



Antimicrobial activity of Plant Extracts (*Curcuma longa*, *Syzygium aromaticum*, *Azadirachta indica*)

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Article info

Received: 15/05/2021

Revised: 28/06/2021

Accepted: 21/07/2021

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Abstract

This research is demonstrated that, the utilization of medicinal plants in Ayurvedic medicine has long been acknowledged for its contributions to human health care systems. This paper underscores the significance of plant-derived formulations in modern medicine, emphasizing their safety and therapeutic advantages over synthetic drugs. With over 1500 herbal preparations available globally, the potential of combining medicinal plants to enhance antimicrobial efficacy against enteric bacterial infections is explored. However, the emergence of multidrug-resistant bacterial strains necessitates urgent exploration of alternative antimicrobial agents. Medicinal plants offer a promising avenue due to their organic antibacterial properties attributed to secondary metabolites. Despite numerous reports on the antimicrobial activity of plant extracts, limited in vitro studies on herbal preparations have been conducted. This study seeks to address this gap by screening the antibacterial potential of herbal products, providing insights into their effectiveness against a spectrum of bacterial pathogens.

Keywords: Plants, Extract, Anti-microbial

Introduction

Many of the medicinal plants used in Ayurvedic medicine can help to improve the human health care system, and formulations made from these plants are valuable in contemporary medicine. Numerous herbal remedies were also discovered by Ayurvedic practitioners to treat a range of conditions (Patwardhan *et al.*, 2003). The main advantages of using medicine produced from plants are that they are significantly more therapeutic and safer than synthetic pharmaceuticals (Pandey *et al.*, 2008). Since ancient times, single and polyherbal preparations have been used to maintain human health due to their wide range of bioactive chemicals (Barrett *et al.*, 1999). More than 1500 herbal concoctions are available for purchase as ethnic traditional remedies or dietary supplements (Handa *et al.*, 2004). WHO and associates (2000) Combinations

of therapeutic plants may broaden the medicines' antibacterial spectrum and boost their efficacy. In underdeveloped nations, diarrheal or enteric illnesses are serious public health issues that cause 3.3–6.0 million child deaths yearly. The main causes of both sporadic and epidemic diarrhea in children and adults are enteric bacteria, which include *Salmonella* spp., *Shigella* spp., *Proteus* sp., *Klebsiella* sp., *Escherichia coli*, *Pseudomonas* sp., *Vibrio cholerae*, and *Staphylococcus aureus*. 5th Programme Report *et al.*, 1985.

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It has recently been shown that numerous human pathogenic microorganisms have become resistant to a number of synthetic medications (Vickers *et al.*, 1999.; De Smet *et al.*, 2002.; Dawson W *et al.*, 2005). According to Bhatia *et al.*, 2010, antimicrobial drugs play a crucial role in lowering the worldwide burden of infectious illnesses. However, because there are fewer, or occasionally no, effective antimicrobial agents available for the infection caused by pathogenic bacteria, the emergence and spread of multidrug resistant (MDR) strains in pathogenic bacteria have become a significant public health threat (Boucher *et al.*, 2009). Therefore, it is crucial to discover novel antimicrobial drugs given the evidence of the fast global spread of resistant clinical isolates. However, even new families of antimicrobial drugs will have a limited lifespan due to the history of the widespread, quick emergence of resistance to newly introduced antimicrobial agents (Marasini *et al.*, 2015). As a potential replacement that might be effective in the treatment of these problematic bacterial infections, a variety of medicinal plants have been identified as significant sources of organic antibacterial compounds (Iwu *et al.*, 1999). According to the World Health Organization (WHO), medicinal plants are the best source of a variety of drugs (World Health Organization, Geneva 2002). Many plants have been used because of their antibacterial qualities, which are caused by phytochemicals generated during the plant's secondary metabolism (Medina *et al.*, 2005). Many secondary metabolites, such as flavonoids, phenolic compounds, alkaloids, and tannins, are found in plants and have been demonstrated to have antibacterial properties in vitro (Djeussi *et al.*, 2013). There is a paucity of published in vitro investigations on herbal preparations, despite numerous reports on the antibacterial efficacy of crude plant extracts that inhibit different bacterial infections. Finding herbal items with antibacterial potential based on illnesses for which there is no medication or just palliative care accessible is urgently needed. In order to manage and prevent bacterial illness, an effort was undertaken to evaluate the antibacterial properties of herbal remedies (Patwardhan *et al.*, 2005., Greensfelder *et al.*, 2000). In this study, we extracted various plant extracts using ethanol

and water as two distinct solvents, and we investigated the antibacterial effects of these extracts on both Gram positive and Gram negative bacteria.

Material and Methods

Plants Used for Antimicrobial Activity

Testing plants against both gram positive and gram negative bacteria allowed researchers to determine their antimicrobial potential. In order to assess their antibacterial activity, the following plants were chosen.

Table: Plants That Are Used to Extract Phytochemicals.

Scientific Name	Family	Common Name	Used Plant Part	Weight
Curcuma longa	Zingiberaceae	Turmeric	Fruits	6g
Syzygium aromaticum	Myrtaceae	Clove	Fruits	6g
Azadirachta indica	Meliaceae	Neem	Leaves	6g

Collections of Greenery

Every plant was gathered from the nearby region. Each plant's designated portions were gathered in order to assess its antibacterial activity.

Phytochemical Extraction from Plants

Three plant samples were chosen for this study based on the folklore surrounding their traditional uses. We bought the plants from the nearby market. Table has a list of the plants along with their common names.

To get rid of dust and other contaminants, plant tissue such as leaves, rhizomes, bulbs, bark, etc., was collected and twice cleaned in distilled water. Following washing, the samples were dried for an entire night at 37°C and then ground with a mortar and pestle to create a paste or powder. Six grams of the material (paste or powder) were placed in two distinct beakers. Two distinct solvents (ethanol and distilled water) each contained 50 milliliters in separate beakers. After that, the bottles were kept at 50°C for 24 hours on a hot plate with a magnetic stirrer. To separate the plant extract from debris, the solutions were centrifuged at 3000 rpm for 15 minutes after 24 hours. Supernatants were carefully moved to fresh, labeled beakers. The extracts were dried at 37°C

in an incubator until a powdery consistency was achieved. The powder was again suspended at a concentration of 1(One) gm/ml in DMSO and autoclaved water. For future research, the finished phytochemical extracts were kept in storage at 4°C.

Preparation of media for antimicrobial activity
The anti-microbial activity was conducted using the nutrient agar medium. For NAM media, the following composition was utilized:

Components	Weight
Peptone	5g
Beef Extract	3g
Sodium Chloride	5g

Agar	15g
Distilled water	1 Lt

Aqueous and alcoholic extractions of some of the often found plants in this area are the focus of the current work. Using harmful bacteria as test bacterium, the antibacterial potential of the aqueous and alcoholic extracts was ascertained. The outcomes are shown in the following manner:

Phytochemicals Extraction

Both ethanol and aqueous solvents were used to extract phytochemicals from plant tissues. Figure 1 (Turmeric), Figure 2 (Clove) and Figure 3 (Neem Bark), are demonstrating the process of phytochemical extraction from several plants.

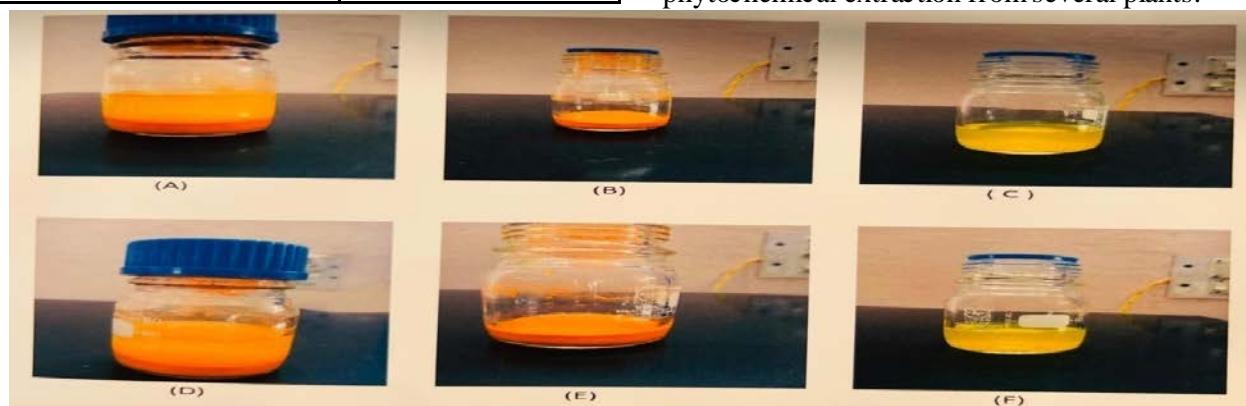


Figure: 1. Turmeric phytochemical extraction in ethanol (D to F) and aqueous (A to C) solvents A and D: Powdered turmeric dissolved in ethanol and water, B and E: After centrifuging, add the turmeric, and then move the supernatant from C to F.

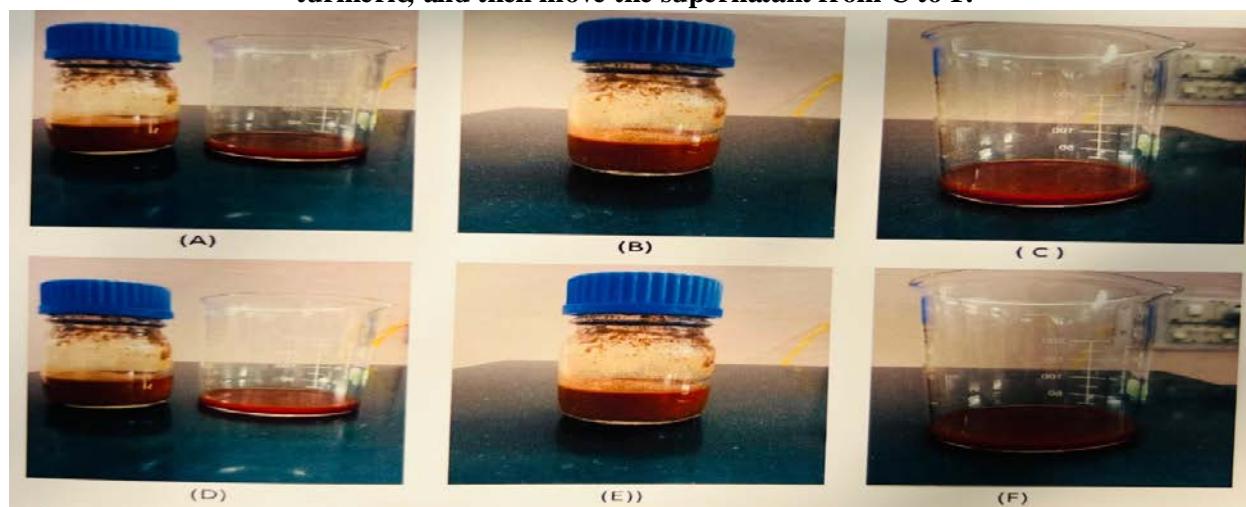


Figure: 2. The phytochemicals from cloves were extracted using ethanol solvent (D to E) and aqueous solvent (A to C). Clove crushing in Mortar Pastel: B and E: Clove powder obtained by centrifugation; C to F: Supernatant obtained through centrifugation.

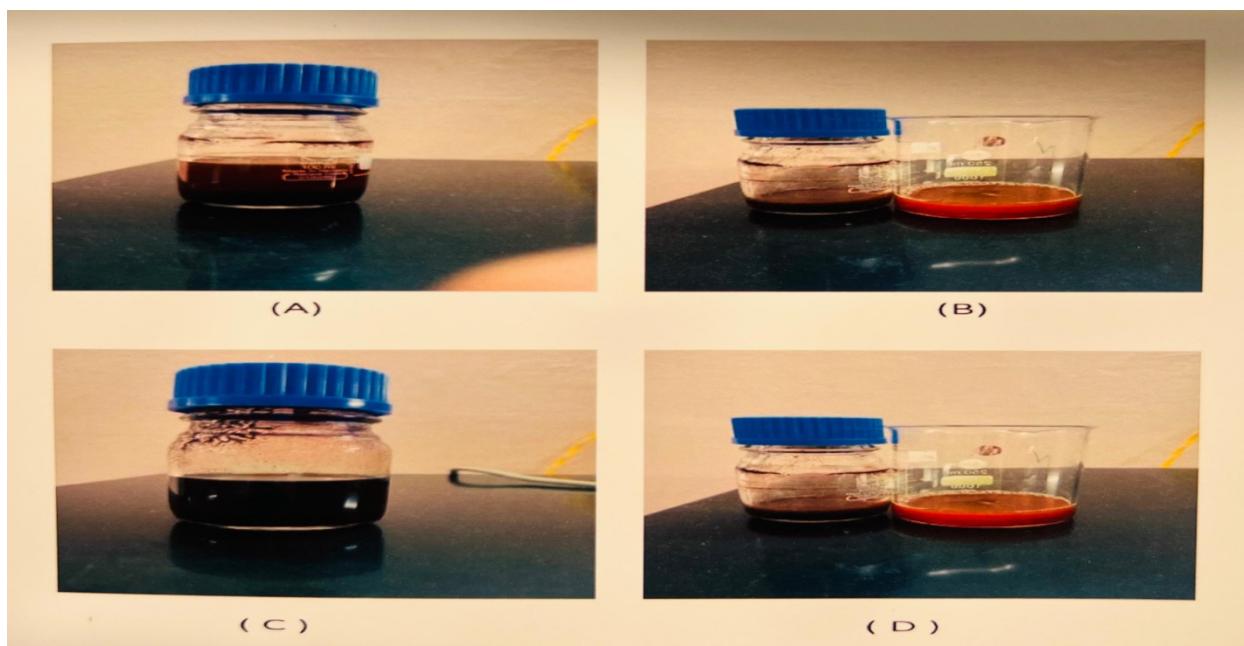


Figure: 3. Phytochemical extraction from neem bark using ethanol (C to D) and aqueous (A to B); A and C: Neem bark is crushed in a mortar and pestle and then dissolved in ethanol and distilled water (B to D: Supernatant after centrifugation).

Test for Antimicrobial Properties of Extracted Phytochemicals

Using *S. aureus*, Enterobacteria, *Acinetobacter*, *Klebsiella*, *B. subtilis*, and *E. faecalis* as test organisms, the antibacterial properties of extracted

phytochemicals were investigated. Scale was used to measure the zone of inhibition in millimeters (mm). Table provides a summary and calculation of the obtained zone of inhibition.

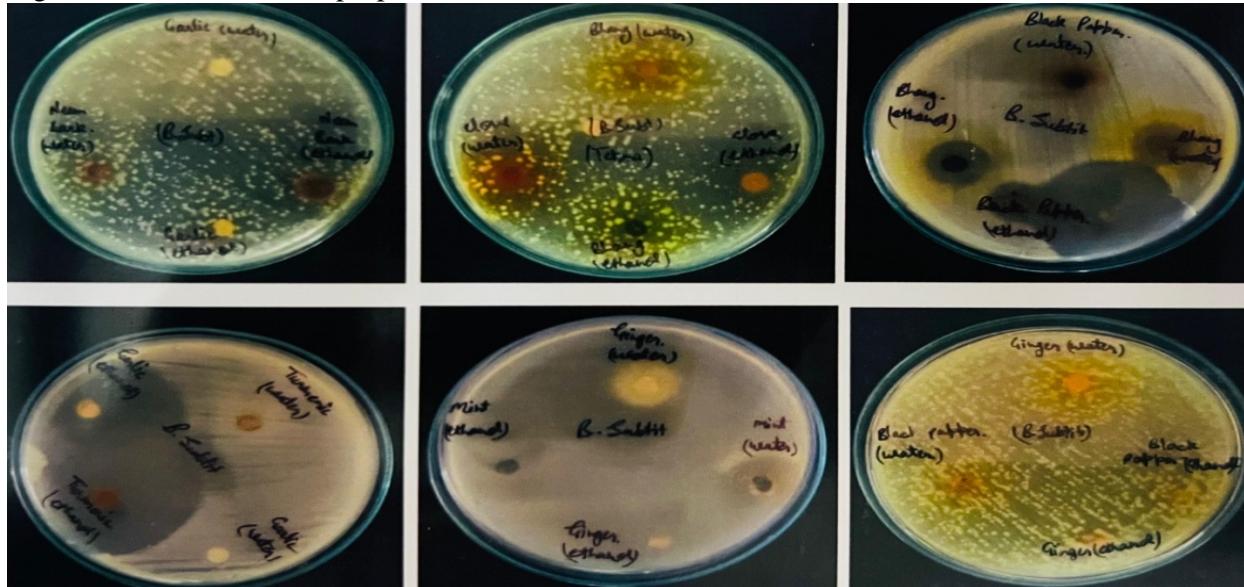


Figure: 4. Antimicrobial test for *B. Subtilis* bacteria using clove, bhang, ginger, black pepper, mint, turmeric, neem bark, and garlic. The positive control in this study was tetracydin.



**Figure: 5. Antimicrobial test of Turmeric, Clove and Neem Bark against bacteria S. Aureus.
Tetracyclin was used as Positive control.**

Table: Zone of inhibition was measured in mm.

Plants name	B. Subtilis	S. Aureus
Turmeric	0	0
Clove	0	8
Neem Bark	12	0
Tetracyclin	20	25

Results and Discussion

The goal of the current study was to ascertain the antibacterial activity of a few widely found indigenous flora. Traditional healers have long utilized the parts and derivatives of these plants as biostimulants, nervine tonics, and for the treatment of both infectious and non-infectious illnesses. Microorganisms play a significant role in health-related disorders or well-being since they affect the skin and several internal organs in both humans and other animals. It is well recognized that a healthy microbiota produces a wide range of vital nutrients. Pathogenic germs' development and lodgement are also inhibited by metabolites that are utilized by normal microbes. Finding out whether any particular plants have any antimicrobial activity becomes crucial when considering their impact on the healthy microbiota and their ability to inhibit pathogenic organisms. Using established protocols, attempts were made to determine the antibacterial activity of plants by aqueous and alcoholic extractions. Three gram positive (*Staph aureus*, *B. subtilis*, and *E. faecalis*) and three gram negative (*Acinetobacter*, *Enterobacter*, and *Klebsiella*) potentially harmful bacterial organisms were tested using bacterial organisms. Since antibiotics are frequently used to treat a wide range of illnesses, the sensitivity of these test organisms to antibiotics was investigated. It was found that some bacterial isolates had become resistant to these

medications. It was discovered that *S. aureus* was resistant to Ampicillin, Ciprofloxacin, Doxycycline, Methicillin, Penicillin, Sulfadiazine, and Amoxycillin. The discovery revealed that *B. subtilis* was resistant to Norfloxacin, *E. faecalis* to Ciprofloxacin, *Acinetobacter* to Ampicillin, *Enterobacter* to Ampicillin, Amoxicillin, and *Klebsiella* to Amoxycillin and Ampicillin. These outcomes support previous research findings. When three plant extracts were tested in this investigation against two pathogenic bacteria for possible antibacterial action, ethanol consistently and prominently exhibited antimicrobial activity greater than that of water extracts (Show in tables). A low concentration of antibacterial chemicals in water extracts may be the cause of their weak antibacterial action; the majority of aqueous extracts were found to be ineffective against all of the microorganisms under investigation. Antibacterial compound(s) or all of the recognized components from plants active against microbes may not be extracted by water extract, or the concentration of antibacterial compounds may be minimal. Furthermore, phytochemicals extracted in ethanol shown promising broad spectrum antibacterial activity against the tested microorganisms.

Conclusion

In conclusion, the plant preparations investigated consistently demonstrated higher antibacterial efficacy in alcoholic extracts compared to aqueous extracts against various microbes. Notably, the alcoholic extract exhibited superior potency against *Staphylococcus aureus* compared to other preparations. This consistent trend underscores the potential of alcoholic extracts as effective antibacterial agents, particularly against *S. aureus* infections. Further research into the mechanisms

underlying this enhanced activity and exploration of broader microbial spectra could enhance our understanding and application of plant-derived antibacterial agents. Overall, these findings highlight the promising role of alcoholic extracts in the development of novel antimicrobial therapies.

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