



Biochemical role of wheat germ oil on biomarkers of oxidative stress and inflammatory response in a rat model of endotoxemia

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ABSTRACT

The main objective of this study was to investigate the protective effect of wheat germ oil on oxidative stress induced by endotoxin in rats. Forty five male rats divided into three equal groups of (15 each). Group I (control): rats received no drugs. Group II (Endotoxin group): rats injected intraperitoneally (i.p) with a single dose of endotoxin (200 mg/Kg b.wt). Group III (wheat germ oil protected group): rats pretreated with wheat germ oil (270 mg/kg b.wt./day) orally for 21 day then injected with a single dose of endotoxin (200 mg/Kg b.wt). Blood samples for serum separation and liver tissues were collected from all animal groups after 1 and 3 hours from the onset of endotoxin injection. Serum tumor necrosis factor- alpha (TNF- α), interleukin-6 (IL-6), L-Malondialdehyde (L-MDA), Nitric oxide (NO) and sialic acid, in addition to liver tissue superoxide dismutase (SOD), glutathione peroxidase (GPx), catalase (CAT), myeloperoxidase (MPO), reduced glutathione (GSH), Glutathione S-transferase (GST) and L-Malondialdehyde (MDA) were determined. The obtained results revealed that, endotoxemia could potentially increase serum TNF- α , IL-6, L-MDA concentrations and markedly decrease NO and sialic acid levels. Moreover, SOD, CAT and MPO activities and GSH concentration in liver tissues were significantly decreased. Pretreatment with wheat germ oil decreased LPS-triggered pathogenic responses and prevented the increase of TNF- α , IL-6 and L-MDA. Moreover, pretreatment with wheat germ oil in endotoxin injected rats responsible for the protection of membrane polyunsaturated fatty acids against lipid peroxidation by increasing the activity of liver tissue antioxidant enzymes GST, SOD and CAT. From the obtained results it could be concluded that, wheat germ oil protects against lipid peroxidation, oxidative stress and decrease the inflammatory response induced by endotoxin injection in rats.

KEY WORDS: Endotoxin, wheat germ oil, Oxidative stress, Inflammatory markers, Antioxidant status.

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1. INTRODUCTION

Lipopolysaccharide (LPS) is a glycolipid component of the cell wall of gram-negative bacteria inducing deleterious effects on several organs including the liver and eventually leading to septic shock and death. Endotoxemia-induced hepatotoxicity is characterized by disturbed intracellular redox balance, excessive reactive oxygen species (ROS) and accumulation inducing DNA, proteins and membrane lipid damages (Sebai et al., 2010). Endotoxin induces shock states in both humans and

animals characterized by fever, hypotension, intravascular coagulation, and finally multi-organ failure system (Sakaguchi and Furusawa 2006). The harmful effects of LPS are, in part, suspected to its ability to induce an oxidative stress status characterized by depletion of endogenous antioxidant enzyme activities such as SOD, CAT and GPx. The pro-oxidant action of the endotoxin is due to its ability to induce excessive ROS and RNS accumulation

leading to cellular injury by impairment of vital macromolecules as protein and lipid (Mallis et al., 2001) resulting in altered membrane fluidity and mitochondrial function (Susana and Ana, 2002). Wheat germ oil is an excellent source of polyunsaturated fatty acids and vitamin E. It is one of the richest natural sources of α -tocopherol, the type of tocopherol with the greatest vitamin E activity. Wheat germ oil has been attributed to reduced plasma and liver cholesterol in animals, improved physical endurance, and delayed aging (Tong and Lawrence 2001). Also, wheat germ oil can reduce oxidative stress, improve lipid metabolism (Singh et al., 2006). Moreover, it had significantly higher protective levels of vitamin E in the blood and liver, conferring greater anti-oxidant protection (Leenhardt et al., 2008). Therefore, in light of above mentioned findings, the aim of the present study was to investigate whether and to what extent wheat germ oil, would provide protection against oxidative stress and inflammatory markers of endotoxemia in rats.

2.2- MATERIALS AND METHODS

2.1. Experimental animals:

Forty five male albino rats, 12-16 weeks old and average body weight 180-220 gm were used in the experimental investigation of this study. The rats were obtained from the Laboratory Animals Research Center, Faculty of Veterinary Medicine, Benha University. Rats were housed in separated wire mesh cages and kept at constant environmental and nutritional conditions throughout the period of experiment. The animals were fed on constant ration and fresh, clean drinking water was supplied ad-libitum. The animals were left 14 days for acclimatization before the beginning of the experiment.

2.2. Wheat germ oil:

Wheat germ oil was obtained from Eminor Trading Company, Egypt .AIKo Natural Products (www.aikochina.com)

(info@aikochina.com). Wheat germ oil was dissolved in propylene glycol and given alone at a dose level of (270 mg/kg b.wt/day), orally for 21 days before endotoxin administration, as a protective antioxidant before endotoxin standard injection (endotoxemia induction).

2.3. Endotoxin (LPS):

Escherichia coli (Serotype O55:B5) is a gram negative rod (bacillus) in the family *Enterobacteriaceae* and is chosen and used in this study to induce endotoxemia. Its white powder, highly soluble in distilled water or in 0.9% saline, solubility clear colourless and endotoxin unit 12,000 units/unit. LPS is manufactured in Sigma Chemical CO. (St.Louis, MO, USA) and purchased from schnellldorf, Germany through the Egyptian International Centre for Import Cairo, Egypt.

2.4. Experimental design:

Rats were randomly divided into three main groups (15 each), placed in individual cages and classified as follow: Group (1): Control normal group: received no drugs, served as control non-treated for all experimental groups. Group (2): Endotoxin group: injected intraperitoneally (i.p) with a single dose of LPS (200 mg/Kg b.wt). Group (3): Wheat germ oil Protected group: given Wheat germ oil as a protective antioxidant before endotoxin standard injection (endotoxemia induction). Wheat germ oil was dissolved in propylene glycol and given alone at a dose level of (270 mg/kg b.wt/day), orally for 21 days before endotoxin administration. Then, at the 22th days, Wheat germ oil was given at the same dose, followed by a single i.p injection of endotoxin. The rats were sacrificed at one hour and 3 hours after endotoxin injection.

2.5. Sampling:

2.5.1. Blood samples:

Blood samples for serum separation were collected after 1 and 3 hour post toxin injection by ocular vein puncture in dry, clean, and screw capped tubes after

overnight fasting from all animals groups (control and experimental groups) twice along the duration of experiment at 1 and 3 hours from the onset of endotoxin injection. Serum was separated by centrifugation at 4000 r.p.m for 15 minutes. The clean, clear serum was separated by automatic pipette and received in dry sterile samples tube, and proceed directly for glucose determination, then kept in a deep freeze at - 20° C until used for subsequent biochemical analysis.

2.5.2. Liver specimens:

Rats were killed by decapitation. The liver specimen quickly removed, cleaned by rinsing with cold saline and stored at -20°C. Briefly, liver tissues was minced into small pieces, homogenized with ice cold 0.05 M potassium phosphate buffer (pH7.4) to make 10% homogenates. The homogenates were centrifuged at 4000 r.p.m. for 15 minute at 4°C. The supernatant was used for the determination of L-MDA, MPO, GSH and antioxidant enzymes.

2.5.3. Biochemical analysis:

Serum TNF- α , IL-6, NO and Liver tissues (L- MDA), GPx, SOD, CAT, GSH, MPO, Sialic acid and GST) were determined according to the methods described by Beyaert and Fiers, (1998), Chan and Perlstein, (1987), Vodovotz, (1996), Mesbah et al., (2004, Gross et al., (1967), Kakkar et al., (1984), Luck, (1974), Moron et al., (1979). Rats Myeloperoxidase ELISA kit (Kamiya Biomedical Company, Cat. No.KT-60345), Sialic acid ELISA kit (Cat.No.CSB-E09605h, Cusabio, Biotech Co, Wuhan, China) and Harada et al., (1987) respectively.

3. RESULTS

3.1. *Table (1): Effect of pretreatment with wheat germ oil on serum biochemical parameters at 1 and 3 hours after endotoxin injection in rats.*

The obtained data (Table 1) revealed that, the mean value of serum AST, ALT, GGT, ALP, 5'-nucleotidase activities, phospholipids, TNF- α , IL-6, total cholesterol and L-MDA concentrations significantly increased in rats injected with LPS after (1hr.and 3hrs.). Meanwhile, Serum NO and Sialic acid significantly decreased in rats injected with LPS after (1hr.and 3hrs.) when compared with control group. Pre-treatment with Wheat germ oil significantly decreased the serum GGT, AST, ALT activities, phospholipids, IL-6, TNF- α and L-MDA after 1 hour and after 3 hours of injection, non-significantly decreased ALP after 1 hour this became significantly after 3 hours of endotoxin injection, significantly decreased the 5'-nucleotidase activity after 1hour and non-significantly decreased after 3 hours of endotoxin injection comparing with the endotoxin group, non-significantly decrease in cholesterol concentration after 1 hour and after 3 hours, meanwhile, significantly increased in NO, Sialic acid concentrations after 1 and 3 hours of LPS injection when compared with endotoxin group. 3.2. *Table (2): Effect of pre-treatment with wheat germ oil on liver tissue biochemical parameters at 1 and 3 hours after endotoxin injection in rats.*

The presented results in Table (2) revealed that, endotoxin treated rats significantly decreased Liver SOD, GPx, CAT, MPO, GST activities and significantly increased GSH concentration after 1 and 3 hours of endotoxin injection in comparison with control group. Pretreatment with Wheat germ oil o significantly increased the liver SOD, GPx, CAT, MPO, GST activities and significantly increased GSH concentration

Table (1): Effect of pretreatment with wheat germ oil on some serum biochemical parameters at 1 and 3 hours after endotoxin injection in rats.

Parameters	Animal Groups Control group	Endotoxin group		Endotoxin + wheat germ oil group	
		Time after endotoxin injection		Time after endotoxin injection	
		1 Hour	3Hours	1 Hour	3Hhours
TNF- α (pg/mL)	89.40 \pm 7.98 ^c	241.83 \pm 16.58 ^a	269.66 \pm 10.37 ^a	94.01 \pm 5.52 ^c	114.12 \pm 8.73 ^c
IL-6 (pg/mL)	43.09 \pm 9.52 ^c	108.97 \pm 4.12 ^{ab}	113.93 \pm 20.62 ^a	42.94 \pm 4.30 ^c	44.48 \pm 6.60 ^c
MDA(mmol/L)	85.52 \pm 4.07 ^{cde}	158.55 \pm 10.67 ^a	135.88 \pm 8.81 ^b	78.37 \pm 2.56 ^{de}	68.90 \pm 5.54 ^e
NO (mmol/L)	38.91 \pm 2.56 ^b	14.94 \pm 1.62 ^c	13.45 \pm 1.78 ^e	38.43 \pm 2.33 ^b	46.61 \pm 1.73 ^a
Sialic acid (nmol/mL)	3.62 \pm 0.18 ^{bc}	1.89 \pm 0.07 ^d	2.26 \pm 0.20 ^d	4.21 \pm 0.15 ^a	4.00 \pm 0.16 ^{ab}

Data are presented as (Mean \pm S.E). Where S.E = Standard error
Mean values with different superscript letters in the same row are significantly different at ($P<0.05$).

Table (2): Effect of pre-treatment with wheat germ oil on some liver tissue biochemical parameters at 1 and 3 hours after endotoxin injection in rats.

Parameter	Animal Groups Control group	Endotoxin group		Endotoxin + wheat germ oil group	
		Time after endotoxin injection		Time after endotoxin injection	
		1 H	3H	1 H	3H
SOD–Tissue(u/gm tissue)	35.13 \pm 1.97 ^a _b	18.67 \pm 1.53 ^c	19.12 \pm 1.58 ^c	35.38 \pm 0.82 ^a _b	37.45 \pm 1.05 ^a
GPX –Tissue (ng/gm tissue)	36.00 \pm 1.54 ^a	14.59 \pm 1.81 ^e	17.73 \pm 1.77 ^{de}	24.52 \pm 1.06 ^c	30.46 \pm 1.016 _b
CAT – Tissue(mmol/gm tissue)	41.00 \pm 1.78 ^a	24.26 \pm 1.13 ^{cd}	22.45 \pm 1.76 ^d	44.38 \pm 1.18 ^a	44.19 \pm 1.97 ^a
MPO – Tissue(ng/gm tissue)	13.15 \pm 0.63 ^a	5.68 \pm 0.87 ^d	6.99 \pm 0.92 ^d	12.29 \pm 1.31 ^a _b	11.62 \pm 0.79 ^{ab} _c
GSH – Tissue(ng/gm tissue)	2.87 \pm 0.08 ^b	4.57 \pm 0.36 ^a	5.10 \pm 0.65 ^a	1.43 \pm 0.06 ^c	2.25 \pm 0.13 ^{bc}
GST – Tissue(ng/gm tissue)	1.21 \pm 0.04 ^a	0.33 \pm 0.03 ^e	0.52 \pm 0.02 ^d	0.85 \pm 0.03 ^b	0.76 \pm 0.04 ^b
MDA - Tissue(mmol/gmtissue)	64.19 \pm 5.86 ^b	144.71 \pm 16.54 _a	134.18 \pm 7.94 _a	68.75 \pm 5.48 ^b	69.45 \pm 4.56 ^b

Data are presented as (Mean \pm S.E). Where S.E = Standard error. Mean values with different superscript letters in the same row are significantly different at ($P<0.05$).

after 1 hour and after 3 hours of endotoxin injection comparing with the endotoxin group.

4. DISCUSSION

Bacterial endotoxin (LPS) is derived from the outer cell wall of gram-negative coliform bacteria, is heat-stable, and is ubiquitous in the environment. The innate immune system recognizes both viable microorganisms and nonviable parts of these organisms (including LPS), which are found in varying concentrations in many indoor and outdoor environments (Braun-Fahrlander et al., 2002). Exposure to LPS during early life induces inflammation and potentially tolerance to subsequent LPS exposure (Natarajan et al., 2008). Bacterial endotoxin is a potent inducer of the host immune system, including the overproduction of numerous pro- and anti-inflammatory cytokines, an increase in oxidative stress, and the induction of nitric oxide synthase (iNOS) and hemeoxygenase-1 (HO-1). Together, these events result in severe tissue injury. Moreover, LPS causes endotoxemia, which is associated with multiple organ failure and is often lethal (Takamiya et al., 2009). Oxidative stress occurred when the physiological balance between radical-generating and radical-scavenging is disrupted in favor of the former. As a result, the production of certain oxidation products is increased. The attack of oxygen free radicals on cellular lipids results in the formation of aldehydic lipid hydroperoxide decomposition products like malondialdehyde (MDA), which is traditionally used as liable marker of lipid peroxidation (Ljubuncic et al., 2000). Reactive oxygen species (ROS) are formed as a normal product of aerobic metabolism in mitochondrial and microsomal enzymatic reactions (Oruc and Uner, 2000). However, these ROS can be generated at elevated rates under pathophysiological conditions (Ahmed et al., 2000). The presented data in Table (1) demonstrated that, endotoxin

injection in rats significantly increased serum TNF- α and IL-6 levels at 1 and 3 hours after endotoxin injection in comparison with control group. A significant decrease in the value of serum TNF- α and IL-6 was reported in wheat germ oil pre-treatment after 1 and 3 hours of endotoxin injection. Similarly, Ibrahim,(2010) concluded that, wheat germ oil pre-treatment led to suppression of TNF- α and IL-6 levels thus support a hepatoprotective role of wheat germ oil in septic conditions. Endotoxin treatment significantly decreased serum nitric oxide and sialic acid concentrations in rats at 1 and 3 hours after injection when compared with control group. These results are agreed with the data of Young et al., (2000) who found that, the release and activity of NO is reduced in LPS, causing dysfunction of the smooth muscle system and reduced vascular relaxation with consequent dysfunction. This dysfunction is attributable to high levels of free radicals that inactivate NO and/or reduce its expression. Also, Hirohashi and Morrison (1996) provided that, LPS was not sufficient to induce NO production in mouse macrophages. Pretreatment with wheat germ oil in endotoxin induced rats exhibited a significant increase in serum NO and sialic acid concentration. Similarly, Garg et al., (2005) showed that, α -tocopherol supplementation increased the antioxidant defense mechanism in LPS rats and provided evidence of its possible therapeutic role in diseases that produce free radicals. As Wheat germ oil is rich with α -tocopherol, it has a dramatic impact on the concentration of free radical nitric oxide in broilers. The obtained data presented in Tables (1 and 2) showed that, intra peritoneal injection of endotoxin to normal rats caused a significant increase in the value of serum and liver tissue MDA concentrations after 1 and 3 hours in comparison with control group. These results are agreed well with those of Köse and Dogan, (1995) who stated that, plasma MDA levels in LPS injected groups were

significantly higher than those of the other groups. In an experimental model, endotoxin injection results in lipid peroxidation and membrane damage in the liver, sepsis and decreasing levels of free radical scavengers (Sakaguchi and Furusawa, 2006). In present study wheat germ oil decrease L-MDA level. Similarly Navarro et al., (1998) showed that, treatment with α -Tocopherol alone caused a decline in MDA level compared with controls, because α -Tocopherol is a lipophilic chain-breaking antioxidant with potent free radical scavenging power. Likely, pretreatment with α -Tocopherol before administration of LPS protected hepatocytes against its toxic effect as reported by Zhang et al., (2001) who confirmed the protective role of α -Tocopherol against lipid peroxidation in isolated mitochondrial membranes. Endotoxin treated rats significantly decreased liver SOD, GPx, CAT, MPO, GST activities and significantly increased GSH concentration after one and three hours of endotoxin injection in comparison with control group. These results agree with the findings of Pilkhwal et al., (2010) who reviewed that, LPS markedly decreased liver SOD activity after 4 hours and this decrease persist after 8 hours after injection indicating oxidative stress in rats as compared with control group. Also, Sushma et al., (2010), observed that, LPS significantly reduced the levels of liver SOD as compared to the control group. SOD is the important antioxidant enzyme having an antitoxic effect against superoxide anion and the less expression of SOD might be an adaptive response and it results in decreased dismutation of superoxide to hydrogen peroxide (Janani and Surapaneni 2010). GPx, an oxidative stress inducible enzyme plays a significant role in the peroxyl scavenging mechanism and in maintaining functional integration of the cell membranes (Chandra et al., 2000). Sushma et al., (2010) reported that, LPS significantly reduced the levels of liver catalase as compared to the control group.

Also, Dutta and Bishayi, (2009) found that, decreased liver catalase activity was observed after LPS administration in mice. It may be suggested that after LPS administration in the mice there may be decreased H_2O_2 in the liver, and not scavenge enough those increased oxidant burden liver tissue so not more catalase expression. This significant decrease in the activity of catalase in LPS injected rats is ascribed as that catalase is the enzyme, which protects the cells from the accumulation of hydrogen peroxide by dismutating it to form water and oxygen or by using it as an oxidant in which it works as a peroxidase (Harishekar, 2012). Myeloperoxidase (MPO) activity is considered as an index for the evaluation of neutrophil infiltration. Bahar et al., 2005 investigate the effect of endotoxin on neutrophil infiltration as an index for the development of inflammation, MPO activity was measured in tissues from endotoxemic animals reported that endotoxemia increased MPO activity in heart and aorta, who added that, endotoxin caused decrease in MPO activity in mesenteric artery. These results suggest that MPO produced by INOS expressed in at least the heart and aorta acts as a pro-inflammatory mediator in this endotoxemia model in rat. On contrary, Haegens et al., (2005) showed that, LPS instillation in a dramatic increase in MPO activity in mice. A significant increase in HO-1 expression and MPO in mice after LPS exposure was observed. The dose of LPS used resulted in dramatic lung neutrophilia in mice with subsequent oxidative stress. Glutathione S-transferase (GST) is a family of enzymes that play an important role in detoxification of xenobiotics. The presented results in table (2) revealed that endotoxin treated rats significantly decreased liver GST after 1 and 3 hours of endotoxin injection who compared with the control group. These results is agreed with the results of Xu et al., (2013) who stated that, LPS injection decrease GST levels in liver tissue of rats. Sushma et al., (2010) stated that, tocopherol

supplementation in LPS- challenged rats increased the SOD level in both pre and post-LPS challenged groups which were supplemented with tocopherol for 15 days. These results may be due to that, α -Tocopherol decreased lipid peroxidation, leading to decreased levels of ($\text{O}_2^{\bullet-}$) and endotoxemia is also accompanied by significant changes in the reductiv oxidative balance of critical target organs(Wang et al., 2004).Likewise, Gilad et al., (1996) indicated that, treatment with vitamin E (pre or post) showed significant restoration in superoxide dismutase activity in liver tissue. Onyema et al., (2006) hypothesized that, the antioxidant enzyme in critical target organs, mainly the liver may be up regulated by the administration of Vitamin E. Moreover, vitamin E treatment (pre or post) showed significant restoration in enhanced glutathione peroxidase activity in liver while no protection was observed in kidney tissue. This may be due to reactions leading to removal of peroxides and aldehydic products of lipid peroxidation with resultant decrease in GSH levels through oxidation and/or conjugation (Mannervik, 1985). Noticeably, Navarro et al., (1998) showed that, treatment with α -Tocopherol alone or pretreatment with α -Tocopherol before LPS administration caused a decline in GPx activity compared with controls. This may be related to that, α -Tocopherol is a lipophilic chain-breaking antioxidant with potent free radical scavenging power. Furthermore, Gilad et al., (1996) recorded that vitamin E treatment (pre or post) significantly reversed depleted Catalase activity in liver tissue. Sushma et al., (2010) concluded that, Catalase activity was significantly increased in tocopherol supplemented and LPS challenged group. On the other hand, Javanbakht et al., (2010) found that, vitamin E could increase erythrocyte catalase activity in sepsis patients accompanied with atopic dermatitis after 60 days from sepsis. Glutathione reduced is a key enzyme of the antioxidative system that protects cells against free radicals. No change of

GSH/GSSG ratio may be contributes to decrease oxidative stress and there is increasing evidence indicating that oxidative stress plays an important role in the pathogenesis of many diseases(Tandogan and Ulusu, 2006). Likewise, Oruc and Uner, (2000) demonstrated that, the increase in specific activity of GSH after LPS administration probably reflects an adaptation to oxidative conditions. Even though, Navarro et al., (1998) investigated that, pretreatment with α -Tocopherol before LPS administration led to a decline in GSH activity, likely because α -Tocopherol is a lipophilic chain-breaking antioxidant with potent free radical scavenging power. The results showed that the protective effect caused by pretreatment with α -Tocopherol then with LPS for 5hours was less than that caused on pretreatment with α -Tocopherol 2 hr. before LPS administration. This may demonstrate the destruction of α -Tocopherol with time (Willmore and Storey, 2007). Furthermore, Muller et al., (1993) indicated that, the specific activity of GSH was elevated after LPS treatment. However, pretreatment with α -Tocopherol 2 hours before administration of LPS reduced the enzyme activity. Gilad et al., (1996) also demonstrated that, treatment with vitamin E (pre or post) showed significant recovery in liver glutathione levels towards the control value. Fernandez-Checa and Kaplowitz, (2005) suggested that, α -tocopherol reduced GSH in rat liver subjected to endotoxemia after exposure for 24 hours from injection.

CONCLUSION: the present study demonstrated that wheat germ oil administration provided an effective protection against endotoxemia and oxidative damage in liver tissue induced by Endotoxin in rats, since wheat germ oil able to ameliorate serum biochemical parameters, enzymatic and non-enzymatic antioxidant defense system in liver tissue. We recommended that, administration of wheat germ oil is very important for protection of different

body tissue, especially liver tissue, against oxidative stress or even inflammation.

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التأثير الوقائي لزيت جنين القمح على الدلالات البيوكيميائية لاجتهاد التأكسدي واستجابة الالتهابات لنموذج التسمم البكتيري في الفئران

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الملخص العربي

في هذه الدراسة تم تقييم التأثير الوقائي والعلاجي لزيت جنين القمح على المؤشرات الحيوية من الأوكسدة والاستجابة الالتهابية الناجمة عن تسمم الكبد من خلال تسمم الدم البكتيري فضلا عن آلية المشاركة في حماية الكبد من مثل هذه الأنواع من التسمم. وقد أجريت هذه الدراسة لأمد قصير مدته ثلاثة اسابيع على عدد خمسة وأربعون من ذكور الفئران البالغة التي تتراوح أوزانها من 180-250 جرام قسمت الى ثلاث مجموعات: المجموعة الأولى (المجموعة الضابطة): اشتملت على 15 فأر غير معالجة. المجموعة الثانية (المجموعة المريضة): اشتملت على 15 فأر تم حقنهم ببكتريا مسممة (الأندوتوكسين) في الغشاء البريتوني بجرعة مقدارها 50 مليجرام لكل 0,250 كيلوجرام من وزن الجسم. المجموعة الثالثة (مجموعة زيت جنين القمح): اشتملت على 15 فأر تم تجرعهم زيت جنين القمح يوميا عن طريق الفم لمدة 21 يوم ثم حقنهم ببكتريا مسممة (الأندوتوكسين) في الغشاء البريتوني بجرعة مقدارها 50 مليجرام لكل 0,250 كيلوجرام من وزن الجسم. العينات: عينات الدم: استمرت التجربة لمدة 3 اسابيع وتم أخذ اول عينات الدم في نهاية الأسبوع الثالث بعد الصيام طوال فترة الليل وتم سحب العينات مرتين بعد ساعة وبعد ثلاث ساعات من حقن الأندوتوكسين. أظهرت النتائج ارتفاع معنوي في عامل تنخر الورم ألفا، انترلوكين 6، مالون داي ألدهيد. وكذلك في أنسجة الكبد زيادة في نشاط إنزيم سوبراكسيد ديسميوتيز، جلوتاثيون بيروكسيداز، إنزيم الكتاليز، مايلوبيروكسيداز -جلوتاثيون اس ترانسفيراز -مالون داي ألدهيد في المجموعة المريضة بعد ساعة وبعد ثلاث ساعات من حقن الأندوتوكسين وانخفاض معنوي في تركيز أكسيد النيتريك ومصل حامض سياليك وأخيرا جلوتاثيون مختزل في نسيج الكبد. من جهة أخرى أوضحت الدراسة انخفاض معنوي في تركيز عامل تنخر الورم ألفا – انترلوكين 6 -مالون داي ألدهيد بعد ساعة وبعد ثلاث ساعات من حقن الأندوتوكسين في مجموعة زيت جنين القمح وكذلك في أنسجة الكبد وجود انخفاض معنوي في نشاط إنزيم سوبراكسيد ديسميوتيز - جلوتاثيون بيروكسيداز - إنزيم الكتاليز - مايلوبيروكسيداز - جلوتاثيون اس ترانسفيراز - مالون داي ألدهيد في المجموعة المعالجة مسبقا بزيت جنين القمح بعد ساعة وبعد ثلاث ساعات من حقن الأندوتوكسين. وزيادة معنويه في تركيز مصل أكسيد النيتريك ومصل حامض سياليك وأخيرا جلوتاثيون مختزل في نسيج الكبد. وأوضحت الدراسة أن استخدام زيت جنين القمح كماده واقية مضادة للأوكسدة ومضادة للالتهابات كان لها دور فعال في حماية أنسجة الكبد من التسمم البكتيري المحدث تجريبيا في الجرذان باستخدام الاندوتوكسن وأدى استخدامه كذلك الى الحفاظ على نسب القياسات البيوكيميائية في الدم والأنسجة لما يقارب النسب الطبيعية. لذلك توصى الدراسة بضرورة استغلال تلك المزايا الهائلة زيت جنين القمح كماده وقائية وعلاجية ومضادة للأوكسدة والالتهابات.

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