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Toxic Effect of Silver Nanoparticles on Some Hematological Parameters and Possible Preventive Role of *Moringa Oleifera* : In Vivo

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Abstract
Toxicity research is conducted on animals in vivo, specifically mammals like rats, since they have a biological configuration similar to humans. Since the last ten years, the total of those research that have studied and analyzed the toxic influences of NPs has jumped up for the presence of these particles in several consumer products for humans . In this study, 24 male rats were divided into 4 groups, and each group comprised 6 rats. The control group (CON) were healthy rats without any treatment. Whereas, (AgNPs) group received 50 µl / kg silver nanoparticles for 30 days, and in (AgNPs + MO) group, rats were co-administered from the combination of AgNPs at a dose of 50 µl / kg with *Moringa Oleifera* at a dose of 300 mg / kg for 30 days. Experimental animals of the (MO) group were treated with 300 mg / kg of MO for 30 days. Some blood parameters were measured using an automatic blood analyzer. Where the rats treated with AgNPs showed a tremendous decrease ($p < 0.05$) in red blood cell counts (RBCs), hemoglobin concentration (Hb), and hematocrit value (Hct), in contrast to white blood cell counts contrasted to the control group. Then, the co-administration of *M. Oleifera* with AgNPs had a significant role and influence on haematological disorders. As a conclusion, these results demonstrated that *M. Oleifera* had a positive effect against the haematological toxicity of AgNPs in male rats.

Keywords: *Silver Nanoparticles, toxicity, blood parameters* .

Introduction

Due to the unique properties of nanotechnology, its application in medicine has made a great revolution in the fields of health for its ability to improve some medical diagnoses as well as treat and prevent diseases[1-4]. Among the metallic nanoparticles are silver nanoparticles (AgNPs) that have antimicrobial, catalytic and other properties, which are firmly involved in medicine and pharmaceutical applications[5-7]. On the other hand, there are growing concerns about the toxicity of nanoparticles to human health[8]. Nanotoxicology is a modern scientific branch that aims to clarify the potential negative effects of nanoparticles and related parameters affecting the cytotoxicity of nanomaterials[9,10]. Herbs are used mainly in medicine for the purpose of treating and preventing many diseases and toxins due to their acceptance in the human body and their few side effects[11-13]. *Moringa Oleifera* belonging to the Moringaceae family is known as the miracle tree, for being rich in nutritional, medicinal and immunity values in humans and animals globally [14-16]. As it contains a large number of essential elements, minerals and amino acids. In many developing tropical countries, it is used to reduce nutritional deficiencies in the human body[17,18]. This study aims to clarify the negative effect of silver nanoparticles on some blood parameters as well as to evaluate the positive role of *M. Oleifera* against the pathological changes caused by AgNPs in some hematological parameters.

Material and Methods

Chemicals

Silver (Ag) Nanoparticles Water Dispersion was purchased from US Research Nano-material , Inc. (HOUSTON , TEXAS,USA). Average particle size (APS) :15 nm, concentration Ag : 5000 ppm. Mono-Nano powder ,color :Tawny. PH :7± 0.5 , purity :

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99.99% . As for *Moringa Oleifera*, it was obtained in powder form from GreeNater PREMIUM, USA, and it was dissolved in a normal saline solution prior to experiment. Rats were given 300 mg/kg [19] daily for 30 consecutive days, orally, via gastric gavage.

Experimental Design

This study was conducted on (24) adult male albino rats with ages ranging between 4-5 months and weights of 190-220 g. Rats were housed in plastic cages and under typical conditions at 25 ± 2 ° C and 12 h light-dark cycles. Animals are permitted access to feed and tap water. They were randomly divided into 4 groups, and each group included 6 rats. Control (CON) healthy group; (AgNPs) group, animals received AgNPs; (AgNPs + MO) , co-treatment group received *M. oleifera* extract plus AgNPs ; (MO) group , rats received *M. oleifera* extract only . The rats received AgNPs at a dose of 50 µl / kg / day intraperitoneally, for the duration of the trial.

Heamatology

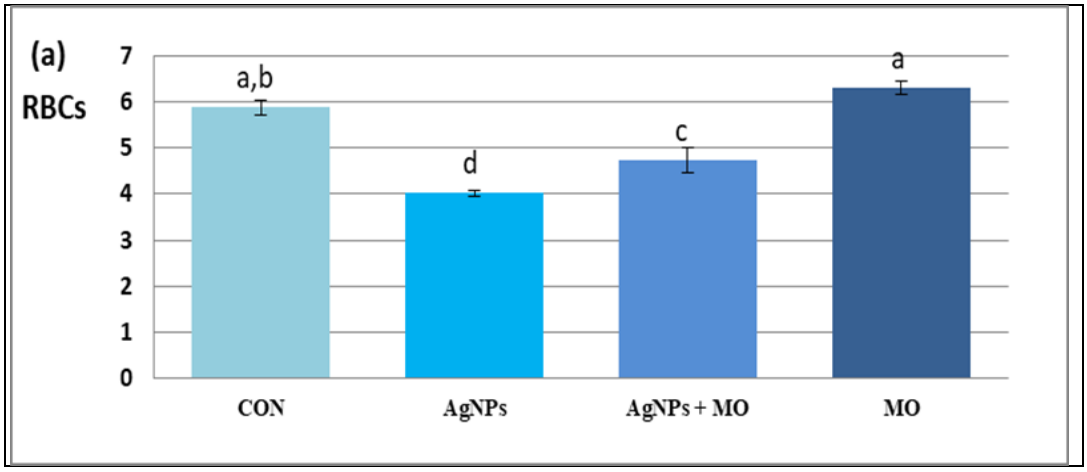
Rats' blood samples were collected, then anticoagulation was done and blood parameters were measured by an automated blood analyzer. Parameters included red blood cell count (RBCs), hemoglobin concentration (Hb), hematocrit value (Htc), as well as white blood cell count (WBCs).

Statistical analysis

The results obtained were based on the mean ± standard deviation (N = 6). The statistical significance analysis between the different groups was done using one-way analysis of variance (ANOVA) adopted to Duncan analysis.

Results

When compared with the control group, AgNPs treated rats showed lower values for RBCs counts, Hb concentration, Htc value . While WBCs counts showed higher values when comparing to the animals of the controlling group. In addition, the results showed that there was ineffective change in hematological parameters with the co-administration of *M. oleifera* extract when compared to toxic rats with AgNPs (P <0.05). As shown in figure (1).



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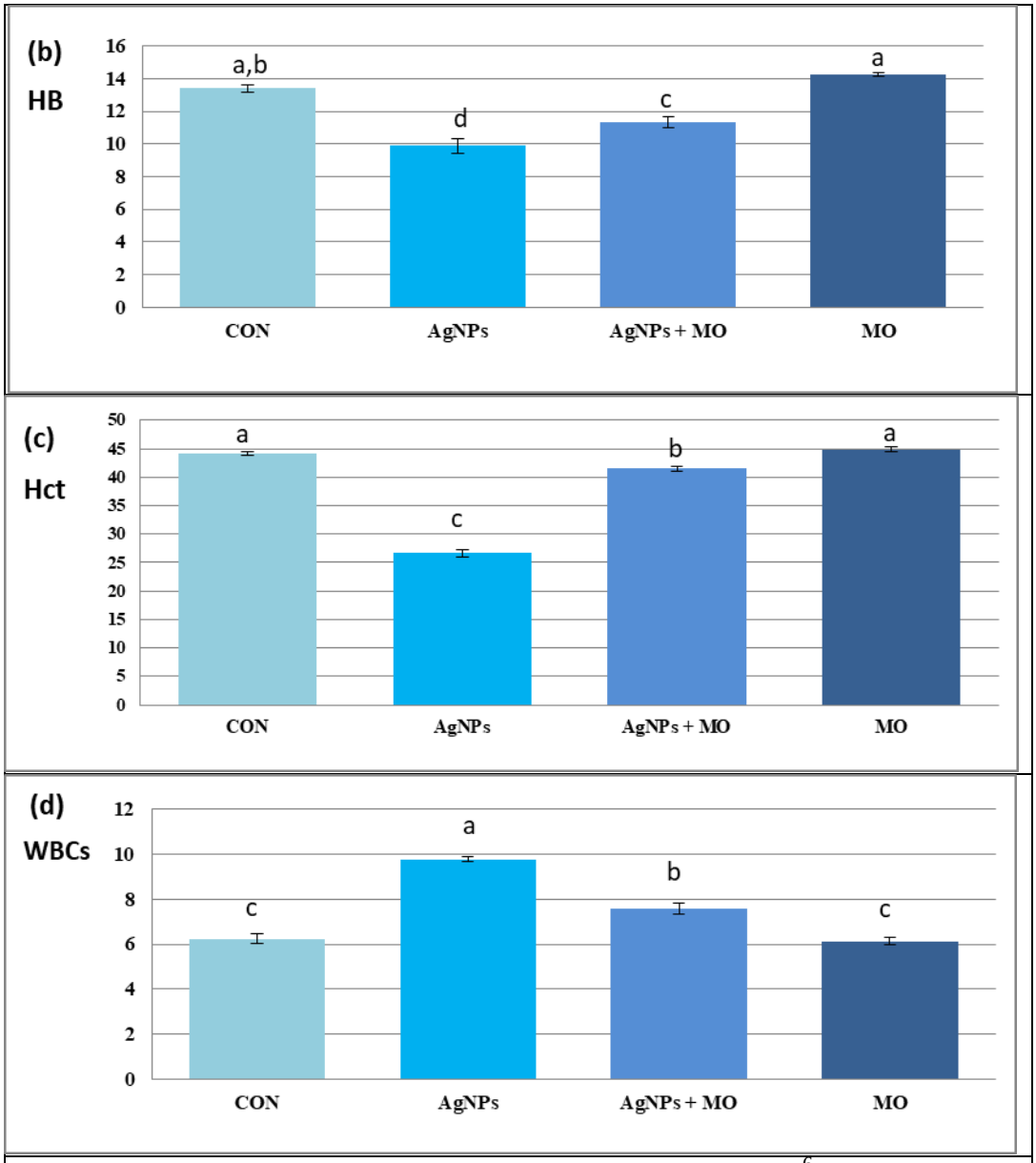

Figure (1): Levels of (a) red blood cells count RBCs ($\times 10^6$ cells / µl), (b) hemoglobin Hb (g / dL),(c) hematocrit Hct value (%), and (d) white blood cells count WBCs ($\times 10^3$ cells / µl) in the blood of the different treatment groups. Data were expressed as mean ± SD (n = 6). $^{a-d}$ various superscript letters denote essential differences within column ($p < 0.05$) test = one-way ANOVA, after hoc = Duncan.

Discussion

Nanotoxicology is an important part of bio-nanoscience, as it investigates the nanoparticles toxicity[20]. Investigating the nanotoxic effects of Ag NPs is essential to the many areas of use for these NPs such as pharmaceutical applications, systems provided for purifying water, and processes of packaging food, making them such a desirable material for humanity [21-23]. The key objective of nanotoxicity research is to determine the properties of NPs that pose a threat to living organisms[24]. Toxic features of NPs can possibly occur when the particle surface cooperates with cellular components depending on the decrease of NPs diameters [25]. At the cellular level, the mechanisms of absorption and the efficiency of NPs are essential factors

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that influence toxicity. NPs pierce the cell through numerous methods including phagocytosis and pinocytosis counting on particle magnitude and surface characteristics. The range from 10 to 100 nm is considered fit for cell uptake[26,27]. On the issue of dosage, it remains unresolved although it is critical to understanding and comparing toxicological data.

The current study revealed a significant reduction in the RBCs count after administration of NPs compared to the control group, as a result of the occurrence of anemia. This indicates the damage of red blood cells and a decrease in the ratio of erythropoiesis . RBCs are created by blood stem cells in the bone marrow through the maturation process with the help of erythropoietin. Therefore, any disturbance in the RBCs count can be caused by the hematopoietic system. As for a decrease in hemoglobin content, it could be associated with a decrease in the erythrocyte count, which in turn indicates the induction of anemia [28,29] . The hematocrit value is a major indicator of blood viscosity. If it is less than normal, then it is an indicator of anemia. This may result from a low red blood cell count, or a low hemoglobin content in all red blood cells, or both. It may result from damage to the myeloid tissue by chemical compounds such as toxins[30]. White blood cells play an essential role in the human body's immunity and defense against antigen invasion. They are usually formed by the body in response to any foreign substance invasion[31].

These hematological parameters showed significant improvement in the AgNPs + *M. Olifera* group when compared to the AgNPs group. This can be attributed to the phytochemical components in the extract as well as the presence of vitamins and minerals. These phytochemical components are well known to be hematopoietic factors that directly affect the production of blood cells in bone marrow [32,33].

Conclusion

This study concluded that silver nanoparticles are toxic to blood cells. The administration of plant extracts of *M. oleifera* to rats was beneficial to the blood, and effective against blood disorders caused by silver nanoparticles.

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