**Article History**
Received: November 9, 2024
Revised: January 7, 2025
Accepted: February 22, 2025
Published: July 09, 2025

Unicross journal of Science, Engineering & Technology (UJOSET), Journal of the Faculty of Physical Sciences, University of Cross River State. Vol 1, Issue 1, 2025.

**Article Type:** Research

# Toxicological Studies of Aqueous Extract of *Psychotria Microphylla* on Serum Total Protein, Albumin, Bilirubin Creatinine & Urea of Albino Rats

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## Abstract

*Psychotria microphylla* Elmer is one of the plants found in the Eastern part of Nigeria used for fishing and prevention of insects from destroying crops and vegetables. It grows well under moist forest, along river and stream bounds. Based on its natural habitat, it is locally known as Akwukwo iyi or Oye (stream plant) in Afikpo South Local Government Area of Ebonyi State Nigeria. This present study was carried out to evaluate the toxicological effects of aqueous extract of *Psychotria microphylla* on some biochemical parameters in albino rats. A total of 25 albino rats weighing between 110-180g were used for this sub-acute toxicity study. The rats were randomly divided into five groups labeled as group A, B, C, D and E with each group containing 5 rats each. Group A, which is the control, was given distilled water only while B to E received 300mg/kg, 500mg/kg, 1000mg/kg and 2000mg/kg respectively of sub-acute doses of aqueous extract of *P. Micropyilla* for 14days. The animals were sacrificed after being anaesthetized with ketamine and blood sample collected via ocular puncture into sample bottles. It was further centrifuge, and the serum was collected for further biochemical analysis. The total protein, albumin, bilirubin, creatinin, and urea were estimated. The result of the effect of *P. microphylla* on total protein, albumin, and biluribin are presented, the differences observed in serum albumin level was not significant ($P>0.05$) when compared with normal control. However, significant increase ($p<0.05$) was observed in total protein and bilirubin level when compared with the normal control. *On the other hand, the result of the effect of P.microphilla on Creatinine, and Urea shows no significant increase ($p>0.05$) in the test group when compared to the control.* This *result therefore indicates that t*he plant may therefore be considered safe at lower doses. However, high concentration of it should be avoided as it may interfere with normal liver function.

**Keywords:** Toxicology, Aqueous extract, Biochemical parameters, *Psychotria microphylla* , Albino rats.

Cite this article as: Ukam E.E, Orji O.U, & Oko F.N. (2025). Toxicological Studies of Equeous Extract of *Psychotria Microphylla* on Serum Total Protein, Albumin, Bilirubin Creatinine & Urea of Albino Rats. *Unicross journal of Science, Engineering & Technology* ***1****(1) pp: 15 -*

## Introduction

Plant poisons have been widely used by traditional societies all over the world as a means of catching fish. Plant extracts are referred to as botanicals and when poisonous to fish are called piscicides (Obomanu *et al.*, 2007), such piscicidal plants contain different active ingredients known as alkaloids, resin, tannin, saponin, nicotine, diosgenin (Obomanu *et al.*, 2005). However, these active ingredients are toxic to fish at high concentrations and wear off within a short time (Burkill, 1985). The botanical piscicides are believed to be more environmentally friendly because they are easily biodegradable (Adewumi, 1991). And leaves no residues in the environment. Also, the use of botanicals has been found to aid fish cropping greatly as it saves time for fishing and increases easy handling. Many plants have been used for fishing, several of which include *Albizia chevalieri*, *Aesculus californica, Chlorogalum pomeridianum, Olax, Careya coccinea, Cleistanthus collinus, Lannea coromandelica, Costus speciosus Lepidagathis alopecueoide* (Michael, 2008; Ukam *et al*; 2024). Many scientific investigators have authenticated a variety of studies and some still undergoing documentation regarding alteration in biochemical components as response to environmental stress and induced toxicant on animals; examples can be seen in the literature report made by some scientific scholars (Saidu *et al.,* 2009; Orji *et al.,* 2014).

*Psychotria microphylla* Elmer is one of the plants found in the Eastern part of Nigeria used for fishing and prevention of insects from destroying crops and vegetables. It grows well under moist forest, along river and stream bounds. Based on its natural habitat, it is locally known as Akwukwo iyi or Oye (stream plant) in Afikpo South Local Government Area of Ebonyi State Nigeria. The local fishermen dig the ground and the whole plant is thoroughly pounded in the hole and pulverized plant materials are applied into the water body where fishing is intended. The fish are harvested once they are stunned and will continue till the next day. They use the extract to spray on vegetable crops against insect infection. Orji *et al.*, (2014) have reported that the plant is very toxic to fish. There is no report on the toxicity of this plant on human or any other animal model. *psychotria microphylla* has also been used in our rural areas where health facilities are limited to treat female reproductive system infections, Bronchitis, Gastrointestinal disturbances, Skin infection, Fever, Headache, Earache, and Eye disturbances (Ibiam *et al* 2014). The leaves, seeds and the roots of this plant also possess various medicinal properties due to the presence of several phytochemical components which are the major bioactive constituents in ethnomedicinal plants (Sangeetha *et al.,* 2013; orji *et al.,* 2014). There is no doubt that Africa is blessed with abundant plants whose medicinal potentials are yet to be tapped. Apart from a sketchy description of the plant and a scientific report made by (Orji *et al.,* 2014) where they indicated that sixteen chemicals identified from both methanol and n-hexane extracts of *P. microphylla* leaves by GC-MSC were found to have various biological activities ranging from therapeutic effects (anti-microbial, anti-cancer, antiandrogenic,

hypocholesterolemic) to toxic effects to aquatic organism, no further work has been reported on its acute toxicity features.

## 2. Materials & Methods

I. Plant Sample Collection and Preparation:

The fresh samples of *psychotria microphilla* were collected from the wild at Afikpo South L.G.A of Ebonyi State, Southeastern Nigeria. The plant was identified and authenticated by Mr. Ozioko of the International Bioresources and Research Centre, Nsukka, Nigeria.

II. Preparation of Lyophilized Aqueous Extract

The samples were washed and shaded, dried and then grounded into fine powder and sifted using 0.25mm sieve. The leaf powder thus obtained was soaked in 1litre of distilled water for 48hours. The stored mixture was filtered through sterile quaze and the filtrate was collected by hand pressure. Further, it was centrifuged at 3000rpm for 10mins and the supernatant filtered through seitz filter (0.2mm). The filtered extract was poured into a special lyophilizing flask. The flask was connected to a vacuum pump and evacuated till dryness and the lyophilized powder used for evaluating the effects of *P. Microphilla* in albino rats.

III. Ethics for the use of laboratory animals

The study adhered to the guidelines for the care and use of laboratory animals as outlined by the Indian Council of Medical Research in 2001 and was approved by the Ethical Committee of Biological Sciences, Ebonyi State University Abakaliki, Nigeria. with the approval number EBSU/FBS/EC/3470.

IV. Experimental Animals

A total of 25 albino rats weighing between 110-180g were used for this sub-acute toxicity study. The rats were obtained from disease free stock of the animal house in Zoology Department, University of Nigeria Nsukka. And brought to the Department of Biochemistry animal house, Ebonyi State University. These rats were allowed to acclimatize for one week under standard condition before the commencement of the experiment. The rats were fed with a normal rat palette. All the rats in both the test and control groups were allowed free access to food and clean water throughout the experimental period. Good hygiene was maintained by constant cleaning and removal of feces and spilling feed from wooden cages daily.

V. Experimental Design

A total of 25 albino rats weighing between 110-180g were used for this sub-acute toxicity study. These 25 rats were further divided into five groups labeled group A, B, C, D and E with each group containing 5 rats each. Group A, which is the control, was given distilled water only while B to E receives the following sub-acute doses of *P. micropyilla* aqueous extract: 300mg/kg, 500mg/kg, 1000mg/kg and 2000mg/kg respectively for 14days. The animals were sacrificed after being anaesthetized with ketamine and blood sample collected via ocular puncture into sample bottles. It was further centrifuge, and the serum was collected for further biochemical analysis.

VI. Estimation of Serum Total Protein Albumin:

Serum levels of total protein and albumin were estimated using the method of Killingsworth (1979) as outline by RANDOX

VII. Estimation of total bilirubin

Total bilirubin was estimated by Spectrophotometric method as described by Nuhu and Aliyu (2008). As outline by randox.

VIII. Determination of the concentration of serum urea and creatinine

The concentration of serum urea and creatinine was determined using the Tietz method (1994) as outlined in the Randox kit.

IX. Statistical Analysis

The data obtained were analyzed using Statistical Package of Social Sciences (SPSS) version 23.0 and the results expressed as mean ± standard error of mean. Significant differences of the result were established by one-way ANOVA and the acceptance level of significance was $p < 0.05$ for all the results.

## 3. Results and Discussion

Many plant poisons have been used in harvesting fish locally in Ebonyi state, Nigeria amongst which is psychotria microphilla which is generally, considered piscicidal because of its toxic potentials to fish. However, there has not been documentation on the effect of this plant on some biochemical parameters of albino rats. It becomes pertinent to determine whether this piscicidal plant could have toxic effect on liver and kidney function parameters of albino rats. The liver being the primary organ for detoxification, distribution and the kidney an organ for excretion,was assessed for the effect of *P. microphylla.*

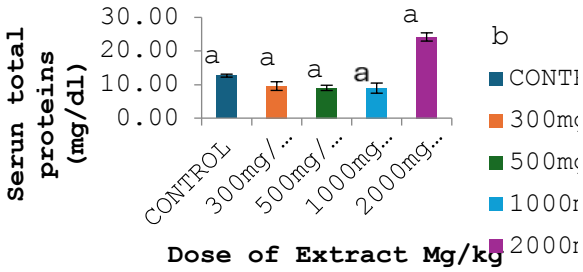


Figure 1 The effect of *Psychotria microphylla* on the serum total protein of albino rat. Data are presented as mean ± standard deviation. Bars with different letters are statistically significant ($P<0.05$).

The results of the effect of *P. microphylla* on total protein, albumin, and biluribin are presented in figure 1, 2 and 3. Our result *showed significant increase ($p < 0.05$) in levels of total protein and total bilirubin respectively in the serum of albino rat.* The differences observed in the mean values of serum albumin level in the test group were not significant ($P>0.05$) when compared to the normal control. However, Significant increase ($p<0.05$) was observed in the result of total protein especially in the group that received the highest dose (2000mg/kg) when compared to both normal control and the groups that received 300mg/kg, 500mg/kg and 1000mg/kg. The result of bilirubin on the other hand showed a dose dependent increase ($p < 0.05$) in the test groups, when compared to the normal control. Although the increase in the mean value of the group that was administered 300mg/kg was not significant ($P>0.05$) when compared with the control.

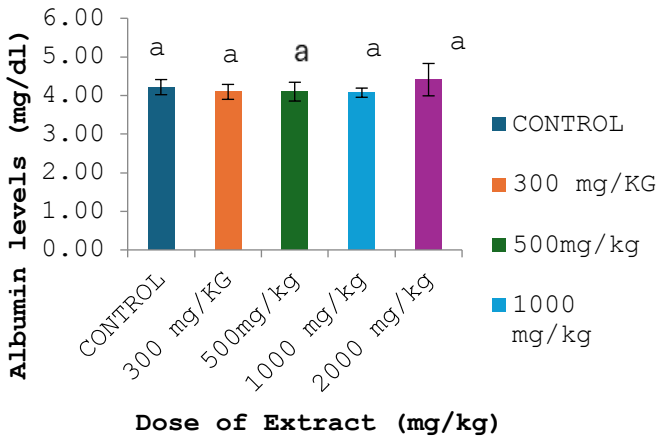


Figure 2 The effect of psychotria microphylla on the Serum albumim of albino rat. Data are presented as mean ± standard deviation. Bars with the same letters are not statistically significant (P > 0.05).

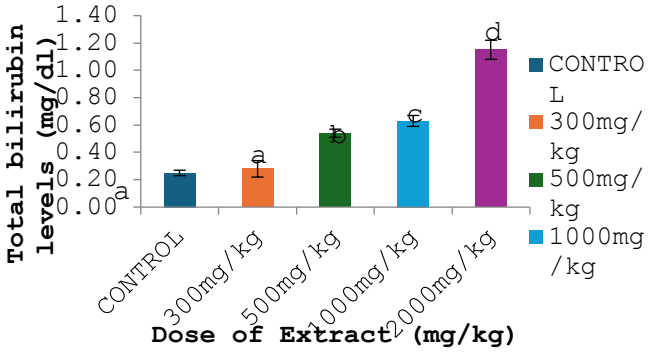


*Figure 3 The effect of Psychotria microphylla on the serum total bilirubin of albino rat. Data are presented as mean±standard deviation. Bars with different letters are statistically significant (P < 0.05).*

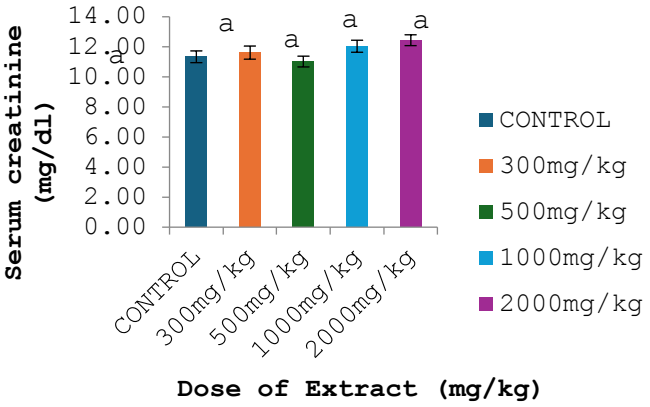


Figure 4 The effect of *P. microphylla* on the serum Creatinine of albino rat. Data are presented as mean ± standard deviation. Bars with the same letters are not statistically significant (P > 0.05).

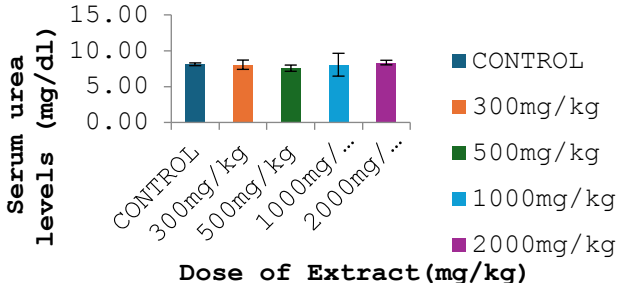


Figure 5 The effect of *P. microphylla* on the Serum Urea level of albino rat. Data are presented as mean±standard deviation. Bars with the same letters are not statistically significant (P > 0.05).

This is like the level of total bilirubin levels in serum of albino rats as reported in work *of* Toxicity Studies of the Crude Aqueous Root Extract of Albizzia chevalieri Harms in Albino Rats (Saidu *et al.,* 2010). *Also,* Duru *et al.,* 2012 made another report on Toxic effect of Carica papaya bark on body weight, hematology, and some biochemical parameters were revealed that direct and total bilirubin of their study increased insignificantly (p > 0.05) in test rats when compared to those of the control rats. This is a somewhat kind of confirmation of hepatic damage since, one of the primary roles of liver is detoxification. Bilirubin is a product of normal haem catabolism, and the liver removes it by conjugating it with glucuronic acid and makes it more water soluble and can be easily removed. Therefore, the elevated levels of bilirubin from our work are an indication of hepatic damage.

Similarly, an abnormally high total protein is indicative of a chronic or ongoing inflammatory disorder mainly of the liver, kidneys or digestive system, but it can also be the result of an inflammatory disorder elsewhere in the body. Diseases such as viral hepatitis or the human immunodeficiency virus or HIV will also cause an elevated total protein level. A high albumin-to-globulin ratio often suggests reduced production of immunoglobulins, which occurs in genetic disorders and leukemia (Buritis 1997). In general, total protein tests examine the amount and ratio of albumin and globulin, which are molecules made of proteins.

In contrast, results in figures 2,4 and 5 show no significant increase ($P > 0.05$) in Serum albumin, creatinine and urea respectively. The results of the effect of *P.microphilla* on Creatinine and Urea are presented in figure 4 and 5 above .The result however shows no significant increase ($p > 0.05$) in the test groups when compared to the normal control. Although there was a slight decrease in the mean value for the group that received 500mg/kg in both creatinine and urea level, however, the decrease was not significant when compared their respective control group. The result however is in accordance with the work done by Obidah, *et al.* 2014, that made a more recent report on a scientific documentation on the effects of Erythrina senegalensis aqueous leaf extract in rats, where they made an observation that the administration of the extract at 0, 50, 150 and 300mg/kg for 4weeks had no effect on rat serum total albumin, blood urea nitrogen and creatinine, they suggests that Erythrina senegalensis aqueous leaf extract has low toxicity in kidney cells, especially when administered orally. In another recent report made by Ukam *et al*; 2024, showed that the administration of N-hexane fraction of *lepidagathis alopecuroides* improved creatinine and urea level in alloxan induced diabetes in rat. The above result which is similar to our work therefore suggests that since the liver and kidney function parameters assessed did not show many alterations except at very high doses, the plant may be considered safe at lower doses.

## Conclusion

The above results showed that the mean values of total proteins, albumin, urea and creatinine were not significant ($P > 0.05$). There was however a significant increase in total bilirubin levels in dose dependent manner. The liver and kidney function parameters assed did not show many alterations except at very high doses. The plant may therefore be considered safe at lower doses. However, high concentration of it should be avoided as it may interfere with normal liver function.

**Acknowledgement**: The authors acknowledge the contributions of Mr. Alfred Ozioko of Bioresource Development and Conservation Programme,Nsukka, Enugu State.

**Conflict of interest:** The authors declare that they have no conflict of interest.

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