



## Evaluation of Bioactive Compounds in Schizomussaenda Henryi Oil by Gas Chromatography Mass Spectrometry (GC-MS)

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### ABSTRACT

This study was carried out to evaluate the bioactive compounds in Schizomussaenda henryi oil by Gas Chromatography –Mass Spectrometry (GC-MS). Oil from Schizomussaenda henryi oil was extracted using steam distillation technique. Evaluation of Schizomussaenda henryi oil reveals the presence of 39 bioactive compounds representing 88.56 %. The major bioactive compounds identified by GC-MS were;  $\alpha$ -Phellandrene (16.31 %), 4-Trifluoroacetyltetradecane (11.92 %), 9,17-Octadecadienal (9.12 %), Palmitoleic acid (8.72 %), Squalene (8.52 %),  $\alpha$ -Panasinene (6.77 %),  $\beta$ -Pinene (5.72 %),  $\beta$ -Phellandrene (5.11 %) and Naphthalene (4.08 %) while Benzene,1,4-dichloro (0.01 %), Hexatriacontane (0.03 %), 2-Ethylhexyl mercaptoacetate (0.07 %), Humulene epoxide II (0.07 %) amongst others were found in lower concentrations. These compounds contain a cocktail of phyto-compounds with therapeutic properties including, anti-inflammatory, immuno-stimulatory, antifungal, antiviral, anti-helminthic, antidiabetic, hepato-protective, gastro-protective, antimicrobial, and antidiabetic. They also show strong antibacterial action against some pathogenic organisms and neutralize the activities of free radicals. They can also reduce the increasing cases of antimicrobial resistance or multi-drug resistance

## INTRODUCTION

*Schizomussaenda henryi*, a flowering plant and erect shrub belongs to the family Rubiaceae. The plant is native to China and has recently been found in Thailand, Pakistan and some parts of India including Rajasthan (Lan et al., 2004). The plant primarily grows in wet tropical biome and has a long history and traditional medicine. It is one of nature's most overlooked medicines and a pharmacy in disguise, the leaves, stems, roots, seeds, flowers all carry unique phytochemicals (Xias and Lian, 1999; Wang and Zhong, 2004). For centuries, traditional healers have used the plant to cure all forms of infections (Deng et al., 2001; Deng, 2002). Inside the leaves are bioactive compounds that go beyond nutrition helping to fight diseases naturally (Liu, 1994; Liu and Ding, 1995). The leaves of *Schizomussaenda henryi* are elliptic or narrowly elliptic with rounded base, up to 10–25 by 2.5–8.0 cm (Li, 1991). Flowers hermaphrodite, heterostylous, 5-merous; flowers long-styled: calyx tube elliptic, 3.22 to 4.36 by 1.86 to 2.76 mm, calyx lobes normal lanceolate, 1.45 to 3.51 mm, petaloid calyx lobes white, ovate, 5.50 to 10.5 by 2.5 to 4.9 cm, with long-petiole 1.5 to 3.3 cm long; corolla salverform, corolla-tube 2.57 to 3.63 cm long, corolla lobes broadly obovate, with long apiculate, orange-yellow; stamens 5, inserted in middle of corolla; anther linear, 3.23 to 4.41 mm long; style 2.63 to 3.46 cm long, stigma bifid, 2.33 to 3.54 mm long; flowers short-styled: calyx and corolla similar to flowers long-styled, stamens 5; anther linear, 3.05 to 4.44 mm long; style 1.87 to 2.55 cm long; stigma bifid, 2.85 to 3.79 mm long (Alejandro et al., 2005; Bremer and Thulin, 1998).

## LITERATURE REVIEW

In folk medicine, it can be used for treating haemorrhoids when applied topically. The root extract of *Schizomussaenda henryi* can be used to treat conjunctivitis, tuberculosis, cough and ease blood pressure. Its leaf infusions are used to treat gastro-intestinal discomfort, respiratory issues and sexually transmitted diseases (Sumei et al., 2006; Long and Li, 2004). The leaves contain a cocktail of bioactive compounds with powerful therapeutic activities like, anti-inflammatory (ITCM, 1988), antifungal, antiviral, hypolipdemic, cardio-protective (Xu et al., 2002), immuno-stimulatory, hepato-protective (ITCM, 1988; Li and Long, 2000), anti-helminthic and antioxidant (Deng et al., 2002). It can also relieve chronic inflammation without harming healthy tissues, lowers blood glucose, show strong antibacterial action against *Escherichia coli* and *Salmonella* spp and protects the liver against oxidative stress (Hedberg, 1993).

Modern research has barely begun to explore the potentials of *Schizomussaenda henryi*. Scientific studies are sparse and robust investigation on its chemical chemistry, mechanisms and safety remains largely unwritten. Therefore, this study is aimed at evaluating the bioactive compounds of *Schizomussaenda henryi* oil by using GC-MS technique.

## METHODOLOGY

### Materials and Methods

#### Site of the Study

This study was undertaken at the Department of Animal Biochemistry, Gandhi College of Agriculture, Rajasthan, India between the month of April to June, 2024.

#### Plant Collection, Authentication and Essential Oil Extraction Technique

Samples of *Schizomussaenda henryi* leaves were collected from different locations in Rajasthan, India and sent to the Department of Taxonomy, Gandhi College, Rajasthan for proper authentication and identification. It was assigned a reference number HD/08C/2024 and shade dried at room temperature for 13 days, pulverized using an electrical blender prior to extraction. The essential oil from the leaf *Schizomussaenda henryi* leaves was obtained by steam distillation with H-Shaped Clavenger apparatus according to the procedure recently published by Hernandez and Alagbe (2025a). Briefly, 500 g of pulverized *Schizomussaenda henryi* leaves was heated in a conical flask contained at a temperature of 60°C for 20 minutes, steam obtained was collected in a condenser. After cooling, mixture of steam and oil was collected in a beaker and later transferred into a separator. The oil obtained was dried over anhydrous sodium sulphate, stored in vials and sent to the laboratory for further examination.

#### GC-MS Analysis of *Schizomussaenda Henryi* Oil

The volatile compounds in *Schizomussaenda henryi* oil was carried out using Perkin Elmer GC-MS (Model GC 2400, USA). 2 mL of *Schizomussaenda henryi* oil was injected via the injection port and equipped with a HP-5ms fused silica capillary column (30 m x 0.25 mm, film thickness 0.25 µm). Analytical conditions were: Oven temperature of 50 °C, with 5 minutes' initial hold, and then to 200 °C at 10 °C/min, with final hold time of 10 minutes; helium was used as carrier gas at a flow rate of 1 mL/min. The mass spectrometry section was operated in EI mode with ionization voltage 85 eV and ion source temperature between 200 - 250 °C. Identification confirmation was by comparison of their mass spectra with reference compounds from the Library of National Institute of Standard and Technology (NIST, 2011) and Willey database compared with mass spectra of published spectra (Adams, 2007).

Table 1. List of Bioactive Compounds Identified in *Schizomussaenda Henryi* Oil By GC-MS

Compounds	Reaction time (min)	% Area
9,17-Octadecadienal	6.07	9.12
2-Ethylhexyl mercaptoacetate	7.11	0.07
2,6-Dimethyldecane	9.06	0.12
Nonane, 3-methyl	9.33	0.49
Hexatriacontane	10.52	0.03
Sulfurous acid, butyl octyl ester	10.95	0.51
N-3-methylButyl acetamide	12.68	0.16
Benzene,1,4-dichloro	12.83	0.01
Benzene,1,2,3-trimethyl	12.97	0.62

Heptadecane, 2,6-dimethyl	13.12	0.09
Bis(2-ethylhexyl) phthalate	14.11	1.42
4- Trifluoroacetoxytetradecane	14.63	11.92
$\beta$ -Phellandrene	15.06	5.11
$\alpha$ -Humulene	15.39	0.58
$\delta$ -Elemene	16.74	0.13
Nonane, 4-methyl	18.02	0.04
Pentadecane	18.35	1.45
$\beta$ -Famesene	18.69	1.94
3-Isothiazolone	18.92	0.45
14-Hexadecena	19.90	0.15
2-methyl-Glutaric acid	21.15	0.06
Ethyl palmitate	21.40	0.31
Palmitoleic acid	21.65	8.72
Trichloroacetic acid, 3-tetradecyl Ester	21.76	0.36
Cyclohexene, 3-1,5-dimethyl-4-hex	23.04	0.57
4-Trifluoroacetoxytetradecane	24.15	0.17
Naphthalene	24.42	4.08
Hexatriacontane	24.68	0.75
$\alpha$ -Phellandrene	25.05	16.31
$\beta$ -Pinene	25.13	5.71
$\delta$ -Cadinene	26.05	0.07
Humulene epoxide II	26.64	0.49
Terpinen-4-ol	26.77	0.60
Squalene	26.95	8.52
Phytol	27.03	0.31
$\alpha$ -Panasinsene	27.55	6.77
$\alpha$ -Copaene	28.05	0.08
$\beta$ -Oplopenone	28.61	0.16
Benzyl benzoate	28.72	0.11
Total	-	88.56
Number of compounds	39	

## RESULT AND DISCUSSION

Result obtained in Table 1 reveals the presence of 39 bioactive compounds representing 88.56 % in *Schizomussaenda henryi* oil. The major bioactive compounds identified by GC-MS were;  $\alpha$ -Phellandrene (16.31 %), 4-Trifluoroacetoxytetradecane (11.92 %), 9,17-Octadecadienal (9.12 %), Palmitoleic acid (8.72 %), Squalene (8.52 %),  $\alpha$ -Panasinsene (6.77 %),  $\beta$ -Pinene (5.72 %),  $\beta$ -Phellandrene (5.11 %) and Naphthalene (4.08 %) while Benzene,1,4-dichloro (0.01 %), Hexatriacontane (0.03 %), 2-Ethylhexyl mercaptoacetate (0.07 %), Humulene epoxide II (0.07 %) amongst others were found in lower concentrations. 9,17-Octadecadienal, palmitoleic acid and squalene were also reported to be found in *Carica papaya* and *Piliostigma thonningii* essential oil (Daniel et al., 2023; Alagbe

et al., 2024). These compounds neutralize free radicals, support blood vessels integrity and support the heart, lungs and nervous system (Hernandez and Alagbe, 2025b; Shittu and Alagbe, 2020). Squalene are potent inhibitors of cancer cell growth which works by blocking the energy supplied inside malignant cell while sparing healthy tissue (Hernandez and Alagbe, 2025a; Singh et al., 2022).  $\beta$ -Pinene and  $\beta$ -Phellandrene have also been reported to be present in *Chenopodium ambrosioides*, *Albizia lebbek* and *Justicia secunda* oil (Hernandez and Alagbe, 2025b; Alagbe, 2025).  $\beta$ -Pinene and  $\beta$ -Phellandrene have been suggested to fuel beneficial gut bacteria producing short chain fatty acid that lowers inflammation (Alagbe 2024).  $\beta$ -Pinene also shields cells from free radicals, curbs inflammation and pains (Ojediran et al., 2024b; Alagbe et al., 2024). Phytol,  $\alpha$ -Humulene and  $\delta$ -Elemene prevents infections, accelerate healing of wounds and also support reproductive and metabolic health (Alagbe et al., 2025; Olaiya et al., 2016). They also protect liver and kidney tissue from oxidative stress, reduce damage from toxins and supporting detoxification (Alagbe, 2024; Velu et al., 2018).  $\beta$ -Famesene, Pentadecane, Nonane, 4-methyl and 3-Isothiazolone have been reported to protect the heart by lowering cholesterol and reducing the risk of neuro-degenerative diseases (Singh et al., 2020; Alhage et al., 2018). 14-Hexadecenal, 2-methyl-Glutaric acid and Ethyl palmitate acts as natural bronchodilator, helps to expel phlegm and reduce airway inflammation (Alagbe, 2023; Seroke et al., 2021). Terpinen-4-ol aids in supporting relaxation and reducing stress induced inflammation (Saha et al., 2021; Kujur et al., 2010). However, the result obtained in this study is in agreement with the reports of (). All the bioactive compounds in *Schizomussaenda henryi* oil work synergistically to calm the digestive tract and ease inflammation throughout the body (Alagbe, 2023). They are also pharmacological power house that block inflammatory pathways, delivers a broad spectrum antimicrobial effects and promote recovery after illness (Alagbe, 2024).

## CONCLUSIONS AND RECOMMENDATIONS

In conclusion, *Schizomussaenda henryi* oil shows promise as a natural antibiotic with neuro-protective potential and antioxidants shielding cells from oxidative stress, protecting liver cells and potentially defending liver tissues from toxins. They are generally regarded as safe (non-toxic), eco-friendly, effective and has no withdrawal period. *Schizomussaenda henryi* oil could also help to reduce the emergence of multi- drug resistance in microorganisms against existing antibiotics.

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