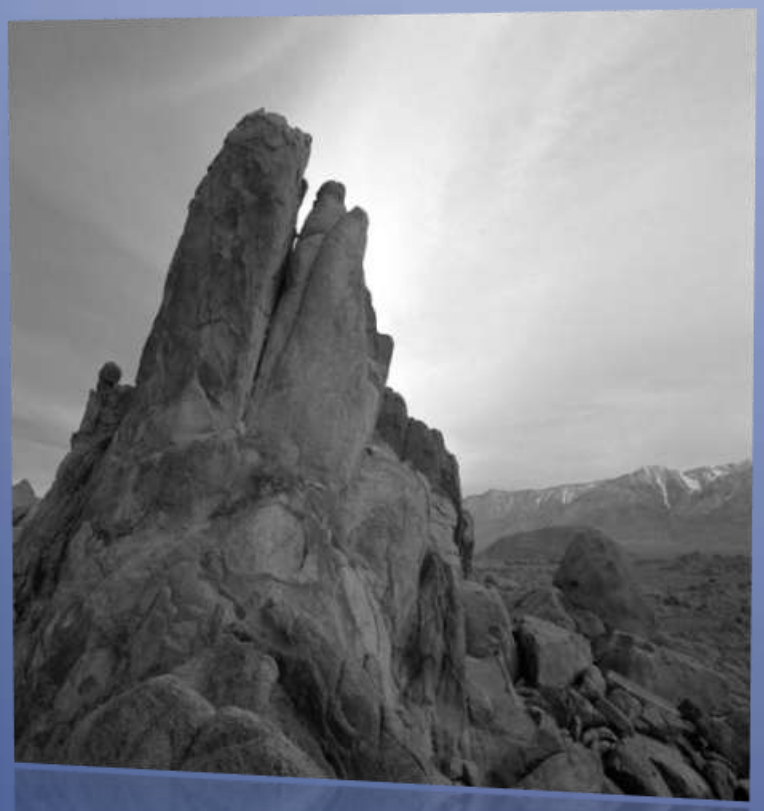

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In-Vitro Study of *Plectranthus Amboinicus* Against Respiratory Pathogens

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ABSTRACT

Plectranthus amboinicus has been reported to be traditionally used for medicine to cure common illnesses such as cough, stomachache, headache and skin infection. The present study is focused mainly on the ability of aqueous and ethanol extracts of the leaves of a traditional Indian medicinal plant against the pathogenic bacteria showing resistance against Ampicillin and *Fluoroquinolones,* isolated from respiratory infections. Different concentrations (20, 10, 5, 2.5, 1.25 µg/ml) of aqueous and alcoholic extracts were tested by agar well diffusion method. It was observed that the both extract were significantly inhibited respiratory resistant pathogenic bacteria against Ampicillin (10µg). The present study is therefore successful in indicating that *Plectranthus amboinicus* is useful in treatment of respiratory disorders associated with bacteria.

KEYWORDS

Plectranrhus amboinicus, Respiratory, Infections, Agar well diffusion

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INTRODUCTION

According to World Health Organization (WHO) more than 80% of the world population rely on traditional medicine for their primary health care needs and found a place in day-to-day life. It is important to note that homeopathy and modern medicine have their roots in medicinal plants. The compounds derived from medicinal plants form the ingredients of analgesics, antibiotics, heart drugs, laxatives, anti-cancer agents, ulcer treatments, contraceptives, diuretics Compounds from plants are referred as plant secondary metabolites, phytochemicals, anti-nutritional factors, plant xenobiotics.

The potential of finding new drugs that may be effective candidates for treating newly developing diseases in humans is great. Herbal molecules are safe and would overcome the resistance produced by the pathogens as they exist in a combined form or in a pooled form of more than one molecules in the protoplasm of the plant cell. Even with the advent of modern or allopathic medicine it have noted that a number of important modern drugs have been derived from plants used by indigenous people[2].

The biological properties are attributed to the occurrence of a wide range of bioactive compounds in the plant extracts as well as an essential oil. Thus, it can be stated that *P. amboinicus* has huge future prospects in meeting the global demand for natural, cost-effective and safer bioactive molecules in pharmaceutical and nutraceutical industries. Though several classes of phytocompounds are isolated and authenticated from this herb, their bioactivity and toxicity studies under in vivo conditions using animal models are limited to only a few compounds. Till now, no scientific evidence is available on the human safety aspects of *P. amboinicus* even though it is used widely in folk medicine. Further, some detailed investigations should be aimed at understanding the effectiveness of these isolated compounds in treating other human illnesses[1].

METHODOLOGY

a. COLLECTION OF PLANT MATERIAL

The fresh plant materials belong to the *Plectranthus amboinicus* were collected from the garden of the SDM college of Ayurveda. (Fig: 1)

b. MEDIA PREPARATION:

As per the standard protocol 100ml of Mueller Hinton Agar medium (Mueller Hinton Agar (MHA)- HIMEDIA M173 and

M063-500G Laboratories Pvt. Ltd. certified for ISO 9001:2008, WHO GMP. Munmai-400086, India.) was prepared, sterilized and about 10-15ml of the medium was poured into petriplates. Petriplates were kept in the refrigerator for other experimental usage[5].

Fig 1 Fresh plant material of *Plectranthus amboinicus*

c. **AQUEOUS ALCOHOLIC EXTRACTION BY COLD MACERATION TECHNIQUE**

Fifty grams of leaves were homogenized using pestle and mortar and mixed with 500ml of distilled water and ethanol separately and the bottles were stopper tightly and were kept in sterile condition for 72 hours with occasional shaking. The content was filtered and evaporated using water bath for 30 minutes [Figure- 2(a-f)]. The obtained plant extract was used to determine antibacterial property of the plant against respiratory disorders associated with bacteria[3.4].

Fig 2a Homogenization

Fig 2b Aqueous and ethanolic contents

Fig 2c Filtration

Fig2d Evaporation using waterbath

Fig 2e Plant extract

Fig: 2f Dilution of extract

d. **ANTIBACTERIAL ASSAY OF *PLECTRANTHUS AMBOINICUS* BY WELL DIFFUSSION METHOD**

The aqueous and alcoholic extract obtained from the *Plectranthus amboinicus* leaves

were subjected to antimicrobial sensitivity test. The Aqueous and alcoholic extraction obtained from the maceration of Plectranthus ambonicus were subjected to Antimicrobial sensitivity test. By well diffusion method, Muller Hinton Agar plate was swabbed with standard McFarland inoculums. Lid was closed for 5 minutes. 6 equidistant wells were made on the plates with the help of sterile cork borer. 100 µl of the Standard (Ampicilline-10µg) and maceration extracts of different concentration (20µg, 10µg, 5µg.2.5µg and 1.25µg) solutions were filled in to the well and labelled accordingly. Plates were incubated in an 37 ºC for 24 hours. After incubation period, the zone of inhibition was measured with a ruler and results obtained were tabulated below (Fig-2g).

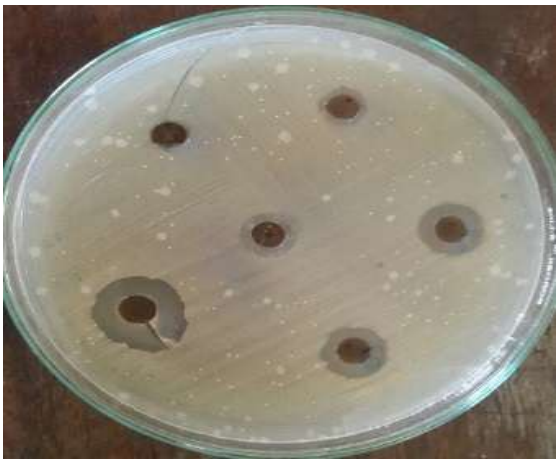


Fig 2g Sensitivity test

RESULTS AND DISSCUSSION

Antibacterial assay:

Aqueous and Alcoholic extraction which is extracted from the *Plectranthus amboinicus* plant revealed the antimicrobial property against multidrug resistant organism such as *E. coli, Klebsiella, Streptococci* spp. and *Staphylococci* spp. With various concentrations. The extract has revealed highly significant antibacterial activity when compared with standard drug (Amp 10µg). Zone of inhibition of 18, 12, 14, 10 mm was observed for different concentrations of 20, 10, 5, 2.5, 1.25 on different organisms.

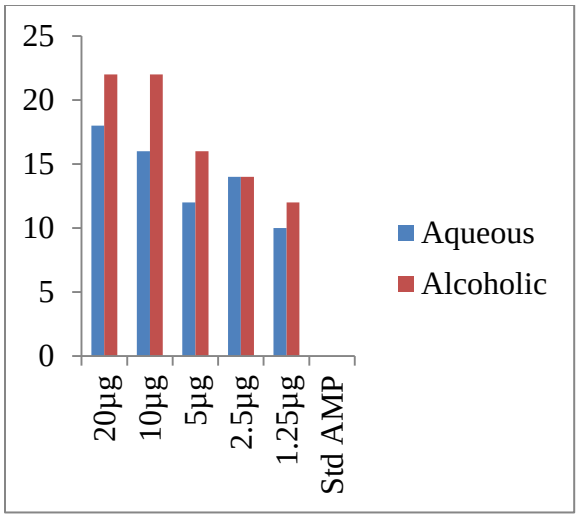
Antibacterial activity against *Streptococci* spp.

Antibacterial assay of aqueous and alcoholic extract showed highly significant zone of inhibition with 22 and 18mm in 20µg concentration and in 1.5µg concentration it had shown 10 and 12mm with respective extract. *Streptococci* spp. were showed resistance to Amp (Table 1). Same has been graphically interpreted (Graph 1).

Table 1 Antibacterial activity against *Streptococci* spp.

Organism	Concentration	Aqueous extract zone of inhibition in mm	Alcoholic extract zone of inhibition in mm
Streptococci spp.	20µg	18	22
	10µg	16	22
	5µg	12	16
	2.5 µg	14	14
	1.25µg	10	12
Standard	Amp(10µg)	R	R

(Note: µg–micro gram mm- millimetre Amp-Ampicillin R- Resistant)



Graph 1 Antibacterial activity against *Streptococci* spp.

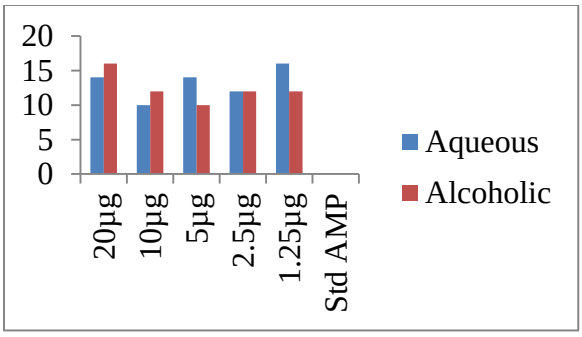
Antibacterial activity against *Staphylococci* spp.

Antibacterial assay of aqueous and alcoholic extract showed highly significant zone of inhibition with 14 and 16mm in 20µg concentration and in 1.5µg concentration it had shown 16and 12mm with respective extract. *Staphylococci spp.* were showed resistance to Amp (Table 2), Same has been graphically interpreted (Graph 2).

Table 2 Antibacterial activity against *Staphylococci* spp.

Organism	**Concentration**	**Aqueous extract zone of inhibition in mm**	**Alcoholic extract zone of inhibition in mm**
Staphylococci spp.	20µg	14	16
	10µg	10	12
	5µg	14	10
	2.5 µg	12	12
	1.25µg	16	12
Standard	Amp(10µg)	R	R

(Note: µg–micro gram mm- millimetre Amp-Ampicillin R- Resistant)



Graph 2 Antibacterial activity against *Staphylococci* spp.

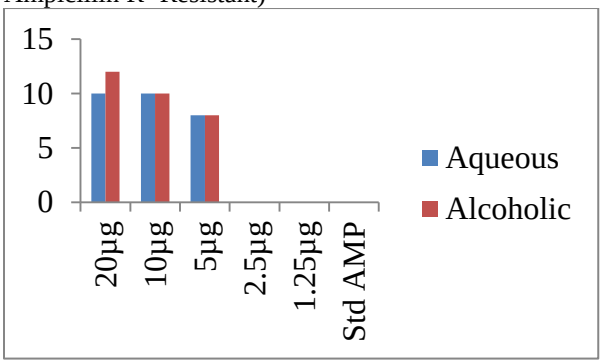
Antibacterial activity against *Klebsiella*

Antibacterial assay of aqueous and alcoholic extract showed highly significant zone of inhibition with 10 and 12mm in 20µg concentration and in 5µg concentration it had shown 8 and 8mm with respective extract. *Klebshiella* Were showed resistance to Amp (Table 3), Same has been graphically interpreted (Graph 3).

Table 3 Antibacterial activity against *Klebsiella* spp

Organism	Concentration	Aqueous extract zone of inhibition in mm	Alcoholic extract zone of inhibition in mm
Klebshiella	20µg	10	12
	10µg	10	10
	5µg	8	8
	2.5 µg	-	-
	1.25µg	-	-
Standard	Amp(10µg)	R	R

(Note: µg–micro gram mm- millimetre Amp-Ampicillin R- Resistant)



Graph 3 Antibacterial activity against *Klebsiella* spp

Antibacterial activity against *E. coli*

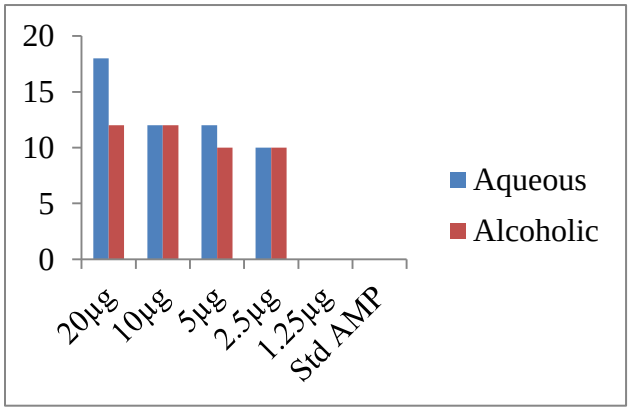
Antibacterial assay of aqueous and alcoholic extract showed highly significant zone of inhibition with 18 and 12mm in 20µg concentration and in 2.5µg concentration it had shown 10 and 10mm with respective extract. *E.coli* Were showed resistance to Amp (Table 4), same has been graphically interpreted (Graph 4).

These findings confirm its traditional medicinal use in the treatment of several infections. The study reveals that the antimicrobial activity of the plant extract is dose dependent.

Table 4 Antibacterial activity against *E.coli*

Organism	Concentration	Aqueous extract zone of inhibition in mm	Alcoholic extract zone of inhibition in mm
E.coli	20µg	18	12
	10µg	12	12
	5µg	12	10
	2.5 µg	10	10
	1.25µg	-	-
Standard	Amp(10µg)	R	R

(Note: µg–micro gram mm- millimetre Amp- Ampicillin R- Resistant)



Graph 4 Antibacterial activity against *E.coli*

CONCLUSION

P. amboinicus is an important aromatic medicinal herb packed with many bioactive constituents and nutrients, which are important for maintaining good health. The plant has shown a wide range of biological properties and proved to be effective in curing respiratory, cardiovascular, oral, skin, digestive and urinary diseases To conclude the study, the extract has demonstrated significant antibacterial activity when compared with standard drug. These actions are exerted through central activity of the plant extract in the respiratory tract. Zone of inhibition of 18,12,14,10 cm was formed for different concentrations of 20,10,5,2.5,1.25µg. These findings confirm its traditional medicinal use in the treatment of several infections. The study reveals that the antimicrobial activity of the plant extract is dose dependent.

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