



PHARMACOGNOSTIC STUDIES AND EXTRACTION OF VOLATILE OIL FROM *CALLISTEMON VIMINALIS* LEAF

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Article Received on
27 Jan. 2019,

Revised on 17 Feb. 2019,
Accepted on 10 March 2019

DOI: 10.20959/wjpps20194-13394

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ABSTRACT

Callistemon Viminalis is commonly known as 'Bottlebrush' plant belonging to family Myrtaceae. It has a got great medicinal importance in ethnic trial communities which are still in practice. The present study deals with the Pharmacognostic studies including examination of morphological and microscopic characters. The volatile oil was extracted by hydro distillation method from leaves of *Callistemon viminalis*. The chemical composition of volatile oil was studied by performing its GC study.

KEYWORDS: *Callistemon viminalis*, Phytochemical evaluation, Pharmacognostic study, Thin layer chromatography.

1. INTRODUCTION

The Genus *Callistemon* R. Br. (commonly known as bottlebrush) belongs to the family Myrtaceae and comprises over 30 species. Myrtaceae is one of the essential oil rich families. The family consists of 130-150 genera. They are woody aromatic trees or shrubs (0.5 m to 7 m tall) widely distributed in the wet tropics, notably Australia, South America and tropical Asia, but are now spread all over the world. *Callistemon* species have attractive narrow foliage and white papery bark. The leaves of *Callistemon* species are lanceolate (3-6 mm wide and 40-70 mm long) in arrangement and very aromatic. The flowers are borne in spikes of about 40-150 mm long with prominent red stamens. Petals are greenish or pale coloured, tiny, inconspicuous and in some cases deciduous. *Callistemon* species are used for forestry, essential oil production, farm tree/windbreak plantings, degraded land reclamation and ornamental horticulture. In China *Callistemon* species, especially *C. viminalis* are used in Traditional Chinese Medicine pills for treating hemorrhoids. *Callistemon* are also used as

weed control and as bioindicators for environmental management. Previous phytochemical investigations of members of this Genus resulted in the identification of C-methyl flavonoids, triterpenoids and phloroglucinol derivatives. Furthermore, piceatannol and scirpusin B isolated from the stem bark of *C. rigidus* from Japan, showed inhibitory effects on mouse α -amylase activity. In addition, antimicrobial, anti-staphylococcal, antithrombin, repellent and nematocidal activities as well as larvicidal and pupicidal values have been reported for the genus. The in vitro antibacterial activity of the essential oils was studied against 12 bacteria strains using disc diffusion and broth microdilution methods. The oils exhibited strong zone of inhibitions against some bacteria such as *S. faecalis* (20.3-24.0 mm), both strains of *S. aureus* (23.0-26.3 mm), *B. cereus* (17.3-19.0 mm) and *S. macrcesens*(11.3-23.7 mm) when compared to standard antibiotics gentamycin and tetracycline used as controls. In this study morphological and microscopical characters, extraction of volatile oil and its Gas Chromatography was carried out.

2. MATERIAL AND METHOD

Identification and Authentication of plant material

The leaves of Bottle brush (*Callistemon viminalis*) is collected from sangola region in the month of January. The leaf of the plant was authenticated from Department of Pharmacognosy, Sahyadri College of Pharmacy, Methwade. The specimen of the plant was kept in the department for future reference.

Extraction of volatile oil (Khandelwal, 2007)

By using clavenger apparatus we have extracted volatile oil. The 100g of *C.viminalis leaves* were crushed were placed in round bottom flask, in which water was added. The heating was switch on and the condensate obtained after 3 hours was collected, which contained water and oil.

GC Analysis

The GC Analysis of the oil was performed on a various 3300 gas chromatogram, using a fused capillary/column (30mm×0.25mm i.d., film, thickness 0.25 μ m) coated with dimethyl siloxane (DB-1). The oventemperature was programmed at 80-2250C at 40C/min, then held isothermally at 2500C. The detector used was FID detector temperature 3000C, injection volume 0.1 μ l and carrier gas nitrogen was used.

3. Pharmacognostic study

Macroscopy and microscopy

The shape, size, color, odor, taste, surface texture and fracture characteristics of the leaves and stems were determined. Microscopy of stem and leaves were studied by taking the transverse section (T.S.). The obtained sections were mounted in glycerine:water, for the identification of various regions. Powder characteristic of the dried stem was separately performed. Phloroglucinol- hydrochloric acid (1:1), iodine and glycerin were used as staining agent.

Morphological study

The leaves of *Callistemon viminalis* were evergreen, lanceolate in shape, alternate in arrangement, entire margin, pinnate venation, very aromatic and stem was grey in color.

Microscopical evaluation

The leaf surface shows the anomocytic stomata which is characteristics of myrtaceae family. Transverse section shows the epidermal layer followed by cuticle layer and vascular bundle (xylem and phloem), pericyclic fibre, collenchyma, unicellular trichomes etc. The transverse section of stem shows epidermal layer, 2-3 layer of cork tissue, 7-8 layer of cortex issue, medullary rays, endodermis, xylem vessels, oil glands, scleridein stellar region and pith at center region (figure 1, 2) observation and result pertaining to specific reagent towards plant tissue were represented.

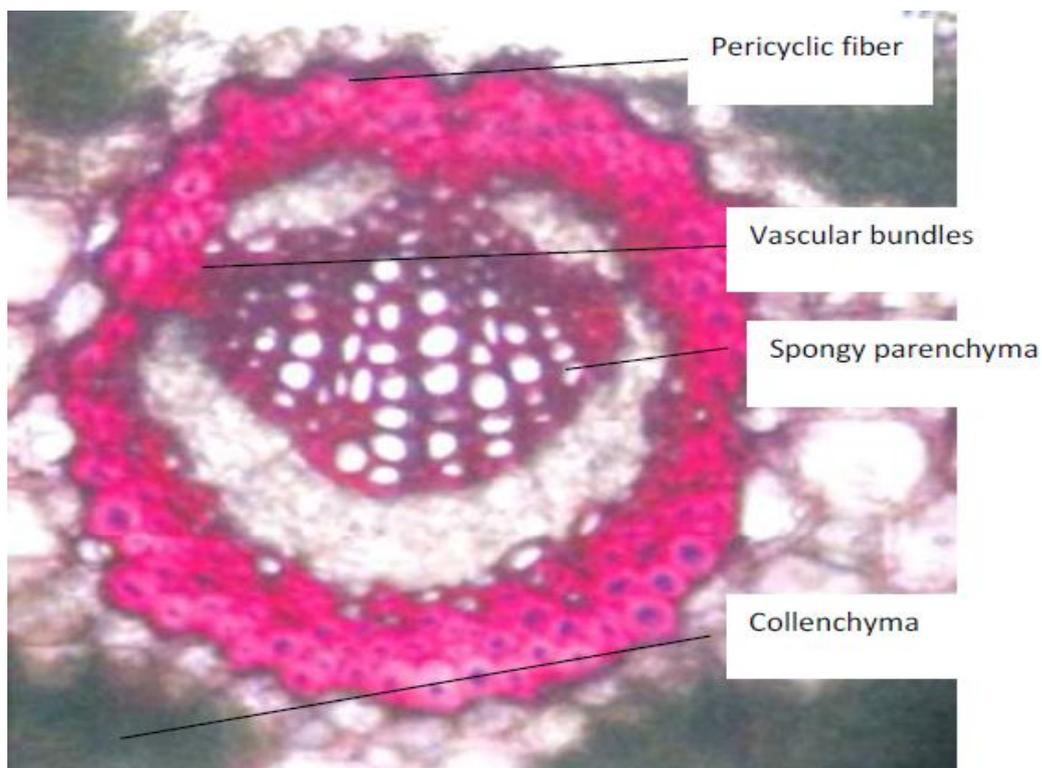


Fig 1: Transverse section of leaves treated with Phloroglucinol: Hydrochloric acid (1:1).

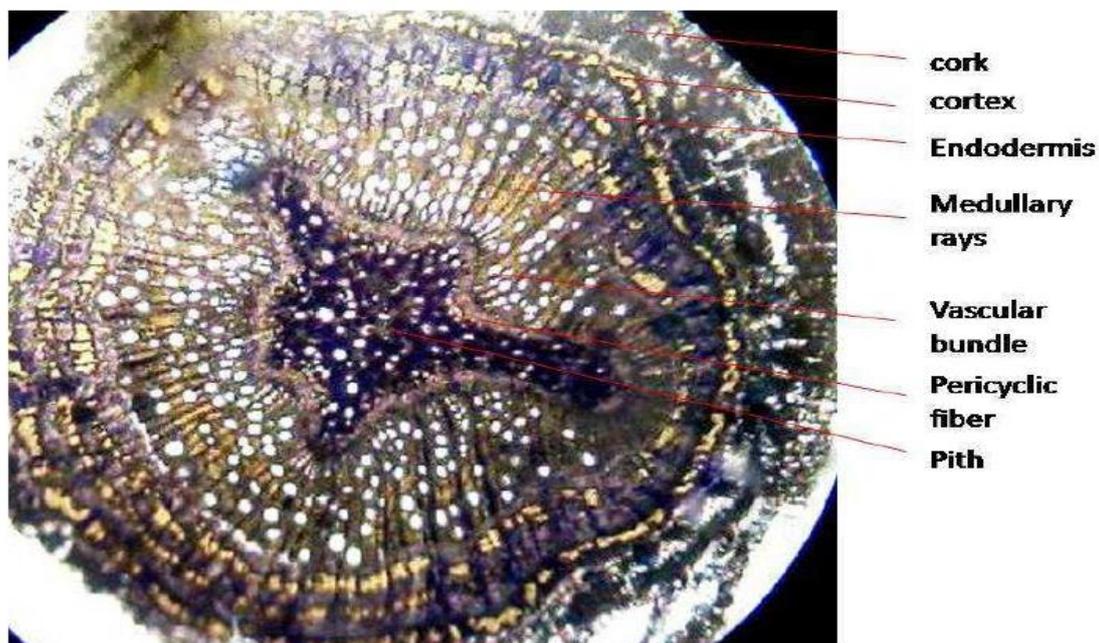


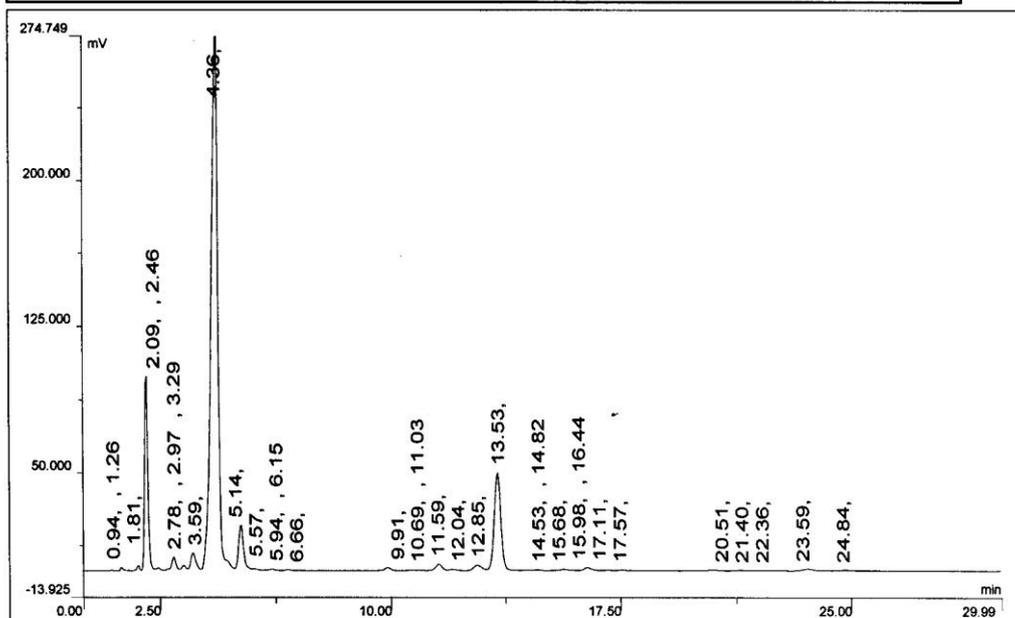
Fig 2: Transverse section of *Callistemon viminalis* stem.

5. GC Analysis

Nirmiti Labs Pvt. Ltd., Mumbai
Name of the Party : Sahyadri College of Pharmacy
Date of Receipt : 13/03/2018

Data File : D:\User1\Mar 2018\048r1303.Dat
Method File : C:\WinchromCrt\CRTdefaultMethod.MET
Sample Name : Bottle Brush Plant Oil **Detector :** FID
Analysis Type : Percent Method **System :** GC

Peak Width	Peak Threshold	Area Reject	Height Reject
4	10	10	10



Area Percent			
Sr.No.	Ret.Time	Area	Area %
No.	Time.	μ volt sec	%
1	0.94	1710.34	0.0277
2	1.26	12028.23	0.1946
3	1.81	13450.97	0.2176
4	2.09	712929.83	11.5320
5	2.46	5477.60	0.0886
6	2.78	1365.83	0.0221
7	2.97	50586.80	0.8183
8	3.29	14506.23	0.2346
9	3.59	88773.49	1.4360
10	4.36	4085280.40	66.0817
11	5.14	233930.86	3.7840

Gas chromatogram of volatile oil isolated from *C.Viminalis*

Sr.No. No.	Ret.Time Time.	Area μ volt sec	Area % %
12	5.57	374.00	0.0060
13	5.94	927.16	0.0150
14	6.15	7425.87	0.1201
15	6.66	5208.91	0.0843
16	9.91	21988.80	0.3557
17	10.69	2595.54	0.0420
18	11.03	216.91	0.0035
19	11.59	44442.40	0.7189
20	12.04	6151.31	0.0995
21	12.85	41124.86	0.6652
22	13.53	738410.40	11.9442
23	14.53	377.03	0.0061
24	14.82	3031.09	0.0490
25	15.68	11896.61	0.1924
26	15.98	1839.45	0.0298
27	16.44	25171.49	0.4072
28	17.11	1716.34	0.0278
29	17.57	4750.86	0.0768
30	20.51	8038.80	0.1300
31	21.40	3082.80	0.0499
32	22.36	2816.86	0.0456
33	23.59	22067.09	0.3569
34	24.84	8467.14	0.1370
		6182162.29	100.0000

Summary

Total Peaks : 34
Multiplication Factor : 1.0000
Dilution Factor : 1.0000

Sample Amount : 100.0000

6. RESULT AND DISCUSSION

The volatile oil from the *C. viminalis* leaves was isolated by using hydrodistillation method. The percentage yield of volatile oil was 1%v/w. The isolated volatile oil was further analysed by means of gas chromatography, as represented in gas chromatogram, which showed 34 peaks indicating 34 constituents presents, in which major chemical constituent found was 1, 8 cineole at retention time 4.36 minutes.

7. CONCLUSION

Volatile oil of leaf contain one important component is 1, 8 cineole the future work is to investigate the biological activities of different plant extract and volatile oil of *Callistemon viminalis* leaf.

8. ACKNOWLEDGEMENT

We are thankful to the Sahyadri College of Pharmacy Methwade, Sangola, Solapur, Maharashtra, India for providing all the necessary facilities.

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