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Amelioration of radiation-induced cellular alterations in rats administered with solvent fractions of methanol leaf extracts of *Adansonia digitata* and *Corchorus olitorius*

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ABSTRACT: Exposure of rats to radiation results in an increase of free radical level from subtoxic (24 µg/mol) to a toxic concentration (120 µg/mol) in the system of rats. Free radical level above 120 µg/mol, leads to dysregulated Nf-kB and Nrf-2, thus exacerbating oxidative stress and cellular alterations in rats. This study investigated the effects of solvents (n-hexane, ethylacetate and n-butanol) fractions of *Adansonia digitata* and *Corchorus olitorius* leaves in radiation-induced Nf-kB and Nrf-2 dysregulation in Cellular System of Rats. A total of 48 rats (198 ± 5.00g) were used in this study and were distributed into 8 groups of 6 each. Group I were fed with rat chow and distilled water only, all other groups were irradiated, such that groups II, III, IV, V, VI, VII, VIII and IX were administered distilled water, n-hexane fractions of *A. digitata* and *C. olitorius*, ethylacetate fractions of *A. digitata* and *C. olitorius*, n-butanol fractions of *A. digitata* and *C. olitorius* and Vitamin-C at 1000 mg/kg body weight respectively. Secondary metabolites screening of *A. digitata* and *C. olitorius* revealed the presence of flavonoids, polyphenol, alkaloids, tannins and saponins. Irradiation significantly (p<0.05) increased Nf-kB, alkaline phosphatase (ALP), alanine transaminase (ALT) and significantly (p<0.05) reduced Nrf-2 and antioxidant capacity. Administration of solvents fractions at 1000 mg/kg bwt significantly (p<0.05) reduced Nf-kB, ALP, ALT and significantly (p<0.05) increased Nrf-2 and antioxidant capacity of rats in the treated groups, such that, n-butanol fraction is the most effective. Data were analysed using analysis of variance and Duncan multiple range test at p<0.05. n-butanol fractions, can therefore be explored as oral remedy against cellular alterations in rats.

Keywords: Free Radical, *Adansonia digitata*, *Corchorus olitorius*, γ -irradiation, Nf-kB, Nrf-2 and cellular alterations, micro nuclei DNA, antioxidant

Introduction

Human exposure to radiation increases free radical level, that is capable of generating reactive oxygen species (ROS) and reactive nitrogen species (RNS) in human (IRES, 2010). ROS/RNS attack cells, tissues, organs and macromolecules (DNA, proteins and lipids) resulting in peroxidation, these are hallmark of oxidative stress and cellular alterations (Jhur-shur, 2009). Oxidative stress catalyzed protein unfolding and misfolding, unregulated activation of 26s proteasomes pathway and dysregulated Nrf-2 and Nf-kB (Said and Aiman, 2014). Free radical at above subtoxic concentration, exacerbate lipid

peroxidation, micronuclei DNA formation and fragmentation, pathological tissue damages and mutations (Fusco *et al.*, 2007). All these ultimately provoke cellular alterations.

Nf- κ B also known as Nuclear Factor Kappa-B, it's a basic leucine containing factor, with a molecular weight of 234kDa. It's mostly conserved in the cytosol as an inactive protein trimer/complex (p50-p52-I κ B) (Scheuring *et al.*, 1999). I κ B prevents the translocation of Nf- κ B to the nucleus, where it binds to the DNA binding region (DBR) of genes, thus blocking gene expression (Talalay, 2003).

Radiation-induced Free radical at or above 120 μ g/mol, mediates phosphorylation of the inactive Nf- κ B trimer (Talalay, 2003). Phosphorylated Nf- κ B is targeted for degradation in the 26S proteasome pathway (Ogbe *et al.*, 2010). I κ B is cleaved-off from Nf- κ B to become an active dimer, and is translocated into the nucleus, where it binds to DNA Binding Region (DBR) of genes expressing for majorly pro-oxidant proteins such as prostaglandins, cyclooxygenase-2, inducible nitric oxide synthase, hexokinase-2 BCL-2, IL-2 BAX and interleukins (Scheuring *et al.*, 1999).

Over expression of these prooxidant proteins (interleukins, prostaglandins, Bcl-2 and BAX proteins) have been implicated in induction of inflammations through ROS production. Overexpression of prostaglandins have been implicated in the growth of cancer (Scheuring *et al.*, 2001). They overwhelmed the endogenous antioxidant capacity, this significantly exacerbate oxidative stress, thus negatively altering the cellular system of rat exposed to irradiation (Patel *et al.*, 2001; Hall and Giaccia, 2006).

The nuclear factor related factor-2 (Nrf-2) is cytosolically repressed by Keap-1. It exists as an inactive trimer in the cytosol (Moi *et al.*, 1994a). Keap-1 prevents Nrf-2 translocation to the nucleus, thus preventing its binding to the antioxidant responsive element (ARE) of DNA of genes expressing for antioxidant enzymes (Moi *et al.*, 1994b).

During less oxidative stress conditions, this condition mediates Nrf-2 activation by phosphorylation, activated Nrf-2 dissociates from Keap-1 and translocates into the nucleus, where it binds to the DNA through the antioxidant responsive region (Patel *et al.*, 2001; Hall and Giaccia, 2006). It is also important to mention that secondary metabolites in plants also activate Nrf-2. Nrf-2 has been implicated in the induction of antioxidant enzymes such as glutathione S-transferase, superoxide dismutase, glutathione peroxidases and haem oxygenase in human system (Itoh *et al.*, 2004). Induction requires a common DNA sequence called antioxidant response element (ARE) that resembles the Nrf-2-binding motif (Nguyen *et al.*, 2003). This study investigated the effects of solvent fractions of n-hexane, ethylacetate and n-butanol fraction of methanol leaves extract of *A. digitata* and *C. olitorius* in gamma irradiated rat's cellular system.

Several synthetic agents have been used, either to scavenge or stabilize free radicals in experimental rats, for example synthetic vitamin E protects the integrity of acetyl choline receptors in normal neurons and prevents toxicity and apoptosis-induced ROS in male rabbits (Kennedy *et al.*, 2001; Behl and Moosmann, 2002). Synthetically produced vitamin C, stabilizes reactive intermediate as well as neutralizes free radical in albino rat (Adaramoye *et al.*, 2001). Most of these synthetic drugs have come with adverse side effects, as well as high cost, even when the money is available, they may not be accessible. Plant and plant products such as vegetables and fruits, contain secondary metabolites (flavonoids, polyphenols, tannins, saponins, glycosides, vitamins) that are cheap, accessible and safe. They mediate and scavenge free radicals and stabilize reactive species intermediate.

A. digitata

A. digitata is a member of the baobab family and commonly known as *Kukah* in Hausa, *luru* in Yoruba and tree of life in English. It's a deciduous tree with four growth phases and produces fruits consisting of a yellow-white pulp, with a floury texture and numerous round seeds, enclosed in a tough shell (Dharmendra *et al.*, 2006). The leaf of the baobab tree is food for many populations in Africa, especially the central region of the continent. It is a commonly used traditional plant consumed in food or used in the treatment of diseases such as anaemia, diabetes, lipid peroxidation disorders, ischemia reperfusion diseases and inflammatory bowel syndrome in South-west Nigeria (Lewanda *et al.*, 2007). The leaves are used as blood booster, anti-asthmatic, antihistamine and anti-tension. The leaves are also used to treat insect bites, guinea worm, internal pains, dysentery, diseases of the urinary tract, ophthalmia and otitis. Baobab leaves are used medicinally as a diaphoretic, astringent, expectorant and as a prophylactic against fever (Scheuring *et al.*, 1999). The carotenoid content of baobab leaves has also

been established to contribute to the prevention of oxidation in the human cell and lots of other diseases.

C. olitorius

Corchorus olitorius (Linn) is a leafy vegetable that belongs to the family *Tiliaceae*, it is known as Jute mallow in English and *Ewedu* in Yoruba. It is an annual herb with a slender stem and an important green leafy vegetable in many tropical areas including Nigeria, Egypt, Sudan, India, Bangladesh, in tropical Asia such as Philippine and Malaysia. The leaves (either fresh or dried) are cooked into a thick viscous soup or added to stew or soup and are rich sources of vitamins and minerals (Branda *et al.*, 2004) with various therapeutic uses in herbal medicine. The leaves extracts have been reported to possess hypotensive, anti-diabetic, anti-malarial, hepatoprotective, anti-microbial, anti-inflammatory, analgesics, anti-diarrhea, anti-viral, anti-carcinogenic and anti-mutagenic properties (Thyagarajan *et al.*, 1988; Odetola & Akojenu, 2000; Sripanidkulchai *et al.*, 2002; Adeneye *et al.*, 2006).

Materials and Methods

Collection of Plant Materials and Authentication

Fresh leaves of *A. digitata* and *C. olitorius* were purchased from 'Ipata' Market, Ilorin, Kwara State, Nigeria. The plants were identified and authenticated at the Herbarium Unit of the Department of Plant Biology, University of Ilorin, Kwara State. Voucher Specimen Number (UICH/001/951 and UITH/002/154) were deposited in the Herbarium respectively.

Preparation of Plant Extracts

Fresh leaves of *Adansonia digitata* and *Corchorus olitorius* were washed with clean water, cut into pieces and dried at room temperature (27°C) for two weeks. The dried plants were grinded using blender (Steelman K207, H.M and co Ltd, China). A known quantity (50 g) of powdered *Adansonia digitata* and *Corchorus olitorius* leaves were extracted in 200 ml Methanol (98.8 %) for 72 hours. The extracts were filtered using Whatman No. 4 filter paper. The filtrates were concentrated using Rotary evaporator (RE, k25, Longfin, Australia).

Preparation of n-hexane, ethylacetate and n-butanol Fractions of *A. digitata* and

n-hexane, ethylacetate and n-butanol partitioned fractions of *A. digitata* and *C. olitorius* were prepared by adopting the method described by Mbaoji *et al.* (2014).

Table 1: Percentage yield of the three solvent partitioned extracts of *A. digitata* and *C. olitorius*

Solvent Fraction	n-Hexane (%)	Ethylacetate (%)	n-Butanol (%)
<i>Adansonia digitata</i>	14.41	12.89	16.21
<i>Corchorus olitorius</i>	16.73	11.45	14.12

Ethical Approval

Ethical Approval was obtained from the University of Ilorin Ethical Committee, University of Ilorin. With the ethical approval number obtained as follow UERC/ASN/2018/1409 on the 13th September, 2018. The study was conducted following the guidelines on the care and use of laboratory animals.

Experimental Animals

A total of fifty-four (54) healthy Wistar rats (198 ± 5.00g) were obtained from the Animal Holding Unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. The animals were housed in clean cages and adequately ventilated house condition. They were allowed free access to rat pellets and clean tap water. The cages were cleaned daily and the study was conducted following the guidelines on the care and use of laboratory animals. Ethical approval was obtained from the University of Ilorin

Ethical Committee, University of Ilorin.

Exposure of Animal to Radiation

Wistar rats were exposed to 6 grey whole-body gamma radiations by the method described by (Kamani *et al.* (2008) and modified by Nwozo and Bello (2014). Radiotherapy Machine with Cobalt 60 was the source of radiation. The exposure was carried out in the Department of Radiotherapy, College of Medicine, University of Ibadan, Ibadan, Nigeria.

Drugs, Chemicals and Assay Kits

The drugs and other chemicals used in the study were commercially obtained from Monobind International, Lake Forest, USA. ELISA kits were products of Cayman Laboratory LTD., USA. All other chemicals will be of analytical grades and prepared in all-glass distilled water.

Secondary Metabolite Constituents of *Adansonia digitata* and *Corchorus olitorius*

Qualitative and quantitative secondary metabolites screening of *A. digitata* and *C. olitorius* leaves were determined by adopting the methods described by Harborne (1993); Sofowora (1993) and Ajayi *et al.* (2010). All determinations were done in triplicates.

Table 2: Administration of the solvent fractions to irradiated rats

Groups	Treatments
I	Non-irradiated, fed with rat chow and distilled water only
II	Irradiated animal and not treated at all
III	Pre-administered with 1000 mg/kg bwt ethyl acetate fraction of <i>A. digitata</i> before exposure to irradiation
IV	Pre-administered with 1000 mg/kg bwt ethyl acetate fraction of <i>C. olitorius</i> before exposure to irradiation
V	Pre-administered with 1000 mg/kg bwt n-butanol fraction of <i>A. digitata</i> before exposure to irradiation
VI	Pre-administered with 1000 mg/kg bwt n-butanol fraction of <i>C. olitorius</i> before exposure to irradiation
VII	Pre-administered with 1000 mg/kg bwt n-hexane fraction of <i>A. digitata</i> before exposure to irradiation
VIII	Pre-administered with 1000 mg/kg bwt n-hexane fraction of <i>C. olitorius</i> before exposure to irradiation
IX	Pre-administered with 1000 mg/kg bwt Vitamin-C before exposure to irradiation

Bwt: body weight.

Preparation of Tissue Homogenate

After sacrifice, the rats were dissected to isolate the liver. The isolated liver was gently cleansed with 1.14 % KCl washing solution to remove blood stains and dirty. The liver was weighed and immediately stored in ice cold 0.25M sucrose solution. 1g of the liver was cut with a clean scalpel, homogenized in ice-cold 0.25M sucrose solution (1:5^{w/v}). The homogenates were cold centrifuge at 12000g for 15 minutes, the supernatant was pipetted into a clean sample tube and stored in the freezer (-5°C) until required for further analysis.

Collection of Blood

Blood was collected by the method described by Akanji and Ngaha (1989).

Collection of Serum

Serum was collected by the method described by Yakubu *et al.* (2003)

Assays

p53, Nf-kB and Nrf-2 assay was carried out by the method adopted by Cayman *et al.* (2003) using the principles of enzyme linked immunosorbent assay (ELISA). Alkaline phosphatase, aspartate aminotransaminase and alanine transaminase assay were carried by the method adopted by Reitman and Frankel (1960b); Reitman and Frankel (1960b); Caboli *et al.* (1965b). Superoxide dismutase, Catalase, Glutathione-s-transferase, Glutathione peroxidase and Glutathione assay were carried out by the method adopted by Haliwel *et al.* (2001); Yhoungh-shur (1980); Abolaji (2015); Lazekhs *et al.* (1982b); Beutler *et al.*(1963).

Statistical Analysis

Data were expressed as mean \pm SEM of six determinations. Duncan Multiple Range Test was used and complemented with Student's T-test. Statistical Package for Social Sciences, version 20.0 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. Statistical significance was set at 95% confidence interval (Mahajan, 1997).

Results and Discussion

Exposure of rats to 6 grey gamma radiation altered cellular system of such rats, by altering cellular parameters (Nf-kB, Nrf-2, ALT, AST, SOD, CAT) associated with physiological functioning of such cellular system (Haliwell, 2001). An increase in free radical level, above physiological concentration (from 20-120ug/mol) is an underlying mechanism through which radiation altered cellular parameters (Simões, 2009). As revealed by the results obtained from this study, radiation significantly ($p < 0.05$) altered Nf-kB, Nrf-2, SOD, CAT, GPx, ALT, ALP and AST level of not treated rat as shown in Figures 1 – 3. The result obtained from the irradiated not-treated group in figure 3a, b and c further alluded to the alteration caused by irradiation as it significantly ($p < 0.05$) reduced SOD, CAT, GPx and GST level as shown figure 3 above in irradiated rats.

Medicinal quality of plants is associated with the bioactive constituents therein the plants, this qualities are the underlying factors responsible for the pharmacological functions of most plant (Baskurt *et al.*, 2009). The influence of *Adansonia digitata* and *Corchorus olitorius* leaves on cellular parameters and metabolism of irradiated rats as shown in this study, have strongly been alluded to the presence of flavonoids, polyphenols, tannins, saponins, alkaloids in the methanolic extract as revealed in Table 1 above. The use of secondary metabolites in plants to regulate cellular metabolism and offer pharmacological protection have long been validated (Olanlokun, 2013). Many plants with medicinal properties have been used in the treatment and protection of cells from cellular alterations. Such as *A. digitata* and *C. olitorius* in this study.

Table 1: Secondary Metabolites Constituents of Methanolic Extract of *A. digitata* and *C. olitorius* leaves

Metabolites	<i>Adansonia digitata</i> (g/dL)	<i>Corchorus olitorius</i> (g/dL)
Alkaloids	78.76 \pm 0.43	72.36 \pm 2.05
Anthraquinone	ND	ND
Cardiac glycosides	ND	ND
Flavonoids	13.42 \pm 1.03	104.81 \pm 0.18
Polyphenols	147.90 \pm 0.68	203.14 \pm 0.32
Phlobatannins	ND	ND
Saponins	16.59 \pm 1.85	22.17 \pm 0.24
Tannins	198.98 \pm 0.14	127.40 \pm 0.16
Terpenoids	42.45 \pm 0.45	21.25 \pm 1.08
Steroids	30.45 \pm 0.51	12.59 \pm 1.85

ND: Not Detected

Nf-Kb

Polyphenols have been validated to upregulate Nrf-2 and down regulate of Nf-kB (Giuliano *et al.*, 1999). Nrf-2 have been implicated in the induction of superoxide dismutase, catalase and Glutathione, whereas Nf-kB mediate their repression (Bossy-Wetzel *et al.*, 2004). The significant ($p < 0.05$) upregulation of Nrf-2 and down-regulation of Nf-kB as shown in figure 1 (a, b and c) and 2 (a, b and c) maybe associated with the presence of significant ($p < 0.05$) amounts of polyphenols in n-hexane, ethylacetate and n-butanol fraction of methanol leaves extract of *Adansonia digitata* and *Corchorus olitorius*, with n-butanol fraction been the most effective of the three solvent as shown in figure 1 and 2 above.

Flavonoids have been implicated in the prevention of malondialdehyde formation in rats cellular system, by impeding the formation of chain of lipid peroxidation (Finkel, 2012). Malondialdehyde is a product of lipids peroxidation, and have been implicated in the genesis of cardiovascular diseases, hypertension, ischemia reperfusion diseases, atherosclerosis etc. (Kuate, 2010), thus the significant ($p > 0.05$) reduction in the level of MDA, as shown in figure 3 a, b and c, in irradiated rat administered with n-hexane, ethylacetate and n-butanol fraction of *Adansonia digitata* and *Corchorus olitorius* leaves, maybe alluded to the presence of significant ($p < 0.05$) amount of polyphenols in the plant as shown in figure 3 above. Even though n-butanol fraction showed the most effective pharmacological effects of the three solvent fractions.

As observed in the irradiated not treated group in Figures 1 – 3, there was significant ($p > 0.05$) reduction in the level of antioxidant enzymes capacity of irradiated rats. This have been implicated in the genesis of cellular alterations in irradiated rat. Farombi and Yhong-sur (2001) revealed that epigallocatechin gallate scavenge free radical produced by administration of aspartame to mice. Thus, the significant ($p > 0.05$) reduction in the antioxidant capacity of rat in this research as shown in figure 1,2 and 3 above, maybe due to radiation-induced free radical in rats.

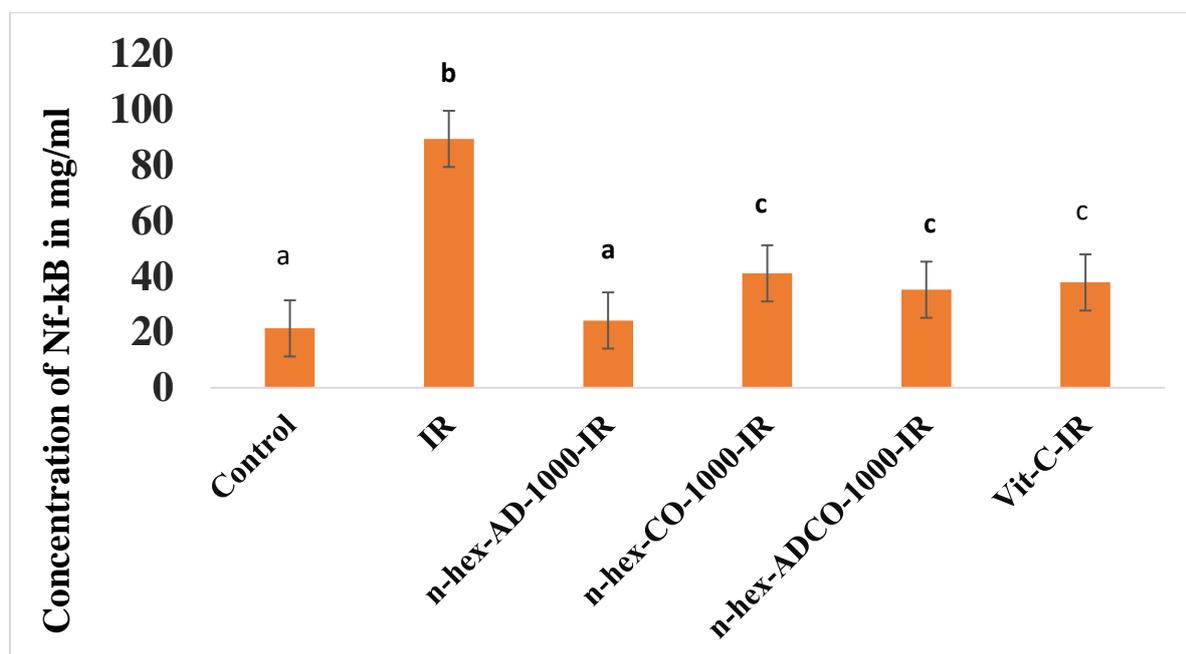


Figure 1a: Effects of n-hexane fraction of Methanol Leaf Extract of *Adansonia digitata* and *Corchorus olitorius* on Nf-kB status of irradiated Rats. Means \pm SEM; n=6, ($P < 0.05$); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

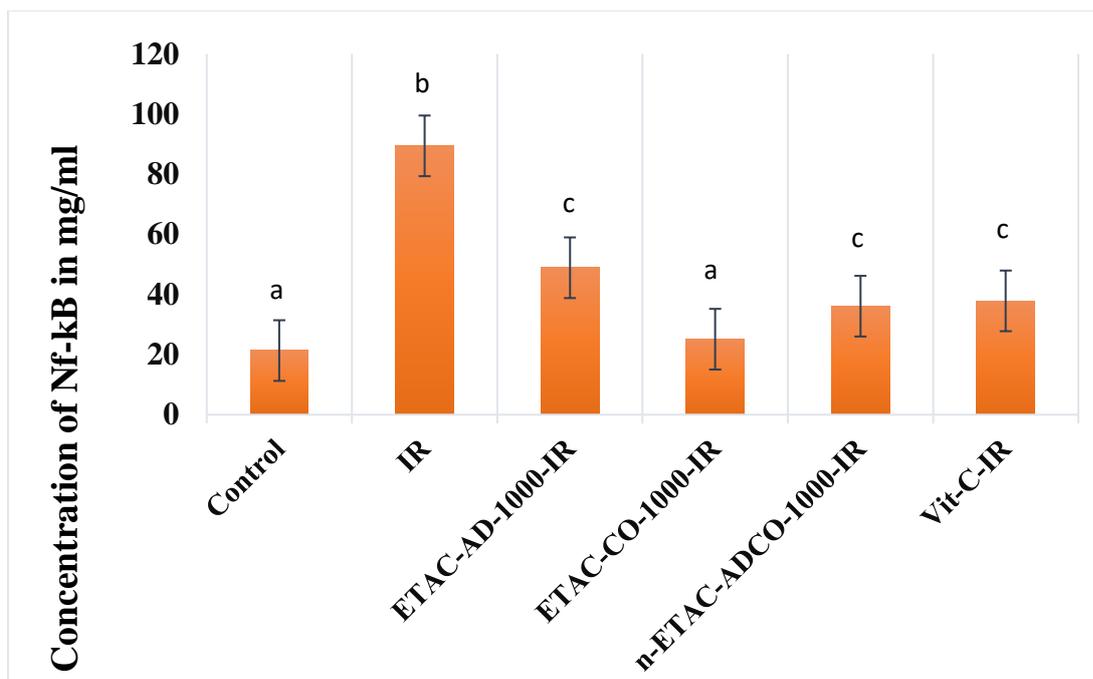


Figure 1b: Effects of ethylacetate fraction of Methanol Leaf Extract of *Adansonia digitata* and *Corchorus olitorius* on Nf-kB status of irradiated Rats. Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

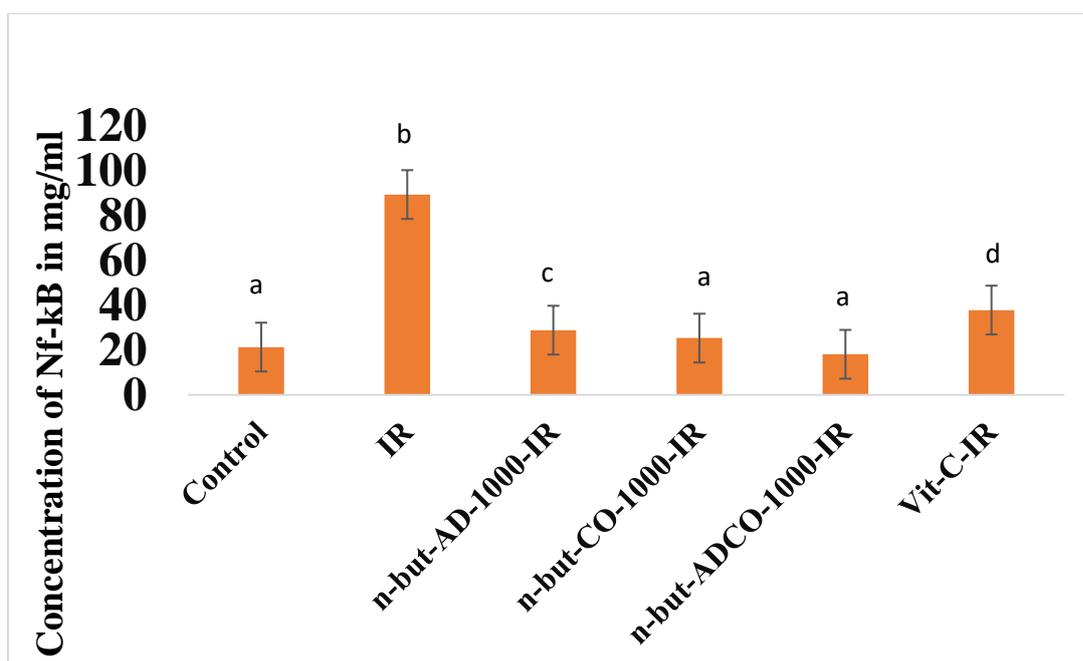


Figure 1c: Effects of n-butanol fraction of Methanol Leaf Extract of *Adansonia digitata* and *Corchorus olitorius* on Nf-kB status of irradiated Rats. Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*; 1000; 1000 mg/ kg body weight.

Nrf-2

Flavonoids have also been implicated in the upregulation of Nrf-2; Nrf-2 mediated the induction and expression of antioxidant enzymes responsible for the scavenging of free radicals in rats exposed to oral anthracene (Olanlokun and Akomolafe, 2013). Alkaloids mediate the expression and induction of Gpx. Gpx is responsible for the stabilization of ROS/RNS generated by benzo(a)pyrene in rabbits (Rushmore, Morton and Pickett, 1991). The significant ($p < 0.05$) increase in SOD, CAT, GST, Gpx and GSH in irradiated rats administered with n-hexane, ethylacetate and n-butanol fractions of methanolic extract of *A. digitata* and *C. olitorius* leaves may be due to the presence of significant ($p < 0.05$) amount of catechin, quercetin and epigallocatechin in the leaves of *A. digitata* and *C. olitorius*. They might also be responsible for the increase in the induction of the antioxidant enzymes of the treated rats groups in this study.

As observed in the irradiated not treated group from Figure 1, there was significant ($p < 0.05$) increase in the level of activated Nf-kB, suggesting that radiation-induced free radicals have exacerbated Nf-kB activations as shown in Figure 1. Nf-kB has been implicated in the expression of pro-inflammatory proteins and suppression of antioxidant enzyme synthesis (Rushmore and Pickett, 1990). The activation of Nf-kB, by irradiation might be responsible for the significant ($p < 0.05$) reduction of SOD, catalase, GPx and GST in irradiated not treated group as shown in Figure 3a, b and c.

Luteolin and eleagnin are flavonoids and polyphenols respectively, they have been validated to prevent the activation of an inactive Nf-kB in the cytosol (Motohashi, 2004). Luteolin and eleagnin also mediated the induction of antioxidant enzymes, that have been implicated to scavenge free radicals that are capable of exacerbating pathological conditions and cellular alterations in rat (Olanlokun, 2013). As revealed by the result from Figure 1a, b and c, irradiation has significantly ($p < 0.05$) exacerbated Nf-kB activation in irradiated rats, but administration of n-hexane, ethylacetate and n-butanol fractions of methanolic extract of *A. digitata* and *C. olitorius* leaves significantly ($p < 0.05$) reduced the activation of Nf-kB in rat exposed to radiation, such that n-butanol fraction is the most effective of the solvent fraction as shown in Figure 1a, b and c.

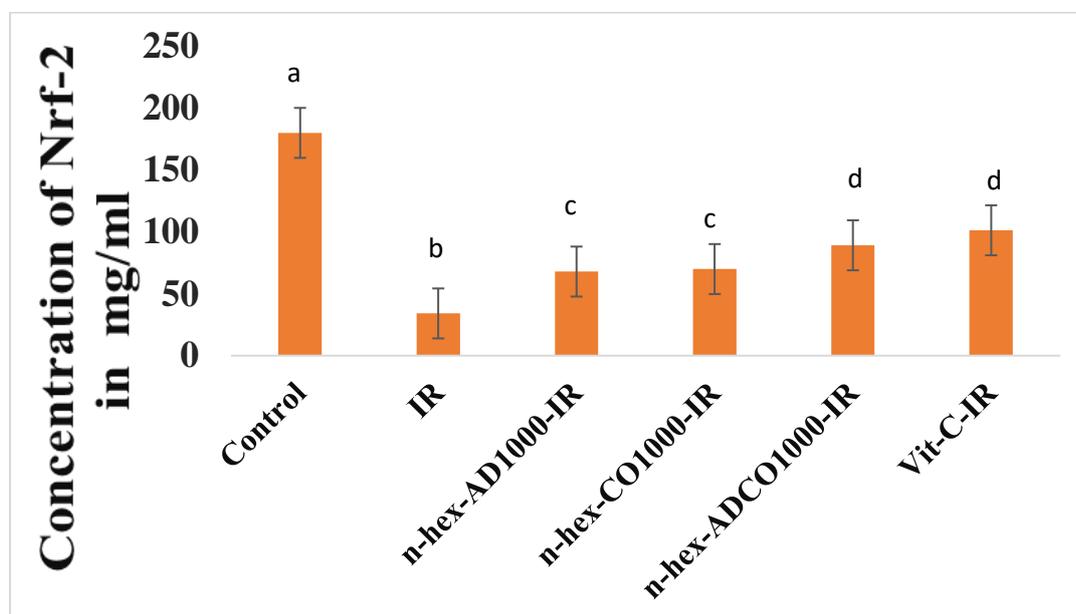


Figure 2a: Effects of n-Hexane, Ethylacetate and n-Butanol Partitioned-fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Nrf-2 status of irradiated Rats. Means \pm SEM; n=6, ($P < 0.05$); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

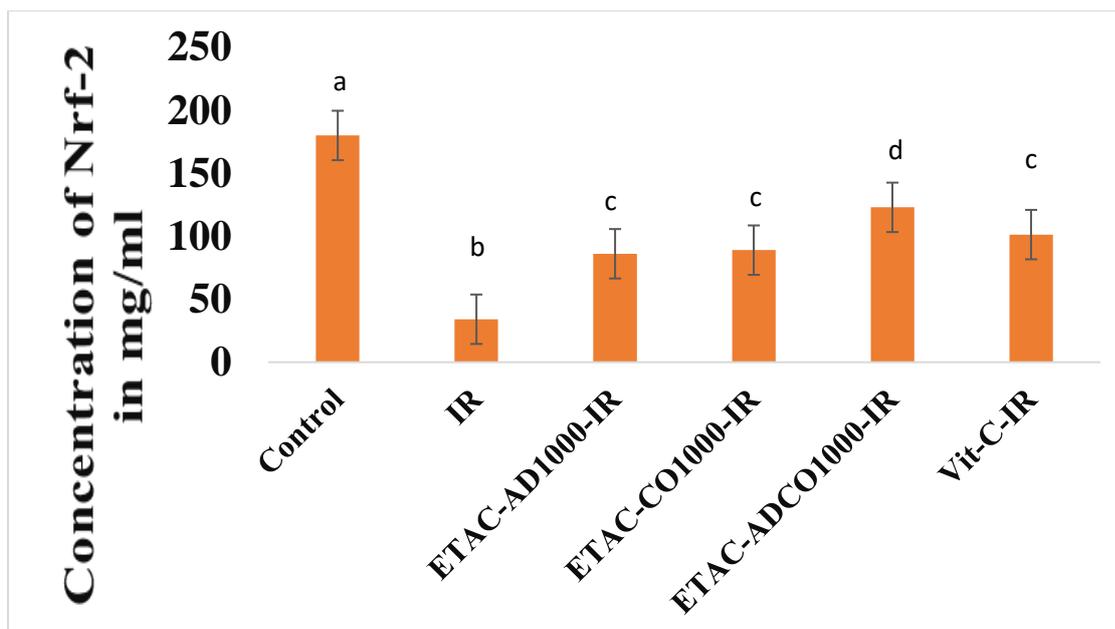


Fig 2b: Effects of Ethylacetate fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Nrf-2 level of irradiated Rats. Means \pm SEM; n=6, (P < 0.05); IR; irradiation, ETAC: ethylacetate, AD: *A. digitata* CO: *C. olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

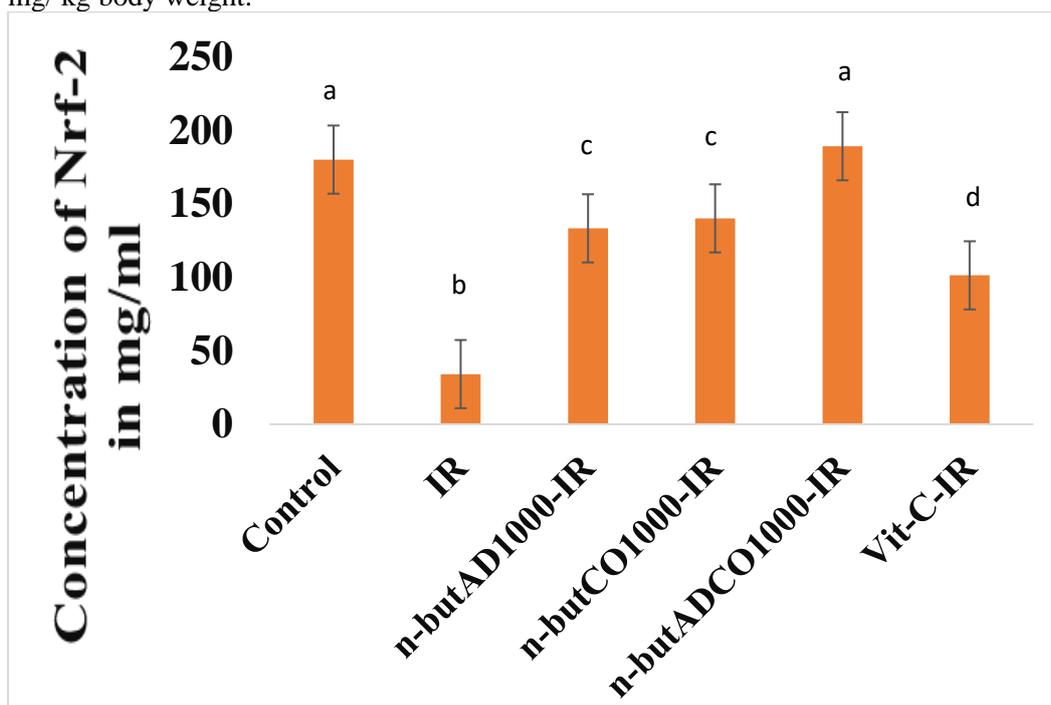


Figure 2c: Effects of n-Butanol fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Nrf-2 level of irradiated Rats. Means \pm SEM; n=6, (P < 0.05), IR; irradiation, n-but: n-butanol, AD: *A. digitata* CO: *C. olitorius*; ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

Antioxidant System

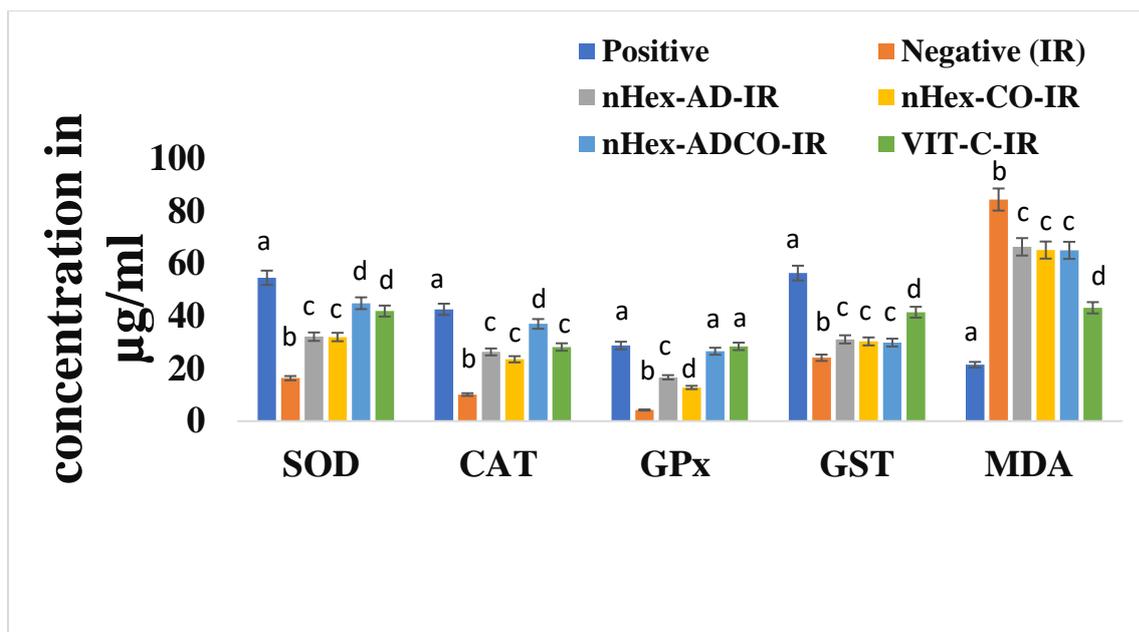


Fig 3a: Effects of n-Hexane Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Antioxidant Capacity of Irradiated Rats

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight. SOD; Superoxide dismutase, CAT; Catalase, GPx; Glutathione peroxidase, GST; Glutathione-s-transferase, MDA: Malondialdehyde

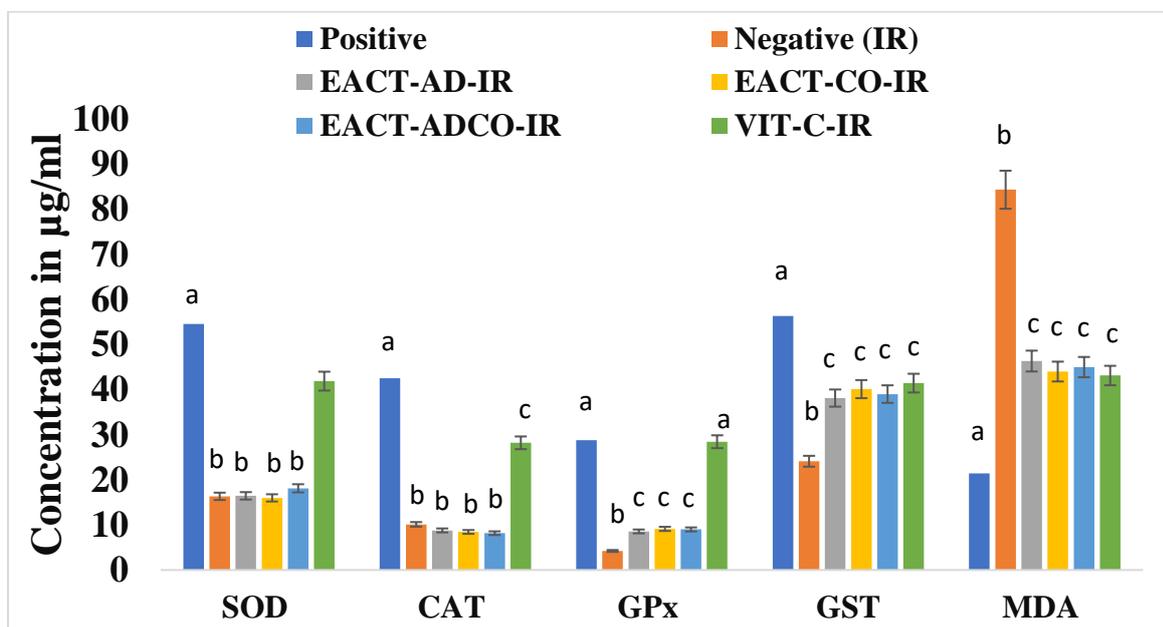


Fig 3b: Effects of Ethylacetate Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Antioxidant Capacity of Irradiated Rats.

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight. SOD; Superoxide dismutase, CAT; Catalase, GPx; Glutathione peroxidase, GST; Glutathione-s-transferase, MDA: Malondialdehyde

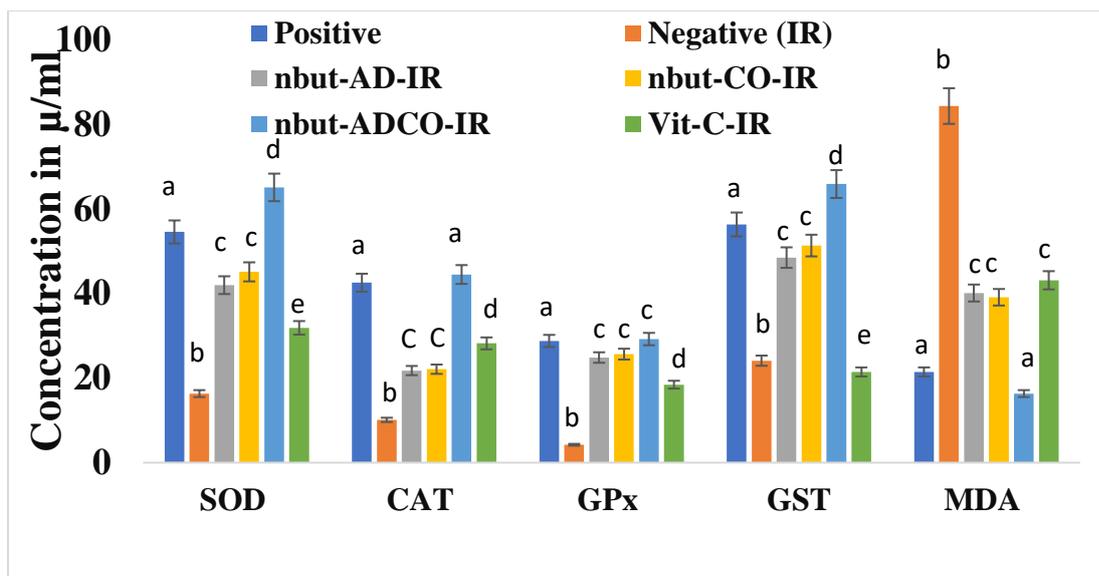


Fig 3c: Effects of n-Butanol Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on Antioxidant Capacity of Irradiated Rats

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight. SOD; Superoxide dismutase, CAT; Catalase, GPx; Glutathione peroxidase, GST; Glutathione-s-transferase, MDA: Malondialdehyde.

Toxicity Profile

As observed in the irradiated not treated group from Figure 4a, b and c, there were significant ($p > 0.05$) increase in the level of serum ALT, AST and ALP of irradiated rats, this suggest, that there is irradiation-induced compromised of the hepatocyte(liver) function (Malomo *et al.*, 2001). This action of free radicals on the liver membrane lead to compromise in functions of the membrane channels, thus resulting in efflux of ALT, AST and ALP out the membrane-bound into the serum stream (Adesokan *et al.*, 2001). The significant ($p < 0.05$) increase in the activities of serum ALT, AST and ALP of irradiated rats as shown in Figure 4a, b and c, maybe due to the exposure of the rats to irradiation, which inturns increased free radical generation and attack on the membrane.

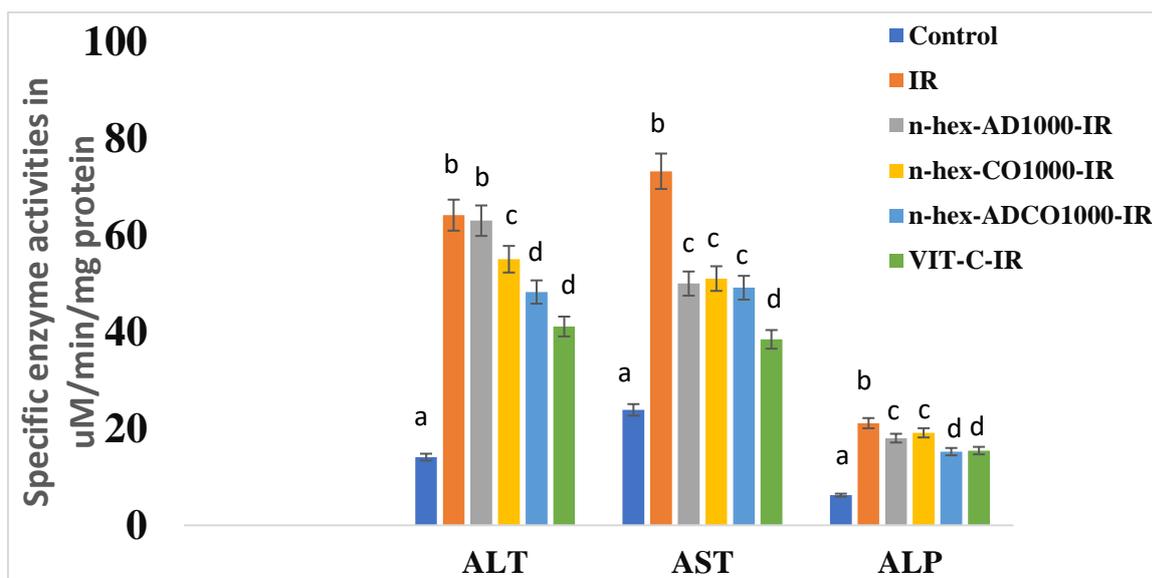


Fig 4a: Effects of n-hexane Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on ALT, AST and ALP of Irradiated Rats

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight.

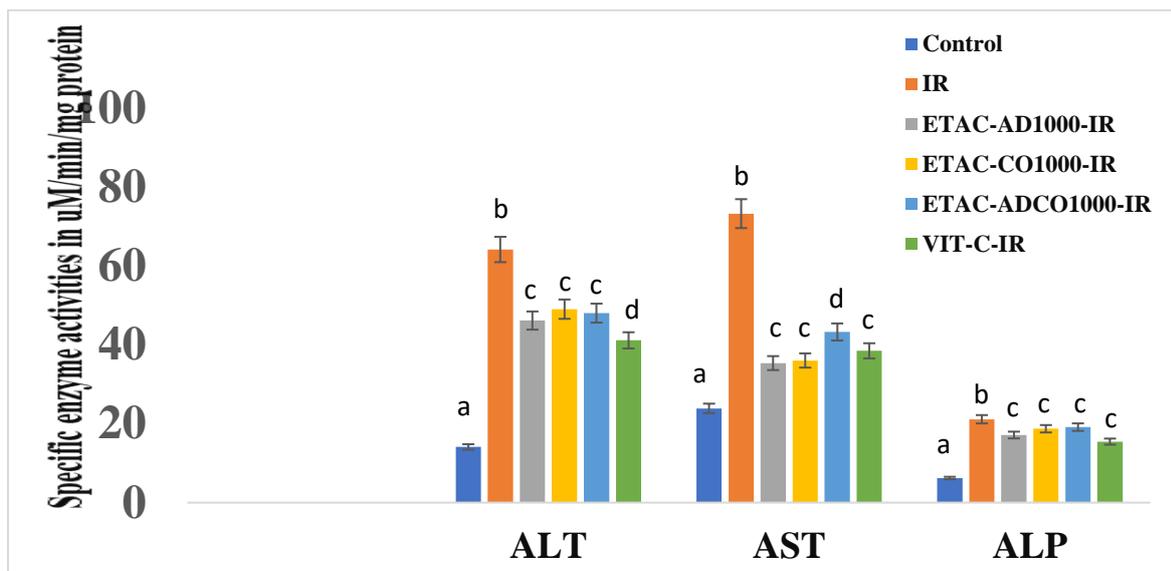


Fig 4b: Effects of Ethylacetate Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on ALT, AST and ALP of Irradiated Rats

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight

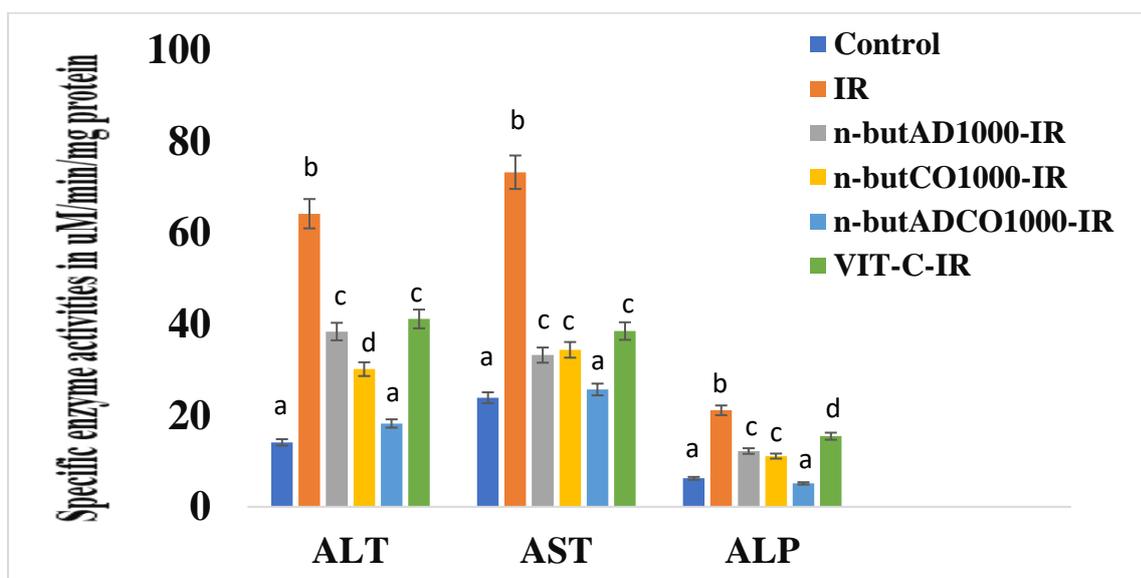


Fig 4c: Effects of n-butanol Fraction of Methanol Leaves Extract of *Adansonia digitata* and *Corchorus olitorius* on ALT, AST and ALP of Irradiated Rats

Means \pm SEM; n=6, (P < 0.05); IR: irradiation, n-hex: n-hexane, AD: *Adansonia digitata*, CO: *Corchorus olitorius*, ADCO: *A. digitata* + *C. olitorius*, 1000; 1000 mg/ kg body weight

The administration of n-hexane, ethylacetate and n-butanol fractions of methanol leaves extract of *A. digitata* and *C. olitorius* to irradiated rats, significantly ($p < 0.05$) reduced the activities of ALT, AST and ALP of the treated group, such that, n-butanol is the most effective of the three solvent.

The significant ($p < 0.05$) reduction in the activities of ALP, AST and ALP in the treatment group, as shown in figure 4a, b and c maybe due to the presence of significant ($p < 0.05$) amount of Flavonoids in the n-hexane, ethylacetate and n-butanol fractions of the methanol leaves extract of *A. digitata* and *C. olitorius* (Djeussi *et al.*, 2013). Astragalins and eleagnins (flavonoids and polyphenols) have been

implicated in the reversal of temporary liver damage in colchicine-induced liver damage in mice (Oduola and Owumi, 2008). Thus, the presence of significant ($P < 0.05$) amounts of flavonoids and polyphenols in n-hexane, ethylacetate and n-butanol fractions of methanol leaves extract of *A. digitata* and *C. oclitorius* might be responsible for the reversal of the damages observed in the administered groups.

Conclusion

At the end of this research, there is no doubt, that radiation induced cellular alteration in rat, but administration of n-butanol fraction of methanol leaves extract of *Adansonia digitata* and *Corchorus oclitorius* is the most effective of the solvent fraction, such that it was able to effectively protect and attenuates radiation-induced cellular alteration in rats. We can therefore conclude, that *Adansonia digitata* and *Corchorus oclitorius* leaves can be explored as oral remedy against radiation-induced cellular alterations.

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Conflict of Interest

There is no conflict of interest on this work be it at the level of research or manuscript submission.

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