



ISSN:1110-0419

Original Article Vol. 62(4) (2024), 31 – 40

• 2023, Faculty of Agriculture, Benha University, Egypt.

DOI: 10.21608/assjm.2025.330402.1321



Effect of Chamomile Extract on Gamma Irradiated Rats

Zeinab H.H.Gad,², Enas Mahmoud Mekawi¹, Mohamed H.M. Abd el-Megid² and Salah M.M. Saad¹

1- Biochemistry Department, Faculty of agriculture, Benha University

2- Natural Products Department, National Center for Radiation Research and Technology(NCRRT), Egyptian

Atomic Energy Authority(EAEA).

* Corresponding Author: <u>dr.zenab2017@gmail.com</u>

Abstract

The chamomile is a good source of phenolic and flavonoid compounds that act as a natural antioxidant. This study was designed to investigate the role of chamomile (CH) against gamma radiation (six gray) induced hepatotoxicity and renal toxicity. Liver markers, kidney function, lipid profile and oxidative stress Enzymes, i.e. MDA, CAT, SOD, and GSH were measured after radiation. The obtained results showed that oral administration of chamomile extract (100mg /kg B.w) orally daily for four weeks , significantly decrements in the levels of transaminase enzyme in the serum of rats , as soon as significantly decrements in the total lipids ,triglyceride and total cholesterol levels in serum of rats. Also improvement in kidney function through decrements in the creatinine and urea levels in serum comparing with irradiated group. Significant decrements in total free radical and MDA in blood and Liver tissues comparing with irradiated group. The present study suggests that chamomile extract has an antioxidant effect to radio protective effect. hence, chamomile is considered as source of natural antioxidant against gamma radiation effects.

Keywords: gamma irradiation, chamomile, liver, kidneys, irradiated rats

Introduction

Ionizing radiation is considered as strong mutagenic and carcinogenic factor, as it produces various free radicals that caused oxidative stress when the rats are exposed to it. It lead to damage the vital compounds in different tissues inside the body, such as the DNA, lipids, protein, and carbohydrate, and damages the liver tissue. (*Xhuti e tal., 2023*). Medical diagnosis, radiotherapy, and various imaging protocols are common sources of radiation exposure. Due to oxidative stress coursed by the generation of reactive oxygen species (ROS) by gamma irradiation Multiple organs are damaged due to oxidative stress and inflammatory caused by the generation of reactive oxygen species (ROS) by γ -irradiation (*Kim et al., 2017*).

Chamomile belongs to the Compositae (*Asteraceae*) family (*Osman etal., 2016*). Since ancient times the plant gained international recognition as one of the most famous medicinal plants (*Park etal., 2017*). Chamomile (*Matricaria chamomilla*) it has been used for thousands of years in ancient Egypt, Rome and Greece as medicinal plant .The chamomile act as scavenging free radical and preventing the oxidative damage on cellular components due to it rich in flavonoids which are antioxidant effect (*Panche etal., 2016*). Chamomile flowers have been identify Over 120 metabolites

including fatty acid, amino acid, polysaccharides, flavonoids, terpenoids and phenolic derivatives. The bioactive chemical compounds in chamomile flowers are quercetin, lutein, a-bisabolol, apigenin and chamazulene (Srivastava etal., 2010). The chemical constituents include coumarins, flavonoids, terpenes, volatile oils, polysaccharides, organic acid and apigenin-7-O-glucoside is the one of the bioactive flavonoids in chamomile, the herb chamomile has been used to heal stomach problems, dermatitis, anticancer ,anti-infective, cramps dermatitis, antiinflammatory ,antithrombotic ,antioxidant hypolipidemic ,hypo-glycemia, antidepressant and neuroprotective (Yun et al., 2023). In traditional medicine, chamomile is one of the most important plants used to treat variety of human aliments such as rheumatism inflammation, and gastrointestinal disorder, as well as to boost the immune system (Amraei et al., 2015). The current study examined the effects of chamomile plant extract (100mg /kg as a natural antioxidant against gamma B.w) irradiated rats by dose six gray.

Materials and Methods

2.1. Materials

Chamomile flowers were obtained from the local market for dried herbal plant, Cairo, Egypt

All of the chemical used in these experiments came from Sigma and Aldrich chemical company of high quality and purity grad

Experimental Animals

Twenty eight male albino rats weighing between 150 and 170 grams were obtained from Faculty of Pharmacy Cairo University, Cairo, Egypt, the animals were kept for two weeks in animal house (Nuclear Research center Atomic Energy, Authority) and were kept on a standard condition of food and water for two weeks

2.2 Methods

2.2.1 Extraction of plants

The flowers of plants were cleaned by hand picking from foreign materials then the plants were crowded and filtered using Whatman NO.1 paper after being shacked overnight with five volume of 95% ethanol for twice repetition. The filtrates was subjected to vacuum distillation at 35C° to remove the solvent using rotary evaporator and obtain antioxidant extract according to (Spingo and Faveri 2007)

2. 2. 2 Determination of total phenolic compound

compounds Phenolic were determined according to (Singleton and Rossi 1965). Added one milliliter of extract to 5 mL distilled water then added 1 mL of folin-cocalteu reagent and 1 mL of sodium carbonate (20%). After 30 minutes of standing in the dark at room temperature, the absorbance was measured using spectrophotometer, at 765 nm UV/Vis spectrophotometer (SM1600 UVvis Spectrophotometers, Azzota, USA). Gallic acid equivalent was used to express the phenolic compound in extract

2. 2. 3 Determination of total flavonoids

Flavonoid were determined according (Meda et al., 2005). Using colorimetric method with aluminum chloride and expressed as quercetin equivalents, added one milliliter of dilute extract to one milliliter of 2% (w/v) ,methanolic AlCl₃, 100 mL 1 M CH₃ COOK, and 2.8 mL distilled water and then left at room temperature for half an hour. At 765nm absorbance was determined using a UV-vis Spectrophotometer (SM1600 UV-vis Spectrophotometers, Azzota, USA).

2. 2. 4 Determination of total antioxidant capacity

2. 2-diphenylpicrylhydrazyl (DPPH·) antiradical test was performed according to the (Blois, 1958) with small adjustment. A (0.004% w/v) in methanol, DPPH stock solution was prepared. After one mg mL⁻¹ of sample in methanol, 0.1 ml of diluted sample was mixed with 3.9 ml of the stock solution and vigorously shaken. After the solution was left in the dark for half an hour ,a UV-Vis spectrophotometer (SM1600 UV-vis Spectrophotometers, Azzota, USA) was used to measure the the absorbance at 517nm. Ascorbic acid was used as standard reference using against the absorbance of the DPPH. and calculated the % of DPPH. de-coloration follows as % of DPPH· de- coloration= $100 \times (A2 - A1/A2)$

Where A1 is the control absorbance and A2 is the sample absorbance.

2. 2. 5 Identification of phenols and flavonoid by HPLC

An agilent 1260 series was used to do HPLC analysis. The eclipse C18 column (4.6 mm x 250 mm i.d., 5 µm) was used to carry out the separation. The mobile phase consisted of water (A) and 0.05% trifluoroacetic acid in acetonitrile (B) at a flow rate 0.9 mL/min. The mobile phase was sequentially programmed using the following linear gradient as: 0 min (82% A); 0–5 min (80% A); 5-8 min (60% A); 8-12 min (60% A); 12-15 min (82% A); 15-16 min (82% A) and 16-20 (82% A). At 280 nm the multiwavelength detector was observed. For every sample solution, the injection volume was 5µL. The temperature of column was kept at 40 °C.

2.2.6.Irradiation

Whole-body of rats exposed to gammairradiation was performed at the National Centre for Radiation Research and Technology (NCRRT), Cairo, Egypt, using a 137Cs Gamma Cell-40 biological irradiator. Animals were exposed to fractionated doses of gamma-radiation, six gray (EL-Shennawy et al., 2016) dose of rat 1Gy/3min at the time of irradiation

2. 2. 7 Experimental Designs

Twenty-eight male albino rats weighing 150-180gm were used. Seven rats in each group

Group 1: (Negative group) un-irradiated rats fed on basal diet for four weeks period.

Group2: (positive control) irradiated rats (six gray) fed on basal diet for four weeks Period .

Group3: un-irradiated rats were fed on basal diet and received chamomile extract 100mg/kg B.w. orally daily for four weeks

Group4: Irradiated rats were fed on basal diet and received chamomile extract 100mg/kg B.w orally daily for four weeks

2.2.8 Blood collection

Samples of blood at two days and at the end of the experiment (four weeks) were taken using heparinized capillary tubes. Each sample was divided into two tubes. First tube contained EDTA for the measurement of total blood count. The second tubes were allowed to clot. serum was separated by centrifuging for 15 min at 3,000 rpm

2. 2. 9. Liver tissue collections

Samples of liver tissue at two days and at the end of the experiment (four weeks) were Anesthesia rats using diethyl ether were collected fresh for glutathione, catalase, superoxide measuring dismutase, malondialdehyde and total free radicals

2. 2. 10. Blood analysis

The biochemical analysis included liver enzyme activities of alanine transaminase (ALT), and aspartate transaminase (AST), were determined according to (Reitman and Frankel 1957) method, kidney functions i.e. creatinine was determined according (Bartels etal., 1972) and urea and was determined according to (Fawcett and Soctt 1960), lipid profile TL (total lipids), TG (triglycerid), and TC (total cholesterol), were determined according to (Zollner and Kirsch 1962), (Fossati and Prencipe. 1982) and (Richmond 1973). and (Allain et al., 1974). respectively, Free radical capacity in blood and tissue of liver (EPR Spectrometer) was determined according to(Gohn, 1986) and (Heckly, 1975). Malondialdehyde (MDA), catalase activity (CAT), superoxide activity (SOD) and glutathione dismutase concentration in serum and liver tissue were determined according to (Satoh, 1978) and (Ohkawa et al., 1979), (Aebi, 1984), (Nishikimi et al., 1972) and (Beutler etal., 1963) respectively 2. 2. 10. Statistical analyses

ANOVA was used for statistical analysis

using SPSS13.0 software, and all data were displayed as mean± *SD* (*SAS*, *1996*).

3. RESULTS A ND D ISCUSSION

3.1. Total phenolic compounds (TP), total flavonoid compounds (TF), and antioxidant activity of ehthanolic chamomile extract

The total phenolic compounds and total flavonoid compounds were determined in the ethanolic extract of chamomile and the results are illustrated in table (1). The total phenolic content

percentages equaled to (1.7%) and the total flavonoids was (1. 2%) these results were slightly lower than **Halimeh** *et al.*, *2020*, results small difference in total phenols and total flavonoids in chamomile between this study and (*Zahra et al.*, *2020*). The different results may be due to the different parts of plants were extracted.

The chamomile (CH) showed strong antioxidant capacity as illustrated in table (1) .the chamomile had (66.43 ± 3.48) % in ethanoic extracts. The polyphenolic and flavonoid contents that had high scavenging free radicals, as these compounds reduce ,and discolor DPPH through their ability to donate hydrogen. Their values were higher than the results obtained by (*Ghoniem et al., 2021*).

3.2 Identification of phenolic and flavonoid Compounds in chamomile extract by HPLC chromatography

Table (2) show that there were 15 phenolic compounds in chamomile ethanolic extract. The main identified compounds and their contents (μ g/mL) were rutin (239.88) followed by chlorogenic acid (101.56), daidzein (42.19) , naringenin (40.57), ferulic acid (33.70), cinnamic acid(14.58), apigenin (11.27), cafic (9.91), gallic acid (6.48), syringic acid (6.30), catechin (3.50), quercetin acid (2.26), vanillin (1.59), hesperetin (1.48), coumaric acid (1.46), The obtained results are in agreement with that (Janmejai and Sanjay 2009).

Table 1. total phenolic content(TP), total flavonoid content(TF) and antioxidant activity of chamomile extracts

phytochemical	Content
TP%	1.7±.09
TF%	1.2±.06
antioxidant activity DPPH ETH%) (66.43±3.48

Table 2. Phenolic and flavonoid compounds (μ g/mL) in chamomile extract analyzed by HPLC chromatography

Contents	Conc. (µg/ml)
Gallic acid	6.48
Chlorogenic acid	101.56
Catechin	3.50
Methyl gallate	0.00
Caffeic acid	2.26
Syringic acid	6.30
Pyro catechol	0.00
Rutin	239.88
Ellagic acid	0.00
Coumaric acid	1.46
Vanillin	1.59
Ferulic acid	33.70
Naringenin	40.57
Daidzein	42.19
Quercectin	9.91
Cinnamic acid	14.58
Apigenin	11.27
Kaempferol	0.00
Hesperetin	1.48

3.3 The Effect of the chamomile extract on liver function of irradiated rats

Data presented in Table 3 show a significant increment in liver enzymes (AST and ALT) levels in the gamma irradiated group treated with six gray after two days and four week after radiation comparing with normal control .The observed increment in serum AST and ALT in gamma irradiated animals in this study may potentially be attributed to the profound physiological impact of irradiation, resulting from either direct interaction between cellular membrane and gamma-rays or radiation-produced free through the action of radicals. Following radiation therapy, alterations in the enzymatic activities may result from the release of enzyme from radiosensitive tissues or from modifications in their synthesis, and they may also be

connected to the widespread destruction of the liver parenchyma ($El - Shahat \ et \ al., 2022$), Generation of free radical consequently, This raises the permeability of the cytoplasmic membrane to organic substance and result in cytosolic enzymes like ALT and AST leaks (*Weiss and Lander 2003*)

The obtained results of the impact of chamomile extract on liver enzymes in gamma irradiated rats and exhibited a significantly improved in liver enzymes levels (Table 3) comparting with control gamma irradiated rats .indicate that the plasma membrane has stabilized as repair of hepatic tissue injured caused by gamma radiation. The obtained results are in agreement with (Maryna et al., 2023)results.

Table 3. effect of the chamomile extract on live	r enzymes level in serum of irradiated rats
--	---

Treatment	AST U/mL		ALT U/m L	
groups	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	139.79±3.8	137.82±2.1 ^b	239.55±6.9	238.04 ± 4.03^{b}
Group (2) +ve control	220.88±2.8 ^a	471.90±7.5 ^a	285.67 ± 5.5^{a}	298.48±4.5 ^a
Group (3) un radiated rats (CH)	135.93 ± 2.2^{b}	136.92 ± 2.1^{b}	234.51 ± 4.2^{b}	236.08 ± 4.08^{b}
Group (4)irradiated rats+orally day chamomile extract (IR+CH)	221.99±5.0 ^a	144.58±2.2 ^b	283.55±3.2 ^a	245.83±4.1 ^b

a Significant change with -ve control <0.05, b significant change with +ve control <0.05 at the same time, CH chamomile extract, IR irradiated

3.4 The Effect of the chamomile extract on kidney function of irradiated rats

A significant increment were noted in serum urea and creatinine, levels in irradiated groups after two days and four weeks compartment with the normal control(Table 4). These result are in agreement with (El-Shahat etal., 2022). The results of this study showed that whole body gammairradiation of rats caused Asignificant increment in the level of serum urea and creatinine. The increment of urea may be due to addition- induced amino acids catabolism. In rats exposed to radiation, the increased rate of protein catabolism is accompanied by a decrease in total protein in the liver and an increase in the level of non-protein nitrogen in the liver and serum as well as increased levels of serum amino acids and ammonia which depends mainly on the protein destruction after irradiation.

Serum creatinine levels are a good indicator of total renal function and glomerular filtration rate. Mammals produce urine as a result of the breakdown of proteins, and the main component of blood is known to be urea. Examined the impact of chamomile extract on gamma radiation-induced damage to serum creatinine and urea levels. Therefore, as a result of lowering the level of creatinine and urea in the rats' serum and shielding the kidneys from oxidative stress caused by gamma irradiation shown as table 4. Significant reduction in urea and creatinine levelS in serum of rats that received chamomile extract, it is possible may be due to the oral administration of the chamomile extract playing a significant role in preventing kidney damage against gamma radiation. These results are in agreement with that reported by Maryna et al., (2023).

Table 4. Effect of the chamomile extract on creatinine and urea levels in serum of irradiated rats

Treatment	Creatinine mg/dL		Urea mg/dL	
Groups				
*	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	0.287 ± 0.006	0.285 ± 0.005^{b}	32.565±0.09	32.757 ± 0.52^{b}
Group (2) +ve control	0.772 ± 0.10^{a}	0.975 ± 0.015^{a}	60.301 ± 0.79^{a}	70.475 ± 1.1^{a}
Group (3) un radiated rats (CH)	0.263 ± 0.004^{b}	0.265 ± 0.005^{b}	31.049 ± 0.51^{b}	31.237 ± 0.49^{b}
Group (4)irradiated rats+orally day	0.770 ± 0.018^{a}	0.300 ± 0.007^{b}	62.327 ± 1.3^{a}	32.250 ± 0.51^{b}
chamomile extract (IR+CH)				

a Significant change with –ve control <0.05 ,b significant change with +ve control <0.05 at the same time, CH chamomile extract,IR irradiated

3.5 The Effect of the chamomile extract on lipid profile in serum of irradiated rats

The profile of serum lipids in normal, irradiated, and received chamomile extract rats is shown in Table 5. Serum TL, TC, and TG, levels were significantly increased in irradiated rats after two days and four weeks comparing with normal control. These results are in agreement with the result reported by Hamza et al., (2013).at has been noticed that radiation caused oxidative stress and free radical generation, which changes lipid metabolism and may be a major reason for hormonal imbalance that causes hypertriglyceridemia, hyperlipidemia, and hypercholesterolemia., moreover, a decrement in the activity of the enzyme cholesterol 7ahydroxylase and an increase in the activity of 3- hydroxyl methyl glutartl COA (HMG-CoA) reductase may be associated with hypercholesterolemia, as may increase the liver capacity for cholesterol biosynthesis. (Moussa et al., 2015) as well as It is possible that inhibition of the activity of lipoprotein lipase causes a decreases in adipose cell uptake of triglycerides, leading to hypertriglyceridemia. (El-Dein et al., 2016).

Such observed data in Table 5 revealed a noticeable protective property for the chamomile extract against gamma radiation damage for serum of total lipids ,triglyceride, and total cholesterol levels, therefore ,it could be concluded that the orally administration plant from chamomile played of the tested important role to decrease the total lipids triglyceride and total cholesterol levels in serum against gamma and protect tissues from oxidative stress from gamma irradiation. The obtained results are in agreement with that reported by *Irena et al.*, (2021).

Table 5. Effect of the chamomile extract on lipids profile in the serum of irradiated rats							
Treatment	_	Total lipids mg/o	đL	Triglycerides	mg/dL	Total cholester	ol mg/dL
Groups		Two days	Four weeks	Two days	Four weeks	Two days	Four weeks
Group (1) v control	/e	992.27±27.4	991.13±15.8 ^b	68.65±0.98	71.19±1.13 ^b	99.70±1.42	101.62 ± 1.62^{b}
Group (2) +ve control		1334.05±17.4 ^a	1440.26±22.9 ^a	91.25±1.18 ^a	95.19±1.35 ^a	121.31±1.57 ^a	131.18±2.03 ^a
Group (3) u radiated rats (CH)	IN	916.28±15.2 ^b	922.30±14.7 ^b	71.50±120 ^b	72.00±1.15 ^b	98.64±1.67 ^b	99.39±1.58 ^b
Group (4) Irradiated rats+ orally day chamomile		1334.69±29.6 ^a	982.40±15.6 ^b	90.30±2.07 ^a	72.51±1.15 ^b	122.37±2.82 ^a	102.94±1.64 ^b

a Significant change with -ve control <0.05 ,b significant change with +ve control <0.05 at the same time, CH chamomile extract,IR irradiated

3.6 Oxidative stress marker

extract (IR+CH)

3.6 .1 The effect of chamomile extract on free radical in irradiated rats

A six gray gamma irradiated caused significantly increment in total free radical in blood and liver tissues of rats after two days and four weeks as shown in Table 6. Production of free radicals may be due to gamma irradiation. When biological material is exposed to radiation, it quickly produces a large amount of reactive oxygen species (ROS), which are mostly produced by the ionizing molecules of water (Reisz etal., 2014). Oral administration of the chamomile extract dose 100mg/kg B.w for four weeks significantly reduced the concentration of total free radicals in both blood and liver tissue in irradiated rats. The accomplished results are in agreement with that reported by *Ismail et al.*, (2022)

```
Table (6) Effect of the chamomile extract on Free Radical in both blood and liver tissue of irradiated rats
```

Treatment	free radical in blood	$(\text{Radicals/g})^{\times 17}$	Free radical in	
Groups			tissue(Radicals/	$(g)^{\times 17}$
	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	1.74 ± 0.04	1.83 ± 0.02^{b}	5.41±0.14	5.78 ± 0.09^{b}
Group (2) +ve control	2.28 ± 0.03^{a}	3.45 ± 0.05^{ab}	$60.84{\pm}0.79^{a}$	40.57 ± 0.64^{ab}
Group (3) un radiated rats (CH)	1.77 ± 0.02^{b}	1.77 ± 0.02^{b}	5.64 ± 0.09^{b}	5.68 ± 0.09^{b}
Group (4)irradiated rats+orally day		1.82 ± 0.02^{b}	60.86 ± 1.36^{a}	6.27 ± 0.17^{b}
chamomile extract (IR+CH)	2.23 ± 0.05^{a}			

a Significant change with -ve control <0.05, b significant change with +ve control <0.05 at the same time, CH chamomile extract, IR irradiated

3.6 .2. The effect of chamomile extracts on malondialdehyde (MDA) on irradiated rats

Table 7 indicates that rats exposed to six gray gamma radiation showed a significant increment in (MDA) in their blood and liver tissue after two days and four weeks. The achieved results are in agreement with that reported by *Harry et al.*, (2023).

These results show a strong correlation between exposure to ionizing radiation and the lipid peroxidation level which is characterized by the formation of MDA .The final outcome of lipid peroxidation in the organism, whether by enzymatic or non-enzymatic processes, is malondialdehyde (MDA), i.e. aldehyde compound (*Jove et al., 2020*). High concentrations of MDA suggest that the process of oxidation occurs in the cell membrane.to defend against attacks by ROS (*Aranda et al.*, 2020).

As a result of increased concentration of MDA an imbalance between the free radical and antioxidant in body that manifests as oxidative stress. The oral administration of the chamomile extracts had to significant decrement (MDA) in serum and liver tissue of irradiated rats as shown in Table 7. This observation may be due to the natural antioxidant found in chamomile plants .The accomplished results are in consistent with those noted by *Dalia et al.*, (2021).

 Table 7. Effect of the chamomile extract on Malondialdehyde (MDA) for both serum and liver tissue of irradiated rats

Treatment	MDA in serum nmol/mL		MDA in liver tissue nmol/mg	
Groups	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	5.117±0.14	5.172±0.08 ^b	0.300±0.007	0.285 ± 0.005^{b}
Group (2) +ve control	11.914±0.15 ^a	9.635±0.15 ^{ab}	0.455±0.006 ^a	0.402±0.006 ^{ab}
Group (3) un radiated rats (CH)	5.272±0.17 ^b	5.312±0.14 ^b	0.295±0.005 ^b	0.297±0.004 ^b
Group (4)irradiated rats+orally day chamomile extract (IR+CH)	11.621±0.28 ^a	5.080±0.08 ^b	0.435±0.011 ^a	0.300±0.007 ^b

a Significant change with -ve control <0.05, b significant change with +ve control <0.05 at the same time, CH chamomile extract, IR irradiated

3.6 .3. The effect of chamomile extracts on superoxide dismutase (SOD) in irradiated rats

The exposed rats to six gray gamma irradiation have shown decrements significantly in SOD in serum and live tissue after two days and four weeks. The obtained results are illustrated in Table 8. The obtained result are agreement with the data noted by *Okeke et al.*, (2022). The observed decrement in the % inhibition of SOD may be connected to the existence of excess ROS. SOD activity declines are indicators of hepatocellular damage and continue to be the most sensitive biomarker of liver damage. These ROS interact with enzyme molecules causing their denaturation and partial inactivation

The observed improvement in the enzymes SOD in group received chamomile extract compared with irradiated group that oxidative stress caused by radiation were mitigated due to the antioxidant activity of phytochemical present in chamomile plant like flavonoids and polyphenols extract. Hence, chamomile extract mechanism of action to scavenging of free radicals may be due to the bioactive molecule hydrogen and regulate of endogenous enzymes. The accomplished results are in same line results obtained by *Ismail et al.*, (2022).

 Table 8 Effect of the chamomile extract on superoxide dismutase (SOD) in both serum and liver tissue of irradiated rats

Treatment	Superoxide Dism	utase (SOD) in	Superoxide Disn	nutase (SOD in
Groups	serum U/mL		liver tissue U/mg	tissue
	Two days	Four weeks	Two days	Four weeks
Group (1) –ve control	1561.00 ± 22.3	1606.07±25.6 ^b	1058.03 ± 23.5	1055.17 ± 16.8^{b}
Group (2) +ve control	1021.31±13.4 ^a	$1105.27 \pm 20.8^{\rm a}$	231.55 ± 2.9^{a}	647.70±10.3 ^a
Group (3) un radiated rats (CH)	1532.49±25.2 ^b	1541.77±24.6 ^b	1033.41±17.6 ^b	1041.78 ± 16.6^{b}
Group (4) irradiated rats +	1020.74 ± 22.1^{a}	1539.57±35.2 ^b	232.68±5.5 ^a	998.05±15.9 ^b
orally day chamomile extract				
(IR+CH)				

a Significant change with -ve control <0.05 ,b significant change with +ve control <0.05 at the same time, CH chamomile extract,IR irradiated

3.6 .4. The effect of chamomile extract on glutathione (GSH) in irradiated rats

A six gray gamma irradiated caused significantly reductions in (GSH) in serum and liver tissue of rats after two days and four weeks as shown in Table 9. The decrement in GSH concentration could be directly related to its use as an antioxidant that occurS in free radicals produced by radiation. The

achieved data are in agreement with that reported by *Okeke et al.*, (2022).

The observed advancement in the level of GSH in serum and liver tissue in irradiated rats has been occurred by chamomile extract. This improvement may be due to natural antioxidant productS (phenolic and flavonoid) in the chamomile extract which act as scavenging free radicalS. These results agree with the results obtained by Ismail *et al.*, (2022).

Table 9. Effect of the chamomile extract on Glutathione (GSH)) in both serum and liver tissue of irradiated rats

Treatment	Glutathione	(GSH) in serum	Glutathione (GS	H) in liver tissue
Groups	nmol/mL		nmol/mg	
	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	4.935±0.12	4.970 ± 0.08^{b}	0.670 ± 0.010	$0.710{\pm}0.010^{\rm b}$
Group (2) +ve control	3.144 ± 0.04^{a}	2.840 ± 0.04^{a}	0.265 ± 0.006^{a}	0.0400 ± 0.00^{a}
Group (3) un radiated rats (CH)	5.098 ± 0.16^{a}	5.135 ± 0.14^{ab}	0.751 ± 0.023^{ab}	0.755 ± 0.020^{a}
Group (4) irradiated rats + orally day	3.251 ± 0.07^{a}	5.172 ± 0.08^{ab}	0.256 ± 0.006^{a}	0.7500 ± 0.010^{a}
chamomile extract				
(IR+CH)				

a Significant change with -ve control <0.05, b significant change with +ve control <0.05 at the same time, CH chamomile extract, IR irradiated

3.6 .5. The effect of chamomile extracts on catalase (CAT) in irradiated rats

The gamma irradiation by six gray courses caused significantly decrements in the catalase enzyme activity in the serum and liver tissue of rats after two days and four weeks as shown in Table 10. This dropping in enzyme activity may be due to the oxidative stress caused by production of free radicals after irradiation. The enzyme is consumed to directly interaction with free radicals. These result are in agreement with that reported by by Okeke et al., (2022)

The observed improvement which is illustrated in Table (10) in enzyme catalase activity after treatment with chamomile extract orally for four weeks after irradiation has occurred by increasing significantly its activity in serum and liver tissue of irradiated rats . This improvement may be due to scavenging free radicalS by phenolic and flavonoid compound in chamomile act as antioxidant products.

Table 10. Effect of the chamomile extract on Catalasein both serum and liver tissue of irradiated rats

Treatment Groups	Catalase(CAT)	in serum mg/dL	catalase(CAT) mg/mg tissue	in liver tissues
	Two days	Four weeks	Two days	Four weeks
Group (1) -ve control	8.42 ±0.11	8.09 ± 0.11^{b}	14.71±0.19	14.9 ± 0.23^{b}
Group (2) +ve control	5.15 ± 0.06^{a}	$2.84{\pm}0.04^{a}$	9.29 ± 0.12^{a}	$7.00{\pm}0.11^{a}$
Group (3) un radiated rats CH)	7.93 ± 0.14^{b}	8.012 ± 0.12^{b}	14.56±0.25 ^b	14. 70±0.23 ^b
Group (4) irradiated rats + orally day chamomile extract (IR+CH)	5.15±0.13 ^a	8.012±0.12 ^b	9.30±0.23 ^a	13.66±0.80 ^b

a Significant change with -ve control <0.05 ,b significant change with +ve control <0.05 at the same time, CH chamomile extract,IR irradiated

Conclusion

From the abovementioned results, generally, it can be observed that the chamomile ethanolic extract contains high amount of phenolic and flavonoid compound. Consequently it has strong antioxidant activity which might have resulted from the phenolic components' synergistic effects. In the present study chamomile ethanolic extract showed antihyperlipidemic activity by acting as a potent reactive oxygen scavenger through its antioxidant compound, it also improved liver and kidney function by reducing free radical and increasing antioxidant enzymes activities when comparing with irradiated groups

Reference

- Aebi, H. (1984). Catalase in vitro, Methods in Enzymology volume 105 121-126
- Allain, C. C., Lucy, S. P., Cicely, S. G., Chan, W. R., and Paul, C. F. (1974). Enzymatic determination of total serum cholesterol, CLIN. CHEM. 20/4, 470-475 *Photobiology B: Biology* Volume 178, January, Pages 489-495

- Amraei, M., Bagrezaei, F., Taghinejad, H. and Mohamadpour, S. (2015). Hydroalcoholic extract of Matricaria chamomilla effectively reduces inflammation induced by xylene in rat. J Bas Res Med Sci. Aug.;2(2):30-6.
- Aranda, R., Cruz, G.A.K., Arancibia, H. A., Y.L., Hernandez, C. E.Y. and Pedraza, C. J. (2020). RONS and oxidative stress: An overview of basic concepts. *Oxygen*, 2, 437-78.
- Bartels, H., Böhmer, M., Heierli, C., (1972). Serum creatinine determination without protein precipitation. Clin Chim Acta 37:193-7
- Beutler, E. and Mary, K. Y. (1963). Erythrocyte glutathione reductase blood. VOL. 21, No. 5
- Blois, M. S. (1958). Antioxidant determination by the use of a stable free radical. *Nature*, *181*(4617), 11-99.
- Dalia, M. A. H. and Aziza, A. S. (2021). Amerolative Influence of chamomile (*Matricaria recutita L.*) on synthetic food additive induced probable toxicity in male albino rats. J. of Food and Dairy Sci., Mansoura Univ., Vol. 12 (7):161-170,
- **El-Dein, E., Anees, L.M. and Aly, S. (2016)**. Effects of a-lipoic acid on c-radiation and lindane-induced heart toxicity in rats. Pak J Zool. 48:1523–1529.
- El-Shahat, A. N., Refaat, G. H., Ashraf, M. M. and Madeha, N. A.(2022). Ameliorative effect of graviola fruit juice on the damaged tissues of gamma-irradiated male rats Pakistan J. Zool., vol. 54(2), pp 583-590
- El-Shennawy, H.M., El Shahat, A.N., Ahmed, A.G and Nahed, A. (2016). Ameliorative effect of silymarin against radiation- induced oxidative stress in the liver of male rats. Pakistan J. zool vol.48(6).pp.
- Fawcett, J.K. and Scott, J.E. (1960.) A rapid and precise method for the determination of urea. Journal of Clinical Pathology, 13, 156-159.
- Fossati, P. and Prencipe, L. (1982). Serum triglycerides determined calorimetrically with an enzyme that produces hydrogen peroxide. Clinical Chemistry, 28, 2077-2080.
- Ghoniem, A.A., Abd El-Hai, K.M.; El-khateeb, A.Y., Eldadamony, N.M. and Mahmoud, E. S.F. (2021). Enhancing the potentiality of trichoderma harzianum against pythium pathogen of beans using chamomile (*Matricaria chamomilla*, L.) flower extract molecules. 26, 1178
- Gohn, A; (1986).ESR and elementary particle applications. John, wiely. New Yourk,
- Halimeh, H., Vahid, N., Sedigheh, A.S. and Bahareh, H.(2020). Antioxidant activity and flavonoid content of *Matricaria Chamomilla* extracts from different. Populations of Iran *Journal of Botanical Research* | Volume 02 | Issue 02 | April

- Hamza, R.G., Mekawey, H.M.S. and El Shahat, A.N. (2013). Antioxidant activity of mulberry (*Morus alba L.*) fruits in male rats exposed to gammaradiation. Arab J Nucl Sci. 46:347–355.
- Harry, N. E., Surniyantoro, T. K., Devita, T., Darlina, Y., Iin Kurnia, H.B. and Yanti, L. (2023).Study of Immune response and malondialdehyde levels in irradiated rats supplemented with curcuma xanthorriza roxb extract Asian Pacific Journal of Cancer Prevention, Vol 24
- Heckly. R. J., Harry ,N. E. S., Teja,, K., Devita ,T. and Darlina Y. I. (1975). "Biological applications of electron spin resonance in free radicals in dry tissues. *Edited by: Swartz HM, Bolton. New York*: Wiley Inter ScienceJR, Borg DC: 5
- Irena, M., Hana, M., Martina ,H., Denisa ,M., Olena, O., Martin, P., Zuzana ,R., Ludmila, K. and Rostisla, V. (2021). The Combination of atorvastatin with silymarin enhances hypolipidemic, antioxidant and anti-Inflammatory effects in a rat model of metabolic syndrome. *Physiol. Res.* 70: 33-43,
- Ismail, Y., Fahmy, D.M., Ghattas, M.H., Ahmed, M.M., Zehry, W., Saleh, S.M. and Aboelmatty, D.M. (2022). Integrating experimental model, LC-MS/MS chemical analysis and systems biology approach to investigate the possible antidiabetic effect and mechanisms of matricaria aurea (golden Chamomile) in type 2 diabetes mellitus. front. Pharmacol. 13:924478. doi: 10.3389/fphar.2022.924478
- Janmejai, K. S. and Sanjay, G. (2009). Extraction, characterization stability and biological activity of flavonoids isolated from chamomile flowers Mol Cell Pharmacol. January 1; 1(3): 138.
- Jove, M., Mota, M. N., and Pradas, I. (2020). The advanced lipoxidation end-product malondialdehyde-lysine in aging and longevity. *Antioxidants (Basel)*, 9, 1-20.
- Kim, H.G., Jang, S.S., Lee, J.S., Kim, H.S. and Son, C.G. (2017). Panax ginseng meyer prevents radiation-induced liver injury via modulation of oxidative stress and apoptosis. J Ginseng Res 41:159–168. https:// doi.org/10.1016/j.jgr.2016.02.006
- Maryna, L., Valentyn, Y. and Victor, R. (2023). Influence of *inula helenium* rhizomes and *MatrIcaria chamomila* inflorescences on the biochemical and physiological parameters in male rats fed a high-fat diet. Rocz Panstw Zakl Hig;74(4):447-458
- Meda, A., Lamien, C. E., Romito, M., Millogo, J. and Nacoulma, O. G. (2005). Determination of the total phenolic, flavonoid and proline contents in Burkina Faso honey as well as their radical scavenging activity. *Food Chemistry*, 91, 571–577.

- Moussa, H.R., Mohammed, A.F.A., Osman, H.G. and Abd-Allah, M.A.N. (2015). Amelioration of oxidative stress induced by gamma irradiation in rats using Spirulina platensis. Eur J Acad Essays. 2:90–97.
- Nishikimi, M., Appaji, N. and Yagi, K. (1972). The occurrence of superoxide anion in the reaction of reduced phenazine methosulfate and molecular oxygen. Biochem. Biophys. Res. Commun., 46: 849-854
- Ohkawa, H., Nobuko, O. and Kunio, Y. (1979). Assay for lipid peroxides in animal tissues by hiobarbituric acid reaction. Analytical Biochemistry, 95, Issue 2, June, Pages 351-358
- Okeke, S.I., Agwu ,K.K., Orji, M.P, Akpa, A.U., Okeke, C.H., and Anaga, A.O. (2022). Radioprotective effects of ethanolic extracts of *Gongronema latifolium* leaf against radiation induced oxidative stress in wistar albino rats International. Journal of Radiation Research, Volume 20, No 1
- Osman, M. Y., Taie, H. A., Helmy, W. A. and Amer, H. (2016). Screening antioxidant, antifungal, and antitumor activities of aqueous extracts of chamomile (*Matricaria chamomilla*). *Egyptian Pharmaceutical Journal*,15 (2), 55-61.
- Panche, A., Diwan, A. and Chandra, S. (2016). Flavonoids: an overview. J nutrit sci. Dec;5 :e47. doi: 10.1017/jns.2016.41 152-160
- Park, E. H., Bae, W.Y., Eom, S. J., Kim, K. T. and Paik, H. D. (2017). Improved antioxidative and cytotoxic activities of chamomile (*Matricaria* chamomilla) florets fermented by Lactobacillus plantarum KCCM 11613P. Journal of Zhejiang University- SCIENCE B,18 (2),
- Reisz, J.A., Bansal, N., Qian, J., Zhao, W. and Furdui, C.M. (2014). Effects of ionizing radiation on biological molecules--mechanisms of damage and emerging methods of detection. Antioxid Redox Signal. PMID 10;21(2):260-92. doi: 10.1089/ars.2013.5489. Epub 2014 Feb 21.: 24382094; PMCID: PMC4060780.
- Reitman ,S. and Frankel, S. (1957). Colorimetric Assay. Amer. J. clin. Path. 28, 56
- Richmond W. (1973). Preparation and Properties of a cholesterol oxidase from nocardia sp. and Its application to the enzy-matic assay of total cholesterol in serum. Clinical Chemistry, 19, 1350-1356
- **SAS (1996).** SAS Procedure Guide. "Version 6.12 Ed". SAS Institute Inc., Cary, NC.

- Satoh K. (1978). Serum lipid peroxides in cerebrovascular disorders determined by a new colorimetric method. Clin Chim Acta 90.; 37-43
- Shennawy ,H.M., El Shahat, A.N., Ahmed, A.G.,and Nahed, A.(2016). Ameliorative effect of silymarin against radiation- induced oxidative stress in the liver of male rats . pakestan j. zool., vol 48(6),pp.1905-1909
- Singleton, V. L. and Rossi, J. A. (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American Journal of Enology and Viticulture*, 16, 144–158.
- Spingo, G. and faveri, D.M. (2007). Antioxidant from grape stalks and mare influence of extraction procedure on yield purity and antioxidant power of the extracts. J.of Food Eng, 78:793-801
- Srivastava, J.K., Shankar, E. and Gupta, S. (2010). Chamomile a herbal medicine of the past with a bright future. Mol Med Rep. Nov;3(6):895-901.
- Weiss, J.F. and Lander, M.R., (2003). Protection against ionizing radiation by antioxidant nutrients and phytochemicals. *Toxicology*, 189: 1-20. <u>https://doi</u>. org/10.1016/S0300-483X(03)00149-5
- Xhuti, D., Irena, A. R., Mahek, M., Linda, M., Kieran, M., Joshua, P. N. and Mark, A. (2023). Tarnopolsky the acute effect of multiingredient antioxidant supplementation following ionizing radiation. Nutrients 15, 207. https:// doi.org/10.3390/nu15010207
- Yun,L. D., Ying, L. , Wang, Q., Feng,J. N., Kun,W.L., Yun,Y. W., Jian, W., Chang,Z. Z. and LiNa, G. (2023). Chamomile: A Review of its traditional uses chemical constituents pharmacological activities and quality control studies .Molecules , 28(1), 133; https://doi.org/10.3390/molecules2801013 3
- Zahra A., Tooba, G., and Helia, H. (2020). Antiinflammatory effects of *Matricaria chamomilla* extracts on BALB/c mice macrophages and lymphocytes. Iran J Allergy Asthma Immunol 19(1):63-73. Doi: 10.18502/ijaai.v19i(s1.r1).2862 doi.org/10.3390/molecules26041178
- Zöllner, N. and Kirsch, K. (1962). Colorimetric method for determination of total lipids. Journal of Experimental Medicine, 135, 545-550

تاثير مستخلص الكاموميل علي جرذان التجارب المعامله بأشعه جاما زينب حامد حماد ²،ايناس محمود مكاوي ¹، مجد حسن عبد المجيد ² , صلاح مصطفي محمود سعد¹ 1- قسم الكمياء الحيويه -كليه الزراعه -جامعه بنها 2- قسم المنتجات الطبيعيه -مركز تكنولوجيا الاشعاع-هيئه الطاقه الذريه – القاهره

تهدف هذه الدراسه الي دراسه تأثير مستخلص الكاموميل علي تحسين وظائف الكبد والكلي المتضرره من تعرضها لاشعه جاما . تم تعريض جرذان التجارب الي اشعه جاما 6 جراي ثم اعطاءها مستخلص الكاموميل 100ملجم /كم من وزن الجسم . تم اخد عينات من دم جرذان التجارب بعد يومان و اربع اسابيع بعد التشعيع وتم تقدير وظائف الكبد والكي بروفايل الدهون وكذلك انزيمات الاكسده والشقوق الحره.وقد وجد ان عند مقارنه النتائج في المجموعه التي تناولت المستخلص بدون التعرض للاشعاع ليس هناك فروق معنويه عند مقارنتها بالمجموعه الضابطه الغير عند مقارنه النتائج في المجموعه التي تناولت المستخلص بدون التعرض للاشعاع ليس هناك فروق معنويه عند مقارنتها بالمجموعه التي تناولت المستخلص بدون التعرض للاشعاع ليس هناك فروق معنويه عند مقارنتها بالمجموعه الضابطه الغير مشععه , وقد ادي التشعيع الي زياده نشاط انزيمات الكبد ومستوي الكرياتينين واليوريا في السيرم وكذلك ارتفاع مستوي الدهون الكليه الجليسريدات الثلاثيه والكوليستيرول الكلي زياده معنويه مقارنه بالمجموعه الضابطه الغير مشععه وانزيمات الاكسده يقابله زياده كبيره في مستوي الشوق الحرد وكبين واليوريا في السيرم وكذلك ارتفاع مستوي الكرياتينين واليوريا في السيرم وكذلك ارتفاع مستوي الدهون الكليه الجليسريدات , وكانت الزياده في مالي يزياده معنويه مقارنه بالمجموعه الضابطه الغير مشععه وانزيمات الاكسده يقابله زياده كبيره في مستوي الشقوق الحره وكانت الزياده في النتيرول الكلي زياده معنويه مقارنه بالمجموعه الضابطه الغير مشععه وانزيمات الاكسده يقابله زياده كبيره في مستوي الشقوق الحره وكانت الزياده في النتيرول الكلي زياده معنويه مقارنه بالمجموعه الضابطه الغير مشععه وانزيمات الاكسده يقابله زياده كبيره في مستوي الشقوق الحره وكانت الزيادة ولي التائج بعد التشعيع بأربعه اسابيع اكبر منها بعد يومان , كذلك اظهرت النائية والكوليستيرول الكلي في الموي الاريات وكانت الزيادي والكوليستيرول الكلي في المجموعه التي تتاولت ، وكانت الزيادة وي الكرياتينين واليوريا في المجموعه الحموي الكرياتي نزمات الاكسده زياده ملحوظه وانخون تركيز الشقوق الحره مستخلص الكاموميل بعد تشعيعها مقارنه بالمجموعه الضابطه المشععه.وزاد نشاط انزيمات الاكسده زياده ملحوظه وانخفض تركيز الشقوق الحره مستخلص الكاموميل الكاموميل ولد الحمول المموميم ملحنا مالمتخلص الكاموميل ليحسين الاضرار النا