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EFFECT OF ETHANOL EXTRACT OF CELERY (*APIUM GRSVEOLENS*) ON SOME BIOCHEMICAL PARAMETERS IN ACETAIMINOPHEN INDUCED WISTAR ALBINO RATS FED WITH FATTY DIET

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ABSTRACT
This study evaluated the effect of celery (*Apium graveolens*) on paracetamol-induced male albino rats fed with high fat diet. 24 wistar albino rats were grouped into four (4) groups each having six (6) animals. Animals in Group 2, 3 and 4 were fed with high fat diet and induced orally with 750mg/kg body weight of Paracetamol(Acetaminophen) afterwhich group 3 and 4 received 200mg/kg and 400mg/kg of celery (*Apium graveolens*) extract respectively and group 2 served as positive control. Biochemical analysis revealed that induction of paracetamol resulted in significant increase ($p<0.05$) in ALT and AST in positive control (1510.67±11.36 and 75.4 ± 3.2) respectively when compared with the normal control groups (715.00±18.4 and 63.5 ± 2.6). However, there was a non-significant ($p<0.05$) decrease in the level of ALP levels in positive control (186.69±4.05). Levels of LDH decreased significantly ($p<0.05$) in positive control as time progressed from 30seconds to 3minutes (1136.51±13.53 to 1195.56±66.53). ALT and AST significantly decreased in extract treated groups 3 and 4 (498.67±17.96 and 553.67±54.63) respectively compared to positive control (1510.67±11.36). ALP levels increased non-significantly ($p<0.05$) in extract treated group 3 and 4 (30.89±1.26 and 30.89±1.99) respectively compared to positive control (29.67±1.00). LDH levels increased significantly ($p<0.05$) in extract treated groups 3 and 4 as time progressed from 30sec to 3mins (1287.78±14.86 and 1205.77±22.16) respectively compared to positive control (1136.51±13.53 to 1195.56±66.53). However, a non-significant difference was observed in positive control group when compared to normal control. Histopathological examinations of liver sections from normal control group shows normal kidney histology while those in group 2 (positive control) showed damage to hepatocytes, however groups 3 and 4 though similar to the control showed signs of inflammatory cells which is indicative of healing process. Findings from these study shows that the extract of Celery (*Apium graveolents*) confers some degree of protective function on the liver and biochemical parameters of paracetamol-induced rats fed with high fat diet.Biochemical analysis revealed that induction of acetaminophen resulted in significant increase ($p<0.05$) in cholesterol and glucose in positive control (301.83±4.82 and 19.03±1.04) respectively when compared with the normal control groups (206.01±2.41 and 39.16±2.08). However, there was a significant ($p<0.05$) decrease in the level of triglycerides, LDL, Cholesterol and Glucose in test groups 1 and 2 when compared to the positive control (188.62±1.47 and 177.02±2.27), (76.95±0.81 and 20.42±0.79), (242.75±4.54 and 201.51±5.56) and (27.18±0.67 and 19.03±1.04) respectively. However, HDL levels increased (208.33±2.14 and 212.99±2.46). Histopathological examinations of kidney sections for normal control group shows normal kidney histology while those in group 2 (positive control) showed necrosis of the renal cortex, however test groups 3 and 4 though similar to the control have fewer glomeruli. Findings from these study shows that the extract of Celery (*Apium graveolents*) confers some degree of protective function on the kidney and biochemical parameters of acetaminophen-induced rats fed with fatty diet.

KEYWORDS: *Paracetamol, Apium graveolens, Histopathological, Cholesterol, Glucose.*

INTRODUCTION
Paracetamol, also known as acetaminophen (APAP), is a medication used to treat pain and fever. It is typically used for mild to moderate pain relief (American Society of Health-System Pharmacists, 2016). There is mixed evidence for its use to relieve fever in children.Paracetamol is also used for severe pain, such as cancer pain and pain after surgery, in combination with opioid pain medication. It is typically used either by mouth or rectally, but is also available by injection into a

vein. Effects last between 2 to 4 hours (Daniel, 2014). Paracetamol is generally safe at recommended doses. The recommended maximum daily dose for an adult is 3 or 4 grams. Higher doses may lead to toxicity, including liver failure. Serious skin rashes may rarely occur. It appears to be safe during pregnancy and when breastfeeding.Nevertheless, many researchers have tried to find ways to reduce the adverse effects associated with paracetamol use through application of medical herbs and different natural products, such as as antioxidants from natural sources.

Celery (*Apium graveolens*) is amedicinal plant in traditional medicine with numerous health benefits. Celery is involved in the prevention of cardiovascular disease, lowering blood glucose in diabetic mice, lowering blood pressure and strengthening the heart (Lans, 2006). Experimental studies report antifungal and anti-inflammatory effects of celery. Celery has an anticoagulant activity. Its root leads to an increase of calcium and decrease of potassium in the heart tissue (Bernard andStiehl, 1986). Essential oil of celery has antibacterial effects. This plant has cooperation in the molecular mechanisms and cellular targets that have a significant effect on the treatment of human cancers (Atta, 1998).

Liver injury or liver dysfunction induced by acetaminophen, chemotherapeutic agents, carbon tetrachloride, alcohol, dantrolene sodium, valproic acid and isonicotinic acid hydrazide, etc., is a major health problem. Recently, liver injury has become one of the major problems in aquaculture; many have been suffering from the ‘‘liver and gall syndrome’’, with the symptom of liver enlargement (up to twofold of original size) and colour change. The causes of this disease are not clear; pathogenic bacteria or viruses have not been identified. Xenobiotic challenge due to drug abuse and environmental pollution may be one of the most important causes of the disease. To prevent and control hepatic diseases, a large quantity of antibiotics and chemicals have been produced which may, in turn, cause problems to consumers.

It has been reported that, among a variety of drugs, acetaminophen (APAP, N-acetyl-4-aminophenol) is the most common cause of drug-induced liver injury (Ostapowicz et al., 2002). APAP is a non-steroidal anti-inflammatory drug (NSAID) commonly known as paracetamol, which has been widely used for over 50 years for the effective treatment as an analgesic and antipyretic at therapeutic doses (Xu et al., 2008). According to FDA; currently, 235 approved prescriptions and drug products contain APAP as an active ingredient. Although APAP is a well-known analgesic and antipyretic, its overdose (3 h following 500 mg APAP/kg body weight) causes acute liver failure (Hinson et al., 1998).

Acetaminophen metabolism generates the toxic metabolite, N-acetyl-p-benzoquinone imine (NAPQI), and it is efficiently detoxified by glutathione (GSH), which is an important cellular antioxidant for detoxification of drugs and foreign chemicals. Therefore, an overdose will saturate the conjugation pathways of GSH and cause depletion of cellular GSH. This subsequently leads to a reduced capacity of GSH to detoxify NAPQI. It has been noted that APAP overdose depletes intracellular glutathione within 1–4 h (Al-Turk and Stohs, 1981; Lores Arnaiz et al., 1995), resulting in accumulation of intracellular reactive oxygen and nitrogen species (ROS/ RNS), causing oxidative/ nitrosative stress and thus enhancing cellular injuries and organ dysfunction including renal damage (Hart et al., 1994). The overproduced NAPQI can covalently bind to the cysteinyl thiol groups of cellular proteins and form protein-(cystein-S-yl)-APAP adducts, which may impair protein functioning. An increased NAPQI also induces/mediates the production of reactive oxygen species. Experimental studies have shown that a toxic dose of APAP produces hepatic necrosis in both humans and animals and also causes oxidative stress by depleting GSH and resulting in mitochondrial damage and cellular death (Agarwal et al., 2011).

Celery (*Apium graveolens*; family, *Apiaceae*) is used in medicine owing to its richness in flavonoids and antioxidant property for the treatment for liver ailments. Studies have indicated that celery lowers blood pressure, regulates heart function and reduces complications of diabetes (MimicaDukic´ and Popovic´ 2007). The healing property of celery is due to its bioactive compounds like rutein, quercetin, luteolin, kaempherol, apigenin and myricetin. (Mimica-Dukic´ and Popovic´ 2007). Existing literature on the protective effect of A. graveolens on APAP-induced hepatotoxicity is little. Thus, we hypothesized that flavonoids rich in celery would prevent APAP-linked abnormalities.

METHODS

Materials

Fresh apium graveolens leaves, gavage, beaker, weighing balance, cotton wool, masking tape, spectrophotometer (Spectrum lab 7555), micropipette, test tube, test tube rack, refrigerator, latex hand gloves, water bath, centrifuge (BL-110), syringes, Dissecting sets, Methylated spirit, stirring rod, pasteur pipette, measuring cylinder, weighing balance, fine test glucometer, fine test strips, plain sample bottles, fluoride oxalate bottles, universal bottles.

Sample Collection

The plant (*celery apium graveolens*) was purchased from Kajuru, Kaduna State, Nigeria and identified by Prof. Alikwe Philip from the faculty of Agricultural Science, Niger Delta University Wilberforce Island Amassoma, Bayelsa State with Voucher number NDU/FA/CS/CEL.001.

Experimental Animals

Twenty four (24) male wistar albino rats weighing from 123-160g were purchased from Vet care pharmaceuticals, Port Harcourt, Rivers state. After acclimatization period of one week, during which the animals were fed with commercial diet (pelletized growers food) and water. the rats were maintained under standard housing conditions (photoperiod: 12h natural light and 12h dark). the rats were divided into four groups, group 1 (control), group 2 (positive control), group 3 (test 1), group 4 (test 2).

Feeds

Group 1 were fed with pelletized growers feed and clean water throughout the experiment (21 days).

Groups 2, 3 and 4 were fed with egg yolk mixed with the pelletized growers feed in a ratio 1:5 after acclimatization (14 days).

Preparation of Extracts

100g of grinded celery apium graveolens was dissolved in 1000ml of ethanol and allowed to stand at room temperature for 72 hours. Afterwards, it was sieved with the aid of a sterile cheese cloth. The filtrate was kept in a water bath at $60^{O}C$ for 48hrs till the ethanol perspired and a paste was formed. The paste was obtained and kept in sample bottle and stored in a refridgerator at $4^{O}C$ ($34^{O}F$) till the time of use. 20g of the extract was dissolved 100ml of normal saline which was administered orally.

Administration of Extract

Ethanolic extract of celery apium graveolens was administered to the experimental animals as stated below;

Group 3 was administered 200mg/kg b/w of the leaf extract.

Group 4 was administered 400mg/kg b/w of the leaf extract.

Blood Collection

On day zero (0), day ten (10) and day fifteen (15), rats were immobilized with a carton before blood collection for fasting blood glucose level using a fine test strip and a fine test glucometer. the tail of the rats was sterilized with a sterile swab dabbed in methylated spirit and lubricated with a brand of petroleum jelly to reduce friction. A gentle and persisitent massage was applied until the tip of the tail became reddish (indicating blood accumulation). A sterile blade was used to make an incision at the tip of the tail and a drop of blood was placed on the tip of the test strip which was already inserted into the glucometer. After blood collection of the tail, the tail was cleaned with a swab dabbed in methylated spirit to prevent infection and then a dry sterile swab to stop the bleeding. On day twenty-one (21) the rats were anaesthesized using chloroform and the blood was obtained using a 5ml syringe directly from the heart. The blood was collected into the sample bottles (Plain bottle and Flouride Oxalate) for triglyceride assay, total cholesterol assay, HDL and plasma glucose assay.

Experimental Design

The healthy male albino rats of wistar strain, after acclimatization for a period of one week were randomly distributed to four (4) groups, with six (6) groups in each group.

Group 1: Normal food + Distilled water + 200mg/kg normal saline.

Group 2: Normal food + Egg yolk + Distilled water + single dose of 750mg/kg b/w nof Acetaiminophen.

Group 3: Normal food + Egg yolk + Distilled water + single dose of 750mg/kg b/w nof Acetaminophen. + 200mg/kg b/w of leaf extract.

Group 4: Normal food + Egg yolk + Distilled water + single dose of 750mg/kg b/w nof Acetaminophen. + 400mg/kg b/w of leaf extract.

Biochemical Assay

Determination of Biochemical Parameters Levels

Determination of Lactate Dehydrogenase Assay (LDH)

Principle

This is an optimized standard method according to the recommendations of the Deutsche Gesellschaft fur Klinische Chemie.

$$\text{Pyruvate} + \text{NADH} + H^{+} \longrightarrow \text{L-lactate} + NAD^{+}$$

Procedure

0.02ml of blood (plasma) of the Wister albino rats was pipetted into test-tubes containing 1ml of the reagent (LDH kit), it was thoroughly mixed, incubated for 0.5 minutes at 37 degree Celsius and the absorbance value (340nm) was measured or read against the reagent blank. It was read again after 1, 2 and 3 min.

R1a: Buffer (Phosphate buffer 50mmol/l, pH 7.5, Pyruvate 0.6mmol/l)

R1b: NADH (0.18mmol/l).

LDH concentration (U/I) = $8095 \times \Delta A$ 340nm

Determination of Aspartate Transaminase (AST) Level

PRINCIPLE:

$$\alpha\text{-oxoglutarate} + \text{L-aspartate} \xrightarrow{\text{AST}} \text{L-glutamate} + \text{Oxaloacetate}$$

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

PROCEDURE

Sodium hydroxide solution (0.4 mol/L) was prepared.

Eight test tubes were arranged in a test tube rack in two rolls, with four test tubes in each roll. Test tubes in the first roll were labeled Reagent blank 1,2,3,4 whereas, test tubes in the second roll were labeled sample 1,2,3,4 respectively.

0.1ml of distilled water and 0.5ml of Reagent 1(buffer) was pipetted into test tubes labeled Reagent blank, 0.1ml of sample and 0.5 ml of Reagent 1 was pipetted into test tubes labeled sample. The mixtures were thoroughly mixed and incubated for exactly 30 minutes at 37˚C. 0.5ml of Reagent 2 (2,4-dinitrophenylhydrazine) was pipetted into all test tubes, mixed thoroughly and allowed to stand for exactly 20 minutes at 25˚C.

Finally, 5ml of Sodium hydroxide was pipetted into all test tubes, mixed thoroughly and taken to a spectrophotometer where the absorbance of the sample (A_{sample}) against the reagent blank was read after 5 minutes, at the wavelength [Hg 546nm].

DETERMINATION OF ALKALINE PHOSPHATASE (ALP) ASSAY

ALP was determined using the method of DeutsheGesellschaft fur KlinischeChemie as outlined in Randox Kit.

Principle

p-nitrophenylphosphate + H_2O $\xrightarrow{\textbf{ALP}}$ phosphate + p-nitrophenol.

Alkaline phosphatase was measured by monitoring the concentration of p-nitrophenol formed with p-nitrophenylphosphate Reagent (R)

R1a: Buffer (Diethanolamine buffer 1mol/l, pH 9.8, L- alanine 200mmol/l, α-oxoglutarate 2.0mmol/l)
R1b: p-nitrophenylphosphate 10mmol/l

Procedure

One vial of R1b was reconstituted with the appropriate volume (10ml) of buffer R1a. The test tubes were labeled with the corresponding samples. The reconstituted Reagent (0.50ml) was added to the sample (0.01 ml). They were mixed and the absorbance read at intervals of 1,2 and 3mins at 405nm.

The calculation was done thus: the activity of ALP in the serum is obtained from table. (See Results Table 1).
2760 X change in absorbance at 405 nm/min.

Determination of Alanine Transferase (ALT) Activity

ALT activity was determined following the principles described by Reitman and Frankel (1957).

Principle

α – Oxidoglutarate + L-Alanine $\xrightarrow{AST}$ L-Pyruvate + oxaloacetate

Table: Assay of ALT activity was done in agreement with the procedure described by Randox laboratory Ltd. United Kingdom.

Phosphate buffer	100mmol/L, $_P$H 7.4
L-alanine	100mmol/L
α-oxoglutarate	2mmol/L
2,4-dinitrophenylhydrazine	2mmol/L
Sodium hydroxide	0.4ml

Procedure

Briefly, 0.1ml of diluted sample was mixed with phosphate buffer, L-aspartate and α-oxoglutarate and the mixture incubated for exactly 30 minutes at 37ºC. 0.5ml of 2,4-dinitrophenylhydrazine was added to the reaction mixture and allowed to stand for exactly 20 minutes at 25ºC. Then 5.0ml of NaOH was added and the absorbance was read against the reagent blank after 5 minutes at 546nm.

ALT assay medium

The test tubes were labelled reagent blank and sample. The reagent blank was prepared with 0.5ml of solution 1 (Randox AST), 0.1ml of distilled water while the test was prepared by diluting 0.1ml of the sample, 0.1ml of solution 1 (Randox AST). then mix and incubate for 30 minutes at 37ºC. into each of the test tubes 0.5ml of solution 2 (Randox AST) was added, mixed and allowed to stand for exactly 20 minutes at 25ºC then 0.5ml of sodium hydroxide was added into the test tube and absorbance was read against the reagent blank after 5 minutes at 546nm.

Glucose Assay

Principle

Glucose was determined after enzymatic oxidation in the presence of glucose oxidase as described by Randox Laboratories limited, England. The hydrogen peroxide formed reacts under the catalysis of peroxidase with phenol and 4-aminophenozone to form a red violet quinoemine dye as indicator.

Procedure

Exactly 10µl of plasma of blood collected of blood collected in fluoride oxalate bottle was transferred into a test tubes containing 1000µl of the reagent. Samples were thoroughly mixed and incubated at 37 for 10 minutes. The absorbance values were read at 540nm using the glucose blank as blank.
Glucose concentration was calculated using
Glucose (mg/dl) = Abs of sample – Abs of standard/Abs of sample X conc. of standard (mg/dl)

Cholesterol Assay

The cholesterol is determined after enzymatic hydrolysis and oxidation. The indicator quinoneimine is formed from hydrogen peroxide and 4-aminoantipyrine in the presence of phenol and peroxidase.

$$\text{Cholesterol ester} + H_2O \xrightarrow{\text{Cholesterol esterase}} \text{Cholesterol} + \text{Fatty Acids}$$

$$\text{Cholesterol} + O_2 \xrightarrow{\text{Cholesterol Oxidase}} \text{Cholesteene-3-one} + H_2O_2$$

$$2H_2O_2 + \text{phenol} + \text{4-aminoantipyrine} \xrightarrow{\text{peroxidas}} \text{Quinoneimine} + 4H_2O$$

Procedure
0.01ml of blood (serum) of the Wister albino rats was pipetted into test-tubes containing 1000µl of the reagent (Total cholesterol kit), it was thoroughly mixed, incubated for 10 minutes at 37 degree Celsius and the absorbance value was measured or read against the reagent blank within 60 minutes at a wavelength of 546nm.

Reagents
R1: Buffer (Pipes buffer 80mmol/l, pH 6.8, 4-aminoantipyrine 0.25mmol/l, Phenol 6mmol/l, Peroxidase ≥0.5U/ml, Cholesterol esterase ≥0.5U/ml, Cholesterol oxidase ≥0.10U/ml).

Calculation
Cholesterol concentration (mg/dl)=$A_{sample}/A_{standard}$× Standard conc. (mg/dl)

Triacylglycerol Assay
Principle
Triacylglycerols were determined after enzymatic hydrolysis with lipases. The indicator is a quinone formed from hydrogen-peroxide, 4-aminophenazene and 4-chlorophenol under the catalytic influence of peroxidase.

Procedure
0.1ml of blood (plasma) of the Wister albino rats was pippetted into test-tubes containing 1ml of the reagent (Triacylglycerol kit), it was thoroughly mixed, incubated for 5 minutes at 37 degree Celsius and the absorbance value was measured or read against the reagent blank within 60 minutes at a wavelength of 500nm.

R1a: Buffer (Pipes buffer 40mmol/l, pH 7.6, 4-chloro-phenol 5.5mmol/l magnesium-ions 17.5mmol/l).
R1b: Enzyme (4-aminophenazone 0.5mmol/l, ATP 1.0mmol/l, Lipases ≥150 U/ml, Glycerol-kinase ≥ 0.4 U/ml, Glycerol-3-phosphate oxidase ≥1.5 U/ml, Peroxidase ≥ 0.5U/ml).

Triacylglycerol concentration (mg/dl)=$A_{sample}/A_{standard}$× Standard conc. (mg/dl)

High Density lipoprotein
Prinicple
Plasma HDL was estimated using the spectrophotometric methods in the kit supplied by Randox laboratories Limited, 55 Diamond Road, Crumlin Country Aantrim, BT29 4QY, United Kingdom.

Procedure
Test tubes were labeled blank, standard, control, test 1, test 2. A total of 24 test tubes were used. 6 for control, 6 test 1 and 6 for test 2.
200µl of sample was pipette into all the test tubes.
500µl of reagent was added to the respective test tubes and mixed.
The mixture was then centrifuged at 3000g for 10minutes to separate the standard supernatant and the supernatant.
100µl of the supernatant was pipette into test tubes and 1000µl of reagent was added and mixed.
The mixture was incubated for 5 minutes at 37^{O}C.
Absorbance of samples was read using the spectrophotometer (S23A techmel and techmel USA) and results recorded.

Calculations
HDL content of sample (mg/dl) = $\frac{Asample}{Astandard} \times Concentration\ of\ standard\ (mg/dl)$
Concentration of Standard = 203mg/dl
Absorbance of Standard = 0.432

Low Density Lipoprotein Cholesterol (LDL)
Principle
The LDL Direct Cholesterol assay is a homogenous method for direct quantitative determination of low density lipoprotein cholesterol (LDL-C) levels in human serum or plasma , without the need for any-line pretreatment or centrifugation steps.

Procedure
The determination of serum LDL cholesterol has been recognized by using Friedwald equation to calculate the LDL cholesterol based on results from three (3) separate assays: Total cholesterol, HDL cholesterol and Triglycerides.

Formula for calculating Low Density Lipoprotein (LDL)
Total Cholesterol – High Density Lipoprotein (HDL) – Triglycerides/5.

Statistical Analysis
SPSS version 17 (SPSS Inc, Chicago, USA) statistical package was used for descriptive statistics and analysis of variance was carried out, while Duncan Multiple Range Test was used to separate the means at P≤0.05.

RESULTS
The effect of *celery apium graveolens* on ALT, ALP, AST and LDH levels in wistar albino rats induced with

paracetamol is given below. Also Presented in Table 4.1 are the mean body weights of the wistar albino rats on Day 3 and before inducing.

Body weight in grams of rats

Table 4.1 Mean body weight of Wistar albino rats on Day 3 and Before Induction.

Groups	On Day 3	Before Inducing
Control	162.33±14.81[a]	135.38±16.05[a]
+Ve control	163.83±16.62[a]	116.78±20.85[a]
Test group 1	145.33±12.31[c]	123.1±7.52[c]
Test group 2	190.83±20.12[d]	160.47±17.43[d]

From the above table it is observed that the weight of wistar albino rats ranged from 145-190g on Day 3 of acclimatization and also the range of weight of the rats before inducing was from 116-160g.

Table 4.2: Effect of *Celery Apium graveolens* on Lactate Dehydrogenase (LDH) level of paracetamol-induced wistar albino rats.

	30sec	1 min	2 min	3 min
Control	1303.45±10.44[a]	1229.46±37.56[a]	1240.39±27.01[a]	1204.31±13.95[a]
+Ve Control	1136.51±13.53[b]	1159.47±31.3[b]	1139.79±6.09[b]	1195.56±66.53[b]
Test group 1	1287.78±14.86[c]	1281.95±17.84[c]	1282.31±12.73[c]	1205.77±22.16[c]
Test group 2	1287.41±4.72[d]	1286.69±11.87[d]	1284.50±16.23[d]	1194.10±8.18[d]

Values are presented as mean±SD (n = 6). means with the same superscript letters on the same column are not statistically different at 95% confidence level (p≤0.05)

Effect of celery apium graveolens on lactate dehydrogenase (LDH) concentration of paracetamol-induced wistar albino rats reveals significant decrease of lactate dehydrogenase concentration as time progresses from 30secs, 1min, 2mins and 3mins. Biochemical analysis revealed that the administration of *Apium graveolens* and induction of acetaminophen resulted in significant decrease (p≤0.05) in Lactate dehydrogenase (LDH) concentration in the positive control at various time intervals, thus, 30secs, 1min, 2mins, 3mins when compared with the normal control groups but the concentration of Lactate dehydrogenase (LDH) of Test grooup 1 and Test group 2 increased respectively at the different time intervals when compared to the positive control.

Table 4.3 Effect of *Celery Apium graveolens* on ALT, ALP and AST of paracetamol-induced wistar albino rats.

	ALT(U/L)	ALP(U/L)	AST(U/L)
Control	715.00±18.4[a]	30.08±1.99[a]	63.5 ± 2.6[a]
+Ve Control	1510.67±11.36[b]	29.67±1.00[a]	75.4 ± 3.2[d]
Test group 1	498.67±17.96[c]	30.89±1.26[a]	72.6 ± 2.4[c]
Test group 2	553.67±54.63[d]	30.89±1.99[a]	71.3 ± 2.3[b]

Values are presented as mean±SD (n = 6). means with the same superscript letters on the same column are not statistically different at 95% confidence level (p≤0.05)

The effect of celery apium graveolens on ALT, AST and ALP of paracetamol-induced wistar albino rats is presented in table 3.3. The results revealed that there was a significant (p<0.05) increase in ALT levels of the positive control (1510.67±11.36) in comparison to the ALT levels of the normal control (715.00±18.4) also from the table above the level of ALT of Test group 1 and Test group 2 decreased significantly (p<0.05) (498.67±17.96 and 553.67±54.63) when compared to the level of ALT of the positive control (1510.67±11.3). ALP and AST levels was observed to have had a non-significant increase (p<0.05).

Histology of the liver of wister albino rat, fed with fatty diet and induced with Acetaminophen.

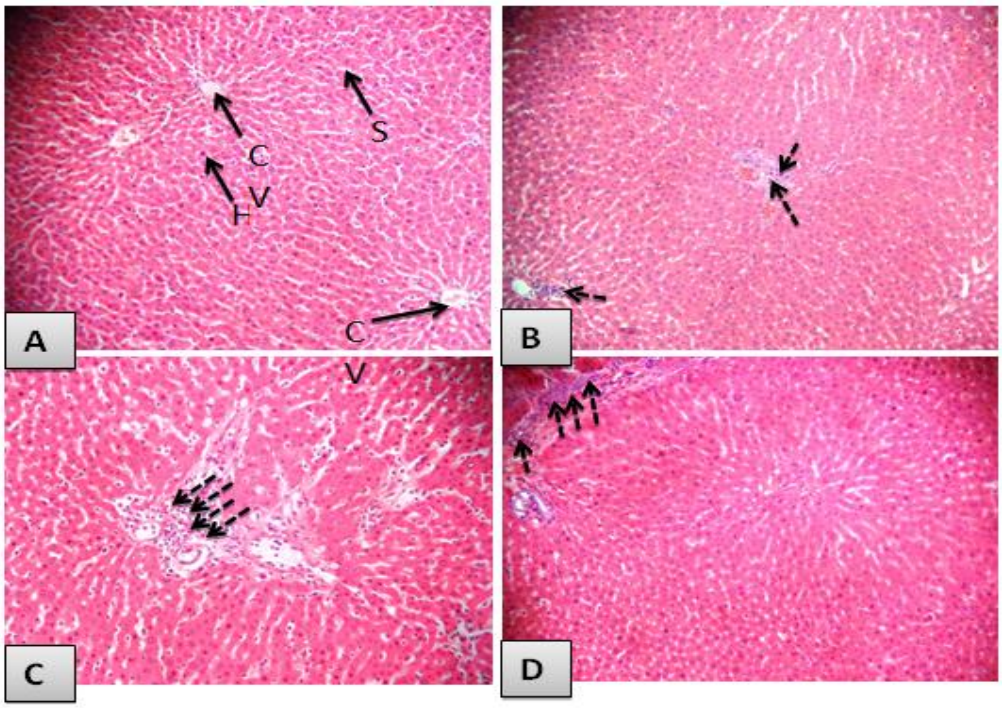

Figure 1: (Photomicrograph of Liver): Group A (Control) shows normal Central vein (CV), with Hepatocytes (H) and sinusoids (S). Groups B- D are characterized by various degrees of inflammatory cells (Dash arrow), which is indicative of healing processes. H&E x100

Results of the effect celery apium on paracetamol-induced wistar albino rats fed with high fat diet is presented in Table 4.3. Presented in Table 3.3is the concentrations of the lipids and sugar level (cholesterol, HDL, triglyceride and glucose) in the control, positive control and test groups. Mean while the weight of the rats before inducing and on day 3 of the research work is presented in Table 4.1 and 4.2 respectively.

Body weight in grams of rats

Table 4.1: Mean body weight of Wistar albino rats on Day 3 and Before Induction.

Groups	Weight(g) On Day 3	Weight(g) Before Inducing
Control	162.33±14.81[a]	135.38±16.05[a]
+Ve control	163.83±16.62[a]	116.78±20.85[a]
Test group 1	145.33±12.31[c]	123.1±7.52[c]
Test group 2	190.83±20.12[d]	160.47±17.43[d]

From Table 4.1, it can be observed that the weight of the rats before inducing ranged from 145-190g, whereas the weight of the rats on Day 3 of acclimatization was observed to have ranged from 116-160g.

Sugar Level and Lipid profile of rats

Table 4.2.Effect of *Celery apium graveolens*on the lipid profile and sugar level of paracetamol-induced Wistar albino rats.

	CHOLESTEROL (mg/dl)	GLUCOSE (mg/dl)	TRIGLYCERIDES (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Control	206.01±2.41[a]	33.27±1.04[a]	192.72±1.20[a]	207.70±1.60[a]	76.95±0.81[a]
+Ve Control	301.83±4.82[b]	39.16±2.08[b]	227.99±10.43[b]	203.29±3.28[b]	45.18±0.77[b]
Test group 1	242.75±4.54[c]	27.18±0.67[c]	188.62±1.47[c]	208.33±2.14[c]	35.09±0.58[c]
Test group 2	201.51±5.56[d]	19.03±1.04[d]	177.02±2.27[d]	212.99±2.46[d]	20.42±0.79[d]

Values are presented as mean ± SDof .means with the same superscript letters on the same column are not statistically different at 95% confidence level (p≤0.05)

The effect of *celery apium graveolens* on the lipid profile and sugar level compared to positive control (+VE) reveals significant decrease of all parameters as shown in table 3.2. Biochemical analysis revealed that induction of acetaminophen resulted in significant increase ($p<0.05$) in cholesterol and glucose in positive control (301.83±4.82 and 19.03±1.04) respectively when compared with the normal control groups (206.01±2.41

and 39.16±2.08). However, there was a significant ($p<0.05$) decrease in the level of triglycerides and HDL in positive control (227.99±10.43 and 203.29±3.28). Levels of Cholesterol decreased significantly ($p<0.05$) in test groups 1 and 2 (242.75±4.54 and 201.51±5.56) respectively compared to positive control (301.83±4.82). Glucose levels decreased significantly ($p<0.05$) in extract treated group 1 and 2 (27.18±0.67 and 19.03±1.04) respectively compared to positive control (39.16±2.08). Triglyceride levels decreased significantly ($p<0.05$) in test groups 1 and 2 (188.62±1.47 and 177.02±2.27) respectively compared to positive control (227.99±10.43). HDL levels increased slightly in test groups 1 and 2 (208.33±2.14 and 212.99±2.46) compared to positive control (203.29±3.28). It was also observed that LDL levels decreased significantly ($P<0.05$) in test groups 1 and 2 (76.95±0.81 and 20.42±0.79).

Effect of Celery *Apium Graveolens* Extract on the Histological Features of the Kidney of Experimental Rats

Histology of the kidney of Wistar albino rat induced with acetaminophen and fed with fatty diet.

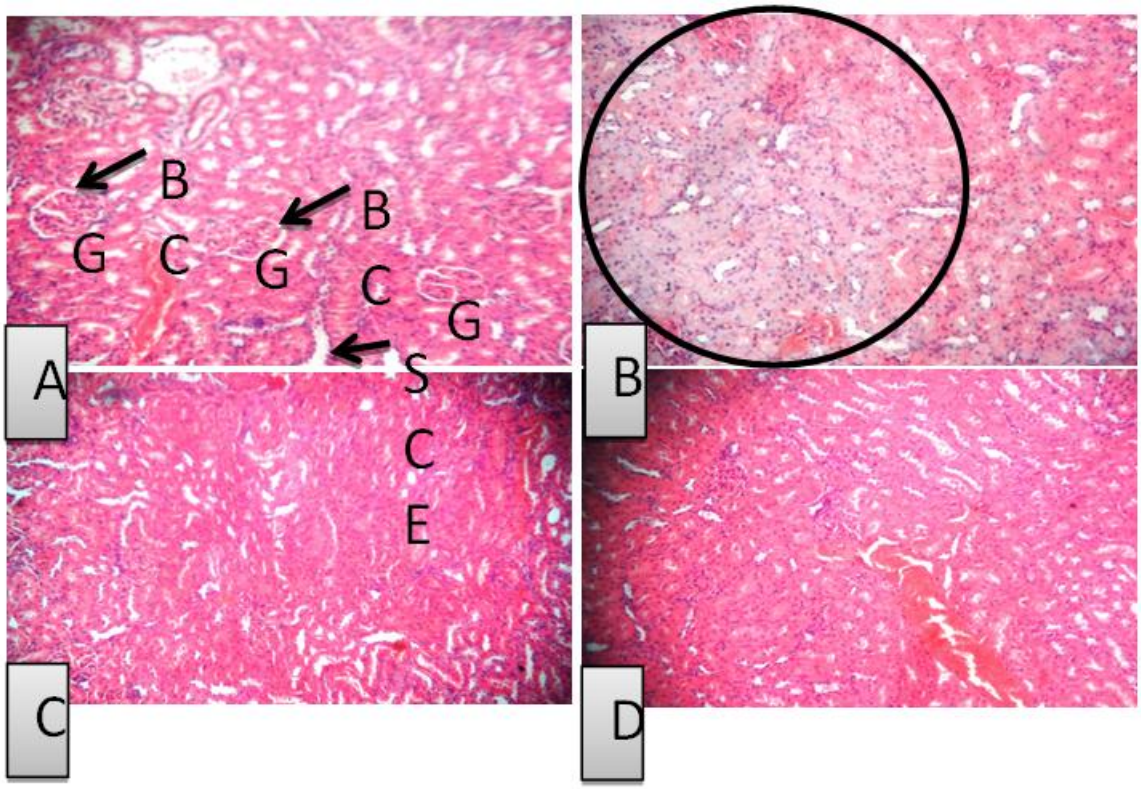

Figure 2: Photomicrograph of Kidney: Group A (Normal control) Shows normal Glomerulus (G), Bowman's capsule (BC) and Simple cuboida Epithelial cells (SCE). Group B shows necrosis of the renal cortex (Circle). However, groups C and D though similar to the control have fewer glomeruli. H&E x100

DISCUSSION

Herbal preparation has been used in many parts of the world since ancient times. In recent years, their popular alternative to modern medicine has increased considerably even in developing countries (Maurya et al., 2004).

In the present study, administration of paracetamol caused liver damage; thus; doses of the extract were chosen to ameliorate the effect of acetaminophen-induced hepatic injury in wistar albino rats. Both biochemical and histological results show that *celery apium graveolens* extract effectively attenuated paracetamol induced hepatic liver injury and the body weight, liver condition, as well as serum enzyme levels (AST and ALP) with the exception of ALT. Acetaminophen which proves highly useful as an experimental liver injury model has been used for years because the lesions caused by this hepatotoxic drug replicate these seen in most cases of liver disease in human, thus causes oxidative stress, lipid peroxidation as well as causing a decrease in antioxidant status (Mehendale, 2010).

Results obtained from this study indicates that there was a significant decrease in AST and ALT levels while a non-significant ($p<0.05$) decrease in ALP levels was observed. The present study revealed that the celery (*Apium graveolens*) extract has no harmful effect on liver function in wistar albino rats rather it ameliorated the hepatic injury induced by paracetamol use in that it reduced the level of AST, ALP and ALT after paracetamol increased their.

Our findings are in agreement with those obtained by (Al-Sa'aidi *et al.,* 2012), who observed decreased in AST, ALT & ALP enzymes and increased LDH content in the liver of rats treated with ethanolic extract of celery (*Apium graveolens*) when compared to positive control. The decrease in AST, ALT & ALP enzymes might due to the biochemical compounds of celery seed as (D-carvon, D-limonen & myrcen), which have biological effects on aminotransferase enzymes (Singh and Handa, 1995), Ahmed *et al.,* 2002 and Taher *et al.,* 2007).

The ability of celery apium graveolens to ameliorate the paracetamol-induced hepatic damage may be due to the presence of phytochemicals such as flavonoids, alkaloids, Medicinal plants are of great importance to the human health of individuals and communities and their medicinal values lie in some chemical substances that produce definite physiological actions on the human body (Edeoga*et al.,* 2005). Plants synthesize hundreds of chemical compounds for functions including defense against insects, fungi, diseases and herbivorous mammals in the example of salicyclic acid, as a hormone in plant defense. Numerous phytochemical with potential and established biological activity have been identified. However, since a single plant contains widely diverse phytochemical, the effect of using a whole plant as medicine is uncertain. Further, the phytochemical content and pharmacological actions of many plants having medicinal potential remain unassessed by rigorous scientific research todefine efficacy and safety (Ahn, 2017). Phytochemicals found in plants are of many kinds but most are in four major biochemical classes: alkaloids, glycosides, polyphenols and terpenes. Phytochemical analysis of *Apium graveolens* indicates the presence of carbohydrates, flavonoids, alkaloids, steroids, and glycosides in the methanolic extract (Khare, 2008).

In this study, the effect of *Celery apium* on the lipid profile and sugar level of paracetamol-induced Wistar albino rats was evaluated. Preliminary analysis of the plasma glucose, cholesterol, HDL and triacyglyceride of the paracetamol induced albino rats showed plasma glucose concentration ranging from 19.02-39.16mg/dl, cholesterol ranged from 201.51-301.83mg/dl, HDL ranged from 203.29-212.99mg/dl and triacylglyceride concentration ranged from 177.02-227.99mg/dl. Also results revealed a significant ($P<0.05$) decrease in LDL levels ranging from 76.95-20.42mg/dl) the effect of celery (*apium graveolens*) was observed to have been concentration dependent.

Continual administration of the extract brought about a significant decrease in the mean plasma glucose, cholesterol, HDL and triglyceride concentration. The decrease was as a result of the nutritional constituents of the extract administered. Results obtained from this study is in line with the study of Al Sa'aidi *et al.,* (2012) who reported decreased glucose, triglyceride and cholesterol concentration in diabetic Wistar albino rats treated with celery *apium graveolens*. Another study, conducted in 2014 by Li et al., (2014) investigated the in vivo and in vitro effect of flavonoids isolated from celery on cholesterol and triglyceride concentration, they reported significant decreases in cholesterol and triglyceride concentrations.

Thus indicating that the ability of the extract to decrease the lipid and sugar profile of the rats in this study suggests its hypolipidemic and hypoglycemic effect.

CONCLUSION

Results obtained in cause of this study revealed *celery apium graveolens* extract effectively attenuated acetaminophen induced hepatic liver injury and the body weight, liver condition, as well as serum levels were improved. Hence, this study has shown that the antioxidant action of *celery apium graveolens* are possible mechanisms to improve liver injury.

In conclusion, continual intake of paracetamol, has a deleterious effect on the body and it induces damage and causes histopathological changes in kidney. Celery *apium graveolens*, was able to ameliorate the harmful effects of paracetamol on the kidney, lipid profile and sugar level in a concentration dependent manner.

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