Ethanolic Extract of *Musa sapientum* (Banana) Spotted Peel Treatment Better Ameliorates CCl_4-Induced Nephrotoxicity than Green or Yellow Peels in Male Wistar Rats

Akpan AE[1], Ekpo AJ[1], *Edagha IA[2]

ABSTRACT

Turning waste to value is trending issue of global importance, and Musa sapientum (Banana) peels are mostly discarded as waste, however spotted peels have been reported to possess rich antioxidants; this study investigated ethanolic extracts of Musa sapientum green, yellow and spotted peels treatment on the kidney histology and renal function parameters following carbontetrachloride (CCl_4) exposure in male Wistar rats. 50 rats (180 - 220g) divided into 10 groups (I - X) of 5 rats per group: Groups I (normal control), Groups (II, IV, V, VI, VII, VIII, IX and X) was injected with CCl_4 intraperitoneally. III (vehicle) received 10% of tween 80 and olive oil. IV and V received 500 and 1000 mg/kg body weight yellow peels extract, VI and VII received 500 and 1000 mg/kg body weight green peels extract, VIII and IX were administered 500 and 1000 mg/kg body weight spotted peels extract, while X received vitamin C 300 mg/kg body weight. Administration was for 21 days. Results showed significant ($p<0.05$) increase in creatinine and urea concentrations in CCl_4-exposed group compared with control and treated groups, also there were significant ($p<0.05$) increase in K^+ concentration in CCl_4-induced group and in groups that received the different peels ethanolic extract at 1000 mg/kg when compared with control. Histology revealed severe distortions in renal tubes, glomerulus and occlusion of urinary space in CCl_4-exposed group compared to control and treated groups. However there were marked nephro-protection in groups IX and X. In conclusion, ethanolic extract spotted peels of Musa sapientum through its antioxidants activity better ameliorates against CCl_4-induced nephrotoxicity in a dose-dependent concentration than the green and yellow peels.

Keywords: CCl_4, Nephrotoxicity, Musa sapientum peels, Renal function, Wistar rats

INTRODUCTION

Carbontetrachloride (CCl_4) is an environmental pollutant that shows toxicity in different organs[1]. CCl_4 is bio-transformed *in vivo* through reductive dehalogenation by the hepatic microsomal P_{450} isoenzymes in the endoplasmic reticulum of hepatic cells resulting in the unstable free radicals of trichloromethyl (CCl_3*) which binds with cellular molecules (nucleic acid, protein, and lipids) impairing crucial cellular processes like lipid metabolism with potential outcome of fatty degeneration, while the reaction between CCl_3* and DNA is thought to function as initiator of hepatic cancer. This radical can react with oxygen to form trichloromethylperoxy radical (CCl_3OO*), which initiates chain reaction of lipid peroxidation by attacking and destroying poly-unsaturated fatty acid[2,3]. However, CCl_4 is distributed at a higher concentration in the kidney than in the liver[4]. The mechanism of CCl_4 renaltoxicity is almost the same as that of the liver, but CCl_4 shows a higher affinity for the kidney cortex which contains predominantly cytochrome P_{450}. Antioxidants such as vitamin C decrease injurious activity of reactive oxygen species (ROS) and free radicals[5].

Peels of various fruits and vegetables are generally considered as waste and are normally discarded, but different studies conducted on peels reveal the presence of important biochemical constituents having activities like antioxidants, antimicrobial, anti-inflammatory which may be useful for pharmaceutical purpose[6]. Zhang and colleagues[7] reported that potential applications of *Musa sapientum* peels depend on its chemical composition, and *Musa sapientum* peels are a good source of polyphenols and carotenoids - phytochemical with antioxidant properties[8]. Gonzalez-Montelongo and co-workers[9] also reported on the antioxidant activities of *Musa sapientum* peels as well as its nutritional composition. Being a climateric fruit the study sought to investigate whether the different ripening stages of banana peels influenced their antioxidant properties, and thus

Department of Biochemistry[1], Faculty of Basic Medical Sciences, University of Uyo, Nigeria
Department of Anatomy[2], Faculty of Basic Medical Sciences, University of Uyo, Nigeria

*Corresponding author: innocentedagha@uniuyo.edu.ng

the experiment assessed banana peels at its physiological matured stage (green peels) as well as at its various stages of ripening (yellow and spotted peels) on carbon tetrachloride induced nephrotoxicity in mature male albino Wistar rats.

MATERIALS AND METHODS

Sample Collection/ Plant Identification and Authentication

Musa sapientum was collected locally from farms within Abak in Akwa Ibom State, Nigeria. Fresh peels of the banana were identified and authenticated as *Musa sapientum* by the Herbarium section of the Department of Botany, University of Uyo with voucher no. Akpan, UUH3158.

Carbon tetrachloride and Vitamin C

Carbon tetrachloride (Poole, Dorset, UK) was used in this experiment, while pharmaceutical grade vitamin C tablets were obtained from Emzor Pharmaceuticals, Lagos, Nigeria. All the kits used for the determination of biochemical parameters were from Randox, UK, and all chemicals and reagents were of analytical grade.

Preparation of Peel Extracts

Musa sapientum were washed under the tap water and their peels were separated from the pulp, cut into small pieces and later sun dried for two weeks. To obtain the yellow peels, the unripe *Musa sapientum* was allowed to undergo the natural ripening process, when properly ripened, the peels were removed and cut into pieces and sun dried. For the spotted peels, the ripened banana was allowed to over ripen thereby developing spots on their peels. The peels were removed and cut into pieces for drying. The dried peels were pulverized using a milling machine and the powdery samples of green, yellow and spotted banana peels which weighed 1860 g, 1866 g and 1497 g respectively were packed into screwed bottles and labeled appropriately. The ethanolic extract of green peel was prepared by soaking the dried powdery sample in 10 litres of 90% ethanol for 48 hours, while ethanolic extracts of yellow and spotted peels were prepared by soaking each of the dried samples in 7.5 litres of 90% ethanol, during which the mixture was stirred intermittently. The extracts were first filtered with cheese cloth then with a Whatman filter paper No. 1 (125mm). The yield of crude extract of green, yellow, and spotted peels weighing 173g, 215g, and 182g respectively were obtained by evaporating the ethanol in a water bath at 40^{o}C and thereafter stored in a refrigerator at about 4^{o}C.

Experimental Animals

50 male albino Wistar rats weighing between 180 - 220g were purchased from the Animal House, Faculty of Biological Sciences, University of Nigeria Nsukka, Enugu State. The rats were kept in standard cages and were fed with rat pellets, and water *ad libitum,* and allowed to acclimatize for 2 weeks in a 12 hour light and 12 hour dark cycle. Animals were randomly selected on the basis of body weight into 10 groups of 5 rats per group, and extract treatment groups were administered 500 and 1000 mg/kg body weight respectively as shown in Table 1 (experimental design), 24 hours after the last administration[10], the experiment was terminated with rats humanely sacrificed via chloroform inhalation. This study conformed to the guide for the care and use of laboratory animals[11].

Table 1: Experimental Design

Groups	Treatment Dosage	Duration
Group I	Control - normal pellets	21 days
Group II	CCl_4 - 10% of tween 80 (5 ml/kg b. wt.) orally and CCl_4(1ml/kg body weight as 0.5 ml CCl_4 + 0.5 ml olive oil)	21 days and 21st day only
Group III	Vehicle -10% of tween 80 (5 ml/kg b. wt.) orally and olive oil (0.5ml/kg body weight). .	21 days and 21st day only
Group IV	CCl_4 + YPE 500mg/kg/day	21 days
Group V	CCl_4 + YPE 1000mg/kg/day	21 days
Group VI	CCl_4 + GPE 500mg/kg/day	21 days
Group VII	CCl_4 + GPE 1000mg/kg/day	21 days
Group VIII	CCl_4 + SPE 500mg/kg/day	21 days
Group IX	CCl_4 + SPE 1000mg/kg/day	21 days
Group X	CCl + vitamin C 300mg/kg/day	21 days

Collection of Blood Sample

24 hours after the last administration, the rats were sacrificed under chloroform anesthesia; incision was made on the abdomen. Blood sample was collected through cardiac puncture using sterile needles and syringes into a labeled sample bottle. Serum was separated by centrifugation at 3000 rpm for 15 mins, and used for the assessment of kidney function tests. The kidney organs were dissected and preserved in 10% buffered formalin for histological studies.

Biochemical Analysis

Determination of Electrolytes

Serum Chloride Ion (Cl^-)

Colorimetric method was used to assay for serum chloride and the absorbance is measured at 480 nM. The intensity of the colour is directly proportional to the chloride concentration in the serum[12].

Serum Potassium Ion (K^+)

Serum potassium ion concentration was determined using colorimetric method. The amount of potassium is determined by using sodium tetraphenylboron in a specifically prepared mixture to produce a colloidal suspension in which the turbidity is proportional to the concentration of potassium in the sample[13].

Serum Sodium Ion (Na^+)

Serum sodium ion concentration was assayed using colorimetric method based on modifications[14,15].

Determination of Serum Urea and Creatinine Levels

Urea and creatinine concentrations were assayed and used to evaluate the functionality of the kidney alongside with electrolytes concentration.

Serum Urea

Urea was determined using kinetic method of urea determination, a modification of the method described by[16]. Urea is catalytically converted to ammonium carbonate by the use of urease. The reaction rate is dependent upon the concentration of glutamic dehydrogenase which catalyses the reaction shown below. The absorbance of NAD^+ is measured at 340 nM and is proportional to the concentration of urease.

Serum Creatinine

Creatinine reacts with picric acid in alkaline conditions to form a coloured complex which absorbs at 510 nM[17]. The rate of the formation of the colour is proportional to the creatinine concentration in the sample.

Histological Assessment of the Kidney: Haematoxylin and Eosin[18]

Kidneys dissected from the male albino Wistar rats were placed in 10% buffered formalin (fixation), and then transferred to a graded series of ethanol (dehydration). On day 1, they were placed in 70% alcohol for 7 hours, then transferred to 90% alcohol and left in the latter overnight. On day 2, the tissues were passed through three changes of absolute alcohol for an hour each then cleared in xylene (clearing). Once cleared, the tissues were infiltrated in molten paraffin wax in the oven at 58°C. 3 changes of molten paraffin wax (impregnation) at one-hour intervals were made, after which the tissues were embedded (embedding) in wax and blocked out. Prior to embedding, it was ensured that the mounted sections to be cut by the rotary microtome were orientated perpendicularly to the long axis of the tissues. Sections were designated "vertical sections". Serial sections of 5μm thick were obtained from a solid block of tissue (microtomy), fixed on clean albuminized slides to prevent sections coming off the slides and later routinely stained with haematoxylin and eosin staining techniques, after which they were passed through ascending grade of alcohol, cleared in xylene and the mountant DPX place on the section and cover-slipped, then allowed to dry at room temperature and observed for histopathological changes under the light microscope (Japan, Olympus - CX31).

Statistical Analysis

The data obtained were expressed as mean ± SEM, and the one way analysis of variance (ANOVA) was used for comparison. The results were subjected to post-hoc test using least significant difference (LSD) multiple comparison. Test values of $p < 0.05$ were considered significant.

RESULTS

Administration of CCl_4 produced significant toxic effects on the renal function, as shown by the data obtained from the parameters investigated (Tables 2-3).

Serum Creatinine

Table 2 shows the effect *of M. sapientum peels ethanolic extract* on CCl_4-induced toxicity in serum creatinine and urea. The creatinine values were significantly ($p<0.05$) increased in CCl_4-induced group compared with the control. The groups administered with the different *Musa sapientum* peels extract dose-dependently had significantly ($p<0.05$) reduced serum creatinine compared to the CCl_4 treated group (group II), with group IV and V showing more significant ($p<0.05$) reduction than groups VI and VII. Similarly, groups VIII and IX showed more significant ($p<0.05$) than groups IV and V as well as groups VI and VII. And also, group X showed significant ($p<0.05$) reduction compared to CCl_4-induced group (group II), though group IX showed more significant ($p<0.05$) reduction compared to group X.

Serum Urea

Administration of CCl_4 to rats significantly ($p<0.05$) increased the level of urea in comparison with the control group. However, the groups administered with the different *Musa sapientum* peels extract dose-dependently and significantly ($p<0.05$) reduced serum urea levels compared to CCl_4 group (group II). The group administered with vitamin C also showed a significant ($p<0.05$) reduction compared to CCl_4 group (group II). Also, groups IV and V showed more significant ($p<0.05$) reduction than groups VI and VII, similarly, groups VIII and IX showed more significant ($p<0.05$) reduction than groups IV and V as well as groups VI and VII. Also groups V, VII and IX showed more significant ($p<0.05$) reduction compared to group X.

Electrolytes

Table 3 shows the effect *of M. sapientum peels ethanolic extract* on CCl_4-induced nephrotoxicity on serum electrolytes.

Sodium (Na^+): There was no significant ($p<0.05$) changes in the serum Na^+ concentrations of CCl_4-induced untreated rats compared with control. However, there was a significant ($p<0.05$) decrease in the group administered with spotted *Musa sapientum* peel extract at 1000 mg/kg (group IX) when compared with CCl_4 group (group II).

Potassium (K^+): There was a significant ($p<0.05$) increased in the serum K^+ concentration of CCl_4-induced untreated rats compared with control. Also, the different banana peels administered at a dose of 500mg/kg body weight significantly ($p<0.05$) reduced serum K^+ when compared with the CCl_4 group (group II). In the same vein, group X (vitamin C) showed more significant ($p<0.05$) reduction in serum K^+ compared with the extract groups.

Chloride (Cl^-): There was a significant ($p<0.05$) increas in the serum Cl^-concentration of CCl_4-induced untreated rats when compared with control. The different banana peels extract though not dose dependently, except groups VIII and IX significantly ($p<0.05$) reduced chloride ion when compared with CCl_4 group, (group II). Vitamin C also showed a significant reduction when compared with CCl_4-induced group (group II).

Table 2: Effect *of M. sapientum Peels Ethanolic Extract* on CCl_4-Induced Nephrotoxicity on Serum Creatinine and Urea

Group	Creatinine (µmol/L)	Urea (mmol/L)
I (control)	67.60±1.12	5.78±0.08
II (CCl_4 only)	143.00±1.22[a]	7.22±0.05[a]
III (OL/TW_{80})	75.40±1.63[a,b]	5.44±0.15[b]
IV (CCl_4 + YPE $_{500mg}$)	70.00±1.30[b]	5.64±0.08[b]
V (CCl_4+ YPE $_{1000mg}$)	59.00±1.22[a,b,c]	4.82±0.12[a,b,c]
VI (CCl_4 + GPE $_{500mg}$)	78.20±3.02[a,b,c]	6.66±0.13[a,b,c]
VII (CCl_4 + GPE $_{1000mg}$)	66.00±1.22[b,d,e]	5.54±0.16[b,d,e]
VIII (CCl_4 + SPE $_{500mg}$)	65.00±1.70[b,e]	5.10±0.14[a,b,c,e]
IX (CCl_4 + SPE $_{1000mg}$)	40.40±1.50[a,b,d,f,g]	4.30±0.18[a,b,d,f,g]
X (CCl_4 + Vit C $_{300mg}$)	56.40±2.73[a,b,c,e,f,g,h]	5.96±0.24[b,d,e,f,g,h]

Table 3: Effect *of M. sapientum Peels Ethanolic Extract* on CCl_4 Induced Nephrotoxicity on Serum Electrolytes

Group	Na^+(mmol/L)	K^+(mmol/L)	Cl^-(mmol/L)
I (control)	143.30±0.33	4.66±0.12	99.70±0.60
II (CCl_4 only)	143.04±0.77	7.10±0.18[a]	101.70±0.82[a]
III (OL/TW_{80})	141.78±0.52[a]	4.70±0.05[b]	99.16±0.62[a,b]
IV (CCl_4 + YPE_{500mg})	142.62±0.64	6.28±0.08[a,b]	98.08±0.27[b]
V (CCl_4+ YPE_{1000mg})	141.68±0.54[a]	8.82±0.13[a,c]	99.50±0.42[b]
VI (CCl_4 + GPE_{500mg})	144.38±0.38[c]	6.23±0.06[a,b]	99.40±0.37[b]
VII (CCl_4 + GPE_{1000mg})	143.52±0.36[d]	8.85±0.15[a,e]	99.78±0.36[b]
VIII (CCl_4 + SPE_{500mg})	142.52±0.32[e]	6.20±0.07[a,b]	100.84±0.57[b]
IX (CCl_4 + SPE_{1000mg})	139.70±0.31[a,b,e,f,g]	8.83±0.14[a,g]	98.54±0.50[b,g]
X (CCl_4 + Vit C_{300mg})	141.74±0.53[a,e,f,h]	5.23±0.05[a,b,c,d,e,f,g,h]	99.40±0.45[b]

Legend: [a]= significantly different from group I ($p < 0.05$), [b]= significantly different from group II ($p < 0.05$), [c]= significantly different from group IV ($p < 0.05$), [d]= significantly different from group V ($p < 0.05$), [e]= significantly different from group VI ($p < 0.05$), [f]= significantly different from group VII ($p < 0.05$), [g] = significantly different from group VIII ($p < 0.05$), [h] = significantly different from group IX ($p < 0.05$).

CCl_4: carbon tetrachloride, OL: olive oil, TW_{80}: tween 80, YPE: yellow peel extract, GPE: green peel extract, SPE: spotted peel extract, Vit C: vitamin C.

Histological Assessment of the Kidney

Photomicrographs showing the renal histoarchitecure of all the treatment groups are shown in Figure 1 (plates 1 to 10).

Group I: Photomicrograph of control kidney without treatment revealed the normal renal histoarchitecture evidenced with average sized glomeruli (G), Bowman's space (BS), and proximal convoluted (PC) tubule H&E x100 (Inference: appears normal).

Group II: Photomicrograph of rats kidney induced with carbontetrachloride showed

derangement of glomeruli (GM) and congested vessels with hyperplasia and reduced proximal convoluted tubule (PC) (Inference: severely affected).

Group III: Photomicrograph of treated with tween 80 and olive oil showed numerous glomeruli (GM), and reduced Bowman's space (rBS). Also seen is reduced proximal convoluted tubule (PC) lumens and foci congested blood vessels (CBV) (Inference: mildly affected).

Group IV: Photomicrograph of rats kidney induced with carbontetrachloride and treated yellow peels of *Musa sapientum* ethanolic extract (500 mg) revealed a well vascularized renal cortex and the glomeruli, patent proximal convoluted tubules and a reduced bowman's space (Inference: severely affected).

Group V: Photomicrograph of rats kidney induced with carbon tetrachloride and treated with yellow peels of *Musa sapientum* ethanolic extract (1000 mg) revealed normal architecture showing normal glomeruli (GM), proximal convoluted tubules (PC), renal artery (RA) and Bowman's space. H&E x100 (Inference: mildly affected).

Group VI: Photomicrograph of rats kidney induced with carbon tetrachloride and treated with green peels of *Musa sapientum* ethanolic extract (500 mg) showed well vascularied glomeruli (GM) with capillaries (C), widened Bowman's space (BS), and proximal convoluted tubules (PC). H&E x100 (Inference: severely affected).

Group VII: Photomicrograph of rats' kidney induced with carbontetrachloride and treated with green peels of *Musa sapientum* ethanolic extract (1000 mg/kg b) showed glomeruli (GM) with decreased Bowman's Space. Also seen are the proximal convoluted tubules (PC) and congested blood vessels (CBV). H&E x 100 (Inference: severely affected).

Group VIII: Photomicrograph of rats kidney induced with carbontetrachloride and treated with spotted peels of *Musa sapientum* ethanolic extract (500 mg/kg) showed normal glomeruli (GM), Bowman's space (BS) and proximal convoluted tubules (PC) (Inference: mildly affected).

Group IX: Photomicrograph of rats kidney treated with carbontetrachloride and spotted peels of *Musa sapientum* ethanolic extract (1000 mg/kg) with normal architecture showing normal glomeruli (GM), Bowman's space (Inference: mildly affected).

Group X: Photomicrograph of rats kidney induced with carbontetrachloride and treated with vitamin C (300mg) showed normal renal tissue of with normal glomeruli (GM), Bowman's space (BS) and proximal convoluted tubules (PC) (Inference: appears normal).

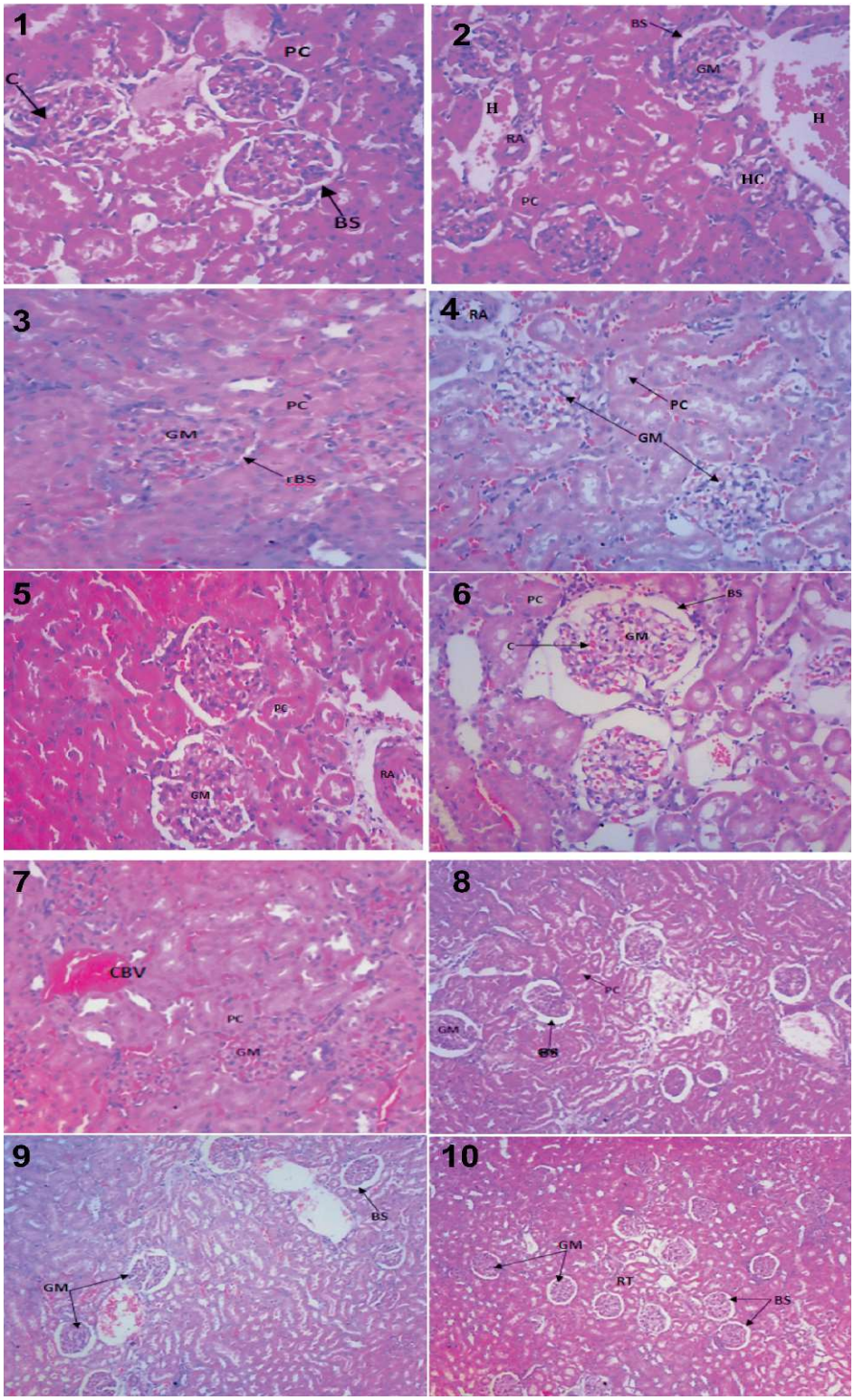

Figure 1: Photomicrograph of rat kidney cross-sections. Plate 1: rats in normal group, Plate 2: rats in CCl4 induced group, Plate 3: rats in tween 80 and Olive oil group, Plate 4: rats treated with yellow *M. sapientum* peels extract at 500 mg/kg, Plate 5: rats treated with yellow *M. sapientum* peels extract at 1000 mg/kg, Plate 6: rats treated with green *M. sapientum* peels extract at 500 mg/kg, Plate 7: rats treated with green *M. sapientum* peels extract at 1000 mg/kg, Plate 8: rats treated with spotted *M. sapientum* peels extract at 500 mg/kg, Plate 9: rats treated with spotted *M. sapientum* peels extract at 1000 mg/kg, Plate 10: rats treated with Vitamin C at 300 mg/kg.

DISCUSSION

The kidney performs the key role of filtering and excreting toxic substances from the body, in addition to helping in the maintenance of osmotic balance. These functions are often impaired in a pathologic kidney[19]. The administration of CCl_4 caused nephrotoxicity as indicated by the significant elevation in serum level of creatinine and urea compared with the normal control group and this result is in agreement with previous findings[20, 21, 22]. From the present study, it is evident that elevation in serum urea and creatinine levels can be attributed to the damage of nephron structural integrity[23]. The ethanolic extract of different peels of *Musa sapientum* at increasing doses, as well as vitamin C significantly lowered urea and creatinine levels compared with the CCl_4-exposed group (group II). This indicates that spotted peels *Musa sapientum* improved renal function in disease rats kidney, as the antioxidant activities of *Musa sapientum* peels as well as its nutritional composition have been reported[9].

All known higher life form require a subtle and complex balance between the intracellular and extracellular environments. In particular, the maintenance of precise osmotic gradients of electrolytes is important. Such gradient regulates hydration of the body, blood pH and are critical for nerve and muscle functions. The concentrations of different electrolytes are kept under tight control via various mechanisms in living organisms[24]. The results from this study showed that there was no significant change in the Na^+ concentration of the CCl_4-exposed untreated group (group II) compared with the control. Significant ($p<0.05$) hyperkalemia was observed in the CCl_4- exposed rats compared with the control, there was also a significant ($p<0.05$) reduction in serum K^+ concentration in the group that received the different peels extract at 500 mg/kg compared with the CCl_4 group (group II). A significant ($p<0.05$) increase in serum K^+ concentration of groups that received the different peels extract of *Musa sapientum* at 1000 mg/kg compared with the CCl_4-induced untreated group (group II) was observed. This could be attributed to the high level of potassium which is known to be present in the *Musa sapientum* peels. Also vitamin C was able to significantly ($p<0.05$) reduce serum K^+ compared to the CCl_4-induced untreated group (group II). A significant ($p<0.05$) increase in the level of serum Cl^- was observed in the CCl_4-induced untreated group compared with the control, though it did not exceed the normal range of serum chloride. Due to hepatorenal injury which accompanies CCl_4 toxicity, the transport function of hepatocytes and nephrotic cells gets disturbed causing plasma membrane leakage, and thereby resulting in an increased enzyme and electrocytes levels in the serum[25].

Diseases of the kidney are as complex as its structure, and four basic morphologic components can be used to assess the endpoints of kidney disease progression namely; glomeruli, tubules, interstitium, and blood vessels. Due to the anatomic and functional interdependence of these renal components, damage to one, almost always affects the others[26]. Histology results (Figure 1; plates I to X are photomicrographs of groups I to X respectively), showed that the histoarchitecture of test group X (CCl_4 + Vit C 300 mg) compared to group II appeared normal, without the features of cellular or membrane distortions. Groups III, V, VIII, and IX showed mild changes when compared with groups II, VI, and VII which presented more severe distortions mainly involving glomerulonephrititis, hypercellularity and inflammatory response characterized by cellular proliferation of mesangial or endothelial cells, leukocytic infiltration of neutrophils, monocytes, and lymphocytes, and formation of crescents the accumulation of cells composed of proliferating parietal epithelial cells and infiltrating leukocytes. Patechial hemorrhages and hemorrhagic pool were also observed in groups II and which will correlate with hematuria. Toxins and drugs can produce renal injury in at least three ways; they may trigger an interstitial immunologic reaction exemplified by the acute hypersensitivity nephritis induced, they may cause acute renal failure, and they may cause subtle but cumulative injury to tubules that takes years to become manifest resulting in chronic renal insufficiency[27]. The histologic changes in the renal morphology can be subdivided into; diffuse - involving all glomeruli, global - involving only a portion of the glomeruli, segmental - affecting a part of each glomerulus and mesangial - affecting predominantly the

mesangial region[26]. However, the mechanisms of kidney drug-induced renal injury are varied and the tubulointestitial compartment is most frequently involved, but in significant cases glomerular and vascular lesions are seen[28] as shown in group II, though with declining distortions in the extract treated groups. In conclusion, bioavailability of rich antioxidants such as phenols and flavonoids compounds in the ethanolic extract of spotted *M. sapientum* peel moderately modulated the renal biochemical and histomorphological alterations caused by CCl_4-induced nephrotoxicity better than green and yellow peels, thus justifying the reported beneficial claims and may be a potential feedstock in animal husbandry and the phenols and flavonoids should be characterized in further studies.

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Conflict of Interest

Authors declare no conflict of interest.

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