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Comparative studies on the effect of some antioxidants on renal dysfunction in rats

Hewaida A.E.Fadel *; Essayed A. Hassanin *; Zakaria E. Rihan* *; Eveleen S. Abd Allah ***

Department of Nutritional Chemistry& Metabolism, National Nutrition `Institute*; Department of Endocrinology, Military Medical academy**; Department of Home Economies, Ain Shams Univerersity** **.

Abstract

The present study is carrying out for investigating the effect of supplementation with some vitamins (A, E&C) as natural antioxidant extracts in renal dysfunction in rates.

Material& Methods: 40 adult male Sprague – Dawley rats (150 – 200 g) divided to two groups. First group: 8 rats were fed on standard diet (S.D.), as a control group. Second group: 32 rats were injected intraperitonial with a single dose of Cis-diammine dichloride Platinum II (CDDP) for inducing renal dysfunction (2.5 mg/Kg) then it was divided to six subgroups each one contained 8 rats. (1): fed on (S.D.) nephrotoxic group. (2): fed on (S.D.) + Vitamin A (15 mg/Kg body weight/day). (3): fed on (S.D.) + Vitamin E (317 I.U. /kg body weight/day). (4): fed on (S.D.) + Vitamin C (280 I.U. /kg body weight/day). The experimental period was four weeks, results were statistically analyzed.

Results: The results proved that groups of nephrotoxicity rats supplemented with Vitamin A,E and C showed significant increase in food intakes, body weight gain and food efficiency ratio (FER) (28.9%, 29.4% and 19.2%), (870.5%, 1615.6% and 409.8%) and (652.5%, 122502% and 327.3%) respectively, compared with nephrotoxic group. The nephrotoxicity rats supplemented with Vitamin A and showed significant reduction in serum vitamin E &kidney glutathione content (9.3% & 47.9%), while nephrotoxicity rats supplemented with Vitamin E&C showed significant increase in serum vitamin E &kidney glutathione content (27.9% & 116.6%), (13.7%&55.8%)respectively. The results showed that nephrotoxicity rats supplemented with Vitamin A,E and C showed significant reduction in serum urea nitrogen and creatinine (48.01% and 55.2%), (52.6% and 60.3%) and (57.0% and 63.04%) respectively. Best results in histopathological examination of kidney were in vitamin A and vitamin C groups.

Conclusion: These results suggest that natural antioxidants could be beneficial as additional therapy in renal dysfunction.

Key words: Natural antioxidants - Renal dysfunction – Nephrotoxicity - Histopathological examination – Kidney function.

Introduction

The kidneys are the main organs of the body through which nitrogenous wastes are excrete in the form of urea (**Shubhangini, 2001**) .The basic functional unit of the kidneys is the nephron. Most kidney diseases attack the nephrons. Causing them to lose their filtering capacity. The two most common causes of kidney disease are diabetes and high blood pressure (**Bruce, 2004**). Any disorder in the kidney results in serious complication of the circulatory system, high blood pressure, anemia, weak bones, poor nutritional health, nerve damage and cause complications may not satisfactory consequences (**Perazella, 2006**). A world wide, the number of patients who receiving renal replacement therapy is estimated at more than 1.4 million, with incidence growing by approximately 8% annually. At the year 2025 the kidney failure patients in the world will be 10 million, 70% of them are there in growing countries. In Egypt there are more than 120000 patients suffering from kidney failure. Driving this increase are population ageing, diabetes mellitus and hypertension, the key risk factors for chronic kidney disease (**Sarah *et al,* 2008**). The specialized researches confirmed that

the last 10 years in Egypt have shown a significant increase in the number of children living with kidney failure between the ages of 2 – 10 years and has become representing 15% of patients with kidney failure (**Sarah *et al,* 2008**). Various environmental agents such as (chemical pesticides, solvents and similar materials), animal venom, certain plants and some drugs are nephrotoxic by producing free radicals such as (O2, RO2, OH, NO2, NO) which can cause kidney damage and dysfunction by starting chain reactions that damage cells (**Staci Nix, 2005**).

Antioxidants are molecules capable of slowing or preventing these chain reactions by removing free radicals intermediates and inhibit other oxidation reactions by being oxidize themselves (**Bjelakovie, 2007**). Moreover, **Saravanan and Nalini, (2007**) demonstrated that treatment with antioxidants offers protection against free radical-mediated oxidative stress in kidney of animals with nephrotoxicity. In addition, **Mohamadin *et al,* (2005**) indicated that oxidative stress plays a role in nephrotoxicity and renal dysfunction in rats. Supplementation with antioxidants could be useful in nephrotoxicity in rats.

Athinson *et aI*. (2007) mentioned that vitamin E is an essential nutrient that functions as an antioxidant in the human body. The body cannot manufacture its own vitamin E and thus it must be provided by foods and supplements. It is more appropriately described as an antioxidant than a vitamin. **Pompella *et al,* (2003)** mentioned that Glutathione has been called the "master antioxidant," in addition to its own potent antioxidant powers, glutathione helps to recycle other antioxidants such as vitamins C and E. Thus, glutathione can help to protect against cancer and other diseases caused by oxidative damage. Glutathione also plays an important role in the regulation of immune cells, and is a potent detoxifying agent. Low levels of glutathione have been associated with hepatic dysfunction, kidney dysfunction, immune dysfunction, cardiac disease, and premature.

Aim of the study

This study is carrying out to comparative study of the effect of supplementation with some natural antioxidant extracts in restricting the renal dysfunction in rats through the following parameters:

1- Investigate of some biological evaluations. 2- Antioxidants level. 3- Kidney function

4 - Histopathological examination of kidney

Material and Methods

Forty adult male Sprague – Dawley rats (150 – 200 g) allowed free access to water and standard diet (S.D.) which was prepared according to modified AIN-93-A (**Reeves *et al,* 1993**) for four days, all rats were individually weighed at the start of the experiment and housed in wire cages. The rats were divided to two groups. First group: 8 rats were fed on standard diet (S.D.), as a control group. Second group: 32 rats were injected intraperitonial with a single dose of Cis-diammine dichloride Platinum II(CDDP)for inducing renal dysfunction(nephrotoxicity) (2.5 mg/Kg body weight) was dissolved in physiological saline solution within one hour before injecting according to (**Iseri *et al.,* 2008**) then it was divided to four subgroups each one contained 8 rats. (1): fed on (S.D.) nephrotoxic group. (2): fed on (S.D.) + Vitamin A (15 mg/Kg body weight/day) according to (**Maria *et al.,* 2007**). (3): fed on (S.D.) + Vitamin E (317 I.U. /kg body weight/day) according to (**Tiu Tian *et al.,* 2005**). (4): fed on (S.D.) + Vitamin C (280 I.U. /kg body weight/day) (**Tiu Tian *et al.,* 2005**).

The experimental period was four weeks.

Biological Evaluation: **-Food Intake**: The total diet consumed per group during the period of the experiment was calculated**.**

-Body Weight Gain: It was calculated as follow: Body weight gain (g)=final weight(g) – initial weight (g).

-Food Efficiency Ratio (FER): It was calculated as mentioned by (**Hosoya, 1980**).

-Relative Organs Weight: After animal sacrificed the internal organs (heart, liver, kidney and spleen) were removed and washed in saline. Then the relative weight of organs was determined according to the method described by (**Chapman *et al.,* 1959**). The kidneys were kept in 10% formalin for the histological study,described by (**Janebova and Zima, 1997**).

Antioxidants level:

-Determination of Serum Vitamin E concentration by HPLC according to the method described by (**Janebova and Zima, 1997**).

- Determination of Kidney Glutathione Content: Glutathione (GSH) was determined according to (**Beutler *et al.,* 1963**).

Kidney function :

- Determination of serum Urea Nitrogen: It was determined by urease-colorimetric method described by (**Tietz, 1990**).

- Determination of serum Creatinine was determined by colorimetric method with deproteinization described by (**Tietz, 1986**).

Histological investigation: Histological examination of kidney was carried out according to the (**Drurg and Wsllington, 1980**). All sections were examined in Faculty of Veterinary, Cairo Univeristy, Egypt.

Statistical analysis :The results were analyzed statistically using means and standard deviation (SD) using SPSS/PC program V17 (2008) .T-test and ANOVA were applied in this study for comparison among mean of different groups according to the method described by (**Kurtz,1983**)

Results

Biological Evaluation Results:

Food intake: Table (1) showed that the nephrotoxicity rats exhibited significant decrease in food intake at (P< 0.01) compared with a control group. However, nephrotoxicity rats supplemented with vitamin A& E exhibit non-significant decrease and nephrotoxicity rats supplemented with vitamin C showed a significant decrease in food intakes compared with a control group and a significant increase at (P< 0.01) compared with nephrotoxic group.

Body Weight Gain: Concerning to the body weight gain in the nephrotoxic group, there was a significant reduction at (P< 0.01) by when compared with a control group. Nephrotoxicity rats supplemented with vitamin A,E& C showed a highly significant reduction in % body weight gain at (P< 0.01) compared with a control group. While comparing these groups with nephrotoxic group, there was a highly significant increase at (P< 0.01).

Food Efficiency Ratio (FER): From table (1), it could be observed that (FER) in the nephrotoxic group showed a significant reduction at (P 0.01) when compared with a control group. On the other hand , in vitamin A&C supplemented group there was a highly significant decrease in (FER) at (P< 0.01) compared with a control group and a highly significant increase at (P< 0.01) by comparing with nephrotoxic group. In vitamin E supplemented

group, there was a highly significant increase in (FER) at (P< 0.01) compared with a control group and a highly significant increase at (P 0.01) when compare with nephrotoxic group.

Organs weight: Table (2) showed that in a nephrotoxic group there was a highly significant increase in the hepato somatic , reno somatic and cardio somatic indeces at (P< 0.01), while a significant reduce in the spleeno somatic index at (P< 0.05) compared with a control group. The nephrotoxicity rats supplemented with vitamin A exhibited a highly significant increase in the relative weight of hepato somatic and reno somatic indeces at (P<0.01) by, while a significant reduction in the relative weight of spleeno somatic and cardio somatic indeces at (P<0, 05) in compared with a control group. Comparing this group with nephrotoxic group showed a highly significant reduction in the hepato somatic, reno somatic and cardio somatic indeces at (P<0.01).

Concerning vitamin E&C treatment group, it recorded a significant increase in the hepato somatic index at (P<0.05), in the reno somatic index at (P<0.01) and a significant reduction in the spleeno somatic index at (P<0.05) compared with a control group. Comparing the same group with the nephrotoxic group showed a significant reduction in the hepato somatic, reno somatic and cardio somatic indeces at (P<0.01).

Antioxidant Levels:

Serum Vitamin E and Kidney Glutathione Content:

Data summarized in table (3) showed that nephrotoxic group exhibited a highly significantly reduction in serum vitamin E concentration and kidney glutathione content at (P<0.0l) when compared with a control group.

As showed in table (3), the nephrotoxicity rats supplemented with vitamin A exhibited

a highly significant reduction in serum vitamin E concentration at (P<0.0l) and non-significant increase in kidney glutathione content when compared with a control group. Comparing this group with nephrotoxic group, there were a highly significant increase in serum vitamin E concentration and kidney glutathione content at (P<0.0l)

As illustrated in table (3), vitamin E& C supplemented group showed a significant decrease in serum vitamin E concentration at (P<0.0l) and significantly increase in kidney glutathione content at (P<0.0l) when compared with a control group.
While, these groups showed a highly significantly increase in serum vitamin Eat (P<0.01) and a highly significant increase in kidney glutathione content at (P<0.0l) when compared with nephrotoxic group.

Renal Function Results:
Table (4) showed that serum urea nitrogen and creatinine levels in the positive control group showed a highly significant elevate at (P<0.01) when compared with the control group.
From the same table, nephrotoxicity rats supplemented with vitamin A, E& C showed a significant increase in serum nitrogen and creatinine at (P<0.01) when compared with the control group. While, in comparing with nephrotoxic group showed a significantly reduction in serum urea nitrogen and creatinine at (P<0.01).

Histopathological Results:
As showed in Fig. (1) microscopically examination of kidney of rats from a control group revealed the normal histological structure of renal parenchyma. Meanwhile, as cleared in Fig. (2) Kidney of rats from nephrotoxic group showed vacuolations of epithelial lining renal tubules in the renal cortex. There was a congestion of glomerular tufts, deposition of protein cast in the bowman's space and in the lumen of renal tubules. From Fig. (3) There was presence of eosinophilic protein cast in the lumen of renal tubules as well as per tubular leucocytic cells infiltration. In addition, a cystic dilatation of renal tubules.
Data presented in table (5) and Fig. (4) Showed that (40%) of rat's kidney from vitamin A group revealed normal renal parenchyma. While, Fig. (5) Showed that (60 %) of the same group revealed homogenous eosinophilic protein cast in the lumen of renal tubules. Moreover, 40% of this group revealed focal area of tubular necrosis associated with leucocytic inflammatory cells infiltration.
From table (5), it could be observed that (20%) of rat's renal from vitamin E group showed no histopathological changes except congestion in glomerular tufts and intertubular renal blood capillaries (Fig.6). There was 40% showed protein cast in the lumen of some renal tubules and atrophy of some glomerular tufts. As showed in table (4) and Fig.(7), 80% of the kidney of rats from vitamin E revealed a cystic dilatation of renal tubules with eosinophilic protein cast in their lumen. While, 20% of the same group showed a focal interstitial nephritis.
From the same table and Fig.(8) it could observed that (60%) from vitamin C group revealed no histopathological changes. Meanwhile, (40%) from the same group showed presence of protein cast in the lumen of renal tubules and focal tubular necrosis associated with leucocytic inflammatory cells infiltration. Moreover, it could be noticed in (Fig. (9) that (40 %) of this group showed a hypertrophy and congestion of glomerular tuft associated with nephritis .

Discussion

Biological Evaluation:
Food intake:. The obtained results of nephrotoxic rats exhibited significant decrease in food intake. These results are in the line with (**Armando *et al.*, 2002 & Aaron *et al.*, 2004**) who stated that rats injected with Puromycin Amino nucleoside to induce nephrotoxicity exhibited significant reduction in food intake at (P< 0.05) than control group.
The present results appear to be in harmony with that of (**Maneesh and Jayalekshmi, 2005**) who stated that Ascorbic acid and Alphatocopherol exhibited an ability to counteract the reduction in food intake of nephrotoxicity rats.
Body Weight Gain: The present reduction in body weight gain of the nephrotoxic group was in agreement with (**Armando *et al.*, 2002 and Aaron *et al.*, 2004)** who stated that rats injected with Puromycin Amino nucleoside inducing nephrotoxicity showed a significant reduction in body weight gain.
These results appear to be agree with that of (**Maneesh and Jayalekshmi, 2005**) who stated that Ascorbic acid and Alpha-tocopherol exhibited an ability to counteract the reduction in body weight gain of nephrotoxic rats.
Food Efficiency Ratio (FER): The present results of FER appear to be agree with that of (**Maneesh and Jayalekshmi, 2005**) who stated that the Ascorbic acid and Alpha-tocopherol exhibited an ability to counteract the reduction in food efficiency ratio of nephrotoxic rats.
-Organs somatic index: The present results were agree with (**Iseri *et al.*, 2007**) who cleared that

Cisplatin which induce kidney and liver dysfunction in rats significantly increase kidney and liver weight. Also, these results are in harmony with (**Saad *et al.*, 2007**) who demonstrate that Cisplatin inducing nephrotoxicity in rats, significant increase the relative weight of kidney. In addition (**Stevan *et al.*, 2002)** showed that chronic administration of (PAN) for inducing nephrotoxicity in rats resulted an increase in kidney weight (2.1 g versus control 1.44 g).

These results of organ somatic indeces appear to be in agreement with (**Craven*et al.*, 2007**) who cleared that treatment with vitamin C (10g /kg b.w. /d) in the drinking water significantly reduced the kidney weight compared with untreated diabetic nephropathy rats. Beside, (**Patricia *et al.*, 2005**) who indicated that kidney weight was significantly higher in all of the diabetic nephropathy groups compared with age-matched controls. Supplementation with vitamin C significantly reduced kidney weight compared with that untreated diabetic nephropathy rats. By contrast, kidney weight in rats that treatment with vitamin E was not different from that untreated diabetic nephropathy rats.

Antioxidant Levels:

Serum Vitamin E and Kidney Glutathione Content:

The present results were in harmony with **Saad *et aL*.(2007)** who demonstrated that serum vitamin E and kidney glutathione content were significant reduce at (P<0.0l) in group of rats injected with Cisplatin-inducing neophrotoxicity when compared with control group. In the same time, the results were in agreement with **Duru *et aL*. (2008**) who demonstrated that there were a significant reduction in serum vitamin E and kidney glutathione content at (P<0.05)in rats injected with Cyclosporine A (CSA) for inducing nephrotoxicity when compared with control group.

The results obtained were partially agree with **Atasayar *et aI*, (2009)** who showed that treatment with antioxidants (vitamin E and vitamin C) in nephrotoxicity rats injected by a single dose of Cisplatin prevented the decrease in kidney glutathione content.

In addition, these results partially agreed with that of **(Mehri *et aI.*, 2005)** who showed that treatment with vitamin C didn't show a significant effect on renal tissue glutathione content of Gentamicin-induced nephrotoxicity in rats. While, Vitamin E treatment prevented the GM -induced reduction in renal tissue glutathione content, also co-administration of vitamin C and E significantly prevented the GM-induced nephrotoxicity demonstrating by preservation of GSH level.

***Omar et al.*,(2012)** cleared that Cisplatin (CP) induced decline of antioxidant enzymes and a decreased level of GSH, Vit. C and Vit. E in hepatic tissue and plasma. Treatment with Vit. C, DPPD and L-cysteine in combination with CP restored the content of GSH, Vit. C and Vit. E to about normal control levels.

Kidney Function :

The present study results were in the line with (**Saad *et al.*, 2007**) who demonstrated that serum urea nitrogen and creatinine levels were significant increase in nephrotoxic rats when compared with control group.

These results were in agreement with **Korkmaz and Kolankaya (2009**) who reported that Ascorbic acid (250 mg/kg,i.p.) treatment significantly reduce the serum creatinine and urea nitrogen levels at (P<0,01) in ischemia-reperfusion rats.

In addition, the present study results were in harmony with **(Mehmet *et al.*, 2005)** who demonstrated that intraperitoneal injection of vitamin C (500 mg/kg b.w.) alone or in combination with vitamin A (195 mg/kg b.w.) significantly decrease the level of serum creatinine and urea nitrogen at (P<0.05) in rats givin lipoploysaccharide (10 mg/kg) (LPS)-induced endotoxemia. As well as the results of present work are partially agreement with**(Ocak *et al.*, 2007)** who stated that vitamin E (1000 mg/kg b.w.i.p.) and vitamin C (200 mg/dl in drinking water) administration significantly decrease the blood urea nitrogen and creatinine levels increased by injection of Vancomycin for inducing nephrotoxicity in rats. Beside, **Yanardag *et al*,(2007)** showed the effect of combination of vitamin C (250 mg/mg/kg.bw), vitamin E (250 mg/kg.bw) and Sodum Selenate (0.5 mg/kg.bw) on Ethanol-induced renal injury in rats. Antioxidants treatment significantly reduced blood urea nitrogen and creatinine levels when compared with untreated rats.

Histopathological Results:

The obtained results were agree with **(Tarladacalisir *et aL*., 2008)** who showed that administration of Cisplatin (5 mg /kg /month, i.v.) plus vitamin C (8 mg/kg/day, i.m.) for 3 month, although the structural damages and morphometric changes were lessened, mononuclear cell infiltration was still observed. While, the study

results partially agree with **Niu *et aL*, (2005)** who demonstrated that treatment of Dahl salt-sensitive (SS) rats on a high-sodium intake with vitamin C (98 mg/d) in the drinking water and vitamin E (111 IU/d) in the food for 5 weeks significantly decreased ($P < 0.01$) glomerular necrosis and renal tubulointerstitial damage.

In addition, the results of the present work were in the line with **(Mehmet *et al*., 2005)** who cleared that in endotoxemic rats treated with vit C, the severity of degenerative changes, shrunken glomeruli and especially tubules and mononuclear cell infiltration were less than in endotoxemic untreated rats and endotoxemic rats treated with vit A. In endotoxemic rats treated with vit A and vit C, degenerative changes in the tubules or glomeruli and mononuclrar cell infiltration were not observed.

Beside, **Korkmaz and Kolankaya, (2009)** mentioned that treatment with Ascorbic Acid reversed the histopathological alterations normally induced by ischemia/reperfusion (I/R). In addition, **Ocak *et al*., (2007)** demonstrated that vitamin E (1000 mg/kg i.m.) and C (200 mg/dl in drinking water) were the most beneficial agent on Vancomycin-induced tubular damage.

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TABLE (1): Effect of supplementation with Vitamin A, E, and C on Food intake, Body weight Gain and Food Efficiency Ratio in Nephrotoxicity Rats.

Groups / parameters		CONTROL	Nephropathy	V.A	V.E	V.C
Food intake (gm/day	Mean	9.36	7.21	9.30	9.33	8.6
	± SD	0.11	0.11	0.20	0.07	0.09
	% change from control	_	- 22.98	- 0.66	- 0.28	- 8.16
	P. value	_	* *	N .S	N .S	* *
	% change from nephropathy	_	_	28.97	29.48	19.23
	P. value	_	-	* *	* *	* *
Body weight gain (gm/28 days)	Mean	18.25	1.27	12.37	21.87	6.5
	± SD	0.70	0.07	0.51	0.64	0.75
	% change from control	-	-93.01	-32.19	19.86	-64.38
	P. value	-	* *	* *	* *	* *
	% change from nephropathy	-	-	874.01	1615.6	409.80
	P. value	-	-	* *	* *	* *
Food efficiency ratio (FER)	Mean	0.06	0.006	0.04	0.08	0.02
	±SD	0.003	0.0003	0.001	0.002	0.002
	% change from control	-	- 90.92	- 31.73	20.20	- 61.24
	P. value	-	* *	* *	* *	* *
	% change from nephropathy	-	-	652.56	1225.2	327.31
	P. value	-	-	* *	* *	* *

- Values are statistically significant at * P< 0.05, * * P< 0.01, (N.S) Non-Significant.

TABLE (2): Effect of supplementation with Vitamin A, E, and C on The Organs somatic indeces in Nephrotoxicity Rats.

parameters \ Groups		control	Nephropathy	V.A	V.E	V.C
Hepato somatic index (gm)	Mean	3.37	4.02	3.71	3.49	3.52
	± SD	0.11	0.07	0.06	0.06	0.07
	% change from contror	_	19.28	10.18	3.51	4.43
	P. value	-	* *	* *	*	* *
	% change from Nephropathy	-	_	-7.63	- 13.22	- 12.44
	P. value	-	-	* *	* *	* *
Reno somatic index (gm)	Mean	0.95	1.46	1.04	1.06	0.94
	± SD	0.04	0.06	0.02	0.03	0.02
	% change from control	-	53.68	9.32	11.20	-0.81
	P. value	-	* *	* *	* *	N.S
	% change from Nephropathy	-	-	-28.86	-27.63	-35.46
	P. value	-	-	* *	* *	* *
Spleeno somatic index (gm)	Mean	0.64	0.60	0.61	0.60	0.62
	± SD	0.02	0.02	0.01	0.03	0.01
	% change from control	-	- 6.06	- 4.18	-5.15	- 2.85
	P. value	-	*	*	N.S	N.S
	% change from Nephropathy	-	-	1.99	0.96	3.41
	P. value	-	-	* *	* *	* *
Cardio somatic index (gm)	Mean	0..30	0..33	0..29	0..29	0.30
	± SD	0.01	0.01	0.004	0.01	0.004
	% change from control	-	11.06	-4.63	-5.15	- 0.61
	P. value	-	* *	*	N.S	N.S
	% change from Nephropathy	-	-	-14.11	-14.58	-10.49
	P. value	-	-	* *	* *	* *

- Values are statistically significant at * P< 0.05, * * P < 0.01, (N.S) Non-Significant.

TABLE (3): Effect of supplementation with Vitamin A, E, and C on Serum Vitamin E Concentration and Kidney Glutathione Content in Nephrotoxicity Rats.

Groups / parameters		control	nephropathy	V.A	V.E	V.C
Vitamin E (µ g / dl)	Mean	501.25	371.62	406.37	475.5	422.62
	± SD	3.53	2.5	6.92	1.85	3.42
	% change from control	_	-25.81	-18.87	-5.08	-15.63
	P. value	_	* *	**	**	* *
	% change from Nephropathy	_	_	9.35	27.95	13.72
	P. value	_	_	* *	* *	* *
Glutathione (m g / g tissue)	Mean	14.60	10.21	15.11	22.13	15.91
	± SD	0.99	0.32	1.04	1.77	1.14
	% change from control	_	-30.04	3.52	51.57	9.003
	P. value	_	**	N.S	**	*
	% change from Nephropathy	_	_	47.98	116.66	55.81
	P. value	_	_	* *	* *	* *

Values are statistically significant at * P< 0.05, * * P< 0.01, (N.S) Non-Significant.

TABLE (4): Effect of supplementation with Vitamin A, E, and C on Serum Urea Nitrogen and Creatinine in Nephrotoxicity Rats.

Groups / parameters			Control	Nephropathy	V.A	V.E	V.C
Urea Nitrogen (mg / dl)	Ref Range	Mean	26.51	69.68	36.22	32.9	29.96
	(8 – 27 mg /dl)	± SD	0.88	0.72	0.87	0.82	1.17
		% change from control	_	162.84	36.63	24.09	13.01
		P. value	_	* *	**	**	* *
		% change from Nephropathy	_	_	-48.01	-52.78	-48.50
		P. value	_	_	* *	* *	* *
Creatinine (mg / dl)	(0.6 – 1.3 mg / dl)	Mean	0.64	2.05	0.91	0.81	0.75
		± SD	0.01	0.14	0.02	0.03	0.06
		% change from Control	_	217.21	41.97	25.72	17.21
		P. value	_	**	* *	**	* *
		% change from Nephropathy	_	_	-55.25	-60.36	-63.04
		P. value	_	_	* *	* *	* *

Values are statistically significant at * P< 0.05, * * P< 0.01.

TABLE (5): Effect of supplementation with Vitamin A, E, and C on kidney histopathological in nephrotoxicity rats

Groups / Parameters	control		Nephropathy		V. A		V. E		V. C	
	N	%	**N**	%	N	%	N	%	N	%
Normal histological structure of renal parenchyma**.**	5/5	100	-	-	2/5	40	-	-	3/5	60
Vacuolations of epithelial lining renal tubules in the renal cortex.	-	-	5/5	100	-	-	-	-	-	-
Congestion of glomerular tufts	-	-	5/5	100	-	-	1/5	20	2/5	40
Deposition of protein cast in the Bowman's space and in the Lumen of the renal tubules.	-	-	5/5	100	-	-	-	-	-	-
Presence of Eosinophilic protein cast in the Lumen of renal tubules as well as peritubular leucocytic cell infiltration	-	-	5/5	100	3/5	60	4/5	80	2/5	40
Cystic dilatation of renal tubules.	-	-	5/5	100	-	-	4/5	80	-	-
Focal area of tubular necrosis associated with leucocytic inflammatory cells infiltration.	-	-	-	-	2/5	40	-	-	2/5	40
Focal interstitial nephritis	-	-	-	-	-	-	1/5	20	-	-
Congestion intertubular renal blood capillaries.	-	-	-	-	-	-	1/5	20	` -	-

Fig. (1): Kidney of rat from negative control group showing the normal histological structure of renal parenchyma.

Fig. (2): Kidney of rat from positive control group showing vacuolations of epithelial lining renal tubules in the renal cortex.

Fig. (3): Kidney of rat from positive control group showing presence of eosinophilic protein cast in the lumen of renal tubules and peritubular leucocytic cell infiltration.

Fig.(4): Kidney of rat from vitamin A group showing apparent normal renal parenchyma.

Fig. (5): Kidney of rat from vitamin A group showing homogenous eosinophilic protein cast in the lumn of renal tubules.

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Fig.(6): Kidney of rat from vitamin E group showing congestion of glomerular tuft and intertubular renal blood capillaries

Fig.(7): Kidney of rat from vitamin E group showing cystic diltation of renal tubules with eosinophilic protein cast in their lumen.

Fig.(8): Kidney of rat from vitamin C group showing no histopathological changes.

Fig.(9): Kidney of rat from vitamin C group showing hypertrophy and congestion of glomerular tuft associated with nephritis.

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دراسات مقارنة على تأثير بعض مضادات الأكسدة الطبيعية فى الحد من الخلل الوظيفى بكلى الجرذان

هويدا عبد الفتاح فاضل*، السيد عبد الخالق حسانين*، زكريا الباز ريحان،ايفيلين سعيدعبد الله*****
قسم كيمياء التغذية والتمثيل الغذائى- المعهد القومى للتغذية*، قسم الغدد الصماء- الأكادمية الطبية العسكرية**، قسم الأقتصاد المنزلى –كلية التربية جامعة عين شمس***

الخلاصة تهدف هذه الدراسة الى مقارنة تأثير بعض مضادات الأكسدة المستخلصة من مصادر طبيعية فى الحد من الخلل الوظيفى بكلى الجرذان

خطوات الدراسة : أجريت الدراسة على 40جرذ (ذكور) من نوع الألبينو سلالة سبراجيو داولى (150 – 200 جم). تم وضع الجرذان فى أقفاص مجهزة و ظروف صحية مناسبة، و تم تغذيتهم على الغذاء القياسى لمدة 4 أيام بهدف التكيف مع البيئة المحيطة، تم تقسيم الجرذان الى مجموعتين رئيسيتين :
- المجموعة الأولى : (8جرذان) تم تغذيتها على الغذاء القياسى (المجموعة الضابطة).
- المجموعة الثانية : (32 جرذ) تم اصابتهم بخلل وظيفى بالكلى عن طريق الحقن بالغشاء البريتونى بمادة Cis-diammine dichloride platinum6 (2.5 ملجم/ كم من وزن الجسم)، ثم تقسيم هذة المجموعة الى

مجموعات فرعية ،يحتوى كل منها على (8 جرذان) كما يلى :

1- مجموعة تم تغذيتها على الغذاء القياسي (المجموعةالمصابة بخلل وظيفى بالكلى).
2- مجموعة تم تغذيتها على الغذاء القياسي + فيتامين (أ) (15 ملجم/ كجم/ اليوم).
3- مجموعة تم تغذيتها على الغذاء القياسي + فيتامين (هـ) (317 وحدة دولية/ كجم/ اليوم).
4- مجموعة تم تغذيتها على الغذاء القياسي + فيتامين (ج) (280 ملجم/ كجم/ اليوم).
- استمرت التجربة لمدة 4 أسابيع. تم وزن الجرذان مرتين أسبوعيا لمدة أسبوعين، ثم مرة أسبوعيا لمدة أسبوعين، وكذلك تم تسجيل الغذاء المأكول يوميا.
- فى نهاية التجربة تم تخدير الجرذان و ذبحها و فصل الأعضاء الداخلية (الكبد، الكلى، القلب، الطحال) وغسلها فى محلول ملحى وتجفيفها ووزنها فى الحال. كما تم فصل أجزاء من كلىالجرذان و حفظها فى محلول فورمالين بغرض الفحص الميكروسكوبى.
- أوضحت نتائج الدراسة أن مجموعات فيتامين(أ) و(هـ)و(ج) أظهرت ارتفاعا ذا دلالة احصائية فى قيم المأخوذ من الغذاء و معدل زيادة وزن الجسم و معامل كفاءة الغذاء مقارنة بالمجموعة الضابطة الموجبة.
- كما أوضحت الدراسة انخفاض ذا دلالة احصائية فى الوزن النسبى للكبد و الكلى والقلب فى مجموعات فيتامين (أ)و (هـ)و(ج) مقارنة بالمجموعة الضابطة الموجبة .
- أوضحت نتائج الدراسة ارتفاعا ذا دلالة احصائية فى مستوى فيتامين هـ فى السيرم وجلوتاثيون الكبد فى مجموعات (هـ)و(ج)
- كما أوضحت أيضا انخفاض ذا دلالة احصائية فى وظائف الكلى فى مجموعات فيتامين(أ)و(هـ)و(ج).
- أظهرت نتائج الفحص الميكروسكوبى لانسجة الكلى للمجموعة المصابة بخلل وظيفى بالكلى العديد من التغيرات بأنسجة الكلى بينما مجموعات فيتامين(أ) و(هـ)و(ج) اظهرت انخفاضا ملحوظا في حدة التغيرات بانسجة الكلى
افضل نتائج الفحص الميكروسكوبي لأنسجة الكلى كانت في مجموعة فيتامين (ج) .
هذه النتائج تشير الى ان المواد الطبيعية المضادة للأكسدة يمكن ان تكون مفيدة كعلاج اضافى للخلل الوظيفى للكلى.