7 CFU kg⁻¹ diet. These values only slightly differed from planned concentrations in experimental diets: 0.2 × 10^9 CFU kg⁻¹ and 1.0 × 10^9 CFU kg⁻¹ of mixtures for group II and III, respectively.

Average body weights of turkey poults registered on the first day of their life and right before each change of feeding mixture as well as at the end of the experiment are presented in Table 4. Body weights of one-day-old turkey poults were typical for the middle-heavy BUT 9 turkey. It should be noticed that from 1st to 42nd day of life, turkey poults were characterised by their body weights near the average values given in breeding instructions for BUT 9 turkeys (B.U.T., 2002), whereas on 56th, 84th and 126th day of their life they were a bit heavier compared to the standard. The average body weight of turkeys in particular groups point out that significant differences between treatments regarding this feature were noted only from 42nd day of birds’ life. Group III, receiving a higher level of Toyocerin® preparation (1 × 10^9 B. toyoi CFU kg⁻¹ diet), had significantly higher body weight than the control and group II (receiving 0.2 × 10^9 B. toyoi CFU kg⁻¹ diet). Statistically significant differences (p ≤ 0.01) in body weight of 84- and 126-day-old turkeys between all examined groups were revealed. The lowest body weights were noticed in the control group and the highest in the experimental group receiving the higher level of Toyocerin®. Contrary to these results, DENLI et al. (2003) did not record any significant improvement of chicken body weight affected by the feed enriched with the probiotic.

From the start of the trial to the end of 3rd week of experiment considerable differences in body weight gain (Table 4) between both groups receiving mixtures with Toyocerin® and control treatment were noted (p ≤ 0.05). In the periods from 7th to 9th and from 10th to 12th week of rearing only group III animals gained weight significantly better. In the last 6 weeks of the trial (week 13–18) not any considerable differences between treatments were noted. The average total weight gain in the whole experiment was the highest in group III – 13,903 g, whereas the control group animals and the birds from the group receiving a smaller dose of probiotic achieved 13,102 and 13,663 g body weight gain, respectively. The differences between particular groups were significant (p ≤ 0.01). Similarly, significant positive influence of probiotic Bacillus subtilis additive to the diet for turkey poults on their body weight gain was noted by BLAIR et al. (2004).

The average feed conversion ratio (FCR 2.57) for the entire study was somewhat higher than announced in standard values for this genetic line. It could be a result of the loose (non-granulated) mixtures used in this trial. This parameter amounted in control group to 2.61 (Table 5). In the experimental groups it was distinctly (p ≤ 0.05) lower by 2% and 4% in group II (0.2 × 10^9 B. toyoi CFU kg⁻¹ diet) and group III (1 × 10^9 B. toyoi CFU kg⁻¹ of diet), respectively. Positive, significant influence of this probiotic (providing B. toyoi at 1 × 10^9 CFU kg⁻¹ feed) on feed conversion ratio were also noted by JEROCH et al. (2004), who carried out the investigations on BIG 6 growing turkeys. Results presented in this study are also similar to those presented by JANKOWSKI et al. (2004), who characterized FCR in the trial with turkeys (BUT 9) with utilization of various growth promoters in the diets. Total FCR in 16 weeks period of the research amounted from 2.54 in group with antibiotic supplementation to 2.67 in control group, while group feeding with probiotic addition (Bacillus toyoi strain) showed the value of this parameter near 2.64. These differences were not significant. Contrary to these results GRASHORN (1998) did not notice any positive influence of probiotic, antibiotic or herbal diet supplement on body weight gains of growing turkeys.

The survival rate of birds in all groups was very good. The highest value of this parameter achieved in group III
reached 97.7%. Only slightly worse (96.2%) was the survival rate of group II, receiving lower level of Toyocerin®, whereas in control group it amounted to 92.4%. The survival rate of all groups presented in this work was much higher than the one achieved in the investigation carried out on turkeys by GUMMI et al. (2006). For birds reared in the intensive system this index amounted to from 82.4 to 89.4%, while for turkeys derived from the semi-intensive system from 76.7 to 96.6%. DJOUVINOV et al. (2005) fed a diet containing probiotic Lactina® to ducklings till 93rd day of their life. They noted that the mortality of birds fed the diet with this probiotic supplementation amounted to 1.7%, while in group without additive it was almost twice as large.

To summarise the whole rearing period on the basis of performance effects, the European Efficiency Factor (EEF) was estimated. Similarly to the above described parameters characterising fattening period, which were the best in group III, also the EEF in this group was the highest and amounted to 427 points. It was lower in group II (409 points). The control group obtained the smallest value (396 points). These results are better or similar to those presented by PUCHAJDA et al. (2000), who compared three genetic lines of turkeys: BIG 6, HLW and Nicholas 700. They reared the birds for 16 weeks and received the following values of EEF: 339, 302 and 397 points, respectively.

The studies dealing the influence of probiotics additive to the diets on haematological and biochemical blood indices are rather scarce. The experiment carried out on growing turkeys by ÇETIN et al. (2005) revealed the increase of haemoglobin, hematocrit and immunological parameters without any influence on leucocyte count when the probiotic Primalec® 454 was used. Contrary to their results in the present experiment significantly (p ≤ 0.05) decreased the level of WBC in blood of turkeys receiving the diet with higher level of Toyocerin® (1 × 10^9 B. toyoi CFU kg^-1) within the first 12 weeks of their age, while hematocrit remained constant (Table 6). Lower level of WBC in blood of turkey females from group III could point to the positive influence of probiotic addition on intestinal microflora by pathogenic microbe exclusion (WIERUP, 2000; AHMAD, 2006). It indicates a better health state of birds receiving Toyocerin®. None of the investigated biochemical parameters of turkey blood serum (Table 6) was modified by probiotic additive except, in tendency, increase of total protein (4.95, 5.01 and 5.06 mmol l^-1 in control and group II and III, respectively) and, in tendency, decrease of total cholesterol (3.25, 3.15 and 3.08 mmol l^-1 in control and both experimental groups, respectively).

### Table 4. Body weight (BW, g) and body weight gain (BWG, g·week^-1) of turkeys in particular groups (n = 120)

<table>
<thead>
<tr>
<th>Age (weeks)</th>
<th>Parameter</th>
<th>Group</th>
<th>SEM</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 (start)</td>
<td>BW</td>
<td>control</td>
<td>56.9</td>
<td>56.7</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td></td>
<td>1.95</td>
<td>1.39</td>
</tr>
<tr>
<td>3</td>
<td>BW</td>
<td>II</td>
<td>654</td>
<td>660</td>
</tr>
<tr>
<td>0-3</td>
<td>BWG</td>
<td>III</td>
<td>199</td>
<td>201b</td>
</tr>
<tr>
<td>6</td>
<td>BW</td>
<td>II</td>
<td>2283A</td>
<td>2298A</td>
</tr>
<tr>
<td>4-6</td>
<td>BWG</td>
<td>III</td>
<td>543</td>
<td>546</td>
</tr>
<tr>
<td>9</td>
<td>BW</td>
<td>II</td>
<td>4429A</td>
<td>4484A</td>
</tr>
<tr>
<td>7-9</td>
<td>BWG</td>
<td>III</td>
<td>194</td>
<td>218</td>
</tr>
<tr>
<td>12</td>
<td>BWG</td>
<td>III</td>
<td>715A</td>
<td>729A</td>
</tr>
<tr>
<td>10-12</td>
<td>BW</td>
<td>III</td>
<td>1011A</td>
<td>1038A</td>
</tr>
<tr>
<td>18</td>
<td>BWG</td>
<td>III</td>
<td>322</td>
<td>327</td>
</tr>
<tr>
<td>13-18</td>
<td>BWG</td>
<td>III</td>
<td>444</td>
<td>531</td>
</tr>
<tr>
<td>0-18 Total gain</td>
<td>BWG</td>
<td>III</td>
<td>1009</td>
<td>1021</td>
</tr>
</tbody>
</table>

A, B, C – values in the same rows with different letters differ significantly at p ≤ 0.01
a, b, c – values in the same rows with different letters differ significantly at p ≤ 0.05.
Conclusion

Toyocerin preparation, containing probiotic of *Bacillus toyoi* strain, added to the growing turkey diets improved their performance – body weight gains and feed conversion ratio, by up to about 3 and 4%, respectively. It also increased considerably the survival rate of animals. The effects of this preparation depended on its level. Feeding diets enriched with Toyocerin decreased the number of leucocytes in turkey blood. The concentration of $1 \times 10^9$ *B. toyoi* CFU kg$^{-1}$ diet can be accepted as the optimum dose.

Summary

The aim of study was to determine the influence of a probiotic containing *Bacillus cereus* var. *toyoi* strain (Toyocerin®) on performance (body weight gain, feed conversion ratio), blood indices and on health of growing heavy turkeys. In total, 360 male turkey poults BUT-9 were used in the experiment. They were randomly allocated to 3 groups, 120 birds each. Group I – control, was fed a diet without antibiotics or other growth promoters, while groups II and III – contained two different levels of Toyocerin preparation: $0.2 \times 10^9$ and $1 \times 10^9$ *B. toyoi*/CFU kg$^{-1}$ feed, respectively.

The production effects achieved by turkeys from control were according to the standards of the used breed. It was proved, however, that the performance of birds receiving Toyocerin®, was considerably better in comparison with control group. Decreased level of leucocyte counts in blood of turkeys receiving diet with probiotic additive was noted. The effects of this preparation depended on its level. The concentration of $1 \times 10^9$ *Bacillus toyoi* CFU kg$^{-1}$ can be accepted as the optimal dose.

Key words

Turkey, probiotic, *Bacillus toyoi*, performance, blood parameters

Zusammenfassung

Einfluss eines Probiotika – Präparates von *Bacillus toyoi* auf die Wachstumsleistung von Puten

Das Ziel der Studie war, den Einfluss einer Zugabe des Probiotikums „Toyocerin® (*Bacillus cereus* var. *Toyoi*) auf Entwicklung (Zuwachsraten, Futterverwertung), Blutparameter und Gesundheitszustand von mittelschweren Puten...
zu prüfen. Im Versuch standen 360 männliche Puten der Linie BUT-9 zur Verfügung, die zufällig auf drei Behandlungsgruppen mit je 120 Stück verteilt wurden. Die Tiere der Gruppe I (Kontrolle) wurden mit einer Alleinfuttermischung ohne Probiotik-Zugabe gefüttert, während die Gruppen II und III Futter mit einem Zusatz an Toyocerin® erhielten: $0.2 \times 10^9$ und $1 \times 10^9$ CFU B. toyoi kg$^{-1}$ Alleinfuttermischung.


### Stichworte

Pute, Probiotika, Bacillus toyoi, Wachstum, Blutparameter

### References


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