



International Journal of Research in Agronomy

E-ISSN: 2618-0618
P-ISSN: 2618-060X
© Agronomy
NAAS Rating (2026): 5.20
www.agronomyjournals.com
2026; 9(8): 1102-1105
Received: 02-06-2026
Accepted: 08-07-2026

Shivakumar SN
Ph.D. Scholar, Department of
Plantation, Spices, Medicinal and
Aromatic Crops, College of
Horticulture, Bengaluru,
Karnataka, India

K Hima Bindu
Former Principal Scientist, ICAR-
Indian Institute of Horticultural
Research, Bengaluru, Karnataka,
India

Debi Sharma
Former Principal Scientist &
Incharge Food Safety Referral
Laboratory, ICAR-Indian
Institute of Horticultural
Research, Hesaraghatta,
Bengaluru, Karnataka, India

K Umesha
Former Dean, College of
Horticulture, Bengaluru,
Karnataka, India

AP Mallikarjuna Gowda
Professor and Senior Farm
Superintendent, ZARS, UAS,
GKVK, Bengaluru, Karnataka,
India

BN Maruthiprasad
Professor & Head, Department of
Plantation, Spices, Medicinal and
Aromatic Crops, College of
Horticulture, Bengaluru,
Karnataka, India

Corresponding Author:
Shivakumar SN
Ph.D. Scholar, Department of
Plantation, Spices, Medicinal and
Aromatic Crops, College of
Horticulture, Bengaluru,
Karnataka, India

Chemical profiling of essential oil from *Piper betle* L. cv. Mysore local by GC–MS/MS

**Shivakumar SN, K Hima Bindu, Debi Sharma, K Umesha, AP Mallikarjuna
Gowda and BN Maruthiprasad**

DOI: <https://www.doi.org/10.33545/2618060X.2026.v9.i8n.6388>

Abstract

Piper betle L. (betelvine) is an economically and culturally important crop valued for its characteristic aroma and diverse phytochemical constituents. The composition of essential oil varies among betelvine cultivars and contributes substantially to their characteristic aroma and biological properties. The present study was undertaken to characterize the volatile constituents of essential oil from *Piper betle* L. cv. Mysore Local using gas chromatography–tandem mass spectrometry (GC–MS/MS). The essential oil was subjected to chromatographic separation using a Shimadzu GC–MS/MS–TQ8040 system, and volatile constituents were identified based on retention indices and mass spectral comparison with the NIST 2015 library. A total of 25 volatile constituents were identified, accounting for approximately 100% of the total chromatographic peak area. Caryophyllene (13.08%) was the predominant constituent, followed by Germacrene D (10.52%), Cubenene (8.95%), β -elemene isomer (6.38%), γ -muurolene (5.77%), a bicyclo [5.2.0] nonane derivative (5.37%) and Spathulenol (5.32%). Other constituents included Eucalyptol, α -Cadinol, (-)-Globulol, β -Ocimene, Viridiflorol and Epicubenol. The results demonstrate a complex volatile profile of *P. betle* cv. Mysore Local, characterized predominantly by sesquiterpene-type constituents. The chemical profile provides useful information for characterization of this cultivar and contributes to the understanding of essential-oil diversity in betelvine.

Keywords: *Piper betle*, betelvine, essential oil, GC–MS/MS, Mysore Local, volatile constituents, chemical profiling

1. Introduction

Piper betle L., commonly known as betelvine, is an important perennial climber belonging to the family Piperaceae and is widely cultivated in tropical and subtropical regions. The leaves are traditionally consumed as a masticatory and are also valued for their culinary, cultural and medicinal significance. Betelvine germplasm comprises numerous cultivars and locally recognized types that differ in morphological, sensory and biochemical characteristics (Chattopadhyay and Maity, 1967) [4].

The characteristic aroma of betel leaves is primarily attributed to their essential oils, which contain diverse volatile constituents, including phenolic and terpene-like compounds (Bajpai *et al.*, 2010a) [2]. Chemical profiling of betelvine leaf essential oils has demonstrated variation in their chemical composition among different landraces, with distinct chemical profiles reported for major betelvine groups (Rawat *et al.*, 1989) [13]. Such variation in volatile composition may contribute to differences in the characteristic aroma and other phytochemical properties of betelvine types. Chemical characterization of essential oils provides a useful approach for documenting variation in volatile composition among plant materials. Gas chromatography coupled with mass spectrometry (GC–MS) is a widely used analytical approach for separation and characterization of volatile constituents based on their chromatographic and mass-spectral properties.

Previous studies have reported considerable variation in the essential-oil composition of *P. betle* and have identified diverse volatile constituents associated with the characteristic chemical profile of betel leaves (Bajpai *et al.*, 2010a; Rawat *et al.*, 1989) [2, 13]. The reported biological properties of betelvine essential oils and their constituents further emphasize the importance of characterizing the volatile fraction (Kumar *et al.*, 2010; Rai *et al.*, 2011) [8, 12].

Although several betelvine types have been characterized for their chemical constituents, cultivar-specific information on essential-oil composition remains valuable for documenting the diversity of volatile compounds. Mysore Local is one of the betelvine types maintained among the germplasm evaluated in the present study. Therefore, the present investigation was undertaken to characterize the essential-oil profile of *Piper betle* L. cv. Mysore Local using GC–MS/MS and to identify its major volatile constituents based on retention-index and mass-spectral information.

2. Materials and Methods

2.1 Plant material

Fresh leaves of *Piper betle* L. cv. Mysore Local were used for essential-oil analysis. The cultivar was maintained as part of the betelvine germplasm evaluated in the study.

2.2 Essential oil extraction and sample preparation

Essential oil from *P. betle* cv. Mysore Local was obtained by hydrodistillation using a standardized procedure developed during the study. The resulting essential oil was subjected to GC–MS/MS analysis for characterization of its volatile constituents.

2.3 GC–MS/MS analysis

The essential oil was analyzed using a Shimadzu GC–MS/MS-TQ8040 system equipped with a benchtop triple-quadrupole mass spectrometer operated in electron ionization (EI) mode. An AOC-20i autosampler was used for sample injection. Chromatographic separation was performed using an SPTM 2560 capillary column (100.0 m length × 0.25 mm internal diameter × 0.2 µm film thickness).

The oven temperature was initially maintained at 100 °C for 5 min, followed by an increase at 4 °C/min to 220 °C, where it was maintained for 30 min, resulting in a total run time of 65 min. Helium was used as the carrier gas at a constant flow rate of 1.00

mL/min. An injection volume of 1 µL was used, with the injector temperature maintained at 220 °C and a split ratio of 40:1. The MS interface and ion-source temperatures were maintained at 220 °C, and the solvent cut time was 13.2 min. The mass scan range was 45–500 m/z, with a sampling speed of 1666.

The relative abundance of individual volatile constituents was expressed as percentage peak area based on the total chromatographic peak area. Compound identification was carried out by comparing retention indices calculated using a homologous series of n-alkanes (C5–C32) with reference information and by matching the acquired mass spectra against the NIST 2015 spectral library.

3. Results

3.1 Essential-oil composition of *P. betle* cv. Mysore Local

GC–MS/MS analysis of the essential oil of *P. betle* cv. Mysore Local revealed 25 identifiable volatile constituents, accounting for approximately 100% of the total chromatographic peak area (Table 1). Considerable variation was observed in the relative abundance of individual compounds (Figure 1).

Caryophyllene was the predominant constituent, accounting for 13.08% of the total peak area, followed by Germacrene D (10.52%) and Cubenene (8.95%). Other major constituents included β-elemene isomer (6.38%), γ-muurolene (5.77%), Bicyclo [5.2.0] nonane, 2-methylene-4,8,8-trimethyl-4-vinyl- (5.37%), and Spathulenol (5.32%).

Eucalyptol (4.50%), α-Cadinol (4.29%), (-)-Globulol (4.21%), β-Ocimene (3.20%), Viridiflorol (3.33%), Epicubenol (2.95%) and Neointermedeol (2.70%) were also detected. Other constituents included Viridiflorene (3.46%), δ-Elemene (1.42%), α-Copaene (2.35%), Linalool (2.22%), Eremophyllene (2.27%), Bicyclogermacrene (2.09%), Safrole (0.81%), Junenol (1.24%), Rosifoliol (1.50%), Methyleugenol (0.69%) and τ-Cadinol (1.36%).

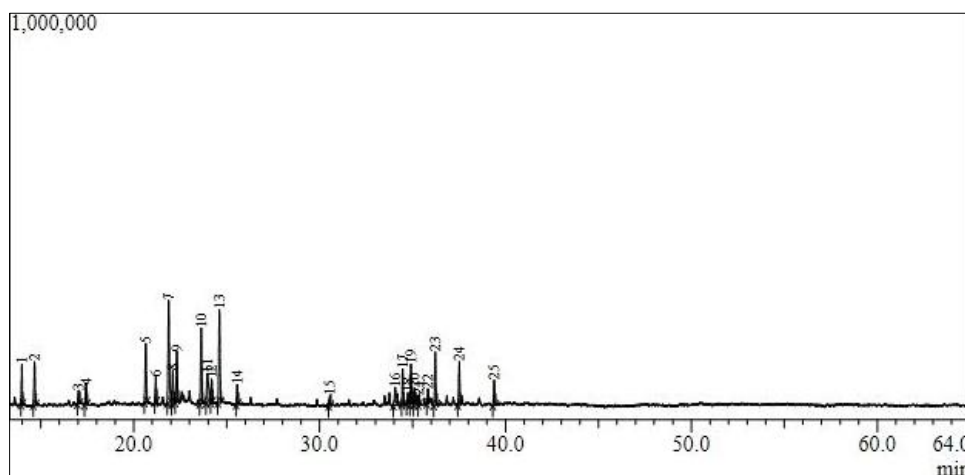


Fig 1: Total ion chromatogram of *P. betle* cv. Mysore Local

Table 1: GC–MS/MS profile of essential-oil constituents of *Piper betle* L. cv. Mysore Local

Peak#	R. Time	Area%	Name
1	13.983	3.2	.beta.-Ocimene
2	14.666	4.5	Eucalyptol
3	17.035	1.42	delta.-Elemene
4	17.439	2.35	.alfa.-Copaene
5	20.649	6.38	Beta elemene isomer
6	21.19	2.22	Linalool
7	21.882	13.08	Caryophyllene
8	22.097	3.46	Viridiflorene

9	22.319	5.77	.gamma.-Muurolene
10	23.631	8.95	Cubenene
11	23.963	5.37	Bicyclo[5.2.0]nonane,2-methylene-4,8,8-trimethyl-4-vinyl-
12	24.194	2.27	Eremophyllene
13	24.609	10.52	Germacrene D
14	25.57	2.09	Bicyclogermacrene
15	30.545	0.81	Safrole
16	34.061	2.95	Epicubenol
17	34.47	3.33	Viridiflorol
18	34.751	1.24	Junenol
19	34.912	4.21	(-)-Globulol
20	35.101	1.5	Rosifoliol
21	35.328	0.69	Methyleugenol
22	35.827	1.36	.tau.-Cadinol
23	36.221	5.32	Spathulenol
24	37.512	4.29	.alpha.-Cadinol
25	39.39	2.7	Neointermedeol
		100	

4. Discussion

The essential oil of *Piper betle* L. cv. Mysore Local exhibited a chemically diverse profile comprising 25 tentatively identified volatile constituents, representing different classes of terpenoid and related compounds. Considerable variation in the relative abundance of individual constituents was observed, with caryophyllene (13.08%), germacrene D (10.52%) and cubenene (8.95%) representing the three major components. Together, these compounds accounted for approximately 32.55% of the total chromatographic peak area, indicating their substantial contribution to the volatile profile of cv. Mysore Local. The occurrence of diverse sesquiterpenes, monoterpenes and phenylpropanoid-related compounds is consistent with the reported chemical complexity of *P. betle* essential oils (Bajpai *et al.*, 2010a; Rawat *et al.*, 1989) [2, 13].

Caryophyllene was the predominant constituent in the essential oil of Mysore Local, accounting for 13.08% of the total peak area. Caryophyllene has previously been reported as an important constituent of *P. betle* essential oils, although its relative abundance varies considerably among genotypes and cultivars. Earlier studies have reported caryophyllene in different *P. betle* materials, including Bangla betel leaves and cured betel leaves (Joseph and Kalesh, 2015; Saini *et al.*, 2020; Madhumita *et al.*, 2019) [7, 14, 10]. The relatively high abundance of caryophyllene observed in Mysore Local therefore contributes substantially to the sesquiterpene-rich character of its essential-oil profile.

Germacrene D was the second major constituent, accounting for 10.52% of the total peak area. Germacrene D has been reported as a prominent sesquiterpene in certain *P. betle* varieties, including Sirugamani-1 and Kapoori (Sugumaran *et al.*, 2011; Chakravarthi *et al.*, 2018) [16, 3]. The relatively high level of germacrene D in Mysore Local is therefore consistent with previous observations demonstrating the contribution of germacrene-type sesquiterpenes to the volatile composition of betelvine.

Cubenene was the third major constituent, representing 8.95% of the total chromatographic area. Other relatively abundant constituents included β -elemene isomer (6.38%), γ -muurolene (5.77%) and spathulenol (5.32%). The occurrence of these compounds indicates that sesquiterpenes and oxygenated sesquiterpenes constitute an important component of the essential-oil profile of Mysore Local. Considerable variation in germacrene-related compounds, muurolenes and oxygenated sesquiterpenes has also been reported among *P. betle* accessions from different geographical regions (Gocchi *et al.*, 2023; Saini *et al.*, 2020) [6, 14].

The presence of γ -muurolene (5.77%) further supports the importance of sesquiterpenes in the essential oil of Mysore Local. Previous studies have reported γ -muurolene in *P. betle* essential oils at variable concentrations, demonstrating differences in its accumulation among betelvine genetic materials (Gocchi *et al.*, 2023; Saini *et al.*, 2020) [6, 14]. Similarly, the detection of oxygenated sesquiterpenes such as spathulenol, α -cadinol, viridiflorol and globulol indicates the chemical complexity of the oil. Comparable oxygenated sesquiterpenes have been reported in *P. betle* accessions from different geographical regions, including δ -cadinol, caryophyllene oxide, spathulenol and α -cadinol.

Monoterpene-related constituents were also detected in the essential oil of Mysore Local. Eucalyptol (4.50%), β -ocimene (3.20%) and linalool (2.22%) were present at lower relative abundances than the major sesquiterpene constituents. Linalool has previously been reported in *P. betle* essential oils, with considerable variation among genotypes. Saini *et al.* (2020) [14] reported relatively high linalool content in Bangla betel, while its occurrence in Sri Lankan accessions has also been documented (Kumaratunge *et al.*, 1999; Saini *et al.*, 2020) [9, 14]. The moderate level of linalool observed in Mysore Local indicates that it contributes to the volatile profile without being a predominant constituent of this cultivar.

Phenylpropanoid-related constituents were detected at comparatively low levels in Mysore Local. Safrole (0.81%) and methyleugenol (0.69%) were present in the analyzed oil. The relatively low abundance of these compounds, compared with the major sesquiterpenes, suggests that the essential-oil profile of Mysore Local is predominantly characterized by terpenoid constituents rather than being strongly phenylpropanoid-dominant. Safrole-rich chemotypes have been reported in several *P. betle* cultivars and geographical types, demonstrating substantial variation in phenylpropanoid composition among betelvine genetic materials (Arambewela *et al.*, 2005; Saxena *et al.*, 2014; Dwivedi and Tripathi, 2014) [1, 15, 5].

The identification of 25 volatile constituents using retention-index information together with mass-spectral library matching provides a useful chemical profile of *P. betle* cv. Mysore Local. The combined use of retention indices and mass-spectral comparison provides greater confidence in compound annotation than reliance on mass-spectral matching alone. However, since authentic reference standards were not used for confirmation of each individual compound, the detected compounds are appropriately described as tentatively identified rather than definitively identified.

The chemical composition of *P. betle* essential oil is known to

vary considerably among cultivars and genotypes. Previous investigations have demonstrated distinct chemical profiles and chemotypes characterized by differences in the relative abundance of sesquiterpenes, monoterpenes and phenylpropanoid-related compounds (Arambewela *et al.*, 2005; Saxena *et al.*, 2014; Sugumaran *et al.*, 2011; Gocchi *et al.*, 2023) [1, 15, 16, 6]. The variation in major volatile constituents reported among *P. betle* genotypes supports the usefulness of essential-oil profiling for documenting chemical diversity within the species. In the present study, the predominance of caryophyllene, germacrene D and cubenene distinguishes the volatile profile observed in Mysore Local from profiles dominated by other compounds in different betelvine genetic materials. Such cultivar-associated differences may contribute to variation in the characteristic aroma and other phytochemical attributes of betelvine. However, direct relationships between individual volatile constituents and sensory characteristics would require dedicated sensory and aroma studies. Overall, the essential oil of *P. betle* cv. Mysore Local was characterized predominantly by sesquiterpenoid constituents, with caryophyllene, germacrene D and cubenene as the major components. The presence of multiple sesquiterpenes, oxygenated sesquiterpenes, monoterpenes and phenylpropanoid-related compounds highlights the chemical complexity of this cultivar. The profile obtained in the present study provides useful baseline information for comparison with other *P. betle* cultivars and for future investigations into the relationships between essential-oil composition, aroma characteristics and biological properties.

5. Conclusion

The essential oil of *Piper betle* L. cv. Mysore Local exhibited a diverse volatile profile comprising 25 tentatively identified constituents. Caryophyllene (13.08%), Germacrene D (10.52%) and Cubenene (8.95%) were the predominant constituents, followed by β -elemene isomer, γ -muurolene, a bicyclo[5.2.0]nonane derivative and spathulenol. The GC-MS/MS profile demonstrated considerable chemical diversity within the essential oil and provided useful information for the chemical characterization of the Mysore Local cultivar. The volatile profile obtained in the present study provides a useful basis for future comparative studies of essential-oil composition among betelvine cultivars.

References

1. Arambewela L, Kumaratunga KGA, Dias K. Studies on *Piper betle* of Sri Lanka. J Natl Sci Found Sri Lanka. 2005;33(2):6-16.
2. Bajpai V, Sharma D, Kumar B, Madhusudan KP. Profiling of *Piper betle* Linn. cultivars by direct analysis in real time mass spectrometric technique. Biomed Chromatogr. 2010;24:1283-1286.
3. Chakravarthi VP, Murugesan S, Arivuchelvan A, Sukumar K, Arulmozhi A, Jagadeeswaran A. GC-MS profiling of methanolic extract of *Piper betle* leaf. J Pharmacogn Phytochem. 2018;7:2449-2452.
4. Chattopadhyay SB, Maity S. Diseases of betelvine and spices. New Delhi: Indian Council of Agricultural Research; 1967.
5. Dwivedi V, Tripathi S. Review study on potential activity of *Piper betle*. J Pharmacogn Phytochem. 2014;3(4):93-98.
6. Gocchi SK, Patra B, Dey SK. Comparative study of bioactive components of betel leaf essential oil. Gradiva Rev J. 2023;9(9):8-20.
7. Joseph P, Kalesh NS. Black pepper leaf oil mass spectrometric variation. Int J Comput Appl. 2015;129(16):40-43.
8. Kumar N, Misra P, Dube A, Bhattacharya S, Dikshit M, Ranade S. *Piper betle* L.: a maligned pan-Asiatic plant with pharmacological prospects. Curr Sci. 2010;99(7):922-932.
9. Kumaratunge KGA, Arambewela LSR, Ekanayake S, Dias K. Preliminary studies on *Piper betle* L. (betel vine). In: Proceedings of the 55th Annual Sessions, Sri Lanka Association for the Advancement of Science; Colombo, Sri Lanka; 1999. p. 216.
10. Madhumita M, Guha P, Nag A. Extraction and GC-MS analysis of betel leaf essential oil. Ind Crops Prod. 2019;138:111578.
11. Parthasarthy VA, Chattopadhyay PK, Bhose TK. Plantation crops. 2nd ed. New Delhi: Oxford and IBH Publishing Co. Pvt. Ltd.; 2009.
12. Rai MP, Thilakchand KR, Palatty PL, Rao P, Rao S, Bhat HP, *et al.* *Piper betle* L. (betelvine), the maligned South East Asian medicinal plant possesses cancer preventive effects: time to reconsider the wronged opinion. Asian Pac J Cancer Prev. 2011;12:2149-2156.
13. Rawat AKS, Tripathi RD, Khan AJ, Balasubrahmanyam VR. Essential oil components as markers for identification of *Piper betle* L. cultivars. Biochem Syst Ecol. 1989;17(1):35-38.
14. Saini S, Nanda S, Dhiman A. GC-MS analysis of bioactives of *Piper betle* Linn. leaf. Curr Bioact Compd. 2020;16(1):24-32.
15. Saxena M, Khare NK, Saxena P, Syamsundar KV, Srivastava SK. Antimicrobial activity and chemical composition of leaf oil in two varieties of *Piper betle* from northern plains of India. J Sci Ind Res. 2014;73(2):95-99.
16. Sugumaran M, Poornima M, Venkatraman S, Lakshmi M, Srinivasanethuvani. Chemical composition and antimicrobial activity of Sirugamani variety of *Piper betle* Linn. leaf oil. J Pharm Res. 2011;4(10):3424-3426.