Hepato-Protective Effect of Ethanol Leaf Extract of Bryophylum Pinnatum on Paracetamol Induce Hepatitis Albino Rats

1Ngobidi KC, 2Igbokwe GE, 3Ajayi AA, 4Otuchristian G, 5Omoboyowa DA, 6Adindu SC

1,3,4,5Science Laboratory Technology Department, School of Science and Technology Akanu Ibiam Federal Polytechnic Unwana, Afikpo Ebonyi State Nigeria

2,6Biochemistry Department, Bioscience Faculty Nnamdi Azikiwe University Awka, Anambra State Nigeria

ABSTRACT

The present study aimed at investigating the hepato protective activity of ethanolic leaf extract of Bryophylum pinnatum of albino rats. The 25 animals used in this study with average body weight of 200g was randomized into 5 groups. Liver damage was induced by oral administration of paracetamol at 100mg/kg body weight once for 7 consecutive days. The extract of the damage was studied by assessing biochemical parameters such as Alanine Transaminase (ALT), Gamma Glutamyl Transferase (GGT), Alkaline Phosphatase (ALT), Total Protein, Albumin, Superoxide Dismutase (SOD), Glutathione Peroxidase (GPx) and Bilirubin using spectrophotometer method. The ethanolic leaf extract of Bryophylum pinnatum (100mg, 200mg and 300mg) were administered orally to the animals induced hepatotoxicity for 7 consecutive days. The result obtained showed significant reduction in serum hepatic enzymes. There was a significant increase (P < 0.05) in the serum total protein, albumin, GPx and SOD. The significant increase of hepatic enzyme and corresponding increase in total protein, albumin, GPx and SOD showed the ability of the extract to restore liver damage caused by paracetamol toxicity and suggest it’s hepatoprotective and antioxidant. It is therefore concluded that ethanolic extract Bryophylum pinnatum has hepatoprotective effect.

INTRODUCTION

Liver is the largest organ in vertebrate body, and plays a major role in metabolic activities such detoxification and as such it is often exposed to maladies (disease or disorder) resulting in a number of clinical syndromes. Many chemicals, foods, drugs and infectious (parasitic, bacterial, viral or fungal) can cause verity of liver disease such as hepatitis, cirrhosis, liver cancer. Hepatitis is a medical condition defined by the inflammation of liver and characterized by the presence of inflammatory cells in the tissue of the organ, hepatitis is acute when it lasts less than six months and chronic when it persists larger. Acute hepatitis can be self-limiting (healing on its own), can progress to chronic hepatitis or rarely, can cause acute liver failure (Bernal et al., 2013). Paracetamol is one of the drug that can cause liver disease such as hepatitis, paracetamol toxicity is caused b excessive use or overdose of the analgesic drug called acetaminophen, the hepatotoxicity results not from paracetamol itself but from one of this metabolites, N-acetyl-p-benzoquione imine (NAPQI) which deplete cells in the liver, leading to liver failure (Raghavendren, et al., 2004).

Numerous phytomedieines has been found to be used for the prevention and treatment of liver disorder or hepatotoxicity example Bryophylum pinnatum is a medicinal plant that possess strong antioxidant activities which are flavonoids, polyphanols, alkaloids, saponins, vitamins (A,C,E & K), carotemoids, minerals, enzymes (glutathione peroxidase), lignins, xanthone and pigment. Due to it’s content, it could be used in the treatment of liver disease which are mainly caused by toxic chemicals or drugs (Pandey et al., 2011). The treatment is aimed to removing the paracetamol from the body and replacing glutathione. Hepatotoxicity caused by paracetamol overdose can be treated with silymarin which serves as an antidote that competes with acetaminophen at its receptors on the cellular membranes.

*Address for correspondence
ngobidikc909f@gmail.com
JUSTIFICATION OF THE STUDY
The justification of this study lies on the need for a more efficacious, cheap, readily available agent for the treatment of a dreadful acute liver toxicity.

AIM OF THE STUDY
To determine the hepatoprotective effect of *Bryophyllum pinnatum* on albino rats induced hepatitis with paracetamol.

OBJECTIVES OF THE STUDY
To determine the serum concentrations of GGT, ALP, ALT, direct and indirect bilirubin, albumin, total protein, to determine the serum levels of SOD and glutathione peroxidase.

MATERIALS AND METHOD
Collection of Plant Material
The leaves of *Bryophyllum pinnatum* was collected authenticated by a taxonomist

Processing of Plant Materials
The plant materials (leaves) were shopped and shade dried at room temperature for 2 weeks. Ground to powder, soaked for 48hours in 95% ethanol, sieved with cheese cloth and the suspension filtered, with whatman paper and was allowed to evaporate to constant weight. The solid extract was stored in an airtight container until used.

Animal Source
A total of 25 healthy adult albino rats weighing about 200 to 250g were used in the study and it was sourced from vertanary medicine department, university of Nigeria Nsukka. The animals was kept in the animal cage and maintained under natural conditions of 12 hours in light and dark cycle. The animals was feed with standard rat feed and portable water *ad libitum* and allowed to acclimatized to the laboratory condition. For 7 days prior to the commencement of the experiment. The experimental animals were handled according to the ethical guidelines suggested by the institutional animal ethics committee (IAEC), Biochemical Research.

Induction of Hepatitis
Hepatitis was induced by oral administration of a single dose of 1000my/kg of paracetamol.

Experiment Design
A total of 25 healthy adult albino rats was used in this study according to the design. The rats were randomized into five (5) groups of 5 animal each.

Group 1: Positive control (Vehicle)
Group 2: Standard drug (Silymarin)
Group 3: 200mg/kg of ethanolic extract of *Bryophyllum pinnatum* by oral administration for 7 days.
Group 4: 300mg/Kg of ethanolic extract of *Bryophyllum pinnatum* by oral administration for 7 days.
Group 5: 400mg/Kg of ethanolic extract of *Bryophyllum pinnatum* by oral administration for 7 days.

Animal Sample Collection
The animal was anesthetized with chloroform, at the end of the administration period, the animals sacrificed, their blood was collected, allowed to clots and spun at 4000rpm for 5 minutes. Sera were collected and finally stored between 0 to 4°C in a refrigerator for further use in biochemical analysis.
Biochemical Analysis


RESULTS

**Alkaline Phosphatase (ALP):** Induced and treated with 100mg/200mg/kg of *Bryophyllum pinnatum* showed no significant decrease compared to the normal control. Induced and treated with 300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with standard drugs.

Induced and treated with 100mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with 300mg/kg weight of *Bryophyllum pinnatum*. As shown fig. 1 below

**Alanine Transaminase (ALP):** Induced and treated with 100/200mg/kg body weight of *Bryophyllum pinnatum* showed no significant decrease when compared to the normal control.

Induced and treated with 300mg/kg body weight of *Bryophyllum pinnatum* showed no significant decrease compared to induced and treated with standard drug. As shown fig. 1 below

**Total Protein:** Induced and treated with 200mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to the normal control. Induced and treated with 300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to the normal control. Induced and treated with 200/300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with standard drug. As shown fig. 2 below

**Albumin:** Induced and treated with 100mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to normal control. Induced and treated with 200/300mg/kg body weight of *Bryophyllum pinnatum* showed no significant decrease compared to induced and treated with standard drugs.

Induced and treated with 200mg/kg body weight of *Bryophyllum pinnatum* showed no significant decrease compared to induced and treated with 300mg/kg body weight of Bryophyllum pinnatum. As shown fig. 2 below

**Direct Bilirubin:** Induced and treated with 100/200/300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to normal control. Induced and treated with 300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with standard drug. As shown fig. 2 below

**Glutathione peroxidase (GPx):** Induced and treated with 200mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to normal control. Induced and treated with 100/300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced to standard drug. Induced and treated with 100mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with 200mg/kg body weight. As shown fig. 3 below

**Superoxide dismutase (SOD):** Induced and treated with 100mg/kg body weight showed of *Bryophyllum pinnatum* showed significant decrease compared to normal control. Induced and treated with 200mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with 300mg/kg body weight. As shown fig. 4 below

**Gamma Glutamyl transferase (GGT):** Induced and treated with 100/200/300mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to normal control. Induced and treated with 200mg/kg body weight of *Bryophyllum pinnatum* showed significant decrease compared to induced and treated with 300mg/kg body weight. As shown fig. 1 below
Fig 1. Effect of ethanol leaf extract of B. pinnatum on activities of Liver marker enzymes of Paracetamol-induced Rats.

NCTRL : Normal Control
PISD : Paracetamol induced Rats treated with standard drug
PIBP 200 mg/kg : Paracetamol induced Rats treated with 100 mg/kg b. w of Ethanol extract of B. pinnatum
PIBP 300 mg/kg : Paracetamol induced Rats treated with 200 mg/kg b. w of Ethanol extract of B. pinnatum
PIBP 400 mg/kg : Paracetamol induced Rats treated with 300 mg/kg b. w of Ethanol extract of B. pinnatum

Fig 2. Effect of ethanol leaf extract of B. pinnatum on activities of Total protein, Albumin and Direct bilirubin level of Paracetamol-induced Rats.
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NCTRL : Normal Control

PISD : Paracetamol induced Rats treated with standard drug

PIBP 200 mg/kg : Paracetamol induced Rats treated with 100 mg/kg b. w of Ethanol extract of B. pinnatum

PIBP 300 mg/kg : Paracetamol induced Rats treated with 200 mg/kg b. w of Ethanol extract of B. pinnatum

PIBP 400 mg/kg : Paracetamol induced Rats treated with 300 mg/kg b. w of Ethanol extract of B. pinnatum

**Fig3.** Effect of ethanol leaf extract of B. pinnatum on activities of Glutathione peroxidase activity of Paracetamol-induced Rats.

NCTRL : Normal Control

PISD : Paracetamol induced Rats treated with standard drug

PIBP 200 mg/kg : Paracetamol induced Rats treated with 100 mg/kg b. w of Ethanol extract of B. pinnatum

PIBP 300 mg/kg : Paracetamol induced Rats treated with 200 mg/kg b. w of Ethanol extract of B. pinnatum

PIBP 400 mg/kg : Paracetamol induced Rats treated with 300 mg/kg b. w of Ethanol extract of B. pinnatum

**Fig4.** Effect of ethanol leaf extract of B. pinnatum on activities of Superoxide dismutase activity of Paracetamol-induced Rats.
Ngobidi KC et al. “Hepato-Protective Effect of Ethanol Leaf Extract of Bryophylum Pinnatum on Paracetamol Induce Hepatitis Albino Rats”

**NCTRL**: Normal Control

**PISD**: Paracetamol induced Rats treated with standard drug

**PIBP 200 mg/kg**: Paracetamol induced Rats treated with 100 mg/kg b. w of Ethanol extract of *B. punnatum*

**PIBP 300 mg/kg**: Paracetamol induced Rats treated with 200 mg/kg b. w of Ethanol extract of *B. punnatum*

**PIBP 400 mg/kg**: Paracetamol induced Rats treated with 300 mg/kg b. w of Ethanol extract of *B. punnatum*

**DISCUSSION**

*Bryophylum pinnatum* has been used traditionally is as herb that is native to India and Africa commonly consumer as beverages n the ancient would and other country known to have medicinal value (Copra et al., 1995).

Paracetamol is a common anagestic used for pain reliever, it has been established that over dose of paracetamol causes liver damage (Raghavendren et al., 2004).

In this study, paracetamol at a high dose was used to induce hepatotoxicity in the animal’s models. This induced hepatotoxicity caused elevation of serum liver enzymes markers (Vries, J.D., 1984). The measurement of activities of enzymes in the tissues and body fluid play a paramount role in disease and investigation of diagnosis (Malomo, 2000). This enzymes such as phosphatases, dehydrogenases and transferees, get body through linkage from disrupted cell membrane in the damaged tissue, (Adaramonye et al. 2008).

The ethanol extract of *Bryophyllum pinnatum* show significant operation of ALT, ALP and GGT when compared with the control. ALT, ALP and GGT which are considered as the specific enzyme markers for liver function here in the extract test growth together with the standard control has no statistical significant with the un-induced and untreated control suggesting that the extract has the ability to restore some of the physiological changes induced by paracetamol hepatotoxicity. In addition to this the plasma membrane interigity of hepatocytes may be said to have been protected by the activity of the extract. This means that the plant extract is hepatoprotective.

**Antioxidant Enzymes**

Reactive oxygen species has been reported to play important roles in disease pathology (Nikhat and Pangey 1996). Biomarkers oxidative stress reflects environmental prooxidant, antioxidant and also serve as surrogate measure of a disease process (Rglinske et al., 1988). The unlinking mechanism of paracetamol induced hepatotoxicity is by oxidative stress, (Bartlett, 2004). The increase oxidative stress leads to depletion of the enzymic and non-enzymic. The administered extract significantly increased depleted level of enzyme antioxidant of SOD, GP x assayed in almost the same level with untreated control showing that the observed un-induced hepatoprotective effect could be due to it’s antioxidant activity. In support of this, the photochemical analysis of the plant extract done in a different studies reveal the presence of Phenolics, Tannin, Autocyanine and Flavonoids. *Bryophylum pinnatum* increases the server level of total protein and albumin which suggest means synthesis of total protein and albumin that accelerate the regeneration process and the protection of the liver cells.

Bilibrubin is the main bi-pigment that is the major product of heme metabolism there was a slight increase in the level of direct bilirubin and extract test group compared with the normal control. This may suggest that the normal control. This may suggest that the extract especially at the 300mg dose may likely have some hemolytic effect on the red blood cell (RBS) but since the movement is not significant one may say that there is no case of either hepatic or hemolytic jaundice.

On the basis of the presence result and the available report, it can be concluded that Ethanolic leaf extract of *Bryophylum pinnatum* has both hepatoprotective effect may be due to it’s antioxidant activities which is as a result of its composition of phytochemcials with antioxidant activities.

Further studies are hereby recommended to investigate the mechanism of the extract and observe the effect, isolate and purity the bioactive compounds in the extract for the purpose of producing drugs for the treatment of chemohepatotoxicity.
REFERENCES


