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Anticonvulsant potentials of ethanolic extract of *Eleusine indica*

Ette Okon Ettebong[1*], Edidiong Etukakpan[2], Augustine Bassey[1]

[1]*Department of Clinical Pharmacology and Therapeutics, Faculty of Clinical Sciences, College of Health Sciences, University of Uyo, Uyo, Nigeria*

[2]*Department of Pharmacology and Toxicology, Faculty of Pharmacy, College of Health Science, University of Uyo, Uyo, Nigeria*

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ABSTRACT

Objective: To assess the anticonvulsant potentials of ethanolic extract of *Eleusine indica*.
Methods: Albino Wistar mice were separated into five groups with six animals in each group and thereafter pretreated with distilled water, various doses of the extract (200–600 mg/kg) and standard drug diazepam (0.5 mg/kg). Thirty minutes later, pentylenetetrazole (70 mg/kg), aminophylline (280 mg/kg) and isoniazid (250 mg/kg) were used to induce convulsions by intraperitoneal administration. These mice were then placed in plexiglas cages and monitored for the occurrence of seizures over a thirty-minute time period. The latency of convulsions, duration of tonic convulsions and mortality protection were recorded. Data obtained were analyzed using GraphPad InStat 3.10.
Results: The results showed that the extract exhibited a dose-dependent increase in the latency of clonic convulsions and decrease in duration of tonic convulsions as compared to the control and these effects were statistically significant ($P < 0.001$). The extract also provided protection against the mortality which was similar to that produced by the standard drug diazepam.
Conclusions: The significant increase in the latency of clonic convulsions and decrease in duration of tonic convulsions caused by the extract show anticonvulsant activity and corroborate with the claims of the traditional use of the plant as an anticonvulsant remedy.

1. Introduction

Epilepsy is a brain disorder characterized by a lasting predisposition to generate epileptic convulsions and by the neurobiologic, cognitive, psychological and social effects of this disease[1,2]. Seizures are the product of the electrical properties of the cerebral cortex. Convulsions occur when excitatory forces exceed inhibitory forces within the network of neurons in the cerebral cortex resulting in a sudden-onset net excitation[3]. The brain controls nearly every function of the body including higher cortical functions. If the affected cortical network is in the visual cortex, the clinical features are visual phenomena. Other areas of the cerebral cortex that can be affected include those associated with senses, taste and motion. When the temporal lobe is involved, deja-vu occurs. The pathophysiological basis of focal-onset seizures differs from the mechanisms underlying generalized-onset seizures. Generally, cellular excitability is increased, but the mechanisms of synchronization differ between these two types of seizure[3]. Some factors in a patient are known to increase the risk of seizure precipitation, including age, metabolic abnormalities (*e.g.* electrolyte imbalance) and head trauma[4]. Natural products and plants already used in traditional medicine can be a good place to search for safer and more effective treatment options. Numerous plants used for the treatment of epilepsy traditionally have been shown to be potent in models of epilepsy and several such plants remain to be scientifically validated. *Leonotis leonurus*, *Mimosa pudica*, *Laggera aurita* and *Synedrella nodiflora* are a few examples[5-7]. *Eleusine indica* (*E. indica*) is used locally to treat convulsion in children among the Ibibios of Nigeria. This study seeks to explore the potential of the extract of *E. indica* as an anticonvulsant remedy.

*Corresponding author: Ette Okon Ettebong, Department of Clinical Pharmacology and Therapeutics, Faculty of Clinical Sciences, College of Health Sciences, University of Uyo, Uyo, Nigeria.
Tel: +2348027900141
E-mail: ettebong@yahoo.com

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2. Materials and methods

2.1. Plant collection

The plant material was collected from Pharmacognosy Farm of the Faculty of Pharmacy, University of Uyo, identified and authenticated by Prof. Margaret Bassey, a taxonomist, and a voucher specimen (UUH1409) has been deposited in the Department of Botany and Ecological Studies.

2.2. Preparation of extract

The plant material was washed after collection, dried under shade for 2 weeks, ground into powder and cold-macerated in 70% ethanol for 72 h and thereafter filtered. The filtrate was dried in vacuo using the rotary evaporator and the dry extract was kept at the temperature of –4 ˚C in a refrigerator until needed.

2.3. Animals

Adult albino mice of both sexes were obtained from animal house of the Department of Pharmacology and Toxicology, University of Uyo. They were housed in standard cages and maintained on guinea feed, and water was given *ad libitum*. All experimental procedures involving animals were conducted in accordance to Organization for Economic Cooperation and Development guidelines and approved by Animal Ethics Committee, Faculty of Pharmacy, University of Uyo.

2.4. Anticonvulsant activities

2.4.1. Pentylenetetrazole (PTZ)-induced convulsion in mice

PTZ-induced convulsion test assessed the anticonvulsant potential of the extract. Thirty mice were divided into five groups with six animals per group. Animals in Group 1 were treated with distilled water (5 mL/kg). Groups 2–4 were administered with extracts of 200, 400 and 600 mg/kg body weight, respectively. Group 5 received 0.5 mL/kg of diazepam. PTZ (70 mg/kg body weight) was given intraperitoneally to induce convulsions. Thereafter, mice were placed in plexiglas cages and monitored for the occurrence of seizures over a 30-minute time period. The time was prior to the onset of tonic convulsions (latency). Duration and mortality protection were recorded[8-10].

2.4.2. Aminophylline-induced convulsions in mice

Thirty mice were randomized and divided into five groups with six animals per group. Animals in Group 1 were administered with distilled water (5 mL/kg). Group 2–4 were given 200, 400, and 600 mg/kg body weight of the extract, respectively. Group 5 received 0.5 mL/kg of diazepam. Convulsions were induced by the intraperitoneal administration of 280 mg/kg body weight of aminophylline. Mice were, thereafter, placed in plexiglas cages and monitored for the occurrence of seizures over a 30-minute time period. The time was prior to the onset of tonic convulsions (latency). Duration and mortality protection were recorded[8,11,12].

2.4.3. Isoniazid (INH)-induced convulsions in mice

Thirty mice were separated into five groups with six animals in each group. In Group 1, animals were treated with distilled water (5 mL/kg). Group 2–4 were given 200, 400 and 600 mg/kg body weight of the extract, respectively. Group 5 received 0.5 mL/kg of diazepam. Convulsions were induced with 250 mg/kg body weight of INH intraperitoneally. Thereafter, mice were placed in separated transparent plexiglas cages (25 cm × 15 cm × 10 cm) and monitored for the occurrence of seizures over a 30-minute time period. The time was prior to the onset of tonic convulsions (latency). Duration and mortality protection were recorded[8,11,13].

2.5. Statistical analysis

Results were expressed as multiple comparisons of mean ± SEM. Significance was determined using One-way ANOVA followed by Tukey-Kramer multiple comparison post test. A probability level of less than 5% was considered significant.

3. Results

The anticonvulsant activity of *E. indica* extract on PTZ-induced convulsion showed that the extract gave a dose-dependent increase in the latency of clonic convulsion as compared to the control. The increase was statistically significant ($P < 0.001$) and comparable to that of the standard drug diazepam (Table 1). A dose-dependent decrease in the duration of tonic convulsions compared to the control was observed and this decrease was statistically significant ($P < 0.001$). The extract in middle and high doses also prevented the mortality for 30 min comparable to the standard drug, diazepam.

Table 1
Anticonvulsant activity of *E. indica* extract on PTZ-induced convulsion.

Group ($n = 6$)	Dose	Latency of clonic convulsion (min)	Duration of tonic convulsion (min)	Mortality (%)
Control	5 mL/kg	0.48 ± 0.03	3.70 ± 0.05	100
Extract	200 mg/kg	1.09 ± 0.01*	2.39 ± 0.05*	30
	400 mg/kg	1.16 ± 0.04*	2.29 ± 0.01*	0
	600 mg/kg	1.33 ± 0.12*	1.63 ± 0.19*	0
Diazepam	0.5 mL/kg	1.74 ± 0.02*	1.42 ± 0.04*	0

Values were represented as mean ± SEM. *: Compared to the control, $P < 0.001$.

On aminophylline-induced convulsion, the extract showed a dose-

dependent increase in latency of clonic convulsion and a decrease in duration of tonic convulsion as compared to the control. These effects were statistically significant ($P < 0.001$). However, the observed effects were much lower than those of the standard drug, diazepam. All the animals treated with the middle and high doses of the extract had protection against mortality as comparable to the standard drug diazepam (Table 2).

Table 2
Anticonvulsant activity of *E. indica* extract on aminophylline-induced convulsion.

Group ($n = 6$)	Dose	Latency of clonic convulsion (min)	Duration of tonic convulsion (min)	Mortality (%)
Control	5 mL/kg	2.26 ± 0.08	17.45 ± 0.39	100
Extract	200 mg/kg	4.35 ± 0.08*	9.57 ± 0.53*	10
	400 mg/kg	4.60 ± 0.17*	9.20 ± 0.44*	0
	600 mg/kg	4.67 ± 0.24*	8.71 ± 0.48*	0
Diazepam	0.5 mL/kg	6.07 ± 0.18*	4.55 ± 0.16*	0

Values were represented as mean ± SEM. *: Compared to the control, $P < 0.001$.

In the INH-induced convulsion, a dose-dependent increase in latency of clonic convulsion and a decrease in the duration of tonic convulsion were observed in extract-treated groups. These results were statistically significant as compared to the control as depicted in Table 3 ($P < 0.001$). The extract gave 100% protection against mortality in all groups except that of the lowest dose, 200 mg/kg.

Table 3
Anticonvulsant activity of *E. indica* extract on INH-induced convulsion.

Group ($n = 6$)	Dose	Latency of clonic convulsion (min)	Duration of tonic convulsion (min)	Mortality (%)
Control	5 mL/kg	17.00 ± 0.41	29.29 ± 0.46	66
Extract	200 mg/kg	18.29 ± 0.28	28.32 ± 0.89	30
	400 mg/kg	19.26 ± 0.30	23.29 ± 0.49*	0
	600 mg/kg	22.28 ± 0.28*	23.04 ± 0.51*	0
Diazepam	0.5 mL/kg	24.54 ± 0.38*	18.10 ± 0.50*	0

Values were represented as mean ± SEM. *: Compared to the control, $P < 0.001$.

4. Discussion

Medicinal plants over the ages have served as sources of readily accessible, inexpensive and effective medication for man. Several medicinal plants are found to possess anticonvulsant potentials and could be used in modern medicine. They need to be assessed for biological activities and validated scientifically[8,14]. This study was to assess the anticonvulsant effects of the ethanolic extract of *E. indica*.

Anticonvulsant activity in an extract is depicted by its ability to delay the onset of convulsions and/or shorten the duration of convulsions[15]. The results of this study show that the extract exhibited a dose-dependent increase in the latency of clonic convulsion as well as a decrease in duration of tonic convulsions in PTZ-, aminophylline- and INH-induced convulsions in mice as compared to the control. Furthermore, like diazepam, the extract gave 100% protection against mortality at 400 mg/kg and 600 mg/kg respectively in the PTZ, aminophyline and INH models within 30 min. Most anticonvulsant agents promote the response of gamma-aminobutyric acid (GABA) by facilitating the opening of GABA-activated chloride channels[8,16]. $GABA_A$ receptors are known to be involved in epilepsy and their direct activation would result in an antiepileptic effect. It is reported that PTZ/INH/aminophylline-induced convulsions occur as a result of inhibition of GABA synthesis and level in the brain[8,17,18]. Therefore, the antiepileptic effect of *E. indica* extract may be due to an increased level of GABA, an inhibitory neurotransmitter in the central nervous system. This is similar to the pharmacological effects of benzodiazepine and it depicts the relevance of the antiepileptic effects of the extract. The anticonvulsant effects of benzodiazepines like diazepam are mostly attributed to the enhancement of the action of GABA[17]. Actually, benzodiazepines bind to the gamma subunit of the $GABA_A$ receptor, for which a structural modification of the receptor results in an increase in $GABA_A$ receptor activity. Benzodiazepines do not substitute for GABA, which bind at the alpha subunit, but increase the frequency of channel opening events, which leads to an increase in chloride ion conductance and inhibition of the action potential[8,19,20]. Therefore, the extract may be interfered with GABAergic mechanism. *E. indica* contains cardiac glycosides, terpenes, flavonoids, tannins and alkaloids[21]. Triterpenic steroids and triterpenoidal saponins are known to possess anticonvulsant properties in seizure models including PTZ[22,23]. Alkaloids, monoterpenes and flavonoids do have protective effects against PTZ-, picrotoxin- and N-methyl-D-aspartate-induced seizures[24-30].

Medicinal potentials of *E. indica* is enormous. Although many compounds have been discovered from *E. indica*, more are still needed to be elucidated.

From the result of this study, it is observed that the ethanolic extract of *E. indica* has potential therapeutic action against convulsions in mice induced experimentally by PTZ, aminophyline and INH. This effect is most pronounced in INH model and least in aminophylline model. The anticonvulsant activity of the plant may be due to the phytochemical constituents. Though the work is not exhaustive, the findings actually corroborated the earlier traditional application of the plant as a remedy for convulsion in children.

Conflict of interest statement

We declare that we have no conflict of interest.

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