



Synergistic Protective Effects of Aqueous Extract of *Sesamum radiatum* Leaves and Ascorbic acid against Paracetamol- induced Hepatotoxicity in Rats

NWEJE- ANYALOWU Paul Chukwuemeka^{1*}, IDAKWOJI Precious Adejoh², OKAFOR Stephen Chiadikaobi³, AJIMA Judith Nnedimkpa⁴, DARA Amarachi Ngozika⁵

¹Department of Biochemistry, Clifford University, Owerri, Abia State, Nigeria
E-mail: nwejeanyalowup@clifforduni.edu.ng

²Department of Biochemistry, Faculty of Biological Sciences, University of Nigeria, Nsukka, Enugu State, Nigeria
E-mail: sirprecious@yahoo.com

³Department of Pharmacology and Therapeutics, Faculty of Pharmaceutical Sciences, Ahmadu Bello University, Zaria, Kaduna State, Nigeria
E-mail: stevechiadi@yahoo.com

⁴Department of Biochemistry, Coal City University, Emene, Enugu State, Nigeria
E-mail: judith.obeta@coalcityuniversity.edu.ng

⁵Department of Biochemistry, Faculty of Science
Madonna University, Elele, Rivers State, Nigeria
E-mail: amarachidara@gmail.com

*Corresponding author: NWEJE- ANYALOWU Paul Chukwuemeka
E-mail: nwejeanyalowup@clifforduni.edu.ng
+234 8038616861

Abstract

Objective: This study investigated the possible protective effect of aqueous extract of *Sesamum radiatum* leaves against paracetamol- induced hepatotoxicity and a possible synergistic effect when it is co-administered with ascorbic acid. **Materials and Methods:** The protective effect of graded doses (200 and 400 mg/kg) of the extract, ascorbic acid (100 mg/kg) and extract (200 mg/kg)/ Ascorbic acid (100 mg/kg) was evaluated against paracetamol (2g/kg) -induced hepatotoxicity in rats. Following treatment, hepatoprotective effect was investigated through the assay of liver function parameters, namely Alanine aminotransaminase (ALT), Aspartate aminotransaminase (AST), and Alkaline phosphatase activities (ALP), total and direct bilirubin, serum protein and albumin concentrations, total cholesterol (T. Chol.), triacylglyceride (TAG), High density lipoprotein (HDL) and Low density lipoprotein (LDL). The liver tissues of the rats were also subjected to histopathological examinations. **Results:** Administration of graded doses of the extract, ascorbic acid as well as the extract/ ascorbic acid co-administration significantly ($p < 0.05$) reduced the activity of enzymes (ALT, AST and ALP), total cholesterol, direct and total bilirubin, T. Chol., TAG and LDL and also significantly ($p < 0.05$) elevated serum levels of HDL, total protein and albumin. Histology of

the liver sections from extract, ascorbic acid- treated, and extract/ ascorbic acid co- treated rats showed reductions in the pathological features compared to the paracetamol-treated animals. The biochemical changes were consistent with histopathological observations suggesting marked hepatoprotective effect of the extract. However, the protective effect of the extract/ ascorbic acid co- administration was much more. **Conclusion:** From the present results we could conclude that aqueous extract of *Sesamum radiatum* leaves possess a promising hepatoprotective effect against paracetamol hepatic damage and when co- administered with ascorbic acid, it provides a synergistic effect that could be useful in the management of liver diseases.

Keywords: *Sesamum radiatum*, hepatotoxicity, histopathological examinations.

1.0 Introduction

The liver is the major organ involved in intermediary metabolism and detoxification/ elimination of toxic substances. In humans, the liver is often exposed to an array of toxic substances through environmental exposure, social factors such as alcoholism, drugs, consumption of contaminated food or during exposure to chemical substances in the occupational environment, all of which can damage and weaken it, eventually leading to diseases like hepatitis or cirrhosis (Zimmerman *et al.*, 1994). Hepatic diseases pose a serious threat to human health worldwide but unfortunately, modern medicine offers few effective treatment options for these diseases (Nyblom *et al.*, 2006). It is therefore necessary to search for drugs of high efficacy and safety to replace or augment the ones currently available for the treatment of liver diseases.

Various studies have reported the hepato-protective properties of Ascorbic acid. Bashandy and Alwasel (2011) reported that Ascorbic acid normalized levels of alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, blood hydroperoxide and malondialdehyde in liver of carbon tetrachloride intoxicated rats. The ability of vitamin C to prevent Carbon tetrachloride induced hepatotoxicity in rats was also reported by (Ademuyiwa *et al.*, 1994; Kataoke *et al.*, 2012). Ascorbic acid was also able to preserved 100% of cell integrity and modulated alanine aminotransferase and aspartate aminotransferase (Grajeda- cota *et al.*, 2004). Similar observation was reported by Amballi *et al.* (2010). He and his co-workers evaluated the effect of Ascorbic acid on short-term hematological and biochemical alterations induced by acute chlorphyrifos in wistar rats. They reported that vitamin C mitigated alterations in serum biochemical parameters via the normalization of alanine amino transferase, aspartate aminotransferase and alkaline phosphatase. Ascorbic acid is reported to form synergy with other agents. One of the visible synergies is between vitamin C and vitamin E. This is supported by the co-administration

of vitamins C and E as food supplement administered for 7 days to ethanol intoxicated rats. This ameliorated ethanol induced hepatotoxicity via normalization of transaminases and inhibition of lipid peroxidation. Ethanol induced histopathological changes were also reversed (Oyinbo *et al.*, 2006).

Sesamum radiatum is a perennial herb found in the tropical areas of Africa and belongs to the Pedaliaceae family (Purseglove, 1974; Hutchinson and Dalziel, 1954). The leaves, seeds and oil serve as food especially in farming communities in Nigeria (Akpan-Iwo *et al.*, 2006). The leaves are used for treating various stomach ailments. The decoction of the leaves is used for the treatment of catarrh, eye pains as well as bruises and erupted skins. The decoction of combined roots and leaves has been reported to have anti-viral and antifungal activity (Gills, 1992). The aqueous extract of the leaves has been found to be rich in phenols, lignans and flavonoids. Sterols were also found to be among its constituents. Despite the many reported pharmacological properties of this plant, its hepatoprotective property is yet to be studied. This study therefore seeks to investigate the hepatoprotective property of the aqueous extract of the leaves of *Sesamum radiatum* and also to investigate a possible synergistic protective effect against paracetamol- induced hepatotoxicity when co-administered with Ascorbic acid.

The experimental intoxication induced by Paracetamol is widely used for modeling liver injury in rats. Hepatotoxicity is connected with severe impairment of cell protection mechanisms. The location of liver injury is defined mainly by the biotransformation of paracetamol, which is cytochrome P-450 dependent. Free radicals initiate the process of lipid peroxidation, which is generally caused by inhibition of enzyme activity. Damage to the liver is not due to the drug itself but to a toxic metabolite (*N*-acetyl-*p*-benzoquinone imine NAPQI) which is produced by cytochrome P450 enzymes in the liver. In normal circumstances this metabolite is detoxified by conjugating with glutathione in phase 2 reaction. In

overdose large amount of NAPQI (Boyd and Bereczky, 1966), is generated which overwhelm the detoxification process and lead to damage to liver cells.

2.0 Materials and Methods

2.1 Materials

2.1.1 Chemicals and drugs

Assay kits for the estimation of serum ALT, AST, ALP, total bilirubin and total protein were commercial kits from Randox Laboratories Limited, United Kingdom. All other chemicals used in this study were of analytical grade and were purchased from Sigma Chemical Co. Ltd (USA) through a local vendor. Ascorbic acid and Paracetamol (500mg) were purchased from a local pharmacy shop.

2.1.2 Animals

Adult Wistar rats of either sex weighing 150–200g were used for this study. They were kept in stainless steel cages under standard laboratory conditions. They were maintained on clean water and standard rodent feed.

2.2 Methods

2.2.1 Plant Collection and Identification

The leaves of *Sesamum radiatum* were collected from a natural habitat in Mararaba Area of Nassarawa State, Nigeria. The plants were identified at the herbarium unit of Biological Sciences Department, Ahmadu Bello University, Zaria and voucher specimens were deposited for future references.

2.2.2 Preparation of Extracts

The leaves of *Sesamum radiatum* were shade-dried for five (5) days and pulverized using an electric blender. One thousand and five hundred (1500) gram of the pulverized leaves was soaked in distilled water for 72-hours. The resulting mixture was filtered using Whatmann filter paper (Size No1) and the extract was concentrated using a free-dryer.

2.2.3 Acute Toxicity Study

The oral median lethal dose (LD50) of the extract was determined in rats according to the method of Lorke (1983).

2.2.4 Experimental Design

A total of 30 rats were weighed and divided into six groups of 5 animals each and treated as follows: Group 1 consisted of normal animals that were administered with distilled water (1 ml), Group 2 was administered distilled water (1 ml), group 3 and 4 were administered 200 and 400 mg/kg of *Sesamum radiatum* extract respectively, while group 5 was administered 100 mg/kg Ascorbic acid +200 mg/kg *Sesamum radiatum* extract daily for 7 days. Group 6 was treated with Ascorbic acid (100 mg/kg) for the same period. Paracetamol 2g/kg, was administered to groups 2-6 on the 7th day. Twenty-four hours after paracetamol administration, animals were sacrificed under light anesthesia. Blood samples were collected by cardiac puncture and used for biochemical assays while the liver was excised for histopathological examination.

2.2.4.1 Biochemical Assays

Serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were measured according to the method of Reitman and Frankel, (1957). Total bilirubin (Jendrassik 1938) and direct bilirubin were also measured. Triglycerides were determined according to Fassati and Prencipe (1982), while total cholesterol was determined according to Fredrikson *et al.*, (1967). Total protein was estimated by the method of Henry, (1964) and albumin were also estimated. All serum biochemical constituents were estimated using commercial kits Randox Laboratories Limited, United Kingdom.

2.2.4.2 Histopathological Examination

Liver of the sacrificed rats were taken from each rat and immersed in 10 % formalin solution. The specimens were then trimmed, washed and dehydrated in ascending grades of alcohol. Samples were then cleared in xylol, embedded in paraffin, sectioned (4-6 microns) and stained with Hematoxylin and Eosin for histopathological examination according to the method of (Sheehan and Harpachak, 1980).

2.2.5 Statistical Analysis

Statistical analysis was carried out using SPSS version 20.0. All the data were expressed as mean \pm SEM and the statistical differences between the means were determined by one way analysis of variance (ANOVA) which was followed by Duncan test and difference between means at $P > 0.05$ were considered significant.

3.0 Results

3.1 Acute Toxicity

The results of acute toxicity studies showed no mortality or physical changes in skin and fur, eyes and mucus membrane, respiratory rate, circulatory signs, autonomic and central nervous system effects up to a dose of 5000 mg/kg of aqueous extract of *Sesamum radiatum*. The oral LD₅₀ of the extract was then taken to be > 5000 mg/kg.

3.2 Effect of the Administration of Aqueous Extract of *Sesamum radiatum* Leaves, Ascorbic Acid and Extract/ Ascorbic Acid Co-administration on Serum Biochemical Parameters

The results of protective effect of aqueous extract of *Sesamum radiatum* and ascorbic acid against paracetamol –induced hepatotoxicity in rats are shown in Tables 1- 2 and Figures 1- 2. The Distilled water+ Paracetamol– treated group showed significant ($p < 0.05$) increase in serum hepatic enzyme levels (AST, ALT and ALP), when compared to control group (Distilled water- treated) (Table 1). Administration of aqueous extract of *Sesamum radiatum* leaves at doses of 200 and 400mg/kg, Ascorbic acid at a fixed dose of 100 mg/kg and co- administration of *Sesamum radiatum* leaves (200mg/kg)/ Ascorbic acid (100 mg/kg) decreased the activities of serum hepatic enzymes. When compared to the normal control

(Distilled water- treated), only the *Sesamum radiatum* leaves (200mg/kg)/ Ascorbic acid (100 mg/kg) co-administration showed no significant ($p > 0.05$) difference (Table 1). The Distilled water+ Paracetamol– treated group also showed significant ($p < 0.05$) increase in Tchol, TAG and LDL with a corresponding significant ($p < 0.05$) decrease in HDL when compared to control group (Distilled water-treated) (Table 2). Treatment with graded doses of aqueous extract of *Sesamum radiatum* leaves (200 and 400mg/kg), Ascorbic acid (100 mg/kg) and co-administration of *Sesamum radiatum* leaves (200mg/kg)/ Ascorbic acid (100 mg/kg) significantly ($p < 0.05$) decreased the serum concentration of Tchol., TAG, LDL and significantly ($p < 0.05$) increased the serum concentration of HDL. Only the aqueous extract of *Sesamum radiatum* leaves (200mg/kg)/ Ascorbic acid (100 mg/kg) co-administration showed no significant ($p > 0.05$) difference in the level of Total Chol., TAG, LDL and HDL when compared to the normal control (Table 2). The Distilled water+ Paracetamol– treated group showed significant ($p < 0.05$) decrease in total protein and albumin and levels (Figure 1) and significant increase in total and direct bilirubin (Figure 2) when compared to control group (Distilled water- treated). Treatment with graded doses of aqueous extract of *Sesamum radiatum* leaves (200 and 400mg/kg), Ascorbic acid (100 mg/kg) and co- administration of *Sesamum radiatum* leaves (200mg/kg)/ Ascorbic acid (100 mg/kg) also significantly ($p < 0.05$) increased the serum total protein and albumin levels (Figure 1) and significantly ($p < 0.05$) decreased the total bilirubin and direct bilirubin levels (Figure 2). Only the co-administration of the aqueous extract of *Sesamum radiatum* leaves (200mg/kg) and Ascorbic acid (100 mg/kg) showed no significant ($p > 0.05$) difference in the serum total protein, albumin, total bilirubin and direct bilirubin concentrations when compared to the normal control (Table 2).

Table 1: Effect of the Administration of Aqueous Extract of *Sesamum radiatum* Leaves, Ascorbic Acid and Extract/ Ascorbic Acid Co- administration on Serum Liver Enzymes

Treatment	AST (IU/L)	ALT (IU/L)	ALP (IU/L)
Group 1	43.49±5.06 ^a	33.17±2.77 ^a	33.17±5.25 ^a
Group 2	91.29±9.80 ^d	98.07±4.28 ^c	91.27±5.56 ^d
Group 3	77.71±5.27 ^c	69.61±9.47 ^b	70.10±3.25 ^c
Group 4	75.47±6.12 ^{bc}	70.16±6.66 ^b	71.33±4.21 ^c
Group 5	41.10±7.23 ^a	41.73±3.53 ^a	35.05±5.33 ^a
Group 6	74.33±4.41 ^{bc}	70.05±4.71 ^b	70.33±4.22 ^c

Data are presented as mean ± SD. Data was analysed by one- way ANOVA followed by Duncan post- hoc test for multiple comparisons, (n=5). Mean values having different lower case alphabets as superscripts are considered significant (p< 0.05) across the columns.

Group 1: Control received 1ml distilled water. Group 2: received 1ml distilled water +Paracetamol. Group 3: received 200 mg/ kg Extract + Paracetamol. Group 4 received 400 mg/ kg Extract + Paracetamol. Group 5: received 200 mg/ kg Extract + 100mg/kg Ascorbic acid +Paracetamol. Group 6: received 100mg/kg Ascorbic acid

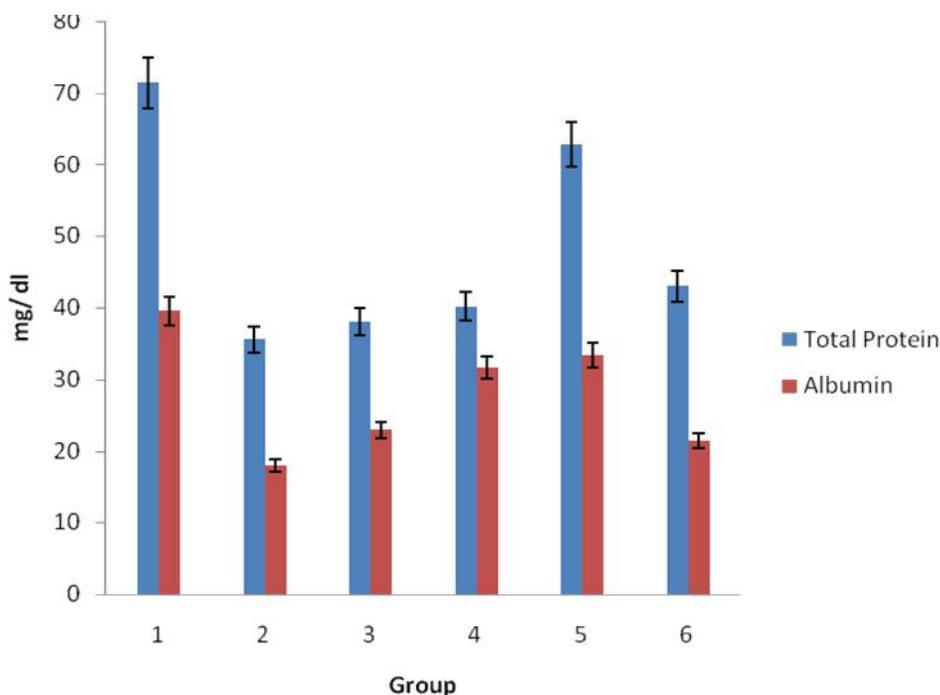


Figure 1: Effect of the Administration of Aqueous Extract of *Sesamum radiatum* Leaves, Ascorbic Acid and Extract/ Ascorbic Acid Co- administration on Serum Total Protein and Albumin Concentrations

Data are presented as mean ± SD. Data was analysed by one- way ANOVA followed by Duncan post- hoc test for multiple comparisons, (n=6).

Group 1: Control received 1ml distilled water. Group 2: received 1ml distilled water +Paracetamol. Group 3: received 200 mg/ kg Extract + Paracetamol. Group 4 received 400 mg/ kg Extract + Paracetamol. Group 5: received 200 mg/ kg Extract + 100mg/kg Ascorbic acid +Paracetamol. Group 6: received 100mg/kg Ascorbic acid

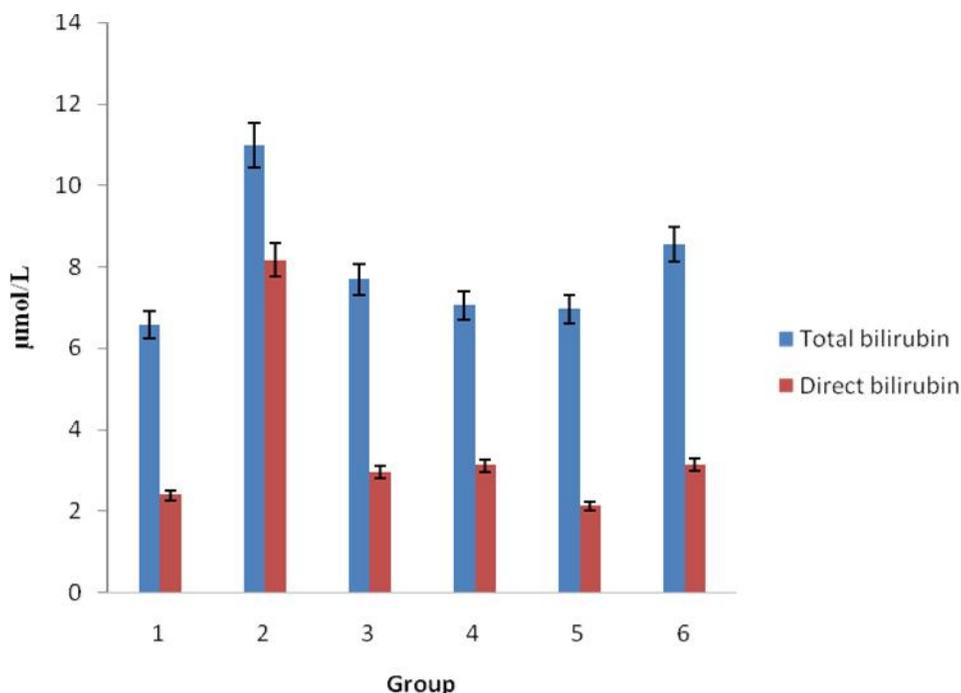


Figure 2: Effect of the Administration of Aqueous Extract of *Sesamum radiatum* Leaves, Ascorbic Acid and Extract/ Ascorbic Acid Co- administration on Serum Total Bilirubin and Direct Bilirubin Concentrations

Data are presented as mean \pm SD. Data was analysed by one- way ANOVA followed by Duncan post- hoc test for multiple comparisons, (n=6).

Group 1: Control received 1ml distilled water. Group 2: received 1ml distilled water +Paracetamol. Group 3: received 200 mg/ kg Extract + Paracetamol. Group 4 received 400 mg/ kg Extract + Paracetamol. Group 5: received 200 mg/ kg Extract + 100mg/kg Ascorbic acid +Paracetamol. Group 6: received 100mg/kg Ascorbic acid

Table 2: Effect of the Administration of Aqueous Extract of *Sesamum radiatum* Leaves, Ascorbic Acid and Extract/ Ascorbic Acid Co-administration on Serum Lipid Profile

Treatment	Tchol (mg/dl)	TAG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Group 1	79.13 \pm 4.48 ^a	72.33 \pm 4.77 ^a	50.26 \pm 3.15 ^e	14.40 \pm 7.80 ^a
Group 2	148.25 \pm 9.58 ^d	135.91 \pm 7.56 ^e	13.30 \pm 0.98 ^a	107.77 \pm 5.99 ^e
Group 3	119.13 \pm 9.99 ^c	116.78 \pm 7.30 ^d	33.17 \pm 4.16 ^b	62.60 \pm 9.08 ^d
Group 4	101.98 \pm 7.20 ^b	83.39 \pm 9.34 ^{ab}	47.97 \pm 5.27 ^{de}	37.33 \pm 6.98 ^{bc}
Group 5	82.17 \pm 7.43 ^a	83.05 \pm 9.41 ^{ab}	49.33 \pm 3.18 ^{de}	16.23 \pm 5.07 ^a
Group 6	102.45 \pm 5.40 ^b	106.18 \pm 7.77 ^{cd}	31.17 \pm 4.22 ^b	50.04 \pm 6.44 ^{cd}

Data are presented as mean \pm SD. Data was analysed by one- way ANOVA followed by Duncan post- hoc test for multiple comparisons, (n=6). Mean values having different lower case alphabets as superscripts are considered significant (p< 0.05) across the columns.

Group 1: Control received 1ml distilled water. Group 2: received 1ml distilled water +Paracetamol. Group 3: received 200 mg/ kg Extract + Paracetamol. Group 4 received 400 mg/ kg Extract + Paracetamol. Group 5: received 200 mg/ kg Extract + 100mg/kg Ascorbic acid +Paracetamol. Group 6: received 100mg/kg Ascorbic acid.

3.3 Histopathological Findings

Histopathological examination of the liver tissues of control group showed radially arranged plates of hepatocytes with no abnormalities (**Plate I**). Distilled water+ paracetamol –treated group showed severe hepatic lesions characterized by necrosis of hepatocytes around the central vein, collapsed central vein with signs of necrosis such as karyolitic, karyorrhexis, pyknosis and eosinophilia of the

cytoplasm (**Plate II**). Administration of 200 and 400 mg/kg aqueous extract of *Sesamum radiatum* leaves showed mild restoration of a normal architecture of liver (**Plates III and IV** respectively). Co-administration of 200 mg/kg aqueous extract of *Sesamum radiatum* leaves/ 100 mg/kg Ascorbic showed complete normalization of liver architecture (**Plate V**). Administration of 100 mg/kg Ascorbic showed mild restoration of the necrotized hepatocytes of (**Plate VI**).

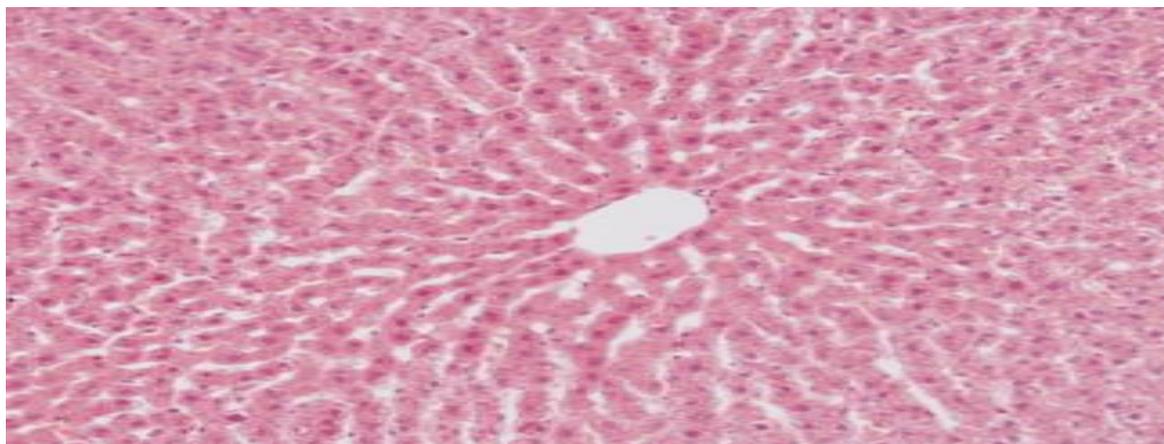


Plate 1: Micrograph of the liver of a normal control rat showing normal arrangement of the central vein, Sinusoids and the hepatocytes. (H and E stain, Mg. × 100)

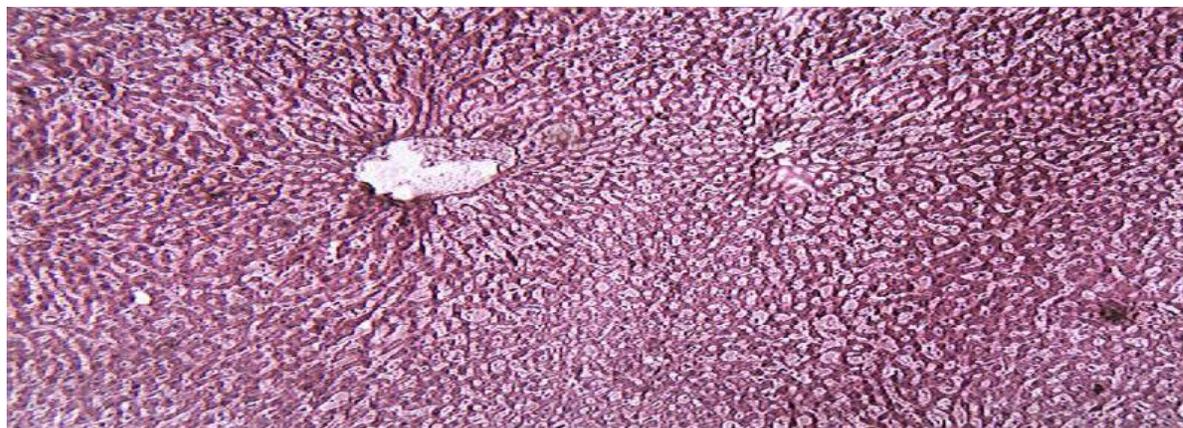


Plate II: Paracetamol induced hepatotoxicity showing centrilobular necrotic areas, hydropic changes, congested sinusoids and hepatocytes with pyknotic nucleus. (H and E stain, Mg. × 100)

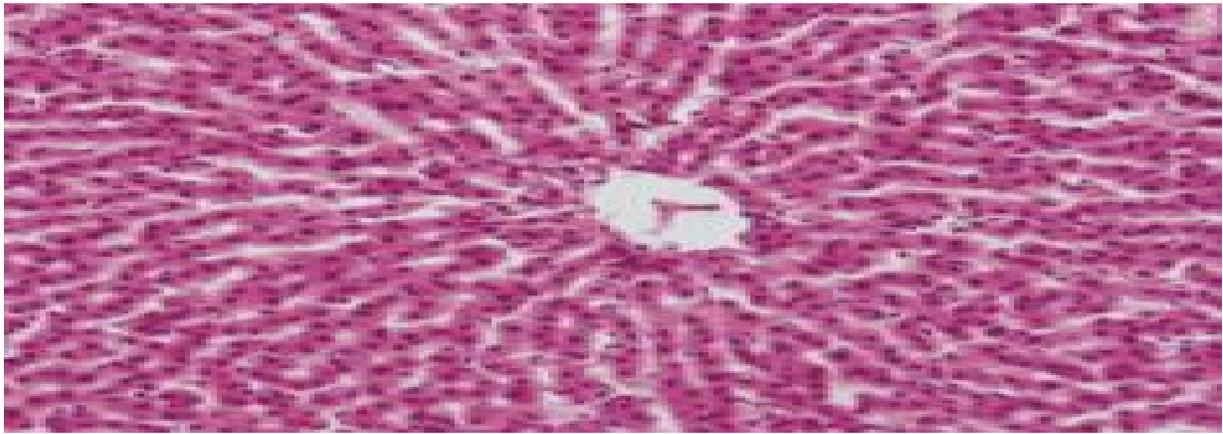


Plate III: Micrograph of the liver of a 200 mg/kg extract treated- group showing mild restoration of the necrotized hepatocytes. (H and E stain, Mg. \times 100)

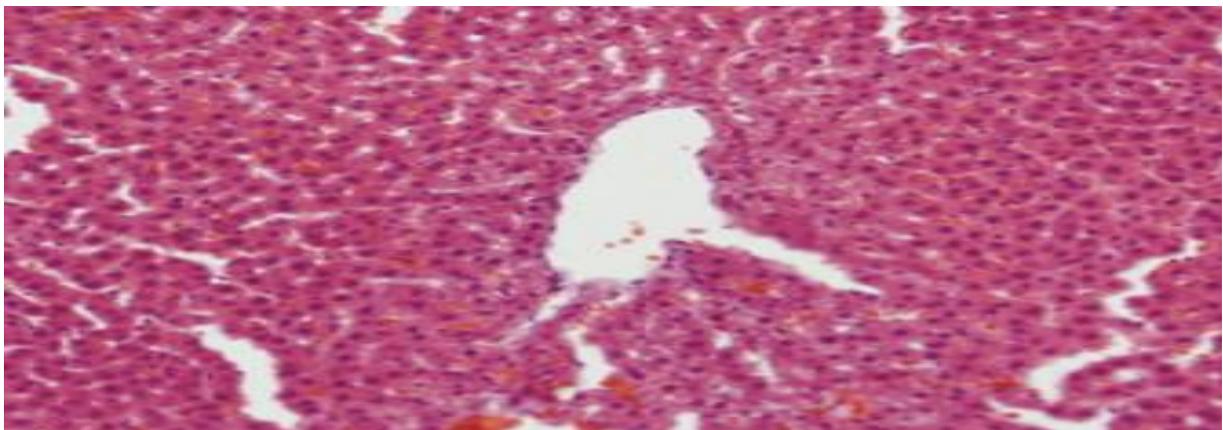


Plate IV: Micrograph of the liver of a 400 mg/kg extract treated- group showing mild restoration of the necrotized hepatocytes. (H and E stain, Mg. \times 100)

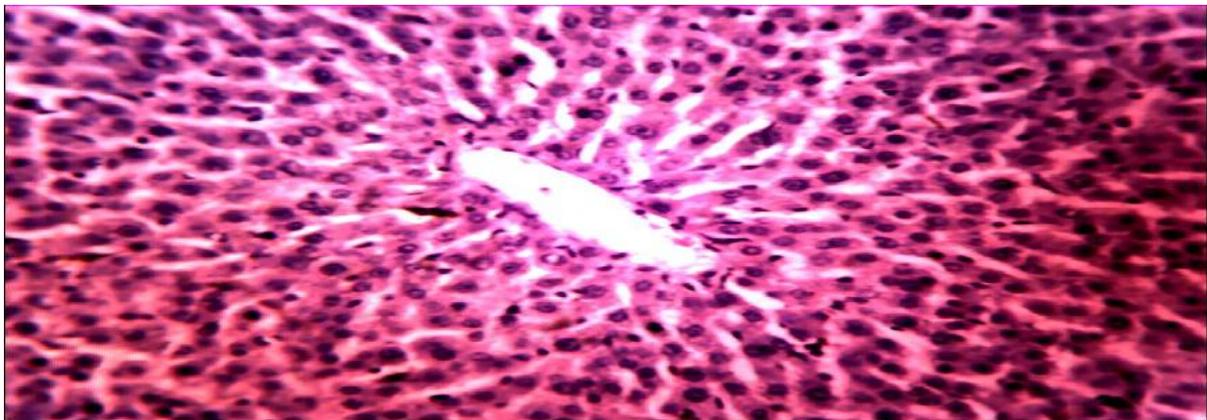


Plate V: Micrograph of the liver of a 200 mg/ kg Extract + 100 mg/kg Ascorbic acid (synergistic) Showing advanced normalization of liver architecture. (H and E stain, Mg. \times 100)

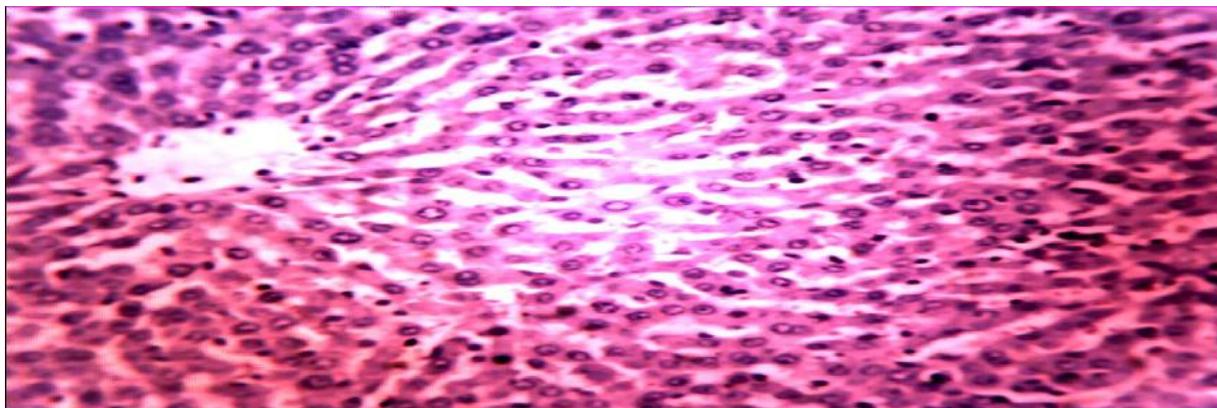


Plate VI: Micrograph of the liver of a 100 mg/kg Ascorbic acid treated- group showing mild restoration of a normal architecture of liver. (H and E stain, Mg. × 100)

4.0 Discussion

Liver is one of the largest organs in human body and the chief site for intense metabolism and excretion. Liver damage is usually associated with defects in its metabolic functions and this pose a challenge to international public health. Many orthodox drugs currently available for the treatment of liver diseases are inadequate and also cause serious side effects (Nyblom *et al.*, 2006). This has necessitated the search for alternative cures particularly traditional herbal medicines that possess hepatoprotective activity. It is generally believed that a single drug cannot be effective for all types of severe liver diseases. Therefore, an effective combination of medicinal plants or medicinal plant and orthodox drug is required. Hepatoprotective activity can be most easily evaluated with the aid of several model systems of liver damage in experimental animals. In this study, the paracetamol- induced hepatotoxicity model was employed.

Protection against paracetamol-induced toxicity has been used as a test for potential hepatoprotective activity by several investigators (Visen *et al.*, 1993). Paracetamol (Acetaminophen) is a widely used analgesic and antipyretic. Adverse effects are usually not common with paracetamol within the therapeutic dose range of 2g - 4g mg taken as three to four separate doses in a day (Koch- weser, 1976). Despite this high margin of safety, paracetamol toxicity remains the leading cause of drug-induced liver failure in the United States (Lee, 2003). Single doses of paracetamol in humans around 15 g carry a great risk of hepatotoxicity (Ameer and Greeblath, 1977),

although doses of as little as 6.2 g may result in liver damage (Bailey, 1980). Overexposure to paracetamol results in fulminating centrilobular hepatic necrosis, which can bridge to the periportal regions of the liver lobule (Mitchel *et al.*, 1973). Paracetamol is primarily metabolized by conjugation with sulfate and glucuronic acid (Cummings *et al.*, 1967) while a small percentage of the dose undergoes bioactivation by cytochrome P450 enzymes to the reactive intermediate N-acetyl *p*-benzoquinoneimine (NAPQI) (Miner and Kissinger, 1979). At non-toxic doses, NAPQI is eliminated from the liver after conjugation with reduced glutathione (GSH) However, at toxic doses; the two main conjugation pathways for paracetamol become saturated, resulting in increased formation of NAPQI. Consequently, detoxification of NAPQI is compromised when existing stores of GSH have been depleted and NAPQI then binds to cellular macromolecules, initiating cell death pathways (Jollow *et al.*, 1973; Potter *et al.*, 1974). This study investigated the possible protective effect of aqueous extract of *Sesamum radiatum* leaves against paracetamol- induced hepatotoxicity and a possible synergistic effect when co-administered with ascorbic acid.

The results of acute toxicity studies revealed that the aqueous extract of *Sesamum radiatum* leaves caused no mortality or physical changes in skin and fur, eyes and mucus membrane, respiratory rate, circulatory signs, autonomic and central nervous system effects in rats up to a dose of 5000 mg/kg. This is an indication that the plant is safe for acute consumption though more studies is required to ascertain its safety when consumed over a long period of time.

Serum ALT, AST, ALP, total and direct bilirubin in plasma have been reported to be a sensitive indicator in liver injury (Molander *et al.*, 1955). Lipid profile, total protein and albumin are also useful markers to ascertain the functional capability of the liver. The decrease in ALT, AST and ALP activities by the aqueous extract of *Sesamum radiatum* leaves, Ascorbic acid and the extract/Ascorbic acid co-administration may be an indication of regeneration process and repair of hepatic tissue damage induced by paracetamol. All the treatments also decreased the elevated total and direct bilirubin levels. Bilirubin, a metabolic product of hemoglobin, undergoes conjugation with glucuronic acid in hepatocytes to increase its water solubility. Determination of serum bilirubin represents an index for assessment of hepatic function, and any abnormal increase in the levels of serum bilirubin indicates hepatobiliary diseases and severe disturbance of hepatocellular function (Martin and Friedman, 1992). Decreased serum bilirubin level following extract treatment indicated the effectiveness of the extract in restoring normal functional status of the liver. The aqueous extract of *Sesamum radiatum* leaves, Ascorbic acid and the extract/Ascorbic acid co-administration also increased the serum level of total protein and albumin. This is suggestive of the stabilization of endoplasmic reticulum leading to the repair of impaired protein synthesis caused by paracetamol with concurrent improvement of Kupffer cells (Suresh Kumar and Mishra, 2008). Paracetamol-induced toxicity might have affected the metabolism of lipids in the liver as suggested by the increased cholesterol levels of rats. Alteration of bio-membrane lipid profile disturbs its fluidity, permeability, activity of associated enzymes and transport system (Cooper *et al.*, 1977) and this could affect lipid transport in the liver. The elevated levels of cholesterol, triglyceride and LDL and decreased level of HDL observed after paracetamol administration was reversed by the protective activity of the extract and ascorbic acid.

Ascorbic acid is considered as highly effective antioxidant. A synergistic effect exists between the extract and vitamin C as seen in the results. The co-administration afforded the best protective effect.

Our observation in the histological examination of hepatic tissues further validates the result of the biochemical studies. There was a dose dependent restoration of the normal architecture of liver tissues of rats administered graded doses of the extract and ascorbic acid. However, a near normal restoration of liver architecture was observed in the liver tissues of rats that received the extract and ascorbic acid co-administration. The mechanism by which the extract induces its hepato-protective activity against oxidative damage caused by paracetamol is not certain. However, it is possible that polyphenolic compounds worked in synergy with ascorbic acid to provide this protection.

5.0 Conclusions

From the present results we could conclude that aqueous extract of *Sesamum radiatum* leaves possesses a promising hepatoprotective effect against paracetamol hepatic damage and when co-administered with ascorbic acid, it provides a synergistic protective effect that could be useful in the management of liver diseases.

References

- Ademuyiwa, O., Adesanya, O. and Ajuwon, O. R. 1994. "Vita-min C in CC14 Hepatotoxicity-A Preliminary Report," *Human Experimental Toxicology*, Vol. 13, No. 2 , pp. 107-109.
- Akpan-Iwo G, Idowu, A. A, Misari, S. M. 2006. Collection and evaluation of sesame (*Sesamum* spp.) germplasm in Nigeria. IGPR/FAO, 142: 59-62.
- Ambali, S. F., C. Onukak, S. B. Idris, L. S. Yaqub, M. Shittu, H. Aliyu and M. U. Kawu. 2010. "Vitamin C Attenuates Short-Term Hematological and Biochemical Alterations Induced by Acute Chlorpyrifos Exposure in Wistar Rats," *Journal of Medicine and Medical Sciences*, Vol. 1, No. 10, pp. 465-477.
- Ameer, B. and Greenblatt, D. J. 1977. Acetaminophen. *Annals of Internal Medicine*; 87:202-209.

- Bailey, B. O. Acetaminophen hepatotoxicity and overdose. 1980. *American Family Physician*; 22:83-87.
- Bashandy S. A., and Alwasel S. H., 2011. "Carbon Tetrachloride-Induced Hepatotoxicity and Nephrotoxicity in Rats: Protective Role of Vitamin C," *Journal of Pharmacology and Toxicology*, Vol. 6, No. 3, pp. 283-292.
- Boyd, E. H, and Berezky GM. 1966. Liver necrosis from paracetamol. *British Journal of Pharmacology*; 26: 606-14.
- Cooper, R. A, Durocher, and J. R, Leslie, M. H. 1977. Decreased fluidity of red cell membrane lipids in a beta lipoproteinemia. *Journal of Clinical Investigation*; 60: 115-121.
- Cummings, A. J., King, M. L. and Martin, B. K. 1967. A kinetic study of drug elimination: the excretion of paracetamol and its metabolites in man. *British Journal of Pharmacology*; 29: 150-7.
- Fassati, P. and Prencipe, L. 1982. Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. *Clinical Chemistry*, 28: 2077.
- Fredrikson, D. S.; Levi, R. L. and Less, R. S. 1967. Determination of cholesterol. *The New England Journal of Medicine*, 276: 148 – 156.
- Gills L.,S (1992). Ethnomedical uses of plants in Nigeria, University of Benin Press, Benin, p. 212.
- Grajeda-Cota, P., M. V. Ramírez-Mares and E. González de Mejía, 2004. "Vitamin C Protects against *in Vitro* Cytotoxicity of Cypermethrin in Rat Hepatocytes," *Toxicology in vitro*, Vol. 18, pp. 13-19.
- Henry, R. J. 1964. Clinical Chemistry. Colorimetric determination of total protein. In: Harper and Row Publ., New York, p 181.
- Hutchinson J, Dalziel J. M. 1954. Flora of Tropical Western Africa, 2nd Edition, volume 1, Part 1. Crown Agents for Overseas Government and Administration, Millbank, London.
- Jendrassik, L. 1938 . Determination of bilirubin. *Biochemical Journal*. 7297: 7281.
- Jollow, D. J., Mitchell, J. R., Potter, W. Z., Davis, D. C., Gillette, J. R. and Brodie, B. B. 1973. Acetaminophen-induced hepatic necrosis. II. Role of covalent binding *in vivo*. *Journal of Pharmacology and Experimental Therapeutics*; 187:195-202.
- Kataoka, H., Y. Nishiyama, K. Yamato, J. Teraoka, Y. Morii, A. Sakoda, Y. Ishimori, T. Taguchi and K. Yama-oka, "Comparative Study on the Inhibitory Effects of Antioxidant Vitamins and Radon on Carbon Tetrachloride- Induced Hepatopathy," *Journal of Radiation Research*, 2012, pp. 1-10.
- Koch-Weser, J. Drug therapy. Acetaminophen. 1976. *New England Journal of Medicine*; 295:1297-300.
- Lee, W. M. Acute liver failure in the United States. *Semin Liver Dis.*2003; 23:217-26. Livingstone, New York.1999; 195-212.
- Lorke, D. 1983. "A new Approach to Practical Acute Toxicity Testing." *Archives of Toxicology* 54: 275-287.
- Martin P., and Friedman L. S. 1992. Assessment of liver function and diagnostic studies. In: Friedman LS, Keefe EB. (Eds.), *Hand Book of Liver Disease*. Churchill Livingstone, Philadelphia, pp. 1–14.
- Miner, D. J. and Kissinger, P. T. 1979. Evidence for the involvement of N-acetyl-p- quinoneimine in acetaminophen metabolism. *Biochemical Pharmacology*; 28:3285-90.
- Mitchell, J. R., Jollow, D. J., Potter, W. Z., Davis, D. C., Gillette, J. R. and Brodie, B. B. Acetaminophen-induced hepatic necrosis 1973. Role of drug metabolism. *Journal of Pharmacology and Experimental Therapeutics*; 187:185-94.
- Molander, D. W, Wroblewski, F, La-Due J. S. 1955. Transaminase compared with cholesterase and alkaline phosphatase an index of hepatocellular integrity. *Clinical Research Proceedings*;3: 20-4.
- Nyblom, H., Björnsson, E., Simrén, M., Aldenborg, F., Almer, S., and Olsson, R. 2006. The AST/ALT ratio as an indicator of cirrhosis in patients with PBC. *Liver International*. 26 (7): 840–845.
- Oyinbo, C. A., W. N. Dare, G. R. A. Okogun, L. C. An-yanwu, N. M. Ibeabuchi, C. C. Noronha and O. A. Okanlawon, 2006. "The Hepatoprotective Effect of Vitamin C and E on Hepatotoxicity Induced by Ethanol in Sprague Dawley Rats," *Pakistan Journal of Nutrition*, 5: 6; 507-511.
- Potter, W. Z., Thorgeirsson, S. S., Jollow, D. J. and Mitchell, J. R. Acetaminophen-induced hepatic necrosis. V. 1974. Correlation of hepatic necrosis, covalent binding and glutathione depletion in hamsters. *Pharmacology*; 12:129-43.
- Purseglove J. W (1974). *Tropical Crops, Dicotyledons*, Longman, London, pp. 430-435.
- Reitman S, Frankel S. Colorimetric method for determination of serum transaminases. *Am J Clin Pathol* 1957;28:56–61.
- Sheehan, D.C. and Hrapchak, B.B. (1980). *Theory and Practice of Histotechnology*. 2nd Ed. The C.V. Mosby company, St. Louis, 205, London, UK.

- Suresh Kumar, S. V, Mishra, S. H. 2008. Hepatoprotective effect of *Pergularia daemia* (Forsk.) ethanol extract and its fraction. *Indian Journal of Experimental Biology*; 46:447- 52.
- Visen, P. K. S., Shukla, B., Patnaik, G. K., Dhawan, B. N. 1993. Andrographolide protects rat hepatocytes against paracetamol-induced damage. *Journal of Ethnopharmacology*; 40:131–136.
- Zimmerman, H. J, Ishak, K. G, MacSween, R, Anthony, P. P, Scheuer, P. J, Burt, A. D. 1994. Hepatic injury due to drugs and toxins, in *Pathology of Liver*. Edinburgh: Churchill Livingstone; 1994. p. 563-634.

Access this Article in Online	
	Website: www.ijarbs.com
	Subject: Biochemistry
Quick Response Code	
DOI: 10.22192/ijarbs.2018.05.06.010	

How to cite this article:

NWEJE- ANYALOWU Paul Chukwuemeka, IDAKWOJI Precious Adejoh, OKAFOR Stephen Chiadikaobi, AJIMA Judith Nnedimkpa, DARA Amarachi Ngozika. (2018). Synergistic Protective Effects of Aqueous Extract of *Sesamum radiatum* Leaves and Ascorbic acid against Paracetamol- induced Hepatotoxicity in Rats. *Int. J. Adv. Res. Biol. Sci.* 5(6): 106-117.

DOI: <http://dx.doi.org/10.22192/ijarbs.2018.05.06.010>