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Evaluating the Potential Effect of γ-Irradiated Dried Paprika Powder against Side Effects Induced by a High-Cholesterol Diet in Male Rats

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Key words
***Capsicum annuum*, Gamma-irradiation technology, Capsaicinoids, Aflatoxins**

ABSTRACT

The present study was conducted to investigate the effect of different doses of γ-irradiation treatment (0, 2, 4, 6 kGy) on total phenolic, capsaicinoids contents, microbial load, and aflatoxin content of dried paprika powder (*Capsicum annuum* L.). Also, to investigate the hypocholesterolemic effect of γ-irradiated paprika powder (GPP) against a high-cholesterol diet (HCD) in rats. The results of γ-irradiation processing (0, 2, 4, 6 kGy) showed that the higher the radiation dose, the higher the level of total phenols, capsaicin, dihydrocapsaicin and total capsaicinoid content. While there is a noticeable decrease in the aflatoxin's contents, total bacterial, yeast and mold load as the radiation dose increases. The results obtained in this study demonstrated that γ-irradiation at dose 6 kGy was proven to be the most effective dose than the lower doses. Supplementation of GPP with HCD for 10 weeks resulted in a reduction in the levels of some lipid contents (total cholesterol, triglycerides, low and very low-density lipoprotein-cholesterol), tumor necrotic factor-alpha and interleukin-6, leptin, glucose, insulin, HOMA-IR (homeostatic index of insulin resistance), lipogenic enzymes activity (glucose-6-phosphate dehydrogenase and malic enzyme), level of malondialdehyde and the activity of some hepatic markers associated with significant elevation in the level of high-density lipoprotein-cholesterol (HDL-C), glutathione content (GSH) and the activity of antioxidant enzymes (SOD and CAT) compared to rats fed with HCD only. In conclusion, the study elucidates that gamma-irradiation technology (6 kGy) could be one of the best solutions for the decontamination of dried paprika powder while retaining its quality and bioactive components, allowing commercial application as a public healthy food product. In addition, the study revealed that γ-irradiated paprika powder (GPP) possesses bioactive constituents that can be used in the treatment of insulin resistance, hypocholesterolaemia, inflammation, and oxidative stress.

INTRODUCTION

Spices are used in a variety of food preparations and are crucial to diets all over the world. Ripe paprika fruit (*Capsicum annuum* L.) is one of the most widely used spices in the world in the dried and powder forms. Owing to its' unique pungency, stimulating taste, colour, and flavour after drying, red paprika powder is used in ethnic foods to improve appetite. Red paprika are considered as a natural source of bioactive chemicals including volatile oils, capsaicinoids, vitamin C, protein, fibre, mineral elements, and antioxidants compounds such as flavonoids, phenolic acids, ascorbic acid, and carotenoids. In addition to its ability to boost immunity and protect against infections, cardiovascular diseases, decongest, weight loss, and cancer (Kim *et al*., 2017, 2020).

The bioactive component capsanthin (major carotenoid) in paprika can plays a protective role against lipid accumulation and avoid hypercholesterolemic disorders in the body as it absorbed into the body and distributed to plasma lipoproteins (Joo *et al*., 2021). Elevation of total cholesterol, LDL-cholesterol and accumulation of lipid enter the portal circulation and reach hepatocytes inducing inflammation and oxidative

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stress (Joo *et al*., 2021). Besides being a risk factor for cardiovascular diseases, hypercholesterolemia can also be harmful liver and different organs in the body (Ongaratto *et al*., 2021).

Paprika like the other spices can be contaminated by a wide range of microorganisms from the soil and windblown dust and by bird droppings and during cultivation, harvesting, storage, and distribution (Balakrishnan *et al*., 2022a). Presences of such microorganisms (bacteria, fungi, and viruses) can cause different diseases in the chili crop leading to reduce the yield of the plant and can be implicated in human illnesses and leading to mortality and morbidity in man. Although, there have been several microbial decontaminating methods, some of them have side effects including ethylene dioxide method could be source of carcinogens, the steam treatment induced degradation of product quality and some of methods could leave pesticide residues (Schottroff *et al*., 2021). Food irradiation is a hygiene process that used ionizing radiation for eliminating or reducing microorganisms and extending the shelf life of some food products without causing nutrient changes or forming harmful substances (Kim *et al*., 2020). The safety of γ irradiation technology has been approved by the World Health Organization (WHO), the United Nations Food and Agriculture Organization (FAO) and US-FDA as a cold sterilization process that can be used to decontaminate spices due to its high penetration into food components without degrading their quality (Balakrishnan *et al*., 2022a). The goal of this study was to evaluate the effectiveness of different doses of γ-irradiation (2, 4 and 6 kGy) on the microbial load, total phenolics, capsaicin, and dihydrocapsaicin of dried paprika powder (DPP). Also, this article aimed to study the hypo-insulinemic, hypo-cholesterolemic, anti-inflammatory and antioxidative activity of γ-irradiated paprika powder (GPP) against high-cholesterol diet (HCD).

MATERIALS AND METHODS

All experiments were carried out at the Egyptian Atomic Energy Authority (EAEA), Food Irradiation Research Department. Paprika powder (*Capsicum annuum* L.) was obtained from local market of spices, grains, and oils (Cairo, Egypt). Chemicals and reagents were purchased from Sigma Chemical Co. (St. Louis, MO, USA).

Gamma irradiation treatment

The Paprika powder was air packaged in polyethylene bags and treated with γ rays at the dose of 2, 4 and 6 kGy, using Indian Gamma Cell (Ge 4000 A)^{60} Co source at a dose rate of 0.8053 kGy/h at the National Centre for Radiation Research and Technology (NCRRT), Egypt. Alanine dosimeter (Traceable to Physical Lab. (UK) are used for measuring the absorbed irradiation dose.

Determination of total polyphenolic and total flavonoids

For quantification of total polyphenol content, the Folin-Ciocalteu's method was used (Singleton *et al*., 1999). The results of total polyphenol content were expressed as mg of gallic acid equivalents per ml of sample (mg GAE/ml) (Zaki *et al*., 2013).

Microbial studies

Total bacterial count (TBC) and yeast and mold count (YMC) were determined by the serial dilution method using pour plate technique (Aneja, 1996). The raw and gamma-irradiated paprika samples (100 g) were grinded in a food grinder without addition of any water or buffer. The homogenized sample of about 1 g was dissolved in 9 mL of 0.1% sterile peptone water. The solution was kept in stirring condition for about 30 min. This solution (1mL) was further diluted by dissolving in 9 mL of sterilized peptone water (0.1%). This way a dilution of 10−3 was obtained i.e., sample was diluted 1000 times followed by pour plating on nutrient agar and potato dextrose agar media to determine TBC and YMC, respectively. After that, the samples were incubated at 35 °C for 2 days for TBC and 30 °C for 4 days for YMC. The colonies so formed were counted and expressed as log cfu/g of sample.

Determination of aflatoxin

Aflatoxins were extracted and cleaned up according to the AOAC official method 2005-08 and LCTech sample preparation and analyses method. High-Performance Liquid Chromatography (HPLC) was used for the separation and quantification of aflatoxin (AFB_1, AFB_2, AFG_1 and, AFG_2,) (Richard, 2000).

Determination of capsaicin and dihydrocapsaicin

The major capsaicinoids (capsaicin and dihydrocapsaicin) in raw and gamma-irradiated paprika were determined by comparison to external reference standards injected under the same conditions. Their identification was based on the retention times measured under identical HPLC conditions while their quantitative determination in the different paprika samples was carried out using the peak areas (Nwokem *et al*., 2010).

Animal groups

Male albino rats Sprague Dawley (170 to 200g body weight (B. WT)) 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. Rats were acclimated to controlled

laboratory conditions for two weeks. Rats were maintained on rodent diet and tap water *ad libitum*. The HCD contained 100 g cholesterol, 30 g propyl thiouracil, and 100 g cholic acid in 1 L of peanut oil (Inoue *et al*., 1990).

Dose level of raw and gamma-irradiated paprika

According to the study of Kanki *et al*. (2003) the no-observed adverse effect level (NOAEL) of DPP was found to be 5% in the diet (0.67 g/rat/day or 2948.4 mg/kg bw/day for male rats).

Experimental design

The study was performed on the adult male rats divided in four groups. Each group comprised 7 animals.

Group 1 (Control group): Control rats fed with normal pellet diet for 10 weeks. Group 2 (HCD group): Rats fed with HCD for 10 weeks. Group 3 (HCD and RPP): Rats fed with HCD plus 5% raw paprika powder (RPP) for 10 weeks (Kanki *et al*., 2003). Group 4 (HCD and GPP): Rats fed with HCD plus 5% gamma-irradiated paprika powder GPP for 10 weeks.

At the end of the experiment (10 weeks), rats were fasted for 24 h and anaesthetized with diethyl ether. Blood samples were collected through heart puncture and allowed to coagulate and centrifuged for to obtain serum for biochemical analysis. Also, liver tissues were removed for biochemical investigation.

Biochemical analysis

Total cholesterol (TC), triglycerides (TG) and high-density lipoprotein-cholesterol (HDL-C) were determined according to the procedure described by Allain *et al*. (1974), Fossati and Prencipe (1982) and Demacker *et al*. (1980), respectively. Low-density lipoprotein-cholesterol (LDL-C) and very low-density lipoprotein-cholesterol (VLDL-C) were evaluated according to Friedwald's formula (Friedwald *et al*., 1972) by the following equations: LDL-C (mg/dl)= TC-(TG/5+HDL-C), vLDL (mg/dl) = TG/5. The activity of serum aspartate transaminase (AST) and alanine transaminase (ALT) was estimated according to Reitman and Frankel (1957), and serum γ-glutamyl transferase (GGT) was assessed according to Rosalki (1975). Detection of serum leptin, tumour necrotic 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. Serum samples were analyzed for glucose (Trinder, 1969) and insulin hormone was determined by radioimmunoassay kit supplied by Diasari, Italy. The homeostatic index of insulin resistance (HOMA-IR) was calculated using the equation: [fasting insulin concentration x fasting glucose concentration x 0.05551] /22.5 (Wallace *et al*., 2004).

The hepatic cytosolic and microsomal fractions were prepared using 0.25 M sucrose/0.5 M EDTA buffer (pH 7.4) and 1.15% KCl solution, respectively. The activities of glucose-6-phosphate dehydrogenase (G6PDH) (Baginski *et al*., 1974), malic enzyme (ME) (Ochoa, 1969), catalase (CAT) (Johansson and Borg, 1988), and superoxide dismutase (SOD) (Minami and Yoshikawa, 1979) were measured in the cytosolic fraction. The concentration of GSH was measured by Beutler *et al*. (1963) and malondialdehyde (MDA) was measured according to Yoshioka *et al*. (1979).

Statistical analysis

Results were presented as mean±SE (n= 7). Experimental data were analysed using one way analysis of variance (ANOVA). Duncan's multiple range test was used to determine significant differences between means. Data were statistically analysed by the aid of Statistical Package of the Social Sciences, SPSS version 25 (copyrighted by IBM SPSS software, USA). Differences between means were considered significant at $P < 0.05$.

RESULTS

The results tabulated in Table I indicated that the total phenols, capsaicin, dihydrocapsaicin and total capsaicinoid content were significantly ($p \leq 0.05$) increased with increasing irradiation doses and was higher in the samples irradiated at 6 kGy dose. The percentage change 7.3%, 10.39%, 9.98% and 9.3%, respectively from the raw sample (14.32 mg GAE/g, 981 mg/kg, 583.1 mg/kg and 1564.1 mg/kg, respectively).

The total bacterial and yeast and mold count Table I, in the control samples were 5.0±0.17 log cfu/g and 3.67 ± 0.11 log cfu/g, respectively. The results revealed that after the radiation treatment, the total bacterial and yeast and mold load decreased significantly ($p \leq 0.05$) in irradiated samples compared to control and was below the detectable levels at an irradiation dose range of 6 kGy. It was found that an average 14.69%, 25.93%, and 30.8 % reduction in aflatoxin was seen in the paprika sample for 2, 4, and 6 kGy, respectively.

As shown in the results under the influence of HCD a significant increase was observed in the serum level of some liver markers (γGT, ALT and AST), glucose, insulin ,homeostatic index of insulin resistance, lipid profile contents (TC, TG, LDL-C, vLDL-C), level of serum adipokines (leptin , TNF-α and IL-6), lipogenic enzyme activity (G6PDH and malic enzyme), and the level of hepatic MDA accompanied by significant reduction in the level of HDL-C, GSH and the activity of SOD and CAT compared to control group (Table II).

Table I. Effect of different doses of γ irradiation on total phenolic, capsaicinoids contents, microbial load (log cfu/g) and aflatoxins (AFB_1, AFB_2, AFG_1, and AFG_2) of *Capsicum annuum.*

Parameters		Radiation dose (kGy)			
		0	2	4	6
Total phenolic (mg GAE/g)		14.32 ± 0.55 [d]	14.56 ± 0.62 [c]	14.81 ± 0.69 [b]	15.37 ± 0.73 [a]
Capsaicin (mg/kg)		981 ± 9.0 [d]	1011± 9.8 [c]	1042 ± 12 [b]	1083 ± 13 [a]
Dihydrocapsaicin (mg/kg)		583.1 ± 5.5 [d]	604.4 ± 6.6 [c]	625.2 ± 6.8 [b]	641.3 ± 7.3 [a]
Total capsaicinoids (mg/kg)		1564.1 ± 15.5 [d]	1615.4 ± 16.3 [c]	1667.2 ± 16.7 [b]	1724.3 ± 19.7 [a]
Microbial load (log cfu/ g)	Total bacterial count	5.0 ± 0.17	3.0 ± 0.15	1.0 ± 0.11	ND
	Yeast mold count	3.67 ±0.11	2.96 ± 0.17	1.52 ± 0.18	ND
Aflatoxins	AFB1	16.55	15.23	13.52	13.07
	AFB2	1.57	1.69	1.31	0.89
	AFG1	1.03	0.93	0.45	0.39
	AFG2	3.59	1.57	1.56	1.38
Total aflatoxins (ppb)		22.74	19.42	16.84	15.73
% Reduction aflatoxins			14.69	25.93	30.8

Table II. Effect of raw and γ-irradiated paprika along with high cholesterol diet on lipid profile, serum adipokinase, leptin, lipogenic enzymes, hepatic markers, insulin, lipid peroxidation and oxidative stress markers in rats.

Parameters	Control	HCD	HCD+ RPP	HCD + GPP (6 kGy)
Lipid profile				
TC (mg/dl)	172.22± 5.83[d]	242.87±6.11[a]	195.11±4.86[b]	183.65±4.72[c]
TG (mg/dl)	119.16±3.79[d]	198.65±4.79[a]	146.72±3.78[b]	135.38±3.54[c]
HDL-C (mg/dl)	53.41±1.59[a]	40.22±1.38[d]	46.51±1.32[c]	48.95 ±1.29[b]
LDL-C (mg/dl)	94.98±3.15[d]	162.92±4.25[a]	119.26±3.91[b]	107.63±3.29[c]
vLDL-C (mg/dl)	23.83±1.73[d]	39.73±1.82[a]	29.34±1.59[b]	27.07±1.53[c]
Adipokinase				
TNF-α (pg/mL)	642.17 ± 21.3[d]	925.55 ± 31.2 [a]	772.45 ± 25.8 [b]	716.69 ± 23.5[c]
IL-6 (pg/mL)	320.45 ± 19.2[d]	491.98± 22.6 [a]	407.35± 20.1 [b]	375.31± 19.8[c]
Leptin, ng/mL	4.75 ± 0.29 [d]	9.86 ± 0.82 [a]	6.94 ± 0.34 [b]	6.17 ± 0.31[c]
Lipogenic enzymes				
G6PDH (µmole/min/mg hepatic protein)	1.97 ± 0.15 [d]	5.85 ± 0.37 [a]	2.35 ± 0.55 [b]	2.15 ± 0.26 [c]
ME (µmole/min/mg hepatic protein)	0.66 ± 0.05 [d]	1.36 ± 0.07 [a]	0.89 ± 0.03 [b]	0.78 ± 0.04 [c]
Hepatic marker				
AST (U/ml)	31.63±0.79 [d]	87.35±2.35 [a]	43.38±0.67 [b]	39.42±1.58 [c]
ALT (U/ml)	29.75±1.42 [d]	90.63±1.42 [a]	47.62±0.83 [b]	38.51±0.75 [c]
γGT (U/ml)	5.13±0.38 [d]	19.86±0.59 [a]	8.21±0.48 [b]	7.39±0.47 [c]
Glucose(mg/dl)	129.26 ± 7.43[d]	231.68 ± 9.72[a]	154.31 ± 7.21[b]	143.85 ± 7.11[c]
Insulin(µU/ml)	7.83 ± 0.52[d]	16.45 ± 0.61[a]	11.26 ± 0.54 [b]	10.35 ± 0.52 [c]
HOMA-IR	2.49 ± 0. 21[d]	9.40 ± 0.36 [a]	4.28 ± 0.24 [b]	3.67 ± 0.22 [c]
Oxidative stress markers				
MDA (n mol/g tissue)	219.86±3.21[d]	506.36±5.13[a]	348.72±4.53[b]	298.75±3.36[c]
GSH (mg/g tissue)	33.19±0.62[a]	18.67±0.54[d]	26.35±0.56 [c]	29.89±0.53 [b]
SOD (U/mg protein)	47.89±0.82[a]	26.53±0.73[d]	36.12±0.64[c]	40.96±0.70[b]
CAT (U/mg protein)	49.63±0.74[a]	30.26±0.62[d]	40.15±0.67[c]	43.93±0.63[b]

HCD, high cholesterol diet; RPP, raw paprika powder, GPP, gamma-irradiated paprika powder, G6PDH, glucose-6-phosphate dehydrogenase; ME, malic enzyme; TNF-α, tumour necrotic factor-alpha; IL-6, interleukin-6, HOMA-IR, homeostatic index of insulin resistance, was calculated as [fasting insulin x fasting glucose concentration x 0.05551]/22.5

Table II showed supplementation of HCD-rats with either RPP or GPP caused a significant reduction in the levels of γGT, ALT, AST, glucose, insulin, HOMA-IR, TC, TG, LDL-C, vLDL-C, lipogenic enzyme activity, leptin, TNF-α, IL-6, and MDA associated with significant elevation in the level of HDL-C, GSH and the activity of antioxidant enzymes (SOD and CAT) compared to rats fed with HCD (Table II).

DISCUSSION

Paprika (*Capsicum annuum* L.) is a rich source of phenols, flavonoids, carotenoids and capsaicinoids (capsaicin and dihydrocapsaicin) that can be used to prevent oxidative damage and myocardial disease. However, the quality of paprika is reduced during drying due to the cross-contamination of microorganisms, insects, and dirt. Gamma irradiation is widely used as a non-thermal, safe, and most effective method for the reduction of spoilage and pathogenic aerobic microorganisms without altering quality criterion (Ayob *et al*., 2021).

In this study, the results revealed that the levels of total phenolic contents and capsaicinoids contents of paprika powder were significantly increased as the radiation dose increase. The increased with radiation treatment was higher in the samples irradiated at 6 kGy dose. The significant elevation in the total phenolic contents could be due to irradiation induces breaking or degradation of larger phenolic compounds into smaller ones and there is also the release of phenolic compounds from glycosidic compounds (Hussain *et al*., 2019). Also, the results obtained that main capsaicinoids analyzed during the study were dihydrocapsaicin and capsaicin and the total capsaicinoids were expressed as a sum of these two pungent components (Ayob *et al*., 2021). The positive correlation between gamma-radiation and the level of dihydrocapsaicin and capsaicin could be attributed to the change in conformation of molecules along with nei'ghboring compounds when subjected to gamma irradiation doses and thus, leading to increase in extraction efficiency. The results in agreement with the results of Iqbal *et al*. (2016) who observed a significant increase in capsaicin and dihydrocapsaicin contents of hot pepper by 4.74% and 4.37%, respectively after gamma-radiation at 6 kGy.

During this study, an obvious reduction has been observed in the total bacterial and yeast and mold load and aflatoxins content (AFB_1, AFB_2, AFG_1, and AFG_2) in irradiated samples compared to control and total bacterial and yeast and mold load was below the detectable levels at an irradiation dose of 6 kGy in agreement with the results of Edae *et al*. (2022). Hussain *et al*. (2019) suggested that the reduction in the total viable microbial cells by γ-irradiation can be related to the ability of gamma rays to break the hydrogen bonds in DNA that are responsible for maintaining the double helical structure of DNA. Therefore, any break in the hydrogen bond leads to complete destruction of DNA and inability of cells to replicate, causing microbial cell death.

Aflatoxins mainly produced by some strains of *Aspergillus flavus*, *Aspergillus parasiticus*, and *Aspergillus nomius* molds are highly toxic and carcinogenic substance that degrades paprika quality and render it unfit for consumption (Balakrishnan *et al*., 2022b). γ radiation has been studied for its efficiency in destroying mycotoxins in agricultural produce and processed foods. Iqbal *et al*. (2013) concluded that aflatoxin contamination decreased as the radiation dose was increased, even though complete removal for aflatoxin was not attained. Balakrishnan *et al*. (2022b) explained that the reduction in aflatoxin by γ radiation can be directly by acting on aflatoxin producing molds under specific conditions to eliminate toxin production, and indirectly by eradicating mold survival which leads to the avoidance of aflatoxin producing fungal growth.

In the present study, the results obtained that the gamma-irradiation of dried paprika powder at dose of 6.0 kGy had significant effect in terms of increasing the total phenolic and capsaicinoids contents and decreasing the total bacterial and yeast and mold load while decreased aflatoxins content. Therefore, GPP at a dose of 6.0 kGy were used to conduct the biological experiment and study its hypo-cholesterolemic effect against HCD.

The results of biological study showed that feeding on HCD induced a significant elevation in lipid profile, lipogenic enzyme activity (G6PDH and malic enzyme), hepatic lipid peroxidation and level of some hepatic markers along with reduction of HDL-C serum levels, GSH content and the activity of hepatic antioxidant enzymes (SOD and CAT) compared to control group. Zălar *et al*. (2022) reported that hyperlipidaemia, along with high LDL-C and low HDL-C serum levels, are key factors in the incidence of coronary heart disease, morbidity, and cerebrovascular mortality. With respect to the study of Adekunle *et al*. (2013), the elevation of serum levels of LDL-C and vLDL-C could be related to transporting of more cholesterol and triglyceride are being transported from the liver to the extra-hepatic tissues. Li *et al*. (2011) suggested that long-lasting high-fat diet leads to lipid metabolism disturbance and hyper-lipidemia by reducing lipid metabolic enzymes such as hepatic lipase and lipoprotein lipase. Xu *et al*. (2004) has also reported that the impaired hepatic lipid homeostasis because of lipid accumulation attributed to the increasing activity of

the enzymes involved in fatty acid biosynthesis in the rats by the dietary cholesterol.

Hypercholesterolemia could increase the hepatic lipid peroxidation by activation of β-adrenergic receptors that could increase lipolysis to yield free fatty acids that are able to uncouple the mitochondrial phosphorylation and further generate free radicals resulted in oxidative damage (Bhandari *et al*., 2011). This oxidative damage leads to hepatic tissue damage causing the release of liver enzymes to the plasma and occurrence of hepatic dysfunction (Dikshit *et al*., 1995). HCD also resulted in hyperinsulinemia, elevation of level of serum adipokines (leptin, TNF-α and IL-6) and insulin resistance which is in agreement with Jung *et al*. (2016) who indicated that abnormal lipid metabolism in obesity is a major cause of dyslipidaemia, insulin resistance, inflammation, and hepatic steatosis. Bhandari *et al*. (2011) reported that leptin might contribute to hepatic steatosis by promoting insulin resistance and by altering insulin signalling in hepatocytes, to promote increased intracellular fatty acids.

Group of rats fed on HCD with either RPP or GPP exhibited significant reduction in the serum lipid profile, glucose and insulin level, insulin resistance, adipokine secretion, lipogenic enzyme activity (G6PDH and malic enzyme), hepatic lipid peroxidation and level of some hepatic markers associated with enhancement of antioxidant status and HDL-C compared to HCD-group. Kim *et al*. (2017) suggested that red paprika and capsanthin have anti-obesity and hepatoprotective capabilities and are effective in ameliorating hepatic steatosis and adipogenesis induced by a high-fat diet in mice. The red pigmentation of paprika fruits (*Capsicum annuum* L.) is related to presence of capsanthin which considered as one of the most powerful antioxidants and has high activity for scavenging free radicals (Kennedy *et al*., 2021). They added that capsanthin belongs to class of carotenoids called xanthophylls that can participate in the primary defence mechanism of HDL against oxidative stress and may also be expected to affect lipid metabolism and/or maintain favourable blood lipid profiles. Several studies revealed that ingestion of paprika, which possesses abundant carotenoids and capsanthin, may modulate lipid metabolism by improving the HDL-C concentrations and alter adipocytokine levels or hepatic gene expression (Jeyakumar *et al*., 2007; Hussein *et al*., 2007).

Kim *et al*. (2017) concluded that the red paprika and capsanthin treatments against high fat diet ameliorated the dysregulation of serum adipokines (leptin, adiponectin, plasminogen activator inhibitor-1 (PAI-1), TNF-α and IL-6) and significantly decreased the activities of the hepatic enzymes involved in FA and TG synthesis (G6PDH and ME). In addition, the red paprika and capsanthin treatments significantly reduced the mRNA expression of leptin and resisting in epididymal white adipose tissue. On the other hand, Chen *et al*. (2017) observed that hypolipidemic effects of paprika may be attributed to its ability to decrease the serum level of TC, LDL-C, and TG, meanwhile increasing serum HDL-C and inhibition of lipid synthesis via suppressing the expression of 3-hydroxy-3-methylglutaryl coen-zyme A(HMG-CoA), cholesterol 7-hydroxylase (CYP7A1) and fatty acid synthetase (FAS).

CONCLUSION

The study's findings demonstrate that gamma-irradiation technology is an effective treatment in microbial decontamination of dried paprika powder while preserving its quality and bioactive components, enabling for its commercial use as a healthy food content. However, this irradiation dose (6 kGy) was not effective in complete reduction of aflatoxins. Finally, the results concluded that GPP exhibited higher significant hypo-insulinemic, hypo-cholesterolemic, anti-inflammatory and antioxidative activity against HCD than RPP and that could be due to the useful effect of gamma-irradiation in enhancing the effectiveness of paprika powder evidenced by increasing in the total phenolic and capsaicinoids contents and decreasing the total bacterial and yeast and mold load and aflatoxins content.

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Ethical statement

Ethical consideration: All animal procedures were carried out following the Research Ethics Commitee for experimental studies (Human and Animal subjects) the National Centre for Radiation Research and Technology (REC-NCRRT), Egyptian Atomic Energy Authority (Cairo, Egyp). Conformed to the ClOMS and ICLAS International Guiding Principles for Biomedical Involving Animals 2012.

Generative AI and AI-assisted technology statement

The authors have declared that no generative AI or AI-assisted technologies were used to create this manuscript.

Statement of conflict of interest

The authors have declared no conflict of interest.

REFRENCES

Adekunle, A.S., Adedeji, A.L., Oyewo, E.O., Adedosu, O.T. and Omotoso, A.T., 2013. Hyperlipidemia induced by atherogenic diet enhanced oxidative stress in the kidney and inflammatory responses: An *in-vivo* study. *Asian J. Nat. Appl. Sci*., **2**: 82-93.

Allain, C.C., Poon, L.S., Chan, C.S., Richmond, W. and Fu, P.C., 1974. Enzymatic determination of total serum cholesterol. *Clin. Chem*., **20***:* 470. https://doi.org/10.1093/clinchem/20.4.470

Aneja K.R., 1996. *Experiments in Microbiology, Plant Pathology, Tissue Culture and Mushroom Cultivation*. 2nd Ed., Wishwa Prakashan, New Delhi, pp. 130-132.

AOAC, 1995. *Official methods of analysis (16th ed.).* Association of Official Analytical Chemists, Arlington VA, USA.

Ayob, O., Hussain, P.R., Suradkar, P. and Naqash, F., 2021. Gamma irradiation and storage effects on quality and safety of Himalayan paprika (Waer). *LWT Fd. Sci. Technol*., **147**: 111667. https://doi.org/10.1016/j.lwt.2021.111667

Baginski, E.S., Foa, P.P. and Zak, B., 1974. Glucose-6-phosphatase. In: *Methods in enzymatic analysis* (ed. H.U. Bergermeyer). Academic Press, New York. pp. 867–880. https://doi.org/10.1016/B978-0-12-091302-2.50072-4

Balakrishnan, N., Yusop, S.M., Rahman, I.A., Dauqan, E. and Abdullah, A., 2022a. Efficacy of gamma irradiation in improving the microbial and physical quality properties of dried chillies (*Capsicum annuum* L.): A review. *Foods*, **11**: 91. https://doi.org/10.3390/foods11010091

Balakrishnan, N., Yusop, S.M., Rahman, I.A., Dauqan, E. and Abdullah, A., 2022b. Effect of gamma radiation and storage time on the microbial and physicochemical properties of dried Byadgi chili (*Capsicum annuum*). *Agriculture*, **12**: 639. https://doi.org/10.3390/agriculture12050639

Beutler, E., Duron, O. and Kelly, B.M., 1963. Improved method for the determination of blood glutathione. *J. Lab. Clin. Med*., **61**: 882-888.

Bhandari U., Kumar V., Khanna N. and Panda B.P., 2011. The effect of high-fat diet-induced obesity on cardiovascular toxicity in Wistar albino rats. *Hum. exp. Toxicol*., 30: 1313-1321. https://doi.org/10.1177/0960327110389499

Chen, X., Ding, Y., Song, J. and Kan, J., 2017. Hypolipidaemic effect and mechanism of paprika seed oil on Sprague–Dawley rats. *J. Sci. Fd. Agric*., **97**: 4242–4249. https://doi.org/10.1002/jsfa.8300

Demacker, P.N., Vos-Janssen, H.E., Hifmans, A.G.M., VaṅtLaar, A. and Jansen, A.P., 1980. Measurement of high- density lipoprotein cholesterol in serum: Comparison of six isolation methods combined with enzymatic cholesterol analysis. *Clin. Chem*., **26**: 1780. https://doi.org/10.1093/clinchem/26.13.1780

Dikshit, M., Rastogi, L., Shukla, R. and Srimal, R.C., 1995. Prevention of ischaemia-induced biochemical changes by curcumin and quinidine in the cat heart. *Indian J. med. Res*., **101**: 31–35.

Edae, B.N., Tufa, M.B. and Muche, A.B., 2022. Effect of gamma radiation on the reduction of aflatoxin in red pepper. *J. Fd. Res*., **11**: 34-39. https://doi.org/10.5539/jfr.v11n4p34

Fossati, P. and Prencipe, L., 1982. Serum triglycerides determined calorimetrically with an enzyme that produce hydrogen peroxide. *Clin. Chem*., **28**: 2077. https://doi.org/10.1093/clinchem/28.10.2077

Friedwald, W.T., Levy, R.I. and Fredrickson, D., 1972. Estimation of concentration of low-density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. *Clin. Chem*., **18**: 499. https://doi.org/10.1093/clinchem/18.6.499

Hussain, P.R., Rather, S.A., Suradkar, P.P. and Ayob, O., 2019. Gamma irradiation treatment of quince fruit (*Cydoniaoblonga* mill): Effect on postharvest retention of storage quality and inhibition of fungal decay. *J. Radiat. Res. appl. Sci*., **12**: 118–131. https://doi.org/10.1080/16878507.2019.1618588

Hussein, G., Nakagawa, T., Goto, H., Shimada, Y., Matsumoto, K., Sankawa, U. and Watanabe, H., 2007. Astaxanthin ameliorates features of metabolic syndrome in SHR/NDmcr-cp. *Life Sci*., **80**: 522–529. https://doi.org/10.1016/j.lfs.2006.09.041

Inoue, Y., Goto, H., Horinuki, R., Kimura, Y. and Toda, T., 1990. Experimental atherosclerosis in the rat carotid artery induced by balloon de-endothelialization and hyperlipidemia. *J. Jpn. Atheroscler. Soc*., **18**: 1147–1154. https://doi.org/10.5551/jat1973.18.12_1147

Iqbal, S.Z., Bhatti, I.A., Asi, M.R., Zuber, M., Shahid, M. and Parveen, I., 2013. Effect of irradiation on fungal load and aflatoxins reduction in red chillies. *Radiat. Phys. Chem*., **82**: 80–84. https://doi.org/10.1016/j.radphyschem.2012.09.015

Iqbal, Q., Amjad, M., Asi, M., Nawaz, A., Khan, S., Aríno, A. and Ahmad, T., 2016. Irradiation maintains functional components of dry hot peppers (*Capsicum annuum* L.) under ambient storage. *Foods*, **5**: 63. https://doi.org/10.3390/foods5030063

Jeyakumar, S.M., Vajreswari, A. and Giridharan,

N.V., 2007. Impact of vitamin A on high-density lipoprotein-cholesterol and scavenger receptor class BI in the obese rat. *Obesity* (Silver Spring), **15**: 322–329. https://doi.org/10.1038/oby.2007.534

Johansson, L.H. and Borg, L.A.H., 1988. A spectrophotometric method for determination of catalase activity in small tissue samples. *Anal. Biochem*., **74**: 331. https://doi.org/10.1016/0003-2697(88)90554-4

Joo, H.K., Lee, Y.R., Lee, E.O., Kim, S., Jin, H., Kim, S., Lim, Y.P., An, C.G. and Jeon, B.H., 2021. Protective role of dietary capsanthin in a mouse model of nonalcoholic fatty liver disease. *J. Med. Fd*., **24**: 635–644. https://doi.org/10.1089/jmf.2020.4866

Jung, U.J., Cho, Y.Y. and Choi, M.S., 2016. Apigenin ameliorates dyslipidemia, hepatic steatosis, and insulin resistance by modulating metabolic and transcriptional profiles in the liver of high-fat diet-induced obese mice. *Nutrients,* **8**: 305. https://doi.org/10.3390/nu8050305

Kanki, K., Nishikawa, A., Furukawa, F., Kitamura, Y., Imazawa, T., Umemura, T. and Hirose, M., 2003. A 13-week subchronic toxicity study of paprika color in F344 rats. *Fd. Chem. Toxicol*., **41**: 1337–1343. https://doi.org/10.1016/S0278-6915(03)00136-4

Kennedy, L.E., Abraham, A., Kulkarni, G., Shettigar, N., Dave, T. and Kulkarni, M., 2021. Capsanthin, a plant-derived xanthophyll: A review of pharmacology and delivery strategies. *AAPS Pharm. Sci. Tech*., **22**: 203. https://doi.org/10.1208/s12249-021-02065-z

Kim, H.K., Jeong, J., Kang, E.Y. and Go, G.W., 2020. Red pepper (*Capsicum annuum* L.) seed extract improves glycemic control by inhibiting hepatic gluconeogenesis via phosphorylation of FOXO1 and AMPK in obese diabetic db/db mice. *Nutrients*, **12**: 2546. https://doi.org/10.3390/nu12092546

Kim, J.S., Ha, T.Y., Lee, S.K.S.J. and Ahn, J., 2017. Red paprika (*Capsicum annuum* L.) and its main carotenoid capsanthin ameliorate impaired lipid metabolism in the liver and adipose tissue of high-fat diet-induced obese mice. *J. Funct. Fds*., **31**: 131–140. https://doi.org/10.1016/j.jff.2017.01.044

Li, C., Xin-Bo, M., Yu-Hong, L., Shi-Cheng, P., Yi-Ping, F. and Min, W., 2011. Effects of persimmon leaf total flavonoid on enzyme of lipoprotein metabolism and antioxidation in hyperlipidemia rats. *Chin. J. Natu. Med*., **9**: 74. https://doi.org/10.1016/S1875-5364(11)60024-1

Minami, M. and Yoshikawa, H., 1979. A simplified assay method of superoxide dismutase activity for clinical use. *Clin. Chim. Acta*, **92**: 337-342. https://doi.org/10.1016/0009-8981(79)90211-0

Nwokem, C.O., Agbaji, E.B., Kagbu, J.A. and Ekanem, E.J., 2010. Determination of capsaicin content and pungency level of five different peppers grown in Nigeria. *N. Y. Sci. J*., **3**: 17-21.

Ochoa, S., 1969. Malic enzyme. In: *Methods in enzymology* (ed. J.M. Lowenstein). Academic Press, New York. pp. 230–237. https://doi.org/10.1016/0076-6879(69)13042-6

Ongaratto, M.A., Endres, M. and Barschak, A.G., 2021. Modelling hypercholesterolaemia in rats using high cholesterol diet Luiza Ferracini Cunha. *Int. J. exp. Path*., **102**: 74–79. https://doi.org/10.1111/iep.12387

Reitman, S. and Frankel, S., 1957. A calorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am. J. clin. Pathol.*, **28**: 56. https://doi.org/10.1093/ajcp/28.1.56

Richard, J., 2000. *Romer labs guide to mycotoxins, vol. 1. Mycotoxins an overview*. Romer Labs, Inc., Union, MO.

Rosalki, S.B., 1975. Gamma-glutamyl transpeptidase. *Adv. clin. Chem*., **17**: 53–107. https://doi.org/10.1016/S0065-2423(08)60248-6

Schottroff, F., Lasarus, T., Stupak, M., Hajslova, J., Fauster, T. and Jager, H., 2021. Decontamination of herbs and spices by gamma irradiation and low-energy electron beam treatments and influence on product characteristics upon storage. *J. Radiat. Res. appl. Sci*., **14**: 380–395. https://doi.org/10.1080/16878507.2021.1981112

Singleton, V.L., Orthofer, R. and Lamuela-Raventos, R.M., 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods Enzymol*., **299**: 152-178. https://doi.org/10.1016/S0076-6879(99)99017-1

Trinder, P., 1969. Determination of blood glucose using 4-amino phenazone as oxygen acceptor. *J. clin. Pathol*., **22**: 246. https://doi.org/10.1136/jcp.22.2.246-b

Wallace, T.M., Levy, J.C. and Matthews, D.R., 2004. Use and abuse of HOMA modeling. *Diabetes Care*, **27**: 1487–1495. https://doi.org/10.2337/diacare.27.6.1487

Xu, G., Pan, L.X., Li, H., Shang, Q., Honda, A., Shefer, S., Bollineni, J., Matsuzaki, Y., Tint, G.S. and Salen, G., 2004. Dietary cholesterol stimulates 15921680373CYP7A1 in rats because farnesoid × receptor is not activated. *Am. J. Physiol.*

Gastrointest. Liver Physiol., **286**: 730-735. https://doi.org/10.1152/ajpgi.00397.2003

Yoshioka, T., Kawada, K., Shimada, T. and Mori, M., 1979. Lipid peroxidation in maternal and cord blood and protective mechanism against activated-oxygen toxicity in the blood. *Am. J. Obstet. Gynecol*., **135**: 372-376. https://doi.org/10.1016/0002-9378(79)90708-7

Zaki, N., Hasib, A., Hakmaoui, A., Dehbi, F. and Ouatmane, A., 2013. Assessment of color, capsaicinoids, carotenoids and fatty acids composition of paprika produced from moroccan pepper cultivars (*Capsicum annuum* L.). *J. natl. Sci. Res*., **3**: 2224-3186.

Zălar, D.M., Pop, C., Buzdugan, E., Kiss, B., Ştefan, M.G., Ghibu, S., Crişan, D., Buruiană-Simic, A., Grozav, A., Borda, I.M. and Mogoşan, C.I., 2022. Effects of colchicine in a rat model of diet-induced hyperlipidemia. *Antioxidants*, **11**: 230. https://doi.org/10.3390/antiox11020230