EFFECT OF DIFFERENT DOSES OF DEOXYNIVALENOL ON METABOLISM IN BROILER CHICKENS

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Abstract

The experiment was conducted to investigate the effect of different doses of deoxynivalenol on plasma indices of broiler chickens. Forty-two one-day-old male broiler chicks were fed 1 of 3 diets containing deoxynivalenol (DON) for 42 d. The diets included: (1) control (0.2 ppm of deoxynivalenol), (2) low level of deoxynivalenol (1 ppm of DON), and (3) high level of deoxynivalenol (3 ppm of DON). Then, all the birds were sacrificed and blood samples for biochemical analyses were collected. The mycotoxin doses in diets were verified using gas chromatography-mass spectrometry. The administration of 1 ppm of DON altered total protein, triglycerides, free glycerol, and potassium levels. Dietary addition of 3 ppm of DON resulted in altered calcium, potassium, total protein, triglycerides, along with free glycerol levels, and aspartate aminotransferase activity. No biochemical parameter, however, responded to increased DON concentration in the diet. The feeding of DON-containing diets did not significantly alter plasma chloride, cholesterol, and albumin levels or aspartate aminotransferase, alkaline phosphatase and lactate dehydrogenase activities. It was concluded that both levels of deoxynivalenol in the diets tested significantly affected protein and lipid metabolism in broiler chicks.

Key words: chicken, deoxynivalenol, mycotoxin, metabolism, blood biochemistry.

Trichothecenes are a structurally diverse group of toxic secondary metabolites produced by *Fusarium* and related species of fungi, which usually contaminate cereal grains in countries with a temperate climate. Consumption of trichothecene-contaminated grains is associated with several clinical findings in both animals and humans. Most trichothecenes cause necrosis and inflammation of the oral cavity. The ‘helicopter disease’ is a manifestation of altered feather structure, growth, and appearance, and is commonly observed in chickens fed trichothecene-contaminated feed.

In this study, we were interested in deoxynivalenol (DON, vomitoxin), a trichothecene toxin, which occurs regularly throughout the world, with possible high contamination levels in cereal grain from various countries (23).

Many studies describe the adverse effects of DON on animal and human health. Indeed, in domestic or laboratory animals, large doses of DON cause feed refusal, decreased weight gain, vomiting, gastrointestinal, and dermal irritation, and immunological alterations. Lower doses of DON have been shown to provoke elevation of serum IgA level and are known to affect cell-mediated and humoral immunity in several animal species (3, 30). The sensitivity to DON varies considerably between species. Poultry is more sensitive to DON and to other trichothecenes than ruminants but less sensitive than swine.

Trichothecenes are well-known inhibitors of protein synthesis (21). They also cause apoptosis, both in vitro and in vivo, in various organs (19). Trichothecenes are also shown to interfere with the metabolism of membrane phospholipids and to increase liver lipid peroxides in vivo (20). In addition, some trichothecenes are shown to alter the activity of serotonin in the central nervous system, which is known to be involved in the regulation of food intake (27).

The aim of this study was to determine the effect of different doses of DON on biochemical variables of growing broiler chickens.

Material and Methods

Forty-two, one-day-old male broiler chicks of a commercial strain Ross 308, Párovske háje, Slovakia, were distributed randomly into groups of 14 chicks in each one. The birds were kept on the floor for the course of the study. Chicks were initially kept at 31°C; then the temperature was gradually lowered by 2°C/week to reach 21°C by the end of week 5, and this temperature was maintained during the experiment. Continuous lighting and water ad libitum were provided throughout the experiment.
The chicks received the diet from the day of hatch to 42 d of age. The three types of diet included control (0.2 ppm of DON) and experimental (1 and 3 ppm of DON). To provide stable dietary content of mycotoxin throughout the experiment, the chickens were fed only one type of poultry feed, i.e. HYD-02. The final diet was obtained by mixing the basal diet (a concentrate supplied by Agrokonzult s.r.o., Slovakia) with 40% addition of maize. Maize used for the control diet contained DON background level of 0.5 ppm, while zearalenone and 15 acetyldeoxynivalenol were below detection limits. Experimental diets contained DON contaminated-maize at the level of 7.5 ppm. Zearalenone and 15-acetyldeoxynivalenol levels in the contaminated maize were 0.3 and 0.6 ppm, respectively. Concentrations of T-2 toxin, isoT-2 toxin, T-2 triol, T-2 tetraol, HT-2 toxin, fusarenon-X, 3-acetyl deoxynivalenol, DAS, scirpentriol, nivalenol, 15 acetylscirpentriol, neosolanil, zearalenol, aflatoxins, and fumonisins were below detection limits in both control and mycotoxin-contaminated maize. The mycotoxin content in the basal diet (the part of HYD-02 diet before addition of 40% portion of control or contaminated maize) were found to be 0.05 and 0.0026 ppm for zearalenone and total aflatoxins, respectively. DON, T-2 toxin, and total fumonisins were below the detection limit of the method used.

All the birds were sacrificed and blood samples were collected. Plasma was separated by centrifugation at 1 600 g for 10 min and stored at – 20°C until analysis.

All experimental procedures with animals were in accordance with European guidelines for care and use of animals for research purpose and they were approved by a local ethics committee.

Mycotoxins in the maize were detected using the GC-MS method (24). The mycotoxin contents in the basal diet were analysed using NOACK ELISA kits with spectrophotometric evaluation.

All biochemical parameters were determined by the colourimetric methods using spectrophotometric kits. The plasma chloride concentration was determined with the use of the method by Kuffer et al. (17); calcium concentration was determined with the use of the method by Ray Sarkar and Chauchan (25); magnesium concentration was measured with the use of the method by Mann and Yoe (18); potassium level was determined with the use of the method by Hiltsmann and Beyer (9); total protein level was determined with the use of the method by Doumas et al. (6); albumin level was determined with the use of the method by Deacon and Dawson (4); triglycerides and free glycerol levels were determined with the use of the method by Koditschek and Umbreit (12). The plasma alkaline phosphatase activity was determined with the use of method by Chromý et al. (11); lactate dehydrogenase activity was determined with the use of the method by Wroblewski and La Due (30), alanine aminotransferase and aspartate aminotransferase activities were determined with the use of the method by Reitman and Frankel (26).

The results are expressed as mean ± S.E.M. Statistical significance was evaluated by one-way ANOVA test.

### Results

Plasma potassium, total protein, triglycerides, and free glycerol were decreased in chickens fed DON-contaminated diet (1 and 3 ppm) (Table 1). Dietary addition of 3 ppm of DON resulted in increased plasma alanine aminotransferase activity. However, the DON-contaminated diet did not significantly alter other biochemical plasma parameters, including chloride, albumin, cholesterol, aspartate aminotransferase, alkaline phosphatase, and lactate dehydrogenase.

### Table 1

Effect of dietary administration of deoxynivalenol (DON) on mean plasma indices in growing broiler chickens

<table>
<thead>
<tr>
<th>Indices</th>
<th>Control</th>
<th>0.2 ppm of DON</th>
<th>1 ppm of DON</th>
<th>3 ppm of DON</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chloride (mmol/L)</td>
<td>104.3 ± 4.88</td>
<td>104.50 ± 3.88</td>
<td>114.40 ± 4.36</td>
<td></td>
</tr>
<tr>
<td>Calcium (mmol/L)</td>
<td>2.17 ± 0.07</td>
<td>1.86 ± 0.19</td>
<td>3.36 ± 0.34</td>
<td></td>
</tr>
<tr>
<td>Magnesium (mmol/L)</td>
<td>0.86 ± 0.05</td>
<td>1.50 ± 0.06</td>
<td>0.34 ± 0.01</td>
<td></td>
</tr>
<tr>
<td>Potassium (mmol/L)</td>
<td>12.01 ± 1.04</td>
<td>5.87 ± 0.22</td>
<td>4.56 ± 0.58</td>
<td></td>
</tr>
<tr>
<td>Alkaline phosphatase (µkat/L)</td>
<td>8.06 ± 0.81</td>
<td>10.05 ± 0.05</td>
<td>10.72 ± 0.12</td>
<td></td>
</tr>
<tr>
<td>Alanine aminotransferase (µkat/L)</td>
<td>0.25 ± 0.02</td>
<td>0.22 ± 0.03</td>
<td>0.47 ± 0.00</td>
<td></td>
</tr>
<tr>
<td>Aspartate aminotransferase (µkat/L)</td>
<td>2.01 ± 0.16</td>
<td>2.15 ± 0.14</td>
<td>2.12 ± 0.09</td>
<td></td>
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<tr>
<td>Lactate dehydrogenase (µkat/L)</td>
<td>2.99 ± 0.12</td>
<td>3.02 ± 0.38</td>
<td>2.88 ± 0.43</td>
<td></td>
</tr>
<tr>
<td>Total protein (g/L)</td>
<td>39.11 ± 1.52</td>
<td>27.04 ± 1.74</td>
<td>27.59 ± 1.92</td>
<td></td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>16.12 ± 0.46</td>
<td>15.60 ± 0.09</td>
<td>15.21 ± 0.32</td>
<td></td>
</tr>
<tr>
<td>Cholesterol (mmol/L)</td>
<td>4.08 ± 0.19</td>
<td>3.54 ± 0.19</td>
<td>3.80 ± 0.34</td>
<td></td>
</tr>
<tr>
<td>Triglycerides (mmol/L)</td>
<td>1.02 ± 0.06</td>
<td>0.36 ± 0.04</td>
<td>0.34 ± 0.02</td>
<td></td>
</tr>
<tr>
<td>Free glycerol (mmol/L)</td>
<td>0.91 ± 0.06</td>
<td>0.27 ± 0.04</td>
<td>0.23 ± 0.02</td>
<td></td>
</tr>
</tbody>
</table>

Significant differences within a row are indicated by the same superscript letter, P<0.01; ± SEM; n = 14.
Discussion

In this experiment, diet containing 1 and 3 ppm of DON were given to growing broiler chickens for 6 weeks. The deoxynivalenol treatment in both doses significantly decreased plasma level of total protein in chicks.

Our results are consistent with those of Kubena et al. (15) who found a decreased total protein level in broiler chicks exposed to a DON contaminated diet (16 mg/kg) from 1 to 3 weeks of age. Bergsjø et al. (2) reported a significant decrease in serum protein and albumin in growing pigs fed a diet containing 3.5 mg/kg of DON. They considered that these effects may be secondary to the reduced feed uptake, but the inhibition of protein synthesis may play a role. Toxic action of DON was thought to consist in the inhibition of protein synthesis (27). Later, Mikami et al. (21) examined the toxicity of DON to porcine hepatocytes. The authors reported that DON reduced albumin secretion from hepatocytes into the medium due to the loss of hepatocytes by apoptosis but also due to the inhibition of protein synthesis. DON has also been reported to reduce serum albumin level in growing piglets fed a diets containing 8.6 mg/kg mycotoxin for 36 d (5).

The toxicity of DON in both concentrations tested was expressed through decreased plasma triglycerides and free glycerol levels. These findings are in agreement with the previous reports of Kubena et al. (14). On the other hand, Accensi et al. (1) reported that DON in low concentrations (0, 280, 560 or 840 µg/kg of feed) did not alter the performance of weanling piglets, which was tested on 34 haematological, biochemical, and immune characteristics, including potassium, chloride, calcium, cholesterol, triglycerides, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, and lactate dehydrogenase. As a general rule, significant biochemical changes were generally observed in animals receiving higher doses of trichotheccenes.

For instance Kubena et al. (16) described decreased plasma triglycerides and cholesterol in White Leghorn chicks fed a 9 and 18 mg/kg of DON contaminated diet for 35 d, and Huff et al. (10) reported a significant decrease in serum triglycerides in chicks fed a diet containing contaminated wheat (16 mg/kg of DON in feed) for 4 weeks. DON has also been reported to increase liver triglycerides and total liver lipid in White Leghorn hens fed a diet containing 0.25 or 0.70 ppm of DON for 86 or 135 d (7).

Dietary addition of 3 ppm of DON resulted in an increased plasma alanine aminotransferase activity, indicating liver damage. DON has also been reported to increase activities of aspartate aminotransferase, lactate dehydrogenase, and γ-glutamyltransferase in broiler chicks fed DON at 15 mg/kg dose, indicating possible tissue damage and leakage of the enzymes into the blood (13). Similar results were observed in horses (10) and piglets (5) fed Fusarium culture material.

In the present study, plasma calcium increased in birds fed only diet containing 3 ppm of DON.

Previous data of Bergsjø et al. (2) indicated a significant decrease in serum calcium and phosphorus in growing pigs fed a diet containing 3.5 mg/kg of DON. DON has also been reported to induce weak hypocalcaemia in rats fed 1 mg/kg of DON for 6 months, suggesting that calcium metabolism disorders during chronic action of mycotoxin could be partially associated with the secondary vitamin D deficiency (28).

Recently, Gouze et al. (8) observed that plasma electrolytes appeared to increase in mice exposed for 4 weeks to low DON doses. The discrepancy between these results and our data could be due to a number of factors, including sensitivity to DON between species, DON concentration, and source, animal, genetic predispositions, sex, nutritional status, and contamination of feed by other mycotoxin.

Our results showed that no other blood biochemical parameters responded to increased DON concentration in the diet. The feeding of DON-contaminated diet did not significantly alter contents of chlorides, albumin, and cholesterol, activities of aspartate aminotransferase, alkaline phosphatase, and lactate dehydrogenase in broiler chicks. Similar results were reported by Dänicke et al. (3) who found that several serum characteristics, including aspartate aminotransferase, were not affected by increasing dietary DON concentrations (0; 2.3; and 4.6 mg/kg) in swine. These data demonstrated that both doses of DON in the diet (1 and 3 ppm) significantly affected protein and lipid metabolism in broiler chicks. Moreover, under field conditions with additional stress factors, the toxicity of DON could be altered, thus might adversely affect the health and performance of poultry.

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References
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