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Oral Toxicity of Magnesium Oxide Nanoparticles, MgO NPs on Liver in Male Rats

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Abstract

Nanotechnology is an emerging technology that has led to the nanomedicine development, which involves nanoparticles that exist in the natural world. Nanoparticle is an ultrafine unit with dimensions measured in nanometers. They are created as a result of human activities, and have recently been used in various biomedical applications, consumer products, and commercially, etc. Magnesium oxide nanoparticles (MgO NPs), among the known metal oxides, have attracted a vast scientific interest. Therefore, human health can be at risk of continuous exposure. However, the toxic effects of MgO NPs should be assessed with their increased applications. The present study aimed to investigate the physiological and histological, alterations induced by the MgO nanoparticles in hepatocytes of male rats through oral route with (250, and 1000) mg/kg for (14, 28, and 56) days. Fifty-four male mature rats, (2.5 - 3 months old) were divided randomly into nine groups of six rats each, gavaged with MgO NPs (which were purchased from US Research Nanomaterials, Inc). The results obtained revealed highly significant increase (p<0.01) in aspartate aminotransferase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), and total bilirubin. The histopathological examinations showed sinusoidal dilatation, atrophy of hepatocytes, depletion of glycoprotein, focal area of necrosis, inflammatory cells infiltration and apoptosis. MgO NPs demonstrated potential harm to liver tissue, and human health.

Keywords: ALP, Apoptosis, Hepatocytes, MgO nanoparticles, Total Bilirubin.

السمية الفموية لجزيئات أوكسيد المغنيسيوم النانوية على خلايا الكبد في ذكور الجرذان

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الخلاصة

تقنية النانو هي تقنية ناشئة أدت إلى تطوير الطب النانوي ، والذي يتضمن الجسيمات النانوية الموجودة في العالم الطبيعي. الجسيمات النانوية عبارة عن وحدة متناهية الصغر بأبعاد تقاس بالنانومتر ، وقد تم إنشاؤها نتيجة للأنشطة البشرية ، وقد تم استخدامها مؤخرًا في العديد من التطبيقات الطبية الحيوية ، المنتجات الاستهلاكية ،الاستخدام التجاري ، إلخ. جذبت جزيئات اوكسيد المغنسيوم النانوية ، من بين أكاسيد المعادن المعروفة ، اهتمامًا علميًا واسعًا. لذلك ، يمكن أن تكون صحة الإنسان في خطر التعرض المستمر. ومع ذلك ، يجب تقييم التأثيرات السامة لـ جزيئات اوكسيد المغنسيوم النانوية مع زيادة تطبيقاتها. هدفت الدراسة الحالية إلى التحقق من التغيرات الفسيولوجية والنسيجية التي تحدثها جزيئات اوكسيد المغنسيوم النانوية في خلايا الكبد في ذكور الجرذان عن طريق التجريع الفموي مع (250 ، و 1000) ملغم / كغم لمدة (24 ، 14 ، 28 ، 56) يومًا. تم تقسيم أربع وخمسون من ذكور الجرذان البالغة بعمر (5.2–3 أشهر) تم تقسيمها عشوائيًا إلى تسع مجاميع من ستة جرذان لكل منها تم تجريعها بـ MgO NPs (التي تم الحصول عليها من US Research Nanomaterials, Inc اظهرت النتائج زيادة معنوية عالية في ناقلة أمين الأسبارتات (AST)، ناقلة أمين الألانين (ALT)، الفوسفاتاز القلوي القلوي(ALP) والبيليروبين الكلي، وأظهرت الفحوصات النسيجية المرضية توسع جيبي ، ضمور في خلايا الكبد ، استنفاذ البروتين السكري ، المنطقة البؤرية للنخر ، تسلل الخلايا الالتهابية ، وموت الخلايا المبرمج. قد يكون لجزيئات اوكسيد المغنسيوم النانوية التأثيرات السامة المحتملة على أنسجة الكبد وصحة الأنسان.

1. Introduction

Nanoparticles are tiny particles that have a diameter between 1-100 nm. Nano means one billionth, a technology at the nanoscale is a diverse field. It is a part of human's everyday life and has improved a potential value to commercial products as in electronics, fabrics, cosmetics, sports, motor industry, deliver chemicals, filter fluids, and food additives. Nanotechnology is an enormous field that studies small feature sizes with broad applications [1]. Nanoparticles have a large surface area to volume ratio which matters as a lot of the uses of the nanoparticles depend more on their surface area than their volume. Besides all of the benefits mentioned above, nanotechnology has some issues as well. Firstly nanoparticles are still relatively new and their effects on the human body are not fully understood yet, although there is some evidence that they cannot cause harm. However, researchers cannot be sure until more thorough testing is done. Some people think that they need to be regulated more strictly and clearly labelling anything that might contain nanoparticles [2].

Nano magnesium oxide nanoparticle (MgO) is an important inorganic white odorless material with assorted properties. It possesses high hardness, high purity, a high melting point and low heat capacity. Due to its unique and excellent optical, electrical, thermal, mechanical and chemical properties, it is the most promising candidate that has various applications in the day-to-day life in clinical, agricultural, environmental, medication and as food additives [3].
The liver is a reddish-brown organ with four lobes of unequal size with the normal weight of about 1.5 kg. It is the largest internal organ and the largest gland in the human body [4]. The liver plays a role in metabolism (Protein synthesis and degradation, red blood cells destruction, hormone production, regulation of glycogen storage and detoxification). The liver produces bile which is very important for digestion. Liver has other functions like producing insulin-like growth factor, iron storage, converting ammonia to urea, producing immune factors and cholesterol synthesis [5].

2. Materials and Methods

2.1. MgO Nano Powder General Description and Preparation

A fine white powder which was composed of > 99% MgO nanoparticles ranging about 20nm in diameter, was purchased from US Research Nanomaterials,

Inc. This nanomaterial had the following properties:
• Nanopowder (MgO) purity > 99%.
• Nanopowder (MgO) APS: 20nm.
• Nanopowder (MgO) APS colour: White.
• Nanopowder (MgO) morphology: Polyhedral.
• Nanopowder (MgO) Bulk Density: 0.145 g/cm3.
• Nanopowder (MgO) True Density: 3.58 g/m3.

Magnesium oxide was prepared in two different doses:
1) 250 mg/kg of MgO NPs (Low dose) rat groups.
2) 1000 mg/kg of MgO NPs (High dose) rat groups.
The powder was blended with deionized water, stirred continuously for several minutes using a vortex. Each rat was dosed with 1cc of the suspension by oral gavage. The dosing was based on its own weight.

2.2. Animal Care and Experimental Design

A total of 60 male Spargue-Dawley albino rats (*Rattus norvegicus*) were used as a mammalian model for analysis of the physiological, and histological parameters. Mature males (2.5 - 3 months old) were purchased and housed in standard plastic cages with a metal network cover under climate-controlled conditions of the animal house with 25±2ºC temperatures and 10:14 light and dark cycle. Rats were checked daily to see if they were healthy. The cages were usually cleaned out every day to reduce their scent, making the rats more relaxed. The rats were provided with clean bedding, food and water as healthy and unstressed rats are essential for good quality scientific research. Water was provided from a bottle and pelleted food from a hopper.

After an adaptation period of one week into this experimental study, fifty-four male rats were randomly divided into nine groups of six rats each. Groups 1, 2, and 3:(Control) did not receive any dose of MgO NPs for 14, 28, and 56 days subsequently, while groups (4, 6, and 8) these experimental groups were administered with 250 mg/kg MgO nanoparticles every 24 hours for 14, 28, and 56 days, respectively, while groups (5, 7, and 9) respectively received (1000) mg/kg every MgO nanoparticles every 24 hours for 14, 28, and 56 days subsequently.

2.3. Samples Collection and Direct Examination

To prevent the animals from going under any stress, they were fully anesthetized by diethyl ether for several minutes before taking blood samples by heart puncture. Eight ml of blood was collected from each rat, 6ml was used to obtain sera (for the physiological study) which was separated by centrifugation at 3000 rpm for 5 min. The samples were then kept at -20ºC until the time for use. The animals were dissected and the liver was cut out and washed with normal physiological saline 0.9% (NaCl), blotted with filter paper, and then kept in the fixative solution (neutral buffered 10% formalin) for histological study.

2.4. Biochemical Tests for Liver Functions

Biochemical tests were performed depending on sandwich enzyme-linked immunosorbent assay (ELISA) technique.
1. Measurement of alkaline phosphatase (ALP) Level: Kinetic method was used to determine ALP levels in rat serum, according to the Human company kit/ 12217/ Germany.
2. Measurement of aspartate aminotransferase (AST) Level: Kinetic method was used to determine GOT levels in rat serum, according to the Human company kit/ 12211/ Germany.

3. Measurement of alanine transaminase (ALT) Level: Kinetic method was used to determine glutamic-pyruvic transaminase (GPT) levels in rat serum, according to the Human company kit/ 12212/ Germany.
4. Bilirubin Blood Test: Serum bilirubin levels were determined by the photometric method, according to the Human company kit/ 10012/ Germany.

2.5. Histological Preparation: The preparation for histological sections was performed according to the grossing method laid down by Suvarna [6]. Grossing is the process in which pathology specimens are examined and trimmed to the proper size and the best part is selected for further microscopic examinations to obtain diagnostic information.

2.6. Statistical Analysis: Statistical Analysis System (SAS) (2012) program was used to perform analysis of variance (ANOVA) and to find the least significant difference (LSD) test with (P<0.01) was used to determine if there was any significant effects among different factors by comparing between the study mean values.

3. Results and Discussion:

Oral administration of low dose of MgO nanoparticles in male rats produced a highly significant increase (p<0.01) in serum ALT level (31.16±0.11, 43.44±0.19 and 56.64±0.19) U/I at 2, 4 and 8 weeks treatment when compared between the treatment groups themselves. Also, the high dose of MgO nanoparticles caused a highly significant increase (p< 0.01) in serum ALT level (39.55 ±0.13, 49.14 ±0.11 and 76.05 ±0.13) U/I at 2, 4 and 8 weeks treatment when compared between the treatment groups themselves. Moreover, it was found that both low and high doses of MgO nanoparticles 2, 4, and 8 weeks of treatment (31.16±0.11 and 39.55 ±0.13), (43.44±0.19 and 49.14 ±0.11) and (56.64±0.19 and 76.05 ±0.13) U/I respectively in male rats produced a highly significant increase (p<0.01) in the serum AST levels compared to the control groups (27.50±0.11, 27.25±0.14, and 27.11±0.19) U/I. (Table 1).

Table 1**:** Effects of MgO NPs on serum ALT level in male rats

Groups	Mean ± SE of ALT			LSD value
	2 Weeks	4 Weeks	8 Weeks	
Control	27.50±0.11 A a	27.25±0.14 A a	27.11±0.19 A a	0.459 NS
MgO NPs: 250 mg/kg	31.16±0.11 A b	43.44±0.19 B b	56.64±0.19 C b	0.500 **
MgO NPs: 1000 mg/kg	39.55 ±0.13 A c	49.14 ±0.11 B c	76.05 ±0.13 C c	0.369 **
LSD value	0.355**	0.452 **	0.517 **	---

**A, B, C represent the difference among groups (within a row) when time is a variable factor, while concentration is a fixed factor.
**a, b, c represent the difference among groups (within a column) when concentration is a variable factor, while time is a fixed factor.
** = High significant effect with a value of p< 0.01.
NS = Nonsignificant effect

Regarding the AST serum levels, the low and high doses resulted in highly significant increased (p<0.01) values (40.85 ±0.13, and 51.62 ±0.16), (55.94 ±0.17, and 62.53 ±0.14), and (71.37±0.15, and 85.02 ±0.15) U/I in male rats respectively as compared to their control groups (33.64 ±0.16, 33.14 ±0.18, and 33.58 ±0.16) U/I respectively (Table 2). Also, the low dose 250 mg/kg of MgO showed highly significant increased (p<0.01) AST levels (40.85±0.13, 55.94±0.17, and 71.37±0.15) U/I during 2, 4 and 8 weeks respectively when compared between the treated groups themselves. The high dose 1000 mg/kg of MgO showed

highly significant increased (p<0.01) AST levels (51.62 ±0.16, 62.53 ±0.14, and 85.02 ±0.15) U/I respectively during 2, 4 and 8 weeks when compared between the treated groups themselves.

Table 2**:** Effects of MgO NPs on serum AST level in male rats

Groups	Mean ± SE of AST			LSD value
	2 Weeks	4 Weeks	8 Weeks	
Control	33.64 ±0.16 A a	33.14 ±0.18 A a	33.58 ±0.16 A a	0.503 NS
MgO NPs: 250 mg/kg	40.85 ±0.13 A b	55.94 ±0.17 B b	71.37±0.15 C b	0.457 **
MgO NPs: 1000 mg/kg	51.62 ±0.16 A c	62.53 ±0.14 B c	85.02 ±0.15 C c	0.455 **
LSD value	0.456 **	0.503 **	0.457 **	---

**A, B, C represent the difference among groups (within a row) when time is a variable factor, while concentration is a fixed factor.
**a, b, c represents the difference among groups (within a column) when concentration is a variable factor, while time is a fixed factor.
** = High significant effect with a value of p< 0.01.
NS = Nonsignificant effect

Table 3 displays the effects of low and high doses of MgO nanoparticles on ALP levels in male rats. It can be seen that 250, and 1000 mg/kg of MgO produced a highly significant increase (p<0.01) in ALP levels (165.73±0.11, 170.25±0.12), (182.14±0.12, 206.26 ±0.12) and (226.37±0.15, 251.87±0.19) U/I during 2, 4, and 8 weeks respectively compared to their respective values in control rats (152.62±0.11, 152.81±0.11, and 152.55±0.16) U/I. However, it was found that the 250mg/kg of MgO significantly increased (p<0.01) ALP levels (165.73±0.11, 182.14±0.12, and 226.37±0.15) U/I during 2, 4, and 8 weeks respectively when compared between themselves. Furthermore, the 1000 mg/kg dose of MgO significantly increased (p<0.01) ALP levels (170.25±0.12, 206.26 ±0.12, and 251.87±0.19) U/I respectively, when compared between the treated groups themselves during 2, 4 and ,8 weeks of exposure to MgO NPs.

Table 3**:** Effects of MgO NPs on serum ALP level in male rats

Groups	Mean ± SE of ALP			LSD value
	2 Weeks	4 Weeks	8 Weeks	
Control	152.62±0.11 A a	152.81±0.11 A a	152.55±0.16 A a	0.385 NS
MgO NPs: 250 mg/kg	165.73±0.11 A b	182.14±0.12 B b	226.37±0.15 C b	0.384 **
MgO NPs: 1000 mg/kg	170.25±0.12 A c	206.26 ±0.12 B c	251.87±0.19 C c	0.449 **
LSD value	0.337 **	0.354 **	0.508 **	---

**A, B, C represent the difference among groups (within a row) when time is a variable factor, while concentration is a fixed factor.
**a, b, c represent the difference among groups (within a column) when concentration is a variable factor, while time is a fixed factor.
** = High significant effect with a value of p< 0.01.
NS = Nonsignificant effect

In the case of serum total bilirubin, both low dose (0.919±0.040, 1.424±0.046, and 1.998±0.171) mg/dl in male rats during 2, 4, and 8 weeks respectively and high dose (1.067±0.170, 1.955±0.041, and 2.787±0.051) mg/dl during 2, 4, and 8 weeks respectively caused highly significantly increased (p<0.01) total bilirubin serum levels, when compared between the treated groups themselves. Also, high and low levels of MgO NPs orally treated male rats displayed highly significantly increased (p<0.01) total bilirubin serum levels (0.919±0.040, 1.067±0.170), (1.424±0.046, 1.955±0.041), and (1.998±0.171, 2.787±0.051) mg/dl respectively during 2, 4 and 8 weeks in comparison to their respective control groups (0.911±0.028), (1.08±0.017), and (1.031±0.039) mg/dl.

Table 4**:** Effects of MgO NPs on serum total bilirubin level in male rats

Groups	**Mean ± SE of Total Bilirubin**			**LSD value**
	2 Weeks	4 Weeks	8 Weeks	
Control	0.911±0.028 A a	1.08±0.017 A a	1.031±0.039 A a	0.308 NS
MgO NPs: 250 mg/kg	0.919±0.040 A a	1.424±0.046 B b	1.998±0.171 C b	0.113 **
MgO NPs: 1000 mg/kg	1.067±0.170 A b	1.955±0.041 B c	2.787±0.051 C c	0.319 **
LSD value	0.089 *	0.317 *	0.342 **	---

**A, B, C represent the difference among groups (within a row) when time is a variable factor, while concentration is a fixed factor.
**a, b, c represent the difference among groups (within a column) when concentration is a variable factor, while time is a fixed factor.
** = High significant effect with a value of p< 0.01.
*= Significant effect with a value of p< 0.05.
NS = Nonsignificant effect

The examination of liver sections from male control groups displayed normal histological structure appearance, and hepatocyte surrounding the central vein (Figure 1). The two weeks of MgO NPs low dose exposure showed simple sinusoidal dilatation and mild atrophy hepatocytes (Figure 2: A). Whereas two weeks of MgO NPs high dose demonstrated congestion of blood vessels and mild depletion of glycoprotein with slight sinusoidal dilatation (Figure 2: B). Moreover, four weeks of low dose showed atrophy of hepatocytes with degenerative changes, dispersed apoptotic cell changes, and mild sinusoidal dilatation (Figure 3: A). Liver sections during the four weeks of high dose marked depletion of glycoprotein with an accumulation of cholesterol (like plant cells) mitotic and dispersive apoptotic cells (Figure 3: B). In the rats liver low dose for 8 weeks marked congestion of the central vein, depletion of glycoprotein and more accumulation of cholesterol with dispersed apoptotic cells (More sinusoidal dilatation) (Figure 4: A). While 8 weeks of high dose exposure to MgO NPs displayed more sinusoidal dilatation, more atrophy of hepatocytes, depletion of glycoprotein, focal area of necrosis, inflammatory cells and infiltration (Figure 4: B).

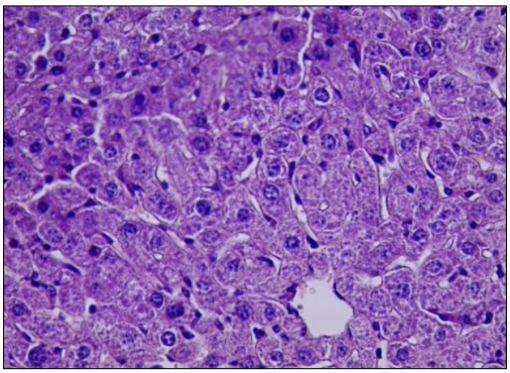

Figure 1**:** The control group of the liver shows normal histological structure appearance, with hepatocytes surrounding the central vein. (40 X) (H&E).

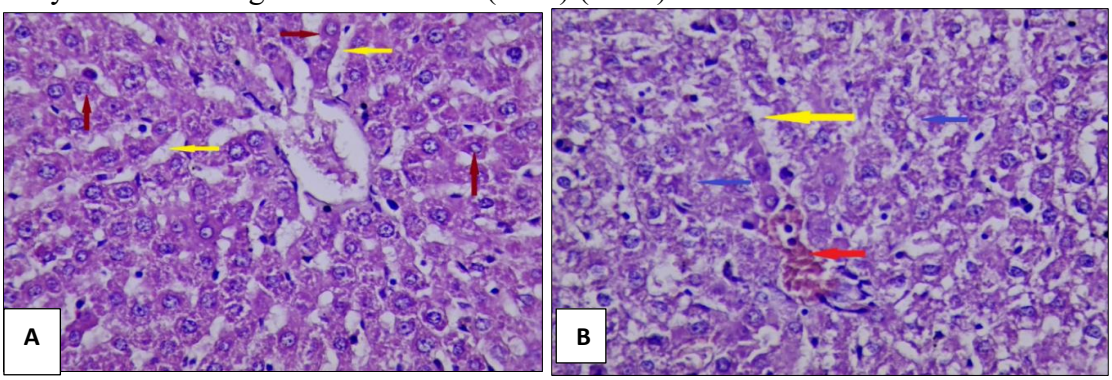

Figure 2: A: The cross-section of the liver shows simple sinusoidal dilatation, mild hepatocyte atrophy in the 2 weeks (low dose) group. (40 X) (H&E).
B: 2 weeks (High dose): Congestion of blood vessels, mild depletion of glycoprotein with slight sinusoidal dilatation. (40 X) (H&E).

Sinusoids Atrophy Depletion Congestion of blood vessels

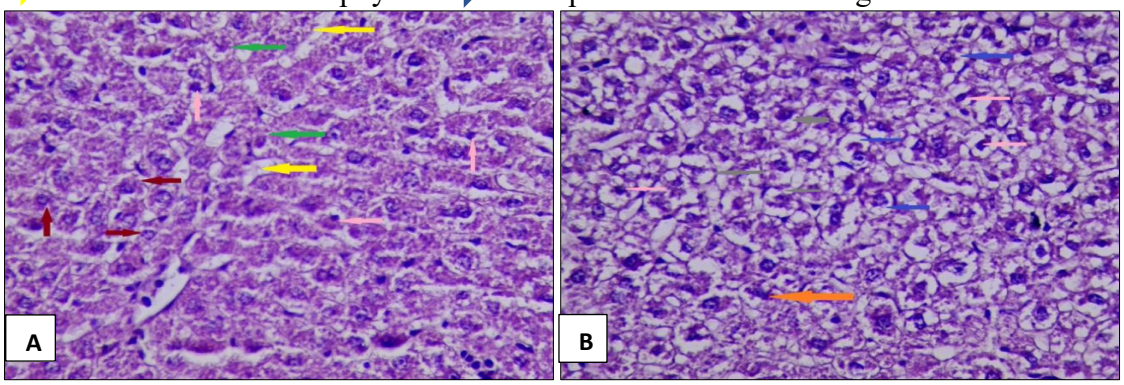

Figure 3: A: The cross-section of the liver in 4 weeks (Low dose) group shows: Atrophy of hepatocyte cell with degenerative changes, dispersed of apoptotic cell changes, mild sinusoidal dilatation. (40 X) (H&E).
B: A cross-section of the liver for 4 weeks (High dose) group shows: Marked depletion of glycoprotein with accumulation of cholesterol (like plant cells) mitotic and dispersive apoptotic cells. (40 X) (H&E).

Atrophy Sinusoids Accumulation of cholesterol
Degeneration Apoptosis Mitosis Depletion

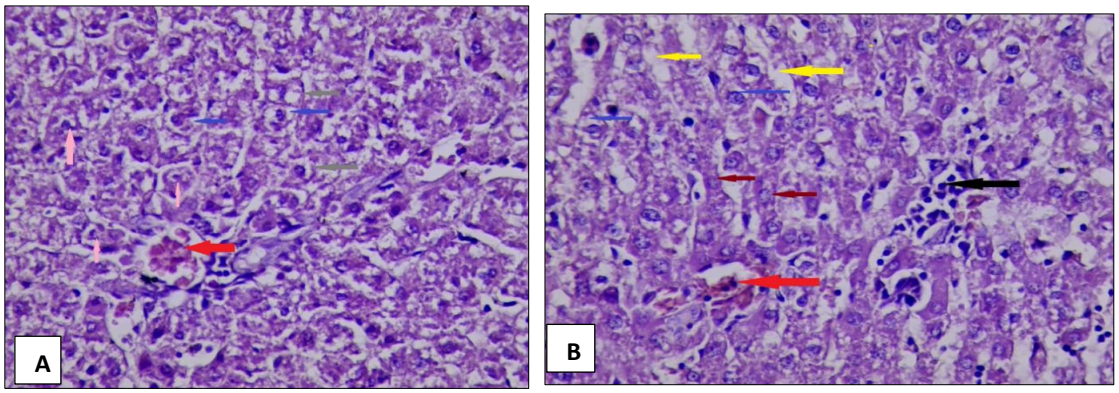

Figure 4: A: In a cross-section of the liver after 8 weeks a (Low dose) group shows: Congestion of central vein, marked depletion of glycoprotein and more accumulation of cholesterol with dispersed apoptotic cells. (More sinusoidal dilatation). (40 X) (H&E).
B: For 8 weeks at (High dose) group, a cross-section of the liver shows: More sinusoidal dilatation, more atrophy of hepatocytes, depletion of glycoprotein, focal area of necrosis, inflammatory cells and infiltration. (40 X) (H&E).

Depletion Sinusoids Accumulation of cholesterol Atrophy
Apoptosis Congestion of blood vessels Necrosis

The study was designed to elucidate how far the low and high doses of MgO NPs are safe to be used in various applications: food additive, dental field, agriculture, electronics, drug delivery and medical treatments.

For this purpose, the estimation of serum liver function parameters, as well as the microscopic examination of the liver were achieved to reveal any abnormal changes in male rats.

In the current study, liver was used to evaluate the cytotoxic effects of MgO NPs as the liver represents the common organ affected by chemical toxicity. The liver has a vital role in the breakdown of most metabolites in the body to maintain normal functioning [7]. Since it was easy for metal oxide nanoparticles to enter the body and with the presence of these nano-size materials in the environment, increase the risk of human exposure and the particles can reach any organ in the body [8]. Nanoparticles distribution is the result of a lot of different processes. Oral administration of MgO NPs passes through the intestine, then their penetration through the intestinal wall, then diffusion to the systemic circulation through different organs before they accumulate in organs and tissues. The maximum accumulation revealed in liver is followed by kidneys, heart, spleen, lungs, and blood [9] . Recently, [10] showed that MgO nanoparticles produce cytotoxic effects in human colon adenocarcinoma (HT29) cells as the results demonstrated a significant increase in apoptosis induction in treated groups of HT-29 with MgO. It is, therefore, better to avoid using them in food stuff and food packaging ingredients. When tested using *in vitro*, three-dimensional (3D) human liver organoid model for 4 weeks results revealed elevated serum GOT and GPT levels, decreased ATP, reduced cell viability and increased reactive oxygen species [11]. The liver has been selected as a priority target organ for oral administration [12]. A 28-day repeated oral administration of MgO NPs in Wistar rats with three different doses (250, 500, and 1000 mg/kg), revealed that MgO NPs accumulated in liver tissue and significantly increased the enzyme levels of GOT, GPT, and ALP in serum [13]. Elevated levels of liver enzymes may be due to the extent of hepatocellular injury, often indicating inflammation or damaged cells leading to a possible leakage of these enzymes into the bloodstream [14]. Plasma membrane functional integrity would be lost resulting in impairment or perturbation of the transport function of hepatocytes resulting in tissue necrosis [15]. Oral administration of MgO NPs

combined with vitamin E or both vitamins (E and C) twice a week for six weeks, increased toxicity, adversely affected blood parameters, elevated Mg bioaccumulation and histological damage to the liver, with a highly significant increase (P<0.001) in serum ROS of male Wistar rats [16]. Hepatocytes injury causes leakage of GPT from the cytoplasm leading to increased serum activity. GOT is present in the cytoplasm and primarily associated with mitochondria. GOT release from the mitochondria requires a severe insult. Therefore, GPT is more readily released when hepatocytes are injured, with high activity level, higher than that of serum GPT. GPT is more specific to liver damage. The largest elevation in GOT activity occurs when hepatocellular necrosis and inflammation occur [17]. Liver mitochondrial enzyme release is considered a strong evidence for hepatic necrosis which is correlated with increased ROS production [18]. Any damage or stress or inflammation that affects the liver can lead to the hepatocytes damage which can essentially release ALP, GOT and GPT in the blood. GPT and GOT elevation means hepatocellular inflammation, rise in levels of bilirubin in conjunction with elevation in transaminase during liver damage or inflammation is a marker index of hepatotoxicity [19]. Whilst studying the biological effects of magnesium oxide NPs size of 5-100 nm and specific surface area of 64,5 m^2/g on human health, generated ROS, DNA damage, interaction with protein structures, destroyed cell membrane, had an impact on proteomic and metabolic profiles and caused mitochondrial dysfunction, in addition to morphological changes and cells death. Toxic effects of magnesium oxide exposure was performed using mathematical models containing data on molecular biological, biochemical, physical, chemical, cytological and ecological properties [20]. Damage that inflicts the functionality of the liver cells can be recognized by the elevation of plasma levels of total bilirubin, albumin, and total proteins even if GPT and GOT activities remained unaltered [21]. In contrast counteractive effects of MgO NPs were observed in this study. They, however, did not show toxicity in other studies. No signs of toxicity or mortality were observed after 14 days of oral exposure at 500 mg/kg of inorganic metal oxides administration to male Wistar rats. The study showed that TiO2, MgO, and ZnO NPs are safe and can be used further [22].

It has been stated that exposure to MgO Nanoparticles with a diameter less than 100 nanometer show no danger [23]. Biosynthesized MgO NPs have suggested nontoxic and bio-functional nanoparticles that possess effective treatment strategies for oxidative-stress-induced tissue damage and thrombosis in both *in vivo* and *in vitro* experimental models [24]. Therefore, MgO NPs are classified as safe and can be used in human food, it has been proven that manganese oxide nanoparticles have antimicrobial properties [25]. The data of a recent study revealed that Cur-MgO nanoparticles conjugation has a potential ameliorative effect on the immunologic, hematologic and hepatic metabolic alterations in type-ll diabetic rats (T2D) [26]. A previous study provides worthy evidences that the MgO nanoparticles can be used as a promising and effective hepatitis treatment in combination with olive and fig extracts for human beings [27]. The biogenically synthesized MgO NPs from pumpkin seed extracts exhibited a significant anticancer efficacy against the ovarian teratocarcinoma cell line (PA-1) [28]. Authors have indicated that nanoparticles (NPs) such as magnesium oxide (MgO) NPs, have few side effects that may have antidiabetic activity. These nanoparticles are also assumed to reduce the damage to the kidneys, liver, pancreas, and reproductive system caused by diabetes, via reducing oxidative stress and increase antioxidant, and glucose utilization [29]. Twenty-five albino female rats received 1 mL of the MgO/ZnO for 30 consecutive days at the respective doses of 1.25, 2.5, 5, and 10 $mg\ L^{-1.}$ The findings indicated that low concentrations of MgO/ZnO core/shell nanoparticles enhanced ROS production which prevented full absorption of MgO/ZnO CS or facilitated their excretion in large amounts, hence lowering its toxicity. Therefore, they are considered safe and desirable for biomedical applications [30].

Histological findings observed that the toxic effects of Mg nanoparticles vary depending on the route of administration, dosage and the duration time of exposure [31]. High concentrations (250 and 500 μg. mL-1) of magnesium oxide NPs administered to Wistar rats under *in vivo* conditions showed congestion in regions of the liver sinusoids, proliferation of bile ductules and apoptotic cells, elevated GOT and ALP levels, whereas there were no significant difference in GPT levels [32]. At a concentration of ≥323.39 μg/mL, MgO nanoparticles induced cytotoxic and genotoxic damage, oxidative damage and cells death which should raise concerns about the safety of MgO nanoparticles used in consumer products [33]. Porous silicon nanoparticles were injected intraperitoneally at a concentration of 1mg/kg, to study the effect of toxicity of these nanoparticles on liver parameters, results of biochemical tests including (GOT), (GPT), and (ALP) were compared with control groups after four weeks, followed by histological analysis of liver sections, results showed non-significant differences in levels (GOT, GPT, ALT) [34]. In a dose-dependent manner, mice received different concentrations (50, 100, 150 and 200 mg/kg) of MgO NPs intraperitoneally and the results exhibited a significant increase (P<0.05) in GOT, GPT and ALP, compared to the control groups. Also histopathological evaluation of the liver showed destruction of hepatocytes, inflammatory cell infiltration, cells division around sinusoidal ducts and cells degeneration around the central vein [35]. Sections of liver from mice treated with 25 mg/kg, 50 mg/kg, and 100 mg/kg ZnO NPs respectively showed histopathological effects, including swelling of hepatocytes at the periphery of lobules and hemorrhagic foci [36]. The suggested mechanism of the nanoparticles toxic effect is revealed in the forms of oxidative stress. As a response of the biological system of the living organisms by producing free radicals, the oxidative stress is pronounced as an imbalance between the formation of ROS and the biochemical mechanisms for detoxifying and repairing of the caused damage. MgO NPs, used in various application fields, simply spread in the environment, terrestrial food chain, and aquatic system, can flow through different levels of the food chain in soil, water, air and a diet containing MgO nanoparticles. Thus evaluating their uptake, toxicity , accumulation, and the traveling through food chain is important to assess their impacts and possible hazards on living organisms [37]. It is considered that metal-based nanoparticles have an impact through a general mechanism on living cells by oxidative stress which eventually leads to cell damage, through the formation of peroxides and free radicals that finally damage DNA, proteins and lipids [38]. Male albino mice were tested for the effects of titanium dioxide nanoparticle suspensions (TiO2) administered intraperitoneally in two doses (150, 600 mg/kg), AST and ALT levels were significantly elevated in all groups exposed, while ALP levels were decreased after fourteen and thirty days of exposure, it was remarkable that TiO2 NPs accumulated in liver and caused histological alterations such as an enlarged dilated portal tract with heavy inflammatory cell infiltration, and a dilated central venule in the liver [39]. An exposure to TiO2 NPS disrupts thyroid and other vital organ functions [40].

4.Conclusion

The results observed that MgO NPs demonstrated potential harm to liver tissue and human health. It has been revealed through different studies that MgO NPs agglomerate in different liver animal models, interact with hepatocytes and may cause oxidative stress . Henceforth, it is necessary to exercise caution when using too many MgO NPs in goods intended for human consumption.

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6. Ethical Approval

Authors have signed ethical consideration approval Ref. No: BCSMU/0122/0004Z. for standards of research involving animals.

7. Conflict of Interest: The authors declare that they have no conflicts of interest.

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