

**Review:**

**Daily used silver nanoparticles induced persisted accumulative genotoxicity and mutagenicity  
"Silver nanoparticles genotoxicity and mutagenicity"**

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**Abstract**

Incredible rapid growth in the uses and applications of Silver (Ag) nanoparticles increases human exposure and the risk of these nanoparticles. Despite the contradictory results obtained on Ag nanoparticles induced toxicity, the genotoxicity and carcinogenicity induced by Ag nanoparticles have been shown in numerous studies. Therefore, the *in vivo* genotoxicity and carcinogenicity of daily used Ag nanoparticles were reviewed in this study.

Keywords: Ag nanoparticles; genotoxicity; carcinogenicity; persisted; accumulative; *in vivo*

**1. Introduction**

Silver (Ag) nanoparticles are promising anti-bacterial / antifungal agents due their potent anti-bacterial activity and have therefore been used in a huge range of products manufacturing such as cosmetics, clothes, air fresheners, surgical and artificial limbs and wound dressings. Along with their potential uses in food packaging to increase the shelf life of the food products (Gottesman *et al.*, 2011; Ribeiro *et al.*, 2013). Moreover, the uses of Ag nanoparticles are not limited to industrial applications, but also extend to environmental uses such as drinking water

disinfection, anti-fouling pools and as antibacterial supplement for existing water paints (Lv *et al.*, 2009; Holtz *et al.*, 2012).

Moreover, Ag nanoparticles are a notable nano-product with potent applications in medicine and hygiene because of their antiviral actions, antibacterial effects and antifungal activity (Elechiguerra *et al.*, 2005; Kim *et al.*, 2007; Mehrbod *et al.*, 2009) as well as they promote wound healing by affecting the cytokine function. In addition, Ag nanoparticles have proven to have antitumor, anti-inflammatory and gene therapy carrier's

effects (Shin et al., 2007; Sriram et al., 2010).

Once Ag nanoparticles enter a cell, they react with various biological molecules such as sugars, proteins, lipids and even nucleic acid since can cross the nuclear membrane and thus interfere directly with the function and structure of genetic DNA (Carinci et al., 2003; Jeon et al., 2011). Consequently, the assessment of engineered nanoparticles toxicity cannot be based on studying the toxicity data from larger particles and comparison of the obtained results. Many studies have been demonstrated the genotoxicity and carcinogenicity of Ag nanoparticles both in vitro and in vivo and these were discussed in the following points.

## 2. Genotoxicity

Genotoxicity of Ag nanoparticles has been studied in numerous studies and discovered that Ag nanoparticles induced genotoxicity is highly dependent on their size and surface charge as the size of nano-Ag particles affects their distribution, uptake, excretion and metabolism while, their surface charges changes their hydrodynamic size distributions and thus affecting nanoparticles agglomeration, absorption and transport within organisms (Renwick et al., 2001; Lockman et al., 2004; Choi et al., 2007).

Generally, the induced damage can occur on the chromosomal, DNA or even on the gene level. The induced DNA damage can be in the form of single- and/or double-strand

breaks, loss of excision repair, cross-linking, alkali-labile sites and point mutations (ECVAM, 2002).

### 2.1 Chromosomal damage

Induction of chromosomal damage by Ag nanoparticles has been studied by several studies using chromosomal aberrations and micronucleus assays and found that nano-Ag induced a dose and time-dependent chromosomal damage (Ordzhonikidze et al., 2009; Patlolla et al., 2010; Tiwari et al., 2011; Mohamed, 2016).

However, the available data on the nano-Ag induced chromosomal damage are contradictory. Several studies uncapped nano-Ag particles did not induce chromosomal damage in Sprague-Dawley rat bone marrow cells after 28 days of oral administration and after 90 days of inhalation exposure using micronucleus assay (Kim et al., 2008; Kim et al., 2011) as well as Ag nanoparticles capped with starch, polyvinyl alcohol and bovine serum albumin were also not genotoxic to zebra fish embryos in the zebra fish model (Asharani et al., 2008; Asharani et al., 2011) and these results are in consistence with the results of Li et al. (2013) study showed that silver nanoparticles coated with either poly vinyl pyrrolidone or silicon isn't genotoxic and didn't cause any increase in the micronuclei or *Pig-a* frequency in mice bone marrow and liver cells.

On contrary, other recent studies demonstrated the induction of chromosomal

damage by Ag nanoparticles as manifested by the significant increases in the levels of structural chromosomal aberrations, the micronuclei frequency and low mitotic index observed in bone marrow cells of rats injected intraperitoneally or orally administered nano-Ag particles indicating potential genetic toxicity of Ag nanoparticles and the need for further characterization of their systemic toxicity, genotoxicity and carcinogenicity (Patlolla et al., 2015).

Furthermore, nano-Ag induced clastogenicity was shown recently by the significant increases in the micronuclei frequencies observed in bone marrow cells of mice sacrificed 24 hours after intraperitoneal injection of the three fractions 1/100, 1/50 and 1/25 of nano-Ag LD50 (Mohamed, 2016).

## 2.2 DNA damage

Because of higher antimicrobial activity of Ag nanoparticles compared to their bulk, their uses and applications highly increased compared with the normal sized Ag and thus increasing human exposure and raised the scientific attention to their genotoxic risk. Numerous studies demonstrated the DNA damage induction by Ag nanoparticles using comet assay in the testicular seminiferous tubules and in the lung of rats and in mouse spleen (Ordzhonikidze et al., 2009; Cho et al., 2013).

Intravenous injection of Ag nanoparticles at the dose level 40 mg/kg b.w increased the induction of single and double

DNA breaks in rats. Furthermore, oral administration of Ag nanoparticles to zebra fish induced DNA damage as revealed by the high levels of  $\gamma$ -H2AX (a marker for DNA double-strand breaks) (Choi et al., 2010; Tiwari et al., 2011). In addition, using RAPD assay nano-Ag has been shown to induce fragmentation of the genomic DNA in vivo even at the dose close to LOAEL (Katsnelson et al., 2013).

Recently, Ag nanoparticles have been shown to weak the stability of sperm chromatin, damage seminiferous tubules and stimulate oxidative DNA damage and thus resulting in disruption of sperm at any stage of cell differentiation as a result of the inhibitory role of Ag nanoparticles on cells proliferation that affect cell cycle causing a significant reduction in the sperm precursors cells or their release into the mid duct of seminiferous tubules (Attia, 2014; Takeda et al. 2009).

Induction of DNA damage by nano-Ag particles has also been demonstrated recently by the significant increases reported in tail length, %DNA in the tail and tail moment in mice injected intraperitoneally with each of the three dose levels of nano-silver (20, 41 or 82 mg / kg) in a dose-dependent manner (Mohamed, 2016; Mohamed, 2017).

However, Li and his colleagues (2013) have shown contradictory results as poly vinyl pyrrolidone- and silicon-coated Ag nanoparticles did not induce any DNA strand breaks demonstrated using the standard comet

assay but the same nanoparticles were found to cause DNA breaks in the enzyme-modified Comet assay in mouse liver cells.

### 3. Mutagenicity

The unique physico-chemical characteristics of Ag nanoparticles in particular their small size enable them to penetrate the biological cell and the nuclear membrane reaching macromolecules molecules: protein, fat and nucleic acids and thus interact with them not only on the chromosomal and DNA level but extend also to the gene level inducing mutations and altering expression level of mutated genes (**El ghor et al., 2014; Mohamed, 2017**).

Recently, Ag nanoparticles coated with polysaccharides have been shown to up-regulate the expression level of heat shock protein 70 as well as elevated the expressions of cell cycle checkpoint p53 protein and cell signaling p38 protein that are involved in the repair pathway of DNA damage in *Drosophila melanogaster* and thus increased and enhanced DNA damage and apoptosis in *Drosophila* exposed in reverse to a non-significant dose-dependent increase observed in hepatic p53 mRNA level after nano-Ag exposure (**Ahamed et al., 2010; Choi et al., 2010; Kobayashi et al., 2009**).

Furthermore, Ag nanoparticles have changed differentially the expression levels of genes in the frontal cortex, caudate nucleus, and hippocampus of the nano-Ag treated mice as Ag nanoparticles upregulated the modified

genes in the caudate, while in the hippocampus the altered genes were down-regulated and thus Ag nanoparticles induced neurotoxicity can be explained by the free radical-caused oxidative stress and alteration of genes expression that mediated apoptosis and neurotoxicity (**Rahman et al., 2009**). Similarly, Ag nanoparticles induced mutations in the p53 and presenilin genes and altered the expression of p53 gene in the liver, kidney and brain of mice injected intraperitoneally with the three dose levels 20, 41 and 82 mg/kg of Ag nanoparticles (**Mohamed, 2017**).

Moreover, oral exposure to Ag nanoparticles even at the low doses also reduced the expression levels of important immunomodulatory genes including MUC3, TLR2, TLR4, GPR43 and FOXP3 altering mucosa associated microbiota and modulating the gut associated immune response and the overall homeostasis of the intestinal tract (**Williams et al., 2014**).

### 4. Conclusion

Unique physico-chemical characteristics of Ag nanoparticles rapidly increase their uses in food, medicine and industry and thus increase their daily human exposure and risks. Despite, the available results on the genotoxicity of daily used Ag nanoparticles are contradictory; the chromosomal and DNA damage and mutagenic effects of Ag nanoparticles were evident in numerous in vivo studies as well as their genotoxic and mutagenic effects are

largely dependent on particles size, the form of nanoparticles, preparation method, assembly grades, incubation conditions, coatings and dosages used. Thus, it is recommended to reduce the uses of Ag nanoparticles alone or to use the natural products with nano-Ag to minimize the side effects of these nanoparticles on humans.

## 5. Conflict of interest

Author declared no conflict of interest

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