

## THE LEAVES OF *Bambusa vulgaris* (L.) AMELIORATE PRO- OXIDANTS INDUCED NEPHROTOXICITY IN RATS - *In vitro*

<sup>1</sup>O. Adeyanju, <sup>\*</sup>2S.F. Akomolafe, , <sup>3</sup>V. Atoki, <sup>3</sup>G. Oboh.

<sup>1</sup>. Department of Chemistry, University of Jos, Jos, Plateau State, Nigeria.

<sup>2</sup>. Department of Biochemistry, Ekiti State University, Ado Ekiti, Ekiti State, Nigeria.

<sup>3</sup>. Department of Biochemistry, Federal University of Technology, Akure, Nigeria.

\*Corresponding Author's e-mail; [purposefulseun@yahoo.co.uk](mailto:purposefulseun@yahoo.co.uk)

### ABSTRACT

*Bambusa vulgaris* are plants associated with a myriad of medicinal uses in different parts of the world and its leaf is used in traditional Asian and African medicine to treat hepatitis, measles and renal disorders. This study sought to investigate the protective effect of aqueous and methanolic extracts of *Bambusa vulgaris* leaves on some pro-oxidants (cisplatin, sodium nitroprusside and FeSO<sub>4</sub>) induced nephrotoxicity in rats *-in vitro*. Aqueous and methanolic extracts of *Bambusa vulgaris* leaves were prepared (1:100 w/v) and used for subsequent analysis. The antioxidant properties and inhibitory effect of the extracts on cisplatin, FeSO<sub>4</sub> and SNP-induced nephrotoxicity in rats were determined *- in vitro*. The incubation of kidney homogenate in the presence of cisplatin and other pro-oxidants induced nephrotoxicity in rats by causing a marked elevation in kidney malondialdehyde (MDA) content. However, this damage was ameliorated by aqueous and methanolic extracts of *Bambusa vulgaris* leaves, suggesting its possible antioxidant and therapeutic properties. Also, the result revealed the highest free radical scavenging potentials, phenolic and flavonoid contents in the methanolic extract. Free radical scavenging potentials of the extracts were found to be proportional to their respective phenolic and flavonoid contents. The results suggest that methanol may be a good solvent of extraction in the exploitation of the antioxidant property of *Bambusa vulgaris* leaves and part of the mechanism through which the *Bambusa vulgaris* leaves protect against oxidative stress and some pro-oxidants induced nephrotoxicity may be through Fe<sup>2+</sup>-chelation, ABTS\*, DPPH, and hydroxyl radical scavenging abilities and reducing power. Thus, *Bambusa vulgaris* leaves could be used for the management of cisplatin toxicity and acute renal damage.

**Keywords:** Oxidative stress; Cisplatin, Nephrotoxicity, Antioxidants, Kidney and *Bambusa vulgaris* leaves.

### INTRODUCTION

Nephrotoxicity involves kidney damage or dysfunction arising from direct or indirect exposure to drugs and industrial or environmental chemicals. Drugs such as cisplatin, cyclosporine and prograf have been reported to induce nephrotoxicity (Sreedeyi and Bharathi, 2010). The

kidney, which is the major route of cisplatin excretion also accumulates cisplatin to a greater degree than other organs (Ronan *et al.*, 2010; Tadagavagi and Reeves, 2010) Oxidative stress, inflammation and apoptosis are some of the mechanisms that explain cisplatin-induced acute kidney injury. A number of strategies have been proposed for the prevention or management of

cisplatin-induced nephrotoxicity with the use of some synthetic drugs. However, these drugs have some associated risks and side-effects (Dong *et al.*, 2010), hence there is a need to search for natural alternatives of plant origin (plant foods/extracts) with little or no side effect.

The use of plants for the prevention and management of diseases have been employed in folklore since time immemorial. Plants have limitless ability to synthesize aromatic substances such as polyphenols, flavonoids and phenolic acids. These compounds exhibit antioxidant properties due to their hydrogen-donating and metal-chelating capacities (Scalbert and Williamson, 2000).

Among popular medicinal plants is *Bambusa vulgaris* (L.) (*B. vulgaris*) which is a member of the Gramineae (Poaceae) family (Kirtikar and Basu, 1990). Different parts of the plant have been used to treat different ailments in folklore. The root is usually burnt and the ash is used to treat bleeding gums and skin infections caused by ringworm (Khare, 2007). Numerous studies have reported the anti-inflammatory, anti-ulcer, anti-diabetic (RathodTaimik *et al.*, 2011) and anti-bacterial activities (Zhang *et al.*, 2010) of the plant. The young shoots are edible and sold as a vegetable mainly in Asia. This specie is also used in traditional Asian and African medicine where young shoots are boiled and used to treat hepatitis, measles and renal disorders (Dransfield and Widjaja, 1995).

Despite the known therapeutic properties of *B. vulgaris*, to the best of our knowledge, scientific information on its protective effects on the renal system is scanty in literatures. We therefore hypothesized that aqueous or methanolic extract of this plant may protect against pro-oxidants induced nephrotoxicity in rats using *in-vitro* model.

## **MATERIALS AND METHODS**

### **Animals**

Male albino rats, weighing 90 – 165 g, were purchased from a private animal colony, Ado-Ekiti metropolis. The rats were maintained at 25°C on a 12 hour light/dark cycle with free access to food and water. They were acclimatized under these conditions for two weeks prior to the commencement of the experiments. The experimental study was approved by the Institutional Animal Ethical Committee of the University of Ado- Ekiti, Nigeria. The handling and use of the rats were in accordance with NIH Guide for the care and use of laboratory animals.

### **Sample collection**

Fresh samples of *B. vulgaris* leaves were obtained from a private farm in Ado - Ekiti metropolis, Ekiti - State, Nigeria. Authentication of the plant was carried out by Mr Ajayi in the Department of Biology, Ekiti State University, Ado – Ekiti, Nigeria. A voucher specimen (UHAE 336) was deposited in the herbarium of the same Department.

### **Aqueous extract preparation**

The plant materials were air dried after which they were homogenized and kept in an airtight container prior to the extraction. About 1.0 g of the sample was soaked in 20 mL of distilled water for 24 hours (Oboh *et al.*, 2007). The mixture was filtered using whatmann No 1 filter paper and the filtrate centrifuged at 805 × g for 10 minutes. The clear supernatant collected is used for the assay.

### **Methanolic extract preparation**

Another sample of about 20 g was soaked in 2000 ml of 80% methanol for 24 hours. The mixture was filtered using whatmann No 1 filter paper and cotton wool into storage container. The filtrate was evaporated under pressure at 40°C and 1.0 g of the

dried sample was later reconstituted in 100 mL of distilled water and used for the assay.

### **Chemicals and reagents**

Thiobarbituric acid (TBA), 1,10-phenanthroline, deoxyribose, gallic acid, and Folin-Ciocalteu's reagent were procured from Sigma-Aldrich, Inc., (St. Louis, MO, USA), trichloroacetic acid (TCA) was sourced from Sigma-Aldrich, Chemie GmbH (Steinheim, Germany), dinitrophenyl hydrazine (DNPH) from ACROS Organics (NJ, USA), hydrogen peroxide, methanol, acetic acid, and FeCl<sub>3</sub> were sourced from BDH Chemicals Ltd., (Poole, England), thiourea, CuSO<sub>4</sub> · 5H<sub>2</sub>O, H<sub>2</sub>SO<sub>4</sub>, sodium carbonate, AlCl<sub>3</sub>, potassium acetate, Tris-HCl buffer, sodium dodecyl sulphate, FeSO<sub>4</sub> and potassiumferricyanide were of analytical grade.

### **Determination of total phenolic content**

The total phenolic content was determined in the extracts using the method of Singleton *et al.* (1999). 2.5 mL of 10% Folin-Ciocalteu's reagent (v/v) was used to oxidize 250 µL of the extracts and neutralized by 2.0 mL of 7.5% sodium carbonate. The reaction mixture was incubated for 40 minutes at 45°C and the absorbance was measured at 765 nm in the spectrophotometer (JENWAY 6305, Barloworld Scientific, Dunmow, United Kingdom). The total phenolic content was subsequently calculated as gallic acid equivalent.

### **Determination of total flavonoid content**

The total flavonoid contents of both extracts were determined using the method of Meda *et al.* (2005). Briefly, 0.5 mL of the sample was mixed with 0.5 mL methanol, 50 µL 10% AlCl<sub>3</sub>, 50 µL of 1 M Potassium acetate and 1.4 mL of distilled water, and allowed to incubate at room temperature (37°C) for 30 minutes. The absorbance of the reaction mixture was subsequently measured at 415 nm and the total

flavonoid content was subsequently calculated as quercetin equivalent.

### **Determination of vitamin C content**

Vitamin C content of the two extracts was determined using the method of Benderitter *et al.* (1998). Briefly, 75 µL DNPH (2 g dinitrophenyl hydrazine, 230 mg thiourea and 270 mg CuSO<sub>4</sub>·5H<sub>2</sub>O in 100 mL of 5M H<sub>2</sub>SO<sub>4</sub>) were added to 500 µL reaction mixture (300 µL of extract with 100 µL 13.3% trichloroacetic acid (TCA) and water). The reaction mixtures were subsequently incubated for 3 hours at 37°C, then 0.5 mL of 65% H<sub>2</sub>SO<sub>4</sub> was added to the medium. The absorbance of the reaction mixture was measured at 520 nm. The vitamin C content of the samples was subsequently calculated as ascorbic acid equivalent.

### **Preparation of tissue homogenates**

The rats were decapitated under mild diethyl ether anesthesia and the kidney were rapidly dissected and placed on ice and weighed. This tissue was subsequently homogenized in cold saline (1/10w/v) with about 10 strokes at approximately 1200 rev/minute in a Teflon glass homogenizer (Mexxcare, mc14 362, Aayu-shi Design Pvt. Ltd., India). The homogenate was centrifuged (KX3400C Kenxin Intl. Co. Hong Kong) for 10 minutes at 3000 × g to yield a pellet that was discarded, and a low-speed supernatant, which was kept for lipid peroxidation assay (Belle *et al.*, 2004).

### **Lipid peroxidation and thiobarbituric acid reactions**

The lipid peroxidation assay was carried out using the modified method of Ohkawa *et al.* (1979). Briefly, 100 µL of the SI fraction was mixed with a reaction mixture containing 30 µL of 0.1M pH 7.4 Tris- HCl buffer, extract (100 µL), and 30 µL of 250µM freshly prepared FeSO<sub>4</sub> as the pro-

oxidant. The volume was made up to 300  $\mu\text{L}$  by water before incubation at 37°C for 2 hours. The colour reaction was developed by adding 300  $\mu\text{L}$  8.1% SDS (sodium dodecyl sulphate) to the reaction mixture, this was subsequently followed by the addition of 500  $\mu\text{L}$  of acetic acid/HCl (pH 3.4) mixture and 500  $\mu\text{L}$  of 0.8% thiobarbituric acid (TBA). This mixture was incubated at 100°C for 1 hour. Thiobarbituric acid reactive species (TBARS) produced were measured at 532 nm and expressed as (%) malondialdehyde (MDA) produced using MDA standard curve (0–0.035mM). Also, for cisplatin and sodium nitroprusside (SNP) -induced lipid peroxidation, the procedure was carried out as outlined above using 1.0 mM and 5 mM of cisplatin and sodium nitroprusside (SNP) as the pro-oxidants respectively.

#### DPPH free radical scavenging ability

The free radical scavenging ability of the extracts against 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical was evaluated as described by Gyamfi *et al.* (1999). Briefly, an appropriate dilution of the extract (1 mL) was mixed with 1 mL of 0.4mM methanolic solution containing DPPH radicals, the mixture was left in the dark for 30 minutes and the absorbance was measured at 516 nm. The control was carried out using 2 mL DPPH solution without the test samples. The DPPH free radical scavenging ability was subsequently calculated thus: DPPH scavenging ability (%) =  $[(\text{Abs}_{\text{con}} - \text{Abs}_{\text{sam}})/\text{Abs}_{\text{con}}] \times 100$ . Where  $\text{Abs}_{\text{con}}$  is the absorbance without the extract and  $\text{Abs}_{\text{sam}}$  is the absorbance of the extracts.

#### Fe<sup>2+</sup> -chelation assay

The Fe<sup>2+</sup> chelating ability of both extracts was determined using a modified method of Minotti and Aust (1987) and Puntel *et al.*, (2005). Freshly prepared 500  $\mu\text{M}$  FeSO<sub>4</sub> (150  $\mu\text{L}$ ) was added to a reaction mixture containing 168  $\mu\text{L}$  of 0.1M Tris-

HCl (pH 7.4), 218  $\mu\text{L}$  saline and the extract. The reaction mixture was incubated for 5 minutes before the addition of 13  $\mu\text{L}$  of 0.25% 1,10-phenanthroline. The absorbance was subsequently measured at 510 nm. The Fe (II) chelating ability was subsequently calculated with respect to the reference (which contains all the reagents without the test sample).

#### Determination of reducing property

The reducing property of the extracts was determined by assessing the ability of the extracts to reduce FeCl<sub>3</sub> solution as described by Oyaizu (1986). A 2.5 mL aliquot was mixed with 2.5 mL of 200 mM sodium phosphate buffer (pH 6.6) and 2.5 mL of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes and 2.5 mL of 10% trichloroacetic acid was added. This mixture was centrifuged at 650 rpm for 10 minutes. 5 mL of the supernatant was mixed with an equal volume of water and 1 mL of 0.1% ferric chloride. The absorbance was measured at 700 nm. The ferric reducing antioxidant property was subsequently calculated using ascorbic acid as standard.

#### 2,2-azinobis (3-ethylbenzo-thiazoline-6-sulfonate) ABTS\* scavenging ability

The ABTS\* scavenging ability of both extracts was determined according to the method of Re *et al.* (1999). ABTS\* was generated by reacting an ABTS\* aqueous solution (7 mmol L<sup>-1</sup>) with K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> (2.45 mmol L<sup>-1</sup>, final concentration) in the dark for 16 hours and adjusting the absorbance 734 - 0.700 nm with ethanol. 0.2 ml of the extract was added to 2.0 ml ABTS\* solution and the absorbance was measured at 734 nm after 15 minutes. The result was reported as trolox equivalent antioxidant capacity.

#### Data analysis

The results of replicate readings were pooled and expressed as mean  $\pm$  standard deviation. One way

analysis of variance was used to analyze the results and Duncan multiple test was used for the post hoc (Zar, 1984). Statistical package for Social Science (SPSS) 16.0 for Windows was used for the analysis. The significance level was taken at  $P < 0.05$ .

## RESULTS

The incubation of the kidney homogenate in the presence of cisplatin caused a significant increase

in the MDA content when compared with the basal kidney homogenate (Figure 1a). However, the two extracts from the plant inhibited MDA production in rat kidney in a dose-dependent manner. Nevertheless, judging by the  $EC_{50}$  values (extract concentration causing 50% enzyme inhibition), methanolic extract of *B. vulgaris* had a significantly higher inhibitory effect on cisplatin-induced lipid peroxidation in the kidney homogenate than the aqueous leaf extract (Table 1).

**Table 1:  $EC_{50}$  values of inhibition of cisplatin,  $FeSO_4$  and Sodium nitroprusside –induced lipid peroxidation by aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**

	Cisplatin- induced	$FeSO_4$ -induced	Sodium nitroprusside -induced
Aqueous extract	$1.29 \pm 0.35^a$	$0.81 \pm 0.02^c$	$0.72 \pm 0.04^a$
Methanolic extract	$0.93 \pm 0.01^b$	$0.69 \pm 0.05^d$	$0.74 \pm 0.13^a$

Values represents mean  $\pm$  standard deviation, number of samples,  $n = 3$ . Values with the same superscript letter along the same column are not significantly ( $p \leq 0.05$ ) different.

Likewise, incubation of rat's kidney tissue homogenate in the presence of  $FeSO_4$  and sodium nitroprusside also caused a significant increase in the kidney malondialdehyde (MDA) content (Figure 1b and 1c). However, both extracts inhibited MDA production content in the kidney in a dose-dependent manner. Judging by the  $EC_{50}$  value, there was no significant difference between the two extracts of the plant for the sodium nitroprusside –induced lipid peroxidation but for  $FeSO_4$  –induced lipid peroxidation, methanolic extract had the highest inhibitory effect compared with the aqueous extract (Table 1).

The findings of our study revealed that the methanolic extract had the highest total phenolic content, while the content of the total phenolic in the two extracts were statistically greater than the standard (Table 2). Furthermore, the total flavonoid content of methanolic extract was higher than that of aqueous extract. Also, the aqueous extract of the plant had appreciable amount of vitamin C content compared to the standard (Table

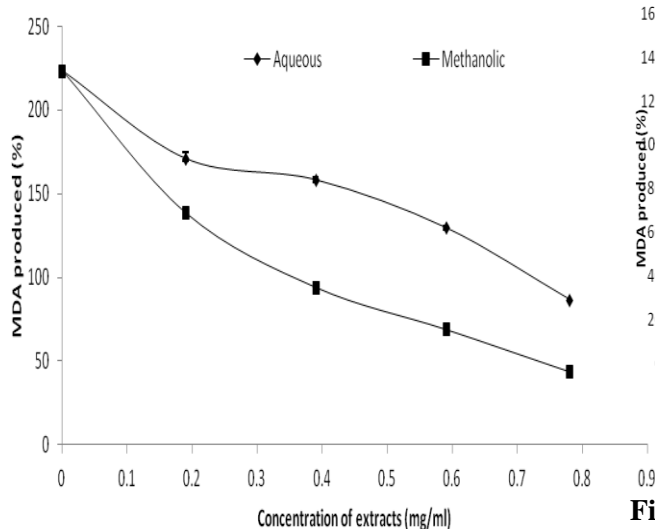
2). The results also revealed that aqueous extract of *B. vulgaris* leaves exhibited significantly higher antioxidant activity compared to the standard. Also, its value was statistically higher than the methanolic extract (Table 2). The trend of inhibition of DPPH radical by the two extracts were concentration dependent (Figure 2). The methanolic extract of the plant exhibited the highest inhibitory effect on DPPH radical which corresponds to its phenolic content. In this study, the ABTS radical scavenging activity of methanolic extract was higher than the aqueous extract, but the two values obtained for the two extracts were lower when compared with the standard (Table 2).

The two extracts chelate  $Fe^{2+}$  in a concentration dependent manner with methanolic extract being a better chelator than aqueous leaf extract (Figure 3). The two extracts of the plant scavenged hydroxyl radicals in a concentration dependent manner. The scavenging ability of the methanolic extract was higher than the aqueous extract (Figure 4).

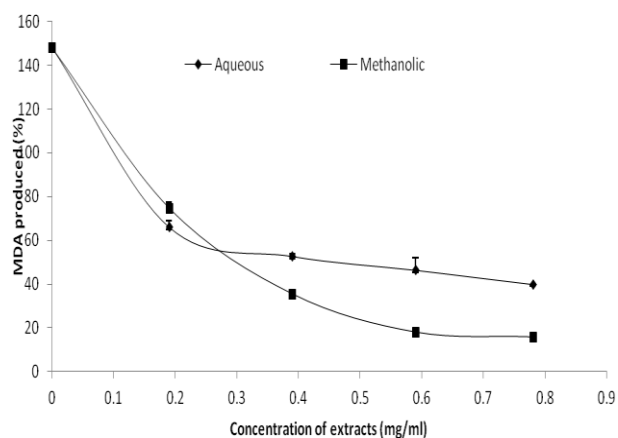
**Table 2: Phytochemical and antioxidant analyses of *Bambusa vulgaris* leaf.**

	Standard	Aqueous	Methanolic
Total phenolic (mg GAE/g)	1.44 ± 0.00 <sup>a</sup>	03.65 ± 0.15 <sup>c</sup>	7.34 ± 0.07 <sup>b</sup>
Total flavonoid (mg QUE/g)	0.28 ± 0.00 <sup>b</sup>	0.71 ± 0.00 <sup>a</sup>	2.39 ± 0.87 <sup>c</sup>
Vitamin C (mg AAE/g)	1.77 ± 0.04 <sup>a</sup>	17.34 ± 0.94 <sup>b</sup>	Not analysed
FRAP (mg AAE/g)	1.77 ± 0.04 <sup>b</sup>	08.75 ± 0.16 <sup>c</sup>	07.88 ± 0.16 <sup>a</sup>
ABTS* (mmol TEAC/g)	1.048 ± 0.02 <sup>c</sup>	0.036 ± 0.00 <sup>a</sup>	0.061 ± 0.01 <sup>b</sup>

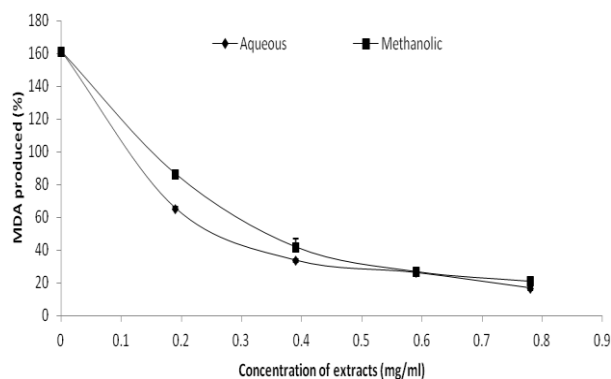
AAE, ascorbic acid equivalent; GAE, gallic acid equivalent; QUE, quercetin equivalent; TEAC, trolox equivalent antioxidant capacity. Values represent mean ± standard deviation of triplicate readings. Values with the same superscript letter along the same row are not significantly ( $p \leq 0.05$ ) different.



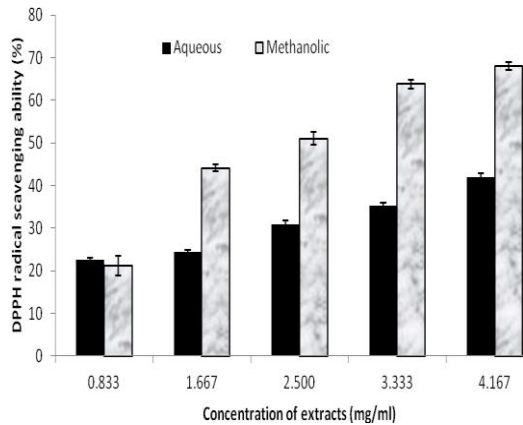
**Figure 1a: Inhibition of cisplatin -induced lipid peroxidation in rat's kidney by aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**



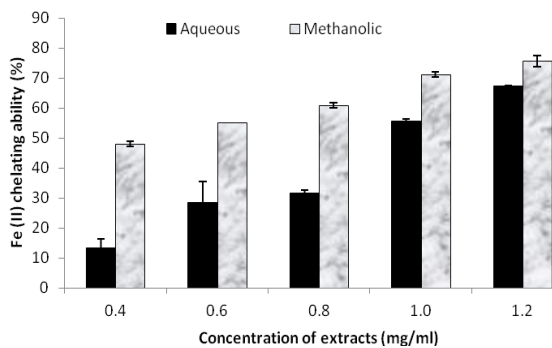
**Figure 1b: Inhibition of FeSO<sub>4</sub> -induced lipid peroxidation in rat's kidney by aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**



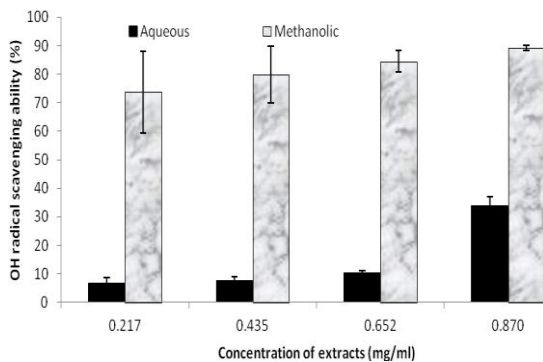
**Figure 1c: Inhibition of Sodium nitroprusside -induced lipid peroxidation in rat's kidney by aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**



**Figure 2: DPPH radical scavenging ability of aqueous and methanolic extracts of *Bambusa vulgaris* leaf**



**Figure 3: Fe (II) chelating ability of aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**



**Figure 4: OH\* radical scavenging ability of aqueous and methanolic extracts of *Bambusa vulgaris* leaf.**

## DISCUSSION

About 25% of the most commonly used drugs in intensive care units are potentially nephrotoxic and are regarded as considerable health and economic burden worldwide (Ciarimboli, 2010). Among these drugs is cisplatin. It has been reported that when used in cancer chemotherapy, cisplatin induces renal impairment and acute renal failure by its induction of reactive oxygen species, tubule interstitial inflammation and apoptosis (Sreedeyi and Bharathi, 2010). Although various studies have reported the protective effect of several agents on cisplatin induced renal toxicity, but the mechanism of nephroprotection remains elusive (Ronald *et al.*, 2010). Thus, this study sought to investigate the protective effect of aqueous and methanolic extracts of *B. vulgaris* leaves on some pro-oxidants (cisplatin, sodium nitroprusside and FeSO<sub>4</sub>) induced nephrotoxicity in rat's kidney – *in vitro*.

The incubation of the kidney homogenate in the presence of cisplatin caused a significant increase in the MDA content when compared with the basal kidney homogenate (Figure 1a). The increase in the kidney MDA content suggests lipid peroxidation. This concurred with earlier studies where the administration of cisplatin caused inflammation and lipid peroxidation (Sreedeyi and Bharathi, 2010; Ronald *et al.*, 2010). However, the two extracts from the plant inhibited MDA production in rat kidney in a dose-dependent manner. Nevertheless, judging by the EC<sub>50</sub> values (extract concentration causing 50% enzyme inhibition), methanolic extract of *B. vulgaris* had a significantly higher inhibitory effect on cisplatin-induced lipid peroxidation in the kidney homogenate than the aqueous leaf extract (Table 1). However, the reduced kidney MDA content of the two extracts in the rat kidney may be a function of the antioxidant properties of the *B. vulgaris* leaves.

Likewise, incubation of rat's kidney tissue homogenate in the presence of FeSO<sub>4</sub> and sodium

nitroprusside also caused a significant increase in the kidney malondialdehyde (MDA) content (Figure 1b and 1c). However, both extracts inhibited MDA production content in the kidney in a dose-dependent manner. Judging by the  $EC_{50}$  value, there was no significant difference between the two extracts of the plant for the sodium nitroprusside –induced lipid peroxidation but for  $FeSO_4$  –induced lipid peroxidation, methanolic extract had the highest inhibitory effect compared with the aqueous extract (Table 1). The protective properties of the plant extracts against sodium nitroprusside induced lipid peroxidation in the kidney could be because of the ability of the antioxidant phytochemicals present in the two extracts to quench or scavenge the nitrous radical and Iron (II) ions produced from the decomposition of sodium nitroprusside. This supports the earlier report correlating the presence of polyphenolic compounds to the antioxidant activity of natural plant products (Oyedemi *et al.*, 2010).

Phenolic substances are pharmacologically active components of plants which are capable of neutralizing free radicals, chelating metal catalysts and inhibiting the activity of oxidizing enzymes in biological systems (Dastmalchi *et al.*, 2007; Foti *et al.*, 1996). They are also capable of regenerating endogenous  $\alpha$ -tocopherol in the phospholipid bilayer of the membrane to its active antioxidant form. This mechanism of antioxidant action confers health beneficial potentials on phenolic substances (Mccall and Frei, 1999; Louli *et al.*, 2004). The findings of our study revealed that the methanolic extract had the highest total phenolic content, while the content of the total phenolic in the two extracts were statistically greater than the standard (Table 2). Furthermore, the total flavonoid content of methanolic extract was higher than that of aqueous extract. Also, the aqueous extract of the plant has a highly significant appreciable amount of vitamin C content compared to the standard (Table 2). The

appreciable level of phenolic compounds in the extracts of the leaves could be responsible for the use of this plant for the treatment of radical related problems such as diabetes, ulcer, skin infection and inflammation (RathodTaimik *et al.*, 2011; Zhang *et al.*, 2010).

A compound with antioxidant property due to its ability to produce reductants usually has reducing capacity (Alothman *et al.*, 2009; Wong *et al.*, 2006). The ability of *B. vulgaris* extracts to reduce ferric ions to its ferrous form is evident in the formation of Perl's blue, which was monitored spectrophotometrically at 700 nm. Our results revealed that aqueous extract of *B. vulgaris* leaves exhibited significantly ( $P < 0.05$ ) higher antioxidant activity compared to the standard. Also, its value was statistically higher than the methanolic extract (Table 2). Nevertheless, our study revealed that *B. vulgaris* is an electron donor that can react with free radicals to convert them into stable products and terminate the chain of reactions that leads to oxidative stress.

The trend of inhibition of DPPH radical by the two extracts were concentration dependent (Figure 2). The methanolic extract of the plant exhibited the highest inhibitory effect on DPPH radical which corresponds to its phenolic content. The strong inhibitory effect on DPPH radical of the two extracts could be linked to polyphenolic compounds which are capable of donating electrons to neutralize free radicals and thus, could be a promising therapeutic agent to treat stress induced pathological conditions.

In this study, the ABTS radical scavenging activity of methanolic extract was higher than the aqueous extract, but the two values obtained for the two extracts were lower when compared with the standard (Table 2). The inhibitory activity of the two extracts on ABTS radical could be assigned to the hydrogen proton donating ability of the extracts to the lone pair of ABTS radical. The donor could be adduced to the polyphenolic contents.



The two extracts chelate Fe<sup>2+</sup> in a concentration dependent manner with methanolic extract being a better chelator than aqueous leaf extract (Figure 3). However, this result is in agreement with the FeSO<sub>4</sub> -induced lipid peroxidation (Figure 1b), phenolic content (Table 2) and antioxidant activity of the extracts, suggesting that Fe<sup>2+</sup> chelation may be one of the possible mechanisms through which antioxidant phytochemicals from *B. vulgaris* leaf extracts prevent lipid peroxidation in tissue by forming a complex with Fe<sup>2+</sup>, thus preventing the initiation of lipid peroxidation.

Hydroxyl radicals have been implicated in the oxidative damage of DNA, proteins and lipids (Walker and Everette, 2009; Mates and Sanchez-Jimenez, 2000). Among the reactive oxygen species, hydroxyl radicals are the most reactive and predominant radicals generated endogenously during aerobic metabolism to initiate cell damage *in vivo* (Spencer *et al.*, 1994). The two extracts of the plant scavenged hydroxyl radicals in a concentration dependent manner. The scavenging ability of the methanolic extract was higher than the aqueous extract (Figure 4). This observation suggests that the extracts of *B. vulgaris* can be used as an alternative remedy to synthetic antioxidants in combating the oxidative activity of hydroxyl radicals.

## CONCLUSION

The incubation of rat's kidney homogenate in the presence of cisplatin and other pro-oxidants induced nephrotoxicity in rats. However, this damage was ameliorated by the aqueous and methanolic extracts of *B. vulgaris* leaves. This suggests that the plants have possible antioxidant and therapeutic properties. The mechanism of antioxidant action was based on the ability of its extracts to donate electrons, chelate Fe<sup>2+</sup>, reduce ferric ions, scavenge ABTS\*, DPPH and hydroxyl radicals. This has justified the use of this plant in folk medicine for the treatment of stress related diseases. The antioxidant potential of the plant was

also dependent on the solvent of extraction which means that methanol may be a good solvent of extraction in the exploitation of the antioxidant property of *B. vulgaris*. Therefore, the leaves of this species may be effective in the management of cisplatin toxicity and acute renal damage.

## Competing interest

The authors declare that they have no competing interests.

## Funding

This research was not funded by any grant received by any of the authors.

## Authors' contributions

The study design was by ASF (Ekiti State University), AO (University of Jos, Jos), OG (Federal University of Technology, Akure). ASF performed the experiments, ASF, AO, OG and AV were involved in the writing of the manuscript. All authors read and approved the final manuscript.

## REFERENCES

- Alothman, M., Bhat, R. and Karim, A. A.** (2009). Antioxidant capacity and phenolic content of selected tropical fruits from Malaysia, extracted with different solvents. *Food Chemistry*. 115: 785-788.
- Belle, N. A. V., Dalmolin, G. D., Fonini, G., Rubim, M. A and Rocha, J. B. T.** (2004) "Polyamines Reduces Lipid Peroxidation Induced by Different Pro-Oxidant Agents," *Brain Research*. 1008(2): 245-251.
- Benderitter. M., Maupoil, V., Vergely, C., Dalloz, F., Briot, F and Rochette, L.** (1998). "Studies by Electron Paramagnetic Resonance of the Importance of Iron in Hydroxyl Scavenging Properties of Ascorbic Acid in Plasma Effects of Iron Chelators," *Fundamental and Clinical Pharmacology*. 12(5): 510-516.
- Ciarimboli, G., Deuster, D., Knief, A., Sperling, M., Holtkamp, M., Edemir, B and Pavenstadt H.** (2010). Cisplatin -induced

- nephrotoxicity in rats. *Pakistan Journal of Pharmaceutical Science*. 21(3): 255-261.
- Dastmalchi, K., Dorman, H. J., Kosarm, D and Hiltunen, R.** (2007). Chemical composition and in vitro antioxidant evaluation of a water soluble Moldavian balm (*Dracocephalum moldavica* L.) extract. *LebensmittelWissenschaft und- Technologie*. 40: 239 - 248.
- Dong, G., Luo, J., Kumar, V and Dong, Z.** (2010). Inhibitors of histone deacetylases suppress cisplatin-induced p53 activation and apoptosis in renal tubular cells. *American Journal of Physiology-Renal Physiology*. 298: F293–F300.
- Dransfield, S and Widjaja, E. A.** (1995). *Bambusa vulgaris* Schrader ex Wendland. Record from Proseabase. Bogor, Indonesia: PROSEA (Plant Resources of South-East Asia) Foundation. <http://www.proseanet.org>.
- Foti, M., Piattelli, M., Baratta, M. T and Ruberto, G.** (1996). Flavonoids, coumarins, and cinnamic acids as antioxidants in a micellar system structure-activity relationship. *Journal of Agricultural and Food Chemistry*. 44, 497–501.
- Gyamfi, M. A., Yonamine, M and Aniya, Y.** (1999). “Free-Radical Scavenging Action of Medicinal Herbs from Ghana: *Thonningia Sanguinea* on Experimentally-Induced Liver Injuries,” *General Pharmacology*. 32(6): 661-667.
- Khare, C. P.** (2007). *Indian Medicinal Plants. An Illustrated Dictionary*. Springer publication. New Delhi, India . pp90.
- Kirtikar, K. R and Basu, B. D.** (1990). *Indian Medicinal Plants, volume-IV*, International Book Distributers, Dehradun, India. pp2724–2727.
- Louli, V., Ragoussis, N and Magoulas, K.** (2004) Recovery of phenolic antioxidants from wine industry byproducts. *Bioresource Technology*. 92, 201–208.
- Mates, J.M and Sanchez-Jimenez, F. M.** (2000). Role of reactive oxygen species in apoptosis: Implications for cancer therapy. *International Journal of Biochemistry and Cell Biology*. 2000; 32, 157–170.
- Mccall, M. R and Frei, B.** (1999). Can antioxidant vitamins materially reduce oxidative damage in humans? *Free Radical Biology and Medicine*. 1999; 26, 1034–1053.
- Meda, A., Lamien, C. E., Romito, M., Millogo, J and Nacoulma, O.G.** (2005) “Determination of the Total Phenolic, Flavon-oid and Proline Contents in Burkina Faso Honey, as Well as Their Radical Scavenging Activity” *Food Chemistry*. 91(3): 571-577.
- Minotti, G and Aust, S. D.** (1987) “An Investigation into the Mechanism of Citrate-Fe<sup>2+</sup>-Dependent Lipid Peroxidation,” *Free Radical Biology and Medicine*. 3(6): 379-387.
- Oboh, G., Puntel, R. L and Rocha, J. B. T.** (2007). “Hot Pepper (*Cap- sicumannum*, *Tepin* and *Capsicum chinese*, *Habanero*) Prevents Fe<sup>2+</sup>-Induced Lipid Peroxidation in Brain—in Vitro,” *Food Chemistry*. 102(1): 178- 185.
- Ohkawa, H., Ohishi, N and Yagi K.** (1979). “Assay for Lipid Per- oxides in Animal Tissues by Thiobarbituric Acid Reaction” *Analytical Biochemistry*. 95(2): 351-358.
- Oyaizu, M.** (1986). “Studies on Products of Browning Reaction: Antioxidative Activity of Products of Browning Reaction Prepared from Glucosamine,” *Japanese Journal of Nutrition*. 44(6): 307-315.
- Oyedemi, S. O., Bradley, G and Afolayan, A. J.** (2010). In vitro and in vivo antioxidant activities of aqueous extract of *Strychnoshenningsii* Gilg. *African Journal of Pharmacy and Pharmacology*. 4: 70-78.

- Puntel, P. R. L., Nogueira, C. W and Rocha, J. B. T.** (2005). "Krebs Cycle Intermediates Modulate Thiobarbituric Reactive Species (TBARS) Production in Rat Brain in Vitro," *Neurochemical Research*. 30(2): 225-235.
- RathodJaimik, D., PathakNimish, L., Patel Ritesh, G., Jivani, N. P.and Bhatt Nayna, M.** (2011). *Phytopharmacological Properties of Bambusaarundinaceas a Potential Medicinal Tree: An Overview*. *Journal of Applied Pharmaceutical Science*. 01 (10): 27-31.
- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M and Rice-Evans, C.** (1999). Antioxidant activity applying an improved ABTS radical cation decolorisation assay. *Free Radical Biology and Medicine*. 26: 1231-1237.
- Ronald, P. M., Raghu, K. T., Ganesan, R and Reeves, W. B.** (2010). Mechanisms of cisplatin nephrotoxicity. *Toxins*. 2: 2490–2518.
- Scalbert, A and Williamson, G.** (2000). Dietary intake and bioavailability of polyphenols. *Journal of Nutrition*. 130(8): 2073 - 2085.
- Singleton, V. L., Orthofer, R and Lamuela-Raventos, R. M.** (1999). "Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent," Academic Press, San Diego.
- Spencer, J. P. E., Jenner, A and Aruoma, O. I.** (1994). Intense oxidative DNA damage promoted by L-DOPA and its metabolites: implications for neurodegenerative disease. *FEBS Letters*. 353 (3):246-444.
- Sreedevi, A., Bharathi, K and Prasad K.V.S.R.G.** (2010). Effect of decoction of root bark of *Berberis Aristata* against cisplatin induced nephrotoxicity in rats. *International journal of Pharmacy and Pharmaceutical Sciences*. 2 2(3): 51–56.
- Tadagavadi, R. K and Reeves, W. B.** (2010). Endogenous IL-10 attenuates cisplatin nephrotoxicity: Role of dendritic cells. *Journal of Immunology*. 185: 4904–4911.
- Walker, R.B and Everette, J. O.** (2009) Comparative reaction rates of various antioxidants with ABTS 430 radical cation. *Journal of Agricultural and Food Chemistry*. 57(4), 1156-1161.
- Wong, C., Li, H., Cheng, K. and Chen, F.** (2006). A systematic survey of antioxidant activity of 30 Chinese medicinal plants using the ferric reducing antioxidant power assay. *Food Chemistry*. 97: 705-711.
- Zar, J. H.** (1984). *Biostatistical Analysis* Prentice-Hall, Inc., Upper Saddle River, NJ, pp 620.
- Zhang, J., Gong, J., Ding, Y., Lu, B., Wu, X., and Zhang, Y.** (2010). Antibacterial activity of water-phase extracts from bamboo shavings against food spoilage microorganisms. *African Journal of Biotechnology*. 9(45): 7710-7717.