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RESEARCH ARTICLE

The Protective Effect of Green Apple Juice on physiological and Biochemical Parameters in Blood of Male Albino Rats Exposed to X- Ray

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Abstract

The present study aimed to investigate the protective effect of green apple juice against the harmful impact resulting from exposure to X- ray in adult male albino rats *Rattus norvegicus* aged (5-7) months, their weight (250- 350)g. 40 male rats were used, they were divided randomly into four groups, 10 rats for each group, as follow: The first group was given standard forage and normal tap water and considered as a control, the second group was exposed to X- ray at about 76 Kv. (reduplicate) for a period of 3 days each 5 seconds, Third and fourth group were treated with green apple juice 2 and 4 ml/Kg. B.W. respectively by gavage tube, and after one hour of treatment, the rats were exposed to X- ray at about 76 Kv. (redupilcate) for a period of 3 days each 5 seconds, the last three groups were given a standard forage and tap water *ad libitum*.

The results showed that X- ray exposure caused a significant decrease at ($P \leq 0.05$) in the haeamoglobin concentration, packed cell volume, total red blood cells count, the concentration of each of total protein, albumin, globulin and uric acid, but a significant increase in total white blood cells count, platelets count, the concentration of urea, creatinine and peroxy nitrate radical in blood of male reats exposed compared with control group (was not exposed). The results also showed a significant decrease in glutathione (GSH) concentration which has a role as antioxidant, with a significant increase in malondialdehyde (MDA) concentration in blood serum of exposed rats as compared with control group, the a above changes indicate the ability of X- ray to induce oxidative stress in male rats.

The results also showed that treating rats with green apple juice 2 and 4 ml/ Kg. B.W. and then exposed them to X- ray causes a significant increase in some of the decreased hematological and biochemical parameters and vice versa, the results also showed that there was a significant increase in GSH concentration a compained with a significant decrease in MDA concentration as compared with the group exposed to X- ray only.

From this study we concluded that the green apple juice has a role protecting against reactive oxygen species that are produced as a result of exposure to radiation through body's cells, which happened as a result of increasing the production of antioxidants and inhibit free radicals in the blood. So we concluded that the green apple juice had the ability to protect against X-ray side effects

Introduction

The environment contains high level of radiation emitted since the emergence of life on earth that is balanced with other environmental factors. Radioactive pollution is considered one of the non physical environmental pollution, that has indirect effect on the human health and the rest of the organisms , however it could be lethal in some cases, or harmful to the extent that makes the human suffer from its effects in the future. Radioactive pollution is an important and complex problem as a result of its proliferation and the frequent use of radioactive materials in various areas such as medicine, agriculture, industry and others (Khader, 2010).

X-rays are considered as invisible and ionized electromagnetic radiations, with a wavelength that varies between (0.01 -10) nm , i.e. a short wavelength short and high frequency , as well as a high-energy that has the ability to ionize materials that pass through it and cause a dangerous chemical imbalance in the living cells during the exposure (Enginar *et al*., 2007; Ma *et al*., 2012) , and with the evolution of scientific applications of X-rays in particular medical applications used in the diagnosis and radiation treatment, in the sterilization and cleansing of medical instruments due to is powerful high permeability more than other radiations, it is the most efficient in the killing of living elements during sterilization, and in scientific researches to develop Medical radiation techniques (Harjan & Hashim, 2009 ; Ma *et al*., 2012).

As well as its use in research and industrial applications in wide areas such as checking the materials used in manufacturing, quality check, in the security field as it the most important equipment used at airports to monitor the baggage and passenger luggage in search for weapons, bombs, and other common applications in our daily lives, which constitutes 90 % of the risk of industrial radiation exposure (IARC,2000; Zschronack, 2007).

X- rays can be harmful, they have biological effects on living organisms in two ways (Ma *et al*., 2012; El-Missiry *et al*.,2012) , the direct method , in which the radiation does a primary ionization , it ionizes the atoms involved in the synthesis of body tissue as a result of the absorption of these atoms to radiation , which leads to the decomposition of molecules and will have major damage on the concentration of a single cell and the entire organism , such as a genetic mutation and cell death (Enginar *et al*., 2007) . In the indirect method, the effect of radiation is not directly transmitted to cells, the radiation produces large numbers of different types of free radicals within the body fluids, especially water (Cadenas & Davies, 2000; Mikkelsen & Wardman, 2003).

These free radicals are toxic chemicals such as the reactive oxygen species (ROS) such as hydrogen peroxide H_2O_2 and reactive nitrogen species (RNS) such as NO_2, that are toxic and harmful to the function of the animal cell and another cells, as it interacts with biomolecules like proteins, nucleic acids and fat in the cell (Zmyslong *et al*. 1998; Gisone *et al*.,2004, Cadet *et al*.,2010, Acharya *et al*.,2010).

Cases of oxidative stress can induce through by X-ray (Ma *et al*.,2012), the exposure to this radiation is strongly related to oxidative stress and free radicals that affect the activity of the cell , particularly the function of cell membrane , as the free radicals have a role in the oxidation of lipid membranes thus losing their functional role which is the permeability of the cell membrane of the liquids, therefore creating more damage in the cell (Stark,1991; Zmyslony *et al*.,1998; Ibrahim *et al*.,2008).

Aim of the Research

Given the exposure of organisms, including the human being to the various types of natural and industrial radiations and their negative effects on the installation of the cell, tissue and organ, some studies attempted to find out the protective effect of some plants against radiation, hence, the present research aims to find out the preventive effect of green apple juice on fighting oxidative stress caused by X- ray in male albino rats.

Materials and Methods

A. Sampling

In this study, the green apples fruit (*Malus domestica*) of the Rosaceae family was used, from the local markets, wash this fruit well, and obtain the extract of apple fruit (juice) a concentration of 100% , the whole fruit was chopped (pulp and rind of the fruit) into small pieces and squeezed by an electric mixer for one minute , and then poured in clean and well sealed glass bottles, to be dosed to the animal on the same day of preparation.

B. X-ray Used

The animals were exposed to the X- ray machine at one of the diagnostic radiology clinics of 12-volt, 76 Kv. and an intensity of 300 ampere at 20 cm distance from the animal placed in a cage. The exposure lasted for 5 seconds (reduplicate).

C. Animals Used

Preparing the Animals

In this study, 40 adult male rats *Rattus norvegicus* were used with ages that ranged between (5-7) months, weights between (250-350)g., all in good health, and obtained from the College of Medicine at the University of Mosul. The animals were placed in plastic cages with metal net covers , especially for breeding rats , and were subjected all along the study to a standardized laboratory conditions of ventilation, temperature, (26 ± 2)ºC , natural light , and then subjected to a probation period of one week for the rats to acclimatzation use to the place . The animals were fed, standard forage and water were available *ad libitum* until the end of the experiment.

Experiment Design

Animals were divided randomly into four groups .10 rats for each group with similar weights, placed in separate cages and were treated as follows:

1. **Control group:** the group includes rats that were given standard forage and normal tap water daily, *ad libitum* for 3 days.
2. **X- ray group only:** the group includes rats that had been exposed to X- ray about 76 Kv. (reduplicate), for a period of 3 days each 5 seconds. Rats were also given standard forage and normal tap water *ad libitum*.
3. **Group of Green apple juice with a dosage of 2 ml/ Kg. B.W. and X- ray :** the group includes rats that were treated with green apple juice 2 ml / Kg. B.W. orally through a gavage tube. Rats were exposed after an hour to an X- ray about 76 Kv. (reduplicate), for a period of 3 days each 5 seconds, they were also given standard forage and normal tap water *ad libitum*.
4. **Group of Green apple juice with a dosage of 4 ml/ Kg. B.W. and X- ray :** the group includes rats that were treated with green apple juice 4 ml / Kg. B.W. orally by a gavage tube. Rats were exposed after an hour to X- ray about 76 Kv. (reduplicate), for a period of 3 days each 5 seconds and, they were also given standard forage and normal tap water.

Note: the first group and the second group were given distilled water daily by a gavage tube for 3 days to offset the stress of keeping rats.

D. Collection and Preservation of Samples

After the treatment of rats and at the end of the period mentioned above, blood was taken from the heart by a heart puncture using a sterile syringe, and dividing the blood by type of examination, 1 ml of blood was put in plastic tubes with lids containing anti coagulant EDTA in order to do blood tests, while the remaining quantity of blood was placed in plain tubes, dry and free of any anticoagulant material. The tubes were left in a room temperature for 20 minutes until the coagulation of blood, and then a centrifuge process was conducted with a speed of 3000 rpm for 15 minutes to get the blood serum for the purpose of doing biochemical tests.

E. Hematology and Biochemical Tests

The whole blood and serum were used to estimate the levels of a number of, blood and biochemical paramerterss using the measurement methods referred to in table (1), as a number of ready tests were used from international companies such as English Randox Company, French Biolabo Company and German Biocon Company, to estimate the concentrations of haemoglobin, biochemical paramerters that include proteins, urea, uric acid and creatinine. The rest of the hematology and biochemical paramerters were estimated using manual methods.

F. Statistical Analysis

Results were analyzed using the complete randomized design (C.R.D.). Differences between the groups were identified using the Duncan multiple range test as results were tested at a concentration of ($P \leq 0.05$), and the use of statistical program ready (SAS) for 2002 (Steel & Torrie, 1980).

Results and Discussion

1. Effect of Treatment on Hematology Parameters

The results in table (2) showed a significant decrease in at probability ($P \leq 0.05$) in the concentration of blood haemoglobin , the packed cell volume (PCV) and the total red blood cells count (RBCs) with a parentage 29% , 26% , 28% , respectively , while it showed a significant increase in the total white blood cells count (WBCs) and platelets count with a parentage 39% 43% respectively in the blood of rats exposed to X - ray only for a period of

3 days, compared with the non - exposed male rats (control group) . The results also showed that exposing rats to X- rays after taking 2 ml and 4 ml / Kg. B.W. of green apple juice, causes a significant increase in the concentration of blood haemoglobin, PCV, RBCs count , while it showed a significant decrease in WBCS count and platelets count compared with a group of male rats exposed to X- rays only, knowing that all these values are not up to the values of the control group.

The results of this study are consistent with the findings of Hashem and El- Sharkawy (2009) from the increase in WBCs count and the platelets count in the blood of rats exposed to the electromagnetic field with a frequency of 50 Hz at a rate of 4 hours per day for 30 days compared with the control group , as these results are consistent with the findings of Abdel Aziz *et al*.,(2010) from a significant decrease in the blood haemoglobin and RBCs count and increase in WBCs count of rats exposed to electromagnetic field with a frequency of 900 MHz compared with the control group . this change in RBCs and WBCs count is the result of its production decrease in the bone marrow (Jandova *et al*.,2005).

These results are also consistent with the findings of Al-Nia´emi *et al*.,(2001) from the decrease of blood haemoglobin and PCV accompanied by an increase in WBCs count of rats exposed to electromagnetic radiation with a frequency of 1 MHz with 1000 and 1200 volt, as the exposure to this radiation caused an abnormal fission in the cells that generate white blood cells in the bone marrow and that this increase is inconsistent with the process of red blood cells formation which causes a decrease in the concentration of blood haemoglobin (Seeley *et al*., 1992). Coskun *et al*.,(2003) noted the decrease of blood haemoglobin concentration, PCV and the increase of WBCs and platelets count which affects the bone marrow when treated with X- rays that lead to the loss of the ability of bone marrow to produce blood cells, especially RBCs, leading to a decrease in the concentration of blood haemoglobin and PCV.

The decrease in the concentration of blood haemoglobin, PCV and RBCs count when exposing rats to X-rays, produces ROS such as H_2O_2 , which results in the oxidation of thiol group (- SH) in the peptide chain of the haemoglobin protein, generating disulfide bonds (Desnoyers, 2000) , and iron ion oxidation(Fe^{+2})in haemoglobin molecule into(Fe^{+3}) , as well as the free radicals attack and crashes the membranes of RBCs, oxidizes the constituent lipids of these membranes, and leading to decomposition (Christopher *et al*., 1990). The decrease in WBCs count in rats exposed to X- rays compared with the control group is attributed to the fact that the animal infects from oxidative stress which led to stimulating the immune system to form defensive cells (Ganong, 1991).

Table (1): The methods used for estimating the number of hematological and biochemical parameters in the present study.

Measured Parameters		Methods used	Reference
Blood components	Blood haemogobin	Drabkin's method.	Drabkin & Austin. 1935
	Packed cell volume	Using the haematocrite reader	Talib & Khurana. 1996.
	Total red blood cells count (RBCs)	Using the hamocytometer and Hayem's solution.	
	Total white blood cells count(WBCs)	Using the hamemocytometer and Turk's solution	
	Platelets count	Using the hameocytameter and ammoninum oxidate solution.	
Biochemicl parameters	Total protein conc.	Biuret method	Gornall *et al*., 1949.
	Albumin conc.	Bromocresol green method	Doumans *et al*., 1971.
	Globulin conc.	According to the following law:- Total protein conc - albumin conc.	Ricterich, 1969
	Urea conc.	Enzymatic method	Fawcett& Soctt, 1960.
	Uric acid conc.	Enzymatic method	Newman & Price, 1999.
	Creatinine conc.	Colorimetirc method	Tietz, 1999.
	Malondialdhyde (MDA) conc.	Thiobarbituric acid modified method	Guidet & Shah, 1989
	Glutathione (GSH) conc.	Modified Ellman's reagent method	Sedlak &Lindsay, 1968.
	Peroxy nitrate radical conc.	Phenol method	Vanuffelen *et al*., 1998.

Table (2): The protective effect of green apple juice on some of hematological parameters in blood male albino rats exposed to X- ray.

Parameters / Treatment groups	Haemoglobin conc.(g/dl)			Packed cell volume (%)			Total red blood cells count (*10^6 cell/ ml^3)			Total white blood cells count (*10^3 cell/ ml^3)			Platelets count (*10^3 plate/ ml^3)		
	*Mean± SE	% Conc.	% Change	Mean± SE	% volume	% Change	Mean± SE	% count	% Change	Mean± SE	% count	% Change	Mean± SE	% count	% Change
Control (Not exposed)	15.40± 0.100 a	100	-	51.0±0.100 a	100	-	8.60± 0.20 a	100	-	9.73± 0.208 d	100	-	813.0± 2.645 d	100	-
Exposed to X-ray only	10.95±0.131 d	71	-29	37.5± 0.360 d	74	-26	6.20± 0.10 c	72	-28	13.53± 0.152 a	139	+39	1159.5±1.307 a	143	+43
Treated with green apple juice 2 ml/Kg. B.W. + exposed to X- ray	11.60± 0.300 c	75	-25	38.5± 0.608 c	76	-24	6.36± 0.208 c	74	-26	12.60± 0.100 b	129	+29	1053.0±6.082 b	130	+30
Treated with green apple juice 4 ml/Kg. B.W. + exposed to X- ray	12.73±0.208 b	83	-17	41.5± 0.435 b	81	-19	7.26± 0.152 b	84	-16	11.96±0.308 c	123	+23	981± 9.539 c	121	+21

*** Mean ± S.E. for three reduplicates.**
The number of animals per group of five
Numbers are preceded by different letters vertically indicate a significant difference at the level of probability (P≤ 0.05) and correct reverse according Duncan test

Table (3): The protective effect of green apple juice on some of biochemical parameters in blood serum of male albino rats exposed to X- ray.

Parameters / Treatment groups	Total protein conc. (g /dl)			Albumin conc. (g/dl)			Globulin conc. (g/dl)			Urea conc. (mg/dl)			Creatinine conc. (mg/dl)		
	*Mean± S.E.	% conc.	% Change	Mean± S.E.	% conc	% Change	Mean± S.E.	% conc.	% Change	Mean± S.E.	% conc.	% Change	Mean± S.E.	% conc.	% Change
Control (Not exposed)	6.60± 0.655 a	100	-	4.90± 0.529 a	100	-	1.70± 0.500 a	100	-	22.65± 0.893 c	100	-	0.46±0.115 c	100	-
Exposed to X-ray only	5.25± 0.278 b	80	-20	4.15± 01.80 b	85	-15	1.10± 0.173 a	65	- 35	34.05± 0.325 a	150	+50	1.45±0.138 a	315	+ 115
Treated with green apple juice 2 ml/Kg. B. W. + exposed to X- ray	5.90±0.754 ab	89	-11	4. 70±0.100 ab	96	- 4	1.20+ 0.700 a	71	- 29	31.20± 0.781 ab	138	+38	1.63±0.104 a	354	+ 254
Treated with green apple juice 4 ml/Kg. B. W. + exposed to X- ray	6.25± 0.132 ab	95	-5	4.95± 0.150 a	101	+1	1.30+ 0. 264 a	76	- 24	29.05± 0.826 b	128	+28	1.03±0.060 b	224	+ 124

*** Mean ± S.E. for three reduplicates.**

The number of animals per group of five

Numbers are preceded by different letters vertically indicate a significant difference at the level of probability (P≤ 0.05) and correct reverse according Duncan test

Table (4): The protective effect of green apple juice on malondialdehyde (MDA), peroxy nitrate concentration and non- enzymatic antioxidants in blood serum of male albino rats exposed to X- ray.

Parameters / Treatment groups	MDA conc. (µmol/ L)			GSH conc. (µmol/ L)			Uric acid conc. (mg/dl)			Peroxy nitrate conc. (µmol/L)		
	*Mean± S.E.	% conc.	% Change	Mean± S.E.	% conc	% Change	Mean± S.E.	% conc.	% Change	Mean± S.E.	% conc.	% Change
Control (Not exposed)	1.38± 0.087 d	100	-	8.15± 0.095 a	100	-	11.10 ± 0.360 a	100	-	61.45± 1.780 c	100	-
Exposed to X- ray only	3.35± 0.095 a	243	+ 143	5.34± 0.078 d	66	-34	6.25 ± 0.050 d	56	-44	155.70 ± 4.233 a	253	+15
Treated with green apple juice 2 ml/Kg. B.W. + exposed to X- ray	2.92 ± 0.098 b	212	+112	6.21± 0.026 c	76	-24	7.40 ± 0264 c	67	-33	88.30 ± 2.594 b	144	+4
Treated with green apple juice 4 ml/Kg. B.W. + exposed to X- ray	1.85 ± 0.045 c	134	+34	7.45 ± 0.104 b	91	- 9	9.10± 0.200 b	82	-18	66.55 ± 1.837 c	108	+8

*** Mean ± S.E. for three reduplicates.**

The number of animals per group of five

Numbers are preceded by different letters vertically indicate a significant difference at the level of probability (P≤ 0.05) and correct reverse according Duncan test.

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Nevertheless, when rats were treated with green apple juice then exposed to X-rays , it led to a significant increase in the concentration of haemoglobin, PCV stacked and RBCs count, accompanied by a significant decrease in WBCs and platelets count, compared with the group exposed to X- rays only, and this is due to the preventive influence of apple juice from ROS and the protection of RBCs from oxidative stress due to the juice that contains active and antioxidants ingredients such as phenolic compounds, flavonoids, carotenoids and vitamins, especially vitamin C , which is one of the most important non-enzymatic antioxidant that works to remove ROS and increase the efficiency of the immune response (Lapidot *et al*., 2002 ; Wolf *et al*., 2003).Vitamin C also increases the vital presence of iron, it also increases its absorption in the gastrointestinal tract (Hallberg *et al*.1989) and stimulates vitamin C to secrete the catalyst for the formation of red blood cells (Erythropoietin) from the kidney, which stimulates the formation of RBCs from the bone marrow (Kassab *et al*.,1992) .

The results ahso showed a decrease of platelets count in blood of rats given green apple juice, and this result is consistent with the findings of Hubbard *et al*., (2003) as the apple contains Querecetin that reduces the number of platelets and inhibit collected. Boyer and Liu (2004) stressed on the role of apple (because it contains Querecetin) in the prevention of heart disease and blood vessels through its due to its effect on the activity of platelets and prevention.

2. Effect of the Treatment on Biochemical Parameters

The electromagnetic field (radiations) penetrates the organisms and affect all members; they change and affect the function of the cell membrane and the distribution of bipolar ions. These changes affect the biochemical processes in the cell, which leads to a change in the biochemical paramerterss and the activity of the enzymes in the serum (Berg, 1993).

The results in table (3) showed a significant decrease in the concentration of each of total protein, albumin and globulin , while the results showed a significant decrease in the concentration of urea and creatinine in the blood serum of male rats exposed to X- rays only for 3 days compared with the control group (non- exposed). And the results showed that the rats were treated with green apple juice and then exposed them to X- rays, led to a significant increase in the concentration of each of the total protein, albumin and globulin , and was accompanied by a significant decrease in the concentration of urea, creatinine , compared with the group of male rats exposed to X-rays only, knowing that all these values are not up to the values of the control group.

The results of this study are consistent with the decrease of albumin and increase of urea and creatinine with the results reached by Hashem and El-Sharkawy (2009), when exposing rats to electromagnetic fields with a frequency of 50 Hz at a rate of 4 hours every day for a period of 30 days compared with the control group. The results of this study are consistent with the study that showed that the exposure to electromagnetic radiation causes significant changes in the concentration of proteins in the blood serum of mice (El- Ashry *et al*., 2008a) , as indicated by Lee *et al*.,(2004) that the exposure to electromagnetic fields affect the biological system by the large production of free radicals , and pointed to the increase of oxidative stress due to exposure , which is the reason to the adverse changes in the cell , leading to a number of enzymatic disorders in the body.

This decrease in the concentration of total protein, albumin and globulin in the blood serum of rats exposed to X-rays, is due to the ability of X- rays (ionized electromagnetic radiation) to generate free radicals (Matkovics, 2003) that have an intense familiarity to interact with biomolecules like proteins and lipids, which breaks the peptide bonds of protein chains and then oxidizes these proteins and decreasing its concentrations (Ray *et al*.,2000; Roche *et al*.,2009). Or due to the increased oxidative stress caused by X- rays that increases the secretion of corticosterone, a hormone of the cortex adrenal gland , and thus increasing the use of amino acids through gluconeogenesis to build glucose from noncarbohydrate sources , this may be a reason of the decreased concentration of proteins in blood serum (William, 1984; Freeman, 1988). The decreased concentration of proteins and increased urea in animals exposed to oxidative stress, may be due to the use of alternative sources of energy in the body like lipids and proteins, as the amino acids compensation for the production of energy increases, which causes the formation of large quantities of urea as an accidental output (Murray *et al*.,2000; Guyton & Hall, 2006).

The albumin is considered among the most sensitive proteins to oxidation , which leads to the decrease of its concentration in the blood serum. Or perhaps to the increase of the proteins and amino acids oxidation, which leads to the increase of the concentration of urea in the blood serum as secondary product (Manna *et al*.,2005; Roche *et al.*, 2009). The decrease of albumin causes the decrease of proteins synthesis and the increase of the rate of proteins catabolism or reduces the formation of ROS, therefore the albumin is a non-enzymatic antioxidant (endogenous), that has an important role in reducing the formation of free radicals through breaking interactions chains and re-repairing damaged biomolecules affected by oxidation, so that explanins the decrease has consumed as an

antioxidant (Suleyman *et al*.,2003), the increase of the concentration of urea and creatinine causes chronic complications in some organs of the body (Guyton & Hall, 2006) .

Treated the rats with green apple juice before their exposure to X- rays , has led to a significant increase in the concentration of the total protein, albumin and globulin , and a decrease in the concentration of urea and creatinine comparing to the group exposed to X-rays only, and this is due to the role of the apple juice in stimulating the process of proteins synthesis in different organs especially the liver , or may be due to several other reasons , such as the activity of antioxidants like the phenolic compounds, flavonoids, carotenoids and vitamin C that is very important and present in the green apple juice, as they all work to reduce oxidative stress by removing free radicals and prevent the oxidation of proteins and amino acids or reduce the exploitation of albumin as an antioxidant (Eberhardt *et al*.,2000; Boyer & Liu 2004). Or the reason could also be due to multiple phenolic compound and carotenoids , such as β - carotene that has the ability to reduce oxidative damage of the renal cells of the renal and renal glomeruli , and protect the liver from oxidative damage and thus reduces disorder and proteins decomposition , which leads to the decrease of urea in blood (Wolf *et al*.,2003; Molina *et al*.,2003; Loru *et al*.,2009). These compounds found in the green apple juice prevent renal failure and maintain glomerular filtration rate within the normal range (Anjaneyulu & Chopra 2004).

3. Effect on Treatment of Lipid Peroxidation and Antioxidants

The results in table (4) showed a significant increase in the concentration of each of malondialdyde (MDA), peroxy nitrate radical, and a significant decrease in the concentration of Glutatthione (GSH) and uric acid in the blood serum of male rats exposed to X- rays only for 3 days compared with the control group. And the results showed that were treated rats with green apple juice with a dosage of 2 and 4 ml / Kg B.W. , then exposing them to X- rays causes a significant decrease in the concentration of MDA and peroxy nitrate, accompanied by a significant decrease in the concentration of GSH and uric acid comparing to the rats exposed to X-rays only.

The results of this study are consistent with the findings of (El-Ashry et al.,2008b; Ibrahim et al.,2008), of a significant increase the concentration of MDA and a significant decrease in the concentration of GSH in the extract of rats livers exposed to different densities of electromagnetic field with a frequency of 50 Hz for two hours during 7 days. Results are also consistent with the results of Mottawie and Ibrahim (2011), that showed an increase in the levels of H_2O_2, MDA, and a decrease in the concentration of GSH and the activity of Glutathione peroxidase (antioxidant enzyme), in the serum and liver of rats exposed to electromagnetic fields with a frequency of 900 Hz.

The compatibility between the high concentration of MDA and low concentration of GSH in the blood serum of rats exposed to X- ray is one of the oxidative stress indicators compared with the control group, due to the start of fatty acids oxidation process that increases the production of H_2O_2, which produces lipid peroxides (Osumi & Hashimoto, 1978; Halliwell & Gutteridge,1984). The increase of the MDA concentration, decrease of GSH concentration and uric acid are due to an imbalance fighting ROS that have an oxidizing impact on the viability of systems destroying these oxidizers, however the exact mechanisms remain unclear.

Several studies showed that the exposure to ionizing radiation or electromagnetic fields has a negative impact on the activity of some enzymatic antioxidant such as the Superoxide dismutase, catalase and peroxidase , as well as its negative impact on the biosynthesis process of glutathione and thiol group which is considered as an antioxidant factor in the GSH structure , and the impact of these radiations on the body's ability to return oxidized form of glutathione (GSSG) into active reduced form (GSH) (Fiorani *et al*.,1998). Erden (1992) suggested to the role of GSH as an antieffects of electromagnetic radiation, as it has an important protective role against the damaged free radicals effects caused by the radiation in the blood serum, lung and brain.

Numerous studies showed that oxidative stress inhibits the activity of the Glucose-6- phosphate dehydrogenase enzyme considered as a Hexose monophosphate pathway enzyme , that leads to interference in the process of reduction of $NADP^+$ (oxidized form) to NADPH (the reduced form) necessary to return reduce oxidized form of glutathione (Meister,1983; Fiorani *et al*.,1998). The increasing concentration of $NADP^+$ in the body leads to the liberation of ferric ion (Fe^{+3}) in particular, which increases the rate of formation of hydroxyl radical through Fenton reaction (Meister, 1983; Zmyslony *et al*.,1998).

The reason of the decrease of uric acid concentration is that it is a non-enzymatic antioxidant (endogenous) that has the ability to inhibit lipid peroxidation process as well as the ability to directly remove free radicals(Rodionov,2003). And the results showed a significant increase in the concentration of peroxide nitrate radical in the blood serum of rats exposed to X- rays , due to an increase in the production of of superoxide cathode radical as a result of oxidative stress caused by the exposure to X- rays , as the peroxide nitrate is formed by the interaction of nitric oxide radical with the superoxide cathode radical due to an increase in the formation of nitric

oxide in rats exposed to X- rays (Pressman *et al*.,2003; Gisone *et al*.,2004) , as the peroxide nitrate can work on attacking the cell membranes of lipids, proteins and nucleic acids, leading to a damage in different cells and an increase of oxidative stress (Denicola & Radi, 2005).

Treated the rats with green apple juice before their exposure to X- rays , has led to a significant increase in the concentration of MDA and peroxide nitrate and a decrease in the concentration of glutathione and uric acid comparing to the group exposed to X- rays only. and this is due to the activity of antioxidants and active compounds that are present in the green apple juice like the phenolic compounds, flavonoids, carotenoids and vitamins especially vitamin C, as they all work to reduce oxidative stress by removing free radicals, these components provide protection against the risk of these radicals, so it is defensive line against oxidative stress and thus lead to the return of these parameters to normal levels (Breinholt *et al*.,2003; Wolf *et al*.,2003; Boyer & Liu 2004).

Conclusions

The researcher concludes that the exposure to X- rays causes clear effects of toxicity on more than one organ in the body, and an imbalance in many physiological and metabolic processes. The green apple juice with a dosage of 2 and 4 ml/Kg. B.W. has positive effects on hematological and biochemical paramerters, and this shows that green apple juice has a preventive ability in removing oxidative stress and toxic effects caused by X- rays.

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