Plant & Fungal Research (2024) 7(2): 2-12
https://doi.org/10.30546/2664-5297.2024.7.2.50.01

December 2024

Comparison of nutritional, phytochemical and bioactive profiles of seeds of the wild forage legume *Centrosema virginianum*

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Abstract: The present study envisages assessing the nutraceutical and bioactive potential of non-germinated and germinated seeds of the wild forage legume *Centrosema virginianum* (L.) Benth. occurring in southwest India. Both types of seeds possess high amounts of carbohydrates and lipids, while proteins, fiber and calorific value are moderate. Qualitative assessment revealed that non-germinated seeds possess more phytochemicals than germinated seeds. Non-germinated seeds possess significantly higher total phenolics and tannins in chloroform extract than methanol extract, whereas this was reversed in flavonoids and vitamin C. In germinated seeds, total phenolics were significantly higher in chloroform than in methanol extract. GC-MS analysis revealed palmitic acid and methyl 9-octadecenoate as major compounds, while ascorbic acid and dotriacontyl isopropyl ether were found in germinated seeds. Among the pigments in acetone extracts of non-germinated seeds, chlorophyll was significantly higher in germinated seeds than non-germinated seeds, while it was reversed in the carotenoids, β-carotene and lycopene. Total antioxidant activity was significantly higher in chloroform extract than methanol extract in non-germinated seeds, while it was reversed in reducing power, ferrous ion chelating capacity and DPPH radical-scavenging activity. In germinated seeds, total antioxidant activity was significantly higher in chloroform than methanol extract, while it was opposite in ferrous ion-chelating capacity. This study strengthened the forage properties of the little-known pasture legume, *C. virginianum*. As this legume is endangered, future studies should focus on its conservation and usefulness in livestock nutrition.

Keywords: *antioxidants, fodder, germinated seeds, legume pasture, pigments, proximal qualities*

INTRODUCTION

Legumes belong to the Fabaceae family, the third largest of flowering plants [Jianhui et al., 2023]. This family consists of several plants that are major food sources such as beans, lentils, peanuts, peas and other pod-bearing plants [Messina, 1999]. Edible legumes are well-known for their richness in fiber, phytochemicals, proteins and vitamins with significant biological activities. Legumes also deliver a range of essential nutrients, such as carbohydrates, dietary fibers, minerals, proteins and vitamins. They are a rich source of proteins compared to cereals owing to the mutualistic association of nitrogen fixing bacteria (e.g., *Rhizobium* spp.) in the roots [Kouris-Blazos, Belski, 2016].

Legumes also provide ample opportunities for the sustainable development of grassland-based livestock production. They contribute to abundant forage yield by substituting input of inorganic nitrogenous fertilizers. They are well adapted to climate change, like drought, heavy rainfall, warmer temperatures, increasing the nutritive value of herbage and raising the efficiency of the conversion of herbage into animal protein [Bär et al., 2020]. Some herbaceous plants frequently employed for animal feeding include *Medicago sativa* L. (alfalfa), *Trifolium pratense* L. (red clover) and *T. repens* L. (white clover). They are also used in ethnic medicine due to their bioactivities, such as anti-inflammatory, antimicrobial, diuretic and estrogenic effects. Application of their active metabolites has been investigated in a number of studies [Ahmed, Zeb, 2020; Parham et al., 2020]. Fabaceae members are the richest in metabolites, with several biologically active compounds (e.g., alkaloids, carotenoids, coumarins, cardiac glycosides, flavones, isoflavones, phenols, phytocyanins, saponins, sterols, terpenoids and tannins with other phenolics) [Tava et al., 2019, 2022; Abhisheka et al., 2022, 2023]. Some species, although restricted geographically, represent precious feed resources [Piano, Pecetti, 2010].

Against the backdrop of climate change, many little-known but useful legumes in the human and

[1]E-mail: munirozturk@gmail.com
Received: 24.09.2024; Received in revised form 07.11.2024; Accepted: 29.11.2024

animal diet could be thoroughly investigated as alternatives to the currently used food and fodder legumes. Their seeds possess sufficient proteins with essential amino acids and carbohydrates to cater to the needs of livestock. For example, alfalfa is used as cubes, fodder, hay, pasture and silage for animals, and it possesses compounds like carotenoids, coumestrants, condensed tannins, flavonoids, saponins and tocols [Sen et al., 1998]. The important substances in legumes are chalcones, flavonoids (e.g., flavonones and isoflavones), which are a group of phytoestrogens [Panche et al., 2016]. Flavonoids are known to substitute estrogen in the postmenopausal period and they can be replaced by natural legume compounds like isoflavones, which are known to reduce the risk as well as side effects of using long-term synthetic or replacement hormones.

Among the less-known legumes, *Centrosema virginianum* (L.) Benth. (butterfly pea) is a perennial climber native to different ecosystems in the southeast United States. *C. virginianum* is also native to Puerto Rico as well as the Virgin Islands. It has one of the highest nitrogen-fixing potentials. This plant has been widely spread in different parts of the world, including the Indian subcontinent. It has been distributed in many habitats including the Western Ghats, disturbed ecosystems, coastal islands, coastal sand dunes, mangroves and shoreline habitats in southwest India. It has been red listed under the category of endangered species. This study emphasizes the importance of the nutritional, phytochemical and antioxidant profiles of non-germinated and germinated seeds of *C. virginianum*.

MATERIAL AND METHODS

Sample collection and processing. Dried pods of *C. virginianum* (Fig. 1) were collected from three locations in the lateritic scrub jungles of the southwest of Karnataka (12°48'N, 74°55'E; 110 m asl) during February 2023. The seeds of *C. virginianum* generally resemble the black gram (*Vigna mungo* (L.) Hepper), but they are flat with different surface shades. The seeds from each collection were separated from the pods (Fig. 2), dried at 50±2°C (until attaining constant weight), powdered using a hand grinder and stored in glass containers. About 5% of aborted and damaged seeds were removed. Seeds from each collection were allowed to germinate on a wet cotton bed. After radical emerges, 2-3 days germinated seeds (Fig. 2) were harvested and sun-dried until they attain moisture <10%, then powdered using a hand grinder and stored in glass containers. They have no dormancy and easily germinate up to 80-90% within 2-3 days. A schematic flow chart of the methodology followed to study *R. versatilis* is represented in Fig. 3.

Figure 1. Morphological characteristics of *Centrosema virginianum*. Scale bar, 1 cm.

Figure 2. Non-germinated (a) and germinated (b) seeds of *Centrosema virginianum*. Scale bar, 1 cm.

Proximal analysis. Proximal qualities such as moisture, protein, total lipids, fiber, ash, carbohydrates (in %) and energy (kJ/100 g dry weight) in dried seed flours of non-germinated and germinated seed samples were determined in three replicates [Abhisheka et al., 2022]. Moisture, crude fiber and ash contents were assessed gravimetrically [AOAC, 1999]. Crude protein was assayed by Lowry's method [Lowry et al., 1951]. Total lipids were estimated based on the method by J. Folch et al. [1957]. Total carbohydrates (%) were calculated using a formula proposed by H.G. Müller and G. Tobin [1980]. The colorific value was calculated based on a formula by S. Ekanayake et al. [1999].

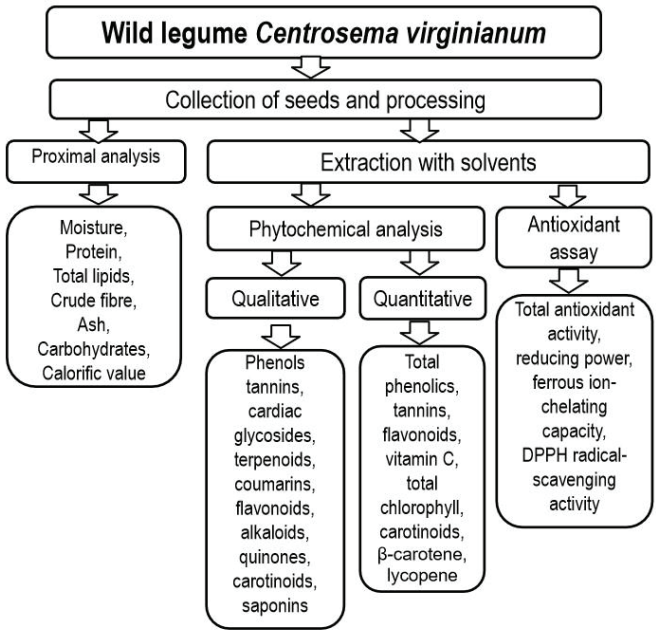

Figure 3. Schematic presentation of the assessment of the seeds of *Centrosema virginianum.*

Qualitative phytochemical analysis. The qualitative composition of biochemical constituents was assessed [Abhisheka et al., 2022]. Five grams of seed flour samples were extracted using two solvents, namely chloroform (30 ml) and methanol (40 ml), on a rotary shaker (150 rpm), for 12 h at room temperature (26±2ºC), then overnight at low temperature (8-10ºC). The extracts were centrifuged to collect the supernatants for qualitative analysis of bioactive components – alkaloids, cardiac glycosides, carotenoids, coumarins, flavonoids, phenols, quinones, saponins, tannins and terpenoids using chemical reactions. The formation of froth and emulsion was studied to ascertain the presence of saponins [Trease, Evans, 1989; Safowora, 1993; Herborne, 1998; Parekh, Chanda, 2007; Soares et al., 2013].

Quantitative phytochemical analysis. Total phenols, tannins, flavonoids and vitamin C of non-germinated and germinated seed flours were determined in three replicates [Abhisheka et al., 2023]. Total phenols extracted with 50% ethanol were determined following the procedure of Rosset et al. [1982] and using gallic acid as the standard J. Rosset et al. [1982]. Gallic acid dissolved in methanol served as a standard to quantify total phenolics in mg gallic acid equivalents (mg GAEs/g). The tannin content of seed samples was determined according to R.Burns [1971]. Tannic acid dissolved in methanol served as a standard to quantify tannin content in mg tannic acid equivalents (mg TAEs/g). The flavonoid content of seed samples was determined by an aluminium chloride colorimetric procedure [Chang et al., 2002]. The standard used was quercetin dihydrate absorbance determined at 415 nm to quantify flavonoid content in mg equivalents per g of seed sample (mg QEs/g). The vitamin C content in seed samples was quantified according to J.H. Roe [1954]. L-Ascorbic acid is used for vitamin C quantification in mg ascorbic acid equivalent per g (mg AAEs/g).

Analysis of bioactive compounds. One gram of seed flour was extracted with ethyl acetate by agitating on a magnetic stirrer up to 8 h, centrifuged (3000 rpm for 15 min) and the supernatant was filtered using a millipore filter (0.2 µ). Bioactive compounds in ethyl acetate extracts of non-germinated and germinated seed flours were assessed using a gas chromatography-mass spectrophotometer (GC-MS: Scion 436-GC Bruker model coupled with a triple quadrupole mass spectrophotometer, Germany). The processes include the fused silica capillary column BR-5MS. Helium gas was used as a carrier at a constant flow (1 ml/min) and 2 µl was injected, split ratio 10:1. The oven temperature of the column was 110ºC hold for 3.5 min, up to 200ºC at the rate of 10ºC/min-no hold up to 280ºC at the rate of 5ºC/min-9 min hold. The injector temperature was 280ºC, while the inlet source temperature was 290ºC and 250ºC, respectively. The mass spectrometer was operated in the positive electron ionization (EI) mode at 70 eV. The solvent delay was 0–3.5 min and the total GC-MS running period was 39 min. The relative percent of compounds present was calculated based on the average peak area of the total areas.

Pigment analysis. Seed flours were extracted in acetone (80%) for the analysis of total chlorophyll by determining absorbance at 630 nm and 664 nm [Nagata, Yamashita, 1992]: Total chlorophyll (mg/100 mg) = $(11.85 \times A_{664}) - (1.54 \times A_{647}) - (0.08 \times A_{630})$. To determine carotenoids, non-germinated and germinated seed flour were extracted in acetone (90%) and read for absorbance (480, 510 and 750 nm) [Nagata, Yamashita 1992]: Carotenoids (mg/100 mg) = $7.6\ (A_{480} - A_{750}) - 1.49\ (A_{510} - A_{750})$. A ratio of 4:6 (v/v) 80% acetone/hexane mixture was used to analyze β-carotene and lycopene contents [Nagat, Yamashita 1992]. β-carotene and lycopene were absorbed at wave lengths of 453, 505 and 663 nm. The extract was centrifuged and the supernatant was taken to observe the absorbance at the respective wavelengths and calculate the quantity of pigments: β-carotene (mg/100 mg) = $(0.216 \times A_{663}) - (0.304 \times A_{505}) + (0.452 \times A_{453})$; Lycopene (mg/100 mg)

$= (0.0458 \times A_{663}) + (0.372 \times A_{505}) - (0.0806 \times A_{453})$.

Antioxidant analysis. Seed flours (0.01–0.05 mg) were extracted using methanol (40 ml) and chloroform (30 ml) on a shaker (150 rpm, 48 h) at laboratory temperature (26±2°C). After centrifuging (3000 rpm), the supernatant was transferred to a pre-weighed Petri plate and allowed to evaporate at laboratory temperature (26±2°C). The mass of the dried extract was determined gravimetrically and the extract was dissolved in a known quantity of chloroform and methanol (1 mg/ml) to assess different antioxidant properties.

The total antioxidant activity (TAA), reducing power (RP), ferrous ion-chelating capacity (FCC) and DPPH radical-scavenging activity (RSA) of chloroform and methanol extracts of seed flours were evaluated [Abhisheka et al., 2023). The total antioxidant activity (TAA) was assessed based on P. Prieto et al. [1999]. The TAA was recorded in µM equivalents of ascorbic acid per g (µM AAEs/g). The reducing power (RP) was determined based on Oyaizu [1986]. Absorbance was measured at 700 nm and higher absorbance denotes increased RP. The ferrous ion-chelating capacity (FCC) was determined based on C.L. Hsu et al. [2003]. The reagents devoid of extract served as control to determine the FCC: Ferrous ion-chelating capacity (%) = (1 - As562/ Ac562) × 100, where, As is the sample absorbance; Ac is the control absorbance. The DPPH radical-scavenging activity (RSA) was evaluated based on R.P. Singh et al. [2002]. The reagents devoid of extract served as a control and the absorbance was read (517 nm) to quantify the RSA: Free radical-scavenging activity (%) = (Ac517 - As517/Ac517) × 100 (where, Ac is the absorbance of the control; As is the absorbance of the sample).

Data analysis. The difference in proximal qualities, phytochemical constituents, pigments and antioxidant activities between non-germinated and germinated seeds was assessed using a t-test [Statistica, Version # 8, 2008].

RESULTS

Proximal qualities. Proximal features of flours of non-germinated and germinated seeds of *C. virginianum* on a dry mass basis are given in Table 1. The moisture content was higher in non-germinated and germinated samples ($p<0.05$). The protein content was not significantly higher in non-germinated than germinated seeds ($p>0.01$). Total lipid content was decreased in germinated seeds ($p<0.01$). Germination decreased the fiber content significantly ($p<0.001$). Ash content was lowered in germinated seeds ($p<0.05$). Carbohydrate contents were also higher in non-germinated seeds ($p<0.05$), as was the calorific value ($p<0.01$).

Usually, protein content decrease owing to the germination of seeds. As seen in *C. virginianum*, a decrease in protein content was seen in the seeds of *Canavalia maritima* Thouars occurring on the coastal sand dunes [Seena et al., 2005a, 2005b; D'Cunha et al., 2009a, 2009b]. Unlike the present study, the total lipid content increased in the germinated seeds of *C. maritima*. Similar to our data, a significant decrease in crude fiber was evident in *C. maritima*. Significant loss of ash quantity reflects the loss of several micro- and macrominerals. The decrease in carbohydrates as well as calorific value is reasonable owing to the utilization of carbohydrates as an energy source for germination and growth.

Phytochemical constituents. Among the 10 classes of phytochemicals qualitatively evaluated in *C. virginianum*, chloroform and methanol extracts of non-germinated seeds showed up to eight constituents (Table 2). In non-germinated seeds, chloroform extract was devoid of terpenoids and quinones, while in methanol extract, alkaloids and saponins were absent. Solvent extracts of germinated seeds showed a lower number of phytochemicals compared to non-germinated seeds. Saponins, quinones, carotenoids and coumarins were not present in chloroform as well as the methanol extract

Table 1. Proximal composition of seeds of *Centrosema virginianum* on a dry mass basis.

	Non-germinated seeds	Germinated seeds
Moisture (%)	7.23±0.26	8.51±0.31*
Proteins (%)	16.43±0.42**	11.6±0.44
Total lipids (%)	5.08±0.06**	4.26±0.13
Crude fiber (%)	3.10±0.04***	1.13±0.13
Ash (%)	2.31±0.03*	2.12±0.17
Carbohydrates (%)	77.96±0.20*	76.11±0.74
Calorific value (kJ/100 g)	408.1±0.63**	403.7±0.22

Note: n=3, mean±SD; t-test: *, 0.05; **, 0.001.

Table 2. Qualitative phytochemical assessment of chloroform and methanol extracts of non-germinated and germinated seeds of *Centrosema virginianum* (+, presence; -, absence).

	Non-germinated seeds		*Germinated seeds*	
	Chloroform	Methanol	Chloroform	Methanol
Alkaloids	+	-	+	+
Saponins	+	-	-	-
Tannins	+	+	+	+
Phenols	+	+	+	+
Terpenoids	-	+	-	+
Flavonoids	+	+	+	+
Quinones	-	+	-	-
Cardiac glycosides	+	+	+	-
Carotenoids	+	+	-	-
Coumarins	+	+	-	-
Total	8	8	5	5

of germinated seeds. Cardiac glycosides and terpenoids were present only in chloroform and methanol extracts of germinated seeds, respectively. Tannins, phenols and flavonoids were present in both the extracts of non-germinated and germinated seeds.

All considered chemical classes were represented either in chloroform or methanol extracts of non-germinated seeds, while only six of them were represented by germinated seeds. This shows that, there will be considerable modification in the phytochemicals, similar to the proximal features, between non-germinated and germinated seeds. It has also been partially reflected in the pigments of the seeds of *C. virginianum*.

Total phenolic content in *C. virginianum* was significantly higher in chloroform than methanol extract in non-germinated seeds, while it was opposite in germinated seeds ($p<0.001$) (Fig. 4). However, the chloroform extract of non-germinated seeds showed the highest peak of total phenolics. The total phenolic content of non-germinated as well as germinated seeds of *C. virginianum* is lower than the non-germinated seeds of maritime *Canavalia cathartica* as well as *C. maritima* [Niveditha, Sridhar, 2014].

Tannin content in *C. virginianum* was significantly higher in chloroform than methanol extract ($p<0.05$), while it was lower in germinated seeds without significant difference. However, the tannin contents of non-germinated seeds of *C. virginianum* are comparable with those of *C. cathartica* as well as *C. maritima* [Niveditha, Sridhar, 2014].

Flavonoids content in *C. virginianum* was significantly higher in methanol than chloroform extract of non-germinated seeds ($p<0.01$), while it was low and not significantly different in germinated seeds.

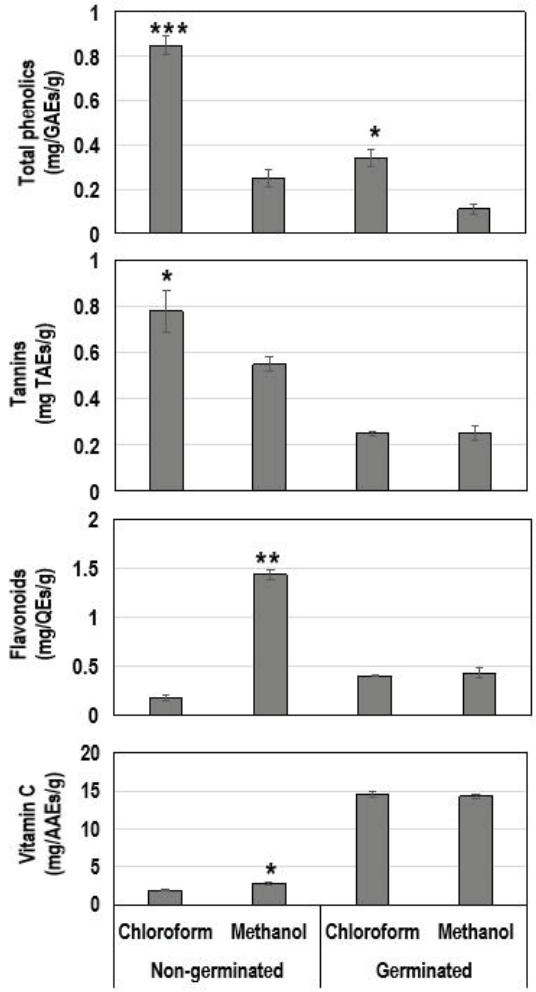

Figure 4. Total phenolics, tannins, flavonoids and vitamin C in chloroform and methanol extracts of seeds of *Centrosema virginianum* (n=3, mean±SD; t-test: *, $p<0.05$; **, $p<0.01$; ***, $p<0.001$).

Vitamin C content was higher in germinated seeds than in non-germinated seeds. Methanol extract of non-germinated seeds showed higher content of vitamin C than chloroform extract ($p<0.05$), while no significant difference was seen between extracts of germinated seeds. The vitamin C contents of the seeds of *C. virginianum* are several folds higher than those of *C. cathartica* as well as *C. maritima* [Niveditha, Sridhar, 2014].

Bioactive compounds and pigments. Ethyl acetate extracts of germinated seeds possess more compounds than non-germinated seeds (27 vs. 25) (Fig. 5). The compounds found between the type of seeds tested were not very overlapping. The major component found in non-germinated seeds was n-hexadecanoic acid (palmitic acid), followed by 9-octadecenoic acid, methyl ester, (E)- (methyl 9-octadecenoate); 9,12-octadecadienoic acid (Z,Z)-; γ-sitosterol and ethyl oleate (Table 3). Palmitic acid is a well-known component of dairy products, meat and oils [Carta et al., 2017]. It is used in the production of cosmetics, mouthfeel, soaps and several foodstuffs. 9,12-Octadecadienoic acid (Z,Z)-, methyl ester has been reported in *Arisaema tortuosum* (Wall.) Schott or cobra lily (Pubchem). The γ-sitosterol serves as an inhibitor of the complement component C1 complex and has the potential to treat diabetes. Ethyl oleate is used as a vehicle for intramuscular drug delivery. Stigmasterol is present in non-germinated seeds, which is an approved food additive in the United Kingdom as well as the European Union [Carbal et al., 2017].

The major components prevailing in germinated seeds were l-(+)-ascorbic acid 2,6-dihexadecanoate, followed by dotriacontyl isopropyl ether (Table 4). Ascorbic acid (vitamin C) is a potent antioxidant used extensively in products. Dotriacontyl isopropyl ether was one of the major compounds in the ethanolic extract of *Sargassum wightii* [Begum, Hemalatha, 2017].

Among the four pigments assessed, chlorophyll showed a significant increase in germinated seeds, while the other three pigments (carotenoids, β-carotene and lycopene) were significantly higher in non-germinated than germinated seeds (Fig. 6).

Antioxidant potential. Total antioxidant activity in *C. virginianum* was significantly higher in chloroform extract than in methanol extract in non-germinated seeds, while it was opposite in germinated seeds (Fig. 7). Such activities in chloroform extracts of non-germinated seeds and methanol extracts of germinated seeds are comparable with maritime *Canavalia cathartica* as well

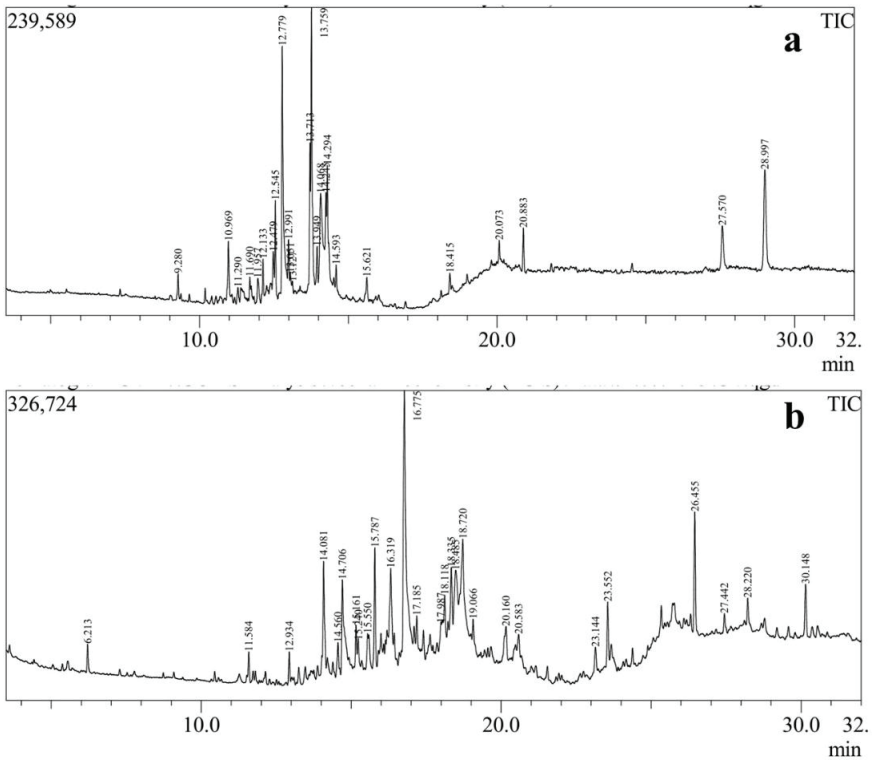

Figure 5. Chromatogram of GC-MS analysis of ethyl acetate extract of non-germinated (a) and germinated (b) seeds of *Centrosema virginianum*.

Table 3. Phytoconstituents identified in the ethyl acetate extract of non-germinated seeds of *Centrosema viginianum* by GC-MS analysis (major compounds are in bold).

Retention time (min)	Peak area (%)	Active principle
9.280	0.89	3-Ethyl-2,6,10-trimethylundecane
10.969	3.11	3-Ethyl-2,6,10-trimethylundecane
11.290	0.92	Hexane, 2,4,4-trimethyl-
11.690	0.89	Sulfurous acid, 2-ethylhexyl hexyl ester
11.957	1.76	2-Isopropyl-5-methyl-1-heptanol
12.133	1.77	Phthalic acid, hept-2-yl isobutyl ester
12.479	1.99	Silane, trichlorooctadecyl-
12.545	3.51	Hexadecanoic acid, methyl ester
12.779	**17.83**	**n-Hexadecanoic acid**
12.991	2.57	Pentadecanoic acid, ethyl ester
13.051	1.13	Decane, 1-iodo-
13.127	0.51	3-Decene, 2,2-dimethyl-, (E)-
13.713	5.25	9,12-Octadecadienoic acid (Z,Z)-, methyl ester
13.759	**14.25**	**9-Octadecenoic acid, methyl ester, (E)-**
13.949	1.81	Triacontanoic acid, methyl ester
14.068	**11.63**	**9,12-Octadecadienoic acid (Z,Z)-**
14.245	4.60	Linoleyl acetate
14.294	**7.34**	**Ethyl Oleate**
14.593	0.92	Heptadecane, 2,6,10,15-tetramethyl-
15.621	0.96	Sulfurous acid, octadecyl 2-propyl ester
18.415	0.89	1,2-Benzenedicarboxylic acid, bis(2-ethylhexyl
20.073	0.85	Eicosanoic acid, methyl ester
20.883	1.94	5,9,13,17-Tetramethyl 4,8,12,16-octadecatetraeno
27.570	3.36	Stigmasterol
28.997	**9.31**	**.gamma.-Sitosterol**

Table 4. Phytoconstituents identified in the ethyl acetate extract of germinated seeds of *Centrosema viginianum* by GC-MS analysis (major compounds are in bold).

Retention time min)	Peak area (%)	Active principle
6.213	0.95	Ethyl 3-acetoxybutyrate
11.584	1.23	Nonane, 5-methyl-5-propyl-
12.934	1.10	Sulfurous acid, 2-ethylhexyl isohexyl ester
14.081	5.29	Eicosane
14.560	1.26	3-Isopropyl-6,10-dimethylundecane-2-ol
14.706	5.68	Tetradecanoic acid
15.161	1.40	Heneicosane
15.240	1.14	Heptadecane
15.550	2.66	2-Methyltetracosane
15.787	4.36	1,2-Benzenedicarboxylic acid, bis(2-methylpro
16.319	3.89	Malonic acid, heptadecyl 4-methylpent-2-yl ester
16.775	**22.31**	**l-(+)-Ascorbic acid 2,6-dihexadecanoate**
17.185	1.24	Heneicosane
17.987	2.19	Carbonic acid, decyl pentadecyl ester
18.118	2.17	7-Hexadecenal, (Z)-
18.335	5.44	Tetrapentacontane
18.485	5.44	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
18.720	**7.82**	**Dotriacontyl isopropyl ether**
19.066	1.51	Hexacosane, 1-iodo-
20.160	2.13	Dodecyl octyl ether
20.583	1.12	Octadecane, 3-methyl-
23.144	1.12	Tridecanol, 2-ethyl-2-methyl-
23.552	2.52	1,2-Benzenedicarboxylic acid, bis(2-ethylhexyl)
26.455	4.51	2,6,10,15,19,23-Pentamethyl-2,6,18,22-tetraco
27.442	0.52	Carbonic acid, decyl nonyl ester
28.220	0.98	3-n-Heptyl-7-methyl-9-(2,6,6-trimethylcycloh
30.148	2.33	Stigmast-5-en-3-ol, oleate

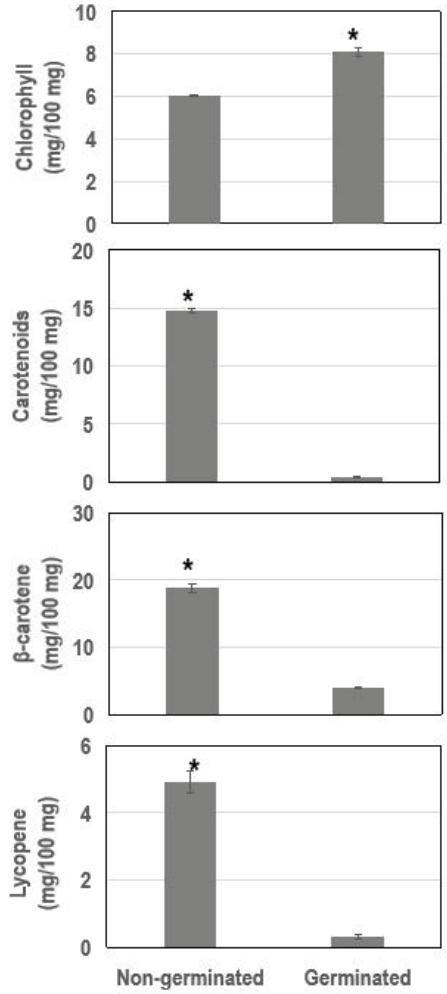

Figure 6. Total chlorophyll, carotenoids, b-carotene and lycopene in the seeds of *Centrosema virginianum* (n=3, mean±SD; t-test: *, p<0.001).

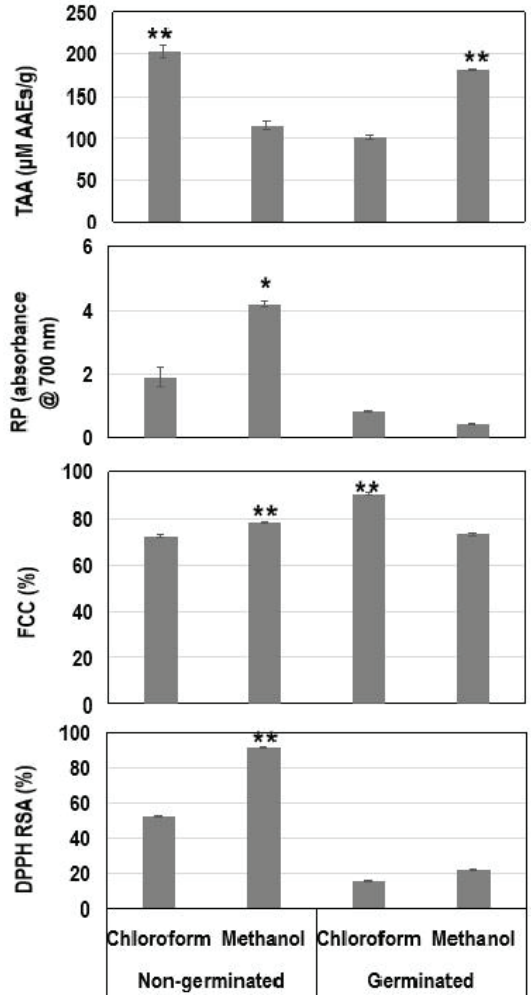

Figure 7. Total antioxidant activity, reducing power, ferrous ion-chelating capacity and DPPH radical-scavenging activity in chloroform and methanol extracts of seeds of *Centrosema virginianum* (n=3, mean±SD; t-test: *, 0.01; **, p<0.001).

as *C. maritima* [Niveditha, Sridhar, 2014]. Reducing power in *C. virginianum* was significantly higher in methanol than chloroform extract in non-germinated seeds (p<0.05), while it was low and did not differ in germinated seeds.

The ferrous ion-chelation capacity in *C. virginianum* was higher in methanol than chloroform extract in non-germinated seeds (p<0.001), while it was opposite for germinated one. The ferrous ion-chelating capacity is several folds higher than that of maritime *Canavalia cathartica* as well as *C. maritima* [Niveditha, Sridhar, 2014].

The DPPH radical-scavenging potential in *C. virginianum* was higher in methanol than chloroform extract of non-germinated seeds (p<0.001), while it was low and did not vary significantly in germinated seeds. The DPPH radical-scavenging activities of chloroform extracts of non-germinated seeds and extracts of both solvents are comparable to those of maritime *Canavalia cathartica* as well as *C. maritima* [Niveditha, Sridhar, 2014]. Methanol extracts of non-germinated seeds showed DPPH radical-scavenging activity considerably higher than that of maritime *Canavalia cathartica* as well as *C. maritima*.

CONCLUSION

Legumes constitute an important source of forage for livestock. The endangered, little-known forage legume *C. virginianum* is a perennial climber widely distributed. The present study strengthened the forage, nutraceutical and bioactive potential of its seeds. Besides its forage value to livestock, there is plenty of

scope to study this legume in terms of conservation (in situ and ex situ), soil fertility, nutritional and medicinal advantages. Due to its wide distribution in southwest India (in lateritic belt, mangroves and coastal sand dune), adaptability, associated microbes, nitrogen fixing ability, improvement of soil fertility and possibility of producing silage, these are the potential areas of future research.

ACKNOWLEDGEMENTS

The authors are indebted to Mangalore University for supporting this study and to the Department of Biosciences for offering the laboratory facilities. We thank Dr. Mahadevakumar, Scientist, Botanical Survey of India, Andaman and Nicobar Regional Centre, Port Blair, India, for technical support. The authors are grateful to the referee for meticulous screening of the draft version of this paper.

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Yabanı yem paxlası *Centrosema virginianum* toxumlarının qida, fitokimyəvi və bioloji aktiv profillərinin müqayisəsi

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Bu tədqiqatın məqsədi Hindistanın cənub-qərbində yayılmış yabanı yem paxlası *Centrosema virginianum* (L.) Benth növünün cücərməmiş və cücərmiş toxumlarının qidalandırıcı və bioaktiv potensialını qiymətləndirməkdir. Hər iki növ toxumda yüksək miqdarda karbohidratlar və lipidlər var, zülal, lif və kalori isə orta səviyyədədir. Keyfiyyətin qiymətləndirməsi cücərməmiş toxumlarda cücərmiş

toxumlara nisbətən daha çox fitokimyəvi maddələrin olduğunu göstərmişdir. Cücərməmiş toxumların xloroform ekstraktı metanol ekstraktı ilə müqayisədə əhəmiyyətli dərəcədə yüksək ümumi fenol maddələr və taninlərə malikdir, flavonoidlər və C vitamini isə azlıq təşkil edir. GC-MS analizi zamanı əsas birləşmələr kimi palmitik turşu və metil 9-oktadekenoat, cücərmiş toxumlarda isə askorbin turşusu və dotriakontil izopropil efiri aşkar edilmişdir. Cücərməmiş toxumların aseton ekstraktlarındakı piqmentlər arasında xlorofil cücərmiş toxumlarda cücərməmişlərə nisbətən xeyli yüksək, karotenoidlərdən β-karotində və likopendə isə tərkibi əksinə olmuşdur. Cücərməmiş toxumların metanol ekstraktı ilə müqayisədə xloroform ekstraktında ümumi antioksidant aktivlik əhəmiyyətli dərəcədə yüksək olmuş, halbuki, dəmir ionlarının xelatlaşdırılması qabiliyyəti və DPPH radikallarını saxlama qabiliyyəti azalmışdır. Cücərmiş toxumlarda ümumi antioksidant aktivlik xloroform üçün metanol ekstraktı ilə müqayisədə əhəmiyyətli dərəcədə yüksək, əksinə dəmir ionlarını xelatlaşdırmaq qabiliyyəti üçün aşağı olmuşdur. Bu tədqiqat az tanınan otlaq paxlası *C. virginianum* növünün yem xüsusiyyətləri haqqında bilikləri artırır. Lobya növünün nəsli kəsilmək təhlükəsi altında olduğunu nəzərə alaraq, gələcək tədqiqatlarda onun qorunmasına və mal-qaranın qidalanmasında faydalılığına diqqət yetirilməlidir.

Açar sözlər: *antioksidantlar, yem, cücərmiş toxumlar, paxlalı otlaqlar, piqmentlər, proksimal keyfiyyətlər*

Сравнение пищевых, фитохимических и биологически активных профилей семян диких кормовых бобовых *Centrosema virginianum*

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Целью настоящего исследования является оценка нутрицевтического и биоактивного потенциала непроросших и проросших семян дикорастущей кормовой бобовой культуры *Centrosema virginianum* (L.) Benth. распространенный на юго-западе Индии. Оба типа семян содержат большое количество углеводов и липидов, тогда как белки, клетчатка и калорийность умеренные. Сравнительная оценка показала, что непроросшие семена содержат больше фитохимических веществ, чем проросшие семена. Непроросшие семена содержат значительно более высокие общие фенольные и дубильные вещества в экстракте хлороформа, чем в метанольном экстракте, тогда как во флавоноидах и витамине С ситуация меняется на противоположную. Анализ ГХ-МС выявил пальмитиновую кислоту и метил-9-октадеценоат в качестве основных соединений, тогда как аскорбиновая кислота и изопропиловый эфир дотриаконтила были обнаружены в проросших семенах. Среди пигментов в ацетоновых экстрактах хлорофилла было больше в проросших семенах, тогда как каротиноидов, β-каротина и ликопина – в непроросших семенах. Общая антиоксидантная активность была значительно выше в экстракте хлороформа, чем в экстракте метанола в непроросших семенах, в то время как она была обратной по восстанавливающей способности, способности хелатировать ионы железа и активности по улавливанию радикалов DPPH. В проросших семенах общая антиоксидантная активность была значительно выше у хлороформа, чем у метанольного экстракта, тогда как по способности хелатировать ионы железа она была противоположной. Данное исследование укрепило информацию о кормовых свойствах малоизвестной пастбищной бобовой культуры *C. virginianum*. Поскольку этот вид бобовых находится под угрозой исчезновения, будущие исследования должны быть сосредоточены на его сохранении и полезности в питании скота.

Ключевые слова: *антиоксиданты, корм, проросшие семена, бобовые пастбища, пигменты, проксимальные качества*