Nutritional Assessment of Invasive Alien Plants as Bioprospecting Resources in Mizoram, an Indo-Burma Mega Biodiversity Hotspot in India

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ABSTRACT

Background: The nutritional value, antinutritional properties, mineral and Vitamin content of six invasive Alien Plant species (IAPs) namely Ageratina adenophora, Ageratina riparia, Chromolaena odorata, Ocimum americanum, Bidens pilosa and Hyptis suaveolens which are used by the local inhabitants in Mizoram, India was analysed. **Materials and Methods:** The proximate composition, antinutritional contents, mineral and Vitamin content were investigated following standard analysis protocols. Tannins, saponins, oxalates, cyanogenic glycoside, and phytate were estimated to ascertain the antinutritional properties of the selected IAPs. Water soluble Vitamins were estimated by HPLC method and results were analysed utilising the Statistical Package SIGMAPLOT 14.5 (Systat Software Inc., USA). Results: Results revealed high protein (1.19-12.70%, dry plant material), carbohydrate (1.55-26.79%, dry plant material) and ash (6.93–12.14%) but low crude fat content (1.12–3.64%) in these IAPs. Highest Vitamin-C content was observed in A. adenophora (28.15 mg/ 100 g dry plant material). The oxalate content was maximum in A. *riparia* (0.29% \pm 0.003%) and least in *O. americanum* (0.17% \pm 0.002%); phytate content ranged from (0.06% \pm 0.001%) in C. odorata to (0.11% \pm 0.007%) in O. americanum. Tannin was detected highest in A. adenophora ($3.82\% \pm 0.164\%$) and least in O. americanum ($1.75\% \pm 0.024\%$). Highly invaded A. adenophora contained mineral in the following order Ca> Zn > K > Mg > Na > Fe >Mn > Cu. **Conclusion:** It can be concluded that these IAPs exhibited high nutritive value (high protein, carbohydrate content; micronutritional components Vitamin-C and minerals) of the examined plants. The study recommends these IAPs could potentially be used as a food or feed additive in Mizoram by locals for sustainable management of invasive plants.

Keywords: Invasive alien plants, Antinutrients, Mizoram, Minerals content, Proximate composition, Vitamin.

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Received: 13-06-2022; Revised: 22-10-2022; Accepted: 06-02-2023.

INTRODUCTION

Invasion by alien plants is one of the most concerning threats to biodiversity, which can irreversibly alter ecosystem and economic services hampering livelihoods leading to major social change.¹ On natural ecosystems, impact of invasive alien plant species (hereafter IAPs) can lead to competition with native species limiting the availability of resources causing degradation of the ecosystem equilibrium as well as agriculture productivity and human health.¹ Protected forest areas in Mizoram, like other areas of Indian Himalayan Regions, are highly susceptible to aggressive alien plant invaders due to high anthropogenic disturbances.



DOI: 10.5530/ijper.57.2s.44

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Recently, 163 alien plants were reported from the state with noxious IAPs like *Ageratina adenophora, Ageratina riparia, Chromolaena odorata, Mikania micrantha, Ageratum conyzoides* and neo-invasives like *Ocimum americanum, Hyptis suaveolens, Bidens pilosa* with their ecological impact to consistent alteration of natural vegetation and ecosystem.² As a remote north-eastern state, Mizoram lacks an effective management framework for plant invasion. Therefore, effective and sustainable management strategy is need of the hour against invasion by alien plants.

Preliminary interaction with the locals of fringe villages around protected areas in Mizoram revealed that they contrast invasion threat by using some IAPs for various purposes like using *Ageratina adenophora, Ageratina riparia, Bidens pilosa, Mikania micrantha* vegetative parts as fodder; *Ocimum americanum* leaves as condiments; *Hyptis suaveolens* vegetative parts paste as appetizer, *Chromolaena odorata* as wound healer and fodder in their livelihood. Hence, it is necessary to identify innovative

Name of the plants	Collection area	Voucher no.	Ethnobotanical uses in Mizoram
A. adenophora	24.301180, 92.748519 23.759338, 93.248397	71535	Leaf decoction is also used as diuretic. Freshly crushed leaf juice is applied on cuts and wounds to stop bleeding.
A. riparia	23.578219, 92.407943 23.729169, 92.534286	71562	Infusion of dried leaf and capitula (8-10 dried capitula per 100 mL) is used as herbal drink to reduce hypertension and as antidiabetic.
C. odorata	22.762641, 93.009445 23.892501, 93.251144	71532	Crushed leaf juice is used topically on cuts and wounds as antiseptic and haemostatic.
H. suaveolens	22.730128, 93.053052 22.746103, 93.059115	71595	Tender shoot tips are used as a flavouring agent in soups and stew. Freshly plucked leaves are boiled and used as a mint-flavoured tea as an appetizer.
B. pilosa	22.851762, 92.779654 22.392340, 92.780809	71598	Used as a fodder where whole plant along with <i>Crassocephalum crepidioides</i> and chopped banana stem is used as pig food.
O. americanum	23.365833, 92.815453 23.578219, 92.407943	71588	Dried inflorescences are used as condiments in local cuisine throughout Mizoram. Dried or fresh leaves are also used as flavouring agent in soup, salads and sauces.

Table 1: Co-ordinates and use of IAPs sample collection from study sites in Mizoram.

and sustainable way of invasive plants management transforming them from "menace" to useful "resources". Although traditionally used native plants have been investigated for their potential value as a source of drugs, nutritional analysis of noxious IAPs as bioprospecting resources have not been explored yet in Mizoram as well as in India. In this context, the present study was designed to understand the utilization potential of some noxious and neo-invasives for bioprospecting which may pave a way to an innovative strategy for sustainable management of alien plant invasion. In this context, our study was designed to perform phytochemical screening viz. nutritional composition, minerals, Vitamins and antinutritional composition of noxious IAPs namely A. adenophora, A. riparia, C. odorata, B. pilosa, O. americanum, H. suaveolens evaluating their use potential as food or fodder in different sectors of human interest adding as a bioprospecting strategy.

Outcome of the present work will significantly enrich the knowledge on use of invasive alien plants, both locally and globally, to control them in a sustainable way and the rationale of the present study is to attract the general public and policymakers' attention on the pharmaceutical potential in these IAPs to induce sustainable management by exploitation of their phytochemicals, which have beneficial effects on human health.

MATERIALS AND METHODS

Study site

Mizoram, a north-eastern state of India (Figure 1) is adjoined by Bangladesh on the western side and Myanmar (Burma) the eastern side sharing a common border with Assam, Manipur and Tripura. As per recent reports,³ forest cover of the state stands at 73.68% of the total geographical area consisting of only 6.75% protected area network. The temperatures usually vary from 18-29°C during summer and 11-24°C during with average annual rainfall of 2160 mm to 3500 mm.³ For the suitability of sample collection for the present work, we have collected the six IAPs from various protected areas in Mizoram (GPS location in Table 1) including Phawngpui National Park (50 km²) and Murlen National Park (200 km²). The natural vegetation of the state is categorised based on its elevation and precipitation viz. tropical (0-900 m), subtropical (900–1800 m) and temperate (1800–3600 m).²

Plant materials

Vegetative parts of fresh plant materials viz. Ageratina riparia (Regel) R.M.King and H.Rob. (Voucher no.71562), Ageratina adenophora (Spreng.) R.M.King and H.Rob. (Voucher no.71535), Chromolaena odorata (L.) R.M.King and H.Rob. (Voucher no.71532), Bidens pilosa L. (Voucher no.71598) (Family: Asteraceae), Hyptis suaveolens (L.) Poit. (Family: Lamiaceae) (Voucher no.71595), and Ocimum americanum L. (Family: Lamiaceae) (Voucher no.71588) were collected from different protected natural vegetation of Mizoram. Information regarding the socio-economic and ethno-botanical uses (Table 1) of the six IAPs was collected along with their co-ordinates and evaluated through a semi-structured interview from the locals with an open-ended questionnaire. Proper identifications of the collected plants were authenticated from Botanical Survey of India, Howrah. One part of the plant samples was preserved at 15°C for water soluble Vitamin estimation while other part was shade dried, pulverised and preserved in an airtight container for further proximate analysis.

Proximate Analysis

The powdered plant vegetative parts were analysed for nutritional composition adhering to the standard analysis

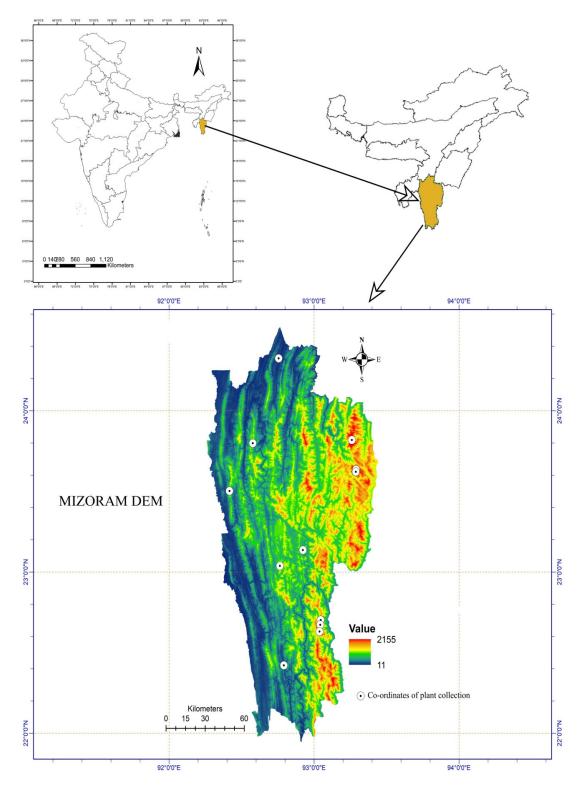


Figure 1: Digital Elevation Map of Mizoram along with study site co-ordinates.

protocols recommended by the Association of Official Analytical Chemists.⁴ Plant samples were continuously heated in a muffle furnace for 5-6 hr at 500°C and the moisture contents of the plants were estimated dehydration of the plant samples at 100°C–110°C in a hot air oven. The experiment for crude fat was analysed in

a Soxhlet apparatus using petroleum ether from moisture-free sample (60°C–80°C) for approximately 6–8 hr. Micro-Kjeldahl method was used for estimation of crude protein⁴. The below mentioned formula was used for the estimation of Total carbohydrate content

Carbohydrate (%) = 100 - [Moisture (%) + Crude protein (%) + Crude fat (%) + Ash (%)]

The values obtained from the abovementioned experiments were multiplied by 4.00, 9.00, and 4.00 for protein, fat, and available carbohydrates, respectively, and summing up the values to get total energy value from each dry plant powder.⁴

Energy value = (% crude protein × 4) + (% carbohydrate × 4) + (% crude fat × 9)

Mineral Content Analysis

The dried plant material was taken in a clean empty pre-weighed silica crucible which was heated using muffle furnace at a temperature of 400°C until it produces smoke. After heating and cooling down, the ash sample from the plant material was soaked in concentrated sulphuric acid and then heated in a heating mantle until fumes of sulphuric acid stopped. Then the crucible filled with plant ash was reheated in a muffle furnace at 600°C until the ash weight became persistent (~2–3 hr). 100 mL of 5% hydrochloric acid (HCl) was added to dissolve the resultant sulphated ash to subject the solution ready for the estimation of minerals using atomic absorption spectroscopy (AAS) (AA 800, Perkin-Elmer Germany) technique. For determination of the minerals by AAS method, standard solution of each plant sample was prepared followed by drawing calibration curves.⁵

Analysis of water-soluble Vitamins using chromatography

The analysis of water-soluble Vitamins was done using chromatography following the method depicted by Seal et al.⁶ Acetonitrile was used as the mobile phase (Solvent A) and aqueous trifluoroacetic acid (TFA, 0.01% v/v) (Solvent B), chromatographic column was thermostatically maintained at a temperature of 22°C, and 20 mL injection volume was also maintained. Changing the proportions of solvent A to Solvent B mixture, gradient elution was conducted. A photodiode array detector ultraviolet (UV) at four separate wavelengths i.e., 210, 245, 275, and 290 nm was used to ascertain all the HPLC chromatograms according to absorption maxima of sample components under analysis. For each sample, 35 min was the total analysis time. Individual value of retention time and spiking with their standard values under similar conditions, each water-soluble Vitamin present in respective plant extracts were identified. The resultant data were meticulously calculated and statistically analysed presented as means ± Standard Error of Means (SEM). The SEM was taken from three independent analysis values.

Antinutritional composition estimation

The method described by Munro and Bassir⁷ were followed for analysing oxalate contents of the IAPs under investigation. Each

plant dried powder was weighed 1 g (in triplicate) followed by mixing with 0.3M HCl and then for 1 hr, stirring with a magnetic stirrer, heated at 50°C for 1 hr. For total oxalate estimation, extracts were diluted up to 100 mL with water. Determination of phytate was performed following Reddy and Love⁸ methods. A total of 100 mL of 2% HCl was added to 1 g dried powder of each sample plant and soaked for 5 hr followed by filtering. Into 25 mL of the filtrate, 5 mL 0.3% ammonium thiocyanate solution was added; followed by titration with FeCl, solution and titration was ceased when a brownish yellow colouration achieved which persisted for 5 min. Using the method of Hudson and El-Difrawi,⁹ the saponin content was detected. The modified vanillin-HCl method as described by Price et al.¹⁰ was used for estimation of Tannins and tannic acid was used as the reference standard. Alkaline titration method⁴ was used to detect and estimate cyanogenic glycoside contents of the sample where the end-point was recorded as permanent turbidity against a black background.

Statistical analysis of the results

To perform suitable accurate statistical analysis, all the results were performed in triplicate tests. Results of the tests were then subjected to univariate analysis of variance which was trailed by Tukey's test ($P \le 0.05$). Obtained statistical results were then tested for principal component analysis (PCA). All the statistical analysis was performed using the statistical package for SIGMAPLOT 14.5 (Systat Software Inc., USA).

RESULTS

Proximate composition

Presence of moisture in edible vegetables is treated as a good source of water and 20% of necessary total water consumption for the human body comes from food. The results demonstrated that these IAPs contained significantly (P < 0.05) high moisture viz. maximum in H. suaveolens (82.57±0.96%) and minimum in O. americanum (9.08±0.11%). The ash content of the various wild edible plants was highest in A. riparia (12.90±0.20%) and the lowest in A. adenophora (6.93±0.067%). The measurement of unrefined protein substances exhibited significantly (P < 0.05) higher protein value in the extracts of A. adenophora (12.70%) \pm 0.07%) and least in the extracts of O. americanum (1.19% \pm 0.015%). Total carbohydrate content exhibited significant (P<0.05) carbohydrate content in A. adenophora (26.79%), O. americanum (9.73%) and B. pilosa (8.69%) compared to low amount A. riparia (3.65%), C. odorata (1.55%) and H. suaveolens (3.08%). The total crude fat content ranged between 1.12-3.64% in these IAPs, with maximum fat content in A. riparia (3.64%) and least amount in B. pilosa (1.12%). All the results for proximate composition were shown in Table 2.

Name of the plants	Ash %	Moisture %	Protein % 6.25 × percentage of N	Carbohydrate %	Crude fat %	Energy content kcal/100 g
A. adenophora	6.93 ± 0.067^{d}	69.37 ± 0.39^{b}	12.70 ± 0.07^{a}	26.79±0.13ª	3.40 ± 0.01^{b}	188.79±0.14ª
A. riparia	12.90±0.20ª	64.74±0.33°	$9.89 \pm 0.17^{\mathrm{b}}$	3.65 ± 0.13^{d}	3.64 ± 0.01^{a}	86.91 ± 0.06^{b}
C. odorata	9.50±0.02°	58.47 ± 0.67^{d}	12.24±0.06ª	1.55 ± 0.03^{f}	2.18 ± 0.03^{d}	74.92±0.07 ^e
H. suaveolens	9.42±0.05°	82.57±0.96ª	9.90 ± 0.17^{b}	3.08±0.03 ^e	2.74±0.03°	76.62 ± 0.03^{d}
B. pilosa	9.29±0.03°	80.49±0.36ª	8.27±0.08°	8.69±0.03°	$1.12 \pm 0.01^{\text{f}}$	77.93±0.04°
O. americanum	12.14 ± 0.14^{b}	9.08±0.11 ^e	$1.19{\pm}0.01^{d}$	$9.73 \pm 0.03^{\mathrm{b}}$	1.81 ± 0.03^{e}	$59.98{\pm}0.07^{\rm f}$

Table 2: Proximate composition of the IAPs.

Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean \pm Standard error of the mean (SEM). Statistical analysis was carried out by Tukey's test at 95% confidence level and statistical significance were accepted at the p < 0.05 level. The superscript letter a, b, c d, e, f denotes the significance of various parameters. Letter a is significant to b, c, d, e, f; N = Nitrogen.

Table 3: Water soluble B-Vitamins and Vitamin C content of IAPs.

Name of the plants	Water-soluble Vitamin (mg /100g dry plant material)							
	С	B ₁	B ₂	B ₃	B ₅	B ₆	B ₉	
A. adenophora	28.43±0.22ª	0.13 ± 0.001^{e}	0.03 ± 0.001^{d}	$0.64{\pm}0.005^{a}$	1.27 ± 0.003^{e}	$1.73{\pm}0.004^{\rm f}$	31.65±0.01ª	
A. riparia	2.93±0.12°	$0.95 \pm 0.005^{\circ}$	$0.23 \pm 0.008^{\circ}$	$0.12 {\pm} 0.005^{\rm b}$	$7.24{\pm}0.003^{a}$	$3.32{\pm}0.003^{d}$	2.14±0.003 ^e	
C. odorata	ND	$0.46 {\pm} 0.004^{d}$	$0.57 {\pm} 0.005^{\rm b}$	ND	$4.73 {\pm} 0.003^{\rm b}$	$4.96 {\pm} 0.003^{\rm b}$	22.96±0.01 ^b	
H. suaveolens	8.76 ± 0.01^{b}	2.34±0.041ª	$0.53 {\pm} 0.005^{\text{b}}$	$0.02 \pm 0.001^{\circ}$	1.92±0.038°	$3.47 \pm 0.002^{\circ}$	3.76 ± 0.02^{d}	
B. pilosa	ND	1.69 ± 0.029^{b}	0.09 ± 0.003^{d}	ND	ND	$8.03{\pm}0.002^{a}$	4.78±0.01°	
O. americanum	ND	$0.38 {\pm} 0.005^{d}$	$0.95{\pm}0.004^{a}$	ND	$1.81{\pm}0.003^{d}$	2.96±0.002 ^e	$1.07{\pm}0.002^{\rm f}$	

Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean \pm Standard error of the mean (SEM). Statistical analysis was carried out by Tukey's test at 95% confidence level and statistical significance were accepted at the p < 0.05 level. The superscript letter a, b, c d, e, f denotes the significance of various parameters. Letter a is significant to b, c, d, e, f. ND =Not Detected

Vitamin composition

Water soluble Vitamins were analysed from IAPs are enumerated in Table 3 and HPLC chromatograms enumerated in Figure 2. Vitamin analysis of the vegetables revealed that vegetative parts of *A. adenophora* had highest values of Ascorbic acid (28.43 ± 0.22 mg/ 100 g), niacin (0.64 ± 0.005 mg/100 g) and folate (31.65 ± 0.01 mg/100 g) but least amount of thiamine (0.13 ± 0.001 mg/100 g), pantothenic acid (1.27 ± 0.003 mg/100 g), and pyridoxine (1.73 ± 0.004 mg/100 g) compared to the five other IAPs. *H. suaveolens* (2.34 ± 0.041 mg/100 g) maximum amount of thiamine and *B. pilosa* had highest pyridoxine (8.03 ± 0.002 mg/100 g) content. *A. riparia* possessed maximum pantothenic acid (7.24 ± 0.003 mg/100 g) content.

Mineral Composition

The minerals like Sodium (Na), Calcium (Ca), Iron (Fe), Potassium (K), Magnesium (Mg), Copper (Cu) and Zinc (Zn) compositions of the IAPs were listed in Table 4. Results of this study revealed that, among the six IAPs examined in this study, *A. adenophora* contained the highest levels of Ca, Zn, Mn and Cu which were significantly (P<0.05) higher recorded as (226.63± 0.033), (71.38 \pm 0.023), (1.23 \pm 0.033) and (0.23 \pm 0.20) mg/100 g DPM respectively. *H. suaveolens* contained the highest level of Na (9.72 \pm 0.020 mg/100 g DPM) and the lowest levels of K which were recorded as (34.65 \pm 0.017 mg/100 g DPM). *B. pilosa* contained the highest levels of Fe and Mg which were observed as (14.60 \pm 0.020 mg/100 g DPM) and (66.82 \pm 0.033 mg/100 g DPM) respectively and the lowest levels of Na recorded as (2.18 \pm 0.067mg/100 g DPM).

Anti-nutritional profile

In the current study, oxalate, phytate, tannin, saponin, cyanogenic glycoside present in vegetative parts of the IAPs were analysed and shown in Table 5. Oxalate was found least amount in *B. pilosa, O. americanum* (0.17% \pm 0.002%) and highest in *A. riparia* (0.29% \pm 0.003%). Phytic acid substance in these IAPs ranged from (0.06% \pm 0.001%) in *C. odorata* to (0.11% \pm 0.007%) in *O. americanum*. Highest amount of tannin was found in the leaves of *A. adenophora* (3.82% \pm 0.164%) and least amount was observed in *O. americanum* (1.75% \pm 0.024%). Among the studied IAPs, maximum saponin was observed in *C. odorata* (4.31% \pm 0.033%) whereas extracts of *A. adenophora* had minimal fixation (0.13% \pm 0.003%). Results also revealed that cyanide components in the

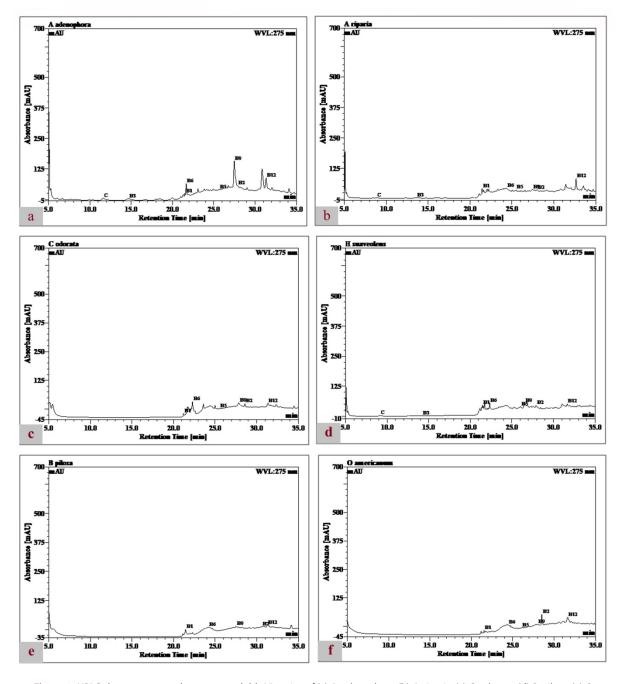


Figure 2: HPLC chromatograms show water-soluble Vitamins of (a) *A. adenophora*, (b) *A. riparia*, (c) *C. odorata*, (d) *B. pilosa*, (e) *O. americanum*, (f) *H. suaveolens*.

studied IAPs are between $1.29\% \pm 0.003\%$ and $2.05\% \pm 0.017\%$ in *C. odorata* and *H. suaveolens* respectively.

Eigen values >1, only the first two principal components were accounted to simplify the analysis of results.

Principal component analysis

The combined proximate composition, Vitamin C, Vitamin B_6 , Vitamin B_9 , Vitamin B_2 and mineral data was subjected to Principal Component Analysis (PCA). The PCA score plots of the plant samples under analysis are shown in Figure 2a (based on all proximate, Vitamin C, B_1 , B_2 , B_3 , B_5 , B_6 and B_9 variables) and their corresponding loading plots are presented in Figure 2b. Although the PCA yielded four Principal Components (PC) with

DISCUSSION

Proximate composition

Moisture analysis results were very close to the moisture contents of some edible and commonly used vegetables like *Cucurbita maxima* (81.2%), *Hibiscus subdarifolia* (85.5%) and *Solanum anguivi* (86.2%) in Mizoram.¹¹ Although high moisture content plant-based food materials are prone to quick decomposition, as the weather condition in Mizoram very rarely cross 28°C on

Table 4: Mineral composition of the IAPs. Name of Minerals content (mg /100g dry plant material) the plants Cu Na Ca Κ Fe Zn Mn Mq A. adenophora 6.54±0.01° 226.63±0.03ª 41.60 ± 0.01^{d} 4.07±0.033° 71.38±0.023^a 1.23±0.033^a 0.23±0.020^a 28.59±0.030° A. riparia 3.35±0.02^e 15.82±0.017^f 56.38±0.02° 9.28 ± 0.020^{b} 2.18±0.020^d 1.02±0.033^b 0.08±0.013° 11.92±0.027e C. odorata 8.66 ± 0.02^{b} 64.93±0.033^d 41.29±0.01^e 3.93±0.020^d 0.96 ± 0.023^{f} 0.77±0.020° 0.17 ± 0.010^{ab} 34.77±0.020b H. suaveolens 9.72±0.02^a 143.96±0.023° 34.65 ± 0.01^{f} 9.21±0.033^b 1.60 ± 0.010^{e} 0.81±0.020° 0.15±0.017^{bc} 25.86±0.037^d B. pilosa 2.18±0.06^f 161.65±0.030^b 61.67±0.03^b 14.60±0.020^a 6.46±0.023° 1.16±0.033^a 0.15 ± 0.010^{bc} 66.82±0.033^a O. americanum 3.91±0.03^d 52.38±0.020e 69.21±0.03ª 1.52±0.030e 15.84±0.027^b 0.36 ± 0.013^{d} 0.13±0.013bc 10.58±0.033^f

Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean \pm Standard error of the mean (SEM). Statistical analysis was carried out by Tukey's test at 95% confidence level and statistical significance were accepted at the p < 0.05 level. The superscript letter a, b, c d, e, f denotes the significance of various parameters. Letter a is significant to b, c, d, e, f.

Table 5. Alternation prome of the larts.							
Name of the plants	Oxalate (%)	Phytate (%)	Tannin (%)	Saponin (%)	Cyanogenic Glycoside (%)		
A. adenophora	0.27 ± 0.003^{b}	0.08 ± 0.001^{b}	3.82 ± 0.164^{b}	0.13 ± 0.003^d	1.62±0.003°		
A. riparia	0.29±0.003ª	$0.06 \pm 0.002^{\circ}$	$2.79{\pm}0.077^{\rm d}$	2.75±0.033°	1.41 ± 0.003^{d}		
C. odorata	0.26±0.002°	$0.06 \pm 0.001^{\circ}$	$2.84{\pm}0.029^{\rm d}$	4.31±0.033ª	1.29±0.003 ^e		
B. pilosa	0.17 ± 0.002^d	0.07 ± 0.003^{bc}	4.17±0.352ª	3.20±0.033°	1.83 ± 0.010^{a}		
H. suaveolens	0.22±0.003 ^e	0.07 ± 0.001^{bc}	3.61 ± 0.043^{b}	2.86 ± 0.033^{b}	2.05 ± 0.017^{b}		
O. americanum	0.17 ± 0.002^{e}	0.11 ± 0.007^{a}	1.75±0.024 ^e	2.88±0.033°	1.82 ± 0.010^{b}		

Table 5: Anti-nutrient profile of the IAPs.

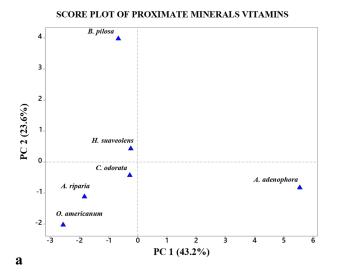
Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean \pm Standard error of the mean (SEM). Statistical analysis was carried out by Tukey's test at 95% confidence level and statistical significance were accepted at the *p* < 0.05 level. The superscript letter a, b, c d, e, f denotes the significance of various parameters. Letter a is significant to b, c, d, e, f.

average, the IAPs plant-based food material storage shelf life must be better compared to other hot humid regions. This study recommends that the vegetative parts of the IAPs should be dried and stored for further use when about to be used for consumption to avoid spoilage by microbial infection.

The level of ash content in a food material represents the number of inorganic compounds including minerals available in food after burning it. Ash contents in the IAPs were higher than the ash contents of some commonly used edible home garden vegetables like *Cucurbita maxima* (2.8%), *Acacia pennata* (7.9%), *Dioscorea esculenta* (1.70%), *Hibiscus sabdariffa* (1.5%) and *Solanum anguivi* (1.2%) in Mizoram.¹¹ The ash content of the IAPs was low compared to five edible wild vegetables reported from NE India.¹² Therefore; the results obtained exhibit significant ash content including high deposit of mineral contents in these IAPs which may contribute a considerable amount of mineral elements in native people's diet.

A plant based food is considered good source of protein if it provides greater than 12% of its calorific value from protein.¹³ Protein-calories malnutrition deficiencies were reported to be one of the major responsible factors in nutritional pathology.¹³ The dietary proteins broken down into amino acids are utilised by the body to integrate the required protein by the body for different functionalities *viz*. antibodies production opposing contamination, for the guideline of enzymes and hormones, for the body tissue and muscle repair.¹⁴ The results of this work revealed that adequate protein is present in IAPs like *A. adenophora* (12.70% \pm 0.07%) and *C. odorata* (12.24% \pm 0.067%) are good source of protein for local inhabitants which may be used to fulfil their daily per capita protein consumption of 54.66 cal.¹⁵ Using *A. adenophora* and *C. odorata* vegetative parts in food formulations may add nutritive value by influencing the quality of protein in the daily diet of local people.

High carbohydrate content observed in *A. adenophora, O. americanum* and *B. pilosa* indicate that these IAPs are a good source of carbohydrate which is an essential component to the body cells as an energy source. The carbohydrate content of *A. adenophora* (26.79% \pm 0.133%) were equivalent or greater than some reported values of regularly used vegetables viz. *Solanum anguivi* (26.95%), *Lycopersicon esculentum* (25.27%), *Solanum melongena* (18.54%) of solanaceae family in Mizoram, India.¹⁶ The carbohydrate content of *A. adenophora* (26.79% \pm 0.133%) were also significantly higher than the carbohydrate content ranged between 1.70 – 13.64% of six wild edible plants consumed by Bodo tribe of Assam, India.¹⁷ The recommended amount of carbohydrate required for children and adults is 130 g per day.



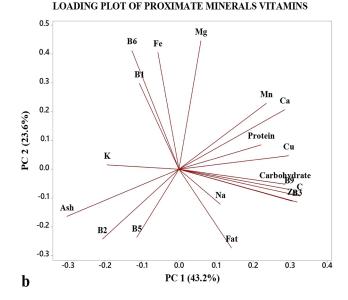


Figure 3: Principal component analysis shows (a) Score plot, (b) Loading plot.

Thus, the results imply that 100 g of these dried plants could provide 1.19–26.79% of the daily carbohydrate requirement.

Crude fat content present in the studied IAPs was in conformity with the findings of works reporting that leafy vegetables are poor sources of lipids.¹² The result was close to that found in six wild edible plants consumed by Bodo tribe of Assam, India.¹⁷ A diet is considered healthy and sufficient when it provides lesser percentage like 1-2% lipid as its caloric energy, as excessive lipid uptake leads to various cardiovascular disorders.¹⁴ Thus, vegetables having low fat content are usually recommended to overweight or obese individuals. Results for lipid content estimation of the IAPs were similar to some of the studies on wild edible plants of India.^{11,12} Absorption of important lipid soluble Vitamins like Vitamin-A and carotene in the body is also amplified by the presence of fat.^{13,14}

Vitamin composition

Vitamins are essential components to maintain the normal metabolic rhythm and homeostasis in the body. Vitamin C is a water-soluble Vitamin and population studies have shown that individuals with high Vitamin C intakes have a reduced risk of chronic diseases viz. cancer, eye, heart disease, diseases and neurodegenerative conditions.¹⁸ Vitamin C content in A. adenophora (28.15 mg/ 100 g DPM) is significantly higher than in the young shoots of bamboo species (3.0-13 mg/100 g DPM)18 which is a staple food of local Mizo people. The elevated level of niacin, folate, thiamine, pyridoxine, pantothenic acid obtained in vegetative parts of A. adenophora, H. suaveolens, B. pilosa, A. riparia exhibit that these IAPs are excellent source of Vitamin B complex. The amount of Vitamin B present in the studied IAPs was higher than some of the wild edible fruits like Meyna laxiflora and Viburnum foetidum from North-East India.12 The amount of Vitamin B detected from this study is considerably higher than some indigenous vegetables in Manipur like Oenanthe javanica, Adhatoda vasica, Allium tuberosum, Parkia roxburghii, Solanum torvum.¹⁹ The results obtained by analysing the IAPs ensure that the required Vitamin B complex supplement to the diet could be fulfilled by using these plant materials in diet.

Mineral Composition

The level of the minerals in highly invaded IAPs A. adenophora and C. odorata were in the following order Ca>Zn>K>Mg> Na > Fe > Mn > Cu and Ca > K >Mg > Na >Fe > Zn > Mn > Cu respectively. According to National Family Health Survey (2020)¹³ report, Mizoram has more than 46.4% of children below five years and 34.8% women between 15-49 years of age affected by anaemia. Therefore, proper dissemination of the information about the importance of these iron rich IAPs might play a role in reducing cases of anaemia in this region. O. americanum contained the highest level of K and the lowest levels of Fe, Mn and Mg. The mineral ion compositions of the IAPs were also relatively higher in all the studied samples than reported in some locally grown solanaceae species¹⁶ in Mizoram and similar to some indigenous vegetables of Manipur, Northeast India.¹⁹ As O. americanum, B. pilosa and A. riparia exhibit high quantity of potassium; their food value may be beneficial as coronary heart diseases are regulated by reducing blood pressure which is influenced by regular dietary intake of potassium.¹⁶ Calcium is involved in cell differentiation, muscle and bone formation, making it an essential mineral ion in human diet.20 Thus, high Cu containing IAPs like A. adenophora (226.63± 0.033) mg/100 g DPM, H. suaveolens (143.96 ± 0.023) mg/100 g DPM and B. pilosa (161.65 ± 0.030) mg/100 g DPM can be a good source of calcium in local diet. H. suaveolens (9.72±0.020) mg/100 g DPM and A. adenophora (6.54 ± 0.013) mg/100 g DPM with high sodium content can also act as a good source of dietary Na which is required for many physiological processes like maintaining body fluid balance and regulating cellular homeostasis and Magnesium is considered as

an important component in circulatory system which is essential for good metabolism²¹ which shows potential use of *B. pilosa* as a source of high Mg (66.82±0.033 mg/100 g DPM). Mn, Fe, Cu and Zn are essential micronutrients required for crucial metabolic processes like DNA synthesis and respiration.¹⁹ Our findings of aforesaid high content of minerals in the studied IAPs support effective utilisation of these plants as a source of minerals or nutrient supplement.

Anti-nutritional profile

Consumption of food products with high antinutrient content has been linked to low bioavailability of nutritional content in the body and these chemical compounds are advantageous for consumption when taken in recommended amounts only.22 Oxalate content obtained from the studied plants are almost identical with some of the regulars vegetables and fruits namely amla (0.296%), almond (0.407%), spinach (0.658%) and amaranth (0.772%).8 As an antinutrient, oxalate at a centralizations value around 45 mg/100, hinder renal calcium assimilation.²² On the other hand, phytate is an antinutrient which binds and hinders the bioavailability micronutrients such as iron, copper, zinc and calcium. Phytic acid substance in these IAPs were equivalent to the results of experiments conducted on rice bean (0.248% and 0.151%) of India.¹⁴ As a secondary phenolic component, Tannins consisting of molecular weights greater than 500 Da are found in bark, fruits and plant leaves.²³ Tannins are capable of precipitating proteins which causes reduced protein component deteriorating the food nutritional quality interfering with iron absorption.²² Low sums below the prescribed acceptable amount found in the studied IAPs may not impose any harmful impact on consumers. Secondary metabolite like saponins interact with erythrocyte membranes by recognizing the cholesterol groups, resulting into haemolysis as well as inhibiting digestive enzymes like trypsin, chymotrypsin, amylase and lipase leading to health disorders related to indigestion.²³ Among the studied IAPs, the amount of saponin was observed lower than the recommended amount.¹⁴ Cyanogenic glycosides are usually found in various plant parts and after hydrolysis produce Hydrogen Cyanide (HCN). This kind of HCN release happens due to enzymatic hydrolysis which causes Cyanide induced toxicity.²² To improve the dietary benefit of foods, continuous expulsion of cyanogenic glycosides is important.14 Results indicate that amount of cyanogenic glycosides in these plants was very low to procure any hindrance and safe to consume as part of the diet. However, the amount of antinutrients present in selected IAPs in this study is much below the destructive amount.²⁴ IAPs like A. adenophora, A. riparia and C. odorata can be treated as a good source of nutritional components and utilized by locals, as these plants have already invaded the protected areas requiring a proper management method.25

Principal component analysis

In Figure 3b, PC1 was negatively associated with Vitamin B_1 , B_2 , B_5 and B_6 ; minerals like K and Fe, ash content whereas positively associated with the Vitamin B_3 , B_9 and C, protein, carbohydrate and fat content, minerals like Mg, Cu, Ca, Zn, Na variables. PC2 was negatively correlated with Vitamin B_2 , B_5 and ash content while positively correlated with the others. The plant parts of *A. adenophora* were clearly separated and distant from all other samples on the right side and *A. riparia* and *O. americanum* on the left side (Figure 3a) as a result of the high contents of water-soluble Vitamins and proximate contents in variable amounts.

CONCLUSION

Tribal People of Mizoram, an Indo-Burma Hotspot region in India, have been using natural forest plants including noxious alien plant invaders and possess significant knowledge on various traditional uses of the IAPs. The outcomes of the study concluded that the studied IAPs are good sources of nutritional value viz. protein, carbohydrate, fat, minerals and natural supplements of Vitamin sources in improving malnutrition problems, combating nutritional human deficiency diseases and could also be treated as a source of drug development for the treatment of various diseases. These IAPs can be used along with commonly consumed vegetables in Mizoram. We must emphasize that the results obtained in this work is the first ever nutritional and antinutrient analysis reported in the literature for the wildly grown IAPs A. riparia collected from Mizoram, India. Along with natural medicinal flora, local people in the fringe villages of protected areas also collect these IAPs to use as food and fodder which can be used as vegetables to supplement their domestic nutritional requirements. As antinutritional values of these plants were observed lower than harmful level, they can be consumed without such concerns. Hence, from these results, it can be stated that these invasive alien plants have high nutritional values, which could be used widely in food applications as dietary vegetables for a person because of their healthful characteristics and used as local sustainable bioresources. These wild consumable IAPs should be embraced as a sustainable management method in huge scale, delivering financial and health benefits for native villagers of Mizoram in near future.

ACKNOWLEDGEMENT

The authors are grateful to Dr A.A. Mao, Director, Botanical Survey of India, Kolkata for encouragement and facility. The authors RS and SSD gratefully acknowledge financial support from the Ministry of Environment, Forest and Climate Change, Government of India for financial support through the National Mission on Himalayan Studies (Project NMHS-2017/LG/01/475).

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

HPLC: High performance liquid chromatography; Na: Sodium; Ca: Calcium; K: Potassium; Fe: Iron; Zn: Zinc; Mn: Manganese; Cu: Copper; Mg: Magnesium; %: Percentage; nm: Nanometer; mg: Milligram; μg: Microgram; SEM: Standard error of Mean; μM: Micromolar; AAS: Atomic absorption spectroscopy; M: Molar; TFA: Trifluoroacetic acid; UV: Ultraviolet; IAPs: Invasive Alien Plant Species; PCA: Principal component analysis; DPM: Dry plant material; ND: Not Detected.

SUMMARY

The present study was designed to assess proximate composition, minerals and Vitamin content, antinutritional properties of six Invasive Alien Plant Species (IAPs) in Mizoram, an Indo-Burma biodiversity hotspot in India. The results of phytochemical screening demonstrated that the selected IAPs are rich source of proteins, carbohydrate, crude fat, minerals as well as possess plentiful Vitamin B-complex and Vitamin-C. The studied six IAPs are recommended to be consumed for improving malnutrition problems and could be developed as source of drugs for the prevention and treatment of diseases for local fringe village dwellers of Mizoram.

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Cite this article: Sengupta R, Dash SS, Seal T. Nutritional Assessment of Invasive Alien Plants as Bioprospecting Resources in Mizoram, an Indo-Burma Mega Biodiversity Hotspot in India. Indian J of Pharmaceutical Education and Research. 2023;57(2s):s381-s390.