

# Study of Histopathological Changes of Silver Nanoparticles in Liver of Coturnix Bird

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## Abstract

Silver nanoparticles (AgNPs) are one of the most widely used particles in the nanotechnology industry because of their antibacterial, antifungal and antiviral properties that enter the blood stream and then organs through inhalation, skin and mouth. This study was conducted to investigate the histological changes caused by silver nanoparticles on the liver of Japanese quail. At this search 100 birds were obtained from the College of Agriculture and Forestry and were distributed into five groups, each of which included 20 birds. The first and second group were dosed orally for 30 days with AgNPs, and the third and fourth groups were dosed for only 3 days, in addition to the fifth group, which is the control group, which was dosed for 30 days with distilled water. Results appeared after 10 days of treatment, changes in the tissue structure of the liver, represented by the expansion of the sinusoids, the severity of Hemorrhage, the emergence of inflammatory focal, thickening and congestion of the blood vessel. The results showed more severe after 30 days of treatment, as the inflammatory focal increased, fatty change of Hepatic cell and an increase in the thickness of the hepatic portal artery and bile ducts. Silver NPs showed histological changes, which were more severe at high concentrations, as it showed vascular congestion, hepatocyte damage, severe Hemorrhage, hepatic portal artery thickness and a Hyperplasia of bile canaliculi. The nanosilver has toxic effects on the liver of quail Japanese.

**Keywords:** AgNPs, quail, liver, histopathology

## 1. Introduction

It is certain that nanomaterials have been produced and used by humans for hundreds of years, but the understanding of some materials as nanostructure was considered relatively recent, and nanotechnology represents 10<sup>-9</sup> as a unit of measurement. Nanomaterials are defined as those materials with a nanometer dimension ranging between 1-100 nanometers [1]. Environmental pollution can be considered one of the most serious problems caused by man to as a result of industrial development, so to solve it using all available technical means, the most important of which is nanotechnology, which controls the properties of the material by forming it to obtain new nanomaterials that clean the environment and rid it of the pollutants that have inflicted on it. Silver particles have distinct physical and chemical properties, so they may interfere with the normal physiological mechanisms of fetuses, developing animals [2, 3]. Due to its small size, it can easily penetrate through the cell membrane [4]. At this, quail was used as a laboratory animal due to its small body size, consumption of a small amount of feed, short life cycle and early sexual maturity, in addition to the low cost of raising it, its adaptation to the appropriate conditions and its resistance to many diseases [5].

## 2. Materials and Working Methods

### Quail source and breeding

100 birds at the age of 10 days were obtained from the College of Agriculture and Forestry, Department of Animal Resources, University of Mosul. They were distributed into five groups in wooden cages. Each cage included 20 birds. The cages were provided with light for 24 hours and spread with sawdust. The birds were left for

five days for the purpose of conditioning. It was treated under field conditions and the birds were fed on a starter feed from the Erbil Feed company for Feed Industry in Iraq-Erbil, with a protein concentration of 17%.

### Experiment design

Silver NPs were used in this study at a size of 100 nm, obtained from the German company Sigma-aldrich Iraqi that a purity of 99.5%, Pcod: 1001151786, concentrations (1 and 2.5) ppm were used that were dosed orally for 30 days with AgNPs (5 and 10) ppm were dosed for 3 days only, in addition to the control group that was dosed for 30 days with distilled water. Birds were dosed orally with 0.5 ml in the morning for 30 days. The birds were explained, and the studied organ, the liver, was taken. And fixed with formalin at a concentration of 10% [6]. The tissue sections were prepared and using hematoxylin and eosin stains based on the method.

## 3. Results

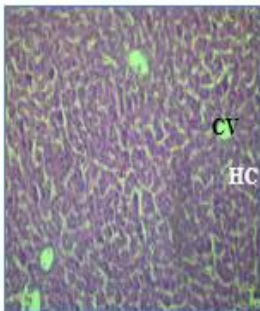
### Histopathological changes after 10 days

The results of the current study showed that the liver tissue of the quail of the control group consisted of the central vein and hepatocytes [Picture \(1\)](#). On the microscopic examination of the tissue sections of the groups with silver nanoparticles 10 days after the dosing. It was observed that histological changes occurred, which were vasodilation of hepatic sinusoids, the appearance of inflammatory focal, the thickening of blood vessels [Picture \(2\)](#). At 2.5 ppm noted focal of monocytic lymphocytes and the thickening of blood vessels [Picture \(3\)](#). The effect of nanosilver at 5 ppm after treatment for 3 days was more Ferocity as it was noted that necrosis focal and blood vessel congestion [Picture\(4A\)](#), And the observation of the severity of hemorrhage between

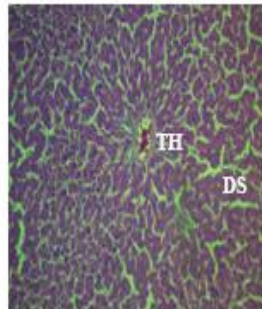
hepatic cell [Picture\(4B\)](#). As for the 10 ppm the tissue sections of the liver showed the presence of necrosis focal [Picture \(5\)](#).

### Histopathological changes after 30 days

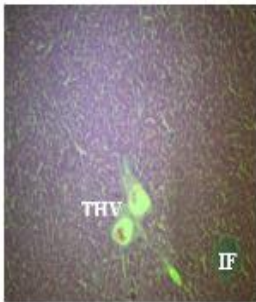
The results of the treatment after 30 days showed the histological structure of the liver of the control group [Picture \(6\)](#). AgNPs showed histopathological effect in a fatty change of hepatocytes at 1 ppm [picture \(7\)](#). While we find at 2.5 ppm an increase in inflammatory focal in the [Picture \(8\)](#). Also both 5 and 10 ppm showed histological changes as vascular congestion, fatty change of hepatocytes and Hemorrhage ([Picture 9-A, B](#)). [Picture-10](#) showed Hemorrhage, congestion, thickening of the hepatic portal artery and a Hyperplasia of bile canaliculi.



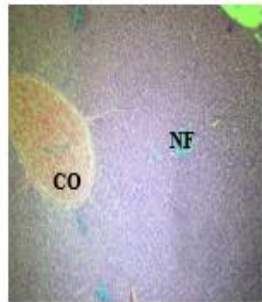
Photomicrography (1) section of liver control group showed central vein(CV), hepatic cell (hc), hematoxylin and eosin stains 100x.



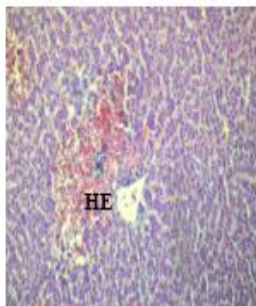
Photomicrography (2) section of liver concentration (1ppm) showed Thickness vesseal (Thv), Dilation sinusoids (Ds), stains H&E, 100X.



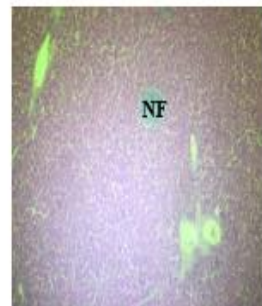
Photomicrography (3) section of liver concentration (2.5ppm) showed Thickness vesseal (Thv), inflammatory focal(IF), H&E, 40X.



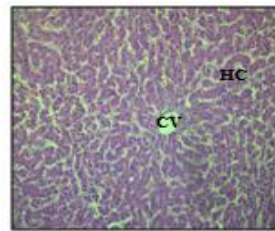
Photomicrography (4-A) section of liver concentration (5ppm) showed Congestion (CO), Necrosis focal (NF), H&E, 40X.



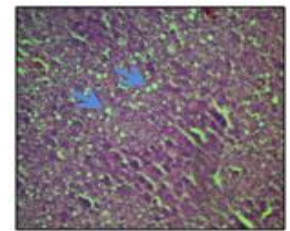
Photomicrography (4-B) section of liver concentration (5ppm) showed Hemorrhage (HE), H&E, 100X.



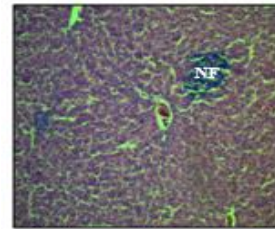
Photomicrography (5) section of liver concentration (10 ppm) showed Necrosis focal (NF), H&E, 10X.



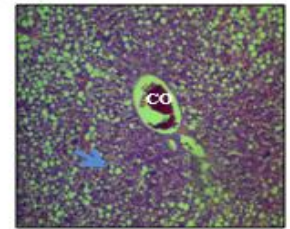
Photomicrography (6) section of liver (5th group) showed Central vein(CV), hepatic cell (hc), H&E, 100X.



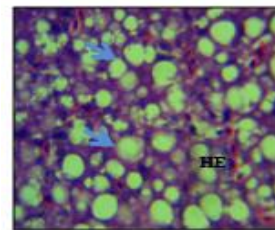
Photomicrography (7) section of liver concentration (1ppm), showed Fatty cytoplasmic vacuolar degeneration (→) H&E, 100X.



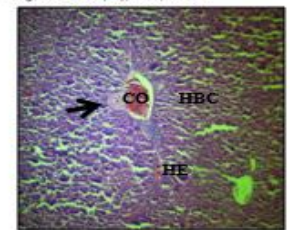
Photomicrography (8) section of liver concentration (2.5 ppm) showed Necrosis focal (NF), H&E, 100X.



Photomicrography (9-A) section of liver concentration(5ppm) showed Congestion (CO) and Fatty cytoplasmic vacuolar degeneration (→), H&E, 100X.



Photomicrography (9-B) section of liver concentration (5ppm) showed Hemorrhage (HE), Fatty cytoplasmic vacuolar degeneration (→), H&E, 400X.



Photomicrography (10) section of liver concentration (10ppm) showed Congestion (CO), Hemorrhage (HE), increased thickness the hepatic portal artery (→), Hyperplasia of bile canaliculi

## 4. Discussion

The results showed that after 10 days of dosing, silver NPs had many pathological lesions on the liver of birds, which showed the expansion of hepatic sinusoids, the presence of necrosis focal, congestion of blood vessels and the presence of hemorrhage between hepatocytes. Sinusoidal expansion returns due to the effect of AgNPs on the functions of mitochondria and the incitation of atrophy of those cells [7]. The researchers [8] also found that the nanoparticles in the liver of albino rats showed a constriction of the hepatic sinusoids and this does not coincide the results of the current study. As well as, it showed which is due to the property possessed by silver nanoparticles and their chemical activity as they have the ability to overcome the barrier of the endothelial layer to enter the peripheral tissues of the vessels in the liver which leads to immune reactions and results in peripheral inflammation in the organs [9]. This result is in contract with the study that was conducted on fish *Clarias gariepinus* liver at a of 100 µg /L. and this does not correspond the findings of the study that was conducted using different concentrations of NPs on the liver of broiler chickens the results showed that AgNPs had no toxic effect on cellular changes in liver [10]. The congestion of the blood vessel is due to the histological change and the occurrence of its expansion [11]. These results agree with the findings [12] who observed the congestion of the blood vessels in the liver of *Oryzias latipes* after exposure to nanoparticles. As [Kim et al. \[11\]](#) showed the cause of hemorrhage, which is due to the acute effect of silver NPs on the walls of blood vessels,

causing their expansion, which leads to the infiltration of red blood cells outside the blood vessel, thus penetrating the spaces between the liver tissue. indicated that the occurrence of thickening indicates an increase in the number of fibers as a result of abnormal tissue growth. After 30 days of treatment, fatty change of hepatocytes appeared, increased necrosis focal, vascular congestion, hemorrhage and increased thickness of the hepatic portal artery and hyperplasia of bile canaliculi. The study conducted by the researchers [13] clarified on the liver of mice treated with 10-200 µg /ml of NPs it showed an increase in inflammatory focal in addition to oxidative stress and cellular programmed death. The researchers [14] evaluated the effects of AgNPs on the liver of rats when it was treated with 200 mg/kg of Silver NPs. A study was conducted on the liver of Japanese quail treated with nanosilver orally 12 mg/L which indicated the occurrence of oxidative stress as a result of the accumulated silver in the liver [15]. Faedmaleki et al. [16] evaluated the effects of silver on liver cells of mice and they were treated with 1-10 ppm, and the results showed the occurrence of oxidative stress, she also indicated that these particles had an inhibitory effect on the growth of cancer cells, and this is not consistent with our results. The size of nanoparticles plays an important role in their uptake by cells [17]. The liver is the most affected by toxic substances, because it has a tissue characteristic that the rest of the body lacks, which is the lining of the blood sinusoids that contain holes with a size of (50-100 nm), so these particles have the great ability to penetrate the endothelial membrane, causing damage to the liver tissue [18]. We suggest doing more studies on the effect of silver nanoparticles on other organs such as the brain and intestines.

## 5. Conclusions

Silver nanoparticles showed histological changes which were more sever at high concentrations as it showed vascular congestion, Hepatic cell damage, severe hemorrhage, hepatic portal artery thickness and a hypetplasia of bile canaliculi we conclude from this study that nanosilver has toxic effects on the liver of quail Japanese.

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## References

- Saleh M. Nanotechnology and a new scientific era. King Fahd National Library Riyadh: 246pp. 2015.
- AbdAlaziz HR, Albaker AA. Histological Effect of Zinc Oxide Nanoparticles on Kidney of Japanese Quail Coturnix coturnixn. Clinical Schizophrenia & Related Psychoses. 2021.
- Handy RD, Owen R, Valsami-Jones E. The ecotoxicology of nanoparticles and nanomaterials: current status, knowledge gaps, challenges, and future needs. Ecotoxicology. 2008;17(5):315-25. <https://doi.org/10.1007/s10646-008-0206-0>
- Hondroulis E, Nelson J, Li C-z. Biomarker analysis for nanotoxicology. In: Biomarkers in Toxicology. Elsevier, 2014. p. 689-95. <https://doi.org/10.1016/B978-0-12-404630-6.00040-3>
- Wilkinson N, Hughes RJ, Aspden WJ, Chapman J, Moore RJ, Stanley D. The gastrointestinal tract microbiota of the Japanese quail, Coturnix japonica. Applied Microbiology and Biotechnology. 2016;100(9):4201-9. <https://doi.org/10.1007/s00253-015-7280-z>
- Suvarna KS, Layton C, Bancroft JD. Bancroft's theory and practice of histological techniques E-Book. Elsevier health sciences, 2018.
- Ji JH, Jung JH, Kim SS, Yoon J-U, Park JD, Choi BS, Chung YH, Kwon IH, Jeong J, Han BS. Twenty-eight-day inhalation toxicity study of silver nanoparticles in Sprague-Dawley rats. Inhalation toxicology. 2007;19(10):857-71. <https://doi.org/10.1080/08958370701432108>
- Sarhan OMM, Hussein RM. Effects of intraperitoneally injected silver nanoparticles on histological structures and blood parameters in the albino rat. International journal of nanomedicine. 2014;9:1505. <https://doi.org/10.2147%2FIJN.S56729>
- Guo H, Zhang J, Boudreau M, Meng J, Yin J-j, Liu J, Xu H. Intravenous administration of silver nanoparticles causes organ toxicity through intracellular ROS-related loss of inter-endothelial junction. Particle and fibre toxicology. 2015;13(1):1-13. <https://doi.org/10.1186/s12989-016-0133-9>
- Tavakoli R, Hashemi S, Davoodi D, Jafari Y, Hassani S. Histopathologic investigation of liver and kidney tissues in broiler chickens fed silver nanoparticles coated on zeolite. Journal of Animal Science Research. 2020;30(2):15-23. <https://doi.org/10.22034/as.2020.11450>
- Kim YS, Song MY, Park JD, Song KS, Ryu HR, Chung YH, Chang HK, Lee JH, Oh KH, Kelman BJ. Subchronic oral toxicity of silver nanoparticles. Particle and fibre toxicology. 2010;7(1):1-11. <https://doi.org/10.1186/1743-8977-7-20>
- Wu Y, Zhou Q. Silver nanoparticles cause oxidative damage and histological changes in medaka (Oryzias latipes) after 14 days of exposure. Environmental Toxicology and Chemistry. 2013;32(1):165-73. <https://doi.org/10.1002/etc.2038>
- Heydarnejad MS, Yarmohammadi-Samani P, Mobini Dehkordi M, Shadkhast M, Rahnama S. Histopathological effects of nanosilver (Ag-NPs) in liver after dermal exposure during wound healing. Nanomedicine Journal. 2014;1(3):191-7. Available from: <http://eprints.mums.ac.ir/5875/>
- Hamad SM, Shnawa BH, Jalil PJ, Ahmed MH. Assessment of the Therapeutic Efficacy of Silver Nanoparticles against Secondary Cystic Echinococcosis in BALB/c Mice. Surfaces. 2022;5(1):91-112. <https://doi.org/10.3390/surfaces5010004>
- Rezaei A, Farzinpour A, Vaziry A, Jalili A. Effects of silver nanoparticles on hematological parameters and hepatorenal functions in laying Japanese Quails. Biological trace element research. 2018;185(2):475-85.

<https://doi.org/10.1007/s12011-018-1267-4>

16. Faedmaleki F, Shirazi FH, Salarian A-A, Ashtiani HA, Rastegar H. Toxicity effect of silver nanoparticles on mice liver primary cell culture and HepG2 cell line. Iranian journal of pharmaceutical research: IJPR. 2014;13(1):235. Available from:

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3985257/>

17. Kettler K, Giannakou C, de Jong WH, Hendriks AJ, Krystek P. Uptake of silver nanoparticles by monocytic

THP-1 cells depends on particle size and presence of serum proteins. Journal of Nanoparticle Research. 2016;18(9):1-9. <https://doi.org/10.1007/s11051-016-3595-7>

18. Loghman A, Iraj SH, Naghi DA, Pejman M. Histopathologic and apoptotic effect of nanosilver in liver of broiler chickens. African Journal of Biotechnology. 2012;11(22):6207-11.

<https://doi.org/10.5897/AJB11.1768>