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An Evaluation of Effects of Black Grape and Ginger Extracts on Hematological Alteration and Lipid Peroxidation of Hepatocyte in Irradiated Albino Mice

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ABSTRACT

Introduction: Exposure to ionizing radiation can trigger adverse biological effects on healthy tissues, such as causing hematological toxicity and potential injury to different organs. Ionizing radiation has sufficient energy to liberate electrons from atoms leaving them with unpaired electrons; hence ionizing them and producing free radicals. In the current study, the risk of exposing to 6 Gy x-irradiation on alteration of some hematological parameters and liver tissue lipid peroxidation activity in albino mice in the presence and absence of black grape and ginger extracts as antioxidants have been investigated.

Material and Methods: Albino mice were exposed to 6 Gy wholebody xirradiation in the absence and presence of black grape and ginger extracts (10mL/Kg).

Results: The results of the present study showed a significant decrease in mice red blood cell count, *hemoglobin*, hematocrit, mean corpuscular volume, and mean corpuscular hemoglobin, following the exposure to 6 Gy x-irradiation ($P \leq 0.05$), while all the mentioned parameters were relatively shifted toward the normal values in the mice that received black grape and ginger extracts as a treatment prior to the radiation. Accordingly, our results demonstrated a noticeable increase in the malondialdehyde analysis (MDA) level in the 6 Gy group, while a slight depletion in the MDA level was observed in the group of mice that received black grape and ginger extracts, compared to that of the control group.

Conclusion: the administration of black grape and ginger extracts prior to irradiation may protect the mice from excess hepatocyte lipid peroxidation and alteration of hematological parameters.

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Introduction

Human exposure to ionizing radiation has increased as it is a central component of medical diagnosis and therapy. Studies showed that nearly half of the patients have been treated with radiotherapy for palliative or curative purposes [1]. However, exposure to ionizing radiation can trigger different primary or secondary adverse biological effects on healthy tissues, such as causing hematological toxicity, immune suppression, and potential injury to the liver and other organs [2, 3]. Ionizing radiation has sufficient energy to liberate electrons from atoms leaving them with unpaired electrons; hence ionizing them and producing free radicals [4]. Free radicals are considered an unstable destructive species causing cellular damages and oxidation of protein, DNA, lipid, and carbohydrate. Moreover, free radicals are produced either from normal metabolic reactions that occur in the body or from exposure to chemicals or radiations. All these factors can accumulate and accelerate tissue-damaging progress and eventually leading to many serious diseases, such as carcinoma, atherosclerosis, and Alzheimer's, especially when these free radicals are produced continuously at high levels [5]. With this background in mind, the scale of cell damage and the impairment of the biological system, all can be regarded as parameters for assessing the effect of radicals produced by radiation.

The hematopoietic system is one of the most essential systems that maintains and organizes mammalian life and is considered to be one of the

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most radiosensitive systems [6]. The short-term effects of ionizing radiation on the hematopoietic system can lead to several risky disorders (e.g., hemorrhage and cytopenia), while the long-term effect may cause most threatening defects, such as leukemia [7].

It has been reported that the liver is considered as another radiosensitive organ [8, 9]. Studies on patients with healthy liver who subjected to ionizing radiation as a part of radiotherapy showed that ionizing radiation causes liver damage, cirrhosis, dysfunction, and unexpected cancer [10, 11]. It has been indicated that the severity of immediate radiation effects depends on the total radiation dose [2]. Clearly, lipid is an organic compound that plays an important role in the cell structure, especially in cell membrane composition; it effortlessly can be affected by radiation and degraded them upon oxidation. When free radicals (R) attack lipid (LH), particularly the unsaturated fatty acid, they produce lipid peroxides (LOOH), aldehydes, and ketones as shown in the following reaction steps [12, 13]. A study showed that an increase in the number of double bonds in the lipid structure results in more lipids' oxidation [14].

$LH + R \rightarrow L\cdot + RH$

$L\cdot + O2 \rightarrow LOO\cdot$

$LOO\cdot + LH \rightarrow LOOH + L\cdot$

$LOOH \rightarrow LO\cdot + LOO\cdot + \text{aldehydes}$

In order to minimize the harmful effects of free radicals, many natural or synthetic antioxidants have been investigated to find safe and effective radio-protecting agents that can scavenge these radicals. Natural products can be selected as one of the non-enzymatic antioxidants due to some components that can attenuate the oxidative stress and consequently reducing apoptosis, such as polyphenols, terpenoids, vitamins, tocopherols, sulfadryls, and metals [15].

Investigations have demonstrated that grape and ginger extracts could be efficient protectors against radicals induced by radiation which are used in the current study [16, 17]. Grape is considered a good antioxidant due to its different phenolic components, such as anthocyanins, catechins, resveratrols, phenolic acids, and procyanidins [18]. Studies showed that the grape seed and skin extracts increase the defense capacity against oxidation. The grape seed extract is currently used as a supplement because of its high phenolic and flavonoid contents [19]. Furthermore, ginger due to the presence of gingerol compounds has the potential of reducing the lipid peroxidation [20]. The extracted gingerols are composed of gingerol, shagaols, gengediols, zingerone, dehydrozingerone, gingerinone, and diarylheptanoids [21, 22].

In the current study, the risk of exposing to 6 Gy x-irradiation on alteration of some hematological parameters and liver tissue lipid peroxidation activity in albino mice in the presence and absence of black grape and ginger extracts as antioxidants has been investigated.

Materials and Methods

Experimental Animals

In the present experimental study, 30 male albino mice with the age range of 10-12 weeks weighting 30-45 gm were obtained from the University of Sulaimaniyah Animal House, College of Education, Department of Biology. The mice were acclimated to the laboratory conditions for two weeks and were accommodated under controlled temperature (23±2°C) and 12 h of the light-dark cycle. Throughout the research period, they had free access to drinking water and standard food. All animals were kept under human care in accordance with the National Institutes of Health guidelines for moral treatment of the laboratory animals.

Experimental Design

The animals were randomly divided into six different groups, five mice in each group. The control and treated groups were as the following: group I (negative control group) received deionized water (10mL/Kg) by oral gavage for eight consecutive days. Group II (6 Gy group) orally received only deionized water for eight days and then were subjected to 6 Gy whole-body x-irradiation which was applied as one-shot dose. Group III (BG-group) orally received only grape extract (10mL/Kg) for eight days without exposure to radiation. Group IV (6 Gy+BG group) orally received black grape (10mL/Kg) for eight days prior to irradiation and then were exposed to whole-body x-irradiation at 6 Gy. Group V (GN-group) orally received only ginger extract (10mL/Kg) for eight days without exposure to radiation. Group VI (6 Gy+GN) received ginger extract (10mL/Kg) for eight days before the irradiation and then were subjected to 6 Gy whole-body x-irradiation.

Ginger and Grape Extract Preparation

The black grapes were grinded in a blender for five min, 100mL of black grape was extracted and then mixed with 25mL of 96% of ethanol for 30 min to increase phenolic extraction. The mixture was warmed for evaporation of the residual ethanol and was returned back the volume to 100mL. At the final step, the extract was filtered using a piece of fabric and was stored at 4˚C [23]. In addition, the ginger rhizome was grinded and filtered using a piece of fabric and was stored at 4˚C [24].

Irradiation

Animals in the 6 Gy, 6 Gy+BG, and 6 Gy+GN groups were kept in a separate plastic container and irradiated to a single dose of 6 Gy whole-body x-irradiation from a 6 MV x-ray linear accelerator machine (Elekta Synergy 6 MV, UK). The absorbed dose rate was 4 Gy/min at a 100cm distance.

Sample Collection and Analysis

After 24 h of exposure to radiation, the mice were sacrificed and their blood and liver tissue samples were immediately collected. Animals were anesthetized using diethyl-ether and blood samples were collected from their inferior vena cava using a sterile syringe. The blood samples were collected in anticoagulant tubes for a complete blood count. Hematological examination includes: red blood cell (RBC) (x 10^6/millimeter3), hemoglobin (Hb) (milligram/deciliter), hematocrit (HCT) (percentage), mean corpuscular volume (MCV) (femtoLiter), mean corpuscular hemoglobin (MCH) (pictogram), red blood cell distribution width (RDW) (percentage) using automatic hematology analyzer Swelab Alfa Standard analyzer Approximately 25mg of liver tissue was weighted out immediately after mice were sacrificed for malondialdehyde analysis (MDA) by reaction with thiobarbituric acid which was colorimetrically quantified . The spectroscopic determination of MDA included instant preparation of sample and standard solutions. A volume of 100 □L sample or standard series solutions were taken, mixed with 100 □L sodium dodecyl sulfate, and then 4mL of the coloring reagent were added. The tubes were capped and put in a boiling water bath for one hour then removed and cooled immediately. The cooled solutions were submitted to centrifugation at 1600x g at 4˚C to clear the solutions. Finally, the solutions were read at 532nm by (EMC-LAB UV-6100PC double beam spectrophotometer / 2nm slit-width.

Statistical Evaluation

The obtained data were statistically analyzed using one-way anova, post hoc analysis in excel. A p-value of less than 0.05 was considered statistically significant.

Results

Some hematological parameters were examined in order to assess the deteriorating effect of x-irradiation on the hematopoietic system of albino mice in the presence of grape and ginger extracts as antioxidants. The results demonstrated a significant decrease ($P \leq 0.05$) in the RBCs count, Hb, HCT, MCV, and MCH in the 6Gy group mice that were exposed to whole-body x-irradiation, compared to those in the control group. However, all parameters were significantly elevated in both BG and GN groups ($P \leq 0.05$). The RDW value showed an increase in the 6 Gy group; however, it showed a slight reduction in BG and GN groups, compared to the control group. The RBC, Hb, HCT, MCV, MCH, and RDW values were relatively improved and shifted toward the normal values ($P \leq 0.05$) in the mice that received black grape and ginger extracts as a treatment prior to the radiation, corresponding to 6 Gy+BG and 6 Gy +GN groups, respectively (Table 1).

After exposing the mice to the 6 Gy x-irradiation, the effects appeared instantly and a decrease was observed in their physical activities. In the current study, the MDA was implicated as a parameter for measuring oxidative stress in the hepatic tissues (e.g., liver) and it was considered less tolerant toward the radical attacks [8]. Accordingly, our results signified a noticeable increase in the MDA level in the 6 Gy group, compared to that of the control group. Moreover, a slight depletion in the MDA level was observed in the group of mice that received black grape and ginger extracts, compared to the control group. Furthermore, the obtained results showed a significant reduction ($P \leq 0.05$) in the MDA level in both treated groups with black grape and ginger extracts following exposure to 6 Gy x-irradiation, compared to the group of mice that exposed to x-irradiation without treatment (Table 2). The findings of the present study suggested that both plants were efficient for fighting radicals induced by ionized radiation.

Table1. Hematological parameters in the control and x-irradiated mice with/without grape juice and ginger extract groups. Data are represented as (mean±SD), (n=5)

Parameters	RBC (x 10^6/mm^3)	Hb (mg/dl)	HCT (%)	MCV (fL)	MCH (pg)	RDW(%)
Control	9.4±0.33	13.5±0.19	46.6±0.06	49.9±0.07	14.3±0.14	16.6±0.07
6 Gy	7.7±0.48	10.75±0.35	36.8±0.20	47.3±0.20	13.7±0.15	17.2±0.14
Grape	9.5±0.40	14.6±0.07	51.9±0.41	53.9±0.41	14.8±0.32	16.02±0.21
Grape+6 Gy	8.1±0.49	11.8±0.03	40.0±0.07	48.8±0.57	14.4±0.35	15.1±0.28
Ginger	8.7±0.14	13.2±0.05	44.0±0.14	51.2±0.09	14.5±0.14	16.1±0.33
Ginger+6 Gy	8.0±0.04	11.5±0.42	38.1±0.04	47.4±0.04	14.2±0.05	14.9±0.10

RBC: red blood cell
Hb: hemoglobin
HCT: hematocrit
MCV: mean corpuscular volume
MCH: mean corpuscular hemoglobinRDW: red blood cell distribution width

Table 2. Malondialdehyde analysis levels in irradiated hepatocytes groups of control, treated, and non-treated with black grape and ginger extracts. Data are represented as (mean±SD), (n=5).

Groups	MDA (µg/gm)
Control	0.168±0.005
6 Gy	0.397±0.0262
Grape	0.166±0.0107
Grape+6 Gy	0.221±0.008
Ginger	0.158±0.0125
Ginger+6 Gy	0.293±0.0179

MDA: Malondialdehyde analysis

Discussion

Despite wide applications of ionizing radiation, it can cause various harmful conditions, such as alterations in the hematopoietic system [25]. In the current research, the RBCs count of the mice decreased at 6 Gy x-irradiation. Similar findings were previously reported by Xu et al. (2012) indicated that after irradiation, the proportion of normal RBCs count reduces [26]. In addition, a comparable alteration was observed by Sancheti et al. (2007) who found a noticeable depletion in the RBCs count in the mice who were exposed to 3 Gy gamma radiations [27]. Drop in the RBC count might be an indicator of increasing erythrocyte permeability and alteration in the cell membrane stability [28, 29, 30]. Radiation induces lipid peroxidation as a result of oxidative stress; blood cells are particularly sensitive to oxidative stress as their plasma membranes contain a high percentage of polyunsaturated fatty acids [31]. Therefore, radiation can damage their cell membranes and this leads to decrease in the RBCs count [32]. The obtained results also noted a significant decrease in Hb, HCT, MCV, and MCH levels in irradiated mice, this was in accordance with the result of a study carried out by Abdelhalim et al. (2015) which reported that the value of Hb, HCT, MCV, and MCH decreased after radiation [33]. These results are also in agreement with earlier findings of Daga et al. (1995) which reported a reduction in the hemoglobin concentration in the gamma-irradiated mice [34]. It can be related to either direct destruction or loss of the RBCs in circulation due to hemorrhage or leakage through capillary walls [35]. The RDW value increased in mice that exposed to 6 Gy x-irradiation, these results were in line with the findings of the study conducted by Mehrotra et a. (2013) which indicated a relationship between radiation and an increase in the RDW value [36]. The slight increase in the RDW value confirmed the presence of variation in the size of RBCs which indicated changes in morphology and deformability of RBCs [33]. Groups of 6 Gy with black grape and ginger extracts showed a significant increase in RBC, Hb, HCT, MCV, and MCH; however, a reduction was observed in the RDW value, compared to 6 Gy group without treatment, which is suggestive of black grape and ginger extracts administration. Black grape and ginger extracts are protective candidate compounds against alteration in some hematological parameters in mice. Moreover, it was reported by El-Desouky et al. (2017) that radiation groups treated with mixture of grape seed extract and green tea showed an increase in all hematological parameters [37]. Singha et al. (2016) investigated the radio-protective action of four grape extracts from different sources, blood samples from healthy volunteers mixed with grape extracts and exposed to 4 Gy radiation [38]. The results showed that the pre-treatment grape extract can ameliorate the deteriorating effect of ionizing radiation on human RBCs in vitro. In a study it was revealed that administration of 10mg/kg or 250mg/kg of ginger hydroalcoholic extract for five consecutive days showed protective features for mice against radiation sickness, gastrointestinal, and bone marrow defects [39].

According to the results of the current research, reduction in the MDA levels followed by administration of black grape and ginger extracts might be due to the fact that these plants possess significant antioxidant properties which can scavenge the oxygen free radicals and consequently inhibit lipid peroxidation and reduce cell death and fibrosis expectancy [38, 40, 41]. In addition, it has been reported that grapes can reduce lipid peroxidation in the hepatocytes produced after exposure to acute irradiation [42]. The antioxidant property of grape and ginger might be due to the presence of some active components, such as polyphenols and flavonoids [40, 41].

Conclusion

Based on the findings of the present study, the administration of black grape and ginger extracts prior to irradiation protects the mice from oxidative stress effects and consequently inhibits excessive hepatocyte lipid peroxidation and alteration of hematological parameters.

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References

1. Jung J, Kim H, Yoon S, Cho B, Kim Y, Kwak J, et al. Targeting Accuracy of Image-Guided Stereotactic Body Radiation Therapy for Hepatocellular Carcinoma in Real-Life Clinical Practice: In Vivo Assessment Using Hepatic Parenchymal Changes on Gd-EOB-DTPAEnhanced Magnetic Resonance Images. International Journal of Radiation Oncology*Biology*Physics. 2018; 102(4):867-74.
2. Guha C, Kavanagh BD. Hepatic radiation toxicity: avoidance and amelioration. InSeminars in radiation oncology. 2011;21(4): 256-63.
3. Lawrence TS, Robertson JM, Anscher MS, Jirtle RL, Ensminger WD, Fajardo LF. Hepatic toxicity resulting from cancer treatment. International Journal of Radiation Oncology* Biology* Physics. 1995;31(5):1237-48.
4. Kobashigawa S, Kashino G, Suzuki K, Yamashita S, Mori H. Ionizing radiation-induced cell death is

partly caused by increase of mitochondrial reactive oxygen species in normal human fibroblast cells. Radiat. Res. 2015; 183: 455‑64.
5. Lobo V, Patil A, Phatak A, Chandra N. Free radicals, antioxidants and functional foods: impact on human health. Pharmacogn Rev. 2010; 4:118‑26.
6. Smirnova OA. Environmental radiation effects on mammals: a dynamical modeling approach. Springer; 2016.
7. Akushevich IV, Veremeyeva GA, Dimov GP, Ukraintseva SV, Arbeev KG, Akleyev AV, et al. Modeling hematopoietic system response caused by chronic exposure to ionizing radiation. Radiation and environmental biophysics. 2011;50(2):299-311.
8. Rösler P, Christiansen H, Kortmann RD, Martini C, Matuschek C, Meyer F, et al. Hepatotoxicity after liver irradiation in children and adolescents. Strahlentherapie und Onkologie. 2015;191(5):413-20.
9. Kim J, Jung Y. Radiation-induced liver disease: current understanding and future perspectives. Experimental & molecular medicine. 2017;49(7):359.
10. Ozasa K, Shimizu Y, Suyama A, Kasagi F, Soda M, Grant EJ, et al. Studies of the mortality of atomic bomb survivors, Report 14, 1950‑2003: an overview of cancer and noncancer diseases. Radiation research. 2011;177(3):229-43.
11. Zielinski J, Garner M, Band P, Krewski D, Shilnikova N, Jiang H, et al. Health outcomes of low-dose ionizing radiation exposure among medical workers: a cohort study of the Canadian national dose registry of radiation workers. International journal of occupational medicine and environmental health. 2009;22(2):149-56.
12. De Freitas R, Augusti P, De Andrade E, Rother F, Rovani B, Quatrin A, et al. Black Grape Juice Protects Spleen from Lipid Oxidation Induced by Gamma Radiation in Rats. Journal of Food Biochemistry. 2012;38(1):119-27.
13. Nimse SB, Pal D. Free radicals, natural antioxidants, and their reaction mechanisms. Rsc Advances. 2015;5(35):27986-8006.
14. Niki E, Yoshida Y, Saito Y, Noguchi N. Lipid peroxidation: mechanisms, inhibition, and biological effects. Biochemical and biophysical research communications. 2005;338(1):668-76.
15. Sree KK, Srinivasan V, Toles R, Jobe L, Seed TM. Nutritional approaches to radioprotection: vitamin E. Military medicine. 2002;167(suppl_1):57-9.
16. Andrade ER, Cruz IB, Andrade VV, Piccoli JC, González-Gallego J, Barrio JP, et al. Evaluation of the potential protective effects of ad libitum black grape juice against liver oxidative damage in whole-body acute X-irradiated rats. Food and chemical toxicology. 2011;49(4):1026-32.
17. Mashhadi NS, Ghiasvand R, Askari G, Hariri M, Darvishi L, Mofid MR. Anti-oxidative and anti-inflammatory effects of ginger in health and physical activity: review of current evidence. International journal of preventive medicine. 2013;4(Suppl 1):36.
18. Zhou K, Raffoul JJ. Potential anticancer properties of grape antioxidants. Journal of oncology. 2012;2012.
19. Grases F, Prieto RM, Fernández-Cabot RA, Costa-Bauzá A, Sánchez AM, Prodanov M. Effect of consuming a grape seed supplement with abundant phenolic compounds on the oxidative status of healthy human volunteers. Nutrition journal. 2015;14(1):94.
20. Rahmani AH. Active ingredients of ginger as potential candidates in the prevention and treatment of diseases via modulation of biological activities. International journal of physiology, pathophysiology and pharmacology. 2014;6(2):125.
21. Samarth RM, Samarth M, Matsumoto Y. Medicinally important aromatic plants with radioprotective activity. Future science OA. 2017;3(4):FSO247.
22. Surh YJ. Molecular mechanisms of chemopreventive effects of selected dietary and medicinal phenolic substances. Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis. 1999;428(1-2):305-27.
23. VÁZquez-Armenta FJ, Bernal-Mercado AT, Pacheco-Ordaz R, Gonzalez-Aguilar GA, Ayala-Zavala JF. Winery and grape juice extraction by-products. InPlant Food By-Products: Apple Academic Press. 2018;157-81.
24. Liu M, Xia X, Chou G, Liu D, Zuberi A, Ye J, et al. Variations in the Contents of Gingerols and Chromatographic Fingerprints of Ginger Root Extracts Prepared by Different Preparation Methods. Journal of AOAC International. 2014;97(1):50-7.
25. De Freitas RB, Augusti PR, De Andrade ER, Rother FC, Rovani BT, Quatrin A, et al. Black grape juice protects spleen from lipid oxidation induced by gamma radiation in rats. Journal of food biochemistry. 2014 Feb;38(1):119-27.
26. Xu D, Peng M, Zhang Z, Dong G, Zhang Y, Yu H. Study of damage to red blood cells exposed to different doses of γ-ray irradiation. Blood Transfusion. 2012;10(3):321.
27. Sancheti G, Goyal PK. Prevention of radiation induced hematological alterations by medicinal plant Rosmarinus officinalis, in mice. African Journal of Traditional, Complementary and Alternative Medicines. 2007;4(2):165-72.
28. Agarwal M, Purohit RK, Chakrawarti A, Basu A, Bhartiya KM. Protective efficacy of Aloe vera against radiation and cadmium induced haematological changes in the Swiss albino mice. Advanced Biotech. 2011;10(10):44-7.
29. Sharma R, Purohit RK, Sharma S, Rao R, Purohit RK. Protective role of liv. 52 against radiation and cadmium induced haematological changes in the Swiss albino mice. InProceedings of the international conference on emerging frontiers and challenges in radiation biology: abstracts 2012.
30. El-Deeb AE, IM AE. The curative effect of some antioxidants on γ-irradiated rats. J. Egypt. Soc. Toxicol. 2006;35:79-89.
31. Abojassim AA, Jaffat HS, Hassan AB. Effects of gamma radiation on some hematological parameters in female rats. Theoretical & Applied Science. 2015(5):101-9.
32. Bloom ML, Simon-Stoos KL. The hemoglobin-deficit mouse: analysis of phenotype and hematopoiesis in the transplant model. Blood. 1997;90(5):2062-7.
33. Abdelhalim MA, Al-Ayed MS, Moussa SA, Al-Shehri AE. The effects of gamma-radiation on red blood cell corpuscles and dimensional properties in rats. Pak. J. Pharm. Sci. 20151;28(5):1819-22.

34. Daga SS, Jain VK, Goyal PK. Radioresponse to leucocytes in peripheral blood mice against gamma irradiation and their protection by Liv. 52. Probe. 1995;34(3):222.
35. Gupta U, Agrawal NK. Prevention of radiation induced hematological alterations in mice by Alstonia scholaris extract. International Journal of Pharmaceutical Sciences and Research. 2013 ;4(4):1453.
36. Mehrotra S, Pecaut MJ, Gridley DS. Effects of minocycline on hematopoietic recovery after whole-body irradiation. in vivo. 2013;27(1):11-28.
37. El-Desouky W, Hanafi A, Abbas MM. Radioprotective effect of green tea and grape seed extracts mixture on gamma irradiation induced immune suppression in male albino rats. International journal of radiation biology. 2017;93(4):433-9.
38. Singha I, Das SK. Grape extract protects against ionizing radiation-induced membrane alterations in Red Blood Cells. 2016.
39. Jagetia GC. Radioprotective potential of plants and herbs against the effects of ionizing radiation. Journal of clinical biochemistry and nutrition. 2007;40(2):74-81.
40. Painuli S, Kumar N. Prospects in the development of natural radioprotective therapeutics with anti-cancer properties from the plants of Uttarakhand region of India. Journal of Ayurveda and integrative medicine. 2016;7(1):62-8.
41. Kiang JG, Fukumoto R, Gorbunov NV. Lipid peroxidation after ionizing irradiation leads to apoptosis and autophagy. InLipid peroxidation. 2012.
42. Nakajima T, Ninomiya Y, Nenoi M. Radiation-Induced Reactions in the Liver−Modulation of Radiation Effects by Lifestyle-Related Factors−. International journal of molecular sciences. 2018;19(12):3855.