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STUDY OF ANTIOXIDATIVE ACTIVITY OF SILVER CHARGED WATER AGAINST GAMMA RADIATION IN SWISS ALBINO MICE

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ABSTRACT

Silver charged water; in which silver is present in nanoparticulate form is used as an Ayurvedic medication from ancient time. Study was done to evaluate the changes in the physiochemical parameters in Silver charged water and biochemical changes after 5 Gy whole body irradiation and its modulation by supplementation of silver charged water on male Swiss albino mice brain. For this, healthy mice from an inbred colony were divided into four groups: (i) Control-vehicle treated (ii) Silver charged water treated-mice in this group were orally supplemented with silver charged adlibitum for one month; (iii) Irradiated (iv) Silver charged water + irradiated- mice in this group were orally supplemented with Silver charged water for one month prior to irradiation. Marked radiation induced changes in the amount of whole brain lipid peroxidation (LPO), Glutathione (GSH) are significantly ($p < 0.001$) ameliorated specially by supplementation of silver charged water prior to irradiation. The results of present study showed that prior supplementation of silver charged water has radioprotective potential as well as neuroprotective properties against the radiation.

Keywords: *Whole Brain, Silver Charged Water, Oxidative Stress, Lipid Peroxidation, Glutathione, Silver Nanoparticles.*

Introduction

The use of atomic energy in the national economy and the incidents of radiation borne hazards on the human population are continuously increasing. Radiotherapy, which is a chief modality to treat cancer, faces a major drawback because it produces severe side effects generated due to damage to normal tissue. Rapidly growing interventional radiological procedures such as dilatation of stenosed vessels or recanalization or vascular Angio anastomoses necessitates a search for radioprotector that has multi dimensional advantages.

The free radicals generated by the action of radiation on water present in human tissues, react with cellular macromolecules, such as DNA, RNA, proteins, membrane, etc, causes cell dysfunction and mortality (Nair *et al.,* 2001). For years researchers have known that free radicals can cause cell degeneration, especially in the brain and leads to oxidative stress in brain. The brain and nervous system are particularly vulnerable to oxidative stress due to limited antioxidant capacity. The brain makes up about two percent of a person's mass but consumes 20 percent of their metabolic oxygen. The vast majority of this energy is used by the neurons (Shulman *et al*., 2004). Researchers studying antioxidant protection of neurons are finding short windows during development of high vulnerability to oxidative stress (Perry *et al.*, 2004). All of these facts tend to indicate that brain should be a tissue that is highly susceptible to oxidative damage.

The rapidly developing field of nanoscience has raised the possibility of using therapeutic nanoparticles in the diagnosis and treatment of human cancers (Yezhelyev *et al.,* 2006). These nanoparticles have been the subject of substantial research in recent years (Li et al., 2001, Iglesias-Silva et al., 2007 and Huang and Yang, 2008).

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Radioprotective potential of nanoparticles has been observed in various studies. Nanoparticles such as cerium oxide nanoparticles, yttrium oxide nanoparticles, carbon nanoparticles, etc. were found to possess antioxidant properties (Rzigalinski, 2005; Ali et al., 2004; Tarnuzzer et al., 2005; Trajkovic et al., 2007; Daroczi et al., 2006; Schubert et al., 2006; Deshpande et al., 2005).

Healing with metals is an ancient Ayurvedic therapy. Silver charged water, in which silver is present in nanoparticulate form is used as an Ayurvedic medication.Gold water, silver water, copper water are water charged with the healing qualities of these metals. It is known that copper, silver and gold have health benefits for the human body. These health benefits can be transferred to the water which comes in contact with these metals. Silver has antibacterial, antiviral (Lara *et al.,* 2010; Sun *et al.,* 2005; Jerley and Margret, 2012) and antifungal properties. Silver nanoparticles have been shown to possess excellent free radical scavenging and antioxidant activities (Banerjee and Narendhirakannan, 2011; Mittal *et al*., 2012; Inbathamizha *et al.,* 2013). Topical use of nanocrystalline silver cream has antiinflammatory and antimicrobial effect against allergic dermatitis as reported in guinea pig and rat model (Bhol *et al*., 2004; Bhol and Schechter, 2007). Antiinflammatory effects of silver nanoparticles have also been reported by Wong *et al.* (2009) and Nadworny *et al.* (2008).

The ionic silver kills harmful bacteria, viral, and fungal organisms just minutes after contact (Robert and Baker, 2009). An antibiotic kills perhaps a half-dozen different disease organisms, but silver kills some 650. Resistant strains fail to develop. Moreover, silver is virtually non-toxic (Powell, 1978). In a study, it was concluded that metal ions, such as silver ions, can destroy germs located both inside and outside of our human cells (Becker *et al.,* 1989)

Silver nanoparticles (AgNPs) are also known for their antimicrobial potential against several other viruses, including hepatitis B, (Lu *et al.*, 2008) respiratory syncytial virus (Sun *et al.,* 2008) and monkey pox virus (Rogers *et al.*, 2008). AgNPs and ions have been shown to possess intrinsic cytotoxic activity (Kim *et al.,* 2007, Baker *et al.*, 2005) and exhibit an enhanced antimicrobial effect when applied on silicon structures (Furno *et al*., 2004). Herpes simplex virus type 1 infection is inhibited by silver nanoparticles capped with mercapto ethane sulfonate (Baram-Pinto *et al.,* 2009).

Through its many biochemical actions, Silver nanoparticles may help in managing some of today's most pressing health concerns, including loss of memory, diabetes, poisoning, tissue wasting, fever, cachexia, urinary disorders, uterine disorder, epilepsy, alcoholism treatment.

Thus, in order to provide a novel insight into radioprotection, the effect of oral administration of Silver Charged Water before irradiation is evaluated, on organ dysfunction with reference to biochemical estimations in brain. The physiochemical parameters in silver charged water are also compared with the normal tap water. The efficacy of Silver Charged Water as nanoparticles, against radiation is studied so that it could be recommended to be used in daily routine by large number of people, could improve the general health, as it is hypothesized that silver water boosts the immune system. And also to evaluate that it could override pills against harmful side effects of radiotherapy.

Materials and Methods

Animals

The Departmental Animal Ethical Committee (DAEC) approved the study. Animal care and handling was done according the guidelines of INSA (Indian National Science Academy, New Delhi, India). Swiss albino mice (*Mus musculus*) (6-8 weeks old, weighing 25±2 gms) from an inbred colony were used for the present study. These animals were maintained under controlled conditions of temperature (25±2 ^{o}C) and light (light: dark, 14 : 10 hrs). Four animals were housed in a poly propylene cage containing sterile paddy husk (procured locally) as bedding through the experiment. They were provided standard mice feed (procured from Hindustan Lever Ltd. Mumbai) and water *ad libitum*.

Chemicals

Thiobarbituric acid was purchased from Sigma, USA. All other chemicals used were of analytical grade.

Source of Irradiation

The cobalt teletherapy unit (ATC-C9) at the Cancer Treatment Center, Radiotherapy Department, SMS Medical College and Hospital, Jaipur, Rajasthan, India, was used for irradiation. Un-anesethesized animals were restrained in well-ventilated Perspex box and whole body exposed to gamma radiation.

Characterization of Silver Nanoparticles

Characterization of silver nanoparticles was performed using scanning electron microscopy (SEM) (Carl Zeiss Evo 18 Special Edition). These techniques are used for determination of particle size, shape, crystallinity, fractal dimensions, pore size and surface area.

Physico-Chemical Analysis of Silver Charged Water

Silver charged water was analyzed to analyse the changes in the physico-chemical properties of water and compared with normal water, taken from the same source. Following parameters were analyzed as per the standard methods of American Public Health Association (1989) and Trivedi and Goel (1986)

- Temperature using Mercury Thermometer
- pH using a pH meter
- Electrical Conductivity with conductivity meter
- Total dissolved solids by Gravimetric method
- Dissolved Oxygen by Winkler's method
- Alkalinity (APHA, 1989)
- Acidity (Trivedi and Goel, 1986)
- Total hardness by EDTA method
- Chlorides by argent metric method

Procedure for Making Metal Charged Water

Silver metal was washed well with clean water. In a steel vessel, 4 to 5 cups of fresh and clean water was taken and the pure silver metal was dropped in this water. The water was kept overnight. The silver metal was removed from the water in the morning and this water was strained through a fine muslin cloth. The resultant water was the metal charged water.

Experimental Design

To study radioprotective efficacy of Silver Charged Water in adult mice, they were divided in to four groups (10 animals of 6 weeks in each):

Group 1 (Control) → Distill water *ad libitum*

Group 2 (Irradiated) → Single dose of 5 Gy of gamma radiation (dose rate of 1.07 Gy/min).

Group 3 (Silver Charged Water Treated) → Silver charged water *ad libitum* for 1 month.

Group 4 (SW + IR) → Silver charged water *ad libitum* for 1 month Single dose of 5 Gy of radiation.

Biochemical Studies

Biochemical estimations were carried out in adult mice whole brain at 7th day post- interval. Following methods were applied for the estimation of various parameters:

- Lipid peroxidation (Okhawa *et al*., 1979).
- Glutathione (Moron *et al*., 1979).

Removal of Brain Tissue

Animals of each group were humanly sacrificed by cervical dislocation and autopsied at various post irradiation intervals *viz,1-30*. To remove brain an incision was given at the sides of the jaws to separate the upper and the lower palates. The upper palate was cut in the middle and after having cleared the surrounding tissues; brain was excised and separated from the spinal cord at the decussation of pyramids. Whole brain was used to estimate various changes in biochemical parameters and cerebellum was fixed in bouins for paraffin sectioning and stained with hematoxyin, eosin for histopathological studies.

Biochemical Assay

- **Lipid Peroxidation (LPO) Assay:** LPO was measured by the method of Buege and Aust [31]. Briefly, 1.2 ml solution of TCA-TBA-HCl prepared in 1:1:1 was added to tissue homogenate (0.8 ml). This final mixture was heated on a water bath for 30 min at 80°C and cooled. After centrifugation the absorbance was recorded at 532 nm using a UV (ultraviolet)-Visible double beam spectrophotometer. The LPO has been expressed as malondialdehyde (MDA) in n mole/gm tissue.

- **Reduced Glutathione (GSH) assay:** The reduced glutathione (GSH) content of tissue samples was determined by the method of Moron *et al.* [32]. Tissue sample was homogenised in the sodium phosphate-EDTA (ethylenediaminetetraacetic acid) buffer then 0.6 M DTNB [5, 5_dithiobis-(2-nitrobenzoic acid)] was added. The optical density of the yellow coloured complex developed by the reaction of GSH and DTNB was measured at 412 nm using a ultraviolet (UV)-vis spectrophotometer. The results were expressed as nano mol/100 mg of tissue.

Statistical Analysis

The results obtained in the present study were expressed as mean ± Standard error of mean. Statistical difference between various groups were analyzed by student t-test and the significance was observed at P<0.001, P<0.005 levels.

Results

The study revealed significant changes in physiochemical properties of silver charged water in comparison to normal water (Table-1). The silver content is found to be 0.006mg/l in silver charged water whereas in normal tap water silver contents were not found. As silver is highly conductive, this energy is being utilized for water charging. Ancient civilizations, such as the Phoenicians and Greeks, knew that storing water and oils within silver containers preserved the freshness of the liquids for long periods of time. According to the National Academies, ionic silver solutions are used to disinfect the potable water on the International Space Station and the Russian "Mir" orbiting station. There are plentiful evidences supporting silver's ability to purify water and disinfect inanimate objects Li *et al*., (2010); Shahverdi *et al*.,2007; Kim *et al*.,2007.

Characterization of silver nanoparticles was performed using scanning electron microscopy (SEM) (Carl Zeiss Evo 18 Special Edition). These techniques are used for determination of particle size, shape, crystallinity, fractal dimensions, pore size and surface area (fig-1). The Morphology and particle size of the nanoparticle could be determined by SEM. The size of siver nanoparticles in silver charged water ranges from 20-200nm. Particle size and size distribution are the most important characteristics of nanoparticle systems (Figure.1). They determine the *in vivo* distribution, biological fate, toxicity and the targeting ability of nanoparticle systems (Panyam *et al*., 2003). Nanoparticles have particle size in the range between 1 and 100 nm and exhibit unique electronic, optical and catalytic properties (Tjong and Chen, 2004). Very often, nanometer-size metallic particles show unique and considerably changed physical, chemical and biological properties compared to their macro scaled counterparts, due to their high surface-to-volume ratio. Nanoparticles may or may not exhibit size-related properties that are seen in fine particles. Size is an important characteristic of nanoparticles because it is the main thing that differentiates them. Experiments on Swiss albino mice showed overall improvement in biochemical parameters when treated with silver charged water pre-irradiation. Irradiation raised the LPO levels by 57% in comparison to the normal group.

Oral supplementation with silver charged water oil prior to irradiation succeeded to bring the LPO level near normalcy (105 %) (Table-1). Oral supplementation of only silver charged water (Group-III) was also able to reduce the base line values approximately by 30 % (69.53%). Glutathione (GSH) content decreased sharply after radiation exposure (Group-II). In Group-IV GSH estimated was raised to the normal level compared to Group-III. Magnitude of such a recovery from oxidative damage was significantly higher (p< 0.001, p<0.005) in SW+IR group as compared to irradiated mice. Only SW treated mice also showed significant increase (p< 0.001) in GSH content as compared to control (Table-1).

Results obtained from this study indicate that silver charged water may act as a prophylactic agent and render protection against radiation induced oxidative stress. Oxidative stress leads to lipid peroxidation, protein and carbohydrate oxidation and metabolic disorders (Sies, 1985, 1986; Pryor and Godber, 1991). The product of LPO such as MDA (malondialdehyde) and 4-hydroxynonenal are toxic to cells (Raleigh, 1985). LPO within the membrane has a devastating effect on the functional state of the membrane because it alters membrane fluidity, typically decreasing it and thereby allowing ions such as Ca+2 to leak into the cell. The peroxyl radical formed through lipid peroxidation attacks the protein membrane and enzymes and reinitiates lipid peroxidation. The preservation of cellular integrity of the cellular membrane depends on protection or repair mechanisms capable of neutralizing oxidative reactions. Inhibition of LPO in cell membrane can be caused by antioxidants (Pryor *et al*., 1976).

In the present study the reduction in the amount of TBARS or MDA equivalents and elevation in the GSH level in the SO treated animals suggests that silver charged water may scavenge the free radicals generated during oxidative stress. GSH, with its sulfhydryl group, functions in the maintenance of the sulfhydryl group of the other molecule (especially protein), acts as a catalyst for disulfide exchange reactions and in the detoxification of foreign compounds like hydrogen peroxide and free radicals. The lesser depletion of brain GSH content in the SW +IR treated group compared to the irradiated group may be an indication of higher availability of GSH, which increases the ability to cope with the free radicals produced by radiation. Decreased brain GSH levels have been reported in neurodegenerative diseases such as Parkinson's disease and Alzheimer's disease, in which oxidative processes contribute to the pathology (Shukitt-Hale, 1998a and b).

The *in vitro* antioxidant properties of the biosynthesized silver nanoparticles from *Syzygium cumini* seed extract as reducing agent have been evaluated and these nanoparticles were found to have higher antioxidant capacity compared to the seed extract of *Syzygium cumini* and thus can be used as potential radical scavenger against deleterious damages caused by the free radicals (Banerjee and Narendhirakannan, 2011). Free radical scavenging and antioxidant activity of silver nanoparticles from Rhododendron dauricum (Mittal *et al*., 2012) have also been evaluated. Free radical scavenging and antioxidant activity of silver nanoparticles from Morinda pubescens (Inbathamizha *et al*., 2013) have been reported.

In-vitro free radical scavenging activity of biosynthesized silver (Ag-NPs) nanoparticles was investigated in a study by Dauthal and Mukhopadhyay, 2013. There is evidence that silver is a natural critical component of our immune system and that low tissue levels associated with a dietary deficiency may result in a relatively weakened immune system, making one more prone to infection. The modern versions of colloidal silver (oligodynamic silver) are shown to have little or no toxicity (www.thesilveredge.com).

Through its many biochemical actions, silver nanoparticles may help in managing some of today's most pressing health concerns, including loss of memory, diabetes, poisoning, tissue wasting, fever, cachexia, urinary disorders, uterine disorder, epilepsy, alcoholism treatment. Available routes of administration of silver nanoparticles include oral, nasal, parenteral or intra-ocular (Mohanraj and Chen, 2006). Silver has also been found to be non-toxic to humans at very small concentrations.

Because of the potential of free-radical-scavenging and antioxidant compounds to act as radioprotectants, silver nanoparticles can be seen to confer radio-resistance to normal cells during ionizing radiation treatment. Silver Bhasm and Silver Charged Water are already in use in Ayurvedic medication in oral form.

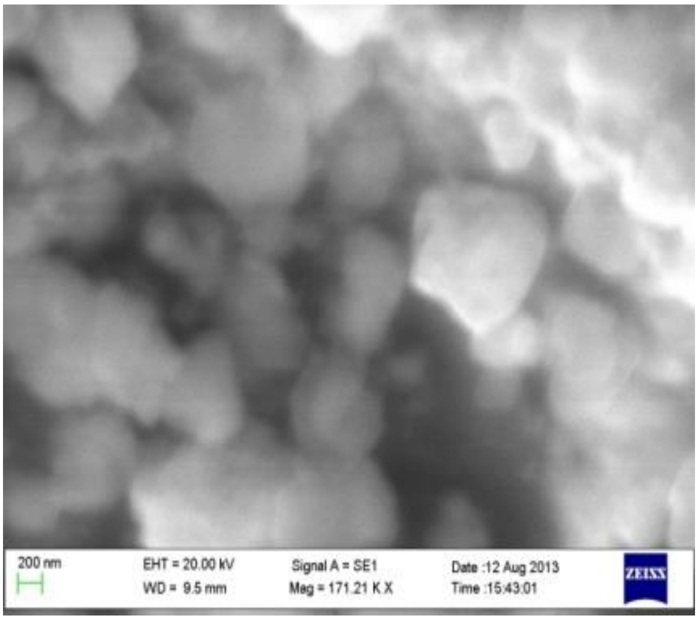
Silver nanoparticles are found to possess antibacterial, antimicrobial and antioxidant capacity and thus the possibility of silver nanoparticles in the form of water suspension to provide protection against damage due to free radicals generated by various toxicants, **cannot be ruled out**.

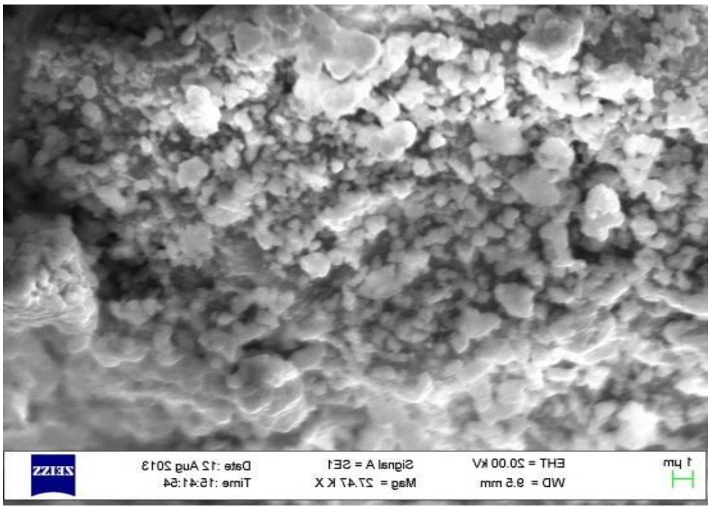
Parameters	Silver Charged Water	Normal Tap water
Silver Content (mg/l)	0.006	Not found
pH using a pH meter	8.02	7.62
Calcium hardness (mg/l)	60	56
Total dissolved solids by Gravimetric method(mg/l)	203	194
Chloride (mg/l)	56	40
Alkalinity (APHA, 1989) (mg/l)	124	196
Acidity (Trivedi and Goel, 1986) (mg/l)	4	4

Table 2: Variation in the LPO (nmol/gm) and GSH (nmol/100mg) Levels at 7th day post irradiation in the presence and absence of Silver charged water

	Normal	Control	SW	SW+IR
LPO	**141.65± 2.118 (100%)**	**222.5± 2.06 (157.07%)**	**98.5±0.594 (69.53%)**	**148.75±1.287 (105%)**
GSH	**18.31±0.987 (100%)**	**8.25±0.509 (45.05%)**	**18.55±0.505 (101.31%)**	**13.07±0.321 (71.38%)**

Characterization of silver nanoparticles Scanning electron microscopy (SEM) (Carl Zeiss Evo 18 Special Edition





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References

1. Nair, C.K.K., Parida, D.K. and Nomura, T., 2001. Radioprotectors in radiotherapy, Journal of Radiation Research. 42, 21–37.
2. Yezhelyev, M.V., Gao, X., Xing, Y., Hajj, A.A., Nie, S., Regan, R.M.O., 2006. Emerging use of nanoparticles in diagnosis and treatment of breast cancer. Lancet Oncol.7, 657–667.
3. Li, Z., Lee, D., Sheng, X., Cohen, R.E., Rubner, M.F., 2006. Two level antibacterial coating with both releasekilling and contact-killing capabilities. Langmuir. 22, 9820–9823.
4. Iglesias-Silva, E., Rivas, J., Isidro, L., Quintela, M., 2007. Synthesis of silver-coated magnetite nanoparticles. J. Non Cryst. Solids, 353, 829-31.

5. Huang, H. and Yang, Y., 2008. Preparation of silver nanoparticles in inorganic clay suspensions. Compos Sci Technol. 68, 2948–2953.
6. Rzigalinski, B.A., 2005. Nanoparticles and cell longevity, Technol Cancer Res Treat. 4, 651-9.
7. Ali, S.S., Hardt, J.I., Quick, K.L., Kim-Han, J.S., Erlanger, B.F., Huang, T.T., Epstein, C. J. and Dugan, L.L., 2004. A biologically effective fullerene (C60) derivative with superoxide dismutase mimetic properties. Free Radic Biol Med. 37, 1191-202.
8. Tarnuzzer, R.W., Colon, J., Patil, S. and Seal, S., 2005. Vacancy engineered ceria nanostructures for protection from radiation-induced cellular damage. Nano Letters. 5, 2573–2577.
9. Trajkovic, S., Dobric, S., Jacevic, V., Dragojevic-Simic, V., Milovanovic, Z. and Dordevic, A., 2007. Tissue-protective effects of fullerenol C60(OH)24 and amifostine in irradiated rats, Colloids Surf B. Biointerfaces. 58, 39-43.
10. Daroczi, B., Gabor, K., Mary, M.F., Jeffrey, W.C., Ulrich, R., Adam, P. and Dicker., 2006. *In vivo* radioprotection by the fullerene nanoparticle DF-1as assessed in a zebrafish model. Clinical Cancer Research. 12, 7086-7091.
11. Schubert, D., Dargusch, R., Raitano, J. and Chan, S. W., 2006. Cerium and yttrium oxide nanoparticles are neuroprotective Biochem Biophys Res Commun. 342, 86-91.
12. Deshpande, S., Patil, S., Kuchibhatla, S.V.N.T. and Seal, S., 2005. Size dependency variation in lattice parameter and valency states in nanocrystalline cerium oxide, Applied Physics Letters. 87 (13), 133113-3
13. Lara, H.H., Ayala-Nunez, N.V., Ixtepan-Turrent, L. and Rodriguez-Padilla, C., 2010. Mode of antiviral action of silver nanoparticles against HIV-1, J Nanobiotechnology. 8, 1.
14. Sun, L., Singh, A.K., Vig, K., Pillai, S.R., Singh, S.R., 2008. Silver nanoparticles inhibit replication of respiratory syncytial virus. J Biomed Biotechnol. 4, 149–158.
15. Jerley, A.A. and Margret, A.A., 2012. Mycosynthesis of silver nanoparticles and antibacteral effects in wound dressing, IJPSR, 3(6), 2.
16. Banerjee, J. and Narendhirakannan, R.T., 2011. Biosynthesis of silver nanoparticles from *Syzygium cumini* (l.) Seed extract and evaluation of their *in vitro* antioxidant activities, Digest Journal of Nanomaterials and Biostructures. 6, 961 - 968.
17. Mittal, A.K., Kaler, A. and Banerjee, U.C., 2012. Free radical scavenging and antioxidant activity of silver nanoparticles synthesized from flower extract of *Rhododendron dauricum*. Nano Biomedicine and Engineering. 4, 118-124.
18. Inbathamizha, L., Ponnub, T. M., Maryc, E.J., 2013. *In vitro* evaluation of antioxidant and anticancer potential of *Morinda pubescens* synthesized silver nanoparticles. Journal of Pharmacy Research. 6 (1), 32-38.
19. Bhol, K.C., Alroy, J. and Schechter, P.J., 2004. Antiinflammatory effect of topical nanocrystalline silver cream on allergic contact dermatitis in a guinea pig model, Clinical and Experimental Dermatology. 29, 282-287.
20. Bhol K. and Schechter, P., 2007. Effects of Nanocrystalline Silver (NPI 32101) in a Rat Model of Ulcerative Colitis, Digestive Diseases and Sciences. 52, 2732-2742.
21. Wong, K.K.Y., Cheung, S.O.F., Huang, L., Niu, J., Tao, C., Ho, C.-M., Che, C.-M., and Tam, P.K.H., 2009. Further Evidence of the Anti-inflammatory Effects of Silver Nanoparticles. Chem. Med. Chem. 4, 1129-1135.
22. Nadworny, P.L., Wang, J., Tredget, E.E. and Burrell, R.E., 2008. Anti-inflammatory activity of nanocrystalline silver in a porcine contact dermatitis model. Nanomedicine: nanotechnology, Biology, and Medicine. 4, 241-251.
23. Perry, S.W., Norman, J.P., Litzburg, A., Gelbard, H.A. (2004): Antioxidants are required during the early critical period, but not later, for neuronal survival. J. Neurosci Res., 78(4): 485-92.
24. Shulman, R.G., Rothman, D.L., Behar, K.L., Hyder, F. (2004): Energetic basis of brain activity: implications for neuroimaging. Trends Neurosci., 27(8): 489-95.
25. Okhawa, H., Ohishi, N. and Yagi, K. (1979): Assay for lipid peroxides in animal tissue by thiobarbituric acid reaction. Anal. Biochem., 95: 351-358.
26. Moron, M.S., Depierse, J.W. and Manrerirk, B. (1979): Levels of GSH, GR and GST activities in rat lung and liver. Biochem. Biophysics Aceta., 582: 67-78.

27. APHA (1989): Standard methods for examination of water and waste water. APHA, AWWA and WPCF, Washington D.C.
28. Trivedi, R.K. and Goel, P.K., 1986. Chemical and Biological methods for water pollution studies, Environmental Publication, Karad.
29. Baram-Pinto, D., Shukla, S., Perkas, N., Gedanken, A., Sarid, R., 2009. Inhibition of herpes simplex virus type 1 infection by silver nanoparticles capped with mercapto ethane sulfonate. Bioconjug Chem. 20, 1497–1502.
30. Furno, F., Morley, K.S., Wong, B., 2004. Silver nanoparticles and polymeric medical devices: A new approach to prevention of infection. J Antimicrob Chemother. 54, 1019–1024.
31. Baker, C., Pradhan, A., Pakstis, L., 2005. Synthesis and antibacterial properties of silver nanoparticles. J Nanosci Nanotechnol. 5, 244–249.
32. Kim, J.S., Kuk, E., Yu, K.N., 2007. Antimicrobial effects of silver nanoparticles. Nanomedicine. 3, 95–101.
33. Rogers, J.V., Parkinson, C.V., Choi, Y.W., Speshock, J.L., Hussain, S.M., 2008. A preliminary assessment of silver nanoparticle inhibition of monkeypox virus plaque formation. Nanoscale Research Letters. 3, 129–133.
34. Lu, L., Sun, R.W., Chen, R., 2008. Silver nanoparticles inhibit hepatitis B virus replication. Antivir Ther. 13, 253–262.
35. Becker Robert, O. M.D., Spadaro, J.A. 1989. Treatment of Orthopaedic Infections with Electrically Generated Silver Ions. J Bone Jt Surg. 197, 60.
36. Powell, J., 1978."Our Mightiest Germ Fighter". Science Digest.
37. Robert, O., Becker, M.D., 2009.The Body Electric, Journal of Longevity. 4(10).
38. Tjong, S. C. and Chen, H., 2004. Nanocrystalline materials and coatings. Materials Science and Engineering: R: Reports. 45, 1-88.
39. Kim, J.S., Kuk, E., Yu, K.N., 2007. Antimicrobial effects of silver nanoparticles. Nanomedicine. 3, 95–101.
40. Li, W.R, Xie, X.B., Shi, Q.S., Zeng, H.Y., Ou-Yang, Y.S., Chen, Y.B., 2010. Antibacterial activity and mechanism of silver nanoparticles on *Escherichia coli*. Appl Microbiol Biotechnol. 85(4), 1115-22.
41. Shahverdi, A.R., Minaeian, S., Shahverdi, H.R., Jamalifar, H., Nohi, A., 2007. Rapid synthesis of silver nanoparticles using culture supernatants of Enterobacteria: A novel biological approach Process Biochemistry. 42, 919–923.
42. Raleigh, J.A. (1985): Radioprotection of membranes. Pharmacology and Therapeutics., 39: 109-113.
43. Sies, H. (1985): Oxidative stress: introductory remarks. In: Oxidative stress (Sies H, ed.), Academic Press, London, pp. 1–8.
44. Sies, H. (1986): Biochemistry of oxidative stress. Angew. Chem. Int. Ed. Engl., 25: 1058–1071.
45. Pryor, W.A. and Godber, S.S. (1991): Noninvasive measures of oxidative stress status in humans. Free Radic. Biol. Med., 10: 177–184.
46. Pryor, W.A., Stanley, J.P. and Blair, E. (1976): Autoxidation of polyunsaturated fatty acids. II. A suggested mechanism for the formation of TBA-reactive material from prostaglandin-like endoperoxides. Lipid, 11: 370-379.
47. Shukitt-Hale, B., Erat, S.A. and Joseph (1998a): Spatial learning and memory deficit induced by dopamine administration with decreased glutathione. Free Radic. Bio. Med., 241: 149-1158.
48. Shukitt-Hale, B., Mouzaks, G. and Joseph (1998b): Psychomotor and spatial memory performance in aging male Fischer 344 rats. Exp. Gerontol., 33: 615-624.
49. Dauthal, P., Mukhopadhyay, M., 2013. In-vitro free radical scavenging activity of biosynthesized gold and silver nanoparticles using *Prunus armeniaca* (apricot) fruit extract Journal of Nanoparticle Research. 15, 1366.
50. Mohanraj, V.J., Chen, Y. 2006. (Eds) Nanoparticles. J Pharmaceutical Research. 5, 561–573.

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