An investigation of the phytochemical richness of fresh *Musa paradisiaca* L. (plantain) stem juice and its anticonvulsant potential on pentylenetetrazole (PTZ)-challenged rats

Sabastine Chinweike Ugwuoke (✉ sabastine.ugwoke.88417@unn.edu.ng)
Department of Biochemistry, University of Nigeria, Nsukka, 410001, Enugu, Nigeria
https://orcid.org/0000-0002-1202-2974

Valentine Odirachukwumma Nwanelo
Department of Biochemistry, University of Nigeria, Nsukka, 410001, Enugu, Nigeria
https://orcid.org/0000-0002-7289-5654

Yusufu Dawoye
Department of Chemistry/Biochemistry, Federal Polytechnic Bali, Taraba, Nigeria

Obiora Celestine Ugwu
Department of Pharmacology, Enugu State University of Science and Technology, P.M.B 01660, Enugu, Nigeria
https://orcid.org/0000-0002-6657-4942

Dionysius Obinna Osuji
Department of Biochemistry, University of Nigeria, Nsukka, 410001, Enugu, Nigeria

Martins Obinna Ogugofor
Department of Medical Biochemistry, Enugu State University of Science and Technology, P.M.B 01660, Enugu, Nigeria
https://orcid.org/0000-0002-9491-7498

Ikechukwu Jacob Okoro
Department of Medical Biochemistry, David Umahi Federal University of Health Sciences, Uburu, Abonyi, Nigeria

Chigozie Paul Odo
Department of Biochemistry, University of Nigeria, Nsukka, 410001, Enugu, Nigeria

Treasure Nneka Nelson
Department of Biochemistry, Michael Okpara University Umudike, 440101, Abia, Nigeria

Chioma Assumpta Anosike
Department of Biochemistry, University of Nigeria, Nsukka, 410001, Enugu, Nigeria
https://orcid.org/0000-0003-4141-3656

Research Article
Abstract

This study was aimed at determining the therapeutic value of fresh *Musa paradisiaca* L. (MP) stem juice as a potential treatment for epileptic convulsions using a pentylenetetrazole (PTZ)-induced seizure model in rats. Six groups of albino rats (n = 4) were involved in the study. Group I was treated with normal saline (p.o), while group II was untreated and group III received diazepam (4 mg/kg, p.o). Group IV, V and VI received 50, 75 and 100% v/v oral dose of MP stem juice, respectively. The treatment lasted for 10 days, followed by PTZ (85 mg/kg b.w, i.p) administration 60 min later. Lethality test and phytochemical screening were conducted. The rats were closely watched and meticulously monitored for seizure manifestations/episodes with the aid of a stop watch. From the results, the MP stem juice up to 100% (v/v) was safe in mice and numerous bioactive compounds were found with phenols being the most abundant (9.46 ± 0.03 mg/g), followed by alkaloids (5.54 ± 0.98 mg/g) and flavonoids (4.27 ± 1.23 mg/g). For the seizure manifestation, three intermittent seizures (episodes 1, 2 and 3) were observed and the stem juice (75 and 100% v/v) significantly (p < 0.05) increased the latency periods of episode 1 tonic and clonic seizures. The stem juice at 50% (v/v) delayed the onset of episode 2 seizures for over 10 minutes more than the untreated group. The groups that received 75 and 100% (v/v) of the stem juice did not experience seizures during the episode 2 as seen in episode 1. The standard and the test groups did not experience seizure during the episode 3.

The findings of this study have demonstrated that the fresh MP stem juice could prevent convulsions by increasing the latencies and decreasing relatively the durations of seizures in PTZ-challenged rats. This study, however, provides the pharmacological evidence for the folk claim behind the use of *Musa paradisiaca* stem juice to manage epileptic convulsions or seizure disorders.

Background

Epilepsy poses a great deal of disease burdens on humans and brings about psychosocial stigma which ruins their inter-personal relationship. It is specifically a chronic neurological disorder which produces recurrent seizures spontaneously, leading to a brief lapse of attention, muscle jerks or convulsions (Fischer et al., 2014). Seizures occur when nerve cells (i.e. neurons) are no longer in control of the electrical signal they fire, which in turn interferes with the passage of signal from one neuron to another (Beck and Elger, 2008). Convulsion is a form of seizure that involves body movement due to increased muscle tone. Seizure leads to changes in behavior, body movements, feelings and consciousness. Two or more seizures that occur at least 24 hours apart are considered to be epilepsy (WHO, 2023). Different types of seizures exist, and with a wide range of symptoms and severity. Most seizures last from 30 seconds to 2 minutes but the one that lasts beyond five minutes is a serious medical issue (Neubauer, 2008). Each year, over 50 million people are affected by seizure disorder globally, out of which over 80% live in Sub-Saharan Africa and Asia where the standard of living is very low (Adewumi et al., 2020). This is presumable due to advanced incidence of antecedent factors such as brain infections, cranial and perinatal traumas as well as infections in these low- and middle-income countries (Ba-Diop et al., 2014). Majority of these risk factors can be reduced or prevented with orthodox medications, but the associated
debilitating side effects are far reaching. According to the World Health Organization (WHO) fact sheet on epilepsy, the premature death risk in people living with epilepsy (PLWE) is thrice that of the general population, whereas over 70% of the PLWE could survive if properly diagnosed and treated [3]. Despite the advances made towards the prevention/treatment of neurological disorders, epilepsy has remained defying to cure. Several licensed antiepileptic drugs (AEDs) have been in use, but have neither cured nor have the disease relapsed completely, and are characterized with cognitive dysfunction and intolerant issues (Foster et al., 2020; Mutanana et al., 2020). Studies have shown that, despite the use of these drugs, up to 30% of the patients continue to suffer seizure crisis (He et al., 2021; Chen et al., 2018). However, the development of more bio-friendly pharmacological preparation that can overcome these limitations associated with existing AEDs becomes an important goal for this study. Natural products obtained mainly from plants are of greatest contribution and their advent showed a remarkable development in the medicinal treatment of various diseases (Newman and Cragg, 2020). These traditional systems of medicine are widely used in developing countries where over 80% of the population solely depends on traditional medicine or folk remedies for primary healthcare need (Tugume and Nyakoojo, 2019). In recent years, several plants have demonstrated high level of potency against seizures/convulsions; and results from the phytochemical and pharmacological studies reviewed that it is due to the presence of bioactive compounds such as flavonoids and other phenolics that have similar mechanism of action as their synthetic counterparts (Edo et al., 2023).

Before now, our forefathers used to manage epileptic convulsions by means of ordinary roots and herbs without having adequate knowledge of the therapeutic implications. For instance, certain herbalists from various parts of Nigeria reportedly claim that Plantain (Musa paradisiaca) stalk juice cures seizure disorders and convulsion. Additionally, the Ezike-Oba people in Igbo-Eze North LGA of Enugu State Nigeria cures febrile (infantile) convulsions by giving a convulsing child, the mother’s urine to drink; other approaches are also exploited such as the use of local palm kernel oil, ripe tomato grown organically and so on (Ugwuoke et al., 2023). This plant shares several physical features with Banana (Musa sapientum) (MS), because they belong to the same Musaceae family, but the MP tree is larger and taller than MS tree, which has thicker trunks and leaves (Reddy et al., 2018). Although, both plants have been used interchangeably for managing seizures/febrile convulsions in different southeastern Nigerian communities, but MP seems to be more preferably used by the majority of the people in Nsukka, Agulu and Obowo in Enugu, Anambra and Imo States, respectively (Ugwuoke et al., 2023). Various parts of MP plant such as roots, leaves and flowers have been used traditionally- the sap is reportedly used as a remedy for dysentery, diarrhea, hysteria and epilepsy; the fruit is eaten as food; the leaf juice for treating wounds, cuts and insect bites; while the leaves have the ability to induce abortion; and a cold infusion of the root is used to cure venereal diseases and anaemia (Ajijolakewu et al., 2021). Additionally, the fruit has reportedly been used as aphrodisiac, antiscorbutic and diuretic agents (Onyenekwe et al., 2013). Medicinal plants’ pharmacological potentials are mainly brought about by the bioactive ingredients or compound in them such as flavonoids, terpenes, saponins, tannins, proteins, alkaloids and phenolic components (Tungmunnithum et al., 2018; Rao et al., 2016).
Previous studies on MP plant have reviewed the presence of phytochemicals such as phenolic compounds, flavonoids; and others including alkaloids, tannins and saponins (Onyenekwe et al., 2013; Gervásio and Batitucci 2023). Furthermore, a recent study has also proven that MP stem juice has the therapeutic ability to prevent seizures by positively influencing the associated biochemical parameters such as GABA, glutamate, GABA-T and brain histology of the experimental rats (Ugwuoke et al., 2023). In this study, however, we aim to explore how the *Musa paradisiaca* stem juice influences the physical manifestation of seizures and to further establish the scientific basis for the folkloric claim.

**Material and methods**

**Material**

**Plant material**

MP stem samples used for this study were collected fresh on February 13, 2022 from the plantain plantation located at Obeke village, Uwani Akpotoro Obimo, Nsukka LGA, Enugu State, Nigeria. The specimen was identified by Mr. Alfred Ozioko, a taxonomist at the international Centre for Ethno-medicine and Drug Development (InterCEDD) Nsukka, Enugu State Nigeria. The voucher specimen was deposited in the herbarium unit of the Botany Department, University of Nigeria, Nsukka for future references.

**Experimental animals**

A total of forty-two (42) animals were used for the experiment, eighteen (18) adult albino mice (18–26 g) and twenty-four (24) adult male Albino rats (120–220 g). They were bred and obtained from the Laboratory unit of the Department of Pharmacology and Toxicology, University of Nigeria, Nsukka. On transfer to the work area, animals were acclimatized for seven days under standard laboratory conditions with free access to standard pellets (Guinea Feeds Plc, Nigeria) and water before the commencement of the experiment. All animal experiments were conducted in compliance with the National Research Council’s Guide for the Care and Use of Laboratory Animals and approved by the Ethical Committee on the use of laboratory animals, Faculty of Biological Sciences, University of Nigeria, Nsukka (Reference No: UNN/FBS/EC/1090).

**Equipment and chemicals**

The equipment and chemicals used for this study were of analytical grade. Some of them were obtained from the Biochemistry Department, University of Nigeria, Nsukka, while others were purchased from the Springboard Research Laboratory, Awka, Anambra State, Nigeria. They include, but not limited to the following:

**Equipment**

Mortar and pestle (Sevico Plast, Nigeria), Volumetric flask, Beaker, test tubes and measuring cylinder (Pyrex, England), Weighing balance (Mettler Toledo PB 602, Switzerland), Whatman filter paper,
separating funnel, water bath, conical flask.

Chemicals/Kits

Pentylenetetrazole (Sigma Aldrich Chem. Co., USA); Diazepam (Hoffman-la Roche, Switzerland); Chloroform, Sodium Hydroxide, Ferric Chloride, 90% Ethanol, Potassium Hydroxide, Deionized/distilled water (STC, Nsukka), Dragendorff’s reagent, Mayer’s reagent, Wagner’s reagent, 10% acetic acid, Fehling solution, diethylether and dil. HCl. Other reagents used for the assays were commercial test Kits and products of Randox, UK, Biovendor, Czech Republic, TECO Diagnostics, USA and Centronic GmbH, Germany.

Methods

Preparation of MP stem juice

A fresh sample of *Musa paradisiaca* (MP) stem was cut and washed free of dirt. It was sliced into smaller fragments and the juice was then extracted by mechanical crushing using a plastic mortar and pestle, followed by filtration. Following the method of dosing by Onyenekwe et al. (2013) in which the dose of 100% (v/v) was prepared from 100 mL of the stem juice in a volumetric flask; 75% (v/v) was prepared from 75 mL of the stem juice made up to 100 mL with distilled water in a volumetric flask; 50% (v/v) was prepared from 50 mL of the stem juice made up to 100 mL with distilled water in a volumetric flask; while 25% (v/v) was made from 25 mL of the stem juice made up to 100 mL with distilled water in a volumetric flask.

Phytochemical analysis of MP stem juice

The qualitative and quantitative screening of the phytochemical constituents of the stem juice was carried out using the methods of Harborne (2013) and Trease and Evans (2002).

Acute toxicity and lethality (LD$_{50}$) study

The oral median lethal doses (LD$_{50}$) of the stem juice were determined in the albino mice using the Organization for Economic Co-operation and Development (OECD) 425 guidelines as reported by Onyenekwe et al. (2013) and Swopna and Karishma (2018).

Experimental design

Pentylenetetrazole (PTZ)-induced seizure model was adopted for this study according to Gupta et al. (1999), involving albino rats (n = 24) which were randomly distributed into six (6) groups of four rats each (n = 4). The animals were administered with freshly prepared MP stem juice by oral intubation. The group I that received saline (p.o) served as the normal control, whereas group II was untreated. The group III (i.e., Standard control) received diazepam (4 mg/kg b.w, p.o); Groups IV, V and VI received 50, 75 and 100%
(v/v) oral doses of MP stem juice, respectively. The treatment lasted for 10 days prior to seizure induction with PTZ.

**Induction procedure**

Seizure induction was done on the last day (i.e. 10th day) of treatment. All the experimental animals, with the exception of the normal control group, were injected with PTZ (85mg/kg b.w, i.p) 45 min after treatment with the stem juice (Gupta et al., 1999). The rats were closely observed and meticulously monitored for seizure manifestations/episodes with the aid of a stop watch.

The experimental protocol was summarized as follows;

- **Group I:** Normal control (Saline only)
- **Group II:** Untreated control (Saline + PTZ administration)
- **Group III:** Standard control (4 mg/kg Diazepam + PTZ administration)
- **Group IV:** 50% (v/v) MP Stem juice + PTZ administration
- **Group V:** 75% (v/v) MP Stem juice + PTZ administration
- **Group VI:** 100% (v/v) MP Stem juice + PTZ administration

**Evaluation of seizure activity (Seizure manifestation)**

Intermittent seizure episodes 1, 2 and 3 were meticulously monitored; tonic and clonic seizure latencies as well as durations were accurately noted by means of a stop watch according to the method of Gupta et al. (1999). Tonic seizure was noted based on the stiffness of the muscles, while clonic seizure was noted based on the twitching or jerking movement of the rats’ muscles.

**Statistical analysis**

Data were analyzed using one-way analysis of variance (ANOVA) with repeated measures using Statistical Product and Service Solution (SPSS) version 21. Results were expressed as mean ± SD and a p-value < 0.05 was considered significant.

**Results**

**Phytochemical screening of MP stem juice**

Table 1 shows that MP stem juice contains alkaloids, flavonoids, phenols, saponins, tannins and steroids, but no terpenoids were detected. However, for their relative abundances, while phenols were found in relatively high abundance (9.46 ± 0.03 mg/g), alkaloids, flavonoids, and tannins were found in moderate concentrations, 5.54 ± 0.98, 4.27 ± 1.23 and 3.64 ± 0.02 mg/g, respectively. Others, including saponins and steroids were found in low concentrations, 1.27 ± 0.01 and 0.84 ± 0.03 mg/g, respectively.
Table 1

<table>
<thead>
<tr>
<th>S/N</th>
<th>Phytoconstituents</th>
<th>Relative Abundance</th>
<th>Amount (mg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>++</td>
<td>5.54 ± 0.98</td>
</tr>
<tr>
<td>2</td>
<td>Flavonoids</td>
<td>++</td>
<td>4.27 ± 1.23</td>
</tr>
<tr>
<td>3</td>
<td>Phenol</td>
<td>+++</td>
<td>9.46 ± 0.03</td>
</tr>
<tr>
<td>4</td>
<td>Saponins</td>
<td>+</td>
<td>1.27 ± 0.01</td>
</tr>
<tr>
<td>5</td>
<td>Tannins</td>
<td>++</td>
<td>3.64 ± 0.02</td>
</tr>
<tr>
<td>6</td>
<td>Steroids</td>
<td>+</td>
<td>0.84 ± 0.03</td>
</tr>
<tr>
<td>7</td>
<td>Terpenoids</td>
<td>ND</td>
<td>-</td>
</tr>
</tbody>
</table>

Values are presented as mean ± SD.

**Key:** Low Concentration (+); Moderate Concentration (++); High Concentration (+++); Not detected (ND)

**Determination of acute toxicity**

Table 2 shows that no death was recorded when MP stem juice was administered to the mice for all the doses (5, 10, 25, 50, 75 and 100% v/v).

Table 2

<table>
<thead>
<tr>
<th>S/N</th>
<th>Groups</th>
<th>MP Stem Juice Dosage (% v/v)</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Group I</td>
<td>5</td>
<td>000</td>
</tr>
<tr>
<td>2</td>
<td>Group II</td>
<td>10</td>
<td>000</td>
</tr>
<tr>
<td>3</td>
<td>Group III</td>
<td>25</td>
<td>000</td>
</tr>
<tr>
<td>4</td>
<td>Group IV</td>
<td>50</td>
<td>000</td>
</tr>
<tr>
<td>5</td>
<td>Group V</td>
<td>75</td>
<td>000</td>
</tr>
<tr>
<td>6</td>
<td>Group VI</td>
<td>100</td>
<td>000</td>
</tr>
</tbody>
</table>

N = 3 for each group; 000 = No death; XXX = death

**Anticonvulsant effect of MP stem juice on PTZ-induced seizure in rats (Episode 1)**

The three graded doses of MP stem juice (50, 75 and 100% v/v) caused an increase in the latency period of PTZ-induced tonic and clonic seizures; and a reduction in the duration of the clonic seizure episode (Table 3).
Medium and high doses, 75 and 100% (v/v) of MP stem juice produced a significant (P < 0.05) increase in the latency periods of tonic seizures in groups V and VI (21.93 ± 8.60 min and 15.22 ± 2.61 min, respectively) when compared to that of group II (4.96 ± 1.19 min). There was non-significant (p > 0.05) increase in the tonic seizure latency in group IV (10.44 ± 1.23 min) compared to that of group III (7.46 ± 5.86 min) but it increased significantly (p < 0.05) with groups V and VI (23.62 ± 9.64 min and 16.07 ± 2.67 min, respectively).

In group IV, the latency period of clonic seizures (11.26 ± 1.38 min) was non-significantly (p > 0.05) higher compared to that of group II (5.44 ± 1.28 min), but was significantly (P < 0.05) higher (23.62 ± 9.64 and 16.07 ± 2.67 min, respectively) with groups V and VI that received 75 and 100% v/v stem juice.

Clonic seizure duration was non-significantly (p > 0.05) reduced in groups IV, V and VI as well as in group III when compared to that of the group II. All experimental rats survived the seizure crises and were also active at the end of episode 1 seizures.

Table 3
Anticonvulsant effect of stem juice of M. paradisiaca on PTZ-induced seizures in adult male albino rats (Episode 1)

<table>
<thead>
<tr>
<th>Groups</th>
<th>Tonic seizure latency (min)</th>
<th>Clonic seizure onset (min)</th>
<th>Clonic seizure duration (min)</th>
<th>Status of animals after episode 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>II</td>
<td>4.96 ± 1.19a</td>
<td>5.44 ± 1.28a</td>
<td>1.31 ± 0.77a</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>III</td>
<td>7.46 ± 5.86a</td>
<td>8.11 ± 6.71a,b</td>
<td>0.65 ± 0.54a</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>IV</td>
<td>10.44 ± 1.23a,b</td>
<td>11.26 ± 1.38a,b</td>
<td>0.64 ± 0.36a</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>V</td>
<td>21.93 ± 8.60c</td>
<td>23.62 ± 9.64c</td>
<td>0.89 ± 0.58a</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>VI</td>
<td>15.22 ± 2.61b,c</td>
<td>16.07 ± 2.67b,c</td>
<td>0.64 ± 0.52a</td>
<td>Alive/Active</td>
</tr>
</tbody>
</table>

Data is expressed as mean ± standard deviation (n = 4). Means with different alphabet as superscript down the column are significantly (p < 0.05) different.

Group I: Normal Control (Saline only)

Group II: Untreated Control (saline + PTZ)

Group III: Standard Control (4 mg/kg b.w diazepam) + PTZ

Group IV: 50% (v/v) MP Stem Juice + PTZ

Group V: 75% (v/v) MP Stem Juice + PTZ

Group VI: 100% (v/v) MP Stem Juice + PTZ
Anticonvulsant effect of MP stem juice on PTZ-induced seizure in rats (Episode 2)

Table 4 shows the second seizure episode in which there were variations in clonic seizure latency, duration, as well as status of the animals at the end of the episode.

The lowest dose of MP stem juice (50% v/v) significantly (p < 0.05) increased the latency period (25.82 ± 1.04 min) of clonic seizures in group IV compared to that of group II (12.90 ± 0.92 min). Similarly, 50% (v/v) of the stem juice caused a significant (p < 0.05) increase in the latency period of group IV (25.82 ± 1.04 min) compared to that of group III (12.72 ± 8.75 min).

Moreover, the duration of the clonic seizure was found non-significantly lower in group IV (0.23 ± 0.18 min) and group III (0.36 ± 0.18 min) compared to that of group II (0.67 ± 0.40 min). In addition, groups V and VI that received 75% (v/v) and 100% (v/v), respectively of the stem juice did not experience or show any form of seizures.

All the experimental rats in each group were alive and also active, except those in group II that were found with some level of weakness.

Table 4

<table>
<thead>
<tr>
<th>Groups</th>
<th>Latency of clonic seizures (min)</th>
<th>Duration of clonic seizures (min)</th>
<th>Status of animals</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>-</td>
<td>-</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>II</td>
<td>12.90 ± 0.92^b</td>
<td>0.67 ± 0.40^b</td>
<td>Alive/Weak</td>
</tr>
<tr>
<td>III</td>
<td>12.72 ± 8.75^b</td>
<td>0.36 ± 0.18^a,b</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>IV</td>
<td>25.82 ± 1.04^a</td>
<td>0.23 ± 0.18^a,b</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>V</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>VI</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
</tbody>
</table>

Data is expressed as mean ± standard deviation (n = 4). Means with different alphabet as superscript down the column are significantly (p < 0.05) different.

NC: No convulsion

Group I: Normal Control (Saline only)

Group II: Untreated Control (saline + PTZ)

Group III: Standard Control (4 mg/kg b.w diazepam) + PTZ

Group IV: 50% (v/v) MP Stem Juice + PTZ
Group V: 75% (v/v) MP Stem Juice + PTZ

Group VI: 100% (v/v) MP Stem Juice + PTZ

**Anticonvulsant effect of MP stem juice on PTZ-induced seizure in rats (Episode 3)**

In Table 5, the third seizure episode is depicted. Only group II rats experienced clonic seizures in this episode. Other groups such as the standard (III) and the test (IV, V and VI) groups did not encounter any form of epileptic seizures.

The experimental rats in each group were alive and active, except those of group II that survived, but were very weak at the end of this seizure episode.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Onset of clonic seizure (min)</th>
<th>Duration of clonic seizure (min)</th>
<th>Status of animals</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>-</td>
<td>-</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>II</td>
<td>19.41 ± 1.21</td>
<td>1.32 ± 0.77</td>
<td>Alive/very weak</td>
</tr>
<tr>
<td>III</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>IV</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>V</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
<tr>
<td>VI</td>
<td>NC</td>
<td>NC</td>
<td>Alive/Active</td>
</tr>
</tbody>
</table>

Data is expressed as mean ± standard deviation (n = 4). Means with different alphabet as superscript down the column are significantly (p < 0.05) different.

NC: No convulsion

Group I: Normal Control (Saline only)

Group II: Untreated Control (saline + PTZ)

Group III: Standard Control (4 mg/kg b.w diazepam) + PTZ

Group IV: 50% (v/v) MP Stem Juice + PTZ

Group V: 75% (v/v) MP Stem Juice + PTZ

Group VI: 100% (v/v) MP Stem Juice + PTZ

**Discussion**
In the present study, the anticonvulsant potentials of freshly prepared MP stem juice were evaluated in adult male albino rats and the stem juice at various doses reduced PTZ-induced seizures as evident in the intermittent seizure manifestations (episodes 1, 2 and 3). In the preliminary studies involving the phytochemical screening, the phenolics content which is relatively more abundant, indicates that MP stem juice is a potent anticonvulsant. Moreover, flavonoids which are the second most abundant in the stem juice have proven to be highly phenolic in nature, and have demonstrated strong anticonvulsant properties in other medicinal plants (Tungumnithum et al., 2018; Celestine et al., 2022; Szwajgier et al., 2017). Other bioactive compound found in MP stem juice at moderate and trace amount such as alkaloids, saponins, tannins and steroids have also been reported in other studies for their anticonvulsant activities (Tungumnithum et al., 2018; Fisseha et al., 2021; Ugwah-Oguejiofor et al., 2023). A similar observation was made for n-butanol (BF) and n-hexane residual aqueous fractions (RAF) of Ipomoea asarifolia leaf which demonstrated strong anticonvulsant effects against PTZ-induced seizures in experimental mice due to its richness in saponins and flavonoids (Chiroma et al., 2022). Flavonoids, an important class of naturally occurring compounds was reported to have a modulatory role in the treatment of neurodegenerative disorders owing to their phenolic nature, since they can disrupt cellular oxidative processes in the nervous system (Panche et al., 2016; Ullah et al., 2020). Some researchers also had it that flavonoids have demonstrated central nervous system (CNS) activities, and intrinsically have strong affinity for GABA_A receptors and anticonvulsive effects (Uddin et al., 2020). However, these bioactive phytochemicals in the MP stem juice, most importantly, flavonoids and phenolics, might have potentially contributed to its anticonvulsant properties.

The acute toxicity test confirmed the safety of MP stem juice as no mortality was recorded up to the highest oral dose of 100% (v/v) of the fresh stem juice in albino mice. This suggests that the MP stem juice is not lethal in mice. In addition, no observable changes were noticed in the behavioural pattern such as posture, mood or motor activities of the mice. However, there were no convulsion crises showing that it is safe to consume, and does not induce convulsion. This study is in consonant with the report of Onyenekwe et al. (2013) who studied the phytochemicals and effect of Musa paradisiaca stem extrude on rat haematological parameters in which all four (4) graded doses (25, 50, 75 and 100% v/v) were found safe amongst the tested animals. Presently, no literature has reported or documented any form of lethality from any part of the Musa paradisiaca plant.

The results of episode 1 show that all the experimental animals experienced tonic and clonic convulsions, characterized by muscles stiffness and twitching (or jerking) movements, respectively. Medium and high doses of the fresh MP stem juice (75 and 100% v/v) remarkably increased the latency periods of tonic/clonic seizures, thereby agreeing with the findings of Mehrzadi et al. (2015), who reported a dose-dependent increase in the latency period of strychnine (STR) - and PTZ-induced seizures in mice following the administration of ethanolic extract of Punica granatum L. seed. The observed increase in the tonic seizure latency also corresponds with the results of the study reporting the prolongation of the latency period to seizure onset in rats administered with 500 mg/kg b.w of both Cichorium intybus and Taaraxacum serotinum (Rehab et al., 2015). The study also agreed with the findings of Uppala et al.
(2012), which evaluated the antiepileptic activity of methanolic extract of *Brassica nigra* seeds in mice. Wang et al. (2021) demonstrated in their study a notable ameliorative effects on PTZ-induced tonic and clonic convulsions by *Amomum tsaoiko* fruit extract. However, this observed increase in the tonic seizure latency period could probably be an indication of a positive interaction of the bioactive compounds in MP stem juice, especially phenolics and flavonoids, with the GABAergic neurotransmission. The marked increase in the tonic seizure latencies detected in groups V and VI as against group III implies that the stem juice exerted more calming or sedative effect on the CNS than diazepam (standard drug) (Al-Snaﬁ et al., 2019). The duration of clonic seizures was shortened following the administration of the fresh MP stem juice, but not to a reasonable extent. The result of this study also showed that MP stem juice at the doses of 75 and 100% (v/v) increased to a greater extent, the latency period of clonic convulsions, implying that the stem juice might possesses neuroprotective effects against glutamate-mediated excitotoxicity as reported for the ethanolic extract of *Bacopa monniera* using different convulsive models (Kaushmik et al., 2011). These results provide additional convincing evidence that MP stem juice embodies great anticonvulsant potentialities.

The episode 2 results showed that only the untreated and standard groups (group II and III), as well as the group that received a low dose of the MP stem juice (group IV) experienced seizures, indicating that the stem juice at high dose possesses more anti-seizure or anticonvulsant activities. The result of this finding corresponds with the work of Al-Snaﬁ (2015) which reported a delay of clonic convulsion onset in experimental rodents at low doses of chloroform and aqueous extracts of *Calotropis procera*. It also agreed with the increased latency of PTZ-induced clonic convulsions in albino mice observed after oral doses of *Otostegia limbata* L extracts (Amin et al., 2022). The observations of this result suggest that the fresh MP stem juice has a particular active compound (s) that confers on it, the ability to delay the onset of clonic seizure. In this episode, at the doses of 75 and 100% (v/v), no clonic seizure was noticed, which suggests that at higher dose of the MP stem juice, convulsion was prevented and abolished completely. In this episode, all the convulsing rats in each group were alive and active, except the untreated group which showed some levels of weakness indicating the magnitude of neurotoxicity possibly caused by the PTZ.

The result of episode 3 demonstrated that only the untreated group (group II) experienced clonic seizures. The standard (group III) and the test groups (groups IV, V and VI) neither encountered nor showed any form of seizure; and the experimental rats in the untreated group were alive, but extremely weak after the episode. Based on this, it could be inferred that the fresh MP stem juice had potent anticonvulsant effects just as diazepam. And the extreme weakness observed in the convulsing rats in the untreated group shows the extent or level of damages done to the rat’s neuronal cells by the PTZ.

**Conclusion**

The findings of this study have demonstrated that the fresh MP stem juice could prevent convulsions by increasing the latencies and decreasing the durations of seizures in PTZ-challenged rats. However, this
study provides pharmacological evidence for the folk claim behind the use of *Musa paradisiaca* stem juice to manage epileptic convulsions or seizure disorders.

**Abbreviations**

MP *Musa paradisiaca*

MS *Musa sapientum*

PTZ Pentylenetetrazole

PLWE People living with epilepsy

AEDs Antiepileptic drugs

CNS Central nervous system

NC No convulsion

**Declarations**

**Acknowledgements**

The Authors acknowledge Prof. B. C Nwanguma, the Departmental head as well as Asso. Prof. P. E Joshua, the coordinator of postgraduate programme, Department of Biochemistry, University of Nigeria Nsukka, for providing the technical support and the facility used to complete this work.

**Author contributions**

SCU and CAA conceptualized the research; SCU, VON, IJO and CAA designed the study and performed the data curation; SCU, IJO, and MOO carried out the experiment; SCU and VON performed the statistical analysis and writing of the original draft; SCU, CAA, VON, OCU and CPO reviewed and edited the work; TNN, DOO and YD were involved in the proofreading and all authors were involved in the proofreading and approval of the final manuscript draft.

**Funding**

No funding was received

**Availability of data and materials**

The datasets of this study is readily available upon request through the corresponding author, S.C. Ugwuoke

**Ethics approval and consent to participate**
The experimental animals used in this study were handled in compliance with the National Research Council's Guide for the Care and Use of Laboratory Animals and approved by the Ethical Committee on the use of laboratory animals, Faculty of Biological Sciences, University of Nigeria, Nsukka (Approval ID: UNN/FBS/EC/1090). All necessary permissions to embark on the study plant were duly obtained from the appropriate authority.

Consent for publication

The authors declare no conflict of interest.

Competing interests

The authors declare no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References


