

Protective properties of vitamin E on hepatic cell injury caused by pregabalin in male rabbits (Biochemical and histopathological study)

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Abstract

This study aimed to reveal the protective properties of vitamin E on liver injury induced by pregabalin (biochemical and histopathological study) in rabbits. Twenty-four local male rabbits were used in this experimental study under suitable conditions of feed and temperature. Rabbits are classified into three groups: Each group 8 rabbits. First group (control group) received distilled water orally daily for 14 days. Second group (pregabalin group) received orally pregabalin 3mg/kg dissolved in normal saline daily for 14 days. Third group was received vitamin E (α -tocopherol) 800 IU orally followed by 3 mg/kg of pregabalin daily for 14 days. The biochemical results showed no significant differences in control group. The ALP, GPT and GOT revealed significant increase in second pregabalin group. The third group showed significant decrease in ALP and GOT and slightly increase in GPT above normal value in control group. The histopathological results showed normal liver histology in control and third group. However, second pregabalin group showed infiltration of inflammatory cells specially leukocytes, dilation of blood sinusoids, portal area, central vein, and presence of activated Kupffer cells.

Keywords: Pregabalin, Biochemical parameters, Histopathological changes, Hepatocytes.

Introduction

There are several types of analgesic medications that are considered preferable for relieving pain, and the most important medications that are used in this field are

gabapentinoids which consider favorite for pain managements and anticonvulsant agent (1). One of Gabapentin important analgesic is pregabalin which used in effectively in post-operative care (2). Pregabalin is similar to gamma amino butyric acid, and is used as

an anti-epileptic and analgesic for nerve pain, It is used as an additional medicine in the treatment of partial epilepsy, whether if it expands to spread throughout the entire body or if it remains confined to one area of the body (3). Antiepileptic drug has important in many studies especially in last ten years. In addition the pregabalin currently used for nervous pain, anxiety and epilepsy (4). Other studies showed anticonvulsant effect produced by pregabalin in combination with another drug. In addition, pregabalin have high effective in the patient with seizures (5). Pregabalin exhibit anticonvulsant effect during experimental seizure in rabbits (6). Prevent colonic seizure induced by some drug as pentylentetrazol (7), and prevent electronic and behavior seizure in rabbits (8). Long term use of pregabalin as anticonvulsant lead to late presence the sings of seizure The pregabalin have protective effect for nerve by reduce injury of brain cortex which result from seizures (9). However the pregabalin have side effect include constipation (10), and injury of liver, accumulation of hemosiderin in tissue and increase liver enzyme and jaundice (11). A-tocopherol or vitamin E Consider important vitamin as antioxidant (12). In rabbits, vitamin E which is lipophilic vitamin. It is become more necessary for growth and general health especially prevent oxidative stress of germ cells and enhanced the fertility. Vitamin E play an important role regulation of gene activity, cellular signaling, initiation of apoptosis and enhancing the function of immune system. Deficiency of vitamin E may be causes lesion in the nervous system (13). The

antioxidant such as vitamin E prevent oxidative stress which consider normal chemical product of nutrition metabolism (14). This study aims to identify the biochemical and histopathological properties of vitamin E on liver injury caused by pregabalin in local male rabbits.

Materials and Methods

The current study, twenty-four local male rabbits were used, five-month-old age, 2400g weight of it. Rabbits were under suitable condition (temperature 22-24 °C and light 12 light vs 12 dark), housed in cages in animal house in college of veterinary medicine, university of Diyala. The rabbits divided into three groups (each group 8 rabbits). Control group was received 1ml orally of normal saline per animal for each day. Group II was received orally 3 mg/kg of pregabalin orally (dissolving in normal saline) for 14 days daily. Group III received vitamin E (α -tocopherol) 800 IU orally followed in 3 hours by 3 mg/kg of pregabalin (dissolving in normal saline) for 14 days daily. The blood sample was collected from jugular vein for biochemical analysis (15). Animal were sedative by xylazine and sacrificed. Tissue sample from the liver for histopathological procedures (16).

Statistical Analysis

Two Way ANOVA single factor TEST was applied for detection the difference of parameters in experimental groups. $p > 0.001$ value statically significant.

Results

The results of biochemical test of liver function showed significant increase in concentration of ALT, GPT and GOT in group animal that received orally 3 mg/kg of pregabalin orally compared to control group. In addition, there is a significant decrease ALP and GOT with slightly increase in GPT third rabbits group received vitamin E (α -tocopherol) 800 IU orally followed hours by 3 mg/kg of pregabalin, (Table 1,2,3,4). In control group, the histopathological study

revealed normal liver tissue including normal portal area, central vein and Kupffer cells (Figure 1). In second group the liver tissue showed infiltration of inflammatory cells specially leukocytes, dilation of blood sinusoids, portal area and central vein, and presence of activated Kupffer cells (Figure 2). In third group, histopathological study revealed normal tissue appearance of liver including and similar to control group (Figure 3).

Table 1: showed value of ALP, GPT and GOT in control group.

Animal number	1	2	3	4	5	6	7	8
ALP TEST (IU/L)	112	112	113	112	114	112	112	113
GPT TEST (IU/ L)	12	12	11	13	13	12	12	12
GOT TEST (IU/ L)	13	12	12	13	12	13	13	12

Table 2: Value of ALP, GPT and GOT in pregabalin group.

Animal number	1	2	3	4	5	6	7	8
ALP TEST (IU/L)	131	131	132	132	132	130	132	132
GPT TEST (IU/ L)	28	28	29	29	30	30	27	29
GOT TEST (IU/ L)	30	32	30	31	31	32	32	32

Table 3: Value of ALP, GPT and GOT in vitamin E and pregabalin group.

Animal number	1	2	3	4	5	6	7	8
ALP TEST (IU/L)	108	108	111	110	110	111	110	109
GPT TEST (IU/ L)	15	14	15	13	16	17	17	17
GOT TEST (IU/ L)	13	11	11	12	13	11	13	14

Table 4: Statistical Analysis of control, pregabalin and vitamin E & pregabalin group high significant difference (p >0.001).

Liver enzyme	Control Group	pregabalin group	Vitamin E & pregabalins group	P value
ALP	112.5000±.26726	131.5000±.26726	109.6250±.41993	0.00000**
GPT	12.0000±0.18898	28.7500±.36596	16.6250±0.32390	0.00000**
GOT	12.6250±0.18298	31.2500±0.31339	12.7500± 0.31339	0.00000**

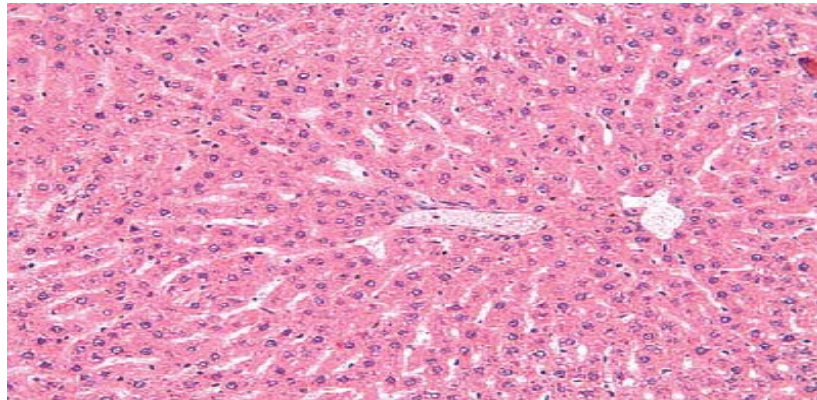


Figure 1: Liver photomicrograph of control group showing normal histological appearance, (H&E stain 40X).

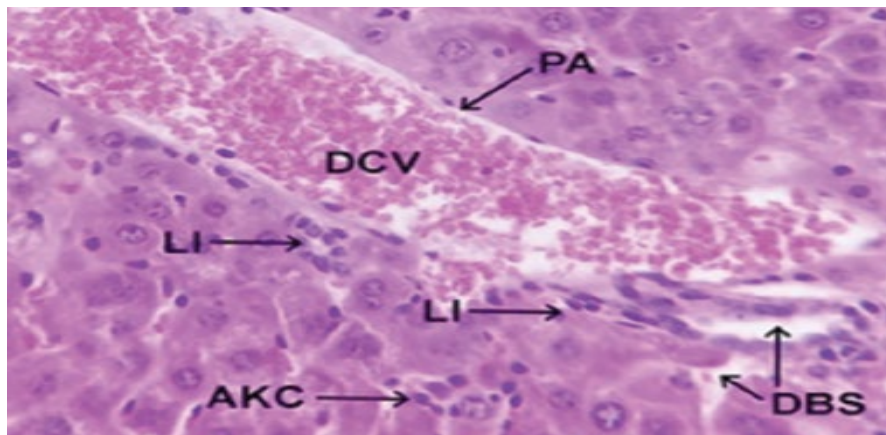


Figure 2: Liver photomicrograph of second pregabalin group after 14 days showing infiltrations of inflammatory cells specially leukocytes (LI), dilation of blood sinusoids (DBS), congestion of portal area (PA) and congestion of central vein (DCV) with presences of activated kupffuer cells (AKC), (H&E stain 40X).

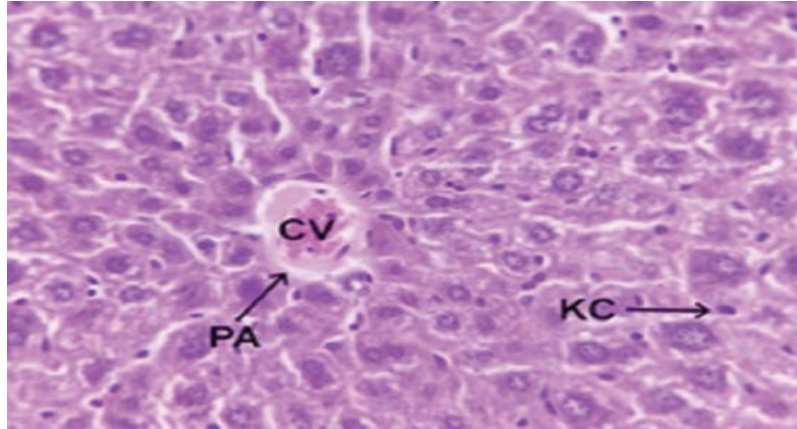


Figure 3: Liver photomicrograph of third Vitamin E and pregabalin group after 14 days later showing normal portal area (PA), central vein (CV) and kupffer cells(kc), (H&E stain 40X).

Discussion

The common uses of antiepileptic causes hepatotoxic injury because of the liver consider the site of metabolism, one of these antiepileptic and or analgesic is pregabalin which is few studies deals with effect of this drug on the liver specially in the rabbits. In current study, the biochemical results of animal of control group showed normal value of ALP, GPT, GOT, and used of pregabalin causes alteration in these parameters. This finding is in agreement with previous study (17). It has been found that there increase of ALT in patient uses of pregabalin suffer from pain in lumbosacral region (11). In this study, the uses of pregabalin lead to elevate of ALP, GPT and GOT which consider indicator of liver injury in rabbits increase the previous enzyme in blood (6). The ALT, GPT and GOT return to normal after administration of vitamin E before 3 hours injection of pregabalin in rabbits. There was no significant differences between first group and third group, this finding might be due to that vitamin E prevent oxidative stress and hepatic cellular

damage by antioxidant activity of it (18). In the present study, control group revealed normal histological appearance of portal area, central vein and Kupffer cells in control group animal. However, the use of pregabalin only in second group showed infiltration of inflammatory cells, dilation of blood sinusoid and presence of active Kupffer cells. This finding is in agreement with previous study (19), which revealed dilated of central vein, infiltration of inflammatory cells and dilation of sinusoids. It has been found that vitamin E help in removal the liver injury caused by long term used of pregabalin by protective the hepatocytes in rabbits (20). Protective properties of vitamin E on liver injury damage caused by cypermethrin the protective role of vitamin E against toxic effect of cypermethrin on the liver. In the current study, the used of vitamin E before the pregabalin lead to enhanced histological appearance of liver compared with only pregabalin group that represent normal tissue structure of liver. The antioxidant activity and similar the histological appearance with control group (21),(22).

Present result in this study in agreement with previous study it has been found that the protective role of vitamin E against pain reducer drug (23). The role of vitamin E to protect liver by prevent lipid peroxidation via reactive oxygen species, decrease the hepatocytes degeneration and establish regeneration of liver cells (24).

Conclusion

Protective role of vitamin E against pregabalin which causes change in biochemical parameters and histopathological changes on liver.

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الخصائص الوقائية لفيتامين اي على الخلايا الكبدية المتضررة بالبريجابالين في ذكور الأرانب (دراسة كيميائية حيوية ونسجية مرضية)

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الخلاصة

هدفت هذه الدراسة إلى الكشف عن الخصائص الوقائية لفيتامين E على إصابة الكبد الناجمة عن عقار البريجابالين (دراسة كيميائية حيوية ونسجية مرضية) في ذكور الأرانب. تم استخدام أربعة وعشرين ذكراً من الأرانب المحلية في هذه الدراسة التجريبية تحت ظروف التغذية ودرجة الحرارة المناسبة. تم تصنيف الأرانب إلى ثلاث مجموعات: كل مجموعة تحتوي على 8 أرانب. تلقت المجموعة الأولى (مجموعة السيطرة) الماء المقطر عن طريق الفم يومياً لمدة 14 يوماً. تلقت المجموعة الثانية (مجموعة البريجابالين) البريجابالين عن طريق الفم 3 ملغم / كغم مذاباً في الماء المقطر يومياً لمدة 14 يوماً. المجموعة الثالثة تم إعطاؤها فيتامين 800 E (α -tocopherol) وحدة دولية عن طريق الفم يليها 3 ملغم/كغم من البريجابالين يومياً لمدة 14 يوماً. وأظهرت النتائج الكيميائية الحيوية عدم وجود فروق ذات دلالة إحصائية في مجموعة السيطرة. كشفت اختبارات ALP و GPT و GOT عن زيادة ملحوظة في مجموعة البريجابالين الثانية. أظهرت المجموعة الثالثة انخفاضاً معنوياً في ALP و GOT وزيادة طفيفة في GPT فوق القيمة الطبيعية مقارنة بمجموعة السيطرة. أظهرت النتائج النسيجية المرضية أن أنسجة الكبد طبيعية في مجموعة السيطرة والمجموعة الثالثة. أما مجموعة البريجابالين الثانية فقد أظهرت ارتشاح الخلايا الالتهابية وخاصة الكريات البيض وتوسع الجيوب الدموية والمنطقة البوابية الكبدية والوريد البوابي المركزي ووجود خلايا كوبفر النشطة.

الكلمات المفتاحية: البريجابالين، الفحوصات الكيميائية الحيوية، التغيرات النسيجية المرضية، الخلايا الكبدية.