Electrophilic, Free Radical and Reactive Oxygen Species Scavenging and Detoxification Potentials of *Lophiraalata* Stem Bark Extract

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ABSTRACT

The electrophilic, free radical and reactive oxygen species scavenging and detoxification potentials of *Lophiraalata* stem bark was evaluated. *L. alata* stem bark effectively scavenged DPPH radical, superoxideion and hydrogen peroxide. It produced 88% scavenging effect of DPPH radical at a concentration of 1.0 mg/ml. Aqueous extract of *L. alata*-stem bark produced 76% and 92% scavenging effect on superoxide ion and hydrogen peroxide respectively at 1.0 mg/ml, which compared favourably with the synthetic antioxidant (butylated hydroanisole and α-tocopherol). A reducing power of *L. alata* stem bark was examined using $K_3Fe(CN)_6$, 2-folds reducing power potentials was exhibited by *L. alata* stem bark when compared with the synthetic antioxidant,butylated hydroanisole. Reactive oxygen species detoxify enzymes; superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase (GRed) were significantly induced by 90, 133, 90 and 172% respectively. While the electrophilic detoxifying enzymes NADPH: quinone oxidoreductase-1 (NQO1), uridyl diphosphoglucuronosyl transferase (UGT), glutathione S- transferase (GST) and epoxide hydrolase (EPh)) were induced by 240, 81, 196 and 281% respectively at the end of the experimental period. In view of these properties, *L. alata* can act as a prophylactic by intervening as electrophilic, free radical and ROS scavenger and detoxifier.

Keywords: *Lophiraalata,* reactive oxygen species, free radicals and electrophile detoxifying enzymes

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INTRODUCTION

Free radicals, specifically reactive oxygen species (ROS), are formed by several different mechanisms. ROS are oxygen ions [singlet oxygen, superoxide ($O_2\cdot$)] or oxygen-containing radicals [hydroxyl, ($OH\cdot$)]. They are generated in response to both endogenous and exogenous stimuli.[1,2,3] ROS and their reaction products [*e.g.* hydrogen peroxide (H_2O_2)] are increasingly recognized as signaling intermediates contributing to adaptive or maladaptive molecular responses.[4] Under physiological concentrations, ROS act as signaling molecules mediating cell growth, migration and differentiation,[5]whereas at higher concentrations, they induce cell death, apoptosis and senescence.[5] Accumulative ROS production is suggested to stimulate oncogenesis via alterations in redox regulated signaling pathways suggesting that the redox state plays a critical role in signal transduction, cellular proliferation, differentiation and apoptosis.[5,6]

Multiple pathways involved in ROS-induced cell death have been proposed. ROS can cause direct injury to proteins, lipids, and nucleic acids, leading to cell death. For example, protein oxidation and nitrosylation (carbonyl, nitration, and nitrotyrosine formation) can impair a wide variety of enzymatic processes and growth factors that can result in marked cellular dysfunction.[7] Lipid peroxidation has been linked to cell death through effects on cellular phospholipids (major cell membrane components) through activation of sphingomyelinase and release of ceramide, which activates apoptosis.[8] Nucleic acid oxidation has been linked with physiologic and premature aging as well as DNA strand breaks, leading to necrosis and/or maladaptive apoptosis.[9] The magnitude of these changes and the cell's ability to repair this damage determines whether the effects

are adaptive or maladaptive. Chemical compounds known to have free radical scavenging properties could effectively protect against this damage only upon their absorption.

Lophiraalata banks ex Gaertn (Ochnaceae) grows as tall trees in the tropical rainforest of Africa. Its stem bark is exploited for medicinal uses and its heartwood is solid as first-grade timber.[10] The local names of the plant are Aba, Akufo (Igbo); Eba (Edo); Ekki (Yoruba); Eleba (Itsekiri); Enwan (Efik); Kabaniko (Boki); Namijin kadai (Hausa); Kuru (Ijaw); Oyben ren-ren (Urhobo). Its medicinal usage includes; anti-inflammatory, antimicrobial and analgesic activity.[11] The stem bark is rich in chalcones of varying structures ranging from mono – hexa-chalcones. The isolated chalcones include; lophirone A-H, bongosin, mbamichalcone, tetraflavonoids among others.[10] In addition, cyanogenic glycoside has been isolated from the stem bark.[12]

Despite the role of chalcone in the amelioration of reactive and oxidative stress, associated carcinogenesis and other related disease; there is no information in the scientific literature that addresses the role of *Lophiraalata* stem bark on free radicals, ROS and electrophilic detoxification. This study thus investigates reactive oxygen species, electrophilic and free radical scavenging and detoxification potentials of *Lophiraalata* stem bark extract.

MATERIALS AND METHODS

Chemicals

Butylated hydroanisole (BHA), hydrogen peroxide, nitroblue tetrazolium (NBT), α, α-diphenyl-β-picrylhydrazyl (DPPH), α- tocopherol, Glutathione disulphide, ephinephrine, glucose-6- phosphate, 1-Chloro-2,4-Dinitrobenzene, Glutathione (Reduced Form), 2,6-Dichlorophenol-Indophenol, β-NADH, β-NAD and Uridine 5'-Diphosphoglucuronic Acid were purchased from Sigma Chemical Co., St. Loius, MO. Glutathione peroxidase assay kit was pocurred from Randox. All other reagents were of analytical grade.

Plant materials

Mallam Muazzam collected *Lophiraalata* stem bark in chaza village, Suleja, Niger State (date: 12-01-10; time: 5.45 pm). The plant sample was authenticated and deposited in the Herbarium unit of Medicinal Plant Research Department, National Institute of pharmaceutical Research and Development, Idu-Abuja, Nigeria with voucher No: 4534.

METHODS

Plant extraction

Plant materials, were chopped and shade dried. The dried plant material was thereafter weighed, extracted in methanol for 96-hrs and concentrated to give 50.03 g residue.

Laboratory animal

Apparently healthy, 3 months old, male albino rats (*Rattus novergicus*) of Wistar strain, weighing 145 ± 2.52 g were obtained from the Animal Holding Unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. They were kept in clean metabolic cages of dimensions 33.0 × 20.5 × 19.0 contained in well-ventilated house conditions (temperature: 28-31 °C; photoperiod: 12 h natural light and 12h dark; humidity: 50-55%) with free access to rat pellets (Bendel Feeds and Flour Mills Ltd., Ewu, Nigeria) and tap water. The experimental protocols were approved by the Ethical Committee on the use and care of animals of the Department of Biochemistry, University of Ilorin, Nigeria. The animals were used according to the NIH Guide for the Care and Use of Laboratory Animals, [13]in accordance with the principles of Good Laboratory Procedure (GLP).[14]

In vitro *free radical and reactive oxygen species scavenging assay*

Scavenging of DPPH radical

The effect of *Lophiraalata* stem bark extract on DPPH radical was estimated as described byYen and Duh.[15] *Lophiraalata* stem bark extract (concentration ranging 0.2 mg/ml – 2.0 mg/ml) was added to 1ml 20 mM DPPH radical in a methanolic solution. The mixtures were shaken and left to stand for 30 min at room temperature. The absorbance of the resulting mixture was measured at 517 nm.

Scavenging of Superoxide ion

The scavenging effect of *Lophiraalata* stem bark extract on superoxide ion was examined by spectrophotometrical measurement of the product formed on reduction of nitro blue tetrazolium as described by Yen and Chen.[16] Briefly, superoxide ion was generated in a non-enzymic system. The reaction mixture contained 1ml of *Lophiraalata* stem bark extract (0.2-1.0 mg/ml) in distilled water, 1 ml of 60 μM PMS in phosphate buffer (0.1M, pH 7.4), 1 ml of 468 μM NADH in phosphate buffer and 1 ml of 150 μM NBT in phosphate buffer, was incubated at ambient temperature for 5 min, and the colour was read at 560 nm against blank samples.

Scavenging of Hydrogen Peroxide

The ability of *Lophiraalata* stem bark extract to scavenge hydrogen peroxide was determined according to the method of Ruch *et al*.[17] Briefly, 4 mM hydrogen peroxide was prepared in phosphate buffer saline (PBS), pH 7.4. *Lophiraalata* stem bark extract 0.2 – 1.0 mg/ml (final concentration) in 4 ml distilled water was added to 0.6 ml hydrogen peroxide solution. Absorbance of hydrogen peroxide at 230 nm was read 10 min later against a blank solution containing *Lophiraalata* stem bark extract without hydrogen peroxide.

Reducing Power

The reducing power of *Lophiraalata* stem bark extract was evaluated using the method of Oyaizu.[18] Varying amount of *Lophiraalata* stem bark extract were suspended in 1 ml of distilledd water and mixed with 2.5 µl of 0.2 M phosphate buffer, pH 6.6 and 2.5 ml of 1% $K_3Fe(CN)_6$. The mixture was incubated at 50 °C for 20 min, 2.5 µl of TCA were added to the mixture. Following centrifugation at 3000 rpm for 10 min, 2.5 µl of the supernatant were mixed with equal amount of distilled water and 0.5 ml of 0.1% $FeCl_3$. The absorbance of the resulting solution was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power.

In vivo *electrophilic and reactive oxygen species detoxification potential study*

Experimental design

Sixty animals were divided into four groups of 15 rats each. The first group received distilled water, while group 2, 3 and 4 received 100, 200 and 400 mg/kg body weight aqueous extract of *Lophiraalata* for 28 days respectively. Five rats were sacrificed 24-hrs after day-1, 14 and 28 days of administration from each treatment group.

Preparation of microsomes and cytosol

Liver was homogenized in sucrose–Tris buffer (0.25 mol/L sucrose, 10 mmol/L Tris–HCl, pH 7.4). Liver microsomes were isolated by the calcium chloride precipitation method.[19]

Enzyme assays

Superoxide dismutase (SOD) and catalase (CAT) were assayed as reported by Misra and Fridovich, and Beers and Sizer respectively.[20,21] GPx and glutathione reductase (GR) assays were also done according to Rotruck *et al* and Mavis and Stellwagen respectively.[22,23] Cytosolic GST and NQO1 activities were assayed according to Habig *et al* and Jaiswal *et al* respectively.[24,25] Microsomal UDPGT and Epoxide hydroxylase were assayed according to Bock *et al* and Depierre *et al* respectively.[26,27]

Statistical analysis

Analysis of variance (ANOVA) followed by Duncan's Multiple Range Test was used to detect any significant differences among different means as well as the interactions between the variables used in this study using StatPlus, 2011. Differences were considered statistically significant at $P < 0.05$.

RESULTS

In vitro *free radical and reactive oxygen species scavengiving study*

Effect of Lophiraalata stem bark extract *on DPPH radicals*

Figure 1 shows the scavenging effect of *Lophiraalata* stem bark extraction DPPH radical. The scavenging effect of *Lophiraalata* stem bark extract produced a marked scavenging effect on DPPH radical in a dose dependent response with the highest percentage of (88.1%) observed for the highest dose. While α-Tocopherol produced a scavenging effect which is almost as effective as *Lophiraalata* stem bark extract, the scavenging effect of BHA could not compare with them with its highest concencentration exhibiting only 73.01%.

Effect of Lophiraalata stem bark extract *on Superoxide ion*

Lophiraalata stem bark extract, BHA and α-tocopherol demonstrated the ability to scavenge superoxide ion

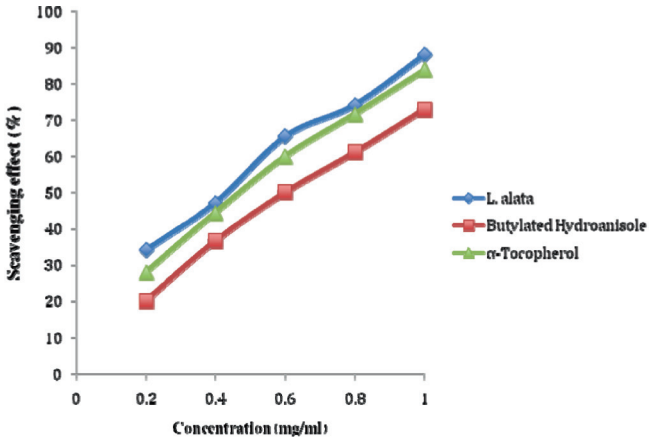

Figure 1. Scavenging effect of aqueous extract of *L. alata* stem bark on DPPH radical

in vitro (Figure 2). The *Lophiraalata* stem bark extract was the most effective of the three with its highest concentration demonstrating 76.1% scavenging effect, while α-tocopherol and BHA have their highest dose exhibiting 72.8 and 63.5% scavenging effect on superoxide ion.

Effect of Lophiraalata stem bark extract *on Hydrogen peroxide*

Hydrogen peroxide was effective scavenge by *Lophiraalata* stem bark extractexhibiting 91.8% scavenging effect at the highest dose used in this study (Figure 3). BHA and α-tocopherol also produce an effective (83.3 and 78.1% respectively) scavenging effect on hydrogen peroxide at the highest dose used in this study (Figure 3).

Reducing Power of Lophiraalata stem bark extract

The reducing power of *Lophiraalata* stem bark extract is shown in Figure 4. A dose dependent reducing power was shown by *Lophiraalata* stem bark extractwith about 3 fold greater than BHA at the higehst dose studied. All doses other than the highest dose of *Lophiraalata* stem bark extractexhibited a greater reducing power than α-tocopherol.

In vivo *electrophilic and reactive oxygen species detoxification potential study*

Antioxidant enzymes

Antioxidant enzymes were significantly ($P < 0.05$) increased following administration of *Lophiraalata* stem bark extract in a dose dependent manner (Tables 1, 2, 3 & 4). The highest dose used produced 89.84, 132.88, 90.00 and 172.35% increase in the liver; SOD, CAT, GPx and GR respectively at the end of the experimental period.

Phase II detoxification enzyme

While the administration of *L. alata* stem bark extract produced 195.55 and 240.89% increase in cytosolic GST

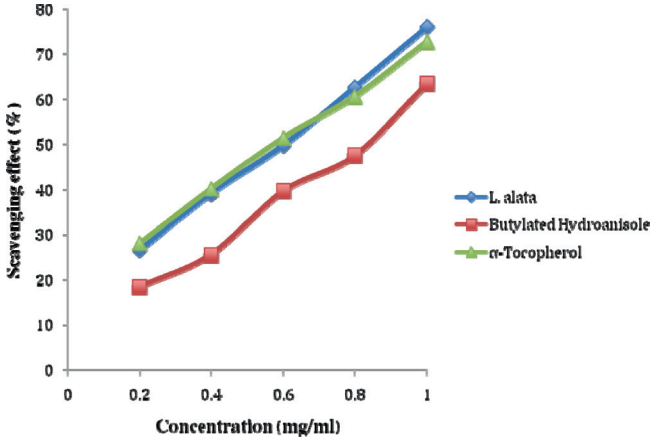

Figure 2. Scavenging of aqueous extract of *L. alata* stem bark on Superoxide ion

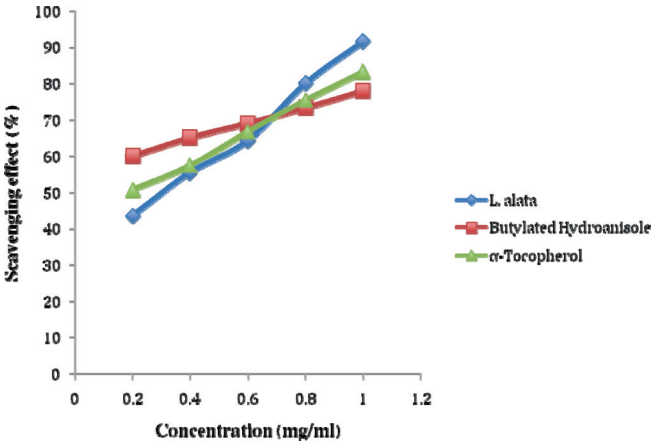

Figure 3. Scavenging effect of aqueous extract of *L. alata* stem bark on hydrogen peroxide

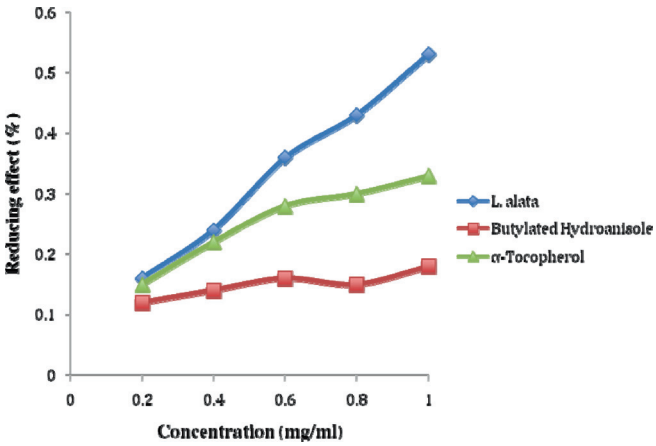

Figure 4. Reducing power of aqueous extract of *L. alata* stem bark on $K_3Fe(CN)_6$

Table 1. Effect of aqueous extract of *L. alata* stem bark on liver superoxide dismutase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	51.2 ± 2.84[a]	52.8 ± 0.87[a]	52.0 ± 1.24[b]
100	69.7 ± 1.05[b]	76.8 ± 4.10[c]	82.7 ± 3.02[d]
200	75.1 ± 1.18[c]	84.3 ± 2.20[d]	90.8 ± 3.16[f]
400	80.3 ± 2.55[d]	89.8 ± 2.36[f]	97.2 ± 2.10[f]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-f) different for each parameter are significantly different ($P < 0.05$).

Table 2. Effect of aqueous extract of *L. alata* stem bark on liver catalase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	28.1 ± 1.23[a]	30.2 ± 2.21[a]	29.2 ± 2.67[a]
100	36.1 ± 1.39[b]	43.2 ± 1.51[c]	52.0 ± 2.71[d]
200	41.5 ± 3.02[c]	53.9 ± 1.73[d]	60.2 ± 4.29[e]
400	48.3 ± 0.31[d]	58.9 ± 0.81[e]	66.9 ± 2.17[f]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-f) different for each parameter are significantly different ($P < 0.05$).

Table 3. Effect of aqueous extract of *L. alata* stem bark on liver glutathione peroxidase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	47.3 ± 1.28[a]	47.9 ± 2.52[a]	47.0 ± 0.13[a]
100	61.2 ± 4.78[b]	66.3 ± 0.11[c]	75.6 ± 4.78[d]
200	67.9 ± 0.32[c]	72.8 ± 5.48[c]	80.1 ± 2.13[d]
400	72.7 ± 2.60[c]	78.7 ± 3.62[d]	89.3 ± 1.05[e]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-e) different for each parameter are significantly different ($P < 0.05$).

Table 4. Effect of aqueous extract of *L. alata* stem bark on liver glutathione reductase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	1.70 ± 0.04[a]	1.72 ± 0.01[a]	1.69 ± 0.05[a]
100	2.13 ± 0.01[b]	2.70 ± 0.05[b]	4.09 ± 0.01[e]
200	2.72 ± 0.07[b]	3.20 ± 0.02[c]	4.18 ± 0.09[e]
400	3.12 ± 0.06[c]	3.66 ± 0.02[d]	4.63 ± 0.03[f]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-f) different for each parameter are significantly different ($P < 0.05$).

Table 5. Effect of aqueous extract of *L. alata* stem bark on liver NADPH:Quinone oxidoreductase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	50.41 ± 3.0[a]	53.12 ± 1.4[a]	51.90 ± 2.1[a]
100	72.63 ± 2.5[b]	95.26 ± 4.8[d]	152.02 ± 4.1[h]
200	85.26 ± 1.6[c]	113.51 ± 1.9[e]	163.58 ± 2.6[h]
400	94.55 ± 3.2[d]	129.82 ± 2.4[f]	171.19 ± 0.6[i]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-i) different for each parameter are significantly different ($P < 0.05$).

Table 6. Effect of aqueous extract of *L. alata* stem bark on liver uridyl diphosphoglucuronysl transferase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	121.5 ± 5.2[a]	122.5 ± 2.6[a]	120.3 ± 1.5[a]
100	138.2 ± 2.9[b]	158.9 ± 3.1[c]	195.5 ± 1.8[f]
200	151.8 ± 2.3[c]	173.5 ± 4.7[d]	209.3 ± 3.7[g]
400	170.1 ± 1.5[d]	191.3 ± 1.9[a]	219.5 ± 2.5[h]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-i) different for each parameter are significantly different ($P < 0.05$).

Table 7. Effect of aqueous extract of *L. alata* stem bark on liver glutathione S transferase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	32.46 ± 0.6[a]	31.04 ± 1.8[a]	33.82 ± 1.1[a]
100	40.19 ± 1.5[b]	55.43 ± 2.0[c]	75.02 ± 2.5[e]
200	51.62 ± 1.1[c]	64.92 ± 1.8[d]	86.46 ± 3.3[f]
400	62.97 ± 2.9[d]	72.06 ± 2.9[e]	95.61 ± 3.8[g]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-g) different for each parameter are significantly different ($P < 0.05$).

and NQO-1 respectively, 80.66 and 280.98% increase in microsomal UGT and EPH were produced by aqueous extract of *L. alata* stem bark extract.

DISCUSSION

Chemical compounds interact at both cellular, biochemical and molecular level, to initiate significant changes in the structure, function and metabolic transformation of all classes of biomolecules, enzymes and metabolic pathways.[28] This interaction at cellular, biochemical and molecular level could be beneficial or detrimental to the cellular well being of the organism. This study thus investigates the electrophilic, free radical and reactive oxygen species scavenging and detoxification role of *L. alata* stem bark.

Table 8. Effect of aqueous extract of *L. alata* stem bark on liver epoxide hydrolase activity (nmol $min^{-1}mg^{-1}$ protein)

Doses (mg/kgbw)	Day 1	Day 14	Day 28
Control	18.4 ± 2.3[a]	17.6 ± 0.8[a]	18.0 ± 0.6[a]
100	22.7 ± 0.3[b]	31.8 ± 1.5[c]	62.4 ± 3.3[g]
200	28.1 ± 1.9[c]	46.6 ± 1.1[e]	66.9 ± 2.5[g]
400	40.4 ± 1.2[d]	58.4 ± 2.3[f]	70.1 ± 2.8[h]

The results are mean ± SD for 5 rats. Enzyme activities are expressed as nmol $min^{-1}mg^{-1}$protein. Values carrying superscripts (a-h) different for each parameter are significantly different ($P < 0.05$).

In vitro *free radical and reactive oxygen species scavenging potentials*

Free radical chain reaction, a common method of lipid peroxidation (LPO), causes some deleterious alterations to cellular molecules, ranging from the polyunsaturated fatty acid of the plasma membrane to the macromolecules located in the cells. Therefore, the scavenging activity on DPPH, H_2O_2, and superoxide ion, as well as the reducing effect on the ferric ion by the aqueous stem bark extract of *L. alata* may be attributed to interference with lipid peroxidation, possibly terminating free radical chain reactions.[29] This could be an electron-donating capability effect of the extract of *L. alata* stem bark, thus forming a stable product and, consequently, terminating free radical chain reaction.

Reactive oxygen species (ROS) hydroxyl (OH·) and superoxide (O_2·) radicals, and hydrogen peroxide (H_2O_2) generated in biological systems are counteracted by the existing enzymic antioxidant system of biological systems (e.g., catalase, superoxide dismutase, glutathione peroxidase, glutathione reductase, and glucose-6-phosphate dehydrogenase), thus preventing damage to cellular macromolecules.[30] This action is, however, exhaustive, and dietary antioxidants can play a significant role in preventing this damage upon absorption *in vivo*. In addition, Dahl and Richardson have implicated singlet oxygen (O·),[31] hydroxyl radical (OH·),[32] and hydrogen peroxide (H_2O_2) (formed from superoxide) to initiate LPO. Thus, the strong scavenging effect of *L. alata* extract on O_2·, OH· as well as H_2O_2 is an implication of its possible potential of terminating LPO, which could arise from superoxide,[15] and the stronger ROS (e.g., singlet oxygen, hydroxyl radical, and hydrogen peroxide).

The ability of *L. alata* stem bark to display a significant reducing power potential on $K_3Fe(CN)_6$, shows its effectiveness to halt the oxidation of cellular macromolecules by oxidizing molecules that could arise from the metabolism of either drugs or toxins.

In vivo *electrophilic and reactive oxygen species detoxification potential*

Cells attempt to fortify the antioxidant arsenal as the first line of defense in response to oxidative stress. Reactive species of oxygen and nitrogen are counteracted, by the enzymic antioxidant system (such as superoxide dismutase, catalase, glutathione peroxidase, and glutathione reductase) in the body, thus preventing damage of cellular macromolecules, via lipid peroxidation, base modification, DNA cross-links and covalent binding to protein.[30] Although, antioxidant enzymes are rapidly, induced in response to oxidative stress, such adaptive survival response is normally transient and prone, to be overwhelmed by excess amount of ROS. Superoxide dismutase prevents oxidative damage by dismutating the O_2 anion to hydrogen peroxide (H_2O_2) and molecular oxygen, while catalase reduces the H_2O_2 generated to water and molecular oxygen. Glutathione peroxidase is also responsible for the conversion of H_2O_2 to water, while the regeneration of cofactor NADPH to keep the redox cycle going is the responsibility of glutathione reductase.

The significant increase in the antioxidant enzymes; SOD, CAT, GRed and GPx (Table 1, 2, 3 and 4) are evident ability of the aqueous extract of *L. alata* stem bark to detoxify ROS in form of O_2, OH· and H_2O_2 via the induction of their detoxification mechanism. We have previously shown that medicinal plants rich in flavonoids and polyphenols could prevent oxidative stress by inducing ROS detoxification enzymes (antioxidant enzymes).[30,33,34,35]

ROS detoxification enzyme induction ability could be adduced to the presence of the numerous chalcone dimers, tetramers as well as flavonoids, which have been shown to induce nuclear translocation of Nrf-2 to mediate antioxidant gene expressions.

Cellular detoxification is crucial for the maintenance of health by providing protection against the daily exposure to various xenobiotics.[36,37] Antioxidant and phase II drug-metabolizing enzymes (NQO1, HO-1, UGT, GST, EPH, among others) act in concert to detoxify and eliminate these harmful xenobiotics. Activation of the phase II enzyme system to detoxify potential carcinogens and reduce inflammation and oxidative stress is an emerging strategy for preventing cancer in both animals and humans.[38,39]

Phase II cytoprotective enzymes play important roles in the detoxification of reactive electrophiles to non-carcinogenic metabolites, and, therefore, sustained induction of these enzymes in humans could alleviate cancer risk in individuals exposed to environmental chemicals.

Persistent induction of phase II electrophilic detoxifying enzymes(NQO-1, GST, UGT & EPH) was shown all through the experimental period, at all doses used. This significant induction by aqueous extract of *L. alata* stem bark shows the ability of the plant to detoxify electrophile and the important role it could play in carcinogenesis prevention and suppression. [30,33,34,35,36,38,39,40] Numerous studies have shown that medicinal plants, as well as phytochemicals capable of inducing phase II electrophilic detoxifying enzymes act as chemopreventive agent. They do this by inactivating xenobiotics and electrophilic carcinogens.

The available data in this study thus shows that *L. alata* stem bark can act as a prophylactic by acting as antioxidant and free radical scavenger as well as enhancing the level of ROS and electrophile detoxification enzymes.

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