ABSTRACT
Phytochemical screening and the effects of oral administration of palm wine extract of *Datura stramonium* on the activity of tissue enzymes and histopathology of rats was investigated. Forty (40) experimental rats of average weight of 100-150g were divided into four groups; A, B, C, and D comprises ten (10) rats each. Subgroups A, B, and C were administered with varying concentrations of palm wine extract of *Datura stramonium*, and group D served as the control group. The animals were kept in separate metabolic cages and fed with rat chow with water provided *ad libitum*. Rats in each group (A, B, C) where given 0.5ml/ kg body weight of the extract per day and sacrificed after day 7, 14, and 21. Rats in the control group (D) were given 0.5ml/ kg body weight of fresh palm wine daily. The enzyme activities acid phosphatase (ACP), aspartate amino transferase (AST) and alanine amino transferase (ALT) were measured. The serum total protein was measured using Biuret method. Histological analysis was carried out on the tissues (kidney, liver and intestine). A significant increase (p<0.05) in the activity of the enzyme (ACP, ALT and AST) and a significant decrease (p<0.05) in total serum protein were observed. Tissue biopsy showed a clear evidence of cell erosion in the treated groups, which was not apparent in the control group.

**KEY WORDS:** *Datura stramonium*, ACP, AST, ALT.
INTRODUCTION

Plants have always been a source of medicine for the treatment of various human ailments. A large portion of the world’s population especially in developing countries depends on the traditional system of medicine for treating variety of diseases. Over 120 pharmaceutical products currently in use are plant derived, and 25% of these were discovered by examining the use of these plants in traditional medicine (Akinrele et al., 1977).

Natural product chemists and phytochemists, recognize that plant species contain a bewildering diversity of secondary metabolites. Individual plant species often contain over a thousand unique chemical entities (for the enzymatic machinery needed to produce compounds upon proper stimulation). The biodiversity of tropical forest plant species, coupled with chemical diversity found within each plant leads to the conclusion that plants are the most valuable source of new bioactive chemical entities (Fansworth and Kaas, 1996).

Over centuries, human beings source information about plants, and use various medicinal plants and herbs that grew in their environment to treat various illnesses. Medicinal plants have emerged as some of the most widely studied plants and significant interest has been shown in their chemistry because of their potential application in medicine (Pandey et al., 2007). Many of these medicinal plants contain chemical constituents that could cause harmful effects to human if abused. Alkaloids occurring in a large amount make these plants poisonous (Gardner and Pfister, 2007). Large quantity of oxalic acid, a protoplasmic poison in the form of oxalates of calcium, sodium and potassium also produces poison (Doaigey, 1991). Some of these plants are poisonous to insects and pests (Sahaf et al., 2007).

The genus “Datura” (Solanaceae) comprises all the night-shades and agricultural plants, including potato, tomato, coffee and pepper. Classification of different species within Datura genus relies heavily on genetic markers, which suggests that this genus has huge variation due to mutation (Fornoni, 2000).

*Datura stramonium*, the most common species within this family, is native to Asia, but is also found in the United States, Canada, and the West Indies. It is popularly called in English: ‘Devil’s trumpet, downy thorn apple, Jimson weed, in Hausa: ‘Zakami’, ‘gegemu’ in Yoruba, Ogoni people knows it as ‘gegami’, and Igala native know it as ‘Jegemi’. It is widespread with higher abundance in temperate, tropical and subtropical regions (Berkov, 2006). Traditionally, *Datura stramonium* has been used for mystic and religious purposes (Ajungla,
2009), and herbal medicine with narcotic effects or to treat asthma (Dessanges, 2001). The seed of *D. stramonium* is smoked to achieve hallucinogenic experiences (Diker *et al*., 2007). Accidental poisoning of humans and animals that consume food sources contaminated with *D. stramonium* in areas where millet, wheat, rye, corn, and bean seeds are used for human consumption, and where *D. stramonium* is the common weed, the grain sometimes is been contaminated with *Datura* seeds (Norton, 2008) and has been reported by Naude and associates (Naudé *et al*., 2005). In Ayurvedic medicine, *D. stramonium* is described as a useful remedy for various human ailments including ulcers, wounds healings, inflammation, rheumatism and gout, sciatica, bruises and swellings, fever, asthma and bronchitis and toothache (Khare, 2007).

Many folk medicine remedies use *D. stramonium* therapeutically. In the Hindu religion, the seed of *D. stramonium* is believed to be associated with the God Shiva, which can promote misuse of the plant on religious occasions, like the Shivaratri and Swasthani Puja (Gaire, 2008). In modern medicine, the therapeutic use of *D. stramonium* is over shadowed by its toxic effects. The administration of large amounts of *D. stramonium* affects the central nervous system with symptoms such as confusion, bizarre behavior, hallucinations and subsequent amnesia, though death by *D. stramonium* poisoning is rare, recovery may take several days (Norton, 2008).

The fruits and seeds of *D. stramonium* have several uses; the spiny fruit is used to card cotton. The calyx base is used in rubbing teeth. A drink made from the seeds is given as intoxicant to Fulani youths (Nigeria) to incite them into the “Sharo contest” or ordeal of manhood (personal communication). Youths in some parts of Plateau State, Nigeria, who use it to perform rigorous work, have claimed that the water extract of the seeds alleviate pains. Seeds along with other substances are used as a remedy for the symptoms of madness based on homeopathic principle, and decoction of seeds is said to be useful in eye diseases. The seeds also constitute the potential source for hyoscine (Anozie, 1986).

**Habitat**

*Datura stramonium* is a shrub like perennial herb, found in India, England and other tropical and subtropical regions. *Datura*, is a mesophyte which grows in tropical and subtropical regions. It is cultivated all around the world for its chemical and ornamental properties. It is also grown for its spiritual values (Khaton and Shaik, 2012).
Phytochemistry

Phytochemical studies of *D. stramonium* have been conducted since the early 1930s. The major phytochemicals isolated from *D. stramonium* are tropane alkaloids, atropine and scopolamine (Jakabova et al., 2012). It is reported that the whole plant contains 0.26% alkaloids. Seeds of *Datura* contain the alkaloid daturine, first isolated, purified and crystallized by Geiger and Hesse, in 1833. Von Planta (1850) pronounced daturine to be identical with atropine, the principal belladonna alkaloid. Ladenburg later differentiated daturine into atropine and hyoscyamine. Schmidt, however, contended that atropine predominates. The seeds contain fatty oil (25%), from which a new fatty acid, daturic acid \((\text{C}_{17}\text{H}_{34}\text{O}_2)\), was isolated. Dohme concluded that the stems contain more alkaloids (0.3% to 0.4%, volumetrically) than even the seeds (0.25% to 0.29%). The seeds contain more alkaloid than the leaves (0.21% to 0.23%, and 0.27% for green leaves) (Khare, 2007).

Berkov et al., (2006) suggested hyoscyamine as the main alkaloid in both diploid and tetraploid hairy root cultures of *D. stramonium*. Iranbakhsh et al., (2006), reported the percentage of atropine and scopolamine in different developmental stages and the parts of the *Datura*. Their study suggested that the root contained lower levels of scopolamine than that of atropine, so for the stem. In stems, atropine was almost three times higher than scopolamine.

However, leaves and seeds contained higher level of scopolamine than that of atropine. Recently, Li et al.,(2012), reported the different alkaloids from *D. stramonium* seeds such as N-trans-feruloyl tryptamine, hyoscyamylactol, scopoletin, umckalin, daturaolone, daturadiol, N-trans-ferulicacyl-tyramine, cleomiscosin A, fraxetin, 1-acetyl-7-hydroxybeta-carboline, and 7-hydroxy-beta-carboline-propionic acid. The chemical structures of major phytochemicals isolated from *D. stramonium* are listed in below;
A colourless crystalline constituent, daturilin has been obtained from the acid of the alcoholic extract of *D. metel* leaves. This compound has been identified as epoxy-(20S, 22S-witha-2, 5, 25-trienolide. These compounds were recognized as withametelin C, D, and E three new withanolide (22 baimantuolouline A, B, and C and the two known withanolides withafastuosin E and withametelin C were isolated from the fraction exhibiting activity for psoriasis from the flower of *D. metel* (Wang *et al*., 2008). The inorganic content of the leaf of *Datura metel* are calcium, magnesium and phosphorous present in the ionic state acid-insoluble fraction trienolide (Siddiqul *et al*., 1992). The structure of Daturilllin is given below;

![Chemical Structure of Daturilllin](image)

**Daturilllin**

A new antibacterial agent 51, 71 dimethyl 61– hydroxy 31, phenyl 3 â- amine â- ß-yne sitosterol has been isolated from *Datura metel* leaves. The structure was established using 13C, 1H NMR, IR and MS spectroscopic data. It displayed antibacterial activity against *Staphylococcus aureus, Pseudomonas aeruginosa, Proteus mirabilis, Solmonella typhi, Bacillus subtilis* and *Klebsiella pneumonia* but could not inhibit *Escherichia coli*. This result supported the use of *Datura metel* in phytomedicine for the treatment of asthma, cough, burns and healing of wounds in Nigeria (Donatus and Ephraim, 2009). In Western Nepal, leaves of *Datura* along with the leaves of *Cannabis sativa* and stem of *Neopicrorhiza scrofulariiflora*, are pounded with water and applied to treat headaches. *Datura* seeds are crushed with grains of rice and taken orally to relieve indigestion. In parts of Central Nepal, fresh leaves are warmed and placed on a sprained body part repeatedly, before going to bed, for the alleged analgesic effect. Juice from the leaves is given with warm milk to expel intestinal worms, specifically tapeworm.

In Nigeria, the seeds are mixed with palm oil and applied to severe cases of insect bites and stings (Egharevba and Ikhatua, 2008). In India, different parts of *Datura stramonium* seeds
are used as a tonic and febrifuge; the leaves are roasted and applied locally to relieve pain (Gorsi and Shahzad, 2002). Women in Pakistan warm up 5 to 8 leaves in low fire, and then tie onto sagging breasts to bust them up. Two to five seeds are added to a cup of green tea to relieve headache (Hussain et al., 2006). Native Americans used *Datura* seed for many years as euphoric agent. Since the 1800s, it was used as a therapeutic agent in Great Britain (Dessanges, 2001).

**Pharmacological properties**
The whole plant, but especially the leaves and seed, is analgesic, antimicrobial, antifungal, antiasthmatic, antispasmodic, antitussive and bronchodilator, hallucinogenic, hypnotic, hypoglycemic, hydriatic and cytotoxic activity.

*Datura stramonium* in phytomedicine is used for the treatment of asthma, cough, burns and healing of wounds.

**Antifungal activity**
Acetone extracts of *D. stramonium* have been reported to have antifungal activity against several fungi including *Penicillium expansum, Aspergillus niger, Aspergillus parasiticus, Colletotrichum gloeosporioides, Fusarium oxysporum, Trichoderma harzianum, Phytophthora nicotiana, Pythium ultimum* and *Rhizoctonia solani*. The MIC of *D. stramonium* extracts ranges from 1.25 to 2.5 mg/mL. The fungicidal effects of the extracts indicate the potential of *D. stramonium* seeds as a natural source of antifungal agent (Mdee, 2009).

**Anti-inflammatory activity**
Ethanol extract of *D. stramonium* leaf showed significant anti-inflammatory activity against carrageenan induced paw edema in rats. In one experiment (Sonika, 2010), 39.43% inhibition of the edema was observed after three hour of oral administration of 200 mg/kg extracts. Maximum activity was observed when the extract was administered in doses of 3-hour intervals. Since the extract of *D. stramonium* inhibited the carrageenan-induced edema that involves the release of histamine and serotonin in the first phase, the inhibitory effect of the extracts could be partly due to inhibition of mast cell mediator release seed (Sonika, 2010).
Hypoglycemic activity

The seed powder of *Datura* was tested for its hypoglycemic activity in normal and alloxan induced diabetic rats. Graded doses (25, 50 and 75 mg/kg body weight) of the seed powder when given to both normal and diabetic rats produced significant reduction in blood glucose at the 8 h. The effect was found to be dose dependent with all treatments at the doses administered. The statistical significance (p<0.0125) was determined (Krishna et al., 2004).

Hallucinogenic property

*Datura* has been documented as a plant with hallucinogenic properties (Kirsten 1986). The hallucinogenic effect of aqueous seed extract of *Datura metel* was evaluated in an experiment carried out on albino rats (Damilare et al., 2010). The Male wistar rats were divided into four groups and were orally administered with aqueous seed extract of 0.2, 0.4, 0.6 and 0.8mg/kg body weight respectively. The treated groups exhibited some behavioral changes such as restlessness, aggressiveness, agitation and disorientation. The effect of the extract on the food and water intake shows a significant decrease (p<0.05) in the 0.6 and 0.8mg/kg extract treated groups as compared to the control. The heart rate increased significantly (p<0.05) in 0.6 and 0.8mg/kg treated groups while the respiratory rate increased in the 0.8mg/kg treated group as compared to the control respectively. The hallucinogenic effect observed may be due to the presence of the alkaloid scopolamine (Damilare et al., 2010).

Anti-asthmatic activity

Asthma relief is attributed to depression or paralysis of the receptive mechanism of the parasympathetic nerves in the bronchi (a known action of solanaceous alkaloids), an effect confirmed by the relaxation produced by the alkaloidal extract from the smoke, on an isolated intercartilagenous portion of a bronchial ring previously contracted by pilocarpine. When smoke is inhaled, it is possible the sticky, resinous substance may help by coating the mucosa and thus lessening the bronchial irritation (Kam and Liew, 2002). *D. stramonium* contains a variety of alkaloids, including atropine and scopolamine, having anticholinergic and bronchodilating activity. Atropine and scopolamine act on the muscarinic receptors by blocking them (particularly the M2 receptors) on airway smooth muscle and submucosal gland cells, which dilate bronchial smooth muscle and ease asthmatic attacks. Charpin et al., (1979) reported that using *D. stramonium* as an antiasthmatic cigarette is an effective bronchodilator in asthmatic patients with mild airway obstruction. However, the exposure of *D. stramonium* to the fetus when a mother uses it for asthma will cause a continuous release.
of acetylcholine, resulting in the desensitizing of nicotinic receptors, which could ultimately result in permanent damage to the fetus (Pretorius et al., 2006).

Anti-epilepsy

Though the antiepileptic activity of *D. stramonium* has not been reported, but combination therapy with other herbs has the protective effect on status epilepticus. An experimental model of status epilepticus was induced in male rats by a single systemic injection of lithium (3 mmol/kg) and pilocarpine (30 g/kg). Rats were then treated with herbal mixture containing *D. stramonium*. One week after the induction of status epilepticus, the rat group treated with extracts of *Scutellaria lateriflora* (Skullcap), *Gelsemium sempervirens* (Gelsemium) and *D. stramonium* (Jimson Weed) displayed no seizure during treatment. The results of this experiment strongly suggest that the appropriate combination of herbs with *D. stramonium* may be helpful as adjunctive interventions to treat epilepsy (Peredery and Persinger, 2004).

Organophosphate poisoning

Since *D. stramonium* contains atropine and other anticholinergic compounds, it is a useful remedy for the central cholinergic symptoms of organophosphate (OP) poisoning. Bania et al., (2004), determined the beneficial effect of *Datura* seed extracts following a severe OP poisoning. According to their experiment, *D. stramonium* seeds were heated in water to make 2 mg/mL atropine solution and administered to male rats as a single intraperitoneal injection 5 minutes before the subcutaneous injection of 25 mg/kg of dichlorvos. Pretreatment with *Datura* seed extracts significantly increased survival in a rat model of severe OP poisoning (Bania et al., 2004).

Antimicrobial activity

The methanol extracts of aerial parts of *D. stramonium* showed the bactericidal activity against Gram-positive bacteria in a dose-dependent manner. However, little or no antibacterial activity was found against *Escherichia coli* and *Pseudomonas aeruginosa* (Eftekhari et al., 2005). Ethanol extract of *D. stramonium* exhibited the highest inhibitory activity against *Klebsiella pneumonia* followed by *Staphylococcus aureus*, with the least activity against *Salmonella typhi*. The aqueous extract showed activity on only *S. aureus*, while *Neisseria gonorrhea* was resistant to both extracts (Shagal et al., 2012). *D. stramonium* was very effective as vibriocidal against various strains of *Vibreo cholera* and *Vibreo parahaemolyticus*. The minimum inhibitory concentration (MIC) value of acetone extracts of
D. stramonium was in the range of 2.5 to 15 mg/mL serving as broad-spectrum vibriocidal agents (Sharma et al., 2009).

**Antioxidant:**
Study revealed that the aqueous extract of D. stramonium contained more phytochemical compounds than ethanol extracts. Antioxidant activities were higher in the plant leaf than the bark. Results suggest that the plant is a natural source of antioxidants and phytochemical quality for antibacterial effectiveness (Akharaiyi, 2011).

**Toxicological details**
D. stramonium is mostly studied for its toxicological properties. Datura poisoning is very common in India, usually involving the seeds. Many cases of unintentional poisoning by D. stramonium species have been reported when taken accidentally, or as decoction prepared from herbal prescription (Hirschmann and De Arias, 1990).

General symptoms of D. stramonium poisoning include delirium, agitation and seizures, mydriasis, blurred vision, photophobia, dry mouth and mucous membranes, extreme thirst, tachycardia, nausea and vomiting, decreased bowel sounds, difficulty swallowing and speaking, hyperthermia, hypertension, loss of consciousness and coma (Oberndorfer et al., 2002). Dugan et al., (1989), reported that ingestion of D. stramonium seed at concentrations of 0.5% or more in the diet produce adverse physiological changes in rats. Bouzidi et al., (2011) reviewed the acute, sub-acute and chronic toxicity studies of alkaloids from the seeds of D. stramonium. According to them, single dose acute toxicity of 10 mg/kg D. stramonium includes decreases in the weight of the liver, spleen and brain, and significant increases in the levels of red blood cells (RBC), hematocrit (HCT), hemoglobin (HGB), and white blood cells (WBC). Similarly, RBC, HGB, HCT and platelet levels were increased in 4-week sub-acute toxicity studies. However, the 120-day chronic toxicity study of D. stramonium alkaloids showed decreased levels of RBC, HCT, HBG and WBC, with a significant increase in liver enzymes. Fatal dosages of D. stramonium toxins occurred with amounts exceeding 10 mg for adults, and 4 mg for children. The amount needed to poison an adult is about 20 seeds, and the estimated LD in an adult is >10 mg atropine or >2 to 4 mg scopolamine (Norton, 2008)

**Tissue Studied**
Tissue studied includes; Liver, Kidney and small intestine.
Enzyme studied

Acid phosphatase
Orthophosphoric-monoester phosphohydrolase (EC 3.1.3.2). It is a non specific phosphomonoesterase that catalyses the hydrolysis of phosphate esters at a pH 4.5 (Yam, 1974).

Acid phosphatase includes all the phosphatases catalyzing the following reaction at an optimal pH below 7:
Orthophosphoric monoester + H₂O → alcohol + H₃PO₄

Occurrence
Acid phosphatase is an enzyme found throughout the body, but primarily in the prostate gland. The male prostate gland has 100 times more acid phosphatase than any other body tissue. Tissues other than prostate have small amounts of acid phosphatase, including bone, liver, spleen, kidney, red blood cells and platelets. Acid phosphatases are present in lysosomes, some extra lysosomal acid phosphatases are also found in many cells (Roy et al., 1971).

Damage to these tissues causes a moderate increase in acid phosphatase levels. Different forms of acid phosphatase are found in different organs, and their serum levels are used as biomarkers for diagnosing disease in the corresponding organs. Acid phosphatase from prostate contributes 1/3rd to 1/2nd of the enzyme activity present in the serum of a healthy male. The source of the remainder of the acid phosphatase in the serum from healthy males and females is not clear. Normal serum value - 1-5 KA units/ 100 ml.

Clinical Significance
The highest levels of acid phosphatase are found in metastasized prostate cancer. It is of clinical importance to differentiate prostatic and non prostatic form of acid phosphatase. The prostatic enzyme is strongly inhibited by dextrorotatory tartarate ions, whereas the erythrocyte isoenzyme is not. Formaldehyde and cupric ions inhibit erythrocyte acid phosphatase but not the prostate acid phosphatase. Diseases of the bone, such as Paget's disease or hyperparathyroidism; diseases of blood cells, such as sickle cell disease or multiple myeloma; or lysosomal disorders, such as Gaucher's disease, also show moderately increased levels of acid phosphatase.
Acid phosphatase activity in serum is used to detect or monitor carcinoma of the prostate. An elevation in the activity of the enzyme has been very useful in the diagnosis of this enzyme (Yam et al., 1979).

**Aspartate amino transferase (AST)**

Transaminases are present in most of the tissues of the body. They catalyze the interconversions of the amino acids and 2-oxoacids by transfer of amino groups. Transaminases are specific for the amino acid from which the amino group has to be transferred to a keto acid. 2-oxoglutarate and glutamate serves as one amino group acceptor and donor pair in all amino transfer reactions. Aspartate amino transferase formerly called glutamate oxaloacetate transaminase (GOT), catalyzes the interconversion of oxaloacetate to aspartate coupled with glutamate to 2-oxoglutarate.

![Diagram of the reaction](image)

**Occurrence**

AST is normally found in the red blood cells, liver, heart, muscle tissues, pancreas and kidney. Elevated levels of AST are usually found in the blood when body tissue or organ such as heart or liver is diseased or damaged.

**Clinical Significance**

AST is found in the tissues but higher in the liver and skeletal muscles. The highest concentration is found in heart muscle cells. Its concentration in healthy human serum is between 0 to 41 unit/L (0 to 41 μmol/mg activity/ 0-41 IU/L), and this is a result of normal death of old and worn out cells, (Moray et al., 1999). In newborns value up to 120 units for AST is considered normal. The amount of AST in the blood is directly related to the extent of tissue damage (Healthwise, 2008). Any damage or injury to the cells of these tissues may cause release of these enzymes along with other intracellular proteins/enzymes into the circulation leading to increase in activities of these enzymes in the blood. Some increases in the activities of both the enzymes are seen after alcohol intake. Although elevated levels of AST is not specific for liver damage or diseases, it is used primarily to diagnose and monitor
the course of heart diseases, heart attack in patients and myocardial infarction (Bertland and Kaplen, 1968). According to Ganong (1995), myocardial infarction is as a result of interrupted blood supply to part of the heart myocardium due to profound changes in the myocardium (myocardial ischemia) resulting in irreversible changes and subsequent death of heart muscle cells. In myocardial infarction high activity of AST is seen in serum. ALT activity is within normal range or slightly increased in uncomplicated myocardial infarction. Rise in AST is seen within 6 to 8 hours of the onset of chest pain, highest level at 18 to 24 hours and returns to pre-infarction levels by 4th to 5th day. There are other superior markers available for myocardial infarction as AST lacks the tissue specific characteristics; its activity may also increases in diseases of other tissues like liver and skeletal muscles. In other conditions like pulmonary emboli, acute pancreatitis, hemolytic disease and gangrene the activity of AST is found to be 2 to 5 times higher than the normal activity.

**Alanine amino transferase (ALT)**

Alanine amino transferase, formerly called glutamate pyruvate transaminase is present in many tissues. This is a non functional enzyme which performs no function in the blood. Its substrate are usually absent in the plasma and it is present in the blood of normal individuals. ALT catalyzes the interconversion of pyruvate to alanine coupled with glutamate to 2-oxoglutarate.

The reactions catalyzes by AST and ALT are reversible but the equilibra of the reaction favor formation of aspartate and alanine respectively. In both the reactions pyridoxal-5- phosphate functions as a prosthetic group in the amino transfer reactions. Normal serum values: ALT (SGPT) - 0-45 IU/L. In newborns value up to and 90 units for ALT is considered normal (Henry, 1974).

**Occurrence**

The enzyme is present in many tissues, especially liver, heart and muscles. The appearance of elevated levels of ALT in the plasma signals possible damage to hepatic tissues (Henry, 1974).
Clinical significance
The activities of ALT are high in tissues especially liver, heart, and muscles. Any damage or injury to the cells of these tissues may cause release of this enzyme along with other intracellular proteins/enzymes into the circulation leading to increase activities of this enzyme in the blood. Some increases in the activities of the enzymes are seen after alcohol intake.

Determinations of activities of AST and ALT in serum in patients with liver diseases like viral hepatitis and other forms of liver diseases with necrosis, give high values even before the appearance of clinical signs and symptoms like jaundice. Activity levels of 20 to 50 fold higher than normal are frequently seen in liver cells damage but it may reach as high as 100 times in severe damage to cells. Highest serum activities are seen between 7th and 12th days and return to normal levels by the 3rd to 5th week (Reir, 1984). In sever tissue damage ALT activity is higher than AST. Some increases in the activities of ALT and AST are seen in extrahepatic cholestasis. In cirrhosis, the level of activities varies with the severity of the disease. It may increase only up to 5 fold of the normal activities. Up to 10 fold increase is seen in carcinoma, of the liver. In both cirrhosis and carcinoma activity of AST is found to be higher than the ALT. Even though the activities of both AST and ALT are elevated in the serum of patients with liver diseases, ALT is more liver specific and increased ALT activity in serum is hardly seen in tissues other than liver cell damage (Aach et al., 1981). In myocardial infarction high activity of AST is seen in serum. ALT activity is within normal range or slightly increased in uncomplicated myocardial infarction. Also, AST and occasionally ALT activity levels are increased in progressive muscular dystrophy and dermatomyositis. Level of AST may go as high as 8 times of the normal. There is no increase in the enzyme activity in the muscle diseases of neurogenic origin. Increased AST activity, 2 to 5 times of normal, is also seen after crushed muscle injuries (Aach et al., 1981).

MATERIALS AND METHODS
Experimental animals
Albino rats were obtained from the animal house, Faculty of Agriculture, Kogi State University, Anyigba.
Collection of plants
The leaves of the plant (*Datura stramonium*) were collected from the wild within the premises of Kogi State University, Anyingba. The plant was identified by Professor S. S. Usman, Department of Biological Sciences.

Enzyme substrate
2, 4-dinitrophenylhydrazine, buffer (phosphate buffer, L-alanine, alpha oxoglutarate), and Biuret reagent were supplied by Randox laboratories limited, United Kingdom. α-naftyl phosphate, buffer (sodium citrate), was supplied by Bio Trust Medics International Limited, United States of America.

Other reagents
Meyers reagent, folin-denis reagent, hydrochloric acid, ammonium hydroxide, sodium hydroxide, ethanol, distilled water, ferric chloride, 10% formalin, chloroform, sulphuric acid.

Equipments
Metabolic cages, spectrophotometer (Biosystems instrument), pre-cooled enamel mortar and pestle, weighing balance, centrifuge, water bath, were all obtained at the Biochemistry Department, Kogi State University, Anyigba. All laboratory glassware used was obtained at the department laboratory.

PHYTOCHEMICAL STUDIES
The spectrophotometric method of Brunner was used for saponin analysis. 1g of finely ground sample was weighed into a 250ml beaker and 100ml of butanol was added. The mixture was shaken vigorously to ensure uniform mixing. Thereafter the mixture was filtered through a Whatman no 1 filter paper into 100ml beaker and 2oml of 40% saturated solution of magnesium carbonate was added. The mixture was filtered through a Whatman no 1 filter to obtain a clear colourless solution. 1ml of the colourless solution was pipette into 50ml volumetric flask and 2ml of 5% ferric chloride was added. Distilled water was added to make up the volume. It was allowed to stand for some time. The absorbance of the sample as well as standard saponin solutions were read after colour development on a spectronic 21D spectrophotometer at a wavelength of 380nm.
\[
\% \text{ Saponin} = \frac{\text{absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Wt of sample} \times 10,000}
\]
II. Screening for phytate (Luca and Markakes, 1975)
The phytic acid was screened using the procedure described by Luca and Markakes. 2g of sample was weighed into 250ml conical flask. 100ml of 2% concentrated HCL was used to soak the sample in the conical flask for three hours. The solution was then filtered through a double layer of filter paper. 50ml of the filtrate was placed into a 250ml beaker and 107ml of distilled water was added into the beaker to give proper acidity.

10ml of 0.3% ammonium thiocyanide solution was added into the solution as an indicator. This was titrated with standard iron, chloride solution. The end colour was slightly brownish yellow and persisted for five minutes the percentage phytic acid was calculated using the formula below;
% phytic acid= X1.19 x 100
Where X= titre value x 0.00195g.

III. Alkaloid determination using Harborne (1973) method
5g of the sample was weighed into a 250 ml beaker and 50 ml of 10% acetic acid in ethanol was added and covered and allowed to stand for 4 hrs. This was filtered and the extract was concentrated on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract 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 residue is the alkaloid, which was dried in an oven at 60°C for 30 minute and weighed. The weight of alkaloid was determined by weight difference and expressed as a percentage of the sample weight analyzed, given by the formula: % alkaloid= (W2-W1)/W X 100
W= weight of sample
W1= weight of empty filter paper
W2= weight of paper + precipitate

IV. Determination of tannins using method of Person (1976)
The folins-denis spectrophotometric method was used. The method was described by Person (1976). 1.0g of sample was dispersed in 10ml distilled water and agitated. The sample was left to stand for 30 minutes at room temperature; it was shaken every 5 minutes. At the end of 30 minutes it was centrifuged. 2.5ml of the extract gotten was dispersed into a 50ml volumetric flask. 2.5ml of standard tannic acid was dispersed into a separate 50ml flask. 1.0ml folins-denis reagent was measured into each flask followed by 2.5ml 0f saturated
sodium carbonate solution. Distilled water was used to make up the mark, and incubated for 90 minutes at room temperature. The absorbance was measured at 250nm in a Genway model 6000 electronic spectrophotometer. Readings were taken with the reagent blank at zero.

V. Determination of oxalate using Fassett (1973) method

Oxalate determination was carried out as described by Fassett (1973). 2g of sample was boiled in 40ml water for 30 minutes. 10ml of Na$_2$CO$_3$ was added and boiled for another 30 minute. The liquid was extracted and the residue was washed with hot water until the wash water stopped showing alkaline reaction. The combination of the wash water and filtrate was concentrated to a small volume and cooled. HCL (1:1) was added drop-wise until the final acid concentration after neutralization was about 1%, a heavy precipitate appeared. The extract was filtered into a 250ml flask and the mark was made up. It was kept over-night, the supernatant was then filtered through a dry filter paper in a dry beaker. A small aliquot of the filtrate was taken int a 400ml beaker and made up to 200ml. 10ml of 10% calcium chloride solution was added and stirred to induce calcium oxalate precipitation. The precipitate was allowed to settle and the clear supernatant was decanted of through whitman No 42 filter paper. The precipitate was dissolved in HCL (1:1). Oxalic acid was precipitated by adjusting the pH with ammonium hydroxide solution. The contents were boiled and allowed to stay over- night. Oxalic acid was determined by titrating against 0.005 KMnO$_4$ solutions. The percentage oxalate was calculated as follows;

\[
1\text{ml of } 0.05N \text{ KMnO}_4 = 0.00225 \text{ anhydrous oxalic acid.}
\]

\[
\% \text{ oxalic acid} = \frac{\text{titre value} \times 0.00225}{2}
\]

\[
= \text{TU} \times 0.1125 , \text{ where TU is titre units.}
\]

VI. Determination of phenolic compounds (Fassett, 1973; Eastwood, 1986)

200\mu l of extract was added to a 50ml volumetric flask containing 25ml of water. 2.5 ml folins-denise reagent was added at the start of the experiment. After 3 minutes 5ml of saturated sodium carbonate was added and the volume was made up by water. The absorbance was measured after 20 minutes at 760nm.

VII. Screening for steroid using method of Brain (1968)

1g of extract was dissolved in chloroform and concentrated sulphuric acid was added. A red colour indicates the presence of steroids.
VIII. Screening for terpenoids using method of Harborne (1973)

5ml of aqueous extract of each plant sample was mixed with 2ml of CHCl₃ in a test tube; 3ml of concentrated H₂SO₄ was carefully added to the mixture to form a layer. An interface with a reddish brown coloration is formed if terpenoids constituent is present.

IX. Flavanoid determination by the method of Bohm and Kocipai-Abyazan (1994)

10g of the plant sample was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper No 42 (125 mm). The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

X. Glycosides determination using Harborne (1973) method

1ml of concentrated H₂SO₄ is prepared in test tube 5 ml of aqueous extract from each plant sample is mixed with 2ml of glacial CH₃CO₂H containing 1 drop of FeCl₃. The above mixture is carefully added to 1ml of concentrated H₂SO₄ so that the concentrated H₂SO₂ is underneath the mixture. If cardiac glycoside is present in the sample, a brown ring will appear indicating the presence of the cardiac glycoside constituent.

Extraction of plant materials

The leaves of Datura stramonium were collected from the wild within the premises of Kogi State University, Anyigba. The leaves were meshed with an enamel mortar and pestle. The leaves were then stored for further use.

Extract preparation

Different quantities of meshed leaves of Datura stramonium (2g, 1.5g, 0.75g) was measured and dissolved in 50ml of fresh palmwine. A stirrer was used to stir the solution to allow uniform mixing. The solution was filtered after a period of one hour using a glass funnel and filter paper. The filtrate from the solution was collected and stored in labeled plastic containers.

Animal grouping

Thirty rats (weighing between 100-150g) were used for this study. The rats were divided randomly into four groups, A, B, C, and D. The treatment groups consisted 3 subgroups designated A, B, and C and these were given 4mg/ml, 3mg/ml and 1.5 mg/ml of the extract...
respectively. While the control group, designated D, received equal volumes of fresh palm wine. Each treatment group consist nine rats while the control group consist three rats.

**Administration**

Palm wine extract of *Datura stramonium* was administered to the treatment groups (A, B, C) once daily. 0.5ml of the different concentrations of the extract was given to specific groups. Animals in the control group (D) were given 0.5ml of fresh palm wine daily. The extracts were administered orally for a period of twenty one days.

**Animal sacrifice**

Three rats from each treatment groups and one from the control group were sacrificed on this stated days (7, 14, 21) starting from the day administration commenced. Day 7 represents animals that were given appropriate dose and sacrificed after seven days, while day 14 represents rats that were given appropriate dose and sacrificed after fourteen days; same is applicable to day 21.

**Collection of blood and preparation of tissue**

The animals were sacrificed twenty four hours after the last dose. The animals were sacrificed by jugular puncture and the blood from the animals were collected into a plain EDTA bottle and centrifuged at a speed of 10,000rpm for five minutes. The serum obtained was used for further analysis. The organs (liver, kidney and intestine) were carefully extracted from the animals and fixed in 10% formalin. The organs were taken for histological examinations.

**Determination of total protein by Biuret method**

Serum total protein determination was carried out by Biuret method (1978).

**Principle**

Cupric ions, in an alkaline medium, interact with protein peptide bonds resulting in the formation of a coloured complex which is proportional to the amount of protein present.

**Methods**

1.0ml serum was collected into each labeled test tubes followed by the addition of 4.0ml Biuret reagent (sample1). The standard was prepared by mixing 4.0ml sample 1 and 1ml of prepared standard (1g/100ml B.S.A). The blank was prepared by mixing 1.0ml of distilled water and 4.0ml Biuret reagent and the absorbance was determined at 540nm.
Table 1: total protein content determination procedure

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>Standard</th>
<th>Blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>1.0</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>Standard serum albumin</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Distilled water</td>
<td>4.0</td>
<td>4.0</td>
<td>1.0</td>
</tr>
<tr>
<td>Biuret reagent</td>
<td>5.0</td>
<td>5.0</td>
<td>5.0</td>
</tr>
</tbody>
</table>

Calculations

Total protein = A sample × concentration of standard

\[
A = \text{absorbance}
\]

Reagent composition

Reagent 1 (Biuret reagent): This is composed of sodium hydroxide (100 mmol/L), Na-K-tartrate (16 mmol/L), Potassium iodide (15 mmol/L), cupric sulphate (6 mmol/L).

Reagent 2 (Blank reagent): this is composed of sodium hydroxide (100 mmol/L), Na-K-tartrate (16 mmol/L).

Determination of acid phosphatase by Hillmann (1977) method

Serum determination was carried out using the modified method of Hillmann (1977).

Principle

Acid phosphatase activity present in the serum was determined by a modified method of Hillmann (1977).

\[
\alpha\text{-naphthyl-phosphatate} + \text{water} \rightarrow \alpha\text{-naphthol} + \text{phosphate}
\]

\[
\alpha\text{-naphthol} + \text{phosphate} + \text{fast red TR} \rightarrow \text{Azo dye}
\]

\[
\alpha\text{-naphthol} \text{reacts with diazoted compound forming a colour with a maximum of absorbance of 450nm. Tartrate is used as a specific of the prostrate fraction.}
\]

Method

1.0ml of sample was measured into a test tube and 0.5ml of sample 1 (sodium citrate pH 5.2) was added. The mixture was equilibrated in a water bath for 10 minutes at 37° C. Sample 2 (\(\alpha\text{-naphthyl-phosphatate}\)) was added. The mixture was incubated for five minutes. The initial absorbance was read at one minute intervals thereafter for 3 minutes. The difference between the absorbance and average absorbance differences per minute (\(\Delta A/\text{min}\)) was then estimated.
Determination of aspartate amino transferase by method of Reitmena and Frankel (1957)

Principle

\[ \alpha\text{-oxoglutarate} + \text{L-aspartate} \rightarrow \text{L-glutamate} + \text{oxaloacetate} \]

AST is measured by monitoring the concentration of oxaloacetate hydrazone formed with 2, 4-dinitrophenylhydrazine.

Method

Test tubes were labeled blank and samples. 0.1ml of serum was pipette into the labeled test tube except the blank. 0.5ml sample 1 (reagent 1) was added to both the blank and test samples, the mixture were incubated for 30min at 37°C. 0.5ml of sample 2 (reagent 2) was added to both the blank and test samples, the mixture were allowed to stand for 20 min at 25°C. 5.0ml of sodium hydroxide was added to stop the reaction; the absorbance of the sample was read against the blank after 5 mins.

<table>
<thead>
<tr>
<th>Table 2: procedure for the determination of AST</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
</tr>
<tr>
<td>Sample 1</td>
</tr>
<tr>
<td>Sample 2</td>
</tr>
<tr>
<td>NaOH</td>
</tr>
<tr>
<td>Total (ml)</td>
</tr>
<tr>
<td>Blank</td>
</tr>
<tr>
<td>-------</td>
</tr>
<tr>
<td>0.1</td>
</tr>
<tr>
<td>0.5</td>
</tr>
<tr>
<td>0.5</td>
</tr>
<tr>
<td>5.0</td>
</tr>
<tr>
<td>6.0</td>
</tr>
</tbody>
</table>

Reagent Composition

Reagent 1: this is composed of phosphate buffer (100mmol/L), L-aspartate (100mmol/L), \(\alpha\)-oxoglutarate (2 mmol/L).

Reagent 2: 2, 4-dinitrophenylhydrazine.

Calculation

The activity of AST in the serum was obtained using the absorbance reading of the calibrator and unknown(s).

\[
\text{Abs. of unknown} \times \text{conc. of calibrator (IU/L)} = \text{AST}
\]

AST values

Determination of alanine amino transferase by method of Reitmen asnd Frankel (1957)

Principle

\[ \alpha\text{-oxoglutarate} + \text{L-alanine} \rightarrow \text{L-glutamate} + \text{pyruvate} \]
Alanine aminotransferase was measured by monitoring the concentration of pyruvate hydrazone formed with dinitrophenylhydrazine.

**Method**

Test tubes were labeled for each sample B (blank) and sample T (test). 0.1ml of serum was pipette into the labeled test tube. 0.5ml Reagent 1 was added to the test samples, the mixture was incubated for 30min at 37°C. 0.5ml of Reagent 2 was added to the test samples, the mixture was allowed to stand for 20min at 25 °C. 5.0ml of sodium hydroxide was added to stop the reaction; the absorbance of the sample was read against the blank after 5mins.

The blank was prepared by adding 0.5ml of reagent1 to the labeled test tube, 0.5ml of Reagent 2 was then added and the mixture was allowed to stand for 20 minutes at 25 °C. The reaction was stopped by addition of 5ml of sodium hydroxide. The absorbance was read at 560nm.

<table>
<thead>
<tr>
<th></th>
<th>Blank</th>
<th>Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>0.1</td>
<td></td>
</tr>
<tr>
<td>Sample 1</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Sample 2</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>NaOH</td>
<td>5.0</td>
<td>5.0</td>
</tr>
<tr>
<td>Total (ml)</td>
<td>6.0</td>
<td>6.1</td>
</tr>
</tbody>
</table>

**Table 3: procedure for determination of ALT**

Calculation

Activity of ALT in the serum was calculated using the absorbance reading of the calibrator and unknown(s).

\[
\text{Abs. of unknown \times conc. of calibrator (IU/L) = ALT}
\]

ALT values

**Reagent composition**

R1: this is composed of Buffer (100mmol/L, pH 7.4), Phosphate buffer (200mmol/L), L-alanine (2.0 mmol/L), α-oxoglutarate (2.0mmol/L). R 2: 2, 4-dinitrophenylhydrazine
RESULTS

Table 4 Phytochemical screening

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Concentration (mg/g)</th>
<th>Percentage (%)</th>
<th>Presence (+)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glycoside</td>
<td>1.15</td>
<td>0.60</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloid</td>
<td></td>
<td></td>
<td>+</td>
</tr>
<tr>
<td>Flavonoid</td>
<td></td>
<td></td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>3.16</td>
<td>0.64</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>20.04</td>
<td>0.84</td>
<td>+</td>
</tr>
<tr>
<td>Phenolic compound</td>
<td>0.64</td>
<td></td>
<td>++</td>
</tr>
<tr>
<td>Phytate</td>
<td></td>
<td></td>
<td>++</td>
</tr>
<tr>
<td>Oxalate</td>
<td></td>
<td></td>
<td>++</td>
</tr>
<tr>
<td>Terpenoid</td>
<td>0.016</td>
<td></td>
<td>++</td>
</tr>
<tr>
<td>Steroid</td>
<td></td>
<td></td>
<td>++</td>
</tr>
</tbody>
</table>

RESULTS OF SERUM ENZYME ACTIVITY

Results were subjected to statistical analysis (one way-ANOVA).

Table 5: Result for total protein

<table>
<thead>
<tr>
<th>Groups</th>
<th>Concentration per days (mg/ml or units)</th>
<th>(day 7)</th>
<th>(day 14)</th>
<th>(day 21)</th>
</tr>
</thead>
<tbody>
<tr>
<td>D (control)</td>
<td>15.77± 0.00&lt;sup&gt;a&lt;/sup&gt;</td>
<td>15.77± 0.00&lt;sup&gt;a&lt;/sup&gt;</td>
<td>15.77± 0.00&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>A(4 mg/ml)</td>
<td>13.23± 1.19&lt;sup&gt;a,b&lt;/sup&gt;</td>
<td>12.45± 0.29&lt;sup&gt;b&lt;/sup&gt;</td>
<td>10.28± 4.26&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>B(3 mg/ml)</td>
<td>12.90± 1.57&lt;sup&gt;a,b&lt;/sup&gt;</td>
<td>11.07± 0.37&lt;sup&gt;c&lt;/sup&gt;</td>
<td>13.46± 0.44&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>C(1.5 mg/ml)</td>
<td>11.03± 1.58&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.30± 0.57&lt;sup&gt;b,c&lt;/sup&gt;</td>
<td>12.76± 1.20&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

a, b- mean with same superscript not significant (p>0.05)

a, b- mean with different superscript is significant (p<0.05)

Table 6: Results for acid phosphatase activity

<table>
<thead>
<tr>
<th>Groups</th>
<th>Activity /Units</th>
<th>7</th>
<th>14</th>
<th>21</th>
</tr>
</thead>
<tbody>
<tr>
<td>D (control)</td>
<td>24.75± 0.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.75± 0.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.75± 0.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>A(4mg/ml)</td>
<td>36.28± 0.89&lt;sup&gt;a&lt;/sup&gt;</td>
<td>28.50± 2.25&lt;sup&gt;a,b&lt;/sup&gt;</td>
<td>35.20± 1.01&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>B (3mg/ml)</td>
<td>36.20± 0.45&lt;sup&gt;a&lt;/sup&gt;</td>
<td>28.37± 2.19&lt;sup&gt;a,b&lt;/sup&gt;</td>
<td>35.43± 1.34&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>C (1.5 mg/ml)</td>
<td>36.61± 0.29&lt;sup&gt;a&lt;/sup&gt;</td>
<td>32.40± 2.55&lt;sup&gt;a&lt;/sup&gt;</td>
<td>30.77± 3.53&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

a, b- mean with same superscript not significant (p>0.05)

a, b- mean with different superscript is significant (p<0.05)
Table 7: Result for aspartate amino transferase activity

<table>
<thead>
<tr>
<th></th>
<th>Activity /Unit</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(day 7)</td>
<td>(day 14)</td>
<td>(day 21)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CONTROL</td>
<td>25.32 ± 0.00a</td>
<td>25.32 ± 0.00a</td>
<td>25.32 ± 0.00a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>26.73 ± 1.48a</td>
<td>25.72 ± 1.78a</td>
<td>25.92 ± 1.89a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>26.40 ± 1.92a</td>
<td>25.32 ± 1.67a</td>
<td>25.93 ± 2.15a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>27.47 ± 1.52a</td>
<td>25.51 ± 1.79a</td>
<td>27.78 ± 0.19a</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a, b- mean with same superscript not significant (p>0.05)
a, b- mean with different superscript is significant (p<0.05)

Table 8: Result for alanine amino transferase activity

<table>
<thead>
<tr>
<th></th>
<th>Activity /Unit</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(day 7)</td>
<td>(day 14)</td>
<td>(day 21)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CONTROL</td>
<td>21.07 ± 0.00a</td>
<td>21.07 ± 0.00a</td>
<td>21.07 ± 0.00a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>25.50 ± 2.45a</td>
<td>24.83 ± 2.77a</td>
<td>20.63 ± 1.21a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>25.29 ± 2.31a</td>
<td>24.57 ± 2.81a</td>
<td>20.25 ± 1.49a</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>24.87 ± 3.10a</td>
<td>23.98 ± 2.94a</td>
<td>22.10 ± 0.94a</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a, b- mean with same superscript not significant (p>0.05)
a, b- mean with different superscript is significant (p<0.05)

RESULT OF HISTOLOGICAL ANALYSIS OF ORGANS OF EXPERIMENTAL ANIMALS (PALMWINE EXTRACT OF DATURA STRAMONIUM)

Fig: 3: section of normal intestinal tissue (control) (H and E X40)
Fig: 4: section of normal kidney (control) (H and E X 100)

Fig: 5: section of normal liver (control) (H and E X 100)

WEEK1 (treatment group)

Fig: 6: section of intestinal tissue (treatment group A) (H and E X 100)
Fig: 7: section of normal kidney (treatment group A) (H and E X 100)

Fig: 8: section of normal liver (treatment group A) (H and E X 100)

Fig: 9: section of intestinal tissue showing mucosal erosion (treatment group B) (H and E X 100)
Fig: 10: section of normal kidney (treatment group B) (H and E X 100)

Fig: 11: section of liver (treatment group B) (H and E X 100)

WEEK 3

Fig: 12: section of intestinal tissue showing mucosal erosion (treatment group C) (H and E X40)
Fig: 13: section of normal kidney (treatment group C) (H and E X400)

Fig: 14: section of liver showing intracellular accumulations (treatment group C) (H and E X400)

Table 9: Result of histological analysis

<table>
<thead>
<tr>
<th></th>
<th>INTESTINE</th>
<th>KIDNEY</th>
<th>LIVER</th>
</tr>
</thead>
<tbody>
<tr>
<td>CONTROL PALMWINE</td>
<td>NORMAL</td>
<td>NORMAL</td>
<td>NORMAL</td>
</tr>
<tr>
<td>WEEK 1 TREATMENT</td>
<td>NORMAL</td>
<td>NORMAL</td>
<td>NORMAL</td>
</tr>
<tr>
<td>WEEK 2 TREATMENT</td>
<td>MUCOSAL EROSION</td>
<td>NORMAL</td>
<td>NORMAL</td>
</tr>
<tr>
<td>WEEK 3 TREATMENT</td>
<td>MUCOSAL EROSION</td>
<td>NORMAL</td>
<td>INTRACELLULAR ACCUMULATIONS</td>
</tr>
</tbody>
</table>

Magnifications: X 40, X 100, X 400

Stains: H&E (Haematoxylin and Eosin)
HISTOLOGIC REPORT

KIDNEY
NORMAL – Normocellular Glomerular tufts displayed on a background containing tubules. No necrosis.

LIVER
NORMAL - Histologic section of liver shows preservation of hepatic architecture. The hepatocytes are arranged as parallel plates, and extend from the portal tracts to the central veins. The sinusoids are not congested. No areas of necrosis haemorrhage or inflammation is seen. No intracellular inclusions are present.

ACCUMULATIONS – Histologic section of liver show hepatocytes with mottled appearance due to presence of cytoplasmic intracellular accumulations (lipid or glycogen).

INTESTINE
NORMAL – Histologic section of intestinal tissue showing lining cells composed of tall columnar cells with interspersed mucous containing goblet cells.

MUCOSAL EROSION - Histologic section of intestinal tissue showing erosion of mucosal lining exposing basally placed glands.

DISCUSSION AND CONCLUSION
The result from the phytochemical screening showed that the plant contains saponin, tannin, oxalate, phenol, steroids, terpenoids, flavonoids and glycoside. The bioactive agents found in the plant showed that, the plant has various potential as a drug and as a stimulant. The presence of flavonoids shows that the plant has anti-inflammatory effects since flavonoids are anti-inflammatory agents (Sonika, 2010).

The presence of alkaloids showed that the plant has anti-inflammatory effects, having anticholinergic and bronchodilating activity. Charpin et al., (1979) reported that using D. stramonium as an antiasthmatic cigarette is an effective bronchodilator in asthmatic patients with mild airway obstruction. The hallucinogenic effect observed may be due to the presence of the alkaloid scopolamine (Damilare et al., 2010).

The animals in the treatment groups displayed aggressive and erratic resistance to handling throughout the duration of this study. The animals of treated group A showed restlessness and...
excitation for a much longer period, followed by animals in group B and then by the animals in group C. At about 20 to 30 minutes after administration, the animals in the treated groups began to show sign of calmness. The animals isolated themselves and avoided response to external events. Dilatation of the pupil was noted upon observing the eyes. Mydriasis is a common consequence of the anticholinergic activity of the biochemical components of the plant, as suggested by Thabet et al., (1999) and Pekdemir et al., (2004). The animals in both the treatment and control groups were frequently checked for any alteration or changes in their behavioral responses.

Following the administration of *D. stramonium* extract to the experimental animals in the treatment groups, the average body weight in each of the experimental groups A, B, C, and D were calculated on day 7, 14 and 21 of the experimental procedures. Anorexia (a reduction in food consumption and body weight) was observed in all the rats in the treatment groups administered 4mg/ml, 3mg/ml, and 1.5mg/ml of the plant extract respectively. Food intake was significantly reduced following the administration of the plant extract in a dose-dependent pattern. Since food consumption was reduced, similar reductions in body weight were observed during the study. Acid, hematoxylin and eosin (H&E) staining techniques revealed that oral administration of aqueous extracts of *Datura stramonium* had deleterious effects on the intestine and liver of the animals of the extract-treated groups since histological analysis showed mucosal erosion of the intestine and intracellular accumulations in the liver of the animals in the treated groups when compared to the control group. However there was no disruption of the integrity of the kidney.

The decrease in body weight of the animals in all the treatment groups as recorded in this study could have been a result of anorexia following oral administration of the aqueous leaf extracts of *Datura stramonium*. Akinlolu and Shokunbi (2010) suggested that decreases in body weight could also occur as a result of the negative impacts of drugs on the normal biological, biochemical, physiological and metabolic processes, with consequent depletion of body protein in the treated animals. The decrease in body weight of the extract treated animals reflected the possible changes in function of the organs that are regulated by the nervous system and metabolic activities. The active constituents of the aqueous leaf extract, mainly tropane alkaloids, prevent the action(s) of the essential neurotransmitter (acetylcholine) in the brain through blockade of its receptors (Henry and Wiseman, 1997).
From the result of serum total protein concentrations obtained (table 5), it was observed that there was a gradual decrease in the serum total protein as dosage increased within the first and second week of administration, while the animals in the third week showed no significant decrease in total serum protein. This may be due to impairment in protein metabolism by the plant extract.

From the result of serum acid phosphatase obtained (table 6), it was observed that there was a gradual increase in serum acid phosphatase of the treatment group when compared to the control. This result suggests that there was an impaired functionality in the liver which caused an increase in the serum acid phosphatase level. This may be due to damage to the prostate gland, which affects the activity of the enzyme (Ngaha et al., 1989).

The result obtained from aspartate amino transferase (table 7), showed that there was no significant difference between the control group and the treatment groups for each week. The result gotten from this table showed that the plant extract may have no toxic effect on the activity of this enzyme at the administered dosage.

The result obtained from Alanine Amino Transferase (table 8), showed that there was no significant difference between the control group and the treatment groups for each week. The result gotten from this table showed that the plant extract has no toxic effect on the activity of this enzyme at the administered dosage.

Although the plant has been reported to have many beneficial and medicinal properties, its side effects (such as hallucinations, psychiatric derangement, disorientation with agitated behavior, etc.) could have been a result of the insults of the chemical components of the plant to the cells of the nervous system. It was observed that the plant has the potential to cause liver and intestinal damage, as well as decrease functionality of enzymes in the liver, and a negative impact on body weight.

Furthermore, there was no adverse effect noticed on physical examination of the experimental animals.

CONCLUSION

The results obtained shows that the plant has deleterious effects when administered in high dosage. The phytochemical screening also showed that the plant could be used as a drug due to the presence of various bioactive agents. The increased serum concentration of hepatic
profiles monitored showed that the plant is deleterious to body organs. Histological analysis revealed the presence of intracellular accumulations (glycogen or lipids) in the liver which may be due to the deleterious effects of the plant extract.

RECOMMENDATION
Further studies should be directed towards isolating the specific component(s) of the plant responsible for the deleterious effects in order to standardize the plant preparation for maximum therapeutic benefit. Further studies should also be carried out on the plant with an increased oral dosage. Other parts of the plants such as the seed and the stem should be screen for potential toxins. The effect of higher dosage on biochemical parameters would be of utmost importance for further research.

REFERENCES


43. Persinger O. MA (2004). Herbal treatment following postseizure induction in rat by lithium pilocarpine: *Scutellaria lateriflora* (Skullcap), *Gelsemium sempervirens* (Gelsemium) and *Datura stramonium* (Jimson weed) may prevent development of spontaneous seizures. Phytother Res. 18(9): 700-705.


