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# Analysis of Antibacterial Action of Common Household Plants - *Barbadensis miller* (Aloe Vera) & *Mentha piperita* (Mint) Against Various Strains of Pathogenic Bacteria and to Evaluate their Role as an Alternative Source of Treatment for Common Ailments

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**Abstract:- The Global concern regarding new multidrug resistant pathogenic microbial strains developing due to increased usage and misuse of antibiotics has paved the way for reviving an alternate plant based safe and effective medicine which targets pathogens without rendering them drug resistant. Plants which are a crucial part of our daily diet tend to possess potent antimicrobial compounds, it is therefore imperative to screen these plants for their antimicrobial activity, thus providing an alternate readily available source of medicine to combat the various pathogenic microbial infectious agents. The present study was carried out to evaluate the antimicrobial activity of plant extracts that we use in day to day lives. The antimicrobial potential of two different plant extracts were screened against six pathogenic microorganisms. The methanolic, ethanolic extracts of *Aloe vera* and *Mentha piperita* were subjected to a test of the antimicrobial properties by agar well diffusion method. The results indicated that the extracts of *Mentha piperita* exhibited potent antimicrobial properties. The highest antimicrobial activity was observed in the methanolic extract of *Mentha piperita* against *Bacillus subtilis* followed by ethanolic extract against *Bacillus subtilis.* Standard drugs were used for comparing the inhibitory action of plant extracts against commercially available antibiotics such as Gentamicin, Penicillin-G and Chloramphenicol against the bacteria. The organic solvents such as n-hexane was also used for testing the antimicrobial activity. The leaf extracts showed moderate to high activity against both Gram positive and Gram negative bacteria. When the antimicrobial properties of both the plants were compared the extract of *Mentha piperita* showed higher antimicrobial properties than *Aloe vera*. This shows that the *Mentha piperita* possesses effective compounds which are responsible for eliminating the pathogenic bacteria, and the antimicrobial activity was due to the presence of various bioactive compounds such as the alkaloids, flavonoids, steroids and phenolic compounds present in the organic extracts.**

## I. INTRODUCTION

Herbs have been in use for their therapeutic properties to alleviate or provide respite from various illnesses and their use can be traced back to the ancient human relatives, the Neanderthals about 60,000 years ago. Plants can act as a potential source for developing new age drugs which can be safe and effective. Till recent times there have been only a few plant based compounds which have been tapped for active use as pharmaceutical medicine and many compounds remain untapped and should be explored for their potential usage [1, 2, 3 ]

Many traditional plant-based medicines have been used for treating various ailments across the globe. In many developing nations, around 80% of the population relies on these traditional herbs and the knowledge has been passed on for generations. People not only find herbs as effective but also low cost and therefore are more popularly used for their affordability. Lately many new age synthetic drugs show debilitating side effects and many resulted in resistant pathogenic strains against the drug or antibiotic. Thus plant origin biologically active compounds are gaining traction in the search for safe and effective natural compounds which have efficacy against the chosen target [ 4, 5]

Crude plant extracts have been in use for centuries. Plant derived compounds display less pronounced drug resistance in pathogens because of the varied mechanisms used by secondary metabolites to target the pathogens. Screening of plant based compounds for their potential activity is therefore very essential to find alternate lines of treatment for the existing antibiotics to prevent development of drug resistant pathogens, as these antibiotics have shown to aggravate the problem of drug resistant pathogens.

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Therefore various plant extracts, herbs and spices are currently being screened to help screen out safe and effective alternatives for antibiotics and synthetic drugs, and also made available as a means of cheap and effective home based remedy to people to treat people themselves for infections [6, 7].

➢ *Aloe Vera -Barbadensis Miller*

Numerous studies have indicated about the antimicrobial, anti-inflammatory and wound healing properties of *Barbadensis miller* (*Aloe*) member of the Liliaceae family. Various phytochemicals that are present include salicylic acid, phenolic compounds such as anthraquinones, fatty acids, steroids, cholesterol, campesterol, beta-sitosterol and lupeol, hormones, auxins and gibberellins for wound healing and are anti-inflammatory. It also possesses 20 amino acids and out of which 7 are essential amino acids. Also contain lignin, an inert compound used in topical preparations for skin penetration and saponins, the soapy substances and about 3% of the gel and have cleansing and antiseptic properties [8, 9, 10] .

Aloe vera is a succulent of the Genus *Aloe*. It has been widely used for its medicinal properties as well as in lotions, cosmetics, ointments and beverages [11, 12]. *Aloe vera* is known to have more than 75 active compounds including anthraquinones and glycosides [13, 14]. The anti-inflammatory action of Aloe has been attributed to lupeol which acts as an antiseptic and analgesic. Auxins and gibberellins in Aloe have also demonstrated anti-inflammatory and wound healing action and the salicylic acid has an anti-inflammatory and antimicrobial action [15, 16]

➢ *Mint- Mentha Piperita*

The *Mentha piperita* plants belong to the family Lamiaceae genus *Mentha*. This family is known to possess many biologically active compounds. The *Mentha* species leaves, flowers, stems and oil extracts are extensively used in herbal teas, tincture, several folk remedies and ointments. Nowadays mint essential oil is being used in food preservatives. Many researchers have vouched for the rubefacient and antispasmodic for muscle pain relief, astringent for cleaning, antipyretic for fever, suppresses catarrh, antiseptic, stimulant, anti-aging activity [17, 18, 19].

The antibacterial activity of mint family is well known, peppermint oil from *Mentha piperita* has displayed antimicrobial action against *Staphylococcus aureus, Streptococcus pyogenes* and *B. subtilis and other* Gram-negative bacteria. The Antibacterial activity has been attributed to the compounds such as pulegone, menthone, menthol, methyl eugenol, carvone, 1, 8-cineole, limonene and b-caryophyllene and phenolic compounds also such as α-pinene, citronellol [19, 20, 21].

According to the World Health Organization (WHO) recent alarming increase in multi-drug resistance pathogenic strains has led to a dire situation where there is a need for discovering new potent compounds which can be used as an alternative for the current commercial antibiotics. Therefore, it is now even more imperative to explore the antimicrobial action from alternate sources. Antimicrobial action of common household plants such as *Aloe vera* and *Mentha piperita* against various pathogenic strains can be evaluated to find out whether they contain any potential antimicrobial compounds. The present report gives us an account of the antibacterial effect of leaves of Aloe vera and Mint plants [22, 23]

## II. MATERIALS AND METHODS

➢ *Collection of Plants:*

Aloe vera and *Mentha piperita*, healthy leaves of these plants were collected from the Sainikpuri, Telangana district, Hyderabad India. The plants were identified by T. Sudha taxonomist, Department of Botany. Sarojini Naidu Vanita Mahavidyalaya, Exhibition Grounds. And the specimens are deposited in the museum of Botany.

Fig 1 Aloe Vera Plant

Fig 2 Mentha Piperita Plant

➢ *Preparation of Plant Extract*

Mature, healthy and fresh leaves of *Aloe vera* were washed under running tap water followed by rinsing with distilled water. Then the leaves were dissected longitudinally and the colorless parenchymatous tissue i.e. the gel was scraped out using a sterile knife without fibers. The gel was ground with methanol with the help of mortar

and pestle. The extracts were filtered using Whatman filter paper No.1 and the filtrate was centrifuged at 5000 rpm for 5 mins.

➢ *Crude Extraction of Organic Solvent*

Weighed 10gm of each plant extract thoroughly mixed with 50 ml of organic solvents like methanol/ethanol and acetone respectively. The mixture thus obtained was filtered through Whattman's filter paper no. 1. The filtrate was concentrated by complete evaporation of solvent at room temperature to yield the pure extract. Stock solution of crude extract was prepared by mixing the appropriate amount of dried extracts with appropriate solvents to obtain a final concentration of 100 mg/ml. Each solution was stored at 4° Celsius after collecting in sterilized glass tubes until used.

➢ *Disc Preparation*

The 5 mm diameter discs were prepared from Whattmans filter paper no.1. and they were immersed in the organic solvents for a minimum of 20 minutes, later the discs were sterilized by using the autoclave at 12°C. After autoclaving the sterilized discs were dried on a hot air oven at 50°C. Similarly, discs for various solvents and control discs were prepared.

➢ *Preparation of Agar Media for the Growth of Bacteria.*

The media used for culturing the microorganisms was Nutrient Agar Medium. It is used for culturing microbes and supports growth of a wide range of non-fastidious organisms. Nutrient agar is used because various types of bacteria as well as fungi can be grown in this medium, agar also supports the growth of bacteria. Nutrient agar medium was prepared by dissolving 28g of nutrient agar powder in 1 liter of distilled water and the pH was adjusted to 7.4. The mixture was heated with constant stirring so as to dissolve the components. After dissolving the mixture, the mixture is autoclaved at 120 degrees for 15 minutes. Once the nutrient agar has been autoclaved, allow the mixture to cool at room temperature before pouring it into the petri-plates before solidifying under aseptic conditions.

➢ *Bacterial Strain Selection*

A total of six bacterial stains such as *Klebsiella pneumoniae, Bacillus subtilis, Staphylococcus aureus, Proteus vulgaris, Pseudomonas aeruginosa, Escherichia coli* were selected to assess the susceptibility patterns against the extracts prepared in the present study. All the strains were collected from the Microbial type culture collection, India. The bacterial cultures were maintained at 37°C. Prior to susceptibility test the microorganisms were reactivated by inoculating them in separate petri-plates containing the growth media. The reactivation was carried out by pour plate method and streak plate method.

➢ *Antibacterial Susceptibility Assay*

Extracts obtained by various processes were evaluated for their potential antibacterial activities by the standard agar well diffusion method. All the extracts were sterilized by a sterile membrane syringe filter.

## III. RESULTS

➢ *Antimicrobial Activity of Aloe vera*

The present study carried out on plant extracts of *Aloe vera* and *Mentha piperita* against the microbial activity revealed positive results. The successive leaf extracts using methanol, ethanol, acetone, which were tested against the pathogenic bacteria at a dose of 100 mg/ml showed the following results.

The results of antimicrobial activity of *Aloe vera* leaf extract tested against pathogenic microorganisms such as *Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Proteus vulgaris,* and *Escherichia coli* are presented in Table 1. The tested extracts showed moderate to strong antimicrobial activity measured based on the zone of inhibition against the selected bacterial strains as seen in Figure 3. The *Aloe vera* plant leaves extracts were prepared in organic solvents such as ethanol and methanol. Ethanol solvent showed maximum zone of inhibition with active metabolites. Extracts of *Aloe vera* showed moderate antimicrobial activity at a concentration of 100mg/ml.

Leaf extracts displayed the zone of inhibition ranging from 3 mm to 9 mm in diameter (Fig. 4-9). Amongst all the six organisms tested, higher inhibitory activity was observed against the organisms *Pseudomonas aeruginosa, Bacillus subtilis and Proteus vulgaris*. The ethanol leaf extract at the dose level of 100 mg/ml showed a moderate zone of inhibition 8 mm against *Pseudomonas aeruginosa* (Fig. 7), followed by 6 mm against *Proteus vulgaris* (Fig. 5) and *Bacillus subtilis* (Fig. 4), 5 mm against *Klebsiella pneumoniae* (Fig. 6) and *Staphylococcus aureus* (Fig. 8), and 3 mm against *Escherichia coli* (Fig. 9). The leaf extract of methanol at the dose level of 100 mg/ml showed the inhibition zone of 6 mm for *Staphylococcus aureus* (Fig. 8)and *Escherichia coli* (Fig. 9), 5 mm for *Bacillus subtilis* (Fig. 4) and *Klebsiella pneumoniae,* 4 mm for *Proteus vulgaris* (Fig. 5), 3 mm for *Pseudomonas aeruginosa* (Fig. 7).

Table 1 Antimicrobial Activity of Leaf Extract of *Aloe vera*

| ***Zone Of inhibition (mm)*** | ***Ethanol*** | ***Methanol*** | ***n-Hexane (Control)*** | ***Acetone (Control)*** | ***Methanol (Control)*** | ***Ethanol (Control)*** |
|---|---|---|---|---|---|---|
| *Bacillus subtilis* | *6mm* | *5mm* | *3mm* | *8mm* | *5mm* | *6mm* |
| *Proteus vulgaris* | *6mm* | *4mm* | *3mm* | _ | *3mm* | *3mm* |

| | | | | | | |
|---|---|---|---|---|---|---|
| *Klebsiella pneumoniae* | *5mm* | *5mm* | _ | *6mm* | *4mm* | *3mm* |
| *Pseudomonas aeruginosa* | *8mm* | *3mm* | *4mm* | *9mm* | *3mm* | *5mm* |
| *Staphylococcus aureus* | *5mm* | *6mm* | _ | _ | _ | *4mm* |
| *Escherichia coli* | *3mm* | *6mm* | _ | *3mm* | *3mm* | *3mm* |

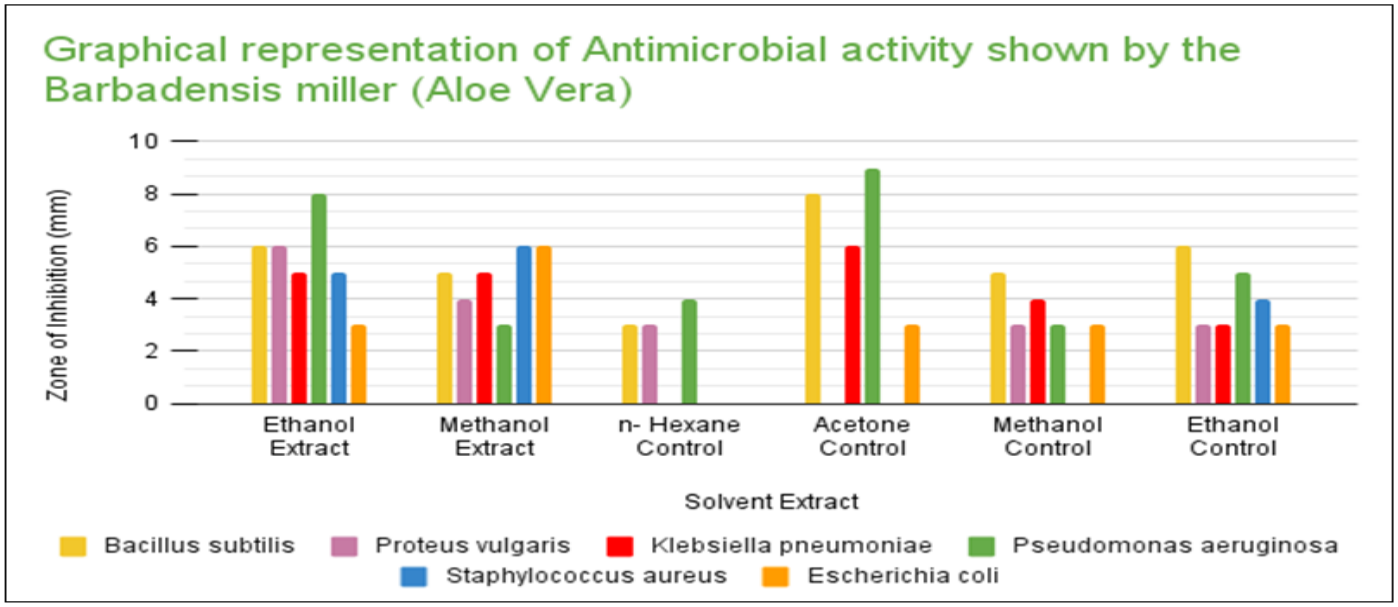


Fig 3 Graphical Representation of Aloe vera Gel Extractions

➢ *Plates Showing Zone of Inhibition for Plant Extract of Aloe vera*

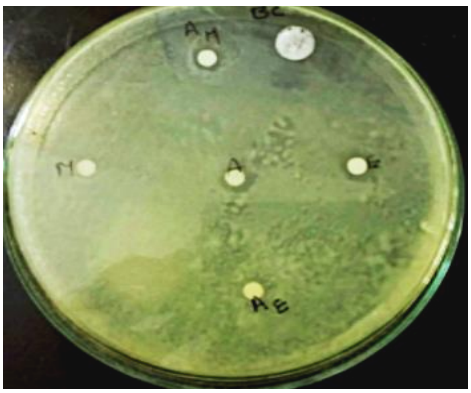

Fig 4 Antimicrobial activity *against Bacillus Subtilis*

Fig 5 Antimicrobial activity *against Proteus Vulgaris*

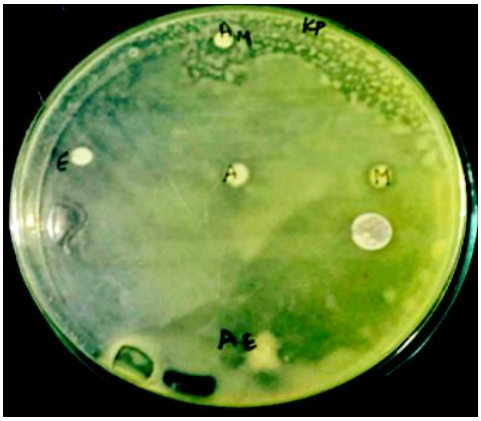

Fig 6 Antimicrobial activity *against Klebsiella pneumoniae*

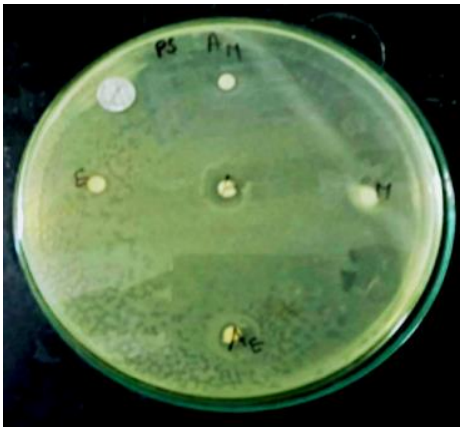

Fig 7 Antimicrobial activity against *Pseudomonas aeruginosa*

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Fig 8 Antimicrobial activity against *Escherichia coli*

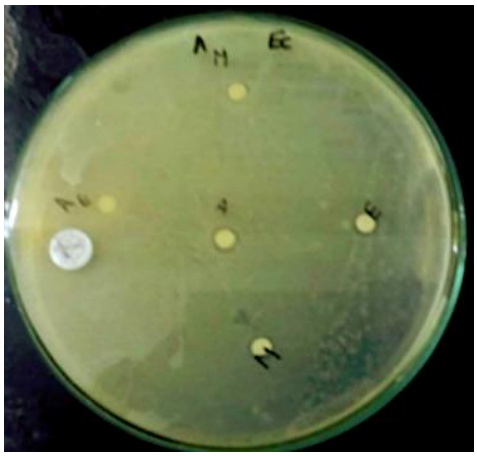

Fig 9 Antimicrobial activity against *Staphylococcus aureus*

➢ *Antimicrobial activity of Mint*

The results of antimicrobial activity of *Mentha piperita* leaf extract tested against pathogenic microorganisms such as *Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Proteus vulgaris,* and *Escherichia coli* are presented in Table 2. The tested extracts showed moderate to strong antimicrobial activity measured based on the zone of inhibition against the selected bacterial strains.

The *Mentha piperita* plant leaves extracts were prepared in organic solvents such as ethanol and methanol. Ethanol solvent showed maximum zone of inhibition with active metabolites. Extracts of *Mentha piperita* showed high antimicrobial activity at a concentration of 100mg/ml.

Leaf extracts displayed the zone of inhibition ranging from 6 mm to 22 mm in diameter (Fig. 11-16). Amongst all the six organisms tested, higher inhibitory activity was observed against the organisms *Bacillus subtilis, Staphylococcus aureus, Escherichia coli and Proteus vulgaris*.

The ethanol leaf extract at the dose level of 100 mg/ml displayed high zone of inhibition 20 mm against *Bacillus subtilis* (Fig. 11), followed by a moderate to high of 10 mm against *Staphylococcus aureus* (Fig. 15) and *Escherichia coli* (Fig. 16), 8 mm against *Klebsiella pneumoniae* (Fig. 13) and *Pseudomonas aeruginosa* (Fig. 14) and 6 mm against *Proteus vulgaris* (Fig. 12).

The leaf extract of methanol at the dose level of 100 mg/ml showed an enhanced inhibition zone of 22 mm for *Bacillus subtilis*, 10 mm each for *Proteus vulgaris* (Fig. 12), *Staphylococcus aureus* (Fig. 15) and *Escherichia coli* (Fig. 16), 6 mm for both *Klebsiella pneumoniae* (Fig. 13) and *Pseudomonas aeruginosa* (Fig. 14).

Table 2 Antimicrobial Activity of *Mentha piperita*

| ***Zone Of inhibition (mm)*** | ***Ethanol*** | ***Methanol*** | ***Methanol (Control)*** | ***Acetone (Control)*** | ***Ethanol (Control)*** |
|---|---|---|---|---|---|
| *Bacillus subtilis* | *20mm* | *22mm* | *5mm* | *5mm* | _ |
| *Proteus vulgaris* | *6mm* | *10mm* | *10mm* | *6mm* | _ |
| *Klebsiella pneumoniae* | *8mm* | *6mm* | _ | _ | *1mm* |
| *Pseudomonas aeruginosa* | *8mm* | *6mm* | _ | *6mm* | _ |
| *Staphylococcus aureus* | *10mm* | *10mm* | *8mm* | _ | _ |
| *Escherichia coli* | *10mm* | *10mm* | _ | *10mm* | _ |

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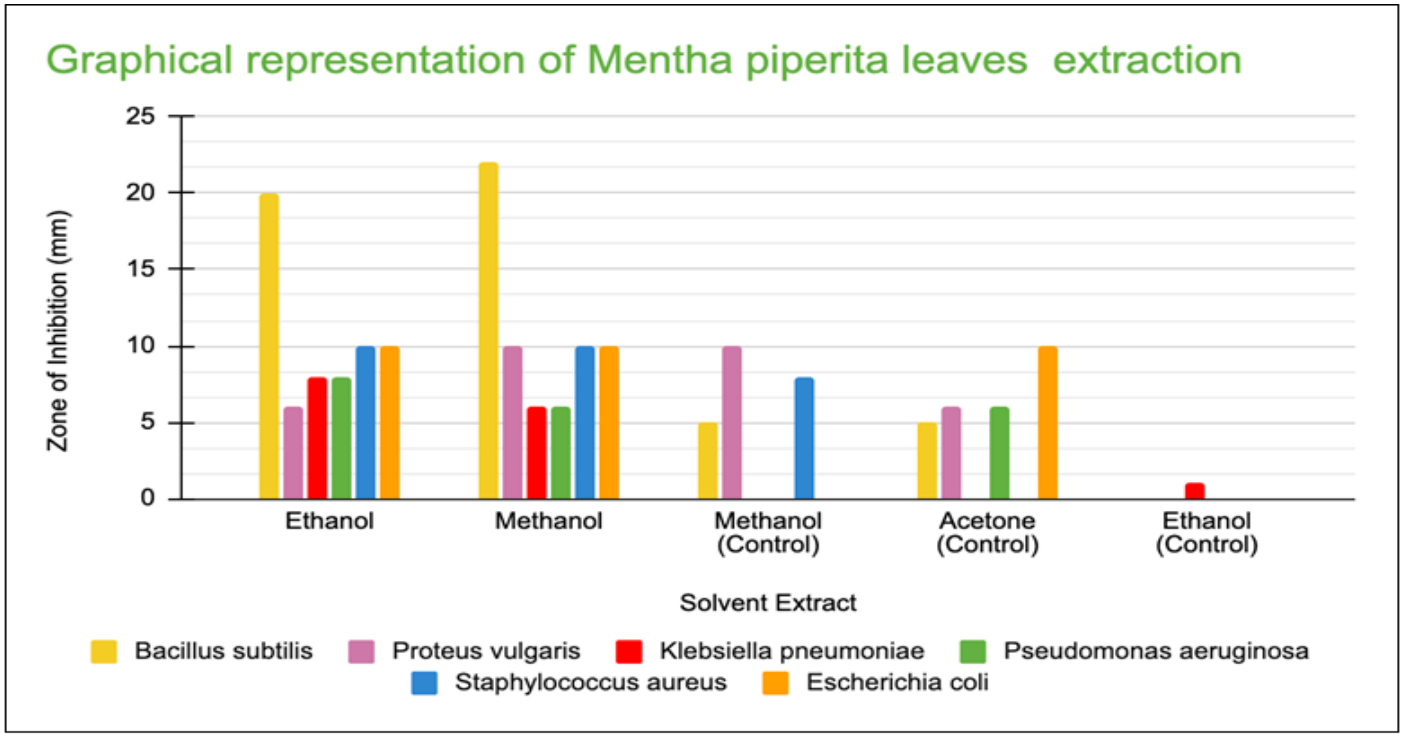


Fig 10 Graphical Representation of *Mentha piperita* Leaves Extraction

- *Plates Showing Zone of Inhibition of Plant Extract of Mentha Piperita*

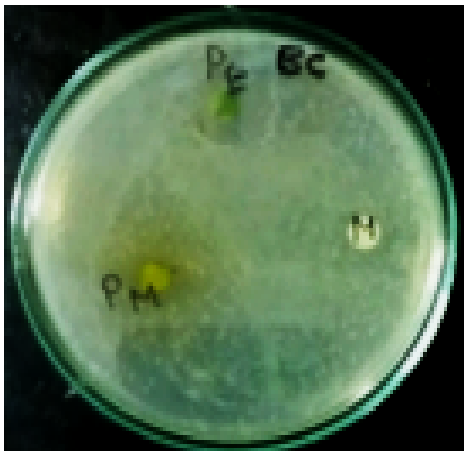

Fig 11 Antimicrobial activity against *Bacillus subtilis*

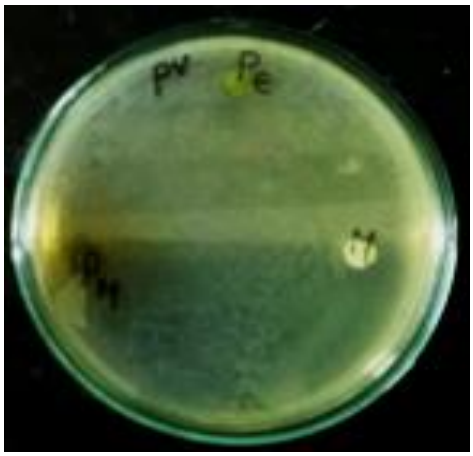

Fig 12 Antimicrobial activity against *Proteus vulgaris*

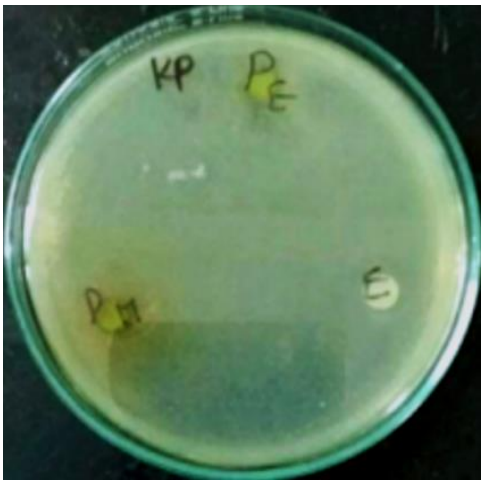

Fig 13 Antimicrobial activity against *Klebsiella pneumoniae*

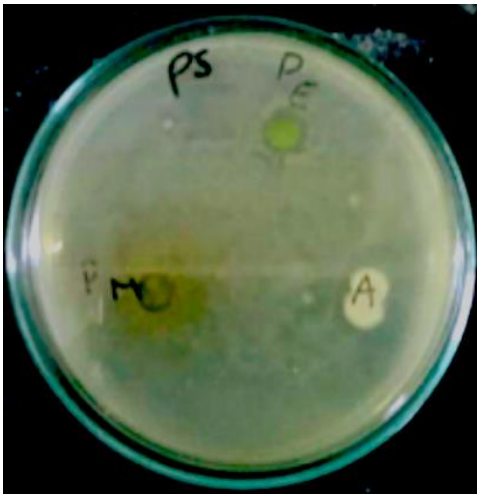

Fig 14 Antimicrobial activity against *Pseudomonas aeruginosa*

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Fig 15 Antimicrobial activity against *Staphylococcus aureus*

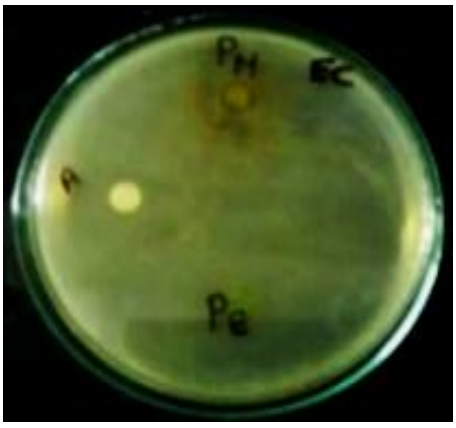

Fig 16 Antimicrobial activity against *Escherichia coli*

The standard drugs were also used for the comparison of the antimicrobial activity, the drugs used were Gentamicin, Penicillin-G and Chloramphenicol. The Commercially available antibiotics displayed moderate to high antimicrobial activity against the bacterial strains (Figures 16-22).

Gentamicin showed the most potent at a dose level of 10 mcg/disc, displaying the inhibition zone of 20 mm for *Bacillus subtilis* (Fig. 17), *Proteus vulgaris* (Fig. 18) and *Staphylococcus aureus* (Fig. 21), 13mm for *Escherichia coli* (Fig. 22), 10 mm for *Klebsiella pneumoniae* (Fig. 19) *and Pseudomonas aeruginosa* (Fig. 20).

Penicillin-G a dose level of 10 units/disc showed the inhibition zone of 15 mm for *Proteus vulgaris* (Fig. 18), 5 mm for *Bacillus subtilis* (Fig. 17) and *Klebsiella pneumoniae* (Fig. 19), *Escherichia coli* 3mm (Fig. 22), *Staphylococcus aureus* 2 mm (Fig. 21), and 1mm for *Pseudomonas aeruginosa* (Fig. 20).

Chloramphenicol a dose level of 30 mcg/disc showed an inhibition zone of 25 mm against *Proteus vulgaris* (Fig. 18), 15 mm for *Pseudomonas aeruginosa* (Fig. 20) , *Staphylococcus aureus* (Fig. 21) and *Bacillus subtilis* 10 mm (Fig. 17), *Klebsiella pneumonia* 5mm (Fig. 19), *Escherichia coli* 2mm (Fig. 22).

Table 3 The Antimicrobial activity of Commercial Antibiotics

| ***Zone Of Inhibition (mm)*** | ***Gentamicin*** | ***Penicillin-G*** | ***Chloramphenicol*** |
|---|---|---|---|
| *Bacillus Subtilis* | *20 mm* | *5mm* | *10mm* |
| *Proteus Vulgaris* | *20 mm* | *15mm* | *25mm* |
| *Klebsiella pneumoniae* | *10 mm* | *5 mm* | *5mm* |
| *Pseudomonas aeruginosa* | *10 mm* | *1 mm* | *15mm* |
| *Staphylococcus aureus* | *20 mm* | *2 mm* | *10mm* |
| *Escherichia Coli* | *13 mm* | *3 mm* | *2mm* |

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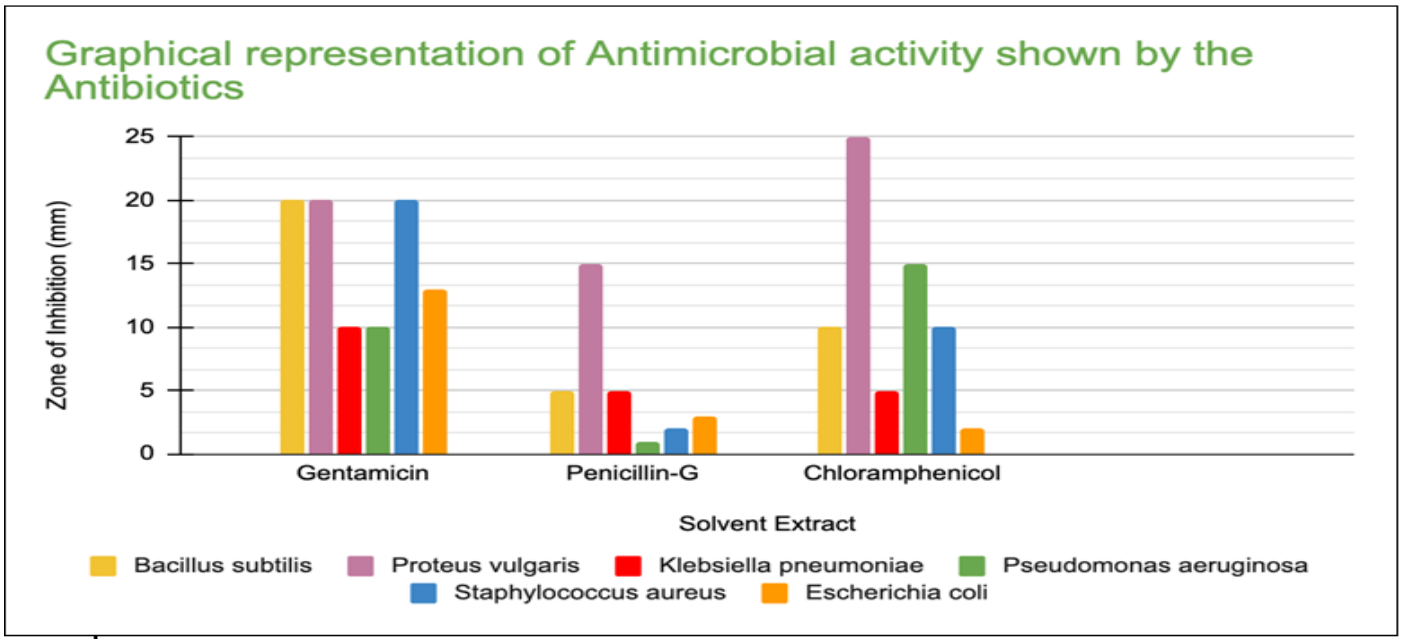


Fig 17 Graphical Representation of Antibiotics

- *Antimicrobial activity of Chloramphenicol, Gentamicin and Penicillin-G*

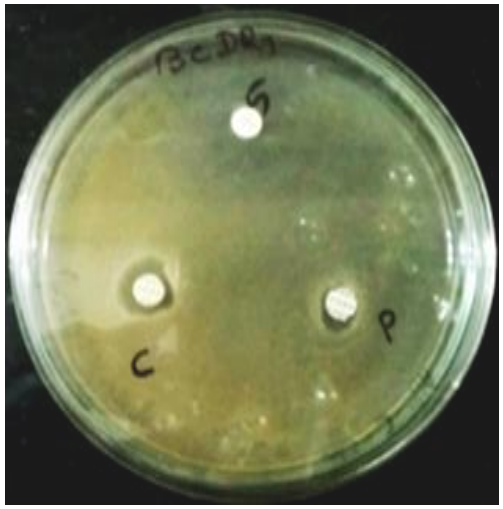


Fig 18 Action against *Bacillus subtilis*

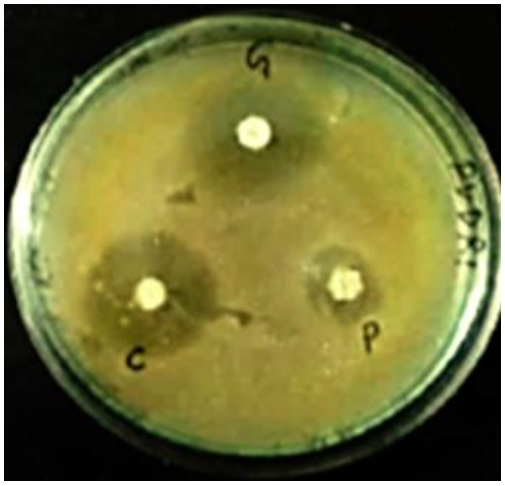


Fig 19 Action against *Proteus vulgaris*

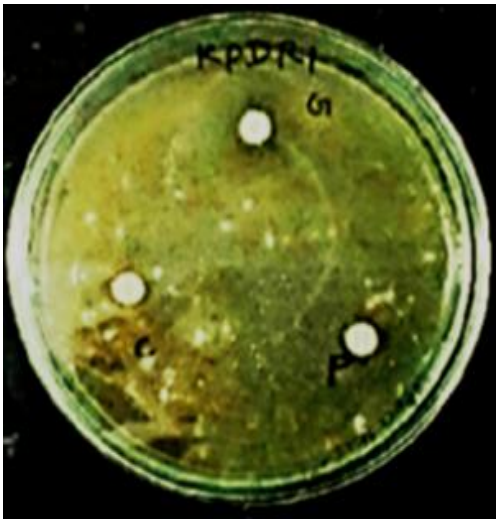


Fig 20 Action against *Klebsiella pneumoniae*

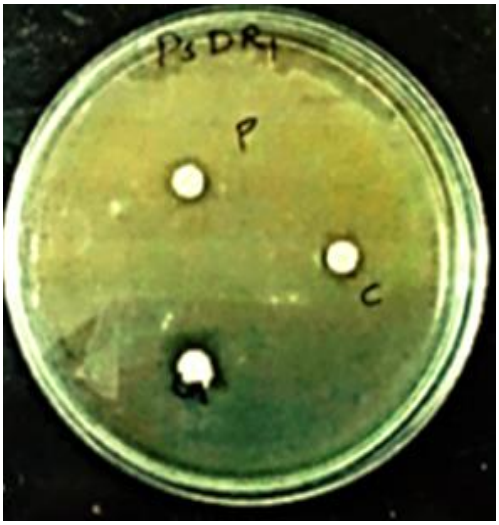


Fig 21 Action against *Pseudomonas aeruginosa*

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Fig 22 Action against *Staphylococcus aureus*

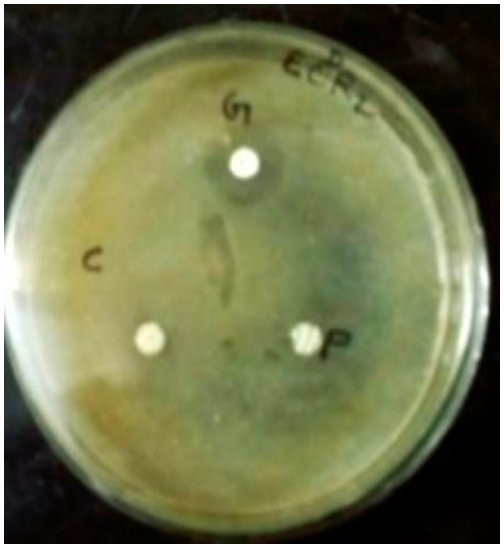

Fig 23 Action against *Escherichia coli*

## IV. DISCUSSION

The solvent extracts of *Mentha piperita* displayed a strong notable antimicrobial activity against *Bacillus subtilis*, and fairly moderate to high action against *Staphylococcus aureus* and *Escherichia coli*, *Klebsiella pneumoniae and Pseudomonas aeruginosa* when compared with the zone of inhibition of standard drugs like Gentamicin, Chloramphenicol, Penicillin-G which had the plant extracts have also shown a zone of inhibition around the discs that were placed in the middle of the culture medium.

*Mentha piperita* showed promising antimicrobial action as compared to solvent extracts of *Aloe vera* which had a low to moderate action against all the pathogenic strains. This study also illustrated the pronounced action of *Mentha piperita* against Gram-positive bacteria such as *Bacillus subtilis and Staphylococcus aureus* and a moderate activity against Gram negative strains such as *Escherichia coli, Klebsiella pneumoniae and Pseudomonas aeruginosa* .

## V. CONCLUSION

In the present study the *Mentha piperita* extracts demonstrated promising antimicrobial activities against the most prevalent microorganisms. Development of herbal medicine for treatment of minor ailments, offering an easy to consume at home remedy, can offer a cheap and safe alternative to the harsh antibiotics which is a work in progress worldwide. Phytochemical screening of different bioactive compounds in the *Mentha piperita* extracts followed by their identification and purification is required in order to develop a more effective drug in purified form. Finally, it can be concluded that the active chemical compounds present in the *Mentha piperita* have an antimicrobial activity against both Gram positive bacteria and Gram negative bacteria and should certainly be used in the treatment of various bacterial infections so as to prevent development of drug resistant microbial strains. Future course of action requires an extensive study of *Mentha piperita* compounds against other potential pathogenic strains of microorganisms, to expand its use in treatment of various other diseases and fully exploit the complete range of bioactive compounds the plant can offer as a safe yet effective alternate means of treatment.

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