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Research Article

Possible effect of wheat germ oil or beta-carotene to enhance kidney recovery processes in irradiated rats

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ABSTRACT

Radiation nephropathy has appeared as a substantial complication of radionuclide radiotherapy. Thus, the requirement for a protective agent against radiation injury is evident. The present study explored the potential effects of beta-carotene (βC) as well as, wheat germ oil (WGO) on gamma-irradiation resulted in oxidative stress and renal nephropathy in rats. Rats were divided into six groups: normal control group, βC group, WGO group, irradiated (IRR) group, βC+IRR group, WGO +IRR group. Oral administration of βC (10 mg /kg) or WGO (80 mg/kg) for 7 days after γ-irradiation (7.5 Gy) have significantly minimized the severity of biochemical changes, which was evidenced by a significant diminution in the level of malondaldhyde (MDA) in kidney, urea, creatinine as well as C - reactive protein (CRP) along with an increase i n Glutathione (GSH) level, superoxide dismutase activity (SOD), urea and creatinine clearance comparing to the corresponding values of irradiated rats ($P < 0.05$). Histopathological examination of kidney tissues further confirms the biochemical records. Therefore, it is concluded that, βC as well as, WGO may reduce radiation nephropathy and enhance recovery processes after radiation exposure.

Keywords: C-reactive protein; Creatinine; Creatine clearance; Radiation nephropathy; Oxidative stress

INTRODUCTION

Radiotherapy, one of the cancer therapies, depends on the generation of reactive oxygen species (ROS) to destroy malignant cells (Borek., 2004) and unfortunately, normal tissues are also injured. Radiation nephropathy defined as kidney damage and lessening of its function by ionizing radiation (Cohen., 2002). Radiation nephropathy is a complication of radionuclide, radiotherapy as well as radiation accidents (Jaggi et al., 2006). Irradiation of the kidney in the course of total body irradiation (TBI) just before bone marrow transplantation, or as a consequence of irradiation of nearby organ, may lead to radiation nephritis. Radiation-induced renal damage is a commonly documented experimental model (Hino et al ., 1993). Although radiotherapy is an essential therapy in controlling the variability of cancers, its harm effects on the normal tissues limits the success of the therapy. Therefore, a new task for medical doctor is to guarantee the patient quality of life by keeping the normal tissue from radiation damage as well as improving anti-cancer efficiency.

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(koukourakis., 2012).

Wheat germ oil contains vitamin E and tocopherols (Zhu et al., 2011), rich in unsaturated fatty acids, mostly oleic, linoleic and α-linoleicacids (Sjovall et al., 2000) and in flavonoids, sterols, octacosanols and glutathione (Zhu et al., 2006). WGO possesses anti-inflammatory and antioxidant properties (Paranich et al., 2000).

Previous studies have confirmed that WGO can diminish oxidative stress (Alessandri et al., 2006), modulate lipid metabolism (Singh et al., 2006), and decrease hyper cholesteremea and hyper glycemic. (Ikmak and Dunford., 2005), and reduces platelet aggregation, thrombus formation (Lass and Sohal., 2000).

β-carotene being the richest source of vitamin A in the human diet (Krinsky and Johnson., 2005). A single molecule of βC can be cleaved in the body to produce two vitamin A molecules. Vitamin A is important in preserving healthy vision (Groff et al., 1995), possess antioxidant biological properties (Riccioni., 2009). It has been shown to reduce singlet oxygen and reduce peroxyl radicals (Wang and Russell., 1999). Hence, the present study was designed to examine whether β C and WGO have any enhancement role in kidney recovery processes after irradiation-induced renal dysfunction *in vivo*. To address our assumption inflammatory marker (CRP), oxidative stress markers and kidney function test were examined

MATERIALS AND METHODS

Chemicals

Beta-carotene was obtained from (MEPACO-MEDIFOOD), Enshas El Raml-Sharkeye-Egypt. It was freshly prepared in sesame oil (El -Habit et al., 2000). The selected dose of βC was 10mg/kg body weight according to (Sarada et al., 2002).

Wheat germ oil was obtained from South Egypt Drug Industries Co. (SEDICO), 6 October City, Egypt, and freshly dissolved in sesame oil. The chosen dose of wheat germ oil was 80mg/kg (Said and Azab., 2006).

Animals

This study was approved to be carried out by the Committee of Scientific Ethics of Faculty of pharmacy, Al-Azhar University, Egypt, following the guidelines of animal usage. Forty eight male rats (150-180 g) were used in this research. They were attained from the animal house of the National Research Centre (Giza, Egypt). Rats were kept under suitable circumstances of controlled humidity, temperature and light. The animals were allowed free access to water and were nourished an ordinary pellet rat diet

Irradiation

Rats exposed to whole body γ-radiation (7.5 Gy) at the National Centre for Radiation Research and Technology (NCRRT), Atomic Energy Authority, Cairo, Egypt; using gamma Cell-40 with a Caesium137. The radiation dose level (7.5 Gy) was delivered at the rate of 0.46 Gy/ min.

Experimental design

Rats were allocated into six groups (each of 8 rats) allocated as follows: Control group: normal non-irradiated animals. Beta-carotene group: this group was administered orally with βC (10 mg /kg) for 7 successive days. Wheat germ oil group: Rats were administered orally with WGO (80 mg/kg) for 7 days. Irradiated group: Rats were subjected to a shot dose of the whole body γ-radiation (7.5Gy). IRR+ Beta-carotene group: this group was subjected to a γ-radiation (7.5 Gy), followed by treatment with βC orally (10 mg/kg) for 7 days. IRR+ Wheat germ oil group: this group was subjected to a single dose of the whole body γ-radiation (7.5 Gy), followed by treatment with WGO oral (80) mg/kg for 7 days. Treatment started 2h after irradiation.

Sample preparation

Urine

Rats were kept separately in metabolic cages at day 6; urine samples were collected every 8 h for 24 h and stored at 4°C. Urine samples from each rat were pooled and assayed. (Stephen et al., 2007)

Blood

Animals weighted, anesthetized by (i.p) injection of urethane at the dose of 1.5g/Kg (Maggi, 1986) and decapitated 7 days after irradiation. Whole blood was collected by heart puncture, centrifuged at 4000 rpm for 10 minutes using a centrifuge (Heraeus Christ, Taiwan).

Kidney

kidney was rinsed with ice cold saline and dried using filter papers, weighed and homogenized in ice cold saline (10% w/v homogenate) using Glas-Col® homogenizer, Terre Haute Indiana, USA, centrifuged by cooling centrifuge (Universal 16 R, Germany) (Becciolini et al., (1972).

Biochemical assays

Weight ratio was determined according to Ferreira et al., (2002) where: KW/BW = kidney weight (g) /body weight (g) x100. Urea and creatinine were assayed by the methods of Fawcett and Soctt (1960); (Jeffe 1886) respectively using kits (Diamond Diagonstics, Egypt). Clearance was calculated according to (Cockcroft and Gault., 1976). CRP was assessed by a standard enzyme linked immunosorbent assay (ELISA) method following the manufacturer's information. Glutathione (GSH) and malondialdehyde (MDA) quantities in renal homogenates were analyzed following the procedure of Beutler et al., (1963); Uchiyama and Mihara (1978), respectively. The activity of SOD was determined following the methods of Minami and Yoshikawa., (1979). Histopathological examinations were done on kidney accordingly to the method of Drury and Wallington (Drury and Wallington., 1976).

Statistical analysis

Obtained results were carried as mean ± SEM of eight animals. The difference between groups was assessed by one way analysis of variance (ANOVA) followed by Tukey's Multiple comparison test using Graph pad software prism (version 6). Statistical significance was deliberated at $p < 0.05$.

RESULTS

Assessment of relative Kidney weight and urine volume

Rats exposed to γ-radiation indicated a significant increase in kidney relative weight along with a decrease in urinary volume as compared to control group ($P < 0.05$). Oral administration of βC, or WGO for 7 days after irradiation significantly decline the kidney weight ratio and increase urinary volume as compared to the irradiated group (Table1).

Renal function markers

Table (2) shows the effects of beta-carotene, wheat germ oil and irradiation (IRR, 7.5Gy) on renal functions. Gamma radiation exposure showed a significant increase in creatinine (serum 140% and urine 130%), urea (serum 140%, urine 130%), accompanied by a significant decrease in creatinine clearance (48 %,) and urea clearance (58%) as compared to control group

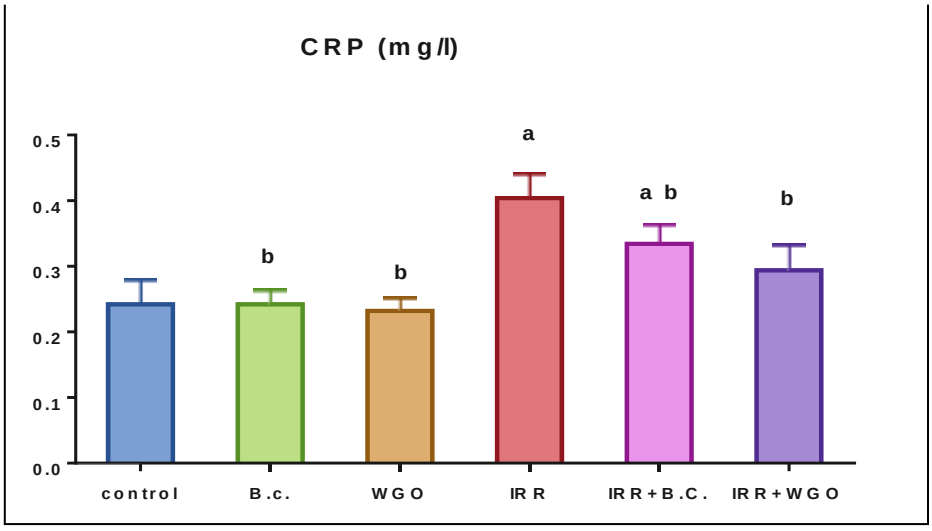

Figure 1: effect of β-carotene, wheat germ oil and irradiation (IRR, 7.5Gy) on CRP.

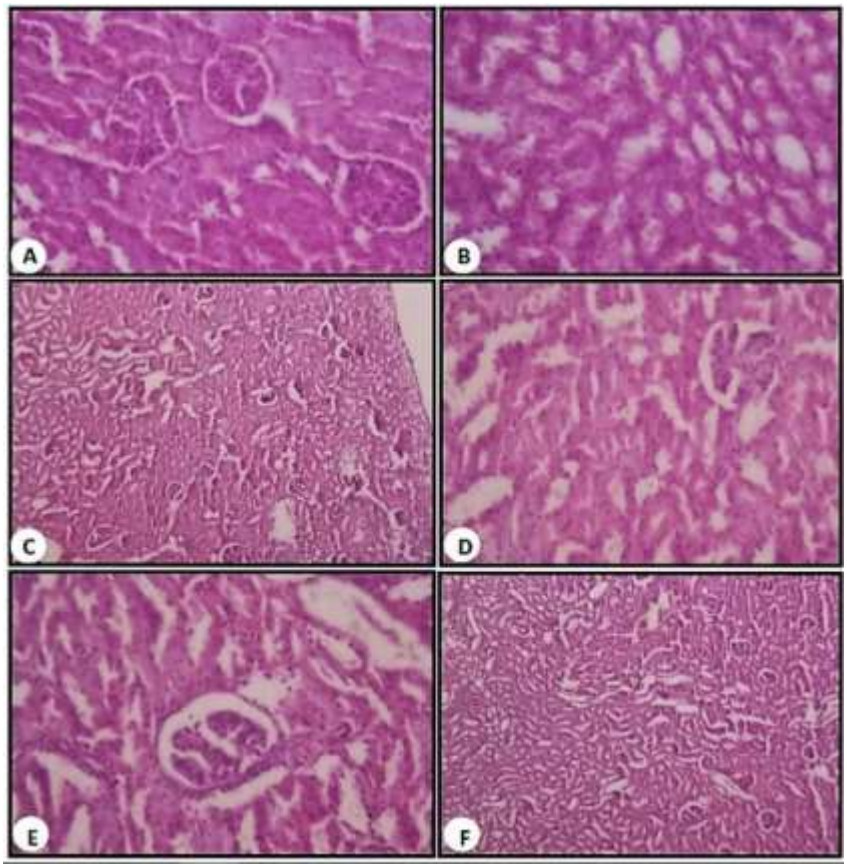

Figure 2: Photomicrograph of hematoxylin and eosin-stained kidney tissue section. Section taken from control group (A&B), control group with wheat germ (C&D), control group with beta carotene (E&F), showing normal structures of kidney with no signs of inflammation.

($P < 0.05$). Oral administration of βC or WGO for 7 days after γ-irradiation significantly decrease the serum and urine creatinine, serum and urine urea but they significantly increase creatinine and urea clearance as compared to the irradiated group.

Inflammatory marker (CRP)

CRP was measured in the serum of different treated groups (Fig:1). The level of CRP displayed a significant rise in the irradiated group (160 %) as compared to control group. Administration of βC or WGO for 7 days after γ- irradiation exerts signi ficant amelioration in the level of CRP as compared to γ- irradiated group.

Oxidative Stress biomarkers

Irradiation-induced oxidative stress in rat kidney was evaluated by assessing MDA, GSH, and SOD activity. As displayed in Table (3) Exposure of rats to γ-radiation showed a significant increase in the level of MDA together with a significant decrease in SOD activity the

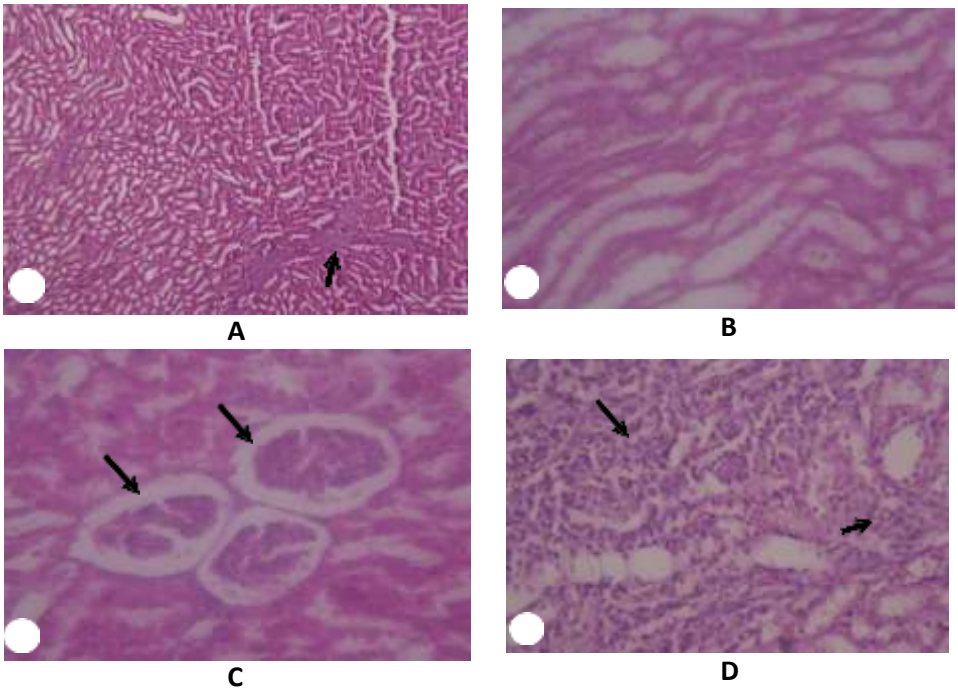

Figure 3: Photomicrograph of hematoxylin and eosin-stained kidney tissue section of irradiated rats showing: (A) the appearance of glomerular collapse (arrow), (B) visible inflammatory cell infiltration (arrows), (C) notice the interstitial fibrosis (arrows) (D) cloudy appearance and lose of cellular architecture.

Table 1: Effect of β-carotene, wheat germ oil and γ-irradiation on relative kidney weight and urinary volume / day.

Parameters	Control	βC	WGO	IRR	IRR+βC	IRR+WGO
Body weight (g)	131.8 ± 3.8	131.9 ± 6.6	132.3 ± 5.9	108.0 ± 3.8^{a}	131.3 ± 4.3	128.3± 3.5
Kidney weight(g)	0.56 ± 0.02	0.56 ± 0.02	0.35 ± 0.02	0.66 ± 0.01^{a}	0.53 ±0.01^{b}	0.52± 0.02^{b}
Weight ratio	0.42±0.009	0.42±0.016^{b}	0.39±0.007^{b}	0.61±0.02^{a}	0.4 ±0.007^{b}	0.39± .004^{b}
Urinary volume (ml)	6.6± 0.3	6.1± 0.3^{b}	6.1 ± 0.18^{b}	3.6± 0.2^{a}	4.3 ± 0.1^{a}	5.1±0.2^{ab}

Data expressed as as mean ± SEM, n = 8. a and b specify significant changes from control and IRR respectively at p ≤ 0.05 using ANOVA followed by Tukey-Kramer as a post ANOVA test.

Table 2: Effect of β-carotene, wheat germ oil and γ-irradiation on renal biomarkers

Parameters	Control	βC	WGO	IRR	IRR+βC	IRR+WGO
Serum creatinine (mg/dl)	0.55±0.009	0.5±0.011^{b}	0.53±0.004^{b}	0.79±0.037^{a}	0.72±0.029^{a}	0.67±0.011^{ab}
Urine creatinine (mg/dl)	52.46±0.95	52.09±1.66^{b}	52.19±1.32^{b}	69.45±2.5^{a}	58.88±1.16^{b}	59.43±1.66^{ab}
Serum urea (mg/dl)	25.08±0.96	24.16±0.48^{b}	24.02±0.45^{b}	35.78±1.4^{a}	30.06±0.51^{ab}	27.3± 0.87^{ab}
Urine urea (mg/dl)	91.66±1.44	93.82±1.74^{b}	92.75±1.76^{b}	122.2±1.96^{a}	103.2±1.36^{ab}	104.9±2.09^{ab}
Creatinine clearance (ml/min)	0.43±0.02	0.41± 0.01^{b}	0.41± 0.03^{b}	0.21±0.007^{a}	0.25±0.01^{a}	0.31± 0.02^{ab}
Urea clearance (ml/min)	0.016±0.001	0.016±0.001^{b}	0.016±0.001^{b}	0.008±0.001^{a}	0.009±0.001^{a}	0.013±0.001^{b}

Data expressed as as mean ± SEM, n = 8. a and b specify significant changes from control and IRR respectively at p ≤ 0.05 using ANOVA followed by Tukey-Kramer as a post ANOVA test.

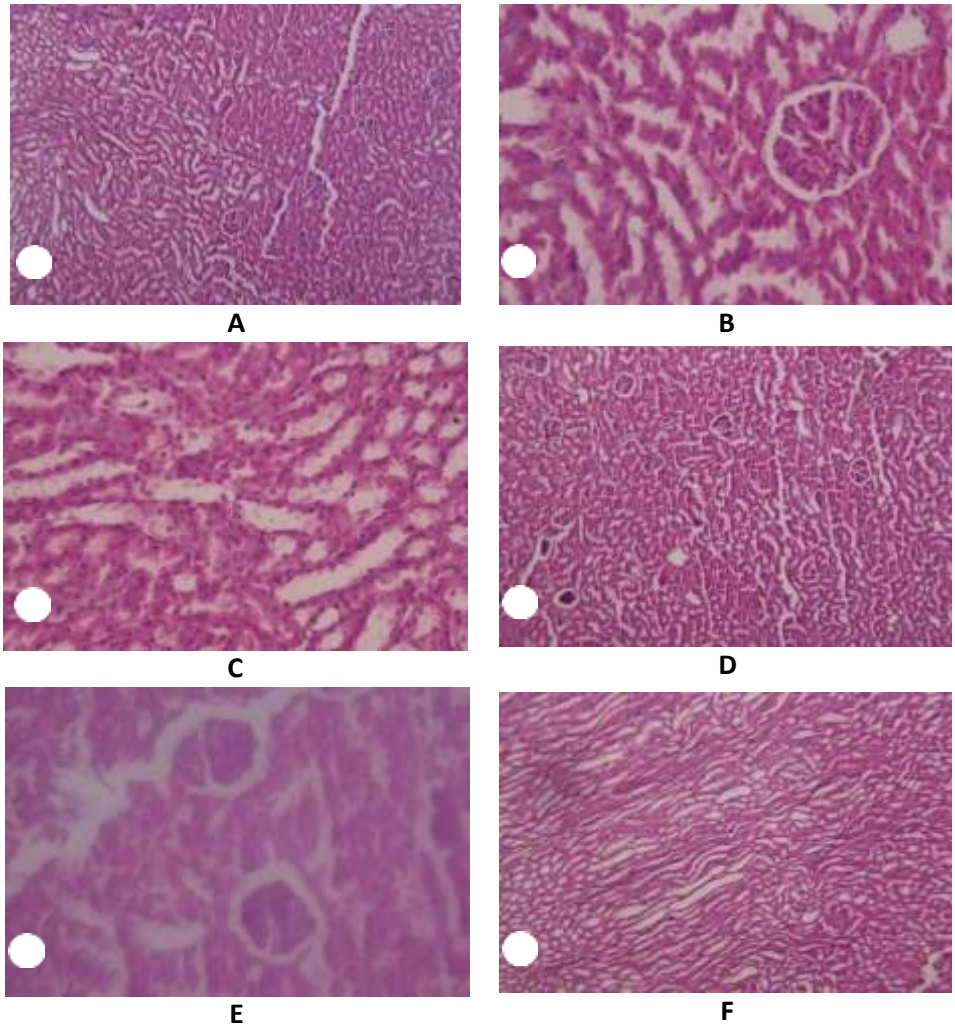

Figure 4: Photomicrograph of hematoxylin and eosin-stained kidney tissue section of irradiated rats treated with wheat germ oil.(A, B, & C), irradiated rats treated with beta carotene (D, E, & F) showing much improvement of the inflammatory process with less glomerular collapse, less inflammatory cellular infiltrate, and less interstitial fibrosis.

Table 3:Effect of β-carotene, wheat germ oil and irradiation (7.5Gy) on MDA, GSH and SOD in rat kidney tissue.

Parameters	Control	βC.	WGO	IRR	IRR+βC	IRR+WGO
MDA level (nmol/g tissue	175.8± 7.8	178± 4.1^{b}	182.5± 3.5^{b}	304.3± 10.4^{a}	224.6± 10.91^{ab}	198.7± 5.9^{b}
GSH level (mg/g tissue)	6.32± 0.5	6.62±0.3^{b}	6.71± 0.17^{b}	3.29± 0.2^{a}	5.42± 0.25^{b}	5.35± 0.24^{b}
SOD activity (µg/g tissue)	89.5± 1.9	87.96± 2.1^{b}	85.97± 1.5^{b}	50.08± 1.7^{a}	78.05± 0.99^{ab}	80.49± 0.6^{b}

GSH level as compared to control ($P < 0.05$). Rats treated with βC or WGO for 7 successive days after γ-irradiation significantly decline the level of MDA, along with an increase in SOD activity and GSH level ($P < 0.05$) compared to the irradiated group.

Histopathological examination

Rats subjected to γ-radiation showed inflammatory cell infiltration, swelling of the glomeruli, interstitial fibrosis, and cloudy appearance as well as loss of cellular architecture. Treatment of irradiated rats with WGO or βC significantly improved the inflammatory process as compared to irradiated group [Fig 2, 3, 4].

DISCUSSION

The prominent radiosensitivity of renal tissue restricts the use of radiotherapy. The occurrence of nephropathy has augmented with the usage of total -body irradiation (TBI) during bone marrow transplantation and radionuclide therapies (Cohen and Mike., 2003). In an effort to find a way of reversing the irradiation damage, two agents were studied in a rat model of radia-

tion nephropathy βC and WGO. Rats are suitable animals for experimental models to estimate renal impairment as they similar to human structure (Kaldir et al., 2008).

The Results of the current study specify a significant increase in weight ratio (kidney weight /body weight) of the irradiated group as compared to the control group. The increase in kidney weight possibly will be due to the hypercellularity in kidneys, especially in inflammatory disorders accompanied with a rise in the number of cells that correlated to cellular proliferation of epithelium cells (Owoeye et al., 2008).

Concerning the radiation effect on urea and creatinine levels, Rats exposed to γ-radiation reveal an increase in urea and creatinine levels as well as severe histopathological changes, including glomerular collapse, inflammatory cell infiltration, and loss of cellular architecture. These data agree with that reported by Adaramoye (2009). The rise in creatinine and urea levels could be attributable to radiation-induced cell membrane destruction leading to the liberation of molecules inside the cell to the blood stream in addition to, changes in amino acid metabolism (Kaplan., 1986). The increase in the level of kidney function biomarkers might indicate the existence of kidney damage. Glomerular filtration rate (GFR) is predictable to reveal the number of healthy glomeruli in the kidney, Thus, GFR is essential to evaluate renal function in medical and experimental models of renal disorder and established as the best indicator of kidney function (Breyer and Qi., 2010). GFR used to evaluate the kidneys' excretory capacity, and achieved clinically from creatinine clearance, by collecting urine samples for 24 hours (El -Minshawy et al., 2010). The results further indicate that rats exposed to ionizing radiation showed a significant decrease in creatinine and urea clearance as compared to normal control rats indicating low GFR and kidney injury. The reduction in GFR, detected in the irradiated rat kidney confirms previous observations (Robbins et al., 1991).

CRP is a sensitive indicator of inflammation and tissue damage; play an important role in damages initiated by radiation (Koc et al., 2003). CRP expression induced in the liver by a numeral cytokine (Shields., 1993), as NF-kB pro-inflammatory cytokines that activated by gamma irradiation (Winyard et al., 1997).

The Data of the current work revealed significant increase in the level of serum CRP following exposure of rats to γ- radiation (7.5 Gy). This might be due to oxidative damage induced by radiation as well as increases in inflammatory activity. Also, diminished filtration (GFR) caused by γ-irradiation could explain the decreased renal clearance of CRP.

In the present study; γ-irradiation caused a noticeable decrease in kidney GSH level and SOD activity along with an increase in the level of MDA indicating oxidative stress in kidney tissue. The obtained results were in agreement with Eroglu (2008). The decrease in SOD activity and GSH level possibly, due to their depletion by the higher creation of ROS (Kregel and Zhang., 2007). Mechanistically, ionizing radiation induced rise in ROS production is believed to be responsible for the rise in the production of lipid peroxidation; MDA (El -Ghazaly and Ramadan., 1996).

The records attained in the present study, verified that administration of WGO for 7 days post γ- irradiation (7.5 Gy) has significantly lessened the alteration of testing biochemical parameters. This is evident by enhancement of cellular antioxidant and GFR; decrease in the level of creatinine and urea as well as a decrease in the inflammatory marker CRP. Histopathological section showed an improvement of the inflammatory process with less glomerular collapse, less inflammatory cellular infiltrate, and less interstitial fibrosis. The possible mechanism that may be responsible for the protection of radiation- induced kidney damage by WGO is the radical scavenging activity intercepting those radicals involved in water radiolysis (Paranich et al., 2000). The scavenger radical activity of WGO may be due its high content of tocopherols (Vitamin E) that increase intracellular glutathione (GSH) stores (Attila et al., 2001). Another possible mechanism of WGO may be due to its contents of linoleic and linolenic acids , the source of both omega-6 and omega-3 essential fatty acids, respectively. These two fatty acids are essential for human metabolism as they cannot be produced by the organism. They are precursors of prostaglandins, which helps in healing of inflammatory processes (Zacchi et al., 2006).

The records from the current study prove that βC; protect the rat kidney from hazardous induced by γ-irradiation. Consistent with the previous study, βC; minimizes free radical damage induced by γ-irradiation (El-Habit et al., 2000) and improve renal function by decreasing plasma creatinine as well as GFR enhancement (Hosseini et al., 2009). Beta-carotene effect against oxidative stress appeared to be mediated through decreasing the pro-oxidants and enhancement of cellular antioxidant activities. Beta-carotene is capable to reduce oxygen radical and counteract lipid peroxyl radicals (Rodrigues et al., 2012). In conclusion: Owing to the data attained in the current work, it could be concluded that WGO and βC by enhancing anti oxidant activities and declining lipid peroxidation, may afford protection against radiation nephropathy by reserving the integrity of tissue utilities and lessen metabolic conditions caused by radiation exposure. Hence wheat germ oil and beta carotene therapy following irradiation could be of value to prevail against normal cell damage in cancer patients.

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REFERENCE

Adaramoye OA, 2010. Protective effect of kolaviron, a biflavonoid from Garcinia kola seeds, in brain of Wistar albino rats exposed to gamma-radiation. Biol& pharm bull, 33: (2), 260-266.

Alessandri C, Pignatelli P, Loffredo L, Lenti L, Del Ben M, Carnevale R, Violi F, 2006. Alpha-linolenic acid–rich wheat germ oil decreases oxidative stress and CD40 ligand in patients with mild hypercholesterolemia. Arterioscler Thromb vasc boil, 26:(11), 2577-2578.

Attila S, Robert C, Michail I, Henry D, Allen I, 2001. Nonlinear behavior of sinusoidally forced pyloric pacemaker neurons. J Neurophysiol, 85: 1623-1638.

Becciolini A, Ravina A, Arganini L, Castagnoli P, De Giuli G, 1972. Effect of ionizing radiation on the enzymes of the intestinal mucosa of rats at different time intervals after abdominal irradiation. Radiat Res, 49: 213-225.

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

Borek C, 2004. Antioxidants and radiation therapy. J. Nutr, 134: 3207S–3209S.

Breyer DM, Qi Z, 2010. Better nephrology for mice and man. Kidney Int, 77: 487 – 489. doi: 10.1038/ki.2009.544.

Cockcroft D W, Gault MH, 1976. Prediction of creatinine clearance from serum creatinine. Nephron, 16: (1), 31-41.

Cohen EP, Mike E, Robbins C, 2003. Radiation Nephropathy. Semin Nephrol, 23: (5), 486-499.

Cohen E P, 2002. Nephritis Radiation. Medicine Journal, 3: (3),1-20.

Drury RAB, Wallington E A, 1976. The nervous system. In Carlton's Histological Technique, ed 4. Oxford University Press, London, 260-262.

El-Ghazaly M, Ramadan L A, 1995. Alteration in some metabolic functions in albino rats after irradiation and possible role of thiola. J Egypt Soc Toxicol, 16: 105-109

El-Habit O, Saada H, Azab K S, Abdel-Rahman M, El-Malah D, 2000. The modifying effect of β-carotene on gamma radiation-induced elevation of oxidative reactions and genotoxicity in male rats. Mutat Res Genet Toxicol Environ Mutagen, 466 :(2), 179-186.

El-Minshawy O, Saber RA, Osman A, 2010. 24-hour creatinine clearance reliability for estimation of glomerular filtration rate in different stages of chronic kidney disease. Saudi J Kidney Dis Transpl, 21:686-93.

Eroglu A, Harrison E H, 2013. Carotenoid metabolism in mammals, including man: Formation, occurrence, and function of apocarotenoids thematic review series: Fat-soluble vitamins: Vitamin A. J Lipid Res, 54 : (7), 1719-1730.

Fawcett J, Scott J,1960. A rapid and precise method for the determination of urea. (1960)", J clin pathol, 13: (2), 56-159.

Ferreira D, Cunha D, Carvalho SF, Antonia DR M, Paula VD, Teixeira A, 2002. Heart Weight /Body Weight Coefficient in Malnourished Adults. Arq. Bras. Cardiol, 78: (4), http://dx.doi.org/10.1590/S0066-782X2002000400005.

Groff J, Gropper S, Hunt S,1995. The water soluble vitamins. Advanced nutrition and human metabolism, 3: 289-297.

Hino O, Klein-Szanto A, Freed J J, Testa J R, Brown D Q, Vilensky M, Knudson A G, 1993. Spontaneous and radiation-induced renal tumors in the Eker rat model of dominantly inherited cancer. Proc Natl Acad Sci, 90: (1), 327-331.

Hosseini F, Naseri M, Badavi M, Ghaffari M, Shahbazian H, Rashidi I, 2009. Protective effect of beta carotene pretreatment on renal ischemia/reperfusion injury in rat. Pak j of biol sci, 12: (16), 1140-1145.

Irmak S, Dunford NT, 2002. Policosanol contents and compositions of wheat varieties. J Agric Food Chem, 53: (14), 5583-5586.

Jaffé M, 1886. Ueber den Niederschlag, welchen Pikrinsäure in normalem harn erzeugt und über eine neue reaction des kreatinins. Z Phys Chem, 10(5): 391-400.

Jaggi JS, Seshan SV, McDevitt MR, Sgouros G, Hyjek E, Scheinberg DA, 2006. Mitigation of radiation nephropathy after internal alpha-particle irradiation of kidneys. Int J Radiat Oncol Biol Phys, 64:1503 - 1512.

Kaldir M, Cosar-Alas R, Cermik T F, Yurut-Caloglu V, Saynak M, Altaner S, Türe M, 2008.Amifostine Use in Radiation-Induced Kidney Damage. Strahlenther Onkol, 184: (7), 370-375.

Kaplan M M, 1986. Serum alkaline phosphatase–another piece is added to the puzzle. Hepatology, 6:(3), 526-528.

Koc M, Taysi S, Buyukokuroglu M E, Bakan N, 2003. Melatonin protects rat liver against irradiation-induced oxidative injury. J Radiat Res, 44: (3), 211 - 215.

Koukourakis MI, 2012. Radiation damage and radioprotectants: new concepts in the era of molecular medicine. Br J Radiol, 85: 313-330.

Kregel K C, Zhang H J, 2007. An integrated view of oxidative stress in aging: basic mechanisms, functional effects, and pathological considerations. Am J Physiol Regul Integr Comp Physiol, 292 :(1), R18-R36.

Krinsky, N. I., & Johnson, E. J, 2005. Carotenoid actions and their relation to health and disease. Molecular aspects of medicine, 26 :(6), 459-516.

Lass A, Sohal RS, 2000. Effect of coenzyme Q(10) and alpha-tocopherol content of mitochondria on the production of superoxide anion radicals. FASEB J ,14 : 87-94.

Maggi CA, Meli A, 1086. Suitability of urethane anesthesia for physiopharmacological investigations in various systems. Part 1: general considerations. Experientia, 42:109–14.

Minami M, Yoshikawa H, 1979. A simplified assay method of super oxide dismutase. Clin Chim Acta, 29: 337-342.

Owoeye O, Malomo A, Elumelu T, Salami A, Osuagwu F, AkinloluA, Shokunbi M, 2008. Radiation Nephritis: Anti-inflammatory Effect of Dexamethasone in Adult Wistar Rats (Rattus norvegicus). Int J Morphol, 26 :(1), 69-74.

Paranich V, Cherevko O, Frolova N, Paranich A, 2000.The effect of wheat germ oil on the antioxidant system of animals. Lik Sprava, 2: 40-44.

Riccioni G, 2009. Carotenoids and cardiovascular disease. Current atherosclerosis reports, 11:(6), 434 - 439.

Robbins M EC , Bywaters T, Rezvani M, GoldingS J Hopewell JW, 1991. The Effect of Unilateral Nephrectomy on the Subsequent Radiation Response of the Pig Kidney. Int J Radiat Biol, 59:(6), 1441-1452.

Rodrigues E, Mariutti L R, Mercadante A Z, 2012. Scavenging capacity of marine carotenoids against reactive oxygen and nitrogen species in a membrane-mimicking system. Mar drugs, 10: (8), 1784-1798.

Said U, Azab K, 2006. Efficacy of wheat germ oil in modulating radiation-induced heart damage in rats. Egypt J. Rad. Sci. Applic, 19: (2), 433-452.

Sarada S, Dipti P, Anju B, Pauline T, Kain A. Sairam M, Selvamurthy W, 2002. Antioxidant effect of beta-carotene on hypoxia induced oxidative stress in male albino rats. J Ethnopharmacol, 79: (2), 149 -153.

Shields M J, 1993. A hypothesis resolving the apparently disparate activities of native and altered forms of human C-reactive protein. Immunol Res, 12:(1), 37-47.

Singh D K, Li L, Porter TD, 2006. Policosanol inhibits cholesterol synthesis in hepatoma cells by activation of AMP-kinase. J Pharm Exp Ther. 318:(3),1020-1026.

Sjovall O, Virtalaine T, Lapvetelainen A, Kallio H, 2000. Development of rancidity in wheat germ analyzed by headspace gas chromatography and sensory analysis. J Agric Food Chem, 48: 3522-3527.

Stephen O A, Abdulkadir A S, Oladepo WD, Thajasvarie N, 2007. Effect of Melatonin on Carbon Tetrachloride Induced Kidney Injury in Wistar Rats. Afr J Biomed Res, 10:153 - 164.

Uchiyama, M, & Mihara M, 1978. Determination of malonaldehyde precursor in tissues by thiobarbituric acid test. Analytical biochemistry, 86: (1), 271-278.

Wang XD, Russell R M, 1999. Procarcinogenic and anticarcinogenic effects of β-carotene. NUTR REV, 57: (9), 263-272.

Winyard P G, Zhang Z, Blake D R, 1993 In: Thomas, C.E., Kalyanaraman, B. (Eds.), Oxygen Radicals and Disease Process, 85–99.

Zacchi P, Daghero J, Jaeger P, Eggers R, 2006. Extraction/fractionation and deacidification of wheat germ oil using supercritical carbon dioxide. Braz J Chem Eng, 23(1): 105-110.

Zhu K X, Zhou H M, Qian H F, 2006. Proteins extracted from defatted wheat germ: nutritional and structural properties. CEREAL CHEM. 83(1): 69-75.

Zhu KX, Lian CX., Guo XN, Peng W, Zhou HM, 2011. Antioxidant activities and total phenolic contents of various extracts from defatted wheat germ. FOOD CHEM, 126:(3), 1122-1126.