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Evaluation of Protective Efficacy of Hydro Alcoholic Extract and Methanol Fraction of *Tamarindus Indica Pod* against Radiation Induced Biochemical and Chromosomal Changes in Swiss Albino Mice

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Abstract: **The objective of the study was to investigate and compare the radioprotective effect of hydroalcoholic extract (HAE) and methanol fraction (MF) of *Tamarindus indica* pod in mice against electron beam radiation. Animals were treated with 100 mg/Kg body weight of HAE and MF of *T. indica* for 15 consecutive days before exposing to 6Gy (sublethal dose) whole body EBR. The irradiation of animals resulted in an elevation in lipid peroxidation and reduction in glutathione, total antioxidants and antioxidant enzymes such as superoxide dismutase, glutathione peroxidase and catalase activities. Irradiated group had shown micronucleus in the bone marrow cells. Treatment of mice with HAE and MF before irradiation, reduced the symptoms of radiation sickness when compared with the irradiated group. Pretreated mice showed a significant reduction in lipid peroxidation followed by elevation in reduced glutathione, total antioxidants and antioxidant enzymes such as superoxide dismutase, glutathione peroxidase and catalase. It also showed a reduction in the micronucleus formation in bone marrow cells. The findings of our study indicate the protective efficacy of HAE and MF on radiation induced biochemical and chromosomal changes in mice may be due to its free radical scavenging and increased antioxidant levels. It also reveals MF, a better protective agent than that of HAE of *T. indica.***

Keywords: Tamarindus indica, **hydroalcoholic extract (HAE), Methanol fraction (MF), Electron beam Radiation (EBR), Radiation sickness.**

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1. INTRODUCTION

Radiotherapy is the most common modality for treating human cancers which has been used since many decades to eradicate cancer. Clinical radiation oncology uses electromagnetic radiation and particulate radiation mostly electrons and to a lesser extent neutrons and protons[1]. Electron beam radiation is a form of ionizing radiation that is generally characterized by its low penetration and high dosage rates. It is a concentrated, highly charged stream of electrons, generated by particle accelerators which are capable of producing beams that are either pulsed or continuous. This high energy electrons are used for various purposes in the field of biology, it is also used in the radiation therapy[2]. The rapid technological advancements of the modern world have increased human exposure to ionizing radiation. In recent years, due to the wide range of applications of ionizing radiation in human life, especially for its effects in cancer treatment, food preservation, agriculture, industrial and energy production, there is an urgent need to develop efficient non-toxic radiation protectants.

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Over the past 50 years, research in the development of radioprotectors worldwide has focused on screening a plethora of chemical and biological compounds [3-5]. The use of certain chemical agents may reduce the ill effects of radiation but the major drawback of chemical compounds has been their high toxicity at the optimum protection dose[6]. Considering, the drawbacks associated with the currently available radioprotective agents several novel approaches exploring & experimenting with the plant kingdom have been tried.

Natural & herbal sources are nontoxic with proven therapeutic benefits & have been utilized, since ancient times for curing various diseases & disorders[7]. A number of medicinal plants evaluated for their radioprotective efficacy have shown protective effects against the damaging effects of ionizing radiation[8]. The traditional Indian system of medicine the *Ayurveda,* gives a detailed account of several disease and their treatments. The majority of the drug and drug formulations used in *Ayurveda* are principally derived from herbs and plants. Plant extracts eliciting radioprotective efficacy contain a plethora of compounds including antioxidants, immunostimulants, cell proliferation stimulators, anti-inflammatory & anti-microbial agents, some of which may act in isolation as well as in combination with other constituents from the same plant. Most studies using phytochemicals have focused on evaluation of radioprotective efficacy of whole extracts or polyherbal formulations & in some cases fractionated extracts & isolated constituents, for their ability to protect against radiation-induced chromosomal aberrations & micronuclei formation [9].

Tamarindus indica L. belongs to the family Fabaceae, is an important food in the tropics. It is a multipurpose tree of which every part finds atleast some use either nutritional or medicinal[10]. It is commonly known as Indian date. *T. indica,* is widely used in traditional medicine in Africa for the treatment of gastrointestinal disorders, gonococci, fever, jaundice and dysentery[11]. It has numerous chemical values and is rich in phytochemicals, and hence the plant is reported to possess antimicrobial [12-17], antivenomic [16]., antioxidant [18-21], antimalarial [22-23], hepatoprotective [24-25], antiasthmatic[26], laxative[27], antidiabetic [28], and anti-hyperlipidemic activity[29]. Several reports indicate that extracts of Tamarind exhibited antimicrobial, anti–inflammatory, anti-diabetic and other pharmacological activities. No published studies are available till date describing the protective role of HAE and MF of *T. indica* pod (whole fruit) against the damaging effects of electron beam radiation on antioxidants and antioxidative enzymes. With this background, the present investigation was carried out to evaluate and compare the protective effect of tamarind pod against radiation induced damage at biochemical and chromosomal alterations in swiss albino mice.

2. MATERIALS AND METHODS

Tamarindus indica fruits were collected in the month of January 2012 from Horticulture Dept garden, Shimoga. The plant material is identified as *T.indica* Linn by Dr. K.G. Bhat, Dept of Botany, Poorna prajna College, Udupi, Karnataka after examination of the specimen.

EXTRACTION: Tamarind pods were thoroughly washed under running tap water and air dried. The fruit pulp was separated, chopped and dried in hot air oven at 40°C for 2 days. The shells, fibres and seeds were separated, dried as above and powdered. Soft part of the pod (Pulp) was macerated with hydroalcohol (70% alcohol) and extracted using shaker incubator for 48 hours. Hard part (Shell, fibers and Seeds) were powdered and extracted using soxhlet apparatus (hot percolation) with hydroalcohol. Later the macerate was filtered through muslin cloth. Both filtrate and extract obtained by maceration and soxhlet extraction respectively were pooled and evaporated using Rotary flash evaporator.

FRACTIONATION: Fractionation was carried out by suspending crude and the consecutive fractions in water separately and partitioning with different organic solvents (hexane, chloroform, ethyl acetate and methanol) in the order of increasing polarity by using separating funnel according to standard procedures[30]. All extracts were stored in an airtight container at 4°C till further use.

EXPERIMENTAL ANIMALS:

Animal care and handling was carried out according to the guidelines set by WHO (World Health Organization; Geneva, Switzerland). This study has been approved by institutional animal ethical committee. Male swiss albino mice *(Mus musculus*) of 6-8 weeks old, weighing 25±5 g, were procured from the Institutional Animal House, K.S Hegde Medical Academy, Nitte University, Mangalore. The animals were maintained under controlled conditions of temperature and light (Light: dark, 10 hrs: 14 hrs.) and were housed in a polypropylene cage containing sterile paddy husk (procured

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locally) as bedding throughout the experiment. They were provided with standard mouse feed and water *ad libitum*. Experimental protocol was approved by the Institutional animal ethical committee.

RADIATION PROCEDURE:

Irradiation was carried out at the Microtron Center, Mangalore University, Mangalore, Karnataka, India. Variable energy accelerator microtron was used for giving radiation. The unanaesthetised animals were restrained in well ventilated Perspex boxes and exposed to sublethal dose (6Gy) of whole body irradiation.

PREPARATION OF DRUG AND MODE OF ADMINISTRATION:

The required amount of HAE and MF of *T.indica* was dissolved in millipore water and administered orally once Daily for 15 consecutive days. The animals were divided into the following groups:

Group I: Control - Animals in this group were administered with distilled water.

Group II: Radiation Control – distilled water + Irradiation.

Group III: Treatment group (HAE+IR) were administered with HAE via oral gavage for 15 consecutive days before irradiation.

Group IV: Treatment group (MF+IR) were administered with MF via oral gavage for 15 consecutive days before irradiation.

BIOCHEMICAL ESTIMATIONS:

The animals treated according to the experimental protocol were euthanized on day 16 and liver tissue was excised. 10% liver homogenate (1g liver in 10mL of Phosphate buffer saline-PBS) was prepared using a glass homogenizer. It was centrifuged at 10,000rpm for 15 minutes at 4°C in a cold centrifuge. The supernatant was collected and used for biochemical estimations of antioxidants (Total Antioxidant Capacity, Reduced Glutathione), lipid peroxidation and antioxidative enzymes (Superoxide Dismutase, Catalase, Glutathione Peroxidase) according to standard protocols using UV-Visible Spectrophotometer.

TOTAL ANTIOXIDANT CAPACITY:

Total antioxidant capacity of tissue homogenate was determined by the phosphomolybdenum method as described by Prierto et al. This assay is based on the conversion of Molybdenum(Mo VI) by reducing agents like antioxidants to molybdenum (Mo V), which further reacts with phosphate in under acidic pH resulting in the formation of a bluish-green coloured complex. The homogenate was treated with 5% TCA and centrifuged to precipitate out the proteins. 100µL of the clear supernatant was transferred into a clean test tube and 1mL of Total Antioxidant Capacity (TAC) reagent containing 0.6M H2SO4, 28mM NaH2PO4 and 4mM ammonium heptamolybdate, was added to it and the mixture was incubated in water bath at 95°C for 90 minutes. A blank was also maintained simultaneously by substituting distilled water instead of sample in the reaction mixture. Following the incubation, the reaction mixture was cooled and the optical density of the greenish blue colour formed was read at 695nm against blank in a UV-Visible Spectrophotometer and TAC was calculated from the standard graph [31].

REDUCED GLUTATHIONE (GSH):

The Glutathione content in tissue homogenate was measured spectrophotometrically according to the method of Beutler et al. This method is based on the development of a relatively stable yellow color, when 5, 5'- dithiobis 2-nitro benzoic acid (DTNB) is added to sulphydryl compounds including glutathione. The diluted samples were treated with 1.5mL of precipitating solution (glacial m-phosphoric acid, EDTA and NaCl per 100mL of distilled water), and kept for 10 minutes for the completion of protein precipitation. The solutions were then filtered through a whatmann No.1 filter paper. 500µL of the filtrate was taken and to this 2mL of phosphate solution (0.3M Na2HPO4) and 250µL of DTNB solution was added. Simultaneously a blank was maintained containing 200µL of distilled water, 300µL of precipitating solution, 2mL of phosphate solution and 250µL of DTNB. The intensity of the yellow color formed was read immediately (within ten minutes) at 412nm against the blank in an UV-Visible Double Beam Spectrophotometer and the GSH concentration was calculated from the standard graph [32].

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LIPID PEROXIDATION:

Lipid peroxidation was measured by the method of Buege and Aust [33].Malondialdehyde (MDA), formed as an end product served as an index of oxidative stress. To the liver homogenate samples, 1mL of TCA-TBA-HCl reagent containing 15% Trichloroacetic acid (TCA) and 0.375% Thiobarbitutric acid (TBA) in 0.25N Hydrochloric acid was added. The samples were kept in boiling water bath for 15 minutes. The reaction mixture was cooled and centrifuged. The supernatant was taken and the optical density of the pink colour formed was read at 535nm in a spectrophotometer. The concentration of malondialdehyde in the sample was obtained by plotting the obtained absorbance against the standard graph. The optical density of the pink colour formed is directly proportional to the concentration of malondialdehyde in the given sample.

GLUTATHIONE PEROXIDASE ACTIVITY:

Glutathione peroxidase activity was measured as described by Rotruck JT et al. The tissue homogenate was mixed with 4mM reduced glutathione. In the presence of GPx, the reduced glutathione converts into oxidized glutathione at 37°C. The leftover reduced glutathione reacts with 5,5-dithiobis 2- nitrobenzoic acid (DTNB). The coloured compound formed absorbs maximally at 412nm[34].

SOD ACTIVITY:

The estimation of superoxide dismutase enzyme was carried out by Beauchamp and Fridovich[35] method. The substrate used for the assay consists of nitro blue tetrazolium chloride (NBT) which reacts with superoxide anions produced upon illumination of riboflavin in the presence of methionine as an electron donor, to produce formazan which is a blue coloured complex. The SOD present in the sample acts on the superoxide anions produced by riboflavin and thereby reduce the net superoxide anions in the substrate leading to decreased production of formazan manifested by decreased intensity of the blue color formed. The decrease in the formation of formazan is directly proportional to the amount of SOD in the sample.

The supernatant of liver homogenate was used for the assay. For each sample analyzed a corresponding control was maintained. A common standard and blank was kept for each set of illumination.

Test: 2.5ml Methionine, 0.3 ml Riboflavin, 0.1mL NBT, 100µl Tissue homogenate.

Control: 2.5ml Methionine, 0.3ml Riboflavin, 0.1ml phosphate buffer 0.05 M (pH 7.8), 100 µl Tissue homogenate.

Standard: 2.5ml Methionine, 0.3 ml Riboflavin, 0.1ml NBT, 0.1 ml phosphate buffer 0.05 M (pH 7.8).

Blank: 2.5ml Methionine, 0.3ml Riboflavin, 0.2ml phosphate buffer 0.05 M (pH 7.8).

The test tubes labelled as Test, Standard and Control were subjected to illumination for 10 minutes in an illumination chamber lined with aluminium foil, and fitted with a 15W fluorescent lamp. Following illumination, immediately the optical density of all the reaction mixtures was read at 560nm. Units of enzyme present in the sample were calculated and expressed as U/mg protein.

CATALASE ACTIVITY:

Catalase activity was measured in terms of decomposition of Hydrogen peroxide (H_2O_2) into water molecule, which was measured as decreased absorbance at 240nm. The diluted tissue homogenate was added to 2ml of 30mM hydrogen peroxide. 2ml of distilled water was maintained as blank and 2ml of hydrogen peroxide served as reagent blank. The decrease in absorbance was followed for 2 minutes at 15 seconds interval at 240nm. The catalase activity was expressed as U/ mg protein i.e., 1µmole of H_2O_2 converted into H_2O in 1 minute [36].

TISSUE PROTEIN CONTENT:

All the samples were analysed for protein content by Biuret method using commercially available kits [37].

MICRONUCLEUS TEST:

The mouse bone marrow micronucleus test was carried out according to the method described by Schmidt[38] for the evaluation of chromosomal damage in experimental animals. The bone marrow from femur was flushed with 5% BSA in

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the form of a suspension into an embryo cup. The cells were dispersed by gentle pipetting and collected by centrifuge at 2000 rpm for 5 min at 4°C. The pellet was resuspended in a drop of BSA and bone marrow smears were prepared. After air drying the smears were stained with May-Grunwald/Giemsa. Micronucleated polychromatic erythrocytes (MnPCEs) and micronucleated nonchromatic erythrocytes (MnNCEs) were observed under microscope. The percentage of micronucleated cells were calculated.

STATISTICAL ANALYSIS:

All Values were expressed as Mean±SD. Comparison between control and treated groups were performed by analysis of Variance (ANOVA) with Tukey HSD. In all these, test criterion for statistical significance was $P<0.05$.

3. RESULTS

Table 1 represents the changes in tissue MDA levels, TAC levels and GSH content in control, irradiated and Pre-treated groups (HAE and MF). The results of lipid peroxidation demonstrated that the changes in MDA levels in irradiated animals were appreciably counteracted by HAE and MF administration. Irradiated animals exhibited significant increase ($P<0.05$) in MDA levels compared to control, whereas the pre-treated groups showed significant decrease ($P<0.05$) in the liver MDA levels. However treatment with MF exhibited much pronounced effect than HAE.

TABLE 1: EFFECT OF TREATMENT WITH TAMARINDUS INDICA EXTRACT & ITS METHANOL FRACTION IN MICE EXPOSED TO ELECTRON BEAM RADIATION

Parameters	Group I	Group II	Group III	Group IV
MDA(µM/g tissue)	1.99±1.01	4.58±0.23	1.58±0.56*	3.04±0.14*
TAC(mg/g tissue)	4.92±0.06	4.06±0.16	4.78±0.13*	5.14±.15*
GSH(mg/g tissue)	1.25±0.04	0.57± 0.09	0.89 ± 0.05*	1.15± 0.03*
GPX(GSH consumed/min/g tissue)	87.54±3.7	68.78±4.15	82.94±2.31*	140.51±6.1*
Catalase (Units/mg protein)	19.89±0.9	10.71±1.55	14.90±1.80*	18.75±2.35*
SOD (units/mg protein)	38.2±0.18	15.4±0.15	20.6±0.8*	29.4±0.42*

*P<0.05. MDA=Melondialdehyde, TAC=Total antioxidant capacity, GSH=Reduced glutathione, GPx=Glutathione peroxidase. Group I= Normal control, Group II =Radiation control, Group III=Treatment Group(HAE), Group IV=Treatment Group(MF).

Changes in the TAC levels showed that radiation significantly decreased ($P<0.05$) the total antioxidant capacity in liver tissue when compared to control group. Administration of HAE and MF to the animals before irradiation caused significant elevation ($P<0.05$) in liver TAC levels when compared to irradiated group.

The study revealed that GSH content decreased significantly in the irradiated group ($P<0.05$) compared to control. The pre treatment groups exhibited significant increase($P<0.05$) in GSH levels when compared to irradiated group. However MF was more effective than HAE.

GPX, Catalase and SOD activities decreased significantly ($P<0.05$) in irradiated group compared to normal controls. The pre treated groups exhibited significant increase ($P<0.05$) in all the antioxidative enzyme activities determined. However

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treatment with MF showed much pronounced effect on the activity of antioxidative enzymes compared to HAE administration.

Table 2 represents increase in micronucleus formation in irradiated group when compared to the control group, whereas administration of HAE and MF before irradiation has brought down micronucleus formation when compared to irradiated group. However MF showd better efficiency in reducing micronucleus formation when compared to HAE .

TABLE 2: EFFECT OF TREATMENT WITH TAMARINDUS INDICA EXTRACT & ITS METHANOL FRACTION ON PROTECTION AGAINST MICRONUCLEUS FORMATION INDUCED BY EBR .

	Group I	Group II	Group III	Group IV
Number of cells counted per 100 cells				
PCE	48.12±2.82	28.8±2.12	37.9±1.48	42.5±2.02
MnPCE	0.04	14±1.41	6±1.41	2 ±0.02
NCE	51.82±1.24	50.2±0.95	54.1±1.6	54.3 ±1.21
MnNCE	0.02	7±1.41	4±1.41	1.2±0.70

PCE=Polychromatic erythrocytes, NCE=Nonchromatic erythrocytes, MnPCE=Micronucleated polychromatic erythrocytes, MnNCE=Micronucleated normochromatic erythrocytes

4. DISCUSSION

Many plants are known to have beneficial therapeutic effects as noted in the traditional Indian system of medicine, Ayurveda. However, they have received little attention for their radioprotective as well as antioxidant activities. Medicinal plants can protect against harmful effects of ionizing radiation. Plants are one of the most important sources of medicine. Today the large number of drugs in use are derived from plants[39].Natural plant extracts or pure compounds are safe ingredients, which do not have any toxic effects. In actively metabolizing cells, there was considerable amount of water apart from the target macro-molecules. EB generates ROS as a result of radiolysis of water. ROS can induce oxidative damage to vital cellular molecules and structures including DNA, lipids, proteins and membranes [40,41].Thus, radiation-induced damage might result in adverse health effects within hours to weeks or delayed effects observable many months after exposure [42].Recently, emerging evidence suggests that oxidative stress is possibly involved in the pathology of some diseases and other inborn errors of lipid and protein metabolism [43].The present study demonstrates that, administration of *T. indica* prior to EB protects against the oxidative stress and tissue damage produced by acute sub lethal doses of EB. The major forms of cellular damage induced by radiation are DNA damage, lipid peroxidation and protein oxidation.

It was hypothesized that during oxidative stress there was tissue damage which on successful antioxidant treatment should delay or prevent the onset of such damage [44].Lipid peroxidation (LPO) is a hallmark of oxidative stress which disrupts the structural integrity of cell membrane and can lead to formation of aldehydes which in turn lead to lipid, protein and DNA damage [45].In the present study, exposure to EB resulted in a significant increase of malondialdehyde (MDA) levels. The increase in lipid peroxidation was shown to be the principal damage induced by radiation in biological membranes[46]. Elevated LPO by radiation exposure could be attributed to formation of free radicals and involvement of free radical induced oxidative cell damage. Thus, increased LPO is suggestive of progressive increase in membrane permeability, disruption of structural and functional integrity of cell organelles.

T. indica extract has shown both *in vitro* and *in vivo* antioxidant property[47]. This shows that extract and fraction combat with oxidative stress caused due to EBR by decreasing the level of MDA and elevating the TAC level and GSH content as well as anti-oxidant enzymes like SOD, Catalase and Glutathione peroxidase either by arresting free radicals formation or by scavenging the free radicals generated.

Depletion of intracellular GSH level has been implicated as one of the causes of radiation-induced damage, while increased levels of this are responsible for the radioprotective action. MF of *T.indica* presupplementation has shown best restoration of GSH when compared to HAE and to the concurrent irradiation controls. This inhibits the radiation-induced lipid peroxidation, thereby protecting against radiation‑induced damage. Ionizing radiation induces lipid peroxidation, which causes DNA damage and cell death[48-49]. Some of the plants *Embelia ribes*[50], *Piper longum, Zinger officinale, Santalum album*[51], *Ocimum sanctum* have been reported to increase GSH. While the other plants *Asparagus racemosus, Glycyrrhiza glabra, Phyllanthus embelica, Boerrhaavia diffusa, Ocimum sanctum, Eclipta alba* have been found to possess *in vitro* antioxidant properties[52].

Bone marrow cells are very sensitive to radiation, which causes chromosomal aberrations and increases the micronucleus formation.The administration of HAE & MF in the present study partially prevented the increase in the micronucleus rate in the bone marrow of these mice, indicating that HAE and MF are anti-mutagenic. This finding is supported by the observation that pre-treatments with LC & LA to γ- irradiation was found to decrease the Mn-PCEs compared with irradiated group[53].

In the present study, the biochemical analysis carried out in the liver homogenate and Chromosomal changes in the bone marrow of Swiss albino mice exposed to sublethal dose of Electron beam radiation ascertained its radioprotective potential.

5. CONCLUSION

The present study demonstrates that, administration of tamarindus indica pod extract & its methanol fraction prior to EB radiation protects against the oxidative stress and tissue damage produced by acute sublethal doses of EB. The major forms of cellular damage induced by radiation are lipid peroxidation, protein oxidation and DNA damage.

Pretreatment with drugs have reduced radiation-induced stress by protecting against the radiation-induced damage at biochemical and chromosomal level. Free radical scavenging, elevation in antioxidant status, GSH levels & reduction in lipid peroxidation appear to be important in providing radioprotection.

Hence our findings provide the evidence that, Tamarindus indica pod extract and its MF are good radioprotective agents particularly MF being the more potent one and hence can be used in the treatment of cancers to prevent damage to the normal tissues.

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REFERENCES

[1] Coia LR & Moyland DJ. Introduction to Clinical Radiation Oncology. Medical Physics Publishing Madison, WI. 1998.

[2] Loevinger R, Karzmark CJ, Weiss Bluth M. Radiation therapy with high Energy electrons. Part I. Physical considerations-10 to 60 Mev, Radiology. 1961;77: 906-27.

[3] Maisin JR. Chemical radioprotection, past, present and future prospects. International Journal of Radiation Biology. 1998; 73: 443–50.

[4] Weiss JF, Kumar KS, Walden TL, Neta R, Landauer MR, Clark EP. Advances in radioprotection through the use of combined agent regimens. International Journal of Radiation Biology. 1990; 57: 709-22.

[5] Weiss JF and Landauer MR. Protection against ionizing radiation by antioxidant nutrients and phytochemicals. Toxicology. 2003; 189: 1–20.

[6] Sweeney TR. A survey of compounds from the antiradiation drug development program of the U.S. Army Medical Research and development command. Government Printing office, Washington, DC Publication. 1979; 308–18.

[7] Fabricant DS, Farnsworth NR. The value of plants used in traditional medicine for drug discovery. *Environmental Health Perspectives*. 2001; 109(1): 69-75.

[8] Jagetia GC, Baliga MS. Influence of the leaf extract of Mentha arvensis Linn.(mint) on the survival of mice exposed to different doses of gamma radiation. Strahlenther Onkol. 2002; 178: 91-8.

[9] Hosseinimehr SJ. Trends in the development of radioprotective agents. Drug Discovery Today. 2007; 12(19-20): 794-805.

[10] Kumar C. S., Bhattacharya S., 2008. Tamarind Seed: Properties, Processing and Utilization.Critical Reviews in Food Science and Nutrition, 48, 1-20.

[11] Souza A., Aka K.J., 2007. Spasmogenic effect of the Aqueous Extract of *Tamarindus Indica L.* (Caesalpiniaceae) on the Contractile Activity of Guinea-Pig Taenia Coli. African journal of traditional, complementary and alternative medicines, 4(3) 261–266.

[12] Muthu SE, Nandakumar S, Roa UA. The effect of methanolic extract of Tamarindus indica on the growth of clinical isolates of Burkholderia pseudomallei. Indian journal of medical research. 2005; 122: 525-8.

[13] Vaghasiya Y, Chanda S. Screening of some traditionally used Indian plants for antibacterial activity against Klebsiella pneumonia. Journal of Herbal Medicine and Toxicology. 2009; 3: 161-4.

[14] Doughari JH. Antimicrobial activity of Tamarindus indica. Tropical Journal of Pharmaceutical Research. 2006; 5(2): 597–603.

[15] Warda S, Gadir A, Mohamed F, Bakhiet AO. Antibacterial activity of Tamarindus indica fruit and Piper nigrum. Research Journal of Microbiology. 2007; 2: 824-30.

[16] Al-Fatimi M, Wurster M, Schroder G, Lindequist U. Antioxidant, antimicrobial and cytotoxic activities of selected medicinal plants from Yemen. Journal of Ethnopharmacology. 2007; 111(3): 657-66.

[17] Melendez PA, Capriles VA. Antibacterial properties of tropical plant fruit from Puerto Rico. Phytomedicine. 2006; 13(4): 272-6.

[18] Siddhuraju P. Antioxidant activity of polyphenolic compounds extracted from defatted raw and dry heated Tamarindus indica seed coat. LWT - Food Science and Technology. 2007; 40: 982-90.

[19] Martinello F, Soaresh SM, Franco JJ. Hypolipemic and antioxidant activities from Tamarindus indica pulp fruit extract in hypercholesterolemic hamsters. Food and Chemical Toxicology. 2006; 44 (6): 810-8.

[20] Vyas N, Gavatia NP, Gupta B, Tailing M. Antioxidant potential of Tamarindus indica seed coat. Journal of Pharmacy Research. 2009; 2: 1705-6.

[21] Osawa T, Tsuda T, Watanabe M, Ohshima K, Yamamoto A. Antioxidative components isolated from the seeds of Tamarind (Tamarindus indica L). Journal of Agricultural and Food Chemistry. 1994; 42: 2671-4.

[22] Norscia I, Borgognini-Tarli SM. Ethnobotanical reputation of plant species from two forests of Madagascar: A preliminary investigation. South African Journal of Botany. 2006; 72: 656-60.

[23] Asase A, Oteng-Yeboah AA, Odamtten GT, Simmonds MSJ. Ethnobotanical study of some Ghanaian anti-malarial plants. Journal of Ethnopharmacology. 2005; 99:273-9.

[24] Tayade PM, Ghaisas MM, Jagtap SA, Dongre SH. Anti-asthmatic activity of Methanolic extract of leaves of Tamarindus Indica Linn. Journal of Pharmacy Research. 2009; 2: 944-7.

[25] Pimple BP, Kadam PV, Badgujar NS, Bafna AR, Patil MJ. Protective effect of Tamarindus indica linn against paracetamol-induced hepatotoxicity in rats. Indian Journal of Pharmaceutical sciences. 2007; 69(6): 827-31.

[26] Prashant M Tayade, Borde SN, Jagtap SA, Patil VP, Vaishnav G, Girbane YR et al. Effects of Tamarindus indica Linn. Against isolated goat tracheal and guinea pig ilium preparation. Pharmacie Globale (IJCP- International Journal of Comprehensive Pharmacy). 2010; 1(2).

[27] Bhat RB, Eterjere EO, Oladipo VT. Ethnobotanical studies from Central Nigeria. Economic Botany. 1990; 44: 382-90.

[28] Maiti R, Jana D, Das UK, Ghosh D. Antidiabetic effect of aqueous extract of seed of Tamarindus indica in streptozototocin-induced diabetic rats. Journal of thnopharmacology. 2004; 92 (1): 85-91.

[29] Maiti R, Das UK, Ghosh D. Attenuation of hyperglycemia and hyperlipidemia in streptozototocin-induced diabetic rats by aqueous extract of seeds of Tamarindus indica Biological & Pharmaceutical Bulletin. 2005; 28 (7): 1172-6.

[30] Yamin B, Sobiya N, Fayyaz MC and Muhammad Z. Antibacterial activity of some selected Medicinal plants of Pakistan. BMC complementary and alternative medicine.2011; 2: 11-52.

[31] Prieto P, Pineda M, Aguilar M. Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: Specific application to the determination of vitamin E. Analytical Biochemistry.1999; 269:337- 41.

[32] Beutler E, Duron O, Kelly BM. Improved method for the determination of blood glutathione. Journal of Laboratory and Clinical Medicine. 1963; 61: 882-8.

[33] Buege JA, Aust SD. Microsomal lipid peroxidation. Methods in Enzymology. 1978; 52: 302-10.

[34] Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG, Hoekstra WG. Selenium: Biochemical role as a component of glutathione peroxidase. Science 1973; 179: 588-90.

[35] Beaurchamp C and Fridovich I. Superoxide dismutase improved assay applicable to acrylamide gels. Analytical biochemistry.1971; 44: 276-87.

[36] Aebi H. Catalase in vitro. Methods in Enzymology. 1984; 105: 121-6.

[37] Thomas L. Clinical Laboratory TH-Books ,Diagnostics.1st ed. Frankfurt:Verlags- gesellschaft. 1998; p.644-7.

[38] Schmidt W. The Micronucleus test. Mutation Research. 1975; 31: 9-15.

[39] Sumit KT, Mandeep KA, Ritu T, Umesh KS. *Ficus racemosa linn*: A boon for ailments of human kind. American Journal of Pharmtech Research. 2012; 2(3): 389-400.

[40] Cadet J, Bellon S, Douki T, Frelon S, Gasparutto D, Muller E, et al., Radiation-induced DNA damage: formation, measurement, and biochemical features. Journal of Environmental Pathology, Toxicology and Oncology. 2004; 23(1): 33–43.

[41] Esterbauer H, Estimation of peroxidative damage: a critical review. Pathologie Biologie.1996; 44(1): 25–28.

[42] Vijayalaxmi, Reiter RJ, Tan DX, Herman TS, Thomas CR. Melatonin as a radioprotective agent: a review. International Journal of Radiation Oncology • Biology • Physics. 2004; 59(3): 639-53.

[43] Onody A, Csonka C, Giricz Z, Ferdinandy P. Hyperlipidemia induced by a cholesterol-rich diet leads to enhanced peroxynitrite formation in rat hearts. Cardiovascular Research. 2003; 58(3), 663–70.

[44] Halliwell B, Whiteman M. Measuring reactive species and oxidative damage in vivo and in cellculture: how should you do it and what do the results mean *g* . British Journal of Pharmacology. 2004; 142(2): 231–55.

[45] Mates JM, Perez-Gomez C, Castro NI. Antioxidant enzyme in human diseases. Clinical Biochemistry. 1999; 32(8): 595-603.

[46] Sun Y. Free radicals, antioxidant enzymes and carcinogenesis. Free Radical Biology and Medicine. 1990; 8(6): 583-99.

[47] Sandesh P, Velu V, Singh R P. Antioxidant activities of tamarind (Tamarindus Indica) seed coat extracts using in vitro and in vivo models. Journal of Food Science and Technology. 2014; 51(9): 1965-73.

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[48] Raleigh JM, Shum FY. Radioprotector in model lipid membranes by hydroxyl radical scavengers: Supplementary role of α‑tocopherol in scavenging secondary peroxy radicals. In: Nygaard OF, Simic MG, editors. Radioprotectors and Anticarcinogens. New York: Academic Press. 1983: 87‑102.

[49] Leyko W, Bartosz G. Membrane effects of ionizing radiation and hyperthermia. International journal of radiation biology and related studies in physics, chemistry, and medicine.1986; 49(5): 743‑70.

[50] Chitra M, Shyamala Devi CS. Protective action of embelin against lipid peroxidation on tumor bearing mice. Fitoterapia 1994; 65: 317‑21.

[51] Banerjee S, Evacuee A, Rao AR. Modulator influence of sandalwood oil on mouse hepatic glutathione s‑transferees activity and acid soluble sulphydryl level. Cancer Letters. 1993; 68: 105‑9.

[52] Joy KL, Kuttan R. Antioxidant activity of selected plant extracts. Amala Res Bull. 1995; 15: 68‑71.

[53] Sally SA, Aziza MH, Nermin E et al. Radioprotective activity of L- Carnitine and α -Lipoic acid against whole body γ-irradiation in rats. Journal of American Science.2010; 6(12): 865-887.