# Chemical composition, antioxidant and antibacterial activities of the essential oils of medicinal plant *Cymbopogonnardus* from Lembang West Java

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***Abstract***
*Different growing places provide a difference in essential oil content that will also affect its biological action. The essential oil was obtained by hydro-distillation from the leaves of Cymbopogon nardus from Lembang West Java. Gas chromatography mass spectrometry (GC–MS) analysis revealed the presence of 53 compounds with the major constituents, including citronella (26.27%), cadinene (6, 97), methyl isoeugenol (5,87%), geranyl acetate (4,41%) and citronellyl propionate (4,97%). The antioxidant activities of the essential oils were determined using 1, 1-diphenyl-2-picrylhydrazyl (DPPH) assay. The results demonstrated that essential oils have antioxidant activities with the EC50% 2.44 µg. mL$^{-1}$.*

*Finally, essential oils showed antibacterial activity against the tested Gram positive and Gram-negative bacteria. The minimum inhibitory concentration of the essential oils ranged from 250 µg. mL$^{-1}$ to 1000 µg. mL$^{-1}$. The essential oil content of plants grown in the highlands such as Lembang is recommended for maximum essential for antioxidant and antibacterial activity*.

**Keywords**: Antibacterial, Antioxidant, Chemical composition, *Cymbopogon nardus.*

## Introduction

Essential oils are oils that hold a distinctive scent and have various uses as preservatives, antioxidants and antimicrobials, which are normally extracted from leaves, branches, roots, stems, fruits and seeds. Essential oils are subtle, aromatic and volatile liquids extracted from various plant parts as secondary metabolites. Secondary metabolites play important ecological and biological roles and are important for plant defense as they often contain antimicrobial and antioxidant properties[9].

The *Cymbopogon* genus is a member of the Gramineae family which is a herb known worldwide for its high essential oil content. They are widespread in all continents where they are employed for several functions. Commercial uses and medical specialties of various species of *Cymbopogon* are well documented.

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Ethno pharmacological evidence suggests that they have a variety of properties that justify their use for pest control, cosmetics and as anti-inflammatory agents. This plant also promises as a potent antitumor and chemopreventive drug. This case of chemotherapy of this genus has been employed as a biomarker for their identification and categorization[1].

*Cymbopogon nardus* L. essential oils, apart from their broad use in food or drinks, are widely involved in perfumery, body care products and soap manufacture. Also important is their pharmaceutical usage.[4] The pharmacological application of *Cymbopogon nardus* is well worked, although studies indicate that other species may also be pharmaceutically useful. Hence this study intends to discuss these species and explore their potential economic importance[10].

## Material and Methods

**Plant material and experimental condition**: Leaves *Cymbopogon nardus* were collected by the Research Institute for Medicinal and Aromatic Plants (BALITTRO), Manoko, District Lembang, West Bandung regency. The species were identified and deposited by Herbarium Jatinangor, Plant Taxonomy Laboratory, Department Biology, Padjadjaran University with voucher number specimens 123/HB/03/2017.

**Essential oil isolation**: Fresh leaves of *Cymbopogon nardus* (1000 g) were isolated with water and steam distillation apparatus for three hours. Subsequently, it yielded 5 mL essential oil (0, 5%). The essential oil samples were tagged and stored at 4°C until being used for analysis.

**Gas chromatography–mass spectrometry (GC–MS) analysis**: Essential oil compositions were determined using Shimadzu GCMS-QP2010 Ultra gas chromatography. The oven temperature was initiated at 60 ºC. Injector and detector temperatures were set to 280 ºC respectively. The analyses were carried out using helium as the carrier gas at a flow rate of 1.31 mL/min. Total flow 264.7 mL/min, pressure 80.2 kPa, linear velocity 41.7 cm/Sec, flow 2.0 mL/min and split ratio 200. The MS was equipped with an ion source temperature 230 ºC, interface time 250ºC, detector gain mode absolute and detector gain 0.80 kV.

**Antioxidant activity (DPPH Radical Scavenging Assay)**: The method was used for the determination of the scavenging activity of DPPH free radical. Different

methanolic dilutions of extracts were mixed with equal volumes of DPPH methanol solution (0.004% w/v). After a 30-minute incubation period at room temperature, the absorbance was read against a blank at 517 nm. Inhibition of free radical by DPPH in percent (I %) was calculated in the following way:

% Inhibition = (AC – AS / AC) × 100

where AC is the absorbance of control reaction (containing the equal volumes of DPPH solution and methanol without any sample) and AS is the absorbance of the sample (Essential oil and standards).

**Antimicrobial activity**: The antibacterial activity of the essential oil was evaluated by microdilution method and tested against 6 bacteria. 4 Gram-positive bacteria were *Staphylococcus aureus* ATCC6538, *Bacillus subtilis* ATCC6633, *Bacillus cereus* ATCC11778 and MRSA respectively. 2 Gram-negative bacteria were *Pseudomonas aeruginosa* ATCC9027 and *Escherichia coli* ATCC8939 respectively. These bacteria were supplied by the Microbiology Laboratory Institute Technology Bandung, Indonesia and stacked away with liquid paraffin wax at 4 °C. All strains were cultured at 37 °C on Mueller-Hinton medium.

## Results and Discussion

**Chemical composition of the essential oil:** A total of 53 components were identified and quantified. The total chemical composition of the essential oil as given in figure 1. The major constituents of the essential oil were citronella (26,27%), δ-cadinene (6,97%), methyl Isoeugenol (5.87%), caryophyllene (5.87%), geranyl butyrate (5.6%), geranyl acetate (4.41%), citronellyl propionate (4.97%), germacrene (2.97%), α-bergamotene (2.84%), eugenol (2.54%), β-element (2.34%), δ-guaiene (1.81%), (-) - isolongifolol (1.7%), farnesol isomer B (1.62%), linalool (1.61%), δ-limonene (1.58%) and other minor components (table 1).

Previous research reported that the chemical content of C*ymbopogon nardus* consists of 35 components[7] and 22 components[11]. The essential oil is a product of secondary metabolites of plants. The chemical content of essential oil profile tends to exhibit different chemical, caused due to ecological factors, geographical condition, age and time of harvesting plants. Differences in chemical properties will have an effect on biological activity to produce a uniform biological activity. Then a major component analysis can be used as an alternative as the basis for determining the quality of essential oils.

The main content of essential oil of *Cymbopogon nardus* is a citronella, citronellol, geraniol, geranial, methyl isoegeunol and neral.[7,10,13]

**Antioxidant activity**: The use of DPPH provides the easy and quick way to evaluate the antioxidant activity.[7] Essential oils were subjected to screening for possible antioxidant activity by the free radical DPPH test system. Stable free radical DPPH is commonly used to determine the ability of a compound to scavenge free radicals. This method is based on the reduction of the methanol solution of DPPH by the presence of hydrogen donating molecules. Reduction of DPPH solution is monitored by measurement of absorption at 517 nm.

*Cymbopogon nardus* essential oil showed the ability of antioxidants to reduce the DPPH Radical with IC50 2.405 µ g/mL. The antioxidant potential of the extracts was also compared with that of ascorbic acid as reference compounds with a well-known capacity to scavenge free radicals. It has been reported also on previous research that families of *Cymbopogon* have antioxidant activity[2,6].

**Antibacterial activity**: In the microwell dilution assay, the essential oil also presented antibacterial activities against all the bacterial strains tested with MIC values of 125 and 2000 µg/mL. The negative control (5% DMSO) did not indicate activity for any of the microorganisms tested. The lowest MIC value was 125 µg/mL of *Bacillus cereus* and the essential oil showed weaker activity against *Bacillus subitilis* with 1000 µg/mL, *Escherichia coli* 500 µg/mL, *Pseudomonas aeruginosa* 2000 µg/mL, *Staphylococcus aureus* 1000 µg/mL and MRSA 2000 µg/mL.

It was reported that essential oils have the antimicrobial activity of.[3,5,12] The research is in line with previous research in which the family has proven *Cymbopogon* antimicrobial activity[13], antibacterial against bacterial *Edwardsiella* spp, *Vibrio* spp, *Escherichia coli*, *Aeromonas* spp, *Salmonella* spp, *Flavobacterium* spp, *Pseudomonas* spp and *Streptococcus* spp. MIC values of the citronella oil ranged from 0244 µg/ml to 0977 µg/ml[12] and antifungal against *Candida* sp with MIC values ranging from 250 to 1000 µ g/Ml[8,10].

## Conclusion

In the present research, the chemical composition, antioxidant and antibacterial activity of the essential oil from the leaves of *Cymbopogon nardus* are reported. Fifty-three compounds were identified and quantified from the leaf essential oil of *Cymbopogon nardus*.

The essential oil demonstrated potent antioxidant with IC50 2,405 µg/mL and antibacterial activity against *Bacillus subitilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and MRSA This result confirms the use of the traditional aromatic plant in the treatment of some infectious diseases. Further research is needed to investigate other potential activities.

**Table 1**
**Essential oil composition of *Cymbopogon nardus* identified by GC-MS.**

| S.N. | Chemical compounds | % |
|---|---|---|
| 1 | Citronella | 26,27 |
| 2 | δ-cadinene | 6,97 |
| 3 | Methyl Isoeugenol | 5,87 |
| 4 | Caryophyllene | 5,87 |
| 5 | Geranyl butyrate | 5,6 |
| 6 | Geranyl acetate | 4,41 |
| 7 | Citronellyl propionate | 4,97 |
| 8 | Germacrene-D | 2,97 |
| 9 | α-Bergamotene | 2,84 |
| 10 | Eugenol | 2,54 |
| 11 | β-Elemene | 2,34 |
| 12 | δ-Guaiene | 1,81 |
| 13 | (-)-Isolongifolof | 1,7 |
| 14 | Farnesol Isomer B | 1,62 |
| 15 | Linalool | 1,61 |
| 16 | δ-Limonene | 1,58 |
| 17 | 1,2,4-Metheno-1H-indene, octahydro-1,7a-dimethyl-5-(1-methylethyl) | 1,5 |
| 18 | geranyl hexanoate | 1,4 |
| 19 | α-Humulene | 1,38 |
| 20 | Citral | 1,23 |
| 21 | γ-Cadinene | 1,06 |
| 22 | Epi-Bicyclosesquiphellandrene | 0,96 |
| 23 | α-Guaiene | 0,9 |
| 24 | trans-β-Farnesene | 0,82 |
| 25 | 3a(1H)-Azulenol, 2,3,4,5,8,8a-hexahydro-6,8a-dimethyl-3-(1- methyl ethyl) | 0,78 |
| 26 | δ-Cadinene | 0,65 |
| 27 | Z-Citral | 0,56 |
| 28 | Naphthalene | 0,56 |
| 29 | Seychellene | 0,55 |
| 30 | cis-Ocimene | 0,48 |
| 31 | β-Ocimene | 0,47 |
| 32 | 1,2,4-Metheno-1H-indene, octahydro-1,7a-dimethyl-5-(1- methyl ethyl) | 0,44 |
| 33 | 2-N-Butyldecalin | 0,39 |
| 34 | β-Myrcene | 0,39 |
| 35 | α-Patchoulene | 0,39 |
| 36 | n-Decanal | 0,38 |
| 37 | Neryl acetate | 0,36 |
| 38 | β-Sesquiphellandrene | 0,35 |
| 39 | β-Patchoulene | 0,35 |
| 40 | Naphthalene, 1,2,3,4,4a,7-hexahydro-1,6-dimethyl-4-(1- methyl ethyl) | 0,34 |
| 41 | Thiogeraniol | 0,31 |
| 42 | (-)-Caryophyllene oxide | 0,29 |
| 43 | α-Cubebene | 0,21 |
| 44 | Germacrene B | 0,2 |
| 45 | Citronellyl butyrate | 0,19 |
| 46 | B-Bourbonene | 0,19 |
| 47 | Neophytadiene | 0,16 |
| 48 | Isomenthone | 0,16 |
| 49 | β-Selinene | 0,15 |
| 50 | Geraniol formate | 0,14 |
| 51 | Geranyl Isovalerate | 0,13 |
| 52 | n-Dodecanal | 0,12 |
| 53 | δ-Elemene | 0,11 |

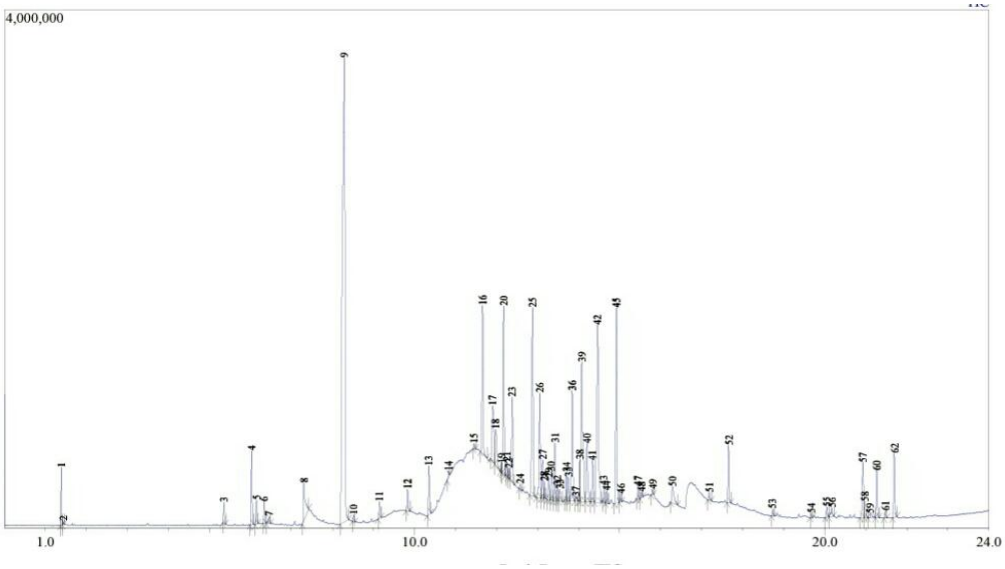


**Figure 1: GC-MS chromatographic profiles of *Cymbopogon nardus***

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