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Protective Effect of *Terminalia muelleri* Extract on Brain of Streptozotocin-Induced Diabetes in Albino Rats

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Abstract

Diabetic neuropathy is one of the complications of diabetes. This study investigated the possibility of reducing neuropathy of STZ-induced diabetic rats by *Terminalia muelleri* extract (TE) and comparing the effect of the extract with the therapeutic effect of pioglitazone (PG) drug. The experimental animals were divided into non-diabetic (normal control), STZ-induced diabetic (diabetic control), TE-treated non-diabetic (200 mg/kg b.wt) (TE-group) TE-treated diabetic (200 mg/kg b.wt) (TE-STZ-group), and pioglitazone-treated diabetic (1.58 mg /kg b.wt) (PG-STZ-group). All treatments were administered orally by oral gavage once daily throughout the 4 weeks of the treatment period. In this study: malonaldehyde, nitric oxide, reduced glutathione and glutathione disulfide were examined as oxidative stress marker in the brain tissue of the experimental rats. The results indicated high oxidative stress in STZ-diabetic groups and reduced oxidative stress of groups treated with TE. The results of norepinephrine, dopamine, gammaamino- butyric acid, brain-derived neurotrophic factor, and Casps-3 also demonstrated the possibility of using TE to attenuate the effects of neuropathy in experimental rats comparable to PG use. This indicated that the TE is promising alternative to chemical treatment with PG drug. This indicated that TE is promising alternative to chemical treatment with PG drug.

Keywords: Diabetic neuropathy; Terminalia; Brain.

1. Introduction

Diabetes mellitus (DM) is one of the most common diseases worldwide. Researchs predicts that the number of patients will reach 366 million patients by 2030. Diabetic polyneuropathy results from Diabetes and occurs in more than 50% of the diabetic patient [1].

Diabetes mellitus is one of the most important metabolic diseases in humans and affects many metabolic systems in the body. Irregular use of insulin by diabetics leads to recurring cases of hypoglycemia and hyperglycemia, which leads to severe developments in the central nervous system [2]. Neuron damage occurs soon after diabetes appears as a result of an increase apoptosis and oxidative stress [3].

Diabetes affects many vital systems in the human body, as diabetic patients are commonly affected by nerve compression syndrome, known as neuropathy [4].

Nerve compression occurs as a result of several hypotheses, including increased concentration of blood glucose, which turns into sorbitol in the polyol pathway and sorbitol leads to nerve swelling due to reduced plasma permeability. The second hypothesis is that the high concentration of glucose leads to the formation of compounds such as Advanced Glycation End Products (AGEs) which in turn reduce the supply of nerve vessels and lead to the destruction of nerve fibers [4]. The effect of diabetes on nerves is described by a group of symptoms called diabetic neuropathy (DN), which appears in late stages of type 1diabetes but appears in early stages in type 2 diabetes. Therefore, it appears in streptozotocin (STZ)-treated rats as a result of insulin deficiency in rat models, which is due to the cellular toxicity of pancreatic beta cells and the production of nitric oxide [5]. Symptoms of neuropathy in STZ- diabetic rats has appeared in the form of short and long-term symptoms including hyperglycemia, insulin resistance, glycosuria, polyphagia, polyuria, polydipsia, and abnormal glucose [1]. Moreover, DN is measured by a set of parameters such as catecholamines and metabolites concentrations, amino acids and oxidized and reduced glutathione [6]. Diabetes leads to the accumulation of some compounds, including amyloid-b, ROS that lead to symptoms of neuropathy, which is due to mitochondrial dysfunction, activation of pro-inflammation and pro-apoptosis [7].

The medicine plants contain many phenolic and polyphenolic compounds that play a major role as antioxidants and that lead to reducing cell damage and death. Numerous recent studies have demonstrated the role of these compounds in improving memory and reducing brain damage to experimental rats [8].

Although there are many studies on the benefits of medicinal plants and their uses in the fields of pharmacy and food, there is an ongoing need to search for new sources for these plants [9]. The *Terminalia* plant is one of these plants recently studied in Egypt and the worldwide [10]. The genus *Terminalia* includes more than 200 species spread in the tropical and semi-tropical regions [11]. It has been used in the traditional medicine because of its high content of phytochemical compounds that have a role as an antimicrobial, antioxidant, anti-diabetic, analgesic effect, and anti-inflammatory [6, 10, 12, 13]. Additionally, many *Terminalia* species have different therapeutic effects, as they have an opposite effect for each of fever, cough, asthma, urinary diseases [14]. Some species of Terminalia such as *Terminalia bellerica* has been widely used in Indian medicine to treat various diseases including diabetes. There is a potential possibility of using termnella to treat hypoglycemia and hyperlipidemia in blood serum of rats with diabetes caused by streptozotocin [12]. Moreover, *Terminalia muelleri* can be considered as a promising medicinal plant as it has shown good results in the treatment of tumors [15]. Children consumed *Terminalia* seeds in some countries, such as Thailand, where many studies mentioned high nutritional properties such as high protein content compared to many legumes as well as high oil content [16]. Etienne, *et al.* [17], showed the important nutritional properties of *Terminalia* and the possibility of using its oil extract for improving the quality of the human diet. They added also that the *Terminalia* oil has been considered as an edible oil because of its high content of unsaturated fatty acids, as well as the high quality of its physical properties.

The present study aimed at evaluating the potential of *Terminalia muelleri* ethanolic extract against some parameters of diabetic neuropathy in blood serum and brain tissue of STZ-induced diabetic rats.

2. Materials and Methods

2.1. Chemicals

All chemicals, standards are used in the experiments were purchased from Sigma Chemical Co. (St. Louis, MO, USA).

2.2. Terminalia Muelleri Extract

The ethanolic extract of *Terminalia muelleri* leaves were prepared according to the method described by Fahmy, *et al.* [18] with some modifications. *T. muelleri* leaves were collected from Zoo botanical garden, Giza city, Egypt. Leaves were washed and air-dried at 37 °C for 48 h. Dried leaves were grounded into powder using electric blinder. One kilogram of *T. muelleri* leaves powder was extracted three times with 80% ethanol. The total extract was concentrated and lyophilized to obtain about 250 g TE powder.

2.3. Animals and Treatments

Thirty female albino- strain rats will be obtained from National Organization Drug Control and Research (NODCAR), Giza, Egypt. Adult animals (120 – 150 ± 5g, 10 weeks age) The experimental protocol was approved by the ethics committee for experimental and clinical studies at the National Organization for Drug Control and Research (NODCAR). The approval number is NODCAR /II/12/2020. The animals were housed in separate stainless-steel cages under controlled conditions at constant temperature (24℃). During adaptation (15 days) and experiment periods (4 weeks), all the animals were allowed free access to water and basal diet and maintained at room temperature. The basal diet was consists of 10 % casein, 10 % corn oil, 5% cellulose, 1 % vitamin mixture, 4 % salt mixture, and 70% corn starch [19].

2.4. Induction of Diabetes

Streptozotocin (STZ) was prepared in freshly prepared 0.01 M citrate buffer, pH 4.5 [20]. Diabetes was induced in rats by intraperitoneal administrating a single dose of STZ (55 mg/kg b.w.). After 48 h, rats with marked hyperglycemia (fasting blood glucose >140 mg/dl) were selected and characterized as diabetic rats.

2.5. Experimental Design

Rats were divided in 5 groups compressing in 6 rats in each group as following:

Group 1: (Normal control, non-diabetic rats) Health animals that given oral administration of 1 mL distilled water throughout the experimental period.

Group 2: (STZ-group, diabetic control) STZ-induced diabetic rats.

Group 3: (TE -group): Healthy rats treated with a daily oral dose of *Terminalia muelleri* extract (200 mg /kg b.wt.) according to Fahmy, *et al.* [21].

Group 4: (TE-STZ-group) Diabetic rats treated with a daily oral dose of *Terminalia muelleri* extract (200 mg /kg b.wt).

Group 5: (PG-STZ-group) Diabetic rats treated with a daily oral dose of pioglitazone (PG drug) (1.58 mg /kg b.wt).

At the end of the experiment, rats were fasted overnight, and blood sample was collected from retrobulbar venous plexus by fine capillary tubes then centrifuged and the obtained serum was used for biochemical analyses. After that, rats were euthanized decapitated and the brain was excised and washed with cold saline and saved at -80˚C until used.

2.6. Biochemical Parameters

Diabetic neuropathy was evaluated by measuring different biochemical parameters in cortex layer of brain tissue. The reduced glutathione (GSH), glutathione disulfide (GSSG) [22], lipid Peroxidase (Malondialdehyde; MDA) [23], and Nitric oxide (NO) [24] in brain tissue were determined as oxidative stress markers.

Catecholamines and metabolites concentrations in brain tissue including norepinephrine (NE) , dopamine (DA), and serotonin (or 5-hydroxytryptamine; 5-HT) were determined in brain tissue according to the method described by Pagel, *et al.* [25].

Amino acids in brain tissue including GABA (gammaamino- butyric acid), glutamate (Glu) and aspartate (Asp) were measured by HPLC as mentioned by Heinrikson and Meredith [26]. Adenosine tri- and mono-phosphate (ATP and AMP) in brain tissue were analyzed [27]. Phosphatidylcholine (PtdCho) was extracted and assayed in brain tissue by HPLC [28].

Acetylcholinesterase (AChE) activity in the brain samples was measured in brain tissue by the method described by Gorun, *et al.* [29]. The levels of brain-derived neurotrophic factor (BDNF) were estimated using a rat-specific immunoassay kit (Rat BDNF ELISA) in brain tissue [3].

Caspase-3 levels were estimated in blood serum using a rat-specific immunoassay kit (Rat Caspase-3 ELISA, from Bioassay Technology Research Co.) according to the manufacturer's protocol [3].

Interleukin 1 (IL1) levels were estimated in blood serum using a rat-specific immunoassay kit (Rat Interleukin 1 ELISA, from Bioassay Technology Research, Co.)[30].

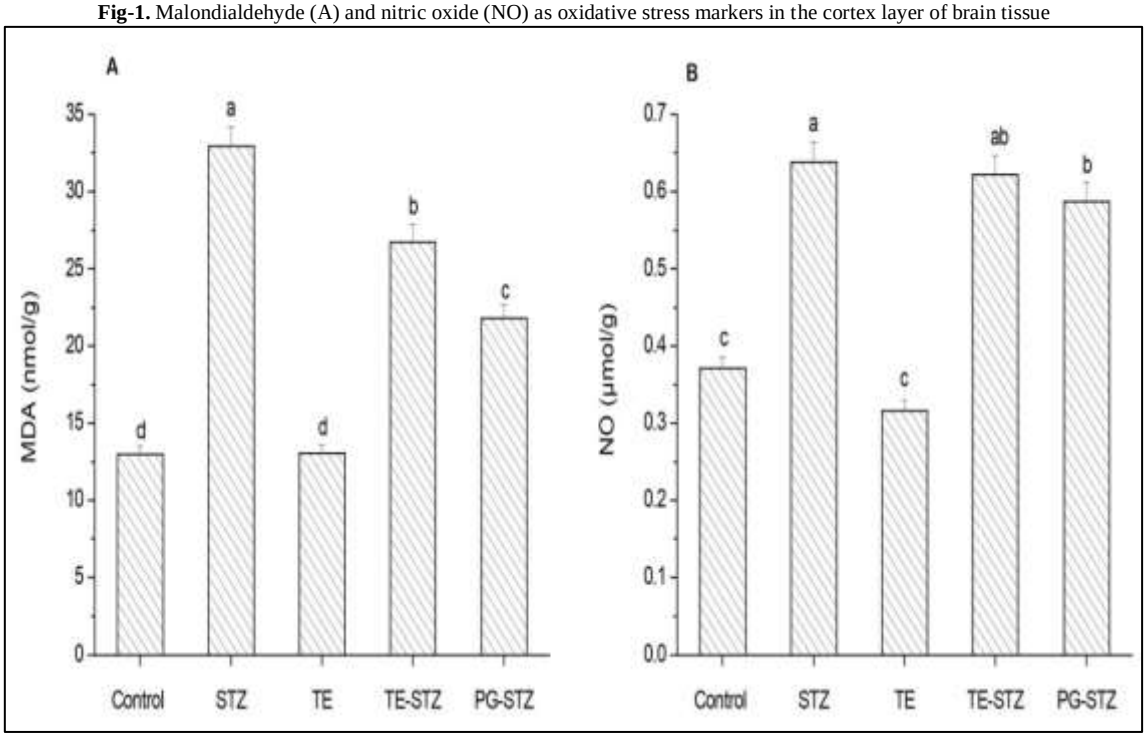
2.7. Statistical Analysis

For each treatment, data from three independent measurements were collected and the mean values ± standard deviations (SD) were determined and analyzed by a one-way ANOVA using 'Proc Mixed' (SAS 8.2, Cary, NC). Differences between treatments were determined by Duncan's method. In all cases, the level of statistical significance was $p < 0.05$. Correlation coefficient matrix was created between different studied variants using statistical analysis tools in Excel program, Microsoft office, USA.

3. Result and Discussion

Oxidative stress due to diabetic neuropathy was evaluated by measuring different biochemical parameters in cortex layer of brain tissue. Malondialdehyde (MDA), and nitric oxide (NO) as oxidative stress markers in brain tissue were determined and illustrated in Fig. 1. Rats treated with STZ showed significant increase in MDA and NO levels as oxidative stress markers in brain tissue comparing to control group (Fig. 1). The non-diabetic rats administrated with TE extract did not show significant changes in either MDA or NO compared with control rats. This finding was in accordance with those mentioned by Salehi, *et al.* [31]. Treatment of STZ-diabetic rats with PG drug or TE extract (in PG-STZ and TE-STZ groups, respectively) led to significant reduction in the values of MDA (Fig. 1). Also, nitric oxide was significantly enhanced due to treatment with PG drug. TE extract led also to slight enhancement in NO value in TE-STZ rats.

Fig-1. Malondialdehyde (A) and nitric oxide (NO) as oxidative stress markers in the cortex layer of brain tissue



Rat groups ($n=6$) were STZ-group: Streptozotocin-induced diabetic rats, TE -group: Healthy rats treated with a daily oral dose of *Terminalia muelleri* extract (200 mg /kg b.wt.), TE-STZ-group: Diabetic rats treated with a daily

oral dose of *Terminalia muelleri* extract (200 mg /kg b.wt), PG-STZ-group: Diabetic rats treated with a daily oral dose of pioglitazone drug (1.58 mg /kg b.wt).

In each parameter in the same column group, values with the different letters were significantly different. (*P<0.05*).

Reduced glutathione (GSH) and oxidized glutathione (GSSG) as oxidative stress markers were determined in the cortex layer of brain tissue. The treatment with STZ led to a dramatic reduction in GSH, while it caused a significant increase in GSSH (Table 1).

Table-1. Reduced glutathione (GSH) and Oxidized glutathione (GSSG) as oxidative stress markers determined in the cortex layer of brain tissue

Groups[$]	GSH (µmol/g)	GSSG (µmol/g)	Total	GSH%
Control	3.52 [a] ± 0.14	0.39 [bc] ± 0.02	3.91 ± 0.08	90.04
STZ-group	2.19 [c] ± 0.8	0.62 [a] ± 0.03	2.81 ± 0.42	77.88
TE-group	3.53 [a] ± 0.14	0.37 [c] ± 0.01	3.89 ± 0.08	90.59
TE,STZ-group	2.67 [b] ± 0.1	0.47 [b] ± 0.02	3.14 ± 0.06	85.08
PG-STZ-group	3.14 [a] ± 0.13	0.54 [a] ± 0.02	3.68 ± 0.08	85.40

Rat groups were described in Fig. 1. Values were expressed as means ±SD (*n=6*). Means in the same column with different letters were significantly different. (*P<0.05*)

Rats treated with TE extract did not display significant changes in the GSH and GSSG compared to rats of control group. The PG-STZ-treatment resulted in increasing the GSH content to become close to the control values without significant difference comparing with the control. On the other hand, TE-STZ-treatment led to attenuate the effect of STZ on GSH, but there is still a significant difference with the control (Table 1). It is well known that the ratio of GSH to GSSG is an indicator on the oxidative stress within cell. In non-diabetic rats in the control and TE-groups the % GSH was more than 90%, which indicates their healthy tissues [32]. The GSH% value of 77.88% in brain tissue of STZ-group ensured exposure of these rats to oxidative stress. The attenuated effect of TE and PG was demonstrated where the GSH% was 85.08 and 85.4 %, respectively.

The Neurotransmitters norepinephrine (NE), dopamine (DA) and serotonin (5-hydroxytryptamine, 5-HT) were determined in cortex layer in brain tissue of the experimental rats (Table 2). Induction of diabetes by STZ (50 mg/kg) caused significant reduction in all studied neurotransmitters in cortex layer. This finding was in agreement with those mentioned by Gallego, *et al.* [33], who demonstrated that administrating STZ in dose mor than 40 mg/kg are associated with a reduction of norepinephrine release in brain tissue . The best treatment for recovering the normal level of all tested neurotransmitters in brain tissue was PG-STZ followed by TE-STZ.

Tables-2. Neurotransmitters (means[*] ±SD) in brain tissue

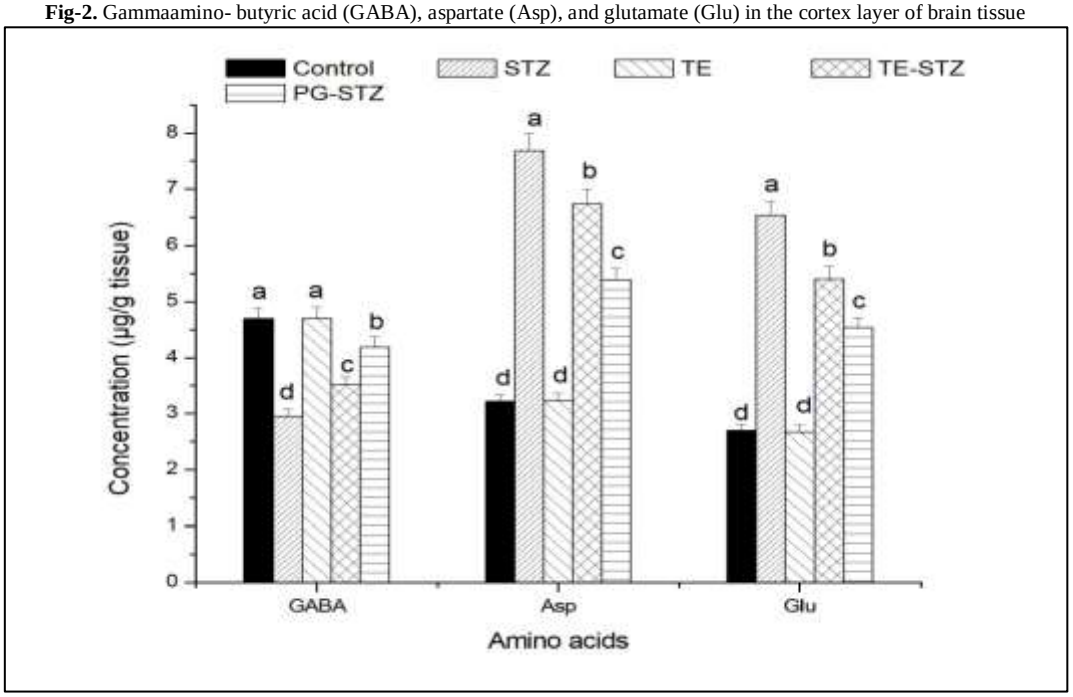
Groups[$]	NE (µg/g)	DA (µg/g)	5-HT (µg/g)
Control	0.386 [a] ± 0.017	1.125[a] ± 0.043	0.397[a] ± 0.017
STZ-group	0.135[d] ± 0.006	0.636[b] ± 0.027	0.18[c] ± 0.007
TE-group	0.394[a] ± 0.017	1.098[a] ± 0.041	0.37 [a] ± 0.015
TE,STZ-group	0.198[c] ± 0.009	0.754[b] ± 0.028	0.239[b] ± 0.009
PG-STZ-group	0.332[b] ± 0.013	1.008[a] ± 0.043	0.317[a] ± 0.014

NE: Norepinephrine, DA: dopamine, 5-HT: 5-hydroxytryptamine (serotonin),
[*]Means in the same row with the same letter indicated no significant difference (*P<0.05*).
[$] as mentioned in Table

Cerebral metabolism is affected by hyperglycemia resulting in changes in the level of brain metabolites such as gammaamino- butyric acid (GABA), aspartate (Asp), and glutamate (Glu) [34]. The obtained results indicated that the STZ-treatment caused reduction in GABA and elevation in Asp and Glu levels in cortex layer of rat brain (Fig. 2). TE-treatment did not significantly change the level of these amino acids compared with control group. Attenuation in STZ negative effect on studied amino acids was noticed in PG-STZ and TE-STZ groups.

The treatment by STZ led to a significant decrease in ATP in brain from 29.2 to 18.6 µg/ g tissue (Fig. 3). This reduction may be due to the reduction effect of STZ on (Na/K)-ATPase activity in the tissues [35]. Moreover, phosphatidylcholine (PtdCho) is the principal and essential phospholipid of plasma and intracellular membranes of mammalian cells, especially in brain. The first step in synthesis of PtdCho is phosphorylating choline to P-choline by aid of choline kinase in the presence of high ATP concentration. In the present study, STZ-treatment led to reduce ATP concentration and consequently PtdCho content in brain tissue. However, the treatment with the PG drug displayed the highest protective effect against STZ, while the treatment of STZ-diabetic rats with the TE extract led to attenuate the negative effect of STZ.

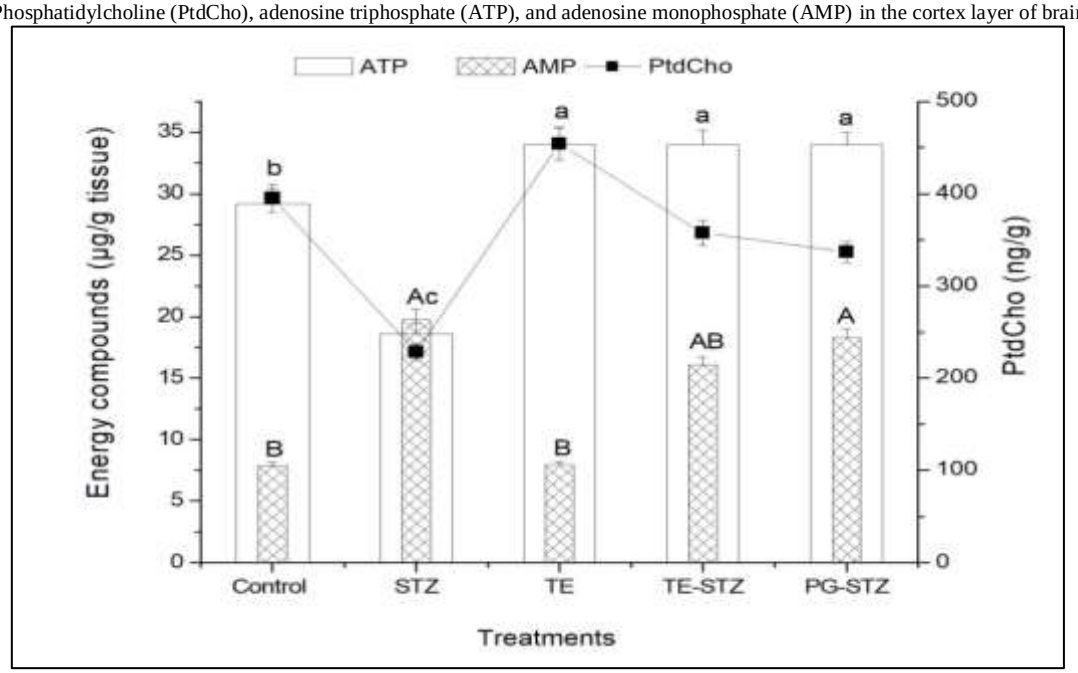
Fig-2. Gammaamino- butyric acid (GABA), aspartate (Asp), and glutamate (Glu) in the cortex layer of brain tissue



Rat groups were described in Fig.1. Values were expressed as means ±SD ($n=6$). In each parameter in the same column group, values with the different letters were significantly different. ($P<0.05$)

STZ-induced diabetic rats showed significant elevation in caspase-3 immunoactivity to be 0.537 pmol/min/mg protein comparing to 0.166 pmol/min/mg protein in nondiabetic rats (Table 3). In agreement with few previous studies we also found relatively increased expression of Casp-3 pro-apoptotic protein [36].

Fig-3. Phosphatidylcholine (PtdCho), adenosine triphosphate (ATP), and adenosine monophosphate (AMP) in the cortex layer of brain tissue



Rat groups were described in Fig.1. Values were expressed as means ±SD ($n=6$). In each parameter values with the different letters were significantly different. ($P<0.05$)

Treatment of non-diabetic rats with TE extract did not show significant change in Casp-3 value. However, TE-treatment of STZ-induced diabetic rats led to significant reduction in the Caps-3 activity from 0.537 to 0.409 pmol/min/mg (Table 3). The best treatment to prevent the increase of Casp-3 activity was PG-STZ followed by TE-STZ. This finding indicated that our treatments led to lowering the expression of Casp-3 and its presence in brain tissue.

As shown in Table 3, the obtained results ensure that, interleukin-1 (IL-1) was expressed in low level in non-diabetic rats in control group (12.94 pg/mg protein) as mentioned in previous literature [1, 36]. In STZ-diabetic rats, IL-1 level was significantly elevated in brain tissue to be 30.44 pg/mg protein. This increase in IL-1 level in the brain of STZ-group indicated the inflammatory response to brain injury due to treatment with STZ [37]. The IL-1 level in TE-STZ group (22.59 pg/mg protein) demonstrated the significant attenuating effect of the ethanolic extract of *Terminalia* leaves. Markedly, there is no significant difference between the attenuated effect of TE and PG. This indicated that the terminalia extract (TE) is promising alternative to chemical treatment with pioglitazone (PG) drug.

Tables-3. Immune-mediators (means* ±SD) in brain tissue

Groups$	Casp-3 pmol/min/mg protein	IL-1 pg/mg protein	BDNF (ng/g tissue)	AChE (U/g tissue)
Control	0.166[d] ± 0.007	12.94[c] ± 0.555	24.55 ± 0.95c	1.27 ± 0.049c
STZ-group	0.537[a] ± 0.023	30.44[a] ± 1.353	72.01 ± 3.199a	3.602 ± 0.16a
TE-group	0.198[d] ± 0.008	12.92[c] ± 0.561	23.5 ± 1.04c	1.328 ± 0.059c
TE,STZ-group	0.409[b] ± 0.017	22.59[b] ± 0.944	46.24 ± 1.884b	2.882 ± 0.117ab
PG-STZ-group	0.297[c] ± 0.012	21.93[b] ± 0.919	43.72 ± 1.895b	2.602 ± 0.113b

Casp-3: Caspase-3 a type of cysteine-aspartic proteases, IL+1: Interleukin, BDNF: brain-derived neurotrophic factor, AChE: Acetylcholinesterase.
*Means in the same row with the same letter indicated no significant difference ($P<0.05$).
$ as mentioned in Fig. 1 .

BDNF level in brain tissue is considered as one of the important factors that could be regulated in diabetes [38]. In the present study, STZ-treatment caused significant increase in the amount of BDNF in brain tissue compared to the control group. Increasing BDNF level in rats treated with STZ was previously demonstrated [39]. Both PG and TE treatments displayed considerable enhancement in BDNF level in brain tissue at the end of the experiment period (Table 3). This result indicated that TE-treatment led to significant regulation in BDNF level in brain to be near to its level in the non-diabetic rats. In this context, Zhou, *et al.* [1] emphasized that BDNF regulation is an important factor in calming the symptoms of neuropathy.

In the present study, STZ-treatment caused significant increase in the amount of AChE in brain tissue compared to the control group. Significant enhancement in BDNF and AChE levels in brain tissue were recorded at the end of the experiment period when rats were treated with PG or TE (Table 3).

To discuss the relationship between all studied markers, correlation coefficient was calculated and presented in Table 4. There is a strong positive relationship between GSH as antioxidant marker and some biochemical parameters including GABA (+1.00), NE (+0.90), 5-HT (+0.99), and PtdCho (+0.89). The increasing of these parameters has been demonstrated previously as a result of increasing GSH in brain cells [34]. On the other hand, strong negative correlation between Casp-3 and GABA (-1.00). This relationship was confirmed by El-Ansary, *et al.* [40] as the results showed an inverse relationship between casps-3 versus GABA, where casps-3 plays an important role during apoptosis and the increase of casps-3 values leads to a clear defect in the mitochondria, which leads to the occurrence of apoptosis and accompanying a marked decrease in the values of GABA.

Table-4. Correlation matrix between the studied biochemical parameters

	GSH	*GSSG*	*MDA*	*NO*	*NE*	*DA*	*5-HT*	*GABA*	*Asp*	*Glu*	*ATP*	*AMP*	*PtdCho*	*Casp-3*	*IL+1*	*BDNF*	*AChE*
GSH	1.00																
GSSG	-0.86	1.00															
MDA	-0.99	0.90	1.00														
NO	-0.87	0.87	0.93	1.00													
NE	0.90	-0.93	-0.96	-0.98	1.00												
DA	0.50	-0.46	-0.55	-0.50	0.58	1.00											
5-HT	0.99	-0.84	-0.99	-0.88	0.91	0.59	1.00										
GABA	1.00	-0.85	-0.99	-0.88	0.90	0.50	0.99	1.00									
Asp	-0.98	0.88	1.00	0.95	-0.96	-0.56	-0.99	-0.98	1.00								
Glu	-0.98	0.91	1.00	0.94	-0.97	-0.55	-0.98	-0.98	1.00	1.00							
ATP	0.66	-0.65	-0.58	-0.34	0.43	-0.06	0.58	0.64	-0.51	-0.56	1.00						
AMP	-0.85	0.95	0.92	0.96	-0.99	-0.60	-0.86	-0.84	0.92	0.93	-0.39	1.00					
PtdCho	0.89	-0.96	-0.90	-0.83	0.88	0.27	0.85	0.88	-0.87	-0.90	0.79	-0.87	1.00				
Casp-3	-1.00	0.87	0.99	0.87	-0.91	-0.57	-1.00	-1.00	0.98	0.98	-0.63	0.86	-0.88	1.00			
IL+1	-0.79	0.77	0.73	0.48	-0.59	-0.25	-0.74	-0.77	0.67	0.72	-0.95	0.56	-0.85	0.79	1.00		
BDNF	-0.97	0.96	0.97	0.87	-0.93	-0.49	-0.95	-0.96	0.95	0.97	-0.72	0.90	-0.96	0.97	0.85	1.00	
AChE	-0.96	0.93	0.99	0.95	-0.98	-0.59	-0.97	-0.96	0.99	1.00	-0.53	0.96	-0.90	0.97	0.70	0.97	1.00

GSH: reduced glutathione, GSSG: glutathione disulfide, MDA: Malondialdehyde, NO: Nitric oxide, NE: Norepinephrine , DA: dopamine, 5-HT: 5-hydroxytryptamine (serotonin), GABA: gammaamino- butyric acid, Asp: aspartate, Glu: glutamate, ATP: Adenosine Tri-phosphate, AMP: Adenosine Mono-phosphate, PtdCho: Phosphatidylcholine, Casp-3: Caspase-3,, IL+1: Interleukin, BDNF: brain-derived neurotrophic factor, AChE: Acetylcholinesterase.
Bold numbers indicated strong correlation relationship (+ or -). Strong relationship (r = -1.0 to -0.7 or 1.0 to 0.7), moderate relationship (r = <-0.7 to -0.5 or < 0.7 to 0.5), weak relationship (r = <-0.5 or 0.5)

4. Conclusion

The obtained results indicated that the neuronal death in brain tissue was referred to the oxidative stress as a result of treatment with STZ. Treatment with Terminalia muelleri extract reduced the harmful effects of oxidative stress resulting in reduction of brain cell apoptosis (casp-3), IL-1, BDNF, AChE and DA. Moreover, TE-treatment led to an increase in NE, 5-HT, ATP and PtdCho. These results confirm that the use of Terminalia extract can be an appropriate treatment for neurological protection and thus preventing neuropathy in the brain in the early stage of diabetes. The study also recommends that future expanded nutrition studies be prepared for the possibility of producing functional foods and oils from the *Terminalia muelleri* for its effective contribution to reducing the effects of neuropathy of diabetics.

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