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Protective effect of Zingiber officinale extract and vitamin C in modulating radiation-induced brain damage in male albino rats

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ABSTRACT

Background*:** Radiation therapy is associated with a risk of long-term adverse effects. Ginger extract has several components that have many biological activities and vitamin c has also been recognized for protection against radiation-induced cell damage. The present study is designed to investigate the possible ameliorating effect of ginger extract and vitamin C on radiation-induced oxidative body damage. Ginger extract and vitamin C were daily given to rats during 14 days before starting irradiation. ***Materials and Method: Rats were exposed to gamma radiation (6 Gray). ***Results:*** the result revealed that the levels of lipid peroxidation measured in brain tissues such as malondialdehyde (MDA), acetylcholinesterase (AChE) were significantly increased, while reduced the activities of superoxide dismutase (SOD), catalase (CAT) and glutathione (GSH) content, dopamine (DA) and serotonin (ST) levels were significantly decreased in the brain homogenate of irradiated rats. Gamma-irradiation (6 Gy) resulted in a significant elevation in inflammatory markers of tumor necrosis factor-alpha (TNF-α), interleukin-6 (IL-6), interleukin-12 (IL-12) and C-reactive protein (CRP) compared to the control group. The rats that were administrated combined treatment with ginger extract and vitamin C showed significantly less severe damage and remarkable improvement in all of the last mentioned parameters when compared to irradiated rats. ***Conclusions:*** According to the results obtained it could be concluded that a combined treatment with ginger extract by its antioxidant constituents, and vitamin C might be a useful candidate against radiation-induced oxidative stress, enzyme activities in the brain and metabolic disorders without any toxicity.

INTRODUCTION

The brain is the control center of the body; it controls all the systems in the body including muscular, respiratory and digestive systems by using a series of chemical, electrical and physical signals from cell to cell. Within the cell, electrical signals are used for transmission, but between cells, chemical signals are used, these chemical signals are called neurotransmitters. Alteration in the metabolism of these neurotransmitters leads to brain dysfunction [1].

Ionizing radiation-related illnesses are one of today's most difficult health complications, having far -reaching medical, societal, and economic implications. With its widespread use in diagnostics and industry, human exposure to ionizing radiation has become unavoidable. Radiation damage is primarily generated by an excess of reactive oxygen species (ROS), such as superoxide anion (O_2•), hydroxyl radical (•OH), and hydrogen peroxide (H_2O_2), which exceed antioxidant levels, causing oxidative stress. Lipid peroxidation, protein oxidation, and antioxidant depletion are the most serious outcomes of oxidative stress. Injury, mutagenesis, carcinogenesis, increased senescence, and cell death may happen if these injuries are irreversible [2].

There are increasing evidences to suggest that many degenerative diseases, such as brain dysfunction, cancer, heart diseases, and weakened immune system, could be the result of cellular damage caused by free radicals, and antioxidants present in human diet may play an important role in disease prevention [3, 4]. Plants have been the companion of man and formed the basis of useful drugs for the treatment of various ailments. The use of plants may be beneficial in protecting against the radiation-induced damage, since they are less toxic than synthetic compounds at their optimum doses [5]. Therefore, screening of plants presents a major avenue for the discovery of new radio protective drugs.

Anticancer potential [6] and antioxidant capabilities [7] of medicinal plants have long been recognized to be employed in the treatment of many ailments. Phytomedicines work through a variety of mechanisms, including antioxidant and

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cytoprotective effects [8]. Headaches, colds, diarrhoea, gastrointestinal problems, nausea, asthma and rheumatic symptoms, arthritis, and muscle pain have all been treated with ginger (Zingiber officinale Roscoe, Zingiberaceae) [9]. It has also been utilized as an anti-inflammatory, cardio-protective, and cancer-fighting drug. Ginger's antioxidant, anti-inflammatory, and antimutagenic activities have been described, and its active components have been linked to apoptosis in skin, ovarian, colon, breast, cervical, oral, renal, prostate, gastric, pancreatic, liver, and brain cancers [10].

Vitamin C has been found to be a powerful antioxidant and free radical scavenger, helping to protect deoxyribonucleic acid, (DNA) and cellular membranes from free radical damage [11]. It is an anti-radiation free radical scavenger that has been found to protect a range of biological systems. The radioprotective properties of ascorbic acid appear to be linked to interactions with free radicals generated by radiation [12]. Vitamin C pretreatment reduces radiation-induced lipid peroxidation [13]. In whole-body gamma rays (rays)-induced wounds, it protected mice from radiation-induced diseases, reduced mortality, and enhanced wound healing [14]. Additionally, it was demonstrated that vitamin C can lessen maternal and paternal transgenerational genomic instability. However, other studies have found conflicting results regarding vitamin C's ability to lessen the detrimental effects of radiation, necessitating more research [15].

Based on the mentioned data, the current study aims at evaluating the protective effects of ginger and vitamin C on the changes induced by Whole-body gamma-irradiation on the brain in male rats. This study is considered one of the first studies that combined both ginger extract (GE) and vitamin C to know their protective role in reducing radiation-induced brain damage.

MATERIALS AND METHODS

Animals

Eighty male adult albino rats weighing around (150±20g) (80 days old adult) were employed during the study. The rats were procured from the Nuclear Research Center's animal house in Inshas, Egypt, and were maintained in plastic cages with metal wire coverings in conventional laboratory settings with a 12-hour light-dark cycle. The rats were given unrestricted access to regular commercial laboratory food and tap water for two weeks prior to the start of the experiment, allowing them to adjust to the laboratory circumstances. The research was approved by the Research Ethics Committee of the National Center for Radiation Research and Technology at Egyptian Atomic Energy Authority (11A/22).

Chemical

Preparation of ginger aqueous extract

Ginger rhizomes were purchased from the local market, and the aqueous extract of Zingiber officinale was prepared as described by Ajith, Aswathy [16]. Fresh ginger rhizomes (1 kg) were washed, peeled, cut into pieces and dried in a hot air oven at 40±2°C. The ginger was then ground using an electric blender, soaked in 2 liters of sterile water for (3, hr.), heated at 60-65°C for 30 min, and then filtered to obtain the crude extract. The process was repeated four times, and all the filtrates were collected, concentrated by evaporation in a rotary vacuum evaporator at 55°C and then dried at room temperature. The powdered extract of ginger was stored at -20°C for future use. The final solution of ginger extract was made by dissolving the ginger powder in a saline solution.

Irradiation procedure

Whole-body gamma-irradiation was performed at the Atomic Energy Authority, Cairo, Egypt, using a Gammacell-40 Carloirradiator cesium-137 source. Animals were acutely irradiated with a single dose of 6 Gy delivered at a rate of 0.713 rad/sec.

Experimental groups

Eighty male albino rats were randomly distributed into eight groups of ten rats each. Group I served as a healthy control group and was orally given normal saline by gavage every day for 14 days. The animals in Group II (the IR group) were exposed to whole-body gamma radiation (IR) (6 Gy).

In Group III, the rats orally received vitamin C (250 mg/kg b.w.) every day for 14 consecutive days. In Group IV, the rats orally received freshly prepared ginger extract (250 mg/kg b.w.) for 14 consecutive days. In Group V, the rats were orally given freshly prepared ginger extract (250 mg/kg b.w.) and vitamin C (250 mg/kg b.w.) every day for 14 consecutive days. Group VI, the rats were orally given freshly prepared vitamin C (250 mg/kg b.w.) every day for 14 consecutive days and then exposed to whole-body gamma radiation (6 Gy) 2hr after the last administration of vitamin C.

In Group VII, the rats were orally given freshly prepared ginger extract (250 mg/kg b.w.) every day for 14 consecutive days and then exposed to whole-body gamma radiation (6 Gy) 2hr after the last administration of ginger. Group VIII, the rats were orally given freshly prepared ginger extract and vitamin C (250 mg/kg b.w.) every day for 14 consecutive days and then exposed to whole-body gamma radiation (6 Gy) 2hr after the last administration of ginger and vitamin C. Twenty-four hours after the last treatment, all animals were anaesthetized with ether, and blood samples were collected from the retro-orbital sinus with non-heparinized haematocrit tubes into clean, dry centrifuge tubes. The centrifuge tubes were allowed

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to sit at room temperature for at least one hour so the blood would coagulate and were then centrifuged at approximately 2500-3000 rpm for 10 min. The supernatant (serum) was transferred into clean Ependorf tubes and stored in a deep freezer at -20°C. The animals were euthanized by cervical dislocation after blood withdrawal.

Brain tissue homogenate preparation

Decapitated animals were sacrificed, and whole brains were taken and cleaned in ice cold isotonic saline. The brain tissue samples were then homogenised in a volume 10 times the weight of the tissue with ice cold 0.1M phosphate buffer (pH 7.4). The homogenized was centrifuged for 15 minutes at 10,000 xg, and aliquots of the supernatant were separated and used for biochemical assays.

Biochemical analysis

Antioxidant markers

Brain concentration of glutathione GSH was determined according to Beutler [17]; the method depends on the reaction of the free SH-group of the reduced glutathione molecule with 5,5'-dithiobis-(2-nitrobenzoic acid) [DTNB] yielding a yellow coloured product (2 nitro-5- thiobenzoic acid) that can be measured colorimetrically at 412 nm. Superoxide dismutase activity (SOD) in the homogenates of whole brain of rats was determined by measuring the inhibition of auto-oxidation of epinephrine at pH 10.2 at 30 °C according to the method of Masayasu and Hiroshi [18]. Catalase (CAT) activity was determined using hydrogen peroxide as substrate according to the method of Rick and Stegbauer [19]. Lipid peroxidation was quantified as malondialdehyde (MDA) according to the method described by Farombi [20]. This method depends upon the reaction of MDA with thiobarbituric acid (TBA) in an acidic medium to give a colored TBA complex which is then extracted with n-butanol and measured colorimetrically at 535 and 520 nm.

Brain biomarkers

Levels of dopamine and serotonin in brain homogenates were measured by Elisa kit (Uscn Life Science Inc. Wuhan). Acetylcholinesterase (AChE) activity was measured according to the method of Ellman, Courtney [21].

Inflammation markers

The levels of serum TNF-α were measured by enzyme-linke immunosorbent assay (ELISA) kit (BMS 622, Vienna, Austria), following the manufacturer's instructions. TNF-a levels were quantified with a commercially available rat TNF-a ELISA kit (Taylor, 2001) [22]. Serum interleukin-6 (IL-6) level was measured according to the method of Beutler *et al.* (1985) [23]. ELISA kit for IL-6 was used in this assay and purchased from Immuno-Biological Laboratories (IBL) Co., Ltd., USA. C-reactive protein was determined by ELISA kit (Bio diagnostic company, Egypt) following the manufacturer's instructions and guidelines.

Statistical analysis

Results are expressed as the mean ± standard deviation (SD). Statistical significant differences were evaluated using analysis of variance one way ANOVA followed by Post Hoc to determine significant differences between means. Differences were considered significant at $P \leq 0.05$ using statistical package system software (SPSS software version 22) [24]. The figures were drawn using Microsoft Excel program.

RESULTS

Whole body irradiation of rats provoked oxidative stress that was demonstrated by a significant increase of MDA level associated with a significant decrease of SOD, GSH and CAT activity, compared to their respective values of the control group during the experimental intervals (figure 1). The oral supplementation of ginger extract and vitamin C to rats during 14 successive days before irradiation via gavages has significantly attenuated the severity of radiation-induced oxidative stress. A significant decrease occurred in the oxidative biomarkers levels. Moreover, a significant increase of antioxidants levels (figure 1) was observed, compared to their respective values of the irradiated rats.

In table 1, the statistical results revealed that rats exposed to 6Gy gamma-irradiation increased the AchE level significantly as compared to the control group. On the other hand, the levels of dopamine and serotonin in brain of the irradiated rats decreased. Treatment of the irradiated animals with GE and vitamin C showed significant differences in AchE, ST and DA levels.

The results, in table 2, show that there were significant elevation in the level of serum TNF-α, IL-6, IL-12 and CRP of gamma-irradiated group compared to the control group. Supplementation of γ-irradiated rats with ginger extract and vitamin C showed a significant decrease in the level of TNF-α, IL-6 IL-12 and CRP relative to γ-irradiated group.

Table 1. Effect of radiation (G2) , vitamin c (G3) ginger extract (G4) vitamin c and ginger extract (G5) administration , vitamin c and radiation(G6), ginger extract and radiation (G7), vitamin c and ginger extract with radiation (G8) compared to Control Group (G1) on brain AChE (pg/mg), ST (ng/mg) and DA (ng/mg) of male albino rats.

Groups	AChE (pg/mg)	ST (ng/mg)	DA (ng/mg)
G1	8.6 ± 1.14^{f}	116.4 ± 4.39^{a}	1.01 ± 0.05^{a}
G2	72.2 ± 4.97^{a}	17.0 ± 6.08^{f}	0.26 ± 0.04^{f}
G3	25.6 ± 4.45^{c}	55.6 ± 3.65^{c}	0.62 ± 0.04^{c}
G4	30.2 ± 3.56^{c}	42.2 ± 5.63^{d}	0.55 ± 0.04^{d}
G5	56.0 ± 6.20^{b}	82.8 ± 6.22^{b}	0.68 ± 0.04^{b}
G6	14.8 ± 3.56^{de}	24.4 ± 4.04^{e}	0.36 ± 0.04^{e}
G7	18.0 ± 3.39^{d}	18.0 ± 4.95^{f}	0.34 ± 0.04^{e}
G8	12.8 ± 1.92^{ef}	29.8 ± 3.83^{e}	0.56 ± 0.06^{d}

Data were expressed as means ± SD. SD: Standard deviation. (a,b,c.....etc): means bearing difference superscripts within the same row are significantly different at ($p<0.05$).

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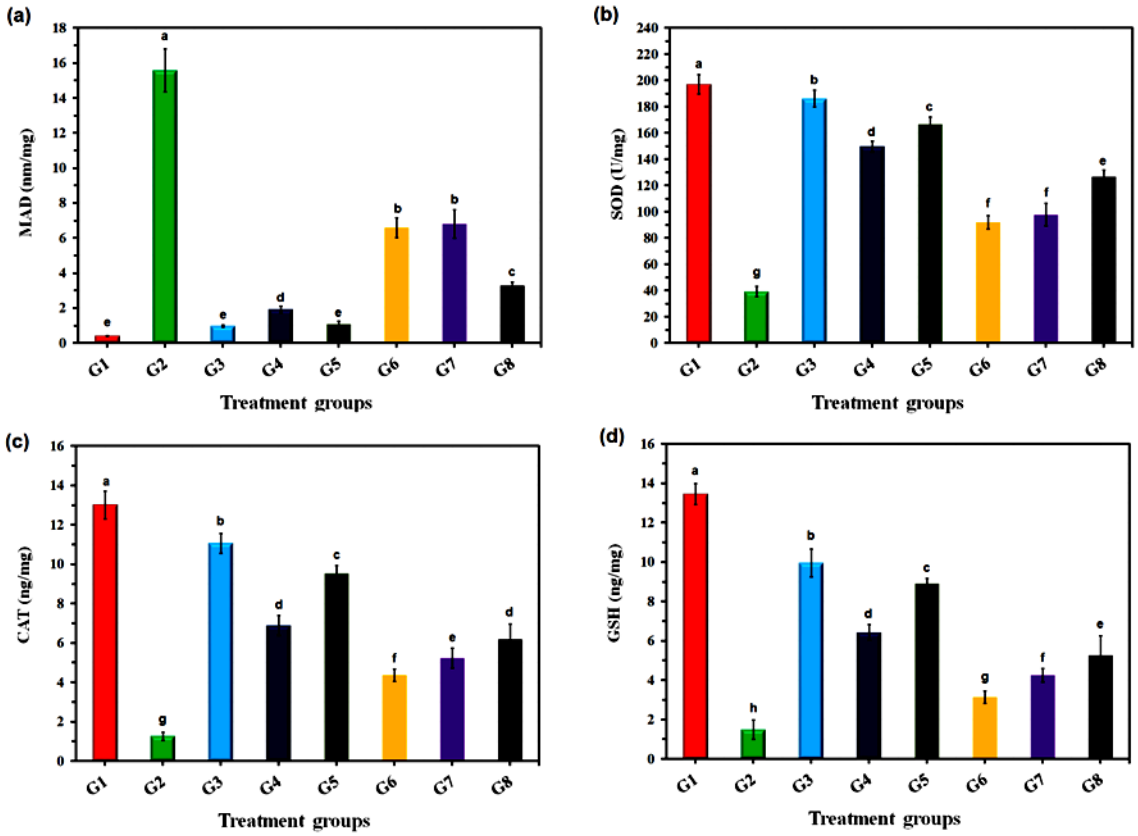

Figure 1. Effect of radiation (G2), vitamin c (G3) ginger extract (G4) vitamin c and ginger extract (G5) administration, vitamin c and radiation(G6), ginger extract and radiation (G7), vitamin c and ginger extract with radiation (G8) compared to Control Group (G1) on brain MAD (nmol/mg), SOD (U/mg), CAT (ng/mg) and GSH (ng/mg) of male albino rats.

Table 2. Effect of radiation (G2), vitamin C (G3) ginger extract (G4) vitamin c and ginger extract (G5) administration, vitamin c and radiation(G6), ginger extract and radiation (G7), vitamin c and ginger extract with radiation (G8) compared to Control Group (G1) on serum TNF (pg/ml), IL-6(pg/ml), IL-12 (pg/ml) and CRP (ng/m) of male albino rats.

Groups	TNF (pg/ml)	IL-6 (pg/ml)	IL-12 (pg/ml)	CRP (ng/m)
G1	26.0±2.74^{f}	131.0±2.92^{h}	2.4±0.36^{f}	34.0±6.44^{g}
G2	104.2±8.26^{a}	584.2±20.2^{a}	27.2±1.92^{a}	144.4±13.9^{a}
G3	39.6±3.71^{e}	240.2±7.16^{f}	5.0±1.58^{e}	55.0±3.16^{e}
G4	42.4±3.05^{e}	262.4±9.91^{e}	9.0±1.58^{d}	69.2±5.26^{d}
G5	30.8±2.77^{f}	153.8±0.78g	4.0±1.58^{ef}	45.0±4.12^{f}
G6	68.4±4.67^{c}	317.4±12.1^{c}	12.8±1.92^{c}	87.0±4.69^{c}
G7	79.4±7.06^{b}	489.2±8.64^{b}	17.6±2.07^{b}	114.6±10.4^{b}
G8	58.0±2.92^{d}	284.4±8.44^{d}	12.2±1.92^{c}	83.4±3.85^{c}

Data were expressed as means ± SD. SD: Standard deviation. (a,b,c.....etc): means bearing difference superscripts within the same row are significantly different at ($p<0.05$).

DISCUSSION

When mammals are exposed to ionizing radiation, they acquire a complicated, dose-dependent cascade of alterations, including organ injury and changes in the structure and function of cellular components [25]. One of the causes of radiation poisoning has been proposed to be oxidative stress and the subsequent formation of reactive oxygen species (ROS) [26]. Because of its high O_2 utilization rate, high content of polyunsaturated fatty acids, which are prone to lipid peroxidation, and high content of iron, which increases the generation of free radicals via Fenton reactions, the brain has been regarded a radiosensitive organ [27]. Furthermore, when compared to other organs, brain tissues are deficient in antioxidants [28]. The ionizing radiation is further harmful than non-ionizing radiation. The radiation effect produced by exposure to ionizing radiation affects people by depositing energy in body tissue which can cause cell damage or cell death. In other cases the cell may survive but become abnormal either temporarily or permanently or an abnormal cell may become malignant [29]. A very small amount of ionizing radiation could trigger cancer in long term even though it may take decades for cancer to appear and other effect with long term is the changes occur in (DNA) called mutations [30]. A variety of oxidative stressors, including radiation, can cause a disruption in pro-oxidant/anti-oxidant systems. As a result of the formation of free radicals, the pro-oxidant and antioxidant activities become unbalanced, resulting in cell death [31]. Low-molecular-weight antioxidant compounds including glutathione (GSH) and antioxidant enzymes make up the antioxidant system. The first line of defense against oxygen-derived free radicals is superoxide dismutase (SOD), which catalyses the dismutation of superoxide anion into H_2O_2. Catalase (CAT) that can break down H_2O_2 into H_2O and O_2. However oxidizing GSH, glutathione

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peroxidase (GSHPx) lowers lipidic or nonlipidic hydroperoxides as well as H_2O_2 [31]. The lowered activity of SOD and CAT, as well as the lower level of GSH in brain tissues, could be attributable to their increased use by ROS. Ahaskar and Sisodia discovered a substantial drop in GSH content in the whole brain after gamma irradiation (5Gy) [32]. Sandeep and Nair [33], found a considerable depletion in the antioxidant system and an increase in lipid peroxides in rats subjected to whole-body gamma-irradiation, which agrees with their findings. The activity of GSH and glutathione peroxidase (GSHPx) in reducing lipid hydroperoxides to stable non-radical lipid alcohols may have resulted in lower levels of GSH in the brains of irradiated animals, or GSH is directly used as an antioxidant in terminating the free radical reaction initiated by irradiation.

GSH depletion is frequently thought to be a sign of increasing oxidative stress, which can lead to tissue damage [34]. The increased creation of ROS, which interacts with enzyme molecules, causing denaturation and partial inactivation, may be the cause of the decrease in GSH levels and activity of SOD and CAT [35].

According to Abdel-Magied and Ahmed [36], a decrease in SOD activity can lead to an increase in super oxide flow in cellular compartments, which could explain the rise in MDA.

The current findings showed that -irradiation resulted in a considerable drop in GSH concentration and activity of SOD and CAT, as well as an increase in MDA activity. This could be due to a greater use of the antioxidant system in an attempt to detoxify free radicals produced by radiation [37]. Radiation-induced cell membrane damage and changes in dynamic permeability of membranes owing to peroxidation may cause a drop in antioxidant enzyme activity, which is followed by the release of intracellular enzymes into the bloodstream [38].

Sharma [39], hypothesized that 6 Gy gamma irradiation resulted in a significant decrease in glutathione content and glutathione peroxidase and catalase activities, as well as a significant increase in malondialhyde (MDA) levels in liver and heart tissues when compared to normal control mice.

The radical chain generation of lipid peroxidation can be disrupted by GSH, which is a powerful reducing agent. Peroxidation of membrane phospholipids is connected to an insufficient GSH content, according to Leedle and Aust [40]. During irradiation, the plasma concentrations of several antioxidants fell considerably [41]. Increased oxidative stress and free radical-induced tissue damage may result from lower levels of physiological antioxidant defense mechanisms.

GSH consumption can be increased by excessive lipid peroxidation. MDA elevation after radiation exposure could be linked to increased antioxidant system consumption in an effort to detoxify radiation -produced free radicals [42], which would explain the drop in SOD and CAT activities.

The decrease in GSH and increase in MDA are consistent with the findings of Abou-Zeid *et al.* [43], who discovered a significant drop in the antioxidant system and an improvement in the lipid peroxides following whole-body irradiation. The natural defense system, which includes GSH and antioxidant enzymes, defends against oxidative harm in its normal condition.

It has been suggested that the synergistic actions of multiple antioxidants are responsible for their powerful antioxidant and disease-prevention activities [44]. In combination with other nutrients, such as ascorbic acid and á-tocopherol, isoflavones may provide protection. Many free radical scavengers are beneficial in the fight against ray-induced nephrotoxicity [45]. According to Chen *et al.* [46], vitamin C decreases cisplatin-induced nephrotoxicity by lowering the generation of reactive oxygen species (ROS).

A substantial increase in total antioxidant content was seen in male Wistar rats subjected to three doses of gamma-ray (2, 4, and 8 Gy) with or without a 10-day pretreatment with ginger extract. These data suggest that whole-body radiation causes kidney damage via oxidative DNA damage and inflammatory reactions, and that these effects can be mitigated by utilizing ginger as an antioxidant and anti-inflammatory agent prior to radiation exposure [47].

The use of GE before and after -radiation helped to reduce the changes in antioxidant levels. GSH, GSHPx, Lipid peroxides (LP), and Nitric Oxide (NO) levels in gamma-irradiated rats treated with GE were nearly identical to control levels. When animals were given GE together with -radiation, the enzymes CAT and SOD exhibited no significant increase in their values. In other research works, mice treated with GE before irradiation showed a decrease in LP and an increase in GSH content [48-50]. After GE administration, the reduction of LP was accompanied by an increase in GSH, which was responsible for the scavenging of radiation-induced free radicals. In a dose-dependent manner, GE was discovered to scavenge OH and O_2 radicals [51]. Increased SOD levels in GE pre-treated animals may limit the production of the hydroxyl radical by dismutazing superoxide radicals [52]. Furthermore, GE increased CAT, which converts H_2O_2 into water, potentially preventing the generation of hydroxyl radicals [53]. One of the key probable mechanisms for the plant's protective activities against radiation toxicity and mortality has been proposed as the antioxidant action of GE [54]. As a result, it appears that GE affects antioxidant responses that are triggered by free radicals released by radiation. It has been demonstrated that gingerol, as a primary component of GE, has anti-inflammatory properties, possesses potent antioxidant, anti-inflammatory, and anti-apoptotic properties [55].

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The ability of ginger extract to scavenge superoxide anion and hydroxyl radicals was discovered. Gingerol, which suppressed the ascorbate/ferrous complex in rat liver microsomes, also reduced the lipid peroxidation [56]. 6-Dehydroshogaol, 6-shogaol, and 1-dehydro-6-gingerdione, all found in ginger extract, reduced nitric oxide generation in activated macrophages [57]. The role of phenolic compounds in giving electrons to H_2O_2 and neutralizing it to water is another probable reason for GE's antioxidative impact [58].

MDA levels were observed to be considerably lowered by VC in the current investigation. The lowered activity of SOD, CAT, and GST, as well as the lower amount of GSH in brain tissues, could be attributable to their increased use by ROS [59]. In rats subjected to whole-body gamma-irradiation, (Bhatia and Jain) found a considerable depletion in the antioxidant system, as well as an increase in lipid peroxides. The activity of GSH and glutathione peroxidase (GSHPx) in reducing lipid hydroperoxide to stable non-radical lipid alcohols may have resulted in lower levels of GSH in the brains of irradiated animals, or GSH is directly used as an antioxidant in terminating the free radical re-action initiated by irradiation. Under normal circumstances, the body's natural defense mechanism, which includes glutathione and anti-oxidant enzymes, can guard against oxidative damage. These endogenous enzymes, particularly SOD, CAT, GST, and GPx, are the major antioxidant system in cells and are responsible for the deactivation of ROS. SOD catalyzes the dismutation of the superoxide ion (O_2•) to H_2O_2 [60]. Gamma-irradiation lowered SOD activity in this study, but it was considerably recovered when rats were administered KV and VC after 8 weeks of exposure. CAT decomposes H_2O_2 into H_2O and O_2in the antioxidant cascade [61]. The effects of kolaviron (KV) and VC on the radiation-induced inhibition of CAT were dramatically improved. Glutathione *S*-transferases (GSTs) are a type of soluble protein that binds xenobiotics to glutathione [62] after glutathionylation, metabolites become more hydrophilic and consequently physiologically inactive. As a result, they are easily eliminated as conjugates in the bile or urine. As a result, this action is thought to be a key process in the etoxification of reactive species. The treatment of VC dramatically reduced GSH depletion in the brains of irradiated rats and increased GST activity, indicating that VC may protect exposed animals against gamma radiation-induced unfavorable effects.

During gamma radiation exposure, the activity of brain antioxidants goes through two phases. To begin with, there is a progressive increase that reaches a maximum, which is thought to be an early adaptive response to increased oxidative stress. Second, when free radical concentrations exceed the capacity of the body to neutralize them, there is a steady reduction [63]. Radiation-induced cell membrane damage and changes in dynamic permeability owing to peroxidation may also cause a drop in antioxidants, which is followed by the release of intracellular enzymes into the bloodstream [64].

Cholinesterases are a vast group of enzyme proteins that are found in both neuronal and non-neuronal tissues [65]. In the current study, irradiation of rats greatly increased acetylcholinesterase activity, an enzyme responsible for acetylcholine breakdown. Gamma irradiation of erythroleukemic K562 cells resulted in an increase in acetylcholinesterase activity, cell differentiation, and cell proliferation.

The current study discovered that in both the protective and therapeutic groups, there was a considerable increase in brain AchE activity. Wattanathorn, Jittiwat [65] previously observed that an alcohol extract of ginger might improve cognitive deficits and protect against brain injury in rats, which supports these findings [59].

According to Wang, [61], ginger aqueous extract increased cholinergic neuron function, inhibited AChE activity, increased the ratio of superoxide dismutase/malondialdehyde (SOD/MDA), and decreased MDA content in the brain, and Ghayur, Gilani [58] found that 70 percent aqueous/methanolic extract of ginger had a combination of muscarinic, Ca++ antagonist, and Butrylcho Both the protective and therapeutic investigations are supported by these findings. As a result, they discovered that all ginger extracts may work as anti-cholinesterase agents. These results are supported by earlier research by Wattanathorn *et al.* who showed that a ginger alcohol extract might lessen cognitive impairments and guard against brain damage in rats [65]. Additionally, ginger was observed to cause vasodilationand has the ability to enhance cerebral blood flow [66].

Serotonin and dopamine are essential for many different neural processes. Numerous neurodegenerative diseases have been shown to be correlated with changes in these neurotransmitter levels [67]. The findings of the current research demonstrated γ- radiation at sub-lethal dose of (6 Gy) caused a substantial reduction in the amount of biogenic amines in the brain tissues. It's possible that molecules involved with the serotonergic and dopaminergic systems have flaws as a consequence of the significant induction of oxidative stress [68]. The decreased levels of neurotransmitters found in this research may be due to radiation-induced damage, which causes a decrease in synthesis and reduced absorption [69]. Additionally, experimental data supports the different biochemical and functional changes in the brain that result from ionising radiation exposure. One of the causes of brain malfunction due to radiation-induced changes in these monoamines' metabolism is possible [70]. According to the current findings, various biogenic

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amines and ROS generation in the radiation-induced rat brain are strongly correlated. Natural antioxidants, in contrast to synthetic medications, are capable of achieving numerous site-specific tasks without being toxic to the host body [71]. These phytochemicals' potent capacity to pass through the blood-brain barrier may be due to their function as neuroprotectors [72]. In our study, rats pretreated with ginger extract and vitamin c for 14 days through orally showed increase in the neurotransmitter level of both dopamine and serotonin in the brain tissues.

In the current study, gamma-irradiation of rats resulted in elevated serum TNF- and IL-6, showing that these cytokines have a role in irradiation-induced damage. Similarly, other reaserchers [59] discovered that after radiation exposure, the production of TNF- and IL-6 was dramatically increased. Wistar rats were exposed to gamma radiation, which resulted in the creation of reactive oxygen species (ROS) that mediated the activation of the transcription factor NF-B. The activation of NF-B causes the expression of pro-inflammatory cytokine genes [IL-1, TNF-, and IL-6] to increase [60].

The activation of tumor necrosis factor alpha (TNF-a) and interleukin-1b (IL-1b) appears to be inhibited by ginger. 6-gingerol inhibited I-kappa B alpha phosphorylation, nuclear factor-kappa B (NF-kB) nuclear activation, and protein kinase C-alpha translocation, all of which reduced TNF-a expression. The synthesis of COX2 was stimulated by TNF-a, as was the activation of NF-kB by TNF-a. Furthermore, through inhibiting the COX-2 and lipoxygenase pathways, as well as inflammation-related pathways, these critical substances reduce the manufacture of prostaglandin and leukotriene. They can reduce inflammation this way [62]. Linalool, one of the main components of coriander essential oil, has strong anti-inflammatory properties, according to Heidari [63], because it suppresses pro-inflammatory mediator expression by reducing necrosis factor (NF)-kappa B. The natural chemicals in ginger have the ability to inhibit the synthesis of cytokines that are triggered by toxins, including TNF-, IL-6, IL-8, IL-2 and IL-1, PLA2, iNOS, COX-2, and PGE2 [73]. The antioxidant and ameliorative properties of ginger extracts may be due to the high quantities of polyphenolic and flavonoid components present [74, 75].

Oxidative stress is one of the factors that plays a key role in many radiation-induced damage pathways. Excessive generation of oxygen radicals affects enzyme function, DNA repair, oxygen usage, lipid peroxidation, and protein oxidation, among other things [64].

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REFERENCES

1. Jia C, Roman C, Hegg CC (2010) Nickel sulfate induces location-dependent atrophy of mouse olfactory epithelium: protective and proliferative role of purinergic receptor activation. *Toxicological Sciences,* ***115****(2): 547-56.*
2. Spitz DR, Azzam EI, Jian Li J, Gius D (2004) Metabolic oxidation/ reduction reactions and cellular responses to ionizing radiation: a unifying concept in stress response biology. *Cancer and Metastasis Reviews,* ***23****(3): 311-22.*
3. Aruoma OI (1998) Free radicals, oxidative stress, and antioxidants in human health and disease. *Journal of the American oil chemists' society,* ***75****(2): 199-212.*
4. Ness AR and Powles JW (1997) Fruit and vegetables, and cardiovascular disease: a review. *International Journal of epidemiology,* ***26****(1): 1-13.*
5. Jagetia GC, Venkatesh P, Baliga MS (2004) Fruit extract of Aegle marmelos protects mice against radiation-induced lethality. *Integrative cancer therapies,* ***3****(4): 323-32.*
6. Tohamy AA, El-Garawani IM, Ibrahim SR, Moneim AEA (2016) The apoptotic properties of Salvia aegyptiaca and Trigonella foenum-graecum extracts on Ehrlich ascites carcinoma cells: The effectiveness of combined treatment. *Res J Pharmaceutical Biological and Chemical Sciences,* ***7****(3): 1872-83.*
7. El-Nabi SH, Dawoud G, El-Garawani I, El-Shafey S (2018) HPLC analysis of phenolic acids, antioxidant activity and in vitro effectiveness of green and roasted Caffea arabica bean extracts: a comparative study. *Anti-Cancer Agents in Medicinal Chemistry (Formerly Current Medicinal Chemistry-Anti-Cancer Agents),* ***18****(9): 1281-8.*
8. Venkateswararao C and Venkataramana K (2013) A pharmacological review on natural antiulcer agents. *J Global Trends Pharmaceut Sci,* ***4****: 1118-31.*
9. Srinivasan K (2014) Antioxidant potential of spices and their active constituents. *Critical Reviews in Food Science and Nutrition,* ***54****(3): 352-72.*
10. Haniadka R, Saxena A, Shivashankara A, *et al.* (2013) Ginger protects the liver against the toxic effects of xenobiotic compounds: preclinical observations. *J Nutr Food Sci,* ***3****(5): 1000226.*
11. Szeto YT, To TL, Pak SC, Kalle W (2013) A study of DNA protective effect of orange juice supplementation. *Applied Physiology, Nutrition, and Metabolism,* ***38(5)****: 533-6.*
12. Suhail N, Bilal N, Khan H, *et al.* (2012) Effect of vitamins C and E on antioxidant status of breast-cancer patients undergoing chemotherapy. *J Clinical Pharmacy and Therapeutics,* ***37****(1): 22-6.*
13. Jagetia GC (2004) Ascorbic acid treatment reduces the radiation-induced delay in the skin excision wound of Swiss mice. *Indian Journal of Radiation Research,* ***1(2)****: 7.*
14. Mallikarjuna Rao K (2004) Ascorbic acid treatment enhances the wound healing in mice exposed to radiation. *Indian J Radiat Res,* ***1****: 24.*
15. Mahdavi M and Mozdarani H (2011) Protective effects of famotidine and vitamin C against radiation induced cellular damage in mouse spermatogenesis process. *Int J Radiat Res,* ***8****(4): 223-230.*

[DOI: 10.61186/ijrr.22.4.1027]

16. Ajith T, Aswathy M, Hema U (2008) Protective effect of Zingiber officinale roscoe against anticancer drug doxorubicin-induced acute nephrotoxicity. *Food and chemical toxicology,* ***46****(9): 3178-81.*
17. Beutler E (1963) Improved method for the determination of blood glutathione. *J lab clin Med,* ***61****: 882-8.*
18. Masayasu M and Hiroshi Y (1979) A simplified assay method of superoxide dismutase activity for clinical use. *Clinica chimica acta,* ***92****(3): 337-42.*
19. Rick W and Stegbauer HP (1974) α-Amylase measurement of reducing groups. Methods of enzymatic analysis: Elsevier; **1974**: 885 -90.
20. Farombi EO (2000) Mechanisms for the hepatoprotective action of kolaviron: studies on hepatic enzymes, microsomal lipids and lipid peroxidation in carbontetrachloride-treated rats. *Pharmacological Research,* ***42****(1): 75-80.*
21. Ellman GL, Courtney KD, Andres Jr V, Featherstone RM (1961) A new and rapid colorimetric determination of acetylcholinesterase activity. *Biochemical Pharmacology,* ***7****(2): 88-95.*
22. Taylor PC, Williams RO, Maini RN (2001) Immunotherapy for rheumatoid arthritis. *Current opinion in immunology,* ***13****(5): 611-6.*
23. Beutler B, Greenwald D, Hulmes J, *et al.* (1985) Identity of tumour necrosis factor and the macrophage-secreted factor cachectin. *Nature,* ***316****(6028): 552-4.*
24. Levesque R (2007) SPSS programming and data management. *A guide for SPSS and SAS Users.*
25. Algeda FR and Ebrahim RM (2020) The efficacy of royal jelly in the restoration of liver injury in irradiated rats. *Egyptian Journal of Radiation Sciences and Applications,* ***33****(2): 77-88.*
26. Tawfik S and Salama S (2008) Possible protective role of carnosine against gamma-radiation-induced cardiac dysfunction in mice. *Egyptian Journal of Radiation Sciences and Applications,* ***21****(2): 341 -56.*
27. Halliwell B (2001) Role of free radicals in the neurodegenerative diseases. *Drugs & aging,* ***18****(9): 685-716.*
28. Toren Fand Nikki JH (2000) Oxidants, oxidative stress and the biology of ageing. *Nature,* ***408****(6809): 239-47.*
29. Verma P, Jahan S, Kim TH, Goyal PK (2011) Management of radiation injuries by Panax ginseng extract. *Journal of Ginseng Research,* ***35****(3): 261.*
30. Ezz MK (2011) The ameliorative effect of Echinacea purpurea against gamma radiation induced oxidative stress and immune responses in male rats. *Australian Journal of Basic and Applied Sciences,* ***5****(10): 506-12.*
31. Pizzino G, Irrera N, Cucinotta M, *et al.* (2017) Oxidative stress: harms and benefits for human health. *Oxidative Medicine and Cellular Longevity,* ***2017****.*
32. Ahaskar M and Sisodia R (2006) Modulation of radiation induced biochemical changes in brain of swiss albino mice by Grewia asiatica. *Asian J Exp Sci,* ***20****(2): 399-404.*
33. Sandeep D and Nair CKK (2012) Protection from lethal and sublethal whole body exposures of mice to γ-radiation by Acorus calamus L.: Studies on tissue antioxidant status and cellular DNA damage. *Experimental and Toxicologic Pathology,* ***64****(1-2): 57-64.*
34. Andreoli SP, Mallett CP, Bergstein JM (1986) Role of glutathione in protecting endothelial cells against hydrogen peroxide oxidant injury. *The Journal of laboratory and Clinical Medicine,* ***108****(3): 182 -9.*
35. Kregel KC and Zhang HJ (2007) An integrated view of oxidative stress in aging: basic mechanisms, functional effects, and pathological considerations. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology,* ***292****(1): R18-R36.*
36. Abdel-Magied N and Ahmed A (2011) Efficacy of clove oil as an antioxidant against radiation risk in male rats. *Journal of Radiation Research and Applied Sciences,* ***4****(3b): 939-55.*
37. Krishna A and Kumar A (2005) Evaluation of radioprotective effects of Rajgira (Amaranthus paniculatus) extract in Swiss albino mice. *Journal of Radiation Research,* ***46****(2): 233-9.*
38. Saada H, Ussama Z, Mahdy A (2003) Effectiveness of Aloe vera on the antioxidant status of different tissues in irradiated rats. *Die Pharmazie-An International Journal of Pharmaceutical Sciences,* ***58*** *(12):* 929-31.
39. Sharma J, Sharma R, Mathur A (2013) Protection of Mouse Liver from Gamma Ray Exposure: A Review. *Int J Pharm Bio Sci,* ***4****(1): 1011-26.*
40. Leedle R and Aust S (1990) The effect of glutathione on the vitamin E requirement for inhibition of liver microsomal lipid peroxidation. *Lipids,* ***25(****5): 241-5.*
41. Shedid SM, Abdel-Magied N, Saada HN (2019) Role of betaine in liver injury induced by the exposure to ionizing radiation. *Environmental toxicology,* ***34****(2): 123-30.*
42. Mansour HH (2013) Protective effect of ginseng against gamma-irradiation-induced oxidative stress and endothelial dysfunction in rats. *EXCLI journal,* ***12****: 766.*
43. Abou-Zeid SM, El-Bialy BE, El-Borai NB, AbuBakr HO, Elhadary AMA (2018) Radioprotective effect of date syrup on radiation-induced damage in rats. *Scientific reports,* ***8****(1): 1-10.*
44. WaNg H, MartiN MW, YiN S (2010) The synergistic effect of daidzein and α-tocopherol or ascorbic acid on microsome and LDL oxidation. Czech Journal of Food Sciences, **28**(5): 385-91.
45. Elkady A and Ibrahim I (2016) Protective effects of erdosteine against nephrotoxicity caused by gamma radiation in male albino rats. *Human & Experimental Toxicology,* ***35****(1): 21-8.*
46. Chen M-F, Yang C-M, Su C-M, Hu M-L (2014) Vitamin C protects against cisplatin-induced nephrotoxicity and damage without reducing its effectiveness in C57BL/6 mice xenografted with Lewis lung carcinoma. *Nutrition and Cancer,* ***66****(7): 1085-91.*
47. Jagetia GC, Baliga MS, Venkatesh P, Ulloor JN (2003) Influence of ginger rhizome (Zingiber officinale Rosc) on survival, glutathione and lipid peroxidation in mice after whole-body exposure to gamma radiation. *Radiation Research,* ***160****(5): 584-92.*
48. Parihar VK, Prabhakar K, Veerapur VP, *et al.* (2006) Effect of sesamol on radiation-induced cytotoxicity in Swiss albino mice. *Mutation Research/Genetic Toxicology and Environmental Mutagenesis,* ***611****(1-2): 9-16.*
49. Parihar VK, Dhawan J, Kumar S, *et al.* (2007) Free radical scavenging and radioprotective activity of dehydrozingerone against whole body gamma irradiation in Swiss albino mice. *Chemico-biological interactions,* ***170(1)****: 49-58.*
50. Haksar A, Sharma A, Chawla R, *et al.* (2006) Zingiber officinale exhibits behavioral radioprotection against radiation-induced CTA in a gender-specific manner. *Pharmacology Biochemistry and Behavior,* ***84****(2): 179-88.*
51. Kim J-K, Kim Y, Na K-M, *et al.* (2007) [6]-Gingerol prevents UVB-induced ROS production and COX-2 expression *in-vitro* and *in-vivo*. *Free Radical Research,* ***41****(5): 603-14.*
52. Bellik Y (2014) Total antioxidant activity and antimicrobial potency of the essential oil and oleoresin of Zingiber officinale Roscoe. *Asian Pacific Journal of Tropical Disease,* ***4****(1): 40-4.*
53. Khader M, Bresgen N, Eckl P (2010) Antimutagenic effects of ethanolic extracts from selected Palestinian medicinal plants. *Journal of Ethnopharmacology,* ***127****(2): 319-24.*
54. Bhatia A and Jain M (2004) Spinacia oleracea L. protects against gamma radiations: a study on glutathione and lipid peroxidation in mouse liver. *Phytomedicine,* ***11****(7-8): 607-15.*
55. Said U (2004) Ne uroprotective properties of rutin in radiation-induced biochemical disorders in brain of rats. *Arab Journal of Nuclear Sciences and Applications,* ***37****(3): 223-31.*
56. Schwenke K, Linow KJ, Zirwer D (1986) Modification of the oligomeric structure of 11 S globulin from sunflower (Helianthus annuus L.) and rape (Brassica napus L.) seeds by succinylation. *Food/ Nahrung,* ***30****(3-4): 263-70.*
57. Wang J, Hang Q, Jia S (2006) Effect of aqueous extract of ginger on vascular dementia in rats. *Journal of Medical Research.*
58. Ghayur MN, Gilani AH, Ahmed T, *et al.* (2008) Muscarinic, Ca++ antagonist and specific butyrylcholinesterase inhibitory activity of dried ginger extract might explain its use in dementia. *Journal of Pharmacy and Pharmacology,* ***60****(10): 1375-83.*
59. Shah KG, Wu R, Jacob A, *et al.* (2009) Human ghrelin ameliorates organ injury and improves survival after radiation injury combined with severe sepsis. *Molecular medicine,* ***15****(11): 407-14.*
60. Godbout J, Berg B, Krzyszton C, Johnson R (2005) α-Tocopherol attenuates NFκB activation and pro-inflammatory cytokine production in brain and improves recovery from lipopolysaccharide-induced sickness behavior. *Journal of Neuroimmunology,* ***169****(1-2): 97-105.*
61. Wang J, Ke W, Bao R, *et al.* (2017) Beneficial effects of ginger Zingiber officinale Roscoe on obesity and metabolic syndrome: a review. *Annals of the New York Academy of Sciences,* ***1398****(1): 83-98.*
62. Naderi Z, Mozaffari-Khosravi H, Dehghan A, *et al.* (2016) Effect of ginger powder supplementation on nitric oxide and C-reactive protein in elderly knee osteoarthritis patients: A 12-week double-blind randomized placebo-controlled clinical trial. *Journal of Traditional and Complementary Medicine,* ***6****(3): 199-203.*
63. Heidari B, Sajjadi SE, Minaiyan M (2016) Effect of Coriandrum sativum hydroalcoholic extract and its essential oil on acetic acid-

[DOI: 10.61186/ijrr.22.4.1027]

induced acute colitis in rats. *Avicenna Journal of Phytomedicine,* ***6*** *(2): 205.*
64. Kurose I, Argenbright LW, Wolf R, Granger DN (1996) Oxidative stress during platelet-activating factor-induced microvascular dysfunction. *Microcirculation,* ***3****(4): 401-10.*
65. Wattanathorn J, Jittiwat J, Tongun T, *et al.* (2010) Zingiber officinale mitigates brain damage and improves memory impairment in focal cerebral ischemic rat. *Evidence-Based Complementary and Alternative Medicine,* ***2011****.*
66. Ghayur MN and Gilani AH (2005) Ginger lowers blood pressure through blockade of voltage-dependent calcium channels. *Journal of Cardiovascular Pharmacology,* ***45****(1): 74-80.*
67. Xu Y, Yan J, Zhou P, Li J, *et al.* (2012) Neurotransmitter receptors and cognitive dysfunction in Alzheimer's disease and Parkinson's disease. *Progress in Neurobiology,* ***97****(1): 1-13.*
68. Maiti P, Manna J, Dunbar GL (2017) Current understanding of the molecular mechanisms in Parkinson's disease: Targets for potential treatments. *Translational Neurodegeneration,* ***6****(1): 6-28.*
69. Zhi W-J, Wang L-F, Hu X-J (2017) Recent advances in the effects of microwave radiation on brains. *Military Medical Research,* ***4****(1): 1-14.*
70. Said U, Saada H, Amin A, *et al.* (2012) Efficacy of hesperidin in modulating radiation-induced brain damage in rats. *J Radiat Res Appl Scien,* ***5****(1): 71-85.*
71. Tan BL, Norhaizan ME, Liew W-P-P, Sulaiman Rahman H (2018) Antioxidant and Oxidative Stress: A Mutual Interplay in Age-Related Diseases. *Frontiers in Pharmacology,* ***9****: 1-28.*
72. Kumar GP and Khanum F (2012) Neuroprotective potential of phytochemicals. *Pharmacognosy Reviews,* ***6****(12): 81.*
73. Zaghlool SS, Shehata BA, Abo-Seif AA, Abd El-Latif HA (2015) Protective effects of ginger and marshmallow extracts on indomethacin-induced peptic ulcer in rats. *Journal of Natural Science, Biology, and Medicine,* ***6****(2): 421.*
74. Danwilai K, Konmun J, Sripanidkulchai B-O, Subongkot S (2017) Antioxidant activity of ginger extract as a daily supplement in cancer patients receiving adjuvant chemotherapy: a pilot study. *Cancer Management and Research, 11-8.*
75. Nasri H, Mortazavi M, Ghorbani A, *et al.* (2012) Oxford-MEST classification in IgA nephropathy patients: A report from Iran. *Journal of Nephropathology,* ***1****(1): 31.*

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