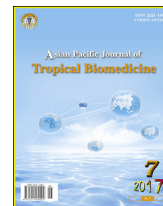




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### Toxicological evaluation and protective effect of ethanolic leaf extract of *Launaea taraxacifolia* on gentamicin induced rat kidney injury

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#### ABSTRACT

**Objective:** To evaluate the toxic potential of *Launaea taraxacifolia* leaf extract (LTE) in rats within 14 d of oral administration and also assess the potential of LTE in protecting against kidney injury induced by gentamicin using rat model.

**Methods:** The protective ability of LTE was done after sub-acute toxicity evaluation has been carried out. Acute Kidney Injury (AKI) was induced by gentamicin at a dose of 160 mg/kg intraperitoneal *i.p.* Parameters and indicators considered include mortality, clinical signs, body and organ weights, haematological and clinical chemistry parameters. Gross examination and histopathological assessment was also done on selected internal organs.

**Results:** There were no treatment-related deaths or changes in clinical signs, haematological and clinical chemistry indices during sub-acute toxicity studies with the exception of creatinine levels. This was confirmed by micrographs obtained from histopathological analysis. Co-administration of LTE with 160 mg/kg of gentamicin (*i.p.*) markedly decreased the levels of urea and creatinine when compared to negative control group. Histological studies of kidney tissues showed an insignificant change in tubular epithelium in LTE plus gentamicin treated group compared to LTE treated only.

**Conclusions:** Data obtained show that ethanolic leaf extract of *Launaea taraxacifolia* is non-toxic within a 14 d administration at a maximum dose of 1000 mg/kg bwt and also possesses the ability to protect against gentamicin-induced kidney damage in rats at a dose of 300 mg/kg bwt.

## 1. Introduction

Gentamicin is amongst the most common antibiotic in the aminoglycoside class that has been approved by many countries for clinical use in managing serious infections caused by aerobic gram-negative bacilli [1]. Nephrotoxicity is a major problem with its usage [2]. Discovering a drug that could be used in the reduction of this toxic effect could enhance its clinical use. Though medications such as deferoxamine, methimazole,

vitamin E, vitamin C, diethyldithiocarbamate, L-histidinol and thymoquinon are used for prevention of gentamicin-induced kidney damage [3], none has proven to be clinically efficient at providing all-embracing protection.

Plant medicine research is an area in the drug discovery process that has proven to be successful with managing several conditions. In this report, a plant commonly referred to as African lettuce that is claimed to possess several medicinal properties [4] was studied. The plant botanically referred to as *Launaea taraxacifolia*, is found in the Tropics and belongs to the family Asteraceae. Previous studies conducted on *Launaea taraxacifolia* leaves showed the presence of flavonoids, tannins, terpenoids, saponins, steroids [5,6].

Though there are reports of bioactivities such as hypolipidemic [7] renoprotective and hepatoprotective in Cisplatin model [8], to date no report is available on *Launaea*

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*taraxacifolia* leaf extracts' ability to protect against gentamicin-induced kidney injury, which is major problem with gentamicin usage. Thus the current study sought to examine the *in vivo* toxic potential of *Launaea taraxacifolia* leaves extract in Sprague-Dawley rats and also assess the ability of the extract in protecting against gentamicin induced kidney damage in rat model.

## 2. Materials and methods

### 2.1. Chemicals, reagents and drugs used

Normal saline, ethanol, EDTA were purchased from BDH Chemicals Ltd Poole England, Gentamicin sulphate (Roche Pharmaceutical Ltd, China).

### 2.2. Collection and preparation of plant material

Fresh tender leaves of *Launaea taraxacifolia* were collected from the Kwame Nkrumah University of Science and Technology (KNUST) Pharmacognosy garden. Samples were identified and authenticated at the Department of Pharmacognosy, Faculty of Pharmacy and Pharmaceutical Sciences, KNUST, Kumasi, Ghana, with a voucher specimen # KNUST/HM1/2012/L060. The fresh leaves were washed and air-dried for three weeks. The dried leaf samples were pulverized and macerated (1: 10 w/v) in 70% ethanol for 3 d. The leaf extract was filtered through a Whatman No 1. filter paper. The extract was concentrated under reduced pressure at 50 °C and the dried extract stored in airtight container and kept at –20 °C in a freezer till usage. The yield obtained was about 34.5%.

### 2.3. Experimental animals and maintenance

Sprague-Dawley (SD) rats were obtained from the Centre for Plant Medicine Research, Akuapem Mampong, Ghana. All experimental protocols were carried out in accordance with guidelines on the use and care of experimental animals as provided by the Organization for Economic Cooperation and Development (OECD) and approved by the Faculty of Pharmacy and Pharmaceutical Sciences Animal Experimentation Ethics committee of the Kwame Nkrumah University of Science and Technology (KNUST), Kumasi, Ghana. Animals were fed on rat feed supplied by Ghana Agro Food Company Tema, Ghana and water *ad libitum*. They were maintained at a 12 h light/dark cycle at (25 ± 2) °C. All animals were allowed to acclimatize to their new environment for two weeks with adequate fresh water and food before the start of any experiment.

### 2.4. Toxicological evaluation in rat model

#### 2.4.1. Acute toxicity assessment in rat model

In this experiment, acute toxicity study was conducted according to the World Health Organization (WHO) guidelines for the evaluation of safety and efficacy of herbal medicines (WHO, 1993). Thirty male rats between (250–300) g were grouped into six with ( $n = 5$ ). Group 1 served as the control and received saline. Groups 2–6 received (10, 100, 300, 1000 and 5000 mg/kg bwt) of extract orally by gavage. Animals were observed every 30 min for 8 h for any abnormal behavioural.

#### 2.4.2. Sub-acute toxicity assessment in rat model

Twenty-five male rats weighing between (250–300) g were grouped into five with ( $n = 5$ ). Group 1 served as the control and received normal saline as vehicle. Groups 2–5 orally received (10, 100, 300, 1000 mg/kg bwt) of extract. The treatments were repeated daily at 10: 00 am GMT for 14 d and observed for any abnormal changes. On day 15, blood samples were taken for haematological and biochemical analysis.

For haematological indices, blood samples were collected into sterile tubes containing EDTA and immediately analysed for TWBC, RBC, HB, PCV, MCV, MCH, MCHC, PLT and LYM using Sysmex Automated Analyser (model KX 21 Kobe, Japan).

Biochemical parameters were performed on blood collected into plain tubes without any anticoagulant. Collected blood samples were centrifuged at 3000 rpm for 5 min to obtain sera. The following biochemical indices namely total bilirubin (TBIL), direct bilirubin (DBIL), aspartate amino transferase (AST), alanine amino transferase (ALT), total protein (TP), albumin (ALB), globulin (GB), alkaline phosphatase (ALP),  $\gamma$ -glutamyltransferase (GGT), urea (URE), creatinine (CR) were measured using Flexor Junior autoanalyzer.

Livers, kidneys, testes, heart and spleen of all rats in experiment were excised, collected and organ weight measured. The body weight of each rat was also assessed during the acclimatization period, once before commencement of dosing, and once on the day of sacrifice after dosing. The relative organ to body weight ratio was then calculated.

### 2.5. Nephroprotective study in rat model

Male Sprague-Dawley rats weighing (150–200) g were selected for use in this experiment. Animals were randomly put into eight groups, with ( $n = 5$ ). Group I was given normal saline for 10 days. Group II that was negative control received normal saline from day 1–10, and also gentamicin sulphate 160 mg/kg bwt (*i.p.*) from day 6–10 simultaneously. Groups III, IV and V were orally given 10, 100 and 300 mg/kg/d (*p.o.*) extract respectively for 10 d. Groups VI, VII and VIII orally received 10, 100 and 300 mg/kg bwt extract respectively for 10 d and simultaneously with 160 mg/kg bwt gentamicin sulphate (*i.p.*) from day 6–10.

Experimental animals were all sacrificed after 10 days of treatment and blood samples collected and processed for measurement of serum creatinine (Cr), urea, total protein and plasma electrolytes. The kidneys were removed and processed for histopathology. The tissues were fixed in neutral buffered formalin, embedded in paraffin wax, cut into 3  $\mu$ m sections and stained with haematoxylin and eosin. They were then viewed under light microscopic (Nikon Eclipse E200, Japan) and captured by Infinity 1 camera microscope under  $\times 10$  magnification.

### 2.6. Data analysis

GraphPad Prism version 5 Software was used for data analysis. One Way ANOVA and Newman – Kuels' post hoc test was used to determine significant difference when compared to vehicle treated control group at 5% level of significance. Data were presented as mean  $\pm$  SEM.

### 3. Results

#### 3.1. Effect of LTE in rats during acute toxicity studies

In the acute toxicity studies, the assessments made included aggressiveness, vomiting, excitement, salivation, sedation, diarrhoea, eating, drinking and death. The treated group showed signs of sedation compared to the control. They exhibited healthy eating and drinking habits. There was no sign of salivation or diarrhoea. Extract doses did not result in lethality over the 24 h period. No latent toxicity was observed after keeping them for extra 14 days.

#### 3.2. Effect of LTE in rats during sub-acute toxicity studies

##### 3.2.1. Body weight

There were no marked changes in the weight of the animals treated with extract (10–1000) mg/kg (*p.o.*), compared to the vehicle treated control category ( $P > 0.05$ ). Meanwhile there was a marked difference in weight of animals at each dose after treatment with extract compared with vehicle control (data not shown).

##### 3.2.2. Effect of LTE on organ to body weight ratio

In assessing the effect of extract on organ to body weight ratio, the target organs including the liver, heart, kidney, testes and spleen did not show significant differences ( $P > 0.05$ ) compared to the vehicle treated control group (Table 1).

##### 3.2.3. Effect of LTE on haematological indices

There were no significant difference in the treatment groups in the total white blood cells (TWBC), red blood cell (RBC) count, haemoglobin concentration (HB), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), platelet (PLT), and lymphocytes (LYM) compared to the vehicle treated control group (Table 2).

##### 3.2.4. Effect of LTE on serum biochemical parameters

The effect of extract doses on serum biochemical indices after 14 days of treatment, revealed no marked differences in albumin, globulin, total protein, direct bilirubin (Dbilirubin), indirect bilirubin (IDbilirubin), total bilirubin (Tbilirubin),

**Table 1**

Effect of extract on relative organ-body weight ratio in rats treated for 14 days during sub acute toxicity studies.

Treatment	Organ-body weight ratio (%)				
	Liver	Heart	Kidney	Testes	Spleen
Control	2.89 ± 0.11	0.56 ± 0.02	0.33 ± 0.02	0.99 ± 0.05	0.21 ± 0.01
10 mg/kg	2.14 ± 0.41	0.51 ± 0.02	0.35 ± 0.06	0.91 ± 0.07	0.22 ± 0.02
100 mg/kg	3.11 ± 0.20	0.53 ± 0.33	0.36 ± 0.04	0.84 ± 0.08	0.22 ± 0.01
300 mg/kg	2.98 ± 0.11	0.51 ± 0.02	0.34 ± 0.01	0.99 ± 0.13	0.22 ± 0.00
1000 mg/kg	2.78 ± 0.51	0.48 ± 0.03	0.32 ± 0.02	0.83 ± 0.07	0.22 ± 0.05

Data presented as mean ± SEM ( $n = 5$ ). Data analysis was done by Newman–Kuels' test in One way ANOVA when compared to vehicle treated control group.

**Table 2**

Haematological parameters of rats treated with the extract during sub acute toxicity studies.

Parameters	Control	10 mg/kg	100 mg/kg	300 mg/kg	1000 mg/kg
TWBC( $\times 10^3/\mu\text{L}$ )	6.867 ± 0.584	9.300 ± 0.873	9.567 ± 0.887	8.733 ± 2.267	7.267 ± 0.176
RBC ( $\times 10^6/\mu\text{L}$ )	7.927 ± 0.145	7.693 ± 0.154	7.590 ± 0.066	7.493 ± 0.159	7.647 ± 0.149
HB (g/dL)	14.870 ± 0.240	13.670 ± 0.318	13.600 ± 0.153	14.100 ± 0.416	13.770 ± 0.318
PCV (%)	48.470 ± 1.214	44.500 ± 1.514	43.77 ± 0.991	45.470 ± 1.934	46.170 ± 0.754
MCV (fL)	61.130 ± 0.406	57.800 ± 1.168	58.000 ± 1.401	60.630 ± 1.272	59.330 ± 0.731
MCH (pg)	18.770 ± 0.088	17.770 ± 0.203	17.930 ± 0.318	18.830 ± 0.145	18.270 ± 0.176
MCHC (g/dL)	30.700 ± 0.306	30.730 ± 0.338	30.930 ± 0.233	31.030 ± 0.433	30.800 ± 0.666
PLT ( $\times 10^3/\mu\text{L}$ )	787.70 ± 57.18	731.30 ± 21.67	831.30 ± 33.38	789.00 ± 87.00	691.70 ± 21.98
LYM (%)	70.800 ± 4.681	75.200 ± 5.237	72.870 ± 0.837	72.870 ± 0.837	69.730 ± 9.427

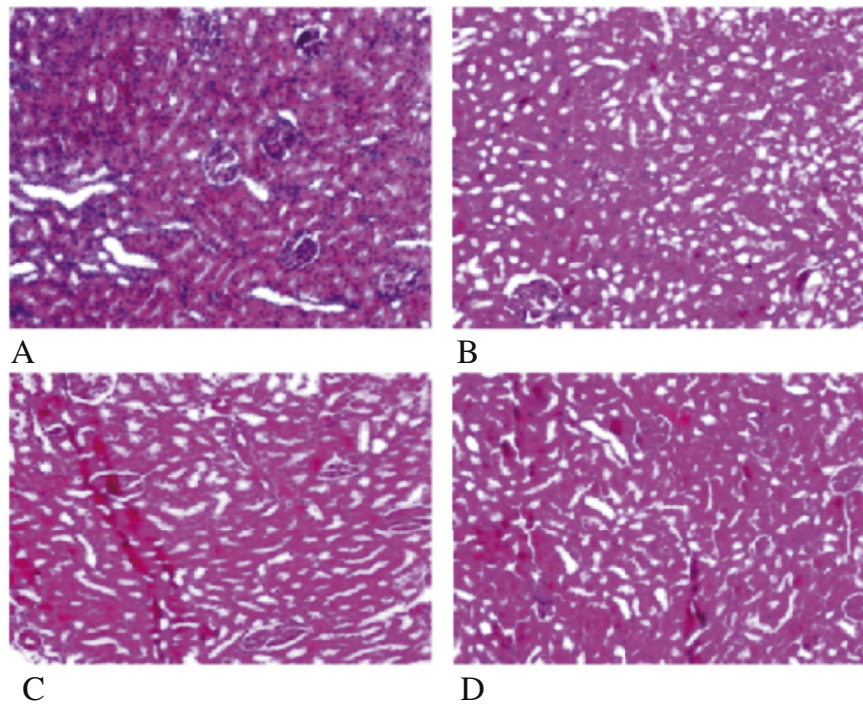
Data are represented as mean ± SEM ( $n = 5$ ). Data analysis was done by Newman - Kuels' test in One way ANOVA when compared to vehicle treated control group.

**Table 3**

Biochemical parameters of rats treated with the extract during sub acute toxicity studies.

Parameters	Control	10 mg/kg	100 mg/kg	300 mg/kg	1000 mg/kg
Albumin (g/dL)	38.87 ± 0.713	34.33 ± 2.173	35.07 ± 1.260	38.80 ± 2.193	35.00 ± 2.401
Globulin (mg/dL)	36.81 ± 2.437	32.93 ± 4.707	34.44 ± 3.226	31.93 ± 0.476	29.40 ± 1.372
Total protein (g/dL)	75.70 ± 2.937	67.27 ± 2.976	69.53 ± 1.994	70.77 ± 1.876	70.10 ± 4.272
ALP (U/L)	689.0 ± 43.21	643.0 ± 57.83	558.3 ± 58.87	504.3 ± 60.51	491.0 ± 54.50
ALT (U/L)	108.3 ± 4.604	73.00 ± 23.76	120.2 ± 4.550	132.1 ± 12.99	155.8 ± 21.02
AST (U/L)	206.6 ± 16.86	146.0 ± 8.807	172.6 ± 9.968	202.0 ± 7.663	184.7 ± 5.686
GGT (U/L)	0.167 ± 0.033	0.133 ± 0.08	0.100 ± 0.100	0.133 ± 0.033	0.100 ± 0.057
Dbilirubin (mmol)	1.100 ± 0.058	1.000 ± 0.058	0.967 ± 0.067	0.700 ± 0.116	0.800 ± 0.173
IDbilirubin (mmol)	0.407 ± 0.134	0.483 ± 0.136	0.683 ± 0.185	0.670 ± 0.085	0.927 ± 0.495
Tbilirubin (mmol)	1.500 ± 0.153	1.467 ± 0.120	1.433 ± 0.088	1.333 ± 0.088	1.367 ± 0.033
Creatinine (mg/dL)	90.10 ± 1.069	70.80 ± 0.924*	66.20 ± 0.945**	76.10 ± 2.050*	71.00 ± 7.578**
Urea (mg/dL)	9.237 ± 0.491	6.393 ± 1.083	6.377 ± 0.157	7.733 ± 0.519	7.130 ± 0.220

Values are represented as Mean ± SEM ( $n = 5$ ). Data analysis was done by Newman–Kuels' test in One way ANOVA when compared to vehicle treated control group. \* $P < 0.05$  compared with control group; \*\* $P < 0.01$  compared with control group.

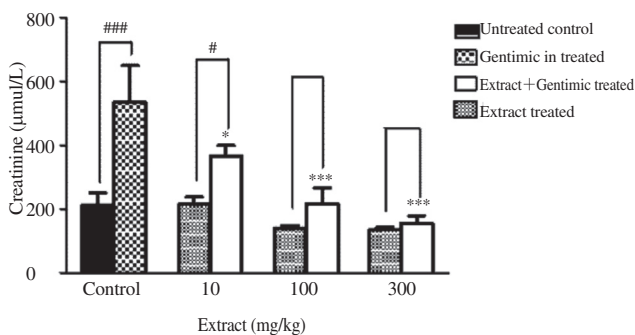


**Figure 1.** Photomicrographs of left kidney tissue of control and extract treated rats during sub-acute toxicity study. A: Control; B: 10 mg/kg; C: 100 mg/kg; D: 300 mg/kg.

alanine aminotransferase (ALT), alkaline phosphatase (ALP) and gamma glutamyl transferase (GGT), compared to the vehicle treated control group. Creatinine was significantly decreased ( $P < 0.05$ ) at doses 10 and 300 mg/kg and also at doses 100 and 1000 mg/kg ( $P < 0.01$ ). Though urea showed a general decrease at all doses, it was statistically insignificant compared to the vehicle treated control (Table 3).

### 3.2.5. Effect of LTE on kidney tissues micrographs

Kidney tissue micrographs of 10, 100 & 300 mg/kg *p.o* extract treated group (Figure 1) revealed no significant changes in kidney cells as evidenced by the absence of tubular degeneration and desquamation, tubular necrosis, mononuclear cells infiltration, intertubular haemorrhage compared to normal control group (Figure 1).



**Figure 2.** Effect of extract on serum creatinine levels of rats after 10 days treatment period by gavage.

The values were expressed as mean  $\pm$  SEM ( $n = 5$ ). \* $P < 0.05$  compared to the vehicle-treated group; \*\*\* $P < 0.001$  compared to the vehicle-treated group by Newman–Kuels' post hoc test in one-way ANOVA; # $P < 0.05$  compared to the vehicle-treated group; ### $P < 0.001$  compared to the vehicle-treated group by Bonferroni post hoc test in two-way ANOVA.

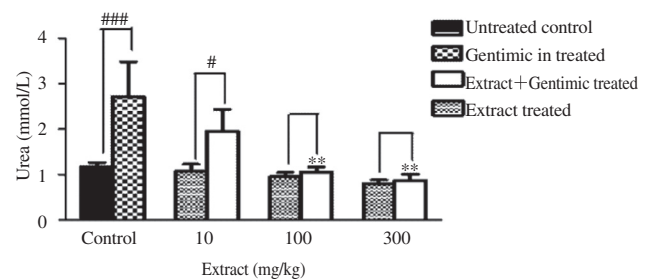
### 3.3. Effect of LTE on gentamicin-induced kidney damage in Sprague–Dawley rats

#### 3.3.1. Serum creatinine levels

Two-way analysis showed that the treatment with gentamicin significantly ( $P < 0.001$ ) increased creatinine levels. However, treatment with extract significantly ( $P < 0.001$ ) inhibited increase in creatinine levels in a dose dependent manner (Figure 2).

#### 3.3.2. Serum urea levels

Two-way analysis showed that the treatment with gentamicin significantly ( $P < 0.001$ ) increased urea levels. However, treatment with extract significantly ( $P < 0.001$ ) inhibited the gentamicin-induced increase in urea levels in a dose dependent manner compared to the vehicle treated control group (Figure 3).



**Figure 3.** Effect of extract on serum urea levels of rats after 10 days oral treatment period.

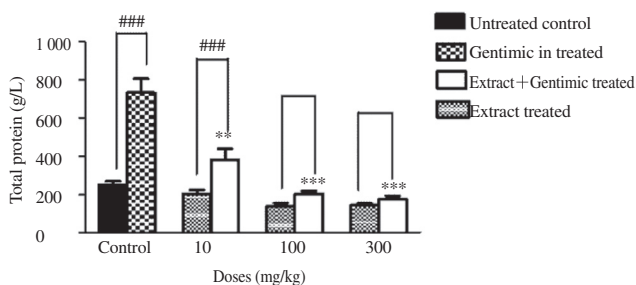
The values were expressed as mean  $\pm$  SEM ( $n = 5$ ). \*\* $P < 0.01$  compared to the vehicle-treated group by Newman–Kuels' post hoc test in one-way ANOVA. # $P < 0.05$  compared to the vehicle-treated group; ### $P < 0.001$  compared to the vehicle-treated group by Bonferroni post hoc test in two-way ANOVA.

### 3.3.3. Serum total protein levels

Two-way analysis showed that the treatment with gentamicin significantly ( $P < 0.001$ ) increased total protein levels. However, treatment with extract significantly ( $P < 0.001$ ) inhibited the gentamicin-induced increase in total protein levels in a dose dependent manner compared to the vehicle-treated control group (Figure 4).

### 3.3.4. Serum electrolyte levels

Two-way analysis showed that the treatment with gentamicin significantly ( $P < 0.001$ ) increased sodium and chloride levels but decreased potassium level. However, treatment with extract significantly ( $P < 0.001$ ) inhibited the gentamicin-induced increase in sodium and chloride levels and showed an increase in potassium levels in a dose dependent manner compared to the vehicle-treated control group (Table 4).



**Figure 4.** Effect of extract on serum total protein levels of rats after 10 days oral treatment period.

The values were mean  $\pm$  SEM ( $n = 5$ ). \*\* $P < 0.01$  compared to the vehicle-treated control group; \*\*\* $P < 0.001$  compared to the vehicle-treated control group by Newman-Kuels' post hoc test by two-way ANOVA. #### $P < 0.001$  compared to the vehicle-treated group by Bonferroni post hoc test by two-way ANOVA.

**Table 4**

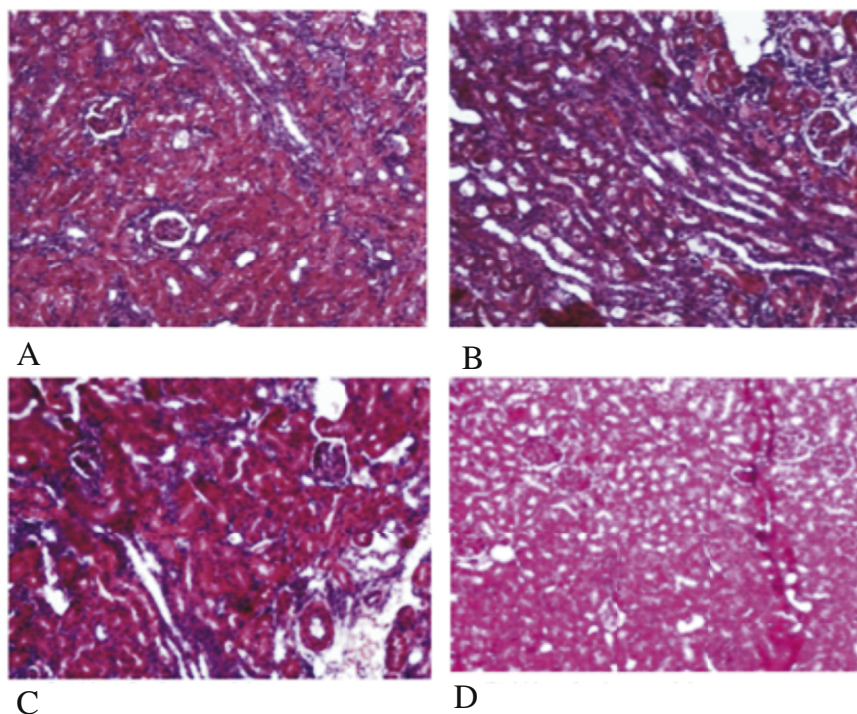
Effect of extract on serum electrolyte levels of rats treated orally for 10 days.

Treatment	Chloride	Potassium	Sodium
Control	124.9 $\pm$ 9.818	1.054 $\pm$ 0.021	139.7 $\pm$ 15.62
10 mg/kg	120.8 $\pm$ 13.25	1.256 $\pm$ 1.202	123.8 $\pm$ 6.391
100 mg/kg	104.4 $\pm$ 3.251	1.276 $\pm$ 0.092	122.0 $\pm$ 5.827
300 mg/kg	102.0 $\pm$ 1.286	1.202 $\pm$ 0.041	112.2 $\pm$ 5.986
GS 160 mg/kg	315.3 $\pm$ 35.70####	0.600 $\pm$ 0.510####	404.4 $\pm$ 46.20####
10 mg/kg & GS	235.1 $\pm$ 35.94*	1.153 $\pm$ 0.098*	288.9 $\pm$ 27.34**
100 mg/kg & GS	122.7 $\pm$ 13.40***	1.448 $\pm$ 0.034**	142.9 $\pm$ 6.882***
300 mg/kg & GS	121.7 $\pm$ 18.98**	1.346 $\pm$ 0.061**	145.6 $\pm$ 9.989***

The values are mean  $\pm$  SEM ( $n = 5$ ). \* $P < 0.05$  compared with control group; \*\* $P < 0.01$  compared with control group; \*\*\* $P < 0.001$  compared with control group; by Newman-Kuels' post hoc test by one-way ANOVA. #### $P < 0.001$  compared with control group by Bonferroni post hoc test by two-way ANOVA.

### 3.4. Photomicrographs of kidney tissue in gentamicin plus LTE treated group

Micrographs of gentamicin sulphate treated group without extract (Figure 5A) exhibited complete tubular degeneration and desquamation, tubular necrosis, mononuclear cells infiltration, intertubular haemorrhage and tubular brush boarder loss compared to extract treated groups (Figure 5 B, C & D). Treatment with gentamicin alone for 5 d showed severe damage to the kidney (Figure 5A). The extract reversed the renal damage caused by gentamicin (Figure 5 B, C and D). Animals treated with gentamicin plus extract showed a protective effect especially at a dose of 300 mg/kg (Figure 5D).



**Figure 5.** Photomicrographs of kidney tissue in gentamicin plus LTE treated group after 10 days.

A: Gentamicin 160 mg/kg; B: 10 mg/kg & gentamicin; C: 100 mg/kg & gentamicin; D: 300 mg/kg & gentamicin.

#### 4. Discussion

Due to the fact that some plant such as the stem bark of *Alstonia boonei*, fruits of *Pithecellobium lobatum*, roots of *Calilepis laureola*, fruit of *Morinda citrifolia* and *Averrhoa carambola* (star fruit) [9,10], have been reported to be toxic to some internal organs at certain doses, there was the need to initially investigate the toxic potential of *Launaea taraxacifolia* leaf. This was done to generally assess the potential health risk of the plant extract in rats. *Launaea taraxacifolia* is used as food and medicine for both humans and animals but has not been scientifically validated. Though a study by Adinortey et al. [5] has shown that *Launaea taraxacifolia* leaves is a good nutritional source, no information is available on the safety profile and protective potential in gentamicin induced kidney damage. This is what necessitated the initial assessment of the potential toxicity of extract of *Launaea taraxacifolia* leaves *in vivo*. The acute toxicity study carried out shows that administration (*p.o.*) of the extract at all doses tested did not cause death in rats. This implies that extract could be of low toxicity when administered orally.

Haematological parameters are usually associated with health status and are of diagnostic importance in clinical assessment. Haematological parameters are good indicators of physiological, pathological and nutritional status. In this study it was observed that, LTE had little or no adverse effect on RBC, Hb, platelet number, haematocrit (HCT) and lymphocyte. This points to the fact that neither the incorporation of haemoglobin into red blood cells nor the morphology and osmotic fragility of red blood cells was altered. Haematocrit is a measure of the volume of blood consisting of solid cells. The HCT which was unaffected by the extract in all the groups indicates that the volume of RBC and WBC in the blood remain constant. However, the total WBC showed an increase compared to the control group suggesting an improvement in the immune system of LTE - treated animals in the toxicity study [11].

Adverse interaction of the plant extract with the major organs would cause cellular constriction and inflammation, which usually reflects in the organ to body ratio [12]. In this study, no significant differences were found in the organ to body weight ratio and this gives an indication of the non-toxic nature of the LTE. Serum biochemical parameters are diagnostic markers of liver and kidney function. The globulin, albumin and total protein levels were not markedly affected by LTE compared to control group during the toxicity study. According to Adeoye and Oyedapo [13] an increase in plasma bilirubin is suggestive of a possible injury to the liver. An excess production of bilirubin may be due to failure of the liver to effectively conjugate it for excretion. In this study, LTE appeared to have caused a decrease in total bilirubin levels of treated animals at all doses used however it was not statistical significant. Plasma creatinine concentration is a better indicator than urea concentration in the first phase of kidney toxicity [14]. High levels of creatinine are found in renal dysfunction or muscle injury [15]. *Launaea taraxacifolia* extract-treated animals showed a significant reduction in serum creatinine, urea and total protein compared to the vehicle-treated group. In the initial toxicity assessment carried out, the extract produced a remarkable decrease in creatinine and urea levels suggesting that the extract could be a possible nephro-protective agent.

This observation prompted further investigation into the potential of *Launaea taraxacifolia* leaves extract in protecting against damage induced by Gentamicin. Meanwhile to the best

of our knowledge there is no scientific report about the protective effect of the ethanolic extract of *Launaea taraxacifolia* leaves on gentamicin induced kidney damage. Nephrotoxicity induced by gentamicin is a complex phenomenon characterized by increase in plasma creatinine and urea levels and severe proximal renal tubular necrosis, followed by deterioration and renal failure [16]. Gentamicin induced renal damage is a popular model used to study the effects of potential nephroprotective drug agents [17-19]. In the present study, *Launaea taraxacifolia* extract-treated group showed a significant improvement in serum creatinine, urea, total protein and electrolyte levels compared to the vehicle-treated group and the toxin control groups. The decrease in these biochemical indices recorded in LTE plus gentamicin treated groups in this study shows that the extract is possibly reversing kidney cell damage induced by the gentamicin.

Measurement of specific ion concentrations ( $\text{Na}^+$ ,  $\text{K}^+$  and  $\text{Cl}^-$ ), are used as potential biomarkers of chemical exposure and effects [20]. The balance of electrolytes is important for normal cellular function. Electrolytes promote fluid balance by maintaining blood volume, facilitation of fluid absorption and generation of impulses. A decrease in electrolyte levels may affect nerve conduction and reduced cell performance [21]. Electrolyte imbalance occurs with increased sodium and chloride and decreased potassium levels in toxin-exposed animals. The imbalance might impede proper nerve conduction and consequently result in death [22]. In this study a significant reticence of gentamicin induced increase in sodium and potassium ions and the reverse in chloride ions levels was observed in LTE treated group.

A relationship between oxidative stress and nephrotoxicity has been well confirmed in many experimental animal models. *Launaea taraxacifolia* leaves extract might have antagonized the biochemical activity of gentamicin on phospholipids in the proximal tubules via possible antioxidant and free-radical scavenging properties that has been reported by Koukoui et al. [7] This could probably also be due to a reduction in injury caused by oxygen-free radicals in the presence of the extract.

Histopathological analysis of gentamicin-treated animals showed that there was significant renal damage in the kidneys. However animals treated with gentamicin plus extracts showed protective effects especially at 300 mg/kg bwt of extract.

The present study indicates that extract of *Launaea taraxacifolia* leaves is not toxic at a dose of 1 000 mg/kg bwt and also possess the ability to protect against gentamicin-induced kidney damage in rats. The data suggest that extract of *Launaea taraxacifolia* could be useful in reducing gentamicin-induced nephrotoxicity.

#### Conflict of interest statement

We declare that we have no conflict of interest.

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