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RP-HPLC based evidences of rich sources of Phenolics and water-soluble vitamins in an annual sedge *Cyperus compressus*

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ABSTRACT

The present study makes an effort to evaluate the content of water-soluble vitamins like ascorbic acid (C), thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), pyridoxine (B6) and folic acid (B9) by HPLC of *Cyperus compressus*. The in vitro antioxidant activity was carried out in different solvent system of varying polarity. The 70% ethanol appeared to be the most potent solvent for extraction of antioxidant compounds. The total phenolic and flavonoid content were found 72.544 ± 0.144 mg/g dm and 12.930 ± 0.144 mg/g dm respectively in the plant. Simultaneous HPLC fingerprinting of some standard phenolic acids and flavonoids in the 70 % aqueous ethanol extract was also carried out. B vitamins ranged between 0.028 to 0.431 mg/100g dm. The results confirm that this annual sedge is an excellent source of antioxidant- based phyto-nutrients, which could be used for the good of human being at the same time it can be used as a vitamin supplement to our diet. The study also provides the scientific basis of traditional significance of *Cyperus compressus*.

Keywords: *Cyperus compressus*, antioxidants, Phenolics, water soluble vitamins, HPLC analysis.

INTRODUCTION

Plants synthesize various phytochemcals such as phenols, flavonoids, alkaloids, tannins, vitamins, carotenoids etc. which protect biotic and abiotic stress mediated oxidative damages [1]. These plant derived phytochemicals have antioxidant property and are also extremely useful in the treatment of many ROS mediated diseases such as atherosclerosis, cancer, liver diseases, neurodegenerative disorder and diabetes [2]. Since these phytochemicals are a part of the normal physiology of the plant cells they are less harmful to mankind and the deleterious effect of synthetic antioxidants can be avoided [3]. Therefore when, oxidative stress is found to be one of the major causes of health hazards plant based medicine has become more important [4]. Since it is a common grazing grass its water soluble vitamins content were assessed. Plant sources are rich in vitamin content and if they could be included to our diet as supplement it would be a cheaper source of nutrition supplement. The antioxidant activity of *C. compressus* is not worked out in detail. Other species of *Cyperus* like *C. rotundus* has indicated presence of strong radical scavenging property [5]. *Cyperus compressus* L. is used to treat cuts and wound [6], the root and stem extract also exhibit insect repellent activities [7].

This paper reports the evaluation of the antioxidant activities of the aerial part extracts from *C. compressus* a along with a HPLC based identification of phenolic compounds and relate them to the folklore uses.

Material and Methods

Plant Materials

C. compressus were collected from various locations of Kolkata, India and were identified from Botanical Survey of India, Howrah. Plant materials were stored at 15^0C and processed for vitamin estimation. Another part was dried, pulverized and stored in an airtight container for the analysis of antioxidant properties, proximate composition and minerals contents.

Extraction of plant material

For the phytochemical extraction, one gram dry sample was extracted separately with 20 ml of methanol, 70% ethanol, chloroform, benzene with agitation for 18–24 h at ambient temperature. The

extracts were subsequently filtered with Whatman #1 filter paper and diluted to 25 ml with corresponding extracting solutions. These solvent extracted materials were analyzed for their antioxidant attributes.

Estimation of total phenolic content

For the extraction and estimation of total phenolic content of the different plant extracts Folin-Ciocalteu procedure was followed [8]. The total phenolic content was expressed as mg/g gallic acid equivalent (GAE).

Estimation of total flavonoid content

For the extraction and estimation of total flavonoid content, the method of Ordonez *et al*. [9] was followed. Total flavonoid contents were expressed as mg/ g rutin equivalent (RE)

Ferric Reducing Antioxidant Power (FRAP) Assay

Total antioxidant capacity of plant extracts was assessed in terms of Ferric Reducing Antioxidant Power (FRAP) and for this the method of Benzie and Strain [11] was followed. The TAC was expressed as trolox equivalent (TE) mg/ g dry mass.

Measurement of reducing power

The reducing power of the extracts was determined according to the method of Oyaizu [10]. Reducing power is calculated as ascorbic acid equivalent (AAE) in mg/ g of dry material

DPPH (2,2-Diphenyl-1-picryl-hydrazyl) assay

For the determination of antioxidant properties using a free radical scavenging assay of DPPH method the process of Blois (1958)[12] was followed.

ABTS [2, 2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)] assay

For the determination of total antioxidant capacity (TAC) using ABTS, the process of Re *et al*. (1999) [27] was followed.

Metal Chelating activity

Metal chelating property

For the estimation of metal chelating property of experimental plant tissue, the process of Lin *et al*. (2009)[14] was followed with slight modifications. Shortly, 1 ml of different solvent extracts (extraction procedure described earlier) was added to a solution of .02 ml 2 mM ferrous chloride and .04 ml 5 Mm ferrozine. The mixture was vigorously shaken and incubated for 10 mins. Absorbance was taken at 562 nm.

Anti-lipid peroxidation assay in linoleic acid system
For the estimation of anti-lipid peroxidation assay experimental plant tissue the process of Amabye (2015)[15] was followed with slight modifications.

RP-HPLC study for the estimation of Phenolic acids and Flavonoids

For RP-HPLC analyses Dionex Ultimate 3000 liquid chromatograph with a diode array detector (DAD) with 5 cm flow cell and with Chromeleon system manager as data processor. Separation of phenolics was achieved by a reversed-phase Acclaim C18 column (5 micron particle size, 250 × 4.6 mm). 20 μL of sample was loaded into the HPLC column.

Fourteen standard phenolic acids having concentration 1 mg/ml (gallic acid, protocatechuic acid, gentisic acid, chlorogenic acid, p-hydroxy benzoic acid, vanillic acid, caffeic acid, syringic acid, p-coumaric acid, ferulic acid, sinapic acid, salicylic acid and ellagic acid) and eight flavonoids having the same concentrations (catechin, rutin, myricetin, quercetin, naringin, naringenin, apigenin and kaempferol) was prepared. The standard and working solutions were filtered through 0.45 µm PVDF-syringe filter and the mobile phase was degassed before the injection to the chromatograph.

Chromatographic analysis of phenolic acids and flavonoids

20 µL of sample was introduced into the HPLC column. The mobile phase contains Solvent A (methanol) and Solvent B (0.5% aq. acetic acid) and the column was thermostatically controlled at 25^0C. Subsequently, a gradient elution was performed by changing the proportion of solvent A to solvent B. Total analysis time per sample was 105 min. HPLC chromatograms were detected at three different wavelengths (272, 280 and 310 nm) using a photo diode array UV detector. Each phenolic compound was identified by its RT value and by spiking with standards under the same conditions. The quantification of phenolic acids and flavonoids in the extract were carried out by the measurement of the integrated peak area.

RP-HPLC study for the estimation of Water Soluble Vitamins

The stock standard solutions of vitamin C, B1, B3, B5 and B6 and were prepared by dissolving each standard in 0.1M hydrochloric acid. For preparation of standard stock solutions of vitamin B9 and B2, each standard were dissolved in 0.1 M sodium hydroxide. Finally, the working standards were prepared from each standard stock solutions by mixing 100 µl mixed vitamins standard (vitamin B9, B5 and B2, C, B1, B6 and B3) with 800 µl phosphate buffer (1M, pH 5.5).

Chromatographic analysis of water soluble vitamins

The chromatographic separation was carried out following the method as described by Seal *et al*. [16] with the mobile phase contains acetonitrile and aqueous trifluoro acetic. A gradient elution was performed by varying the proportion of two solvents. Detected was done by using a photo diode array UV/detector at four different wavelengths (210, 245, 275 and 290 nm).

RESULTS

Extraction yield, total phenolics and flavonoids content

Table 1 shows percentage yield of the different solvent extracts of *C. Compressus*. The extraction yield of these samples varied from

0.500% to 8.885 % (Table 1). 70 % hydro-ethanol proved to be better solvent for the plant studied. The process of extraction solubilises the phenolic compounds of the plant sample studied and hence it is an essential part of phenol estimation. In this study four different solvent systems of varying polarity viz. 70% hydro-ethanol, methanol, chloroform and benzene were used as extracting solvent**.** Total phenolic *content (GAE g^{-1} dm) of the extract showed a wide variation for C. Compressus*, ranging from 72.544 – 17.735 mg GAE/ g dm (Table 1)**.** The flavonoid content is expressed as mg rutin equivalent (RE)/ g dm. Maximum flavonoid content was observed in 70% ethanol extract (Table 1). Flavonoids are widely distributed group of plant phenolic compounds responsible for the antioxidant activity of the plants.

Table 1: Extractive capacities (% extractive yield), Total phenolic content (mg GAE/ g dm) and Flavonoid content (mg RE/ g dm) of *C. compressus* in different solvent system

Extractive value and quantification of secondary metabolite	Extracts of *C. compressus* in different solvent system			
	70% ethanol	Methanol	Chloroform	Benzene
Extractive Value (% extractive yield)	8.885 ± 0.057	1.998 ± 0.059	1.497 ± 0.016	0.500 ± 0.064
Total phenolic content (mg GAE/ g dm)	72.544 ± 0.144	30.000 ± 0.555	26.410 ± 1.282	17.735 ± 0.407
Total flavonoid content (mg RE/ g dm)	12.930 ± 0.162	9.286 ± 0.040	8.480 ± 0.092	7.161 ± 0.053

Each value in the table was obtained by calculating the average of three experiments (n=3) and data are presented as Mean ± SEM.

Reducing property and FRAP

Reducing property is expressed as mg ascorbic acid equivalent (AAE)/ g dm. The reducing property of the solvent extract can be ranked as 70% ethanol > Methanol > Chloroform> Benzene (Table 2). Reducing activity in 70% hydro ethanol (27.430 ± 0.508 mg AAE/ g dm) and methanol extract (24.257 ± 0.181 mg/ g dm) were comparable. Reducing property was also studied by FRAP and is expressed as µ mole trolox equivalent (TE)/ g dm and is represented in Table 2. The results indicate that both hydro ethanol extract (2.196 ± 0.002 µ mole TE/ g dm) and methanol extract (2.142 ± 0.004 µ mole TE/ g dm) showed almost comparable results.

Table 2: Reducing activity (mg AAE/ g dm), FRAP (µ mole TE/ g dm) and Metal chelating activity (% inhibition/ g dm) of *C. compressus* in different solvent system

Parameters	Extracts of *C. compressus* in different solvent system			
	70% ethanol	Methanol	Chloroform	Benzene
Reducing activity (mg AAE/ g dm)	27.430 ± 0.508	24.257 ± 0.181	18.043 ± 1.255	11.329 ± 0.242
FRAP (µ mole TE/ g dm)	2.196 ± 0.002	2.142 ± 0.004	1.496 ± 0.001	0.466 ± 0.003
Metal chelating activity (% inhibition/ g dm)	40.295 ± 0.246	33.171 ± 0.373	27.729 ± 0.229	15.343 ± 0.768

Each value in the table was obtained by calculating the average of three experiments (n=3) and data are presented as Mean ± SEM.

Metal chelating property

Metal chelating ability is represented as % inhibition of metal ions/ g dm and is represented in Table 2. The hydro-ethanol extract (40.295 ± 0.246 %) showed promising metal chelating property (Table 2).

Radical scavenging property

The antioxidant capacities using DPPH and ABTS assays of various extracts from *C. compressus* are shown in Tables 3. In the DPPH method, the free radical scavenging capacities of the plant studied ranged between 4 – 22% for *C. compressus*. In the ABTS method, the free radical scavenging capacities of the plant studied ranged between 5 – 55% for *C. compressus*. Maximum radical scavenging activity was observed with 70% ethanol and the activity decreased with decreasing polarity.

Table 3: Radical scavenging activity (% inhibition/ g dm) of *C. compressus* in different solvent system using DPPH and ABTS

Radical Scavenging Property	Extracts of *C. compressus* in different solvent system			
	70% ethanol	Methanol	Chloroform	Benzene
DPPH	22.923 ± 0.328	13.907 ± 0.154	5.563 ± 0.043	4.450 ± 0.300
ABTS	55.243 ± 0.271	54.518 ± 0.226	26.172 ± 0.154	5.371 ± 0.128

Each value in the table was obtained by calculating the average of three experiments (n=3) and data are presented as Mean ± SEM.

Anti-lipid peroxidation property

Of the four different extracts compared maximum activity was observed in 70% hydro ethanol and methanol extract for both the plants studied. Based on this anti-lipid peroxidation assay was studied in hydro ethanol and methanol extract and is represented in Table 4 as % inhibition of lipid peroxidation. Amongst the several factors that lead to deteriorated food products, one of the most concerned is lipid auto-oxidation**.**

Table 4: Anti-lipid peroxidation assay (% inhibition/ g dm) of *C. compressus* in different solvent system

% inhibition of lipid peroxidayion	**Extracts of *C. compressus* in different solvent system**	
	70% ethanol	Methanol
	29.133 ± 0.003	20.536 ± 0.397

Each value in the table was obtained by calculating the average of three experiments (n=3) and data are presented as Mean ± SEM.

RP-HPLC based qualitative and quantitative profiling of some pharmacologically important phenolic acids and flavonoids from hydroethanolic extracts of *C. compressus*

A typical HPLC chromatogram of the mixture of twenty one standard phenolic acids and flavonoids (mentioned in Materials and Methods) recorded at 280 nm is presented in fig. 3. The 70% ethanol extract of *C. compressus* revealed the presence of vanillic acid, ferulic acid, rutin, myricetin, quercetin and apigenin at 260 nm as depicted in chromatogram in Fig. 4. The quantification data of phenolics and flavonoids in *C. compresus* as represented in Table 5 represented as mg/ 100g dm.

Table 5: Phenolic acids and flavonoids in 70% ethanol extract of *C. compressus* (mg/ 100g dm)

Phenolic acids and flavonoids	**Amount mg/100gm dry plant material**	**Phenolic acids and flavonoids**	**Amount mg/100gm dry plant material**	**Phenolic acids and flavonoids**	**Amount mg/100gm dry plant material**
Gallic acid	ND	Caffeic acid	ND	Rutin	0.761 ± 0.002
Protocatechuic acid	ND	Syringic acid	ND	Ellagic acid	ND
Gentisic acid	ND	p-Coumaric acid	ND	Myricetin	2.607 ± 0.001
p-Hydroxy benzoic acid	ND	Ferulic acid	0.218 ± 0.001	Quercetin	0.055 ± 0.001
Catechin	ND	Sinapic acid	ND	Naringenin	ND
Chlorogenic acid	ND	Salicylic acid	ND	Apigenin	2.163 ± 0.001
Vanillic acid	0.190 ± 0.001	Naringin	ND	Kaempferol	ND

Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ± SEM

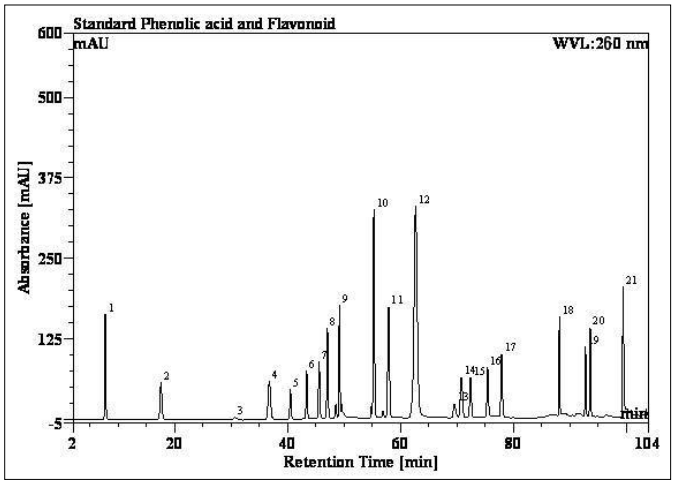

Figure 3: HPLC Chromatogram of mixture of Standard phenolic acids and flavonoids

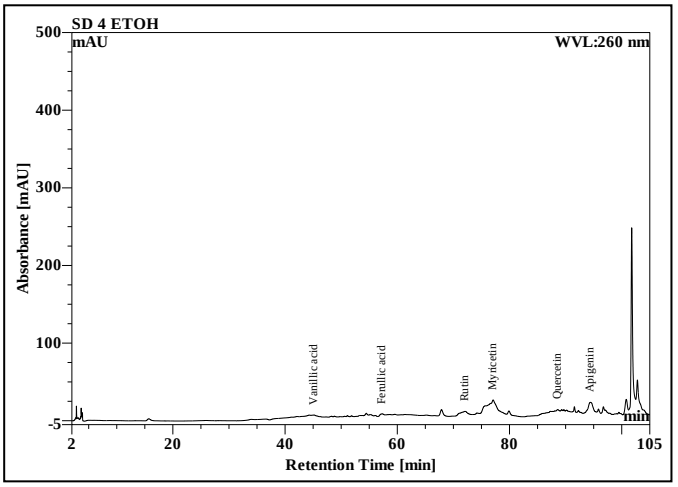

Figure 4: HPLC chromatogram of fruits of *C. compressus* showing phenolic acids and flavonoids

Quantification of water soluble vitamins

Fig 3 represents HPLC chromatogram of the all standard vitamin mixture recorded at 275 nm. The vitamin content is depicted in Table 6 represented as mg/ 100g dry plant material. The HPLC chromatogram is represented in fig. 4 and shows clear separation at 275 nm.

Table 6: Comparative account of water soluble vitamins (mg/ 100 g dry plant material)

C	**B1**	**B2**	**B3**	**B5**	**B6**	**B9**
ND	0.016 ± 0.001	0.431 ± 0.005	0.229 ± 0.008	ND	0.028 ± 0.001	0.09 ± 0.001

Each value in the table was obtained by calculating the average of three experiments (n=3) and data are presented as Mean ± SEM.

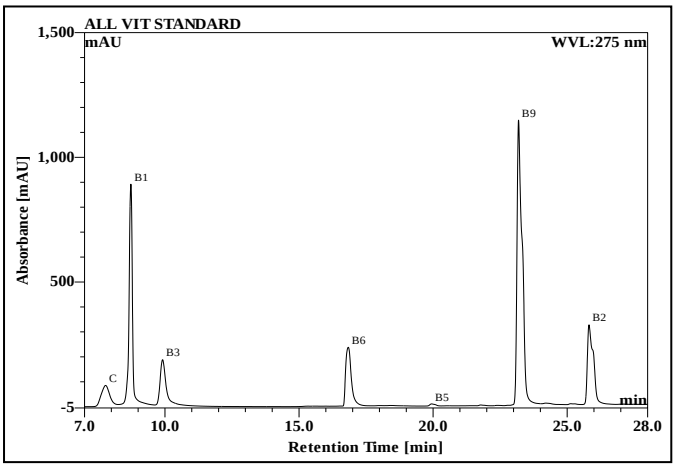

Figure 3: HPLC Chromatogram of mixture of Standard vitamin (C)Ascorbic acid; (B1) Thiamine; (B3) Niacin; (B6) Pyridoxine; (B5)Pantothenic acid; (B9) Folic acid; (B2) Riboflavin.

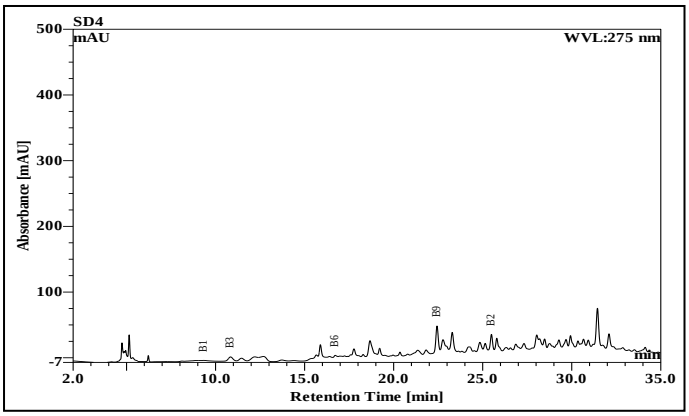

Figure 4: HPLC chromatogram of fruits of *C. compressus* showing water soluble vitamins Thiamine; (B3) Niacin; (B6) Pyridoxine; (B5) Pantothenic acid; (B9) Folic acid; (B2) Riboflavin.

DISCUSSION

The polarity of the solvent, nature of chemical present and parts used for extraction are the important factors that affect extraction yield [17]. Based on our results reported here, the highest extraction yield for antioxidants was found with hydro-alcoholic solvents. This indicates that most of the components in *C. compressus* are water-soluble. Chloroform and benzene appeared to be less effective in extracting antioxidant compounds from the plants studied. Addition of water to pure solvent increased the polarity of ethanol and facilitated the extraction of antioxidant compounds of wide range of polarity [18].Hydro-alcohol has been reported to be more efficient for extraction of antioxidant over pure solvent system [19]. Antioxidant activity was maximum 80 % aq. methanol from plant materials like rice bran, wheat bran, citrus peel, coffee beans and guava leaves [20]. Absolute methanol proved to be a better extracting solvent in case of wild leafy vegetables [21] and wild edible fruits [22]. In the present analysis, the variation in the extract yields from the plant sample differed due to different polarity of the polyphenolic or other antioxidant compounds or their association with other molecules [23]. The amount of the antioxidant is also affected by extraction procedure, temperature and time [24-25].

Polyphenols particularly the flavonoids have the ability to scavenge ROS and also to chelate the transition metal ions necessary for the generation of OH· through fenton reaction [26-27]. It is also suggested that polyphenolic compounds through their redox-regulatory property can inhibit cancer [28]. The maximum phenolic content in the hydro ethanol extract of *C. compressus* indicated the high polarity of the phenolics. Results of the present study showed that among all the solvent extracts; the hydro-ethanol extracts had the highest phenolic content and it decreased with decreasing polarity. Polar solvents such as aqueous methanol/ethanol are often better solubiliser of phenolic compounds as compared with absolute methanol/ethanol [20-22, 24-25]. However, non-polar solvent like ethyl acetate have also been demonstrated to extract phenolic compounds in higher amounts from onion and citrus peel [29-32].

Flavonoids are one of the widely distributed group of phenolic compounds responsible for defence and antioxidant activity of the plants. Flavonoids show wide array of pharmacological and biochemical activities including radical scavenging properties [33-35]. The flavonoid content was maximum in 70% ethanol and it decreased with solvent polarity. The flavonoid content was maximum in 70% ethanol and it decreased with solvent polarity. Similar observation was made in case of *Torilis leptophylla* where the highest amount of flavonoid content was analyzed in ethanolic extract, followed by that in methanol and benzene [36]. The quantity of flavonoids from the solvent extract correlates to the nature of flavonoids present in the plant sample.

The presence of phytonutrients with antioxidant properties in the plant sample reduces Fe^{+3} to Fe^{+2} which is the reflection of the antioxidant potential of the sample. Fe^{+3} promotes free radical formation by Fenton reaction and also facilitates lipid peroxidation. The reducing property indicates that the plant sample can reduce the intermediates of lipid peroxidation processes through electron donation [37]. Highest reducing property is observed in the hydro alcoholic extracts of both the plants which can be attributed to its high content of phenolics in those extracts. Maximum FRAP activity was also observed to be highest in the 70% ethanol extract. The reducing power generally corresponds to high phenolic content [38].

Metal ions are major initiators of lipid peroxidation that results in deterioration of food [39]. The catalysis of metal ions also contributes to free radical formation that correlates with incidents of degerative diseases [40]. The most effective transition metal ions working as pro-oxidants in plant tissue are the ferrous ions [41]. In the present study, the chelating ability of *C. compressus* extracts was investigated. Chelating capacity of the extracts decreased with the decreasing polarity. Chelating ability can be attributed to the high phenol and flavonoid content in these extract.

Maximum radical scavenging activity was observed with 70% ethanol and the activity decreased with decreasing polarity. Methanol extracts showed maximum radical scavenging activity in fruit powder of citron and blood orange followed by acetone and ethanol extracts [42]. Similar observations were made for green coffee extracts where methanol extract exhibited maximum radical scavenging activity followed by acetone [43]. The data suggests that polar components like phenol and flavonoids of the plant studied contributed to the radical scavenging activity. The flavonoid aglycones would give higher antioxidant capacity than flavonoid glycosides [44].

Lipid auto-oxidation is the major factors that results in rancidity of fat based food products and decrease shelf life of food [45]. To protect food against oxidative degradation food additives are used. Synthetic

antioxidants, e.g., BHT, BHA, are added in food products to suppress lipid peroxidation for quality improvement and stability. Synthetic antioxidants on the other hand have several side effects. The phenolic compounds and other chemical components may suppress lipid peroxidation through by inactivating the free alkyl radicals and scavenging the metal ions [46]. In addition, lipid peroxidation of cell membrane is associated with various pathological events such as atherosclerosis, inflammation and liver injury [47]. So addition of plant based phytonutrients reduces the risk of side effects and consumption of which is also beneficial to human.

Rutin is a flavonol with biological effects, such as antidiabetic effect [48] and anticancer activity [49] and can potentially be used as a therapeutic agent. Vanillic acid is used as a flavouring agent. Studies also indicate that vanillic acid has a hepatoprotective effect through its suppressive action on immune-mediated liver inflammation in concanavalin A-induced liver injury [50]. Ferulic acid is present in both the plant extract and is known for its anti-microbial, anti-inflammatory, anti-cancer activities etc. Presence of ferulic acid justifies the application of both the plant on cuts and wounds. Myricetin has been proven to posses anticancer, antidiabetic and anti-inflammatory activity [51]. Quercetin display anti-cancer [52] anti-histamine, as also anti- degenerative properties [53], that is primarily due to their antioxidant activities. Apigenin, a flavone, reduce the risk of several degerative disorders including neurological, cardiac, and gut [54].

Thiamine (B1) is essential for energy production, carbohydrate metabolism and nerve cell function [55]. Amount of B1 quantified is (0.016 ± 0.001 mg/100g). Thiamine has been shown to occur in some common vegetables like beans (0.132mg/100gm), cauliflower (0.073 mg/100gm), spinach (0.076mg/100gm) [56]. *C. Compressus* contained maximum amount of Riboflavin (B2) (0.431 ± 0.005 mg/100g).The data obtained from this study is comparable to some common vegetables like spinach (0.24 mg/100g), green beans (0.12 ± 2 mg/100g, potato (0.023 ±1 mg/100g) [56]. Vitamin B3 (0.229 ± 0.008 mg/100g), B6 (0.028 ± 0.001 mg/100g) and B9 (0.090 ± 0.001 mg/100g) is also found in good amount. Vitamin B3 plays an important role in DNA repair and fat metabolism [55]. Folic acid (B9) plays an important role in DNA synthesis and repair [55]. It has been observed that vitamin B6 possess strong antioxidant activity in linoleic acid based lipid peroxidation. Vitamin B1, B2 and B9 though acted as prooxidants at initial stages of lipid peroxidation but acted as strong antioxidant in later stages [57]. Based on the above data it is suggested that *C. Compressus* act as a good source of food preservative at the same time a considerable good source of vitamin B.

CONCLUSION

Based on the above findings it can be ascertained that *C. Compressus* can be exploited as a source of natural antioxidant and vitamin B. Further, the knowledge of antioxidant and nutraceutical potential of the leaf tissue will be useful in designing strategies that will maximize the effective and practical utilization of *Cyperus compressus* L.

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REFERENCES

1. Sowndharajan K, Jinse MJ, Arunachalam K, Manian S. Evaluation of *Merremia tridentates* (L) Hallier f. for in vitro antioxidant activity. Food Science and Biotechnology 2010; 19(3):663-669.
2. Halliwell B. Antioxidants and human diseases: a general introduction. Nutrition Reviews 1997; 55:44-52.
3. Madsen HL, Bertelsen G. Spices as antioxidants. Trends Food Science Technology 1995; 6:271-277.
4. Padmaja M, Sravanthi M, Hewalath KPJ. Phytopharmacoloy, Evaluation of Antioxidant activity of 2 Indian medicinal plant. Journal of phytology 2010; 3(3):86-91.
5. Kilani-Jaziri S, Bhouri W, Skandrani I, Limem I, Chekir LG, Ghedira K. Phytochemical, antimicrobial, antioxidant and antigenotoxic potentials of *Cyperus rotundus* extracts. African Journal of Botany 2011; 77:767-776.
6. Dangol DR, Gurung SB. Ethnobotany of Tharu tribe of Chitwan district, Nepal. International Journal of Pharmacology 1991; 29:203-209.
7. Morimoto M, Fujii Y, Komai K. Antifeedants in Cyperaceae: Coumarins and quinines from *Cyperus* spp. Phytochemistry 1999; 51:605-6089.
8. Singleton VL, Rossi JA. Colorimetry of total phenolics with Phosphomolybdic-phosphotungstic acid reagents. American Journal of Enology and Viticulture 1965; 16:144-158.
9. Ordonez AAL, Gomez JG, Vattuone MA, Isla MI. Antioxidant activities of *Sechium edule* (Jacq.) Swart extracts. Food Chemistry, 2006; 97:452-458.
10. Oyaizu M. Studies on product on browning reaction prepared from glucose amine. Japan Journal of Nutrition 1986; 44:307-315.
11. Benzie IF, Strain JJ. The ferric reducing ability of plasma (FRAP) as a measure of ‘‘antioxidant power’’: the FRAP assay. Analytical Biochemistry 1996; 239:70-76.
12. Blois MS. Antioxidant determination by the use of of a stable free radical. Nature 1958; 181:1199-1200.
13. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cationdecolorization assay. Free Radical Bio Medicine 1999; 26:1231-1237.
14. Lin SC, Chang CMJ, Deng TS. Enzymatic Hot Pressurized Fluids Extraction of Polyphenolics from *Pinus taiwanensis* and *Pinus morrisonicola*. Journal of Taiwan Institute of Chemical Engineers 2009; 40:136-142.
15. Amabye TG. Evaluation of Physiochemical, Phytochemical, Antioxidant and Antimicrobial Screening Parameters of *Amaranthus spinosus* leaves. Natural Product Chemistry Research 2015; 4:199-203.
16. Seal T, Chaudhuri K, Pillai B. Nutraceutical and antioxidant properties of *Cucumis hardwickii* Royle: A potent wild edible fruit collected from Uttarakhand, India. Journal of Pharmacognosy and Phytochemistry 2017; 6(6):1837-1847.
17. López A, Rico M, Rivero A, de Tangil MS. The effects of solvents on the phenolic contents and antioxidant activity of *Stypocaulon scoparium* algae extracts. Food Chemistry 2011; 125:1104-1109.
18. Uma DB, Ho CW, Wan WM, Aida WM. Optimization of extraction parameters of total phenolic compounds from Henna (*Lawsonia inermis*) leaves. Sains Malaysiana 2010; 39(1):119-128.
19. Spigno G, Tramelli L, De Faveri DM. Effects of extraction time, temperature and solvent on concentration and antioxidant activity of grape marc phenolics. Journal of Food Engineering 2007; 81:200-208.
20. Anwar F, Jamil A, Iqbal S, Sheikh MA. Antioxidant activity of various plant extracts under ambient and accelerated storage of sunflower oil. Grasses Aceites Sevilla 2006; 57:189-197.
21. Seal T, Pillai B, Chaudhuri K. Effect of solvent extraction system on the antioxidant activity of some selected wild leafy vegetables of Meghalaya state in India. International Journal of Pharmaceutical Science and Research 2013a; 4(3):1046-1051.
22. Seal T, Chaudhuri K, Pillai B. Effect of solvent extraction system on the antioxidant activity of some selected wild edible fruits of Meghalaya state in India. Journal of Chemical and Pharmaceutical Research 2013b; 5(1):276-282.

23. Hsu B, Coupar IM, Ng K. Antioxidant activity of hot water extract from the fruit of the Doum palm, *Hyphaene thebaica*. Food Chemistry 2006; 98:317-328.
24. Siddhuraju P, Becker K. Antioxidant properties of various extracts of total phenolic constituents from three different agroclimatic origins of drumstick tree (*Moringa oleifera* Lam.) leaves. Journal of Agricultural and Food Chemistry 2003; 51:2144-2155.
25. Sultana B, Anwar F, Przybylski R. Antioxidant activity of phenolic components present in barks of barks of *Azadirachta indica*, *Terminalia arjuna*, *Acacia nilotica*, and *Eugenia jambolana* Lam. trees. Food Chemistry 2007; 104:1106-1114.
26. Sahreen S, Khan MR, Khan RA. Evaluation of antioxidant activities of various solvent extracts of Carissa opaca fruits. Food Chemistry 2010; 122:1205-1211.
27. Florence OJ, Adeolu AA, Anthony JA. Comparison of the nutritive value, antioxidant and antibacterial activities of *Sonchus asper* and *Sonchus oleraceus*. Records of Natural Products 2011; 5(1):29-42.
28. Gursoy N, Sarikurkcu C, Cengiz M, Solak MH. Antioxidant activities, metal contents, total phenolics and flavonoids of seven *Morchella* species. Food and Chemical Toxicology 2009; 47:2381-8.
29. Abdille MH, Singh RP, Jayaprakasa GK, Jens BS. Antioxidant activity of the extracts from *Dillenia indica* fruits. Food Chemistry 2005; 90:891-896.
30. Peschel W, Sanchez-Rabaneda F, Dn W, Plescher A, Gartzia I, Jimenez D, *et al*. An industrial approach in the search of natural antioxidants from vegetable and fruit wastes. Food Chemistry 2006; 97:137-150.
31. Rehman ZU. Citrus peel extract- A natural source of antioxidant. Food Chemistry 2006; 99:450-454.
32. Li Y, Guo C, Yang J, Wei J, Xu J, Cheng S. Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. Food Chemistry 2006; 96:254-260.
33. Bravo L. Polyphenols: chemistry, dietary sources, metabolism and nutritional significance. Nutrition Reviews 1998; 56:317-333.
34. Pourmorad F, Hosseinimehr SJ, Shahabimajd N. Antioxidant activity, phenol and flavonoid contents of some selected Iranian medicinal plants. African Journal Biotechnology 2006: 5(11):1142-1145.
35. Prasad KN, Yang B, Dong X, Jiang G, Zhang H, Xie H. Flavonoid contents and antioxidant activities from Cinnamomum species. Innovative Food Science and Emerging Technology 2009; 10:627-32.
36. Saeed N, Khan MR, Shabbir M. Antioxidant activity, total phenolic and total flavonoid contents of whole plant extracts *Torilis leptophylla* L. BMC Complementary and Alternative Medicine 2012; 12:221-232.
37. Yen GC, Chen HY. Antioxidant activity of various tea extracts in relation to their antimutagenicity. Journal of Agriculture and Food Chemistry 1995; 43:27-32.
38. Seal T. Antioxidant Activities of some Wild Vegetables of North-Eastern Region in India and Effect of Solvent Extraction System. 2014; 6(5):315-319.
39. Gordon MH. The mechanism of antioxidant action in vitro. In: Hudson BJF (ed), Antioxidants. Elsevier Applied Science, London, New York, 1990, 1-18.
40. Halliwell B, Murcia HA, Chirco S, Aruoma OI. Free radicals and antioxidants in food an in vivo: what they do and how they work. CRC Critical Reviews in Food Science 1995; 35:7-20.
41. Yamaguchi T, Takamura H, Matoba T, Terao J. HPLC method for evolution of the free radical-scavenging activity of foods by using 1,1-dicrylhydrazyl. Bioscience Biotechnology and Biochemistry 1998; 62:1201-1204.
42. Jayaprakasha GK, Patil BS. *In vitro* evaluation of the antioxidant activities in fruit extracts from citron and blood orange. Food Chemistry 2007; 101:410-418.
43. Ramalakshmi K, Kubra IR, Rao JM. Antioxidant potential of low-grade coffee beans. Food Research International 2008; 41:96-103.
44. Heim KE, Tagliaferro AR, Bobilya DJ. Flavonoid antioxidants: Chemistry, metabolism and structure-activity relationships. Journal of Nutrition and Biochemistry 2002; 13(10):572-84.
45. Gutensperger B, Hammerli-Mier DE, Escher FE. Rosemary extract and precooking effects on lipid oxidation in heat sterilized meat. Journal of Food Science 1998; 63:955-957.
46. Mathew S, Abraham TE. *In vitro* antioxidant activity and scavenging effects of *Cinnamomum verum* leaf extracts assayed by different methodologies. Food chemistry and Toxicology 2006; 44:198-206.
47. Singh HP, Kaur S, Negi K, Kumari S, Saini V, Batish DR. Assessment of in-vitro antioxidant activity of essential oil of Eucalyptus citriodora (lemon-scented Eucalypt; Myrtaceae) and its major constituents. LWT Food science and Technology 2012; 48:237-241.
48. Srinivasan K, Kaul CL, Ramarao P. Partial protective effect of rutin on multiple low dose streptozotocin induced diabetes in mice. Indian Journal of Pharmacology 2005; 37:327-328.
49. Lin JP, Yang JS, Lin JJ, Lai KC, Lu HF, Ma CY, *et al*. Rutin inhibits human leukemia tumor growth in a murine xenograft model in vivo. Environmental Toxicology 2012; 27(8):480-484.
50. Kim SJ, Kim MC, Um JY, Hong SH. The beneficial effect of vanillic acid on Ulcerative Colitis. Molecules. 2010; 15:7208-7217.
51. Ong KC, Khoo HE. Biological Effects of Myricetin. General Pharmacology 1997; 29(2):121-126.
52. D'Andrea G. Quercetin: A flavonol with multifaceted therapeutic applications. Fitoterapia 2015; 106:256-71.
53. Seal T. HPLC determination of phenolic acids, flavonoids and ascorbic acid in four different solvent extracts of *Zanthoxylum acanthopodium*, a wild edible plant of Meghalaya. International Journal of Pharmacy and Pharmaceutical Sciences 2016; 8(3):103-109.
54. Mohammad A, Elham KK. Medicinal uses and chemistry of flavonoid contents of some common edible tropical plants. Journal of Paramedical Science 2013; 4:119-38.
55. FAO/WHO. Human vitamin and mineral requirements. Report of a joint FAO/ WHO expert consultation, Bangkok, Thailand, 2001.
56. Watada AE. Vitamins C, B9 and B2 contents of stored fruits and vegetables as determined by High Performance Liquid Chromatography. Journal of the American Society of Horticultural Science 1987; 112(5):794-797.
57. Higashi-Okai K, Nagino H, Yamada K, Okai Y. Antioxidant and prooxidant activities of B group vitamins in lipid peroxidation. Journal of University of Occupational and Environmental Health 2006; 28(4):359-368.

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