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A STUDY ON RATIONALE FOR ETHNOPHARMACOLOGICAL USES OF *BLUMEA LACERA* (BURM.F.) DC COLLECTED FROM DIFFERENT GEOGRAPHICAL LOCATIONS IN INDIA

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Abstract

India's biodiversity is full of nature's perspective. In respect to the plant species of this biodiversity, it is very important to do scientific study of wild plants in which useful phytoalexin compounds are present in abundance. Hence, in the present study, we mainly aimed to establish rationale for ethno pharmacological uses of *Blumea lacera* (Burm.f.) DC accessions collected from difference geographical locations in India. The quantification of precocene and caryophyllene oxide contents in water and ethanolic extract of different accessions of *Blumea lacera* (Burm.f.) DC collected from different geographical locations in India was caried out using HPLC methods. The results of quantification of Precocene I and II in different accessions of *Blumea lacera* delineated that the *Blumea lacera* accession CVP-05-04 contains highest quantity of Precocene I i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%). The results of quantification of Caryophyllene oxide in different accessions of *Blumea lacera* revealed that *Blumea lacera* accession CVP-05-04 contains highest quantity of Caryophyllene oxide i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%). In conclusion, presence in highest quantities of Precocene I and Caryophyllene oxide in CVP-05-04 accession of *Blumea lacera* would be responsible ethnopharmacological activities of *Blumea lacera* accession CVP-05-04. Hence, CVP-05-04 variety of *Blumea lacera* could be explored for ethnopharmacological uses.

Keywords: *Blumea lacera*, Ethnopharmacological uses, Geographical locations, Precocene, Caryophyllene oxide

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

Introduction

Indian flora has been at the forefront of addressing all kinds of health problems for centuries. Climate diversity of India has been the major reason for the variation in quantity and quality of chemicals present in the same plant. In such a situation, scientific observation and validation of elite chemotype of concern medicinal plants on the basis of geographical diversity has become necessary so that industrial demand can be easily fulfill with minimally spent.

Blumea lacera (Burm.f.) DC is an erect, leafy, herbaceous annual plant under Asteraceae family. It is a well-known ethno-botanically explored medicinal plant.[1] Rainy season is the favorable condition for the full growth of Blumea and is a cosmopolitan habitat in the tropical and sub-tropical zones of Asia. *Blumea* is well documented in ayurveda as a potent medicine for several ailments.[2] Whole plant parts of *Blumea* contain an essential oil that has cineol, fenchone, contanol and camphor.[3] It is very famous for Nagi camphor that contains important useful bio-active compounds as β-caryophyllene, precocene and α-humulene,[2,4] and that has potent tick repellent property.[5]

Earlier research studies on *Blumea lacera* have been observed several types of secondary metabolites under phenols, flavonoids, terpene, sterols groups.[6,7] As per modern medicine, whole plant parts of *Blumea lacera* are used in various human health problems as diuretic, anti-inflammatory, anti-microbial, anti-dysentric, anthelmintic, cholera and respiratory infections.[8,9]

In view of the above facts and based on geographical diversity *Blumea lacera* plant, research has been done to collect *Blumea lacera* from various locations of Indian gangetic plains for the investigation and to establish rationale for ethno pharmacological uses of *Blumea lacera* (Burm.f.) DC accessions collected from difference geographical locations in India.

Materials and Methods

Collection of plant material

Different germplasms of *Blumea lacera* were collected from the natural site between May to December 2022 from different locations of Gangetic plains of India. The plant sample was identified and authenticated by Dr. A K S Rawat, Former Chief Scientist, Pharmacognosy Division, CSIR–NBRI, Lucknow. A passport data of each accession has been prepared depicting information about the altitude, latitude, longitude, phytogeographical zones etc (Table 1).

Table 1. Brief passport data sheet of *Blumea lacera* (Burm.f.) DC

Sample code	Date of collection	Phytogeographical Zone	Location/ Dist./State	Altitude (m)	Latitude (N)	Longitude (E)
CVP-05-01	08/05/2017	MGP	NBRI-LKO, UP	113	26º 55'00″	80º59'00″
CVP-05-02	11/05/2017	MGP	Gaya, Bihar	114	24˚44′55.76″	84˚56′37.44″
CVP-05-03	15/05/2017	MGP	Rajgir, Bihar	96	25˚1′03.35″	85˚29′52.11″

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

CVP-05-04	12/08/2017	LGP	Ranchi, JH	2081	$23^0$25'00.02"	$85^0$19'00.31"
CVP-05-05	15/08/2017	LGP	Garhwa, JH	1143	$23^0$59'29.49"	$83^0$47'10.66"
CVP-05-06	17/08/2017	LGP	Parasnath, Jharkhand	3742	$23^0$57'53.65"	$86^0$08'40.74"
CVP-05-07	23/08/2017	MGP	Bhagalpur, Bihar	39	25°20′52.08″	86°58′56.74″
CVP-05-8	08/12/2017	LGP	Howrah, WB	8	22°35′44.77″	88°15′49.10″
CVP-05-9	09/12/2017	LGP	Kakdwip, WB	5	21°52′45.30″	88°11′38.15″
CVP-05-10	11/12/2017	LGP	Kharagpur, WB	268	22°20′23.99″	88°11′38.15″
CVP-05-11	19/12/2017	MGP	Katihar, Bihar	36	25°33′07.37″	87°34′18.70″
CVP-05-12	22/03/2018	LGP	Barakar, JH	435	23°44′22.37″	86°49′05.45″

LGP, Lower Gangetic Plain, **MGP**, Middle Gangetic Plain; UP, Uttar Pradesh; JH, Jharkhand; WB, West Bengal

Preparation of plant extract

The plant material was manually screened for any impurities and dried in shade. For complete drying, it was kept in hot air oven at 45°C and then powered with an electric grinder. The coarse powder was subjected to ethanol extraction. Extracts were continuously stirred for 6 h and kept at room temperature up to 18 h. The process was repeated up to complete extraction. The extract was filtered and concentrated under vacuum in a rotatory evaporator (Buchi Rotavapor, Switzerland) at 40°C. The extract was finally freeze-dried and stored at 4°C for further use. 10 mg/mL of the extract were used for HPTLC studies. The standard compound Caryophyllene oxide procured from Sigma-Aldrich (USA).

Validation of Elite Bio-active compound (Chemotypic profiling)

The extract was redissolved in methanol and filtered prior to HPTLC analysis. Chromatography was performed on Merck HPTLC precoated silica gel 60G F254 (20 × 10 cm) plates. Methanolic solution of samples and standard compound precocene I of known concentration were applied to the layers as 6 mm-wide bands positioned 10 mm from the bottom and 15 mm from the side of the plate, using a CAMAG Linomat V automated TLC applicator with nitrogen flow providing a delivery speed of 150 nL s^{-1} from the application syringe. These conditions were kept constant throughout the analysis of samples. Following sample application, the layers were developed in a CAMAG twin-trough glass chamber which was pre saturated with mobile phase of hexane–chloroform (6:4) till the proper separation of bands up to 8 cm height. After development, the layers were dried with an air dryer. Precocene I was quantified using a CAMAG TLC Scanner model 3 equipped with CAMAG winCATS IV software. The following scan conditions were

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

applied: slit width, 6 mm × 0.45 mm; wavelength, 300 nm; and absorption–reflection mode. In order to prepare calibration curves, stock solution of precocene I (0.1 mg/mL) was prepared and various volumes of the solution were analyzed by HPTLC; calibration curves of peak area *vs.* concentration were also prepared.

Statistical analysis

All physiological measurements were carried out in triplicates by using three independent plants for each treatment. CropStat program developed at IRRI, Philippines was used for analysis of variance (ANOVA) of experiments. The treatment means were compared by least significant difference (LSD) test at a significance level of $p \leq 0.05$.

Results

The results of yield of water and ethanolic extract of *Blumea lacera* variety CVP-05 collected in different locations was represented in Table 2. Results depicted that the yield of water extract of *Blumea lacera* accession CVP-05-09 collected from Kakdwip, West Bengal was found to highest 27.42% followed by 22.93% (Howrah, WB) and 22.58% (Ranchi, JH) for water extract of *Blumea lacera* accessions CVP-05-08 and CVP-05-04 respectively. Similarly, the yield of ethanol extract of *Blumea lacera* accession CVP-05-03 collected from Rajgir, Bihar was found to highest 24.14% followed by 20.59%, and 17.29% for water extract of *Blumea lacera* accessions CVP-05-09 (Kakdwip, WB) and CVP-05-01 (Lucknow, UP) respectively.

Table 2. Extractive yield for whole plant of *Blumea lacera* accessions using different solvents

Sample code (CVP-05)	Location	Yield of Water Extract (%)	Yield of Ethanol Extract (%)
CVP-05-01	Lucknow, UP	20.06	17.29
CVP-05-02	Gaya, Bihar	16.92	13.64
CVP-05-03	Rajgir, Bihar	17.24	24.14
CVP-05-04	Ranchi, JH	22.58	17.25
CVP-05-05	Garhwah, JH	17.44	12.38
CVP-05-06	Parasnath, JH	12.52	6.42
CVP-05-07	Bhagalpur, Bihar	20.01	14.80
CVP-05-08	Howrah, WB	22.93	16.03
CVP-05-09	Kakdwip, WB	27.42	20.59
CVP-05-10	Kharagpur, WB	15.48	12.71
CVP-05-11	Katihar, Bihar	21.31	15.65
CVP-05-12	Barakar, JH	17.49	13.06

UP, Uttar Pradesh; JH, Jharkhand; WB, West Bengal

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

The results of quantification of Precocene I and II in different accessions of *Blumea lacera* was represented in Table 3. Results revealed that the *Blumea lacera* accession CVP-05-04 contains highest quantity of Precocene I i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%). However, Precocene II was not detected in any of the selected accessions of *Blumea lacera* for this research investigation.

Table 3. Quantification of Precocene I and II in different accessions of *Blumea lacera*

Sample code	**Precocene I (%)**	**Precocene II (%)**
CVP-05-01	0.026	Not detected
CVP-05-02	0.032	Not detected
CVP-05-03	0.044	Not detected
CVP-05-04	0.053	Not detected
CVP-05-05	0.017	Not detected
CVP-05-06	0.010	Not detected
CVP-05-07	0.025	Not detected
CVP-05-08	0.042	Not detected
CVP-05-09	0.018	Not detected
CVP-05-10	0.032	Not detected
CVP-05-11	0.033	Not detected
CVP-05-12	0.047	Not detected

The results of quantification of Caryophyllene oxide in different accessions of *Blumea lacera* was represented in Table 4 and Figure 1 & 2. Results revealed that *Blumea lacera* accession CVP-05-04 contains highest quantity of Caryophyllene oxide i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%).

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

Table 4. Quantification of Caryophyllene oxide in different accessions of *Blumea lacera*

Sample code	Caryophyllene oxide (%)
CVP-05-01	0.026
CVP-05-02	0.032
CVP-05-03	0.044
CVP-05-04	0.053
CVP-05-05	0.017
CVP-05-06	0.010
CVP-05-07	0.025
CVP-05-08	0.042
CVP-05-09	0.018
CVP-05-10	0.032
CVP-05-11	0.033
CVP-05-12	0.047

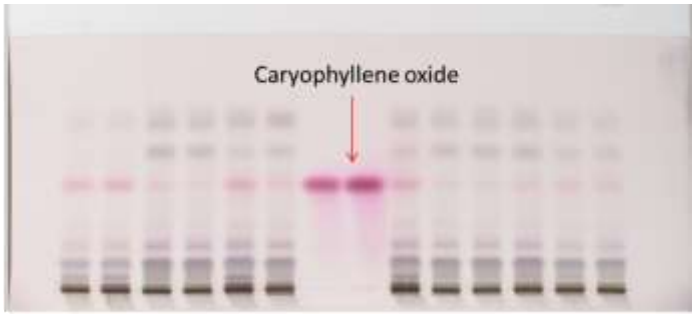

Figure 1. Confirmation of Caryophyllene oxide in CVP-05 accessions of *Blumea lacera* on TLC plate

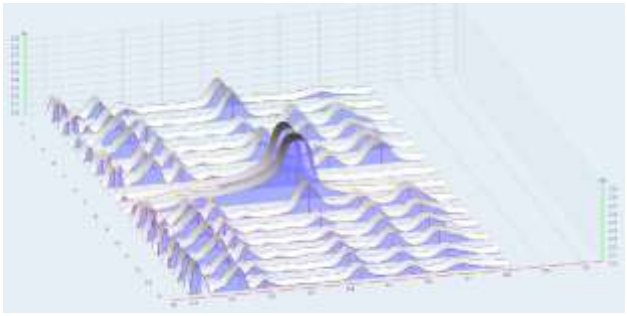

Figure 2. Densitometric profiling of Caryophyllene oxide in CVP-05 accessions of *Blumea lacera*

Discussion

The main objective of the present study is to establish rationale for ethno pharmacological uses of *Blumea lacera* (Burm.f.) DC accessions collected from difference geographical locations in India. The reported phytochemical and pharmacological findings are useful as supporting evidence to interpret and validity of uses of *Blumea lacera*. A plant which is used in one area in the treatment of disease whether similar plant is used in the different area in the treatment of

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

same disease. Use in other area presumably increases likelihood that the plant is active against the illness. The ethno medicinal uses reported from other areas support the folk use, the plant is ranked as possessing highest degree of confidence. The use of *Blumea lacera* for anal fissure and piles/hemorrhoids is corroborated with its antibacterial, anti-inflammatory and analgesic property and also with ethno botanical reports of its use on cuts, wounds, injury, bleeding etc. However similar use has been reported by inhabitants of Noakhali and Nashik districts by Bhowmik et. al. (2014) and Kakulte et. al. (2014).[10,11] Therefore, the use of *Blumea lacera* get support from earlier reports of ethnobotanical work.

In our study, the yield of water extract of *Blumea lacera* accession CVP-05-09 collected from Kakdwip, West Bengal was found to highest 27.42% followed by 22.93% (Howrah, WB) and 22.58% (Ranchi, JH) for water extract of *Blumea lacera* accessions CVP-05-08 and CVP-05-04 respectively. Similarly, the yield of ethanol extract of *Blumea lacera* accession CVP-05-03 collected from Rajgir, Bihar was found to highest 24.14% followed by 20.59%, and 17.29% for water extract of *Blumea lacera* accessions CVP-05-09 (Kakdwip, WB) and CVP-05-01 (Lucknow, UP) respectively. The results of quantification of Precocene I and II in different accessions of *Blumea lacera* delineated that the *Blumea lacera* accession CVP-05-04 contains highest quantity of Precocene I i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%). The results of quantification of Caryophyllene oxide in different accessions of *Blumea lacera* revealed that *Blumea lacera* accession CVP-05-04 contains highest quantity of Caryophyllene oxide i.e., 0.053% followed by CVP-05-12 (0.047%) and CVP-05-03 (0.044%).

Various research investigators in the literature revealed that that different phytochemical of *Blumea lacera* reported to be responsible for different pharmacological activities of *Blumea lacera*. Satyal et. al. (2015) reported the presence of Lachnophyllum ester which possess antibacterial and antifungal properties. Many bioactive phytochemicals are present in *Blumea lacera* and its allied species. These chemicals are also present in the plants belonging to different taxa. These phytochemicals are showing promising pharmacological activities therefore it is necessary to record their biological activities so as to evaluate medicinal value of *Blumea lacera*. Citral (Gernial) is present in the oil extracted from *Blumea lacera*. It is used as a flavoring agent in confectionary, soft drink and cosmetics such as after shaves and body lotions. Germacrene ($C_{15}H_{24}$) are a class of volatile organic hydrocarbons specially sesquiterpenes have been reported to have antimicrobial.[12]

β-farnesene is present in *Blumea lacera*. Santos et. al. (2013) detected β-farnesene from essential oil from Drimys angustifolia (Winteraceae) reported to have antibacterial property. β-caryophyllene is a natural bicyclic sesquiterpene abundantly found in essential oil from various spices, fruits, medicinal plants and as well from *Blumea lacera*. It is approved by US Food and Drug Administration and European agencies as food additive, taste enhancer and flavoring agent and termed as phytocannabinoid.[13] Sharma et. al. (2016) reported its anti-inflammatory, antimicrobial activites.[14] Khandekar et.al. (2013) and Salisu et. al. (2015) revealed presence of important chemicals such as α-carotene, β-carotene and phenolic glycosides from *Blumea lacera*.[15,16] α-amyrin is a pentacyclic triterpene present in this plant has been reported to show

ISSN PRINT 2319 1775 Online 2320 7876

 UGC CARE Listed (Group -I) Journal Volume 11,Iss 12, 2022

anti-inflammatory activity reported by Singh et. al., (2015).[17] The main constituents of essential oil is thymoquinoldimethyl-ether. Essential oil contains interesting components such as (-)-borneol and (-)-camphor which is used in perfumery. The oil possesses antibacterial, antifungal activity. The plant and its defatted extract exhibited anti-inflammatory activity. (+)-borneol exerted remarkable antihyperalgesic effects in a mouse model of oxaliplatin-induced neuropathic pain.

Conclusion

In conclusion, presence in highest quantities of Precocene I and Caryophyllene oxide in CVP-05-04 accession of *Blumea lacera* would be responsible ethnopharmacological activities of *Blumea lacera* accession CVP-05-04. Hence, CVP-05-04 accession of *Blumea lacera* could be explored for ethnopharmacological uses.

Author Contribution Statement

Pragyan Paramita Sinha and Kamal Kant Patra were involved in the execution of all the field collection & laboratory work and data analysis. A K S Rawat was contributed his valuable guidance as plant identification. Keshamma E contributed in her guidance on laboratory skills for research work and in preparation of the present manuscript. All authors have read and approved the final manuscript before its submission.

Conflict of Interest

None to declare.

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