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Protective role of biosynthetic silver nanoparticles in broilers with aflatoxicosis through histopathological study of spleen

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Abstract

Aflatoxin (AF) is considered a problematic issue in poultry farms. A novel bio-green synthesis nanoparticle technique is newly introduced in the poultry industry, thus this study aimed to investigate the harmful effects of aflatoxin on the histological structure of broilers' spleen as well as its toxicity on the immune system through the study of CD4+ and CD8+ expression and determination of the silver nitrate nanoparticles (AgNP) protective role against aflatoxin. Forty-five broiler chicks were divided into three groups. T1 control, T2 birds were treated with AF 70 ppb, and T3 birds were treated with AF and silver nanoparticles 150 ppm for 21 days. The result of histological examination in T2 revealed progressive pathological alteration in the red and white pulp with regressive pathological lesions in the splenic trabeculae and central artery sclerosis and white pulp regeneration with edema and congestion in T3. The descriptive chart analysis was used for pathological lesions, showing that the percentages for the red pulp in the three groups were 76, 71, and 73 and for the white pulp 24,29, and 27, respectively. Furthermore, there was a significant decrease of CD4+ and CD8+ expression in the splenic tissue of the broiler in T2 in contrast to T1 and T3. This study concluded that biosynthetic silver nanoparticles can reduce the histological effects and immunotoxicity of aflatoxin, and the descriptive and semi-quantity analysis of the histopathological lesions are essential modern methods in significantly evaluating the results of histological examination.

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Introduction

Aflatoxicosis is a carcinogenic and toxic condition that affects growth and poultry public health as a result of ingestion of feed that is contaminated with aflatoxins, which fungal agents produce as *Aspergillus flavus* and *A. parasitics* which are colonized in the grain and crop under high humidity and temperature (1,2). Aflatoxins B1(AFB1) are absorbed from contaminated feed through the duodenum of poultry and then transported via blood circulatory into the liver. In the liver, it transformed, under the influence of enzymes such as cytochrome P450, into 8,9-epoxide, which is a more electrophilic and highly effective metabolite and can bond with phospholipids, protein, and nucleic acids, leading to several genetic disorders in metabolic signaling, and cell architecture alterations (3). The lymphatic system (organs and vessels) has protective roles against infectious agents, and the spleen is the main secondary lymphoid tissue in all birds (4,5). The spleen plays a critical role in birds' immune system because avian lymphatic arteries and nodes are underdeveloped. The splenic T cell is an important cell that reflects the alignment of mature T cells in the tissue. This composition determines the biological activity of these mature T cells and ultimately affects the body's cellular immune function. Previous research has indicated that AFB1 induces a harmful impact on the spleen tissue, such as

decreasing the number of CD4+ and CD8+T-cells and inducing mutation in the lymphocyte (6,7). Nanotechnology has attracted considerable attention for years because it has many vital applications in medicine, engineering, chemistry, physics, and biology (8). Nanoparticles have unique characteristics in contrast to large bulk particles, and these novelty features are related to the particles' variable-specific properties, such as morphology, size, and distribution. Silver nanoparticles (AgNPs) have been prepared in physical, chemical, and biological methods. Among the prepared methods, some advantages were chosen, such as the chemical reduction method due to synthetic nanoparticles without aggregation, high production, low synthesis cost, and easy and specific conditions. Silver nanoparticles have unique properties such as performance preference and dimensional structures, photoelectric, and catalytic properties, thus it has been implicated in wide applications in many fields (9), such as in medicine and industry such as antifungal (10), antibacterial (11), anti-parasite (12) and antiaflatoxine (13).

Several methods have been used for inactivating, declining concentration, and destroying the mycotoxin, including chemical, physical, and recently biological ones, so this experiment aims to investigate the histological effects of aflatoxin in the spleen as well as evaluate its immunotoxicity on T cells (CD4+ and CD8+) through immunohistochemistry, furthermore study the antagonist activity of AgNPs on the aflatoxin in broiler as well as application the descriptive and semi-quantities mathematical methods in histological examination.

Materials and methods

Ethical approval

The approval from the Scientific Ethical Committee on Animal Experimentation at the College of Veterinary Medicine, University of Mosul, UM.VET.2023.045 was dependent on the conduction of this study.

Housing and animal management

In this study, forty-five unsexed broiler Ross 308 chicks one day old were obtained from a local hatchery. They were then transported to a 1.5*2 m^2 cage, their housing facility at the College of Veterinary Medicine- University of Mosul. The farm underwent a process of decontamination using formaldehyde gas, which was generated by mixing potassium permanganate powder with 40% formalin. The decontamination was carried out in a well-ventilated chamber, and each chamber partition was illuminated with 200-watt electric bulbs to maintain a light and dark cycle of 23 hours of light and 1 hour of darkness per day. A layer of pristine straw, measuring 4 centimeters in depth, was carefully spread over each cell to form a deep litter. Furthermore, each compartment was equipped with a suitable feeder and waterer supplier. The chicks were weighed upon their arrival at the farm. Birds were administered Newcastle disease and avian influenza vaccines via subcutaneous injection (Introvert), and they had free access to water and food throughout the 21-day experiment (14).

Silver nanoparticles

The silver nanoparticles that have been used in this study were obtained from the University of Mosul, College of Veterinary Medicine- Department of Pathology and Poultry Diseases; the size of AgNP was 40nm (15).

The fungal strain (*Aspergillus flavus*)

To obtain Aflatoxin (AFB1), the fungal strain *Aspergillus flavus* was taken from the University of Mosul, College of Science (16). The fungal strain was inoculated in the rice medium and kept in an incubator for at least two weeks at 28°C. After that, the rice stream is heated to inhibit fungal growth, dried, ground, and mixed with the broiler ration (17).

Experimental design

The total number of boilers (Ros 308 = 45) was randomly divided into three groups (treatments), and each one of them was subclass into three cages, five birds /cages, as follows; The first group (T1): broilers were fed with basal ration for 21 days. The second group (T2): broilers were fed with basal ration with aflatoxin at a dose of 70 ppb for 21 days. Third group (T4): broilers were fed with basal ration with aflatoxin at dose 70 ppb and silver nanoparticles 150 ppm for 21 days (18).

Histopathological examination

After a 21-day of exposure to AF, the broilers were anesthetized and sacrificed using the cervical displacement procedure**; the** spleen was collected from each treatment and fixed with formalin 10%, dehydration, clearance with xylene, and then embedded in wax paraffin. Tin sections of 5 micrometers were fixed to the slide and stained with hematoxylin and eosin (H&E) (19). The lesions were categorized in table 1 (20), and the scoring assessed these lesions - grading system as no lesions =0, minimal =1, mild = 2, moderate = 3, and severe =4 (21).

Descriptive chart analysis

A point grid of (17x23) was systematically pragmatic to histological figures derived from five distinct slides, each captured at magnification 100X within every delineated group to assess the proportion of white and red pulps. The enumeration of points overlaying the areas corresponding to white and red pulp was executed, with a standardized total of 391 points uniformly distributed across each evaluated region. Subsequently, the mean for each group was calculated, and the capsule and trabeculae were excluded (23).

Table 1: Immunotoxicity categories in the spleen of the broiler with aflatoxicosis (22)

Reaction	Descriptive splenic area	Lesions criteria
Progressive	Peri arteriolar lymphoid sheath	Decreases or increased (size, number of follicles and lymphocytes)
	Marginal zone	Decreases or increased (size and number of lymphocytes)
	Red pulp	Increased hematopoietic cell
Regressive	Follicles	Decreases or increased (no of lymphocytes and germinal centers)
	Architecture	Increase fibrosis and necrosis

Immunohistochemical analysis

Al-Ali and Al-Sabaawy (24) reported the immunohistochemical procedure involving thin sections of the spleen were affixed onto a layered polylysine glass slide, which was then stained with a monoclonal antibody specific for CD8+ (anti-rat) and CD4+ (anti-rat CD4+, Novocastra, RTU-CD4-1F6, Cat. Number PA0427). The product called the CD8+ antibody from Novocastra is ready to use and has the catalog number PA0183. The severity of immunohistochemical staining can be scored as usual or negative (-), mild (+), moderate (++), and severe (+++) (25).

Statistical analysis

Every experiment was run three times, and the mean and standard deviation of the findings were reported. The t-test was used to determine how the research factors affected the biofilm. The statistical analysis was conducted utilizing SPSS version 26. When $P \leq 0.05$, the differences were deemed significant (26).

Results

Histopathological examination

The microscopic examination revealed normal splenic structure (Figure 1) and the histologic criteria in the spleen of broiler exposed to aflatoxin at 70 ppb for 21 days; these criteria ranged from circulatory disturbances, cell injury, proliferation, depilation with cell alteration. The pathological lesions involve mild edema with red pulp degeneration. There was a minimal foamy arteriolar wall with severe depletion of the periarteriolar sheath (Figure 2). Furthermore, there was congested red pulp and necrosis in the splenic trabeculae (Figure 3), and other mild histological alterations in the spleen were fibroblast proliferation which leads to thickening of trabeculae as well as minimal degeneration of tunica intima of the trabeculae arterioles (Figure 4). Furthermore, minimal vascular disturbances characterized by sclerosis of the central artery with moderate dilatation of venous sinuses (Figure 5). Histological examination of the spleen in the broiler with aflatoxin (70 ppb) and silver nanoparticles (150 ppm) for 21 days reveal moderate white pulp regeneration, mild edema, and venous sinuses congestion (Figure 6).

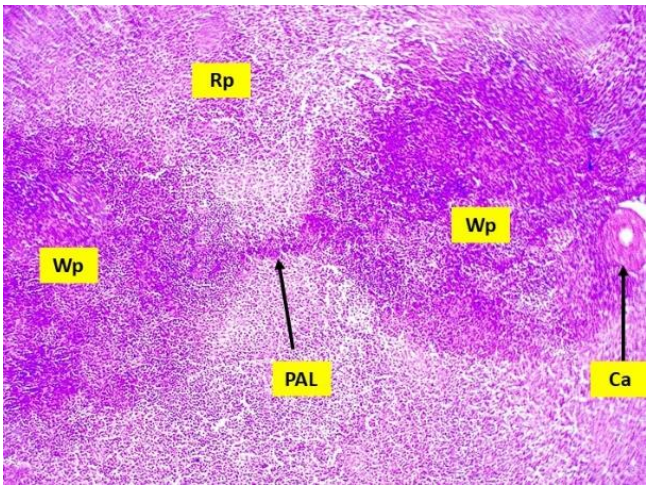

Figure 1: Histological examination of normal splenic structure in the broiler, white and red pulps (WP and RP), respectively, Central artery (Ca) and Periarterial lymphoid sheath (PAL), H&E, 100X

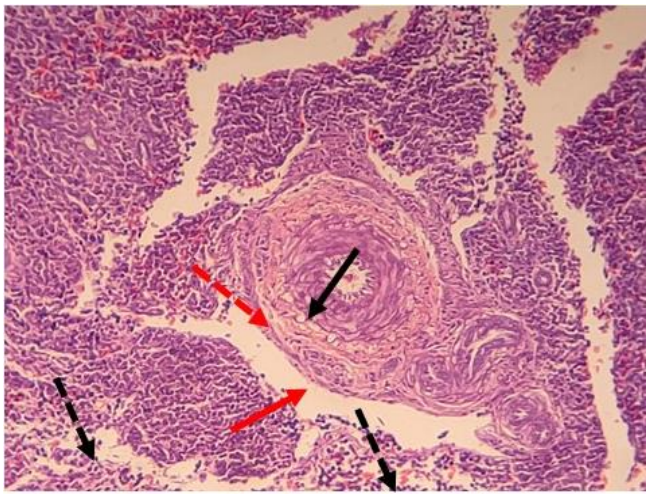

Figure 2: Histological examination of the spleen of a broiler exposed to aflatoxin (70 ppb) for 21 days reveals red pulp degeneration (black dot arrow), foamy arterial wall (black arrow), depletion cells of the periarterial sheath (red dot arrow), and edema (red arrow), H&E, 100X.

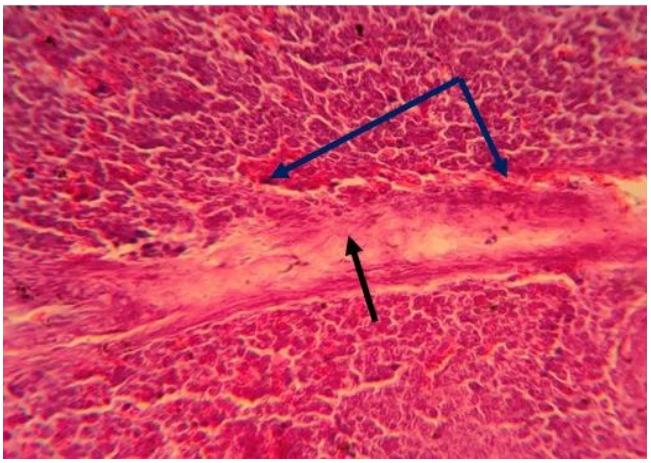

Figure 3: Histological examination of the spleen in the broiler with aflatoxin (70 ppb) for 21 days reveals red pulp congested (blue row) with necrosis in the splenic trabeculae, H&E, 400X.

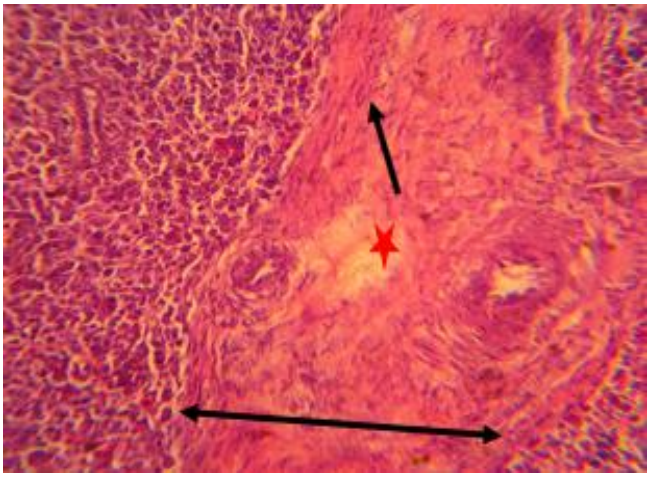

Figure 4: Histological examination of the spleen in the broiler with aflatoxin (70 ppb) for 21 days revealed fibroblast proliferation (black row), edema (red star), and thickening of the splenic trabeculae (two head black row), H&E, 400X.

Descriptive chart analysis

The descriptive chart of histopathological lesions revealed that aflatoxin caused more destruction in the white pulp, and the percentage reached 29% while the red pulp was 71%, in contrast to the control group 24 and 76% for white and red pulp, respectively, while the proportion descriptive means for red pulp in the spleen of broiler exposed to aflatoxin with silver nanoparticles revealed 27% while the percentage of white pulp was 73% (Figure 7).

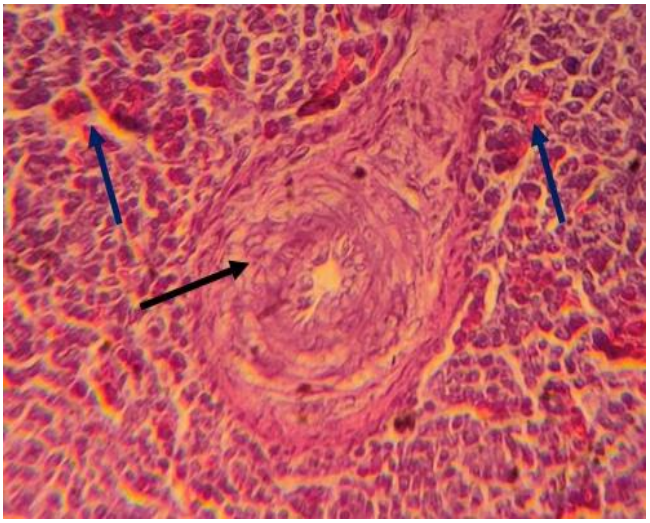

Figure 5: Histological examination of the spleen in the broiler with aflatoxin (70 ppb) for 21 days revealed central artery sclerosis (black row) with dilatation of venous sinuses (blue row), H&E, 400X.

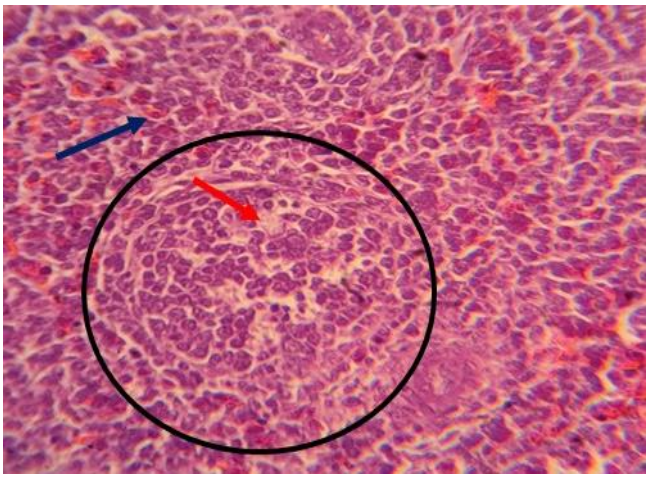

Figure 6: Histological examination of the spleen in the broiler with aflatoxin (70 ppb) and silver nanoparticles for 21 days revealed white pulp regeneration (black circle) with slight edema (red row), and venous sinuses congestion (blue row), H&E, 100X.

Immunohistopathology semi quantities analysis

The spleen serves as the immune organ. The T-cell subsets, including CD4+ and CD8+ lymphocytes, are located within the parenchyma of the spleen. The exposure to AF at a dose of 70ppb impacts T cell expression of both (CD4+ and 8+) at score (+) and grade- mild. This study demonstrated that supplementing broilers with 150 ppm of silver nanoparticles resulted in an improved expression of T lymphocytes (CD4+ and CD8+) at score (++/ moderate) as shown in figure 8.

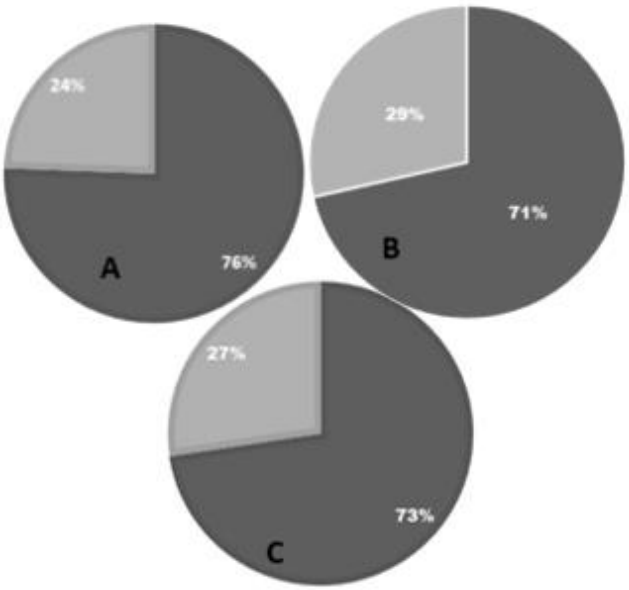

Figure 7: Descriptive analysis. A: proportion of white and red pulps in the spleen of broiler in the control group (%); B: proportion of both two pulps in the spleen of broiler with aflatoxin (%); and C: proportion of splenic white and red pulps of broiler treated with aflatoxin and silver nanoparticles (%).

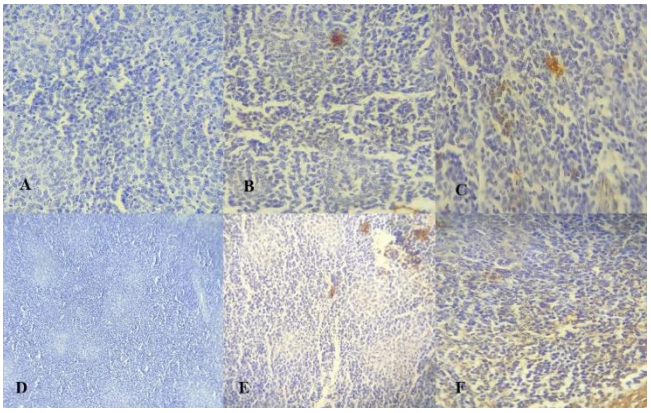

Figure 8: Immunohistochemistry examination revealed negative staining of CD4+ and CD8+ (A and D), minimal expression of CD4+ (B) and CD8+ (E) in the spleen of broiler treated with aflatoxin, mild expression of CD4+(C) and CD8+(F) in in the spleen of broiler treated with Aflatoxin and silver nanoparticles, 400x.

The semi-quantities analysis and descriptive chart is the modern mathematical method for detection the significance pathological alteration between variable groups, so the results of this investigation demonstrate a significant elevation in CD4+ and CD8+ cells in the spleen ($P \leq 0.05$) of broiler in control group 9.20 and 2.40, respectively in compare to first and second treatments, with a significant elevation in the result of semi-quantities of immune expression of CD4+ and CD8+ cells in the spleen of broiler in the second treatment 6.20 and 5.20, respectively (Figure 9).

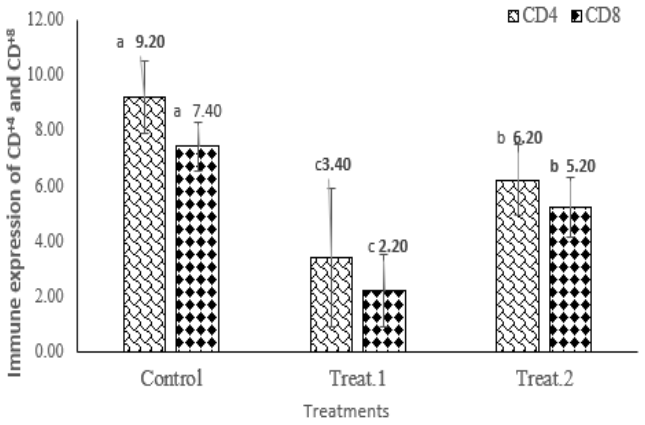

Figure 9: Descriptive chart of aflatoxin and silver nanoparticles in the immune expression of CD+4 and CD+8 lymphocytes in the spleen of broilers.

Discussion

Aflatoxins are a specific category of mycotoxins, the second metabolites formed by fungi in food. Aflatoxin B1 (AFB1), poultry, which is very susceptible to the harmful effects of AFB1 and can manifest as either acute or chronic conditions, leading to reduced meat and egg production, immunosuppression, and decline resistance to diseases (27,28). The main typical pathological alterations were investigated in the spleen of the broiler at 21 days of age with aflatoxicosis congested and degeneration of red pulp with depletion cells of the periarterial sheath, edema, and decreased number of lymphocytes in the follicle. These pathological lesions are similar to the results of a previous study (29,30), which demonstrated blocked proliferation of T and B cells in the spleen of Aflatoxin broilers. The descriptive chart is one of the best and most recent methods in histopathological diagnosis; descriptive planning is one of the best and most modern methods of pathological diagnosis, as it was observed through this study the percentage of red and white pulp in both groups (31).

The spleen is a vital organ in the immunological system. T lymphocyte subset as CD4+ and CD8+ cells are the primary population cells in the spleen parenchyma. The immunohistochemistry analysis of this study shows that the lymphocyte cell expression both for the CD4+ and CD8+ was declined in the spleen of broiler with Aflatoxicosis at dose 70 ppb (21). The thymus and T cells migrate to the spleen, undergo cell division, insight the spleen, and are subsequently transported to the peripheral circulation and

lymphatic organs. The CD4+ lymphocytes are considered helper/inflammatory T cells, which respond to external antigens in conjunction with major histocompatibility complex (MHC) class II molecules. On the other hand, CD8+ T cells respond to internal antigens in conjunction with MHC class I molecules and typically act as cytotoxic T cells. Previous studies have shown that AFB1 causes a decrease in the development and growth of the thymus (32). The results of this study investigated whether there was a reduction in mature splenic T lymphocytes due to dietary AFB1, which agreed with the result of a previous study (33). Thus, the decrease in the population of the T cells in splenic tissue may be attributed to both inhibited thymus growth and reduced propagation of T cells in the spleen. Several studies have shown that AFB1cause mitochondria injury (34,35), damage and loss of the integrity of the lymphocyte cell membranes, also causes a reduction in the RNA and DNA synthesis, and this causes inhibition in both migration and growth of T cells (36,37) and triggering apoptosis in splenocytes. In addition, red pulp was congestion correlated with a reduction in the proportions of CD4+ CD8+ T cells. Reports indicated that a lack of oxygen, resulting from congestion, significantly reduced the growth of T cells (38-41).

Combination of Aflatoxin and AgNPs combined with improvement of immune expression, the results supported by previous research conducted by Jasim and Al-Taee (18), $AgNO_3$ can absorb aflatoxins leading to reduce the exposure concentrations of mycotoxins in chicks (42), also it considered as an effective antifungal agent that inhibits the growth of fungi and inhibits the production of mycotoxins. The unique criteria of $AgNO_3$ nanoparticles play a vital role in the adsorption of aflatoxin, or the attraction with different charges, in addition, it has been active substances from plant extracts that enter the synthetic processes of nanomaterials. With all of these features, $AgNO_3$ nanoparticles are crucial in interacting with aflatoxins activity (43), and these reasons support the immune improvement results in this current study through the improvement of gene expression of CD4+ and CD8+ T cells.

Conclusion

It is concluded from this study that the toxic effect of aflatoxin on the spleen leads to insight alteration (progressive and regressive cellular reaction). It was inferred, through the use of the descriptive chart analysis, that these alterations are progressive as depletion cells of the periarterial sheath, splenic arterial sclerosis, and circulatory disturbances and don't cause deleterious effects in the spleen architecture, also from the results of this study indicates that aflatoxin is an immunotoxin lead to depilation of T cells. That silver nitrate nanoparticle is essential in reducing the toxic effect of aflatoxin and protective splenic tissue.

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Conflict

The authors declare no conflict.

Reference

1. Medina A, Rodriguez A, Magan N. Effect of climate change on *Aspergillus flavus* and aflatoxin B1 production. Front Microbiol. 2014;5:348. DOI: 10.3389/fmicb.2014.00348
2. Kumar A, Pathak H, Bhadauria S, Sudan J. Aflatoxin contamination in food crops: Causes, detection, and management: A review. Food Prod Process Nutr. 2021;3:17. DOI: 10.1186/s43014-021-00064-y
3. Wang Y, Wang X, Li Q. Aflatoxin B1 in poultry liver: Toxic mechanism. Toxicon. 2023;233:107262. DOI: 10.1016/j.toxicon.2023.107262
4. Vali Y, Gumpenberger M, Konicek C, Bagheri S. Computed tomography of the spleen in chickens. Front Vet Sci. 2023;10:1153582. DOI: 10.3389/fvets.2023.1153582
5. Sabourin PJ, Price JA, Casbohm SL, Perry MR, Tuttle RS, Rogers JV, Rowell KS, Estep JE, Sabourin CL. Evaluation of acute immunotoxicity of aerosolized aflatoxin B1 in female C57BL/6N mice. J Immunotoxicol. 2006;3:11-20. DOI: 10.1080/15476910500468635
6. Korkmaz D, Kum S, Eren U. The effects of vitamin E on T cell subsets and immunoglobulin-containing plasma cells in the spleen of heat-stressed broiler chickens. Med Weter. 2023;79(06):6778-2023. DOI: 10.21521/mw.6778
7. Abedi A, Talebi E. effect of aflatoxins on poultry production and control methods of destructive influence. ARPN J Agric Biol Sci. 2015;10(12):441-446. [available at]
8. Tabassum N, Vidyasagar GM. Synthesis, characterization, and antimicrobial activity of silver nanoparticles using *Santalum album aqueous* seeds extract. Int J ChemTech Res.2016;9(05):352-8. [available at]
9. Chen SY, Huang MT, Pender SF, Ruslin M, Chou HH, Qu KL. The application of silver nanoparticles on developing potential treatment for chronic rhinosinusitis: Antibacterial action and cytotoxicity effect on human nasal epithelial cell model. Mater Sci Eng C. 2017;80:624-630. DOI: 10.1016/j.msec.2017.03.189
10. Yu A, Wang Q, Wang J, Chang C. Rapid synthesis of colloidal silver triangular nanoprisms and their promotion of TiO2 photocatalysis on methylene blue under visible light. Catal Commun. 2017;90:75-78. DOI: 1016/j.catcom.2016.11.004
11. Xue B, He D, Gao S, Wang D, Yokoyama K, Wang L. Biosynthesis of silver nanoparticles by the fungus *Arthroderma fulvum* and its antifungal activity against genera of Candida, Aspergillus and Fusarium. Int J Nanomed. 2016;11:1899-1906. DOI: 10.2147/IJN.S98339
12. Kamyar SH, Mansor AB, Mohsen Z, Wan M, Zin WY, Nor AI, Parvaneh SH, Mansour GH. Synthesis and characterization of silver/ montmorillonite/chitosan bionanocomposites by chemical reduction method and their antibacterial activity. Int J Nanomed. 2021;6:271-284. DOI: 10.2147/IJN.S16043
13. Mohammed SA, Ali AA. Effect of selenium nanoparticles against protoscoleces of *Echinococcus granulosus* in vitro and hydatid cysts in mice. Iraqi J Vet Sci. 2022;36(I):195-202. DOI: 10.33899/ijvs.2022.135838.2535
14. Maty HN. Impact of sorbitol and l-carnitine on stimulating thyroid hormone, triiodothyronine, and adenosine triphosphate levels in broilers. Iraqi J Vet Sci. 2023;37(3):589-590. DOI: 10.33899/ijvs.2022.135305.2464

15. Jasim YJ, AL-Taee SK. Evaluation of the Antifungal activity of green biosynthesis silver nanoparticles using pomegranate peel extract. IOP Conf Ser Earth Environ Sci. 2023;2031:1315-1755. [available at]
16. Al-Dabbagh RQ. Isolation and identification fungi from foods produced by some local factories in Mosul City [master's thesis]. Biology / Botany: Mosul University; 2019. 72 p.
17. Al-Shawabkeh K, Herzallah S, Al-Fataftah A, Zakaria H. Effect of aflatoxin b1 contaminated feed on broiler chickens' performance and meat content of conjugated linoleic acid. Jordan J Agric Sci. 2009;5(3):314-323. [available at]
18. Jasim YJ, Al-Taee SK. Evaluation of the role of green synthesis silver nanoparticles as adsorbents and protective agents for broilers tissue treated with aflatoxin. Iraqi J Vet Sci. 2023;37(3):675-681. DOI: 10.33899/ijvs.2023.136771.2614
19. Jaber MT, Al-Jumaa M, Al-Taee SK, Nahi HH, Al-Hamdany MO, AlSalh MA, Al-Mayahi B. Bioaccumulation of heavy metals and histopathological changes in muscles of common carp (*Cyprinus carpio* L.) in the Iraqi rivers. Iraqi J Vet Sci. 2021;35(2):245-249. DOI: 10.33899/ijvs.2020.126748.1368
20. Elmore SA. Enhanced histopathology of the spleen. Toxicol Pathol. 2006;34(5):648-55. DOI: 10.1080/01926230600865523
21. Prakoso YA, Puspitasari, Rini CS, Aliviameita A, Salasia SO, Kurniasih, Ikram AD, Walalangi B, Utama KP, Al Huda MF, Su'udiyah NA. The role of *Sauropus androgynus* (L.) merr. leaf powder in the broiler chickens fed a diet naturally contaminated with aflatoxin. J Toxicol. 2018;2018:1-18. DOI: 10.1155/2018/2069073
22. Saied AF, Al-Taee SK, Al-Taee NT. Morphohistopathological alteration in the gills and central nervous system in *Cyprinus carpio* exposed to lethal concentration of copper sulfate. Iraqi J Vet Sci. 2022;36(4):981-989. DOI: 10.33899/ijvs.2022.132781.2131
23. Ungor B, Malas MA, Albay S. The proportions of the white and red pulps of the human fetal spleen. J Saudi Med. 2006;27(9):1315. [available at]
24. Al-Ali SA, Al-Sabaawy HB. Morphology and immunohistochemistry analysis of broiler intestine treated with immunostimulant agents. Iraqi J Vet Sci. 2023;37(4):893-898. DOI: 10.33899/ijvs.2023.137991.2758
25. AL-Taee SK. Immunohistochemically and semi-quantities analysis of carp gills exposed to sodium thiosulfate. Iraqi J Vet Sci. 2024;38(1):191-198. DOI: 10.33899/ijvs.2023.140746.3086
26. SAS Institute. SAS Statistical Guide for personal computers. 3rd ed. USA: Cary Inc.; 2014. 466 p.
27. Fouad AM, Ruan D, El-Senousey HK, Chen W, Jiang S, Zheng C. Harmful effects and control strategies of aflatoxin B_1 produced by *Aspergillus flavus* and *Aspergillus parasiticus* strains on poultry: Review. Toxins. 2019;11(3):176. DOI: 10.3390/toxins11030176
28. Al-Taee ZT, Saeed MG. Correlation incidence between infectious bursal disease and aflatoxicosis in broiler chicken farms in Nineveh province, Iraq. Iraqi J Vet Sci. 2023;37(1):183-190. DOI: 10.33899/ijvs.2022.133881.2315
29. Omar NA. Effect of some aflatoxins on a lymphatic organ (spleen) of male albino rats (histopathological study). Egypt Hosp Med. 2012;48:357-367. DOI: 10.21608/EJHM.2012.16240
30. Li H, Guan K, Zuo Z, Wang F, Peng X, Fang J, Cui H, Zhou Y, Ouyang P, Su G, Chen Z. Effects of aflatoxin B1 on the cell cycle distribution of splenocytes in chickens. J Toxicol Pathol. 2019;32(1):27-36. DOI: 10.1293/tox.2018-0015
31. Hasan AA, Altaey OY, Sultan GA. Morphological, histological, and histochemical study of the adult golden hamster (*Mesocricetus auratus*) spleen. Open Vet J. 2023;13(3):253-261. DOI: 10.5455/OVJ.2023.v13.i3.1
32. Guo SN, Liao SQ, Su RS, Lin RQ, Chen YZ, Tang ZX, Wu H, Shi DY. Influence of Longdan Xiegan decoction on body weights and immune organ indexes in ducklings intoxicated with aflatoxin B1. J Anim Vet Adv. 2012;11:1162-1165. [available at]
33. Ezzat GM, Meki AMA, Meligy FY, Omar H, Nassar AY. Antiapoptotic and chemotaxis-stimulating effects of poly (D, L-lactide-co-glycolide)-chitosan and whey proteins against aflatoxicosis-induced splenic and thymic atrophy. Mol Biol Rep. 2023;50(12):9805-9824. DOI: 10.1007/s11033-023-08902-7
34. Alpsoy L, Yildirim A, Agar G. The antioxidant effects of vitamin A, C, and E on aflatoxin B1-induced oxidative stress in human lymphocytes. Toxicol Ind Health. 2009;25(2):121-7. DOI: 10.1177/0748233709103413
35. Mary VS, Theumer MG, Arias SL, Rubinstein HR. Reactive oxygen species sources and biomolecular oxidative damage induced by aflatoxin B1 and fumonisin B1 in rat spleen mononuclear cells. Toxicol. 2012;302(2-3):299-307. DOI: 10.1016/j.tox.2012.08.012
36. Groopman JD, Kensler TW. The light at the end of the tunnel for chemical-specific biomarkers: Daylight or headlight?. Carcinogenesis. 1999;20:1-11. DOI: 10.1093/carcin/20.1.1
37. Meissonnier GM, Pinton P, Laffitte J, Cossalter AM, Gong YY, Wild CP, Bertin G, Galtier P, Oswald IP. Immunotoxicity of aflatoxin B1: Impairment of the cell-mediated response to vaccine antigen and modulation of cytokine expression. Toxicol Appl Pharmacol. 2008;231(2):142-9. DOI: 10.1016/j.taap.2008.04.004
38. Wang F, Shu G, Peng X, Fang J, Chen K, Cui H, Chen Z, Zuo Z, Deng J, Geng Y, Lai W. Protective effects of sodium selenite against aflatoxin B1-induced oxidative stress and apoptosis in broiler spleen. Int J Environ Res Public Health. 2013;10(7):2834-44. DOI: 10.3390/ijerph10072834
39. Robbins JR, Lee SM, Filipovich AH, Szigligeti P, Neumeier L, Petrovic M, Conforti L. Hypoxia modulates early events in T cell receptor-mediated activation in human T lymphocytes via Kv1.3 channels. J Physiol. 2005;564:131-143. DOI: 10.1113/jphysiol.2004.081893
40. Al-Baker A, AlKshab AA, Ismail HK. Effect of silver nanoparticles on some blood parameters in rats. Iraqi J Vet Sci. 2020;34(2):389-395. DOI: 10.33899/ijvs.2020.165812
41. Al Dujaily AH, Mahmood AK. The effectiveness of biogenic silver nanoparticles in the treatment of caprine mastitis induced by *Staphylococcus aureus*. Iraqi J Vet Sci. 2021;35(I-III):73-78. DOI: 10.33899/ijvs.2021.131415.1946
42. Wijnhoven SP, Peijnenburg WM, Herberts CA, Hagens WI, Oomen AG, Heugens EW, Roszek B, Bisschops J, Gosens I, Van De Meent D, Dekkers S, Dejong WH, van Zijverden M, Sips A, Geertsma RE. Nano-silver - a review of available data and knowledge gaps in human and environmental risk assessment. Nanotoxicol. 2009;3(2):109-38. DOI: 10.1080/17435390902725914
43. Stroka J, Maragos CM. Challenges in the analysis of multiple mycotoxins. World Mycotoxin J. 2016;9:847-861. DOI: 10.3920/WMJ2016.2038

الدور الوقائي لدقائق الفضة النانوية الحيوية في الفروج اللاحم المصاب بتسمم الافلا من خلال الدراسة النسجية المرضية للطحال

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الخلاصة

يعتبر الأفلاتوكسين قضية إشكالية في مزارع الدواجن. أدخلت حديثا الجزيئات النانوية والمصنعة بتقنية التصنيع الحيوي الأخضر المبتكرة الى صناعة الدواجن وبالتالي تهدف هذه الدراسة إلى تحديد التأثيرات الضارة للأفلاتوكسين على التركيب النسجي للطحال في الفروج اللاحم

بالإضافة إلى سميته على الجهاز المناعي من خلال التعبير الجيني لعنقود التمايز الرابع والثامن وتحديد الدور الوقائي لجسيمات نترات الفضة النانوية ضد الأفلاتوكسين. تم تقسيم خمسة وأربعين فرخ من الفروج اللاحم إلى ثلاث معاملات معاملة السيطرة وفي المعاملة الثانية تم معاملة الطيور بالافلاتوكسين ٧٠ جزء في المليون وفي المعاملة الثالثة تم معاملة الطيور بالافلاتوكسين وجزيئات الفضة النانوية ١٥٠ جزء في المليون لمدة ٢١ يوماً. أظهرت نتيجة الفحص النسجي في مجموعة المعاملة الثانية تغييرات مرضية تراجعية في اللب الأحمر والأبيض مع آفات مرضية متقدمة في الحويجزات الطحالية وتصلب الشريان المركزي وتجدد اللب الأبيض مع وذمة واحتقان في المعاملة الثالثة، تم استخدام التحليل الوصفي للآفات المرضية إذ تبين أن النسب المئوية للب الأحمر في المجموعات الثلاث كانت ٧٦، ٧١، ٧٣وللب الأبيض ٢٤،٢٩،٢٧ على التوالي. بالإضافة الى الانخفاض المعنوي في التعبير الجيني لعنقود التمايز الرابع والثامن في النسيج الطحالي للفروج اللاحم في مجموعة المعاملة الثانية بالمقارنة مع مجموعة السيطرة والمجموعة الثالثة. يستنتج من هذه الدراسة بان جزيئات الفضة النانوية المصنعة حيويا يمكن أن تقلل من التأثير النسجي والسمي المناعي لسموم الافلاتوكسين، وان التحليل الوصفي وشبه الكمي طرق حديثة ومهمة في التقييم المعنوي نتائج الفحص النسجي.