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EFFECT OF PIOGLITAZONE AND FLUOXETINE ON NEUROPATHIC PAIN INDUCED EXPER- IMENTALLY IN DIABETIC RATS

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ABSTRACT

Diabetic neuropathic pain, an important microvascular complication in diabetes mellitus, is recognized as one of the most difficult types of pain to treat due to underlying neural pathological changes such as inflammation and fibrosis. It is not possible to use a single drug as a first-line treatment for diabetic peripheral neuropathic pain. The aim of this work is to study the neural pathological changes and tissue levels of proinflammatory cytokines, TNF- α and IL-6 that may underlie the different types of neuropathic pain (tactile allodynia and thermal hyperalgesia) and to explore the effects of pioglitazone and/or fluoxetine on these changes. Sixty adult male white albino rats were assigned into two main groups, diabetic group with induction

of diabetes by single intra-peritoneal injection of streptozotocin with high cholesterol diet and non-diabetic group fed on normal diet. Each main group was divided into subgroups (n=6 rats each) as follows: [I] control, with no treatment; [II] subjected to peripheral sciatic nerve ligation (PSL) only with no treatment; [III] PSL with pioglitazone treatment; [IV] PSL with fluoxetine treatment and [V] PSL with pioglitazone and fluoxetine combined treatment. All subgroups were tested before, at day 7 and day14 after PSL for tactile allodynia and thermal hyperalgesia followed by measurement of nerve tissue levels of TNF- α and IL-6, quantification of collagen deposition and macrophages counting. We found that PSL significantly increased inflammatory cell infiltration

mainly macrophages and collagen deposition with significant increase of nerve tissue levels of TNF- α and IL-6 in both groups. These changes were associated with significant increase of tactile allodynia and thermal hyperalgesia. Administration of pioglitazone and/or fluoxetine significantly decreased both macrophages infiltration and collagen deposition and nerve tissue levels of TNF- α and IL-6. These effects were associated with significant attenuation of tactile allodynia and thermal hyperalgesia produced by PSL in both diabetic and non-diabetic groups but fluoxetine alone had weaker effect in diabetic group. These results suggested that macrophages infiltration and collagen deposition with associated elevation of tissue proinflammatory cytokines could be a cause of neuropathic pain and administration of pioglitazone and fluoxetine can attenuate neuropathic pain by abolishing these changes.

INTRODUCTION

Neuropathic pain is characterized by pain in the absence of a stimulus and by reducing nociceptive thresholds so that normally innocuous stimuli produce pain (Scholz et al., 2007). Diabetic neuropathic pain, an

important microvascular complication in diabetes mellitus, is recognized as one of the most difficult types of pain to treat (Chong et al., 2007). The management of peripheral diabetic neuropathy consists of excluding other causes of painful peripheral neuropathy, maximizing diabetic control and using medications to alleviate pain (Chong et al., 2007). It is not possible to nominate a single drug as the first-line treatment for diabetic peripheral neuropathic pain. Numerous studies using animal models have proposed candidates for therapeutic targets to reduce neuropathic pain. However currently, there are no good pharmacotherapies for neuropathic pain (Park et al., 2007). Pro-inflammatory cytokines and the mRNA of TNF- α and IL-1 β increased in the brain associated with pain-associated behavior in the rat models of neuropathic pain (Watkins et al., 2003). Peroxisome proliferator-activator receptor (PPAR) is a ligand activated transcription factor belonging to a nuclear hormone receptor superfamily, containing three isoforms (α , β/δ , and γ) (Lebovitz et al 2001). PPAR γ plays a critical physiological role as a primary lipid sensor and regulator of lipid metabolism.

Thus, its ligands are clinically used for treatment of some diseases, including type 2 diabetes (Luna-Medina et al 2005). Some studies evaluated that; pioglitazone attenuates the development and maintenance of allodynia and hyperalgesia in mice with neuropathic pain due to peripheral nerve injury (Goldberg et al., 2005). Antidepressant drugs are reported to be used as co-analgesics in clinical management of migraine and neuropathic pain (Singh et al., 2001). There is increasing recognition that norepinephrine (NE) and serotonin (5-HT) reuptake inhibitors (NRIs and SRIs) are efficacious in treating some types of pain. Studies have not systematically evaluated the relative activity at the NE and/or 5-HT transporter required for maximal efficacy in rodent pain models (Leventhal et al., 2007). Some proinflammatory cytokines contribute to neuropathic pain TNF- α and IL-6 are expected mediators underlying tactile allodynia and thermal hyperalgesia induced by peripheral nerve injury (Moalem et al., 2006). Thus, the aims of the present work were to explore the effects of pioglitazone or/and fluoxetine on tactile allodynia and thermal hyperalgesia in neuropathic pain as well as on the patho-

logical changes induced experimentally in non-diabetic and diabetic rats and to clarify their possible neuroimmune mechanism of action.

MATERIALS AND METHODS

1- Drugs and chemicals:

- Streptozotocin (STZ) powder creamy white: (Sigma Chemicals Co., U.S.A).
- Urethane (Ethyl Carbamate); (Prolabo, Paris) white crystals.
- Fluoxetine powder (Misr Co., Egypt)
- Pioglitazone powder (Unipharma, Egypt)
- Carboxy-methyl cellulose (powder) (El Nasr Pharmaceutical Chemicals Co.)
- Ray Bio ® Mouse IL-6: Ray Biotech.U.S.A.
- MEDGENIX TNF- α EASIA Kit.

2-Animals:

Adult male albino rats (n=60), weighting 150-200 g. They were brought from (Experimental Animal Breeding Farm, Helwan - Cairo) .All animals were housed in controlled laboratory condition at 20 -25C in a 12h light/dark cycle and had free access to standard laboratory chow (El-Nasr Company, Abou-Zaabal, Cairo, Egypt) and water. They have acclimatized for one week and were caged (6/cage) in fully ventilated

room (at room temperature) in pharmacology department, Benha Faculty of Medicine. All experimental protocols were approved by the committee of Benha University.

3 – Study Design:

After acclimatization for 1 week, the rats were assigned into 2 groups' non-diabetic and diabetic group. Each group was subdivided into five experimental groups, 6 rats each and treated for 14 days as follow:

Group (I): Normal control group. Fed normal diet, did not receive any drugs but the sciatic nerve of right lateral hind limb in rats was exposed, and not subjected to ligation.

Group (II): None-treated peripheral sciatic nerve ligation (PSL) group. Fed 40%-cholesterol diet. Peripheral sciatic nerve subjected to ligation according to the modified method of Seltzer Z et al. 2004.

Group (III): Pioglitazone-treated group. Received pioglitazone (25mg/kg/day) by gavage according to Maeda et al., 2008.

Group (VI): Fluoxetine-treated group. Received fluoxetine

(30mg/kg/day) by subcutaneous injection (Jett et al., 1997).

Group (V): Pioglitazone+fluoxetine-treated group. Received pioglitazone (25mg/kg/day) by gavage and fluoxetine (30mg/kg/day) by subcutaneous injection.

Diabetes was induced by a single intraperitoneal injection of streptozotocin (STZ 60 mg/kg) dissolved in cold 0.1 mole citrate buffer (pH 4.5) after fed the rats with high fat diet for 2 weeks as a model for type 2 diabetes (Islam et al., 2007) 72 hours after STZ injection, diabetes was confirmed in rats by showing blood glucose levels increased to > 300 mg/100 ml (Ganda et al., 1976). The blood glucose concentration was measured using a glucometer from blood samples obtained by tail prick.

4- Surgical procedure

The procedure used for induction of neuropathic pain was the model adopted by Seltzer Z et al., 2004. Rats were anaesthetized with urethane (1.5-1.75 g/kg I.P). The left sciatic nerve was exposed at the upper-thigh level, and the dorsal third to half of the sciatic nerve was

tightly ligated. The wound was then sutured and the rats were allowed to recover in their home cage.

5 -Behavioral tests

All the behavioral data were recorded before surgery, at day 7 and day 14 for all subgroups. The paw pressure threshold in response to normal innocuous mechanical stimuli was measured by using an analgesimeter (Randall et al., 1957). Rats were situated on the platform under the situation point, so that this is at 5mm distance. Once the rats are prepared we activate the start pedal. The values on the display will increase in a progressive way and once the selected algesic response has been reached (shaking of the stimulated rat, vocalization etc.), we free the start pedal. The force (g) being exerted at the moment of freeing the start pedal is considered the end-point of the test. Once the pedal is released, the motor will turn counter-wise, at the same time the value shown at the digital display is transmitted through the RS232 serial connector.

On the other hand, thermal sensitivity was determined by measuring hind-paw withdrawal latencies to a radiant heat stimulus according to

Hargreaves et al., 1988 just before and after 7 days in non-diabetic groups, and 14 days after STZ injection with sciatic nerve ligation.

6 -Estimation of serum TNF- α and IL-6:

After the behavioral testing on day 7 and day 14 post-surgery, all rats were deeply anesthetized with sodium pentobarbital and the sciatic nerve was dissected (1 cm in length, including the ligation region). We determined the change in expression of tumor necrosis factor- α (TNF- α) and interleukin-6 (IL-6) in the sciatic nerve

a- Estimation of TNF- α by a sandwich ELISA with (MEDGENIX TNF- α EASIA Kit).

TNF- α was determined using a commercially available kit according to the manufacture instructions.

b- Estimation of IL-6 by ELISA technique using (Ray Bio @ Mouse IL-6):

By following the manufacture instruction according the protocol of Howord et al., 1992.

7- Histopathological examination:

After the behavioral testing on day 7 and 14 post-surgery, all rats were deeply anesthetized and the sciatic nerve was dissected, fixed with 10%formaldehyde; embedded in

paraffin, cut at 5 μ m, stained with hematoxylin and eosin (H&E) and with Masson trichrome and examined. The pathologist was unaware of the treatment protocol.

a- Quantification of Fibrotic Areas

Quantitative analysis of sciatic nerve collagen fibers deposition in masson trichrome-stained tissues was performed by morphometric analysis (James et al., 1990). A total of 10 fields were randomly chosen per mouse and images were taken with a digital camera mounted on a CX41 Olympus optical microscope. Collagenous areas stained with masson trichrome were extracted and analyzed using the NIH Image software. The extent of fibrosis was expressed as the percentage of the stained area relative to the total area. The percentages obtained from the 10 fields were expressed as the mean \pm standard deviation.

STATISTICAL ANALYSIS

Data are shown as mean \pm S.E.M. The results were analyzed by one way analysis of variant (ANOVA) followed by t-test or turkey test with $p < 0.05$ selected as the criterion for statistical significance. The histopathological data were analyzed and

compared using Student's t-test or Mann-Whitney's U test.

RESULTS

1- Behavior test :

Peripheral sciatic nerve ligation (PSL) induced significantly tactile allodynia ligation which calculated as the rats showing hind paw withdrawal in response to lower weight (grams) pressure applied to its hind paw. Administration of pioglitazone (25mg/kg, orally) daily and combination of pioglitazone and fluoxetine once daily alleviated the tactile allodynia in non-diabetic and diabetic groups. Also, administration of fluoxetine (30mg/kg, s.c) once daily significantly attenuated tactile allodynia in non-diabetic rats but, could not successfully improve tactile allodynia in diabetic rats.

Regarding the thermal hyperalgesia, it was observed that peripheral sciatic nerve ligation-induced thermal hyperalgesia as sciatic nerve ligation groups had significantly shorter withdrawal latencies, and this was reversed by pioglitazone administration and combination of pioglitazone and fluoxetine in non-diabetic and diabetic rats. On the other hand, administration of fluoxetine (30mg/kg,

s.c) improved thermal hyperalgesia elicited by peripheral nerve injury in comparison to the control group in non-diabetic rats. But administration of fluoxetine (30mg/kg, s.c) produced no significant improvement in thermal hyperalgesia in diabetic rats (table 1).

2- Production of inflammatory cytokines:

TNF- α and IL-6 were at a low level in the control group. Sciatic nerve ligation was associated with marked increase in the expression of both cytokines TNF- α and IL-6 in all examined nerve tissue on day 7 and 14. Pioglitazone and fluoxetine, sup-

pressed significantly the elevation of both cytokines in all examined tissues (table 2).

3- Histopathological examination

Sciatic nerve ligation produced inflammatory cell infiltration with increased collagen fibers deposition in the injured sciatic nerve in comparison to normal control group. Administration of pioglitazone and /or fluoxetine showed less inflammatory cell infiltration with significant reduction of collagen deposition in both groups non-diabetic group. But fluoxetine alone show lesser improvement in diabetic group ($P < 0.006$ vs. 0.0009 in non-diabetic group (Figure 1A, 1B)

Table (1): showing effects of pioglitazone and fluoxetine on tactile allodynia and thermal hyperalgesia which enhanced by peripheral sciatic nerve ligation.

Groups	Threshold (g)		Latency (S)	
	Non-diabetic groups	Diabetic groups	Non- diabetic groups	Diabetic groups
Normal control group	340.3 \pm 14	310.2 \pm 16	6.8 \pm 1.6	5.9 \pm 2.2
Sciatic nerve ligation group (PSL)	152.8 \pm 8 P1<0.05	90.8 \pm 8 P1<0.05	3.2 \pm 0.14 p1<0.05	3.1 \pm 0.12 p1<0.05
Pioglitazone group (25mg/kg)	260.4 \pm 18 P2<0.05	270.4 \pm 10 P2<0.05	4.9 \pm 0.1 p2<0.05	4.7 \pm 0.1 p2<0.05
Fluoxetine group (30 mg/kg)	280.6 \pm 20 p3<0.05	160.8 \pm 13 p3>0.05	3.8 \pm 0.4 p3<0.05	3.8 \pm 0.19 p3>0.05
Pioglitazone (25mg/kg)+Fluoxetine(30mg/kg)	305.1 \pm 23 P4<0.05	290.6 \pm 14 p4<0.05	5.6 \pm 0.5 p4<0.05	4.8 \pm 0.13 p4<0.05

P1 compares values in control groups with normal values

P2 compares values in pioglitazone group with PSL group

P3 compares values in fluoxetine group with PSL group

P4 compares values in pioglitazone (25mg/kg)+fluoxetine (30mg/kg) with PSL group

Table (2): showing effects of pioglitazone and fluoxetine on sciatic nerve levels of TNF- α and IL-6.

Groups	Non diabetic		Diabetic	
	TNF- α	IL-6	TNF- α	IL-6
Normal Control group	0.25 \pm 0.01	0.08 \pm 0.001	0.03 \pm 0.001	0.12 \pm 0.01
PSL group	0.72 \pm 0.013 P1<0.05	0.25 \pm 0.011 P1<0.05	0.82 \pm 0.011 P1<0.05	0.29 \pm 0.012 P1<0.05
Pioglitazone group (25mg/kg)	0.45 \pm 0.03 P2<0.05	0.16 \pm 0.013 P2<0.05	0.35 \pm 0.03 P2<0.05	0.22 \pm 0.011 P2<0.05
Fluoxetine group (30 mg/kg)	0.46 \pm 0.02 P3<0.05	0.18 \pm 0.002 P3<0.0	0.51 \pm 0.014 P3<0.05	0.21 \pm 0.012 P3<0.05
Pioglitazone (25mg/kg)+Fluoxetine(30mg/kg)	0.42 \pm 0.004 P4<0.05	0.13 \pm 0.005 P4<0.05	0.46 \pm 0.03 P4<0.05	0.17 \pm 0.005 P4<0.05

P1: Significant difference from normal control group
 P2 compares values in pioglitazone group with PSL group
 P3 compares values in fluoxetine group with PSL group
 P4 compares values in pioglitazone (25mg/kg) + fluoxetine (30mg/kg) with PSL group

Table (3): showing effects of pioglitazone and fluoxetine on % of collagen deposition in different groups compared to the normal control group

Groups	Non diabetic		Diabetic	
	% Fibrosis	P-value	% Fibrosis	P-value
Normal Control group	3.9 \pm 0.16		3.9 \pm 0.16	
PSL group	9.23 \pm 0.73	P1<0.0001	9.53 \pm 0.32	P1<0.0001
Pioglitazone group (25mg/kg)	6.93 \pm 0.82	P2<0.0057	7.12 \pm 0.91	P2<0.0008
Fluoxetine group (30 mg/kg)	6.02 \pm 0.95	P3<0.0009	7.83 \pm 0.93	P3<0.006
Pioglitazone(25mg/kg)+ Fluoxetine (30mg/kg)	4.34 \pm 0.72	P4<0.0001	5.021 \pm 0.63	P4<0.0001

P1: Significant increased collagen deposition from normal control group
 P2 compares % fibrosis reduction in pioglitazone group with PSL group
 P3 compares % fibrosis reduction in fluoxetine group with PSL group
 P4 compares % fibrosis reduction in pioglitazone (25mg/kg) + fluoxetine (30mg/kg) with PSL group

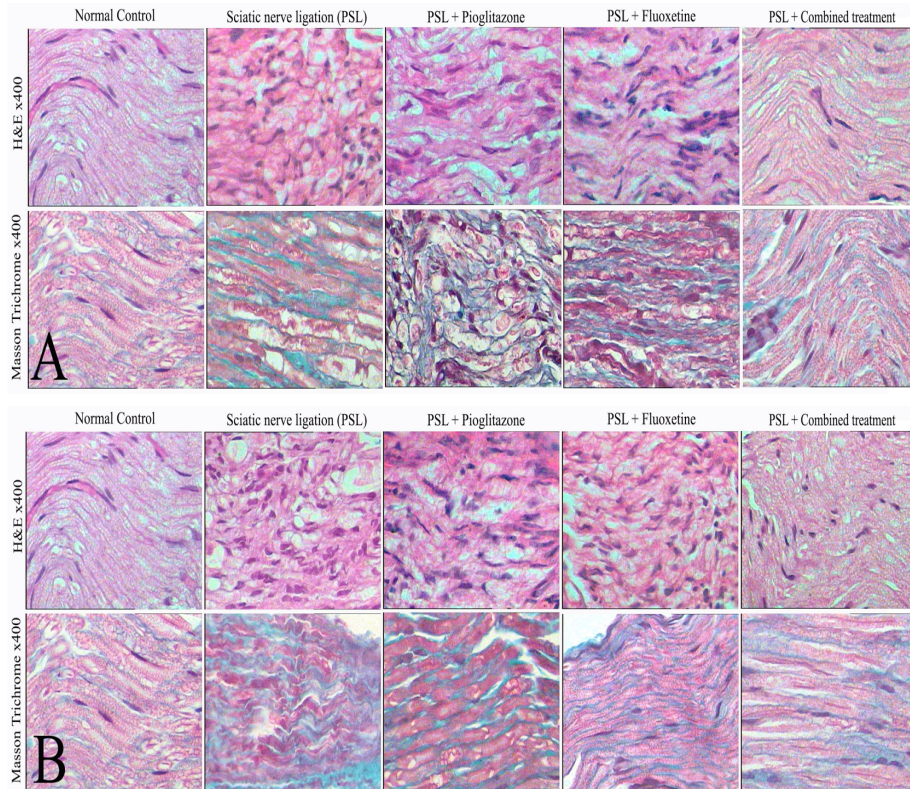


Figure (1): Effects of pioglitazone and fluoxetine on histopathological changes in rats with: A) Non-diabetic peripheral sciatic nerve neuropathy. B) Diabetic groups.

Sciatic nerve ligation produced inflammatory cell infiltration with significant increase in the collagen fibers in the injured sciatic nerve in comparison to normal control group. Administration of pioglitazone and /or fluoxetine decrease the inflammatory cells and significantly reduced the amount of collagen in diabetic and non-diabetic groups but fluoxetine alone showed milder improvement in diabetic group.

DISCUSSION

Results of the present work revealed that administration of pioglitazone (25mg/kg/day) attenuated tactile allodynia and thermal hyperalgesia. These results are in agreement with Maeda et al., (2008) who found that pioglitazone attenuated the development and maintenance of allodynia and thermal hyperalgesia in mice with neuropathic pain due to peripheral nerve injury. Also, Papans et al., 2011 reported that pioglitazone has been shown to improve experimental diabetic neuropathy and alleviate neuropathic pain. Wiggins et al., 2008 observed that rosiglitazone reduced oxidative stress and prevented the development of thermal hypoalgesia in streptozotocin-induced diabetic mice. In addition, Churi et al., 2008 found that PPAR δ ligand-induced activation of spinal PPAR-gamma rapidly reverses nerve injury-induced mechanical allodynia. On contrast to these finding, Shibata et al., 2000 reported that troglitazone was less effective in controlling neuropathy in the Zucker diabetic fatty (ZDF) rats. This difference in our results may be due to uses of different doses and different species of animals.

In the present study, fluoxetine significantly attenuated tactile allodynia and thermal hyperplasia. These effects are in line with those reported by Kesim et al., 2006 that fluoxetine showed significant antinociceptive effect in non diabetic mice, but they could not successfully show this effect in diabetic mice. In disagreement with these findings, Sounvoravong et al., 2007 who stated that fluoxetine itself lacks antinociceptive properties in diabetic and sciatic nerve ligation mice, as model of neuropathic pain. Jett et al., 1997 added that fluoxetine (3-30mg/kg, s.c) did not inhibit either hyperalgesia or allodynia in formalin model and the L5/L6 spinal nerve ligation model of neuropathic pain.

Some proinflammatory cytokines contribute to neuropathic pain. TNF- α and IL-6 are accepted mediators underlying tactile allodynia and thermal hyperalgesia induced by peripheral nerve injury (Moalem et al., 2006). Sciatic nerve injury increases the levels of both cytokines in the sciatic nerve, dorsal root ganglion, and spinal cord (Murphy et al., 1995, Deleo et al., 1996, George et al., 1999, and Ignatowski et al., 1999 which agrees with the present study.

Transcription of both cytokines is regulated by transcription factor binding sites within the promoter. It has been reported that promoter activities of both TNF- α and IL-6 are driven by binding of NF- κ B, AP-1, and STAT, primary proinflammatory transcription factors (Chinetti et al., 2000). We therefore, tested whether pioglitazone and fluoxetine blocked partial sciatic nerve ligation induced up-regulation of both cytokines. Administration of pioglitazone and fluoxetine after PSL reduced increases in the expression level of both cytokines in all examined tissues. These results strongly suggest that pioglitazone and fluoxetine alleviates tactile allodynia and thermal hyperalgesia, at least in part, through inhibiting the upregulated proinflammatory cytokines. These results are in congruent with those of Maeda et al., 2008 who reported that pioglitazone alleviates neuropathic pain through attenuation of proinflammatory cytokine upregulation by peroxisome proliferative activated receptor gamma stimulation.

Finally, histopathological examination of sciatic nerve showed inflammatory cell infiltration with increased amounts of collagen after

partial sciatic nerve ligation. Administration of pioglitazone and fluoxetine decreased the histopathological changes in the sciatic nerve sections of the treated diabetic and non diabetic rats. These results are in line with Takahashi et al., 2011 who reported that, rosiglitazone treatment in the early phase of neuropathic pain significantly alleviated the development of tactile allodynia by regulating macrophage infiltration and production of proinflammatory molecules at the inflamed site.

In conclusion, pioglitazone in combination with fluoxetine may be an effective combination in attenuating diabetic and non-diabetic neuropathic pain through inhibiting the over expression of pro-inflammatory cytokines with subsequent decrease in inflammatory cell infiltration and nerve fibrosis.

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الملخص العربى

تأثير تآزر الفلوكستين مع بيوجليتازونى على السيطرة على السكر واللمس وفرط الالتهام الحرارى فى الفئران الغير مصابة بالسكرى والمصابة بالسكرى والمعرضة للإصابة بالتهاب العصب الطرفى.

أمانى نصر عبد الهادى إبراهيم و عمرو عوض الكارف

قسمى الفارماكولوجى - كلية طب بنها

جامعة بنها وقسم الباثولوجى - كلية طب المنصورة

آلام الأعصاب الطرفية هو مشكلة هامة وخاصة بين مرضى السكرى. والعلاقة الدقيقة بين السيطرة على سكر الدم، وتحسين شدة إعتلال الأعصاب المحيطية مع السكرى لاتزال مثيرة للجدل .

الهدف : من هذه الدراسة هو إستكشاف آثار بيوجليتازونى (٢٥مجم/كجم، بالفم) وفلوكستين (٣٠مجم/كجم، تحت الجلد)، والجمع بين الدواءين إما فى جرعة منخفضة (١٠مجم/كجم) أو ارتفاع جرعة بيوجليتازونى إلى (٢٥مجم/كجم) وفلوكستين (٣٠مجم/كجم، تحت الجلد) تعطى مرة واحدة يومياً لمدة ٧ أيام و ١٤ يوماً على اللمس وفرط التآلم الحرارية فى نموذج ألم الأعصاب السكرى وغير السكرى فى الفئران وآلية العمل الممكنة ورصد آثارها على مستويات السكر فى الدم. وقياس عامل تآكل الأورام ألفا فى أنسجة الأعصاب فى الفئران المصابة بالسكرى وغير السكرى .

تم تقسيم الفئران إلى مجموعتين :

احدهما غير مصابة بالسكرى والأخرى مصابة بالسكرى عن طريق الحقن داخل الصفاق بجرعة واحدة من الستيريتوزوتوسين (٦٠ مجم/كجم) بعد تغذية الفئران مع حمية عالية الدهون (٤٠٪) من السرعات الحرارية والدهون) لمدة أسبوعين باعتباره نموذج لمرض السكرى من النوع الثانى. وأكد السكرى فى الفئران

من خلال أظهار زيادة مستويات السكر في الدم بعد ١٨ ساعة و ٧٢ ساعة صيام بعد الحقن . وقد تم قياس تركيز السكر في الدم باستخدام عينات دم من وخز الذيل وقياسها بالكلومتر .

ثم تم تقسيم الفئران الغير مصابة والمصابة بالسكري إلى خمسة مجموعات :

المجموعة الأولى :

مجموعة المراقبة وتشمل ٦ فئران .

المجموعة الثانية :

المجموعة المصابة بالتهاب الاعصاب الطرفى بدون أى علاج وتشمل ٦ فئران .

المجموعة الثالثة :

مجموعة البايوجليتازون (٢٥مجم/كجم) وتشمل ٦ فئران .

المجموعة الرابعة :

مجموعة الفلوكسيتين (٣٠ مجم/ كجم) وتشمل ٦ فئران

المجموعة الخامسة :

مجموعة البايوجليتازون (٢٥ مجم/ كجم) والفلوكسيتين (٣٠ مجم/ كجم) وتشمل ٦ فئران .

الفئران لمصابة وغير المصابة بالسكري خضعت لربط العصب الوركى . ثم تم قياس اللمس باستخدام جهاز قياس الألم قبل وبعد الجراحة . وتم تقييم فرط التألم الحرارية الساخنة باستخدام لوحة فى كل من الفئران المصابة بالسكري وغير مصابة بالسكري . وتم قياس مستوى السكر فى الدم لكل المجموعات كما تم قياس وزن الفئران قبل وبعد الجراحة . ثم تمت التضحية بالفئران بعد ١٤ يوم من العملية وتم فحص مجهرى لتحديد التغيرات الباثولوجية المصاحبة فى أنسجة الاعصاب وتعيين مستوى عامل تآكل الأورام ألفا فى أنسجة الأعصاب المصابة وغير مصابة بالسكري

وقد تبين الآتى من النتائج :

١- الإلتهاب الأعصاب الطرفى أدى إلى فرط الألم الناتج عن اللمس وفرط التألم الحرارى لمدة أسبوعين . أما اعطاء بيوجليتازون وحده وفلوكستين وحده وكلا الدوائين معا لمدة ٧ أيام للفئران الغير مصابة بالسكري أدى إلى وهن التألم الناتج عن اللمس ووهن التألم الحرارى، ولكن فلوكستين وحده لا يظهر بنجاح هذه الآثار فى الفئران المصابة بالسكري بعد ١٤ يوما من ربط العصب الطرفى .

٢- وأما اعطاء بيوجليتازون وحده وفلوكستين وحده كان له آثار ضئيلة على سكر الدم فى الفئران المصابة بالسكرى وغير المصابة بالسكرى ولكن مزيج من بيوجليتازونى (٢٥مجم/كجم) وفلوكستين (٣٠مجم/كجم) كان لها آثار كبيرة فى سكر الدم فى الفئران المصابة بالسكرى .

٣- أن زعطاء البيوجليتازون أدلى إلى زيادة وزن الفئران أما اعطاء الفلوكستين فأدى إلى فقدان للوزن فى كلا من الفئران المصابة بالسكرى وغير مصابة بالسكرى .

٤- مستوى عامل تآكل الأورام ألفا كان مرتفعاً فى أنسجة أعصاب الفئران المصابة وغير مصابة بالسكرى والمصابة بالتهاب الأعصاب الطرفى بالمقارنة بالمجموعة الضابطة. أما اعطاء بيوجليتازون وحده وفلوكستين وحده وكلا الدوائين معاً لمدة ٧ أيام للفئران الغير مصابة بالسكرى أدى إلى إنخفاض مستوى عامل تآكل الأورام ألفا .

٥- التغيرات الباثولوجية المصاحبة فى أنسجة الأعصاب كانت فى صورة زيادة فى الخلايا الملتهمة وزيادة فى ترسب الأنسجة الليفية فى الأعصاب والتي تحسنت كثيراً عند المعالجة بمزيج من بيوجليتازونى (٢٥مجم/كجم) وفلوكستين (٣٠مجم/كجم) بالمقارنة باعطاء كلا منهما على حدة .

الخلاصة :

هذا يشير إلى أن بيوجليتازونى وفلوكستين يخفف آلام الأعصاب فى الفئران على حد سواء وذلك من الممكن عن طريق توهين خلوى وإنخفاض مستوى عامل تآكل الأورام ألفا والتي أدت إلى تحسن فى التغيرات المصاحبة فى الأنسجة وكان هناك ارتباط بين مستويات السكر فى الدم والتسكين على الرغم من أن الجمع بين جرعات عالية من كلا من الدوائين يسبب إنخفاض ملحوظ فى مستوى السكر فى الدم. كما أن اضافة الفلوكستين إلى بيوجليتازون منع زيادة الوزن التى يسببها البيوجليتازون فى الفئران .