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Complex toxin binder mycotox® ng influence on the hepatotoxic effect of aflatoxin B_1 in experimental treated goslings

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Abstarct. *The aim of the present investigation was to evaluate the effects of aflatoxin B_1 and Mycotox NG applied either independently or together, on blood total protein, albumin, blood glucose, total bilirubin, triglycerides, cholesterol, enzyme activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (AP), gamma-glutamyl transferase (γ GT), lactate dehydrogenase (LDH) and changes in liver morphology. At the same time, the potential of supplementation of feed with a mycosorbent (Mycotox NG) was evaluated. Experiments were carried out with 40 1-day-old Toulouse geese from mixed sexes divided into one control and three treatment groups (n=10). Groups were as followed: Group I – control (0 mg/kg AFB_1 not supplemented with Mycotox NG); Group II (0.5 g/kg Mycotox NG), Group III (0.5 mg/kg AFB_1) and Group IV (0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox NG). In this study, commercially available geese of Toulouse strain were reared from day one to forty two days in the deep litter system of management and the birds were divided into four groups. Normal feed tested free of aflatoxin (AFB_1), was given to the control (Group - 1). 0.5 g/kg Mycotox was supplemented with the feed to Group 2, Aflatoxin (0.5 mg/kg feed) was supplemented with the feed to Group 3 and Mycotox Ng (0.5 g/kg feed) + 0.5 mg/kg feed AFB_1 was supplemented with the feed to Group 4. The duration of the experiments was 42 days. The monitored blood chemical parameters were analysed on post treatment days 21 and 42. In birds treated only with AFB_1, (group III) increased blood activities of studied enzymes. At the same time, blood total protein, albumin, cholesterol, glucose and triglycerides were reduced as compared to controls. The observed histopathological changes in the liver consisted in various extent of dystrophy (congestion, vacuolar and granular dystrophy, round cell proliferation, necrobiotic changes, hyperplasia of gallbladder epithelium). The addition of mycosorbent (Mycotox NG) to the feed of Groups IV reduced substantially the changes in blood chemistry and the severity and frequency of liver histological lesions. The addition of mycosorbent (Mycotox NG) to the feed of Groups IV reduced substantially the changes in blood chemistry and the severity and frequency of liver histological lesions.*

Keywords: aflatoxicosis B_1, chemical parameters, liver lesions, Mycotox NG, Toulouse geese

Introduction

Liver is the organ responsible for the metabolism and detoxication of a number of xenobiotics in the animal and human body. It performs different functions associated with metabolism, and detoxication of various toxic substances and narcotics (Parsai et al., 2014).

Poultry and mycotoxins have been closely linked over the last few decades as they are one of the most sensitive animal species to the toxic effects of mycotoxins (Solis-Cruz et al., 2019). Aflatoxins were first isolated in the 1960s after a lethal outbreak in turkeys (Blount, 1961) and liver cancer in rainbow trouts (Rucker et al., 2002) fed rations containing peanut and cotton meal. Aflatoxins are bioactive substances whose chemical structure differs a little (Cullen and Newberne, 1993), and are produced by molds from the *Aspergillus* genus (*Aspergillus flavus* and *Aspergillus parasiticus*). These fungi are widespread in nutrients as wheat, corn, rice, nuts, peanuts, dried fruits and spices (Rucker et al., 2002). Aflatoxins are involved in the food chain mainly after ingestion through the alimentary tract of men and animals (Aycicek et al., 2005). Aflatoxicosis is a nutritional toxicosis occurring after consumption of foods contaminated with aflatoxins. Two clinical forms of aflatoxicosis are known in animals – acute, manifested with severe liver damage resulting in death and chronic, with symptomatic course (Cullen and Newberne, 1994). Among all identified aflatoxins (AF), AFB_1, AFB_2, AFG_1 and AFG_2 are the four main forms found in naturally contaminated feeds. The toxicity of AFG_2, AFB_2 and AFG_1 is equal to 10%, 20% and 50% of that of AFB_1 respectively (Leeson et al., 1995). Among these metabolites, AFB_1 is the most dangerous and abundant mycotoxin (Pitt and Miller, 2017). Aflatoxin B_1 is the most toxic and predominant form in feed ingredients (Monbaliu et al., 2010). The AFB_1 has a range of biological toxicities, including acute hepatic toxicity, teratogenicity, mutagenicity, carcinogenicity and immunosuppression (Peng et al., 2015).

The toxic effects of aflatoxins in domestic poultry are manifested with anorexia, apathy, reduced growth performance, reduced feed intake, increased feed conversion ratio, reduced egg production, altered visceral organs' relative

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weights, increased death rates (Bailey et al., 1998; Kubena et al., 1998; Manfi, 2012; Kana et al., 2014); anaemia (Al-Daraji et al., 2004); reduced humoral and cellular immunity (Gabal and Azzam, 1998, Oguz et al., 2003); lower blood serum concentrations of total protein, albumin, cholesterol, triglycerides, glucose (Zhao et al., 2010; Fani Makki et al., 2014; Kana et al., 2014); increased blood total and direct bilirubin (Fani Makki et al., 2014); increased activity of various enzymes – aspartate aminotransferase (AST), alkaline phosphatase (AP), gamma-glutamyl transferase (γ GT) and lactate dehydrogenase (LDH) (Kaki et al., 2012; Kalpana et al., 2012; Fani Makki et al., 2014; Kana et al., 2014; Azizpour and Moghadam, 2015). Histopathological changes in the liver consisted in hepatomegaly, ochre yellow colour, haemorrhages, vacuolar (fatty) dystrophy of hepatocytes, proliferation of neutrophils and mononuclear cells, proliferation of the biliary duct epithelium, necrotic alterations in the parenchyma, fibrosis (Ekhlas, 2012; Ramdas et al., 2013; Azizpour and Moghadam, 2015).

The sensitivity of the different fowl species to the toxic effects of aflatoxins is different. Most sensitive are growing ducklings, goslings, pheasants and broiler chickens (Leeson et al., 1995). From public health point of view, aflatoxins are dangerous for the health of poultry meat consumers as they accumulate and could provoke neoplastic growths (Ishfaq et al., 2014).

Different methods for elimination, inactivation and reduction of bioavailability of aflatoxins in feeds have been implemented: physical (separation, heat, radiation), chemical (ammonia, ozone, sulfites, hypochlorites), biological (bacteria, yeasts) and nutritional (vitamins, minerals). Most of them have at least two disadvantages: high cost of decontamination of feeds and failure to ensure a complete removal of aflatoxins without losing a great part of the nutritional value of feeds (Méndez-Albores et al., 2007; Eshak et al., 2010). One of the most practical methods nowadays is the use of non-nutritional mycosorbents as aluminosilicates, bentonites, clinoptiolites (Miazzo et al., 2000; Oguz and Kurtoglu, 2000; Oguz et al., 2003), activated charcoal (Khadem et al., 2012) or bioproducts: yeast cultures (*Sacchromyces cerevisiae*) (Abdel-Wahhab et al., 2002). Toxin-binders reduce the toxic effects of mycotoxins through binding them and reducing their absorption by the gastrointestinal tract and their bioavailability in blood (Kana et al., 2014).

The present experiment aimed to evaluate the hepatotoxic effects of aflatoxin B_1 through follow-up of changes in concentrations of liver-specific blood parameters and liver morphology. The possibility for effective alleviation of toxic effects of AFB_1 by feed supplementation with the mycosorbent Mycotox NG (Ceva Sante Animale, France) was also evaluated.

Material and methods

Experimental design

Experiments were carried out with 40 1-day-old Toulouse geese mixed sexes divided into one control and three treatment groups (n=10): Group I – control (0 mg/kg AFB_1 not supplemented with Mycotox NG); Group II (0.5 g/kg Mycotox NG containing micronised yeasts, montmorillonite, thymol) (Ceva Sante Animale, France); Group III (0.5 mg/kg AFB_1) and Group IV (0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox NG). The duration of the experiments was 42 days. The monitored blood chemical parameters were analysed on post treatment days 21 and 42. All birds were fed a complete compound feed according to their age category (starter and grower).

The aflatoxin B_1 used in this experiment was produced by *Aspergillus flavus* (99% purity) purchased from Sigma-Aldrich, Germany. All goslings were kept under optimum microclimatic parameters according to requirements of Ordinance, 44/20.04.2006.

Blood samples were collected from *v. metatarsalis medialis* on days 21 and 42 in sterile heparinised vacutainers (FL medical, Italy) for analysis of ALT, AST, AP, γGT, LDH, total protein, albumin, blood glucose, total bilirubin, triglycerides and total cholesterol. Within 30 min after blood collection, blood samples were centrifuged at 1.500×g for 10 min. Plasma was harvested and stored at -20°C until analysis. All biochemical analytes were assayed on an automated biochemical analyser BS–120 (Mindray, China).

After the end of the experiment, control and experimental goslings were euthanized by cervical dislocation, in compliance with Ordinance No. 20 on the minimum requirements for the protection and welfare of experimental animals and requirements to objects for use, cultivation and/ or supply (State Gazette 87/9/11/2012). Liver specimens for histological examination were fixed in 10% formalin, dehydrated in an ascending ethanol series, embedded in paraffin and stained with haematoxylin/ eosin. The experiment was approved by the Bulgarian Food Safety Agency – permission No 201/07.01.2018.

Statistical analysis

The statistical analysis of data was done by one-way ANOVA, and the level of statistical significance – with the Tukey- Kramer test ($p<0.05$). Results are presented as mean ± standard error of the mean (SEM).

Results

Blood chemistry analysis

The toxic effects of AFB_1 with or without Mycotox NG on the values of the traced chemical parameters in the blood on days 21 and 42 are presented in Tables 1, 2 and 3.

Total protein, albumin and glucose: The effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma total protein, albumin, glucose, cholesterol and tryglicerides are presented in Table 1. Plasma total protein, albumin and glucose in geese from group III were statistically significantly lower than controls on day ($p<0.001$). On the 42nd day, the changes were more pronounced ($p<0.001$). The addition of mycosorbent to the feed reduced partly the deleterious toxin effects ($p<0.05$-0.01) analytes.

Table 1. Effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma total protein, albumin, glucose, cholesterol and tryglicerides in geese

Groups	Total protein, g/l		Albumin, g/l		Glucose, mmol		Cholesterol, mmol/l		Tryglicerides, mmol/l	
	day 21	day 42	day 21	day 42	day 21	day 42	day 21	day 42	day 21	day 42
I	50.21± 0.96	49.54± 1.26	29.2± 1.02	27.1± 0.94	19.57± 0.78	21± 0.90	5.68± 0.15	5.60± 0.10	2.05± 0.13	1.90± 0.18
II	51.17± 1.12	48.01± 1.38	30.3± 0.89	28.1± 1.05	19.45± 0.80	20.82± 0.38	5.81± 0.19	5.56± 0.11	1.99± 0.14	1.89± 0.08
III	40.69± 0.74[1c,2c]	37.33± 1.64[1c,,2a]	21.8± 0.89[1c,2a]	17.8± 0.72[1c,2c]	12.67± 0.46[1c,2c]	11.8± 0.40[1c,2c]	3.84± 0.10[1c,2c]	3.66± 0.11[1c,2c]	1.54± 0.08[1a,2a]	1.05± 0.15[1b,2b]
IV	45.91± 1.26[1a,2b,3b]	42.20± 01.57[1c,2a,3b]	24.9± 1.09[1a,2b]	21.7± 1.02[1b,2c]	15.82± 0.59[1b,2b,3a]	14.09± 0.64[1a,2c,3c]	4.96± 0.11[1b,2ab3c]	4.40± 0.12[1c,2c,3c]	1.74± 0.07	1.43± 0.08

*Data are presented as mean±SEM; n=10 geese per group; [a]p<0.05; [b]p<0.01; [c]p<0.001; 1 – control group (0 mg/kg AFB_1 not supplemented with Mycotox® NG)) vs group II (0.5 g/kg Mycotox® NG); 2 – vs experimental group III (0.5 mg/kg AFB_1); 3 – vs experimental group IV (0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox® NG)

Triglyceride and cholesterol: As seen from Table 1, triglyceride and cholesterol concentrations in group III were reduced 21 and 42 days after treatment (p<0.01-0.001) as compared to controls (p<0.01-0.001). The addition of mycosorbent to the feed of group IV partially reduced the deleterious effects of AFB_1 on cholesterol values during both monitoring periods (p<0.01-0.001) and at the same time had a preventive effect on triglyceride value (p>0.05).

Total bilirubin: Effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma total bilirubin in geese Table 2 demonstrated statistically significantly higher values in group III treated at dose of 0.5 mg/kg AFB_1 on days 21 and 42 (p<0.001). The deleterious effects of AFB_1 on blood bilirubin was attenuated in group IV by the addition of 0.5 g/kg Mycotox NG (p<0.05-0.001).

Table 2. Effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma total bilirubin in geese

Groups	Total bilirubin, µmol/l	
	day 21	day 42
I	7.65±0.34	7.23±0.33
II	7.77±0.34	7.21±0.22
III	5.56±0.37[1c,2c]	4.66±0.16[1c,2c]
IV	6.32±0.22[1a,2a]	5.63±0.17[1c2c]

*Data are presented as mean±SEM; n=10 geese per group; [a]p<0.05; [b]p<0.01; [c]p<0.001; 1- control group (0 mg/kg AFB_1 not supplemented with Mycotox® NG)) vs group II (0.5 g/kg Mycotox® NG); 2- vs experimental group III (0.5 mg/kg AFB_1); 3- vs experimental group IV (0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox® NG)

Table 3. Effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma AST, ALT and γ-GT, LDH and AP

Groups	AST (U/L)		ALT (U/L)		γGT (U/L)		LDH (U/L)		AP (U/L)	
	day 21	day 42	day 21	day 42	day 21	day 42	day 21	day 42	day 21	day 42
I	39.1± 2.27	37.2± 2.07	21.3± 1.36	22.9± 1.25	14.7± 1.55	19.8± 1.86	281.1± 9.11	307.0± 13.43	966.4± 8.35	966.2± 6.37
II	37.8± 1.43	37.9± 1.37	19.8± 0.57	22.9± 1.38	13.0± 1.08	20.6± 1.31	284.5± 8.85	308.0± 11.90	971.8± 4.12	966.7± 3.70
III	51.7± 1.44[1c,2c]	64.4± 2.08[1c,2c]	30.5± 1.45[1c,2c]	36.9± 2.97[1c,2c]	27.8± 1.66[1c,2c]	34.3± 1.68[1c,2b]	353.3± 13.61[1c,2b]	449.7± 23.74[1c,2c]	1310.1± 47.02[1c,2c]	1483.4± 25.83[1c,2c]
IV	47.0± 2.47[1a,2c]	55.2± 2.29[1c,2c, 3a]	25.8± 1.23[1a,2b,3a]	31.2± 1.47[1a,2b]	20.1± 0.79[1a,2b,3b]	28.6± 1.23[1b,2a,3b]	330.7± 14.72[1a,2b]	369.0± 10.82[1a,2a,3b]	1110.1± 42.90[1a,2b,3b]	1308.4± 25.83[1c,2c,3c]

*Data are presented as mean±SEM; n=10 geese per group; [a]p<0.05; [b]p<0.01; [c]p<0.001; 1- control group (0 mg/kg AFB_1 not supplemented with Mycotox® NG)) vs group II (0.5 g/kg Mycotox® NG); 2- vs experimental group III (0.5 mg/kg AFB_1); 3- vs experimental group IV (0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox® NG)

Enzymatic activites: Table 3 shows the effects of aflatoxin B_1 applied independently or combined with Mycotox NG on blood plasma AST, ALT and γ-GT, LDH and AP. The toxic effects of AFB_1 on liver function was demonstrated by elevated enzyme activity in group III. The activities in the group IV that received aflatoxin B_1 and mycosorbent was lower to this one in the groups treated only with AFB_1 (p<0.01-0.001). There were no statistically significant changes in studied blood biochemical analytes between the group supplemented with mycosorbent (Group II) and untreated chickens.

Morphological studies

Gross changes in the liver of goslings from the group treated at 0.5 mg/kg AFB_1, are presented in Figure 1. The liver of goslings from the group treated with 0.5 mg/kg AFB_1 was characterised with altered colour of the organ, e.g. much lighter than normal (yellow-brown) due to fat deposition in hepatocytes, with very frail consistency, spattered with striated haemorrhages and petechiae (Figure 1A). The gallbladder was overfilled with cloudy dark green fluid, with multiple floccules.

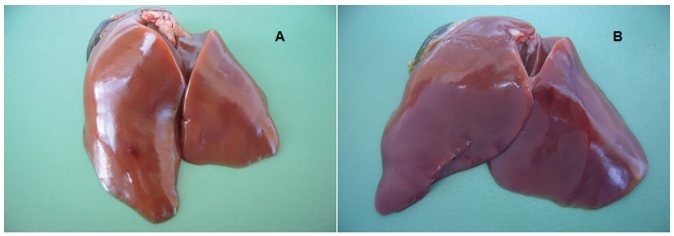

Figure 1. Liver of goslings from the group treated at 0.5 mg/kg AFB_1, showing fatty dystrophy and haemorrhages (A) and liver of goslings from the group treated at 0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox® NG with a brown-reddish colour and yellowish areas at some places (B)

In goslings treated with 0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox NG liver colour was not as intense ochre, with occasional yellowish areas (Figure 1B). The liver of control birds and receiving only Mycotox NG did not show any gross changes in either colour or consistency.

Pathomorphology of liver in goslings treated with 0.5 mg/kg AFB_1 was outlined with granular and fatty degeneration of hepatocytes (Figure 2). The nuclei of cells exhibited chromatolysis (loss of stainable substance), with hyperchromatosis of nuclear membranes. The capillary endothelium was activated, with generalised hyperaemia in organ lobules (Figure 3). At some areas perivascular mononuclear and connective tissue proliferation were seen (Figure 4). In the liver of goslings treated with 0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox NG pathomorphological changes were considerably less intense. Capillary endothelium was activated, a weak perivascular proliferation was also observed (Figure 5).

Birds fed the standard feed only (control group) and receiving only Mycotox NG (Group II) did not exhibit gross changes in normal liver architectonics.

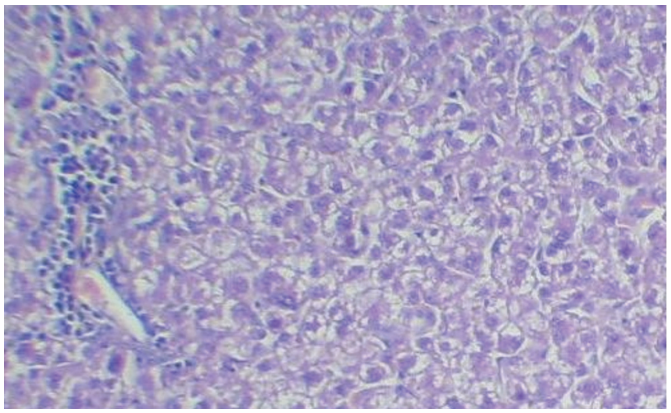

Figure 2. Liver of goslings from the group treated at 0.5 mg/kg AFB_1. Granular and fatty degeneration of hepatocytes. H/E. Bar 20 µm

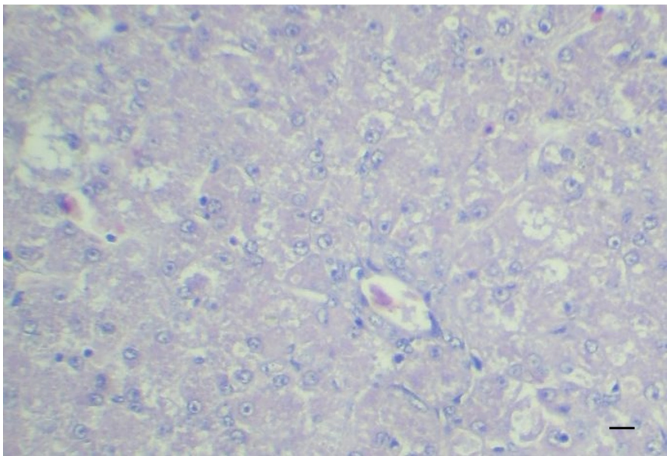

Figure 3. Liver of goslings from the group treated at 0.5 mg/kg AFB_1. Hyperchromatosis of nuclear membranes of hepatocytes and chromatolysis, focal necrosis and substantially activated capillary endothelium. H/E. Bar 20 µm

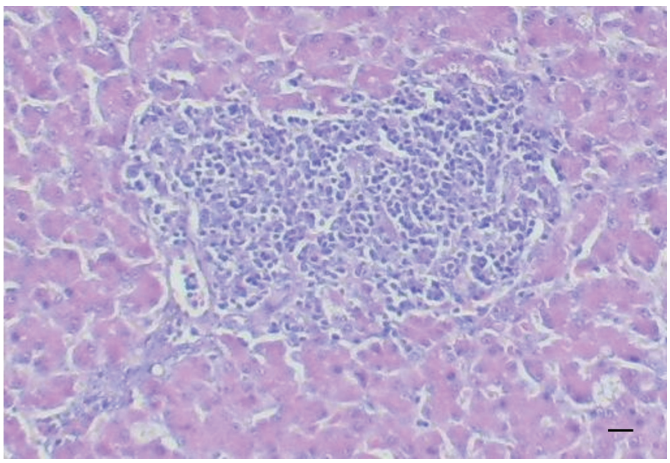

Figure 4. Liver of goslings from the group treated at 0.5 mg/kg AFB_1. Mononuclear and connective tissue proliferation. H/E. Bar 20 µm

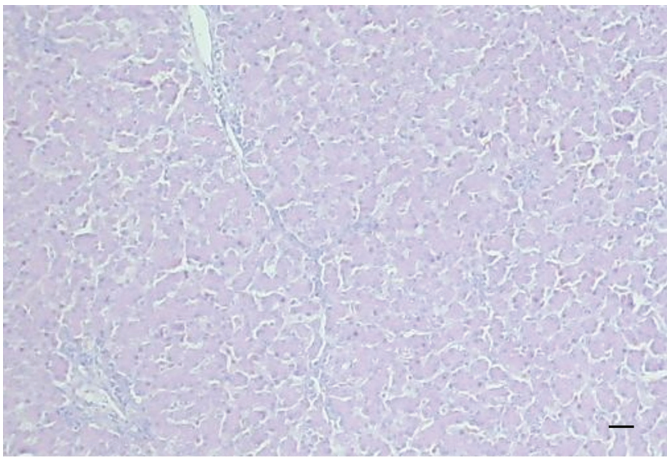

Figure 5. Liver of goslings from the group treated at 0.5 mg/kg AFB_1 and 0.5 g/kg Mycotox Ng. Activation of capillary endothelium and weak perivascular proliferation. H/E. Bar 20 µm

Discussion

Aflatoxin B_1 is among the most commonly encountered poultry feed contaminants, incurring economic losses to poultry industry (Rizzi et al., 1998). The present results showed statistically significant elevation in activities of

amino transferases (AST, ALT and γGT), LDH and ALP. The high activities of AST, ALP, γGT, LDH and ALT in blood are bioindicators of liver damage, was also established by Kubena et al. (1998), Abdel-Wahhab et al. (1999), Miazzo et al. (2000), Nowar et al. (2001), Oguz et al. (2003), Mohamed and Mohamed (2009), He et al. (2010), Zhao et al. (2010), Kana et al. (2014), Parsai et al. (2014), Pappa and Padmalatha (2014) and Wade et al. (2018). These enzymes are located into the cytoplasm and mitochondria of hepatocytes and when the stuctural integrity of the liver is damaged, they pass into the blood plasma (El-Nekeety et al., 2011).

Lower blood levels of total protein and albumin observed in the present experiment agree with the results of others (Shi et al., 2009; Zhao et al., 2010; Kana et al., 2014; Lakkhawar et al., 2016; Wade et al., 2018). The lower protein biosynthesis and dystrophic changes of liver cause reduction in plasma protein concentrations observed in this study (Tessari et al., 2010; Shahzad et al., 2014). Aflatoxins impair the protein synthesis by forming adducts with DNA, RNA and proteins (Busby and Wogan, 1984), RNA synthesis inhibition, reduction of DNA-dependent RNA polymerase activity (Cullen and Newberne, 1994) and inhibition of amino acid transport and mRNA transcription (Wade et al., 2018). Hypoproteinaemia can probably is a result of reduced food intake (Kana et al., 2014; Lakkhawar et al., 2016).

Bilirubin is a secondary product of haemoglobin conversion, filtered through kidneys and then, conjugated to glucuronic acid in the liver and excreted with the bile. Increased total bilirubin results from impaired liver function in aflatoxicosis (Rustemeyer et al., 2010). Hyperbilirubinaemia is a very sensitive test for detection of the functional integrity of the liver, as well as for assessment of dystrophy severity consequently to aflatoxicosis (Rizvi and Shakoori, 2000; Banu and Muthumary, 2010).

The present study established lower plasma cholesterol and triglycerides in turkey broilers treated with AFB_1. They result from aflatoxin-impaired lipid metabolism in the liver (Shi et al., 2009).

Lower blood glucose in the present experiment was due to lower intake of food and/or reduced activity of enzymes involved in carbohydrate metabolism and glycogenolysis (glucose-6-phosphatase and fructose-1,6-bisphophatase) as a result of liver dystrophy or overproduction of insulin (Soliman et al., 2008; Zhao al., 2010). Aflatoxins inhibit the protein synthesis and thus, the excess of amino acids could be used to meet the energy needs of the body and maintenance of blood glucose levels, bit impaired gluconeogenesis could cause lower blood glucose levels (Soliman et al., 2008). Hypoglycaemia could probably result from enhanced glycolysis or reduced absorption of glucose through the intestines (Semenov et al., 2016).

Our observations on liver morphology changes are in line with data reported by Espada et al. (1992), Oguz et al. (2003), Kumar and Balachandran (2009), Zhao et al. (2010) and Ekhlas (2012). Liver dystrophy was due to damage of main cell macromolecules (lipids, proteins, DNA). They result from oxidative stress induced by aflatoxins, a mechanism provoking oxidative damage in DNA and lipid peroxidation (Mohamed and Mohamed, 2009). On the other hand, the accumulation of calcium in hepatocytes provokes mitochondrial dysfunction and reduced rate of adenosine triphosphate synthesis, resulting in morphological disturbances of liver structure (Quesada et al., 2000; Fatemi et al., 2006). The liver is a target organ for the toxic effect of AFB_1, as in the liver aflatoxins undergo bioactivation to reactive 8,9-epoxide, which binds to DNA and proteins. As a result, the normal liver structure is damaged (Pasha et al., 2007). Aflatoxin B_1 is cytotoxic for hepatocytes and inhibits their proliferation (Abdel-Wahhab et al., 2007). The observed histopathological changes in the liver parenchyma are due to the accumulation of fats in hepatocytes following impaired lipid metabolism, which in turn is caused by inhibited synthesis of phospholipids and cholesterol causing impairment of liver lipid transport (Espada et al., 1992).

Compared to control group of birds, the addition of 0.5 g/kg Mycotox NG to the feed of Group IV reduced partly the deleterious effects of aflatoxin B_1 on blood concentrations of studied biochemical blood parameters and the extent of histological liver changes. It is presumed that most of aflatoxin molecules are absorbed by feed ingredients in the gastrointestinal tract causing less adverse effects on the histostructure of target organs and blood biochemical parameters (Azizpour et al., 2015). These data are in concordance with those reported by other authors having used brewers' yeasts (*Saccharomyces cerevisiae*) (Che et al., 2011) and bentonite (montmorillonite) (Shi et al., 2009). The process of binding of aflatoxins with toxin-binders is based on the principle of electric polarity. The negative polarity of mycotoxins is bound by the positive polarity of the toxin binder, so the toxins are immobilised and eliminated from the animal body (Kana et al., 2014).

Conclusion

The results of the present study showed that the supplementation of geese feed with AFB_1 (group III) had a negative effect on liver function as seen from the increased blood enzyme activities of AST, ALT, LDH, γ GT and AP. At the same time, the tested dose of aflatoxin B_1 provoked increase plasma total bilirubin and reduction of total protein, albumin, blood glucose, triglycerides and cholesterol. The single addition to the ration of group III of aflatoxin B_1 induced specific histological changes in the liver. The addition of 0.5 g/kg Mycotox NG to poultry diets contaminated with 0.5 mg/kg AFB_1, was capable to alleviate the severity of changes in analysed parameters and to reduce the severity of histological lesions induced by the aflatoxicosis.

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