Anti-convulsant activity of ethanol extract of *Enantia chlorantha* stem bark on rodents

Augustine Bassey, Paul Nwafor and Jude Okokon

**Abstract**

*Enantia chlorantha* is a medicinal plant that is used all over Nigeria. The Ibibios of South–South Nigeria use the stem bark of this plant for the treatment of febrile illnesses, stomach upset, toothache and convulsions. This study evaluated the anti-convulsant activities of the extract in rodents. Other pharmacological investigations such as anti-inflammatory, analgesic, anti-pyretic, anti-ulcer and studies were also carried out. Anti-convulsant effect of the extract was assessed using a modified method of Vellucci and Webster (1984) on overnight fasted adult albino mice (25–30g) of both sexes. The extract significantly (p<0.01–0.001) delayed the onset of clonic and tonic convulsions in mice induced by PTZ and exerted significant (p<0.01) delay in the onset of clonic and tonic convulsions caused by picrotoxin when compared to control. The above results provide support for the traditional use of *Enantia chlorantha* stem bark in treating convulsions in South-South Nigeria. Further characterization of the extract is recommended.

**Keywords:** Enantia chlorantha, stem bark, ethanol extract, tonic clonic convulsions

1. **Introduction**

*Enantia chlorantha* is a fair sized ornamental forest tree that can grow to heights of 30m. It grows in dense shade and may be recognized by its bright yellow slash and conspicuous black fruits [1]. It is located in the West African region and extends from southern Nigeria to Gabon, Zaire and Angola. It is commonly called African white wood, Moambe Jaune and Annikia chlorantha. Locally, the name varies from place to place [2]. The Ibibios of Akwa Ibom call it Uno eto; the Yoruba’s call it Osupupa or dokita Igbo. The Edo people refer to it as Erenbav bogo while Ikale and Boki tribes refer to it as Osumolu and Kakerim respectively. The family is Annonaceae and the specie is chlorantha. There are over 40 different species that grow in Nigeria as is seen by the various names that it is called and the varied uses to which it is applied. *Enantia chlorantha* is used to treat a wide variety of conditions using its roots, stem bark, fruit and leaves. In Akwa Ibom it is used to treat malaria fever, typhoid fever, jaundice, dysentery, wounds, infections, high blood pressure and convulsions. It has been used also as anti-viral, anti-candidal and for gastroenteritis [3, 4]. A decoction of the stem bark in illicit gin is usually taken to relieve painful and swollen joints, fever, headache and toothache. We aim by this work to use various models to evaluate its anticonvulsive properties and possible mechanism of action.

2. **Materials and Methods**

2.1 **Plant material**

The plant *Enantia chlorantha* was collected in January 2012 in Uyo the capital city of Akwa Ibom State, Nigeria. It was identified and authenticated by a Taxonomist in the Department of Botany and Ecological Studies, University of Uyo, Akwa Ibom State Nigeria. A voucher specimen (voucher number UUH 018/13) was deposited with the herbarium of the Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo.

2.2 **Extraction**

The plant stem bark was washed and partially air dried for about 2 weeks at ambient temperature (25 °C ± 1 °C). It was then ground to powder using pestle and mortar. The pulverized sample was divided into two parts. One part was cold macerated in 70 % ethanol at room temperature for 72 hr and then filtered. The filtrate was dried in a rotary evaporator at 40 °C. This extract is referred to as crude. The other part was successively and gradiently macerated for 72 hr at room temperature in the following solvents: n-hexane, chloroform, ethyl acetate, methanol and water to obtain different fractions. The crude extract and the fractions were stored in the freezer at -4 °C until required.
2.3 Acute toxicity study of the extract

The method of Lorke (1983) was used to determine the LD$_{50}$ of the extract in Swiss albino mice. The extract was administered to three groups of mice containing three (3) mice each at a dose range of 100-1000 mg/kg, (i.p). The animals were observed for physical signs of toxicity and the number deaths in each group within 24 hr were recorded. The animals were fasted for 24 hr prior to the experiment but allowed water ad libitum. The LD$_{50}$ was calculated as the geometric mean of the maximum dose producing 0 % mortality (A) and the minimum dose producing 100 % mortality (B). $LD_{50} = \sqrt{AB}$

2.4 Phytochemical screening

The phytochemical screening of the extract was carried out according to the methods of [5-7]. The following bioactive compounds were screened for their presence: saponins, tannins, and alkaloids. Others were flavonoids, antraquinones, cardiac glycosides and reducing sugars.

2.5 Animal stock

Swiss albino mice weighing 25-30 g of both sexes were used for the experiments. They were obtained from the Department of Pharmacology Animal House in the University of Uyo, Uyo. The animals were housed in standard plastic cages at room temperature and moisture under naturally illuminated environment of 12:12 h dark/light cycle. They were fed with pelleted feeds (Bendel Feeds) and allowed water ad libitum.

2.6 Evaluations of anti-convulsive properties

2.7 Anti-convulsant activity of extracton Pentylenetetrazol and Picrotoxin-induced convulsions in mice Anti-convulsant effect of the extract was assessed using a modified method of Vellucci and Webster (1984) on overnight fasted mice. Adult albino mice (25 – 30g) of both sexes fasted for 24 hr were randomly divided into seven(7) groups of six (6) animals each. Group I served as control and received normal saline (10mg/kg), Groups 2–4 received extract (32.40, 64.80 and 96.20 mg/kg), respectively. Group 5 was treated with phenytoin (40mg/kg). Groups 2–4 received extract (32.40, 64.80 and 96.20 mg/kg) respectively. Group 7 received phenytoin (40 mg/kg) and the extract (64.80 mg/kg) which was administered one hour before induction of convulsion. Seizure was induced in each set of mice with PTZ (70 mg/kg i.p) and picrotoxin (4 mg/kg). The onset of Convulsion and the mortality rate was recorded and compared with the respective control group. The ability of the plant extract to prevent or delay the onset of the hind limb extension exhibited by the animals was taken as an indication of anti-convulsant activity [8-10].

### Table 1: Anti-convulsant activity of the extract on PTZ-induced convulsion

<table>
<thead>
<tr>
<th>Drug extract</th>
<th>Dose (mg/kg)</th>
<th>Time for Onset of Convulsion (min)</th>
<th>Time of Death (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (normal saline)</td>
<td>0.2ml</td>
<td>0.47 ± 0.55</td>
<td>1.22 ± 0.10</td>
</tr>
<tr>
<td>32.40</td>
<td>0.93 ± 0.17</td>
<td>1.39 ± 0.10</td>
<td></td>
</tr>
<tr>
<td>64.80</td>
<td>1.26 ± 0.03*</td>
<td>2.68 ± 0.26*</td>
<td></td>
</tr>
<tr>
<td>96.20</td>
<td>3.33 ± 0.09*</td>
<td>5.66 ± 0.19*</td>
<td></td>
</tr>
<tr>
<td>Phenobarbital</td>
<td>30.00</td>
<td>3.65 ± 0.18*</td>
<td>6.15 ± 0.09*</td>
</tr>
<tr>
<td>Extract + Phenobarbital</td>
<td>64.80+30.00</td>
<td>1.48 ± 0.02*</td>
<td>3.14± 0.07*</td>
</tr>
</tbody>
</table>

Data are represented as mean ± SEM.

Significant at *p*< 0.05, **p**< 0.001, ***p**< 0.001 when compared to control (n=6).
Table 2: Anti-convulsant activity of the extract on picrotoxin-induced convulsion

<table>
<thead>
<tr>
<th>Drug Extract</th>
<th>Dose (mg/kg)</th>
<th>Time for Onset of Convulsion (min)</th>
<th>Time of Death (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (normal saline)</td>
<td>0.2ml</td>
<td>2.90 ± 0.25</td>
<td>3.85 ± 0.44</td>
</tr>
<tr>
<td>Extract</td>
<td>32.40</td>
<td>3.10 ± 0.30</td>
<td>6.50 ± 0.40</td>
</tr>
<tr>
<td>Extract</td>
<td>64.80</td>
<td>3.88 ± 0.17</td>
<td>8.81 ± 0.20</td>
</tr>
<tr>
<td>Extract</td>
<td>96.20</td>
<td>5.53 ± 0.25</td>
<td>11.58 ± 0.42</td>
</tr>
<tr>
<td>Phenobarbital</td>
<td>30.00</td>
<td>6.04 ± 0.24</td>
<td>11.34 ± 0.06</td>
</tr>
<tr>
<td>Extract + Phenobarbital</td>
<td>64.80+30.00</td>
<td>4.67± 0.71</td>
<td>9.48 ± 0.02</td>
</tr>
</tbody>
</table>

Data are represented as mean ± SEM. Significant at *p< 0.05, **p<0.01, ***p< 0.001 when compared to control (n=6).

4. Conclusions

From this study, it can be concluded that the stem bark of *Enantia chlorantha* has observable anticonvulsant effects. Evaluation of sub-chronic toxicological properties showed that in low doses, the extract has proven relatively safe for use in animals employed for these experiments. This is demonstrated in the biochemical parameters and post mortem histology of vital organs. Observable pharmacologically active phytochemical constituents of *Enantia chlorantha* may be responsible for the effects noted in the study. Therefore, this plant may be a good candidate for developing new treatment for convulsions. Conflict Of Interest: the authors hereby declare that we have no conflict of interest where this manuscript is concerned.

5. References