

SYZYGIUM AROMATICUM I. AND THEIR EFFECT ON SEIZURED CHICKENS

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ABSTRACT

Cloves (*Syzyguim Aromaticum* L.) represent very interesting plant that has been used for many medicinal purposes such as treatment of convulsions due to its potent antioxidant activity. The purpose of this study is to investigate the anti-convulsant effect of aqueous extract of *Syzyguim Aromaticum* L. (cloves) at the doses (100,200,300 mg/kg), sodium valproate (200mg/kg) act as reference standard on serum total antioxidant capacity (T-AOC), gamma amino butyric acid (GABA) concentration and cholinesterase (ChE) activity in serum and brain, electrolytes (sodium Na⁺, potassium K⁺, chloride Cl⁻ and total calcium Ca⁺²), glucose and total protein in serum of chicks after induction of seizures by pentylenetetrazole (PTZ), a plant used in traditional medicine as anticonvulsant. 72 chicks were recruited in this study were randomly divided into 6 groups of 12 each. Negative control received normal saline, positive control was given only PTZ, group III was given sodium valproate orally. The rest of three groups were given the extract at previous concentrations given through oral role. Phytochemical analysis of the aqueous extract of cloves revealed the presence of psychoactive compounds such as flavonoids, cardiac glycoside, tannins, and alkaloids. The present work suggest that the aqueous extract of plant had increased level of T-AOC,GABA and ChE in serum of chicks, which were decreased due to the susceptibility to PTZ induced seizure, and decreased the concentration of Na⁺ and glucose in the serum. We reported that the cloves extract had quite low effect against PTZ induced seizure in spite of their positive effect on

different biochemical parameters in serum; this may be due to the low concentration of doses used, which may be inadequate to produce obvious behavioral results.

INTRODUCTION

Ethnomedicine or phytotherapy is defined as the science of using herbal remedies to treat the disease, have remained as a component of health care system of many societies in spite of the availability of well alternative therapy (1). Plants are the major sources of drug in the modern as well as traditional system of medicine throughout the world. Plants and its phytoconstituents are used to prevent and cure various disorders like epileptic seizures (2). Drug-resistance is very important clinical problem in case of epilepsy. This phenomenon is observed in about 1/3 of patients and is defined as seizures that continue to occur despite treatments with at least three appropriate antiepileptic drugs at maximum tolerated doses (3). The ideal anti-seizure drug causes suppression in all seizures without appearing side effects. While the drugs used nowadays fail to control seizure activity in some patients, and cause unfavorable side effects (4). The plants are used efficiently as a natural treatment for different diseases with fewer side effects and lesser drug interactions. Thus the herbal drugs make anticonvulsant treatment more rational and patient friendly. There is a growing interest between phytochemical constituents of plant and their pharmacological activities. So the proper uses of technological advances helped the traditional medicine which has become a necessity in order to promote the research in phytopharmacological field (2). Standard antiepileptic drugs usually caused side effects in about 50% or more of treated patients, such as hepatic failure, serum sickness and hepatitis (5). Herbal drugs used for treatment of epilepsy in traditional medicine are acting at target side having same mechanism of action as that of synthetic drugs (1) and (6). With the introduction of allopathic drugs, the use of crude drugs from medicinal plants is on the decline and subsequently this traditional knowledge may be lost in the near future (1). The introduction of new plant gives hope that the proportion of patients with uncontrolled epilepsy will substantially decrease.

Epilepsy is one of the major brain disorders worldwide. This term is derived from the Greek word *epilepsia*, which means "falling sickness" and may be called "seizure", or

"convulsion" (7). Epilepsy is a medical condition that produces recurrent seizures. The condition is characterized by repeated seizures or fits (7) and, (8). Seizure is uncontrolled electrical activity in the brain, which may produce a physical convulsion, minor physical signs, thought disturbances, or a combination of symptoms. There are different mechanisms through which seizures can develop in the normal or pathologic brains include, decrease of inhibitory mechanism due to GABA, enhancement of the excitatory mechanism and enhancement of endogenous neuronal burst firing by enhancing voltage dependent calcium currents (9). The type and symptoms of seizures depend on where the abnormal electrical activity takes place in the brain, what its cause, and such factors as the patient's age and general state of health. Hyperactivity of excitatory neurotransmitters systems, insufficient GABA_A-receptor-mediated neurotransmission, and defect in intrinsic properties of neuronal membranes are remaining as the most important mechanisms of seizures (10). Seizures can be caused by head injuries, brain tumors, lead poisoning, maldevelopment of the brain, genetic and infectious illnesses, and fevers. In fully half of the patients with seizures, no cause can yet be found. Cellular imbalance can contribute to the disturbed function of the ligand- and voltage-dependent sodium, potassium, chloride and calcium channels, resulting in epileptiform activity (10).

Syzygium Aromaticum L. (synonym: *Eugenia caryophyllata*) from the Mirtaceae family commonly known as clove, is an median size tree (8-12 m), native from the Maluku islands in Indonesia. (11). *Syzygium Aromaticum* L. is a common plant has been employed for centuries as medicinal plant, traditionally there cloves are used as anticonvulsant, analgesic, anesthetic, anti-Alzheimer, antibacterial, anticancer and anti-inflammatory, antioxidant, antispasmodic and neuroprotective properties (12) and (13).

In order to get more insight into the effect of aqueous extract of (*Syzygium Aromaticum* L.) on T-AOC, GABA, ChE activity, electrolytes, glucose and total protein in seizure, we performed a comparative analysis of previous parameters with one of the most effective antiepileptic drug for certain types of seizure.

MATERIAL AND METHODS

1. Preparation of plant aqueous extract:

The dried cloves of *Syzyguim Aromaticum* L. were purchased from a local market in Dohuk, Kurdistan region/ Iraq. The specimen was reduced to a fine powder and successively extracted according to the method described by Sushma *etal.*, (2012) (14). The powder were suspended in soxholet apparatus, at 25g/250ml of distilled water for (7-8 hrs.), the extract was concentrated by rotary evaporator at 60°C for about 3hrs. /L under reduced pressure. After that the crude extract produced was let overnight in dark room to further evaporate then preserved in airtight and dark bottle and kept in refrigerator (4 °C) and used during 5-10 days. For the preparation of samples at concentrations 100,200, 300 mg /kg the weighed amount of extract was dissolved in distilled water and administered to animals orally using gavages needle.

2. Animals used:

We enrolled (72) chicks of either sex, were obtained from local hatchers, Duhok province/Kurdistan Region/ Iraq. Their ages were one day and all the animals were acclimatized to laboratory condition for two weeks before commencement of experiment, weighing about (200-300g). The chicks were maintained in a well ventilated room with normal day light photoperiod (light/dark cycle) in cages. The chicks were fed with standard chick food and water was given *ad libitum*.

3. Experimental design:

The (72) were randomly divided into 6 groups, each group composed of 12 chicks. The first group served as (negative control) and was injected subcutaneously with normal saline. The second group (positive control) was injected subcutaneously with freshly prepared solution of PTZ at 80mg/kg (15). The third group was treated orally with antiepileptic drug (sodium valproate) at 200mg/kg (16) for 6 days. The remaining three groups were received cloves aqueous extract of *Syzyguim Aromaticum* L. at concentration 100,200, 300 mg /kg respectively through oral route for 6 days. On the 6th day, seizure was induced to all the groups of chicks except the first one using PTZ subcutaneously in the neck area 30 minute after administering the

treatments. Animals were observed for a period of 30 minutes after PTZ injection for presence or absence of seizures. The Anti-seizure property of the plant extract was assessed by its ability to prevent or delay the onset of convulsions. Onset time, latency of convulsions, the number of animals protected from seizure and % mortality against PTZ induced convulsions were measured.

4. Serum and brain sampling:

Blood samples were collected after overnight fasting at the 6th day after 3 hours from PTZ injection (17) from the jugular vein (18). The blood was collected in the tubes without anticoagulant, and then left for 30 minutes at room temperature. The serum was separated by centrifuge at 5000 rpm for 10 minutes, the supernatant was collected, aliquot and stored at -20°C until use (19) for estimation of T-AOC, GABA, ChE activity, electrolytes, glucose and total protein. At the end of experiment chicks were scarified after slaughtering, whole brain was dissected and stored in plastic container, kept at -20°C until use (20) for determination of GABA and ChE activity.

5. Phytochemical analysis of plant:

The preliminary qualitative phytochemical screening carried out on the crude aqueous extract of cloves to reveal the presence of pharmacologically active constituents as flavonoids, cardiac glycoside, reducing sugar, tannins, saponins and alkaloids was carried out by standard methods as described by Alagpulinsa (2010) and Enemor (2012) (21) and (22).

6. Biochemical analysis:

The quantitative estimation of some serum and brain constituents was undertaken for all chicks groups. T-AOC in serum, GABA in serum and brain was determined by ELISA using standard kit (YH Bioresearch Lab. /China). Electrometric estimation of serum and brain ChE activity was measured according to the method described by Mohammed (2007) (23). Serum Na⁺, K⁺ were determined photometrically by spectrophotometer using standard kit (Gesellschaft/ Germany) according to the method of Trinder (1951) and Hillman and Beyer (1967) respectively (24) and (25), Cl⁻ and Ca⁺² were determined also colorimetrically using standard kit (Biolab/ France) according to the method of Zall et al., (1956) and Moorhead and Briggs (1974) respectively (26) and (27). Serum glucose level was determined enzymatically using

spectrophotometer and standard kit (Biolab/ France) according to the modified method described by Trinder (1969) (28). Finally total protein in the serum was determined colorimetrically using standard kit (Biolab/ France) according to the method of Gornall et al., (1949) (Biuret method) (29).

7. Statistical analysis:

Results were presented as means \pm standard error of mean (S.E.M). The significance of differences among the group assessed using one way analysis of variance (ANOVA) the test followed by Duncan test. Differences were considered significant statistically when $P < 0.05$.

RESULTS

1. Phytochemical Analysis of *Syzygium Aromaticum* L. Extract:

The result in the table (3.1) mention The preliminary phytochemical screening of extract revealed the presence of flavonoids ,cardiac glycosides, tannins and alkaloids compounds in the aqueous extract of plant cloves used in experiment while absence of reducing sugar and saponins in the extract.

Table (3.1): Results of Phytochemical Analysis of *Syzygium Aromaticum* L. Extract.

Constituents	Result	Reaction
Lead acetate of flavonoids	Positive	Orange color observed
Cardiac glycoside test	Positive	Reddish- brown layer
Reducing sugar test	Negative	No change in color
Test of tannins	Positive	Blue-dark precipitate observed
Saponins test	Negative	No foam observed
Test of alkaloids	Positive	Orange to red precipitate

2. Anti –seizure activity:

PTZ produced seizure in all the chicks after 3 minutes from their injection, the signs remained to 20 minutes and the mortality rate is occur in about 50% of animals.

Sodium valproate (200mg/kg) had abolished the seizures and offered 100% protection against PTZ. The extract had a weak anticonvulsant activity against PTZ-induced convulsion. Pretreatment with aqueous extract of cloves at 100 and 200 mg/ kg delayed the onset of seizure produced by PTZ (reduced the duration of seizures) (appear after 5 minutes and remain to 30 min) and increased the threshold for convulsions compared with positive control. In 300mg/kg group the onset of seizure begin after 7 minutes and remain to 36 minutes, all the concentration of extracts decrease the number of deaths and increase in the duration of death time.

3. Effect of cloves of *Syzyguim Aromaticum* L. Aqueous Extract on Biochemical Parameters:

3.1 Effect of cloves of *Syzyguim Aromaticum* L. aqueous extract on serum T-AOC, serum and brain GABA and ChE activity in PTZ treated chicks compared with controls.

In the table (3.2) treatment of chicks with PTZ lead to decrease of T-AOC when compared with negative control group but not reach the significant level. While sodium valproate causes significant increase in T-AOC compared with negative and positive control groups. Treatment of chicks with plant extract at 100,200, 300 mg /kg B.W concentration lead to increasing in the level of T-AOC in comparison with positive control and the increasing correlated with the concentration of the extract.

Level of GABA in the serum was decreased in positive control group in compared with negative control group, treatment of chicks with antiepileptic drugs and plant extract at different concentrations caused significant increase in GABA compared with positive control group. Concentration of GABA in the brain increased only in plant extract treated group at concentration 300 mg /kg compared with positive control group (table 3.2).

ChE activity was decreased in the serum in PTZ treated group compared with normal group, antiepileptic drug and plant extracts lead to increase the level of ChE compared with PTZ treated group. Whereas ChE activity in the brain as shown in the table (3.2), in drug treated group and all treated groups of aqueous extract did changed in compared with negative and positive control groups.

Table (3.2): Effect of cloves of *Syzyguim Aromaticum* L. aqueous extract on serum T-AOC, serum and brain GABA and ChE activity in PTZ treated chicks compared with controls.

Parameters	Groups					
	Negative control	Positive control	Sodium valproate 200mg/kg	Plant extract 100mg/kg	Plant extract 200mg/kg	Plant extract 300mg/kg
T-AOC (U/ml)	DC 5.79	D 4.69	B 7.34	BC 6.77	AB 7.68	A 9.08
GABA serum (ng/ml)	A 22.53	C 6.91	B 11.45	B 10.97	B 11.57	B 13.32
GABA brain (ng/ gm wet tissue)	AB 264.6	B 234.1	AB 263.5	B 217.8	AB 272.8	A 340.9
ChE serum (Δ pH/30 min.)	A 0.61	B 0.49	A 0.67	A 0.61	A 0.62	A 0.66
ChE brain (Δ pH/30 min.)	A 0.52	A 0.46	A 0.51	A 0.52	A 0.51	A 0.54

N=12 chicks in each group, values are expressed as mean +SE,

Different letters refer to the significant differences at level $P < 0.05$.

3.2 Effect of cloves of *Syzyguim Aromaticum* L. aqueous extract on serum electrolytes in PTZ treated chicks compared with controls.

Table (3.3) summarizes the effect of extract on serum electrolytes and total Ca^{+2} ions.

Na^{+} ion showed significant Increase in positive control group compared with negative control group, while in drug treated group and in all three concentration of extract was decreased. No appreciable changes of K^{+} ion have shown among negative, positive and drug treated groups. The 100 and 300 mg /kg of plant extract display significant increasing in comparison with negative and positive control groups. The Cl^{-} ions level as shown in table (3.3). Positive control did not changed in compared with negative control and also drug treated group and all treated group of aqueous

extract did not changed in compared with positive control. Induction of seizure in chicks cause no significant changes in total Ca^{+2} compared with negative control group, sodium valproate display significant decreasing in comparison with negative and positive control groups and all treated groups of aqueous extract did not shows significant differences in compared with positive control group

Table (3.3): Effect of cloves of *Syzyguim Aromaticum* L. aqueous extract on serum electrolytes in PTZ treated chicks compared with controls.

Parameters	Groups					
	Negative control	Positive control	Sodium Valproate 200mg/kg	Plant extract 100mg/kg	Plant extract 200mg/kg	Plant extract 300mg/kg
Na^+ ion (mmol/L)	B 150.2	A 164.9	B 155.1	C 134.0	C 133.1	C 134.6
K^+ ion (mmol/L)	CD 5.94	CD 6.09	D 5.42	A 11.09	BC 7.93	B 8.36
Cl^- ion (mmol/L)	A 125.9	A 125.6	A 125.1	A 134.2	A 127.6	A 125.7
Ca^{+2} ion (mmol/L)	A 11.95	A 12.02	B 9.49	AB 11.48	A 12.62	AB 10.75

N=12 chicks in each group, values are expressed as mean +SE.

Different letters refer to the significant differences at level $P < 0.05$.

3.3. Effect of cloves of *Syzyguim Aromaticum* L. aqueous extract on serum glucose and protein in PTZ treated chicks compared with controls.

No difference was observed in the concentration of glucose concentration between negative and positive control groups. The concentration of glucose was significantly higher in the sodium valproate treated group in comparison with controls and the plant extract treated groups. Treatment of chicks with aqueous extract caused marked decrease in the glucose concentration compared with PTZ treated group only (table 3.4). For total protein, no significant changes have been shown between negative and positive controls. Sodium valproate and plant extracts at 200 and 300 mg /kg treated

groups exhibited significantly lower levels of total protein compared to positive control group (table 3.4).

Table (3.4): Effect of cloves of *Syzygium aromaticum* L. aqueous extract on serum glucose and total protein in PTZ treated chicks compared with controls.

Parameters	Groups					
	Negative control	Positive control	Sodium valproate 200mg/kg	Plant extract 100mg/kg	Plant extract 200mg/kg	Plant extract 300mg/kg
Glucose (mg/dl)	BC 263.0	B 307.9	A 383.2	CD 209.6	D 179.9	CD 201.0
Total protein (g/dl)	A 3.16	A 3.11	B 2.64	A 2.99	C 2.25	BC 2.61

N=12 chicks in each group, values are expressed as mean +SE.

Different letters refer to the significant differences at level P< 0.05.

DISCUSSION

Injection of PTZ at 80 mg/kg cause appearance of seizure signs in 100% of animals. Prevention the seizures induced by PTZ in laboratory animals is the most commonly test used for characterizing anti-convulsive drugs. The test is assumed to identify anticonvulsant drugs effective against seizures as PTZ produces convulsions. The antiepileptic drug should prevent or increase the threshold for convulsions (30). The mechanism by which PTZ exert its convulsive action is by interacting with GABA neurotransmitter at GABA_A-receptors (30)The chicks group which treated with sodium valproate didn't show any signs of seizure and the drug offered complete protection against PTZ .Sodium valproate is thought to produce their effects by inhibiting voltage dependant Na⁺ channels (31). The extract of cloves failed to prevent seizures signs in the dose range tested in this study, although prolonged seizure latency and decreased mortality rate were founded. Despite of presence of previous studies refer that the plants extract which contain flavonoids and alkaloids shows anticonvulsant property in chicks (32) and (33).

Epileptic patients have variably altered status of free radical scavenging system activities (antioxidant system) and electrolytes (34). The mechanisms of epilepsy are not well established. Several studies suggested that the membrane lipid peroxidation due to increase in free radicals or decrease in activities of antioxidant defense mechanisms and body electrolytes involved in some forms of epilepsies and also to increase the recurrence of seizure (35). Injection of PTZ lead to slightly decrease the level of T-AOC in the serum of chicks compared with negative control group which injected with normal saline but not significantly. This study revealed a significant increase in the antioxidant marker as T-AOC which was evident in sodium valproate treated group compared with positive control group (table 3.2) which agrees with many studies (36) and (37). In contrast, Hamed *et al.*, (2004) and Schulpis *et al.*, (2006) (34) and (38) reported decrease of T-AOC level in epileptic patients treated particularly with sodium valproate in compared with the normal control. Plonka *et al.*,(2011) (36) observed that the activity of some antioxidant enzymes are higher in patients treated with valproate in comparison to controls and concluded that valporic acid modulate and regulates activity of antioxidant enzymes. Zhang *et al.*, (2012)(39) mention that antioxidants supply is important for brain functions and prevention of neurological diseases. Our results showed elevated T-AOC in all groups treated with cloves extract in different concentration (100,200,300 mg/kg) versus positive control group and the increases have correlation with increasing the concentration of extract. Pérez-Jiménez *et al.*,2010 (40) mention that clove contain high amount of antioxidant compounds, and Shan *et al.*,(2005) (41), was highlighted the huge potential of clove as free radical scavenger. In this study the phytochemical analysis of plant extract revealed the presence of flavonoids and many references indicated antioxidant and anti-convulsant activity of flavonoids (42).

In PTZ treated group, GABA levels significantly decreased in serum compared with negative control group. Some studies showed similar observation (43) and (44). PTZ produce its effect to induce seizure by inhibiting the activity of GABA at their receptors. GABA is an important inhibitory neurotransmitter which has a bad role in epilepsy. The inhibition of GABA and there neurotransmission role attenuates and enhances convulsion. It is well known that seizures can be generated in response to imbalance between excitatory (glutamate) and inhibitory (GABA) influences which called neurotransmitters systems together with changes in intrinsic neuronal properties

can lead repetitive neuronal discharges, hyper excitability and seizure. Enhancement of excitatory transmission and simultaneous failure of inhibitory mechanisms together with changes in intrinsic neuronal properties results in repetitive neuronal discharges (45) and (46). Antiepileptic drug (sodium valproate) restored the GABA level in serum that coincides with the previous study (43). Enhancers of GABA-ergic transmission comprise a large group of new generation antiepileptic drugs (47). The discovery of valproic acid efficacy in treatment of epilepsy created a new therapeutic paradigm. This drug was thought to be effective in enhancing GABA-ergic neurotransmitter in brain, and became one of the first drugs in which a mode of action was studied (48). *Syzygium Aromaticum* L. extract at the doses of (100,200,300 mg/kg) showed a significantly increased in GABA content in the serum dose dependently. These results may be attributed to the presence of flavonoids in the extract which enhance GABA function and productions through their enzymes system modulators (49) with contribute the presence of tannins and alkaloids of aqueous extract (43). Flavonoids administration orally or i.p in rats treated with PTZ, reduced convulsive response and suggested that the flavonoids have anticonvulsant activity against PTZ (15). Since the extract delayed the occurrence and decreased the duration of convulsions induced by PTZ, it is possible that the anticonvulsant effects might be due to enhancement of GABA-mediated inhibition and/or inhibition of Ca^{+2} currents.

The amount of GABA in brain tissue were unaltered among all groups of experiment except the 300mg /kg extract treated group exhibit significantly elevated GABA amount compared with PTZ treated group. The extract may be exhibited anticonvulsant activity against seizures induced by PTZ when used in high concentrations probably by increasing the brain contents of GABA.

Chick's treatment with PTZ and induction of seizure cause significant decrease in the activity of ChE in serum compared with normal chicks. This goes with the study of Ali *et al.*, 2012 (17), and also in compatible with the results of Seedo, 2015 (43). According to the results of this study PTZ cause inhibition of ChE activity which may be accompanied by accumulation of acetylcholine (a neurotransmitter) at the nerve endings and causing hyper stimulation of nervous system producing seizure. In this study using standard antiepileptic drug (sodium valproate) cause significant increase in the activity of serum ChE compared with positive control group. Significant increases of serum ChE has been reported in epileptic patients treated with different

anticonvulsant drugs (50). Previously published results suggest that sodium valproate produce liver injury and hepatotoxicity lead to elevation of serum liver enzymes (51). Turecki *etal.*,(52) refer that increased serum ChE is considered an important marker of liver diseases associated with insulin resistance which cause elevation of glucose level in sodium valproate treated patients. Aqueous extract of cloves at 100,200,300mg/kg resulted in significant increase of serum ChE activity; this result may be one of the benefits of this plant to prevent the accumulation of high quantity of acetylcholine and producing nervous signs.

This work suggests that aqueous extract of cloves may possess antiepileptic properties that may be due to restoring the T-AOC, GABA and ChE in the serum of chicks. These results support the traditional uses of the plant in the treatment of convulsions. However more studies and experimental analysis are required for a definitive explanation and dose which must be used for treatment of seizure.

The study showed a significant elevation of serum Na^+ ion in positive control group by induction of seizure with PTZ with no changes have been observed in K^+ ion compared with negative control . Escamilla *etal.*, 2007 (53) reported increased in Na^+ ion level while K^+ ion not changed in rats when treated with PTZ. Whereas in contrast with study carried out by Seedo, 2015 (43) who recorded significant decrease in Na^+ ions and increase in K^+ in chicks treated with PTZ. There are several studies suggested that the body electrolytes have a vital role for enabling seizure conditions to produce; and routine laboratory estimation of serum Na^+ , K^+ , and Ca^{+2} are essential for the understanding and management of epilepsy (35). Chicks treated with sodium valproate and PTZ showed significant decrease in Na^+ ions in serum with no changes in the K^+ ions compared with the group treated with PTZ only (positive control) the present findings are somewhat agree with Seedo, 2015 (43) who found an increase of Na^+ ions, while no changes were observed in K^+ ions in convulsive chicks treated with sodium valproate .Sodium valproate block and inhibit voltage-gated sodium channels when activated (31). These data suggests that the drug decreases neuronal excitability through decreasing the persistent Na^+ current. The extract of plant at the three different concentrations which used in these study revealed significant decrease of Na^+ compared with positive control and drug treated groups, and caused significant increase of K^+ ion except in 200mg/ kg compared with positive control. The plant may be work like sodium valproate by inhibiting sodium channels and decrease

neuronal excitability, and acts as a potassium channel opener that produce opening of voltage activated K^+ channels in neurons.

In the present study, no changes detected in the Ca^{+2} ions in the serum of positive control group and among all aqueous extract treated groups .On other hand sodium valproate treated group revealed significant decrease in Ca^{+2} ions compared with positive control group. The present result are agree with the study of Abdul Wahid, 2010 (54) who mention there are no changes in Ca^{+2} ions in untreated epileptic patient compared with normal control, but disagree with the same study when say there are elevation in Ca^{+2} ions in epileptic patient s treated with sodium valproate in comparison with the control. Sodium valproate cause reduction in the concentration of Ca^{+2} ions in the serum by reducing calcium influx at nerve terminals thereby affect on the excretion of excitatory neurotransmitters in "hyper-excited" neurons, restoring them to normal physiological state (55). Kaur and Goel (56) refer the *Berberis Vulgeris* plant has anticonvulsant activity which was due to its calcium channel antagonist action.

PTZ injection in chicks not produced significant differences in serum glucose, Seedo, 2015 (43) also founded same results. Significant increase in serum glucose level was shown in sodium valproate treated group, the result is in agreement with the study of Ali, 2010 (57) who found an increase of serum glucose in epileptic rats treated with antiepileptic drug, and in agreement with Seedo, 2015(43) who referred in a previous study the treatment of chicks with valproate caused increase glucose in the serum. This result may be explained by serious complications occur during the uses of sodium valproate which produce pancreatitis and acute necrotic hepatitis and affect on insulin synthesis and secretion (58). Administration of chicks with aqueous extract cause significant decrease of serum glucose at all three concentrations which used compared with positive control group, also the level of glucose become less than their level in the negative control group. Numerous herbs and spices have been found to reduce blood glucose levels, among these herbs, *Syzyguim Aromaticum* L. and several studies have reported that *Syzyguim Aromaticum* L. exerts different pharmacological actions, including hypoglycemic and antioxidant (59). Jung *etal.*, 2012 (60) also mention to hypoglycemic effect of *Syzyguim Aromaticum* L. extract in mice and explain that this finding may be due to lower insulin resistance.

Sodium valproate displayed significant decrease in total protein level compared with negative and positive controls. Both concentrations 200 and 300 mg/kg of plant extract causes significant decrease in total protein concentration in the serum of PTZ treated chicks and may indicate protein catabolism, in contrast to Adam *et al.*, 2013 (61) who observed increase in total protein concentration in rats during treatment them with aqueous extract of *Syzygium Aromaticum* L. at 400 and 800 mg /kg.

The result obtained from this research confirmed the aqueous extract of (*Syzygium Aromaticum* L.) had low efficacy potential as anticonvulsant at studied concentrations in spite of containing phytochemical compounds mainly flavonoids, tannins and alkaloids which reported as active substances for anticonvulsant activity (62) that may be useful in the management of seizure. Further experiments will be required to uses of larger doses and identify the active molecules which present in the extract and their mechanism(s) of action.

نبات القرنفل وتأثيره على الدجاج المستحثة بنوبات الصرع

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الخلاصة

يعتبر نبات القرنفل من النباتات المهمة جدا حيث يستخدم للعديد من الاغراض الطبية مثل علاج الاختلاجات نتيجة لأمتلاكه فاعلية قوية كمانع للأكسدة. لقد تم إجراء هذه الدراسة لمعرفة تأثير المستخلص المائي لنبات القرنفل كمضاد للاختلاجات عند استخدامه بثلاثة تراكيز مختلفة (١٠٠, ٢٠٠, ٣٠٠ ملغم/ كغم من وزن الجسم) وكذلك دواء الصوديوم فالبرويت كمادة قياسية على مستوى إجمالي مضادات الأكسدة، تركيز كاما أمينو بيوتاريك أسيد ، فعالية أنزيم الكولين إستريز في مصل الدم ونسيج الدماغ ، الايونات (الصوديوم ، البوتاسيوم، الكلووريد والكالسيوم الكلي) وكذلك الكلوكوز والبروتين الكلي في مصل دم افراخ الدجاج بعد ان تستحث نوبات الصرع بواسطة البنبتلين تترازول ، حيث يستخدم هذا النبات شعبيا كمضاد للاختلاجات (التشنجات). استخدم ٧٢ فرخ دجاج في هذه الدراسة قسمت عشوائيا الى ٦ مجاميع كل مجموعة مكونة من ١٢ حيوانا. المجموعة الاولى مجموعة السيطرة السليمة عوملت بمحلول الملح الفسيولوجي، المجموعة الثانية أستحث فيها المرض بحقنها بالبنبتلين تترازول، المجموعة الثالثة عولجت بدواء الصوديوم فالبرويت عن طريق الفم المرض، المجموعة الرابعة والخامسة والسادسة عولجت بمستخلص نبات القرنفل وبالتراكيز السابقة الذكر عن طريق الفم. التحليل الكيميائي للمستخلص المائي للنبات بين أحتواءه على الفلافوناييد، الكاردياك كلايكوسايد،

، التانينات والألكلويدات. أظهرت نتائج هذه الدراسة بأن المستخلص المائي للنبات أدى إلى ارتفاع مستوى إجمالي مضادات الأكسدة، كما أمينو بيوتاريك أسيد، فعالية أنزيم الكولين إستريز في مصل دم الافراخ بعدما حصل انخفاض لهم نتيجة المعاملة بالبنتيلين تترازول واستحداث النوبات، كذلك أدت المعاملة بالمستخلص إلى انخفاض مستوى أيون الصوديوم (Na^+) والكلوكوز في مصل الدم. إستناداً إلى نتائج هذه الدراسة تم الوصول إلى أن المستخلص المائي للقرنفل له تأثير طفيف على نوبات الصرع المستحثة بالبنتيلين تترازول بالرغم من تأثيره الايجابي على مستوى العديد من القيم الكيموحيوية في مصل الدم وهذا قد يعود إلى قلة تركيز الجرعة المستخدمة والتي قد تكون غير كافية لأظهار نتائج واضحة في سلوك الحيوان.

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