EVALUATING THE ANTICONVULSANT ACTIVITY OF THE HYDRO-METHANOL LEAF EXTRACT OF *Icacina trichantha* Oliv. IN MICE

BY

Faith Isoken IRANUWEN (Miss)

LSC2009814

PHYSIOLOGY AND PHARMACOLOGY TECHNIQUES

DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY

FACULTY OF LIFE SCIENCE

UNIVERSITY OF BENIN

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A PROJECT WORK SUBMITTED TO THE DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY, FACULTY OF LIFE SCIENCE, UNIVERSITY OF BENIN, BENIN CITY, EDO STATE, NIGERIA. IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE BACHELOR DEGREE (BSc.) IN SCIENCE LABORATORY TECHNOLOGY DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY (PHYSIOLOGY AND PHARMACOLOGY TECHNIQUES) FACULTY OF LIFE SCIENCES, UNIVERSITY OF BENIN, BENIN CITY

OCTOBER, 2025

CERTIFICATION

This is to certify that this project work titled "EVALUATING THE ANTICONVULSANT ACTIVITY OF THE HYDRO-METHANOL LEAF EXTRACT OF *Icacina trichantha* Oliv.IN MICE" was carried out by Faith Isoken IRANUWEN with Matriculation Number LSC2009814, of the Department of Science Laboratory Technology, Faculty of Life Sciences, University of Benin, Benin City.

____________________	____________
Mrs. E. Okpakpor (Project Supervisor)	Date
____________________	____________
Dr. P.O. Alonge (Project Coordinator)	Date
____________________	____________
Prof. J.O. Osarunwense (Head of Department)	Date
____________________	____________
External Examiner	Date

DEDICATION

I dedicate this work to God Almighty, for his grace to finish strong. It was you Lord in every step of the way. Thank you so much.

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I would like to express my heartfelt gratitude to God Almighty, for His undying love and unending provision towards me.

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ABSTRACT

Epilepsy, a brain disorder associated with recurrent seizure. Although, epileptic drugs exist, about 30% of patient have drug-resistants or experience adverse effects, emphasizing the need for safer alternatives. *Icacina trichantha* Oliv.is a medicinal plant which belongs to the family Icacinaceae, used traditionally in the treatment of epilepsy. However, there is limited scientific evidence supporting such claims. This study evaluates the anticonvulsant potential of *I. trichantha*.in mice. Fresh leaves were collected, air-dried, pulverized, and extracted using 1:1 water and methanol solvent to obtain H-MLE of *I. trichantha* after concentrating to dryness. Phytochemical screening was carried out using Sofowara 1993 and Harborne 1973. For anticonvulsant activity, 25 mice were randomly allotted into 5 groups of n = 5 for both MES and PTZ induced seizures. Group I received 10 ml/kg deionized water, groups II – IV received graded doses of the plant extract 100, 200 and 400 mg/kg and group 5 received 100 mg/kg phenobarbitone for MES and 5 mg/kg diazepam for PTZ and was observed for the presence of seizures. Phytochemical screening revealed the presence of flavonoids, alkaloids, tannins, terpenoids, phenolics, saponins, and carbohydrates. Results obtained revealed that H-MLE had a significant $P < 0.001$ at 400 mg/kg in onset of seizure and $P < 0.0001$ at 400 mg/kg in duration of seizure in the PTZ induced seizure. However, there was no significant result $P > 0.05$ in the MES. This study supports its use in the treatment of epilepsy in traditional medicine which could be due to presence of secondary metabolites.

CHAPTER ONE

INTRODUCTION

1.1 GENERAL OVERVIEW OF EPILEPSY

Globally, over 50 million people live with epilepsy, making it one of the most common chronic brain disorders. Epilepsy is a chronic neurological condition characterized by the brain's ongoing tendency to produce seizures without an obvious cause. In simple terms, it means the brain can sometimes have sudden bursts of abnormal activity that lead to seizures, even when there's no immediate trigger. The International League Against Epilepsy defines epilepsy as either having two or more unprovoked seizures that occur more than 24 hours apart (Fisher *et al.,* 2022).

1.1.1 Seizure

Seizure is essentially a sudden burst of abnormal electrical activity in the brain, which can cause a wide variety of symptoms from brief lapses in awareness, like staring spells, to intense, uncontrollable movements of the body. When these seizures happen repeatedly over time without a clear cause, the condition is known as epilepsy. It's this ongoing pattern of unpredictable brain activity that defines the disorder (Beghi, 2020).

1.1.2 Factors That Can Cause Epilepsy

Epilepsy can arise from a variety of underlying factors, many of which affect how the brain functions. Genetics play a significant role. Certain gene mutations can make a person more prone to seizures (Xavierarnau *et al.,* 2023).

Brain injuries, especially those caused by trauma, are another common cause and can lead to what's known as post-traumatic epilepsy (Ngadimon *et al.,* 2022).

Infections like meningitis and neurocysticercosis are major triggers in low-income areas, where access to healthcare and early treatment may be limited (Balestrini *et al.,* 2021). Problems around the time of birth, such as a lack of oxygen to the baby's brain, can also increase the risk of developing epilepsy later on (Ghosh, 2023). However, in many individuals, no clear cause can be identified; this is referred to as cryptogenic epilepsy (Beghi, 2023).

1.1.3 How It Affect Life

Living with epilepsy often means navigating a life filled with unpredictability. Since seizures can strike without warning, it's common for individuals to feel anxious, fearful, or even self-conscious especially in public settings. This constant uncertainty can impact a person's confidence in everyday activities like driving, working, attending school, or socializing (Chen *et al.,* 2023).

Many people with epilepsy need to take daily medications to manage their seizures, but these treatments can come with side effects such as drowsiness, mood swings, or memory difficulties, which may make concentrating on tasks at work or school more challenging. Emotionally, some individuals may feel isolated or hesitant to talk about their condition, worrying that others might not understand or might judge them unfairly (Devinsky *et al.,* 2023).

Unfortunately, in some cultures and communities, outdated beliefs and misinformation about epilepsy still exist, leading to stigma, discrimination, and social exclusion. This can make the emotional and social burden of the condition just as difficult to manage as the medical side (Fisher *et al.,* 2023).

Epilepsy doesn't just impact the individual, it can deeply affect families as well. For parents of children with epilepsy, there's often a constant sense of worry, and the stress of anticipating the next seizure can place strain on relationships and family dynamics. Still, it's important to remember that many people with epilepsy go on to lead full, meaningful, and joyful lives. With proper medical treatment, emotional support from family and friends, and effective seizure management, the condition can often be kept under control allowing individuals and their loved ones to thrive (Tong *et al.,* 2024).

1.1.4 Epilepsy Treatment

Epilepsy is managed through several treatment options, but for most people, antiseizure medications (ASMs) are still the main and most effective approach. Recent research shows that with regular and consistent use, more than 70% of individuals with epilepsy can gain good control over their seizures. Thanks to medical advances, newer ASMs are showing promising results not only in reducing seizure frequency but also in minimizing side effects. For example, medications like cenobamate and brivaracetam were particularly effective for treating focal epilepsy, offering better seizure control and being easier on the body than many older drugs (Tong *et al.,* 2024).

For about 30% of people with epilepsy, seizures continue despite taking medication this is known as drug-resistant epilepsy. When that happens, other treatment options may be explored. One approach that has shown real promise, especially in children, is the ketogenic diet, a special high-fat, low-carbohydrate eating plan. Although it may sound unusual, this diet can help reduce the frequency and severity of seizures in some individuals when medications alone aren't enough (Li *et al.,* 2023).

When medications and dietary changes don't provide enough relief, doctors may turn to more advanced options like surgery or device-based therapies. For some individuals, brain surgery such as removing a small area of brain tissue where seizures start (like in a temporal lobectomy) can lead to a significant drop in seizures or even complete seizure freedom. In addition, certain medical devices that stimulate the brain or nerves have proven effective over time. Techniques like vagus nerve stimulation (VNS) and responsive neurostimulation (RNS) use small implanted devices to help control abnormal brain activity. VNS, for example, has been in use for over 17 years and has been shown to cut seizure frequency by half or more in about 50% of patients with a number of people even becoming seizure-free (Fisher *et al.,* 2024).

Newer treatment options are also emerging, including cannabidiol (CBD), a compound derived from the cannabis plant. CBD has received official approval for use in certain severe childhood epilepsy conditions, such as Lennox-Gastaut syndrome and Dravet syndrome. Studies have shown that CBD can make a real difference in reducing seizures for children with these challenging forms of epilepsy. While it's currently approved for specific cases, ongoing research suggests it may have even broader potential for helping others with epilepsy in the future (Devinsky *et al.,* 2023).

1.1.5 Orthodox Drugs Involved in Epilepsy

When it comes to managing epilepsy, orthodox or conventional medicine mainly relies on antiepileptic drugs (AEDs), also known as antiseizure medications (ASMs). These drugs help reduce or completely stop seizures in many people by balancing the electrical activity in the brain. Some of the most commonly prescribed medications today include; Carbamazepine, Valproic acid (or sodium valproate), Phenytoin, Lamotrigine, Levetiracetam, Topiramate, Brivaracetam, Clobazam etc. Each of these medications works in its own way, and doctors choose the right one based on several factors like the type of seizures a person has, their age, any other health conditions they may have, and how their body responds to the medication.

Take levetiracetam, for example. It’s a popular choice for both children and adults because it generally doesn’t interact much with other medications and does a good job of controlling seizures. However, like all medications, it’s not without side effects. Some people may experience mood changes, such as irritability or feelings of depression, while taking it (Chen *et al.,* 2023).

Valproic acid is often very effective in treating generalized seizures, making it a common choice for many patients. However, like all medications, it can come with side effects. Some people may experience weight gain, hair thinning, or hand tremors. In rare cases, it can affect liver function, which is why regular monitoring is important. It’s also important to note that valproic acid isn’t considered safe during pregnancy, as it can increase the risk of developmental issues in the baby. For this reason, it's usually avoided in women who are pregnant or planning to become pregnant (Verrotti *et al.,* 2023).

Newer antiseizure medications like brivaracetam and cenobamate are becoming more widely used, especially for people with drug-resistant epilepsy. These newer options often provide better seizure control for some individuals and may come with fewer side effects compared to older medications. Still, they're not completely without drawbacks. Even the latest drugs can cause issues like fatigue, dizziness, or sleep disturbances. In general, some of the more common side effects seen with antiepileptic drugs include; Feeling drowsy or overly tired, Dizziness or a sense of being light-headed, Difficulty with memory or concentration, Mood changes, including irritability or depression, Weight gain or weight loss, Skin rashes or allergic reactions, Changes in liver or kidney function (usually checked with routine blood tests). Because everyone responds differently, it often takes time to find the medication that offers the best balance of seizure control and manageable side effects (Tong *et al.,* 2024).

Epilepsy medications are grouped into different classes depending on how they work to stabilize the brain's electrical activity. While these drugs don't cure epilepsy, they play a key role in managing the condition by reducing or preventing seizures through various mechanisms in the brain. For instance, sodium channel blockers such as carbamazepine, phenytoin, lamotrigine, and lacosamide help by slowing down overly active nerve signals that can trigger seizures. Among them, lamotrigine is often praised for being effective in seizure control while causing fewer problems with thinking or memory compared to some other medications (Chen *et al.,* 2023).

Another group of epilepsy medications works by boosting the brain's natural calming chemical, GABA (gamma-aminobutyric acid). GABA helps quiet down overactive brain activity, which can reduce the likelihood and severity of seizures. Drugs like phenobarbital, clobazam, valproic acid, and tiagabine increase GABA activity to help manage seizures. Among them, clobazam has been especially helpful for children with epilepsy; it provides strong seizure control while causing less drowsiness compared to older GABA-based medications (Verrotti *et al.,* 2023).

Some epilepsy medications help by controlling calcium channels in brain cells. This action prevents the release of certain chemicals that can trigger seizures, helping to keep brain activity more stable. Medications like ethosuximide, gabapentin, and pregabalin work this way. Ethosuximide, in particular, remains a go-to treatment for absence seizures in children because it works specifically on that seizure type and tends to have manageable side effects (Tong *et al.,* 2024).

Some antiepileptic medications work in more than one way to control seizures, which can make them especially helpful for people who haven't responded well to other treatments. Valproic acid, topiramate, and zonisamide are examples of these multi-action drugs. Because they act on several brain pathways, they can be quite effective but they may also bring a broader range of side effects. For example, topiramate is known to help control seizures, but it can sometimes affect memory and concentration in certain individuals (Tong *et al.,* 2024).

1.1.6 Classes of People That Suffers From Epilepsy

Epilepsy can affect people at any age, but certain groups are more at risk than others. Infants and young children are especially vulnerable, often due to brain development issues or complications during pregnancy or birth. At the other end of the age spectrum, older adults particularly those who have had strokes, head injuries, or other neurological problems also face a higher risk. According to a U.S. health study from 2013 to 2018, adults aged 40 to 59 were found to be almost twice as likely to develop epilepsy compared to younger adults. The study also noted that poor sleep or sleep disorders can significantly raise the risk of seizures, showing just how important good sleep is for brain health (Yang *et al.,* 2023).

1.1.7 Diagnosis of Epilepsy

Diagnosing epilepsy usually begins with a thorough discussion between the doctor and patient. The doctor will ask detailed questions about the person's medical background, as well as any symptoms or seizure-like events they've experienced. This helps build a clearer picture of what might be happening in the brain and guides the next steps in evaluation (Chaplin, 2022). As part of the evaluation for epilepsy, doctors often perform a physical and neurological exam to look for signs of brain-related issues such as memory difficulties, changes in reflexes, or problems with muscle coordination. To directly assess brain activity, an electroencephalogram (EEG) is commonly used. This test involves placing small sensors on the scalp to record the brain's electrical signals. If a standard EEG doesn't capture enough information, especially if seizures are infrequent, doctors may recommend extended monitoring over several days, either at home or in a hospital setting, to increase the chances of detecting abnormal brain activity (Yusuf *et al.,* 2024).

Doctors often use brain imaging tools like MRI or CT scans. These scans help identify visible problems in the brain such as scar tissue, tumors, or abnormal blood vessels that could be triggering seizures. MRI is typically preferred because it provides more detailed images. For more complex cases, especially when surgery is being considered, doctors may use advanced imaging techniques like magnetoencephalography (MEG) or metabolic scans such as PET and SPECT. These tests help pinpoint the exact areas in the brain where seizures start. In addition, blood tests and genetic screening may be used particularly if epilepsy begins early in life or appears to run in families. These tests can help rule out other medical conditions or identify inherited genetic factors that might be contributing to the seizures (Dahl-Hansen *et al.,* 2018).

1.1.8 Globally Is CNS Disease the Major Leading Cause Of Death

Diseases affecting the central nervous system (CNS) aren't the leading cause of death globally, but they do cause a major burden in terms of long-term disability. By contrast, cardiovascular diseases especially heart attacks and strokes remain the top cause of death worldwide, accounting for around 17.9 million deaths each year (Roth *et al.,* 2020).

However, neurological conditions such as stroke, dementia, epilepsy, and migraines have a widespread impact on quality of life. As of 2021, these disorders affected over 3.4 billion people globally and were responsible for about 443 million disability-adjusted life years (DALYs) , the highest burden of any disease group (Joseph *et al.,* 2017). This highlights their significant role in long-term health challenges, even if they aren't the top killers.

Epilepsy is one of the most common disorders of the central nervous system (CNS). While it's not a leading cause of death, conditions like heart disease still top that list. It affects a large number of people worldwide, with an estimated 52 million individuals living with epilepsy as of 2021. According to the 2021 Global Burden of Disease study, epilepsy was ranked among the top ten neurological conditions contributing to disability-adjusted life years (DALYs) , a measure of both years lost due to illness and early death. Alongside conditions like stroke, migraine, and dementia, epilepsy is part of a group of neurological disorders that together accounted for 443 million DALYs, making them the most burdensome group of diseases globally in terms of long-term health impact (Feigin *et al.,* 2021).

Although conditions like stroke may lead to more deaths, epilepsy continues to be a major cause of long-term disability, especially in low- and middle-income countries, where over 80% of the global epilepsy burden is concentrated. In these regions, limited access to diagnosis, treatment, and ongoing care often worsens the impact of the condition, making it a significant public health concern despite its lower mortality rate. Although death rates from epilepsy have slightly decreased over time, the number of people living with the condition continues to rise. This increase is largely driven by secondary epilepsy, which develops as a result of other health issues like head injuries, brain infections, or complications around birth. These underlying causes are especially common in areas with limited access to healthcare, contributing to the growing burden of the disease worldwide (Manole, 2023).

1.1.9 Animals Models for Carrying Out Epilepsy In The Laboratory

Scientists often use animal models to better understand epilepsy and how to treat it. These models are carefully designed to reflect different types of epilepsy seen in humans, allowing researchers to explore how seizures begin, how they affect brain function, and which treatments might work best. One widely used method involves giving animals a substance called pentylenetetrazole (PTZ). PTZ works by blocking GABA receptors, which are responsible for calming brain activity. When these receptors are blocked, it causes generalized seizures similar to what some people with epilepsy experience. This model helps researchers test new medications aimed at reducing excessive brain activity and controlling seizures more effectively (Yang *et al.,* 2024).

To study focal or partial seizures, researchers often use a method called "kindling." This involves applying small, repeated electrical pulses to specific parts of the brain, such as the amygdala or hippocampus. Over time, this gentle stimulation makes the brain more sensitive and more likely to have seizures, mimicking the gradual development of chronic epilepsy seen in some people. The kindling model helps scientists understand how localized seizures form and progress, and it's especially useful for testing treatments aimed at this specific type of epilepsy (Lee *et al.,* 2024).

To better understand temporal lobe epilepsy, researchers often use chemicals like pilocarpine or kainic acid in lab models. These substances overactivate brain cells, triggering long-lasting seizures. After this initial phase, the animals usually go on to develop repeated, spontaneous seizures, similar to what happens in people with chronic epilepsy. This model allows scientists to study not just how seizures occur, but also how epilepsy changes the brain over time. It's especially useful for testing treatments that aim to slow or prevent the progression of the disease itself, rather than just controlling individual seizures (Khan *et al.,* 2023).

1.1.10 Principles behind the Animal Models

Some research models are specifically designed to study drug-resistant epilepsy, where typical seizure medications aren't very effective. One such model is the 6 Hz seizure model, which uses low-frequency electrical stimulation to trigger seizures that don't respond well to standard treatments. Because it closely mimics the challenges faced by people with treatment-resistant epilepsy, this model is especially valuable for testing newer drugs like cenobamate or levetiracetam that may offer better seizure control when other options fail (Rodríguez-Almaraz *et al.,* 2023).

Beyond models that use chemicals or electrical stimulation, researchers also use genetic models to study epilepsy. A well-known example is the GAERS rat, which naturally develops absence seizures briefly, starting episodes that closely resemble those seen in some people with epilepsy. Because these seizures occur without external triggers, GAERS rats are especially valuable for exploring inherited forms of epilepsy and testing treatments that target the underlying genetic causes (Bhandari *et al.,* 2023).

Even small, simple animals like zebrafish and fruit flies play an important role in epilepsy research. Zebrafish larvae respond to seizure-inducing chemicals in ways that are surprisingly similar to mammals, and because their bodies are transparent, researchers can easily watch how brain activity changes in real time. Fruit flies with gene mutations linked to epilepsy are also useful for exploring the basic genetics of the condition. They allow scientists to quickly screen large numbers of potential drugs. Each model has unique advantages. Some are ideal for studying short-term seizure activity, while others are better for understanding chronic epilepsy or drug resistance. By carefully choosing the right model for their research goals, scientists can improve the development of safer, more targeted epilepsy treatments (Hernandez *et al.,* 2024).

1.2 SCOPE OF THE STUDY

This study aims to find out if the hydro-methanol leaf extract of *Icacina trichantha* Oliv. can help prevent or reduce seizures in laboratory mice. To carry out this research, we'll observe the mice's behavior and test the extract's effects using two trusted seizure models: one triggered by pentylenetetrazole (PTZ) and the other by maximal electroshock (MES). These two models were chosen because they mimic different types of seizures (PTZ) for absence seizures and (MES) for tonic-clonic ones and they're widely used in early research to test whether new substances might help prevent seizures (Thomé *et al.,* 2023).

The study will also look into what natural compounds are present in the extract, to help figure out which ones might be behind its seizure-fighting effects. The study will also include a safety test to find out how much of the extract could be harmful to the mice, helping determine whether it's safe enough to explore further as a possible treatment (Okeke *et al.,* 2021).

This study is being carried out only on animals (mice) at this stage, and doesn't yet involve testing on humans. In this study, we're only using the leaves of *Icacina trichantha* and extracting them with a mix of water and methanol, based on the idea that this method can pull out a wide range of useful compounds from the plant (Hosseinzadeh *et al.,* 2021).

This study doesn't go as far as breaking down the extract to find individual compounds or investigating how it works at the molecular level, like how it interacts with brain receptors or affects brain chemicals. Even so, the results from this study will offer early scientific support for the traditional use of *Icacina trichantha* in managing seizures, and could lay the groundwork for future research to identify the active compounds, understand how they work, and eventually test them in humans (Lattanzi and Trinka, 2022; Zuberi *et al.,* 2021).

This study covers the process of preparing the plant extract using a mix of water and methanol, a method known to pull out a wide variety of natural compounds like alkaloids, flavonoids, tannins, saponins, and terpenoids which have shown promise in affecting the brain and nervous system in similar plants (Okeke *et al.,* 2021).

The results from this study are anticipated to provide valuable preclinical evidence supporting the ethnomedicinal use of *Icacina trichantha* in seizure management. These findings may also serve as a foundation for future investigations into its pharmacodynamic mechanisms and its potential as a lead compound in the development of novel antiepileptic drugs (Zuberi *et al.,* 2021; Okeke *et al.,* 2021).

1.3 Significance of The Study

Epilepsy is still one of the most widespread brain disorders around the world, and it tends to impact people in low and middle income countries the most where getting reliable and affordable treatment can be a real challenge (Zuberi *et al.,*2021).

Even with all the progress in developing epilepsy medications, about 30% of people still suffer from seizures that don't respond well to treatment or they experience unwanted side effects from the drugs they take (Lattanzi and Trinka, 2022).

This study matters because it looks into the science behind the traditional use of *Icacina trichantha* Oliv. a medicinal plant commonly used in West Africa to treat seizures and similar health issues (Okeke *et al.,*2021).

By using well-known laboratory tests like the pentylenetetrazole (PTZ) and maximal electroshock (MES) seizure models, it gives clear evidence-based insight into whether the hydro-methanol leaf extract of the plant might help prevent or reduce seizures (Thomé *et al.,* 2023).

This study also adds to the growing support for using plant-based treatments as possible options for developing new epilepsy drugs. After all, many of the medicines we use today came from nature, and medicinal plants are still a valuable source of powerful, healing compounds (Hosseinzadeh *et al.,* 2021).

If shown to be both effective and safe, *Icacina trichantha* could become a promising starting point for developing new antiepileptic drugs or offer a more affordable treatment option, particularly in areas where access to standard medications is limited. These findings will also help build the scientific understanding of the plant by either supporting or questioning its traditional use based on actual experimental results. In the end, this study supports the global push to expand treatment choices for epilepsy, encourages the responsible use of traditional remedies backed by science, and helps close the treatment gap especially in communities with limited access to healthcare (Devinsky *et al.,* 2018).

1.4 Aim of The Study

The aim of this study is to evaluate the hydro-methanol leaf extract of *Icacina trichantha* Oliv. in albino mice using maximal electric shock and chemically induced epilepsy using pentylene tetrazole.

1.5 Objectives of Study

1. To carry out extraction of the plant extract
2. To carry out phytochemical screening of the plant extract
3. To evaluate the anticonvulsant activity of the plant

CHAPTER TWO

LITERATURE REVIEW

2.1 *Icacina trichantha*

Icacina trichantha Oliv., from the Icacinaceae family, is a hardy, drought-resistant perennial plant that grows naturally across West and Central Africa, especially in Nigeria, Ghana, and Cameroon. Despite being underutilized, it holds cultural importance and is recognized by different local names such as "Urumbia" in Igbo, "Gbegbe" in Yoruba, and "False yam" in English due to its yam-like tubers (Wahua and Awogbayila, 2024).

The plant plays a vital role in many rural communities, where it's valued not just as traditional medicine but also as a source of food especially during times when food is hard to come by. It thrives naturally in tropical savannas and areas where forests meet grasslands, and people usually gather it from the wild rather than grow it on farms (Wahua and Awogbayila, 2024).

Icacina trichantha has a distinctive physical structure, starting with a woody base that supports long, climbing stems. Its leaves are broad and lush, and it produces round-shaped fruits. Beneath the ground, the plant develops large tubers sometimes reaching sizes comparable to regular farm-grown yams (Ekeke *et al.,* 2021).

A nutritional breakdown of the leaves and tubers shows that the leaves are especially rich in protein, fiber, and essential minerals. They contain about 27% protein, over 43% crude fiber, and are packed with calcium, potassium, and iron making them quite nutritious. On the other hand, the tubers are more energy-giving, with nearly 74% carbohydrates and a decent amount of protein at around 11%, making them a good food source during periods of high energy demand. These results show that both the leaves and tubers of the plant can play an important role in meeting nutritional needs, especially in areas where food is scarce. The study also found that the plant contains a variety of natural compounds like alkaloids, tannins, flavonoids, saponins, terpenoids, and glycosides which suggests it may have medicinal benefits as well (Alawode, 2024).

Advanced testing including phytochemical screening and GC-MS analysis revealed nine key compounds in the chloroform extract of *Icacina trichantha* tuber peels. Among the most abundant were undecane (making up 43.3%), 2-hexanone (23.3%), and dodecanoic acid ethyl ester (6.2%). The extract also contained bioactive substances like 9-octadecynoic acid and triarachine, which are known for their potential health benefits In a study by (Otuokere *et al.,* 2022).

These compounds have shown a wide range of healing possibilities; they've been found to fight off bacteria, viruses, and even certain types of disease-causing microbes. Interestingly, computer-based studies (in silico) suggest that some of them may also block key proteins of the SARS-CoV-2 virus, pointing to their potential use in modern antiviral treatments (Otuokere *et al.,* 2022).

Researchers have also looked into the essential oils found in *Icacina trichantha* leaves, examining both fresh and air-dried samples. The fresh leaves produced about 0.39% oil and contained 17 different compounds, including 9-oxabicyclo[6.1.0]nonane, oleic acid, and phytol. Although the air-dried leaves yielded slightly less oil (0.33%), they were notably rich in linoleic acid (34.6%) and gamma-sitosterol. These oils proved to be quite effective against fungal infections, especially strains like *Candida albicans* and *Candida tropicalis*, showing strong antifungal activity at low concentrations (MIC around 0.20 mg/mL). In addition, the oils displayed impressive antioxidant properties, with their ability to neutralize free radicals closely matching that of ascorbic acid, a well-known antioxidant (Olubomehin *et al.,* 2024).

Tests on the antioxidant and anti-inflammatory properties of *Icacina trichantha* revealed impressive results. The methanol extract of the leaves was packed with beneficial plant compounds, showing a high phenolic content and strong ability to neutralize harmful free radicals comparable to known antioxidants. The tuber extract also showed solid antioxidant power, based on its Ferric Reducing Antioxidant Power (FRAP) value. Notably, the ethyl acetate extract from the leaves stood out for its anti-inflammatory effect in lab tests. It helped stabilize cell membranes and, at lower doses, even performed better than the commonly used drug indomethacin (Alawode, 2024).

2.1.1 Taxonomy

Kingdom: Plantae

Phylum: Tracheophyta

Class: Equisetopsida

Subclass: Magnoliidae

Order: Icacinales

Family: Icacinaceae

Genus: Icacina

Specie: Trichantha

(Ekeke *et al.,* 2021)

2.1.2 Botanical Description

Icacina trichantha Oliv. is a hardy, long-living shrub or small tree from the Icacinaceae family, known for its sturdy, woody structure. It usually reaches a height of about 2 to 4 meters and is easily recognized by its large, swollen underground root system, which grows deep into the soil. This thick, tuberous root is often valued for its traditional use in food and medicine, thanks to its nutritional and healing qualities. *Icacina trichantha* has unique physical characteristics in both its leafy and flowering parts, which make it quite easy to identify in the wild. These distinct features not only support its use in traditional practices but also add to its potential value in modern medicine (Alonso *et al.,* 2022)

Leaves: The leaves of *Icacina trichantha* are simple and arranged alternately along the stem, typically taking on a broad oval to elliptical shape. They vary quite a bit in size, usually measuring between 10 to 30 cm long and 6 to 15 cm wide. The tips taper to a point (acuminate apex), while the base is either rounded or slightly heart-shaped. Leaf edges are smooth (entire margins). On the top side, the leaves are smooth and dark green, while the underside tends to be lighter in color and may have fine hairs, especially along the veins. The leaf veins follow a pinnate pattern, with a clear central vein (midrib) and smaller side veins that curve gently toward the edges. Each leaf is supported by a sturdy petiole (leaf stalk) that ranges from 2 to 6 cm in length. The internal structure of the leaf shows a thick outer layer (cuticle) and spongy parenchyma tissue beneath, both of which help the plant retain moisture and provide structural strength, especially in dry or harsh conditions (Ekeke *et al.,* 2021).

Flowers: The flowers of *Icacina trichantha* are small, greenish-yellow in color, and contain both male and female parts. They grow in loose clusters at the tips of branches or in the angles between the stem and leaves. Each flower is made up of five sepals and five petals arranged in a balanced, circular pattern. There are also five stamens positioned between the petals, and the ovary sits above the base of the flower (superior), with three chambers fused together each containing a single ovule. These flowers typically bloom during the rainy season in their natural environment and rely on insects for pollination. The structure of the flowers is well-suited for attracting a variety of pollinators, which helps improve the plant’s chances of successful reproduction, especially in the mixed forest and savanna regions where it naturally grows (Wahua and Awogbayila 2024).

Stem: The stem of *Icacina trichantha* is thick, upright, and woody, and in mature plants, it can grow as tall as 4 meters. It tends to have a moderate number of branches and is wrapped in bark that's usually smooth or slightly rough with a brownish tone. The internal structure of the stem features well-organized vascular bundles, with strong, lignified xylem tissues that help transport water efficiently throughout the plant. These tough tissues also give the stem the strength it needs to stay upright and support its overall growth (Ekeke *et al.,* 2021).

Root: *Icacina trichantha* has a distinctive root system made up of large, fleshy tuberous roots that grow deep into the ground. These tubers act as storage centers, packed with starch and beneficial plant compounds. On the outside, the tubers are rough and brown, but when cut open, they reveal a soft, yellowish-white interior with a mealy texture, highlighting their nutritional and medicinal value. Anatomically, the tuber of *Icacina trichantha* is made up of soft parenchyma cells filled with starch granules, along with thick-walled storage tissues. This internal structure not only boosts its nutritional content but also helps the plant survive dry conditions by storing energy and moisture underground (Wahua and Awogbayila 2024).

Tubers: The tubers, which form a major part of the root system, are usually round or slightly elongated in shape and can grow quite large, often weighing several kilograms. They're commonly harvested for both traditional medicine and as a food source. Rich in beneficial plant compounds like alkaloids, saponins, flavonoids, and terpenoids, these tubers are especially valued for their ability to fight bacteria and neutralize harmful free radicals, making them important for both health and nutrition. The tubers have special structural features like a thick outer layer and large storage cells that help them survive tough environmental conditions. These same features also make them useful in traditional medicine and as a food source, showing how nature and culture have both shaped their importance (Alawode, 2024).

Plate 1: The leaves of *Icacina trichantha oliv.* (Wahua and Awogbayila 2024)

2.1.3 Origin of the plant

Icacina trichantha originally comes from the moist tropical regions of West Africa, particularly the transitional zones between forest and savanna found in places like Benin and southern Nigeria. Although the plant was first officially described by Daniel Oliver back in 1868, more recent research, especially ecological and anatomical studies has helped deepen and clarify what we know about its natural origins. Morphological and anatomical studies conducted on *Icacina trichantha* populations in Nigeria have confirmed that this species, along with other members of the *Icacina* genus, is endemic to secondary forest ecosystems and woodland savannas in southern Nigeria. Researchers noted that the plant thrives in sandy to lateritic soils, within annual precipitation ranges of 800 to 1,500 mm, and is commonly found at altitudinal gradients reaching up to 1,000 meters above sea level (Ekeke *et al.,* 2021).

Icacina trichantha is native to the moist savanna zones of West Africa, where it's commonly found growing in alongside plants that are well adapted to cope with seasonal dry spells and moderate levels of rainfall (Wahua and Awogbayila 2024).

According to the 2024 update from Kew's Plants of the World Online, *Icacina trichantha* is confirmed to grow naturally from Benin through to southern Nigeria, where it's described as a climbing shrub that thrives in wet tropical environments. When considered alongside other recent studies from 2021 and beyond, the evidence clearly shows that this plant is both native and unique to the forest-savanna transition zones of West Africa. Its natural habitat and regional spread have been well documented through up-to-date botanical and ecological research.

2.1.4 Traditional Uses

Icacina trichantha holds great importance in West African traditional medicine and food culture, where it's widely used for both healing and nutritional purposes.

The tuber is commonly eaten during periods of food shortage or famine because it's rich in starch, making it a reliable source of energy when other food options are limited. In Nigeria and Ghana, people often dry and grind the tuber into flour, which they use to prepare porridge or thicken soups. While the raw tuber has a bitter taste and contains some toxic compounds, traditional processing methods like drying, soaking, or boiling help eliminate the bitterness and make it safe and more pleasant to eat (Wahua and Awogbayila 2024).

Traditionally, boiled extracts made from the tuber and leaves of *Icacina trichantha* are used as natural remedies to treat digestive problems like diarrhea, dysentery, and stomach aches. In traditional medicine, healers often give water-based extracts of the tuber to help fight bacterial infections. The leaves are usually crushed and applied directly to the skin to treat wounds, boils, and rashes, thanks to their soothing and germ-fighting properties. These healing effects are believed to come from natural compounds like alkaloids, flavonoids, and tannins, which are known for their ability to combat microbes and support skin repair (Che *et al.,* 2016).

Icacina trichantha is also used traditionally to help manage malaria and fevers. People often boil the leaves or tubers to make a herbal drink that's taken by mouth to lower body temperature and relieve symptoms of fever (Ekeke *et al.,* 2021).

Another key traditional use of *Icacina trichantha* is in managing conditions like high blood pressure and diabetes, where parts of the plant are used to help regulate blood sugar and support heart health. In many rural communities, older adults often take small amounts of tuber extracts as part of their daily routine to help keep their blood pressure and blood sugar in check. This long-standing practice aligns with recent research that has confirmed the plant's natural ability to lower blood sugar and reduce blood pressure (Wahua and Awogbayila 2024).

In certain communities, *Icacina trichantha* holds spiritual meaning beyond its physical uses. People sometimes bury the tuber near the entrance of their homes or along farm boundaries as a protective charm to ward off evil or bad luck. This practice is deeply rooted in traditional beliefs and cultural symbolism rather than any known medical function (Ekeke *et al.,* 2021).

2.1.5 Pharmacological Reports On *Icacina trichantha* leaf extract

Recent scientific research on extracts from the leaves of *Icacina trichantha* has validated many of its traditional uses by demonstrating key biological effects. These include strong antioxidant activity, the ability to fight off harmful microbes, reduce inflammation, relieve pain, and promote wound healing all of which support its long-standing role in folk medicine (Alawode, 2024).

Antioxidant Activity

One of the most thoroughly explored medicinal benefits of *Icacina trichantha* leaf extract is its antioxidant capacity. Studies have shown that both methanol and water-based extracts of the leaves are rich in polyphenolic compounds such as flavonoids, tannins, and phenolics that help neutralize harmful free radicals in the body. When tested using standard antioxidant assays like DPPH and Ferric Reducing Antioxidant Power (FRAP), the methanol extract showed strong free radical scavenging activity, exceeding 70% at a concentration of 100 µg/mL comparable to the effects of vitamin C. These results indicate that *Icacina trichantha* could play a protective role against diseases linked to oxidative stress. By reducing the buildup of harmful free radicals, the plant's compounds may help lower the risk of conditions like atherosclerosis, cancer, diabetes, and neurodegenerative disorders such as Alzheimer's (Alawode, 2024).

Antimicrobial Properties

Studies have shown that both ethanol and water-based extracts of *Icacina trichantha* leaves are effective against several harmful microorganisms, including *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Candida albicans*. Among the two, the ethanol extract showed more powerful antimicrobial action, producing clear zones of inhibition between 12 and 20 millimeters, and minimum inhibitory concentrations (MICs) ranging from 100 to 250 µg/mL indicating its strong potential to suppress the growth of these pathogens. Through GC-MS analysis, researchers identified key bioactive compounds in *Icacina trichantha* leaves, including phytosterols like stigmasterol and β-sitosterol, fatty acids such as oleic and linoleic acid, and triterpenes. These natural compounds are thought to play a major role in the plant's antimicrobial activity. Their presence helps explain why the leaves have traditionally been used to treat infections and promote wound healing, giving scientific backing to those long-standing medicinal practices (Olubomehin *et al.,* 2024).

Wound-Healing Activity

Applying *Icacina trichantha* leaf extract directly to the skin has shown encouraging results for wound healing. In studies using rats with skin wounds, those treated with an ethanolic extract of the leaves experienced quicker wound closure, faster skin regeneration, and improved formation of healthy tissue compared to untreated animals. These results suggest the extract may help speed up the body's natural healing process. In a study involving rats with skin wounds, those treated with an ethanol-based extract of *Icacina trichantha* leaves healed more quickly than untreated rats. The wounds in the treated group closed faster, new skin formed more rapidly, and the overall tissue repair was more organized and healthy, showing the extract's potential to support and speed up natural wound healing. Microscopic examination of the healed tissue showed increased collagen production and the formation of new blood vessels, both of which are vital for proper wound healing. These effects are believed to be driven by flavonoids and tannins in the extract, as these natural compounds help boost fibroblast activity key cells involved in tissue repair while also protecting the wound from infection (Wahua and Awogbayila 2024).

Anti-Inflammatory and Analgesic Effects

The leaf extract of *Icacina trichantha* has been shown to have both anti-inflammatory and pain-relieving effects. In one study, rats given the extract by mouth showed a clear reduction in swelling in a standard inflammation test (carrageenan-induced paw edema), with greater effects at higher doses. In another test that mimics pain (acetic acid-induced writhing), the extract noticeably reduced the number of pain-related reactions, suggesting it can help relieve discomfort as well as reduce inflammation. The pain-relieving effect of *Icacina trichantha* leaf extract was also evident in the hot-plate test, which points to its ability to influence the central nervous system's response to pain. Researchers believe this effect comes from naturally occurring compounds in the plant such as alkaloids, flavonoids, and terpenoids that help block the production of inflammation-triggering substances like prostaglandins and cytokines (Otuokere *et al.,* 2022).

Phytochemical Basis of Activities

Icacina trichantha contains a rich mix of naturally active compounds such as alkaloids, saponins, flavonoids, steroids, tannins, glycosides, and terpenoids. These substances don't just work individually; they often act together, enhancing each other's effects. This synergy likely explains why the plant shows such a wide range of health benefits in both lab-based (in vitro) and animal (in vivo) studies (Olubomehin *et al.,* 2024).

2.1.6 Phytochemistry of *Icacina trichantha*

The chemical constituents of *Icacina trichantha* leaf extracts obtained successively using n-hexane, ethyl acetate, and ethanol were thoroughly analyzed using gas chromatography and mass spectrometry (GC/GC-MS). Preliminary phytochemical screening of the crude extracts confirmed the presence of several biologically active compounds, including tannins, flavonoids, phenols, and glycosides. These groups of compounds are widely recognized for their therapeutic potential and are likely contributors to the plant's traditional medicinal uses. In evaluating the plant's antibacterial properties, the study tested all three extracts against gram-negative bacteria: *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella oxytoca*. Results showed that the n-hexane and ethyl acetate extracts were more effective than the ethanol extract in inhibiting the growth of these bacterial strains. The extracts were also assessed for antioxidant capacity using a spectrophotometric DPPH assay, which measures the ability to neutralize free radicals. Among the extracts, the hexane fraction demonstrated the strongest antioxidant activity, with an IC_{50} value of 0.21 g/mL significantly lower (and therefore more potent) than the IC_{50} values of the ethyl acetate (3.917 mg/mL) and ethanol extracts (4.812 mg/mL). For the first time, the study identified key compounds responsible for these biological effects: stearic acid was the major constituent in the hexane extract (30.74%), oleic acid dominated the ethyl acetate extract (36.04%), and erucic acid was the most abundant in the ethanol extract (29.01%). These compounds, either individually or in combination, are believed to contribute significantly to the observed antibacterial and antioxidant activities of the plant (Otun *et al.,* 2015).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Plant Collection and Identification

Fresh *Icacina trichantha* leaves were sourced from a local market in Ovia North East Local Government Area of Benin City, Nigeria. The plant was formally identified by Prof. E.I. Aigbokhan, with verification and voucher number UBH-1185 provided by Dr. H.A. Akinigbosun, both of the Department of Plant Biology and Biotechnology at the University of

Benin. After collection, the leaves were carefully washed, air-dried, and ground into a fine powder. The resulting material was stored in an airtight container until it was ready for use in further analysis.

3.2 Experimental Animals

Albino Wistar mice, weighing between 15-25g, were obtained from the animal facility located within the Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Benin, Benin City, Nigeria. The animals were kept in standard plastic cages and had free access to water and feed (Livestock feeds Ltd Ibadan Nigeria) and were exposed to natural lighting and room temperature. This study was approved by the Faculty of Science Laboratory Technology Research Ethical Committee with reference number UNIBEN/FSLT/0023.

3.3 Preparation of Plant Extract

Fresh *Icacina trichantha* leaves were thoroughly rinsed under running water to remove dirt, and then air-dried under controlled conditions. Once fully dried, the leaves were ground into fine powder. A total of 100 grams of the powdered plant material was measured using a digital balance and transferred into a clean maceration container. To extract the bioactive compounds, the powder was soaked in a 1:1 mixture of methanol and water (600 ml each). The container was sealed with parafilm to prevent contamination and shaken vigorously to mix the contents well. The mixture was left to stand for 72 hours, during which it was shaken intermittently to enhance extraction. After three days, the liquid mixture was filtered separating the liquid extract (filtrate) from the solid plant residue (chaff), which was discarded. The filtrate was then poured into sterilized stainless-steel trays and dried in a hot-air oven set at 53°C until a solid extract formed. Once dried, the extract was carefully scraped off and transferred into a clean universal bottle.

3.4 Dose Preparation of Extract

For the dose of 100mg/kg, 42mg/kg of the extract was weighed and dissolved in 3mls of deionized water. For the dose of 200mg/kg, 72mg/kg of the extract was weighed and dissolved in 3mls of deionized water. For the dose of 400mg/kg, 141mg/kg of extract was weighed and dissolved in 3mls of deionized water. They were stored in a fridge for future use. Administration to the experimental animals was via oral route using an oral gastric tube. The animals were weighed before the administration to get the volume to be administered to each animal.

3.5 Experimental Designs

After 14 days of acclimatization to laboratory conditions, the Wistar albino mice
were allotted into four (4) groups of n = 7.

Group I served as the control and was administered de-ionized water.

Group II was administered a dose of 100 mg/kg hydro-methanol extract of *Icacina trichantha*

Group III was administered a dose of 200 mg/kg hydro-methanolic extract of *Icacina trichantha*

Group IV was administered a dose of 400 mg/kg hydro-methanolic extract of *Icacina trichantha*

All administrations were via the oral route with the aid of an orogastric tube.

3.6 Phytochemical Screening

Simple chemical tests were carried out on the crude powdered sample and the methanol extract according to standard procedures to identify the phytochemical constituents (Stahl, 1973; Sofowora, 1982; Harborne, 1998; Evans, 2002).

Approximately 5 g of the crude powdered sample was boiled with 75 mL of distilled water for 30 minutes. The solution was filtered hot and allowed to cool. The filtrate obtained was used to carry out the following tests.

3.6.1 General Tests for Alkaloids

Two drops of Dragendorff's reagent was added to 2 mL of the filtrate.

Two drops of Wagner's reagent was added to 2 mL of the filtrate.

Two drops of Hager's reagent was added to 2 mL of the filtrate.

Two drops of Mayer's reagent was added to 2 mL of the filtrate.

3.6.2 Tests for Carbohydrates

Molisch's Test

To 2 mL of filtrate was added 2 drops of 1% alcoholic naphthol followed by 2 mL of concentrated sulphuric acid at a slanting position.

3.6.3 Tests for Reducing Sugars

Fehling's Test

To 2 mL of filtrate was added 2 drops of Benedict's reagent (a mixture of equal volumes of Fehling's solution A and B). The resulting solution was heated over a boiling water bath for 3 minutes.

Tollen's Test

Dilute ammonium hydroxide solution was added drop wisely to silver nitrate solution containing a few drops of 10% sodium hydroxide until the precipitate of silver oxide almost completely dissolved. Few drops of the filtrate was then added to the mixture above.

Keller Kiliani's Test for Deoxysugars

To 2 mL of filtrate was added few drops of dilute acetic acid containing a trace of 5% ferric chloride. The resulting mixture was transferred to the surface of concentrated sulphuric acid.

Test for Saponins

Frothing Test

The filtrate (1 mL) was diluted with 10 mL distilled water and shaken vigorously for one minute.

Fehling's Test

To 10 mL of the filtrate was added 5 mL of dilute H_2SO_4. The mixture was boiled for 15 min, filtered and cooled. 2.5 mL of the filtrate was made alkaline with 20% NaOH solution and boiled with 0.1 mL each of Fehling's solutions A and B for 2 minutes.

Lieberman Burchard's Test for Steroidal saponins or Phytosterols

A mixture of 1 mL chloroform and few drops of acetic anhydride was added to 2 mL of the filtrate. To the final mixture was added 2 drops of concentrated sulphuric acid.

3.6.5 Test for Tannins

Gelatin Test

To 2 mL of the filtrate was added 2 mL of 1% gelatin solution in 10% NaCl.

Test for Terpenoids

Salkowski Test

The filtrate (5 mL) was mixed with 2 mL of chloroform and concentrate H_2SO_4 was carefully added (drop wise) to form a layer.

Test for Phenolic compounds

Ferric chloride Test

To 2 mL of filtrate was added 5 mL of distilled water followed by 2 drops of 5% ferric chloride solution. A blank test was done by adding 2 drops of 5% ferric chloride solution to 5 mL of distilled water.

Folin Ciocalteu's Test

To 5 mL of filtrate was added 0.5 mL 10 % folin ciocalteu's phenol reagent followed by 5 mL of 7% Na_2CO_3.

Test for Flavonoids

Alkaline reagent Test

To 2 mL of filtrate was added few drops of 20% sodium hydroxide solution followed by few drops of dilute hydrochloric acid solution.

Lead acetate Test

Few drops of lead acetate solution was added to 2 mL of the filtrate.

Aluminium chloride Test

The filtrate (3 mL) was shaken with 0.1 mL each of 1% $AlCl_3$ solution and 1 M CH_3COOK solution. The mixture was allowed to stand for 30 min.

Test for Anthraquinone Derivatives

Bontreger's Test

The filtrate (2 mL) was shaken with 2 mL of petroleum ether. The ether layer was washed with 2 mL distilled water and then shaken with dilute ammonia solution.

Test for Proteins

Xanthoproteic Test

Few drops of concentrated nitric acid was added to 2 mL of the filtrate.

Ninhydrin Test

Two drops of Ninhydrin solution was added to 2 mL of the filtrate.

3.7 Maximal Electro Shock

The mice were weighed, marked and placed in 5 groups of 5 mice each. The necessary calculations were made for the concentration and volume to be administered to each mice were done. Diazepam was administered interperitoneally and the *Icacina trichantha* extract was administered orally. 3-minute interval was taken after each administration and the time of administration was noted. A controlled electric shock using electrodes were placed on their ears. This was to look out for the mice to stiff,,extended movement of the bag legs ,which they did.

3.8 Pentylene Tetrazole induced epilepsy

The mice were weighed, marked and placed in 5 groups of 5 mice each. The necessary calculations were made for the concentration and volume to be administered to each mice were done, and a 3-minute interval was taken too after each administration. The standard drug, diazepam were administered interperitoneally and the extract were administered orally first, and after 30 minutes of administration, the mice were all induced with 70mg of PTZ interperitoneally and were observed for onset, type, duration and severity of seizures.

3.9 Data Analysis

The results are presented as mean values with the standard error of the mean [SEM], denoted as mean ± SEM, with 'n' indicating the number of experimental animals per group. Group comparisons were conducted using one-way analysis of variance [ANOVA] followed by Tukey's post hoc test. Statistically analysis was performed using GraphPad Prism software version 5. [GraphPad Software, USA]. A significance level of $P < 0.05$ was considered indicative of a statistically significant difference between the compared groups.

CHAPTER FOUR

RESULTS

Table 4.1: Qualitative phytochemical constituents of hydro-methanol extract of *I. trichantha*

TEST	INFERENCE
Flavonoid	+
Phenolics	+
Saponins	+
Terpernoids	+
Tannins	+
Alkaloids	+
Carbohydrates	+
Reducing Sugars	+
Assay Sugars	+
Proteins	_

- absent, + present

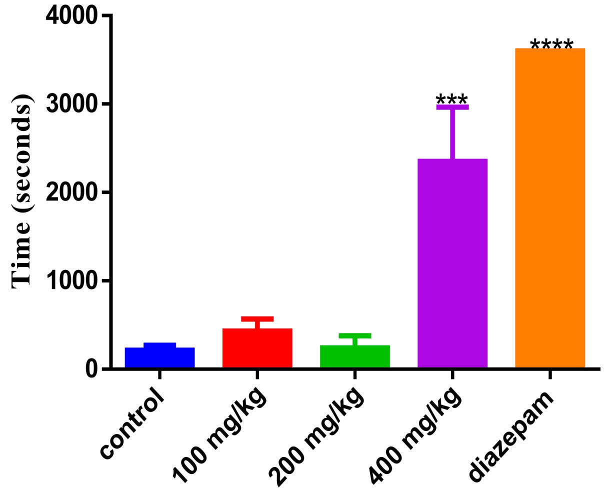

Figure 1: Effect of the hydro-methanol leaf extract of *Icacina trichantha* on the onset of seizure.

There is a significant increase at 400mg/kg when compared to the control ($P < 0.001$). Data are represented as mean ± SEM, n = 5.

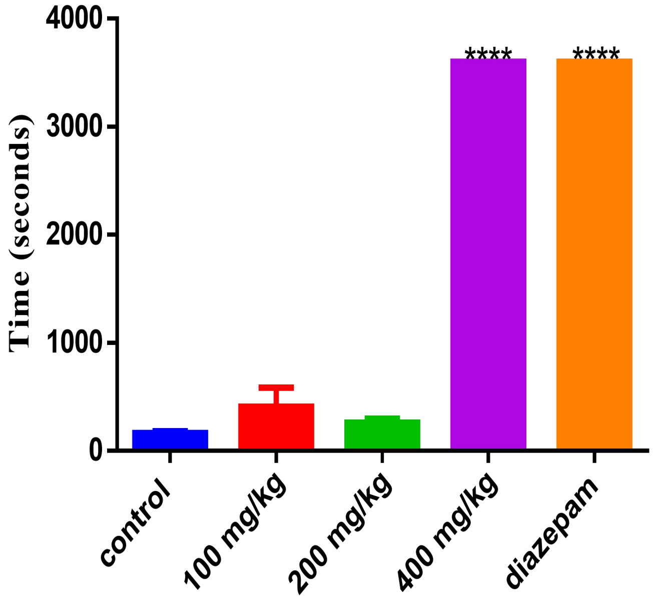

Figure 2: Effects of the hydro-methanol leaf extract of *Icacina trichantha* on the duration of seizure.

There is a significant increase at 400mg/kg when compared to the control ($P < 0.0001$). Data are represented as mean ± SEM, n = 5.

Table 4.2: Effect of the Hydro-Methanol Leaf Extract of Icacina Trichantha on Maximal Electro-Shock in Mice

TREATMENT	% PROTECTION
Control	0
100 mg/kg	0
200 mg/kg	0
400 mg/kg	0
Phenobarbitone 30 mg/kg	100

CHAPTER FIVE

5.1 DISCUSSION

The hydro-methanol leaf extract of *I. trichantha* produced a distinct dose-related anticonvulsant effect in the experimental seizure model. Animals in the control group developed seizures quickly, and these episodes were brief, indicating the absence of protection. Administration of 100 and 200 mg/kg of the extract produced only a mild prolongation in seizure onset with limited protective influence. In contrast, treatment with 400 mg/kg resulted in a pronounced delay in the initiation of seizures and a substantial reduction in their duration. The magnitude of this effect closely resembled that of diazepam, a standard reference anticonvulsant, implying that the extract harbors bioactive constituents capable of modulating neuronal excitability and suppressing abnormal electrical discharges in the brain (Li *et al*. 2024).

The hydro-methanol leaf extract of *Icacina trichantha* showed noticeable anticonvulsant activity in the pentylenetetrazole (PTZ) model, but not in the maximal electroshock (MES) model. These contrasting results are biologically reasonable, since the two experimental models target distinct underlying mechanisms of seizure generation.” PTZ induces clonic and myoclonic seizures mainly by blocking GABA receptor–mediated inhibition, thereby increasing neuronal excitability. When a compound prevents seizures in this model, it is generally interpreted as evidence that it may strengthen GABAergic signaling, influence chloride channel conductance, or otherwise elevate the seizure threshold. This test is also responsive to agents with antioxidant or neuroprotective actions that can dampen the excitotoxic processes initiated by PTZ (Nasehi *et al*., 2021).

The MES model simulates tonic–clonic seizures triggered by an external electrical stimulus, and it is especially useful for identifying compounds that block seizure propagation by stabilizing voltage-gated sodium channels or suppressing high-frequency neuronal firing (Loscher, 2022).

The lack of protection in the MES model suggests that the extract does not effectively interfere with the ionic processes required to block electrically induced tonic seizures. The response profile observed here showing activity in the PTZ model but not in MES implies that the bioactive compounds in *Icacina trichantha* are more likely to act on GABAergic or related neurochemical pathways rather than on sodium channel–dependent processes. Comparable patterns have been documented with other plant extracts, where crude fractions protected against PTZ-induced seizures but showed no effect in MES until further purification revealed sodium channel–blocking constituents (Belayneh *et al*., 2021).

Phytochemical screening of the extract showed the presence of flavonoids, phenolics, saponins, terpenoids, tannins, alkaloids, carbohydrates, and sugars, while proteins were not detected. These phytochemical groups can raise the seizure threshold against convulsants that act through GABA antagonism, but they do not necessarily stabilize neuronal membranes under conditions of intense electroshock. Dosage may also be a contributing factor, as the levels effective in the PTZ model might have been too low for the MES test, which typically requires higher doses or more purified compounds with potent sodium channel–blocking activity (Belayneh *et al.,* 2021).

The results obtained from the PTZ model may also relate to the role of oxidative stress. PTZ-induced seizures are closely linked to excessive production of reactive oxygen species and subsequent neuronal injury, and plant extracts with antioxidant properties often counter this by delaying seizure onset or reducing their severity (Rajabian *et al*., 2021).

MES-induced seizures arise almost immediately after the electrical stimulus, providing little opportunity for antioxidant defenses to confer meaningful protection. the findings. *I. trichantha* extract may be beneficial against seizure types associated with impaired

GABAergic signaling or oxidative stress, but it is less likely to provide protection in tonic–clonic seizures that depend on sodium channel blockade. Future studies should focus on fractionating the extract to isolate constituents active in the MES model, evaluating antioxidant enzyme activity and GABA levels following treatment, and conducting pharmacokinetic analyses to confirm central nervous system exposure.

These secondary metabolites are widely recognized for their ability to support brain health and contribute to seizure control through multiple pathways. Flavonoids and phenolic compounds act as powerful antioxidants, protecting neurons from oxidative stress that is often associated with seizure onset and progression. Alkaloids are particularly important for their effects on the central nervous system, as many interact with neurotransmitter systems such as GABA, a major inhibitory pathway that helps prevent abnormal neuronal firing. Alkaloids represent another key class of compounds identified in the extract. They are well recognized for their influence on the central nervous system, with several known to interact directly with major neurotransmitter systems, including GABA and glutamate (Zhang *et al.,* 2023). Saponins and terpenoids are also reported to possess sedative and anticonvulsant effects, likely by enhancing inhibitory signaling in the brain (Zhang *et al.,* 2024).

A number of flavonoids have been shown to interact with GABA receptors—the same receptor sites targeted by benzodiazepines like diazepam—allowing them to produce anticonvulsant and anxiety-reducing effects. Terpenoids have been noted for their sedative and anticonvulsant properties, effects that are thought to arise from their ability to influence ion channel activity and interact with neurotransmitter receptors. Saponins are a diverse group of naturally occurring compounds that have been linked to both calming and protective effects on the nervous system. Their neuroprotective role is thought to stem from their ability to stabilize neuronal membranes, reduce excitotoxicity, and modulate signaling pathways involved in cell survival. In the context of seizure control, these properties are particularly important because they may help limit excessive neuronal firing and protect brain cells from damage caused by oxidative stress and recurrent seizures. Additionally, saponins have been suggested to enhance inhibitory neurotransmission, which contributes to their sedative and anticonvulsant potential. By acting through these multiple mechanisms, saponins strengthen the overall anticonvulsant activity of the extract and provide further evidence of its therapeutic promise in managing epilepsy and related neurological disorders (Tabassum *et al.,* 2023). Tannins, with their additional antioxidant properties, may further reinforce neuronal stability and resilience under conditions of stress (Obese *et al.,* 2021).

Findings indicate that the anticonvulsant activity observed in the study can be attributed to the bioactive constituents present in the extract. At a dose of 400 mg/kg, the extract produced effects closely resembling those of diazepam, suggesting that *I. trichantha* holds considerable potential as a natural source of seizure-modulating agents. Its beneficial action is most likely achieved through a combination of antioxidant protection and interactions with neurotransmitter pathways, particularly the GABAergic system, which plays a central role in suppressing excessive neuronal excitation. These results not only validate the plant's traditional application in seizure management but also underscore its promise as a candidate for further research and possible development into therapeutic options for epilepsy and related neurological conditions.

5.2 CONCLUSION

The hydro-methanol leaf extract of *I. trichantha* at 400mg/kg used in this study demonstrated a distinct anticonvulsant effect, which indicates that the hydromethanol leaf extract of *Icacina trichantha* reduced PTZ-induced seizures but the plant has no significant effect in the induced MES seizures.

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