Available Online through
www.ijpbs.com (or) www.ijpbsonline.com

IJPBS |Volume 4| Issue 2|APR-JUN|2014|35-46

Research Article

Pharmaceutical Sciences

EFFECTS OF L-CARNITINE ON SERUM LEPTIN AND LIPID PROFILE IN ALLOXAN-INDUCED DIABETIC RABBIT

*** Mohammed Daood Mahmood,** Wahda Basheer Al-Youzbaki,*** Bassam N. Aziz**

***BSc Pharmacist; MSc Pharmacology, Assistant Lecturer, Department of Pharmacology, College of Pharmacy - University of Mosul – Iraq**

****MBChB ; MSc; PhD Pharmacology , Assistant Professor, Department of Pharmacology , College of Medicine - University of Mosul – Iraq.**

***** BSc (Veterinary Medicine), MSc; PhD Physiology, Assistant Professor, Department of Anesthesia, Mosul Technical Institute, University of Mosul – Iraq.**

***Corresponding Author Email: wahdayouzbaki@yahoo.com**

ABSTRACT

Objective: *To evaluate the effect of L-carnitine supplementation on serum leptin, lipid profile and body weight (BW) in alloxan- induced type 1 diabetes mellitus (T1DM) experimental animal model and to assess whether adding L-carnitine to insulin in the treatment regimen of T1DM will have beneficial effects as adjuvant therapy.*

Methodology: *This is a Pharmacological Conventional Non Randomized Trial conducted on animals at the animal room of the health departments of Technical Institute/ Mosul Fertility that belongs to Technical Institute of Mosul / Iraq, from the period of the 15th of April 2011 to the 15th of April 2013. A total of 36 apparently healthy adult male rabbits of a local strains weighing between 980-1450 gm and 4–6 months of age were included in the study and 24 of them were made diabetic via injection of alloxan monohydrate. Twelve healthy rabbits and 24 diabetic rabbits were randomly allocated into 6 groups of 6 animals each, as follows: Group 1 (Healthy control + DW), Group 2 (Healthy + L-carnitine), Group 3 (Diabetes + DW), Group 4 (Diabetes + Insulin), Group 5 (Diabetes + L-carnitine) and Group 6 (Diabetes + L-carnitine + Insulin). The blood samples were collected from all groups three times; Zero time (before induction of diabetes), Stage I (one month after diabetes induction) and Stage II (one month after treatment) to measure serum leptin and lipid profile by commercial kits.* ***Results:*** *This study demonstrated that administration of alloxan (SC) in a dose of (150mg/kg BW) had led to a significant increase in serum levels of TC, TG and LDL-c whereas serum HDL-c, leptin and body weight decreased significantly. Administration of L-carnitine (IM) in a dose of 500 mg/kg BW over a period of 30 days caused a significant decrease in mean serum TC, TG and LDL-c level and significant increase in HDL-c but no significant variations in mean serum leptin and BW in healthy and diabetic rabbits taking L-carnitine in comparison to healthy controls and diabetics tacking DW, respectively. Administration of insulin SC in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days caused a significant increase in serum leptin and HDL-c serum level, but a significant decrease in mean serum TC,TG and LDL-c level in comparison to stage I within the same diabetic animal group. Administration of L-carnitine IM in a dose of 500mg /kg BW combined with insulin SC in a dose of 10 IU to diabetic rabbits caused a more significant decrease in serum TC,TG and LDL-c level and a more significant increase in HDL-c but no significant variations in serum leptin and BW in diabetic rabbits taking the two treatments in comparison to diabetic rabbits taking insulin only.*

Conclusion:

Based on current research it can be concluded that L-carnitine supplementation may have a beneficial effects in ameliorating diabetic complications as an adjuvant therapy to insulin.

KEY WORDS: *carnitine, leptin, lipid profile, body weight.*

INTRODUCTION

Although insulin has became one of the most important therapeutic agents known to medicine to treat diabetes mellitus, the search for compounds with novel properties to deal with the disease condition is still in progress and there is a continuing effort to find insulin substitutes, secretagogues, adjuvant or sensitizers from synthetic or plant sources for the treatment of diabetes[1].

Carnitine is an endogenous amino acid derivative which plays a key role in energy metabolism, produced in the kidneys and liver using amino acids L-lysine and L-methionine, as substrates[2] and derived from meat and dairy products in the diet[3]. Carnitine transports long-chain acyl groups from fatty acids into the mitochondrial matrix, so they can be broken down through β-oxidation to acetyl CoA to obtain usable energy via the citric acid cycle[4]. Carnitine supplementation has been recently approved by the US Food and Drug Administration (FDA) not only for the treatment, but also for the prevention of carnitine depletion, therefore carnitine is considered as essential micronutrient[5].

Patients with diabetes (particularly those who are insulin dependent or have disease-related complications) seem to be at increased risk for carnitine deficiency [6]. The decrease in carnitine level may be attributed to insulin deficiency, excess glucagons[7], increased urinary excretion of carnitine in diabetic patients compared to controls[8]. Moreover, recent evidence suggested that carnitine requirements increase under conditions of sustained metabolic stress such as in DM and had linked carnitine deficiency with developing insulin resistance due to intracellular accumulation of acyl-CoA derivatives[9].

As yet, studies investigating the effect of carnitine supplementation on serum leptin, lipid profile, and body weight in T1DM are very little or absent, not only in the human but also in the experimental animals[10].

The aim of this study was to assess the effect of L-carnitine supplementation serum leptin, lipid profile (TC, TG, LDL-c, HDL-c), and body weight in T1DM experimental animals and to assess whether adding L-carnitine to insulin in the treatment regimen of T1DM will have beneficial effects as adjuvant therapy.

Animals and Methods

The approval of the study protocol by an ethic committee has been obtained from the local health committee of College of Medicine – University of Mosul – Iraq.

This is an Pharmacological Conventional Non Randomized Trial conducted on animals in the animal room of the Health Departments of Technical Institute/ Mosul, Foundation of Technical Education from 15th April 2011 to 15th April 2013. A total of 36 apparently healthy adult male rabbits of a local strains weighing between 980-1450 gm and 4–6 months of age which were obtained from local market and housed in animal room of the health departments of Technical Institute/ Mosul. Animals kept in metal mesh cages (50x50x60 cm), in small (6 rabbits/cage) groups per cage at room temperature and allowed free access to tap water and food (green grasses and laboratory chow). Antihelmintic drug (Ivermectin 2mg/kg) subcutaneously was given to act against internal and external parasites. All the animals were observed for 30 days before the beginning of the experiment to acclimatize under the same laboratory conditions (temperature, light, and humidity) and to exclude any possibility of abnormal behavior and disease[12].

Diabetes was induced in a total of 24 overnight fasted rabbits by injection of alloxan monohydrate which was dissolved in sterile physiological saline solution immediately before being used at a dose of (150mg / kg BW). Alloxan can induce fatal hypoglycemia as a result of massive pancreatic insulin release and to avoid the hypoglycemic shock, the animals were provided with 5% dextrose solution after 6 h of alloxan treatment. Induction of diabetes was tested after 72 h and the animals were allowed one week for the stabilization of blood glucose level. At seventh day, animals having a blood glucose level higher than 300 mg/dl were considered diabetic and used for the for the study. The diabetic state was monitored by periodic tests for hyperglycemia by colorimetric assay along the next 30 days.

After a period of one month, twelve healthy rabbits and 24 diabetic rabbits were allocated into 6 groups of 6 animals each, as follows:

1. Six healthy rabbits were given distilled water (IM) once daily for one month period, served as control group.
2. Six healthy rabbits were given 500mg /kg BW of L-carnitine (IM) once daily for one month period.
3. Six diabetic rabbits were given distilled water (IM) once daily for one month period, served as control group.
4. Six diabetic rabbits were given 500mg /kg BW of L-carnitine (IM) once daily for one month period.
5. Six diabetic rabbits were given 10 IU/Kg BW insulin SC once daily for one month period.
6. Six diabetic rabbits were given both 500mg /kg BW of L-carnitine (IM) and 10 IU/Kg BW insulin SC once daily for one month period.

Blood Sampling were performed three times for all groups as the following:

1. When all groups are normally healthy after one month of acclimatization under the same laboratory conditions (temperature, light, and humidity) and to exclude any possibility of abnormal behavior and disease [11].
2. After one month of establishment of diabetes mellitus taking in consideration the normal physiological changes in the control groups.
3. After one month of drug administration.

Blood samples (5 ml) were drawn after an overnight fasting healthy control and diabetic rabbits. All rabbits were lightly anesthetized with ketamine hydrochloride 30 mg/kg and xylazine 3 mg/kg IM. Intracardial puncture was performed and 5 ml blood was pooled into a clean dry plain tube without anticoagulant. Serum samples obtained were stored at -20oC to be analyzed thereafter for measuring of:

1. Serum TC was measured, using the enzymatic colorimetric method by UV-VIS Spectrophotometer instrument, and TC kit from (BIOLABO), France.
2. Serum TG was measured, using the enzymatic colorimetric method by UV-VIS Spectrophotometer instrument, and TG kit from (BIOLABO), France.
3. Serum HDL-c was measured, using the enzymatic colorimetric method by UV-VIS Spectrophotometer instrument, and HDL-c kit from (BIOLABO), France.
4. Serum LDL-c was measured, using the Friedewald equation[12].
5. Serum Leptin was measured, using the ELISA technique (enzyme linked immunosorbent assay), by Chromate instrument, and Rabbit Leptin ELISA kit (LOT; C0202190666) from CUSABIO, China.

Statistical Analysis: The data obtained in the current study was analyzed using statistical package for social sciences (SPSS) (version 12).Standard statistical methods were used to determine the mean and standard error. Unpaired t-test was used to compare between healthy and diabetic rabbits and healthy control and healthy taking L-carnitne rabbits. One way ANOVA and Post Hoc (Duncan) Test were used to identify statistical difference through comparison within and among groups.

RESULTS

Table (1) shows that administration of alloxan (SC) in a dose of (150mg/kg BW) had led to significant increase in serum levels of TC, TG and LDL-c whereas serum HDL-c, leptin and body weight decreased significantly in the diabetic group compared to the control group.

Table (1): Comparison between mean serum level of measured biochemical parameters between healthy and diabetic rabbits (Stage I)

Parameters	Mean ± SE		P-Value*
	Healthy rabbits (n=12)	Diabetic rabbits (n=24)	
TC (mmol/L)	2.53 ± 0.13	4.29 ± 0.19	0.000
TG (mmol/L)	1.62 ± 0.08	2.55 ± 0.09	0.000
HDL-c (mmol/L)	0.85 ± 0.07	0.52 ± 0.02	0.014
LDL-c (mmol/L)	1.10 ± 0.04	2.60 ± 0.07	0.000
Leptin (pg/ml)	275.94 ± 28.91	142.33 ± 7.07	0.000
BW (gm)	1383.91 ± 21.76	1029.66 ± 17.14	0.018

*** Using unpaired t-test**

Table (2) demonstrates that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male local rabbits, over a period of 30 days (stage II) caused a significant decrease in mean serum TC level in comparison to (Zero time) and stage I within the same healthy animal group (group 2). Results also shows that insulin administration (SC) in a dose of
10 Iu to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TC level in comparison to stage I within the same diabetic animal group (group 4). Furthermore, the administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TC level in comparison to stage I within the same diabetic animal group (group 5). Also, the administration of L-carnitine (IM) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TC level in comparison to stage I within the same diabetic animal group (group 6).

Table (2): Effects of L carnitine with or without insulin on serum TC level in healthy and diabetic rabbits

Group / Stage	Mean ± SE					
	TC level (mmol/l)					
	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)	Group 4 (n=6)	Group 5 (n=6)	Group 6 (n=6)
Zero time before Induction of Diabetes	2.74 ± 0.07	2.71 ± 0.08 **a**	2.69 ± 0.31 **b**	2.71 ± 0.23 **b**	2.74 ± 0.03 **c**	2.75 ± 0.37 **b**
Stage I One month after Diabetes Induction	2.39 ±0.25	2.68 ± 0.07 **a**	4.28 ± 0.36 **a**	4.30 ± 0.43 **a**	4.48 ± 0.01 **a**	4.11 ± 0.61 **a**

Stage II One month after Treatment	2.62±0.23	1.77 ± 0.07 **b**	4.10 ± 0.53 **a**	2.28 ± 0.08 **b**	3.22 ± 0.03 **b**	1.68 ± 0.07 **c**
P-Value*	**NS**	0.025	0.032	0.000	0.000	0.014

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters vertically (a), (b), (c) indicate significant difference.

Table (3) demonstrates that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male local rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TG level in comparison to (Zero time) and stage I within the same healthy animal group (Group 2). Results also shows that insulin administration (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TG level in comparison to stage I within the same diabetic animal group (group 4). Furthermore, the administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TG level in comparison to stage I within the same diabetic animal group (group 5). Also, the administration of L-carnitine (IM) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum TG level in comparison to stage I within the same diabetic animal group (group 6).

Table (3): Effects of L carnitine with or without insulin on serum TG level in healthy and diabetic rabbits

Group / Stage	Mean ± SE TG level (mmol/l)					
	Group 1 (n=6)	**Group 2 (n=6)**	**Group 3 (n=6)**	**Group 4 (n=6)**	**Group 5 (n=6)**	**Group 6 (n=6)**
Zero time before Induction of Diabetes Stage I	1.76 ± 0.13	1.58 ± 0.24 **a**	1.65 ± 0.20 **b**	1.58 ± 0.17 **b**	1.51 ± 0.04 **c**	1.57 ± 0.07 **b**
One month after Diabetes Induction Stage II	1.62 ± 0.07	1.63 ± 0.16 **a**	2.57 ± 0.27 **a**	2.66 ± 0.14 **a**	2.54 ± 0.07 **a**	2.44 ± 0.24 **a**
One month after Treatment	1.86 ± 0.17	0.89 ± 0.03 **b**	2.72 ± 0.27 **a**	1.60 ± 0.18 **b**	2.01 ± 0.03 **b**	1.08 ± 0.03 **b**
P-Value*	**NS**	0.000	0.018	0.000	0.000	0.001

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters vertically (a), (b), (c) indicate significant difference.

Table (4) demonstrates that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male local rabbits, over a period of 30 days (stage II) caused a significant increase in serum HDL-c level in comparison to (Zero time) and stage I within the same healthy animal group (group 2). Results also shows that insulin administration (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant increase in serum HDL-c level in comparison to stage I within the same diabetic animal group (group 4). Furthermore, the administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant increase in serum HDL-c level in comparison to stage I within the same diabetic animal group (group 5). Also, the administration of L-carnitine (IM) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant increase in serum HDL-c level in comparison to stage I within the same diabetic animal group (group 6).

Table (4): Effects of L carnitine with or without insulin on serum HDL-c level in healthy and diabetic rabbits

Group / Stage	HDL-c level (mg/dl) Mean ± SE					
	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)	Group 4 (n=6)	Group 5 (n=6)	Group 6 (n=6)
Zero time before Induction of Diabetes	0.87±0.09	0.85±0.01 b	0.88±0.07 a	0.88±0.09 a	0.93±0.01 a	0.92±0.04 b
Stage I One month after Diabetes Induction	0.86±0.18	0.84±0.03 b	0.61±0.08 b	0.50±0.02 b	0.48±0.02 b	0.49±0.03 c
Stage II One month after Treatment	0.83±0.14	1.17±0.02 a	0.50±0.09 b	0.90±0.10 a	0.79±0.02 a	1.22±0.02 a
P-Value*	NS	0.002	0.019	0.000	0.024	0.000

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters vertically (a), (b), (c) indicate significant difference.

Table (5) demonstrates that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male local rabbits, over a period of 30 days (stage II) caused a significant decrease in serum LDL-c level in comparison to (Zero time) and stage I within the same healthy animal group (group 2). Results also shows that insulin administration (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum LDL-c level in comparison to stage I within the same diabetic animal group (group 4). Furthermore, the administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum LDL-c level in comparison to stage I within the same diabetic animal group (group 5). Also, the administration of L-carnitine (IM) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant decrease in serum LDL-c level in comparison to stage I within the same diabetic animal group (group 6).

Table (5): Effects of L carnitine with or without insulin on serum LDL-c level in healthy and diabetic rabbits

Group / Stage	LDL-c level (mg/dl) Mean ± SE					
	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)	Group 4 (n=6)	Group 5 (n=6)	Group 6 (n=6)
Zero time before Induction of Diabetes	1.08±0.04	1.18±0.05 a	1.05±0.05 b	1.17±0.12 b	1.10±0.04 c	1.23±0.32 b
Stage I One month after Diabetes Induction	1.05±0.05	1.16±0.08 a	2.49±0.17 a	2.77±0.10 a	2.75±0.07 a	2.37±0.13 a
Stage II One month after Treatment	0.93±0.02	0.49±0.04 b	2.51±0.10 a	1.16±0.12 b	1.75±0.08 b	0.42±0.01 c
P-Value*	NS	0.020	0.028	0.002	0.032	0.018

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters vertically (a), (b), (c) indicate significant difference.

Table (6) demonstrates that administration of L-carnitine (IM) alone in a dose of 500mg /kg body weight to healthy male local rabbits, over a period of 30 days (stage II) caused no significant variations in serum leptin level in comparison to (Zero time) and stage I within the same healthy animal group (group 2). Results also shows that insulin administration (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant increase in serum leptin level in comparison to stage I within the same diabetic animal group (group 4). Furthermore, the administration of L-carnitine (IM) in a dose of 500mg /kg body weight to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused no significant variations in serum leptin level in comparison to stage I within the same diabetic animal group (group 5). Also, the administration of L-carnitine (IM) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days (stage II) caused a significant increase in serum leptin level in comparison to stage I within the same diabetic animal group (group 6).

Table (6): Effects of L carnitine with or without insulin on serum leptin level in healthy and diabetic rabbits

Group / Stage	Leptin level (pg/ml) Mean ± SE					
	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)	Group 4 (n=6)	Group 5 (n=6)	Group 6 (n=6)
Zero time before Induction of Diabetes	271.91±51.43	265.46±46.66	274.85±28.10 **a**	276.96±26.63 **a**	286.91±28.12 **a**	266.68±28.96 **a**

Stage I One month after Diabetes Induction	276.25±51.88	275.63±31.40	145.75±11.31 **b**	146.23±41.13 **b**	152.50±11.98 **b**	124.85±16.75 **b**
Stage II One month after Treatment	270.90±18.92	274.73±27.65	157.66±15.23 **b**	248.35±13.03 **a**	154.68±23.36 **b**	247.03±23.48 **a**
P-Value*	**NS**	**NS**	0.021	0.004	0.010	0.001

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters vertically (a), (b), (c) indicate significant difference.

Table (7) demonstrate that L-carnitine (IM) in a dose of (500mg/kg) , over a period of 30 days, caused no significant variations in body weight in healthy rabbits taking L-carnitine (Group 2) in comparison to healthy taking D.W (Group 1). Table (7) shows that insulin administration (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days caused a significant increase in body weight in diabetic rabbits taking insulin (Group 4) in comparison to diabetic rabbits taking D.W (Group 3). Table (7) also demonstrate that L-carnitine (IM) in a dose of (500mg/kg) , over a period of 30 days, caused no significant variations in body weight in diabetic rabbits taking L-carnitine (Group 5) in comparison to diabetic rabbits taking D.W (Group 3). Also, administration of L-Carnitine (I.M) in a dose of 500mg /kg BW combined with insulin (SC) in a dose of 10 IU to alloxan induced diabetic rabbits, over a period of 30 days caused no significant increase in body weight in diabetic rabbits taking the two treatments (Group 6) in comparison to diabetic rabbits taking insulin only (Group 4).

Table (7): Effects of L carnitine with or without insulin on body weights among the study groups (stage II)

Group / Parameter	Mean ± SE						P-Value*
	Group 1 (n=6)	**Group 2 (n=6)**	**Group 3 (n=6)**	**Group 4 (n=6)**	**Group 5 (n=6)**	**Group 6 (n=6)**	
Body Weight (gm)	1592.83±43.68 a	1584.5±29.00 a	837.50±83.71 c	1475.83±39.01 b	884.50±43.18 c	1422.33±21.16 b	0.000

*** Using one way analysis of variance and the differences were calculated by Duncan test**
Different letters horizontally (a), (b), (c) indicate significant difference.

DISCUSSION

The present study showed that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male local rabbits, over a period of 30 days caused a significant reduction in serum TC,TG and LDL-c level and significant increase in HDL-c in comparison to healthy controls. This finding is in agreement with other studies on rats[13] and in human[14].

In contrast, these results disagree with Van Weyenberg et al., (2009)[15] who found that L-carnitine supplementation for 7 days caused no significant effects on TG level and non-esterified fatty acids concentration. These differences in the results may be explained by different routes of L-carnitine administration, since in this study carnitine was administered parentally in a dose of 500mg /kg BW whereas in Van Weyenberg et al., (2009)[15] study carnitine was supplemented in much lower doses (4 g / whole ponies body).

The present study shows that administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced TIDM rabbits, over a period of 30 days caused a significant reduction in serum TC, TG and LDL-c level and significant increase in HDL-c level in comparison to diabetic controls. This is in agreement with other studies[16,17] who found that L-carnitine treatment for streptozotocin (STZ) induced diabetic rats significantly lowered TC and TG levels. The results of the present study also agree with studies on T2DM patients who found that L-carnitine significantly improved the lipid profile[18]. This may be accounted to that the administration of L-carnitine can increase β-oxidation especially in carnitine deficient patients[4].

The present study disagree with the study of Bazotte and Lopes-Bertolini, (2012)[10] who found that carnitine supplementation caused no significant changes in levels of TC, HDL-c and LDL-c but a significant reduction in TG in alloxan induced diabetic rats. This is because of the difference in the dose of carnitine administered which was 200-400 mg/kg/1.day-1 while in this study the dose administered was 500 mg/kg/BW. Also in contrast with Derosa et al., (2003)[19] study who found no significant decrease in serum TG nor in serum HDL-c and with Rahbar et al., (2005) [20] study who even found significant increase in serum TG but no increase of HDL-c in diabetic patients after receiving L-carnitine .This may probably linked to period of carnitine administration, dose, routes of drug administration or the type of the diseased metabolic conditions.

L-carnitine increases β-oxidation to an extent, that the resultant acetyl-coA inter Krebs's cycle and result in more production of energy by burning fats , so increase utilization of cellular fatty acids, and removal of abnormal fat on cellular membrane. In another word the physiological role of carnitine rose to the hypothesis that, this compound could act as a fat burner by optimizing fat oxidation and a consequently reducing its availability for storage[4].

The present study showed that administration of L-carnitine (IM) alone in a dose of 500mg /kg BW to healthy male rabbits, over a period of 30 days caused no significant differences in serum leptin levels in comparison to healthy controls. This finding is in agreement with other study[21] which found that L-carnitine supplementation to rats receiving protein or energy restricted diets has not significantly modified the leptin concentrations. In contrast with our results serum leptin levels were found to be decreased significantly in Iossa et al., (2002) study[3] which may be due to the age related variations in body composition since the reduction in leptin levels was noted in only (24 months) old rats and not in young (2 months) old. While serum leptin level was found to be increased in Hishem et al., (2006) [13] and Van Weyenberg, (2009)[15] studies after carnitine supplementation orally, This difference from our study could be due to different concentrations of carnitine reaching blood according to the differences in the route of administrations because bioavailability of carnitine orally is about 15% [22].

The present study shows that administration of L-carnitine (IM) in a dose of 500mg /kg BW to alloxan induced hyperglycemic rabbits, over a period of 30 days caused no significant variations in serum leptin concentrations in comparison to diabetic controls. This is in line with Ruggenenti et al., (2009)[23] study who investigated the effects of carnitine infusion on leptin levels in T2DM patients and they found that leptin level did not changed appreciably at different time points compared with baseline. In contrast with our results serum leptin levels were found to be decreased significantly in mice after carnitine supplementation[24]. This could be a result of differences in the metabolic status in the studied experimental animal model caused by differences in animal diet nutrition since in the study on mice[24], the mice were fed high-fat diet which is known to induce metabolic disturbances and insulin resistance...

The present study showed that administration of L-carnitine (IM) alone in a dose of 500mg/kg BW to healthy male rabbits, over a period of 30 days caused no significant variations in body weight in comparison to healthy controls. This finding is in agreement with Kordi et al., (2012) study[25] which was conducted on dminis roach (Rutilus rutilus caspicus) juveniles to investigate the effect of dietary L-carnitine supplementation on growth performance and found that giving L-carnitine 500, 1000 and 2000 mg /kg diet, over a 70-day period showed no significant changes in weight gain and weight gain

percentage. Also results in this study are in line with other studies[3,14] which found that L-carnitine supplementation had no demonstrable effect on growth and body weight in animal and human subjects.

In contrast with our results body weight were found to be decreased significantly in some studies[3,13,26] .This difference from our study could be due to different concentrations of carnitine reaching blood according to the differences in the route of administrations because bioavailability of carnitine orally is about 15% [22]. While body weight were found to be decreased significantly other studies[21,27] after carnitine supplementation in animal and human subjects. This differences could be a result of difference in the metabolic status among the study subjects since patients shared in Crill et al., (2006) study[27] were premature infants while those shared in Onbasilar and yalcin (2009) study[21] were on energy restricted diet regimens.

The present study found that administration of L-carnitine (IM) in a dose of 500mg /kg body weight to alloxan induced hyperglycemic rabbits, over a period of 30 days caused no significant variations in body weight in comparison to diabetic controls. This finding is in agreement with Bazotte and Lopes-Bertolini, (2012) study[10] who found that the L-carnitine (200 or 400 mg.kg-1.day-1) supplementation during one or four weeks to non-diabetic and alloxan induced – diabetic rats did not change water and food intake and body weight.

In contrast with our results body weight was found to be decreased significantly in Mun et al., (2007) study[24] which found that carnitine supplementation to high-fat diet mice significantly reduced the elevated body weight during the treatment period. This could be a result of differences in the metabolic status in the studied experimental animal model caused by differences in animal nutrition since in Mun et al., (2007) study[24], mice were fed high-fat diet which is known to induce metabolic disturbances and insulin resistance. The result of this study and other results, which agree with it may be explained by the fact that carnitine supplementation may corrected the carnitine deficiency state encountered in DM and the significant decrease in total fat mass; came with significant increase in total muscle mass with no differences in final body weight [14].

CONCLUSION

L-carnitine IM in a dose of (500mg/kg) , over a period of 30 days, caused a significant decrease in serum TC,TG and LDL-c level and significant increase in HDL-c but no significant variations in serum leptin and body weight in diabetic rabbits. This may have beneficial effects in ameliorating diabetic complications as an adjuvant therapy to insulin. These results may or may not be applicable to humans, so further research is recommended to determine whether similar effect would result in humans or not.

REFERENCES

1. Zhang H, Tan C, Wang H, Xue S, Wang M. Study on the history of Traditional Chinese Medicine to treat diabetes. Europ J Integ Med 2010; 2: 41–46.
2. Steiber A, Kerner J, Hoppel C. "Carnitine: a nutritional, biosynthetic, and functional perspective". Mol Aspects Med 2004; 25 (5–6): 455–473.
3. Iossa S, Maria P, Lilla L, Raffaella C, and Giovanna L: Acetyl-L- Carnitine supplementation differently influences nutrient partitioning, serum leptin concentration and skeletal muscle, mitochondrial respiration in young and old rats. J Nutr 2002; 132: 636-42.
4. Mayes PA, Botham K M, Bender D (2003). Lipid transport and storage, Oxidation of fatty acids, Gluconeogenesis and control of blood glucose .In: A Lange Medical Book Harper's Illustrated Biochemistry. Murry RK, Granner DK, Mayes PA , Rodwell V W (editors), 26th edition ,Torento ,London: pp153-205.
5. Seim H, Eichler K, Kleber H L (2001). Carnitine and its precursor, gamma-butyrobetaine. In: Nutraceuticals in Health and Disease Prevention. Kramer K, Hoppe P, Packer L, eds. New York: Marcel Dekker, Inc. pp: 217-256.
6. Mamoulakis D, Galanakis E, Dionyssopoulou E, Evangeliou A, Sbyrakis S. Carnitine deficiency in children and adolescents with type 1 diabetes. J Diabetes Comp 2004; 18: 271-274.
7. Lee Y, Heo YR, Cha YS. The plasma and urinary carnitine system in Korean diabetic patients. Nutrl Sci 2005; 8: 97-103.
8. El-Hattab AW, Li FY, Shen, J, Powell BR, Bawle, EV, Adams DJ et al., Maternal systemic primary carnitine deficiency uncovered by newborn screening: clinical,

biochemical, and molecular aspects. Genet Med 2010; 12: 19-24.
9. Noland RC, Koves TR, Seiler SE, Lum H, Lust RM, Ilkayeva O et al., Carnitine insufficiency caused by aging and over nutrition compromises mitochondrial performance and metabolic control. Cell Metab 2012 May 2; 15(5): 764-777.
10. Bazotte RB, Lopes-Bertolini G. Effects of Oral L-carnitine and DL-carnitine supplementation on alloxan-diabetic rats. Braz Arch Biol Technol 2012; 55 (1): 81-88.
11. Koolhaas J (1999): The laboratory Rabbits. The UFAW handbook on the care and management of laboratory animals. 7th, ed. Vol. 1: pp 313.
12. Friedewald WT, Levy RJ, Fredrickson DS. Estimation of the concentration of LDL-c in plasma without use of the preparative ultracentrifuge. Clin Chem 1972; 18: 499-502.
13. Hishem NE, El-Zawahry BH, El Nakeeb SMS, Ahmad LA. Influences of short –term aerobic exercise and supplementation of carnitine with or without choline on body weight, serum leptin an carnitine as well as lipid status in male rats. Egyp J Hosp Med 2006; 25: 597– 609.
14. Pistone G, Finocchiaro G, Dell'Arte S, Leotta C, Marino A. Levocarnitine administration in elderly subjects with rapid muscle fatigue: effect on body composition, lipid profile and fatigue. Drugs Aging 2003; 20(10): 761-767.
15. Van Weyenberg S, Buyse J, Janssens GP. Increased plasma leptin through l-carnitine supplementation is associated with an enhanced glucose tolerance in healthy ponies. J Anim Physiol Anim Nutr (Berl) 2009 Apr; 93(2): 203-208.
16. Heo YR, Lee Y, Cha YS. L-carnitine dministration improves lipid metabolism in streptozotocin-induced diabetic rat. Nutr Sci 2002; 5: 3-8.
17. Patel J, Goyal R, Bhatt P. Beneficial effects of Levo-carnitine on lipid metabolism and cardiac function in neonatal streptozotocin rat model of diabetes. Int J Diabet Metabol 2008; 16: 29-34.
18. Derosa G, Maffioli P, Ferrari I, D'Angelo A, Fogari E, Palumbo I, Randazzo S, Cicero AF. Comparison between orlistat plus l-carnitine and orlistat alone on inflammation parameters in obese diabetic patients. Fundam Clin Pharmacol. 2011 Oct; 25(5): 642-651.
19. Derosa G, Cicero AF, Gaddi A. The effect of L-carnitine on plasma LP(a) levels in hypercholesterolemic patients with type 2 diabetes mellitus. Clin Ther 2003; 25: 1429-1439.
20. Rahbar AR, Shakerhosseini R, Saadat N, Taleban F, Pordal A, Gollestan B. Effect of L-carnitine on plasma glycemic and lipidemic profile in patients with type П diabetes mellitus. Eur J Clin Nutr 2005; 59(4): 592-596.
21. Onbasilar I, Yalcin S S. Changes in serum leptin concentrations in rats according to malnutrition models and effects of carnitine supplementation on catchup growth. Revue Méd Vét 2009; 160 (7): 362-369.
22. Rebouche CJ. Kinetics, pharmacokinetics, and regulation of L-carnitine and acetyl-L-carnitine metabolism. Ann NY Acad Sci 2004; 1033: 30-41.
23. Ruggenenti P, Cattaneo D, Loriga G, Ledda F, Motterlini N, Gherardi G et al., Ameliorating hypertension and insulin resistance in subjects at increased cardiovascular risk; Effects of Acetyl-L-Carnitine therapy. Hyperten 2009; 54: 567-574.
24. Mun EG, Soh JR, Cha YS. L-carnitine reduces obesity caused by high-fat diet in C57BL/6J mice. Food Sci Biotechnol 2007; 16: 228-233.
25. Kordi H, Imanpour M R, Sedaghat S. Effect of L-carnitine supplementation on growth performance and carcass composition of caspian roach (Rutilus rutilus Caspicus). World J Fish Marine Sci 2012; 4 (4): 396-399.
26. Ardekani H M, Shevazad M, Chamani M, Aminafshar M, Arani ED. The effect of L-carnitine and low crude protein supplemented with crystalline essential amino acids diets on broiler chickens. Annals of Biolog Research 2012; 3 (2): 1085-1093.
27. Crill CM, Christensen ML, Storm MC, Helms RA. Relative bioavailability of carnitine supplementation in premature neonates. J Parenter Enteral Nutr 2006; 30: 421-425.

***Corresponding Author:**
Dr. Wahda Basheer Al-Youzbaki, PhD
Pharmacology
E-mail: wahdayouzbaki@yahoo.com