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Review

Toxic Effects of Metallic Nanoparticles on Rat's Spleen; a Review

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Abstract

Metallic nanoparticles play a key role in the development of new technologies in recent years. Metallic nanostructures are now being manufactured using a wide range of elements including copper, zinc, titanium, magnesium, gold and silver. These are increasingly used in commercial and biomedical applications and this has led to an increase in the likelihood of nanoparticles interactions with both terrestrial and aquatic ecosystems, as well as the possibility of exposure and damages that happen to animal's health. This review article focuses on toxicity of silver, copper and gold nanoparticles that affect the spleen in rat model. The nanoparticles produce reactive oxygen species (ROS) that results in oxidative stress, altering cell membrane integrity, vacuolation of cells with ill-defined white pulp, altered architecture and congestion of red pulp and multinucleation of spleen cells. Silver nanoparticles cause more severe effects on rat's spleen as compared to copper and gold nanoparticles. Further research is required to study the impact of other nanoparticles (lipid nanoparticles, polymeric nanoparticles etc.) on the spleen of rat model.

Keywords: nanoparticles, toxicity, white pulp, red pulp, spleen, rat model

Introduction

Nanoparticles (NP) are extremely small particles, ranging from 1 to 100 nanometers in size. The features, forms, or dimensions of NPs categorize them in a variety of distinct groupings. Some of the well-known classes of NPs are carbon-based NPs, metal NPs, ceramics NPs, semiconductor NPs, polymeric NPs, lipid-based NPs [1] as demonstrated in Fig. 1.

Nanoparticles are in a position to have significant and extensive applications, primarily in the field of biomedicine, due to their tiny size and huge surface-to-volume ratio. Although the small size of engineered nanoparticles has been linked with highly desirable properties (mechanical, electrical, and chemical) for specific uses, it is likely that these same desirable properties will also be associated with unwanted biological or toxicological reactivity. The small size of nanoparticles raises questions about the safety of biomedical nanoparticle usage [2].

Copper, zinc, titanium, magnesium, gold and silver are some of the elements that are now being utilized in the production of a variety of metallic nanostructures [3]. Studies have found a correlation between the particle size of certain metal-based nanoparticles (e.g., Ag, Au, and Cu) and toxicity, even if the same material is relatively inert in its bulk form. This is despite the fact that metal nanoparticles have received attention

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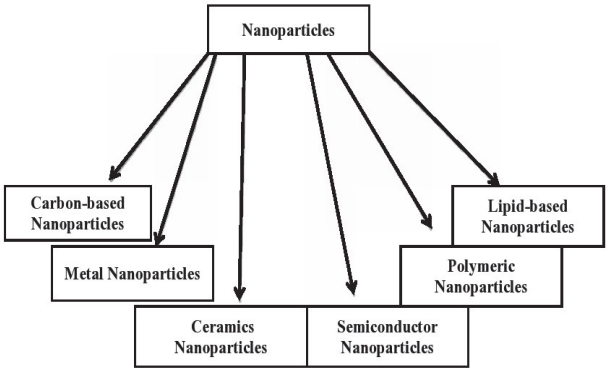

Fig. 1. Classes of Nanoparticles.

due to the widespread applications they have in the medical, consumer, industrial, and military fields [4]. In addition, there is a scarcity of evidence about the safety and toxicity profiles of metal nanoparticles' [5].

Taking into consideration the nanoparticles tiny size, huge surface area to volume ratio, and chemical reactivity, as well as the biological activity of the nanoparticles, raises the possibility that the concerns about potential health dangers are not wholly unwarranted. Studies have shown that nanoparticles have the potential to disrupt normal physiology by interacting with biomolecules in living cells, which can lead to negative effects at the cellular, subcellular, and protein levels. Additionally, nanoparticles may be able to cross the blood-brain barrier and cause neurotoxicity [5].

Silver nanoparticle (AgNP), which possesses potent broad-spectrum antibacterial properties, strong permeability, and little drug resistance, was used in the production of a variety of antibacterial medical products, such as toothpaste, gynecologic suppository, and wound dressing. These antibacterial medical products were designed to treat infections caused by bacteria. It is estimated that the only United States generates between 2.8 and 20 tons of AgNPs every single year. The alteration of around 14% of the goods that contain AgNPs has the potential to release these nanoparticles. Concern has been raised over the potentially harmful consequences that might result from prolonged exposure to AgNPs [6].

Copper nanoparticles (Cu NPs) and their oxidized form, copper oxide nanoparticles (CuO NPs), are commonly employed as additions in cattle and poultry feed, polymers and plastics, and lubricants for metallic coatings [7]. It is well established that nano-Cu induces the generation of reactive oxygen species within the body, hence leading to oxidative damage. After moving through the gastrointestinal tract, Jani et al., found that nanoparticles might reach lymphocyte nodes in the liver and spleen [8]. After being taken in by the splenic macrophages via the process of endocytosis, the nanoparticles will clump together to create cytotoxic aggregates [9, 10, 11].

When compared to other nanomaterials, gold nanoparticles (AuNPs) have a higher stability and are more biocompatible. This makes them one of the leading options for usage in biomedical applications. They are simple to synthesize and describe, and they are able to be conjugated with a wide variety of chemical and biological components in order to provide diagnostic, therapeutic, and delivery systems that are biocompatible, targeted, and under control. The destiny of AuNPs after they have served their medicinal function as well as the possible dangers they represent to both human and environmental health, however, be determined [12].

The most notable quality of metal nanoparticles is that, when used as a carrier, it may improve the organ enrichment of ions. This property not only enables the widespread employment of metal nanoparticles in the treatment of cancer but also in the development of biomedical imaging technologies. On the other hand, once nanoparticles have gathered in the organs, it may take a long time for them to be cleared out, and they may have a harmful impact if they are allowed to remain there. As a result, concerns have been raised regarding the possibility of danger posed by the utilisation of nanoparticles in medical applications [13].

Structure and Significance of Spleen

The spleen is an organ that range in color from dark red to blue-black and is found in the left cranial abdomen. The spleen can have a variety of gross appearances and can range in size, as a result it does not include any afferent lymphatic channels. It is made up of two unique parts, the red pulp and the white pulp, each of which have their own individual functions and morphologies [14].

The red pulp acts as a filter for the blood, removing foreign particles as well as damaged or defective erythrocytes [15]. Additionally, it serves as a repository for iron, erythrocytes and platelets in the body. It is a location of hematopoiesis in rodents, notably in fetal and neonatal animals of the species [14]. The white pulp, which is anatomically analogous to a lymph node and contains T cell and B cell zones, is found inside the fruit (the latter are also called follicles) [15].

Additionally, the spleen is the biggest secondary lymphoid organ in the body [16, 17] comprising approximately one-fourth of all the lymphocytes in the body and being responsible for the initiation of immunological responses to blood-borne antigens. The white pulp that surrounds the central arterioles is responsible for performing this job. The periarteriolar lymphoid sheath (PALS), the follicles, and the marginal zone are the three subcompartments that make up the white pulp [14]. The spleen's primary role is to remove dead, dying, and necrotic cells from the body, in addition to its contribution to the immune response of the body as a whole [9].

In addition to its role in the immune system, the spleen plays a crucial role in a number of other physiological processes, including iron homeostasis. Red blood cells that have become senescent or ruptured are collected in the spleen, where their haemoglobin content is converted into free iron. This provides the majority of the iron that is required for the process of erythropoiesis. [18, 19]. Extra medullary hematopoiesis occurs extensively in the adult spleen as well [17]. When the bone marrow needs to produce more blood cells due to an infection, anaemia, or a hereditary blood problem, or because of pregnancy, hematopoietic stem cells (HSC) move into splenic niches and begin differentiating into blood cells [20]. The spleen stores red blood cells, platelets, thrombocytes, plasmablasts, and long-term memory B cells [21].

Studies conducted in vitro indicated that AgNP hinder embryonic stem cell development and have a detrimental impact on cells due to their influence on cellular metabolism and membrane integrity [22].

Exposure Pathway of Nanoparticles

Animals can be exposed to NPs intentionally or unintentionally via organs that directly interact with the environment, such as the respiratory tract via inhalation, the skin via penetration, or the gastrointestinal tract via ingestion [23-25] as shown in Fig. 2. In addition, intravenous injections are one of the most common protocols utilized in the field of nanomedicine in order to access the tissues that are the focus of treatment. There have been reports of skin tissue exposure following the use of NPs as wound dressings. [26]. NPs have the ability to cross the barriers in the lungs and enter the bloodstream, where they may then spread throughout the body and enter organs such as the spleen [26-29]. Despite their diminutive size, nanoparticles are very hazardous [30].

Mechanism of Toxicity of Nanoparticles

NPs can have a variety of toxicological consequences, including at least one of the following: ROS generation within or outside the cell, NP produced ROS altering cell membrane integrity, particle dissolution influencing cellular function following NP internalisation, mechanical injury to subcellular units, or NP dissolution outside the cell, which can compromise its integrity [31]. The most of these events result in oxidative stress and the initiation of an inflammatory response in the spleen. [26]. The toxicity of metallic NPs, such as Ag, Cu and gold has been characterized by their ability to increase ROS generation, which results in oxidative stress and, in some models, triggers apoptosis, oxidative DNA damage, membrane cell damage, DNA adducts, gene mutation, and protein oxidation, all of which lead to cytotoxicity and genotoxicity in spleen cells [26, 32] as shown in Fig. 3.

Effects of Silver Nanoparticles on Rat Spleen

The investigation of the toxicological consequences of silver nanoparticles has taken place despite the fact that their expanding usage in biological applications provides a number of benefits. It is possible for

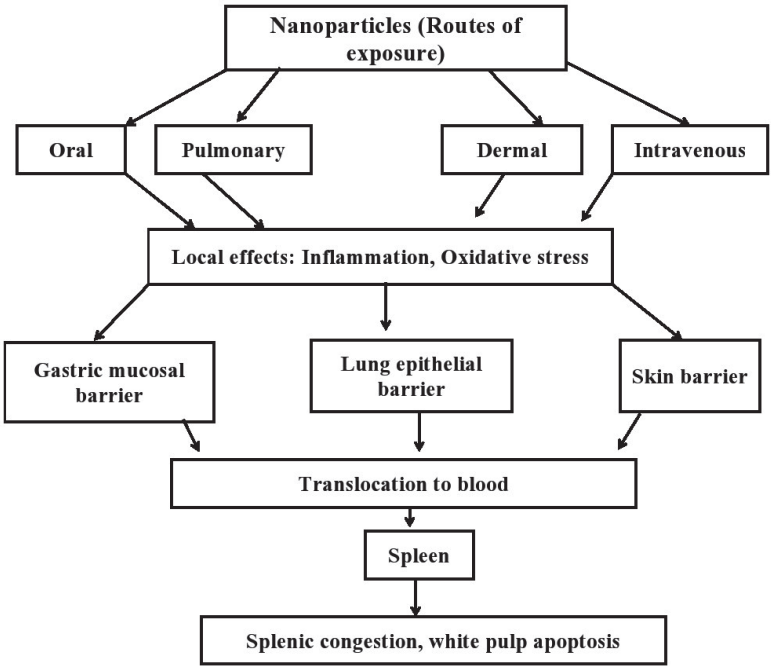

Fig. 2. Schematic representation of toxicity of silver nanoparticles following various routes of exposure.

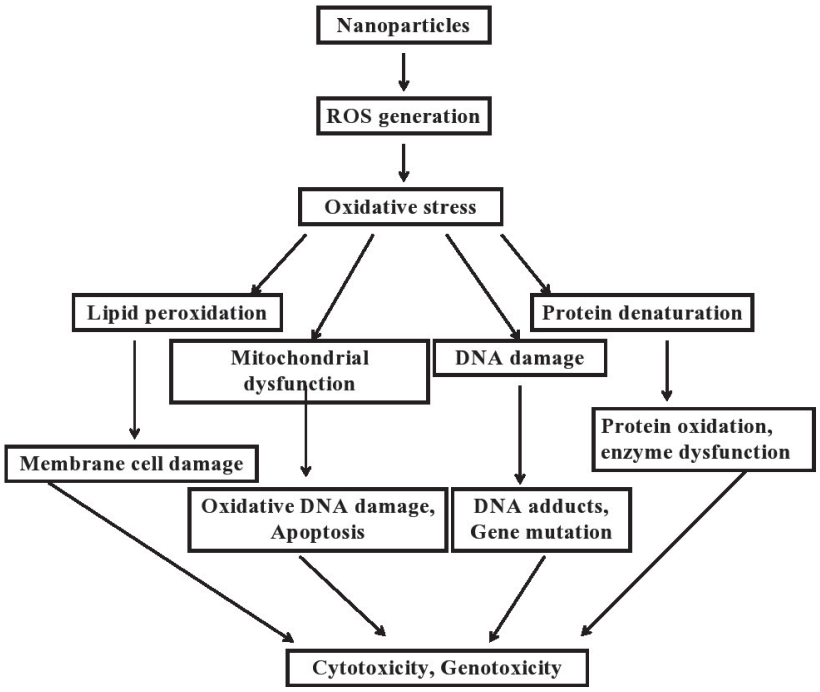

Fig. 3. Mechanism of Toxicity of Nanoparticles.

silver nanoparticles to enter the body by a variety of routes and to collect in a variety of tissues and organs, including the spleen [26]. In comparison to the toxicity of other nano sized metals, the toxicity of silver nanoparticles is significantly higher due to the elevated generation of reactive oxygen species (ROS) and the leakage of the enzyme lactate dehydrogenase [33]. Numerous variables, including the physical and chemical characteristics of the silver nanoparticles, the environment and contact interactions have been shown to affect the toxicity of silver nanoparticles [34].

Histopathological abnormalities were seen in the spleen as a result of exposure to AgNPs. These changes included vacuolation of cells with ill-defined white pulp, altered lymphoid architecture and congestion of red pulp. It was shown that oxidative stress and inflammation were to blame for the histopathological toxicity alterations found in the spleens of albino rats [35]. Alterations in the spleen's color and atrophy were seen in rats after exposure to silver nanoparticles. Histopathological examination revealed that a high-dose treatment with silver nanoparticles led to the development of adverse effects on the spleen in a particular patient group. An assessment by a pathologist revealed that there was damage to the tissues, blood loss, cell necrosis, and apoptosis [30].

The spleen of rats that had been treated with AgNPs showed structural changes that were dose-dependent. These alterations included a reduction in the size of the white pulp follicles, as well as a depletion of lymphocytes, degeneration, and apoptosis. The spleen had structural changes that were consistent with an inflammatory response and oxidative stress after exposure to AgNPs [36]. An investigation by histopathology revealed congestion, bleeding, cellular degradation, apoptosis, and necrosis in the tissue of the liver and kidneys, in addition to lymphocytic depletion with growing visible macrophages in the spleen [37].

Effects of Copper Nanoparticles on Rat Spleen

There is still insufficient evidence on the possible risks of exposure to Cu-based NPs compared to other NPs, despite the broad uses and expanding availability of nanoproducts containing copper. [38]. Cu-based NPs cause cytotoxic effects in a variety of cell types, including epithelial cells, that are accompanied by an increase in reactive oxygen species [39]. In HL60 cells, copper nanoparticles (NPs) exhibited a greater toxicity than its oxide nanoparticles (CuO NPs) [40]. But few studies have examined in vivo toxicity, and those that have found biochemical and histological changes in the liver, kidney and spleen following a single or brief exposure to CuNPs [41, 42].

Nano-Cu produced alterations in structure and function of the spleen, additionally oxidative stress and inflammation were detected that promoted the activation of key regulatory pathways. Nano-Cu has also influenced the numbers of B and NK cells, as the number of B cells fell, the expression level of antibodies for IgA, IgG, and IgM were dramatically lowered. Nano-Cu significantly interfered with cellular and humoral immune processes in rat spleens, that leads to lower disease resistance in animals [9]. Rats treated

with Cu NP exhibited the presence of multinucleated cells in the spleen [7].

Effects of Gold Nanoparticles on Rat Spleen

Due to its numerous beneficial qualities, including adjustable sizes, simple production, easy modification and strong optical properties, gold nanoparticles have been employed extensively in the biomedical area [43]. The findings on the toxicity of gold nanoparticles are currently contradictory. According to some researchers, these particles are biocompatible and have minimal toxicity [44, 45]. While others highlighted their severe toxicity due to their physicochemical characteristics and the ROS they produce, which causes oxidative stress [26, 46, 47].

Exposure to AuNPs may occur during their development and synthesis or during application like direct ingestive intake, intravenous injection and waste disposal. Other exposure pathways include dermal absorption, inhalation, through implants, adhesion of airborne and surface contaminants that become difficult to detect and dermal absorption. It is generally believed that AuNPs accumulate at large levels in the liver and spleen, further affecting the body [48].

AuNPs accumulated in all of the tissues, although they were more prevalent in the mononuclear phagocyte system (also called the reticuloendothelial system). The spleen were found to have the highest level of accumulation. The majority of the AuNP-containing macrophages in the spleen were found in the red pulp of mice, with rats having a significantly larger concentration of AuNP-containing macrophages than mice [12, 49]. In most cases, reports describe the effects of AuNP on the immune system as being either minor or anti-inflammatory. AuNP has the ability to prevent the activation of inflammatory reactions that were generated by IL-1; however, the effect was size-dependent on AuNP [50]. After receiving AuNP, the spleen had a much higher number of genes that had been down regulated as opposed to those that had been upregulated. The genes that have been down regulated include those that are associated to responses to external stimuli, responses to injury, defensive responses, wound healing, and blood coagulation [49].

Conclusion

This article analyzed the research on the toxicity of metallic nanoparticles after exposure via inhalation, instillation, oral, cutaneous, and intravenous routes/pathways into rat's spleen. There is a lack of knowledge on the possible impacts of NP exposure on animal models and the mechanisms of action of NPs are currently poorly understood. To develop Nano medical technology into clinical practice, Nano toxicological studies are essential. This article concludes that silver nanoparticles causes more toxicity than copper and gold nanoparticles hence causing severe effects on rat's spleen.

Conflict of Interest

The authors declare no conflict of interest.

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