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Phytochemical profiling and antibacterial potential of *Peonia emodi* Extract

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Peonia emodi, a native plant species found in the Swat and Shangla districts, was investigated in this study to evaluate its phytochemical composition and antibacterial potential. Various parts of the plant were subjected to extraction, and the resulting extracts were analyzed using spectroscopic techniques. The analysis unveiled the presence of alkaloids, phenols, and sterols as the primary phytochemical constituents, with the highest concentration observed in the root of Peonia emodi. Subsequently, antibacterial tests were conducted to assess the efficacy of Peonia emodi extracts against Gram-positive bacterial strains. The results demonstrated that aqueous extracts of Peonia emodi exhibited remarkable antibacterial activity against the tested Gram-positive bacterial strains. Conversely, the ethanolic extract showed no inhibitory activity against these bacteria. This comprehensive study highlights the potential of Peonia emodi as a valuable source of bioactive compounds with significant antibacterial properties, particularly in its aqueous extracts. These findings provide promising insights into the medicinal and pharmaceutical applications of Peonia emodi and underscore the importance of further research to harness its therapeutic potential.

Keywords: **Antibacterial, Antifungal, Gram positive, pesticides, herbicide.**

INTRODUCTION

The relationship between plants and humans is as old as the relationship between human civilization, like Indian, Chinese, Roman and Greek civilization. These civilizations are known for many and many different purposes (Ahmad & Sher, 2004; Brantner & Grein, 1994; Ghosh et al., 2008; Nascimento et al., 2000). Approximately in the whole universe there exists 4, 22,127 plants in which 35,000-70,000 species have medicinal uses. The Pakistani floor contains 6000 Types of flowering plants including 500550 for medical use. Have been used. Many of them are available in Swat and Malakand territory. *Scino-Japanese* is very rich in biodiversity. 6.16% of Pakistani flora contain medicinal species (Aw, 1966; Baltekin et al., 2017; Khan et al., 2019; Roth et al., 1997). Plants create organic compounds that are not directly related to the primary functions of normal growth, and reproduction called secondary metabolism They are commonly used for biological purposes in biology. This type of metabolite is commonly found in many organisms or cells (Agbor et al., 2011; Khan et al., 2005: Alpinar et al., 2009; Marwah et al., 2007). All members in genus Paeonia are perennial shrubs or herbs, most of them are herbs with tallness 0.82-3.28 feet and some of them are woody shrubs having height 0.82-11.48. They have thick and storage root for storing water and minerals, some members are tufted due to crown make adventitious buds while other have stolon's while leaves are the largest compounds and alternate, deeply lobed having no glands and stipule and have anomocytic stomata (Ahmad & Sher, 2004). The genus Paeoniais common and endemic to temperate, cold region occurred in five regions in north round about half of sphere all over the world including Central Asia, Eastern Asia, the west Himalaya, Mediterranean and North America near to the "Pacific Ocean". Most of the species are found in Europe and Asia_ two are confined to western North America (Krause & Bedard, 2008). About more than 262 compounds have been isolated from Paeoniaceae. They are classified

into seven classes including mono terpenoid glucoside, flavonoid, tannins, stilbenoids, tri terpenoids, steroids, paeonols and phenols (Ekren et al., 2013).

MATERIALS AND METHODS

The whole process consists of three stages. The first step involves the selection, identification and collection of plants. The second step involves the extraction, separation and separation of the material from the electrode. In the third step, the application of wavelengths was demonstrated by various methods, such as UV visible, IC and NMR spectroscopy.

Material Used.

In this study, the *Peonia emodi* plant was selected from their natural habitat. Commercial classification is used to isolate acetone, CH_3OH, N-hexane, $CHCl_3$, n-butanol, diethylamine, distilled water and ethyl acetate solvents. Rotary steam (Bochi R210), separate fireplace, bakers, glasses, reagent bottles, tripod for iron, measuring cylinder, dummy, Petri dish, digital scale, chain plate, column, TLC card (pre-coated silica gel), mill, UV lamp and driver was used for various purposes.

Culturing and Activities Processing.

Antibacterial activity and culturing of Bacteria.

Bacterial microorganisms were worthy in nutrient media. 28 mg of nutrients were dissolved in 900 ml of distilled water. The medium was then stirred for 15 minutes. After sterilization, the medium was then transferred to an autoclave with a Petri dish, approximately 12 psi at high pressure and for up to 50 minutes. After the sterilization process, the laminar flow chamber was washed and cleaned with methylated alcohol, and then under UV light for 7 minutes. Then the media were placed in a patriotic dish to become tough and hard. Then the bacterial spot was applied to a solid carrier using a stick. Then a 5 mm sterile filter disk was placed on each petri dish. 25 µL of the prepared sample was transferred to a filter disk. Positive controls (ciprofloxacin as antibiotics) were used as standard, while negative dimethyl sulfoxide (DMSO). Dietal sulfoxide was allowed to stand for some time until it was completely dispersed. The plate (Petri dish) was incubated at 37 ° C for 24 hours. After incubation time, the incubation zone was measured in millimeters with the help of a ruler. On average all sides were confirmed. Antibacterial activity mechanism was reported by (Bauer, AW et al., 1966)

Antifungal activity and culturing of Fungi.

Potato dextrose agar (PDA) was used as media for antifungal activity. 28 grams of PDA dissolved in 900 mL distilled water. Then dissolve the solution for 14 minutes, stirring thoroughly. The petri dish and media were then transferred to Autoclave for sterilization, with high pressure at 15 psi and high temperature at 121 ºC for 46 minutes. After the sterilization process, the Laminar Gash cabinet was cleaned for some time to kill the microbes (germs) by foundation and ethanol and UV light. The patri dish was completely dry. Then the media was cooled down a bit. The media was poured into a 20 ml Petri dish and solidified. Decent cookies were pulled onto solid media with sterile shrubs, and then sterile filter disks were used on the media. 25 µL of the prepared sample was transferred to a filter disk. The positive control fluconazole was used on average, while the negative control was dimethyl sulfoxide, which contained each sample on a separate filter disk in the petri dish. The petri dish (plate) was baked at 37 ° C for 48 hours. After the incubation phase, the injection zones were calculated in millimeters with the help of a ruler and average cost documents were collected from each side.

RESULTSAND DISCUSSION

In the pharmaceutical industry, plants are recognized for their wide variety of structural diversity and wide range of pharmaceutical activities. Biologically active compounds present in plants are called phytochemicals. It is derived from various parts of the phytochemical plant such as leaves, saplings, seeds, seed coat, flowers, roots and mattresses and is used directly as a source of pharmaceutical agents. Numerous plants, squeeze them or purify them. The active ingredients can act as allelo chemicals in other plants and thus may be candidates for application for agricultural purposes.

Phytochemistry describes a number of secondary metabolic compounds present in plants. Plants are a repository of naturally occurring chemical compounds and structurally diverse bioactive molecules. Depending on the biotic compounds from plants and their quantitative and qualitative estimation for the detection of new biomolecules, they may be used directly by the pharmaceutical and agrochemical industry or as lead molecules for the synthesis of more powerful molecules.

Antibacterial activities Gram positive and Gram-negative:

Antibacterial (Gram positive) activities.

Pakistan, like most developing countries, has a strong tradition of treating herbs. Its rural population is still dependent on the indigenous system of medicine for their health issues. Therefore, it was interesting to conduct a scientific review and to determine the utility of the aerial parts of a medicinal plant, which is commonly used by traditional practitioners. We present, in the present study, a screening of airborne parts of this plant for in vitro biological activities including anti-fungal, anti-bacterial and antiviral activities. The extracts of the plants were mostly effective against bacterial and fungal species, and some showed the highest activity. The aqueous extracts of *Parania amodi* showed excellent activity against Gram-positive bacterial strains, eg, *Bacillus subtilis, Enterococcus faecalis, Streptococcus pyogenes, Staphylococcus aureus, Staphylococcus epidermidis,* whereas Asiatic activity against acetate. Great activity shown. *Paranium emodi's* unknown and chloroform extract showed zero results against gram-positive strains, while chloroform and unknown compounds showed the lowest activity against

gram-positive bacteria, as shown in Table 1. Plants contain various nutrients, anti-nutrients, and other secondary metabolites, which are known to play important roles in their defense mechanisms. These compounds are also reported as a potential source of antioxidants and anti-microbial agents.

The results obtained from the present study indicate that *P. amiodi* can be useful as a natural herbicide and can be a source of biotic agrochemicals results are given in table 1.

Table 1: Activities results for different organisms.

S. No	Organisms name	Unknown	Ethyl acetate C4H8O2	Chloroform CHCl3	WATER
01	MRSA	_	_	_	_
02	Bacillus subtilis	_	_	_	+
03	Enterococcus faecalis	_	+	_	+
04	Streptococcus pyogenes	_	+	_	+
05	Staphylococcus aureus	_	_	_	+
06	Staphylococcus epidermidis	_	_	_	+

Antibacterial (Gram positive) activities.

Because modern drugs are expensive in developing countries, the antimicrobial activity of ethno-medicinal plants is still valid. They will find the way to antimicrobial weapons recommended by phytochemical therapists. It has been found that the use of hexane, ethanol and chloroform as solvents is more effective in removing active compounds. For Gram-negative bacteria, *Pavonia amodi* root and root, ethanol extract proved to be the most effective as shown in the table 2. The most effective of Amody's water supply was against *Kudosila pneumonia, Escherichia coli, Salmonella enterica, Enterobacter aerogens, Proteus. Mirabilis, Shigella bodi, Salmonella paratyphi A, Seritia marcescens, Shigella sonnei, Salmonella tethe and Pseudomonas aeruginosa,* respectively. Ethyl acetate extract in our study is also effective against specific strains of Gram-negative bacteria, such as *Klebsiella pneumoniae, Salmonella enterica, Shigella bidi and Pseudomonas aeruginosa.* While chloroform and unknown compounds have no effective results against Gram-positive bacteria in our study. Extensive study was done by it to obtain the methanolic extract from whole plants of *Pinea amodi* tested in vitro biological activities, including anti-fungal, anti-bacterial and pesticides. Aconitum accumulates and it experiences anti-inflammatory, anti-oxidant activity.

Table 2: Antibacterial (Gram positive) activities.

S. No	Organisms	UNKNOWN	Ethyl acetate C4H8O2	Chloroform CHCl3	WATER
01	Klebsiella pneumoniae	_	+	_	+
02	Escherichia coli	_	_	_	+
03	Salmonella enterica	_	+	_	+
04	Enterobactor aerogenes	_	_	_	+
05	Proteus mirabilis	_	_	_	+
06	Shigella boydii	_	+	_	+
07	Salmonella paratyphi A	_	_	_	+
08	Serratia marcescens	_	_	_	+
09	Shigella sonnei	_	_	_	+
10	Salmonella typhi	_	_	_	+
11	Pseudomonas aeruginosa	_	+		_

Antifungal Activities.

Ethnic extract extracted from the aerobic part of *P. amodi* was found to be effective in a variety of activities, including anti-fungal, anti-bacterial, pesticide, phytotoxic, and hemagglutination activities. *Paranoia amodi* root and root bark, ethanolic extract, did not find any inhibitory activity against cookies, as shown in Table 3. All four excerpts from amodia have no significant effect on fungal species, but water extracts have shown positive effects against *A. flavus* species. Nowadays, there has been an increase in public demand for more eco-friendly methods so as not to spoil food and feeding cookies. The goal of our study was to explore the anti-fungal potential of *ranonculisi* in different regions. It can be used for the biological control of cookies. This may be due to the fact that plant sources contain many chemical compounds besides vitamin C, E and Carotenoids and their synergistic effects may increase antioxidant capacity. It was found that the ethnic extract from *P. amodi* leaves showed excellent (83%) radical warping activity using the DPPH test.

figure 1, shows comparable activity in different solvents, such as ethyl acetate, chloroform, water, comparison shows that this activity is higher for a given microbes than for ethyl acetate and chloroform. Roots and rhizomes are used for the treatment of back pain, irritation and epilepsy, and are also used as tonic, emetic, laxative, cleansing blood and colic, while the seeds are cleansing. The roots are used to help with headaches, dizziness, vomiting and pregnancy.

Table 3: Antifungal Activities.

S.NO	Organisms	UNKNOWN	Ethyl acetate C4H8O2	Chloroform CHCl3	WATER
01	A. flavus	–	–	–	+
02	A. niger	–	–	–	–
03	A. terreus	–	–	–	–
04	C. albicans	–	–	–	–
05	C. kruseii	–	–	–	–
06	C. kefyr	–	–	–	–
07	C. tropicalis	–	–	–	–
08	Crptococcal neoformans	–	–	–	–
09	Rhizomucor	–	–	–	–

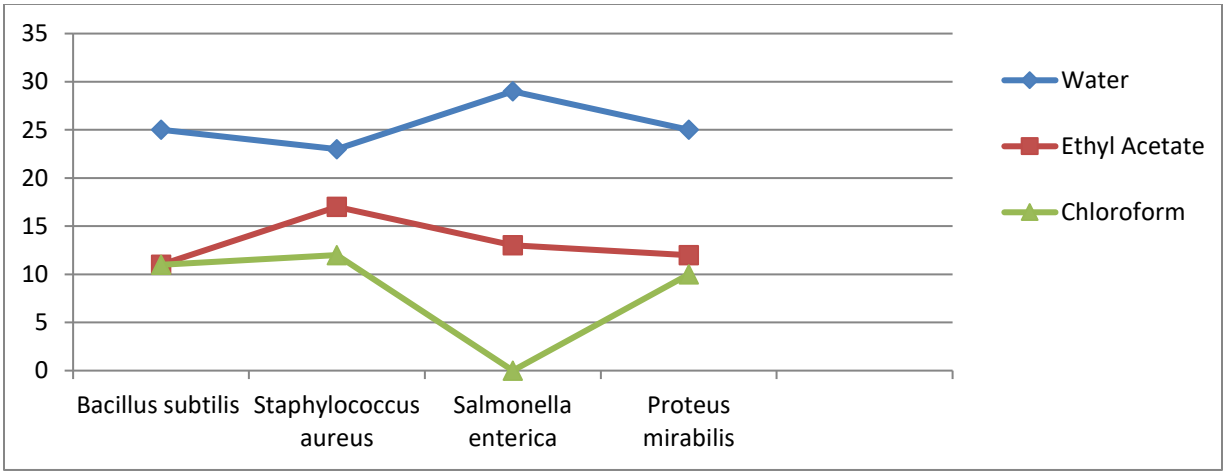

Figure 1: Shows comparable activity in different solvents, such as ethyl acetate, chloroform, water,

Minimal inhibitory concentration.

The antibacterial activity of the extracts obtained from the plant under the study shown in the table 4. 6 Gram +ve bacteria and 11 Gram –ve were used as a test organism. The test sample that contain antibacterial agents inhibited the growth of bacterial stain producing a zone of inhibition. According to our study the unknown compound extracted from *p. emodi* show no growth inhibition for all the stain of Gram +ve and G –ve bacteria except from *Bacillus subtilis.* So, the unknown compound is considered the minimal inhibitory concentration for most of the bacteria. Chloroform extracts also show a minimal inhibitory effect against the different strain of bacteria, ie, except from *Bacillus subtilis and Staphylococcus aureus* of Gram +ve stain and *Proteus mirabilis and Shigella sonnei* of Gram –ve stain. The compound ethyl acetate has a maximum inhibitory concentration against Gram +ve stain, while it can also inhibit the growth of most of the Gram –ve stain except from the *Salmonella paratyphi A* and *Serratia.*

Table 3: Minimal inhibitory concentration

Serial NO.	Organisms name	UNKNOWN (1)	Ethyl acetate C4H8O2 (2)	Chloroform CHCl3 (3)	WATER (4)
	Gram +ve				
01	MRSA	-	100	-	50
02	***Bacillus subtilis***	200	200	200	50
03	***Enterococcus faecalis***	-	200	-	100
04	***Streptococcus pyogenes***	-	200	-	100
05	***Staphylococcus aureus***	-	50	200	100
06	***Staphylococcus epidermidis***	-	200	-	50
	Gram -ve				
07	***Klebsiella pneumoniae***	-	50	-	100
08	***Escherichia coli***	-	200	-	100
09	***Salmonella enterica***	-	200	-	25
10	***Enterobactor aerogenes***	-	200	-	100
11	***Proteus mirabilis***	-	200	200	100
12	***Shigella boydii***	-	50	-	100
13	***Salmonella paratyphi A***	-	-	-	100
14	***Serratia marcescens***	-	-	-	100
15	***Shigella sonnei***	-	200	200	50
16	***Salmonella typhi***	-	200	-	50
17	***Pseudomonas aeruginosa***	-	15	-	100

DISCUSSION:

Peonia distributed in China were qualitatively and quantitatively analyzed by high performance liquid chromatography - diode array detection (HPLC-DAD) and HPLC-Q-TOF-MS. A total of 21 metabolites were isolated and identified, including monoterpene glycosides, phenols, paeonols, tannins, and flavonoids. Our results show that the composition and content of secondary metabolites in the roots of these species were different. In some species, the main bioactive metabolites (monoterpene glycosides, paeonols, and tannins) in the roots of *Paeonia* extracts were similar to or even higher than those of *P. lactiflora* and *P. anomala* subsp. *veitchii*, which can be used as potential germplasm resources for extracting specific drugs (Yang et al., 2020). It has been listed as a critically endangered plant species. and *P. emodi* has been shown to have antioxidant, nephroprotective, lipoxygenase inhibitory, cognition and oxidative stress release, cytotoxic, anti-inflammatory, antiepileptic, anticonvulsant, haemaglutination, alphachymotrypsin inhibitory, hepatoprotective, hepatic chromes, and pharmacokinetics of carbamazepine expression (Kumar et al., 2023). Inhibitors of Nitric Oxide Production, three compounds isolated from *Paeonia* showed significantly suppressed nitric oxide production (Ding et al., 2012). A few of the compounds showed significant cytotoxicity against a panel of human cancer cell lines, against MCF-7, HT-29, M-14 (Ambrož et al 2016). *Paeonia emodi* ethanol extracts block the growing size of ordinary duckweeds, and were mildly

active to kill the flour's red beetles. No blockage of the growing size of bacterium and fungus should be established and no common poisonous was examine in brine shrimp, mention that the use is may be safe (Denzler et al., 2010).

CONCLUSION

The roots of *paunia emodi* were extracted with solvents from different fields (i.e., hexane, chloroform, ethyl acetate, methanol). All the extracts were screened for their respective phytochemical profiles and their bacterial activity. Phytochemical screening revealed the presence of alkaloids, phenols and sterols present at the root of the *paranoia emodi* plant. These different crude extracts were analyzed in vitro for anti-bacterial activity. The highest activity of the root system was shown by ethyl acetate and water. Various Peoniamia raw materials were tested for antifungal and antibacterial activity by disinfection. These different crude extracts were used against different Gram and Gram + bacteria. Water and ethylene acetate were high in activity against *Bacillus subtilis, Enterococcus faecalis, Streptococcus pyogenes, Staphylococcus aureus, Staphylococcus epidermidis.* Different raw periods of *Paranium emodi* were used against different fungal species, such as *A. flavus, A. Terios, C. albicans, C. Croceae, C. Kieffer, C. Tropicals, Cryptococcal neoformans, Rhizomacor*. *A. flavos* show a positive activity in the middle of the water while the other species show negative effects. This means that the plants are *Paeonia amodi*.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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AUTHOR CONTRIBUTION

SUR and NA presented the idea, Both SUR and NA supervised the work, SK did the experiment, AK and IU wrote the manuscript. IU and PI reviewed and reform the manuscript. All authors read and approved the final version.

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