BENHA MEDICAL JOURNAL bmj
Print ISSN 1110-208X
Online ISSN 2357-0016

Original article

The Potential Association between Diabetes Mellitus Type 2 and Depression- Role of L-Carnitine (An Experimental Study on Adult Albino Rats)

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Received:
Accepted:

Abstract

Background: Comorbid diabetes and depression are significant public health burdens as the consequences of both diseases are worsened by each other. **Aim of the study**: This work was designed to explore the potential effect of L-carnitine (LC) alone and in combination with paroxetine and dapagliflozin in depression-like behaviour diabetic rats **Material and Methods:** Rats were randomized into 8 groups (6 rats for each): control group, diabetic-untreated group, Pre-diabetic LC treated group received prophylactic LC (300mg/kg/day, orally) at start of 7th week, LC-treated diabetic group: received LC (300mg/kg/day, orally) for 6 weeks after induction of diabetes, paroxetine-treated diabetic group: received paroxetine (20 mg/kg/day orally) for 6 weeks after induction of diabetes., dapagliflozin-treated diabetic group: received dapagliflozin (1.5 mg/kg/day orally) for 6 weeks after induction of diabetes., LC + dapagliflozin-treated diabetic group: received LC and dapagliflozin for 6 weeks after induction of diabetes., and LC + paroxetine-treated diabetic group: received LC and paroxetine for 6 weeks after induction of diabetes. **Results:** Treated groups showed significant improvement in all parameters: body weight, fasting blood glucose, fasting insulin level, HOMA-IR, pro-inflammatory cytokines (INF-γ and IL1-β), malondialdehyde (MDA), super-oxide dismutase (SOD), cortisol level, serotonin, and dopamine, assess of depressive behavior by; open field test, tail suspension test, and improvement in the histopathological changes of the brain. **Conclusions:** L-carnitine is a promising agent in the management of diabetes mellitus type-2 associated with depression either singly or in combination with dapagliflozin and paroxetine

Keywords: Diabetes Mellitus Type 2; Depression; L-Carnitine; paroxetine; dapagliflozin.

Introduction

Diabetes has emerged as one of the most serious and common chronic diseases of our times, causing life-threatening, disabling, and costly complications [1]. According to estimates, around 536.6 million people (10.5%) worldwide aged between 20 and 79 years had diabetes in the year 2021. The prevalence of diabetes is expected to rise to 783.2 million (12.2%) by 2045 [2].

Egypt is ranked ninth in the prevalence of diabetes worldwide according to International Diabetes Federation (IDF) with a prevalence of 15.2% in the adult population in early 2020. The prevalence is expected to cont rising to more serious levels [3].

Depression is one of the most severe and common psychiatric disorders across the world. It is characterized by persistent sadness, loss of interest or pleasure, low energy, worse appetite and sleep, and even suicide [4].

The prevalence of depression, anxiety, and stress in people with T2-DM were 13.6% – 33.8%, >35%, and 20% – 25%, respectively. These statistics have been obtained from studies in Middle East countries [5].

Depression and T2DM share common biological origins through hypothalamic-pituitary-adrenal (HPA) axis deregulation, autonomic nervous system (ANS) hyperactivity, and inflammatory processes [6]. In response to physiological or psychological stressors, the HPA axis is activated, resulting in the secretion of corticotrophin-releasing hormone (CRH) from the hypothalamus, which stimulates the anterior pituitary gland to release adreno-corticotropic hormone (ACTH). ACTH then stimulates the release of cortisol from the adrenal gland [7]. Similarly, chronic stress over-activates the sympathetic nervous system (SNS) and increases catecholamine release. High cortisol and catecholamine levels impair insulin binding to its receptor, leading to insulin resistance and the development of hyperglycemia [8].

Hyperglycemia is a possible reason for hippocampal atrophy. There is an inverse relationship between blood sugar level and hippocampal volume, which is detected by glycated hemoglobin (HbA1c) level. Prolonged hyperglycemia or fluctuating glucose causes neuronal damage by activating the polyol pathway, which induces oxidative stress and increases the formation of advanced glycation end products (AGEs) [9].

L-carnitine has been shown to improve glucose tolerance and insulin sensitivity by different mechanisms including, improved mitochondrial oxidation of the long chain acyl CoA since their accretion is linked to insulin resistance; increasing the intra-mitochondrial acetyl-CoA/CoA which is positive for pyruvate dehydrogenase complex (PDHC) activity; improving expression of enzymes in the glycolytic and gluconeogenic; improved expression of genes in the insulin signaling cascade; improved signaling cascade for insulin-like growth factor-1 (IGF-1) axis and IGF-1. L-carnitine has been linked to the prevention of toxic effects caused by beta-amyloid (Aβ) and improves symptoms of Alzheimer's disease. The neuroprotective effects of carnitines could be related to the reduction of amyloid-related mitochondrial dysfunction and the reduction in reactive oxygen species (ROS) levels [10]. L-

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carnitine also has been shown to increase monoamine neurotransmitters (such as serotonin (5-HT), dopamine (DA), and noradrenaline (NE)), which can induce an antidepressant effect. Unlike some currently used antidepressants that may take up to 2-3 weeks to take effect, L-Carnitine can start working within just 2-3 days [11].

Dapagliflozin is a selective and reversible inhibitor of the sodium-glucose transporter 2 (SGLT2). SGLT2 proteins are expressed in the proximal convoluted tubule (PCT) of the kidneys where they are responsible for glucose and sodium reabsorption from the glomerular filtrate [12]**.** Paroxetine has always been the first choice of antidepressant for clinicians as a selective serotonin reuptake inhibitor (SSRI) [13]**.**

The present work was conducted to study the prophylactic and therapeutic effect of LC alone and in combination with paroxetine and dapagliflozin on T2-DM rats with depressed behavior changes by measuring the following parameters: body weight, fasting blood glucose, fasting insulin level, HOMA-IR, pro-inflammatory cytokines (INF-γ and IL1-β), malondialdehyde (MDA), superoxide dismutase (SOD), cortisol level, serotonin, and dopamine, assess of depressive behavior by; open field test, tail suspension test, and histopathology of brain.

Materials and methods

It is a pilot study conducted during the period from January 2023 to July 2023.

Animals:

Forty-eight adult male albino rats weighing between 150-200 g obtained from Experimental Animal Breeding Farm, (Helwan-Cairo) were used in this study. All rats were kept 7 days for adaptation before the experiment and housed in metabolic cages at room temp. 25oC± 2 and 12 hours dark/ light cycle with free access to water and a balanced diet ad libitum. All procedures were carried out in compliance with ethical standards, as approved by the Ethical Committee of the Faculty of Medicine, Benha University{M.S.29.11.2022}.

Drugs and Chemicals:

Streptozotocin (STZ), was purchased from Sigma-Aldrich Chemical Company (St. Louis, Mo., USA). LC (300 mg tab., Amriya, Egypt). Paroxetine (EVA Pharm, Egypt). Dapagliflozin: (PHARMA CARE EGYPT for trading agency). The biochemical analysis was performed using standard kits. The chemicals used in the histopathological study were all high analytical grade.

Methods:

Rats were randomized into 8 groups (6 rats for each). Diabetes was induced by high-fat diet (HFD) for 16 weeks and STZ (single dose of 25mg/kg/ I.P) in the 10th week of the study [14]. Blood was extracted from the tail vein to measure the fasting glucose concentration 72 h after STZ injection. Rats with blood glucose levels higher than 250 mg/dl were accepted as being diabetic.

Control group: (saline, orally). **Diabetic-untreated group** (HFD for 16 weeks+ STZ 25mg/kg once, i.p.in the10 week) [14]. **Pre-diabetic LC-treated group** (LC (300mg/kg/day orally) at start of 7th week till the end of experiment) [15]**. LC-treated diabetic group** (LC (300mg/kg/day, orally) for 6 weeks after induction of diabetes) [15]**. Paroxetine-treated diabetic group** (paroxetine (20

mg/kg/day orally) for 6 weeks after induction of **diabetes)** [16]**. Dapagliflozin-treated diabetic group** (dapagliflozin (1.5 mg/kg/day orally) for 6 weeks after induction of diabetes) [17]. **LC + dapagliflozin-treated diabetic group** (LC (300mg /kg/day orally) + dapagliflozin (1.5 mg/kg/day orally) for 6 weeks after induction of diabetes). **LC + paroxetine-treated diabetic group** (LC (300mg/kg/day orally) + paroxetine (20 mg/kg/day orally) for 6 weeks after induction of diabetes).

Assessment of behavior changes: through open field test and tail suspension test

Measurement of glycemic biochemical parameters

Fasting serum glucose level was measured in serum using ACCU –CHEK Active apparatus (Mannheim Germany). Fasting serum insulin level was measured in serum by ELISA. Insulin resistance was calculated using the homeostatic model assessment of insulin resistance (HOMA-IR) as described by [18]. Using the following equation: HOMA-IR = fasting glucose (mg/dl) × fasting insulin (µIU/mL)/405.

Measurement of Brain tissue parameters:

Interferon (INF-γ) [19] and Interleukin 1 beta (IL-1 beta) [20] were measured by ELISA kits according to the manufacturer's protocol, level of Malondialdehyde (MDA) was measured by using a double sandwich ELISA technique[21], level of superoxide dismutase was measured by using Purified Rat SOD antibody[22], Dopamine ELISA kit applies the competitive enzyme immunoassay technique [23], serotonin[24] and cortisol (CORT) [25] apply the quantitative sandwich enzyme immunoassay technique.

Histopathological examination

Brain tissue samples were collected and fixed overnight in 10% formalin at 4 °C. Following proper fixation, dehydration of the specimens was done in ascending grades of ethyl alcohol. The specimens were embedded in paraffin wax. Five-micron sections were cut. Brain sections were stained with hematoxylin and eosin (H and E) to study the general histological structure of brain tissue [26].

Statistical Analysis

The collected data were summarized in terms of mean ± Standard Deviation (SD). Comparisons between the different study groups were carried out using the one-way analysis of variance (ANOVA) followed by post hoc tests using the LSD method using the Statistical Package for Social Science (SPSS program, version 19.) Chicago IL USA, 2000). P value < 0.05 was considered statistically significant [27].

Results:

Induction of DM by high-fat diet and STZ resulted in a significant increase in blood glucose, insulin, HOMA-IR, IL1-β, INF-γ, MDA, cortisol, and a significant decrease in body weight, SOD, dopamine and serotonin levels as shown **(table 1,2)**

Regarding the behaviour changes: central square crossing, latency in 5 minutes and immobility time in 6 minutes were significantly higher in untreated diabetic group than the normal control group (P<0.05). The number of lines crossing and rearing in 5 minutes were significantly lower in the untreated diabetic group than the normal control group as shown **(table 3)**

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Regarding the treated groups there was a significant improvement in biochemical parameters and behaviour tests as shown **(table 1, 2, 3)**
There is also significant improvement in histopathological brain changes compared to diseased group as shown **(figure 1: B, C, D, E and F and figure 2: B, C, D, E and F)**

However, the combination between both LC and dapagliflozin group and LC and paroxetine group produced more improvement in all these parameters. If compared to normal group as shown **(table 1, 2 and 3, figure1: A, G and H and figure 2 A, G and H)**

Table 1: Effects of LC, paroxetine and dapagliflozin treatment separately and in combination on body weight, blood glucose, insulin, HOMA-IR, brain tissue level of IL1-β, INF-γ, MDA, and SOD levels in STZ -induced diabetes in rats at the end of the study

	Body weight (gm)	Glucose (Mg/dl)	Insulin (µIU/ml)	HOMA-IR
G1: Control group	295.0±11.18	98.20±6.18	5.92±0.23	1.43±0.09
G2: Untreated diabetic group	224.2±3.77 [a]	318.8±18.46 [a]	25.22±2.11 [a]	19.81±1.46 [a]
G3: Pre-diabetic LC-treated group	255.0±3.81 [a, b]	190.6±6.19 [a, b]	20.58±0.54 [a, b]	9.68±0.27 [a, b]
G4: LC-treated diabetic group	245.2±3.96 [a, b, c]	154.8±5.67 [a, b, c]	10.6±0.69 [a, b, c]	4.04±0.21 [a, b, c]
G5: Paroxetine-treated diabetic group	275.2±3.96 [a, b, c, d]	146.4±3.05 [a, b, c]	14.48±0.66 [a, b, c, d]	5.23±0.25 [a, b, c, d]
G6: Dapagliflozin-treated diabetic group	265.2±3.96 [a, b, c, d, e]	160.0±6.20 [a, b, c, e]	17.06±0.21 [a, b, c, d, e]	6.74±0.31 [a, b, c, d, e]
G7: LC + dapagliflozin-treated diabetic group	280.0±7.91 [a, b, c, d, f]	142.4±5.08 [a, b, c, d, f]	7.94±0.59 [a, b, c, d, e, f]	2.79±0.30 [a, b, c, d, e, f]
G8: LC + paroxetine-treated diabetic group	285.4±4.16 [a, b, c, d, e, f]	140.0±3.54 [a, b, c, d, f]	9.50±0.53 [a, b, c, d, e, f, g]	3.28±0.14 [a, b, c, d, e, f]
	IL 1 beta (Pg / g tissue)	**INF – γ (ng / g tissue)**	**SOD (u/g tissue)**	**MDA (Nmol/g tissue)**
G1: Control group	10.06±5.80	9.66±1.0	69.02±2.79	7.58±0.88
G2: Untreated diabetic group	75.12±5.53 [a]	55.70±2.55 [a]	18.24±3.80 [a]	45.06±3.96 [a]
G3: Pre-diabetic LC-treated group	41.36±1.40 [a, b]	31.18±1.23 [a, b]	23.48±1.25 [a, b]	22.40±1.03 [a, b]
G4: LC-treated diabetic group	24.92±1.27 [a, b, c]	19.96±1.03 [a, b, c]	36.40±1.59 [a, b, c]	14.02±0.65 [a, b, c]
G5: Paroxetine-treated diabetic group	30.16±2.85 [a, b, c, d]	23.74±1.94 [a, b, c, d]	30.18±1.55 [a, b, c, d]	16.44±2.06 [a, b, c, d]
G6: Dapagliflozin-treated diabetic group	35.30±1.46 [a, b, c, d, e]	27.12±0.63 [a, b, c, d, e]	26.84±0.55 [a, b, c, d, e]	19.84±0.65 [a, b, c, d, e]
G7: LC + dapagliflozin-treated diabetic group	13.72±0.51 [b, c, d, e, f]	12.68±0.65 [a, b, c, d, e, f]	53.86±3.62 [a, b, c, d, e, f]	8.14±0.52 [b, c, d, e, f]
G8: LC + paroxetine-treated diabetic group	19.50±1.38 [a, b, c, d, e, f, g]	16.44±0.80 [a, b, c, d, e, f, g]	42.32±2.45 [a, b, c, d, e, f, g]	11.16±0.78 [a, b, c, d, e, f, g]

Data are presented as mean ± SD a: sig if compared to G1 b: sig if compared to G2 c: sig if compared to G3 d: sig if compared to G4 e: sig if compared to G5 f: sig if compared to G6 g: sig if compared to G7.

Table 2: Effects of LC, paroxetine and dapagliflozin treatment separately and in combination on brain tissue level of cortisol, dopamine and serotonin levels in STZ -induced diabetes in rats at the end of the study

	Cortisol Pg / g tissue	**Serotonin ng/ g tissue**	**Dopamine ng/ g tissue**
G1: Control group	13.54±1.52	63.62±3.19	50.12±2.41
G2: Untreated diabetic group	78.96±6.68 [a]	15.26±2.23 [a]	11.16±1.10 [a]
G3: Pre-diabetic LC-treated group	43.52±1.42 [a, b]	21.86±1.01 [a, b]	18.08±0.24 [a, b]
G4: LC-treated diabetic group	27.86±1.10 [a, b, c]	33.70±1.64 [a, b, c]	26.84±1.12 [a, b, c]
G5: Paroxetine-treated diabetic group	33.28±3.03 [a, b, c, d]	28.30±1.42 [a, b, c, d]	22.60±1.46 [a, b, c, d]
G6: Dapagliflozin-treated diabetic group	37.68±1.57 [a, b, c, d, e]	25.28±1.44 [a, b, c, d, e]	20.44±0.77 [a, b, c, d, e]
G7: LC + dapagliflozin-treated diabetic group	17.36±0.59 [a, b, c, d, e, f]	49.54±2.83 [a, b, c, d, e, f]	39.60±1.83 [a, b, c, d, e, f]
G8: LC + paroxetine-treated diabetic group	22.78±1.06 [a, b, c, d, e, f, g]	40.0±1.87 [a, b, c, d, e, f, g]	31.88±1.52 [a, b, c, d, e, f, g]

Data are presented as mean ± SD. a: sig if compared to G1 b: sig if compared to G2 c: sig if compared to G3 d: sig if compared to G4 e: sig if compared to G5 f: sig if compared to G6 g: sig if compared to G7.

Table 3: Effects of LC and paroxetine and dapagliflozin treatment separately and in combination on open field test and tail suspension test in STZ -induced diabetes in rats at the end of the study

	Central crossing	**Latency sec**	**Line crossing**	**Rearing**	**Immobility time**
G1: Control group	4.2±0.84	0.0±0.0	77.8±3.7	23.0±2.24	80.6±4.16
G2: Untreated diabetic group	0.2±0.45[a]	4.4±0.55[a]	18.2±3.49[a]	1.4±0.55[a]	156.6±5.55[a]
G3: Pre-diabetic LC-treated group	1.0±0.71 [a]	1.6±0.55[a, b]	36.6±1.14[a, b]	8.8±0.84[a, b]	107.8±5.07[a, b]
G4: LC-treated diabetic group	2.0±0.71 [a, b]	1.0±0.71[a, b]	42.6±5.59[a, b]	10.8±1.92[a, b]	128.8±3.11[a, b, c]
G5: Paroxetine-treated diabetic group	1.6±1.14 [a, b]	1.0±1.0[a, b]	55.6±1.67[a, b, c, d]	11.4±2.41[a, b, c]	109.6±9.40[a, b, d]
G6: Dapagliflozin-treated diabetic group	0.4±0.55 [a, d, e]	2.2±1.3[a, b, d, e]	27.2±2.28[a, b, c, d, e]	3.8±0.84[a, b, c, d, e]	101.0±3.39[a, b, d, e]
G7: LC + dapagliflozin-treated diabetic group	2.4±0.89 [a, b, c, f]	0.4±0.55[b, c, f]	61.2±10.16[a, b, c, d, f]	15.2±1.92[a, b, c, d, e, f]	91.2±6.46[a, b, c, d, e, f]
G8: LC + paroxetine-treated diabetic group	1.8±0.84 [a, b, c, f]	0.4±0.55[b, c, f]	53.4±7.23[a, b, c, d, f, g]	14.2±1.92[a, b, c, d, e, f, g]	110.2±4.15[a, b, d, f, g]

Data are presented as mean ± SD. SD a: sig if compared to G1 b: sig if compared to G2 c: sig if compared to G3 d: sig if compared to G4 e: sig if compared to G5 f: sig if compared to G6 g: sig if compared to G7.

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Histopathological changes of the brain

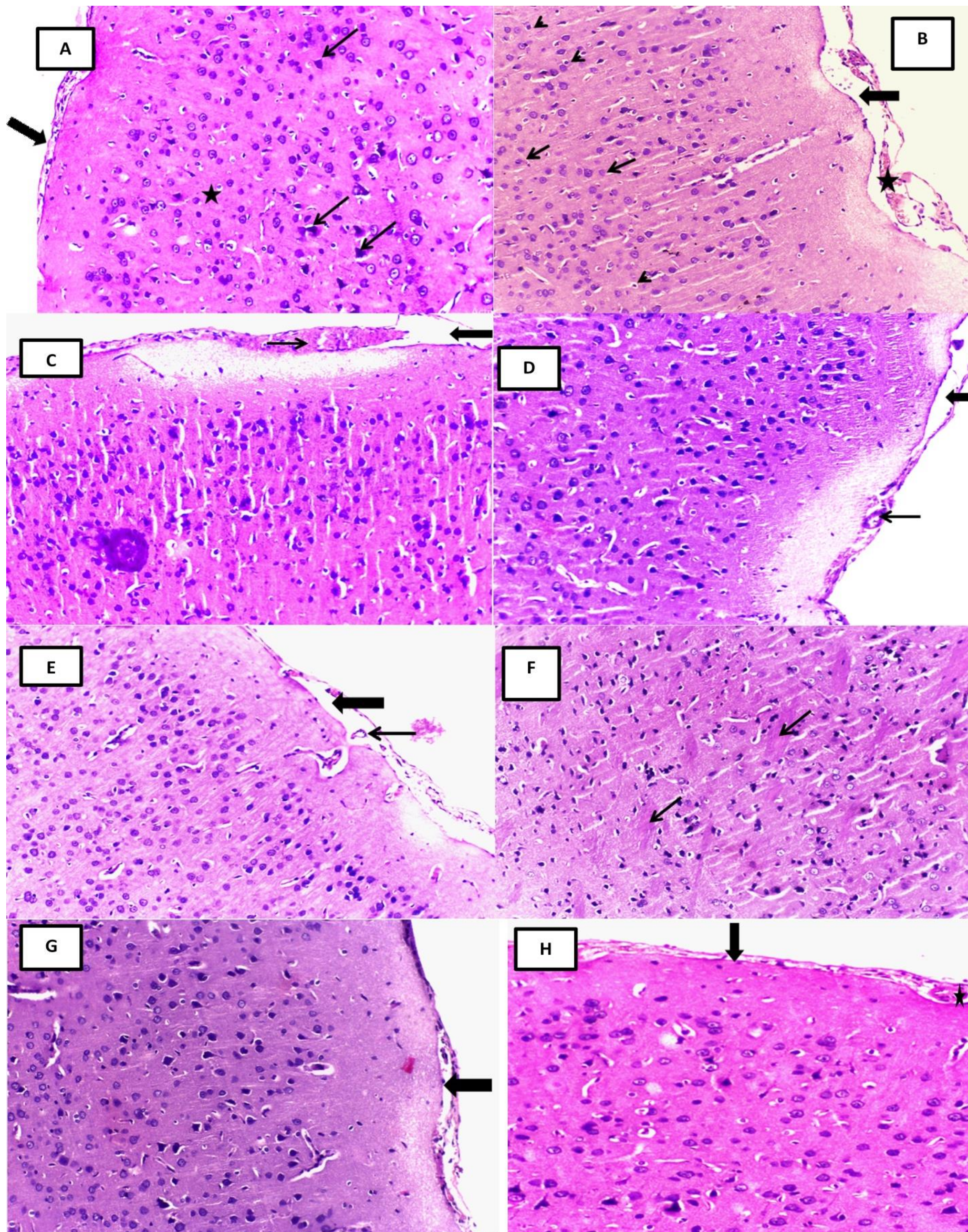

Figure (1): (A): A photomicrograph of a cut section in the cerebral cortex of the brain of a control rat (group I) Showing the Pia matter (Thick arrow) and normal cerebral cortex with normal pyramidal cells (Thin arrows) and surrounding neuropil (Star) (x200). **(B):** A photomicrograph of a cut section in the cerebral cortex of the brain of an untreated diabetic rat group 2 showing separation of Pia matter (Thick arrow), congested blood vessels (star), degenerated (Thin arrows) and apoptotic (arrow heads) pyramidal cells (×200). **(C):** A photomicrograph of a cut section in the cerebral cortex of the brain of pre-diabetic l-carnitine treated group (group III) Showing mild improvement of pathological changes in the form of improved separation of the pia matter (Thick arrow), and mild congestion (Thin arrow) (×200). **(D):** A photomicrograph of a cut section in the cerebral cortex of the brain of l-carnitine treated diabetic group (group IV) showing mild improvement of pathological changes in the form of improved pia matter separation (Thick arrow) and normal blood vessels with no congestion (Thin arrow) (×200). **(E):** A photomicrograph of a cut section in the cerebral cortex of the brain of paroxetine treated diabetic group (group V) showing decreased separation of the pia matter (Thick arrow), and normal non congested blood vessel (Thin arrow) (x200). **(F):** A photomicrograph of a cut section in the cerebral cortex of the brain of dapagliflozin treated diabetic group (group VI) showing improved gliosis (arrows) (×200). **(G):** A photomicrograph of a cut section in the cerebral cortex of the brain of L-carnitine + dapagliflozin treated diabetic group (group VII) showing moderate improvement in pia matter separation (Thick arrow) (×200). **(H):** A photomicrograph of a cut section in the cerebral cortex of the brain of L-carnitine + paroxetine treated diabetic group (group VIII) showing minimal pia matter separation (Thick arrow), normal non congested blood vessels (Star), with normal non degenerated pyramidal cells (Thin arrow) (x200).

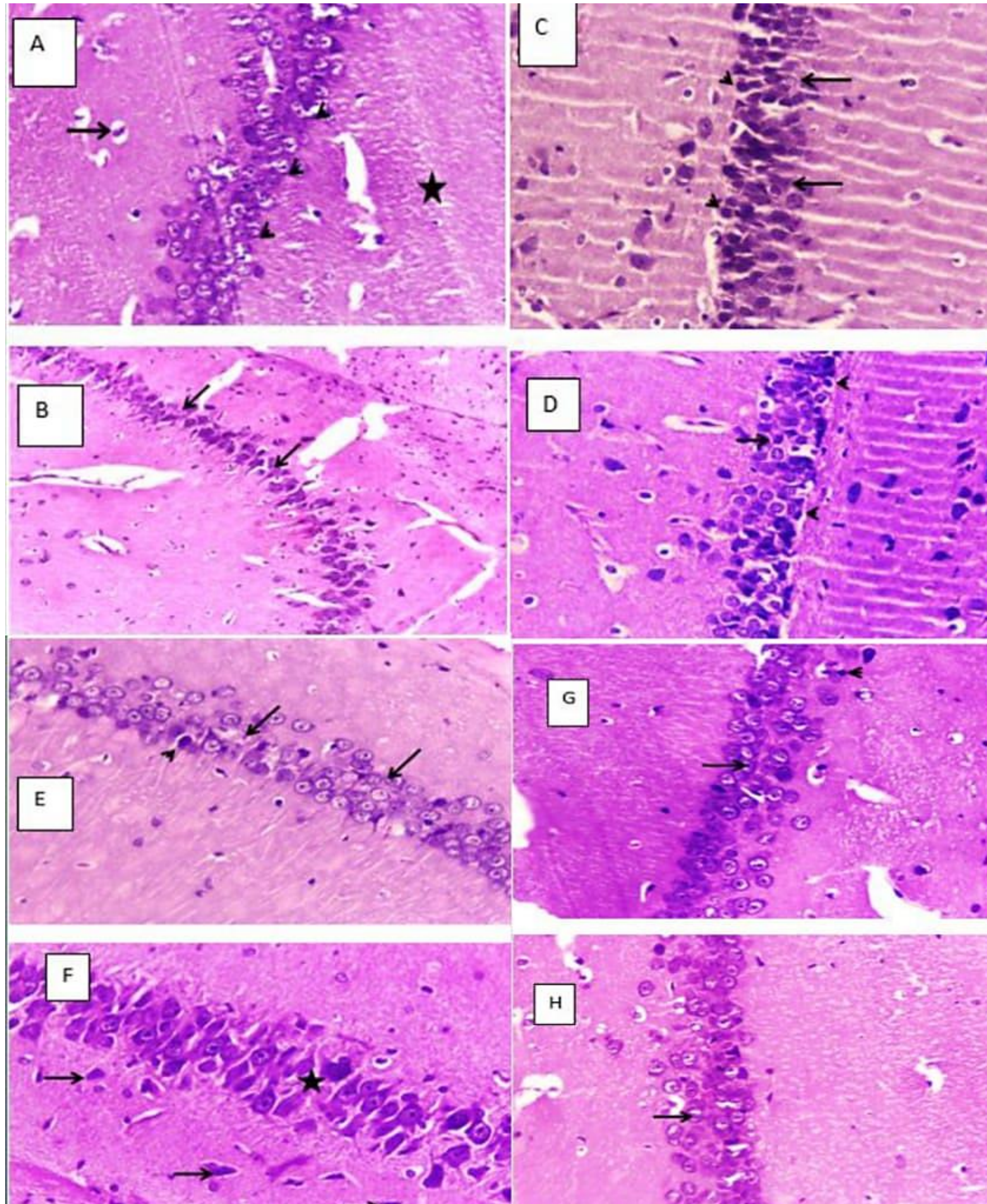

Fig 2:(A): A photomicrograph of a cut section in the hippocampus of the brain of a control rat (group I) showing normal dentate gyrus with inner large pyramidal cells [arrow], middle granule cell layer [arrow heads], and outer dendritic molecular layer [star] (x400). **(B):** A photomicrograph of a cut section in the hippocampus of the brain of an untreated diabetic rat (group II) showing decreased thickness of the middle granular cell layer (arrows) (×200). **(C):** A photomicrograph of a cut section in the hippocampus of the brain of pre- diabetic l-carnitine treated group (group III) showing near normal thickness of the middle granular cell layer [arrows] with minimal pyknotic nuclie [arrow heads] (×400). **(D):** A photomicrograph of a cut section in the hippocampus of the brain of l-carnitine treated diabetic group (group IV) showing near normal thickness of the middle granular cell layer [arrows] with decreased number of pyknotic nuclie [arrow heads] (×400). **(E):** A photomicrograph of a cut section in the hippocampus of the brain of paroxetine treated diabetic group (group V) showing normal thickness of the middle granular cell layer [arrows] with marked decrease in the number of pyknotic nuclie [arrow head] (×400). **(F):** A photomicrograph of a cut section in the hippocampus of the brain of dapagliflozin treated diabetic group (group VI) showing near-normal thickness of the middle granular layer (star) and normal pyramidal cells (arrows) (×400). **(G):** A photomicrograph of a cut section in the hippocampus of the brain of L-carnitine + dapagliflozin treated diabetic group (group VII) showing improvement in the middle granular cell layer thickness (arrow) and minimal pyknosis (arrow-head) (×400). **(H):** A photomicrograph of a cut section in the hippocampus of the brain of L-carnitine + paroxetine treated diabetic group (group VIII) showing near normal thickness of the middle granular cell layer without pyknosis (arrow) (×400).

Discussion

The present work revealed that induction of diabetes in rats by HFD and STZ resulted in a significant increase in FBG, plasma insulin, HOMA-IR index, IL1-β, INF-γ, MDA, cortisol, and a significant decrease in body weight, SOD, dopamine, serotonin levels, and alteration of behaviour in the form of a significant increase in immobility time, and latency time and a significant decrease in central crossing, rearing, and number of line crossing in STZ -induced diabetes in rats at the end of the study. Also changes in histopathological findings in the brain as compared with the control group were seen.

These findings agree with [28] who reported that HFD-STZ-induced T2-DM is associated with the same change in parameters. More so, [29] reported that HFD and STZ induced T2-DM rats exhibited depression-like behaviors and anxiety after 12 weeks. It was associated with increased expression of pro-inflammatory cytokines like IL-6.

In this study, it was found that administration of l-carnitine, paroxetine and dapagliflozin in diabetic rats for 6 weeks after induction of diabetes showed a significant decrease of the levels of fasting blood glucose, serum insulin, and HOMA-IR ratios. With a significant increase in body weight, pro-inflammatory cytokines (INF-γ and IL1-β), oxidative stress markers (MDA and SOD), cortisol, serotonin, and dopamine levels, improve alteration of behaviour in the form of a significant increase in central crossing, line crossing, and rearing and a significant decrease in latency, and immobility time. But combined treated groups resulted in more significant improvement than other treated groups.

A study done before [30] revealed that administration of L-carnitine in diabetic and glucose intolerance patients can significantly reduce FBG, HbA1c, HOMA-IR, CRP, TNF-α, weight, BMI (basal metabolic index). However, there was no significant effect observed on serum insulin level.

L-carnitine supplementation decreased fasting blood glucose in STZ-induced diabetic rats. These findings suggest that L-carnitine is an important potent agent to attenuate diabetes deteriorating effects in diabetic patients [31].

Carnitine administration reduced insulin resistance in mice fed a HFD devoid of affecting their consumption or weight. Carnitine enhanced insulin-induced glucose metabolism in diabetic and obese mice [32]**.** Carnitine can improve glucose metabolism by means of several mechanisms. First, the enhancement of mitochondrial oxidation of long chain-Acyl-CoA, which accumulation produces insulin resistance in muscle and heart. Second, inducing changes in glycolytic and gluconeogenic enzymes. Third, modifying the expression of genes related to the insulin signalling cascade, finally, improving glucose utilization by heart [33]**.**

Alhasaniah[32] reported the administration of L-carnitine caused a decrease in oxidative stress marker (MDA level) in male albino rats, and also decreased pro-inflammatory status, improving the oxidative stress.

Nicholson and co-workers [11 reported that LC is an effective drug in the treatment of mental disorders such as depression by playing an important role in

neurotransmitter regulation including monoamine, 5-methyl-hydroxytryptamine, and dopamine (DA). Moreover, long-term use of LC supplementations can reduce the GABA level in the hippocampus of mice which is considered to be related to the pathological mechanism of depression.

This was shown that treatment with dapagliflozin increased body weight significantly [34].

In contrast to our results, it was shown that dapagliflozin reduces total body weight. Because dapagliflozin increases urinary glucose excretion, this weight decrease could result from reduced body fat secondary to caloric loss or from fluid loss secondary to osmotic diuresis or from a combination of both factors [35]. Ramirez-Rodrigues and colleagues [36] found that administration of dapagliflozin decreases blood glucose levels and increases insulin sensitivity. Regarding to anti-inflammatory action of dapagliflozin, ElMahdy and others [37] showed that dapagliflozin has an anti-inflammatory role and decreases interleukins. A recent hypothesis for the probable mechanism involved in this anti-inflammatory action includes the restoration of autophagy, which leads to anti-apoptotic, antioxidant, and anti-inflammatory effects [38].

A study done in 2019 [39] showed that dapagliflozin decreased the level of MDA while the level of SOD was significantly increased. The possible mechanism was that dapagliflozin reduced the expression of inflammatory cytokines by activation of the M2 macrophage phenotype.

Sodium-glucose co-transporter 2 (SGLT2) inhibitors, such as dapagliflozin have been shown to alleviate depressive symptoms in DM patients by reduction of serum ACTH levels [40]. On the other hand, patients with major depressive disorders (MDD) have a dysfunction in the hypothalamic-pituitary-adrenal (HPA) axis which results in an increase in the level of this hormone [41]. Therefore, it is assumed that this group of drugs may have a positive effect on improving depressive symptoms. The antidepressant effect of dapagliflozin might be related to the increase of monoamine levels, neurotrophic factors, and and-inflammatory cytokines [42].

Fava, 2000 [43] found that paroxetine may be more likely to cause weight gain than other SSRIs during long-term treatment. The possible mechanisms for SSRI-induced weight gain are recovery from depression or clinical improvement, appetite increase/carbohydrate craving, and changes in serotonin 5HT2c receptor activity.

It was reported that SSRIs can induce hypoglycaemia by various mechanisms, including increasing insulin sensitivity, interfering with the metabolism of sulfonylureas, and reducing gluconeogenesis [44]. Paroxetine inhibits insulin secretion at least via decreasing intracellular 5-HT and insulin biosynthesis [45]. A study [46] showed that administration of antidepressants resulted in a significant decrease in IL-1 beta level. According to Dionisie and co-workers [47]; paroxetine decreases the neuro-inflammation by the reduction of blood or tissue cytokines or regulating complex inflammatory pathways: nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB), inflammasomes, peroxisome proliferator-activated receptor gamma (PPARγ). Also, SSRIs show these effects in association with an antidepressant action.

It was shown that paroxetine has significant relief of depression-like behaviours, decrease in the MDA level,

increase in superoxide dismutase and catalase activities, and increase in serotonin transporter 5-HTT and norepinephrine transporter (NET) expression. The results may suggest that the anti-depressive effect of paroxetine is at least partly related to reversing oxidative stress imbalance and elevating the expression of 5-HTT and NET [48].

It was shown that SSRIs have been found to decrease cortisol levels and normalize HPA axis activity [49].

The behavioural effects of acetyl-l-carnitine may involve central mechanisms, e.g., cholinergic neurotransmission in the brain**.** This agrees with Nicholson and co-workers [11] who revealed that the most studied potential mechanisms of carnitine in improving symptoms of depression include the neuroplasticity effect, oxidative stress, and neurotransmitter regulation.

These results agree with Ripoll and others' [50] who approved that paroxetine decreased immobility time in tail suspension test in depressed rats. Observed that administration of dapagliflozin was able to decrease the motor disabilities observed in the open field test [51].

Conclusions

L-carnitine has prophylactic and therapeutic effects on T2-DM and depression-like behavior induced by DM. Its effect involves many mechanisms; including neurotransmitter regulation including monoamine, 5-methyl-hydroxytryptamine, and dopamine, reducing the GABA level, and as a hypoglycemic effect, it leads to weight loss and improved blood sugar regulation. LC alone or combined with paroxetine or dapagliflozin could improve biochemical changes in diabetes and pathological changes in the brain in depression. LC could be considered as a promising agent for the prophylactic and therapeutic effect on depressed patients on top of T2-DM. Paroxetine has an anti-diabetic effect. Dapagliflozin could be used for depressed diabetic patients.

Recommendations

Further studies are needed for Study more neuroprotective and hypoglycemic mechanisms of l-carnitine, detection of the optimal dose, toxic dose and duration of use of l- carnitine, and detection of the optimal dose of combination of l-carnitine, dapagliflozin, and paroxetine

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To cite this article: Abeer A. Abdel-Hameed, Ahmed S. Mohamed, Omaima M. Abdullah, Mahitab E. Gahgah, Abeer A. Eldeeb.The Potential Association between Diabetes Mellitus Type 2 and Depression- Role of L-Carnitine (An Experimental Study on Adult Albino Rats). BMFJ XXX, DOI: 10.21608/bmfj.2025.344849.2292

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