HISTOLOGICAL LESIONS OF THE LIVER IN CHICKEN’S OCHRATOXICOSIS

CARMEN SOLCAN, I. COMAN, GH. SOLCAN

Faculty of Veterinary Medicine Iasi
carmensolcan@yahoo.com

Summary

The aim of the study was to evidantiate the histological lesions of the liver in experimental ochratoxicosis of broiler chickens. Investigations were made on 60 broiler chickens divided in 4 groups: E1, E2, E3 and control. The ochratoxin A was gived orally, in vegetal oil suspension, each day (from D1 to D21 of life) in the following doses: 1ppm for E1, 5ppm for E2 and 20ppm for E3. Groups of 5 chickens from both experimental and control group were euthanasiad at 7, 14 and 21 day of life, to study histological lesions. In E1 group after 7 and 14 days of poisoning with ochratoxin the hepatosteatosis and slight glycogenosis was observed. After 21 days of poisoning, the glycogenosis and other degenerative changes of the hepatic cells were more evident. In E2 group, degenerative lesions induced by glycogen storage into hepatic cells were observed both at 7th, 14th and at 21th day. Hepatic cells were surcharged with glycogen. In E3 group the liver distrophy was more marked than in E1 and E2 group. Some regions with hepatic cells without nucleus or completely degenerated alternating with nucleolated cells surcharged with glycogen were observed. Proliferation of biliary ducts epithelium was also observed in E3 group, after 21st day of experimental ochratoxicosis. This can suggest a hepatocarcinogen effect of ochratoxin.

Ochratoxin A (OTA) is a nephrotoxic/carcinogenic mycotoxin, produced by several Aspergillus- and Penicillium-strains. It also have a hepatotoxinc and immunosuppressive effect. The aim of the study was to evidantiate the histological lesions of the liver in an experimental ochratoxicosis pattern of broiler chickens from 1st to 21st day of life.

Materials and methods

Investigations were made on 60 broiler chickens divided in 4 groups: E1, E2, E3 and control. The ochratoxin A was gived orally, in vegetal oil suspension, each day (from D1 to D21 of life) in the following doses: 1ppm for E1, 5ppm for E2 and 20ppm for E3. Control group received only vegetal oil. All the groups received the same fodder and water, free of mycotoxins, ad libitum. Groups of 5 chickens from both experimental and control group were euthanasiad at 7, 14 and 21 day of life, to study histological lesions. The liver fragments were prepared for histological exam by paraffin embedding and stained by HEA, PAS and Pappenheim method.

1 Researches financed by the grant CEEX Neoprev 147/2006
Results and discussions

In control group the liver has a normal structure, the lobulae being undelimited by conjunctive septa, being indentified on the basis of centrolobular venula and the Kiernan spaces near it. Hepatic cells have a polygonal shape, with 1 or 2 nuclei, with nucleolus (Fig 1.).

In E1 group after 7 and 14 days of poisoning with ochratoxine the hepatosteatosis and slight glycogenosis was observed (fig.2). Ochratoxins perturb calcium homeostasia affecting lipid oxydation, inhibiting the activity of mithochondria and ARNt synthetase (Fink-Gremmelset all., 1995). Ochratoxins does inhibit formation of Aminoacetyl ARNt synthetase directly, inhibiting the production of ATP in mithochondria (needed for aminoacids activation) and so inhibit the synthesis of proteins.

After 21 days of poisoning with ochratoxine, the glycogenosis and other degenerative changes of the hepatic cells are more evident, as mentioned other authors (3, 6, 8).

Hepatic cells degeneration induced by ochratoxins is realized by the inhibition of protein kinase, enzyme initiating the the glycogen fosforilation system. Ochratoxine A affect firstly adenosine 3'- 5' AMPc dependent by the proteinase, responsible by the initiation of the enzymatic cascade leading to glycogenolise. Those results confirm the morphologic criteria of glycogenosis from glycogen storage disease, type X which is a AMPc deficiency, dependent by a preoteinkinase. In phisiological conditions those enzymatic cascade is the target of some hormons like glucagon and epinephrine which stimulate the adenilcyclase involved in producing of secondary mesagers AMPc (8).
In E2 group, which received 5ppm OA per day, degenerative lesions induced by glycogen storage into hepatic cells were observed both at 7th, 14th and at 21th day. Hepatic cells were surcharged with glycogen, which in usual staining methods was displaced at one of cell's pole (glycogen lavage). Synusoid capillaries and venulae were ectazied.

![Image of liver](image1.png)

Fig.2. Liver of chickens; 14th day of ochratoxicosis. E1 group. Col PAS x200

In E3 group (receiving 20ppm OA daily) the liver distrophy was more marked than in E1 and E2 group. Some regions with hepatic cells without nucleus or completely degenerated alternating with nucleolated cells surcharged with glycogen were observed. In Kiernan spaces the venulae were enlarged and biliary channels show proliferations of the epithelium (Fig. 4, 5, 6).

The activity of glycolytic enzymes is reduced, whereas those of gluconeogenic enzymes are increased. The diabetogenic effect of ochratoxin A was thought to be due to inhibited synthesis and/or release of insulin from pancreatic cells, thereby suppressing glycolysis and glycogenesis and enhancing gluconeogenesis and glycogenolysis (7).

Proliferation of biliary channel epithelium was also observed in E3 group, after 21st day of experimental ochratoxicosis, even if this lesion is most common in aflatoxicosis. This can suggest a hepatocarcinogen effect of ochratoxin A. Is well known that ochratoxin A causes renal tumours. The hypothesis that DNA damage induced by ochratoxin A is due to oxidative stress represents an alternative explanation supported by several experimental observations. An unusually large number of DNA adducts (up to 30 individual adducts) was formed from ochratoxin A in low yields in various experimental systems (5). Some of these results are consistent with a major role of oxidative stress in the toxicity of ochratoxin A. For
example, antioxidants prevent the induction of DNA damage by ochratoxin A in mice (2). Induction of renal toxicity, oxidative stress due to mitochondrial dysfunction, and persistent cell proliferation represent an alternative mechanism for the renal carcinogenicity of ochratoxin A. The toxin is known to induce oxidative stress and the formation of hydrogen peroxides. In addition, mechanisms linked to long-term renal toxicity and oxidative stress is known to play an important role in tumour induction in rat kidney (1, 4, 5, 7).

![Liver of chickens; 21th day of ochratoxicosis. E2 group. Col PAS x400](image)

Using some inductors or inhibitors of biotransformation enzymes (including cytochrome P-450, cycloxygenase, lipoxigenase, glutathione S transpherase) Pfohl-Leszkowicz şi Castegnaro (2005) demonstrated that ochratoxine is biotransformed in genotoxic derivates which modify the structure of DNA. Some metabolits of ochratoxins were isolated from tissues and cells treated with cu mycotoxins, like lactone (OP-OTA) and quinone (OTQ) which are genotoxic (5).
Fig. 4. Liver of chickens; 14th day of ochratoxicosis. E3 group. Col PAS x400
Glycogenosis

Fig. 5. Liver of chickens; 21th day of ochratoxicosis. E3 group. Col PAS x400
Zones of intense hepatic degeneration (glycogenosis).

Fig. 6. Liver of chickens; 21th day of ochratoxicosis. E3 group. Col PAS x400
Proliferation of biliary duct epithelium.
Conclusions

1. The histological lesions of the liver in experimental ochratoxicosis pattern of 21 days of broiler chickens were studied. In E1 group, receiving 1 ppm ochratoxin each day, after 7 and 14 days of poisoning the hepatosteatosis and slight glycogenosis was observed. After 21 days of poisoning, the glycogenosis and other degenerative changes of the hepatic cells were more evident.

2. In E2 group, which received 5ppm OA per day, degenerative lesions induced by glycogen storage into hepatic cells were observed both at 7th, 14th and at 21th day. Hepatic cells were surcharged with glycogen.

3. In E3 group, receiving 20ppm OA per day, the liver distrophy was more marked than in E1 and E2 group. Some regions with hepatic cells without nucleus or completely degenerated alternating with nucleolated cells surcharged with glycogen were observed. Proliferation of biliary ducts epithelium was also observed in E3 group, after 21st day of experimental ochratoxicosis. This can suggest a hepatocarcinogen effect of ochratoxin A.

References


