

## Analysis of anti-bacterial and phytochemical screening by using different *Anisomeles malabarica* samples

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### Abstract

The aqueous leaf, flower, stem and boiled leaf extracts of *Anisomeles malabarica* were evaluated for antibacterial screening by disc diffusion methods along with their phytochemical screening. The *Anisomeles malabarica* extracts are sensitive to five pathogenic organisms such as *Staphylococcus aureus*, *Staphylococcus epidermis*, *E.coli*, *Pseudomonas aeruginosa* and *Bacillus substilis* except *Proteus vulgaris* by disc diffusion methods. The phytochemical screening to identify the active compounds.

**Keywords:** *Anisomeles malabarica*, Antibacterial activity, Phytochemical screening.

## 1. Introduction

India is known worldwide for its Ayurvedic treatment. Herbal plants have high medicinal values as they have the rich source of Bioactive compound and have no side effects. The bioactive compound from the herbal plant reduce the virulence of the microorganisms, thereby preventing and protecting from the infections.

*Anisomeles malabarica* (L) is a traditional medicinal plant, distributed throughout India. It has been used in folk medicine for the treatment of cancer, liver disorder, stomach ailment, fever, cold and cough<sup>1</sup>.

*Anisomeles* belong to *Lamiaceae* family is represented by 45 genera and 574 species with 256 endemic species<sup>2</sup>. It is an Aromatic, densely pubescent, perennial herb, 1.2-2m in height belonging to the family Lamiaceae. The common vernacular names of the plant are Malabar catmint, chodara, Bhutan Kusham, Peyameratti etc.<sup>3</sup>.

Plant have been known to synthesize active secondary metabolites such as tannins, steroids, phenolic compounds etc, found in essential oils with established potent antimicrobial activities, which indeed has formed the basis for their applications in some pharmaceuticals, alternative medicines and natural therapies<sup>4</sup>. The antimicrobial compound from plant may inhibit bacterial growth by different mechanisms than those presently used<sup>5</sup>. The aim of the studies have identified and isolate the main active ingredients in the plants responsible for this antimicrobial activity against the pathogens.

## 2. Materials and Methods

**2.1 Collection of Plant Materials:** Fresh, young plant of *Anisomeles malabarica* L was collected from Thekamalai near Vaiyampatti, dry rocky region of Tiruchirappalli district, Tamil nadu. The leaves, stem and flowers were separated from the collected plant. Then it were air dried in shade for 15 days and then pulverized to fine powder for further analysis.

### 2.2 Antibacterial screening

**2.2.1 Microbial strains used:** Different microbial strains were used to evaluate the antimicrobial effect of which three were Gram positive bacterial strains (i.e) *Staphylococcus aureus*, *Staphylococcus epidermis*, *Bacillus substilis* and three were from Gram negative bacterial strains (i.e) *Escherichia coli*, *Pseudomonas aeruginosa*, and *Proteus vulgaris*. The strains were obtained from MTCC, Chandigarh in India and maintained on agar slants.

**2.2.2 Disc diffusion method:** Disc diffusion method was carried out for antibacterial susceptibility testing according to the standard method to assess the presence of antibacterial activities of the plant extracts. Muller Hinton Agar plates were prepared. Overnight Muller Hinton broth culture of test organisms were seeded over the Muller Hinton plates using sterile cotton swab so as to make lawn culture. The discs which had been impregnated with aqueous extracts of leaf, stem, flower and boiled leaf extracts were placed on the Muller Hinton agar plates with the negative control disc and subjected to antibacterial screening. The plates were then incubated at 37°C for 18 to 24 hours depending on the species of bacteria used in this test. After the incubation, the plates were examined for inhibition zone.

**2.3 Phytochemical Analysis:** A qualitative phytochemical test to detect the presence of alkaloids, flavonoids, saponins, Tannins, steroid, glycosides, oils and fats, phenolic compounds, protein, Amino Acids, Gums and mucilage and Carbohydrates were carried out using standard procedure<sup>6</sup>.

### 3. Results

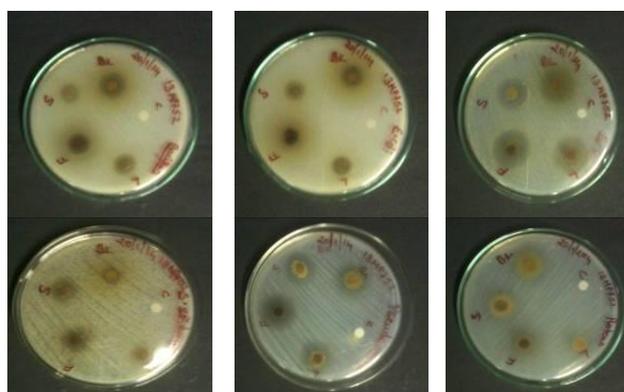
The bacterial infection affects human throughout their life. The infection depends on the nature of the organisms and interacts with the human body. Antibacterial activity was assayed invitro by disc diffusion method against six bacterial strains. The antimicrobial activity of the aqueous various parts of plant extracts of *Anisomeles malabarica* was more effective against *Staphylococcus aureus* (L-24mm,F-23,S-20,BL-21mm), *Staphylococcus epidermis* (L-10mm,F-11,BL-11mm), *E.coli*(L-9mm,F-9,BL-10mm), *Pseudomonas aeruginosa*(L-20mm,F-19mm) and *Bacillus subtilis* (L-9mm,S-12,BL-10mm). Where as no microbial activity against *Proteus vulgaris*. The bacterial strains revealed the zone of Inhibition (Table 1 and figure 1).

**Table 1. Zone of Inhibition formed by aqueous extracts of *Anisomeles malabarica* against bacterial Strains**

S. No	Sample	Bacterial Strains	Zone of Inhibition in Diameter (mm)				
			SV	L	F	S	BL
1	<i>Anisomeles malabarica</i> Leaf, Stem, Flower and Boiled leaf extracts	<i>S.epidermis</i>	20	10	11	-	11
2		<i>Paeruginosa</i>	20	20	19	-	-
3		<i>S.aureus</i>	20	24	23	20	21
4		<i>E.coli</i>	20	9	9	-	10
5		<i>Proteus vulgaris</i>	20	-	-	-	-
6		<i>Bacillus substilis</i>	20	9	12	-	10

L- Leaf; F-Flower; S- Stem; BL- Boiled leaf; SV-Standard value  
Table Value  $\chi^2(0.05)=3.841$  ,chi-square value significance at 5% level

**Fig.1 Anti-Microbial Activity by Disc Diffusion Method**



In earlier studies, Antibacterial activity of field grown *Anisomeles malabarica* was screened against gram positive and gram negative bacteria such as *E.coli*, *S.aureus*, *P.mirabilis*, *Paeruginosa*, *K.pneumoniae*. Both extracts showed varying of inhibitory effects. The inhibitory effects of extracts were directly propotional to the increasing concentration of field grown leaf<sup>7</sup>. Investigated including its wide spectrum of activity, wide ethanolic and methanolic extract of *Anisomeles malabarica* showed less activity compared to that of aqueous extract<sup>8</sup>.

The chi-square values obtained respectively which was less than the calculated table value.  $\chi^2(0.05)=3.841$  at 5% level of significance. Above results lead to the conclusion that the data was consistent with the hypothesis, the diameter of inhibition zone obtained from the observed data showed the similarities with experimental data.

In present study, the aqueous extracts of *Anisomeles malabarica* were subjected to suitable chemical tests to confirm the presence of Phytochemical compounds. The Table-2 showed the following phytochemical compounds such as Alkaloids, flavonoids, saponins, Tannins, sterols, Glycosides, Oil and fats, phenolic compound, Carbohydrates, protein and Amino acid ,Gums and mucilage were present.

**Table 2 : Phytochemicals Analysis**

S. No	Sample	PHYTOCHEMICALS	Interpretation			
			L	F	S	BL
1	<i>Anisomeles malabarica</i> Leaf, Stem, Flower and Boiled leaf extracts	Alkaloid	+	+	+	+
2		Flavonoids	+	+	-	+
3		Saponins	+	+	+	+
4		Tannins	+	+	+	+
5		Sterols	+	+	+	+
6		Oil and Fats	-	+	+	+
7		Phenolic Compounds	+	-	-	-
8		Protein & Amino Acid	+	+	+	+
9		Gums and Mucilage	+	+	+	+
10		Carbohydrates	-	+	-	+
11		Glycosides	+	-	-	-

(+) Presence; (-) Absence; L- Leaf; F-Flower; S- Stem; BL- Boiled leaf

#### 4. Discussion

In the earlier study by Remya Mohanraj *et al*<sup>o</sup> (2012) indicate the methanol extract of *Anisomeles malabarica* leaf has good antibacterial, while that of hexane ethyl acetate extract have less antibacterial activity against four pathogenic bacteria *K.pneumonia*, *S.aureus*, *V.cholerae* and *Aeroginosa*. The preliminary phytochemical screening of *Anisomeles malabarica* methanol leaf extract revealed the presence of phenolic in high amount followed by protein and carbohydrate in trace.

The presence of such phytochemicals may be correlated with the facts that aqueous extracts showed maximum activity against the bacterial strains. The active constituents of plants usually interfere with growth and metabolism of microorganisms in a negative manner.

#### 5. Conclusion

The present study showed the antibacterial activity of the leaf, flower, stem and boiled leaf extracts of *Anisomeles malabarica* against pathogenic organisms. Hence this plant can be use in folk medicine for the treatment of stomach ailment, fever, cold, liver disorder and cough. So the infected people continuously take the juice for 2 days. Immediately recovered that problem.

Futher studies are needed to isolate pure compounds from this plant extracts and to establish the mode of action of the isolated compounds.

#### References

1. Vijayalakshmi R, Ranganathan R, Chemopreventive effect of *Anisomeles Malabarica* whole plant extracts during DMBA induced hamster Buccal pouch carcinogenesis. *Asian Journal of Pharmaceutical & Clinical Research* 2012; 5(3): 185-188.
2. Erik S, Tarikahya B, Turkiye Florasi Uzerine, Kebikec, 2004; 17: 139-163.
3. Neeraj Choudhary, Amit Kumar, Naveen Bimal, Krishna Reddy Bar. A Review of Phytochemistry and Biological activities of *Anisomeles Malabarica*. *Journal of herbal Science* 2012; 1(1): 1-9.
4. Hammer KA, Carson CF, Riley TV. Antimicrobial activity of essential oils and other plant extracts. *J App microbiol* 1999; 86(6): 985-900.
5. Eloff JN. Which extract should be used for the screening and isolation of antimicrobial components from plants? *I ethopharmacol*, 1988; 60(1): 1-8.
6. Brindha P, Sasikala B, Purushothama K. Phytochemical analysis of *E. alba*. *BEER* 1981; 3(1): 84-96.
7. Kavitha T, Nelson R, Thenmozhi R, Priya E. Antimicrobial Activity and Phytochemical analysis of *Anisomeles Malabarica (L) R*. *Journal of Microbiology & Biotechnology Research* 2012; 2(1): 1-5.
8. Suresh Subramanian, Suriyavathana Muthakrishnan. Antimicrobial activity of various Extracts of leaf and wood of *Anisomeles malabarica*. *International Journal of current research* 2013; 5(3): 547-600.
9. Remya moharaj, Someshwar Nath, Pankaj jha. Bioactivity studies on *Anisomeles malabarica (AM) R.BR*. *IJPI'S Journal of Biotechnology and Biotherapeutics* 2012; 2(9): 1-8.