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Hepatoprotective Potential of Alkaloid Extracts from *Vitex doniana* and *Ficus thonningii* Leaves in Alloxan-induced Diabetic Rats

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Authors' contributions

This work was carried out in collaboration among all authors. Author OCN conceptualized and designed the study. Author AIA carried out the analyses of the study and also wrote the manuscript. Author AUM managed the literature searches, author NOO managed the statistical analysis while author OLO wrote the protocol of the study. All authors read and approved the final manuscript

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ABSTRACT

Aim: This study sought to evaluate the hepatoprotective potential of alkaloid extracts from *V. doniana* and *F. thonningii* leaves on alloxan-induced diabetic rats.
Materials and Methods: Fresh leaves of *V. doniana* and *F. thonningii* were obtained from a local market Nkwagu in Abakaliki local government area of Ebonyi State. They were dried and pulverized to fine granules using manual grinder. Crude alkaloid was extracted using standard method. The acute oral toxicity of both plants was determined. Forty adult male albino rats were induced intraperitoneally with alloxan. The rats were grouped into eight groups of five animals per group: Groups 1 and 2 were treated with 200 and 400 mg/kg body weight of *V. doniana* respectively, groups 3 and 4 were treated with 200 and 400 mg/kg body weight of *F. thonningii* respectively, group 5 was treated with 100 mg/kg of *V. doniana* + 100 mg/kg of *F. thonningii*, group 6 was treated with 200 mg/kg of *V. doniana* + 200 mg/kg of *F. thonningii*, group 7 animals were not

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induced with alloxan and untreated (normal control) while group 8 were induced with alloxan but not treated (diabetic control). The extracts were administered to the animals orally for 30 days. Biochemical analyses were assayed using standard methods.
Results: Alloxan-induced diabetes greatly caused hepatotoxicity but was ameliorated by alkaloid extracts from *V. doniana* and *F. thonningii* leaves.
Conclusion: Results showed that alloxan induction damaged the liver but the effects were ameliorated by extracts of *V. doniana* and *F. thonningii*, thus, making both plants hepatoprotective. However, extract of *F. thoningii* seems to be more potent.

Keywords: Alloxan-induced diabetes; Ficus thoningii; hepatoprotective potential; Vitex doniana.

1. INTRODUCTION

Diabetes Mellitus (DM) is a chronic disease caused by inherited and/or acquired deficiency in production of insulin by the pancreas, or by ineffectiveness of insulin produced. Such a deficiency results in increased concentration of glucose in the blood, which in turn damages many of the body's systems in particular the blood vessels and nerves [1]. As the number of the people with diabetes multiplies worldwide, the disease has taken an ever-increasing share of national and international health care budgets. It is projected to become one of the world's main disablers and killers within the next 25 years. Regions with greatest potential are Asia and Africa, where DM rates could rise to two-to-three-folds compared with the present rates. Apart from currently available therapeutic options, many herbal medicines have been recommended for the treatment of diabetes.

Liver is the major organ which plays key roles in processing critical biochemical and physiological phenomena including metabolism and detoxification of endogenous and exogenous compounds, such as drugs and xenobiotics, homeostasis, growth, energy and nutrient supply [2]. Hepatic injury could occur by hepatotoxic agents such as drugs, alcohol, hydrocarbon and viral infections [2]. Liver diseases like jaundice, cirrhosis and fatty liver have been public health concern across the world [3]. Prevalence of chronic liver disease worldwide is 18.5% and cirrhosis is 4.5 to 9.5% while 2 million people die each year. In terms of medication, conventional or synthetic drugs are limited. Moreover they can have serious side effects [2]. Due to this fact, a huge number of medicinal plants have been used to figure out hepato-protective activities [4]. Approximately 160 phytochemical constituents originated from 101 plants have been reported to be potentially hepato-protective [2]. At present, medicinal plants have been a vital source of treatment of liver [4].

Alkaloids, widely existing in natural plants, are compounds containing nitrogen atoms. Most alkaloids are pharmacologically active ingredients in many medicinal plants due to their significant physiological activity. Many alkaloids can be extracted from natural plant materials and purified by modern separation techniques [5]. Recent studies showed that simple alkaloid extracts have efficacy against viruses including *herpes simplex* [6] and human immunodeficiency virus (HIV) and tumors [7].

Vitex doniana (Fig. 1) a member of Verbenaceae family is a medium-sized deciduous tree with a heavy rounded crown and a clear bole up to 5m. It is widely distributed in the Eastern and Western parts of Nigeria. The plant commonly called Black plum (English), Dinya (Hausa), Oriri (Yoruba), and Ucha koro (Igbo), is a deciduous ever green tree, usually 4-8 metres high occasionally up to 15 metres with a dense rounded crown [8]. Medical applications include treatment against mental illness, rheumatism, as anthelmintic and tranquilizer, gastrointestinal disturbance, urinary ailments and so on [9]. Also Kilani [10] have assessed the stem bark of *V. doniana* for antibacterial activity and establish its efficacy in the management of dysentery and gastroenteritis infections. The root is used for treatment of gonorrhoea, and women drink a decoction of it for backaches [9]. Phytochemical analysis of the various parts of the plant extract revealed the presence of saponin, tannins, phenols cardiac glycosides, flavonoids, sterols and triterpenes as well as high concentration of sodium, Potassium, Calcium, Iron, Phosphorus and Sulphur [11]. Furthermore, Njoku et al. [12] has reported the antidiabetic potential of its leaves on alloxan-induced diabetic rats.

Ficus thoningii (Fig. 2) also known as common wild fig is an evergreen tree with a rounded to spreading, dense crown; it can grow 6 - 21 metres tall [13]. The plant often begins life as an epiphyte, growing in the branch of another tree; as it grows older it sends down aerial roots which, when they reach the ground quickly form roots and become much thicker and more vigorous. They supply nutrients to the fig, allowing it to grow faster than the host tree. The aerial roots gradually encircle the host tree, preventing its main trunk from expanding, whilst at the same time the foliage smothers the foliage of the host. Eventually the host dies, leaving the fig to carry on growing without competition. It eventually becomes a stilt-rooted, banyan-like tree with multiple ascending trunks and massive wide-spreading branches [14]. The bark is important in local medicine, and it is used in treating colds, sore throat, dysentery, wounds, constipation, nosebleed and to stimulate lactation. Extracts of the bark are used in baths as a treatment of nervous illnesses, tuberculosis, paralysis and leprosy. The latex is used for wound fever [15]. The milky latex is dropped into the eye to treat cataracts. An infusion of the root and fibre is taken orally to help prevent abortion. The powdered root is taken in porridge to stop nose bleed. Furthermore, Njoku et al. [12] has reported the antidiabetic potential of its leaves on alloxan-induced diabetic rats.

Fig. 1. *Vitex doniana* leaves [12]

Fig. 2. *Ficus thoningii* leaves [12]

Diabetes mellitus is a chronic disease with serious health complications and is now one of the most common non-communicable diseases globally. It is the fourth leading cause of death in most developed countries. Recently, there has been a growing interest in anti-diabetic agents from natural products especially those derived from plants. Alloxan-induced diabetes has been reported to compromise hepatic integrity [16,17]. Therefore, this study sought to evaluate the hepatoprotective potential of alkaloid extracts from *V. doniana* and *F. thonningii* leaves on alloxan-induced diabetic rats.

2. MATERIALS AND METHODS

2.1 Extraction of Alkaloid from the Plants

Fresh leaves of *V. doniana* and *F. thonningii* were obtained from a local market Nkwagu in Abakaliki local government area of Ebonyi State. They were identified by Prof. F. N. Mbagwu, a plant taxonomist at the Department of Plant Science and Biotechnology, Imo State University, Owerri, Nigeria. They were washed thoroughly in running water to remove contaminants. They were separately air-dried at room temperature for two weeks. The dried leaves were pulverized to fine granules using manual grinder. Crude alkaloid was extracted according to Obadoni and Ochuko [18]. 500 g of each plant leaf sample was weighed separately into a 250 ml beaker and 200 ml of 10 % acetic acid in ethanol was added and covered and allowed to stand for 6hrs. This was filtered with Whatman filter paper No.1. The extract was concentrated on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added to the extract drop wisely until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The separate residues were further tested using Dragondoff's reagent which gave an orange colour solution to confirm the presence of alkaloid. The extracts were dried and stored in the refrigerator for further analysis.

2.2 Acute Oral Toxicity Studies (LD_{50})

The acute oral toxicity study was conducted using the limit doses test of up and down procedure according to organization for economic and cultural development [19,20]. Fifteen Wistar rats weighting between 150 and

200 g were used for this experiment. They were acclimatized for 7 days and grouped into 5 of 3 rats each. Animals in group A were administered 1000 mg/kg *V. doniana*, those in group B were administered 3000 mg/kg *V. doniana*, those in group C were administered 1000 mg/kg *F. thoningii*, those in group D were administered 3000 mg/kg *F. thoningii*, while those in group E were administered 1500 mg/kg *V. doniana* + 1500 mg/kg *F. thoningii*. They were observed for 48 hours, and thereafter for 7 days. Animals were weighed daily and recorded. They were observed for signs of acute toxicity, weight changes, morbidity and mortality. The behavioral changes and other changes observed in the experimental rats were recorded according to OECD [19] guidelines.

2.3 Experimental Design

A total of 40 male albino rats with body weight ranging from 160 to 180 g were used for this study. They were acclimatized for seven days to Laboratory condition. They were kept in plastic cages and fed with commercial rat chow and supplied with water *ad libitum.* The rats were used in accordance with NIH Guide for the care and use of laboratory animals; NIPRD Standard Operation Procedures (SOPs). After the acclimatization period, the rats were injected with alloxan monohydrate dissolved in sterile normal saline in a dose of 120 mg/kg body weight intraperitoneally [21]. After 72 hours of the injection, rats with fasting blood glucose (FBG) at or above 200 mg/dL were considered diabetic.

2.4 Grouping of Animals

The animals were randomly assigned to 8 groups of 5 rats each, and treated as follows:

Group 1: Diabetic rats treated with 200 mg/kg body weight of *V. doniana* (VD_1).
Group 2: Diabetic rats treated with 400 mg/kg body weight of *V. doniana* (VD_2).
Group 3: Diabetic rats treated with 200 mg/kg body weight of *F. thonningii* (FT_1).
Group 4: Diabetic rats treated with 400 mg/kg body weight of *F. thonningii* (FT_2).
Group 5: Diabetic rats treated with 100 mg/kg body weight of *V. doniana* and100 mg/kg body weight of *F. thonningii* ($VD+FT_1$).
Group 6: Diabetic rats treated with 200 mg/kg body weight of *V. doniana* and 200 mg/kg body weight of *F. thonningii* ($VD+FT_2$).
Group 7: (Normal control) which received feed and water only.
Group 8: (Diabetic control) which was induced with alloxan without treatment**.**

Weight of animals was recorded at intervals. After 30 days of orally administering the animals daily with the plant alkaloid extracts, the animals were fasted overnight and sacrificed. Blood samples for biochemical analyses were collected through ocular puncture and gently dispersed into plain sample bottles. The Sera samples were obtained after clotting of the blood via centrifugation**.**

2.5 Determination of Biochemical Parameters

Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) activities were determined using Randox commercial Enzyme kits according to the method of Reitman and Frankel [22]. Alkaline Phosphatase (ALP) activity was determined by Phenolphthalein Monophosphate method described by Babson et al*.,* [23]. Total Protein concentration was carried out using Biuret method described by Henry et al. [24]. Estimation of albumin was done by bromocresol green (BCG) method described by Doumas et al. [25]. Total bilirubin concentration was determined by diazo method described by Royden and Alfred [26].

2.6 Statistical Analysis

Data were subjected to analysis of variance using Graph Pad Prism (version 6). Results were presented as Mean ± Standard deviation. One way analysis of variance (ANOVA) was used for comparison of the means. Differences between means were considered to be significant at $p<0.05$.

3. RESULTS

Response and effect on the body weight of rats treated for 7 days with 1000 $mgkg^{-1}$and 3000 mg kg^{-1} leaf extracts of *V. doniana* and *F. thoningii* is shown in Table 1. The results showed that the high doses induced progressive and sustained weight loss from 13.71%-16.84% after 7 days of oral administration (Table 1). The results showed that all rats treated with the 3000 $mgkg^{-1}$ limit dose of the leaf extracts were hypo-reactive to external stimuli such as touch in the first 30 min

to 1hr post administration and subsequently became active and exhibited normal behavior throughout the 7 days observation period. The limit test dose of 3000 mgkg-1 did not cause any mortality or any major acute toxicity. Thus, the LD_{50} of the leaf extracts is greater than 3000 mg/kg.

4. DISCUSSION

Evaluation of liver function is very important when analyzing toxicity of drugs and plant extracts because of its relevance for the survival of the organism [27]. Increased activities of alkaline phosphatase (ALP), aspartate transaminase (AST) and alanine transaminase (ALT) have been reported to be an indicator of hepatotoxicity or liver damage/diseases [2]. Studies on the alterations of these enzymes might reflect the metabolic abnormalities and cellular injuries in some organs. The liver and kidney have extremely important function in detoxification and excretion of metabolic wastes and xenobiotics [3,27]. Exposure to toxic chemicals causes alterations in some tissue enzyme activities [4].

The results of acute toxicity study indicated that the LD_{50} of the leaf extracts of *V. doniana* and *F. thoningii* is greater than 3000 mg/kg body weight (Table 1). The limit test dose is primarily used in situations where the experimenter has information indicating that the test material is likely to be non-toxic or of low toxicity [19]. Thus, the non-lethal effects produced with the high doses of this extracts is an indication that the leaf extracts of *V. doniana* and *F. thoningii* are relatively safe on acute oral exposure. It can therefore be concluded that *V. doniana* and *F. thoningii* leaf extracts is non-toxic which is in agreement with the American society for testing and materials [28], that any chemical substance with LD_{50} estimate greater than 3000-5000 mg/kg (oral route) could be considered of low toxicity and safe. OECD [19] also recommended the use of limit test dose with LD_{50} greater than 5000 mg/kg (oral route) as having low acute oral toxicity. This implies that the leaf extracts of both plants are relatively safe.

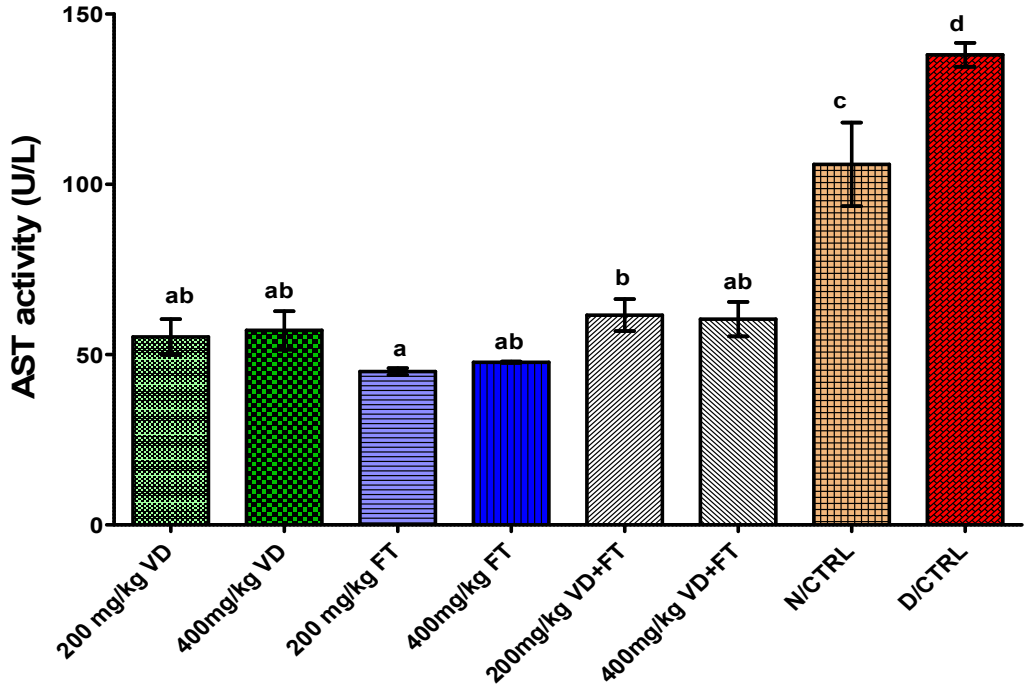

Fig. 3. Aspartate aminotransferase (AST) activity (U/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant ($p<0.05$); Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

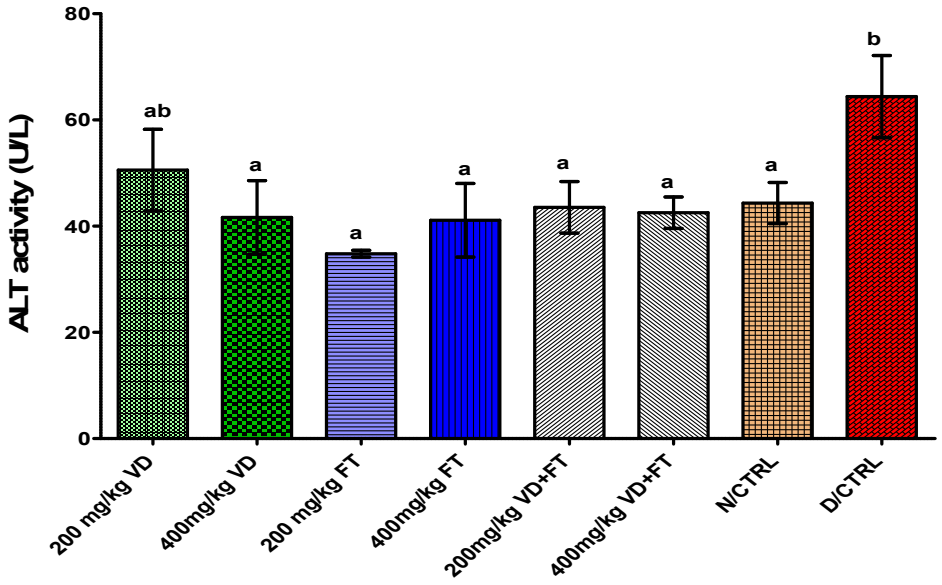

Fig. 4. Alanine aminotransferase (ALT) activity (U/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant (p<0.05); Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

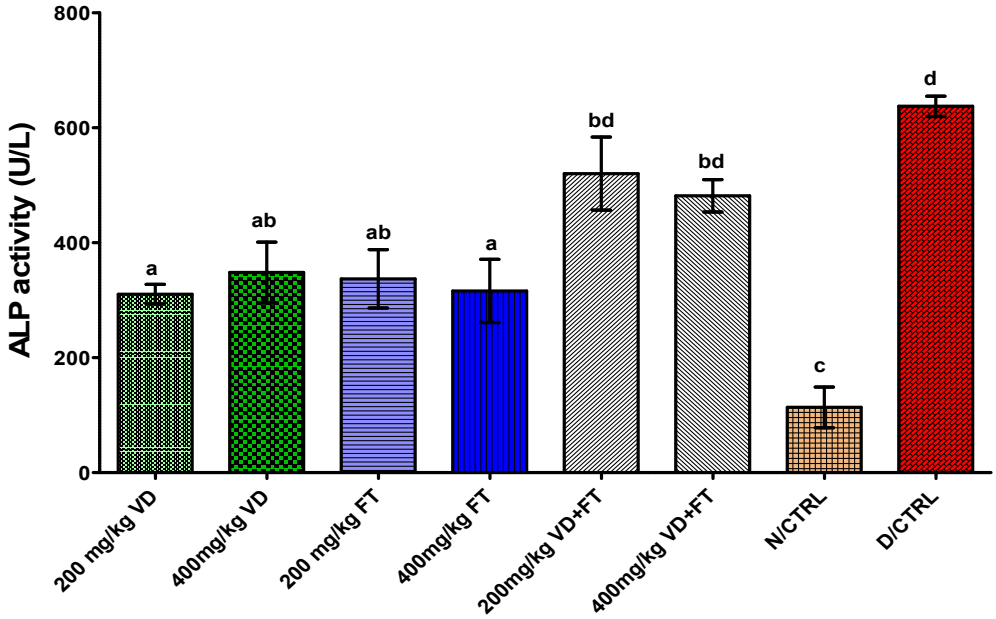

Fig. 5. Alkaline Phosphatase (ALP) activity (U/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant (p<0.05); Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

Table 1. Acute oral toxicity of rats exposed to *V. doniana* and *F. thoningii*

Animal Groups	Treatment	Changes in body weight from day 0 to day 7 (%)	Number of Death recorded
A	1000 mg/kg of *V. doniana*	16.14	0
B	3000 mg/kg of *V. doniana*	13.73	0
C	1000 mg/kg of *F. thoningii*	13.71	0
D	3000 mg/kg of *F. thoningii*	16.84	0
E	1500 mg/kg of *V. doniana* + 1500 mg/kg of *F. thoningii*	15.50	0

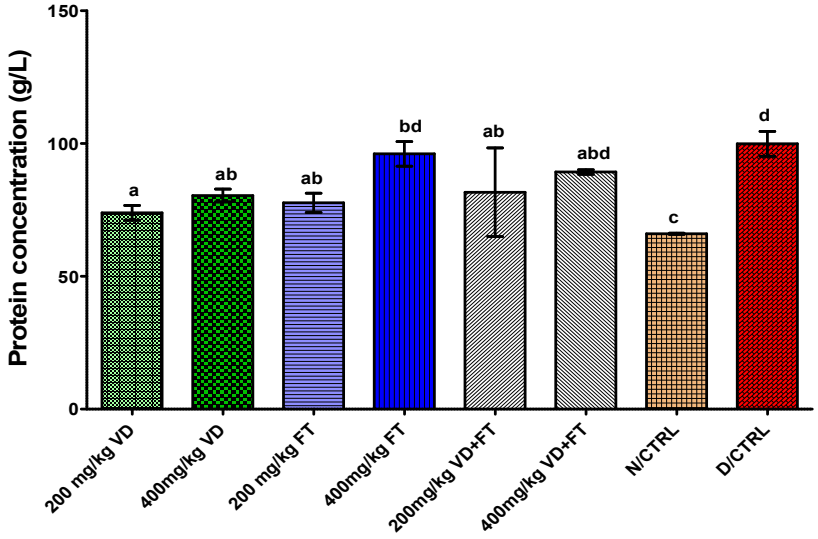

Fig. 6. Total protein concentration (g/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant ($p<0.05$); Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

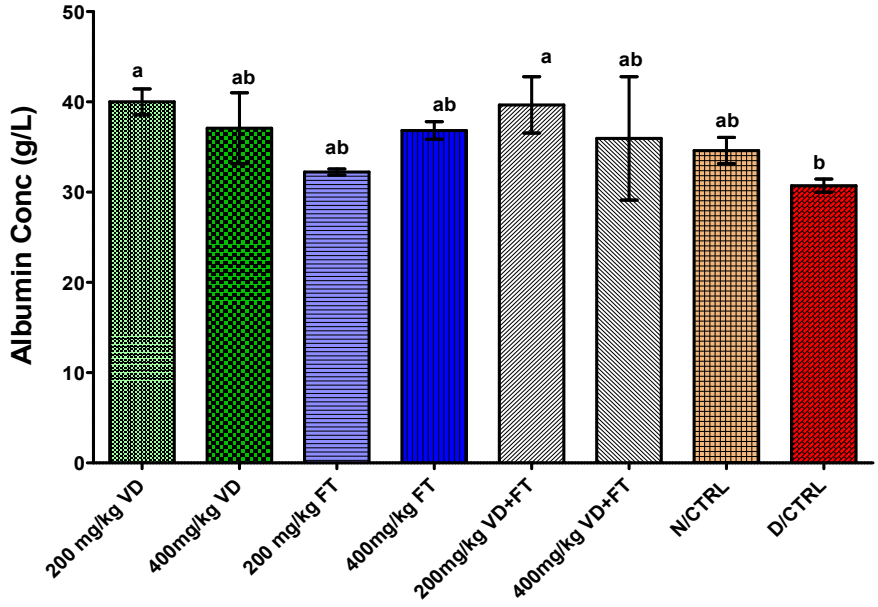

Fig. 7. Albumin concentration (g/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant (p<0.05).Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

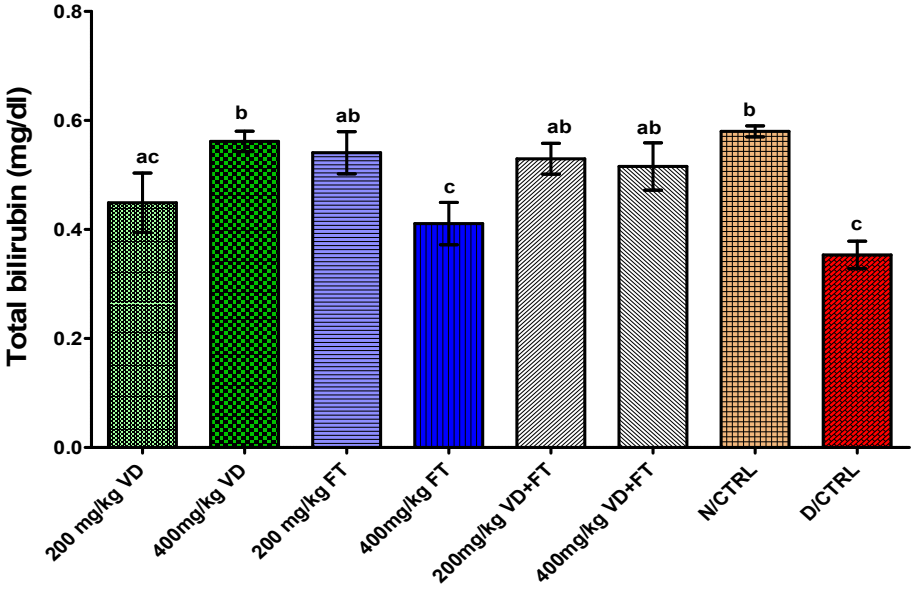

Fig. 8. Total Bilirubin concentration (g/L) of diabetic albino rats administered single and combined doses of alkaloid extracts of *V. doniana* (VD) and *F. thoningii* (FT)

Results are presented as mean ± standard deviation. Bars bearing different letter(s) are statistically significant (p<0.05); Legend: N/CTRL = Normal Control; D/CTRL = Diabetic Control

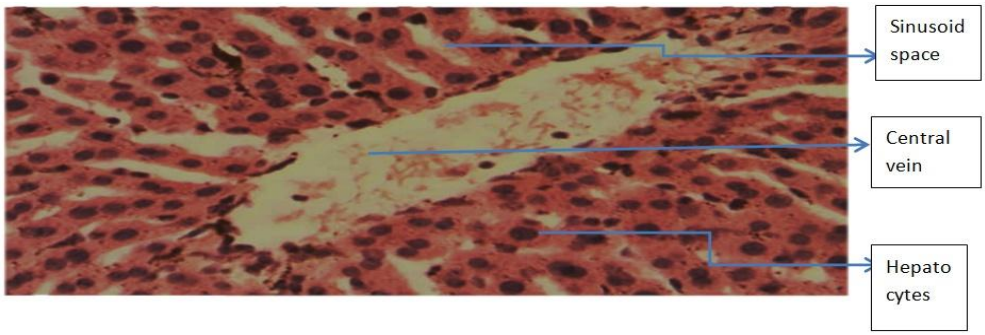

Fig. 9. Liver tissue of normal control group showing normal slightly slanted central vein, normal round hepatocytes and narrowed sinusoid space

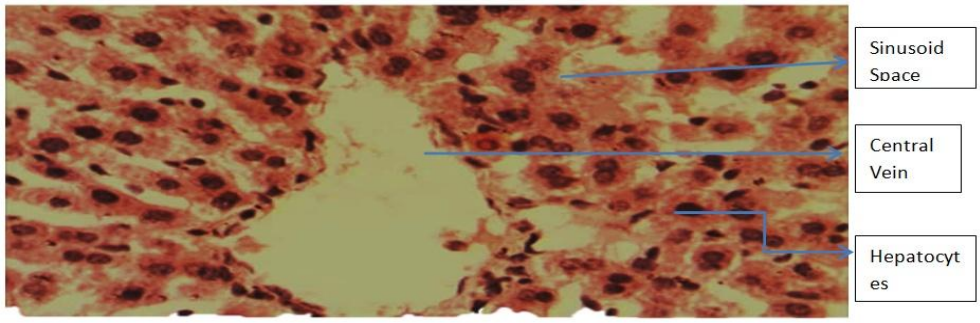

Fig. 10. Liver tissue of diabetic control group showing highly abnormal morphology mostly on the sinusoid spaces

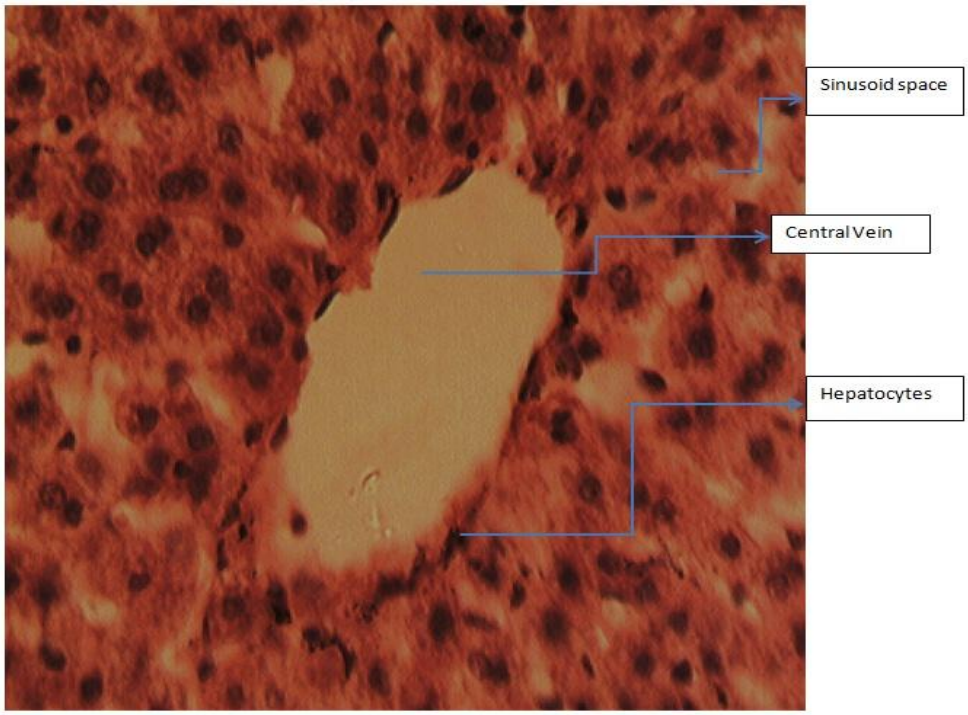

Fig. 11. Liver tissue of group treated with 200 mg/kg alkaloid extract of *V. doniana* showing restoration of central vein, hepatocytes and sinusoid space.

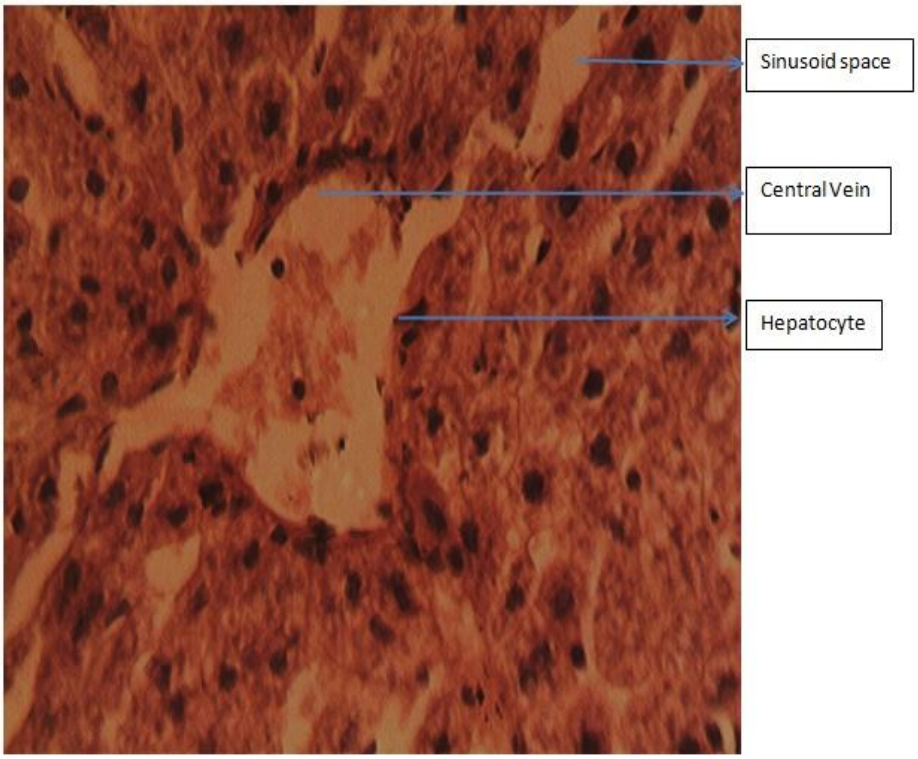

Fig. 12. Liver tissue of group treated with 400 mgkg^{-1} alkaloid extract of *V. doniana* showing irregular central vein morphology, wide sinusoid space and hepatocytes on the central vein.

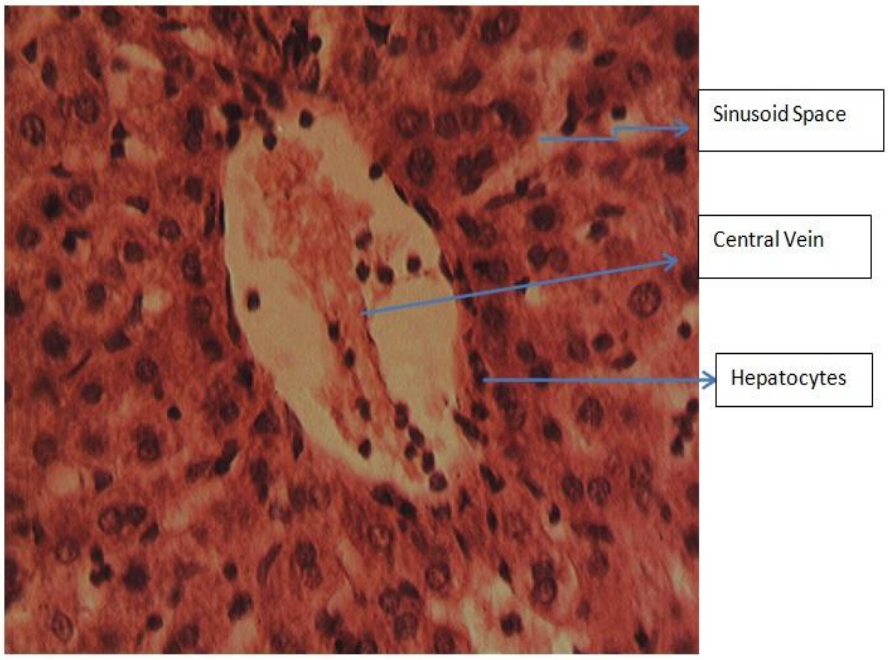

Fig. 13. Liver tissue of group treated with 200 mgkg^{-1} alkaloid extract of *F. thoningii* showing abnormalities on the central vein having traces of hepatocytes on it

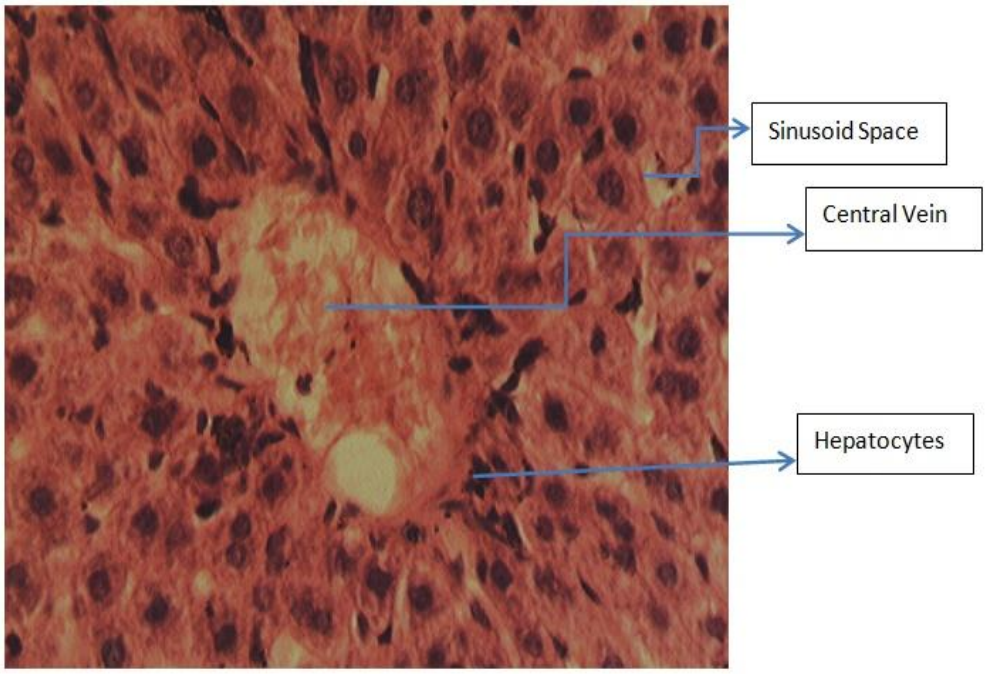

Fig. 14. Liver tissue of group treated with 400 mgkg^{-1} alkaloid extract of *F. thoningii* showing mild regeneration of the central vein, and abnormal hepatocyte structure

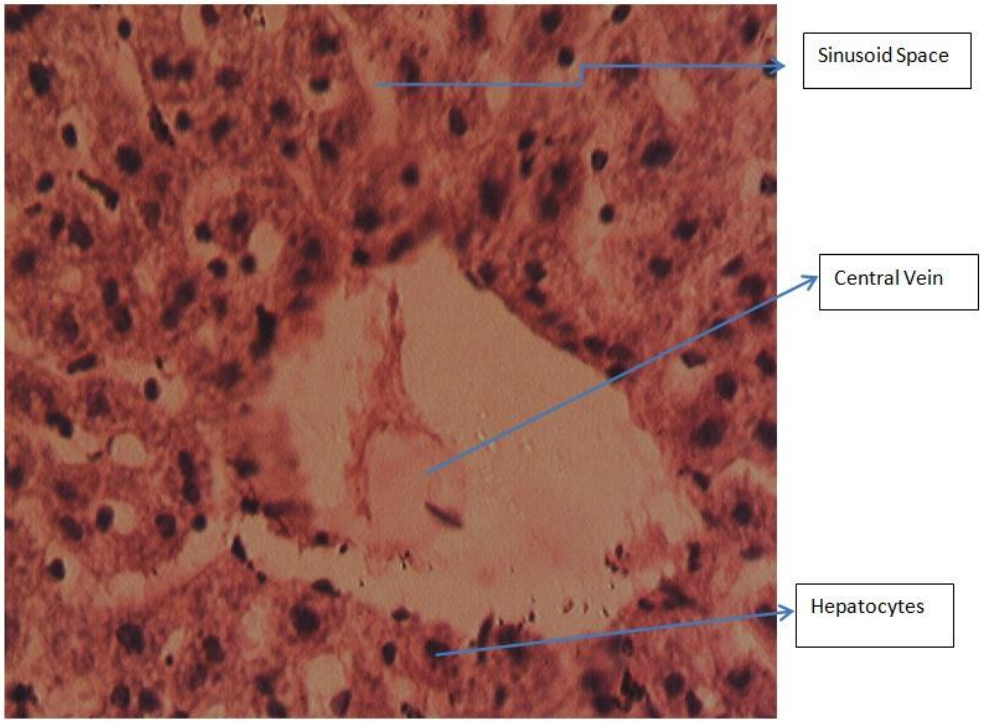

Fig. 15. Liver tissue of group treated with 100 mgkg^{-1}alkaloid extract each from *V. doniana* and *F. thoningii* showing regeneration of the central vein, normal hepatocyte and wide sinusoid space

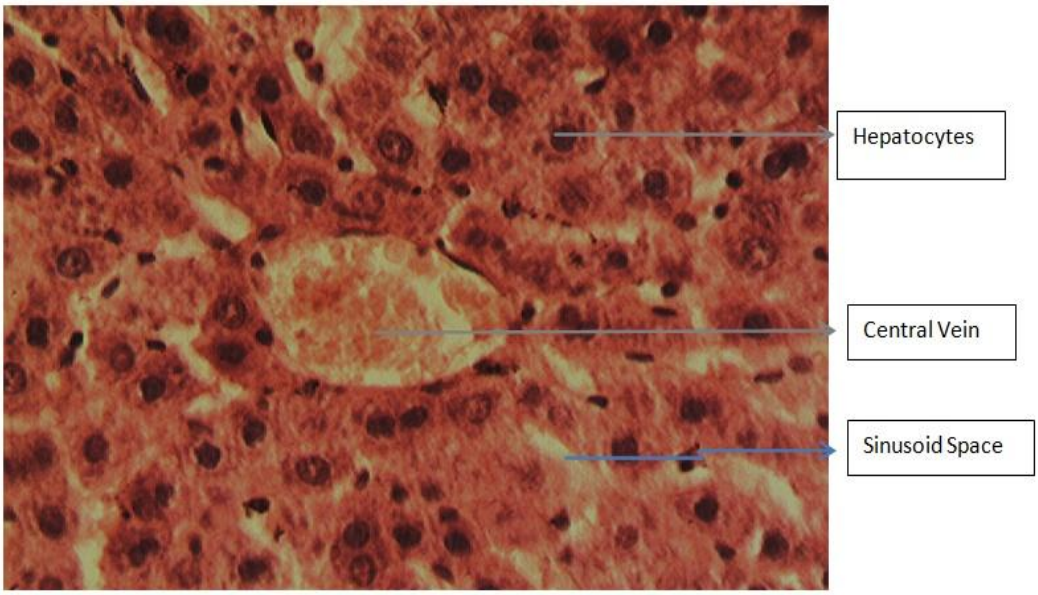

Fig. 16. Liver tissue of group treated with 200 mgkg^{-1} alkaloid extract each from *V. doniana* and *F. thoningii* showing regeneration of the central vein, normal hepatocytes and sinusoid space

The results of hepatic enzyme activities are presented in Figs. 3-5. The significant elevation in the activities of ALP, AST and ALT in the diabetic control rats when compared with normal control at $p<0.05$ might indicate hepatic tissue damage [29]. This result is similar to the findings of Airaodion et al. [30,31] who reported an elevation in the activities of hepatic enzymes when animals were exposed to ethanol and hydrocarbon respectively. The mechanism of elevation of these enzymes by alloxan in this study might be similar to that of ethanol and hydrocarbon. The enzymes found within liver tissues are released into the bloodstream following cellular necrosis and cell membrane permeability and are thus used as diagnostic measure of liver damage [32]. Damage to structural integrity of tissues is always reflected by an increase in some of these enzymes in the serum, probably through leakage from the altered cell membrane structure [33]. Therefore, the corresponding increase in serum ALP activity following induction of diabetes confirms damage to the plasma membrane, leading to compromise of its integrity [34]. Loss of ALP activity from the tissue may hinder adequate transportation of required ions or molecules across the cell membrane [35]. It may also affect other metabolic processes such as the synthesis of nuclear proteins, nucleic acids, phospholipids, and cleavage of phosphate esters that require the enzyme [36]. The aminotransferases (ALT and AST) considered in this study are useful ‘‘marker’’ enzymes of liver cytolysis [37]. The enzymes occupy a central position in the metabolism of amino acids as they help to retain amino groups (or to form new ones) during the degradation of amino acids. Increase in serum AST and ALT activities following induction of diabetes (Figs. 3 and 4 respectively) confirms damage to the liver cells, due to compromise of cellular integrity [38]. The ability of the extracts of *V. doniana* and *F. thoningii* to lower the activities of these studied serum marker enzymes in diabetic rats may be attributed to its bioactive alkaloid content [39]. This result is similar to the findings of Airaodion et al. [40,41] who reported the hepatoprotective potentials of *Parkia biglobosa* and *Talinum triangulare* leaves respectively against ethanol-induced oxidative stress due to the presence of phytochemicals in these plants. It also corresponds to the findings of Ogbuagu et al. [42] who reported the prophylactic propensity of methanolic extract of *Vernonia amygdalina* leaves against acute ethanol-induced oxidative stress in Wistar rats. Thus, *V. doniana* and *F. thoningii* alkaloid extracts can be said to have hepatoprotective properties.

In this study, serum total protein and albumin concentrations decreased significantly in diabetic control group when compared with those of normal control animals at $p<0.05$ (Figs. 6 and 7

respectively). This observation may be attributed to numerous effects of hyperglycemia in the alloxan-induced diabetic rats. Hyperglycemia increases gluconeogenesis and as such leads to excess protein breakdown as well as excess loss of nitrogen resulting in negative nitrogen balance [43]. A decline in total serum protein level in diabetes have been attributed to inhibition of oxidative phosphorylation which leads to decrease in protein synthesis, increase in catabolic processes and reduction in protein absorption [44]. The results showed that administration of the alkaloid extracts of *V. doniana* and *F. thoningii* caused a remarkable increase in the serum total protein and albumin levels in the diabetic treated rats. These observations may be due to the presence of some compounds which help in the provision of a reserved store of protein [45].

Administration of alloxan resulted in an increase in total bilirubin levels of the diabetic control animals when compared with that of the control group at $p<0.05$ (Fig. 8). The results showed that oral administration of the alkaloid extracts from *V. doniana* and *F. thoningii* significantly lowered the total bilirubin levels in the diabetic rats. This observation indicates that hyperglycemia can enhance protein glycation. The increased total bilirubin level is an indication that there is increased haemoglobin destruction or that there is impairment in the liver function relative to haemoglobin metabolism [46,47].

Histological tissue images obtained in this study showed the normal structural features of hepatocytes, central vein and narrowed sinusoid space in the normal control rat group (Fig. 9). Widened sinusoid space and glycogen accumulation were significantly apparent in the diabetic control group (Fig. 10). Regenerations occurred in the alkaloid extract treated diabetic groups (Figs. 11-16).

Undiabetic normal control rat liver tissue showed normal cellular hepatocyte architecture, narrowed sinusoid space and central vein (Fig. 9) while the liver tissue of diabetic control rat showed cellular abnormalities with irregular hepatocyte, widened sinusoid space, congestion and dilatation of central vein (Fig. 10). When diabetic rats were treated with 200 $mgkg^{-1}$ alkaloid extract of *V. doniana*, the liver tissue showed restoration of central vein, hepatocytes and sinusoid space (Fig. 11). Treatment with 400 $mgkg^{-1}$ alkaloid extract of *V. doniana* resulted in irregular central vein morphology, wide sinusoid space and hepatocytes on the central vein (Fig. 12). Administration of 200 $mgkg^{-1}$ alkaloid extract of *F. thoningii* to diabetic rats resulted in abnormalities on the central vein having traces of hepatocytes on it (Fig. 13). Administration of 400 $mgkg^{-1}$ alkaloid extract of *F. thoningii* to diabetic rats resulted in mild regeneration of the central vein, and abnormal hepatocyte structure (Fig. 14). Administration of 100 $mgkg^{-1}$ alkaloid extract from *V. doniana* and 100 $mgkg^{-1}$ alkaloid extract from *F. thoningii* to diabetic rats showed regeneration of the central vein, normal hepatocyte and wide sinusoid space (Fig. 15). Administration of 200 $mgkg^{-1}$ alkaloid extract from *V. doniana* and 200 $mgkg^{-1}$ alkaloid extract from *F. thoningii* to diabetic rats also showed regeneration of the central vein, normal hepatocytes and sinusoid space (Fig. 16).

Alloxan induction causes a diabetic state in experimental animals and at certain doses will cause hepatic tissue damage. This result is similar to the damage of the liver by monosodium glutamate reported by Ogbuagu et al. [48]. The liver is an important organ in the body and has a central role in the metabolism of substances that could be toxic in the body. This makes the liver an organ that is susceptible to metabolic system disorders. Indications of disorder can be seen in changes in liver histologic structures such as abnormalities on the central vein, sinusoids, and hepatocyte morphology. Damage to liver cells can be temporary or permanent. In the case of temporary damage, the liver cells will undergo regeneration as a form of adaptation process. This temporary change can be assisted or achieved via treatment [49].

5. CONCLUSION

Alloxan induction caused a diabetic state in experimental animals and at certain doses will cause hepatic tissue damage. The liver is an important organ in the body and has a central role in the metabolism of substances that could be toxic in the body. This makes the liver an organ that is susceptible to metabolic system disorders. Indications of disorder can be seen in changes in liver histologic structures such as abnormalities on the central vein, sinusoids, and hepatocyte morphology. The ability of the alkaloid extracts of *V. doniana* and *F. thoningii* to regenerate full morphology of the liver tissues revealed that it is also hepatoprotective. However, extract of *F. thoningii* seems to be more potent.

CONSENT

Not Applicable.

ETHICAL APPROVAL

Not Applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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