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Investigation of Antioxidant Compositions and Antioxidative Activities of Ethanol Extract of *Alstonia boonei* Stem Bark

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A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of article.

Abstract

Background: Oxidative stress is the major mechanism of action employed by many drugs, xenobiotics and toxicants that generate reactive metabolites in the body to elicit adverse reactions when there are insufficient antioxidants to neutralise the excess reactive metabolites.

Objectives: This study evaluated antioxidant compositions and antioxidative activities of ethanol extract of *Alstonia boonei* stem bark with the view of understanding its ameliorative potentials on oxidative stress. **Materials and Methods:** Thirty-six rats divided into 6 groups of 6 rats each was used for this study. Group 1 served as normal control, group 2-6 were carbon tetrachloride-induced oxidative stress which served as treatment groups. Group 2 was untreated, group 3 was treated with silymarin (100 mg/kg) while group 4-6 were treated with the extract 100, 200 and 500 mg/kg body-weight for 28 days respectively. The animal study and antioxidant compositions of the extract were analysed using standard methods.

Results: The extract was found to rich in antioxidant components such as phenols, flavonoids, β-carotene and lycopene. The extract showed high DPPH (2, 2-diphenyl-1-picrylhydrazyl) and nitric oxide radical scavenging activities and high ferric reducing antioxidant power with increasing concentrations. The significant ($P < 0.05$) reduction in antioxidant enzymes activities and increased level of lipid peroxidation caused by the carbon tetrachloride induction were significantly ($P < 0.05$) reversed, in the extract-treated groups relative to the normal control. **Conclusions:** These findings suggest that the extract possesses antioxidative properties and can be used in the management of oxidative stress and its related disease conditions.

Keywords: Antioxidant components, *Alstonia boonei* stem bark, Antioxidative stress, Lipid peroxidation, Antioxidant enzymes.

INTRODUCTION

Oxidative stress occurs when there are excess amounts of free radicals or reactive oxygen species in the body and insufficient antioxidants defence system to attenuate the damaging effects of the free radicals on tissues, membranes, organs and vital macromolecules such as lipids, proteins and macromolecules (Uroko *et al.*, 2019a). Free radicals are normal by-products of aerobic metabolic processes in living organisms. They are required in small concentrations as part of the innate immune system but their excessive production in the body could lead to many pathological conditions including diabetes, atherosclerosis, hepatic and kidney

injuries, and cancers such as prostate and colon cancers (Seifried *et al*., 2007; Moshaie-Nezhad *et al*., 2018, Uroko *et al*., 2019b). In the body, endogenous and exogenous antioxidants including non-enzymatic (flavonoids, phenols, ascorbic acids, lycopene, β-carotene, and vitamin "E" and enzymatic antioxidants such as catalase, superoxide dismutase and glutathione peroxidase counter the actions of free radicals and protect the body from adverse effects of oxidative stress (Chand *et al*., 2018).

Carbon tetrachloride (CCl_4) is one of the chemical toxicants commonly used for induction and investigation of oxidative stress in the animal model. In the liver, carbon tetrachloride is metabolised to highly reactive trichloromethyl and trichloromethyl peroxyl radicals by cytochrome P450 enzymes. The reactive species could effectively attack functional biomolecules like nucleic acids, proteins and lipids thereby damaging them and besides cause lipid peroxidation (Manibusan *et al*., 2007). The peroxyl radicals generated from CCl_4 breakdown has been reported to inactivate and deplete endogenous antioxidant enzymes, thus making it inevitable for the supply of exogenous antioxidants to ameliorate adverse effects of free radicals (Cordero-Pérez *et al*., 2013).

Alstonia boonei is a medicinal plant of West African origin which belong to the family *Apocynaceae* and has spread almost all over the tropical rain forests in Africa and beyond. *A. boonei* extracts possess a wide range of medicinal properties including antimalarial, antipyretic, analgesic, anti-inflammatory, anthelmintic, diuretic, antipsychotic and antifertility effects (Fabiyi *et al*., 2012; Adotey *et al*., 2012). Report from previous studies shows that its stem bark extract has been used to treat fever, painful micturition, and chronic diarrhoea, rheumatic pains and as anti-snake venom (Adotey *et al*., 2012). In Nigeria, *A. boonie* is commonly called "Egbu-ora" by the Igbos, "Awun" by the Yorubas (Osadebe, 2002). It has been reported that *A. boonei* stem bark extract is rich in mineral elements and phytochemicals like alkaloids, tannins, saponin, flavonoids and ascorbic acid (Osadebe, 2002). However, there is no sufficient scientific evaluation of antioxidant components and antioxidant activities of the stem bark extract of *A. boonei* and its possible ameliorative effects on oxidative stress. This study was designed to evaluate antioxidant compositions and antioxidant activities of ethanol extract of *Alstonia boonei* stem bark with the view of understanding its ameliorative potentials on carbon tetrachloride-induced oxidative stress in rats.

METHODOLOGY

Collection and preparation of plant material

The Fresh stem bark of *A. boonei* was collected from Abia State University, Law campus Umuahia, Abia State – Nigeria. It was identified and authenticated by Dr Ibe K. Ndukwe of the Department of Forestry and Environmental Management, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. The stem bark of *A. boonei* was hand-picked, washed under clean running water to remove dirt and debris, it was then sliced into smaller pieces and dried under shade at room temperature for 4 Weeks. On drying, the sample was pulverized into a coarse powder, weighed and stored in a dry, clean, sterile container ready for extraction.

Extraction of *A. boonei* stem bark

Five hundred gram (500 g) of the coarsely ground *A. boonei* stem bark was macerated in 1.5 L of absolute ethanol in a clean, sterile and air-tight container for 72 h. It was filtered with Whatman no 1 filter paper and concentrated on a water bath at a temperature of 50^{o}C till all the ethanol added evaporated completely. The concentrated extract was weighed and its percentage yield calculated. The extract was then stored in a desiccator for use in the experiment.

Experimental animals

Male Wistar albino rats (36) weighing 100 – 120 g were purchased from the Animal House, Department of Zoology and Environmental Sciences, Faculty of Biological Sciences, University of Nigeria, Nsukka, Nigeria. The rats were placed on a standard laboratory diet and water *ad libitum* and acclimatised to laboratory condition under a 12 h light-dark for 7 days before the commencement of the experiment fully.

Chemicals and reagents

All the chemicals and reagents used for this study were of analytical grade and were sourced from JHD (Gunsgdong Guandghua Chemicals, China), Sigma Aldrich (Germany) and RANDOX Laboratories RandoxR Ltd (Ardmore, United Kingdom).

Induction of oxidative stress in a rat model using carbon tetrachloride

Oxidative stress was induced in the rats by intraperitoneal injection of carbon tetrachloride in olive oil (2:1) 2 mL/kg body weight on day 1, 7, 14 and 28 of the experiment which was later confirmed by the level of lipid peroxidation in the rats.

Experimental design

The rats were divided into six groups randomly (6 rats per group). Group 1 served as normal control, group 2 served as CCl_4 control, group 3 served as silymarin control, group 4, 5 and 6 were treated with low, moderate and high doses (100, 200 and 500 mg/kg body weight) of the ethanol extract of *A. boonei* stem bark by oral administration for 28 days. Group 1 are normal rats without CCl_4 induction, group 2 are CCl_4 induced untreated, group 3 were CCl_4 induced treated with silymarin (100 mg/kg body weight) while group 4, 5 and 5 were CCl_4 treated with low, moderate and high doses of ethanol extract of *A. boonei* stem bark for 28 days, respectively. On day 1, 7, 14 and 28 of the experiment, treatment was given to the rats 1 h after CCl_4 induction. After the last administration of CCl_4 and treatment of the 28th day, the rats fasted overnight. Blood samples were collected and the rats were sacrificed with the aid of chloroform.

Determination of antioxidant components in ethanol extract of *A. boonei* stem bark

The β-carotene and lycopene were determined according to the methods of Nagata and Yamashita (1992). Phenolic in the extract was quantified according to the method of Velioglu *et al*., (1998). Flavonoid content in the extract was determined according to the method of Harborne (1998). Ascorbic acid was determined according to the method of Klein and Perry (1982).

***In vitro* antioxidant assay of ethanol extract of *A. boonei* stem bark**

Ferric reducing antioxidant power was measured as described by Oyaizu (1986). The nitric oxide scavenging activity of the ethanol extract of *A. boonei* stem bark was determined according to the method described by Marcocci *et al*. (1994). The ability of ethanol extract of *A. boonei* stem bark to scavenge stable DPPH (2, 2-diphenyl-1-picrylhydrazyl) free radical was evaluated as described by the method of Hatano *et al*. (1988).

RESULTS

The percentage yield obtained from the extraction of 500 g of a coarsely ground sample of *A. boonei* stem bark was found to be 6.82 % equivalent to 34.1 g of the extract. This showed that *A. boonei* stem barks are rich in polar phytoconstituents which are extractable with ethanol to a larger extent.

The results of antioxidants composition of ethanol extract of *A. boonei* stem bark indicate a high content of antioxidant components with phenols (as determined by total phenols) and lycopene as the most abundant and least abundant antioxidant components in the extract, respectively (Table 1).

Assay of glutathione peroxidase (GPx) activities

Glutathione peroxidase activities were assayed as outlined in the method of Ursini *et al*. (1985). **T**his assay is based on the ability of glutathione peroxidase to reduce oxidized glutathione to its reduced form with concomitant oxidization of NADPH to $NADP^+$ thereby causing a decrease in the absorption capacity of NADPH at 340 nm. Reduction of NADPH absorbance at 340 nm is proportional to glutathione peroxidase activity.

Assay of superoxide dismutase (SOD) activities

Superoxide dismutase (SOD) activities were assayed according to the method of Xin *et al*. (1991). In this assay, superoxide dismutase inhibits autoxidation of adrenaline. Thus, superoxide by the action of xanthine oxidase causes oxidation of adrenaline to adrenochrome and the concentration of adrenochrome generated per superoxide introduced increases with pH and with increasing concentration of adrenaline which is proportional to superoxide activity.

Assay of catalase (CAT) activities

Catalase activity was assayed according to the method described by Aebi, (1983). In this method, catalase catalyses the breakdown of hydrogen peroxide to water and oxygen, leading to a decrease in absorption of hydrogen peroxide at 240 nm which is proportional to catalase activity.

Determination of malondialdehyde (MDA) concentrations

The concentration of malondialdehyde (MDA), a lipid peroxidation product was determined spectrophotometrically using the method of Wallin *et al*. (1993). In this method, thiobarbituric acid reacts with malondialdehyde produced from lipid peroxidation given a red or pink colour with maximum absorption at 532 nm. The absorption observed at 532 nm is proportional to the concentration of malondialdehyde liberated from lipid peroxidation.

Table 1: Antioxidant components present in ethanol extract of *A. boonei* stem bark

Antioxidant components	*A. boonei* stem bark
Total phenols (mg/g)	282.20±3.920
Flavonoid (mg/100g)	414.58±2.300
β - carotene (mg/100g)	0.92±0.006
Lycopene (mg/100g)	0.25±0.001
Vitamin C (mg/g)	2.50±0.920

Values are presented as mean ± standard deviation (n = 3)

The results in Figure 1 show nitric oxide scavenging activities of ethanol extract of *A. boonei* and cucumin (standard control) which indicated a dose-dependent increase in both nitric oxide scavenging activities of the extract and cucumin, respectively. The extract and cucumin had the highest percentage nitric oxide inhibition of 64% and 82.6 % respectively at 100 µg/mL concentration. The EC_{50} obtained for the ethanol extract was 83.6µg/mL was higher than the EC_{50} of 56 µg/mL obtained for cucumin which is a known antioxidant.

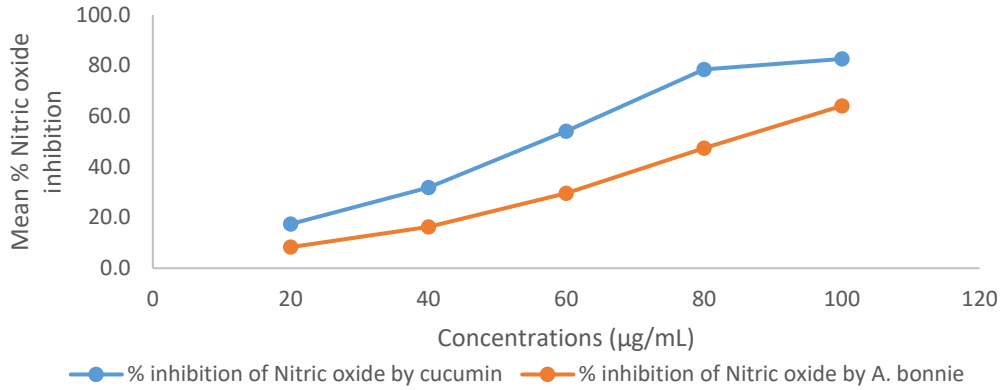


Fig. 1: Nitric oxide scavenging activities of ethanol extract of *A. boonei* stem bark relative to cucumin

The data in Figure 2 show DPPH free radical scavenging activities of ethanol extract of *A. boonei* stem bark and vitamin C. From the results, it was observed that the extract possesses DPPH free radical scavenging activity which increases with the increasing concentrations of the extract similar to the DPPH free radicals scavenging activity exhibited by vitamin C, a standard antioxidant. The extract exhibited higher DPPH free radical scavenging activity at lower concentration (0.02 mg/mL) than vitamin C but at 0.03 mg/mL, both the extract and vitamin C had equal DPPH free radical scavenging activities. However, at increase concentration above 0.03 mg/mL, vitamin C had higher DPPH free radical scavenging activity than the ethanol extract of *A. boonei* stem bark. At a concentration of 0.02 mg/mL, the ethanol extract of *A. boonei* stem bark and vitamin C exhibited the least percentage DPPH inhibition of 35.9 and 44.3 %, respectively while at 0.1 mg/mL concentration the ethanol extract and vitamin C exhibited the highest percentage DPPH inhibition of 60.5 and 89.2 %, respectively.

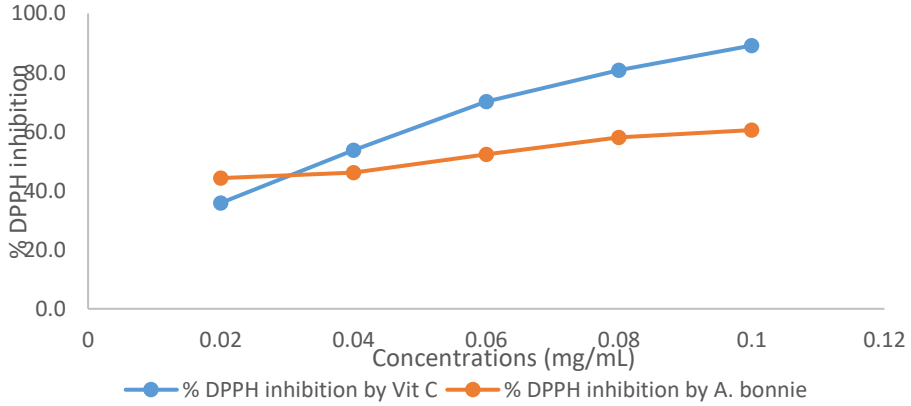


Fig. 2: Percentage DPPH free radical scavenging activities of ethanol extract of *A. boonei* stem bark relative to vitamin C

The results in Figure 3 show ferric reducing antioxidant power (FRAP) of ethanol extract of *A. boonei* stem bark relative to antioxidant activities of vitamin C and rutin (standard controls) which are potent antioxidants. The extract exhibited high ferric reducing antioxidant power been much higher with increasing concentrations of the ethanol extract of *A. boonei* though lower than ferric reducing antioxidant power observed for vitamin C and rutin. Ferric reducing antioxidant power of vitamin C was generally higher than that observed for rutin and the ethanol extract, respectively.

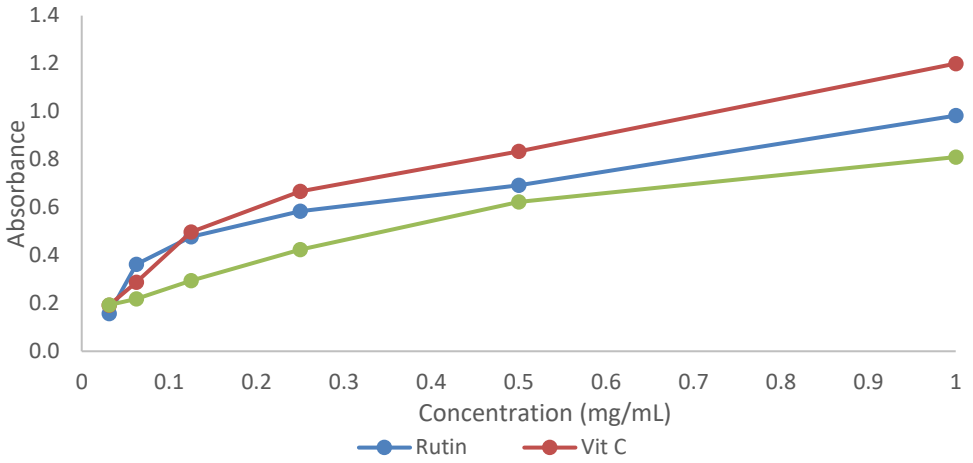
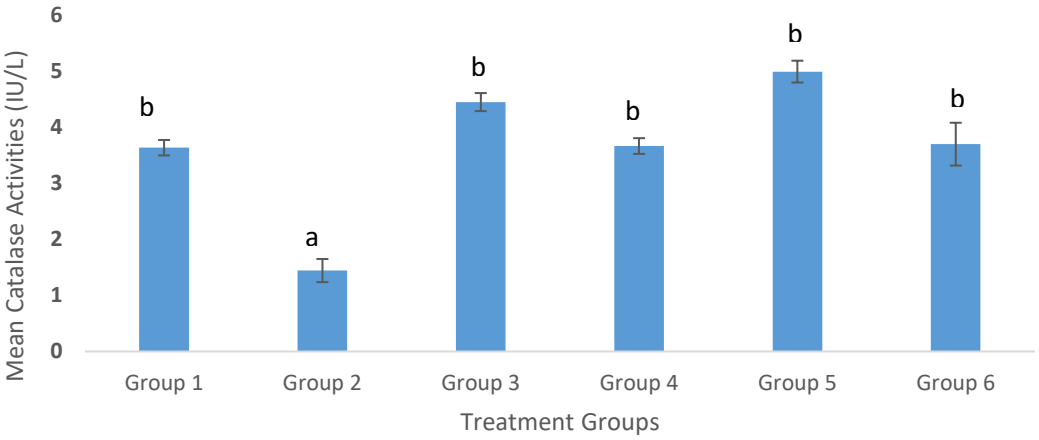


Fig. 3: Ferric Reducing Antioxidant Power (FRAP) of ethanol extract of *A. boonei* stem bark relative to vitamin C and Rutin

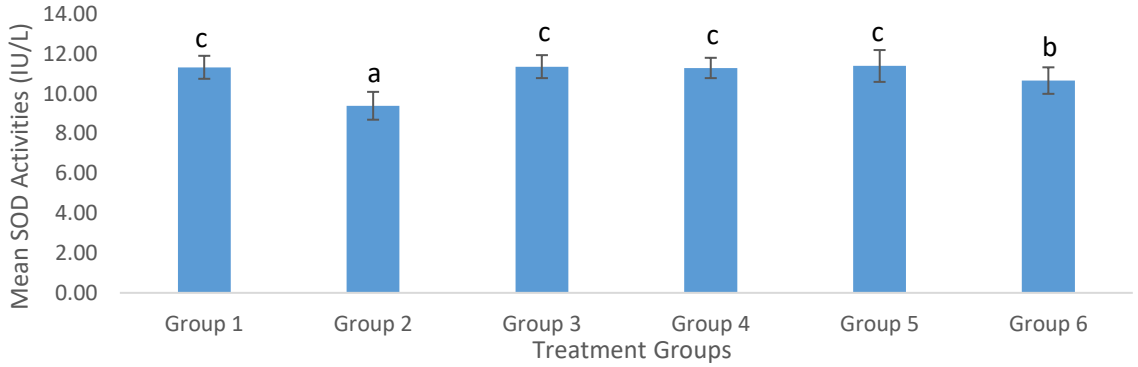
The data in Figure 4 show the effects of ethanol extract of *A. boonei* stem bark on the catalase activities of carbon tetrachloride-induced oxidative stress in rats. It was observed that group 2 rats that were carbon tetrachloride-induced without treatment showed significant ($P < 0.05$) decrease in catalase activities when compared with other carbon tetrachloride-induced rats treated with either silymarin or ethanol extract of *A. boonei*. The catalase activities observed in group 1, group 3 – 6 were significantly ($P < 0.05$) higher than the catalase activities of the negative control (group 2) that were CCl_4 induced. The normal control rats (group 1) and group 4 – 6 that were induced and treated with graded doses of the extract showed no significant ($P > 0.05$) difference in their catalase activities relative to group 3 that was CCl_4 induced but treated silymarin.



Each bar represent mean ± standard deviation (n = 6)
Bars withdifferent superscripts are significantly different (P < 0.05)

Fig. 4: Catalase activities of rats induced oxidative stress with carbon tetrachloride treated with ethanol extract of *A. boonei* stem bark

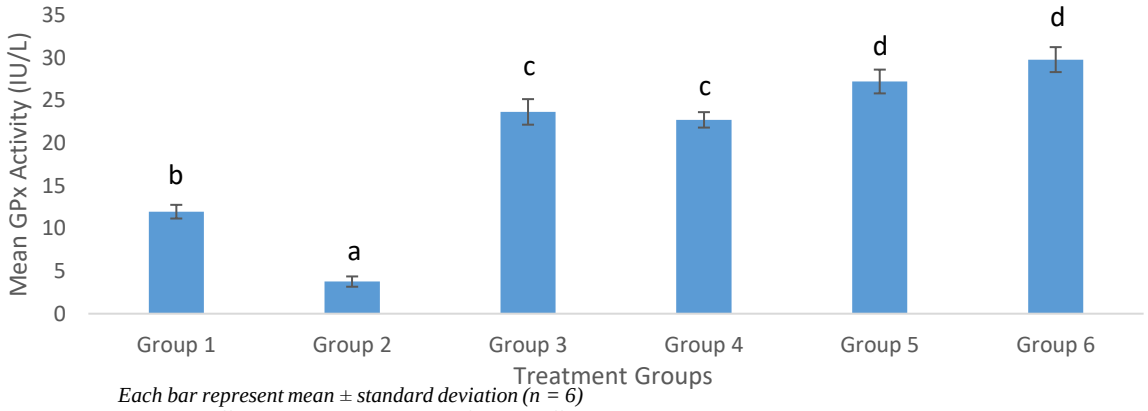
The superoxide dismutase (SOD) activities observed in rats induced oxidative stress with carbon tetrachloride show that the negative control (group 2) had significantly ($P < 0.05$) lower SOD activities when compared with the normal control (group 1), silymarin treated group 3 and ethanol extract of *A. boonei* treated groups, respectively (Figure 5). There was no significant ($P > 0.05$) difference observed in SOD activities of the normal control, positive control (group 3), group 4 and 5 treated with 100 and 200 mg/kg body weight of the ethanol extract, respectively. However, group 6 treated with 500 mg/kg body weight of the extract had SOD activities significantly ($P < 0.05$) higher than the negative control but significantly ($P < 0.05$) low when compared with the normal control and groups treated with silymarin and ethanol extract, respectively.



Each bar represent mean ± standard deviation (n = 6)
Bars withdifferent superscripts are significantly different (P < 0.05)

Fig. 5: Superoxide dismutase (SOD) activities of rats induced oxidative stress with carbon tetrachloride treated with ethanol extract of *A. boonei* stem bark

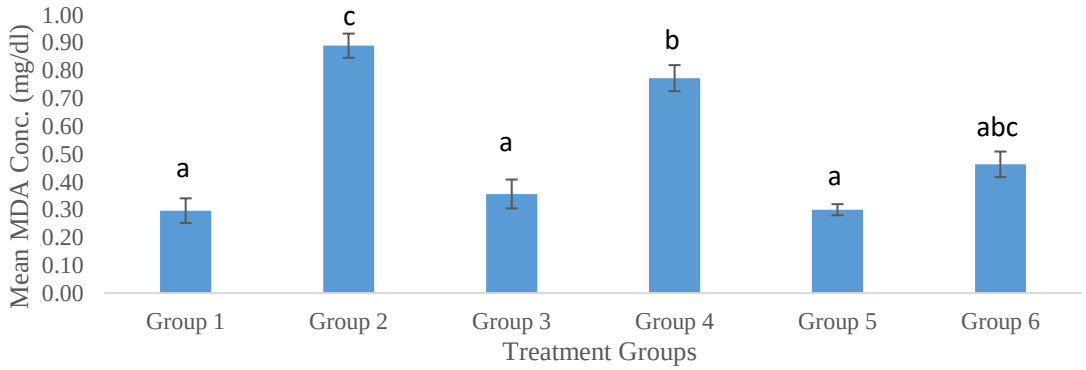
The results in Figure 6 show glutathione peroxidase (GPx) activities of rats induced oxidative stress with carbon tetrachloride which indicates that group 2 (negative control) rats that were oxidative stress-induced untreated had significantly (P < 0.05) lower GPx activities when compared with the normal control (group 1). Group 3 (positive control) rats that were oxidative stress-induced, treated with 100 silymarin and group 4, 5, and 6 rats that were oxidative stress-induced, treated with 100, 200, and 500 mg/kg body weight of ethanol extract, respectively showed significant (P < 0.05) increase in GPx activities relative to group 1 and 2 respectively. Group 5 and 6 rats treated with moderate and high doses of ethanol extract exhibited significant (P < 0.05) increase in GPx activities when compared with the positive control treated with 100 mg/kg body weight of silymarin.



Each bar represent mean ± standard deviation (n = 6)
Bars withdifferent superscripts are significantly different (P < 0.05)

Fig. 6: Glutathione peroxidase (GPx) activities of rats induced oxidative stress with carbon tetrachloride treated with ethanol extract of *A. boonei* stem bark

The data in Figure 7 show the malondialdehyde (MDA) concentrations in rats that were induced oxidative stress with carbon tetrachloride. From the figure, it was observed that group 2 rats induced oxidative stress untreated and group 4 that were oxidative stress-induced, treated with 100 mg/kg body weight of ethanol extract had significant (P < 0.05) increase in MDA concentration when compared with the normal control (group 1). Group 3 rats treated with 100 mg/kg body of silymarin, group 5 and 6 rats treated moderate and high doses of ethanol extract, respectively showed no significant (P > 0.05) increase in MDA concentrations relative to the normal control and significantly (P <0.05) lower than the MDA concentration of the negative control (group 2).



Each bar represent mean ± standard deviation (n = 6)
Bars withdifferent superscripts are significantly different (P < 0.05)

Fig. 7: Malondialdehyde (MDA) concentrations of rats induced oxidative stress with carbon tetrachloride treated with ethanol extract of *A. boonei* stem bark

DISCUSSION

This study evaluated antioxidant compositions and antioxidative activities of ethanol extract of *Alstonia boonei* stem bark with the view of understanding its ameliorative potentials on carbon tetrachloride-induced oxidative stress in rats. The high antioxidant components detected in the ethanol extract could be attributed to its richness in phytochemicals with well-established antioxidant activities. The antioxidant constituents in the extract indicate that it could effectively ameliorate excess free radicals generated in the body under oxidative stress when administered in right concentrations. The high levels of total phenols and vitamin C in the extract suggest that they are the major antioxidant components in the extract and could be responsible for its antioxidant activities. Vitamin C, also known as ascorbic acid is a potent antioxidant, soluble in an aqueous medium that synergies with vitamin E and most antioxidant components to scavenge free radicals generated from both endogenous and exogenous sources to prevent damage to proteins, DNA and vital organs in the body. In synergy with vitamin E, vitamin C help to regenerate α-tocopherol and maintain substantial amounts of antioxidants that play a vital role in protection against oxidative stress in the body (Kojo, 2004; Naziroglu and Butterworth, 2005). Low level of vitamin C in the body could accelerate the rate of oxidative stress as this result to decrease in free radical scavenging capacity, α-tocopherol regeneration and disruption of iron metabolism. Antioxidant compounds including phenols and flavonoids can chelate free radicals such as hydroxyl groups and metal ions which could prevent negative effects associated with the free radical attack on vital biomolecules and thus, promote good health (Kumar *et al.*, 2013). The high antioxidant compounds detected in this extract are in agreement with the findings of Baiano and Del (2016) who had obtained similar results in vegetables.

The dose-dependent increase in the *in vitro* antioxidant activities of ethanol extract as demonstrated by percentage nitric oxide and DPPH inhibition and ferric reducing antioxidant power observed in this study could be attributed to antioxidant components present in the extract. It further indicated that the extract possesses high antioxidant activities at high concentration than lower concentrations which are comparable to vitamin C, rutin and cucumin which are well-known antioxidant compounds. Plant extracts rich in antioxidant components such as phenols, flavonoids, β-carotene, lycopene and vitamin C possess antioxidant activities and are capable of donating a hydrogen atom to reduce DPPH radical as demonstrated by this study and are in line with findings of Rajesh and Natvar (2011). The results of this study are in agreement with the previous findings by Daffodil *et al*. (2012) that plant extracts antioxidant components like phenolics, vitamin C and flavonoids are capable of scavenging DPPH radical due to their high redox potentials and available free hydroxyl group (OH), that donates hydrogen and remove the extra electron. The high nitric oxide scavenging activities exhibited by the extract in a dose-dependent manner could be attributed to the ability of its antioxidant components in reducing the amounts of nitrite ions produced in the body. The strong antioxidant activities of ethanol extract demonstrated by its ability to scavenge DPPH and NO· radicals to a larger extent in line with the findings of Omowumi *et al*. (2017) who had reported that its aqueous leaves extract exhibited high DPPH, H_2O_2 and NO radical scavenging activities. This indicates that its polar constituents are responsible for its observed antioxidant activities. Most plant extracts rich in antioxidant components contribute immensely to the

reduction of oxidative stress through their free radicals scavenging activities and stabilization or maintenance of the concentrations of circulating endogenous antioxidant enzymes. They are capable of repairing or destroying damaged biomolecules due to negative effects of oxidative stress. The antioxidant components present in the ethanol extract of *A. boonei* stem bark are responsible its antioxidant activities which are in agreement with the findings of Rozina *et al*. (2013) who reported that antioxidant constituent in their plant extracts depleted oxygen level that would have reacted with nitric oxide and thus prevent further production of nitrite.

The oxidative stress associated with carbon tetrachloride administration is primarily due to free radical activity of trichloromethyl radical (CCl_3·) generated from its metabolic breakdown (Ohata *et al.*, 2008; Uroko *et al*., 2019). Oxidative stress is capable of depleting circulating antioxidant enzymes, inactivation, shorten their lifetime, accelerate lipid peroxidation and many vital biomolecules are vulnerable to oxidative stress resulting from excess free radicals in the body. In the body, both non-enzymatic and enzymatic antioxidant defence systems work synergistically to protect the body oxidative damage from the excess free radical attack. The significant reduction in the enzymatic antioxidants (catalase, superoxide dismutase and glutathione peroxidase) activities of the negative control (group 2) rats that were carbon tetrachloride-induced untreated could be attributed to oxidative stress resulting from excess trichloromethyl radical (CCl_3·) generated from its metabolism. These antioxidant enzymes are proteins and gene products of their respective genes (DNA) which are also vulnerable to oxidative attack when excess free radicals generated in the body overwhelmed the action of antioxidant defence systems in the body. The administration of carbon tetrachloride caused a sudden rise in the free radical attack on the biomolecules resulted in the rapid depletion of the antioxidant enzymes than they are synthesized and the inevitable decrease in their concentrations and activities. Thus, genes coding for these antioxidant enzymes in the untreated groups may have suffered oxidative damage and were unable to code for their respective antioxidant enzymes to a greater extent.

The significant increase in the activities of catalase, superoxide dismutase and glutathione peroxidase enzymes in groups treated with the ethanol extract of *A. booeni* stem bark indicated the recovery of the rats from oxidative stress caused by the CCl_4 induction and could be attributed to the antioxidant activities exhibited by the extract. The ethanol extract of *A. booeni* stem bark possesses potent antioxidant activities comparable with silymarin that served as standard control possibly due to effects of phenols, flavonoids, vitamin C, β-carotene and lycopene with proven antioxidant activities detected in it. The ethanol extract could have caused an increase in the catalase, superoxide dismutase and glutathione peroxide activities by stabilization of the circulating concentrations in blood system mediated by the antioxidant activities of the non-enzymatic antioxidants in the extract. The extract could have also increased activities of these enzymes in the extract-treated groups via induction of genes coding for each of these enzymes leading to their increased expression. The *in vivo* and *in vitro* antioxidant activities demonstrated by this ethanol extract of *A. booeni* stem bark suggest that the plant extract could play a vital role in ameliorating oxidative stress, health conditions and adverse health conditions associated with it when administered adequately in the right concentration.

Furthermore, rats treated with moderate and high dose ethanol extract of *A. booeni* stem bark respectively had significantly lower malondialdehyde (MDA) concentrations as the normal control rats. These observations suggest that the ethanol extract of *A. booeni* stem bark was effective in preventing lipid peroxidation in rats due to its antioxidants and free radical scavenging activities. It showed that the antioxidant activities of the ethanol extract of *A. booeni* stem bark are better demonstrated when administered in moderate or high concentrations rather than a low concentration to protect biomolecules like enzymes, proteins, DNA lipid or cell membranes from oxidative damage and disrupt normal cell functions. The antioxidant components in this extract could have efficiently interacted with free radicals generated by CCl_4 induction (trichloromethyl and trichloromethyl peroxyl radicals) decreased their production, induced their rapid removal, and prevented them from attacking and destroying functionally important biomolecules. However, the high level of MDA observed in the CCl_4 induced untreated negative control (group 2) could be attributed to the effects of oxidative stress on biomolecules as there were insufficient antioxidants which could have reacted with radicals generated from CCl_4 metabolism. Thus, most vital organs, proteins, DNA, lipids and cell membranes could have suffered extensive oxidative as demonstrated by elevated levels of lipid peroxidation product (MDA) due to insufficient antioxidant defence system to scavenge free radical and prevent lipid peroxidation. The low level of MDA concentrations observed in this study may be attributed to the high content of lycopene, vitamin C and antioxidants components in the extract which are in line with previous findings that dietary ingestion of sufficient amounts lycopene reduces lipid peroxidation and its associated adverse health effects (Uroko *et al*., 2019a; Gary *et al*., 2001)

CONCLUSION

The findings of this study show that ethanol extract of *A. boonei* stem bark is rich in antioxidant components and possesses sufficient antioxidative activities capable of preventing and ameliorating oxidative stress and its associated health consequences.

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