

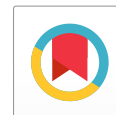


## GC-MS Analysis and Antimicrobial Evaluation of *Oldenlandia Corymbosa*

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### Abstract

The aim of the present study is finding out the bioactive chemical constituents and to evaluate the antimicrobial activity of the methanol extract of *Oldenlandia corymbosa* medicinal plant. This study involves the preliminary phytochemical screening separation and identification of phytochemicals. The methanolic extract was subjected to GC-MS analysis. Alkaloids, tannins, glycosides, terpenoids, steroids, flavonoids, and saponins are found in the plant extracts. The extract also was tested for antimicrobial activity by disc method. The extract of the plant showed antimicrobial activity against both gram (+) and gram (-) bacteria.

**Keywords:** Antimicrobial activity; GC-MS analysis; *Oldenlandia Corymbosa*; Phytochemical screening; Therapeutic use.

### 1. INTRODUCTION

Medicinal plants have a wide variety of chemical constituents and some of them have the ability to inhibit the growth of microorganisms (Elhoussine derwich *et al.* 2011). Medicinal plants are the source of great economic value in the Indian subcontinent. In recent years secondary plant metabolite, phytochemicals have been extensively investigated as a source of medicinal agents (Okoli *et al.* 2009). Today natural products derived from plants are being tested for presence of new drugs with new modes of pharmacological action (Charles *et al.* 2011).

However, synthetic medicine can cause side effects and as a result people are more favorable to use natural compounds obtained from plants. About 20,000 plant species are used for medicinal purposes (Abdel-Ghani *et al.* 2001). Hence the present study has been

attempted to study the GC-MS analysis and antimicrobial activity of *Oldenlandia corymbosa* plant.

### 2. MATERIALS & EXPERIMENTAL METHODS

Fresh parts of plant *Oldenlandia corymbosa* were collected at kolli hills in Namakkal district, Tamil Nadu. The plant materials were identified by botanically. The plant materials were dried until all the water molecules evaporated and plants became well dried for grinding. After drying, the plant materials were grinded well using mechanical blender into fine powder and transferred into the sealed container with proper labeling.

Crude plant extract was prepared by Soxhlet extraction method. About 50 g of powdered plant material was uniformly packed into a thimble and extracted with 250 ml of methanol solvent is used separately. The process of extraction continues for 15 hours or till the solvent in siphon tube of an extractor become colorless. After that the extract was taken in a beaker and kept on hot plate and heated at 30-40 °C till

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**Table 1. Phytochemical components identified in the alcoholic extract of *Oldenlandia corymbosa***

Name of the Test	Phytochemical constituents	Methanol Extract
Mayer's Test	Alkaloids	++
Dragon-draff Test		++
Wagner Test		++
Molish Test	Carbohydrates	-
Fehling Test		+
Benedict Test		-
Foam Test	Saponins	+
Lead Acetate Test	Tannins	+
Ferric chloride Test	Pseudo tannins	Condensed Tannin
Ammonia Test	Chlorogenic acid	+
Salkowaski Test	Steroidal Glycosides	-
H <sub>2</sub> SO <sub>4</sub> Test	Anthocyanin	-
Liebermann's Burchard Test	Steroidal Glycosides	+
H <sub>2</sub> SO <sub>4</sub> Test	Saponins glycosides	+
Ammonia Test	Flavonoids	++
Shinoda's Test	Flavones	-
Ferric chloride Test	Phenolic compounds	++
Sodium chloride Test	Coumarin	+
Borntrager's Test	Anthracene Glycoside	-

**High : +++****Present : +****Moderate : ++****Absent : -**

all the solvent gets evaporated. Dried extract was stored in refrigerator at 4 °C.

The dried extract was injected to the GC-MS equipment and the sample was analyzed using standard procedure (Lalitha *et al.* 2012). The antimicrobial activity was determined using disc diffusion method (Zahir Hussain and Aruna, 2010; Hosamani *et al.* 2012) by measuring zone of inhibition in mm and comparing with standard drugs *Ciproflaxin* and *fluconazole*. The extract was also tested for the presence of bioactive compounds by qualitatively using standard methods (Suresh G. Killedar and Harinath, 2011).

### 3. RESULTS & DISCUSSION

Qualitative preliminary screenings of extracts were performed initially with different chemical reagents to detect the phytoconstituents present in each extract. The extracts showed the presence of alkaloids, carbohydrates, saponins, tannins, glycosides, flavonoids and phenolic compounds. The results are presented in table 1.

### 4. GC-MS ANALYSIS

GC-MS studies reported the methanol extracts of the whole plant of *oldenlandia corymbosa*. The various plant phytochemical components are found in the plant of *oldenlandia corymbosa*. They are listed in table 3.

The peak is found at RT 5.44 with a peak area of 1.67 %. It shows N-Propylethylenediamine. This compound is an alkaloid with a molecular formula C<sub>5</sub>H<sub>14</sub>N<sub>2</sub> and molecular weight 102.18. (Fig 1) Amine compound shows antimicrobial and antimalarial activities. Alkaloids play some metabolic role and control development in living system (Sunita Dalal and Sudhir K. Kataria, 2010).

The peak at RT 8.12 with a peak area of 45% corresponds to the compound is Acetylcyclohexane. The molecular formula and molecular weight of this compound is C<sub>8</sub>H<sub>14</sub>O and 126.2 respectively. Acetylcyclohexane compound belongs to alkane group. It has antioxidant activities.

Table 2. GC-MS analysis for *Oldenlandia corymbosa* methanolic extract

S.No	RT	Name of the compound	Molecular formula	Molecular Weight	Peak area %
1	3.3845	2,4-Diisocyanato-1-methylcyclohexane	C <sub>9</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub>	180.21	0.0589
2	3.4931	1H-Imidazole, 2-ethenyl-	C <sub>5</sub> H <sub>6</sub> N <sub>2</sub>	94.12	0.0427
3	3.5731	13C-Bicyclo[1.1.0]butane	C <sub>4</sub> H <sub>6</sub>	54.2	0.0511
4	3.9732	2-Methyl-2-oxazoline	C <sub>4</sub> H <sub>7</sub> N O	85.1	0.035
5	4.0246	4-methyloxazole	C <sub>4</sub> H <sub>5</sub> N O	83.2	0.0567
6	4.7618	N,N-Dimethyl hydroxylamine	C <sub>4</sub> H <sub>9</sub> NO <sub>5</sub>	151.12	0.168
7	5.0591	2-Cyclobutene-1-carboxamide	C <sub>5</sub> H <sub>7</sub> NO	98.15	0.1964
8	5.442	N-Propylethylenediamine	C <sub>5</sub> H <sub>14</sub> N <sub>2</sub>	102.18	1.6671
9	5.6649	pentyl	C <sub>5</sub> H <sub>11</sub>	71.14	0.2006
10	5.8021	2-Methylcyclopentanone	C <sub>6</sub> H <sub>10</sub> O	98.14	0.6922
11	5.9792	2(5H)-Furanone, 5-methyl-	C <sub>5</sub> H <sub>6</sub> O <sub>2</sub>	98.21	0.0712
12	6.1221	1H-Pyrrole, 2,3-dimethyl-	C <sub>6</sub> H <sub>9</sub> N	95.15	0.0566
13	6.3278	1-Amino-2-propanol	C <sub>3</sub> H <sub>9</sub> NO	75.11	0.0423
14	6.545	1-Propanol, 2-methyl-	C <sub>4</sub> H <sub>10</sub> O	74.12	0.6643
15	0.0279	2-Propenamide	C <sub>3</sub> H <sub>3</sub> NO	71.20	0.0279
16	0.0473	2-Hydroxycyclohexanone	C <sub>6</sub> H <sub>10</sub> O <sub>2</sub>	114.14	0.0473
17	0.0434	Acrylaldehyde	C <sub>3</sub> H <sub>4</sub> O	56.12	0.0434
18	8.1167	Acetylcyclohexane	C <sub>8</sub> H <sub>14</sub> O	126.2	0.0468
19	8.1625	2,4-octadienol	C <sub>8</sub> H <sub>14</sub> O	126.22	0.1045
20	8.231	3-Hexen-2-one, 3,4-dimethyl-	C <sub>8</sub> H <sub>14</sub> O	126.21	0.0428
21	8.3739	Trimethylene imine	C <sub>3</sub> H <sub>7</sub> N	57.09	0.041
22	8.4025	Azetidine	C <sub>3</sub> H <sub>7</sub> N	57.09	0.1001
23	8.6368	1-Propene, 3-azido-	C <sub>3</sub> H <sub>5</sub> N <sub>3</sub>	83.05	0.023
24	8.6654	2-Heptenal, 2-methyl-	C <sub>8</sub> H <sub>14</sub> O	126.12	0.0371
25	8.9569	2,2,6,6-D4-cyclohexanone	C <sub>6</sub> H <sub>6</sub> D <sub>4</sub> O	102.70	0.2076
26	9.0655	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	C <sub>6</sub> H <sub>8</sub> O <sub>4</sub>	144.13	2.6146
27	9.437	trans-2-Pentalal	C <sub>5</sub> H <sub>8</sub> O	84.06	0.1862
28	9.5342	Acetamide, N-methyl-	C <sub>3</sub> H <sub>7</sub> NO	73.16	0.1288
29	9.7513	3,3,3-Trifluoro-2-methylpropene	C <sub>4</sub> H <sub>5</sub> F <sub>3</sub>	110.15	0.3514
30	9.9342	p-vinylphenol	C <sub>8</sub> H <sub>8</sub> O	120.18	1.0893
31	10.042	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	C <sub>6</sub> H <sub>7</sub> NO <sub>3</sub>	141.32	0.9815
32	10.231	Methyl 3-hydroxytetradecanoate	C <sub>15</sub> H <sub>30</sub> O <sub>3</sub>	258.45	0.8202
33	10.654	1-Tetradecyne	C <sub>14</sub> H <sub>26</sub>	194.36	0.301
34	10.728	2H-Imidazol-2-one, 1,3-dihydro-1,4,5-trimethyl-	C <sub>8</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub>	168.19	0.4363
35	11.288	2-Methoxy-4-vinylphenole	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	150.17	0.8479

S.No	RT	Name of the compound	Molecular formula	Molecular Weight	Peak area %
36	11.4259	Acetamide, N-methyl-	C <sub>3</sub> H <sub>7</sub> NO	73.09	0.0657
37	12.5518	2,5-Dimethylpyrazine	C <sub>6</sub> H <sub>8</sub> N <sub>2</sub>	108.14	0.401
38	13.9235	2-Propynamide	C <sub>3</sub> H <sub>7</sub> NO	73.32	0.1744
39	14.335	Aziridine, 1-(2-buten-1-yl)-, (Z)-	C <sub>6</sub> H <sub>11</sub> N	97.09	0.506
40	14.3922	p-Hydroxybenzoic acid	C <sub>7</sub> H <sub>6</sub> O <sub>3</sub>	138.14	0.5859
41	14.6665	2-Cyanocyclopentanone	C <sub>6</sub> H <sub>7</sub> NO	109.18	0.2492
42	14.8551	Benzeneethanamine	C <sub>8</sub> H <sub>11</sub> N	121.64	0.1542
43	15.4152	Propynamide	C <sub>3</sub> H <sub>3</sub> NO	69.02	0.1426
44	15.6553	3-Deoxy-d-mannonic lactone	C <sub>6</sub> H <sub>10</sub> O <sub>5</sub>	162.06	2.5728
45	16.524	n-Butylamine	C <sub>4</sub> H <sub>12</sub> N	74.178	0.1085
46	21.3363	2-Propenenitrile, 2,3,3-trifluoro-	C <sub>3</sub> F <sub>3</sub> N	107.04	0.063
47	26.5029	Anthranilic acid, N-methyl- butyl ester	C <sub>14</sub> H <sub>19</sub> N O <sub>5</sub>	282.16	0.1258
48	26.9773	2-Ethylacridine	C <sub>15</sub> H <sub>13</sub> N	207.19	1.0527
49	27.8917	Silicic acid, diethyl bis(trimethylsilyl) ester	C <sub>10</sub> H <sub>28</sub> O <sub>4</sub> Si <sub>3</sub>	296.52	2.4851
50	27.9546	Cuvan 80	C <sub>6</sub> H <sub>18</sub> O <sub>3</sub> Si <sub>3</sub>	222.48	1.4888
51	28.8748	Cyclotrisiloxane, hexamethyl-	C <sub>6</sub> H <sub>18</sub> O <sub>3</sub> Si <sub>3</sub>	222.46	1.0931
52	29.2405	1,3-Bis(trimethylsilyl)benzene	C <sub>12</sub> H <sub>22</sub> Si <sub>2</sub>	222.48	0.9437

The peak at RT 9.067 and peak area 26% is 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- which is a flavonoid compound. The molecular formula and the molecular weight of this compound is C<sub>6</sub>H<sub>8</sub>O<sub>4</sub> and 144.13, respectively. This flavonoid is known to possess most medicinal activities like antidiabetic, anti-inflammatory, antidermatic, antileukemic and anticancer activities. It has been recognized that flavonoids show antioxidant activity and their effects on human nutrition and health are considerable. The mechanisms of action of flavonoids are through scavenging or chelating process.

The amine group of compounds n-Butylamine is observed at peak at RT 16.524 with a peak area of 10%. This compound has the molecular formula C<sub>4</sub>H<sub>12</sub>N

and molecular weight is 74.145. It shows anti-malarial and anti-inflammatory activity.

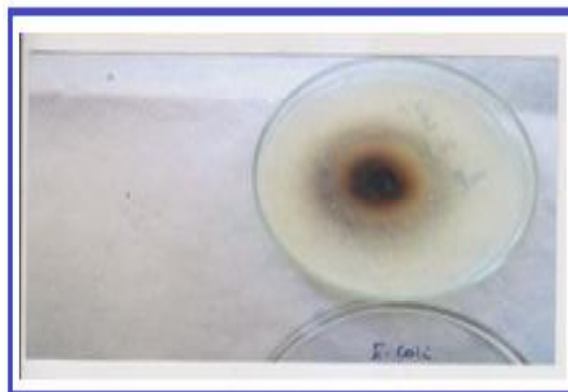
The peak at RT 6.12 with peak area 5% shows 1H-Pyrrole, 2,3-dimethyl this compound is Pyrrole derivative compound. The molecular formula is C<sub>6</sub>H<sub>9</sub>N and molecular weight of this compound is 95.14. This compound has antibacterial and anti-inflammatory activity.

## 5. ANTIMICROBIAL ACTIVITY

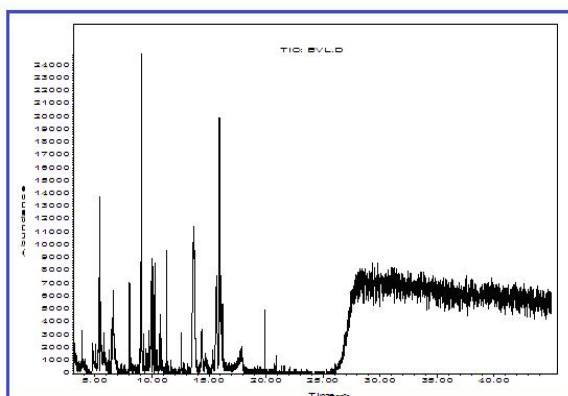
The extract was subjected to antimicrobial activities by disc diffusion method (Zahir Hussain et al. 2010). Methanolic extract of *Oldenlandia corymbosa* plant shows antimicrobial activity against the tested

**Table 3. Antibacterial activity of methanol extract of *oldenlandia corymbosa***

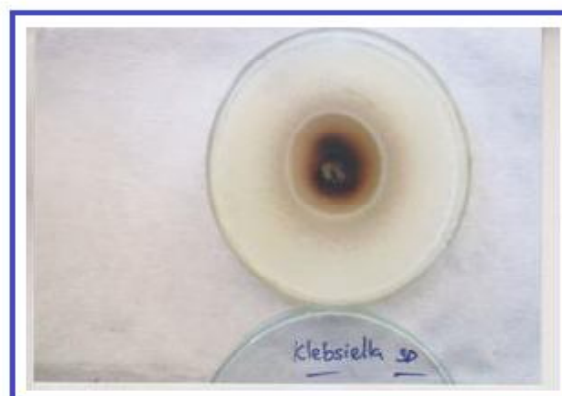
Organism	Zone of inhibition (mm)
<i>Bacillus sp</i>	27
<i>Escherichia coli</i>	32
<i>Klebsiella sp</i>	33
<i>Proteus sp</i>	22
<i>Pseudomonas sp</i>	26
<i>Staphylococcus aureus</i>	28
<i>Candida albicans</i>	13
<i>Aspergillus niger</i>	12
Control	15



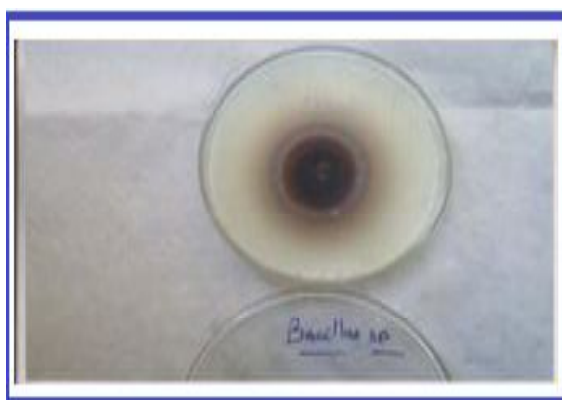
**Fig. 2b: Zone of inhibition measured in 32mm**



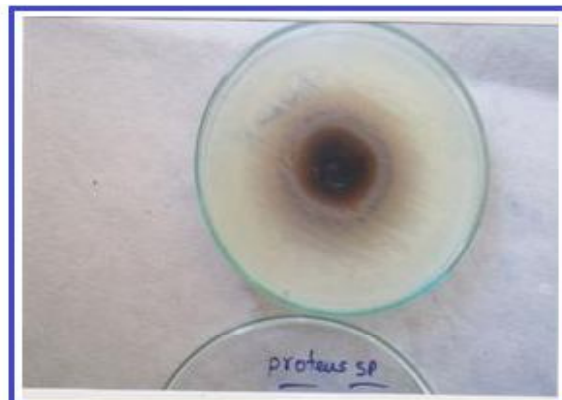
**Fig. 1: Methanol extracts of *Oldenlandia Corymbosa* (GC-MS Analysis)**



**Fig. 2c: Zone of inhibition measured in 33mm**



**Fig. 2a: Zone of inhibition measured in 27mm**



**Fig. 2d: Zone of inhibition measured in 22mm**

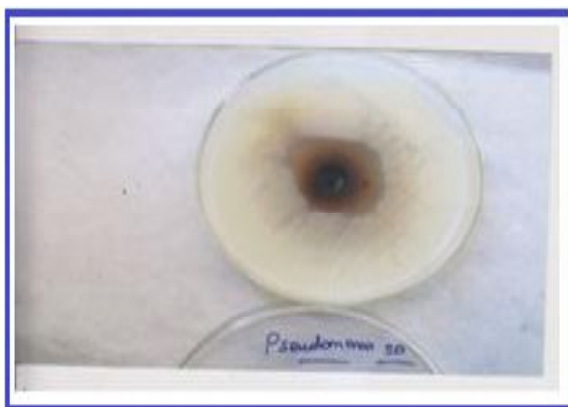


Fig. 2e: Zone of inhibition measured in 26mm

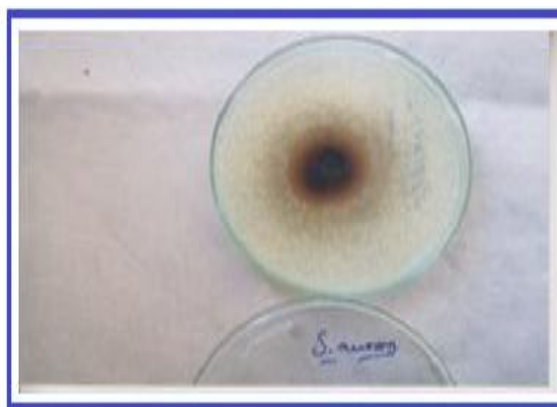


Fig. 2f: Zone of inhibition measured in 28mm



Fig. 2g: Zone of inhibition measured in 15mm (Ciprofloxacin)

organisms in the order of *Bacillus* (27mm), *Escherichia coli* (32mm), *Klebsiella* (33mm), *Proteus* (22mm), *Pseudomonas* (26mm), *Staphylococcus aureus* (28mm), *Candida albicans* (13mm) and *aspergillus niger* (12mm). In case of fungi, antimicrobial activity against tested organisms was in the order of *C. albicans* (10mm) and *A. niger* (9mm). The maximal antibacterial activity is observed against *Klebsiella* while maximal antifungal activity is found against *candida albicans* which are shown in Fig 1. and the values are represented in Table 2.

## 6. CONCLUSION

The methanolic extracts of plant contains many bioactive chemical constituents including alkaloids, glycosides, terpenoids, steroids, flavonoids, and tannins. In the present study fifty two phyto chemical constituents have been identified from ether extract of the whole plant of *Oldenlandia corymbosa* by GC-MS analysis. The extract of *oldenlandia corymbosa* is subjected to antimicrobial activity against tested organisms both gram(+) and gram(-) namely

*bacillus, Escherichia coli, klebsiella, proteus, pseudomonas, staphylococcus aureus, candida albicans, aspergillus niger*. The maximal bacterial activity is found in *klebsiella* species and fungi activity is found in *candida albicans*. The presence of various bioactive compounds justified the use of whole plant for various ailments by traditional practitioners.

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