



Comparative investigations of the effects of *Kanwa* (Trona) and *Tokansenyi* (plant potash) on liver and kidney of albino rats

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ABSTRACT

Kanwa (Trona) and *Tokansenyi* (Plant potash) are food tenderizers that are commonly used in developing countries especially. Their advantage in improving texture of food and in health cannot be over emphasized. However, long-term consumption of these salts has some health implications. Therefore this study is to compare the effects of these two salts on albino rats so as to determine which one is safer. Albino rats were grouped into three groups, with the first group as control, second group contains water with three grams of *Tokansenyi* and the third group contains water with three grams of *Kanwa*. Blood samples for liver and kidney function test were collected after six weeks. Organs (liver and kidney) were also collected for histological studies. The salts element composition were analysed using Inductively Coupled Plasma – Mass Spectrophotometry (ICP-MS). At end of the period of treatments no mortality was recorded and no any visible sign of toxicity. The results obtained for liver and kidney enzymes concentration AST, ALT and ALP; for both treatments shows significant difference ($P < 0.05$) as compared with the control. The total bilirubin concentration for blood of albino rats treated with *Kanwa* shows significant difference ($P < 0.05$). The concentration of urea in both treatments also shows significant difference ($P < 0.05$) as compared with control. The histological results for liver treated with *kanwa* revealed a sinusoid with bodies stained with deep red colouration, enlarged nuclei and distorted cord-like arrangement of hepatocytes. The histological results for kidney treated with *Kanwa* also revealed a degenerated collecting duct and inflamed glomeruloid. The elemental composition of both salts showed similar concentration for most of the elements including the toxic ones. However, some elements like Mo, Mn and Co showed higher concentration in *Tokansenyi* as compared to *Kanwa*. The study revealed that *Tokansenyi* is safer for the liver and kidney as compared with *Kanwa* (Trona). The study has shown that *Tokansenyi* is relatively safer than *Kanwa*.

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Keywords: *Kanwa* (Trona), *Tokansenyi* (Plant Potash), albino rats, liver, kidney.

INTRODUCTION

Kanwa is the Hausa name for dry lake salt (Helga et al., 2017), also erroneously called potash, even though it has low

concentration of potassium as compared to sodium. It is found mostly in Northern parts of Nigeria, especially, Kano and Borno states and some other neighbouring countries, such as Chad and Niger (Ajiboye et al., 2013;

Imafidon et al., 2016; Okoye et al., 2016). *Tokansenyi* is the Hausa name for potash, a trade name for crude potassium carbonate obtained by leaching the ashes of burnt plants and animal bones with water and evaporating the resulting solution to dryness. With impurities it is brownish or blackish in colour while in pure state it is whitish. Its' chemical configuration is K_2CO_3 with molecular weight of 138.7 KgMol^{-1} . It is translucent (granular) or white odourless deliquescent solid known in the anhydrous and hydrated forms (Shagal, 2011).

Kanwa is widely used in West Africa and particularly in Cameroon, Ghana and Nigeria (Ekosse, 2010). In the gastro intestinal system, *Kanwa* is used as an antacid stomachic for the relief of constipation and flatulence (Ibeme, 2012). *Kanwa* is also a liver stimulant. In the renal system, *Kanwa* induces alkaline diuresis (i.e increased urination) and dilates blood vessels to enhance renal blood flow. In the respiratory system, *Kanwa* induces secretion of respiratory mucosa to act as an expectorant. Generally, *Kanwa* induces diaphoresis by dilating blood vessels of the skin with subsequent cooling effect that may feel helpful in febrile conditions. In traditional concoctions and for culinary purposes *Kanwa* serves as tenderizer, thickener, seasoning, potentiating adjunct and preservative. Ancient Egyptians used *Kanwa* solutions as preservative in mummification (Ibeme, 2012).

Tokansenyion the other hand is believed to be a non-purgative substance in preparing crude palm oil and African salad popularly known as *Abacha* (Lim and Zaharah, 2000). Okoye et al. (2016) found that *Tokansenyi* have high pH and contains varying amounts of other nutrients such as calcium (Ca), phosphorus (P), and magnesium (Mg). Studies has also shown that *Tokansenyi* is an effective fertilizer and liming material for increasing soil fertility, pH, and nutrient uptake by crops such as maize and cassava (Ojeniyi et al., 2009). However, few studies have demonstrated its effect in the body following its use as food tenderizer. This study is aimed at comparing effect of *Tokansenyi* and *Kanwa* in albino rats.

METHODS AND MATERIALS

Samples collection

Kanwa were bought from FarinGada market in Jos north LGA while *Tokansenyi* were bought from Fwangkwak village in Kerang District of Mangu LGA, Plateau State.

Preparation of samples

Both *Kanwa* and *Tokansenyi* were weighed out 3.5 g into 100 ml of pipe borne water. They are always freshly prepared before administering to the albino rats.

Experimental animals

A total of 15 male albino rats weighing between 250-288 g were purchased from the animal house of University of Jos. The albino rats were then subdivided into three groups of five albino rats per group with Group one as control, Group 2 as test 1 and Group 3 as test 2. These were then kept under standard environmental conditions.

Chemicals

All chemicals used were of the analytical grade. Kits used for ALT, AST, ALP, total protein, albumin, total bilirubin, urea and creatinine were obtained from Randox Laboratories (Crumlin, Co Antrim, Spain). Others were products of BDH Laboratories (BDH Chemicals Limited, Poole, England) (Imafidon et al., 2016).

Administration of dissolved salts

The *Kanwa* and *Tokansenyi* were administered to the albino rats orally, for; Group one (control) were fed with growers mash and normal pipe borne water, Group two (test 1) were fed with growers mash and pipe borne water with dissolved *Kanwa* (3.5 g/100 ml) and Group three (test 2) were fed with growers mash and pipe borne water with dissolved *Tokansenyi* (3.5 g/100 ml). The albino rats were fed with these foods and water for six weeks before sacrificing them.

Blood sample collection

Six weeks after treatment with *Kanwa* and *Tokansenyi*, blood samples were collected by direct cardiac puncture and placed into sterile containers with or without anticoagulant. The blood were kept to clot and

then centrifuged at 4000 rpm for 10 minutes. The sera were then used for liver and kidney function test using the method described by Mohammed et al. (2014) with little modification.

Organs sample collection

The albino rats were sacrificed and their kidney and liver were dissected and stored in containers with 10% formalin solution for histological study (Chinedum et al., 2014; Muhammed et al., 2014).

Tissue processing for histological study

The tissues were chemically fixed and transferred to a cassette (a container designed to allow reagents to freely act on the tissue inside). To dehydrate the tissues, the cassettes were then immersed in multiple baths of progressively more concentrated ethanol (10%, 40%, 70% and 90%). Thereafter, they were immersed into toluene and later paraffin. The processed tissues were then taken out of the cassettes and set out in a mould. Additional paraffin was then added to make a 'paraffin block' attached to the outside of the cassettes. The paraffin blocks containing the tissues were then sectioned using a microtome. The paraffin section was then removed by floating on a hot water bath. The tissues were then mounted on glass slides and stained using a combination of hematoxylin and eosin to produce the contrast needed to visualize the tissue with microscope.

Elemental analysis

The target elements in *Kanwa* was determined using PerkinElmer ELAN 9000 Inductively Coupled plasma Mass Spectrophotometry (ICP-MS) while the target elements in *Tokansenyi* was determined using NexION 300 Inductively Coupled plasma Mass Spectrophotometry (ICP-MS) in Acme Laboratory Canada.

Statistical analysis

All data were expressed as mean \pm SD (n = 3). One-way analysis of variance (ANOVA) was used to test for difference among all the groups. A p-value of <0.05 was considered statistically significant.

RESULTS

Liver and Kidney function

The results AST for *Kanwa* treated albino rats were 174.0 ± 5.0 u/l while *Tokansenyi* albino rats treated were 171.0 ± 3.0 u/l as compared with control 36.0 ± 1.0 u/l. The increased concentration of AST in albino rats treated with the salts show about five times what was obtained from control albino rats ($p < 0.05$). ALT results are 77.0 ± 2.0 u/l for albino rats treated with *Kanwa*, 85.0 ± 1.0 u/l treated with *Tokansenyi* and 38.0 ± 1.2 u/l for control. The increased concentration of ALT in albino rats treated with the salts show more than two times the concentration obtained from control albino rats ($p < 0.05$). Likewise, the concentration of ALP in the albino rats treated with the salts shows about two times higher concentration than the control ($p < 0.05$) as shown in Table 1.

The result of total bilirubin for albino rats treated with *Kanwa* shows more than two times lower concentration as compared with control ($p < 0.05$), while for albino rats treated with *Tokansenyi* were found to be lower but not significant ($p > 0.05$). Creatinine, Albumin and Total Protein are not significant ($p > 0.05$) between albino rats treated with *Kanwa* and *Tokansenyi* as compared with control. Urea concentration in albino rats treated with *Kanwa* were found to be about two times that in the control ($p < 0.05$), while for those treated with *Tokansenyi* were found to be more than two times that in control ($p < 0.05$) as shown in Table 2.

The results in Table 3 shows that Na^+ , K^+ , Cl^- and HCO_3^- are not significant ($p > 0.05$) between albino rats treated with *Kanwa* and *Tokansenyi* as compared with control. For Na^+ concentration in albino rats treated with *Kanwa* shows to be slightly lower than that found in albino treated with *Tokansenyi*, even when it has been established that *Kanwa* has more Na^+ than *Tokansenyi*. K^+ was found to slightly higher in albino rats treated with *Tokansenyi* than in those treated with *Kanwa* as shown in Table 3.

Figure 1 shows the slide for Control Liver of albino rat at magnification X400. The down arrows in the slide shows normal nuclei with a normal cord like arrangement of

hepatocytes. While the down arrows shows presence of kupfer cell within the sinusoid.

Figure 2 shows the slide of Liver of albino rat treated with *Tokansenyi* at magnification X400. The down arrow a normal cord like arrangement of hypatocytes whereas, the right arrow normal nuclei within the hepatocyte.

Figure 3 shows the slide of Liver of albino rat treated with *Kanwa* at magnification X400. The right arrow shows that the sinusoids contain within them bodies stained deep red. The up arrow shows that nuclei are larger than normal and distortion of the cord like arrangement of the hepatocytes.

Figure 4 shows slide of control Kidney of albino rat at magnification of X400. The up arrow shows a normal glomerulei while the right arrow shows normal nuclei within the collecting duct.

Figure 5 shows slide of Kidney of albino rat treated with *Tokansenyi* at magnification of X400. The down arrow shows a normal glomerulei while the right

arrow shows normal nuclei within the collecting duct.

Figure 6 shows slide of Kidney of albino rat treated with *Kanwa* at magnification X400. The up arrow shows cells of the collecting duct appear degenerated. Left arrow shows glomeruloid inflammation, whereas, down arrows normal collecting duct with normal opening which are few.

The results in Table 4 shows that K^+ is higher in *Tokansenyi* than in *Kanwa*. And Na^+ is higher in *Kanwa* than in *Tokansenyi*.

In Table 5, Fe concentration in *Kanwa* is higher than in *Tokansenyi*, Mo and Ca are higher in *Tokansenyi* than in *Kanwa*. There was no significant difference ($p > 0.05$) in concentration of Cu and Zn in both salts.

In Table 6, Pb concentration in *Kanwa* is slightly higher than in *Tokansenyi* but not significant ($p > 0.05$). The element Co and Mn concentration in *Tokansenyi* are significant ($p < 0.05$) as compared to their concentration in *Kanwa*. The concentration of Cadmium in both salts is the same.

Table 1: Showing concentration of enzymes of liver and kidney of albino rats.

	AST (SGOT) (u/l)	ALT (SGPT) (u/l)	ALP (u/l)
Control	36.0 ± 1.0	38.0 ± 1.2	150.0 ± 3.0
<i>Kanwa</i>	174.0 ± 5.0 ^a	77.0 ± 2.0 ^a	245.0 ± 5.0 ^a
<i>Tokansenyi</i>	171.0 ± 3.0 ^a	85.0 ± 1.0 ^a	274.0 ± 4.0 ^a

The parameters were taken in triple. Values with superscripts are significant at $p < 0.05$. ± Shows standard deviation.

Table 2: Showing concentration of other parameters for liver and kidney function.

Treatment	Total Bilirubin (µmol/l)	Creatinine (g/l)	Urea (g/l)	Albumin (g/l)	Total Protein (g/l)
Control	0.10±0.01	32.0±1.0	23.0±1.0	4.0±0.5	6.9 ± 0.1
<i>Kanwa</i>	0.04±0.01	34.0±2.0	45.0±2.0 ^a	4.3±0.3	7.5 ± 0.5
<i>Tokansenyi</i>	0.08±0.01	38.0±2.0	48.0±4.0 ^a	4.6±0.4	7.8 ± 0.5

The parameters were taken in triplet. Values with superscripts are significant at $p < 0.05$. ± Shows standard deviation

Table 3: Showing concentration of electrolytes.

Treatment	Na ⁺ (mmol/l)	K ⁺ (mmol/l)	Cl ⁻ (mmol/l)	HCO ₃ ⁻ (mmol/l)
Control	145.0±2.0	5.5±0.3	106.0±0.3	31.0±0.7
Kanwa	139.0±1.0	5.9±0.2	100.0±1.0	25.0±0.9
Tokansenyi	142.0±2.0	6.1±0.1	100.0±1.0	23.0±1.0

The parameters were taken in triplet; ± Shows standard deviation

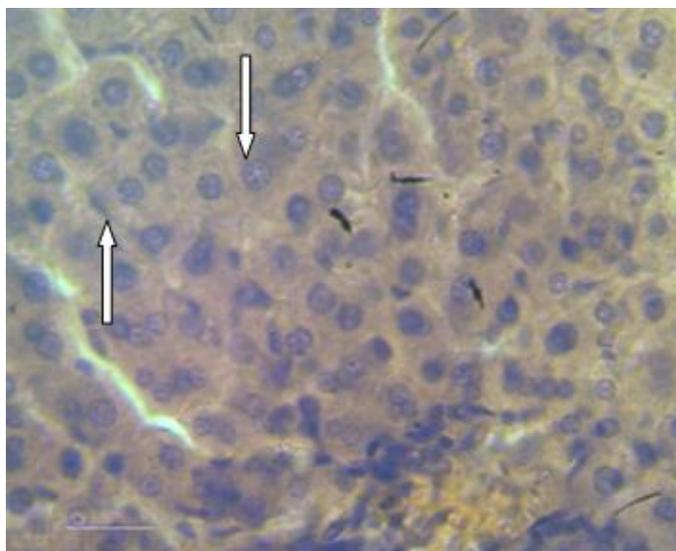


Figure 1: Slide of Control Liver of albino rat x400.

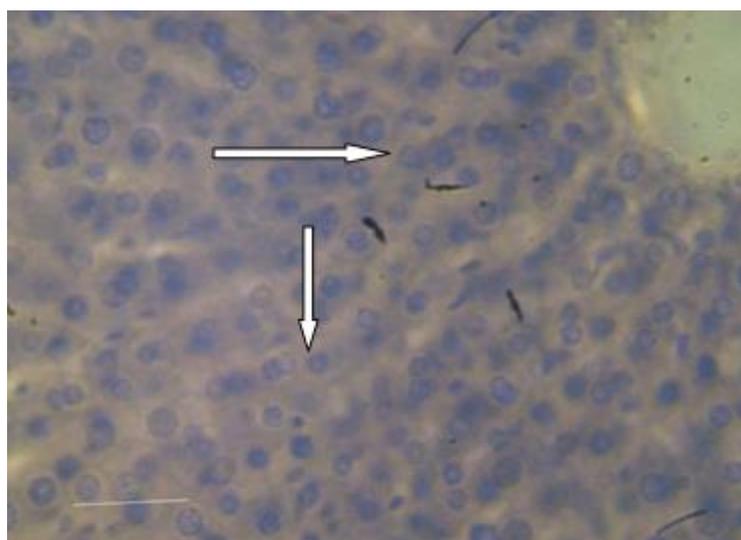


Figure 2: Slide of Liver of albino rats treated with *Tokansenyi* (Plant Potash) x400.

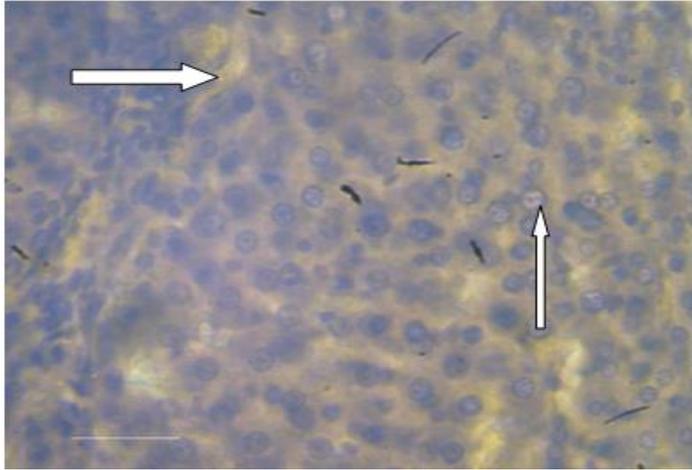


Figure 3: Slides of Liver of albino rats treated with *Kanwa* (Trona) x400.

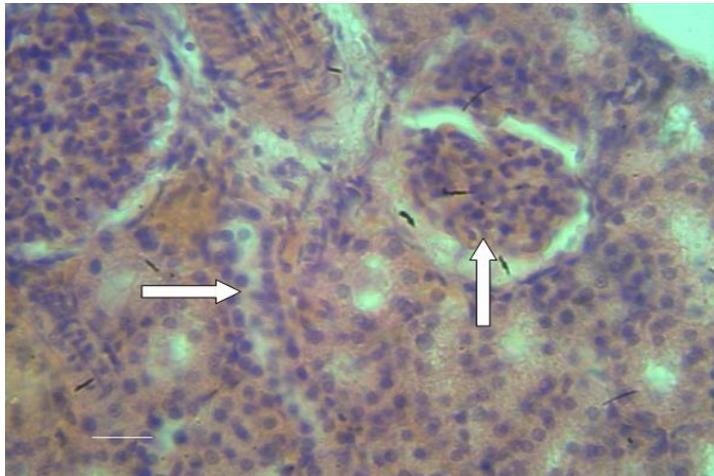


Figure 4: Slide of control Kidney of albino rats x400.

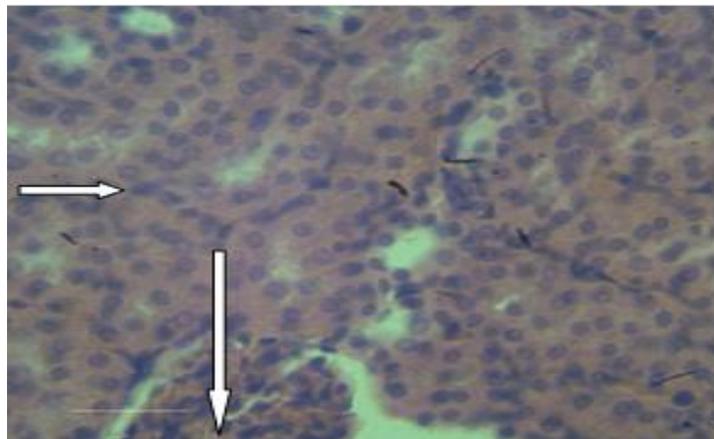


Figure 5: Slide of Kidney of albino rats treated with *Tokansenyi* (Plant Potash) x400.

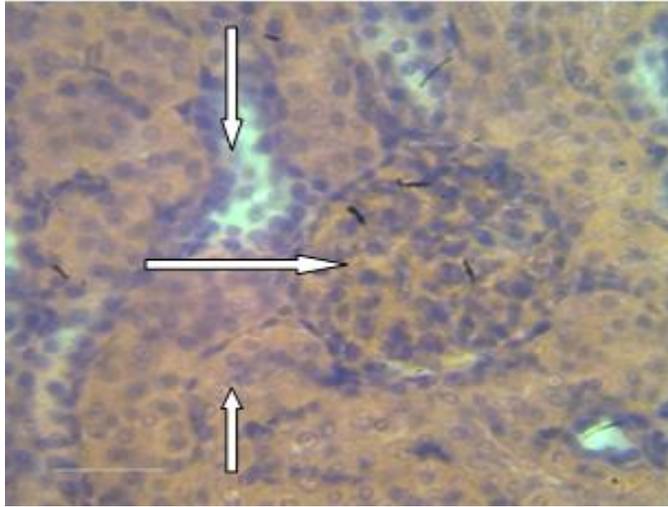


Figure 6: Slides of kidney of albino rats treated with *Kanwa* (Trona) x400.

Table 4: Showing concentration Na and K in *kanwa* and *Tokansenyi*.

Treatment	K (ppm)	Na (ppm)
<i>Kanwa</i>	2.41.0±0.1	>5.0 ^a
<i>Tokansenyi</i>	>10.0 ^a	0.11±0.01

> greater than± Standard deviation ppm parts per million Samples were analysed in triplets;
Values with superscripts are significant at p < 0.05.

Table 5: Showing some essential elemental composition of *kanwa* and *Tokansenyi*.

Treatment	Fe(ppm)	Cu (ppm)	Mo (ppm)	Zn (ppm)	Ca (ppm)
<i>Kanwa</i>	0.10±0.01 ^a	8.09±0.1	3.68±0.3	2.2±0.01	0.04±0.01
<i>TokanSenyi</i>	0.04±0.00	8.07±0.11	9.82±0.5 ^a	3.2±0.02	0.13±0.01 ^a

Samples were analysed in triplets± Standard deviation ppm parts per million Values with superscripts are significant at p < 0.05.

Table 6: Showing some elemental composition of *kanwa* and *Tokansenyi*.

Treatment	Pb (ppm)	Co (ppm)	Mn (ppm)	Cd (ppm)
<i>Kanwa</i>	0.40±0.01	0.40±0.01	15.0±0.5	<0.01
<i>Tokansenyi</i>	0.32±0.01	1.56±0.02 ^a	25.0±0.7 ^a	<0.01

Samples were analysed in triplets ± Standard deviation; ppm: parts per million; < less than;
Values with superscripts are significant at p < 0.05.

DISCUSSION

Kanwa and *Tokansenyi* are food tenderizers that are commonly used in developing countries. The salts have advantages both in improving texture of food and in health (Nasiru et al., 2011; Danbauchi et al., 2014; Okoye et al., 2016). But continuous consumption of these salts has its corresponding health effect. This brings aim of the study, to identify which is safer, *Tokansenyi* or *Kanwa*.

The elemental analysis of the two salts depicted why the two salts cannot give the same result based on composition and concentration of the elements. *Kanwa* has far high concentration of Na⁺ than K⁺ whereas *Tokansenyi* has far high concentration of K⁺ than Na⁺ (Table 4). *Kanwa* contained high concentration of Fe as compared to *Tokansenyi*. While *Tokansenyi* has high concentration of Mo, Ca, Co and Mn as compared to *Kanwa*. Among the toxic element Pb is slightly in *Kanwa* than in *Tokansenyi* (Table 5 & 6). These results agreed with the findings of Imafidon et al. (2016) who obtained similar results in his elemental analysis.

The no mortality and visible sign of toxicity observed during the period of feeding and salts intake in water by the albino rats, could be justified by why the effects are usually gradual on consumers. However, the assessment of albino rats administered with fixed concentration of *Kanwa* and *Tokansenyi* revealed significant increase in kidney and liver enzymes (Table 1). This indicates that at these concentrations, these tenderizers may be toxic especially when administered on long-term basis.

The enzymatic activity of ALT, AST and ALP were studied to investigate Liver and Kidney malfunctions (Table 1). Significant increases in AST, ALT and ALP activities were observed in the albino rats treated with *Kanwa* and *Tokansenyi* as compared with control. This demonstrates that the concentration given, has the possibility of liver and kidney damage on long-term exposure. Similar increases in ALP, ALT and AST activities have also been reported on

administered of *Kanwa* (Ajiboye et al., 2013; Imafidon et al., 2016). AST have also been reported to be elevated in disease condition such as pulmonary infarction, acute infarction and progressive muscular dystrophy (Sood, 2009). The reports of Bankole et al. (2015) and Ebadan et al. (2014) who observed renal toxicity following administration of *Kanwa* further supports the argument.

Section of liver of albino rat treated with *Kanwa* shows the sinusoids contained within their bodies stained deep red and the nuclei of the liver were larger than normal and distortion of the cord-like arrangement of the hepatocytes as compared to control of albino rat liver treated with *Tokansenyi*. The kidney of albino rat treated with *Kanwa* shows collecting ducts that appear degenerated and glomeruloid inflamed as compared to control and kidney of albino rat treated with *Tokansenyi* as shown in Figures 1 – 6. These results agreed with the finding of Okoye et al. (2016).

Conclusion

In conclusion, the histological results for liver and kidney for albino rats treated with *Kanwa* demonstrates its slight effects on the organs as compared with those treated with *Tokansenyi*. In comparing the two salts shows that *Tokansenyi* is relatively safer than *Kanwa*.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

The conception and design of the study, interpretation of data, drafting the article and final approval of the version to be submitted was by RJK, FOI, FEU and SSG. were responsible for the acquisition of data and analysis and also contributed to the interpretation of data.

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