

# Effect of Nanoparticles and Ionization Radiation on Same Oxidant and Antioxidant Enzymes

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## Abstract

Ionizing radiation effect on atoms and molecules of living organisms by formation of free radicals because it has high energy radiation which causes gain or loss one electron or more. In last year's there are a biggest interest for using nanoparticles in medicine as a drug delivers or cancer treatments, as its will knew. The present research was focused to evaluate the effects of X- ray and nanoparticles (alone or together) on level of Malondialdehyde (MDA), it is the final product of oxidative reactions, and, on the other hand, alleviate these effects by addition of vitamin – E (as anti- oxidant) in blood samples in-vitro. Results which were yield from this research indicated that level of MDA had been significantly increased when blood samples exposed to X- rays but its values were significantly dropped in the presence of nanoparticles of titanium dioxide (TiO<sub>2</sub>) in a comparison with those non- treated blood samples (control samples). It was well found that levels of MDA were markedly decrease in the samples treated with V- E after exposure them with both X-ray and nanoparticles. From those observations, it appear that exposure of living tissues to X-rays and nanoparticles causing production of free radicals (oxidative stress) and these adverse effects can be limited and ameliorated by treatment with antioxidant vitamins.

**Keywords:** TiO<sub>2</sub>NPS (titanium dioxide nanoparticles), ROS (Reaction Oxygen Species) MDA (Malondialdehyde enzyme)

## Introduction

Free radicals are naturally considered to be intracellular interactions from the metabolic processes of the cell to produce energy after Glucose or Cholesterol oxidation reactions, rapid free radicals enable to form other compounds mostly toxins or they interact with particles in cells. Free radicals are unstable atoms or molecules with free outer electrons. This makes them highly reactive because free electrons always strive to form a stable bond. This stabilization involves gaining an electron from another molecule, triggering a chain reaction. Such reactions are omnipresent in the human body, but under certain circumstances can damage biomolecules. There are many different sources of free radicals within cells and

the environment. In aerobic organisms, free radicals are produced during and through normal metabolic processes. Various in vitro and in vivo studies show that free radical formation can be triggered by nanoparticles (fullerenes, carbon nanotubes, quantum dots, emission particles)<sup>12,13</sup>. Nanoparticles can be taken up actively (phagocytosis) by certain cells (macrophages) and initiate ROS formation<sup>14, 15</sup>. Passive cellular uptake of particles has also been documented. X- Ray is one of the most common electromagnetic radiation that characterised with high frequency and short wavelength, these radiation are employed to be applicator in wide range of science fields such as medicine<sup>[1]</sup>, biology and industries. Radiotherapy has been used in treatment of many disease in particular cancerous diseases and oxidative

stress (OS) [2]. The Oxidative stress is an imbalance between production of reactive oxygen species (ROS) and antioxidant components within living cell that can mediate the damage of biological macromolecules including lipids, proteins and nucleic acids [3,4]. ROS are commonly called oxidants that are harmful for living tissues, among the ROS are superoxide anion ( $O_2^-$ ), hydrogen peroxide ( $H_2O_2$ ) and the hydrogen radical ( $OH^\cdot$ ) as well as nitrogen reactive species (NRS) [5]. Reactive oxygen species can be removed and inhibited by antioxidant that are enzymatic and non-enzymatic, such as catalase and vitamins [6] such as hypertension, hypercholesterolemia, and diabetes, are associated with oxidative stress. These observations and further data derived from a plethora of investigations provided accumulating evidence that oxidative stress is decisively involved in the pathogenesis of endothelial dysfunction and atherosclerosis. Several enzymes expressed in vascular tissue contribute to production and efficient degradation of reactive oxygen species, and enhanced activity of oxidant enzymes and/or reduced activity of antioxidant enzymes may cause oxidative stress. Various agonists, pathological conditions, and therapeutic interventions lead to modulated expression and function of oxidant and antioxidant enzymes, including NAD(P) Malondialdehyde (MDA) is the most common product of lipid peroxidation because of ROS reaction [7]. Nanoparticles were synthesized and employed in different fields of science, such as medical, biological and clean energy, these nanoparticles have been documented to induce oxidative stress because the production reactive oxygen species. [8]

### Materials and Methods

Method of preparing and adding nanoparticles ( $TiO_2$ ):

**1-** Titanium dioxide particles  $TiO_2$  in a synthesised as in [9]

**2-** Normal Saline with concentration 20 micrograms per 10 ml (it is dissolved and placed in a water bath for 15 seconds to ensure that the

nanoparticles in normal saline are distributed correctly)

### Sampling and addition method:

**1-**Collecting 20-30 venous blood samples from healthy donors, with an amount of 15 ml of blood per person

**2-**Samples are placed in tubes containing anticoagulant

**3-**Blood samples are placed in the centrifugation apparatus to separate the plasma from the blood for a of 15 minutes at 3000 cycles per minutes

**4-**blood plasma samples sprated to 7 test tubes (1 ml each tube), the samples classified as follows:

**a-**The first sample is considered control sample (without any addition).

**b-**The second sample we add 40  $\mu$ l of NPSTio2 in plasma of blood to study the effect of NPSTio2

**c-**The third part is left without additives for irradiation, but it is also treated like the rest of the samples

**d-** the fourth part, put NP<sub>5</sub>Tio<sub>2</sub> of 40  $\mu$ l and radiate it to study the effect of NPSTio2 and irradiation together

**e-**The fifth part, added 20  $\mu$ l of therapeutic vitamin E to be used as antioxidant, not pure vitamin E

**f-**The sixth part, add 20  $\mu$ g of vitamin E and 40 microliters of NPSTio2 to study the effect of vitamin and nano together.

**g-**The seventh part, add 20 $\mu$ l of the NPSTio2 and 40 $\mu$ g of Vitamin.E and left it around 24h to rest and after that irradiated with X-rays about 10 Gy to study the effect of radiation and the vitamin together

A chemical solution is applied to all parts of the samples except for the control sample, and this chemical solution works as a kit to measure

malondialdehyde, as it has been manually prepared in a chemical manner as described In <sup>[10]</sup> hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and this kit is placed in studied and appropriate quantities for the purpose of the tests and measurements that are made on all the samples above

\* After the process of preparing the samples and left for 24 hours in appropriate conditions, away from moisture and kept at 4° C and far from the sun's rays,. The samples were examined by using a UV-visible spectroscopy device to found the absorbance for each one separately and study the differences between the different sample treatments described for the purpose applying absorbance value to the (MDA) equation and measurement.

### Results and Discussion

MDA is considered to be a measure of free radicals

in living cells as the time period for formation and dissolution of free radicals is very short, MDA is used as a measure of the percentage of free radicals in the body in the following research. MDA and increased enzyme measurements are the result of an increase in free radicals resulting from metabolic processes in the living cells of the human body.

Note that ionizing X-ray works to greatly increase Malondialdehyde (MDA) activity and that the MDA enzyme is responsible for creating a free radicals in the human body, which means that the radiation increases free radicals ,also noted that the samples are reduced by the presence of Tio<sub>2</sub>NPS, but by the presence of Tio<sub>2</sub>NPS and ionizing X-rays together, we notice that there is a slight increase in the value of enzyme activity.

**Table (1) the activity of Malondialdehyde at (control) and Measurement the effect of NPs-Tio<sub>2</sub>, vitamin and x-ray each one at time and both to together.**

| S+V.E+X-ray | S+TiO <sub>2</sub> -NP<br>+V.E | S+V.E       | S+TiO <sub>2</sub> -NP<br>+X-ray | S+X-ray   | S+TiO <sub>2</sub> -NP | Control    |                           |
|-------------|--------------------------------|-------------|----------------------------------|-----------|------------------------|------------|---------------------------|
| 2.169±0.353 | 0.654±0.196                    | 0.165±0.059 | 1.75±0.197                       | 3.15±0.27 | 0.69±0.221             | 1.45±0.177 | Average                   |
| 1.501       | 0.83507                        | 0.253       | 0.839                            | 1.163     | 0.939                  | 0.752      | SD                        |
| -50%        | 55%                            | -25%        | -21%                             | -117%     | 52%                    |            | Change<br>from<br>control |

Note that the MDA enzyme decreased in the presence of nanoparticles.

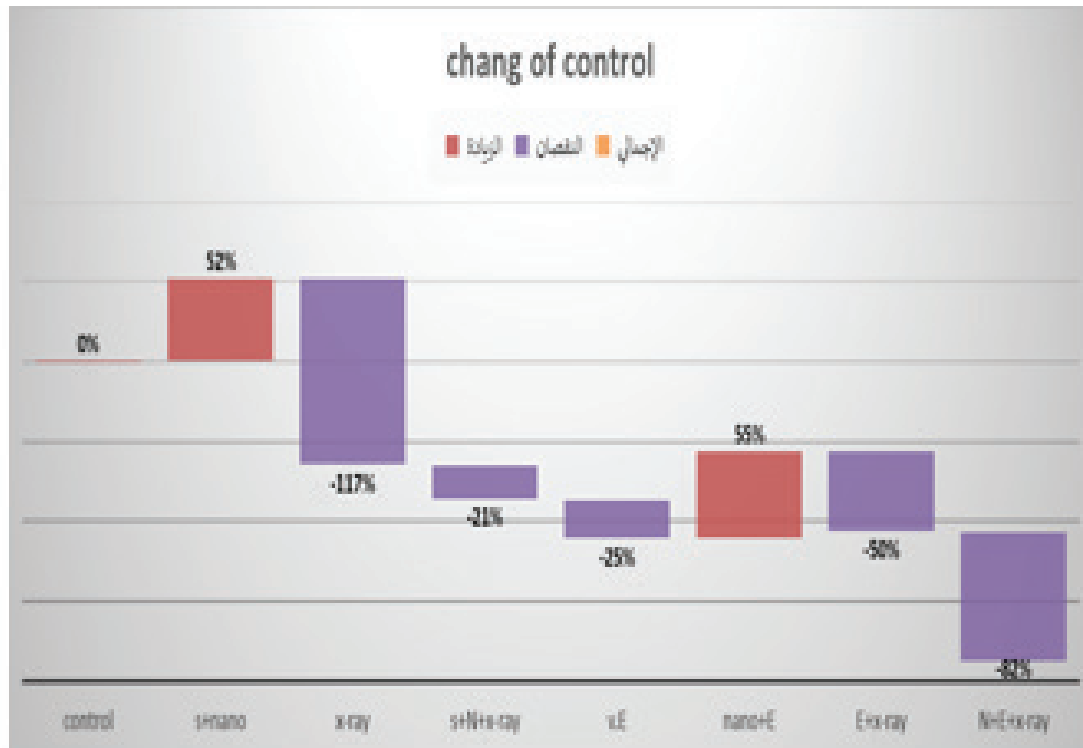


Fig1 (change of control)

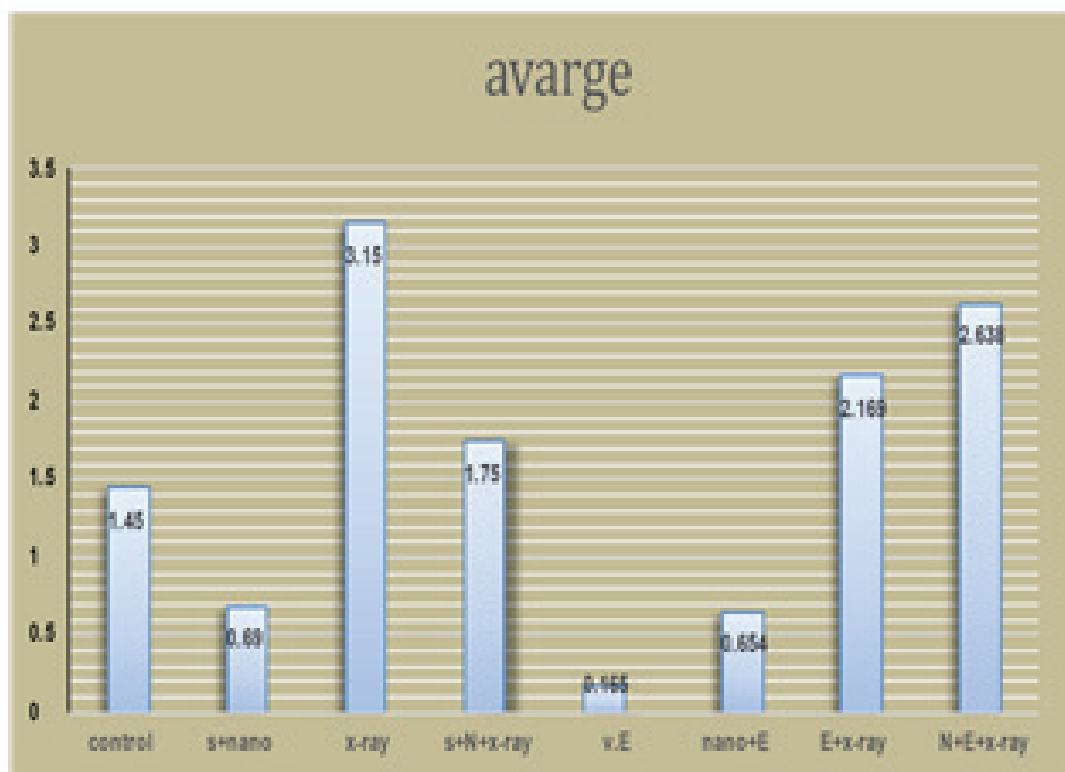


Fig2: Average of changes

But it increased a lot when it was irradiated with 10Gy of x-rays because the x-rays increase the rate of production of free radicals, meaning that (MDA is an enzyme that results from the increase of free radicals). A slight increase in the presence of titanium dioxide and X-ray particles. We note that it decreased in the presence of vitamin E because vitamin E works to soothe and reduce free radicals. And it decreased slightly with the presence of nano-dioxide titanium + vitamin E (because the presence of nanoparticles reduces the effect of vitamin E). It also increased with the presence of (vitamin E and X-rays). Significant increase in (Nano-dioxide titanium, vitamin E and x-ray), in the cells of the human body, the use of ionization radiation (IR) was witnessed a wide applications in deferent fields of science such as medicine, diagnostic, methods and therapeutic options <sup>[11]</sup>. Recent studies was conducted by <sup>[12]</sup> confirmed that decrease or minimize the adverse stress by addition of vitamin D and melatonin hormone whose functions are ameliorating of oxidative stress and they have protective effect to minimize side effects of ionizing radiation. It is well documented the deterioration effects of (IR) are induce oxidative stress when attack living tissue, since, the (IR) causes ionization chemical structures including organic and non-organic structures of living cells <sup>[13]</sup>. Previous experimental study was conducted on rat animal in this study 1000 GY does of ionization radiation was exposed to head of animals the obtaining results induced results of lipid peroxidation which manifest itself by increment of carbonyl and methylene group of lipid component as well as decrease member fluidity, strength of hydrogen bonds located in phosphate groups <sup>14</sup>

In addition, another previous experimental research investigate the effects of X-rays of antioxidant system of rabbits, the study involved in poseur of exposure of animals to 550-rad- X-rays with administration

Antioxidant vitamin (E and C) and antioxidant minerals (Manganese, Zinc, and copper) the results

of this study showed increase of Malondialdehyde (MDA) and decrease of glutathione peroxidase in irradiated group without antioxidant (vitamin and minerals) but those group which administered with antioxidants do not affected with oxidative stress after irradiation because reinforcement of antioxidant system <sup>15</sup>. In conclusion, the results which are recorded above, it obviously appear that ionizing radiation and nanoparticles, alone or combined, can increase levels of free radicals but the application of synthetic antioxidant vitamins have ability to ameliorating and alleviate these effects

**Financial Disclosure:** There is no financial disclosure.

**Conflict of Interest:** None to declare.

**Ethical Clearance:** "All experimental protocols were approved under the College of science and carried out in accordance with approved guidelines".

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