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Review article

ZnO Nanoparticles Impact on Organ Systems in Rats: A Comprehensive Exploration of Diverse Exposure Pathways

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Abstract

The synthesis and utilization of nanomaterials with precise spatial dimensions on the nanoscale are pivotal in the field of nanotechnology. In recent years, metal oxide nanoparticles have become increasingly common, raising concerns in both scientific community and the general public about their potential harm to the environment and living organisms. Despite this, there are still significant debates and misconceptions regarding the adverse effects and mechanisms of these nanoparticles. To facilitate their safe and responsible use, it is imperative to gain a comprehensive understanding of their adverse effects. This review aims to provide an overview of the biological fate of zinc oxide (ZnO) nanoparticles in rats through various exposure routes, shedding light on their toxicological consequences and the underlying mechanisms of toxicity. Despite the fact that ZnO nanoparticles have a propensity to target organs such as the liver, kidneys, and lungs, it is noteworthy that higher concentrations of zinc are detected in these tissues following exposure via various routes. The liver plays a central role in the metabolism of ZnO nanoparticles. Multiple exposure routes, including oral, intraperitoneal, intravenous, and intratracheal routes, have been shown to induce liver damage, along with adverse effects on the kidneys and lungs when exposure occurs via airways. A significant toxicological mechanism associated with ZnO nanoparticles involves the generation of reactive oxygen species (ROS) and the subsequent initiation of oxidative stress. ROS production can result from both excessive release of Zn^{+2} ions and the particulate effect stemming from the semiconductor or electronic properties of ZnO nanoparticles. The potential for surface coatings and modifications holds the promise of further expanding the range of biomedical applications for ZnO nanoparticles, opening up exciting possibilities for futuristic medical treatments, including targeted drug delivery, advanced imaging techniques, and diagnostics.

Keywords: Albino rats, intravenous route, airway exposure, potential infection, hepatic toxicity.

1. Introduction

Nanotechnology is a term used to define areas of science and engineering in which phenomena occurring at nanoscale dimensions are used in the design, characterization, manufacture, and applications of materials, structures, devices, and systems [1]. It has the potential to revolutionize the medical research industry and open up new fields for the betterment of humanity [1,2,3]. The three main categories of nanotechnology are nanotools, nanodevices, and nanostructured materials. Nanotechnology relies on a wide range of techniques, such as computer modeling, surface science, supramolecular chemistry, nanolithography, synthetic approach, and analytical tools. Nanoelectronics, nanospintronics, nanosensors, nano-optical electronics, and nano-drug delivery systems are all examples of nanodevices.

Figure 1. Risks associated with pharmaceutical ZnO utilization [4].

Some examples of nanostructured materials are nanowires, nanoparticles, fullerene, carbon nanotubes, graphene, nanocomposites, thin solid films, nano-patterned surfaces, and supramolecular systems [5]. Different morphologies of ZnO nanoparticles, such as nanoflake, nanoflower, nanobelt, nanorod, and nanowire, have been reported [6, 7, 8].

ZnO is a white inorganic substance which is soluble in acidic or alkaline solutions but insoluble in water. It doesn't naturally occur in large amounts [9]. There are numerous ways to make ZnO nanoparticles, including thermal evaporation, gas evaporation, hydrothermal, the vapor-liquid-solid process, self-combustion, simple thermal sublimation, and green synthesis [10]. Moreover, the simplicity, cost-effectiveness, and eco-friendly nature of green synthesizing ZnO nanoparticle synthesis using extracts from different plants and fruits have garnered significantly attention and interest [11]. Several issues have been highlighted by the extensive and extended use of ZnO at pharmaceutical levels in pig husbandry, including ZnO nanoparticle's advantages are lost with excessive or prolonged ZnO intake, which also increases the risk of harmful consequences [12, 13]. Due to the excessive Zn buildup in animal organs such the pancreas, liver, and kidney [14], which is susceptible to Zn excess [13].

ZnO nanoparticles their increasing application has prompted safety concerns [15]. Size, surface properties, solubility, and mode of exposure are the most important factors in determining the toxicity of metal oxide nanoparticles. The biological fate and toxicity of ZnO nanoparticles upon exposure via various mechanisms must be understood.Workers are most likely to be exposed to ZnO nanoparticles since they are used as food additives and packaging materials ingestion, in addition to the more common routes of exposure (inhalation and skin contact) [16]. If you consume ZnO nanoparticles orally, they will dissolve in your stomach's acidic environment (pH 1.5-2.0) and Zn^{+2} will be absorbed into your blood [17]. ZnO nanoparticles are likely absorbed in both ionic and particle forms after oral administration to rats [18]. After being injected intraperitoneally, ZnO nanoparticles are taken up by the body as ions rather than particles. After oral and intraperitoneal administration, Zn^{+2} is taken up by the liver via the first-pass effect and subsequently redistributed. By dissolving in the acidic lining fluid of the lungs, ZnO

nanoparticles are able to cross the alveolar membrane and enter the circulation after being inhaled [19, 20]. Inhaled ZnO nanoparticles pass the alveolar barrier and enter the circulation because they dissolve in the acidic lining fluid of the lungs. Skin has a pH that varies from the surface to the stratum corneum, which may lead to the dissolution of topically administered ZnO nanoparticles and the dermal absorption of Zn [21].

ZnO nanoparticles are particularly useful in biomedical applications due to their unique properties, such as their size similarity to biomolecules, availability of functionality over wide surfaces, and quantum size impact. ZnO quantum dots can be used for medicine delivery and bioimaging thanks to their high biocompatibility, low toxicity, and good stability [22]. The potential health benefits of ZnO nanoparticles as an antibiotic, nutritional supplement, and food additive are also the subject of ongoing study [23] characteristics are present in ZnO Nanoparticles [24, 25]. They are employed to get rid of aquatic vegetation Applications like medication delivery, cancer prevention, diabetes prevention, anti-diabetic, anti-bacterial, and agronomic properties [26]. Although ZnO is employed for targeted drug delivery, its cytotoxicity limitation has not yet been overcome [27]. They exhibit stronger antibacterial effects than chemically produced ZnO nanoparticles [28, 29, 30]. Additionally, they have been used in the production of rubber, paint, water purification, protein adsorption, and dentistry applications. piezoelectric and pyroelectric that is immune to all types of eradication methods, including physical, chemical, and mechanical ones [31].

2. Different Exposure Routes

Emerging pollutants having ecological and toxicological consequences on individuals, communities, and diverse ecosystems, manufactured nanoparticles, and more especially metal oxide nanoparticles, are finding ever-increasing uses in industrial and consumer products [32, 33].

2.1 Intraperitoneal Administrations

According to Li et al. [34] mice were injected intraperitoneally with ZnO nanoparticles (average size 93 nm) at a dose of 2.5 g/kg, and the findings revealed that Zn accumulated in all organs except the brain (blood-brain barrier). Zn concentrations were highest in the liver, then in the spleen, the lungs, the kidneys, and finally the heart cardiac vascular dysfunction as shown in Figure (2). According to Lin et al. [35] ZnO nanoparticles (size 47.8 nm, dosage 10 mg kg^{-1}) accumulated in the liver, lung, kidneys, spleen, and heart 6 hours after a single intraperitoneal injection. Amara et al, studied the effect of intraperitoneal injection of ZnO nanoparticles (size 20-30 nm, dosage 25 mg kg^{-1}) and they were found no accumulation of Zn in the liver or kidneys of the rats [36]. Elshama et al. [36] found that long-term intra-peritoneal injection of ZnO nanoparticles generated histological and ultrastructural abnormalities in the brains and spinal cords of rats, with the severity of these changes dependent on the dosage and the generation of reactive oxygen species [37].

2.2 Oral Administrations

Baek et al. [18] studied the ZnO Nanoparticles (20 and 70 nm) accumulated in the kidneys, liver, and lungs of rats after a single oral treatment Zn levels in all these organs were were significantly increased 6-24 hours after a low dosage (50 and 300 mg kg^{-1}) of ZnO nanoparticles . After two days of receiving a substantial dosage (2000 mg kg^{-1}), considerable buildup occurred in the liver and kidneys; however, by day seven, levels had returned to normal (about 20 g/g in the liver and around 10 g/g in the kidneys). Earlier research has demonstrated that, upon acute oral exposure, nano-forms of certain particles are more hazardous than their micro-counterparts. Liu N et al. [17] found the level of Zn content accumulation in the heart, liver, spleen, lungs, kidneys, and brain of exposed mice, after receiving a single oral dosage of 45 mg kg^{-1} of ZnO nanoparticles (size 27.54.1 nm), The tissue distribution pattern of ZnO nanoparticles was found to be different from that of $ZnCl_2$ with a greater concentration in the lungs and a lower concentration in the kidneys and liver which leads to oxidative stress as will as DNA damage shown in Figure (3). ZnO nanoparticles (size 40 nm, dosage 134.2-536.8 mg $kg^{-1} day^{-1}$) were distributed to the liver and kidneys in rats

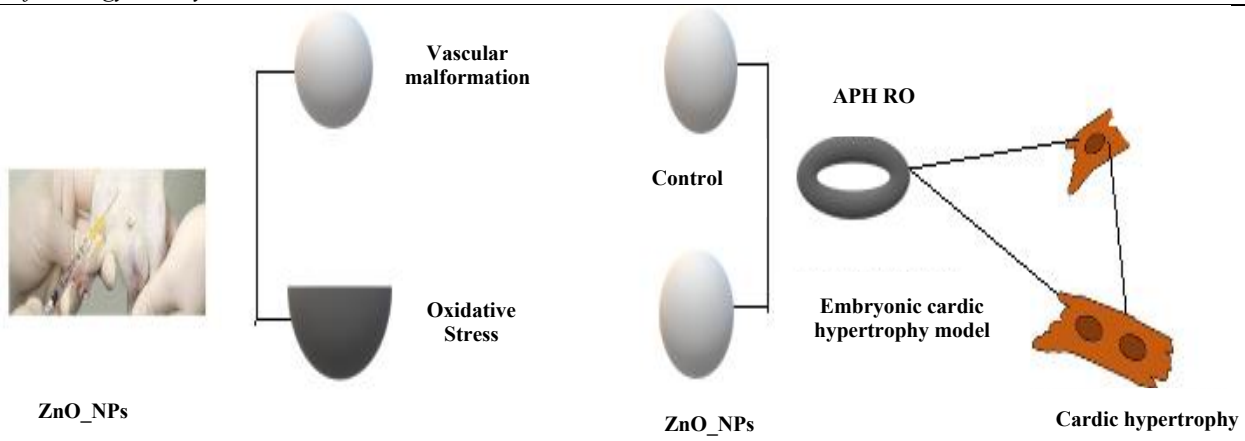

Figure 2. Intraperitoneal ZnO nanoparticles induces vascular malformation and oxidative stress.

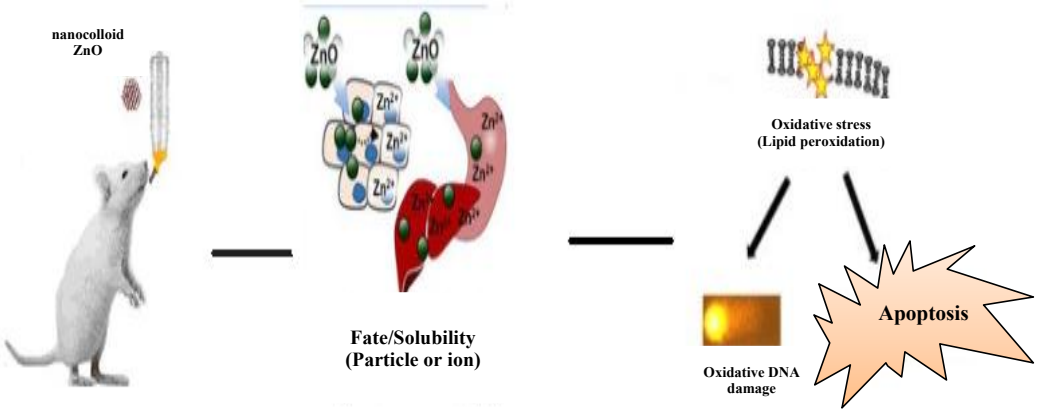

Figure 3. Oral ingestion leading to stomach digestion-induced apoptosis and DNA damage.

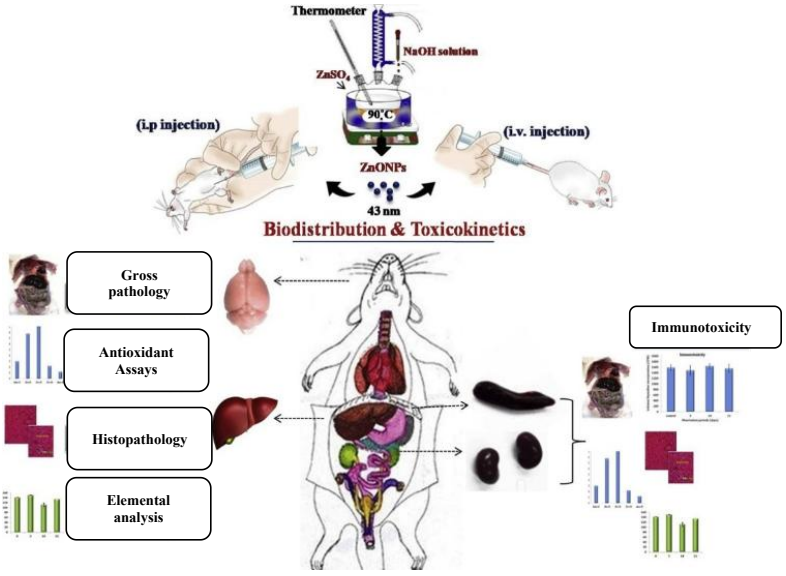

Figure 4. Intravenous and intraperitoneal injection of ZnO nanoparticles induces acute toxicity in vital organs including liver, lungs, kidneys, and other body systems [38].

given repeated oral doses for 13 weeks, although lung distribution was not mentioned.

After being given ZnO nanoparticles (size 30 nm, dose 300 mg kg^{-1}) orally for 14 days in a row, mice saw their liver Zn content significantly increase and their kidney Zn content somewhat increase [16]. According to Choi et al. [15] a single oral dose of either 3 or 30 mg kg^{-1} of ZnO nanoparticles showed that these particles were predominantly distributed in the liver, kidneys, and lungs. Following oral administration, it was observed that ZnO nanoparticles had limited absorption in the gastrointestinal tract (GIT) and were primarily excreted in feces. [39].

Pasupuleti et al. [40] considered that ZnO nanoparticles accumulated in the liver of mice after 14 days of oral treatment of 30 nm ZnO nanoparticles, and they also caused oxidative stress. Sprague Dawley (SD) rats were also given oral administration of ZnO nanoparticles for 14 days. They discovered that rats treated with low doses of nanoparticles had higher rates of lesions in the liver, pancreas, heart, and stomach than rats treated with high doses; however, high dosages of the micro-sized nanoparticles produced more lesions than the low one.

2.3 Intravenous Administrations

Lee J et al. [41] were used rats for checking the effects of intravenous injections of 5, 10, and 20 mg kg^{-1} of ZnO nanoparticles on dams and fetuses from gestation day 6 to day 20. Twenty dams in the 20 mg kg^{-1} treatment group lost two of them while under treatment. In treated dams, hematological analysis and serum biochemistry revealed dose-dependent damage. Tubular dilatation in the kidneys, extremely hemopoiesis in the liver, and multifocal mixed cell infiltration and thrombosis in the lung were all discovered by histopathological study of treated dams.

Yeh et al. [38] when injected intravenously into mice, radioactive ZnO nanoparticles that emit gamma rays were mostly localized in the lungs, with some also found in the organs responsible for digestion and detoxification. The distribution of ZnO nanoparticles at 24 hours after injection showed the largest amounts in the lungs and liver. ZnO nanoparticles (size 10 and 70 nm, dosage 120 g $mouse^{-1}$) were injected intravenously and studied for their effects on mice over time (days, weeks, and months).

Radioactive ZnO nanoparticles that release gamma rays were predominantly located in the lungs after being injected intravenously into mice, with some also identified in the organs responsible for digesting and detoxification. After being administered intravenously into mice, radioactive ZnO nanoparticles that produce gamma rays were found mostly in the lungs, with some being found in the digestive and detoxifying organs as shown in Figure 4 were given a single intravenous injection, and by the next day, nanoparticles could be found throughout the mice's bodies, including their blood, liver, spleen, lungs, brain, and heart [38].

Fujihara J et al. [42] were administered intravenously ZnO nanoparticles (size 58.5 nm, 0.2 mg kg^{-1}) to mice, and their short-term tissue distribution in the lungs, liver, kidneys, and spleen was evaluated for up to 1 hour after administration Liver and lung Zn levels peaked 5 minutes after the treatment, kidney, and spleen Zn levels peaked 15 minutes after the administration, and tissue Zn levels peaked 1 hour after the dose. They also reported that Zn tissue accumulation over time 6 days following intravenous injection of ZnO nanoparticles (0.05 or 0.2 mg kg^{-1}). Zn levels were only substantially higher in the kidneys after one day at 0.05 mg kg^{-1} compared to the control group. Zn content was considerably elevated in the liver and spleen after just one day and six days at a level of 0.2 mg kg^{-1}.

2.4 Inhalation Exposure

Vysloužil et al., [43] were reported that ZnO nanoparticles (size 374.2 nm, dosage 6.46104 and 1.93106 particles/cm^3) to enter rats organs from ambient air at two different doses, with the lower dose considerably increasing Zn content in the liver and at higher dose significantly increasing Zn content in the lungs. Konduru et al., were injected intratracheally ZnO nanoparticles (size: 4.62.5 nm, dose: 1 mg kg^{-1}) into rats and their tissue distribution was mapped out. The transfer of 65Zn to the skeletal muscle, bone, kidneys, liver, and skin occurred on day 2. It was injected into the skin, bones, and muscles on

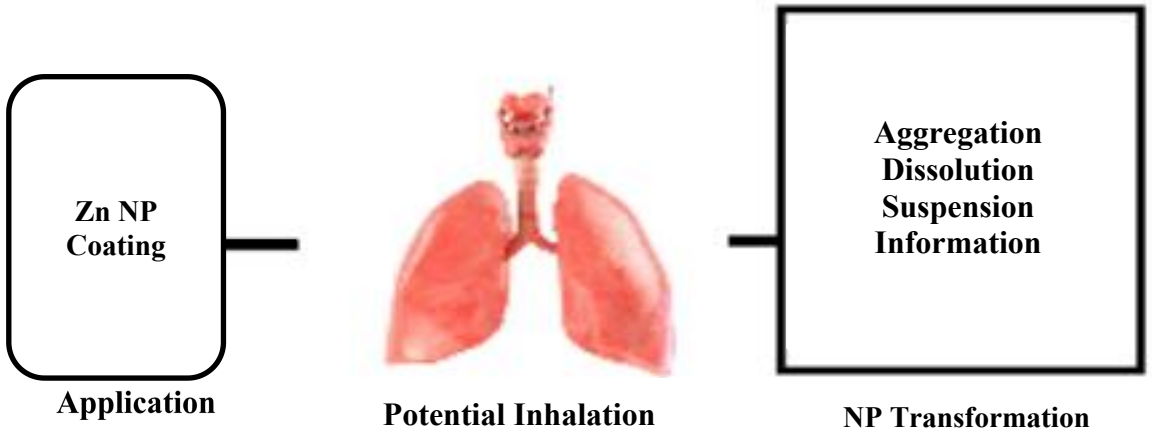

Figure 5. Inhaled ZnO nanoparticles directly induce lung injury.

days 7 and 28 [44]. Wang et al., were injected intratracheally ZnO nanoparticles (size 4218 nm, dosage 2.5 mg kg^{-1}) in the lungs and liver of mice [19]. Depending on the particle size and solubility, ZnO nanoparticles induced inflammatory and fibrotic responses in the tracheobronchial and alveolar tissues after inhalation. Lung fluid is acidic, so when ZnO nanoparticles were dissolved in it, their concentration increased and they were hazardous to the lungs [45]. Fujihara J., ZnO nanoparticles inhalation experiments revealed negligible lung cytotoxicity, histopathologic alterations, or pulmonary inflammation. ZnO nanoparticles have likely been dissolved in the respiratory system following inhalation if there is a higher Zn content in the BAL fluid and lungs. The toxicity of ZnO nanoparticles was significantly influenced by the exposure concentration, exposure mode, and time post-exposure. To conclude that ZnO nanoparticles had low sub-chronic toxicity via inhalation, exposure for 13 weeks at a cumulative dose of 10.9 mg kg^{-1} resulted in increased lung cellularity, but other markers of toxicity did not differ from sham-exposed animals [46].

3. Toxicological effects ZnO nanoparticles on various organs of Rats

ZnO nanoparticles have been shown to exert harmful effects on different cells, including membrane damage, an inflammatory response, DNA damage, and apoptosis. According to recent research, the release of Zn^{+2} ions is what causes ZnO nanoparticles to be poisonous.

3.1 Effects on Body Weight and Organ Weight

Treatment of rats orally with 536.8 mg kg^{-1} day^{-1} for 13 weeks resulted in a reduction in body weight [47] Rats (size 12-90 nm) and mice (size 205 nm) were given 30.3 mg kg^{-1} intraperitoneally on a daily basis for 28 days, and 1, 10, and 100 mg kg^{-1} intraperitoneally daily for 14 days [48]. Rats and mice have been used as examples. After a single oral dosage of 2000 mg kg^{-1} (size 20 and 70 nm), rats lost a small amount of weight. At 14 days post-exposure, mice given a single intragastric dose (size205 nm, 100 mg kg^{-1}) and rats given repeated intraperitoneal and intravenous exposure (size1290 nm, dose 30.3 mg kg^{-1} for 14 and 28 days) showed decreased organ weight of the heart, liver, spleen, lungs, kidneys, and brain [48]. After 400 mg kg^{-1}/day of nanoparticles were administered to the dams, their body weight dramatically fell. They also ate less after receiving 200 and 400 mg kg^{-1} /day of nanoparticles, and after 400 mg kg^{-1} /day of nanoparticles, their liver and adrenal gland weights also rose [49].

Jo et al. [49] were exposed rats to ZnO nanoparticles (500 mg kg^{-1} bw) with a size smaller than 100 nm. Additionally, the zinc concentration in dams and offspring's bodies was assessed. Rats given nano-ZnO treatment had lower pup weights, fewer live births, and higher fetal resorption rates. The liver and kidney of puppies as well as the mammary tissue of mothers were given ZnO nanoparticles. These findings suggest that nanoscale ZnO nanoparticles [50]. Wang et al. [50] found that 50 and 500 mg kg^{-1} nano ZnO illustrated increases in body weight while 5000 mg kg^{-1} showed declines in body weight, indicating that high dosages of ZnO nanoparticles in the diet

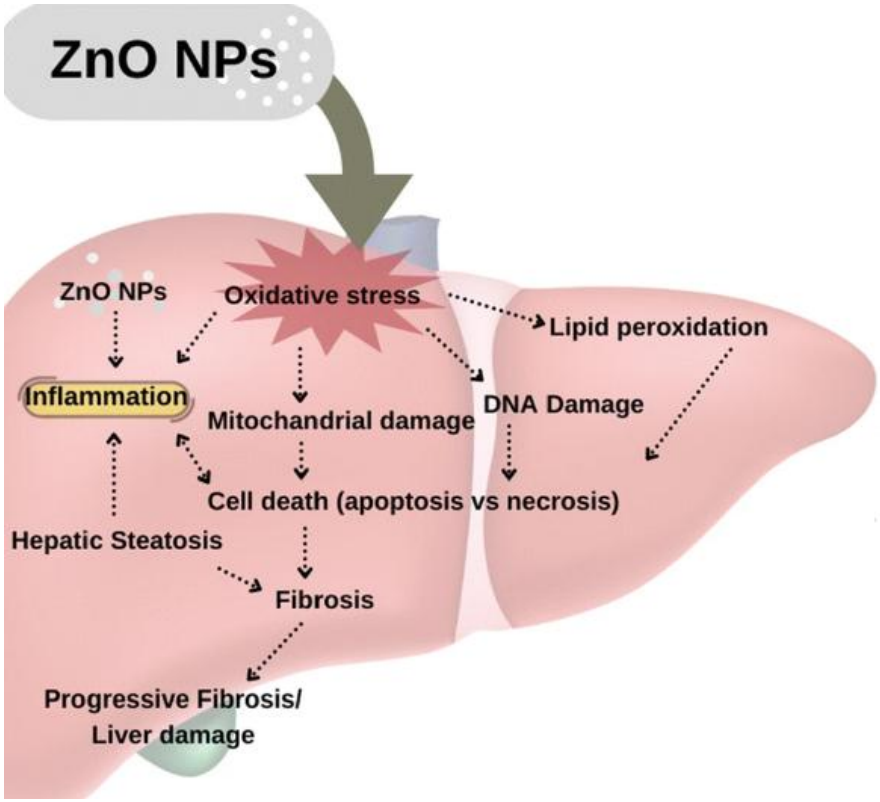

Figure 6. Graphical representation of the effects of ZnO nanoparticles on the liver [51].

could have toxicological effects [52]. The rise in the relative organ weights of the pancreas, brain, and lung at 5000 mg kg^{-1} ZnO nanoparticles may be largely attributed to the decrease in body weight [53].

3.2 Effects on Liver Tissues

After entering the body through any of the various methods, ZnO nanoparticles may function as a key target organ for the liver, which is the main organ of metabolism. The ability of ZnO nanoparticles to induce apoptosis and genotoxicity in human liver cells (HepG2), as well as the underlying molecular mechanisms of its cellular toxicity. Investigations were done on the part that dissolution plays in the toxicity of ZnO nanoparticles [54]. Given that inhalation is the primary method of exposure to ZnO nanoparticles in the workplace, pulmonary toxicity caused by ZnO nanoparticles has come under increased scrutiny. Acute pulmonary inflammation, chronic inflammation, altered metabolisms, histological abnormalities in the lungs, and airway irritation were among the toxicity outcomes caused by ZnO nanoparticles that were previously documented in vivo investigations [55].

ZnO nanoparticles, regardless of how they enter the body, preferentially accumulate in the liver. ZnO exposure has been linked to hepatic damage, suggesting that the liver, the body's biggest detoxifying organ, is vulnerable to xenobiotic-mediated damage which is shown in Figure (6). ZnO nanoparticles caused increased levels of the enzymes aspartate transaminase (AST), alanine transaminase (ALT), and lactate dehydrogenase (LDH) in rats and mice after a single oral dosage [56]. Alkaline phosphatase (ALT) levels were shown to be increased in studies using a single intraperitoneal dosage of ZnO nanoparticles. When a single dosage of ZnO nanoparticles was given orally to mice [23]. Histopathological changes in the liver have been reported after a single oral dosage of ZnO nanoparticles in mice, including extensive hepatic edema, vacuolization, cellular necrosis, congestion, and fibrosis. glycogen buildup [34].

Table 1. Toxicological effects ZnO nanoparticles on the livers, kidneys, and lungs of rats.

Organs	Routes	Effects	References
Livers	Oral, IM, IV, IP	Abnormal rise of blood liver enzymes • AST, ALT, ALP, and LDH and Histopathological changes	[19,23,34,48,57,58]
	Oral, IV	causes significant liver enlargement, vacuolization, cellular necrosis, congestion, and glycogen buildup.	[34,58]
	Oral	Low-dose apoptotic liver alterations and single-dose IP localized inflammation	[23]
	IM	The hepatic sinusoid can be partially dilated	[42]
Kidneys	IP	Elevated levels of kidney injury marker BUN and creatine phosphokinase	[35, 59]
	Oral	Histopathological alterations were seen that decreased total kidney glutathione levels.	[58]
	IP	Resulting in necrosis, edema, and hydropic degeneration	[16]
	SC, IP	causes tubular dilation, Focal interstitial edema, and inflammation	[23]
Lungs	IP	Markers for oxidative stress and inflammation were found to be elevated, including lipid peroxide, heme oxygenase-1 (HO-1), and -tocopherol in the lungs	[60]
	IT	Extreme alveolar desquamation caused by massive acute lung inflammation caused	[61]
	IT	lung and systemic inflammation, dyslipidemia, and elevated blood HO-1 levels.	[20]
	IP, IT	Induces edema, lymphoid cell infiltration, and increased bronchiole epithelial cell proliferation and hypertrophy, induced pulmonary fibrosis and inflammation	[19]
	IV, IT	can cause aortic damage; cause serious inflammation, significant hyperemia in the alveoli, and edema;	[20,58]
	IP	causes mild interstitial inflammation.	[23]
	IV		[42]

System localized infarction at high dosages and early apoptotic changes in the liver were seen after a single intraperitoneal injection of ZnO nanoparticles in rats. We found that 1 day after a single intravenous injection (0.2 mg kg^{-1} of ZnO nanoparticles in mice, the hepatic sinusoid was slightly dilated and the Zn concentration in liver was 10.1-8.6 g/g [39].

3.3 Effects on Kidney Tissues

Yan G et al. [62] examined the biochemical compositions of urine and kidney samples from rats that received doses of 100, 300, and 1000 mg kg^{-1} of ZnO nanoparticles over a 14-day period, using a 1H nuclear magnetic resonance (NMR) technique. The results show that ZnO nanoparticles can disrupt energy metabolism and result in mitochondrial and cell membrane impairment in the rat kidney, which may contribute to ZnO nanoparticles-induced nephrotoxicity.

Rani V et al. [63] treated the rats with ZnO nanoparticles (50 mg kg^{-1}). Their histopathological studies also showed that the morphology of the liver cells had improved. ZnO nanoparticles may provide protection by selectively intoxicating proliferating

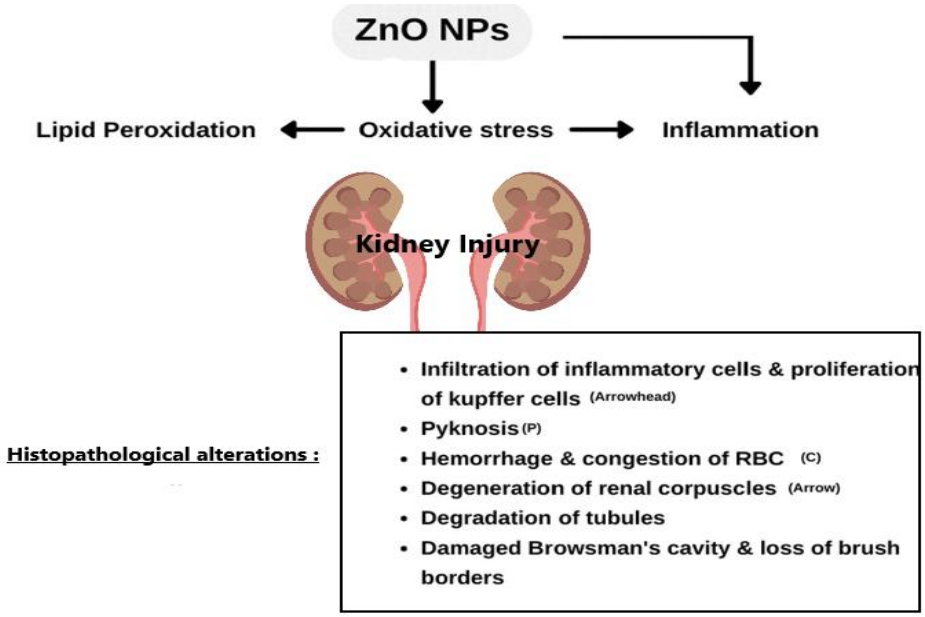

Figure 7. Flowchart depicting kidney damage caused by ZnO nanoparticles [51].

tissue, such as the adenomatous islands developed in the liver. The amelioration of DMN-induced toxic effects may also involve zinc metallothionein (Zn-MT), which is induced by ZnO nanoparticles. The major mechanism underlying ZnO nanoparticles' protective properties is still their ability to reduce oxidative stress. ZnO nanoparticles accumulated in the tested organs including kidneys, suggesting that the kidney could be one of the major target organs for the ZnO nanoparticles induced toxicity [35]. The most recent research on the nephrotoxicity caused by ZnO nanoparticles in several animal models. Bowman's gap increases, the distal convoluted tubule is destroyed, there is intratubular protein deposition, inflammatory cells are infiltrated, and there are capillaries clogged between the tubules, among other histological alterations in the kidney. Histological examination of the kidney and serum biochemical analysis [64]. The kidneys, due to their high blood supply and ability to concentrate toxins, are especially susceptible to xenobiotics and are the preferred accumulation site for ZnO nanoparticles following oral ZnO exposure (600 milligrams or 1 gram per kilogram of body weight every day for 5 days) total glutathione in the kidneys is significantly reduced, suggesting functional impairment to kidney tissue [59].

The rats had elevated levels of blood urea nitrogen (BUN) and creatinine (Cre) 6 hours after receiving a single intraperitoneal injection of ZnO nanoparticles (dosage 10 mg kg^{-1}), These are biochemical markers of kidney damage and a Zn level of around 70 g/g in the kidneys. In contrast, we found that mice given a single intravenous injection of ZnO nanoparticles (0.2 mg kg^{-1}) did not develop any pathological alterations to the kidneys, including an increase in BUN or Cre levels. In comparison, the Zn content in kidneys was 8.6 1.0 g/g [57]. ZnO nanoparticles (size 30 nm, dosage 300 mg kg^{-1}) were given orally to mice over the course of 14 days, and Zn content was measured in kidneys to be about 40 g/g, indicating that the tubules had enlarged as a result. After receiving a single intraperitoneal dose of ZnO nanoparticles (size 20 nm, dose 100 g/mL daily for 14 days), the kidneys of mice were inflamed and developed focal interstitial edema, as determined by a pathological investigation. Khorsandi et al. [65] reported that inhaled ZnO nanoparticles cause renal irritation to last for a long time. Malonaldehyde, H_2O_2, and NO concentrations in the kidney were reduced in DMN (2 l/100 g body weight/rat)-treated rats when ZnO nanoparticles (50 mg kg^{-1} body weight/rat) were administered. The healing of oxidative DNA damage and less apparent histological abnormalities in the

kidney lend weight to these findings. ZnO nanoparticles are thought to be harmful to renal tissue, yet their high therapeutic and antioxidative properties aid in lessening the rat kidney damage caused by dimethylnitrosamine (DMN) [66].

3.4 Effects on Lung Tissues

Inhalation is the most common route of ZnO nanoparticles exposure on the job. Studies on the toxicity and injury caused by ZnO nanoparticles to the lungs have mostly involved exposure by inhalation, intratracheal instillation, or intranasal administration. Researchers have shown that inhaling ZnO nanoparticles is the most lethal route of exposure [67]. Due to the acidic nature of lung lining fluid, ZnO nanoparticles degrade and release Zn^{+2}, leading to ROS-induced inflammation, necrosis, and cell death [68]. High levels of oxidative stress were seen in rats after receiving a single intratracheal injection of ZnO nanoparticles (size 21 nm, dosage 70 g mL^{-1}), as evidenced by an increase in lung lipid peroxide, heme oxygenase-1 (HO^{-1}), and -tocopherol. The ZnO nanoparticles also stayed there in the lungs and continued to release Zn^{+2} [60]. Jacobsen et al. [61] were injected ZnO nanoparticles (size 12–3 nm, low dose–0.3 mg kg^{-1}) into the trachea of mice, and the animals afterward experienced massive acute pulmonary infarction, excessive desquamation of alveolar barrier epithelial cells, and death, and histological alterations (including edema, eosinophilic granuloma, lymphoid cell infiltration, and enhanced proliferation and hypertrophy of bronchiole epithelial cells). An increase in the lung weight to body weight ratio, as well as histological abnormalities such as pulmonary fibrosis and inflammation, were seen 7 days after intratracheal injection of ZnO nanoparticles in mice. Huang et al., studied the effects of ZnO nanoparticles inhalation in mice (dosage 2.5 mg/m3, 5 hours/day for 5 days) and hypothesized that ZnO nanoparticles inhalation may lead to the development of allergic airway inflammation [69]. Figure 5 shows the toxicity effect of in lungs.

Saptarshi et al. [70] investigated the 24 hours inhalation of ZnO nanoparticles (size 30 nm, dose 5 mg kg^{-1}), mice showed signs of pulmonary inflammation as evidenced by an increase in eotaxin mRNA expression in lung tissue and the release of pro-inflammatory cytokines in their blood. Histopathological abnormalities in the lungs of mice were seen after a single oral exposure to ZnO nanoparticles. These abnormalities included severe inflammation, vascular damage figure (8) severe hyperemia in the alveoli, and edema [71]. Cho W-S et al. [72] administered ZnO nanoparticles intratracheally to rats at two different dosages (50 and 150 cm2/rat). They conducted assessments at 24 hours, one week, and four weeks to evaluate the dose-dependent and time-dependent responses. Eosinophilia, airway epithelial cell proliferation, goblet cell hyperplasia, and lung fibrosis were all brought on by ZnO nanoparticles. Chronic bronchocentric interstitial lung fibrosis was linked to elevated myofibroblast accumulation and positive transforming growth factor. The fundamental source of ZnO nanoparticles-induced various progressive severe lung damage is a pH-dependent breakdown of ZnO nanoparticles inside phagosomes. Wang, D et al. [19] were given varied doses of ZnO nanoparticles (200, 400, 800 g/kg) to mice. Animal mortality, organ/body weight ratios, hematological, blood biochemistry, and histopathology were used to determine the acute toxicity of the substance. Malondialdehyde levels in the lung homogenates also rose. In addition, it was found that the lungs had undergone inflammatory and hyperplastic alterations.

4. Therapeutic Mechanism of ZnO Nanoparticles

The exact toxicological mechanisms of ZnO nanoparticles are yet unclear. Nanoparticles have various physical and chemical characteristics, which may be related to their potential toxicity because their surface area is proportionately bigger than that of larger particles [73].When ingested, ZnO nanoparticles break down and release Zn^{+2}. Some research has put forward the theory that Zn^{+2} is responsible for toxicity effects of ZnO nanoparticles [74]. Fukui et al., injected intratracheally ZnO nanoparticles stayed in the lungs of rats, where they constantly produced Zn^{+2} and caused severe oxidative stress. ZnO nanoparticles induced a rise in 8-hydroxydeoxyguanosine (8-OHdG), a major ROS product and a widely used marker for

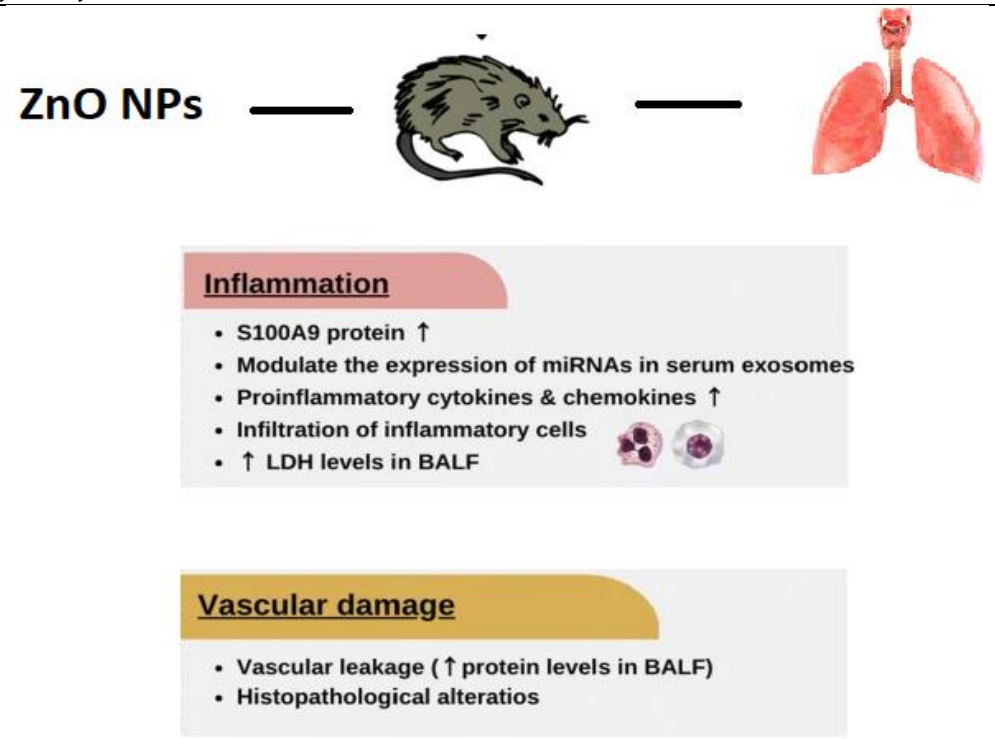

Figure 8. Lung, vascular damage, and histopathological alterations induced by ZnO nanoparticles [51].

oxidative DNA damage, whether injected intratracheally or breathed. ZnO nanoparticles are capable of producing high amounts of free radicals, which can lead to oxidative damage [60]. Li YS et al. [75] found that 8-OHdG was highly accumulated in the lungs after intratracheal installation of ZnO nanoparticles containing lipopolysaccharides. Possible involvement of oxidative stress in inflammation; this can lead to DNA damage and cell death, or apoptosis. They postulated further that ZnO nanoparticles may cross the blood-air barrier and harm the liver in this way. Previous research examined how much 8-OHdG was excreted in the urine following a single intravenous dose of ZnO nanoparticles. The concentration was significantly higher after day one and decreased dose-dependently over the next six days. Serum superoxide dismutase levels were considerably elevated at 24 and 48 hours after intravenous injection of 0.2 mg kg^{-1} ZnO nanoparticles. An in vitro study found that both ZnO nanoparticles and Zn^{+2} entered cells. Zn^{+2} impacts enzyme balance, transcription factors, and signaling pathways, whereas nanoparticles induce cell inactivation, oxidative stress, mitochondrial damage, and intracellular Ca^{+2} excess. Comparing the effects of ZnO nanoparticles and bulk ZnO on astrocytes revealed that both were toxic, but that astrocytes exposed to ZnO nanoparticles had more ROS production and caspase activity than those exposed to bulk ZnO [76]. Tang et al. [77] found that after a week of oral treatment of ZnO Nanoparticles at 100, 300, and 600 mg kg^{-1}, mRNA expression for cytochrome P450 1A2 (CYP1A2) was downregulated, whereas expression for cytochrome P450 2C11 and CYP3A4 was upregulated, and pathological abnormalities were observed in liver and kidney tissues.

5. Conclusion

ZnO nanoparticles are rapidly distributed throughout the body and are efficiently eliminated. They are primarily absorbed in ionic form, with some in particle form. Importantly, these nanoparticles do not tend to accumulate in tissues over an extended period. Regardless of the exposure method, higher concentrations of Zn were detected in the key target organs for ZnO nanoparticles, including the liver, kidneys, and lungs. The liver is the primary site of accumulation for ZnO nanoparticles, and exposure through various routes led to histological changes and liver damage. Following a single oral or intraperitoneal

dose, kidney injury was observed. Notably, Lung damage was assessed using intratracheal instillation and inhalation exposure methods. The primary toxicological mechanism associated with ZnO nanoparticles involves the generation of substantial oxidative stress, characterized by the production of significant levels of reactive oxygen species (ROS). ROS generation is attributed to two main factors: the release of Zn^{+2} ions from ZnO nanoparticles and the particulate effect resulting from the semiconductor or electronic properties of ZnO nanoparticles. One effective strategy to mitigate the toxicity of these particles is by coating the surface of ZnO nanoparticles with silica, which effectively suppresses ROS production and Zn^{+2} ion release.

Data Availability statement

The data used to support the findings of this study are available from the corresponding author upon request.

Conflicts of Interest

All authors declare that, they have no conflict of interest.

Author Contributions

All authors participated in the initial draft creation, reviewed the manuscript, and contributed to the editing process.

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