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Protective effect of irradiated-Ficus carica dried leaf against hepatic-damage induced by ethanol in male rats

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► ***Original article***

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ABSTRACT

Background: This study aimed to evaluate the effects of gamma rays and electron beams (5kGy) on the chemical content of dried fig leaves. In addition, the hepatoprotective effect of irradiated-dried fig leaves powder (GFLP) against ethanol (EtOH)-induced hepatotoxicity in rats was investigated. ***Materials and Methods:*** The 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity, sugar, total phenolic, total flavonoid content, phytate, and tannin contents of either raw or irradiated dried fig leaves were detected in this study. For the experimental study, 28 male rats were divided into four groups; Group C, control; Group 2: received an oral dose of ethanol (0.5 ml/100 gm b.wt. /day /5 weeks) and Groups 3-4: rats fed daily (60 min after EtOH injection) on either raw or irradiated fig leaves powder (4% of the weight of the diet). ***Results:*** It was observed from the results that gamma-irradiation (5 kGy) resulted in a higher increase in DPPH radical scavenging activity, sugar, total phenolic and total flavonoid content, and a higher reduction in phytate and tannin contents than e-beam irradiation. The results of the experimental study revealed that treatment of EtOH-rats with GFLP resulted in a significant reduction in the serum blood alcohol level, the activity of some liver enzymes, level of tumor necrotic factor-alpha, interleukin-6, malondialdehyde, and lipid profile contents associated with significant elevation in the level of high-density lipoprotein-cholesterol, hepatic total antioxidant capacity and glutathione content relative to EtOH-group. ***Conclusion:*** The enhanced antioxidant activities of Ficus carica leaves through irradiation could be applied as a natural antioxidant source in foodstuff.

INTRODUCTION

Liver ailments are normally related to numerous reasons including genetic inheritance, poisonous ingestion, viral infection, and immoderate alcohol abuse. Among these reasons, each acute and persistent alcohol intake is a harmful social, economic, and clinical problem. Excessive alcohol consumption is a causal component in an extensive variety of multi-organ pathology, together with alcoholic liver disease (ALD) [1]. Previous research has counseled that mitochondrial damage, overproduction of free radicals, and oxidative damage are essential pathogenic occasion's withinside the development of ALD. The severity of this ailment will increase withinside time and dose-dependent manner with alcohol consumption and ranges initially from steatosis and steatohepatitis to fibrosis and cirrhosis [2]. Untreated alcoholic liver ailment becomes hepatocellular carcinoma, frequently inflicting mortality. Antioxidants and bioactive compounds from natural fruits and plants have been pronounced to ameliorate the impact of oxidative stress and infection in liver-associated illnesses without causing any side effects [2].

Therefore, introducing nutritional foods with high levels of antioxidants can enhance the efficient and cost-effective therapy of free radical disorders while avoiding the toxicities and unintended side effects produced by traditional drugs [2]. Figures (*Ficus carica*); belongs to the family Moraceae; is of tropical and subtropical plant cultivated for its nutritive and therapeutic characteristics around the world [3]. Figure leaves, bark, tender shoots, fruits, seeds, and latex have been utilized as folk medicine utilized in the treatment of Jaundice, diarrhea, and nutritional anemia. Figure trees are frequently pruned to stimulate their growth. As a result, a lot of discarded branches and leaves are accumulated each year, causing environmental pollution and waste of resources [3]. According to some research, it was discovered that these leaves contain higher levels of total phenol and flavonoid than the stem bark and fruit, which can operate as powerful antioxidants [4]. Many flavonoids content (rutin, quercetin, and anthocyanin) found in fig leaves are known to have a variety of pharmacological activities that can be used in the treatment of hepatotoxicity, cardiovascular diseases, osteoporosis, diabetes, and diarrhea [4]. Also, fig leaves have been found to contain sugars, pectin, tannins, vitamin C, and trace elements. As a result, utilizing fig waste leaves to their full potential

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is a practical means of realizing resource utilization and preventing environmental contamination [3].

The production of dried figs involves exposing the figs to multiple sources of fungal infection, making it a particularly "risky" operation. Both the dried figs' surface and inside cavities may contain toxic fungus. The crucial time for the contamination of dried figs is during the fruit's ripening stage on the tree [5]. One of the contemporary techniques for food preservation is food irradiation, a type of physical therapy that uses ionizing radiation (such as gamma rays and electron beam radiation) to stop the growth of undesired biological organisms or reduce their population [6]. International organizations such as the World Health Organization (WHO), the Food and Agriculture Organization (FAO) of the United Nations, the International Atomic Energy Agency (IAEA), and Codex Alimentarius have validated the safety of this technology. By removing or destroying mycotoxins while maintaining the food's organoleptic properties and safety aspects, food radiation processing has the potential to increase food shelf life [6].

This study aims to study the effectiveness of either gamma-irradiation or electron beam processing, as one of the safe sterilizing techniques, on some chemical contents of dried fig leaves. As well as, to investigate the hepatoprotective effect of irradiated fig leaves against Ethanol induced-liver toxicity in rats in a trial to provide a natural, and cost-effective therapeutic source to protect against alcoholic-liver diseases.

MATERIAL AND METHODS

All experiments were carried out in 2022 at the Egyptian atomic energy authority, food irradiation department. Dried Fig leaves were purchased from a local store of spices, grains, and oils (Cairo, Egypt). under the shade. A mortar and pestle were used to crush the dried leaves. Until usage, the powdered leaf sample was kept in an airtight container. The Sigma Chemical Co. (St. Louis, MO, United State agency) supplied the chemicals and reagents.

Gamma Irradiation treatment

Dried fig leaves powder was transferred into polyethylene bags and treated with gamma rays at the doses of 5 kGy, using Indian Gamma Cell (Ge 4000 A) 60Co source at a dose rate of 0.8053 kGy/h at the National Centre for Radiation Research and Technology (NCRRT), Egypt.

Electron beam irradiation (EBI)

Electron beam irradiation at the doses of 5 kGy was carried out at the NCRRT using an electron beam accelerator (model: ICT, VIVIRAD CO, France). This accelerator has a maximum energy of 3 MeV, a beam current of 30 MA, a beam power of 90 KW, a scan width of 90 cm, and a distance between the scanner and the conveyor system of 53.0 cm. The Department of Radiation Protection and Dosimetry at the National Center for Radiological Research and Technology (NCRRT) did extensive dose mapping in compliance with Egyptian requirements.

Determination of sugar, total phenolic, and Total flavonoid content

Free sugar contents of raw and irradiated samples were determined by high-performance liquid chromatography (HPLC), after an extraction procedure previously described by the study of Pereira *et al.* [7]. Dried sample powder (1.0 g) was spiked with melezitose as internal standard (IS, 5mg/mL), and extracted with 40mL of 80% aqueous ethanol at 80°C for 30min. The resulting suspension was centrifuged at 15,000 g for 10 min. The supernatant was concentrated at 60°C under reduced pressure and defeated three times with 10 mL of ethyl ether, successively. After concentration at 40°C, the solid residues were dissolved in water to a final volume of 5mL and filtered through 0.2µm Whatman nylon filters. The compounds were identified by chromatographic comparisons with authentic standards. Quantification was performed using the internal standard method and sugar contents were further expressed in g/100g of dry weight (dw).

Either raw or radiated samples were put in water, shaken (10 g/L, v/w) for 15–20 min, and then filtered using the Whatman filter paper. The total phenolic content of each leaf sample was determined by the Folin-Ciocalteau method as developed by Otles and Yalcin [8]. To 50 µL of water extract or standard solution, 250 µL of Folin-Ciocalteau reactive was added. This mixture was kept at room temperature for 5 minutes in a dark environment. At the end of this time, a 750L of 7 percent Na_2CO_3 solution was added. In this approach, the hydroxyl groups in phenolics could deliver H to water. Pure water was used to dilute the mixture to 5 mL. The mixture was then kept at room temperature for 120 minutes in a dark environment to react. At 760 nm, the absorbance of the samples and standards were measured. Instead of the 50L extract, an 80 percent methanol solution was added to the blank solution. A calibration curve was constructed using gallic acid equivalent standards to determine total phenolic content.

The flavonoid content of leaf samples was evaluated by the procedure of Surana *et al.* [9]. 0.50 mL of leaf powder extracts were carefully measured in a test tube. The test tube was then filled with a 0.1 mL aluminum chloride solution, 1.50 mL methanol, 0.1 mL potassium acetate solution, and 2.8 mL distilled water mixed. Sample blanks for extract and rutin standard dilutions (10-100 g/ml) were generated in the same method but with distilled water rather than aluminum chloride solution. The

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solutions were filtered using Whatman filter paper (No. 1) to determine the absorbance. The absorbance ratios were compared to blanks at 510 nm. 1 mg rutin per gram of extract was used to calculate the overall flavonoid content.

Determinations of tannins and phytate contents

A-The tannins were determined by the Folin-Ciocalteu method [10]. About 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water and 0.5 ml of Folin-Ciocalteu phenol reagent, 1 mi of 35% sodium carbonate solution and diluted to 10 ml with distilled water. The mixture was shaken well and kept at room temperature for 30 min. A set of reference standard solutions of tannic acid (20, 40, 60, 80, 100 µg/ ml) were prepared in the same manner as described earlier. Absorbance for test and standard solutions was measured against the blank at 700 nm with a UV/ Visible spectrophotometer. The estimation of the tannin content was carried out in triplicate. The tannin content was expressed in terms of mg of tannic acid equivalents/ g of dried sample.

B-Phytates were estimated using the method described by Sotelo *et al.* [11]. One gram of each sample was extracted with 10 mL of 3 % trichloro acetic acid to precipitate the phytate as ferric phytate with 10 ml of 0.1 % ammonium ferric sulfate. The ferric phytate is converted to ferric hydroxide and sodium phytate by boiling with 10 mL (0.5 M) sodium hydroxide. The precipitate was dissolved with 1 mL of 0.65 M HCl and phytate was determined using a spectrophotometer at 519 n m. Phytic acid, Dodecasodium salt (Sigma Chemical Co. (St. Louis, MO, USA)) was used as standard.

Determination of radical scavenging activity

The radical scavenging activity (RSA) of raw and irradiated samples was measured using the method of Valko *et al.* [12] by dissolving about 24mg of 1,1-diphenyl-2-picryl hydrazyl (DPPH) in 100 mL of methanol for making the stock solution. Filtration of DPPH stock solution using methanol yielded a usable mixture with an absorbance of around 0.973 at 517 nm. In a test tube, 3 mL DPPH workable solutions were combined with 100 µL of either raw or irradiated sample extract. Three milliliters of the solution containing DPPH in 100 µL of methanol is often given as a standard. After that, the tubes were kept in complete darkness for 30 min. The absorbance was therefore determined at 517 nm. The following formula was used to compute the percentage of antioxidants or RSA

% of antioxidant activity= [(Ac−As) ÷Ac] × 100

Ac: -Control reaction absorbance; as: -Testing specimen absorbance.

Animals

Male rats (200-230g body weight (B. WT), 8 weeks) were purchased from the Egyptian Holding Company for Biological Products and Vaccines (Cairo, Egypt) and used for the different investigations carried out in the present study. For two weeks the rats were acclimated to controlled laboratory conditions. Rats were maintained on a stock rodent diet and tap water that were allowed *ad libitum*. All animal procedures were carried out following the Research Ethics Committee for experimental studies (Human & Animal subjects) at the National Centre for Radiation Research and Technology (REC-NCRRT), Egyptian Atomic Energy Authority (Cairo, Egypt). Conformed to the CIOMS and ICLAS International Guiding Principles for Biomedical Involving Animals 2012.

Grouping of animals

The animals were randomly divided into 4 groups, each consisting of 7 rats. Group C: rats fed on a balanced diet and served as control, ethanol control (EtOH): rats were administered an oral dose of ethanol everyday between 10:00 AM and 11:00 AM for 5 weeks (0.5 mL C_2H_5OH/100 gm body weight) [13]. Group EtOH-RFLP: rats were administered an oral dose of ethanol and fed on the raw fig leaves powder (RFLP) by 4% of the weight of the diet [4], after 1 hour of alcohol administration. Group EtOH-GFLP: rats were administered an oral dose of ethanol and fed on the gamma-irradiated fig leaves powder (GFLP) by 4% of the weight of the diet [4], after 1 hour of alcohol administration.

At the end of the experimental period (5 weeks), after 24 hrs. From the last dose animals from each group were sacrificed. Blood samples were withdrawn by cardiac puncture after a slight analyzation of rats using diethyl ether and allowed to coagulate and centrifuged to get serum for biochemical analysis. The liver was removed and processed for biochemical studies.

Biochemical analysis

During the experimental period (5 weeks), the blood was taken from the tail vein 1 h after administration of either ethanol only or ethanol with raw or irradiated-dried fig leaves. Blood alcohol levels (BAL) were measured using the alcohol dehydrogenase kit from Sigma Chemical Co. (St. Louis, MO, USA) [14]. The activity of serum aspartate transaminase (AST) and alanine transaminase (ALT) was estimated according to Reitman and Frankel [15] and serum γ-glutamyl transferase (γGT) was assessed according to Rosalki [16]. Detection of serum tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6) was performed by ELISA technique (BioSource International, Camarillo, CA, USA) according to the manufacturer's instructions by using Rayto Microplate Reader RT-2100C at absorption wavelength 450 nm. Total cholesterol (TC), triglycerides (TG), and high-density lipoprotein-

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cholesterol (HDL-C) were determined according to the procedure described by Allain *et al.* [17] Fossati and Prencipe [18] and Demacker *et al.* [19], respectively. Low-density lipoprotein-cholesterol and very Low-density lipoprotein-cholesterol were evaluated according to Friedwald's formula [20] by the following equations: LDL-C (mg/dl) = TC-(TG/5+HDL -C), vLDL (mg/dl) = TG/5. Hepatic tissues (100 mg tissue/ml buffer) were homogenized in 50 mM phosphate buffer (pH 7.2; St. Louis, MO, USA); the homogenates were then centrifuged at 1,200× g for 15 min and the supernatant was used for determination of malondialdehyde concentration (MDA) by the method of Yoshioka *et al.* [21], total antioxidant capacity (TAC) by Mahfouz *et al.* [22] and glutathione content (GSH) as described by Beutler *et al.* [23].

Statistical analysis

Results were presented as mean ± SE (n = 6). Experimental data were analyzed using one-way analysis of variance (ANOVA). Duncan's multiple range test was used to determine significant differences between means. Data were statistically analyzed with the aid of Statistical Package of the Social Sciences, SPSS version 25 (copyrighted by IBM SPSS software, United State Agency). Differences between means were considered significant at P<0.05.

RESULTS

The results in table 1 indicated that there was a significant elevation in the level of sugar content (glucose and fructose), total phenolic, and total flavonoid content of raw *F. carica* dried leaves by both irradiation sources. The percent elevation induced by gamma irradiation is higher than that induced by e-beam irradiation as follows; Glucose (17.43% and 11.9%, respectively), fructose (12.9% and 9.67%, respectively), TPC (33.5% and 23.5%, respectively) and total flavonoid (23.7% and 12.9%, respectively). Whereas the level of phytate and tannins reduced under the effect of gamma-radiation by 11.9% and 9.56%, respectively, and under the effect of e-beam by 7.44% and 7.13%, respectively compared to the non-irradiated sample. The DPPH scavenging activity of raw samples (75.49 ± 0.30%) was increased under the effect of gamma-irradiation and e-beam by 12.14% and 6.6 %, respectively (table 1).

As shown in table 2, the ethanol concentration for EtOH-rats treated with RFLP or GFLP dose (60 min after alcohol) has a statistically significant difference from that for the EtOH group (treated with alcohol only). The ethanol concentration was significantly decreased by RFLP by about 22.9-25.08 % from the 1st to 5th weeks compared to the EtOH group while its concentration was significantly reduced by GFLP by about 25.2-27.9% during the 5 weeks (experimental period).

Table 1. The phytochemical composition and DPPH scavenging activity of raw and irradiated F. carica dried leaves.

	Raw	Ionizing radiation (5 KGy)	
		Gamma-rays	Electron Beam
Glucose (g/100gm dw)	22.37±0.92 ^{c}	26.27±0.86 ^{a}	25.05±0.64 ^{b}
Fructose (g/100gm dw)	9.82±0.35 ^{c}	11.09±0.52 ^{a}	10.77±0.49 ^{b}
Total phenolic (mg GAE/g)	129±3.61 ^{c}	172.25±4.6 ^{a}	159.32±4.4 ^{b}
Total flavonoid (mg QUE/g)	42.74±2.4 ^{c}	52.87±2.3 ^{a}	48.27±2.5 ^{b}
Tannins (mg Tannic acid /g DW)	6.17±0.11 ^{a}	5.58±0.12 ^{c}	5.73±0.09 ^{b}
Phytate (mg/g DW)	3.36±0.05 ^{a}	2.96±0.07 ^{c}	3.11±0.06 ^{b}
DPPH scavenging activity (%)	75.49±0.30 ^{c}	84.66 ± 0.47 ^{a}	80.48±0.51 ^{b}

Values are means of three replicates (± SD). Values in the same row with different superscripts are significantly different at P<0.05. DPPH: 1,1-diphenyl-2-picryl hydrazyl

Table 2. Effect of raw and gamma-irradiated-dried fig leaves on blood ethanol level in rats from the 1st to 5th weeks.

Time (Week)	Ethanol Conc. (mg/100ml)				
	EtOH	EtOH-RFLP		EtOH-GFLP	
		Value	%Change	Value	%Change
1	384.9±7.6 ^{a}	296.6±5.6 ^{b}	-22.9%	281.4 ±4.9 ^{c}	-26.8%
2	692.3±6.8 ^{a}	521.1±6.2 ^{b}	-24.7%	499.1 ±5.7 ^{c}	-27.9%
3	884.5±7.4 ^{a}	694±6.9 ^{b}	-21.5%	662±6.2 ^{c}	-25.2%
4	1164±11 ^{a}	872±8.7 ^{b}	-25.08%	858±7.6 ^{c}	-26.2%
5	1273±13.5 ^{a}	965±10.1 ^{b}	-24.1%	943±9.2 ^{c}	-25.9%

EtOH: Ethanol; RFLP: raw fig leaves powder; GFLP: gamma-irradiated fig leaves powder. Values are expressed as means ± S.E. (n=7). Values in the same row with different superscripts are significantly different at P<0.05.

As observed from the results in this study, administration of EtOH to rats resulted in a significant elevation in the serum activities of AST, ALT, and GGT, serum level of TNF-α and IL-6, TC, TG, LDL-C, vLDL-C, and concentration of hepatic MDA associated with a remarkable decrease in the level of hepatic a TAC and GSH content when compared to the control group (tables 3-5).

Table 3. Effect of ethanol administration along with raw or irradiated-dried fig leaves on liver enzyme activity, TNF-α, and IL-6 levels in male rats.

	Control	EtOH	EtOH-RFLP	EtOH-GFLP
AST (U/ml)	35.65± 1.62^{d}	57.33± 2.82^{a}	47.27± 1.58^{b}	41.13± 1.67^{c}
ALT (U/ml)	31.52± 1.63^{d}	53.19± 1.78^{a}	40.37± 1.23^{b}	37.52± 1.15^{c}
γGT (U/ml)	5.47± 0.46^{d}	13.51± 0.73^{a}	8.26± 0.65^{b}	6.81± 0.39^{c}
TNF-α (pg/mL)	627.5± 17.3^{d}	915.6± 25.7^{a}	753.8 ± 23.4^{b}	701.9± 22.5^{c}
IL-6 (pg/mL)	323.7± 18.4^{d}	488.9± 21.8^{a}	403.5± 20.5^{b}	375.4± 18.6 ^{c}

EtOH: Ethanol; RFLP: raw fig leaves powder; GFLP: gamma-irradiated fig leaves powder; AST: aspartate transaminase; ALT: alanine transaminase, γGT: γ-glutamyl transferase; TNF-α: tumor necrosis factor-alpha; IL-6: interleukin-6. Values are expressed as means ± S.E. (n=7). Values in the same row with different superscripts are significantly different at P<0.05.

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Table 2. Statistical data of the radiographic parameters (kVp and mAs values) and patient anthropometric data for selected X-ray examinations.

	Control	EtOH	EtOH-RFLP	EtOH-GFLP
TC (mg/dl)	166.27± 6.72^{d}	237.39± 5.77^{a}	188.72± 7.28^{b}	172.90± 5.37^{c}
TG (mg/dl)	118.36± 3.46^{d}	179.92± 3.65^{a}	138.92± 3.32^{b}	129.97± 2.85^{c}
HDL-C (mg/dl)	50.41± 1.48^{a}	37.18±1.54^{d}	43.11± 1.46^{c}	46.01± 1.67^{b}
LDL-C (mg/dl)	92.21± 2.75^{d}	164.23± 3.36^{a}	117.91± 3.17^{b}	100.90 ±2.79^{c}
vLDL-C (mg/dl)	23.66± 1.68^{d}	35.98±1.72^{a}	27.70± 1.55^{b}	25.99± 1.62^{b}

Values are means of three replicates (± SD). Values in the same row with different superscripts are significantly different at P<0.05. DPPH: 1,1-diphenyl-2-picryl hydrazyl

On the other hand, treatment of EtOH-rats with either RFLP or GFLP resulted in a significant reduction in the serum activities of some liver enzymes, lipid profile contents (TC, TG, LDL-C,vLDL-C), lipid peroxidation (MDA) and level of inflammatory factors (TNF-α and IL-6) associated with an obvious elevation in level of hepatic a TAC and GSH content when compared to EtOH-group (tables 3-5).

DISCUSSION

The fruits and vegetables, rich in antioxidants and different micronutrients, defend against varied types of xenobiotics-induced hepatic injury and DNA damage [24]. Ethanol is a potent toxicant with the ability to disturb the cellular antioxidant defense system and induced damage to cell membranes [2]. This study was designed to evaluate the effect of gamma- and e-beam irradiation on some chemical contents of dried Fig leaves. As well as to study the hepatoprotective effect of irradiated-dried fig leaves against ethanol-induced hepatoxicity in rats.

The data in table (1) revealed that the sugar contents are about 32.19 g/100g, which makes dried fig a very energetic product. According to the study of Faleh *et al.* [25] the results in this article indicated that glucose and fructose were found to be the principal monosaccharides in the dried fig leaves. Veberic *et al.* [26] revealed that Fructose and glucose were found to be the dominant sugars in fig and sucrose was found at a very low level. The level of glucose and fructose content of dried fig leaf was significantly increased with gamma- and e-beam irradiation by 17.43%, 11.9%, 12.9%, and 9.67%, respectively. This increase in sugar content could be attributed to the degradation of higher polysaccharides such as starch and cellulose into lower monosaccharides due to the breaking of glycosidic bonds [27].

Also, the results cleared that dried fig leaf contains different amounts of total phenols (129 ± 3.61 mg GAE/g DW) and total flavonoids (42.74 ± 2.4 mg QUE/g). Osowe *et al.* [28] reported that *F. carica and F. exasperata* could be good sources of phenols when supplemented in the dietary intake of humans and livestock. In this study, gamma-irradiation (5 kGy) resulted in a higher increase in TPC (172.25±4.6 mg GAE/g DW) and total flavonoids (52.87±2.3mg QUE/g) than e-beam irradiation (159.32±4.4mg GAE/g DW and 48.27±2.5mg QUE/g, respectively). Hwang *et al.* [29] indicated that gamma rays were the most effective irradiation source for improving the antioxidant activity than electron beams and X-rays. Jamshidi *et al.* [30] reported that a noticeable increase in the TPC because of ionizing irradiation could be due to the breakdown of larger phenolic compounds into smaller ones and the liberation of phenolic compounds from glycosides. The elevation in the total flavonoids could be explained that the easy breakdown of active properties from the irradiation-degraded complex structures [31].

Also, the results revealed that the DPPH scavenging activity of the raw sample (75.49 ± 0.30%) was significantly elevated by gamma-irradiation (84.66 ± 0.47%) higher than its elevation by than e-beam irradiation (80.48 ± 0.51%). The results agreed with the results of Hwang *et al.* [29] who concluded that electron-beam irradiation was observed to be least effective at increasing the DPPH scavenging activity than gamma rays. Lee *et al.* [32] proposed that enhanced antioxidant activity was most likely due to the newly formed compounds produced by irradiation.

The obtained results (table 2) indicated that the *F. carica* dried leaf contains different amounts of anti-nutrition compounds such as phytate (6.17±0.11 mg/g DW) and tannins (3.36±0.05 mg Tannic acid /g DW) in the agreement with the results of Osowe *et al.* [32]. Phytates in food are known to bind with essential minerals such as calcium, iron, magnesium, and zinc in the digestive tract, resulting in mineral deficiencies [33]. Phytates in food are known to bind with essential minerals such as calcium, iron, magnesium, and zinc in the digestive tract, resulting in mineral deficiencies [33]. Tannins are plant polyphenols, which can form complexes with metal ions and with macro-molecules such as proteins and polysaccharides [34]. The level of phytate and tannins was significantly reduced by gamma-irradiation by a value higher than the reduction induced by e-beam irradiation. The decrease in tannin content by ionizing irradiation has been related to the generation of the hydroxyl and superoxide anion radicals induced chemical degradation [35]. The reduction in phytic acid during the radiation process might be due to the chemical degradation of phytate to the lower inositol phosphates and inositol by the action of free radicals produced by radiation or might be due to cleavage of the phytate ring itself [36].

From the results, it was observed that e-beam irradiation was least effective than gamma rays in increasing the antioxidant properties of dried fig

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leaves in the irradiated treatments. Thus, according to these observations, gamma-irradiated dried fig leaves were used to perform the biological experiment and study their efficacy against ethanol-induced hepatotoxicity.

The results obtained that the ethanol concentration of rats treated with RFLP or GFLP after 60 min of alcohol administration was significantly reduced during the experimental period compared to the EtOH group. This reducing effect could be due to the presence of fructose and glucose in fig leaves. Shi *et al.* [37] suggested that fructose and glucose could inhibit the absorption of ethanol in mice's gastrointestinal tract or enhance the elimination of ethanol in intoxicated mice. Keegan and Batey [38] found that carbohydrate supplementation (fructose or glucose) enhanced ethanol elimination without affecting alcohol dehydrogenase activity, indicating that a change in hepatic alcohol dehydrogenase was not the cause of the alterations in the rate of ethanol removal after carbohydrate intake. As a result of gamma-irradiated dried fig leaves containing a higher percentage of glucose and fructose, it had a more significant effect than raw fig leaves.

Administration of ethanol led to hepatotoxicity indicated by elevated serum levels of ALT, AST, and GGT in addition to disturbed oxidant-antioxidant status evidenced by elevated serum MDA and reduced level of hepatic TAC and GSH content when compared to the control group. These results agreed with Jang *et al.* [39] who indicated that chronic ethanol administration resulted in a clear hepatotoxicity as evidenced by the increased plasma AST and ALT and a significant decrease in the activities of the hepatic antioxidant enzymes SOD and CAT. Sallie *et al.* [40] revealed that the elevation in serum levels of AST, ALT, and ALP has been attributed to the damaged structural integrity of the liver because they are cytoplasmic in location and released into circulation after cellular damage.

In comparison with the control group, the serum level of TNF-α and IL-6, TC, TG, LDL-C, and vLDL-C was significantly increased by EtOH administration. Ganapathi *et al.* [41] reported that acetaldehyde (ethanol active metabolite) enhances the overproduction of free radicals, induction of oxidative stress, lipid accumulation, and inflammatory process that finally results in alcoholic liver hepatotoxicity.

On the other hand, treatment of EtOH-rats with either RFLP or GFLP resulted in significant reduction in serum activities of some liver enzymes, lipid profile contents (TC, TG, LDL-C, vLDL-C), lipid peroxidation (MDA) and level of inflammatory factors (TNF-α and IL-6) associated with an obvious elevation in concentration of HDL-C and level of hepatic a TAC and GSH content when compared to EtOH-group. The studies of Gilani *et al.* [42] denoted that dried *F. carica* contains alkaloids, flavonoids, coumarins, saponins, and terpenes that could be responsible for the radical-scavenging activity in alcohol toxicity. Tawfik and Alhejy [43] concluded that the presence of different phenolic and antioxidant compounds in figs in dried figs may enhance its ability in decreasing total cholesterol levels, low-density LDL cholesterol and triglycerides, reduced liver enzymes AST and ALT significantly and increased high-density cholesterol HDL. Vinson *et al.* [44] recommended adding more dried fig fruits and leaves that contain a lot of important antioxidants to the diet by nutrition experts to take advantage of the efficiency of these natural compounds in protecting against various diseases.

CONCLUSION

In this study, the gamma-irradiated dried fig leaves (5 kGy) were chosen to study their effect against liver damage caused by ethanol in male rats instead of using the dried fig leaves treated with e-beam, because the results of this study showed that irradiation processing by e-beam was least effective than gamma-rays in enhancing the antioxidant properties of dried fig leaves.

In addition, treatment of EtOH-rats with GFLP induced a higher significant hepatoprotective effect against EtOH damage than RFLP which could be attributed to the effectiveness of gamma-rays in increasing DPPH scavenging activity and the total phenolic and total flavonoids, as well as, decreasing the level of some anti-nutritional factor (Phytate and tannins) of dried fig leaf powder.

Statement of conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this article.

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