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# Effect of induced epilepsy on some biochemical parameters in female rats

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**Abstract**

The activity of cholinesterase and some biochemical parameters of blood such as glucose, cholesterol and phospholipids were estimated in 52 epilepsy induced females of Wister albino rats. Animals of this experiment were divided into two groups, group (I) regarded as control and group (II) administrated subcutaneously by pentylenetetrazole 100mg/kg and divided in to three sub-groups according to the time of samples collection 3 hrs, 24 hrs and 1 week. The results revealed that epilepsy induction caused a significant inhibition of serum cholinesterase activity 3 hrs after induction while in the brain, the activity of cholinesterase was significantly increased after 24 hrs Serum glucose level was significantly elevated after 3 hrs and 24 hrs of induction, total cholesterol and phospholipids were not changed. From the results obtained in this study, it can be concluded that epilepsy caused significant changes in cholinesterase activity in brain and serum in addition to the glucose level in the serum.

***Keywords:*** Epilepsy; Pentyleneterazole; Cholinesterase.
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# تأثير استحداث الصرع على المعايير الكيموحيوية في ءاناث الجرذان المختبرية

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**الخلاصة**

استهدفت هذه التجربة معرفة تأثير الصرع المستحدث بأستخدام مادة البنتلين تترازول على فعالية انزيم الكولين استريز في مصل الدم ونسيج الدماغ وكذلك بعض المقاييس الكيموحيوية كالكلوكوز، الكولستيرول والدهون الفسفورية في مصل دم (٥٢ انثى جرذ) من نوع Wister albino. شملت التجربة مجموعتين من الجرذان، المجموعة الاولى مثلت مجموعة السيطرة والمجموعة الثانية فقد استحدث فيها الصرع عن طريق حقنها بالبنتلين تترازول بجرعة ١٠٠ ملغم/كغم من وزن الجسم تحت الجلد، وقسمت الى ثلاثة مجاميع ثانوية اعتماداً على وقت جمع الدم وقتلها بعد استحداث الصرع وهي (٣، ٢٤ ساعة واسبوع). أوضحت النتائج ان استحداث الصرع في الجرذان ادى الى حدوث انخفاض معنوي في فعالية انزيم الكولين استريز في مصل الدم بعد ٣ ساعات من استحداث الصرع في حين ادى الصرع الى ارتفاع معنوي في فعالية الانزيم في نسيج الدماغ بعد ٢٤ ساعة، اما مستوى كلوكوز مصل الدم فقد شهد ارتفاعاً معنوياً بعد ٣ و ٢٤ ساعة من بدء استحداث المرض، بينما لم يؤدي استحداث الصرع في الجرذان الى حصول اي تغييرات معنوية في مستويات الكولستيرول والدهون الفوسفورية في مصل الدم. نستنتج من هذه التجربة ان استحداث الصرع في اناث الجرذان ادى الى حدوث تغييرات معنوية في مستويات انزيم الكولين استريز في كل من مصل الدم ونسيج الدماغ بالاضافة الى حدوث تغييرات معنوية في مستوى الكلوكوزفي الدم.

## Introduction

Epilepsy is a devastating chronic neurological disorder that affects about 0.8% of the population worldwide. The clinical hallmark of epilepsy is recurrent seizures, which consist of neuronal synchronized discharges (1). An epileptic seizure is characterized by excessive and/or hyper synchronous and usually self-limited activity of neurons in

the brain (2). Epilepsy is also defined as a status of recurrent seizures (3). During a cluster two or more seizures occur within a 24 hour time span (4). A seizure of 30 minutes or longer duration or recurrent seizures without resumption of baseline central nervous system function are called status epilepticus (2,5).

The word 'Epilepsy' is derived from the Greek word 'Epilepsia', which in turn can be broken into epi (upon) and lepsis (to take hold, or seizure) (6,7). Epilepsy can affect people of any age (8), but is more common in children (9). Epilepsy increases the person's risk of premature death by about two to three times compared to the general population (10).

Epilepsy can be put into two main groups according to its causes; 1- Idiopathic (Primary) Epilepsy: It is the most common type of epilepsy, which means the cause can not be identified. Such seizures are usually between age 5 and 20 years, but can occur at any age (10,11). 2- Symptomatic (Secondary) Epilepsy: This is occurring when there is a known structural cause for why a person's epilepsy has started. The cause could be brain damage from a loss of oxygen, or trauma during birth, a sever blow to the head, a stroke that starves the brain of oxygen (causes brain tissue abnormally excitable), infection such as encephalitis, meningitis, or a scar, abscess, tumor in the brain (9-12).

Epilepsy can occur in animals other than in humans. Canine epilepsy is often genetic; epilepsy in cats and other pets is rare, likely because there is no hereditary component to epilepsy in these animals. It is not known precisely how common canine epilepsy is occurring, but studies show that it affects 4-14% of dog. It is more common in certain breeds, including Beagles, Dachshunds, German Shepherds, and Golden Retrievers (13).

Pentyleneterazole (PTZ) has been used in several trails to cause generalized seizures in animal models (14), simulating the effects generated in human beings with epilepsy (15). It is considered non-competitive GABA antagonist (GABA-$_A$ receptor antagonist) (16). So, PTZ is used for induction of experimental epilepsy (14). Seizures are induced by acute or chronic (repeated) PTZ administration (16).

Induction of epilepsy in male rats showed no change in the activity of serum cholinesterase (ChE) (17) while (18) reported that serum ChE level increased significantly in treated epileptic patients compared to the control.

Abnormal glucose levels, whether too high or too low, can cause seizure. The problem is especially pertinent, whose blood glucose levels can fluctuate widely over the course of a day, as a result of variation in insulin levels in case of diabetes, or other metabolic factors (19). In idiopathic or generalized epilepsy, serum cholesterol concentration decreased, especially just before a seizure. On other hand, some epilepticus showed increase of serum cholesterol (20).

Pilocarpine administration in adult rats and the resulting status epilepticus produced a significant decrease in brain ChE activity (21) while a significant increase in brain ChE activity was observed in rats with cobalt induced epilepsy (22).

Because most of the researches concerned with epilepsy were done in the male rats and also in patients treated with antiepileptic drugs, we suggest studying this disease in female without giving any drug to animals, so the objective of this study is to investigate the effect of induced epilepsy solely without any treatment on the biochemical parameters in blood and tissues of female rats.

## Material and methods

### Animals

The study was performed on (52) females Wister albino rats, with mean body weight about (160-250 gm) and average age of (2-2.5 months). The animals were housed in groups of (5-6 per cage), in a room with a controlled light/dark cycle (12 hrs light /12 hrs dark) at (22 ± 2°C) and were allowed free access to diet and tap water during the entire experimental period.

### Experimental design

52 rats were divided into 2 groups: group 1 included 10 rats were served as control and injected with a corresponding amount of saline (0.9%). Group 2 included 42 rats were induced epilepsy by administrating subcutaneously with PTZ in a single dose of 100mg/kg B. W. This group was divided into 3 subgroups according to the time of blood collection after induction of epilepsy: 3 hrs group (14 rats), 24 hrs group (14 rats) and 1 week group (14 rats).

### Induction of epilepsy in rats

PTZ (Sigma, Germany) was dissolved in saline at 100mg/ml and administered to rats subcutaneously (S.C.) under the loose skin behind the neck in a single dose (100 mg/kg B.W. in a volume of 0.1 ml/100 gm B.W.) (23). Control animals were treated with a corresponding amount of saline (0.9%). Animals were observed after injection with PTZ for 3 hrs to observe the occurrence of seizer activity. Approximately 6-28 min. after PTZ injection, most of the animals entered status epilepticus (42.8% of PTZ-treated animals were died due to status epilepticus).

### Samples collection and biochemical analysis

Blood samples were collected from the retro-ocular vein (24) into clear dry centrifuge tubes after 3 hrs, 24 hrs, and 1 week allowed clotting; serum was separated after centrifugation at 3000 rpm for 15 minute. Serum and brain ChE activity were measured electrometrically according to the method described by (25), serum glucose level was

enzymatically measured using standard enzymatic kit (Spinreact, Spain) (26), total cholesterol was measured enzymatically using standard enzymatic assay (Biolabo reagents, France) (27) and phospholipids was estimated according to (28).

**Results**

In the serum, after induction of epilepsy there was a significant (P<0.05) inhibition of ChE activity noted after 3 hrs while after 24 hrs and 1 week there was no significant change compared with control group. Glucose level showed a significant (P<0.05) increase after 3 hrs and 24 hrs compared with control group, But after 1 week the increase in glucose level was not significant comparing with control group. No significant changes in both Total cholesterol and phospholipids concentration in all experiment periods compared with control (Table 1).

In the brain, the activity of ChE enzyme was significantly increased after 24 hrs of the experiment. This result was significantly different from those of the control at P<0.05. While after 3 hrs and 1 week the increase of the enzyme activity was not significant in comparing with control group (Table 2).

Table 1: Effect of inducing epilepsy on some biochemical parameters in serum of female rats.

| Biochemical parameters | Control | After inducing epilepsy | | |
|---|---|---|---|---|
| | | 3 hrs | 24 hrs | 1 week |
| ChE activity (Δ pH/30 minutes) | 0.52 ± 0.01 A | 0.42 ± 0.02 B | 0.53 ± 0.02 A | 0.59 ± 0.04 A |
| Glucose (mg/dl) | 88.01 ± 4.56 C | 143.94 ± 10.14 B | 186.98± 19.96 A | 108.67± 7.01 C |
| Total cholesterol (mg/dl) | 112.25 ± 17.01 A | 123.69 ± 11.40 A | 89.92 ±5.46 A | 110.37 ± 12.22 A |
| Phospholipids (mg/dl) | 167.90 ± 12.14 A | 178.08 ± 10.15 A | 148.03 ± 4.86 A | 166.23 ± 10.87 A |

Number of rats10 in control and 7 in other groups, Values are expressed as mean ± SE.
Different letters in the same row refer to the significant differences at level P<0.05.

Table 2: Effect of inducing epilepsy on ChE activity in the brain of female rats.

| Biochemical parameters | Control | After inducing epilepsy | | |
|---|---|---|---|---|
| | | 3 hrs | 24 hrs | 1 week |
| ChE activity (Δ pH/30 minutes) | 0.14 ± 0.01 B | 0.19 ± 0.02 AB | 0.27 ± 0.06 A | 0.14 ± 0.02 B |

Number of rats10 in control and 7 in other groups, Values are expressed as mean ± SE.
Different letters in the same row refer to the significant differences at level P<0.05.

**Discussion**

This study indicates that epilepsy induction in female caused a significant decrease in the activity of serum ChE after 3 hrs (Table1). (29) stated that inducing epilepsy in rats by pentroxifylin injection leading to decrease the ChE activity after 24 hrs of the experiment. Epilepsy induction by administration of carbofuran leading to a maximum inhibition of ChE activity by 82-90% in rats, carbufuran induced neuronal hyperactivity blocks pathways associated with oxidative damage in neurons (29) While (18) showed that serum ChE was significantly increased in a group of epileptic patients (male and female) treated with anticonvulsant drugs in different ages. (17) refered that epilepsy induction in male rats did not change the activity of ChE in serum.

Epilepsy induction caused a significant increase in serum glucose level after 3 hrs and 24 hrs with no significant change in its level after 1 week. This result is in agreement with the study by (30) who found that in adult rats susceptibility to clonic and tonic clonic-induced seizures was positively correlated with blood glucose concentration, as the increased glucose concentration was associated with proconvulsant effects and also in agreement with the results of (17) who observed a significant increase in the serum glucose level after 3 hrs and 24 hrs of epilepsy induction as compared with their levels before epilepsy induction. After 1 week, the level of glucose decreased and returned to normal pre-inducing level. (31) indicated that plasma glucose level increase at 3 min. after the onset of epilepsy induction and elevated to peak after 10 min of seizure in rats induced to epilepsy by intense sound exposure. These results were disagree with (32) whose study indicated that spontaneous epileptic rats showed decrease in serum glucose level due to the frequent occurrence of tonic convulsions and wild jumping associate with low body weight. Administration of glucose to rats prior inducing seizures by exposure to Atmosphere Absolute Oxygen (ATA) lead to increase time-to seizure by 90% because glucose offered partial protection to rats then blood glucose levels were consistently elevated in rats following oxygen exposure (33).

In the present study, serum total cholesterol and phospholipids levels were not changed in comparing with control levels (Table 1). This study is not in consistent with the study carried by (32) who found that serum cholesterol and phospholipids levels decreased significantly in spontaneous epileptic rats. (17) observed both total cholesterol and phospholipids levels decreased significantly after 3 hrs and 1 week from inducing epilepsy in male rats as compared with results before induction.

Inductions of epilepsy in rats caused increase the activity of ChE in the brain after 24 hrs these result is disagree with the study of (21) who indicated that administration of pilocarpine induced status epilepticus and leading to a significant decrease in brain ChE activity in adult rats and of (17) who found that induction of epilepsy does not change the activity of ChE in the male rats. Cerebral ChE decreased reversibly in rats soman-induced to epilepsy treated with donepezil and procyclidine (34).

At conclusion, inducing epilepsy in female rats lead to changes in blood ChE and glucose level in the serum and brain tissue in female rats. More prospective studies are needed to clarify the causes around these changes in blood parameters in females compared with males and why there are differences between male and female.

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