

The protective effect of kefir as a probiotic on reproductive potential of male rats

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ABSTRACT

Nowadays foods are not purposed to only accept hunger and to give vital nutrients for humans, but also to protect and treated nutrition-related diseases and ameliorate physical and mental status of the consumers. In this respect, probiotic play a prominent role. Thence this study aimed to estimate the chemical constituents of kefir and kefir grain and identify the potential protective effect of probiotic (yoghurt/ kefir milk / kefir grain) to diminish the risk of lead (Pb) intoxication on male rats. Biological study was administered on forty two male rats which allotted into 6 groups involved negative control group (7 rats) , normal group consumed kefir milk and four groups which treated 50 mg of lead/kg body weight per day as testicular toxicant and classified into positive control group (+ve) , subgroups : yoghurt, kefir milk and kefir grain subgroups. The results revealed that all experimental treatments enhanced liver and kidney functions , antioxidant parameters and testosterone hormone levels, luteinizing hormone (LH) and follicle stimulating hormone level (FSH) the most prominent positive effects were perceived in the group that consumed kefir milk and kefir grain. These acquired results are assured by the histopathological examination. In the present study, the protective effect of kefir milk and kefir grain against lead toxicity in rats was become well. Therefore, this study recommends that consume kefir and supplemented the products with kefir grains to prevent the damage caused by lead contamination.

Key words: *probiotic- yoghurt- kefir milk –kefir grain – lead acetate-Sexual Toxicity –antioxidant.*

INTRODUCTION

Probiotics is marketed as functional foods, where they are consumed for their positive benefits in the digestive system and systemic areas such as vagina, liver or bloodstream. probiotics vie with microbial that can cause disease for restricted number of receptors existent on the surface of the intestinal epithelium (Martta et al., 2007 and Kengang et al.,2014) and steadiness of intestinal penetrability wall, therefore limitation existence of pathogens and removing strange antigens by producing short fatty acids to feed the cells lining of the colon, lifting the acid in the colon, and producing antibiotic-like fluids that act as a protective barrier against harmful bacteria, and improve the immunity system (Markowiak and zewska , 2017).

Probiotics is living microorganisms, on being utilized in sufficient quantity, Benefits are granted on healthiness host like enhancement of the capacity intestines and amelioration of the

lactose sensitivity and decrease the value of lipids in serum. The cells of the intestinal bacteria can bind by cholesterol and bile acids, which inhibits the absorption of total cholesterol and reduces its level in the blood. The bacterial cells actively process the bile salts and release about half of the bile from their salts that reach the small intestine. This is followed by an increase in the output of bile salts during the waste, which in turn reflects the improvement of lipid rate in the blood (Kumar et al. 2012). Yoghurt and kefir were considered as functional foods containing probiotics. Protein, phosphorus, calcium, thiamin (vitamin B1), riboflavin (vitamin B2), niacin, and vitamin B12, are found in yoghurt at an excellent rate, Yoghurt is a valuable source of folic acid, zinc and magnesium (Madhu et al., 2013).

Kefir is potential antioxidants which react directly with an extensive variety of species responsible for oxidative damage. The antioxidant capacity of kefir may be attributed to their proton- contribute capacity, their

decreasing force and superoxide dismutase (SOD)-like action as represented through 1,1-diphenyl-2-picrylhydrazyl (DPPH) and superoxide radical searching and lipid per-oxidative restraint comes about. Accordingly, kefir is probably the most important dietary supplement because of its effective role as a natural antioxidant for humans. (Nurliyani et al., 2015).

Lead is a non-essential element that found in nature. It is known that heavy metal ions cause a great risk to the environmental system in very low doses on account of their high toxicity stability and accumulation and transfer in food chains considered among the main environmental pollutants, along with mercury and cadmium, which is one of the most harmful effects on human (Haryanto, 2016). Sub-chronic oral lead intoxication may affect testes and body organs for numerous years even in absenteeism of continual exposure (Adhikari et al., 2001).

Accumulation of lead in tissues was due to cellular damage by oxidative stress after excessive formation of reactive oxygen species and decreased in the activities of the cellular antioxidant system (Gandhi et al., 2017). Lead exposure related with decreased enzyme activities in testis and disruption of sperm formation and histoarchitecture (Boscolo et al., 1988). In addition, BaSalamah et al., (2018) found that rats which chronically the exposed to Pb, showed a significantly increase in tissue contents of the heavy metal that concur with significantly reduce in rate of 25-OH VD and Ca²⁺ in serum .

Co-administration of probiotics alone or with dandelion extract has the ability to prevention of testicular toxicity induced by arsenic in rats and improved kidney and liver functions, while the dandelion extract alone did not occur this (Al-Damegh et al., 2014). Rosa et al., (2014) demonstrated that kefir supplementation with high and normal dose(3.5&.7 ml) for 1

month in Wister rats did not observe injurious effects in growth, hematology, and blood chemistry in rats, thus consumption of kefir is safe. Milk kefir showed marked curative benefits in HCV patients by lowering viral load, mitigating the altered liver function and amelioration of lipid profile. The ameliorative effects of milk kefir in HCV patients may be due to its antioxidant, anti-inflammatory and immune-stimulatory effects (Morsy et al., 2014).

Lead is a toxic heavy metal that raise the level and freeing of free radicals and reduces the activity of the enzymatic and non-enzymatic antioxidant system to respond to oxidative stress. The goal of this study was to explore the protective effect of kefir against the effect of lead on oxidative stress and damage in rat testis. Because of, probiotic has the ability to act as antioxidants; also, it is the main source of calcium and vitamin D. Previous studies noted that the role of calcium and vitamin D in the removal of toxicity resulting from

lead. The researchers had observed that preventive effect for vitamin D against Pb-induced testicular and renal damage by antioxidative and anti-inflammatory mechanisms (BaSalamah et al., 2018).

MATERIALS & METHODS

Materials:

Forty two male albino rats of Sprague Dawley strain were bought from Helwan ranch of experimental animals, Helwan, Egypt. Mean weight of rats ranged from 110 to 115g. The basal diet was implemented in accordance with (NRC, 1995). Fresh cow's milk was taken from the herd of the dairy cattle at Faculty of Agriculture, Mansoura University, and Mansoura, Egypt. Lead acetate, all chemical Diagnostic kits were obtained from El-Gomhoria Company, El-Mansoura city, Egypt.

Cultures: Yoghurt starter culture (YC-X11 DIP 50u) Lactobacillus delbrueckii subsp contains and Streptococcus thermophilus. bulgaricus was gotten from Chr. Hansens

Laboratories, Denmark and activated at 42°C utilizing 12% sterilized reconstituted skim milk. After incubation at 42°C for 4-5 h, the obtained working culture was freshly utilized. Kefir grains were kindly provided by the Food Technology Research Institute, Agricultural Research Centre, Giza, Egypt.

Methods:

Kefir preparation

Kefir grains were cleaned with distilled water and vaccinated in whole cow's milk during incubation at room temperature. The kefir milk was made by inoculation of (2% w/v) kefir grains into pasteurized milk (milks heated to 95 °C for 15 min and immediately cooled to 23 °C) and incubated at 24 °C for 24 hours, then the grains were taken away from the fermented milk by filtration with a plastic sifter, washed, and kept for following preparation (Kefir was added at different concentrations and incubated for several hours. Then, the concentration and number of incubation hours were selected

according to the lowest percentage of alcohol produced by fermentation). This procedure was reshaped day by day all through the 45 day of the trial time frame.

Production of Yoghurt

Cow's milk (protein 3.72%, fat 3%, total solids 12.3% and acidity 0.17%) was utilized then heated to 95°C for 15 min. and after that quickly cooled to 45°C. The working yoghurt culture was added at the percentage of 2% (w/v). The inoculated mix was packed into 2.0 kg plastic cups and incubated at 43°C. The incubation was finished at pH 4.5. At this point, the yoghurt was stored in a refrigerator (5±1°C).

Analytical Methods:

Approximate chemical compositions of kefir milk and kefir grain (moisture, ash, fat and crude protein) were estimated by the techniques for the (A.O.A.C. 2007). Whereas, total carbohydrates were estimated by subtracting the difference from initial weight of the samples

follows, as: Carbohydrates% = 100 - (% moisture + % protein + % fat +%ash). Total phenols and total flavonoid were estimated by **Slinkard and Singleton (1977) and Zhishen et al.,(1999)** . Alcohol was determined in the fermented milks according to **Caputi et al., (1968)**.

Experimental design:

Animals were kept in the laboratory in plastic cages under constant temperature (24±2 C). All rats were nourished on basal diet only for seven days before beginning the trial for acclimatization. After acclimatization period, Forty two male rats were allotted into 6 groups (7rats/treatment) at random; the negative control (C-) group was fed the basal diet only. Group Normal kefir milk (N.K.M) fed on basil diet plus kefir milk dosage of 1.8 mL for 45 day. The rest of the rats except (7 rats) were administered lead acetate 50 mg/kg rat according to (**Owolabi et al., 2012**). Positive control (C+) group was fed the basal diet in plus lead acetate orally once in a

day for 35 days. While the other 3groups were fed diets plus yoghurt (Y) , Kefir milk (K.M) and Kefir grain (K.G). The rat yoghurt and kefir milk dosage of 1.8 mL/day once in a day for 45 days according to **Punaro et al.,(2014)** . Kefir grain was included at 40g/kg of standard diet according to **Can et al., (2012)**. Lead acetate 50 mg/kg.bw were given on 10th day, one hour after the yoghurt, kefir milk and Kefir grain administration for 35 days.

Growth performance and feeding were noticed by enrollment daily feed intake, body weight gain and feed efficiency ratio (FER) as per **Chapman et al., (1950)** as mean daily body weight gain divided by mean daily food intake .The rats were sacrificed after 45 days blood collected and centrifuged at 3,000 rpm for 15 min to get serum, and testis were compiled for biochemical analysis and histopathological examination.

Biochemical analysis of serum:

Uric acid was assessed in serum as indicated to the method

by **Fossati et al., (1980)**. Urea was assessed according to **Patton and Crouch (1977)**. Creatinine was measured by **Bonsens and Taussky (1984)**. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were measured by **Reitman and Frankel (1957)**. Serum testosterone and follicle stimulating hormone levels (FSH) in the serum were dictated by a catalyst connected immunosorbent measure (ELISA) utilizing business packs as depicted by **Wilke and Utley (1987) and Ballester et al., (2004)**. Luteinizing hormone (LH) was measured by radioimmunoassay as indicated by the strategy portrayed by **Schams and Karg (1969)**.

Detection of oxidative in testes tissue:

Reduced glutathione (GSH) was resolved by **Sedlak and Lindsay (1968)**. Lipid peroxidation (malondialdehyde) was estimated as portrayed by **Ohkawa et al., (1979)**. Superoxide dismutase SOD activity was measured by **Marklund (1985)**.

Histopathological examinations were performed on testicular tissues as described by **Bancroft et al., (1996)**.

Statistical analysis:-

Results obtained from this study have been described as mean \pm standard deviation (mean \pm SD).Data were analyzed using one way classification, analysis of variance (ANOVA) test. The differences between means were tested for significance using least significant difference (LSD) test at <0.05 (**Snedcor and Cochran, 1967**).

RESULTS & DISCUSSION

Moisture, Protein, fat, ash, and carbohydrates of Kefir milk, Kefir grain yoghurt were identified and schedule in Table (1). It was plainly that Moisture , Protein, fat, ash , carbohydrates, total phenol, total Flavonoid, pH and Ethanol were 87.62%, 3.03%, 3.15%, 0.043% , 5.77% , 0.84 mg/100g , 0.16 mg/100g , 4.9 and 0.065 respectively for Kefir

milk while Moisture , Protein, fat, ash , carbohydrates , total phenol and total Flavonoid of Kefir grain were 9.53%, 10.75%, 7.88%, 2.03%.18.71%, 2.03 mg/100g and 0.88 mg/100g respectively. Constituents of kefir milk from fat, protein, carbohydrates and ash was similar to **Hallé et al.,(1994)**, while , **Libudzisz and Piatkiewicz (1990)** noted that kefir milk contain ethanol of 0.035 – 2.000% . A study done by **Garrote et al., (1997)** found that chemical composition of kefir grains were 2 g kg-1 lipid, 30 g kg-1 .protein, 7 g kg-1 ash, 60 g kg-1 sugars and 890-900 g kg-1 water. However, **Libudzisz and Piatkiewicz (1990)** reported that dray mass of fresh grain 10 -16 g/100g, 30g/100g protein and 25-50g carbohydrate. Thence, this variation in values obtained can be attributed to the different kefir species.

Data in table (2) illustrated that, rats in normal control and normal plus kefir milk groups had a significant increase in values of body weight gain ,feed intake

and feed efficiency ratio compared to rats intoxicated by lead acetate control (+ ve) and probiotic groups; yogurt, kefir milk and kefir grain. A probiotic groups showed a significant higher values of the feed intake ,body weight gain and feed efficiency ratio compared to control (+ ve) group. The results of the other studies showed that treatment with lead acetate caused decrease in the growth rate in experimental animals when lead acetate was orally supplementation , which has adverse effects on body weight gain and feed intake and liver function experimental animals (**Ali et al., 2010; Seddik et al., 2010**). Lead acetate causes dysfunction in the absorption and metabolism of some important nutrients so that might be the cause of body weight loss (**Marija et al., 2004**). Comparable results were likewise reported by **Ibrahim et al., (2012)**, " Lead causes lowering in growth rate for rats because of could be related by some factors, one of that is imbalance metabolism created by impairing zinc status in zinc-

dependent enzymes which are vital for many metabolic processes" .Whilst, **Goni,et al., (2016)** explained that whole milk might be useful in reducing the body burden of lead toxicity.

From results in table (3), it could observe that there was significant improvement in uric acid, urea and creatinine in all groups that feed on probiotics to study their protective effect., Uric acid, creatinine, , urea level AST and ALT were significantly elevated in control (+ve) group comparing with normal control group. However protective effect group with yogurt, kefir milk and kefir grain significantly reduced these parameters compared to positive group. It appears from the present results that kefir milk and kefir grain were safe and improves renal and liver functions.

Daily consumption of 500 mL of kefir was reported to stabilize metabolism, positively effect on liver, gallbladder, and renal function, and blood circulation (**Liu et al., 2006**). Studies have resulted that kefir was more

effective than vitamin E by increasing levels of glutathione, glutathione peroxidase and reduced lipid peroxidase in the rat model of injury oxidation caused by CCl₄ (**Güven and Gülmez.,2003**) .

Çenesiz et al., 2008 observed the positive biochemical effects of kefir on oxidative stress; the antioxidant effect on the formation of abnormal coldness in the colon caused by azoxymethane was higher in rats consumption kefir. **Alsayadi et al., (2013)** assured that water kefir have antioxidants action it a best scavenger of free radicals from body. For that, it is possible that kefir can restrain the degradation of the mitochondrial membrane,

Mogheth et al., (2017) found that kefir was characterized by protection against hepatic toxicity caused by CCl₄ and an antimicrobial activity against pathogenic microorganisms. These protective effects were demonstrated through its anti-inflammatory effect and inhibition of CCl₄ movement and improved

liver capacity. In this way, kefir may be applied in clinical application in the avoidance as well as treatment of liver danger. Probiotic supplements have attracted the attention of the researchers' consequent to its prospective to detoxify the gastrointestinal tract. A study done by **Imani Fooladi et al., (2013)** reported that healthy status of the gastrointestinal tract is in a symbiotic relation with that of the liver. This is consequent to the vital role by the microflora resident of the intestinal lumen to the function of the hepatocytes, in which alterations to per se may reason liver dysfunctions.

Results in table (4) indicated that administration of lead acetate in positive control group showed significant diminishing in serum testosterone (T), Follicle Stimulating Hormone (FSH) and luteinizing hormone (LH) compared to (-ve) control group. All groups that feed on probiotics to study their protective effect (yoghurt, kefir milk and kefir grain) had a significantly elevated in serum testosterone,

FSH and LH levels compared with the positive control group.

Lead reproductive toxicity may be associated with the extreme creation of Reactive Oxygen Species (ROS), an issue that has received more attention lately. ROS, inhibits enzyme reactions, inhibits the creation of sulfhydryl antioxidants, destroys nucleic acids and hinders DNA repair, and additionally, starting lipid peroxidation in cellular membranes. Lead promotes the generation of hydrogen peroxide and induces oxidative stress (**Vaziri and Khan 2007**).

These information might be because of the impacts of oxidative stress and dopamine on male sexual capacity, the larger part of microorganisms demonstrate cancer prevention agent conduct, wiping out the abundance oxygen free radicals and creating superoxide dismutase, or glutathione (**Songisepp et al., 2004**). The yogurt microorganisms *Lactobacillus delbrueckii* and *Streptococcus thermophiles*

repressed peroxidation of lipids by rummaging the responsive oxygen radicals, for example, hydroxyl radical, or hydrogen peroxide (Ling and Yen 1999). BaSalamah et al., (2018) confirmed that vitamin D3 significantly minify Pb-induced tissue damage in testis and kidney. Tissue regeneration or safeguard was related by significant raise of anti-oxidative indications. Henceforth, it can be hypothesize that yoghurt and kefir treatment can offer protection against pb-induced oxidative stress in vivo because of probiotic was rich source of vitamin D.

Results in table (5) showed that oral administration of lead acetate in control(+) group revealed significant reduction in superoxide dismutase (SOD) glutathione (GSH), and increase in malondialdehyde level compared to (-ve) control group. All groups that feed on probiotics to study their protective effect (yogurt, kefir milk and kefir grain) enhanced the active of testicular (SOD), malondialdehyde and (GSH) antioxidant enzymes

compared to positive control group. These outcomes were in concurrence with **Ozcan et al., 2009**, they reported that on supplementing diet by kefir with or without lead, (SOD) and (GSH) levels became resembles control group and Vit. E raised by kefir supplementation. Accordingly, the oxidative damage induced by lead and supported oxidative system may be indeed preventing by kefir.

Vitamins A , conjugated linoleic acid (CLA), B-carotene , vit.E and coenzyme Q10 are the main antioxidants in milk fat and other compounds with antioxidant characteristics comprise vitamin D3, phospholipids and probably, 13-methyl-tetradecanoic acid (**German and Dillard 2006; Baldi and Pinotti 2008**). Vitamin E (a-tocopherol) and conjugated linoleic acid (CLA) are the more active antioxidants in milk fat. The antioxidant strength of vitamin E, shown by its capability to deactivate singlet oxygen, is reinforced by B-carotene and vitamin A which are most active than vitamins E and C. Vitamins A, E and D3 are properties by

high levels of thermal stability and they stay active in each dairy products without of the utilized thermal treatment method (Claeys et al., 2014). Mustafa and Abdullah (2009) concluded that probiotic decreased lead acetate absorption and reduced its toxic action on studied parameters in chicks.

The results obtained in table (3, 4&5) referred to it is possible to note that there is a link between dysfunction in the liver and kidney function and also the lack of hormone testosterone and can be consistent with Sinclair et al., 2015 said that in men with cirrhosis, decline in testosterone value in parallel with the severity of liver disease deterioration. Testosterone levels (T) are generally low in men with advanced liver disease, and gradually decrease with increased acuteness of liver disease (Handelsman et al.,1995) various clinical symptoms of advanced liver disease, like as anemia, sarcobinia, bone disease, and gynecomastia are also a phenomenon in gonadal

insufficiency, thence, decrease in T levels may has recently been specified as an independent prophetic indication in cirrhosis(Grossmann et al., 2012), moreover, liver diseases in human are related with several hormone disorders, like as decreased serum levels of cortisol, T3, FSH , testosterone and insulin in males (Barreca et al., 1983; Kolster et al., 1990), thence , this study recommends further studies illustrating this relationship.

Histopathological Results:

Results obtained from this study were assured by the histopathological examination. Pict.(1) shows that the testis of rat from the control (- ve) group showed normal seminiferous tubules normal spermatogenic cells and the tubules contain mature spermatid. Meanwhile, the testis of rat from the control (+ve) group was exhibiting destruction of some spermatogenic cells of the seminiferous tubules with interstitial edema as shown in Pict. (2) the testis of normal rat from kefir milk group revealed normal

structure of seminiferous tubules as shown in Pict. (3). Testis of rat from yogurt group showing, few degeneration changes in the spermatogenic cells with separation of the seminiferous tubules as shown in Pict. (4).The testis of rats from kefir milk revealing normal spermatogenesis with few reductions of mature sperm in seminiferous tubules as shown in Pict. (5). the testis of rats from kefir grain showing activation of spermatogenesis and mature spermatid as shown in Pict. (6). Lead occurred testicular damage in rats; This is supported by **Jegede et al., (2015) and Anjum and Reddy (2015)**, in which they stated ,Chronic consuming of Pb led to interstitial degeneration and reduce numbers of viable Sertoli, Leydig and germ cells simultaneously, that shown in testicular tissue with significantly decrease serum testosterone. This might be due to the ability of Lead ions to generate reactive oxygen species competent of soft tissue break-down according to **(Dare et al., 2012)**. Effect of lead toxic on

testes is attributed to the output of extreme reactive oxygen species (ROS), **(Mariola et al., 2004)**. These results coped with **Aprioku and Obianime (2014)** they noted that toxicity pb led to atrophy in testis and ultimately affected on their physiological function. The improvement in histopathological examination results of groups which consumed probiotic agree with **(Amdekar and Singh, 2016)**, they found that the intake of *Lactobacillus acidophilus* would protect the testis from damage caused by oxidation. The treatment of *Lactobacillus acidophilus* can be detected by removing the free radicals that have been created, thus it has the ability to protect the testis from oxidative damage. This improvement in histopathological results can be attributed to probiotics and its contains of vitamin A and D and E, and its effective role as a natural antioxidant, is congruent with this notion **Momeni et al.,2012** for they found that vitamin E has the ability to improving the histopathological changes caused

by sodium arsenite in the testis , vit. E alone promoted the height and the average of diameter of seminiferous tubules and decreased the average diameter of seminiferous tubule's lumen in comparing by normal control animals. Is consistent with this notion, vitamin E is called an anti-sterility vitamin

CONCLUSION

Present study indicates that pb-induced testicular toxicity in male rats. The pretreatment of pb-intoxicated groups with kefir restored the lipid peroxidation, the antioxidant enzymes status and the histoarchitecture of pb-damaged testes. The protective effect of kefir may be because of its antioxidant property and detoxification capacity.

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Table (1): Chemical composition (g/100g) of Kefir milk and Kefir grain

Sample	Moisture %	C. protein %	T. lipid %	Ash %	T. Carb. %	T. phenol mg/g	Flavonoid mg/g	PH %	Ethanol %
Kefir milk	87.62	3.03	3.15	0.043	5.77	0.84	0.16	4.9	0.065
Kefir grain	9.53	10.75	7.88	2.03	18.71	2.03	0.88	---	-----

Table (2): Effects of yogurt, kefir milk and kefir grain supplementation on biological parameters on rats intoxicated by lead acetate

Groups Variable	Weight gain (g)	Feed intake (g/d)	FER
C-	55.80a±2.49	12.70a±0.51	0.10a±0.002
N.K. M	55.36a±2.23	12.74a±0.32	0.10a±0.002
C+	19.20d±2.17	7.80c ±0.39	0.05e±0.005
Y	37.97c±0.81	13.18a±0.31	0.06d±0.002
K. M.	39.80c±3.27	11.82b±0.41	0.07c±0.005
K.G.	46.80b±3.19	11.86b±0.40	0.09b±0.005

Values in each column showed as average having different letters (a, b,...) were significant .

C-: Normal control

N.K. M: Normal+ kefir milk **C+**: positive control

Y: yogurt

K. M.: kefir milk

K.G.: kefir grain

Table (3): Effects of yogurt, kefir milk and kefir grain supplementation on kidney functions and liver functions of rats intoxicated by lead acetate

Groups Variable	Uric acid mg/dl	Creatinine mg/dl	Urea mg/dl	AST (U/ml)	ALT (U/ml)
C-	1.97 ^c ±0.12	0.58 ^c ±0.02	29.12 ^c ±0.81	27.17 ^d ±1.55	17.83 ^d ±1.61
N.K. M	1.93 ^c ±0.18	0.58 ^c ±0.06	29.20 ^c ±0.69	27.05 ^d ±2.54	17.75 ^{d±} 1.39
C+	3.81 ^a ±0.33	3.42 ^a ±0.57	47.47 ^a ±2.35	55.48 ^a ±2.02	36.22 ^a ±2.41
Y	2.65 ^b ±0.47	1.70 ^b ±0.34	32.91 ^b ±1.68	48.42 ^b ±7.00	26.52 ^b ±2.07
K. M.	2.45 ^b ±0.31	1.47 ^b ±0.41	29.55 ^c ±1.06	32.10 ^c ±3.81	20.58 ^c ±1.00
K.G.	2.06 ^c ±0.11	0.66 ^c ±0.10	29.84 ^c ±0.86	27.50 ^{cd} ±2.22	19.73 ^{cd} ±1.83

Values in each column showed as average having different letters (a, b,...) were significant

C-: Normal control

N.K. M: Normal+ kefir milk **C+**:positive control

Y: Lead acetate + yogurt

K. M.: Lead acetate + kefir milk

K.G.: Lead acetate + kefir grain

Table (4): Effects of yogurt, kefir milk and kefir grain supplementation on sexual hormonal parameters on rats intoxicated by lead acetate.

Groups	FSH ng/mL	LH ng/mL	T ng/mL
Variable			
C-	155.60ab ±6.43	4.54a ±0.11	25.52ab ±0.43
N.K. M	160.88a ±5.60	4.58a ±0.04	26.54a ±2.29
C+	101.46e ±1.48	1.41 e ± 0.03	11.26e ±0.46
Y	122.94d ±1.90	2.99d ±0.04	16.83d ±0.41
K. M.	138.69c ±0.44	4.01c ±0.03	22.07c ±0.63
K.G.	149.39b ±0.41	4.19b ±0.29	24.97b ±0.12

Values in each column showed as average having different letters (a, b,...) were significant

FSH: Follicle Stimulating Hormone level

LH: Luteinizing hormone T: testosterone.

C-: Normal control

N.K. M: Normal+ kefir milk **C+:** positive control **Y:** Lead acetate +yogurt

K. M.: Lead acetate +kefir milk

K.G.: Lead acetate + kefir grain

Table (5): Effects of yogurt, kefir milk and kefir grain supplementation on antioxidant parameters on rats intoxicated by lead acetate.

Groups	MDA (nmol/g)	SOD (u/g)	GSH (nmol/g)
Variable			
C-	30.20c ±1.12	2.42b ±0.24	1.90bc ±0.24
N.K. M	28.19d ±1.25	3.02a ±0.41	2.32a ±0.30
C+	43.74a ±1.93	0.83d ±0.03	0.91e ±0.04
Y	36.36b ±1.42	1.83c ±0.19	1.33d ±0.04
K. M.	30.30c ±1.18	2.36b ±0.34	1.74c ±0.19
K.G.	30.92c ±1.97	1.89c ±0.14	2.04b ±0.24

Values in each column showed as average having different letters (a, b,...) were significant

MDA:- Malondialdehyde **SOD**:- Superoxide dismutase **GSH**:- Glutathion

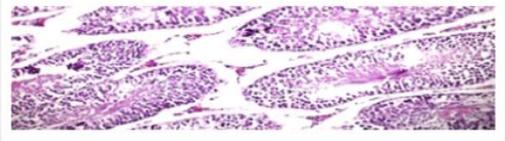
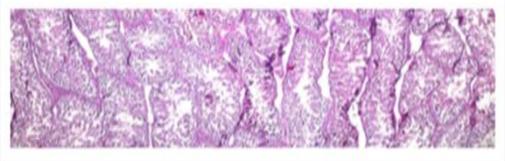
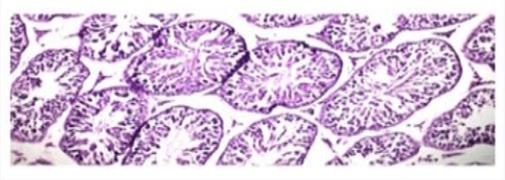
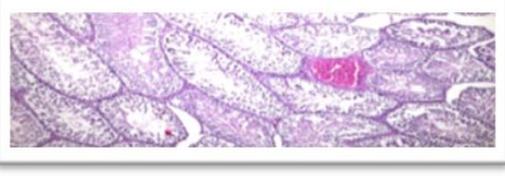
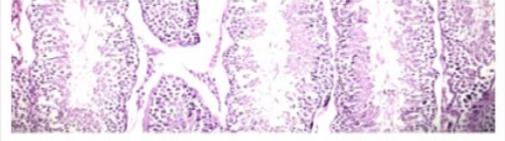
C:-Normal control

N.K. M: Normal+ kefir milk **C+**:positive control **Y**: Lead acetate + yogurt

K. M.:- Lead acetate + kefir milk

K.G.:- Lead acetate + kefir grain

Histopathological Examination of the Testes

<p>Pict.(1) the control (-ve) group showing normal seminiferous tubules, normal spermatogenic cells and the tubules contain mature spermatid.</p>	
<p>Pict.(2) the control (+ ve) group exhibiting destruction of some spermatogenic cells of the seminiferous tubules with interstitial edema. H & E X 200</p>	
<p>Pict.(3) normal rat from kefir milk group showing normal seminiferous tubules, normal spermatogenic cells and the tubules contain mature spermatid.</p>	
<p>Pict.(4) yoghurt group showing, few degeneration changes in the spermatogenic cells with separation of the seminiferous tubules. H & E X200</p>	
<p>Pict.(5) kefir milk group revealing normal spermatogenesis with few reduction of mature sperm in seminiferous tubules H & E X 200</p>	
<p>Pict.(6) , kefir grain group showing activation of spermatogenesis and mature spermatid tubules H & E X 200</p>	

التأثير الوقائي للكهفير كبروبيوتيك علي القدرة التناسلية في ذكور الجرذان

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الملخص العربي

ليس الهدف من الغذاء اليوم اشباع الجوع وتوفير العناصر الغذائية الضرورية للإنسان فقط ، ولكن أيضا للوقاية و العلاج من الأمراض المرتبطة بالتغذية وتحسين الحالة الجسدية والعقلية للمستهلكين. ففي هذا الصدد، اصبح للبروبيوتك دورا بارزا. وقد تم اجراء هذا البحث بهدف تقدير المكونات الكيميائية للبن الكهفير و حبيبات الكهفير وكذلك التعرف التأثير الوقائي المحتمل للبروبيوتيك (حليب الزبادي / لبن الكهفير / حبيبات الكهفير) للحد من خطر التسمم بالرصاص على ذكور الجرذان . وقد أجريت الدراسة البيولوجية على اثنين وأربعين من جرذان التجارب قسمت الى ستة مجموعات هم المجموعة الضابطة السليمة (٧ فئران تم تغذيتهم على الغذاء القياسي طول مدة التجربة) ومجموعه سليمة تتناول الوجبة القياسية مع لبن الكهفير واربعة مجموعات اخري من الجرذان تتناول ٥٠مجم اسيتات رصاص لكل كيلو جرام من وزن الجرذان منها المجموعة الضابطة الموجبة و الزبادي ولبن الكهفير وحبوب الكهفير. أظهرت النتائج المتحصل عليها تحسن ملحوظ في وظائف الكبد والكلية والنشاط الإنزيمي المضاد للأكسدة وكذلك بالنسبة لمستوى الهرمونات الذكرية التستوستيرون LH ,FSH لدي جميع مجموعات الدراسة مقارنة بالمجموعة الضابطة الموجبة وخاصة في مجموعة لبن الكهفير وحبوب الكهفير وقد تم تأكيد هذه النتائج من خلال الفحص الهيستوباثولوجي وقد اتضح من هذه الدراسة ،الدور الوقائي لحليب الكهفير وحبوب الكهفير ضد السمية الجنسية الرصاص في الجرذان. أوصت الدراسة بضرورة تناول الكهفير وتدعيم بعض المنتجات الغذائية بحبوب الكهفير نظرا لأهميته التغذوية ونشاطه المضاد للأكسدة وذلك لمنع الضرر الناتج عن التلوث بالرصاص بحوالي ٢٠٠ مل من حليب الكهفير وحبوب الكهفير ٣٠ جم يوميا .

الكلمات المفتاحية: الكهفير – حبوب الكهفير – الزبادي – البروبيوتيك - أسيتات الرصاص – السمية الجنسية