HEPATOPROTECTIVE ACTIVITY OF ETHANOLIC STEM EXTRACT OF *HOMALIUM LETESTUI* AGAINST THIOACETAMIDE-INDUCED LIVER INJURY

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ABSTRACT

There is an immensely growing demand for research into plants that is of benefit in protecting and/or healing vital organs in the body. *Homalium letestui* plant is a plant of folkloric importance that is used in management of several ailments. The hepatic protective ability of ethanolicextract of the plantagainst thioacetamide induced liver injury in rats was investigated. Thirty six (36) male adult abino rats were divided into 6 groups of six rats per group. Group 1 received normal saline 10 mg/kg, group 2 received thioacetamide 200 mg/kg and group 3 were administ3.ered Silymarin 100 mg/kg. Animals in group 4, 5 and 6 received 250, 500 and 750 mg/kg respectively of ethanol stem extract of *Homalium letestui* plant. Administration was carried out for 8 days after which animals were sacrificed. Examination of liver function parameters and histopathological observations were used to assess hepatoprotective activity of the stem extract. There was significant ($p \leq 0.05$) decrease in level of liver enzymesAST, ALT and ALP, increase in endogenous level of total cholesterol, total and direct bilirubin and decreases in total protein and albumin levels when compared to control. Histopathological examination agrees with that of chempathology results suggesting the plant may have organ protective ability. The extracts result was also comparable to that of standard drug used. The result of the study shows that the plant may possess hepatoprotective activity which supports it use in traditional medicine as an antidote.

KEYWORDS: *Homalium letestui*, hepatoprotective, thioacetamide

Introduction

Reactive oxygen species (ROS) include a number of chemically reactive molecules derived from oxygen[1]. Some of those molecules are extremely reactive, such as the hydroxyl radical, while some are less reactive (superoxide and hydrogen peroxide). Intracellular free radicals, i.e., free, low molecular weight molecules with an unpaired electron, are often ROS and vice versa and the two terms are therefore commonly used as equivalents. Free radicals and ROS can readily react with most biomolecules, starting a chain reaction of free radical formation. Antioxidants are the compounds which terminate the attack of free radicals and thus reduce the risk of these disorders[2]. Free radicals or highly reactive oxygen species are formed by exogenous chemicals or endogenous metabolic processes in the human body. These are capable of oxidizing bio-molecules viz nucleic acids, proteins, lipids and DNA and can initiate different degenerative diseases like neurological disorders, cancer, emphysema, cirrhosis, atherosclerosis, arthritis etc[3]. Almost all organisms are protected up to some extent from free radical damage with the help of enzymes such as super-oxide dismutase, catalase and antioxidant compounds viz. ascorbic acid, tocopherol, phenolic acids, polyphenols, flavonoids and glutathione. It has been reported that antioxidant supplements or dietary antioxidants protect against the damaging effects of free radicals[4]. Presently, much attention has been focused on the use of natural antioxidants to protect the human body especially brain tissues from the oxidative damage caused by free radicals. In last two decades, several medicinal plants have

shown such effectiveness through the traditional methods of psychoneuropharmacology[5].

The liver is a vital organ of the digestive system present in vertebrates and some other animals. The liver is the only human internal organ capable of natural regeneration of lost tissue; as little as 25% of a liver can regenerate into a whole liver. Regeneration is very rapid. The liver will return to a normal size in one to two weeks following the removal of greater than 50% of its weight[6].This is predominantly due to the hepatocytes re-entering the cell cycle. It has a wide range of functions, including detoxification, protein synthesis, and production of biochemicals necessary for digestion[7].There are numerous method of chemically – induced models of hepatotoxity; this include CCl_4 - induced hepatotoxity, thioacetamide - induced hepatotoxicity, galactosamine - induced hepatotoxicity, alcohol - induced hepatotoxicity and Paracetamol - induced Hepatotoxicity[8].

Homalium letestui is a plant of various tradomedical importance. *Homalium letestui* occurs from Senegal east to the Central African Republic and south to western DR Congo and Cabinda (Angola).. The fruits are showy and the young leaf-flush is red before turning green. The tree is thus attractive and worthy of cultivation[9].In Ivory Coast sap from the bark is used in enemas for the treatment of generalised oedemas while macerate from the bark are rubbed over the area[10]. Stem bark and root are used in various decoctions by the Ibibios of the Niger Delta of Nigeria to treat stomach ulcer, malaria and other inflammatory diseases and also as an aphrodisiac by the Yorubas of Western Nigeria[11].Okokon[12] reported the presence of α-terpineol, Vanillin, 4-phenyl isocoumarin, 3,4,5-trimethoxy phenol, 2-Coumaranone, and xanthones in the stembark extract of *H. letestui*. Also, antiplasmodial[11], antidiabetic[13], anti-inflammatory and analgesic[12], cellular antioxidant, anticancer, and antileishmanial[12], depressant and anticonvulsant[14] antibacterial[15], *in vitro* antioxidant activity against DPPH[15], antiulcer[13] and antidiarrheal[13] activities of the plant have been established. The hepatoprotective ability of ethanol stem extract of *Homalium letestui* plant was studied against carbon thioacetamide induced liver injury in order to provide scientific explanation for its use in traditional medicine.

Materials and methods

Plants collection

Homalium letestui (stem) was collected in a forest in Uruan area, Akwa Ibom State, Nigeria. It was identified and authenticated by Dr. Margaret Bassey of Department of Botany and Ecological Studies, University of Uyo, Uyo, Nigeria.Hebarium specimen (FPUU 382) was deposited at Department of Pharmacognosy and Natural Medicine Herbarium.

Extraction

The stem was washed and shed dried for two weeks. The dried plant material was then cut into smaller pieces and grounded to powder. The powdered material was macerated in 70% ethanol. The liquid filtrate was evaporated to dryness *in vacuum* 40C using rotary evaporator. The ethanol extract was stored at -4^{O}C until used.

Animals

Adult male white albino rats were obtained from the University of Uyo animal house. They were maintained on standard animal pellets and water *ad libitum*. Permission and approval for animal studies were obtained from the College of Health Sciences Animal Ethics committee, University of Uyo.

Animal treatment

Thirty six (36) rats were weighed and divided into six groups with 6 animals per group. Treatment was as follows: Group 1 consisted of normal animals that were administered with normal saline (10 ml/kg) for eight days, Group 2, the organotoxic group, recieved normal saline 10 ml/kg for eight days. Group 3 served as the standard group and rats in this group were administered 100 mg/kg body weight of silymarin orally for 8 days, while groups4, 5 and 6 were administered p.o with 250, 500 and 750 mg/kg of *H. letestui* stem extract respectively daily for 8 days. On the 8th day the animals in group 2-6 were administered 200 mg/kg bw of thioacetamide dissolved in saline orally. Twenty hours later all animals were weighed again and sacrificed under light diethyl ether vapour.

Hematological study

Blood samples were collected from each rat by cardiac puncture immediately after the animals were sacrificed under diethyl ether anesthesia, using 21 gauge (21 G) needles mounted on a 5 ml syringe into ethylene diamine tetra-acetic acid (EDTA) - coated sample bottles for analyzed. Hematological parameters such as full blood count (FBC), hemoglobin, (Hb), packed cell volume (PCV), platelet concentration (PLC) and Total and differential white blood cell count (WBC). These parameters were analyzed using automatic hematological system.

Evaluation of the protective effect of the extract against thioacetamide-induced liver injury on biochemical parameters and histology of liverof rats

Serum was separated from the blood of each animal sacrificed and the sera were stored at -20°C until used for biochemical determinations such as total protein, albumin, aspartate aminotransferase(AST),alanine aminotransferase (ALT),alkaline phosphatase(ALP), total cholesterol,total and direct bilirubin. The determinations were done spectrophotometrically using Randox

analytical kits according to standard procedures of manufacturer's protocols[16]. The livers of the animals were surgically removed, weighed and a part of each fixed in 10% formaldehyde for histological processes.

Statistical Analysis and Data Evaluation

Data obtained from this study were analyzed statistically using Students't-test and ANOVA (One - way) followed by a post test (Tukey-Kramer multiple comparison test). Differences between means were considered significant at 5%, 1% and 0.1% level of significance i.e $P \leq 0.05$, 0.01 and 0.001.

RESULT

Effect of Treatment with Ethanol Stem Extract of *Homalium letestui* on the Hematological Parameters of Rats with Thioacetamide-Induced Hepatotoxicity.

The administration of thioacetamide (200 mg/kg bw) did not affect RBC, WBC, platelet count and haemoglobin concentration as well as PCV, basophils and lymphocytes percentages when compared to normal control. However, there was significant ($p<0.01$-0.001) increase in the percentage of neutrophils in thioacetamide-treated rats and those of rats pretreated with middle and high doses of the extract (500 and 750 mg/kg). Eosinophils percentage was significantly ($p<0.05$ - 0.001) reduced in both thioacetamide and extract treated groups (Table 1)

Evaluation of Effect of *Homalium letestui Stem* on Liver Function Test of Thioacetamide - Induced Liver Injury in Rats

Administration of thioacetamide (200 mg/kg bw) to rats caused a significant ($p<0.001$) elevation of enzymes levels such as AST, ALT , ALP, total cholesterol, total and direct bilirubin and decreases in total protein and albumin levels when compared to control. There was observable significant ($p<0.01$ - 0.001) non – dose dependent decreases of these enzymes levels and that of total cholesterol, total and direct bilirubin in the groups pre-administered with the stem extract *of H. letestui* (500 – 1000 mg/kg bw) when compared with the thioacetamide group. Total protein and albumin levels were significantly ($p<0.05$ - 0.001) elevated dose-dependently in the groups pre-treated with the stem extract when compared to the thioacetamide group (Table 2).

Disarrangement of normal hepatic cells with centrilobular necrosis, hyperplasia, vascular and cellular degeneration, polymorphonuclear aggregation, inflammation and fatty degeneration was observed in the thioacetamide treated rats (Figure 1). The liver sections of the rats treated with stem extract of *H. letestui* (250 - 750 mg/kg) showed signs of protection as was evident by the reduction/ absence of inflammatory cells, vascular congestion and degeneration, cellular degeneration, necrosis and vacuoles, while the liver sections of the rats treated with silymarin (100 mg/kg) showed significant reduction in fatty degeneration and absence of necrosis and inflammation (Figure 1 and 2).

DISCUSSION

Thioacetamide (TAA) is a hepatotoxic agent that is converted by the liver enzymes to highly reactive toxic S-oxide derivatives which cause centrolobular necrosis. These metabolites interfere with the movement of RNA from the nucleus to the cytoplasm which leads to membrane injury, therefore they reduce the number of viable hepatocytes as well as rate of oxygen consumption[17]. Moreover, TAA decreases the volume of bile and its content (bile salts, cholic acid and deoxycholicacid). Rodents intoxicated with TAA have been demonstrated as good experimental model of liver cirrhosis and fibrosis, and have been used for evaluation of antihepatotoxic drugs[18]. It is well known that the mechanism of liver toxicity by TAA involved free radical chain reactions. Proteomic analysis of TAA - induced hepatotoxicity and cirrhosis in rat livers have revealed that administration of TAA down-regulates the enzymes of the primary metabolic pathways such as fatty acid beta-oxidation, branched chain amino acids and methionine breakdown, whereas it up-regulates proteins on the other hand, that are related to oxidative stress and lipid peroxidation[19]. Under oxidative stress conditions, the cascade of reactions induced by pro-oxidant leads to the degeneration of macromolecules such as nucleic acids, proteins and lipids. In the liver, the peroxidation of membrane lipids is the starting point of tissue necrosis.

In this work, the haematological effect of the ethanol extract of the *Homalium letestui* stem extract on thioacetamide – induced toxicity in rat was investigated. The results demonstrated that thioacetamide exposure was associated with slight increase in Hb, PCV, neutrophils and slight decrease in lymphocytes, monocytes and RBC. The result is in line with Muddasir[20] that acute TAA administration caused neutrophilia, thrombocytosis as well as increased hemoglobin concentration and reduction of erythrocytic count. The depression in RBCs count and rise in Hb contents and PCV recorded in the present work is suggest megaloblastic RBCs and could be as a result of disturbed hematopoiesis, destruction of erythrocytes, and reduction in the rate of their formation and/or their enhanced removal from circulation. According to Travlos[21], consistent erythrocyte damage could be related to direct oxidative injury to the red cells by the chemicals or to the pitting function of the spleen. Leucopenia is a clinical manifestation with decrease in circulating white blood cell (WBC). From the present study, the pretreated group had improved level of lymphocytes and

monocytes, while there were improvements in some of the haematological parameters. Neutrophils and its derived cytokines play a crucial role in the development and manifestation of inflammation. The stimulation of neutrophils can lead to the production of oxygen-derived free radicals also called reactive oxygen species (ROS) that cause further cellular damage[22]. The formation of free radicals and cytotoxic oxygen metabolites probably impart a key role in various types of tissue degeneration and pathology such as aging, cancer and retinal degeneration[23]. In the present study, after intraperitonial injection of TAA, there was significant elevation in the neutrophil count (neutrophilia) when compared to control which may be due to the free radicals resulting from TAA metabolism which caused liver injury and a proportion ofthese free radicals liberated into the blood may also affect the circulating cells and induce a significant change in their number. This significant neutrophilia might reflect its involvement in inflammation by forming various reactive oxygen species (ROS), inflammatory, metabolic and myloproliferative disorders, tissue necrosis, acute hemorrhagia, malignant tumors or due to rapid release of young cells from the bone marrow[24]. Persistent increase in the neutrophil level of all extract group was observed in this work which may suggest that the extract could not effectively reverse the effect of elevation in neutrophil count induced by thioacetamide administration. It could also be that increase in neutrophils in pretreated groups may be a potentiation in immunological response to the toxicant[25].

A marked reduction of serum total protein and albumin levels were observed in the thioacetamide treated group when compared with the normal healthy animals. *Homalium letestui*-treatment caused significant improvements in the levels of these proteins in the pretreated group. Parallel findings were also previously reported[26]. Also, the levels of plasma total protein in all the extract-treated rats were encouragingly above that of Silymarin-treated rats, indicating that the plant can be of immense clinical importance in the management of liver diseases. Hepatic factors (ALP, ALT, and AST) were significantly increased in the thioacetamide administered rats, which agrees with previously described work[27]. Although, ethanol stem extract of *Homalium letestui* showed significant prospect in the liver function test it did not reduce the elevated liver enzymes levels to normal as in the normal control in the doses administered. Hematological and liver function parameter results were supported by the histopathological analysis of the organotoxic group that the plant possesses hepatoprotective activity.

Limitation

The study is expected to be beneficial to humans but had limitation of using animals for the work.

Conclusion

Results of hematological, biochemical and histological analysis showed that the plants has hepatic protecting ability against thioacetamide induced liver injury in rat, which is of immense potential in preventing and treating liver disease.

Conflict of Interest

There is no potential conflict of interest in this research work.

Reference

1. Halliwell, B. (1996). Antioxidants in Human Health and Disease. *Annual Review of Nutrition.* 16:33–50.
2. Rice-Evans, C.A., Miller, N.J. and Paganga, G. (1996). Structure Antioxidant Activity Relationships of Flavonoids and Phenolic Acids. *Free Radical Biology and Medicine*, 20, 933–56.
3. Halliwell, B. and Gutteridge, J. M. (1984) Oxygen Toxicity, Oxygen Radicals, Transition Metals and Disease. *Biochemical Journal*, 219, 1–14.
4. Prior, R. L. and Cao, G. (1999). Variability in Dietary Antioxidant Related Natural Product Supplements: The Need for Methods of Standardization. *Journal of American Nutraceutical Association*, 2, 46–56.
5. Dhawan, B. N., Koslow, S. H., Murthy, R. S. and Coelho GV. (2005). Decade of the Brain: India/USA Research in Mental Health and Neurosciences Rockville: *National Instituteof Mental Health*, 197-202
6. Dieter Häussinger, ed. (2011). *Liver Regeneration*. Berlin: De Gruyter. p. 1–350.
7. Shneider, B. L. and Sherman, P. M. (2008). *Pediatric Gastrointestinal Disease*. Connecticut: PMPH-USA. p. 751.
8. Eliwa, H. A., El-Denshary, E. S., Nada, S. A., Elyamany, M. F., Omara, E. A. and Asaaf, N. (2014). Evaluation of the Therapeutic Effect of Whey proteins on the Hepatotoxicity Induced by Paracetamol and Alcohol Co-administration in Rats. *International Journal of Progressive Pharmaceutical Research*, 3, 295-314.
9. Aubréville, A. (1959) De quelques Césalpiniées africaines. *Bulletin de la Société Botanique de France,* 104, 495–498.
10. Bouquet, A. and Debray, M. (1974). *Medicinal Plant of the Ivory Coast*. Trav Doc Orstom, 32. Pp 441.

11. Okokon, J.E., Ita, B. and Udokpoh, A.E. (2006). Antiplasmodial Activity of *Homalium letestui*. *Phytotherapy Research*, 20, 949–951.
12. Okokon, J. E, Okokon, P. J, Dar Farooq, A. and Choudhary, M. I. (2013). Anti-inflammatory and Antinociceptive Activities of *Homalium letestui*. Pharmaceutical Biology. 1459-66.
13. Okokon, J.E., Antia, B.S. and Ita, B.N. (2007). Antidiabetic Effects of *Homalium letestui* (Flacourtiaceae) in Streptozotocin Induced Diabetic Rats. *Research Journal of Medicinal Plants*, 1, 134–138.
14. Okokon, J. E. and Davies, K. (2014). Psychopharmacological Studies of *Mammea africana* Stem Bark Extract. *The Journal of Phytopharmacology*, 3, 204–213.
15. Ita, B. and Ngochindo, R. (2014). Fatty Acid Composition, Antioxidant and Antimicrobial Activity of *Homalium letestui* Stem. *Australian Journal of Basic and Applied Sciences*, 8, 416-422.
16. Reitman, S. and Frankel, S. (1957). Glutamic – Pyruvate Transaminase Assay by Colorimetric Method. American Journal of Clinical Pathology, 28, 56.
17. Müller, A., Machnik, F., Zimmermann, T. and Schubert, H. (1988). Thioacetamide-induced cirrhosis-like liver lesions in rats—usefulness and reliability of this animal model. *Experimental Pathology*, 34, 229–236.
18. Brandon, E., Schrum, L. W, Schmidt, C. M. and McKillop, I. H. (2012). Rodent Models of Alcoholic Liver Disease of Mice and Men. *Alcohol*, 46, 715-25.
19. Low, T.Y., Leow, C.K., Salto-Tellez, M. and Chung, M.C. (2004): A Proteomic Analysis of Thioacetamide - Induced Hepatotoxicity and Cirrhosis in Rat Rivers. *Proteomics*, 4, 3960-3974.
20. Muddasir, S., Rajinder, R., Pawan, A. S., Kumar, V. H., Verma, W.A and Payen, E. S. (2013). Hepatoprotective Mechanisms of *Ageratum conyzoides* L. on Oxidative Damage Induced by Acetaminophen in Wistar Rats. *Human Experimental Toxicology*, 65, 429 – 437.
21. Travlos, G.S., Morris, R.W., Elwell, M.R., Duke A., Resenblum, S. and Thompson, M. B. (1996). Frequency and Relationships of Clinical Chemistry and Liver and Kidney Histopathology Findings in 13-week Toxicity Studies in Rats. *Toxicology*, 107, 17–29.
22. Bartosz, G. (2000). Free Radicals in Biology and Medicine, *Cell Biology International*. 24, 764 - 770.
23. Shapiro, H., Ashkenazi, M., Weizman, N., Shahmurov, M., Aeed H. and Bruck R. (2006). Curcumin Ameliorates Acute Thioacetamide-Induced Hepatotoxicity, *Journal of Gastroenterology and Hepatology*. 21 (2). Pp. 521.
24. Doi, K., Kurabe, S., Shimazu, N. and Inagaki, M. (1991). Systemic Histopathology of Rats with CCl4-Induced Hepatic Cirrhosis, *Laboratory Animals*, 25, 21-25.
25. Sicherer, S. and Sampson, H. A. (2010). Allergen and Immunological Consequences. *Journal of Allergy and Clinical Immunology*. 125 (22), 116-25.
26. Kumar, A. (2012). A Review on Hepatoprotective Herbal Drugs. *International Journal of Reasearch in Pharmaceuticals*, 2, 92-102.
27. Alshawsh, M. A., Abdulla, M. A., Ismail, S. and Amin, Z. A. (2011). Hepatoprotective Effects of *Orthosiphon stamineus* Extract on Thioacetamide-Induced Liver Cirrhosis in Rats. *Evidence-Based Complement and Alternative Medicine*, 6, 1-6.

Appendix: Tables and Figures

Table1:Effect of treatment with ethanol stem extract of *Homalium letestui* on the hematological parameters of rats with thioacetamide -Induced hepatotoxicity.

Parameters Treatment Dose (mg/kg)	RBC (X 10^{12}/l)	PCV (%)	Hb (g/dl)	WBC (X 10^{9}/l)	Neutrophils (%)	Lymphocytes (%)	Monocytes (%)	Eosinophils (%)	Basophils (%)	Platelets (%)
Normal control	4.95±0.13	46.8±2.60	15.5±0.86	5.62±0.54	41.5±5.99	45.5±6.17	7.00±1.80	4.33±2.02	0.00±0.00	141.6±20.91
THA +Dist. Water	4.08±0.25	47.8±2.13	15.8±0.72	5.16±0.45	48.33±3.28^{c}	42.3±3.63	5.66±1.43^{c}	2.33±0.80^{a}	0.20±0.20	144.5±15.87
Silymarin 100 mg/kg + THA	5.15±0.85	57.8±3.33	17.25 ±1.10	4.80±0.43	42.1±6.96	48.6±7.00	5.16±2.35^{d}	2.00±0.85^{a}	0.00±0.00	154.0±22.37^{a}
HL. 250mg/kg + THA	5.11±0.28	54.8±1.77	18.8±0.59	4.66±0.48	52.6±8.14^{c}	41.3±3.63	5.00±1.36^{c}	1.00±0.81^{c,e}	0.00±0.00	156.5±13.01^{c}
HL. 500mg/kg+ THA	4.80±0.16	49.0±2.12	16.3±0.70	5.85±0.69	44.3±5.30	45.8±4.84	5.0±1.73^{c,d}	3.83±0.85^{c}	0.00±0.00	189.3±27.56^{ce}
HL. 750mg/kg+ THA	5.11±0.39	54.3±3.32	18.16±7.68	5.10±0.78	49.1±7.68^{c}	40.8±6.38	5.0±1.65^{c}	4.33±1.05^{a}	0.00±0.00	133.1 ±16.66^{c}

Data were expressed as mean SEM. significant at ap< 0.05, bp< 0.01, cp< 0.001 when compared to control. dp< 0.05, ep< 0.01, fp< 0.001 when compared to paracetamol . n = 6.

Table 2: Effect of *H. letestui* on liver function of thioacetamide –induced liver injury in rats

PARAMETERS/ TREATMENT	TOTAL PROTEIN (g/dl)	ALBUMIN (g/dl)	TOTAL BILIRUBIN (mg/dl)	CONJUGATED BILIRUBIN (mg/dl)	AST (IU/L)	ALT (IU/L)	ALP (IU/L)	TOTAL CHOLESTEROL Mmol/L
Normal control		4.45±0.59	2.98±0.28	1.23±0.13	114.0±3.65	38.66±3.74	178.83±14.35	3.93± 0.29
THA +Dist. Water	2.95±0.58^{c}	2.58±0.22^{c}	7.41±0.25^{c}	2.53±0.13^{c}	170.1±3.25^{c}	86.62±5.20^{c}	281.32±10.37^{c}	7.53±0.45^{c}
Silymarin 100 mg/kg + THA	5.73±0.37^{f}	4.12±0.39^{f}	3.75±0.34^{f}	0.85±0.13^{f}	151.82±4.92^{cd}	61.50±6.11^{bd}	202.8±10.24^{cf}	4.05±0.39^{f}
Ext. 250 mg/kg + THA	6.13±0.84^{f}	4.67±0.20^{f}	5.16±0.39^{af}	1.60±0.05^{d}	163.1±6.22^{c}	80.30±10.32^{c}	224.0±6.19^{cf}	5.67±0.53^{cf}
Ext. 500 mg/kg+ THA	6.36±0.98^{f}	4.11±0.19^{e}	4.56±0.42^{af}	1.00±0.15^{f}	132.16±4.33^{cf}	80.66±6 .25^{c}	214.6±3.49^{cf}	6.63±0.14^{bf}
Ext. 750 mg/kg+ THA	6.25±1.05^{f}	4.15±0.33^{f}	4.35±0.25^{f}	0.86±0.06^{b}	150.5±4.35^{ad}	84.50±9.80^{c}	239.5±7.32^{ce}	6.46±0.11^{f}

Data were expressed as mean SEM. significant at ap< 0.05, bp< 0.01, cp< 0.001 when compared to control.dp< 0.05, ep< 0.01, f< 0.001 when compared to paracetamol . n =6

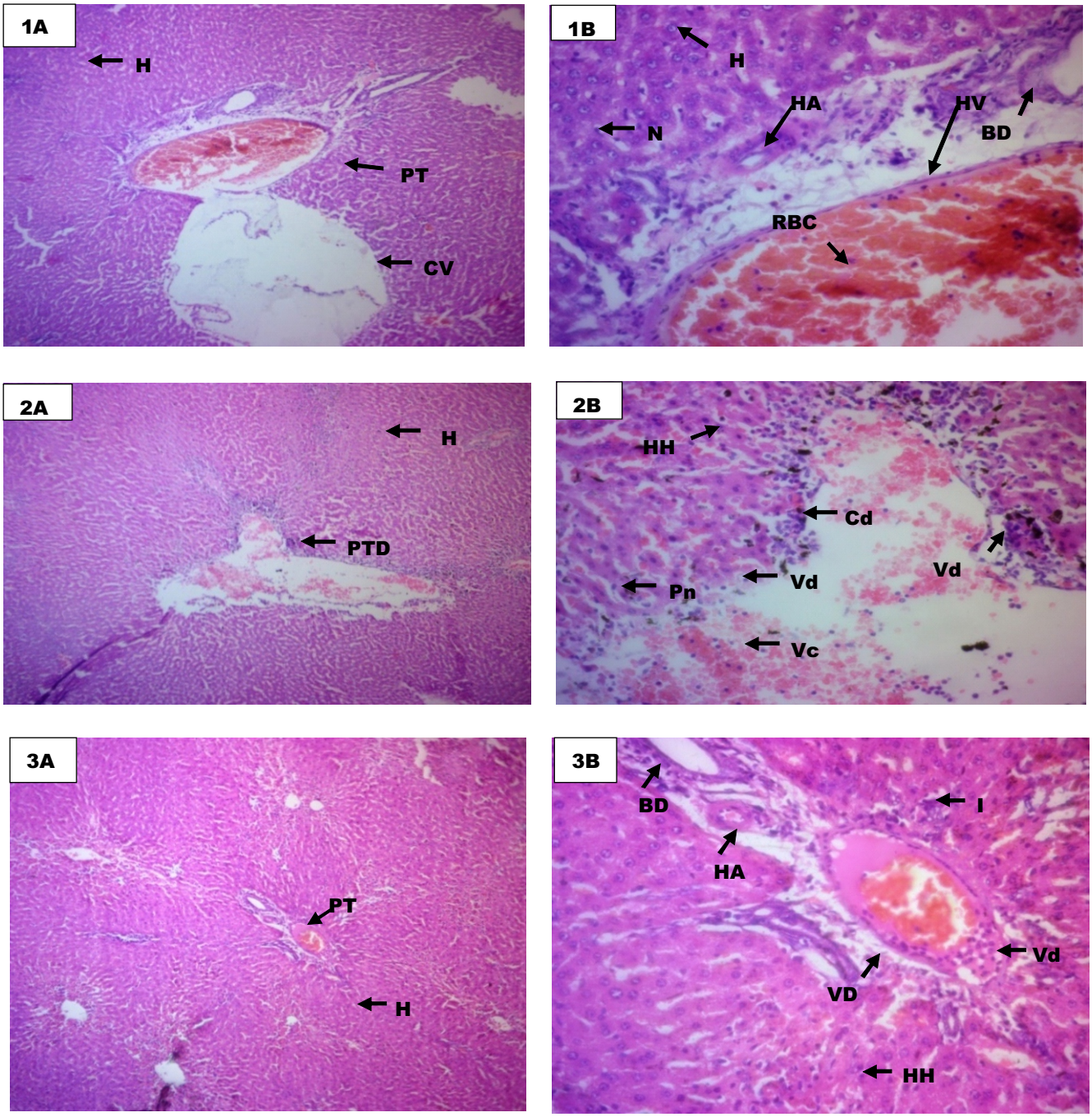

Figure 1: Histological sections of Livers of rats treated with Normal saline 10 ml/kg bw (1),Thioacetamide 200 mg/kg bw (2) and Silymarin100 mg/kg bwand Thioacetamide 200 mg/kgbw(3) at magnification A (x100) and B(x400) using H&E technique.

Keys: Bile duct (BD) Cellular degeneration (Cd), Portal triad (PT), Inflammation (I), Portal triad degeneration (PTD), Cellular Degeneration, Vascular congestion (Vc), Hepatic artery (HA), Vascular degeneration (Vd), Hepatocytic hyperplasia (HH), Hepatic artery (HA), Pyknotic nucleus (Pn)

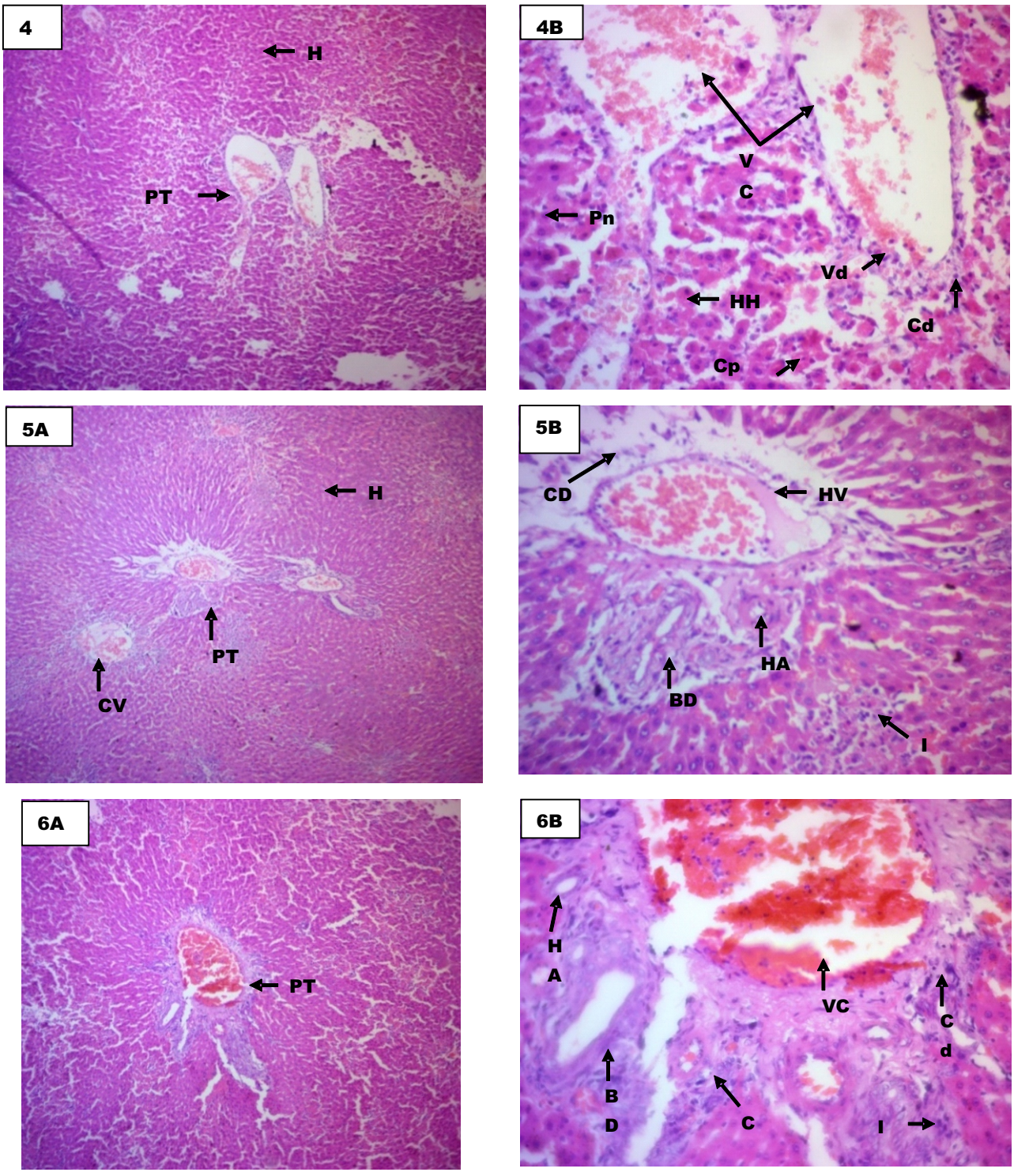

Figure 2: Histological sections of Livers of rats treated with *Homaliumletestui*250 mg/kg bw (4) and Thioacetamide,*Homaliumletestui*500 mg/kg bw and Thioacetamide 200 mg/kg bw (5) and *Homaliumletestui*750 mg/kg bwand Thioacetamide 200 mg/kgbw (6) at magnification A (x100) and B(x400) stained with H&E technique.

Keys: Bile duct (BD) Cellular degeneration (Cd), Inflammation (I), vascular degeneration (Vd), Hepatocytic hyperplasia (H), Hepatic arterty (HA), Pyknotic nucleus (Pn), Portal triad (PT) and Vascular congestion (Vc)