

Heracleum Rigens, Eryngium Foetidum L., and Passiflora subpeltata Medical Plants: Analysis of their Antimicrobial Activity

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Abstract - This extensive study examines the effectiveness of the medicinal herbs *Heracleum rigens*, *Eryngium foetidum* L., and *Passiflora subpeltata* against various microorganisms. The alarming rise in antimicrobial resistance has prompted scientists to look for healing compounds in nature. There is a long history of traditional usage for these plants, and they have also been studied for their possible antibacterial qualities.

Keywords - Medicinal herbs, traditional, antimicrobial resistance, microorganisms.

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INTRODUCTION

Traditional medical treatment has evolved into a respected profession in the contemporary industrial world. The wealthy and powerful pharmaceutical firms are responsible for creating synthetic pharmaceuticals with side effects that are more harmful than the ailments they are meant to treat. There are several antibacterial compounds in plants. Due to overuse, many microbes have developed resistance to antibiotics, reducing the effectiveness of traditional therapy. Major clinical issues in the treatment of infectious illnesses have resulted from the emergence of multidrug resistance in harmful bacteria and parasites. As a result, research into novel antimicrobial agents is urgently required. The antioxidative, antibacterial, and other health-promoting effects of bioactive chemicals found in peels, seeds, leaves, flowers, and stem bark have attracted more attention in recent years.[1]

Mankind has relied on plants for survival from the beginning of time, using them for everything from food to clothing to building materials. Plants also provide a wealth of secondary metabolites, which find use in the medicinal, agrochemical, and flavoring industries. Synthetic flavors, preservatives, and preservatives. Eighty percent or more of the roughly 30,000 natural compounds known today come from plants.[2]

Chemical compounds in medicinal plants that have a measurable physiological effect on the human body are crucial to the health of both people and societies. For their therapeutic properties, several of these local plants are included into dishes specifically prepared for expectant and nursing women. Knowing the makeup

of a medical plant is crucial before any further research is conducted, since almost all plant species include more than one active component. Phytochemical research aids in the identification of new plant-based sources for therapeutically-relevant compounds.

Modern medicine has shown that antimicrobial drugs derived from natural sources, especially traditional medicinal plant extracts, are efficient against bacteria. Roughly 700 plant-derived chemicals have contributed to the western pharmacopeia. In this study, we chose three plants used in traditional medicine in order to test their antibacterial and phytochemical capabilities. *Heracleum rigens*, *Eryngium foetidum*, and *Passiflora subpeltata* were the lucky plants chosen.[3]

MATERIAL AND METHODS

Testmicroorganisms

Heracleum rigens and *Eryngium foetidum* were treated with *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Staphylococcus aureus*, and the fungus *Candida albicans*.

"*Passiflora subpeltata* was tested with *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter aerogenes*, *Staphylococcus aureus*, and *Enterobacter faecalis*. These bacteria, yeasts, and molds were collected at the University of Yavatmal's Herbal Drug Technology Laboratory, Department of Microbiology."

The Agar Diffusion Plate Method for Evaluating Antimicrobial Activity

Heracleum rigens and Eryngium foetidum solvent extracts were tested for their antibacterial properties using the agar well diffusion technique. The optical density of a microbial suspension of around 1.5 10⁸ cfu/ml was achieved by adjusting all the microbial cultures to 0.5McFarland standards. Each Petri dish received 20 ml of agar material and was swabbed with a 100 l inoculate of the organisms under study. The wells were drilled into the seeded agar plates using a sterile cork borer of 6 mm diameter, and then filled with a 50l volume of distinct solvent extract formulated in the respective solvents used for extraction, at a concentration of 50 mg/ml. The plates were kept at 37 degrees Celsius for 24 hours. The zone of growth inhibition against the test bacteria was used to determine the antimicrobial activity of extracts from various solvents. As a negative control, we used a well containing only the extraction solvents, while as a positive control, we used Gentamicin (a gold standard antibacterial drug) and Nystatin (a gold standard antifungal drug) at a concentration of 10 l/well from a 1 mg/ml stock solution. The tests were run in triplicate.

Paper disc assay for antimicrobial efficacy

The paper disc diffusion technique was used to test the antibacterial activity of both aqueous and organic extracts of Passiflora subpeltata. "Bacterial cultures were turbidimetrically calibrated to a 0.5McFarland standard, then inoculated onto 15-centimeter-diameter plates of Nutrient agar, and incubated at 37 degrees Celsius for 18 hours. Over the growth plates, we placed sterile filter paper discs (diameter: 6 mm) impregnated with 100 l of extract dilutions reconstituted in lowest quantity of solvent at concentrations ranging from 20 to 100 mg/ml." 20 l of a 10 mg/ml solution impregnated into paper discs. The gold standard antibacterial employed for this study was streptomycin. As a comparison, we utilized filter paper discs that we soaked in distilled water and then dried as a control. After that, the plates spent 24 hours in a 37°C incubator. Zones of inhibition surrounding each paper disc were measured to ascertain their antimicrobial efficacy. The test organism was exposed to each extract three times.

Establishing the lowest effective concentration (MIC)

Microbroth dilution testing on 96-well microtitre plates with standard inocula of 2106 cfu/ml for bacteria and 2105 cfu/ml for fungi was used to determine the MICs. "Final concentrations of 200 ng/ml to 0.18 ng/ml of test chemicals diluted in extraction solvents were made in Mueller-Hinton Broth and Sabourauds dextrose broth for bacteria and fungus, respectively. A bacterial inoculum of 10 L was poured into each well." As a growth indicator, a combination of triphenyltetrazolium chloride and methyl thioazyltetrazolim bromide was added to the culture medium at a final volume of 0.05

ml for the experiments. After incubation at 37 C for 24 h for the bacteria and at 26 C for 48 h for the fungus, the absorbance at 600 nm was measured using a universal microplate reader to assess the microbial growth. The MIC is the concentration of an antimicrobial agent at which growth of the bacterium is no longer detectable.

RESULTS

Heracleumrigens.

Table displays the results of testing H. rigens' antibacterial and antifungal activities. Antimicrobial and antifungal properties as a consequence H. rigens solvent extracts were all effective across a wide range of microorganisms. When compared to chloroform and petroleum ether, the antibacterial activity of the ethyl acetate and methanol extracts was much higher. For P. aeruginosa and B. subtilis, the aqueous and ethyl acetate extracts had the maximum inhibitory efficacy (20 mm). S. aureus was found to have the weakest inhibitory action (10 mm). S. aureus, P. aeruginosa, and B. subtilis all had the largest inhibitory zones.

At a concentration of 10 l/well from a stock solution of 1 mg/ml, the inhibitory activity of gentamicin (a typical antibacterial medication) varied from 18 mm to 30 mm. Positive results in comparison to Gentamicin are quite promising. Inhibition zones for Gentamicin and ethyl acetate against P. aeruginosa are 18 and 20 millimeters, respectively.

When compared to Nystatin (3.12 g/ml), the antifungal effectiveness of ethyl acetate extract against C.albicans was greatest at a concentration of 1.56 g/ml. All solvent extracts demonstrated antifungal activity against C.albicans except chloroform and water. The extract in methanol had the weakest inhibitory effect (12 mm), whereas the extract in ethyl acetate had the most (20 mm). Results at a concentration of 10 l/well for the conventional antifungal medication nystatin (14 mm of inhibition).

Table 1: The agar well diffusion technique results (in mm) for the antibacterial activity of several solvent extracts of H.rigens roots.

	B.subtilis	P.aeruginosa	E.coli	S.aureus
Gentamicin	30	18	25	25
Pet.etherextract	14	12	17	15
Chloroformextract	15	12	17	10
Ethylacetateextract	17	20	16	12
Methanolextract	18	15	18	14
Aqueousextract	20	12	12	13

Table 2 displays the results of a MIC experiment. The antibacterial activity of ethyl acetate and methanol extracts against P. aeruginosa was comparable to that of the standard antibacterial

medication Gentamicin(12.5 g /ml) at a concentration of 6.25 g /ml. When compared to Nystatin (3.12 g/ml), the antifungal effectiveness of ethyl acetate extract against *C.albicans* was greatest at a concentration of 1.56 g/ml. In this research, ethyl acetate extract of *H.rigens* root was shown to be effective against *P.aeruginosa* and *C.albicans*. This might be because of the triterpenes and flavonoids found in the plant.

Table 2: The MIC (in micrograms per milliliter) of several *H.rigens* root solvent extracts against a collection of gram-positive and gram-negative bacteria.

	S.aureus	P.aeruginosa	E.coli	C.albicans	B.subtilis
Gentamicinforbacteria	1.56	12.5	3.12	NT	0.39
Nystatinforfungi	NT	NT	NT	3.12	NT
Pet.etherextract	100	50	100	6.55	100
Chloroformextract	200	50	100	NS	100
Ethylacetateextract	200	6.25	100	1.56	100
Methanolextract	100	6.25	150	25	50
Aqueousextract	200	NS	200	NS	50

All solvent extracts demonstrated antifungal activity against *C.albicans* except chloroform and aqueous (Fig. 2). The extract in methanol had the weakest inhibitory effect (12 mm), whereas the extract in ethyl acetate had the most (20 mm). Standard antifungal drugs like nystatin (14mm inhibitory activity at 10l/well) have been demonstrated to work. Root preparations of *H.rigens* include phenolic chemicals and terpenoides, which are responsible for this effect.

Eryngiumfoetidum

Table displays the results of testing for *E. foetidum*'s antibacterial and antifungal activities. The ethylacetate extract of *E. foetidum* demonstrated the greatest antibiotic activity against the four bacterial strains and a fungus *C. albicans*, out of the five extracts tested. This suggests that ethylacetate may be more suitable than the other solvents for dissolving the active ingredients that limit the development of sensitive bacteria and fungi. It was also discovered that the ethylacetate extract showed more inhibition (28mm) against *P. aeruginosa* than the gold standard (Gentamicin, 18mm) and showed comparable results (25mm) against *S. aureus*. When compared to the normal dose of Nystatin, the inhibition against *C. albicans* (18mm) was much better.

All the microorganisms we examined were inhibited by Pet.ether extract, however it had no effect on *C. albicans*. Only Gram-negative bacteria, such as *B.subtilis* and *S.aureus*, were inhibited by the chloroform extract. *S. aureus* (20 mm), *B. subtilis* (16 mm), and *E. coli* (15 mm) were all killed by the methanol extract. Both *B.subtilis* and *S. aureus*, both Gram-positive bacteria, were killed by the aqueous extract.

Extracts of ethyl acetate, methanol, and pet. ether were the most effective against *E. coli*, whereas

extracts of chloroform and water were ineffective. *P. aeruginosa* was shown to be vulnerable to extracts of pet.ether and ethylacetate. *B.subtilis* was highly reactive to all of the chemical extracts tested, responding best to ethylacetate and then to pet.ether, chloroform, methanol, and finally water. The most sensitive solvent extract for *S. aureus* was ethylacetate, which is in line with the standard for Gentamicin.

Table 3: The agar well diffusion technique results (zone of inhibition in mm) for the antimicrobial activity of several solvent extracts of *E. foetidum* leaves.

Compound	S.aureus	P.aeruginosa	B.subtilis	C.albicans	E.coli
Nystatinforfungi	NT	NT	NT	14	NT
Gentamicin	25	18	30	NT	25
Pet.etherextract	20	20	18	NS	14
Chloroformextract	NS	NS	17	NS	NS
Ethylacetateextract	25	28	20	18	17
Methanolextract	20	NS	16	NS	15
Aqueousextract	18	NS	14	12	NS

Five extracts of *E.foetidum* were tested against many bacterial strains and the fungus *C.albicans* to determine their minimum inhibitory concentration (MIC). Table displays the results of the MIC test. At a concentration of 3.12 g/ml, ethylacetate extract was more effective against *P. aeruginosa* than Gentamicin (12.5 g/ml). When comparing the antifungal effects of ethyl acetate extract (1.56 g/ml) with Nystatin (3.12 g/ml), the latter shown to be more effective against *C.albicans*.

Table 4: *E. foetida* leaf extracts' minimum inhibitory concentration (MIC; g/ml) against microorganisms and *Candida albicans*.

Compound	P.aeruginosa	B.subtilis	C.albicans	S.aureus	E.coli
Nystatinforfungi	NT	NT	3.12	NT	NT
Gentamicinforbacteria	12.5	0.39	NT	1.56	3.12
Pet.etherextract	-	3.12	3.12	3.12	-
Chloroformextract	100	100	-	-	--
Ethylacetateextract	3.12	50	1.56	3.12	50
Methanolextract	-	200	-	3.12	200
Aqueousextract	-	100	12.5	50	-

Passiflorasubpeltata

Table 5 displays the results of a MIC experiment. Both the methanol and ethyl acetate extracts were shown to be effective against all of the test microorganisms. The 2.0 g/ml concentration of methanol extract of leaves and fruits was the most effective against *S. aureus*, *E. faecalis*, and *E. aerogenes* compared to Streptomycin's 2.5 g/ml, 3.5 g/ml, and 3.0 g/ml MIC values. Leaves and fruit extracts of both methanol and ethyl acetate were

shown to be effective against E.aerogenes, S.aureus, and E.faecalis in this investigation.

Table 5: The MIC (in micrograms per millilitre) of P.subpeltata leaf and fruit extracts in ethyl acetate and methanol against a panel of gram-positive and gram-negative bacteria.

	Extract	MIC(µg/mL)				
		S.aureus	E.coli	E. faecalis	K. pneumoniae	E.aerogenes
Leaves	Methanol	2.0	2.0	2.0	2.5	2.0
	Ethylacetate	2.5	2.5	2.5	3.0	2.5
Fruits	Methanol	2.0	2.0	2.0	2.5	2.0
	Ethylacetate	3.5	2.5	3.0	2.5	3.5
Standard	Streptomycin	2.5	2.0	3.5	1.5	3.0

Positive and negative effects on humans have led to widespread usage of alkaloids in medicine. Alkaloids may be found in a variety of plant tissues and often have their own unique chemical structures. Some alkaloids have been shown to have medicinal properties, including pain relief, inflammation reduction, and stress tolerance enhancement. The presence of alkaloids in the plants has been linked to a reduction in asthma symptoms by several researches.

Flavonoids are phenolic compounds that have been hydroxylated. Protecting against allergens, inflammation, free radicals, platelet aggregation, bacteria, ulcers, hepatotoxins, viruses, and cancers are just some of the many biological roles flavonoids play. Their capability to compound with bacterial cell walls likely accounts for their action. Flavonoids with a higher lipophilicity may potentially cause membrane disruption in microorganisms. Several types of gram-positive bacteria and fungi are vulnerable to their effects.

DISCUSSION

Infectious illnesses kill many people every year all across the world, but especially in poorer nations. Because of their unique chemical activity profile, natural materials provide many possibilities for the development of innovative additives and pharmacological treatments. The antibacterial efficacy of thousands of plant species has been investigated. The great majority of species have not yet been evaluated. Many drugmakers are investigating plant-based medicines in response to the prevalent notion that these are safer and more dependable than the more expensive synthetic pharmaceuticals.

New antimicrobial chemicals, notably those effective against bacterial infections, are derived from plant extracts. The current work's findings corroborate the existence of elements with documented pharmacological and physiological activities.[4]

Klebsiella pneumoniae is a Gram-ve bacteria, meaning that it is a rod-shaped, non-motile, encapsulated,

facultative anaerobe. As a result of inflammation, haemorrhage, and cell death, it may lead to detrimental alterations in human lungs. Carbapenem-resistant Klebsiella pneumoniae is one example of the rising tide of bacteria that have developed resistance to antimicrobial drugs. Acknowledge that K. pneumoniae is becoming resistant to carbapenems.[5]

Gram-negative bacteria are more difficult to kill than Gram-positive bacteria because their cell walls are thicker and more resistant to antibacterial medicines, hence all three medicinal plants were evaluated for their ability to inhibit Gram-negative bacteria. Ethanolic extract, on the other hand, showed no bactericidal action in investigations.[6]

Similar to the work, we used the ethyl acetate extract of selected plants to test against bacteria like E. coli, E. aerobacter, K. pneumoniae, S. aureus, and E. faecalis and found maximum inhibiting activity. Traditional usage, such as diuretic, anti-inflammatory, edematous, diarrheic, dropsy, and antispasmodic, may have some validity in the antibacterial activities shown in the current study's extracts, which include alkaloids, steroids, and flavonoids.[7]

Antimicrobial medicines that are both environmentally benign and physiologically efficacious are urgently required. Based on the results of this investigation into the antibacterial activity of plants, ethyl acetate extract was shown to be the most effective against E. coli, while chloroform extract was the least effective.[8]

Positive effects were seen for almost all tested plant extracts. Eryngium foetidum ethyl acetate extract had the greatest zone of inhibition, measuring 28 mm at a MIC of 3.12 g/ml. Drug-resistant Staphylococcus strains have been documented, and our findings are consistent, who found that antistaphylococcal properties may be found in naturally occurring compounds. The kind of solvent employed has a significant impact on the accuracy of predicting natural chemicals from plant extracts. Organic solvents showed more antibacterial activity against test microorganisms than aqueous extracts, contrary to the practises of conventional practitioners who utilised water as a solvent. [9]

CONCLUSION

Antimicrobial activity of phenolic compounds have been described. Tannin refers to a class of polymeric phenolic compounds that may be used to precipitate gelatin out of solution or to tan leather. They may be found in the bark, wood, leaves, fruits, and roots of a plant. Tannins have been linked to a variety of physiological effects in humans, including the activation of phagocytic cells, host-mediated tumour activity, and a variety of anti-infective effects. Possible mechanisms of their antibacterial activity

include interference with microbial adhesions, enzymes, cell envelope, transport-proteins, etc.

Terpenoids were shown to have wound healing effects. Some of the most common species responsible for wound infection include staphylococcus aureus, escherichia coli, pseudomonas aeruginosa, streptococcus pneumonia, and streptococcus pyogenes, Both bacteria and fungi are inhibited by terpenoids . They are quite effective against Bacillus subtilis and Staphylococcus aureus, but less so against Gram-negative bacteria and Candida albicans .

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