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# Antibacterial and GC-MS analysis of stem and leaf of *Premna paucinervis* (C.B. Clarke) gamble (*Lamiaceae*)- An endemic and rediscoverded species

**Steffy Francis[1*], V Anand Gideon[2], S John Britto[3]**

[1] Research Scholar, Department of Botany, Bishop Heber College, (Affiliated to Bharathidasan University), Tiruchirappalli, Tamil Nadu, India

[2] Associate Professor and Head, Department of Botany, Bishop Heber College, (Affiliated to Bharathidasan University), Tiruchirappalli, Tamil Nadu, India

[3] The Rapinat Herbarium and Centre for Molecular Systematics, St. Joseph's College, (Affiliated to Bharathidasan University), Tiruchirappalli, Tamil Nadu, India

**Abstract**
The present study aims to bring out the bioactive compounds of *Premna paucinervis* and evaluate the antibacterial activities of its crude extracts. The antibacterial activity was determined in the extracts using zone of inhibition method and were tested against *B. cereus, B. subtilis, E.coli, E.aerogenes, K.pneumoniae, P.mirabilis, P.vulgaris, P.aeruginosa, S.paratyphi, S.marcescens, S.aureus, S.pneumoniae* and *V.cholerae*. Zone of inhibition of the extracts were compared with that of control for antimicrobial activity. The results of the leaf extract showed notable inhibition of the bacterial growth against the tested organisms. The microbial activity of the *P. paucinervis* might be due to the presence of various secondary metabolites. Hence, this plant can be used to identify the specific bioactive compounds which may serve as leads in the development of new antimicrobial agents. Gas Chromatography- mass spectrometry (GC- MS) analysis of stem and leaf extracts showed presence of 45 and 65 compounds respectively. N-Hexadecanoic acid, Oleic Acid, Stigmasta-5, 20(22)-dien-3-ol and 9, 12-Octadecadienoic acid (Z, Z) - were the major bioactive components found in the stem extract. 2-Propenoic acid, 3-(4-hydroxy-3-methoxyphenyl)-, methyl ester, 2-Methoxy-4-vinylphenol, 6-Hydroxy-4, 4, 7a-trimethyl-5, 6, 7, 7a-tetrahydrobenzofuran-2(4H)-one and Benzofuran, 2, 3-dihydro- were the major components found in the leaf extract.

**Keywords:** antibacterial, GC-MS analysis, methanolic extract, *Premna paucinervis*

## Introduction

Plants are very useful sources of various bioactive compounds which have direct or indirect use in the treatment of various human ailments (Dogra, 2015) [4]. The knowledge of their healing properties has been transmitted over the centuries within and among human communities. Active compounds produced during secondary metabolism are usually responsible for the biological properties of plant species used throughout the globe for various purposes, including treatment of infectious diseases (Singh, 2015) [10]. The chemical features of these constituents differ considerably among different species. Gas Chromatography- mass spectrometry (GC-MS) method used for the analysis of plant extracts can be an interesting tool for testing the amount of active principles used in drugs, pharmaceutical or food industry (Gomathy, 2015) [6]. Products derived from plants may potentially control microbial growth in diverse situations and in the specific case of disease treatment. Numerous studies have aimed to describe the chemical composition of these plant antimicrobials and the mechanisms involved in microbial growth inhibition, either separately or associated with conventional antimicrobials (Singh, 2015) [10]. *Premna* species are well known for their medicinal properties and have been used in Indian traditional system of medicine especially for diarrhoea, stomach and hepatic disorders. The various biological activities including antioxidant, antibacterial, anti-inflammatory, cytotoxic and hepatoprotective have been displayed both at extract and pure compound level (Rekha, 2015) [8]. During the taxonomic revision of Indian Verbenaceae, Rajendran and Daniel (2002) recognized 31 species and 6 varieties of *Premna.* Recently, Prabhu Kumar *et al*. (2013) [7] reported the discovery of a new species *Premna rajendranii* from Chinnar and Madukkarai hill ranges of Western Ghats in Kerala and Tamilnadu. Apart from this, a research team comprising Robi, Augustin, Sasidharan and Udayan (2013) [9] rediscovered an endemic and rare species of *Premna viz*., *Premna paucinervis* (C. B. Clarke) Gamble from the Vagamon hills along the South Western Ghats of Kerala after a lapse of 140 years of its original type collection by R.H. Beddome (1872) from Anamalais, Western Ghats (Tamilnadu) (Kumar, 2013; Roby, 2013; Bose, 2014; Francis, 2019) [7, 3, 5]. The genus *Premna*, earlier included in the family Verbenaceae, was recently transferred to the Lamiaceae family based on molecular data (A.P.G.IV. 2009) [1].

## Materials and Methods

### Collection and Authentication

The plant material was collected from Nelliyampathy forest, Palaghat, Kerala and authenticated by Dr. S. John Britto S.J., at the Rapinat Herbarium and Centre for Molecular Systematics, St. Joseph's College (Autonomous), Tiruchirappalli. The voucher specimen (RHT 68492) was deposited for future references.

**Extraction**

The stems and leaves were shade dried, powdered using mechanical grinder and stored in air tight containers. The stem and leaf samples were immersed in different solvents (Acetone, Ethanol, Methanol and Aqueous) for extraction and kept in rotary shaker for 72 hours at room temperature. The extracts were filtered using muslin cloth and then used for antimicrobial and GC-MS analysis.

**Anti-bacterial Activity**

**Test Micro-Organisms**

13 Bacterial strains were used in the study namely *Staphylococcus aureus* (MTCC # 3163), *Escherichia coli* (MTCC# 199), *Klebsiella pneumoniae* (MTCC # 3040), *Pseudomonas aeruginosa* (MTCC # 2474), *Salmonella paratyphi* (MTCC # 734), *Vibrio cholera* (ATCC # 14104), *Enterobacter aerogenes* (MTCC # 2990), *Streptococcus pneumonia*e (ATCC # 7066), *Bacillus subtilis* (MTCC # 441), *Bacillus cereus* (ATCC # 4342), *Proteus vulgaris* (MTCC # 1771), *Proteus mirabilis* (MTCC # 1429) and *Serratia marcescens* (MTCC # 2645). These pathogenic micro-organisms were obtained from Rapinat Herbarium and Centre for Molecular Systematics, St. Joseph's College Tiruchirappalli, Tamil Nadu. All the test bacterial strains were maintained on nutrient agar media at 4 °C.

**Preparation of Disc**

6 mm discs were prepared and sterilized in autoclave and were soaked in the different extracts - Acetone, Distilled water, Ethanol and Methanol. The standard drug streptomycin was used as control.

**Determination of Antibacterial Activity**

Antibacterial activities of *P. paucinervis* extracts were determined by disc diffusion method (Bauer, 1996) [2]. Nutrient agar was prepared for the study. Each plate of nutrient agar was swabbed with different bacterial strain using sterile cotton swab. The soaked dried discs were placed on the surface of each inoculated plate. The plates were allowed for diffusion for half an hour and then transferred to incubator at 37˚C for 24 hours. Standard disc of Streptomycin was also placed as positive control. The antibacterial activity of *P. paucinervis* stem and leaf extracts was determined by measuring the diameter of zone of inhibition in mm.

**GC-MS Analysis**

The GC-MS analysis of the extractions was performed using a Hewlett Packard 5890 II GC equipped with a FID detector and HP-5 ms capillary column (30m´ 0.25m, film thickness 0.25µm). For GC-MS detection, an electron ionization system was used with ionization energy of 70 eV. Helium was used as a carrier gas at a constant flow rate of 1ml/min. Injector and MS transfer line temperature were set at 220 and 290°C respectively. Column temperature was initially at 50°C, and then gradually increased to 150°C at 3°C/min rate, held for 10 min and finally increased to 250Vc at 10Vc/min. Diluted methanol sample of 1.0µl was injected manually in the split less mode. The components were identified based on the comparison of their relative retention time and mass spectra with those of Wiley 7N Library data and standards of the main components.

**Results and Discussion**

The leaf extracts showed maximum antibacterial activity against 12 bacterial strains (Table 3, fig.4, 5). Stem extracts showed lower antibacterial activity compared to leaf extracts (Table 2, fig.3). Moreover, ethanol and methanol extracts showed better activity compared to the acetone extract. Aqueous extract of *P. paucinervis* showed no activity. Thus, it can be concluded that the leaves of *P. paucinervis* have significant antimicrobial activity.

**Table 1:** Antibacterial activity of *Premna paucinervis* stem

| Bacterial strain | Acetone | Ethanol | Methanol | Aqueous | Antibiotic |
|---|---|---|---|---|---|
| *Proteus mirabilis* | 0 | 9.3±1.15 | 8.3±0.58 | 0 | 24.7±0.06 |
| *Proteus vulgaris* | 7.7±0.58 | 8.7±1.15 | 7.7±0.58 | 0 | 22±1 |
| *Pseudomonas aeruginosa* | 8.7±0.58 | 8.7±1.15 | 8±1 | 0 | 14±1.73 |
| *Salmonella paratyphi* | 8±1 | 7.3±0.58 | 8±0 | 0 | 15.3±0.58 |
| *Serratia marcescens* | 0 | 10±1 | 10±1.73 | 0 | 15.7±0.33 |
| *Staphylococcus aureus* | 0 | 8.7±1.15 | 8±1 | 0 | 19.7±1.53 |
| *Vibrio cholerae* | 15.7±0.58 | 11±2 | 9±1 | 0 | 19.7±2.52 |

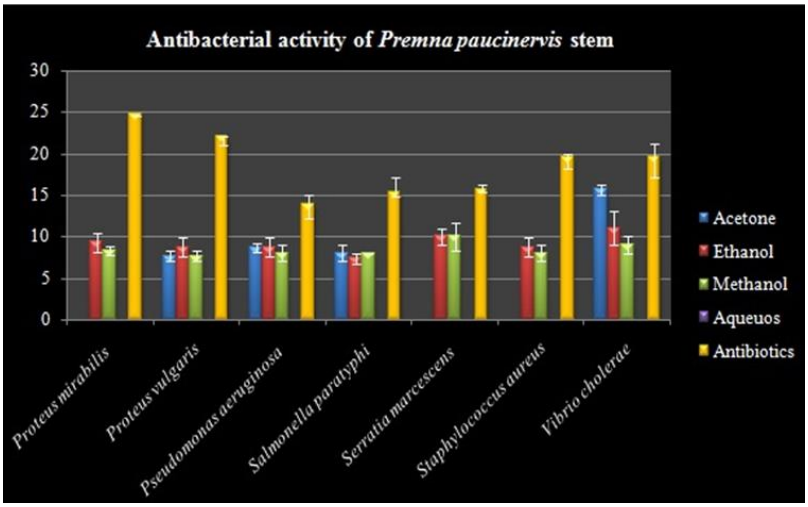



**Fig 1:** Antibacterial activity of *Premna paucinervis* stem

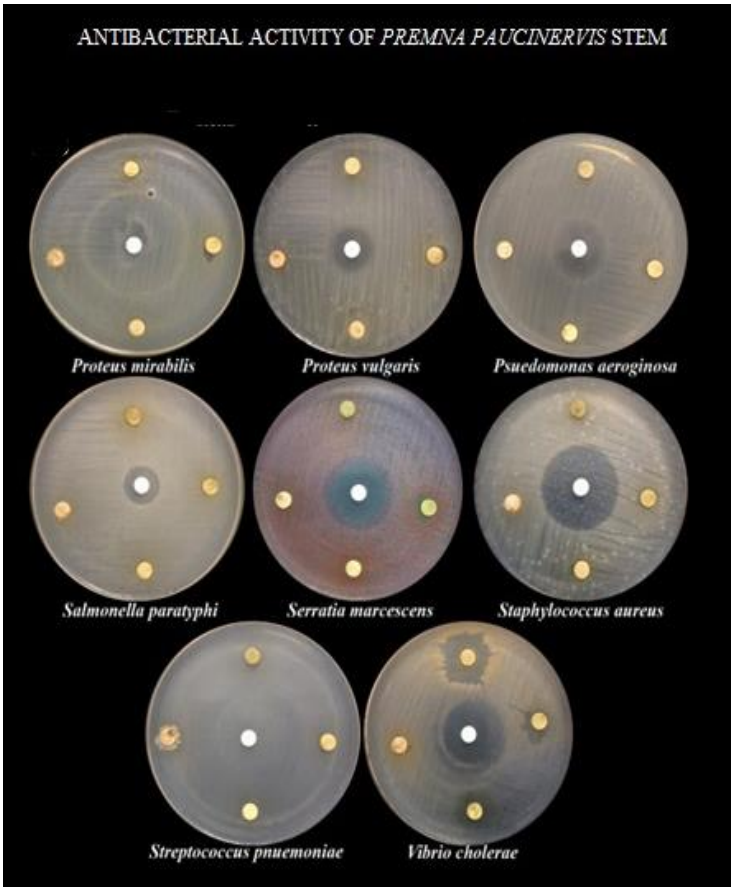



**Fig 2:** Antibacterial activity of *Premna paucinervis* stem

**Table 2:** Antibacterial activity of *Premna paucinervis* leaf

| Bacterial strains | Acetone | Ethanol | Methanol | Aqueous | Antibiotic |
|---|---|---|---|---|---|
| *Bacillus cereus* | 13.7±1.5 | 11.3±0.58 | 10±1 | 0 | 24.7±0.58 |
| *Bacillus subtilis* | 13±1 | 15.3±0.06 | 16±0.1 | 0 | 24.7±3.1 |
| *Enterobacter aerogenes* | 16±1 | 8.7±1.15 | 9.7±0.58 | 0 | 26.7±1.53 |
| *Escherichia coli* | 11.7±0.58 | 17.3±1.53 | 8.7±1.15 | 0 | 24.7±1.53 |
| *Klebsiella pneumoniae* | 8±0.58 | 17.3±2.52 | 13±1 | 0 | 26±1.73 |
| *Proteus mirabilis* | 8±0.58 | 13±1 | 13.7±1.53 | 0 | 26±2 |
| *Proteus vulgaris* | 13.3±1.53 | 14±1.73 | 7.3±0.58 | 0 | 24.7±0.58 |
| *Pseudomonas aeruginosa* | 8±1.15 | 15.3±1.15 | 8.7±0.58 | 0 | 18.3±1.53 |
| *Salmonella paratyphi* | 8.3±1.53 | 8.3±0.58 | 12±1.53 | 0 | 26.3±1.53 |
| *Serratia marcescens* | 11±1 | 13.3±1.53 | 8.7±1.53 | 0 | 28.7±0.58 |
| *Staphylococcus aureus* | 0 | 13.7±2.1 | 9.7±0.58 | 0 | 26.7±1.15 |
| *Vibrio cholerae* | 17.3±1.53 | 11.3±1.15 | 15.7±2.1 | 0 | 25.3±0.58 |

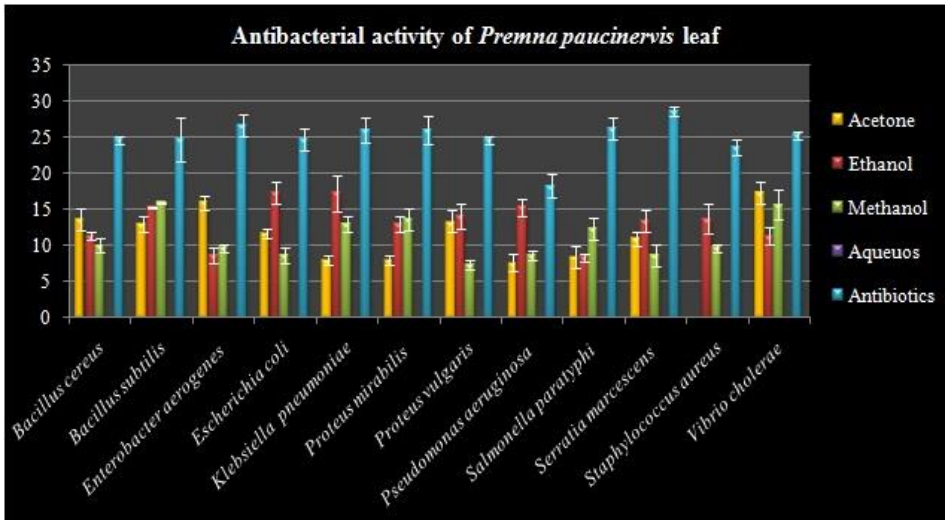



**Fig 3:** Antibacterial activity of *Premna paucinervis* leaf

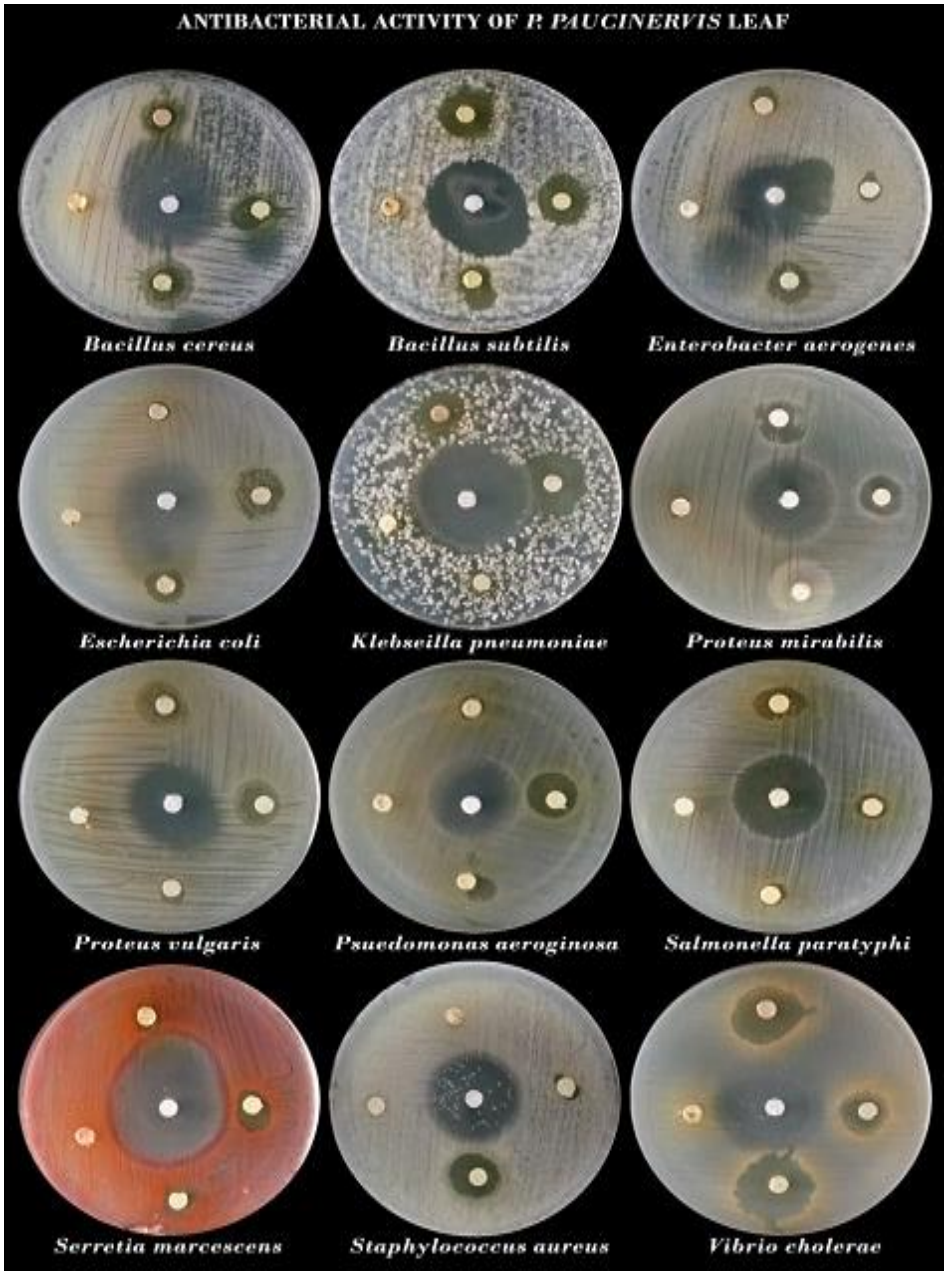



**Fig 4:** Antibacterial activity of *Premna paucinervis* leaf

The GC-MS analysis of stem and leaf of *Premna paucinervis* showed the presence of various phytochemicals which possess bioactivity. Methanol stem extract contained about 45 phytochemical compounds among which n-hexadecanoic acid (19.98%), oleic Acid (9.75%), stigmasta-5, 20(22)-dien-3-ol (5.79%) and 9, 12-octadecadienoic acid (Z, Z)- (5.67%) were the major bioactive components. Besides caryophyllene oxide, aromadendrene oxide-(2), tetradecanoic acid, spiro [4, 5] decan7-one, 1,8-dimethyl-8,9-epoxy-4-isopropyl-, 2,6,10-trimethyl,14-ethylene-14-pentadecne, 2-pentadecanone, 6,10,14-trimethyl-, hexadecanoic acid, methyl ester, 9,12,15-octadecatrienoic acid, methyl ester, 2-hexadecen-1-ol, 3,7,11,15-tetramethyl-, [r-[r*,r*-(e)]-, octadecanoic acid, nerolidyl acetate and ergost-5-en-3-ol, (3.beta.,24r)- were also present in the stem extract which are used as flavour ingredient, diuretic, lowering cholesterol, antioxidant, anti-inflammatory, ant arthritic, anticancer, antiviral and antimicrobial agent. (Table 3) (Fig 5) Methanol leaf extract of *P. paucinervis* showed presence of 65 phytochemical compounds among which 2-propenoic acid, 3-(4-hydroxy-3-methoxyphenyl)-, methyl ester (8.06%), 2-methoxy-4-vinylphenol (6.21%), 6-hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one (4.34%) and benzofuran, 2,3-dihydro- (3.32%) were the major bioactive components. Benzofuran-2-carboxaldehyde, benzoic acid, 4-hydroxy-3-methoxy-, methyl ester, 2(4H)-benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-, 3-hydroxy-.beta.-damascone, megastigmatrienone, alpha.-cadinol, tetradecanoic acid, n-hexadecanoic acid, sinapic acid methyl ester, 2-hexadecen-1-ol, 3,7,11,15-tetramethyl-, [r-[r*,r*-(e)]-, dichloroacetic acid, tridec-2-ynyl ester, octadecanoic acid, benzyl. beta.-d-glucoside, hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester and 1,4-butanediol 2,3-divanillyl- were also present in the leaf extract which are used as flavour ingredient, diuretic, hepatoprotective, antioxidant, anti-inflammatory, antiarthritic, anticancer, anthelmintic, anti-myocardial ischemia, antiviral and antimicrobial agent. (Table 4) (Fig 6)

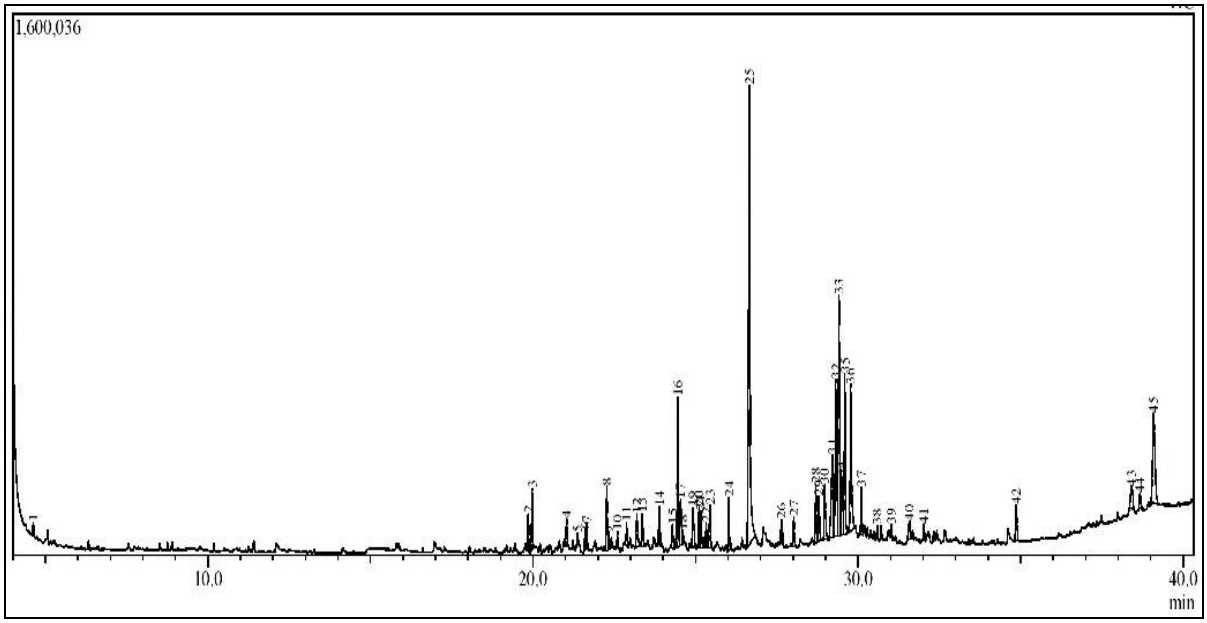



**Fig 5:** GC-MS analysis of *Premna paucinervis* stem extract

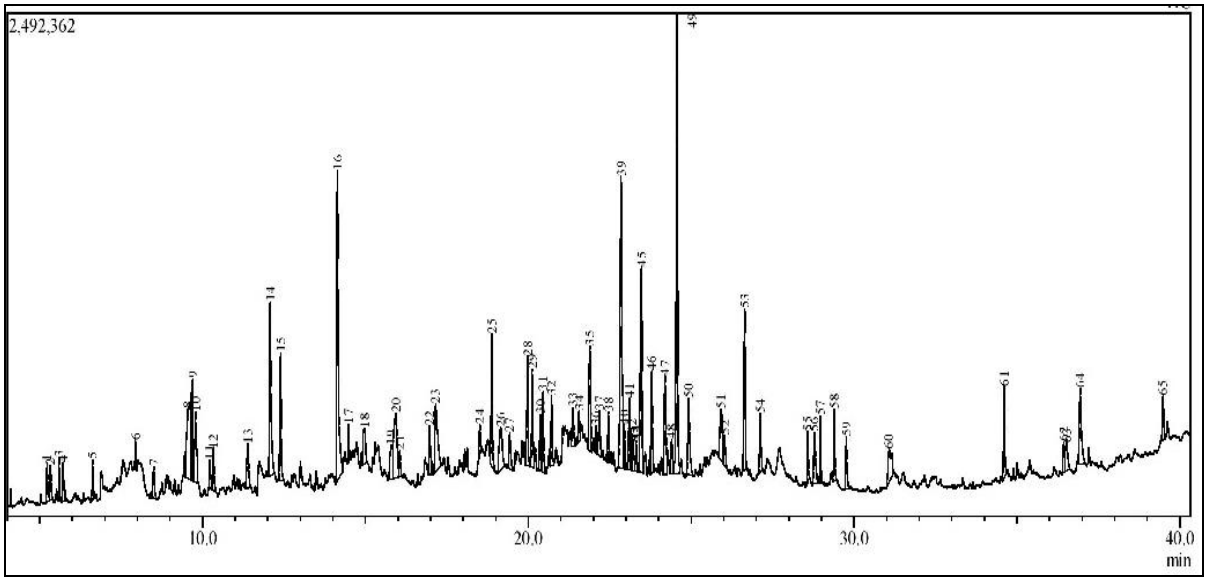
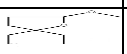
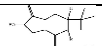
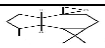
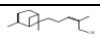
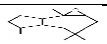
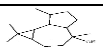
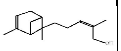
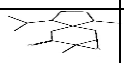
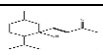
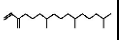
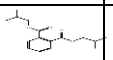
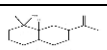
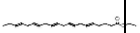
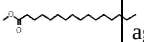
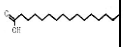
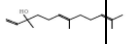
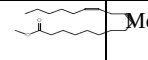
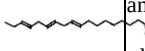
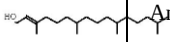
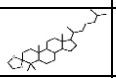
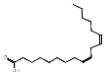
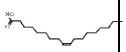
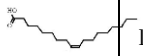
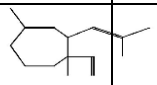
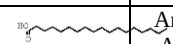
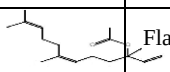
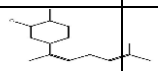
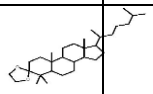
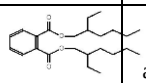
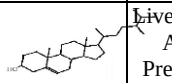
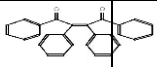
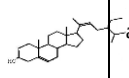



**Fig 6:** GC-MS analysis of *Premna paucinervis* leaf extract

**Table 3:** GC-MS analysis of *Premna paucinervis* stem extract

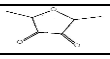
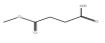
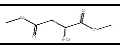
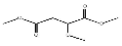
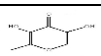
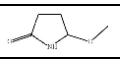
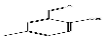
| S. No. | Name | MF | MW (g/mol) | RT | Area % | Structure | Activity |
|---|---|---|---|---|---|---|---|
| 1 | 2-Methoxy-1-oxa-spiro[4.4]nonane | $C_9H_{16}O_2$ | 156 | 4.67 | 0.29 |  | - |
| 2 | (-)-Spathulenol | $C_{15}H_{24}O$ | 220 | 19.905 | 1.05 |  | Volatile oil component, plant metabolite, anaesthetic and vasodilator agent. |
| 3 | Caryophyllene oxide | $C_{15}H_{24}O$ | 220 | 20.03 | 1.71 |  | Antifungal |
| 4 | Caryophylla-4(12),8(13)-dien-5.alpha.-ol | $C_{15}H_{24}O$ | 220 | 21.08 | 0.69 |  | - |
| 5 | (1aR,3aS,7S,7aS,7bR)-1,1,3a,7-Tetramethyldecahydro-1H-cyclopropa[a]naphthalen-7-ol | $C_{15}H_{26}O$ | 222 | 21.405 | 0.35 |  | - |
| 6 | Aromadendrene oxide-(2) | $C_{15}H_{24}O$ | 220 | 21.63 | 0.64 |  | Antimicrobial, antiviral |

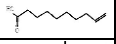
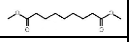
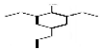
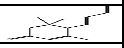
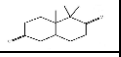
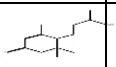
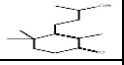
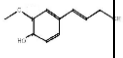
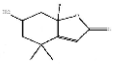
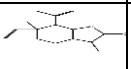
| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 7 | (-)-5 Oxatricyclo[8.2.0.0(4,6)]dodecane,,12-trimethyl-9-methylene-, [1r-(1r*,4r*,6r*,10s*)]- | $C_{15}H_{24}O$ | 220 | 21.7 | 0.69 |  | Flavoring agent |
| 8 | Tricyclo[7.3.0.0(3,8)]dodec-1-en-12-one, (Z)-3,8-cisoid-8,9-2-cyano-9-.beta.-methyl- | $C_{14}H_{17}NO$ | 215 | 22.31 | 1.94 |  | - |
| 9 | Bergamotol, Z-.alpha.-trans- | $C_{15}H_{24}O$ | 220 | 22.36 | 0.46 |  | Perfumes and odorants |
| 10 | 1,3B,6,6-tetramethyldecahydro-1h-cyclopropa[7,8]azuleno[4,5-b]oxirene | $C_{15}H_{24}O$ | 220 | 22.65 | 0.43 |  | - |
| 11 | 1,1,4,7-Tetramethyldecahydro-1h-cyclopropa[e]azulen-4-ol | $C_{15}H_{26}O$ | 222 | 22.925 | 0.53 |  | Biomarker |
| 12 | Tetradecanoic acid | $C_{14}H_{28}O_2$ | 228 | 23.275 | 1.34 |  | Antifungal, Antioxidant, cancer preventive, nematicide, hypercholesterolemic, Lubricant |
| 13 | Bergamotol, Z-.alpha.-trans- | $C_{15}H_{24}O$ | 220 | 23.43 | 1.16 |  | Perfumes and odorants |
| 14 | Spiro[4,5]decan7-one, 1,8-dimethyl-8,9-epoxy-4-isopropyl- | $C_{15}H_{24}O_2$ | 236 | 23.945 | 1.48 |  | Anti-inflammatory |
| 15 | 4-(1-Hydroxy-2-isopropyl-5-methylcyclohexyl)-3-butyn-2-one | $C_{14}H_{22}O_2$ | 222 | 24.355 | 0.92 |  | - |
| 16 | (3aR,4R,7R)-1,4,9,9-Tetramethyl-3,4,5,6,7,8-hexahydro-2H-3a,7-methanoazulen-2-one | $C_{15}H_{22}O$ | 218 | 24.505 | 4.49 |  | - |
| 17 | 2,6,10-Trimethyl,14-ethylene-14-pentadecne | $C_{20}H_{38}$ | 278 | 24.6 | 1.75 |  | Anti-inflammatory, antimicrobial, antiproliferative, plant metabolite and algal metabolite |
| 18 | 2-Pentadecanone, 6,10,14-trimethyl- | $C_{18}H_{36}O$ | 268 | 24.655 | 0.3 |  | Flavoring agent |
| 19 | 1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester | $C_{16}H_{22}O_4$ | 278 | 25.005 | 1.63 |  | - |
| 20 | Neointermedeol | $C_{15}H_{26}O$ | 222 | 25.14 | 1.42 |  | Flavoring agent |
| 21 | Ethyl 5,8,11,14,17-icosapentaenoate | $C_{22}H_{34}O_2$ | 330 | 25.23 | 1.14 |  | - |
| 22 | Thunbergol | $C_{20}H_{34}O$ | 290 | 25.385 | 0.67 |  | - |
| 23 | Thunbergol | $C_{20}H_{34}O$ | 290 | 25.51 | 1.42 |  | - |
| 24 | Hexadecanoic acid, methyl ester | $C_{17}H_{34}O_2$ | 270 | 26.075 | 1.45 |  | Anti-inflammatory agent, decreases blood cholesterol |
| 25 | n-Hexadecanoic acid | $C_{16}H_{32}O_2$ | 256 | 26.835 | 19.98 |  | Anti-inflammatory agent, antioxidant, hypocholesterolemic nematicide, pesticide, anti-androgenic flavor, hemolytic,5-alpha reductase inhibitor, potent mosquito larvicide |
| 26 | 1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl- | $C_{15}H_{26}O$ | 222 | 27.725 | 0.97 |  | - |
| 27 | 1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-, [s-(z)]- | $C_{15}H_{26}O$ | 222 | 28.075 | 0.77 |  | - |
| 28 | 9,12-Octadecadienoic acid (Z,Z)-, methyl ester | $C_{19}H_{34}O_2$ | 294 | 28.75 | 1.62 |  | Moisturiser for skin, nails, and hair |
| 29 | 9,12,15-Octadecatrienoic acid, methyl ester | $C_{19}H_{32}O_2$ | 292 | 28.855 | 1.55 |  | Antibacterial, anticancer, anticandidal, antiinflammatory, hypocholesterolemic, hepatoprotective, |

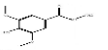
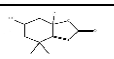
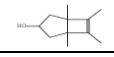
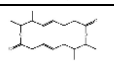
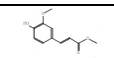
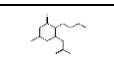
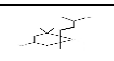
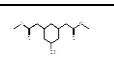
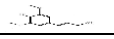
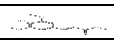
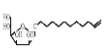
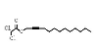
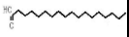
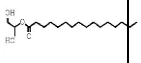
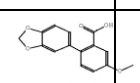
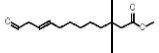
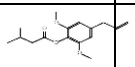
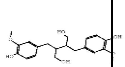
| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | nematicide, insectifuge antihistaminic, antiarthritic, anticoronary, antieczemic antiacne, 5-Alpha reductase inhibitor and antiandrogenic |
| 30 | 2-Hexadecen-1-ol, 3,7,11,15-tetramethyl-, [r-[r*,r*-(e)]]- | $C_{20}H_{40}O$ | 296 | 29.045 | 2.12 |  | Antimicrobial, anti-inflammatory, anticancer, diuretic |
| 31 | Cholestan-3-one, 4,4-dimethyl-, cyclic 1,2-ethanediyl acetal, (5.alpha.)- | $C_{31}H_{54}O_2$ | 458 | 29.265 | 4.24 |  | - |
| 32 | 9,12-Octadecadienoic acid (Z,Z)- | $C_{18}H_{32}O_2$ | 280 | 29.365 | 5.67 |  | Anticancer agent, essential for the synthesis of various hormones like prostaglandins, thromboxanes, and leukotrienes |
| 33 | Oleic Acid | $C_{18}H_{34}O_2$ | 282 | 29.47 | 9.75 |  | An excipient in pharmaceuticals, lowering cholesterol and reducing inflammation |
| 34 | Oleic Acid | $C_{18}H_{34}O_2$ | 282 | 29.555 | 2.56 |  | An excipient in pharmaceuticals, lowering cholesterol and reducing inflammation |
| 35 | 1-Cycloheptene, 1,4-dimethyl-3-(2-methyl-1-propene-1-yl)-4-vinyl- | $C_{15}H_{24}$ | 204 | 29.68 | 4.83 |  | - |
| 36 | Octadecanoic acid | $C_{18}H_{36}O_2$ | 284 | 29.895 | 5.65 |  | Anti-inflammatory, Antimicrobial and antiarthritic |
| 37 | Nerolidyl acetate | $C_{17}H_{28}O_2$ | 264 | 30.16 | 1.45 |  | Flavor and fragrance agent |
| 38 | cis-Z-.alpha.-Bisabolene epoxide | $C_{15}H_{24}O$ | 220 | 30.635 | 0.39 |  | - |
| 39 | 2H-Pyran, 2-(2-heptadecynyloxy)tetrahydro- | $C_{22}H_{40}O_2$ | 336 | 31.07 | 0.44 |  | - |
| 40 | Cholestan-3-one, 4,4-dimethyl-, cyclic 1,2-ethanediyl acetal, (5.alpha.)- | $C_{31}H_{54}O_2$ | 458 | 31.63 | 0.71 |  | - |
| 41 | 1-Heptatriacotanol | $C_{37}H_{76}O$ | 536 | 32.075 | 0.42 |  | Enzyme inhibitor, anti-hypercholesterolemic effects |
| 42 | Bis(2-ethylhexyl) phthalate | $C_{24}H_{38}O_4$ | 390 | 34.935 | 1.36 |  | Apoptosis inhibitor and medical devices |
| 43 | Ergost-5-en-3-ol, (3.beta.,24r)- | $C_{28}H_{48}O$ | 400 | 38.49 | 1.31 |  | Liver disease, jaundice, Artherosclerosis, Precursor of anabolic steroid boldenone |
| 44 | 2-Butene-1,4-dione, 1,2,3,4-tetraphenyl-, (Z)- | $C_{28}H_{20}O_2$ | 388 | 38.735 | 0.44 |  | - |
| 45 | Stigmasta-5,20(22)-dien-3-ol | $C_{29}H_{48}O$ | 412 | 39.235 | 5.79 |  | Antioxidant, antibacterial activity, anti-inflammatory, antiarthritic antiasthma, diuretic |

MF: Molecular Formula, MW: Molecular Weight, RT: Retention Time

**Table 4:** GC-MS in leaves of *Premna paucinervis*

| S. No. | Name | MF | MW (g/mol) | RT | Area % | Structure | Activity |
|---|---|---|---|---|---|---|---|
| 1 | 5-Methyl-isoxazolidin-3-one | $C_4H_7NO_2$ | 101 | 5.3 | 0.61 |  | - |
| 2 | 2(5H)-Furanone | $C_4H_4O_2$ | 84 | 5.395 | 0.55 |  | Appetite depressant, immunosuppressive agent |
| 3 | 1,2-Cyclopentanedione | $C_5H_6O_2$ | 98 | 5.66 | 0.49 |  | Synthesis of chemical probes for selective labeling of sulfenic acid proteins |
| 4 | 3-Thietanol | $C_3H_6OS$ | 90 | 5.775 | 0.61 |  | Harmful if it comes in contct with the skin, if inhaled and swallowed |
| 5 | Pentanoic acid, 4-methyl-, methyl ester | $C_7H_{14}O_2$ | 130 | 6.695 | 0.56 |  | Insecticide, flavoring agent, |
| 6 | 3-Hydroxy-4,4-dimethyldihydro-2(3h)-furanone | $C_6H_{10}O_3$ | 130 | 7.985 | 0.33 |  | Used to make perfumes |
| 7 | 2,5-Dimethylfuran-3,4(2H,5H)-dione | $C_6H_8O_3$ | 128 | 8.58 | 0.62 |  | - |
| 8 | 4-Methyl-n-(2-oxo-4-vinyl-tetrahydro-furan-3-yl)-benzenesulfonamide | $C_{13}H_{15}NO_4S$ | 281 | 9.65 | 3.78 |  | - |
| 9 | Butanedioic acid, monomethyl ester | $C_5H_8O_4$ | 132 | 9.73 | 2.36 |  | Beverage industry, primarily as an acidity regulator |
| 10 | Butanedioic acid, hydroxy-, dimethyl ester | $C_6H_{10}O_5$ | 162 | 9.86 | 1.68 |  | - |
| 11 | Butanedioic acid, methoxy-, dimethyl ester | $C_7H_{12}O_5$ | 176 | 10.29 | 0.45 |  | Flavoring agent, Pigments, solvents, Viscosity adjustors |
| 12 | 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- | $C_6H_8O_4$ | 144 | 10.415 | 0.69 |  | Skin irritation, eye irritation |
| 13 | 5-Methoxypyrrolidin-2-one | $C_5H_9NO_2$ | 115 | 11.47 | 1.06 |  | Antifungal agent |
| 14 | Benzofuran, 2,3-dihydro- | $C_8H_8O$ | 120 | 12.21 | 3.32 |  | Antiviral, antibacterial, anti-inflammatory, antiangiogenic and antimitotic activities |
| 15 | 1H-Pyrrole-2,5-dione, 3-ethyl-4-methyl- | $C_7H_9NO_2$ | 139 | 12.5 | 2.06 |  | - |
| 16 | 2-Methoxy-4-vinylphenol | $C_9H_{10}O_2$ | 150 | 14.31 | 6.21 |  | Antimicrobial, antioxidant, anti-inflammatory, analgesic, anti-germination |
| 17 | cis-2,3-Epoxyoctane | $C_8H_{16}O$ | 128 | 14.535 | 0.51 |  | - |
| 18 | 2-Cyclopenten-1-one, 3-methyl- | $C_6H_8O$ | 96 | 15.05 | 1.09 |  | - |
| 19 | 5-Oxo-pyrrolidine-2-carboxylic acid methyl ester | $C_6H_9NO_3$ | 143 | 15.845 | 1.18 |  | - |
| 20 | 2-Furanmethanol, 5-ethenyltetrahydro-.alpha.,.alpha.,5-trimethyl-, cis- | $C_{10}H_{18}O_2$ | 170 | 16.025 | 2.81 |  | - |
| 21 | Benzaldehyde, 4-hydroxy-3-methoxy- | $C_8H_8O_3$ | 152 | 16.125 | 0.43 |  | - |
| 22 | Benzofuran-2-carboxaldehyde | $C_9H_6O_2$ | 146 | 17.035 | 1.02 |  | Synthesis of isoxazolines, antifungal agent |
| 23 | 2-Hydroxy-5-methylbenzaldehyde | $C_8H_8O_2$ | 136 | 17.325 | 2.41 |  | Radiopharmaceuticals for diagnostic imaging |
| 24 | Benzoic acid, 4-hydroxy-3-methoxy-, methyl ester | $C_9H_{10}O_4$ | 182 | 18.555 | 0.43 |  | Anticancer |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 25 | 2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)- | $C_{11}H_{16}O_2$ | 180 | 19.005 | 2.07 |  | Antimicrobial activity, Flavor and fragrance agents |
| 26 | 9 Decenoic acid | $C_{10}H_{18}O_2$ | 170 | 19.27 | 1.63 |  | - |
| 27 | 2,5-Dimethoxy-4-ethylamphetamine | $C_{13}H_{21}NO_2$ | 223 | 19.52 | 0.7 |  | Psychotropic agent |
| 28 | Nonanedioic acid, dimethyl ester | $C_{11}H_{20}O_4$ | 216 | 20.06 | 1.84 |  | - |
| 29 | Phenol, 2,6-dimethoxy-4-(2-propenyl)- | $C_{11}H_{14}O_3$ | 194 | 20.18 | 1.61 |  | Sweet and spicy tasting compound. |
| 30 | 4,6,7-Trimethyl-2h-azepine-2,5(6h)-dione | $C_9H_{11}NO_2$ | 165 | 20.415 | 1.06 |  | Used to produce detergents and biodiesel |
| 31 | 3-Hydroxy-.beta.-damascone | $C_{13}H_{20}O_2$ | 208 | 20.545 | 1.23 |  | 17-beta-hydroxysteroid dehydrogenase inhibitor, Aryl-Hydrocarbon Hydroxylase inhibitor, Testosterone Hydroxylase Inducer, Beta 2 Receptor agonist, Beta adrenergic Receptor Blocker, Beta adrenergic agent, Beta Glucuronidase Inhibitor, Anti-inflammatory, Cancer protective efficacy |
| 32 | Megastigmatrienone | $C_{13}H_{18}O$ | 190 | 20.785 | 1.1 |  | Antioxidant |
| 33 | .alpha.-Cadinol | $C_{15}H_{26}O$ | 222 | 21.45 | 0.76 |  | Anti-fungal, hepatoprotective, remedy for drug resistant tuberculosis. |
| 34 | 7-Oxabicyclo[4.1.0]heptan-3-ol, 6-(3-hydroxy-1-butenyl)-1,5,5-trimethyl- | $C_{13}H_{22}O_3$ | 226 | 21.6 | 0.64 |  | - |
| 35 | 2,6-Naphthalenedione, octahydro-1,1,8a-trimethyl-, cis- | $C_{13}H_{20}O_2$ | 208 | 21.955 | 2.26 |  | - |
| 36 | (E)-2,6-Dimethoxy-4-(prop-1-en-1-yl)phenol | $C_{11}H_{14}O_3$ | 194 | 22.12 | 0.45 |  | Flavoring agent |
| 37 | 2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl- | $C_{13}H_{22}O_2$ | 210 | 22.25 | 0.93 |  | - |
| 38 | 2-Cyclohexen-1-one, 3-(3-hydroxybutyl)-2,4,4-trimethyl- | $C_{13}H_{22}O_2$ | 210 | 22.505 | 0.72 |  | - |
| 39 | (E)-4-(3-Hydroxyprop-1-en-1-yl)-2-methoxyphenol | $C_{10}H_{12}O_3$ | 180 | 22.935 | 7.34 |  | - |
| 40 | 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one | $C_{11}H_{16}O_3$ | 196 | 23 | 0.58 |  | Anti-inflammatory |
| 41 | 2-Propenoic acid, 3-(4-hydroxyphenyl)-, methyl ester | $C_{10}H_{10}O_3$ | 178 | 23.145 | 1.48 |  | Potential biomarker |
| 42 | 2(3H)-Benzofuranone, 6-ethenylhexahydro-3,6-dimethyl-7-(1-methylethenyl)-, [3S-(3.alpha.,3a.alpha.,6.alpha.,7.beta.,7a.beta.)]- | $C_{15}H_{22}O_2$ | 234 | 23.195 | 0.63 |  | - |
| 43 | Tetradecanoic acid | $C_{14}H_{28}O_2$ | 228 | 23.275 | 0.58 |  | Antitumer, anticancer agents, larvicidal and repellent activity, surfactant, opacifying agent, |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | texture enhancer, emollient, cleansing agent and emulsifier |
| 44 | Benzoic acid, 4-hydroxy-3,5-dimethoxy-, hydrazide | $C_9H_{12}N_2O_4$ | 212 | 23.365 | 0.48 |  | - |
| 45 | 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one | $C_{11}H_{16}O_3$ | 196 | 23.55 | 4.34 |  | Anti-inflammatory |
| 46 | 2,3-Bis(1-methylallyl)pyrrolidine | $C_{12}H_{21}N$ | 179 | 23.87 | 1.85 |  | - |
| 47 | Endo-1,5,6,7-Tetramethylbicyclo[3.2.0]hept-6-en-3-ol | $C_{11}H_{18}O$ | 166 | 24.265 | 2.14 |  | - |
| 48 | 1,9-Dioxacyclohexadeca-4,13-diene-2-10-dione, 7,8,15,16-tetramethyl- | $C_{18}H_{28}O_4$ | 308 | 24.45 | 0.7 |  | Antibacterial |
| 49 | 2-Propenoic acid, 3-(4-hydroxy-3-methoxyphenyl)-, methyl ester | $C_{11}H_{12}O_4$ | 208 | 24.66 | 8.06 |  | Antibacterial |
| 50 | 2-Butyl-5-methyl-3-(2-methylprop-2-enyl)cyclohexanone | $C_{15}H_{26}O$ | 222 | 25.055 | 1.88 |  | - |
| 51 | 7-Oxabicyclo[4.1.0]heptan-3-ol, 6-(3-hydroxy-1-butenyl)-1,5,5-trimethyl- | $C_{13}H_{22}O_3$ | 226 | 25.99 | 1.61 |  | - |
| 52 | (3-Hydroxy-5-methoxycarbonylmethyl-cyclohexyl)-acetic acid, methyl ester | $C_{12}H_{20}O_5$ | 244 | 26.1 | 0.67 |  | - |
| 53 | n-Hexadecanoic acid | $C_{16}H_{32}O_2$ | 256 | 26.71 | 2.64 |  | Anti-inflammatory agent, antioxidant, hypocholesterolemic nematicide, pesticide, anti androgenic flavor, hemolytic,5-alpha reductase inhibitor, potent mosquito larvicide |
| 54 | trans-Sinapyl alcohol | $C_{11}H_{14}O_4$ | 210 | 27.2 | 1.12 |  | Plant metabolite |
| 55 | Sinapic acid methyl ester | $C_{12}H_{14}O_5$ | 238 | 28.635 | 0.75 |  | Antioxidant |
| 56 | 10-Undecenyl hexofuranoside | $C_{17}H_{32}O_6$ | 332 | 28.895 | 1.19 |  | - |
| 57 | 2-Hexadecen-1-ol, 3,7,11,15-tetramethyl-, [r-[r*,r*-(e)]]- | $C_{20}H_{40}O$ | 296 | 29.02 | 0.87 |  | Antimicrobial, anti-inflammatory, anticancer, diuretic |
| 58 | Dichloroacetic acid, tridec-2-ynyl ester | $C_{15}H_{24}Cl_2O_2$ | 306 | 29.455 | 1.06 |  | Antimicrobial, antioxidant |
| 59 | Octadecanoic acid | $C_{18}H_{36}O_2$ | 284 | 29.84 | 0.83 |  | Anti-inflammatory, Antimicrobial and antiarthritic |
| 60 | Benzyl. beta.-d-glucoside | $C_{13}H_{18}O_6$ | 270 | 31.215 | 1.39 |  | Antioxidant, plant metabolite |
| 61 | Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester | $C_{19}H_{38}O_4$ | 330 | 34.685 | 1.36 |  | Anti-inflammatory, antioxidant and anthelmintic |
| 62 | 4-Methoxy-4',5'-methylenedioxybiphenyl-2-carboxylic acid | $C_{15}H_{12}O_5$ | 272 | 36.47 | 0.55 |  | - |
| 63 | Methyl 12-oxo-9-dodecenoate | $C_{13}H_{22}O_3$ | 226 | 36.625 | 0.9 |  | - |
| 64 | 6-Methoxyeugenyl isovalerate | $C_{16}H_{22}O_4$ | 278 | 37.095 | 1.98 |  | - |
| 65 | 1,4-butanediol, 2,3-divanillyl- | $C_{20}H_{26}O_6$ | 362 | 39.555 | 0.76 |  | Antioxidant, antimicrobial activity, anti-myocardial ischemia effects |

MF: Molecular Formula, MW: Molecular Weight, RT: Retention Time

## Conclusion

The plant extracts remarkably inhibited the growth of tested bacterial strains. The antibacterial and GC-MS analysis of *P. paucinervis* confirmed the presence of many bioactive compounds. Thus this study reveals that *P. paucinervis* may serve as a potential source of medicine due to the presence of these compounds. Further research on *P. paucinervis* is necessary for finding the active principles and their mode of action.

## Acknowledgment

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## References

1. Angiosperm Phylogeny Group. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG III. Botanical Journal of the Linnean Society. 2009; 161(2):105-121.
2. Bauer AW, Kirby WMM, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. American Journal of Clinical Pathology. 1996; 45(4ts):493-496.
3. Bose VL. Pharmacognostic and phytochemical analysis of *Premna serratifolia* L. in Kerala. University of Calicut. Kozhikode. India, 2014, 17.
4. Dogra KS, Chauhan S, Jalal JS. Assessment of Indian medicinal plants for the treatment of asthma. Journal of Medicinal Plants Research. 2015; 9(32):851-862.
5. Francis S, Gideon VA, Britto SJ, Dessy VJ. A review on two endemic species of genus *Premna* and their conservational importance, Journal of Drug Delivery and Therapeutics. 2019; 9(4-s):666-669.
6. Gomathi D, Kalaiselvi M, Ravi kumar G, Devaki K, Uma C. GC-MS analysis of bioactive compounds from the whole plant ethanolic extract of *Evolvulus alsinoides* (L.) L. Journal of Food Science and Technology. 2015; 52(2):1212-1217.
7. Kumar KM, Sunil kumar T, Sreeraj V, Thomas B, Balachandran I, Antony VT *et al*. A new species of the genus *Premna* L. (Lamiaceae) from Western Ghats of India. Webbia. 2013; 68(2):127-31.
8. Rekha K, Richa PK, Babu S, Rao M. A phytochemistry of the genus *Premna*: a review. International Journal Pharmaceutical and Chemical Sciences. 2015; 4(3):317-25.
9. Robi AJ, Augustine J, Sasidharan N, Udayan PS. Rediscovery of an endemic and rare *Premna paucinervis* (Verbenaceae) from the Western Ghats after a century, Rheedea. 2013; 23(1):10-12.
10. Singh R. Medicinal plants: A review. Journal of Plant Sciences. 2015; 3(1-1):50-55.