Coated and Uncoated Magnetic Iron Oxide Nanoparticles: Toxicity Assessment and Properties of Non Accumulation in Biological Tissues

Murtadha Aabdulhasan Aldhalemi[1], Azhar Jawad Shanshool[2], Ali A. Taha[3]

[1]*Department of Food Science, University of Kufa, Kufa, Iraq.*
[2]*Department of Food Science, University of Baghdad, Baghdad, Iraq.*
[3]*Applied Sciences Department, University of Technology, Baghdad, Iraq*

KEYWORDS
Magnetic Iron Oxide Nanoparticles; Coated Nanoparticles; liver; kidneys; Rat testicles ;Arab Gum; Nanomaterials

ABSTRACT
Background: There are an increasing number of studies on magnetic nanofeeders with multiple applications in agriculture and food processing. Among the magnetic separation oxides, iron oxide has emerged as an indispensable tool in nanoscale differentiation, especially bio-nanoscale. This is attributed to the characteristics of such as size, shape, magnetism and coexistence. Aim of the study: The aim of this study feeding experiment is to prove the non-toxicity of magnetic nano-iron oxides, whether coated when dosed with distilled water or uncoated in milk or distilled water. Subject, materials and methods: Materials: magnetic iron oxide nanoparticles was obtained from SkySpring Nanomaterials (U.S.A.) at a size of 30 nm. Acacia gum (Gum Arabic) is prepared from Thomas Baker (Indian), Sodium hydroxide from Riedel-de Haën (Germany), Potassium phosphate dibasic from CDH (Indian), Potassium phosphate mono basic from HIMEDIA (Indian),Potassium bromide from AVONCHEM(UK), MNPs are characterized by various tools to examine their physicochemical properties. The size of NPs plays a major role in exhibiting different physicochemical properties, and even a small difference in their nanoscale dimensions can alter their properties. Some of the instruments used for their characterization are Atomic Force Microscopy (AFM), Energy Dispersive X-ray Diffraction (EDXD), Field-Emission Scanning Electron Microscopy (FE-SEM), Fourier Transform Infrared Spectrometer. Results: After the completion of the experimental period for this study (28 days), the laboratory animals were sacrificed using ether anesthesia. The animals were then dissected using dissection tools to remove these organs. The organs were weighed using a sensitive balance after being washed with 0.9% normal saline solution and dried with filter paper [1][2]. The liver, kidneysamples were collected in pre-labeled plastic containers and preserved in 10% formalin until histological sectioning could be performed. Tissue sections were performed for all organs - liver, kidney and they were placed under a microscope. The results showed no damage to these organs. Conclusion: The findings suggest that both arabic gum-coated and milk-coated magnetic iron oxide nanoparticles, at the tested doses and treatment durations, did not induce observable tissue damage or pathological alterations in the kidneys or liver of the treated rats.

1. Introduction

Nanoparticles (NPs) that possess magnetic properties are referred to as magnetic nanoparticles (MNPs) and can be manipulated by external magnetic fields for a myriad of applications [3–5]. MNPs have several other technological applications in biomedicine such as magnetic resonance imaging, drug delivery and magnetic hyperthermia [6–8]. Magnetic properties of nanoparticles (NPs) are dominated by two main features [9–11]. finite-size effects (single-domain, multi-domain structures and quantum confinement) and surface effects, which results from the symmetry breaking of the crystal structure at the surface of the particle, oxidation, dangling bonds [12,13]. Surface effects become significant as the particle size decreases because the ratio of the number of surface atoms to the core atoms increases [11][14]. It is well established that several magnetic properties such as magnetic anisotropy, magnetic moment per atom, Curie temperature, and the coercivity field of NPs can be different than those of a bulk material [15–17]. In most medical applications, the preferred size of the nanoparticles is typically around 10–50 nm[18–21] Pure iron nanoparticles can be synthesized, but their sensitivity to oxidation due to strong dipole-dipole attractive interaction and surface reactivity, is a major drawback for biomedical applications. Depending on the application, magnetic nanoparticles may be linked into larger conglomerates to increase the overall magnetic moment. Iron nanoparticles coated and stabilized with coverages and consequently have been found to be quite thermodynamically stable under physiological conditions without any toxic effects on the cell[22–24] Stable Enzymatic Nanoparticles from Nucleases, Proteases, Lipase and Antioxidant Proteins with Substrate-Binding and Catalytic Properties.Limited membrane permeability and

biodegradation hamper the intracellular delivery of the free natural or recombinant enzymes necessary for compensatory therapy. Nanoparticles (NP) provide relative protein stability and unspecific endocytosis-mediated cellular uptake[25–27]. Therefore, this study aims to demonstrate whether the body can get rid of magnetic nanoparticles, whether coated or uncoated, when administered with distilled water or milk[9][28].

2. Materials and Methods

Materials: magnetic iron oxide nanoparticles was obtained from SkySpring Nanomaterials (U.S.A.) at a size of 30 nm. Acacia gum (Gum Arabic) is prepared from Thomas Baker (Indian), Sodium hydroxide from Riedel-de Haën (Germany), Potassium phosphate dibasic from CDH (Indian), Potassium phosphate mono basic from HIMEDIA (Indian), Potassium bromide from AVONCHEM(UK),

Characterization of MNPs

MNPs are characterized by various tools to examine their physicochemical properties. The size of NPs plays a major role in exhibiting different physicochemical properties, and even a small difference in their nanoscale dimensions can alter their properties. Some of the instruments used for their characterization are Atomic Force Microscopy (AFM), X-ray Diffraction (X-ray), Field-Emission Scanning Electron Microscopy (FE-SEM), Fourier Transform Infrared Spectrometer (FT-IR)[29].

Surface Modification of MNPs with GA

The surface of the Fe3O4 nanoparticles was coated with gum arabic by mixing 0.5 g of nanoparticles with 50 ml of GA solution (5 mg. L-1). Then, the mixture was sonicated for 30 minutes at room temperature. The gum arabic coated magnetic nanoparticles that formed were recovered from the reaction solution by placing a magnet under the bottle, followed by washing the magnetic nanoparticles with distilled water several times. Finally, they were dried in an oven at 40°C for 24 hours[18][30].

The Nutritional Experiment

Preparation of Laboratory Animals and Conducting the Experiment:

The nutritional experiment was conducted on male Albino rats obtained from the animal house at the University of Kufa, aged 4-5 weeks, with an average weight ranging between 110-120 grams. A total of 28 rats were divided into four groups, ensuring that their weights were as equal as possible. Each group consisted of seven individuals. The rats were housed in locally manufactured plastic cages with dimensions of 15x20x27 cm (height-width-depth, respectively), which are similar to the dimensions mentioned in the references. Each cage contained one rat. The experiment was carried out under controlled standard conditions, with a temperature of 22 ± 2°C, relative humidity of 55 ± 5%, and a 12-hour light-dark cycle. Food and distilled water were provided ad libitum according to the needs of the animals, and the bedding was changed every two days[19][31].

Table 1: illustrates the division of the experimental animal groups used in the nutritional experiment and the treatments administered to them:

No.	Symbol	Treatment
1	CO	Negative control group, administered distilled water only
2	A1	Positive control group, administered MNPs with distilled water
3	A2	Positive control group, administered GA@MNPs with distilled water

4	A3	Positive control group, administered MNPs in skim milk

The treatments were administered orally using a gavage, a tool specifically designed for rats. Histological Study of Experimental Animal Organs (Liver, Kidney), Collection of Liver, Kidney Samples. After the completion of the experimental period for this study (28 days), the laboratory animals were sacrificed using ether anesthesia. The animals were then dissected using dissection tools to remove these organs[32,33] The liver, kidney samples were collected in pre-labeled plastic containers and preserved in 10% formalin until histological sectioning could be performed.To perform histological detection of magnetic nanoparticles, the target organs in this study, preserved in formalin solution, were extracted and washed with water. They then underwent a series of processes based on previous studies described by Lee et al.[34,35].

Samples were sectioned and tissue sections were stained. Tissue sections were photographed using a MEIJI light microscope, with results recorded at 100X and 400X magnification. The microscope was equipped with a Canon high-resolution digital camera.

2. Results and Discussion

Field-Emission Scanning Electron Microscopy (FE-SEM)

FE-SEM is a technique that visualizes the shape and size of nanoparticles [36,37]. The images of the magnetic iron oxide nanoparticles (MNP) in Figure 1 reveal a homogeneous structure with spherical shapes.

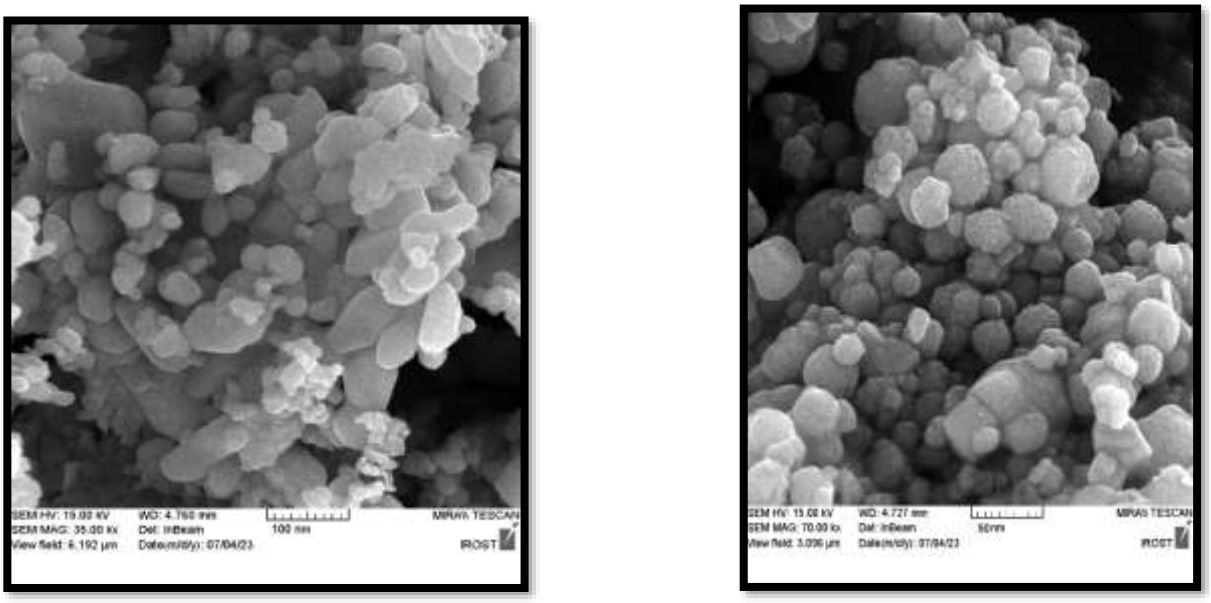

Figure (1): The Shape and size of magnetic iron oxide nanoparticles (MNPs) using Field-Emission Scanning Electron Microscopy (FE-SEM) analysis .

The surface change of magnetic nanoparticles after coating with gum arabic GA was studied using FESEM analysis. From Figure (2), it is clear showing how much the average of the total diameter of the GA@MNP, as it increased after being coated with gum arabic compared to the uncoated one, and this is consistent with

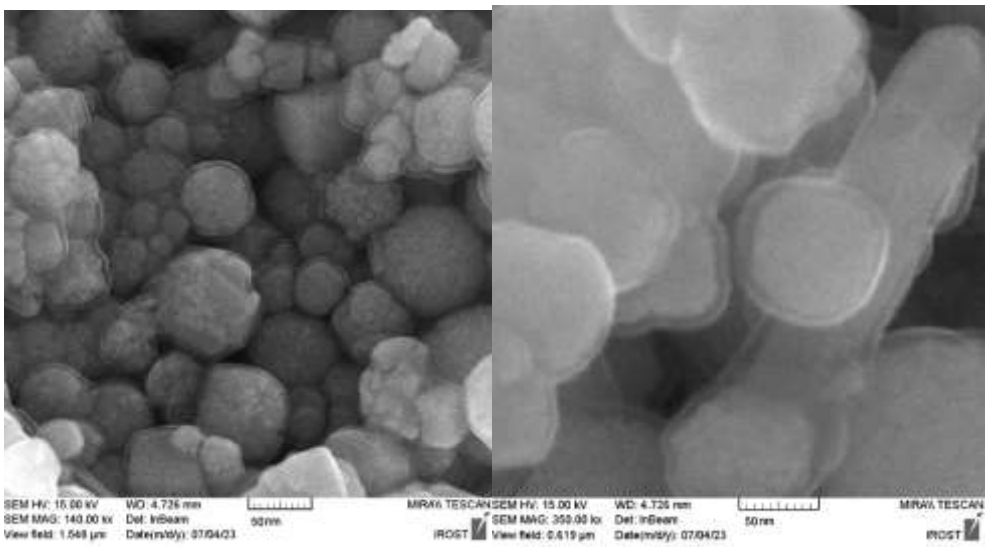

Figure (2): Shape and size of magnetic iron oxide nanoparticles (MNPs) after coating with gum arabic GA (GA@MNP using Field-Emission Scanning Electron Microscopy (FE-SEM) analysis.

The surface change of magnetic nanoparticles after coating them with gum Arabic (GA) was studied using FE-SEM analysis. Figure (2) shows the extent of increase in the average total diameter of magnetic nanoparticles (GA@MNP) after coating them with gum Arabic compared to the uncoated ones. Hassan Hussein *et al* [38].

Fourier-transform infrared spectroscopy (FTIR) analysis:

(FTIR) Fourier-transform infrared spectroscopy analysis is a suitable method to determine the functional groups present in the organic compounds in the sample[11,18].

FTIR analysis was performed on magnetic nanoparticles of iron oxides Fe3O4 to determine the functional groups and bonds present in the sample. For this purpose, FTIR spectra of the nanoparticles were obtained. Using the KBr-Pellet method. Figure 3 (A) shows the FTIR of MNP particles, gum arabic, MNP coated with gum arabic, GA-MNP. It is shown in Figure 3 that the wave number was from cm-1 005 to cm-1 4000. Accordingly, the characteristic peak of the spectrum for magnetic nano-oxides of iron, i.e. Fe-O, occurs at cm-1 586, which is the bond for the magnetite (MNP) [19,20].

In the FTIR spectrum of gum arabic, Figure 3 (A), the infrared characterization feature indicates the presence of the absorption band that appears at 1030 cm^{-1}, which is attributed to the stretching vibration of carbon dioxide, while the absorption band at 1610 cm^{-1} is due to the stretching vibration. C=O in gum arabic. The absorption band that appears at 2922 cm^{-1} is attributed to C-H stretching in the gum arabic structure. It is noted from this that the distinct absorption bands attributed to gum arabic in the FTIR spectrum of GA@MNP [39–41] Therefore, a conclusion can be drawn that GA has succeeded in coating magnetic nanoparticles.

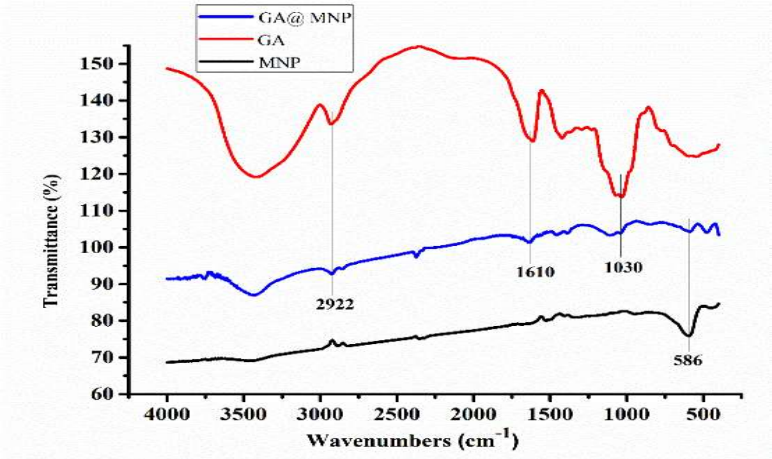

Figure 3: Fourier-transform infrared spectroscopy (FTIR) spectrum of MNP, gum arabic, gum arabic-coated MNP GA@MNP

XRD analysis

Figure 4 shows the XRD patterns of Fe3O4 MNPs, which agree well with the crystal structure pattern of magnetite [42,43]. Characteristic peaks could be identified at 2θ values of 30.20, 35.6, 43.25, 53.75, and 57.15, which corresponded to the crystal planes (114), (322), (82), (48), (121) of unmodified Fe3O4. These results are in line with the findings of Dawn et al[44]). After encapsulating the MNPs using gum arabic and conducting an XRD spectrum, which indicates that the surface functionalization did not change the crystalline nature, and this is consistent with what was indicated by Ali et al [29].

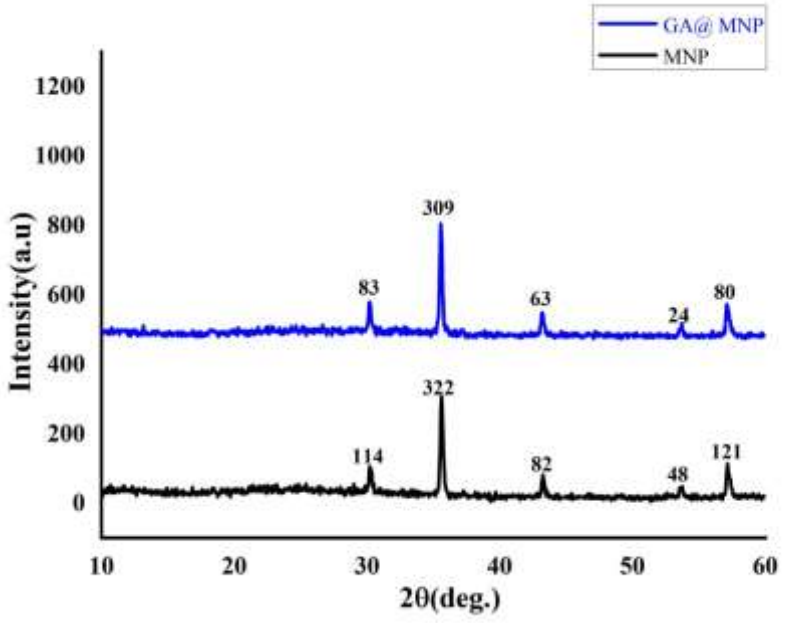

Figure 4: XRD analysis patterns of Fe3O4 MNPs

Study Results

The results of this study demonstrated the successful coating of MNPs with Arabic gum, attributed to the biocompatibility of Arabic gum, which is one of its advantages. This process is environmentally friendly as it prevents the oxidation of MNPs. The GA@MNPs meet the requirements for biological applications due to their non-aggregation properties, protection and stabilization of MNPs, and lack

of toxicity. Furthermore, analyses using FE-SEM (Field Emission Scanning Electron Microscopy) and AFM (Atomic Force Microscopy) indicated a change in the external appearance of the MNPs as a result of the coating.

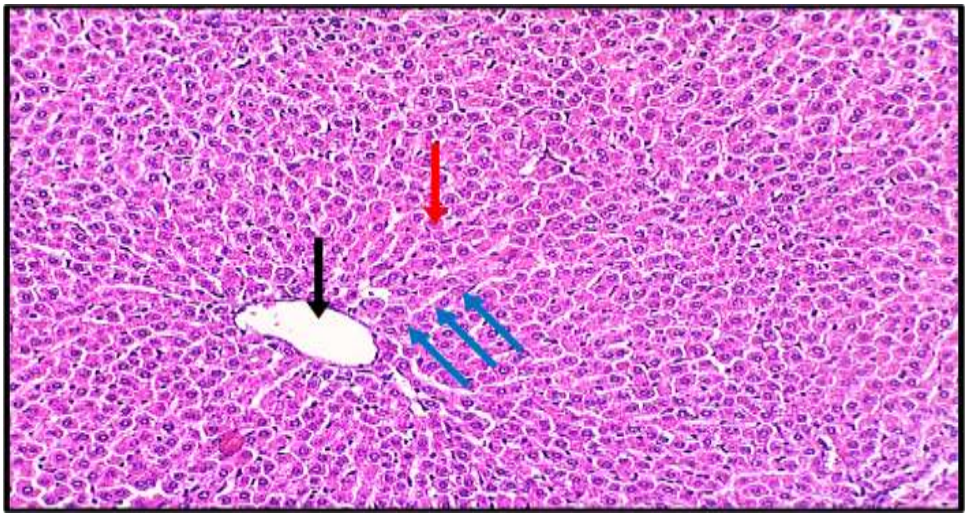

Figure 5: Histological Section of the Liver in the Negative Control Group Rats

The liver tissue shows the normal structure without any detectable pathological lesions.

- The black arrow indicates the central vein in the hepatic portal area.
- The red arrow points to normal hepatocytes in the liver tissue.
- The blue arrows highlight the hepatic sinusoids, which are spaces between liver cells where blood flows.

The tissue was stained using Hematoxylin and Eosin stains. The histological section was photographed using a light microscope and a digital camera at 10x magnification.

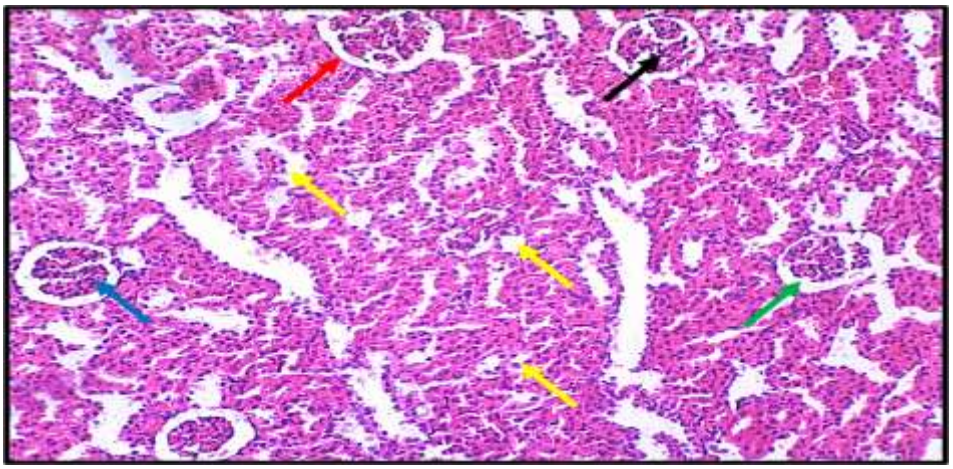

Figure 6: Histological Section of the Kidney in the Negative Control Group Rats

The kidney tissue shows the normal structure without any detectable pathological lesions.

- The black arrow indicates the normal renal corpuscle.
- The red arrow points to the renal capsule, which surrounds the renal corpuscle.
- The blue arrow indicates the network of glomerular capillaries within the renal corpuscle.
- The green arrow shows the Bowman's space, where glomerular filtration occurs in the kidney.

- The yellow arrows highlight the renal tubules, which transport filtered waste products.

The tissue was stained using Hematoxylin and Eosin stains. The histological section was photographed using a light microscope and a digital camera at 10x magnification.

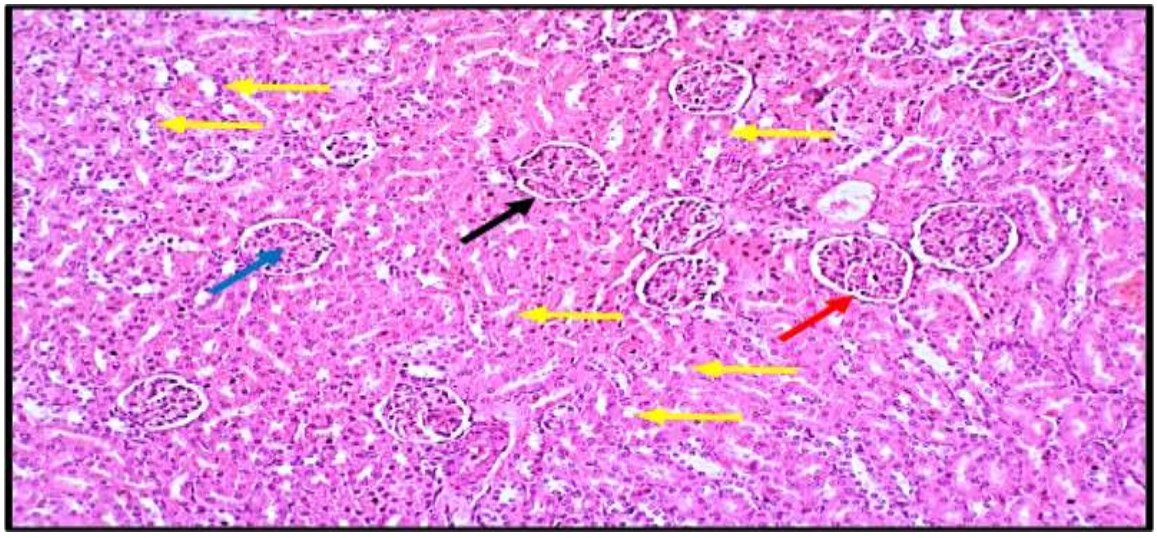

Figure 7: Histological section of the kidney in the negative control group rats

The kidney tissue shows the normal structure without any detectable pathological lesions.

- The black arrow indicates the normal renal corpuscle.
- The red arrow points to the renal capsule, which surrounds the renal corpuscle.
- The blue arrow indicates the network of glomerular capillaries within the renal corpuscle.
- The green arrow shows the Bowman's space, where glomerular filtration occurs in the kidney.
- The yellow arrows highlight the renal tubules, which transport filtered waste products.

The tissue was stained using Hematoxylin and Eosin stains. The histological section was photographed using a light microscope and a digital camera at 10x magnification.

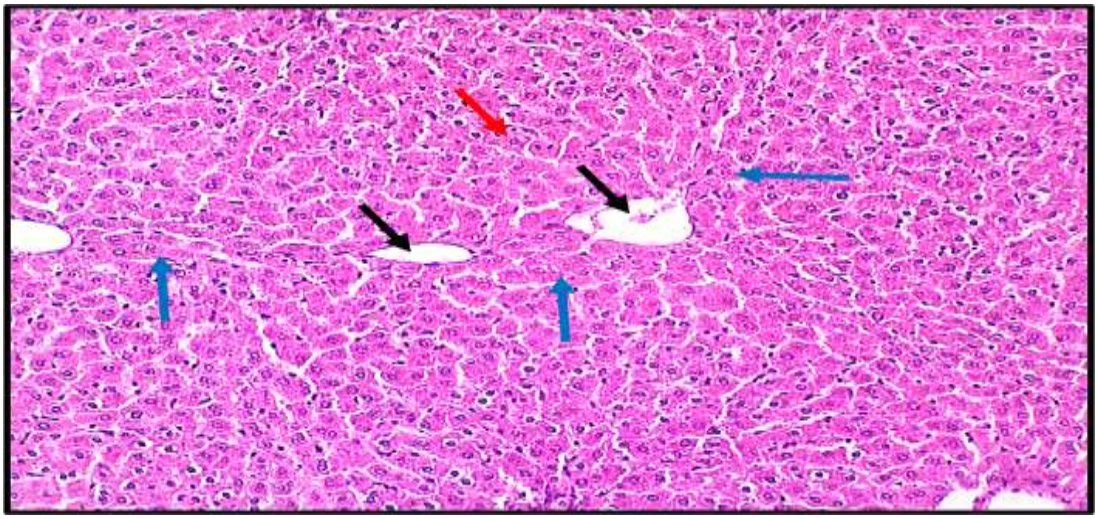

Figure 8: Histological Section of the Liver in Rats Treated with Magnetic Iron Oxide Nanoparticles (Dose: 5 mg/kg)

The liver tissue shows a normal structure without any detectable pathological lesions.

- The black arrows indicate the central vein in the portal area of the liver.

- The red arrow points to normal liver cells (hepatocytes) in the liver tissue.
- The blue arrows indicate the hepatic sinusoids, which are spaces lined by hepatocytes in the liver tissue.

The tissue was stained using Hematoxylin and Eosin stains. The histological section was photographed using a light microscope and a digital camera at 10x magnification.

Evaluation of the dose-dependent effect of magnetic iron oxide nanoparticles (Fe3O4 MNPs) on liver tissue indicated that administration of Fe3O4 at a dose of 5 mg/kg was not associated with pathological changes in the tissue. McClements et al (2017)[1]. Regarding oral administration, magnetic iron oxide nanoparticles did not accumulate in tissues or cause toxicity. the study presented by Parivar et al. (2016 Other studies have confirmed that oral administration of Fe3O4 did not cause accumulation of magnetic nano-iron oxides in tissues or cause toxicity [4].

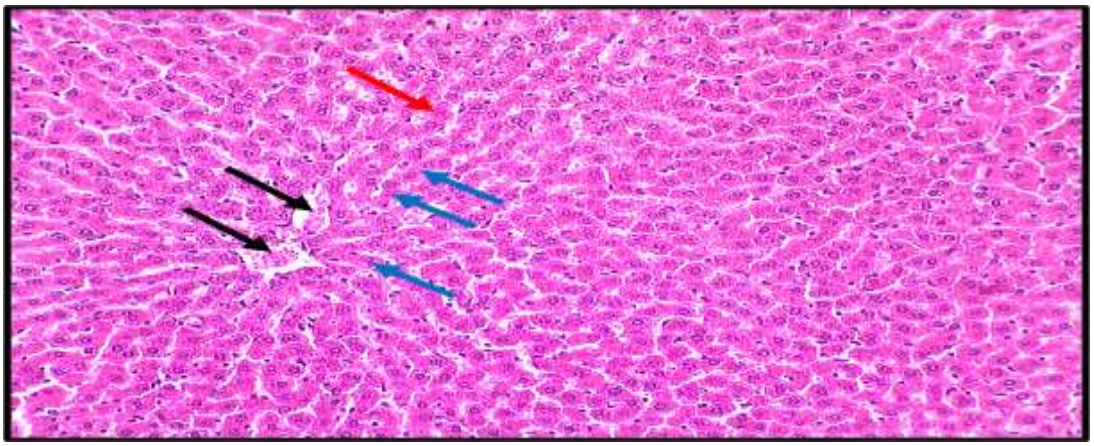

Figure 9: Histological Section of the Liver in Rats Treated with Arabi Gum-Coated Magnetic Iron Oxide Nanoparticles (Dose: 5 mg/kg)

The liver tissue exhibits a normal structure without any detectable pathological lesions.

- **Black arrows:** Central vein in the portal area of the liver.
- **Red arrow:** Normal hepatocytes (liver cells) within the liver tissue.
- **Blue arrows:** Hepatic sinusoids, which are spaces formed by aligned hepatocytes in the liver tissue.

The tissue was stained using Hematoxylin and Eosin stains. The histological section was captured using a light microscope and a digital camera at 10x magnification.

The figure(9) shows the effect of the dose of magnetic iron oxide nanoparticles coated with gum arabic (Fe3O4) GA@MNPs on liver tissues, as no pathological changes were observed in the tissues. Gum arabic acts as a protective layer around the magnetite particles, reducing their interaction with cells and tissues[45,46]. The mechanism by which gum arabic mitigates magnetite accumulation in the liver can be attributed to its physicochemical properties. Gum arabic forms a protective layer around magnetite nanoparticles, changing their surface charge and hydrophilicity. This modification enhances the dispersion of nanoparticles in biological fluids and facilitates their removal from the body through renal pathways, thus reducing the likelihood of their accumulation in the liver [47], it was confirmed that coating these particles prevents their aggregation and increases the body's ability to get rid of them.

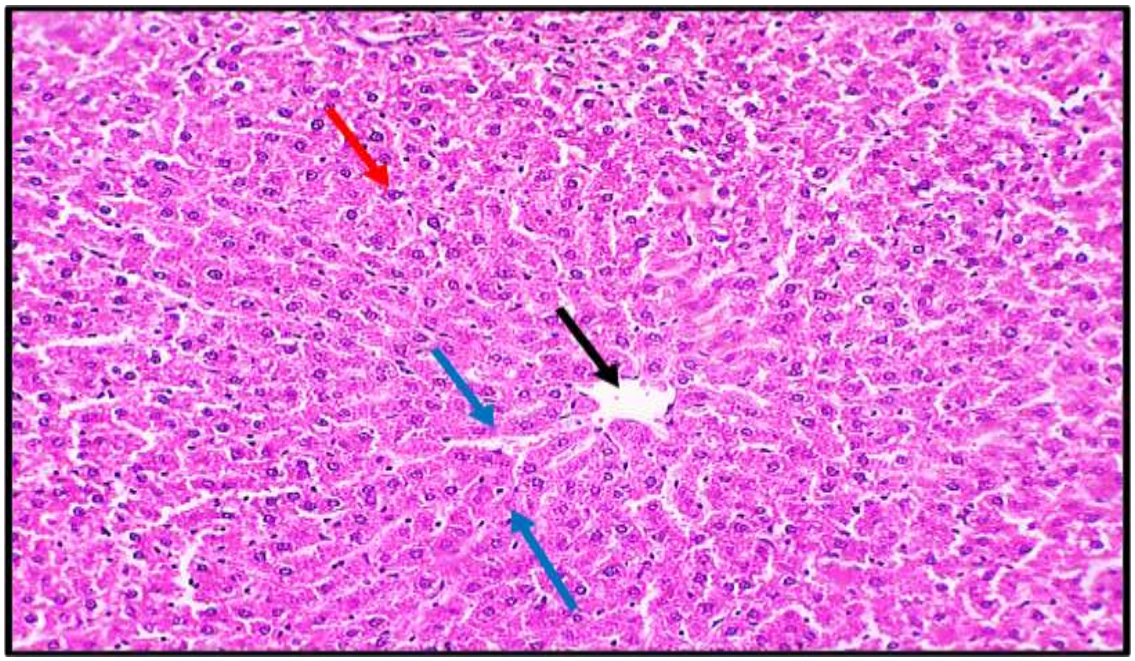

Figure 10: Histological Section of the Liver in Rats Treated with Magnetic Iron Oxide Nanoparticles Coated with Milk (Dose: 5 mg/kg)

The liver tissue displays a normal structure with no identifiable pathological abnormalities.

- **Black arrow:** Central vein in the portal area of the liver.
- **Red arrow:** Normal hepatocytes (liver cells) in the liver tissue.
- **Blue arrows:** Hepatic sinusoids, spaces formed by aligned hepatocytes in the liver tissue.

The tissue was stained with Hematoxylin and Eosin. The histological section was captured using a light microscope and a digital camera at 10x magnification.

Figure (10) shows that there is no damage to the liver tissue as a result of dosing magnetic nanoparticles at a dose of (5 mg/kg) with milk, as the latter's proteins work by adhering to the walls of these particles and forming an outer layer that prevents the aggregation of particles and works to disperse them, thus making them safe [48]. Also, the study conducted by Huang et al. [42], using milk casein attached to the walls of magnetic nano-iron oxides, delivered the drug safely orally. Other studies have also been conducted that prove the safety of these particles when dosed in milk, as the coating consisting of a layer of casein on the surface of the particles shows non-toxicity and biological compatibility, the coating consisting of milk casein gives the magnetic nanoparticles long-term stability. In another study, the issue was not limited to its non-toxicity, but rather went beyond that, as these particles were used with milk to treat iron deficiency anemia (IDA) in the body. Kumari et al[7].

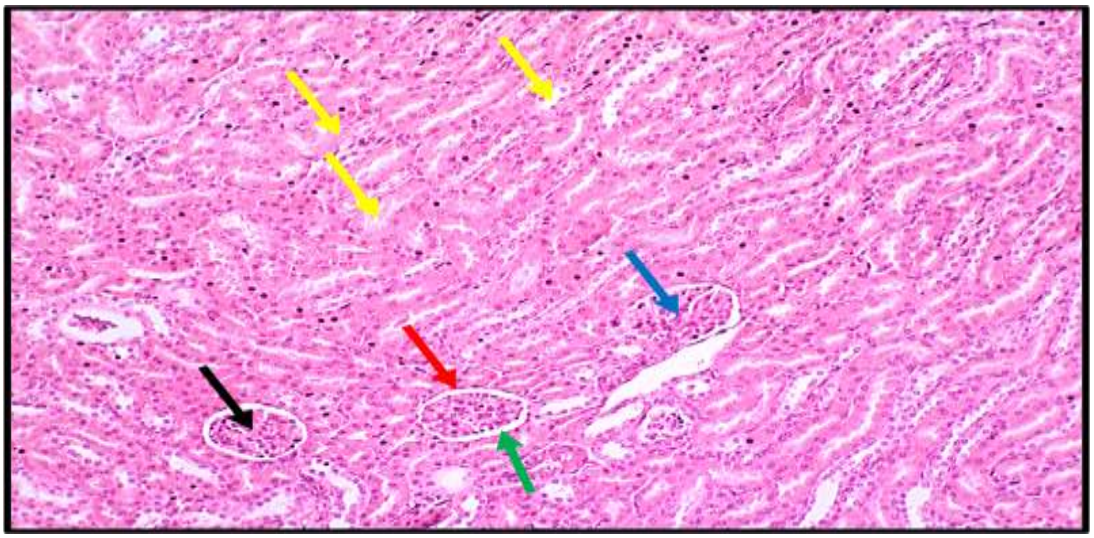

Figure 11: Histological Section of the Kidney in Rats Treated with Magnetic Iron Oxide Nanoparticles (Dose: 5 mg/kg)

The kidney tissue shows no detectable pathological lesions.

- **Black arrow:** Normal renal corpuscle.
- **Red arrow:** Capsule of the renal corpuscle.
- **Blue arrow:** Network of fine blood vessels (glomerulus) within the renal corpuscle.
- **Green arrow:** Bowman's capsule, through which renal filtration occurs.
- **Yellow arrows:** Renal tubules responsible for transporting filtered wastes.

The tissue was stained with Hematoxylin and Eosin. The histological section was captured using a light microscope and a digital camera at 10x magnification.

Figure (11) shows the kidney tissue of animals dosed with magnetite, and it was noted that there was no obvious damage to the kidney tissue. A study conducted by Madkor et al [45]. confirmed that lead toxicity on the kidneys was reduced using uncoated magnetic nanoparticles of iron oxides. Another study confirmed that these particles are safe and do not cause changes to the kidneys over time, As for the size, several studies have confirmed that the size of the magnetic nanoparticles targeted in this study can be eliminated by the kidneys [49].

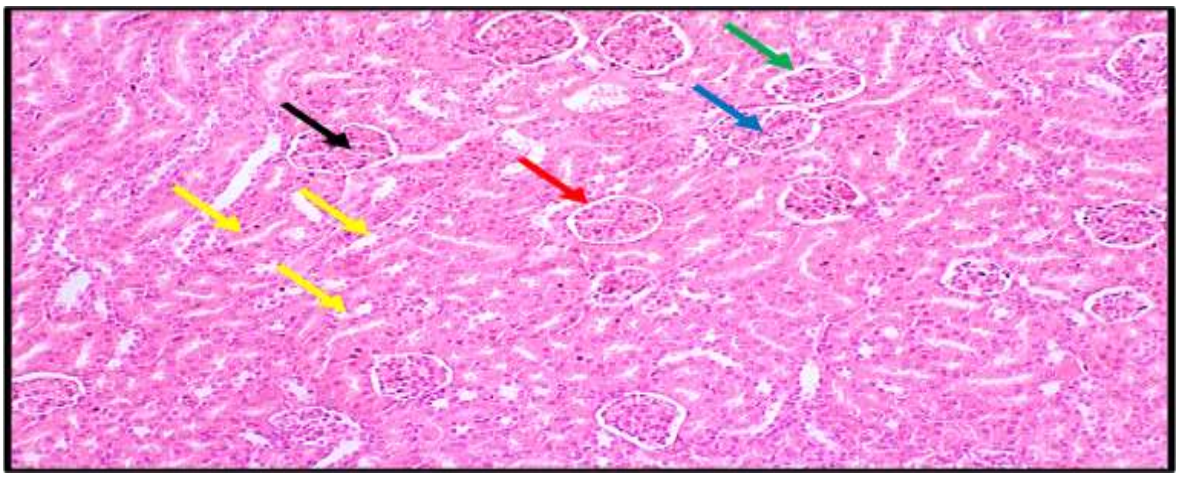

Figure 12: Histological Section of the Kidney in Rats Treated with Arabi Gum-Coated Magnetic Iron Oxide Nanoparticles (Dose: 5 mg/kg)

The kidney tissue exhibits a normal structure without any detectable pathological lesions.

- **Black arrow:** Normal renal corpuscle.
- **Red arrow:** Capsule of the renal corpuscle.
- **Blue arrow:** Network of fine blood vessels (glomerulus) within the renal corpuscle.
- **Green arrow:** Bowman's capsule, through which renal filtration occurs.
- **Yellow arrows:** Renal tubules responsible for transporting filtered wastes.

The tissue was stained with Hematoxylin and Eosin. The histological section was captured using a light microscope and a digital camera at 10x magnification.

Figure (12) shows the kidney tissue after dosing with magnetic nano-iron oxides coated with gum arabic. It was noted that there was no damage resulting from these nanoparticles, as the coating maintains the dispersion of the magnetite magnetic nanoparticles and prevents their aggregation and makes them safe, non-toxic, and does not cause any damage to the tissue[49]. Another study also showed that the process of coating with gum arabic eliminates the damage that may result from uncoated magnetic nanoparticles. It also confirmed that coating with gum arabic works to prevent the potential negative effects of these nanoparticles on tissues [47].

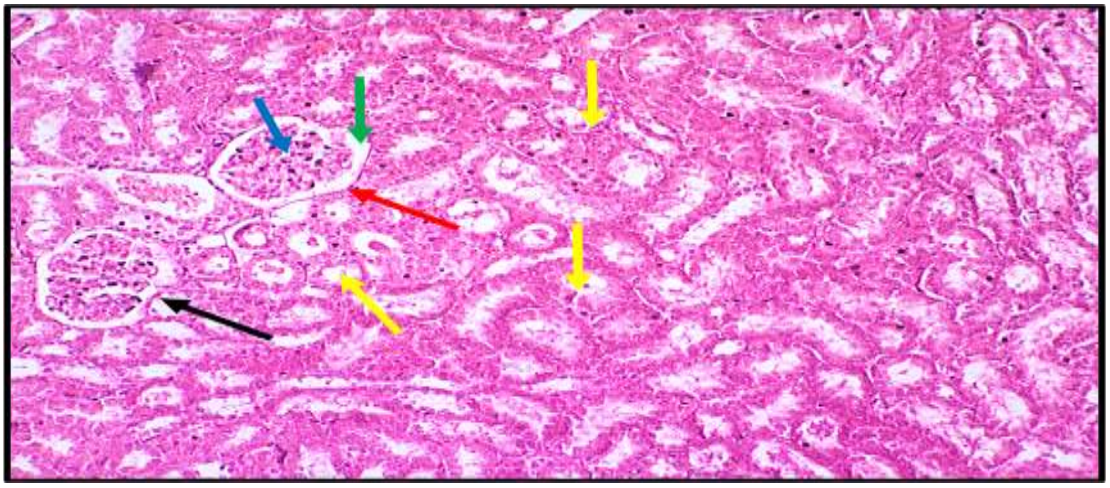

Figure 13: Histological Section of the Kidney in Rats Treated with Milk-Coated Magnetic Iron Oxide Nanoparticles (Dose: 5 mg/kg)

The kidney tissue exhibits a normal structure without any detectable pathological lesions.

- **Black arrow:** Normal renal corpuscle.
- **Red arrow:** Capsule of the renal corpuscle.
- **Blue arrow:** Network of fine blood vessels (glomerulus) within the renal corpuscle.
- **Green arrow:** Bowman's capsule, through which renal filtration occurs.
- **Yellow arrows:** Renal tubules responsible for transporting filtered wastes.

The tissue was stained with Hematoxylin and Eosin. The histological section was captured using a light microscope and a digital camera at 10x magnification.

Figure 13 shows that there was no harm to the magnetic nanoparticles that were dosed after mixing them with milk, as the milk proteins work to coat the magnetic nanoparticles, causing them to stabilize and not aggregate, i.e. maintaining their dispersion, thus being biocompatible [49,50].

3. Conclusion

In this study, two axes were accomplished: the first, magnetic iron oxide nanoparticles were coated with gum arabic, and the second axis was treating laboratory rats with magnetic iron oxide nanoparticles coated with gum arabic or milk, at specific doses and treatment periods. Histological examination of kidney and liver tissues showed no detectable pathological lesions in all treatment groups. Overall, these results indicate that the magnetic iron oxide nanoparticles coated with gum arabic and coated with milk, at the tested doses and treatment periods, did not cause significant tissue damage or pathological changes in the kidney or liver of treated rats.

Conflict interest

Neither the authors nor the researchers involved in this study have any financial or personal interests that could potentially lead to conflicts of interest. The research was conducted with a focus solely on contributing to the scientific knowledge and understanding within the field.

Funding

This entire study wasn’t funded.

Acknowledgement

The authors thank the Ministry of Science and Technology

Reference

[1] D.J. McClements, H. Xiao, Is nano safe in foods? Establishing the factors impacting the gastrointestinal fate and toxicity of organic and inorganic food-grade nanoparticles, Npj Sci. Food. 1 (2017) 6.

[2] Y.S. Pestovsky, A. Martínez-Antonio, The use of nanoparticles and nanoformulations in agriculture, J. Nanosci. Nanotechnol. 17 (2017) 8699–8730.

[3] M.D. Ruaa, S.A. Alsahib, I.F. Ascar, Synthesis, characterization and cyclazation of pyran using Ag2O nanoparticle from natural source “ginger,” Iraqi J. Agric. Sci. 52 (2021) 1171–1184.

[4] S. Chamorro, L. Gutiérrez, M.P. Vaquero, D. Verdoy, G. Salas, Y. Luengo, A. Brenes, F.J. Teran, Safety assessment of chronic oral exposure to iron oxide nanoparticles, Nanotechnology. 26 (2015) 205101.

[5] A.K. Al-Jubouri, N.H. Al-Saadi, M.A. Kadhim, Anti-inflammatory and anti-bacterial activity of copper nanoparticles synthesized from Myrtus communis leaves extract, Iraqi J. Agric. Sci. 53 (2022) 698–711.

[6] A.M. Al-Ghareebawi, B.N. Al-Okaily, O.M.S. Ibrahim, Characterization of zinc oxide nanoparticles synthesized by olea europaea leaves extract (part L), Iraqi J. Agric. Sci. 52 (2021) 580–588.

[7] R. Vakili-Ghartavol, A.A. Momtazi-Borojeni, Z. Vakili-Ghartavol, H.T. Aiyelabegan, M.R. Jaafari, S.M. Rezayat, S. Arbabi Bidgoli, Toxicity assessment of superparamagnetic iron oxide nanoparticles in different tissues, Artif. Cells, Nanomedicine, Biotechnol. 48 (2020) 443–451.

[8] M. Esmaili, H. Dezhampanah, M. Hadavi, Surface modification of super paramagnetic iron oxide nanoparticles via milk casein for potential use in biomedical areas, J. Biomol. Struct. Dyn. 39 (2021) 977–987.

[9] R.N. Suhail, S.A. Mustafa, E.E. Al-Obodi, Effeciency of silver nanoparticales as antibacterial against Aeromonas hydrophila isolated from infected common carp, Iraqi J. Agric. Sci. 53 (2022) 589–597.

[10] A. Ali, T. Shah, R. Ullah, P. Zhou, M. Guo, M. Ovais, Z. Tan, Y. Rui, Review on recent progress in magnetic nanoparticles: Synthesis, characterization, and diverse applications, Front. Chem. 9 (2021) 629054.

[11] M.T. Riwad, M.K. Alag, Role of nano and metallic boron foliar nutrition on water stress reducing in sweet corn yield and its components, Iraqi J. Agric. Sci. 54 (2023) 1421–1432.

[12] O.S. Hamid, S.S. Mahmood, THE SYNERGISTIC EFFECT OF GOLD NANOPARTICLE LOADED WITH CEFTAZIDIUM ANTIBIOTIC AGAINST MULTIDRUG ERSISTANCE PSEUDOMONAS AERUGINOSA, Iraqi J. Agric. Sci. 52 (2021) 828–835.

[13] Z.R.J. Al-Malikshah, I.J. Abdulrasool, Spraying nano chitosan loaded with NPK, nettel and green tea extracts as a tool for improvement potato productivity, Iraqi J. Agric. Sci. 55 (2024) 175–185.

[14] M.A.M. Al Rubaee, S.J. Hamodi, F.M. Hussein, RESPONSE OF PHYSIOLOGICAL AND HISTOLOGICAL TRAITS TO ENZYME SUPPLEMENTATION AND SOAKING OF DIETS CONTAINING CORN AND WHEAT OF THE LAYNING HENS, Iraqi J. Agric. Sci. 47 (2016).

[15] H.N. Mustafa, I. Al-Ogaidi, Efficacy of zinc sulfide-chitosan nanoparticles against bacterial diabetic wound infection, Iraqi J. Agric. Sci. 54 (2023) 1–17.

[16] K. Parivar, F.M. Fard, M. Bayat, S.M. Alavian, M. Motavaf, Evaluation of iron oxide nanoparticles toxicity on liver cells of BALB/c rats, Iran. Red Crescent Med. J. 18 (2016).

[17] A.K. Chhillar, J.S. Rana, Enzyme nanoparticles and their biosensing applications: a review, Anal. Biochem. 581 (2019) 113345.

[18] E. Alzahrani, Gum Arabic-coated magnetic nanoparticles for methylene blue removal, Int. J. Innov. Res. Sci. Eng. Technol. 3 (2014) 15118–15129.

[19] M. Fernández, M. López-Jurado, P. Aranda, G. Urbano, Nutritional assessment of raw and processed faba bean (Vicia faba L.) cultivar major in growing rats, J. Agric. Food Chem. 44 (1996) 2766–2772.

[20] S.A. Mohamed, M.H. Al-Harbi, Y.Q. Almulaiky, I.H. Ibrahim, R.M. El-Shishtawy, Immobilization of horseradish peroxidase on Fe3O4 magnetic nanoparticles, Electron. J. Biotechnol. 27 (2017) 84–90.

[21] R. Chapman, M.H. Stenzel, All wrapped up: stabilization of enzymes within single enzyme nanoparticles, J. Am. Chem. Soc. 141 (2019) 2754–2769.

[22] A. Kumari, A.K. Chauhan, Iron nanoparticles as a promising compound for food fortification in iron deficiency anemia: a review, J. Food Sci. Technol. 59 (2022) 3319–3335.

[23] S. Rathee, A. Ojha, A. Upadhyay, J. Xiao, V.K. Bajpai, S. Ali, S. Shukla, Biogenic engineered nanomaterials for enhancing bioavailability via developing nano-iron-fortified smart foods: advances, insight, and prospects of nanobionics in fortification of food, Food Funct. 14 (2023) 9083–9099.

[24] T. Kuang, Y. Liu, T. Gong, X. Peng, X. Hu, Z. Yu, Enzyme-responsive nanoparticles for anticancer drug delivery, Curr. Nanosci. 12 (2016) 38–46.

[25] J. Sangeetha, J. Philip, The interaction, stability and response to an external stimulus of iron oxide nanoparticle–casein nanocomplexes, Colloids Surfaces A Physicochem. Eng. Asp. 406 (2012) 52–60.

[26] R.S. Madkor, N. Abdelaleem, A. Abdeen, A.E. El-Mashed, S. Ibrahim, Ameliorative role of green synthesized iron oxide nanoparticles using Punica granatum extract against lead acetate hepato-renal toxicity in albino rat, Benha Vet. Med. J. 45 (2023) 11–17.

[27] M. Li, G. Zhao, W.-K. Su, Q. Shuai, Enzyme-responsive nanoparticles for anti-tumor drug delivery, Front. Chem. 8 (2020) 647.

[28] H.A. Jebur, J.M. Auda, Evalution of antimicrobial activity of partial purified bacteriocin from local isolate of Bacillus licheniforims HJ2020 MT192715. 1., Iraqi J. Agric. Sci. 51 (2020).

[29] I.H. Ali, M.Z. Bani-Fwaz, A.A. El-Zahhar, R. Marzouki, M. Jemmali, S.M. Ebraheem, Gum Arabic-magnetite nanocomposite as an eco-friendly adsorbent for removal of lead (II) ions from aqueous solutions: equilibrium, kinetic and thermodynamic studies, Separations. 8 (2021) 224.

[30] K.F.M. Horamani, A.O.I. Sarmamy, IMPACT OF SOIL STERILIZATION FROM DIFFERENT LOCATIONS, WHEAT SEEDS CLEANING AND THEIR INTERACTIONS ON WEED CONTROL, AND YIELD AND YIELD COMPONENTS, IRAQI J. Agric. Sci. 55 (2024) 894–904.

[31] M. Cayetano-Cruz, C.F. Coffeen, J. Valadez-García, C. Montiel, I. Bustos-Jaimes, Decoration of virus-like particles with an enzymatic activity of biomedical interest, Virus Res. 255 (2018) 1–9.

[32] M. Güneş, K. Aktaş, B. Yalçın, A.Y. Burgazlı, M. Asilturk, A.E. Ünşar, B. Kaya, In vivo assessment of the toxic impact of exposure to magnetic iron oxide nanoparticles (IONPs) using Drosophila melanogaster, Environ. Toxicol. Pharmacol. 107 (2024) 104412.

[33] O. V Morozova, N.A. Barinov, D. V Klinov, Stable enzymatic nanoparticles from nucleases, proteases, lipase and antioxidant proteins with substrate-binding and catalytic properties, Int. J. Mol. Sci. 24 (2023) 3043.

[34] E.K. Slewa, J. Azhar, Use nanotechnology in capsulation omega-3 fatty acid to improve its thermal stability and use it to enrich pasteurized milk, Pakistan J. Biotechnol. 15 (2018) 33–43.

[35] A.M.H.H. Al-Khafaji, K.D.H. Al-jubouri, DEVELOPMENTAL CONTROL OF SOME PHYSIOLOGICAL FACTORS ON REPRODUCTIVE BIOLOGY AND RUDIMENTARY EMBRYOS PHENOMENON IN CARROT SEEDS, IRAQI J. Agric. Sci. 55 (2024) 1038–1047.

[36] E. Mohammadi, D. Qujeq, H. Taheri, K. Hajian-Tilaki, Evaluation of serum trace element levels and superoxide dismutase activity in patients with inflammatory bowel disease: translating basic research into clinical application, Biol. Trace Elem. Res. 177 (2017) 235–240.

[37] Q.S. Atwan, N.H. Hayder, Eco-friendly synthesis of Silver nanoparticles by using green method: Improved interaction and application in vitro and in vivo, Iraqi J. Agric. Sci. 51 (2020) 201–216.

[38] L.A. Yaaqoob, Evaluation of the biological effect synthesized iron oxide nanoparticles on Enterococcus faecalisa, Iraqi J. Agric. Sci. 53 (2022) 440–452.

[39] M. Varamini, H. Zamani, H. Hamedani, S. Namdari, B. Rastegari, Immobilization of horseradish peroxidase on lysine-functionalized gum Arabic-coated Fe3O4 nanoparticles for cholesterol determination, Prep. Biochem. Biotechnol. 52 (2022) 737–747.

[40] R. Hasan Huseen, A. A Taha, A. M Abdul Hussien, Cytotoxicity and antibacterial activities of coated and non-coated magnetic nanoparticles, J. Nanostructures. 11 (2021) 698–710.

[41] T. Jeyapragasam, R. Saraswathi, Electrochemical biosensing of carbofuran based on acetylcholinesterase immobilized onto iron oxide–chitosan nanocomposite, Sensors Actuators B Chem. 191 (2014) 681–687.

[42] D.I. Lama, S.I. Neamah, Metabolic responses to in vitro in drought-tolerant in a Cucurbita pepo L. elicited by salicylic acid and zinc oxide nanoparticles, Iraqi J. Agric. Sci. 54 (2023) 1223–1233.

[43] S. Toth-Manikowski, M.G. Atta, Diabetic kidney disease: pathophysiology and therapeutic targets, J. Diabetes Res. 2015 (2015).

[44] N. Malhotra, J.-S. Lee, R.A.D. Liman, J.M.S. Ruallo, O.B. Villaflores, T.-R. Ger, C.-D. Hsiao, Potential toxicity of iron oxide magnetic nanoparticles: a review, Molecules. 25 (2020) 3159.

[45] C. Wenck, N. Meier, E. Heinrich, V. Grützner, F. Wiekhorst, R. Bleul, Design and characterisation of casein coated and drug loaded magnetic nanoparticles for theranostic applications, RSC Adv. 14 (2024) 26388–26399.

[46] M. McMenamin, J. Kolmer, I. Djordjevic, F. Campbell, H. Laurenson-Schafer, J.L. Abbate, B.M. Abdelgawad, A. Babu, T. Balde, N. Batra, WHO Global Situational Alert System: a mixed methods multistage approach to identify country-level COVID-19 alerts, BMJ Glob. Heal. 8 (2023) e012241.

[47] U.S. Ezealigo, B.N. Ezealigo, S.O. Aisida, F.I. Ezema, Iron oxide nanoparticles in biological systems: Antibacterial and toxicology perspective, JCIS Open. 4 (2021) 100027.

[48] H.H. Zahraa, H.A. Rasheed, CYTOTOXIC EFFECT OF ASPARAGINASE PRODUCE FROM E. COLI AND CONJUGATED WITH ZNO NANOPARTICLES ON CANCER CELL LINES, Iraqi J. Agric. Sci. 54 (2023) 1664–1678.

[49] X. Yao, K. Xu, M. Shu, N. Liu, N. Li, X. Chen, K. Nishinari, G.O. Phillips, F. Jiang, Fabrication of iron loaded whey protein isolate/gum Arabic nanoparticles and its adsorption activity on oil-water interface, Food Hydrocoll. 115 (2021) 106610.

[50] F. Liu, Y. Peng, Y. Qiao, Y. Huang, F. Song, M. Zhang, F. Song, Consumption of flavonoids and risk of hormone-related cancers: a systematic review and meta-analysis of observational studies, Nutr. J. 21 (2022) 27.