Free Radical Scavenging and Anti-lipid Peroxidative Effects of a Hydro-ethanolic Extract of the Whole Plant of *Synedrella nodiflora* (L.) Gaertn (Asteraceae)

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ABSTRACT

Background: *Synedrella nodiflora* (L.) Gaertn. (Asteraceae), a native Ghanaian shrub, has been used for the treatment of epilepsy, hiccup and threatened abortion. The present study aimed at investigating the possible mechanisms of antioxidant effects of the hydro-ethanolic extract of the whole plant. **Methods**: Total phenolic content was determined using the Folin-Ciocalteau assay and the antioxidant capacity by the phosphomolybdenum method. The antioxidant activity was evaluated by the 1, 1-diphenyl-2-picryl-hydrazyl (DPPH) scavenging assays, reducing power assay and inhibition of linoleic acid peroxidation and lipid peroxidation in pentylenetetrazole (PTZ)- kindled rat brains. **Results:** The extract (0.1-3.0 mg ml^{-1}) was found to contain phenolic compounds which could be responsible for the antioxidant properties since the coefficient of correlation between the Total Phenolic Content (TPC) and the Total Antioxidant Capacity (TAC) was high (r^2 = 0.9908). Both *n*-propyl gallate (0.001-0.03 *mg* ml^{-1}), a reference antioxidant and the extract (0.1-3 mg ml^{-1}) exhibited antioxidant properties by reducing Fe^{3+} to Fe^{2+} in the reducing power test, scavenged DPPH free radicals and effectively inhibited linoleic acid autoxidation and also inhibited lipid peroxidation in PTZ-kindled rat brains. **Conclusions:** These findings suggest that hydro-ethanolic extract of *Synedrella nodiflora* contains antioxidant principles which may contribute to its traditional use in epilepsy management.

Key words: *Synedrella nodiflora*, antioxidant, linoleic acid, autoxidation, *n*-propyl gallate, phosphomolybdenum, Folin-Ciocalteau, DPPH.

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INTRODUCTION

Synedrella nodiflora (L.) Gaertn. (Asteraceae) is an annual herb which grows to about 60-120 cm high. It is a native tropical American weed but now dispersed pan-tropically, and occurring throughout the West African region. In Ghana, the foliage is readily eaten by livestocks whereas, in Indonesia, the foliage is eaten as a vegetable by some indigenous tribes. In Ghanaian traditional medicine, the whole plant is boiled and the aqueous extract drunk as required for the treatment of epilepsy. The leaves are used for the treatment of hiccup and threatened abortion.[1] The plant has also been extensively used in Nigeria for cardiac problems, wounds and for stopping bleeding.[2] In Malaysia and Indonesia, the plant has also been used for the treatment of headaches, earaches, stomachaches and in embrocation for rheumatism.[3] The whole plant extract have been reported to possess potent anti-inflammatory,[4] antibacterial and antioxidant[5] and antinociceptive.[6] Free radicals and associated lipid peroxidation are implicated in epilepsy and inflammation[7] and the role of antioxidants in the management of conditions caused by oxidative stress have been reported.[8-11] We have demonstrated the anticonvulsant effects of the hydro-ethanolic extract of the plant in our laboratory. To date, there is little scientific evidence of the possible antioxidant mechanisms of the plant and its role in the traditional uses of the plant. This study reports the free radical scavenging and anti-lipid peroxidative properties of the hydro-ethanolic extract of the whole plant of *Synedrella nodiflora*.

MATERIALS AND METHODS

Animals

Sprague-Dawley rats (150-200 g) were purchased from Noguchi Memorial Institute for Medical Research, University of Ghana, Legon, and maintained in the Animal House of the Department of Pharmacology, Kwame Nkrumah University of Science and Technology (KNUST), Kumasi. The animals were housed in groups of six in stainless steel cages (34 × 47 × 18 cm) with soft wood shavings as bedding, fed with normal commercial pellet diet (GAFCO, Tema), given water *ad libitum* and maintained under laboratory conditions (temperature 24-28 °C, relative humidity 60-70%, and 12 hour light-dark cycle). All procedures and techniques used in these studies were in accordance with the National Institute of Health Guidelines for the Care and Use of Laboratory Animals (NIH, Department of Health Services publication No. 83-23, revised 1985). The protocols for the study were approved by the Departmental Ethics Committee.

Plant collection

The whole plant of Synedrella nodiflora was obtained from the Botanical Gardens, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana in August 2007 and authenticated by the Department of Herbal Medicine where a specimen voucher, FP/08/025, has been kept.

Preparation of extract

The plant samples collected were air-dried for seven days, powdered. Two kilograms of the powdered was cold-macerated with 70 % v/v of ethanol. The hydro-ethanolic extract was then evaporated to a syrupy mass under reduced pressure, air-dried and kept in a dessicator. A 7% w/w yield was obtained. This is subsequently referred to as the extract or SNE.

Phytochemical Analysis and TLC

Chemical tests and TLC were carried out on the extract for the qualitative determination of phytochemical constituents and retardation factor values using standard procedures as described by Trease and Evans.[12]

Drugs and Chemical

The drugs and chemicals used in this study include: Folin-Cicocalteu's phenol reagent (EMD Chemicals Inc., Gibbstown, NJ, USA); sodium bicarbonate, sulfuric acid, sodium phosphate, ammonium molybdate, potassium ferricyanide, tannic acid, ferric chloride, *n*-propyl gallate, linoleic acid (BDH, Poole, England); trichloroacetic acid, thiobarbituric acid and DPPH (Sigma-Aldrich, St. Louis, MO, USA)

Total Phenol Assay

The total soluble phenols present in the extract was quantitatively determined by colorimetric assay using the Folin-Cicocalteu's phenol reagent [13] with some modifications. Tannic acid (0.01-0.3 mg ml^{-1}) was used as the reference drug. The test drug (1 ml) was added to 1 ml of Folin-Cicocalteu's phenol reagent (diluted five folds in distilled water). The content of the test tube was mixed and allowed to stand for five minutes at 25 °C in the incubator. 1 ml of 2 % sodium bicarbonate solution was added to the mixture. The reaction mixture was then incubated at 25 °C for 2 hours. It was then centrifuged at 1238 g-force for 10 min to obtain a clear supernatant. The absorbance of the supernatant was then determined at 760 nm using a Cecil CE 2040 spectrophotometer (Cecil Instrument Limited, Milton Technical Centre, England). Distilled water was used as the blank. The total phenols were expressed as tannic acid equivalents using the GraphPad Prism for Windows version 4.02 (GraphPad Software, San Diego, CA, USA).

Total Antioxidant Capacity Assay

The assay is based on the reduction of Mo (VI) to Mo (V) by the extract and subsequent formation of a green phosphate/Mo (V) complex at acid pH.[14] The tubes containing extract (0.1-3 mg ml^{-1}) and reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate) were incubated at 95 °C for 90 min. After the mixture had cooled to room temperature, the absorbance of each solution was measured at 695 nm against a blank. The antioxidant capacity was expressed as ascorbic acid equivalent.

Reducing Power

The reducing capacity of the extract (0.1-3 *mg* ml^{-1}) in methanol was determined using the method of Fe^{3+} - reduction to Fe^{2+}[15] as previously described,[16] using *n*-propyl gallate (0.001-0.3 mg ml^{-1}) as a reference antioxidant. The drug/extract (1 ml) was mixed with 2.5 ml of 0.2 M sodium phosphate buffer (pH 6.6) and the 2.5 ml of 1 % potassium ferricyanide solution in a test tube. The mixture was incubated at 50 °C for 20 min.

Following this, 1.5 ml of 10 % trichloroacetic acid solution (TCA) was added to the incubated mixture, and centrifuged at 3000 rpm for 10 min. The supernatant (2.5 ml) was then mixed with 2.5 ml distilled water and 0.5 ml of the 0.1 % ferric chloride solution ($FeCl_{3(aq)}$) in a test tube. The absorbance was then measured at 700 nm. Distilled water was used in place of the test drug and this was used as the blank. Data was presented as concentration - absorbance curves and the EC_{50} (concentration that gives 50% of maximal response) determined.

Free Radical Scavenging Assay

The experiment was carried out as previously described by [17] with a few modifications. The extract (0.1-3 mg ml^{-1} in methanol) was compared with n-propyl gallate (0.001-0.03 mg ml^{-1} in methanol) as a reference free radical scavenger. The test drug/extract was centrifuged at 3000 rpm for 10 min and the supernatant collected. The supernatant (1 ml) was added to 3 ml of methanolic solution of DPPH (20 mg l^{-1}) in a test tube. The reaction mixture was kept at 25 °C for 1 h in an orbital shaker (BoroLabs, Aldermaston Berkshire, EC). The absorbance of the residual DPPH was determined at 517 nm in a Cecil CE 2040 spectrophotometer (Cecil Instrument Limited, Milton Technical Centre, England). Methanol (99.8%) (1 ml) was added to 3 ml DPPH solution, incubated at 25 °C for 1 h and used as control. Methanol was used as the blank. The results were expressed as % DPPH scavenging effect against concentration and the EC_{50} determined.

Linoleic acid auto-oxidation

The method as previously described was used with slight modifications.[18] The extract (0.1-3 *mg ml*$^{-1}$ in 96% ethanol) was compared with *n*-propyl gallate (0.001-0.03 *mg ml*$^{-1}$ in 96% ethanol) as a reference antioxidant. Briefly, a mixture of the sample (2 ml), 2.052 ml of 2.51% linoleic acid (in 96% ethanol), 4 ml of 0.05M phosphate buffer (pH 7.0) and 1.890 ml of distilled water was put into a test tube with a screw cap and placed in an oven at 40 °C in the dark for seven (7) days.

To 2ml of the sample solution, which was prepared and incubated as described above, was added 2 ml of 20% trichloroacetic $acid_{(aq)}$ solution and 1 ml of 0.6 % thiobarbituric $acid_{(aq)}$ solution. This mixture was placed in boiling water bath for 10 min and after cooling, was centrifuged at 1238 g-force for 10 min. The absorbance of the supernatant was measured.

The % inhibition of linoleic acid autoxidation was calculated as follows

$$\% \text{ Inhibition} = \frac{(C_0 - C_1) - (D - D_0)}{(C_0 - C_1)} \times 100$$

Where

- $\mathbf{C_0}$: is the degree of lipid peroxidation in the absence of antioxidant.
- $\mathbf{C_1}$: is the underlying lipid peroxidation before the initiation of lipid peroxidation.
- **D**: is any absorbance produced by the extract or the drug being tested.
- $\mathbf{D_0}$: is the absorbance of the extract/drug alone

Lipid peroxidative assay in PTZ-Kindled rat brain homogenates

This test was done to assess the role of lipid peroxidation in PTZ- induced kindling and the beneficial role of SNE which has demonstrated in vitro antioxidant properties. To induce kindling, a 35 mg kg^{-1} dose of pentylenetrazole (PTZ) was injected i.p. every 48 h into SNE or diazepam treated male Sprague-Dawley rats (200-300 g) and a rat was considered fully kindled after showing generalized tonic-clonic convulsions after two consecutive PTZ administrations. On day seven after kindling had been achieved, the rats were challenged with 35 mg kg^{-1} of PTZ and the entire event was also recorded. On the following day after the 35 mg kg^{-1} PTZ challenge, kindled rats were sacrificed by decapitation and forebrain was dissected out and homogenized (100 mg ml^{-1}) in ice-cold 0.1 M phosphate buffer (pH 7.4) using Ultra-Turrax T 25 homogenizer (IKA Labortehnic, Staufen, Germany). Brain homogenate (2.5 ml) was mixed with 1 ml phosphate buffer and mixture was then incubated in an orbital shaker incubator (BoroLabs, Aldermaston Berkshire, EC) at 37 °C for 1 h.

To assay for thiobarbituric acid reactive substances (TBARS), 0.1 ml of the incubated reaction mixture was taken into a test tube containing 1.5 ml of 10% trichloroacetic acid (TCA), and allowed to stand for 10 min. Then the tubes were centrifuged at 4000 rpm for 10 min. The supernatant was separated and mixed in a tube containing 1.5 ml of 0.67% thiobarbituric acid (TBA) in 20% acetic acid. The mixture was heated in a hot water bath at 85 °C for 1 h to complete the reaction and allowed to cool. The intensity of the pink-colored complex formed was measured at 535 nm in a spectrophotometer (Cecil CE 7200 spectrophotometer, Cecil Instrument Limited, Milton

Technical Centre, England). The Absorbance decreases with increasing ability to inhibit lipid peroxidation. Phosphate buffer was used as blank throughout the experiment.

The percentage inhibition of lipid peroxidation was then calculated from the following equation:

$$\% \text{ Inhibition} = \frac{(C_{PTZ} - C_O) - (FRM - C_O)}{(C_{PTZ} - C_O)} \times 100$$

Where:

1. **C_{PTZ}**: is the degree of lipid peroxidation in the kindled but untreated rats
2. **Full Reaction Mixture** (**FRM**): is the underlying lipid peroxidation of the treated rats
3. **C_O**: is the absorbance of the unkindled and untreated rats

Data was presented as % inhibition of lipid peroxidation against concentration and the EC_{50} (concentration that produces 50% of the maximal effect of drug) for each drug determined from concentration-response curves using GraphPad Prism for Windows version 4.02 (GraphPad Software, San Diego, CA, USA)

Data Analysis

GraphPad Prism Version 5.0 for Windows (GraphPad Software, San Diego, CA, USA) was used for all statistical analyses. Data are presented as mean ± SEM $P \leq 0.05$ was considered statistically significant in all analysis. The graphs were plotted using SigmaPlot for Windows Version 11.0 (Systat Software Inc., Germany)

RESULTS

Phytochemical Analysis and TLC

The phytochemical screening of SNE revealed the presence of alkaloids, saponins, tannins and reducing sugars. The TLC also revealed five different spots with varying colours and retardation factors (table 1)

Table 1. Thin Layer Chromatography of SNE

Shape	Colour	Retardation Factor (Rf) value
Round	Green	0.35
Round	Orange	0.68
Round	Green	0.75
Round	Yellow	0.88
Oval	Orange	0.90

Total Phenol Assay of SNE

The phenol content of tannic acid (0.01-0.3 mg ml^{-1}) increased with increasing concentration (r^2 = 0.9901) (Figure 1). The extract SNE (0.1-3 mg ml^{-1}) also showed a concentration dependent increase in phenolic content expressed as tannic acid equivalent

Antioxidant Capacity of SNE

The total antioxidant capacity of ascorbic acid (0.01 - 0.3 mg ml^{-1}) increased with increasing concentration (r^2 = 0.9771) (Figure 2A). The extract (0.1-3 mg ml^{-1}) also showed a concentration dependent increase in total antioxidant capacity expressed as ascorbic acid equivalent (Figure 2).

The antioxidant capacity of SNE was strongly dependent on the total phenolic contents as revealed by the high coefficient of correlation (r^2 = 0.9908) (Figure 3).

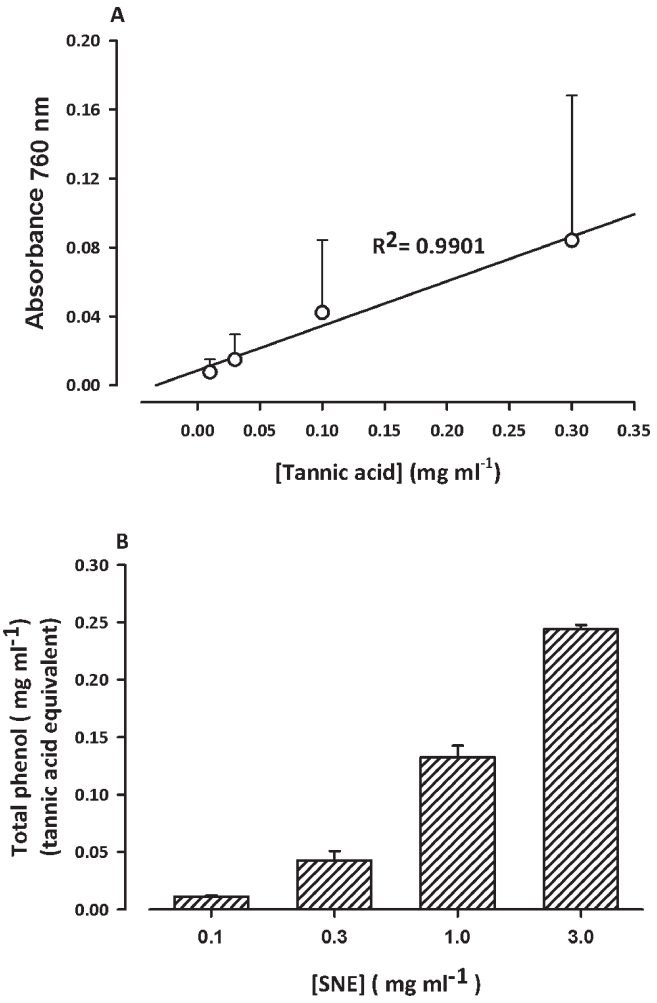

Figure 1. (A) Absorbance of tannic acid (0.01-0.3 mg ml^{-1}) and (B) total phenolic substances present in various concentrations of SNE (0.1-3 mg ml^{-1}) expressed as tannic acid equivalent. Each point in A and columns in B represent the mean ± s.e.m (n = 5)

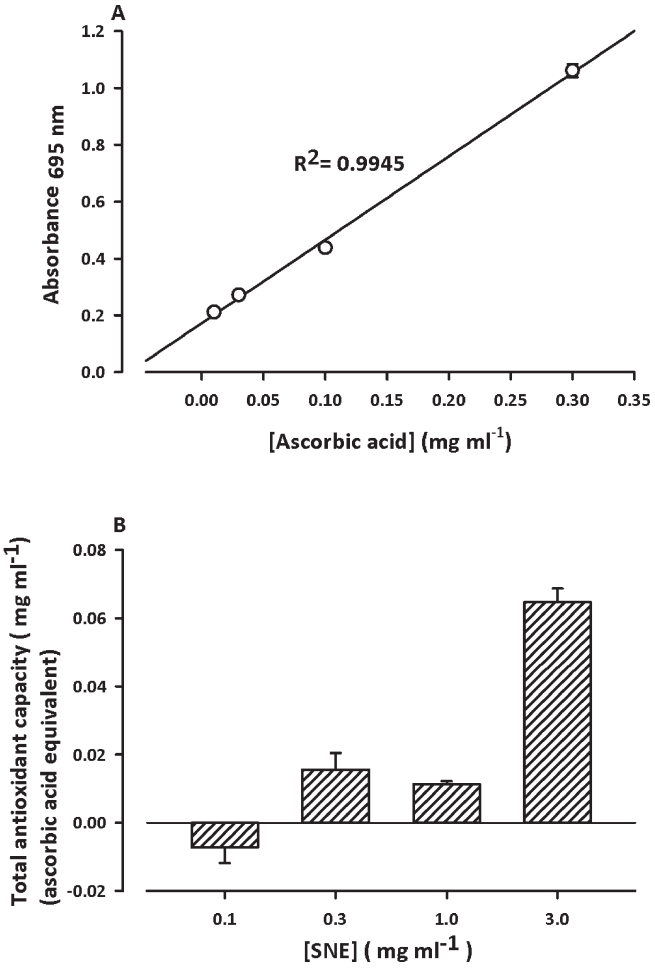

Figure 2. (A) Absorbance of ascorbic acid (0.01-0.3 mg ml^{-1}) and (B) total phenolic substances present in various concentrations of SNE (0.1-3 mg ml^{-1}) expressed as ascorbic acid equivalent. Each point in A and columns in B represent the mean ± s.e.m (n = 5).

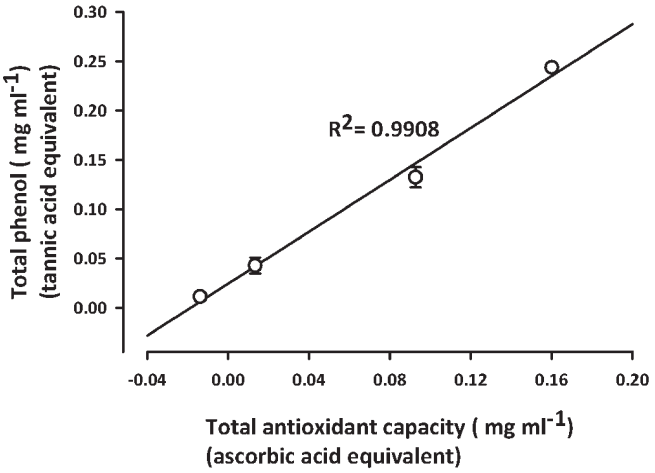

Figure 3. A linear regression of the total phenolic content expressed as tannic acid equivalents and total antioxidant capacity expressed as ascorbic acid equivalents of SNE (0.1-3 mg ml^{-1}). Each point represents the mean ± s.e.m (n = 5)

SNE has Reducing Power

The extract (0.1-3 mg ml^{-1}) and the reference antioxidant n-propyl gallate (0.001-0.03 mg ml^{-1}) dose dependently reduced Fe^{3+} to Fe^{2+} resulting in concentration dependent increase in absorbance (Figure 4). SNE was found to be about 18 fold less potent than *n*-propyl gallate the reference antioxidant (table 2).

SNE has Free Radical Scavenging Effect

Both *n*-propyl gallate (0.001-0.03 mg ml^{-1}) and the extract SNE (0.1-3 mg ml^{-1}) exhibited dose dependent free radical scavenging activity (Figure 5). The rank order of potency (defined by EC_{50} in mg ml^{-1}, Table 1) showed that *n*-propyl gallate was 100 fold more potent than SNE.

SNE inhibited linoleic acid auto-oxidation

All the test samples used, *n*-propyl gallate (0.001-0.3 mg ml^{-1}), and the extract (0.1-3 mg ml^{-1}) showed concentration dependent ability to inhibit the autoxidation of linoleic acid lipid peroxidation (Figure 6). *n*-propyl gallate was found to be twice as potent as SNE.

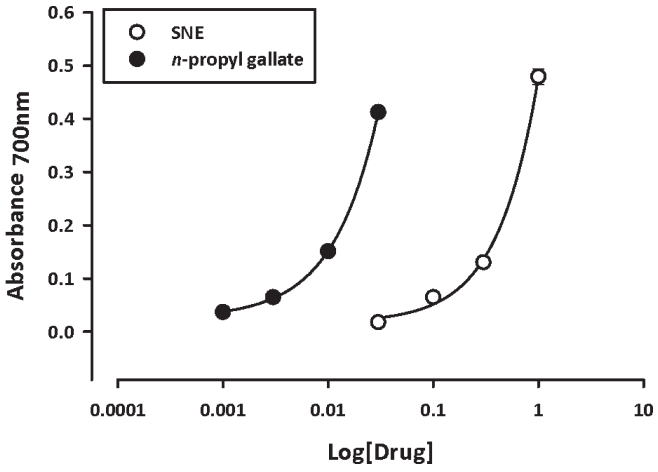

Figure 4. Reducing power of SNE (0.1-3 mg ml^{-1}) compared to n-propyl gallate (0.001-0.3 mg ml^{-1}). Each point represents the mean ± s.e.m.(n = 5)

Table 2. EC_{50} of test drugs in the reducing power, DPPH scavenging and lipid peroxidation assays

Drug	EC_{50} (mg ml^{-1})		
	Reducing Power	DPPH Scavenging × 10^{-3}	Linoleic autoxidation × 10^{-3}
SNE	459.7	315.9	14.35
n-Propyl gallate	26.1	3.221	8.954

Lipid peroxidative assay in PTZ-Kindled rat brain homogenates

SNE (100-1000 *mg kg*$^{-1}$) dose-dependently inhibited lipid peroxidation in the PTZ-kindled rats whereas diazepam (0.1-1.0 *mg kg*$^{-1}$) gave an opposite effect (Figure 7).

DISCUSSION

The present study demonstrates the free radical scavenging and anti-lipid peroxidative effects of the hydro-ethanolic extract of *S. nodiflora in vitro*. The results confirm the earlier report that extracts from the plant possess antioxidant and provides additionally useful scientific data.

The phytochemistry of *S. nodiflora* revealed the presence of steroids, reducing sugar, alkaloids, phenolic compounds and aromatic acids and this confirmed what researchers found in similar extracts of the plant.[19] These plant constituents have been found to possess antioxidant properties. Alkaloids such as quinoline alkaloids and β-carboline, and tannins and pseudotannins (catechin) are known to be potent free radical terminators.[20-21] Thus the potent antioxidant properties of *S. nodiflora* may be related to the presence of these constituents.

Polyphenols (electron-rich compounds) have the ability to go into electron-donation reactions with oxidizing agents to form stable species [22] and thus inhibit or delay the oxidation of different biomolecules.[16,23] Hence various plant phenols such as vitamin E (α-tocopherol), exhibit antioxidant properties.[22-24] Phenolic antioxidants are potent free radical terminators and this is thought to be due to the ability to donate hydrogen to free radicals and their presence is a good marker of potential antioxidant activity. The high potential of phenolic compounds to scavenge free radicals may be explained by their phenolic hydroxyl groups. Detection of phenols in the *S. nodiflora* extract was a preliminary evidence of its possible antioxidant activity (5). The total phenol was assayed based on the reduction of phosphomolybdate-phosphotungstate salts to form a blue complex that is detected quantitively at 700 nm. The total phenolic content of the extract expressed as the tannic acid equivalents increased concentration dependently.

The phosphomolybdenum method of assay of the total antioxidant capacity was based on the reduction of Mo (VI) to Mo (V) by the antioxidant compound and the formation of a green phosphate/ Mo (V) complex with a maximal absorption at 695nm. Chemical constituents

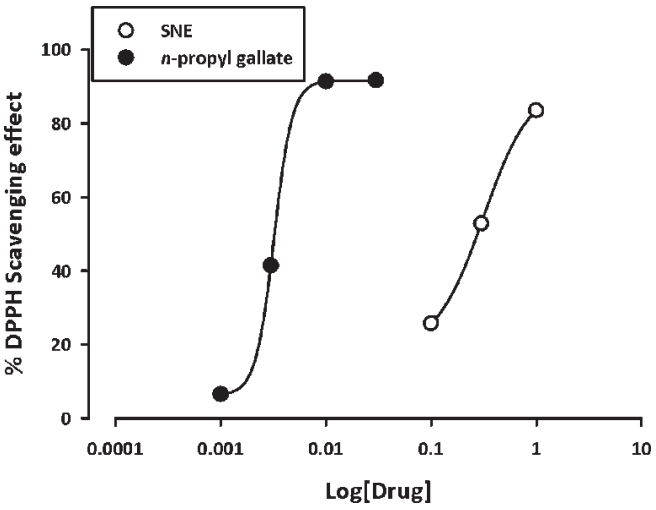

Figure 5. Free radical scavenging ability of SNE (0.1-3 mg ml^{-1}) compared to *n*-propyl gallate (0.01-0.3 mg ml^{-1}) in the DPPH scavenging assay. Each point represents the mean ± s.e.m (n = 5).

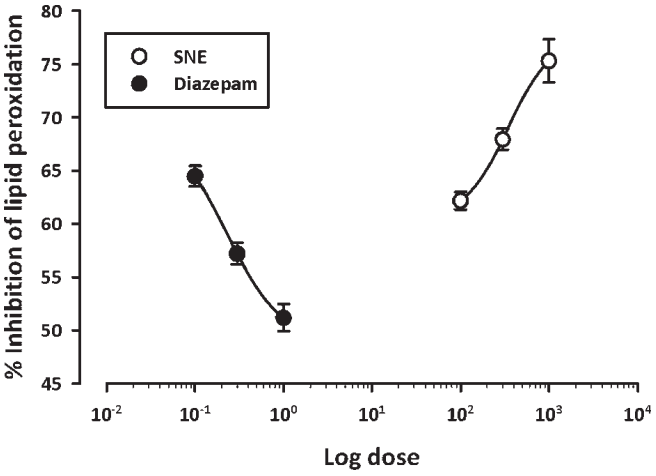

Figure 6. Percentage inhibition of linoleic acid autoxidation by SNE (0.1-3.0 mg ml^{-1}) compared to *n*-propyl gallate (0.001-0.3 mg ml^{-1}). Each point represents the mean ± s.e.m (n = 5).

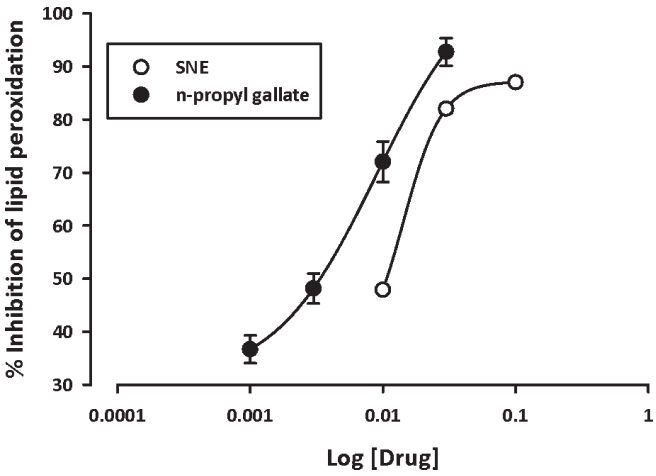

Figure 7. Percentage inhibition of PTZ induced lipid peroxidation in PTZ kindled rats by SNE (100-1000 *mg kg*$^{-1}$) and diazepam (0.1-1.0 *mg kg*$^{-1}$). Each point represents the mean ± s.e.m (n = 5).

that might contribute to the total antioxidant capacity includes carotenoids, flavonoids and cinnamic acid derivatives.[25] Also a high correlation was obtained between the total antioxidant capacity and the total phenol content suggesting that the phenols detected may basically be responsible for the antioxidant effect found in SNE.

The 1, 1-diphenyl-2-picryl-hydrazyl (DPPH) is used as a reagent to evaluate free radical scavenging activity of antioxidants. It is a stable free radical and accepts an electron or hydrogen radical to become a stable diamagnetic molecule.[26] The reduction capability of DPPH is determined by the decrease in the absorbance at 517 nm induced by antioxidants. This test system is a valid for the primary characterization of the scavenging potential of compounds.[27] The extract's ability to concentration- dependently reduce DPPH forming yellowish-coloured diphenylpicylhydrazine suggest that SNE is a free radical scavenger and acts so by donating an electron or hydrogen radical.

The extract showed a concentration dependent increase in its reducing power. This measurement, as described by Oyaizu (1986), describes the Fe^{3+} to Fe^{2+} transformation in the presence of the extract. Iron has the ability to gain and loss electrons (i.e. (Fe^{2+} «Fe^{3+}) very easily, thus a common catalyst of oxidation reaction. The release of iron from the breakdown of red blood cells can be detrimental to cellular membranes because of the pro-oxidation effects it can have. Other authors have observed a direct correlation between antioxidant activity and reducing power of certain plant extracts and have associated this effect with the presence of reductones.[28-29] Reductones are also reported to react with certain precursors of peroxides, thus preventing peroxide formation. However activity of antioxidants has been attributed by various mechanisms, among which some of them are prevention of chain initiation, binding of transition metal ion catalysts, decomposition of peroxides, prevention of continued hydrogen abstraction, reductive capacity, and radical scavenging.[30]

S. nodflora showed an ability to inhibit lipid peroxidation in linoleic acid antoxidation assay. Polyunsaturated fatty acids (PUFAs) are abundant in cellular membranes and low density lipoprotein.[31] Peroxidation of PUFAs by free radicals yields aldehydes such as thiobarbituric acid reacting substances (TBARS) which have been widely accepted as a general marker of free radical production.[32] The most commonly measured TBARS is malondialdehyde (MDA) (33). Lipid peroxidation is considered a marker of oxidative stress.[34] Linoleic acid autoxidation assay was chosen due to it relatively simple and cost effective methodology.[18] The ability of SNE in inhibiting lipid peroxidation suggests a potent antioxidant property and this may contribute to its mechanism of action.

Also, SNE was observed to dose-dependently inhibit lipid peroxidation in brains from PTZ kindled rats. Thus suggesting the anticonvulsant effect of SNE in the kindling may be partly due to anti-lipid peroxidative effects. It is reported that free radical generation due to the increased activity of the glutamatergic transmitter plays a fundamental role in neuronal cell death of the PTZ kindling in rats.[35-40] There are evidence indicating a linkage between the generation of clonic–tonic seizures and the increased formation of free radicals in the brain.[39] Thus, some antioxidants such as ascorbic acid have been shown to be exert anticonvulsant effect in pilocarpine induced SE through the reduction of lipid peroxidation content and increased catalase enzyme activity.[41] Thus the role of antioxidants as adjunct support to the treatment of epilepsy are been explored.[10-11,42] The hydro-ethanolic extract of *S. nodiflora* has demonstrated potent anticonvulsant effects in both acute and chronic murine models of experimental epilepsy (unpublished data), anti-inflammatory [43] and analgesic properties [6] and this observed free radical scavenging and anti-lipid peroxidative effects may contribute to the overall mechanisms employed by the extract.

The ability of SNE to scavenge free radicals, possess reducing capacity and inhibit lipid peroxidation suggests that SNE is an antioxidant that can be classified as preventive, scavenging or chain-breaking.

CONCLUSION

The findings from the study clearly demonstrate significant antioxidant effects of *S. nodiflora* extract which may play a role in the traditional uses of the plant.

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LIST OF ABBREVIATIONS

DPPH	1, 1-diphenyl-2-picryl-hydrazyl (DPPH)
EC_{50}	Effective Concentration that gives 50% of maximal response

NIH	National Institutes of Health
PTZ	Pentylenetetrazole
PUFAs	Polyunsaturated fatty acids
SNE	*Synedrella nodiflora* extract
TAC	Total Antioxidant Capacity
TBA	Thiobarbituric acid
TBARS	Thiobarbituric acid reactive substances
TLC	Thin Layer Chromatography
TPC	Phenolic Content

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