Effects of Crude Aqueous Extracts of Pyrenacantha Staudtii and Trichilia Monadelpha on Visceral Organs of Albino Mice

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DOI: 10.36348/sjls.2020.v05i05.001 | Received: 27.04.2020 | Accepted: 07.05.2020 | Published: 12.05.2020

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Abstract

Pyrenacantha staudtii and Trichilia monadelpha are herbal plants used indiscriminately to treat malaria by local people in Nigeria. Hence the need to examine the effects of these plants on the visceral organs of mice in order to determine their safety use. Crude aqueous extracts of the plants were prepared. Different concentrations such as 500mg/Kg, 800mg/Kg, 1000mg/Kg, 2000mg/Kg and 4000mg/Kg of the extracts were separately administered to mice in different groups. After 24h post-administration of the extracts, the mice were sacrificed to examine the histology of their visceral organs. No mortality was recorded among the treated mice 24h post-administration of the extracts. There was no lesion on the lung and heart tissues of the treated mice. The extracts of P. staudtii caused necrosis in the liver and kidney tissues at high concentration. But no necrosis was observed in the liver and kidney tissues of mice administered with T. monadelpha extract. Generally, there were cellular infiltrations in the liver and kidney tissues of the mice administered with the extracts of both P. staudtii and T. monadelpha. No lesion was seen in the tissues of untreated mice. Both P. staudtii and T. monadelpha appeared to be toxic at high concentration. Indiscriminate use of these plants without standard measurement should be discouraged.

Keywords: Pyrenacantha staudtii, Trichilia monadelpha, toxicity, plant extract, histology, viscerals.

INTRODUCTION

Men learnt to exploit plants for medicine, almost as early as they cultivated them for food [1]. Traditional medicine has not only played a vital role in providing healing but has also contributed to the discovery of most pharmaceutically active substances in plants [2, 3] which have been used in the commercial production of drugs. Medicinal plant species also show diversity in their parts used for the treatment of diseases, with leaves, roots and barks being the most commonly harvested parts used in herbal medicine preparation [4]. Nigeria has rich flora diversity and many of the plant species are used by some local people for medicinal purposes. Some of the plant species are used for malaria treatment across all ethnic and cultural groups in the country [5].

Pyrenacantha staudtii and Trichilia monadelpha were among the herbal plants used for the management of malaria in Ekiti State, Nigeria [6]. Both P. staudtii and T. monadelpha have been reported to possess antimalarial properties [7, 8]. But these plants are used indiscriminately without any standardized measurement or dosage to treat malaria [6]. This necessitated the design of this study with the aim to determine the safety use of these herbal plants in antimalarial recipes among the local people in Nigeria. Observing the histological changes in the visceral organs of laboratory animals to a drug in an acute toxicity test is one of the criteria used to determine the safety use of drug in humans. Hence the need to examine the histology of the visceral organs of mice administered with the crude aqueous extracts of P. staudtii and T. monadelpha in an acute toxicity test.

METHODS AND MATERIALS

Collection and Identification of Plants

P. staudtii and T. monadelpha were collected from Ajowa farm at Ido-Ekiti in Ekiti State, Nigeria. The plants were identified and authenticated at the Herbarium of Forestry Research Institute of Nigeria (FRIN), Ibadan where voucher specimens were deposited with the following numbers: Pyrenacantha staudtii, Icanaeaceae (FHI No 108805) and Trichilia monadelpha, Meliaceae (FHI No 108844). The parts of the plants used for the study were the leaves of Pyrenacantha staudtii and stem bark of Trichilia
monadelpha in accordance with the way they are used by the local people [6].

Preparation of Crude Aqueous Extracts of the Plants

The parts of the plants (i.e. leaves and bark) were sorted to remove contaminants, dead matter or sand particles and they were air-dried inside a ventilated room before grounded into a coarse powder. The coarse powder (200g) was extracted using distilled water for 48 hours at room temperature. The extract was filtered to obtain a filtrate which was concentrated to dryness over a water bath [7], [8]. Appropriate concentrations from each of the plant extracts were made in water and used in the experiment.

Acute Toxicity Tests of Crude Aqueous Extracts of Pyrenacantha staudtii and Trichilia monadelpha in mice

Adult Swiss albino mice of average weight of 25g were divided into six groups for each experimental test of P. staudtii and T. monadelpha extracts. Each cage of the six groups consisted of five mice. The mice were labeled using dilute picric acid. The acute toxicity tests of the crude aqueous extracts of P. staudtii and T. monadelpha were separately carried out. The crude extract of the plant with the concentration of 500mg/Kg, 800mg/Kg, 1000mg/Kg, 2000mg/Kg and 4000mg/Kg body weight of mice were separately administered to the mice in the respective Group 1-5 via an oral route with the aid of a cannula. Mice in Group 6 which served as a control were given water as placebo. Animals were observed for 24 hours for any changes. After 24 hours of the administration of plant extracts, the animals were sacrificed to collect the visceral organs which were lung, heart, liver and kidney for histological examination as described by Gu et al. [9]. The organs were carefully removed and fixed in 10% neutral buffered formalin. Afterwards, the organs were gradually dehydrated in ethanol solution at different concentrations. They were immersed in xylol, and then embedded in paraffin. Serial sections, 5μm thick was prepared and stained with hematoxylin-eosin on a microscope slide.

RESULTS AND DISCUSSION

No mortality was recorded in all the mice after the oral administration of the crude aqueous extracts of both P. staudtii and T. monadelpha 24 hours post-administration even at the highest concentration of 4000mg/Kg body weight of mice. This indicates that the oral median lethal dose (LD₅₀) of the two plants extracts were greater than 4000mg/Kg body weight. This result was similar to the findings of Akuodor et al. [10] where no mortality was reported with mice administered with 5000mg/Kg of ethanolic extract of a plant, Verbena hastate. The LD₅₀ of Alstonia boonei, a plant commonly used in antimalarial recipes in Southwest Nigeria was also estimated to be ≥ 5000mg/Kg body weight [11]. Using LD₅₀ to ascertain the safety use of a testing plant extract in acute toxicity test may not be satisfactory enough to indicate that such plant extract is non-toxic.

The heart tissues of both the treated and the untreated animals (control) did not show any visible lesion. However, the lung tissues of the mice administered with 800mg/Kg of P. staudtii extract showed moderate congestion of the inter-alveolar septum while those administered with 1000, 2000 and 4000mg/Kg showed moderate interstitial congestion. But no lesion was seen on the lung tissues of mice administered with 5000mg/Kg of P. staudtii extract and the mice administered with various concentrations of T. monadelpha extract. The lung tissues of the untreated mice also did not show visible lesion. However, the effects of the two plant extracts on liver tissues were significant. At concentrations of 500 and 800mg/Kg, P. staudtii extract caused a mild periportal cellular infiltration by mononuclear cells. Also, the portal canal of the liver was moderately congested. The effect was however very significant at higher concentrations. At 1000mg/Kg, there was a diffuse hepatic degeneration and necrosis, with severe periportal cellular infiltration by mononuclear cells. At 2000 and 4000mg/kg, the liver tissues showed severe hepatic necrosis with severe portal congestion and periportal cellular infiltration by mononuclear cells (Figure 1).

![Fig-1: Micrograph of liver tissues of mice administered with P. staudtii extract. (a) Tissues show no lesion. (b) Arrows show the mild periportal cellular infiltration. (c) Arrows show severe hepatic necrosis with diffuse and periportal cellular infiltration by mononuclear cells](image)
In the case of *T. monadelpha*, the extract caused severe portal congestion and periportal cellular infiltration on the liver tissues by mononuclear cells even at 800mg/Kg (Figure 2). There was moderate portal congestion and periportal cellular infiltration by mononuclear cells in the liver tissues of the mice administered with 500mg/Kg. However, unlike the extract of *P. staudtii*, the extract of *T. monadelpha* did not cause necrosis at any concentration even at 4000mg/Kg body weight of the experimental animals. No lesion was seen on the liver tissues of the untreated animals.

The kidney tissues of the mice administered with 4000mg/Kg of *P. staudtii* extract showed multiple foci of interstitial cellular infiltration, some tubules are necrotic while those of 2000mg/Kg showed severe cortical congestion and severe interstitial cellular infiltration. Mice that were administered with 1000mg/Kg only showed moderate renal cortical congestion and no visible lesions were seen in the mice administered with 800mg/Kg, 500mg/Kg and the untreated mice (Figure 3). However, at concentration of 4000mg/Kg of *T. monadelpha* extract, the kidney tissues only showed a mild renal cortical congestion (Figure 3). But no visible lesion was seen in the kidney tissues of the mice administered with 500, 1000 and 2000mg/Kg body weight of mice.

![Fig-2: Micrograph of liver tissues of mice administered with *T. monadelpha* extract](image)

(a) Tissues show no lesion in the untreated mice.
(b) Arrows show moderate portal congestion and periportal cellular infiltration by mononuclear cells.
(c) Arrows show severe portal congestion and periportal cellular infiltration by mononuclear cells

![Fig-3: Micrograph of kidney tissues of mice administered with 4000mg/Kg of extracts of *T. monadelpha*, *P. staudtii* and the untreated mice](image)

(a) Arrows show mild renal cortical congestion.
(b) Arrows show multiple foci of interstitial cellular infiltration.
(c) Tissues show no lesion in the untreated mice

Toxicity studies are considered very important in drug development while considering the indiscriminately use of herbal medicines by local people without due consideration to the harmful effects that could possibly associate with the use of such herbs [12, 13]. After 24 hours of treatment with the crude aqueous extracts of *P. staudtii* leaves, there were treatment-related changes in the histological structure of the liver and the kidney tissues between the control and the treated groups. Even though that there was no significant damage in the heart and lung tissues of the experimental animals, the hepatic degeneration, severe portal congestion and necrosis, with severe periportal cellular infiltration observed in the liver tissues of
animals administered with *P. staudtii* extract suggest the toxicity properties of the plant at a high concentration. The infiltration by mononuclear cells might be due to a response elicited by the immune system of the animals. This caused the inflammation of the liver otherwise referred to as hepatitis. Drug-induced allergic hepatitis was thought to be mediated by type IV immunological reactions [14].

The fact that some cells of the liver were necrotic showed the toxic nature of the plant extract at a high concentration. Phytochemical analysis of the ethanol extract of *P. staudtii* leaves showed the presence of alkaloids (benzylisoquinoline), glycosides, saponins, carbohydrate, tannins, flavonoids and resins [15, 16]. Detoxification of these compounds by liver at a high concentration such as 4000mg/Kg might have resulted to the inflammation observed. This is because chemicals absorbed following ingestion pass almost exclusively into the hepatic portal vein and are transported to the liver for detoxification. The kidney tissues were also affected showing cellular infiltration still confirming the toxic nature of this plant at a high concentration. Indiscriminate use of this plant for the treatment of malaria by local people without any standard measurement as reported [6] is therefore unacceptable.

However, the aqueous extract of *T. monadelpha* appeared to be less toxic. The histology of the heart and lung tissues showed no lesion to suggest any toxic effect on these tissues even at a high concentration of 4000mg/Kg. Although, lesions were observed on the liver and kidney tissues of the experimental animals treated with *T. monadelpha* at concentration of 4000mg/Kg, but there was no necrosis as compared to *P. staudtii*. A study carried out with *T. monadelpha* on the sperm indices and testosterone level of male albino rats showed the toxic effect but rather promoted the mobility and viability of the sperms [17].

**CONCLUSION**

The extract of *P. staudtii* appeared to be more toxic than *T. monadelpha*. Use of these herbal plants by humans need to be standardized. Although, *T. monadelpha* appeared to be safer than *P. staudtii* but indiscriminate consumption of these herbal plants by the local people should be discouraged.

**ACKNOWLEDGEMENTS**

The author thanks Dr. O. O. Aina of Department of Veterinary Anatomy, University of Ibadan, and Ibadan, Nigeria for interpreting the histology results of the animal tissues.

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