Research Article

Antibacterial and antifungal prospective of cypress vine (*Ipomoea quamoclit* Linn)

Kushnood Ur Rehman¹, Muhammad Hamayun², Gulzad Ahmad¹, Wali Samad Khan², Sayyar Muhammad³, Rani Gul⁴ and Tabassum Yaseen^{5*}

1. Department of Botany, Islamia Collage Peshawar-Pakistan

2. Department of Botany, Abdul Wali Khan University Mardan, 23200-Pakistan

3. Department of Chemistry, Islamia Collage Peshawar-Pakistan

4. Department of Agriculture, Bacha Khan University Charsadda Khyber Pakhtunkhwa-Pakistan

5. Department of Botany, Bacha Khan University Charsadda Khyber Pakhtunkhwa-Pakistan

*Corresponding author's email: tabassumyaseen@bkuc.edu.pk

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Abstract

The study established out to find the anti-fungal and anti-bacterial action of *Ipomoea quamoclit*. We focus on the study to find fractions, which are active against bacterial and fungal species. The fractions aqueous, chloroform, n-hexane, methanolic extract and ethyl acetate was obtained and were tested against the selected four fungal and four bacterial species which are very common in different hospitals of Khyber Pakhtunkhwa. The n-hexane fractions inhibit the growth of bacterial strains (46-48%) which was the highest inhibition zone and the lowest activity shown by crude methanolic extracts (38-44%) against all the selected bacterial strains. However, in ant-fungal activity, the highest activity shown by crude methanolic extracts (35-65%) against the four selected fungal species and lowest activity displayed by the aqueous fractions. The other fractions also shown some moderate activity but were significant.

Keywords: Antibacterial; Antifungal; Cypress vine (*Ipomoea quamoclit* Linn); Inhibition

Introduction

From the initiation of this world, plants have a prominent role in the civilization of human beings. Some use for ornamental purpose while other for medicinal purpose. In such medicinal plants, *Ipomoea quamoclit* is a native species of America and spread throughout the Europe in 1500s. In both areas, it is studying for medical purposes. The species is reported from some regions of India. From some new reports, it was clear that it is also an ornamental species and people growing it in their homes and gardens. *I. quamoclit* also called morning glory and

member of family Convolvaceae. The climber flowers of the plant form an attractive and heart catching property in horticulture areas. At lower latitude across the world it is common and also touching the Ontario of Canada [1-3].

From many decade plants are either used directly as a traditional medicine or an alternative medicine against several diseases caused by pathogens [4, 5]. Such knowledge spread with the help of conferences or writings across the world [4]. In different countries different traditional centers were open which based on plants like in china,

Chinese medicines of china, Siddha, Unani medicines India and Unani dawakhana in Pakistan [6]. A survey was carried out by World Health Organization that about 80% people across the world depended upon medicinal [5, 7]. The plant medicines are very prominent because they possess either less or no side effect and very active against pathogens [8]. The bioactive substances such as flavonoids, alkaloids, tannins, glycosides, protein, amino acids, phenols and many other secondary metabolites [9]. Many readings were also conducted to find such biologically active substances in medicinal plants which play a very crucial role against disease causing agents but still many plants and their constituents were not known up to now [10]. Therefore different researchers are busy in their findings. In such regards the same study was also carried out to find the antifungal and antibacterial activity of *Ipomoea quamoclit* Linn (Cypress vine).

Material and methods

Collection and processing of plant specimen

Different areas of the Khyber Pakhtunkhwa were visited for the collection of *Ipomoea quamoclit* Linn plant. The collected plants were washed and dried in shadow followed by oven drying in the oven. The completely dried plants were powdered with the help of grinder. Then the powder stored at cool place before extraction.

Preparation of extraction and fractions

Cold maceration method was used for the extraction of the active metabolites. One and half of the powdered plant material was dipped in two liter of ethanol and incubated for 5 days at 40°C. The material filtered thrice and a clear filtrate was obtained. The filtrate was subjected for evaporation via rotatory evaporator at 40°C. The obtained extract was dried and then dissolved in 100ml distilled water. The solution was the fractionate by using different organic solvents including ethanol, chloroform, n-

hexane, methanol and ethyl acetate by using separating funnel. All the fraction thus obtained were concentrated by rotatory evaporator and designate for that solvent fraction.

Media preparation

Agar well diffusion method was used during antibacterial activity. One liter distilled water was to liquefy 25 g of Luria Broth, PH of Miller powder was put at 7.0. The media put in autoclave in 250ml flask. The selected four bacterial strains were introduced into the flask and kept overnight at 150RPM at 37°C. After that agar converted into solid form and by using sterilized borer five holes were tunneled. The inoculum was introduced into the tunneled. The bacterial and fungal species were selected due to their frequent occurrence in local hospitals of Khyber Pakhtunkhwa (KPK) and also showing resistance to different drugs.

Test for bacterial strains

Among the four selected bacterial strains three were gram positive and one strain is gram negative i.e MRSA (*Methicillin resistance staphylococcus aureus*), *streptococcus mutans* and *staphylococcus aureus*. And the gram negative is *Serratia marcescens*.

Measurement of zones of inhibition

Dimethyl sulfoxide 20mg/ml was used as a negative control in which the extracts were dissolved. While the Cefotaxime (standard antibiotics) was used as a positive controller. The plant fraction about 75µl were introduced into the wells of petri dish and that petri dishes were positioned in the incubator at 37°C for twenty four (24) hours. When the incubation period was completed then the diameter of each transparent zone was measured. The experiment was repeated again and again to calculate the standard data.

Test for fungal strains

During antifungal activity the four fungal strains were selected i.e *Fusarium oxysporum*, *Aspergillus flavus*,

polyspondylium pallidum, and *Alternaria alternata*.

Results and discussion

The research study conducted to evaluate the anti-fungal and anti-bacterial potential of the cypress vine plant (*Ipomoea quamoclit* Linn.) plant. The results revealed that this plant is a very effective source for the inhibition of the growth of the tested species of both fungi and bacteria. The plant can be investigated for active metabolites to evolve the anti-bacterial and anti-fungal drugs through biochemical and biophysical essays.

Antibacterial activities of *Ipomoea quamoclit* Linn

In the current research work five fractions of *I. quamoclit* Linn extracts were used to know their potential use against the selected bacterial strains that are *Staphylococcus aureus*, *Streptococcus mutans* and Methicillin-resistant *Staphylococcus aureus* (MRSA) and *Serratia marcescens*. These species were selected for the current experiments on the basis of their frequent pathological reports from hospitals of Khyber Pakhtunkhwa. Results of the anti-fungal activity as shown in (Table 1 and Fig.1) showed that all the fractions were active against the selected bacterial strains. The growth of Methicillin-resistant *Staphylococcus aureus* (MRSA), *Streptococcus mutans*, *Staphylococcus aureus* and *Serratia marcescens* was inhibited by the crude methanolic extract with 10.0mm, 12.0mm, 09.0mm and 11.0mm zones of inhibition respectively. The *n*. hexane extracted fractions inhibited the

growth of *Staphylococcus aureus*, *Streptococcus mutans*, *Serratia marcescens* and Methicillin-resistant *Staphylococcus aureus* (MRSA) with 12.0mm, 13.0mm, 14.0mm and 10.0mm zones of inhibition respectively. The chloroform fractions were produced 11.0mm, 14.0mm, 07.0mm and 14.0mm zones of inhibition against *Staphylococcus aureus*, *Streptococcus mutans*, *Serratia marcescens* and Methicillin-resistant *Staphylococcus aureus* (MRSA) respectively. The ethyl acetate fraction was most active against *Streptococcus mutans* (ZOI=10.0mm) followed by Methicillin-resistant *Staphylococcus aureus* (ZOI=09.0mm), *Staphylococcus aureus* (ZOI=08.0mm) and *Serratia marcescens* (ZOI=08.0mm). The aqueous extracted fraction inhibited the growth of tested bacterial strains in the order of Methicillin-resistant *Staphylococcus aureus* (ZOI=08.0mm), *Staphylococcus aureus* (ZOI=05.0mm), *Serratia marcescens* (ZOI=05.0mm) and *Streptococcus mutans* (ZOI=04.0mm) respectively. Globally several hundred genera of medicinal plants are used as the main sources of anti-bacterial drugs [11, 12]. According to [13] in the previous century most of the faculties in the medicine sector switched their concern from natural to synthetic drugs. But for the last few decades this trend is shifting in the reverse direction [14, 15]. In a survey 25% of the drug prescriptions in 35 countries are plants derived in non-modified or little modified [16-19].

Table 1. Antibacterial activity of the crude extracts of *Ipomoea quamoclit*

Anova: Two-Factor Antibacterial						
Summary	Count	Sum	Average	Variance		
MRSA	6	87	14.5	70.3		
<i>S. marcescens</i>	6	76	12.66667	69.46667		
<i>S. mutans</i>	6	82	13.66667	82.66667		
<i>S. aureus</i>	6	92	15.33333	126.6667		
Standard	4	123	30.75	21.58333		
Cr. Met. Ext	4	54	13.5	4.333333		
n- hexane	4	41	10.25	2.916667		
CHCl3	4	39	9.75	4.916667		
EtOAc	4	63	15.75	4.916667		
Aqueous	4	17	4.25	2.916667		
ANOVA						
Source of variation	SS	df	MS	F	P-value	F crit
Rows	23.45833	3	7.819444	1.15796	0.358338	3.287382
Columns	1644.208	5	328.8417	48.69724	9.6E-09	2.901295
Error	101.2917	15	6.752778	-	-	-
Total	1768.958	23		-	-	-

Figure 1. Antibacterial efficacy of *Ipomoea quamoclit* Linn**Antifungal activities of *Ipomoea quamoclit* Linn**

Effects of the anti-fungal activity as shown in (Table 2 & Fig. 2) showed that the n-hexane extracted sample inhibited the growth of

Fusarium oxysporum completely. Chloroform and Ethyl acetate inhibited the growth of *Polysphondylium pallidum*. While the aqueous extract showed significant activity against *Aspergillus flavus* and

Alternaria alternate. Crude methanolic extract showed good results against *Polysphondylium pallidum*. Trend in results revealed that *Fusarium oxysporum* is comparatively more sensible to the n-hexane extracts, *Polysphondylium pallidum* to chloroform and Ethyl acetate extracts while *Aspergillus flavus* and *Alternaria alternate* to the aqueous extracts of plants. The usage of medicinal plants as anti-fungals is quite old practice performed by human beings. These result in the production of important compounds that are used to develop drugs against fungal pathogens. One of the positive aspects of these drugs is this that these drugs have very little adverse/side effects on the human health [5, 20]. The demand for the development anti-fungal drugs has enormously increased as it is established that among others fungal pathogens are causing serious losses in the quantity, quality, shelf life and mortality of the crop plants [21]. In

regard to humans, fungi as the cause of diseases offer a new aspect of human pathology [22]. In this regard medicinal plants are proved to be the sources for the discovery of new drugs [23, 24]. In the developing countries, which face the problems of the quantity and quality of modern drugs, medicinal plants derived compounds have recently gained much importance and their old traditional uses are valued much more nowadays [25]. The current study revealed that antifungal activity of the wild plants proved to be much more significant and can be a very suitable substitute of the modern medicines. Similar findings have been reported by [26], for the fungus *Alternaria alternate*. Similarly [6] publicized that the crude methanolic extract of the medicinal plants showed maximum anti-fungal properties which is in agreement with our work. Similar results with other medicinal plants were reported by [27, 28].

Table 2. Two factor ANOVA of antifungal

Anova: Two-Factor antifungal						
Summary	Count	Sum	Average	Variance		
A.flavus	6	220	36.66667	1129.067		
A. alternate	6	209	34.83333	1286.167		
F. oxysporum	6	234	39	1033.2		
P. pallidum	6	243	40.5	908.3		
Standard	4	400	100	0		
Cr. Met. Ext	4	141	35.25	9.583333		
n- hexane	4	76	19	88.66667		
CHCl3	4	88	22	60		
EtOAc	4	158	39.5	25		
Aqueous	4	43	10.75	164.9167		
ANOVA						
Source of variation	SS	df	MS	F	P-value	F crit
Rows	112.8333	3	37.61111	0.605546	0.621516	3.287382
Columns	20852	5	4170.4	67.14419	9.94E-10	2.901295
Error	931.6667	15	62.11111	-	-	-
Total	21896.5	23		-	-	-

Figure 2. Antifungal efficacy

Conclusion

It is evident from the results of anti-bacterial and anti-fungal activities that the selected plant i.e. *Ipomoea quamoclit* Linn possessed very significant properties. The crude methanolic and ethanolic.

Authors' contributions

Conceived and designed the experiments: KU Rehman, G Ahmad, WS Khan & S Muhammad, Performed the experiments: G Ahmad, WS Khan & S Muhammad, Analyzed the data: KU Rehman & M Hamayun, Contributed materials/ analysis/ tools: R Gul & T Yaseen, Wrote the paper: KU Rehman.

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