

HISTOLOGICAL EFFECTS OF LONG TERM ADMINISTRATION OF PILIOSTIGMA THONNINGII STEM BARK ON THE LIVER OF ADULT WISTAR RATS

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ABSTRACT

The histological effects of long term administration of *piliostigma thonningii* commonly used for the treatment of dysentery, fever, respiratory ailments, snake bites, hookworm and skin infections on the liver of adult wistar rats was carefully investigated. The rats of both sexes (N=21), with an average weight of 180g were randomly assigned into two test groups (n_1 & $n_2=14$) and control group ($n_3=7$). Animals in the test group A received the extract of *Piliostigma thonningii* stem barks at a single dose of 200mg/kg body weight daily, animals in test group B received a single dose of 400mg/kg body weight daily for twenty eight days through the orogastric tube administration while the control animals received equal volume of distilled water through the same route and for the same period. The rats were fed with grower's mash obtained from Edo Feeds and Flour Mill Limited, Ewu, Edo state, Nigeria. The rats were sacrificed by cervical dislocation on the twenty nine day of the experiment and the liver was carefully dissected out and quickly fixed in 10% formal saline for further routine histological study. The findings indicated that the liver in the test groups (groups A&B) showed some level of distortion and disruption of the cytoarchitecture with some marked congestion of blood mainly at the central vein of the liver. The presence of perivascular polymorph nuclei cells and congestion of blood in the sinusoidal space of the liver was also observed in the tested groups with that of group B more marked. Long term administration of extract of *Piliostigma thonningii* stem barks may therefore have an adverse effect on the liver of adult wistar rats.

Keywords: *Piliostigma thonningii*, liver, Wistar rats.

INTRODUCTION

Herbal medicines are one type of complementary and alternative medicine whose use has dramatically increased over the past decade, growing from 2.5-12% in community survey (Roy-Byrne et al., 2005). A variety of plants or materials derived from plants have been used for the prevention and treatment of diseases virtually in all cultures. Herbs have been used as sources of food and medicinal purposes for centuries and this knowledge have been passed from one generation to another (Adedapo et al., 2005). Pravin, (2006) reported that about 70% of the human population is dependent (wholly or partially) on plant-based medicines and the World Health

Organization (WHO) estimated that 80% of the world population presently uses herbal medicine for some aspect of primary health care (WHO, 2008). The potential of herbal medicines and medicinal plant research results in health care is no longer in doubt, having gained recognition in several nations of the world and the World Health Organization (WHO).

Piliostigma thonningii schum is an angiospermae in the subclass os dicotyledonae in the order of Rosales, suborder to leguminosae and the family, Caesalpiniodeae. It is a shrub like tree with pinnate or bipinnate leaves whose

flower are showy, slightly irregular with five petals or fewer (Akobundu, 1986). In Nigeria, it bears such local names depending on the medicinal use in that vicinity. In Ibo, it is called 'okpo atu', Yoruba, 'Abefe'; Hausa, 'kalgo'; Nupe, 'Baffin'; Gwari, 'Tawin'; while the Senegal called it Niama or Niamia and the Togolese called it 'Ewe kio'. The seeds of the *Piliostigma thonningii* have been reported to be eaten by African antelope and elephant while farmers in the lower savannah region used the seed as fodder for cattle during winter months (Djuma, 2003).

Different parts of *Piliostigma thonningii* have been described traditionally (Djuma, 2003). Its root and twig have been used for the treatment of: dysentery, fever, wound infection, jaundice, chicken pox, respiratory ailments, snake bites, hookworm and skin diseases. Various preparations of the plant have been used to treat fevers and bacteria infections, to arrest bleeding, as a laxative, anthelmintic and as anti-inflammatory agents (Igoli et al., 2002; Togola et al., 2005; Igoli et al., 2005). Some pharmacological properties of *Piliostigma thonningii* such as its antibacterial has been established

(Ogundaini, 1999; Ibewuiké et al., 1997); anthelmintic (Fakae et al., 2000) and anti-inflammatory properties (Ibewuiké et al., 1997).

The liver is a large glandular organ located in the abdominal region. It is involved in the detoxification of most substances (Burkitt and Health, 1993). It is lined by endothelial cells that contain the kupffer cells, which are typical macrophages (Junqueira *et al.*, 1998). In addition to the endothelial cells, the sinusoids contain phagocytic cells known as the kuffer cells which are found in the surface of the endothelial cells (Junqueira *et al.*, 1998). In humans, the majority of drugs administered are eliminated by a combination of hepatic metabolism and renal excretion (Katzung, 1998). Since the liver is susceptible to injuries particularly in situations of toxicity, it would therefore be worthwhile to examine the effects of *Piliostigma thonningii* stem bark on the liver of adult wistar rats. The purpose of this study is to evaluate the possible effects of *Piliostigma thonningii* stem bark on the microanatomy of the liver of adult wistar rats.

MATERIALS AND METHODS

Animals care ethics

The School of Basic Medical Sciences, University of Benin granted approval before the work began. Twenty one adult wistar rats of both sexes with average weight of 180g were randomly assigned into three groups: A, B & C of seven rats each in a group. Group A & B served as test groups (n_1 & $n_2=14$) while group C ($n_3=7$) served as the control group. The rats were obtained and maintained in the Animal Holding of the Department of Anatomy, School of

Basic Medical Sciences, University of Benin, Benin city, Edo State, Nigeria. The animals were caged in wooden cages with raised wire floors based on their sex to avoid pregnancy, fed *ad libitum* with grower's mash obtained from Edo Feeds and Flour Mill Limited, Ewu, Edo State, Nigeria.

Preparation and administration of Piliostigma Thonningii stem bark.

Piliostigma thonningii stem barks were obtain within the university of Benin

premises and taken to the Department of Pharmacognosy, Faculty of Pharmacy, University of Benin for proper identification. The stem barks were washed free of debris and dust particles and were air dried at ambient sun temperature for two weeks. The dried stem barks were reduced in size using wooden mortar and pestle. The samples were further pre-dried in the oven at 25°C for 5 hours. It was immediately pulverized using plant milling machine in the Department of Pharmacognosy. 100g of the sample was extracted using soxhlet apparatus and subjected to ethanolic extraction for 10 hours and later concentrated in -vacuums and transferred into suitable container for freeze dried ready for the experiment. All preparations were performed at the Department of Pharmacognosy, Faculty of Pharmacy, University of Benin, Edo state, Nigeria.

Treatment of animals

Animals in the test group A received the extract of *Piliostigma thonningii* stem barks at a single dose of 200mg/kg body weight daily, animals in group B

RESULTS

The photomicrograph of the liver in the control group showed normal histological features. The section indicated normal hexagonal boundary of the hepatic lobules been defined by portal tract and sparse collagen fibres (Figure 1). The microanatomy (H & E) of the tested liver showed some level of distortion and disruption of the

received a single dose of 400mg/kg body weight daily for twenty eight days through the orogastric tube administration while the control animals received equal volume of distilled water through the same route and for the same period. The rats were sacrificed by cervical dislocation on the twenty nine day of the experiment and the abdominal region was quickly opened and the liver dissected out and fixed in 10% formal saline for routine histological techniques.

Histological study

The tissues were dehydrated in an ascending grade of alcohol (ethanol), cleared in xylene and embedded in paraffin wax. Serial sections of 7 microns thick were obtained using a rotatory microtome. The deparaffused sections were stained routinely with haematoxyline and eosin (Drury *et al.*, 1976). Photomicrographs of the desired results were obtained using research photographic microscope in the Department of Anatomy, School of Basic Medical Sciences, University of Benin, Benin city, Edo State, Nigeria.

cytoarchitecture with some marked congestion of blood mainly at the central vein of the liver. The presence of perivascular polymorph nuclei cells and congestion of blood in the sinusoidal space of the liver was also observed in the tested groups with that of group B more marked (Figure 2 & 3,).

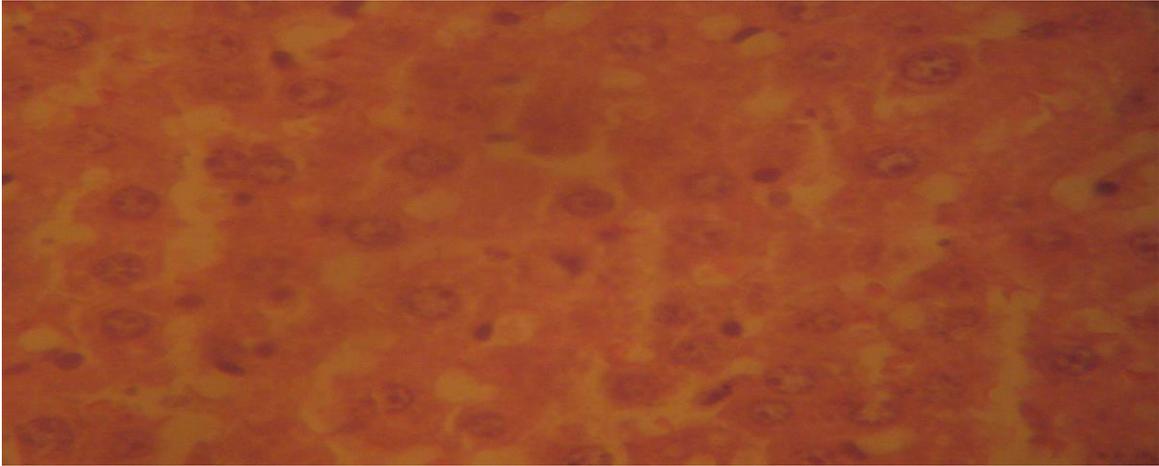


Plate 1: Control section of liver (H & E Method x400)

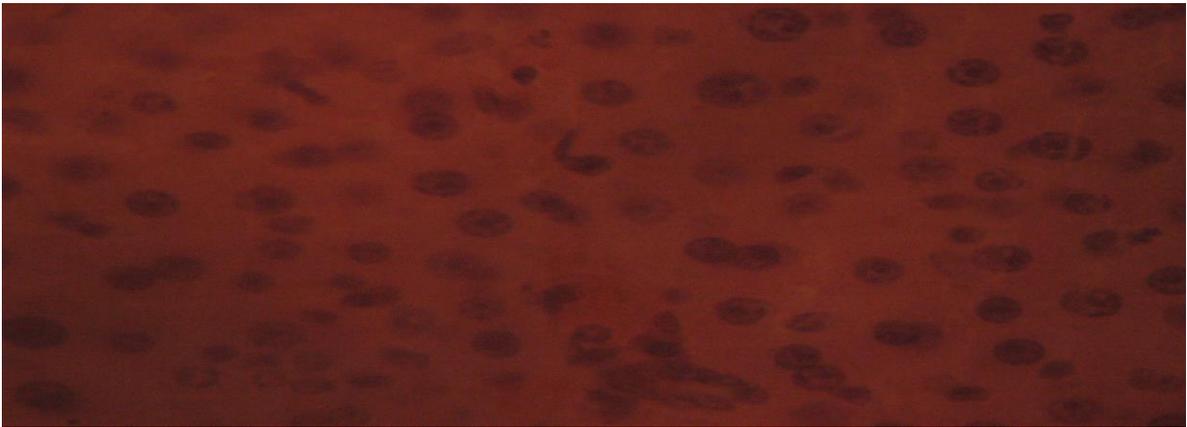


Plate 2: Treated section of group A liver (H & E Method x400)

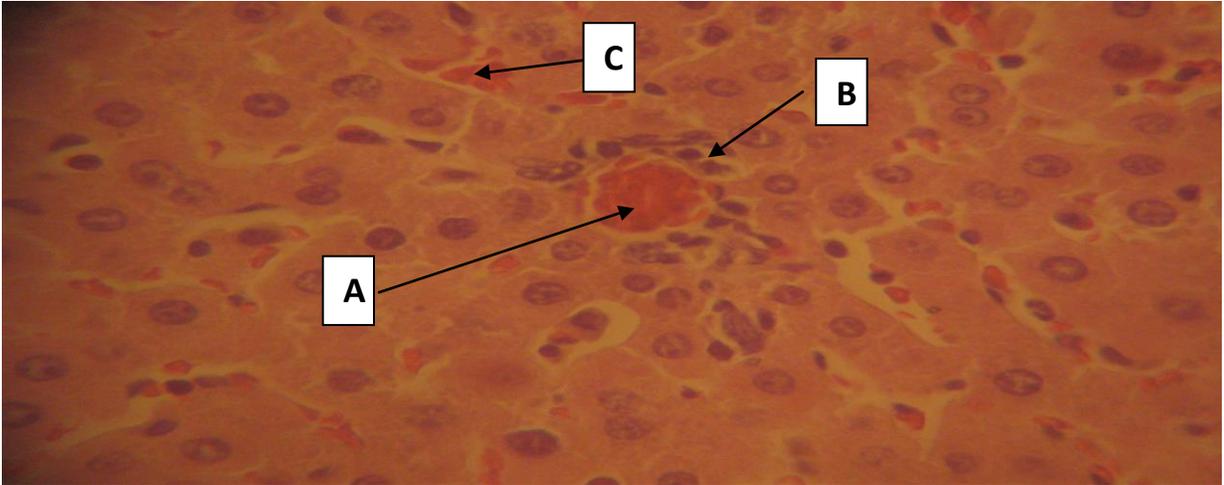


Plate 3: Treated section of group B liver showing A= congested blood in central vein, B perivascular polymorphonuclei cells, C = congested sinusoidal space. (H & E Method x400)

DISCUSSION

The findings indicated that the photomicrograph of the liver in the control group showed normal histological features. The section indicated normal hexagonal boundary of the hepatic lobules been defined by portal tract and sparse collagen fibres. The microanatomy (H & E) of the tested liver showed some level of distortion and disruption of the cytoarchitecture with some marked congestion of blood mainly at the central vein of the liver. There is the presence of perivascular polymorph nuclei cells and congestion of blood in the sinusoidal space of the liver was also observed in the tested groups with that of group B more marked.

The result obtained in this experiment may be probably due to the chronic administration of aqueous extract of *Piliostigma thonningii* stem bark on the tested liver. It appeared that chronic administration of aqueous extract of *Piliostigma thonningii* stem bark is not as harmless as generally believed. The distortion and disruption of the cytoarchitecture of the liver observed in this experiment may have been associated with the functional changes that could be detrimental to the health status of the animals. The observed changes in *Piliostigma thonningii* stem bark tested liver might be due to the cytotoxic effect of *Piliostigma thonningii* on the liver. Ischemia or pharmacologic disruption of cellular transporters can cause swelling of parenchyma of any organ. The pharmacologic disruption of the liver weights caused by *Piliostigma thonningii* stem bark extract is a cardinal feature of the results of this experiment. The obvious signs of distortion and disruption of the cytoarchitecture with some congestion of blood in the central vein of the tested liver in this

experiment may have been due to the cytotoxic effects of *Piliostigma thonningii* stem bark on the tested liver. These findings implicated *Piliostigma thonningii* as a possible precipitant of liver disease by causing congestion of blood in the microanatomy of the liver. Pathological or accidental cell death is regarded necrotic and could result from extrinsic insult to the cell as osmotic, thermal, toxic and traumatic effects (Farber *et al.*, 1981). The process of cellular necrosis involves disruption of membranes, as well as structural and functional integrity. Cellular necrosis is not induced by stimuli intrinsic to the cells as in programmed cell death, but by an abrupt environmental perturbation and departure from the normal physiological conditions (Martins *et al.*, 1978). In cellular necrosis the rate of progression depends on the severity of the environmental insults. The greater the severity of the insults, the more rapid the progression of neuronal injury (Ito *et al.*, 1975). The principle holds true for toxicological insult to the brain and other organs (Martins *et al.*, 1978). It may be inferred from the present study that higher dose and prolonged administration of extract of *Piliostigma thonningii* stem bark may result in increased toxic effects on the liver. The result obtained in this experiment is in consonance with the work carried out by Enaibe *et al.*, (2007) where it was reported that administration of *Turnera diffusa* to a matured wistar rats resulted in the distortion and disruption of the renal cortical structures, reduced size and number of the renal corpuscles and some degree of cellular necrosis in the histology of the kidney. In this experiment, *Piliostigma thonningii* may have acted as toxin to the cells of the liver thus resulting in the

distortion and disruption of the cytoarchitecture of the liver with some marked congestion of blood mainly at the central vein of the liver. The presence of perivascular polymorph nuclei cells and congestion of blood in the sinusoidal space of the liver was also observed in the tested groups with that of group B more marked.

This is a suggestive to the fact that prolong administration of aqueous extract of *Piliostigma thonningii* stem bark might be carcinogenic.

In conclusion, our findings indicated that long term administration of aqueous extract of *Piliostigma thonningii* stem bark resulted in some level of

distortion and disruption of the cytoarchitecture with some marked congestion of blood mainly at the central vein of the liver. The presence of perivascular polymorph nuclei cells and congestion of blood in the sinusoidal space of the liver was also observed in the tested groups with that of group B more marked. Long term administration of aqueous extract of *Piliostigma thonningii* stem bark may therefore have an adverse effect on the liver of adult wistar rats. It is recommended that further studies aimed at corroborating these observations be carried out.

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