COMPARATIVE EFFECTIVENESS OF TINOSPORA CORDIFOLIA LEAVES AND STEMS AGAINST THREE DIFFERENT BACTERIAL STRAINS

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Abstract: Antibiotic resistance is prevalent in today’s world, with harmful bacteria becoming resistant to a broad range of antibacterial agents. As a result, there is an urgent need in today’s society to discover a natural antibiotic. In herbal medicine, plant species with medicinal value are used to treat a variety of illnesses that are caused by microbial infections. Using the stem and leaves of Tinospora cordifolia, the researchers in this study evaluated the antibacterial activity of the two plant parts in terms of stem and leaves. Three bacterial strains were acquired in lyophilized form from the Microbial type culture collection centre in Chandigarh, India, and were revived using Nutrient agar. Ethanol was used to extract bioactive chemicals from the plant species. The extracts of both plant sections had antibacterial activity against all of the bacterial strains tested, although there was a significant variation in their antibacterial effectiveness between the two plant parts.

Keywords: Antibacterial, Ethanol, Resistance, Tinospora cordifolia

INTRODUCTION

Antibiotics with the fewest possible adverse effects are the most important need in today's world of increasing antibiotic resistance. It is only in the lap of mother nature that such antibiotics with minimal side effects may be discovered. More than 4000 years ago, herbal remedies have been employed in the treatment of a variety of illnesses, and their effectiveness has been documented in many Indian, Chinese, and Egyptian texts. According to the World Health Organization, herbal products are used by about 80% of the world's population. It has also been discovered that the globe has about 21,000 plant species, many of which are capable of curing a wide range of diseases. Herbal medicines are regarded as the safest medications since they have no or few adverse effects, which makes them extremely popular in many cultures across the world, including the United States. The second benefit of utilising herbal treatment is that it is not restricted to any one gender or age group (NHP, 2016). Antibacterial resistance is a significant source of worry today, and finding a pathogen-specific therapy that is effective while having no or minimum adverse effects would be beneficial. In herbal medicine, plants of various kinds are used to treat a variety of illnesses. Often, the whole plant is utilised, but occasionally just a part of the plant, such as the seed, stem, bark, leaf, flower, or roots, is used to treat the condition. The antibacterial properties of many plants have been shown. In addition to being an herbaceous vine that belongs to the family Menispermaceae and is used to cure a variety of diseases (Kumar et al., 2020), Tinospora cordifolia is also a medicinal plant. It is known by a variety of names, including Guduchi, Giloy, and others (Saha and Ghosh, 2012). The plant is a huge deciduous shrub with many extended knotted branches that grows widely across the landscape. This plant is also known as heart-leaved moonseed because its leaves are heart-shaped and the fruit is reddish in colour. Tinospora cordifolia flowers are unisexual, tiny in size, and look greenish-yellow when the plant is leafless, indicating that the plant is in bloom (Sinha et al., 2004). Male flowers are arranged in clusters, while female blooms are usually single in appearance. A variety of plant-derived active components have been extracted from the different sections of the plant's body, including alkaloids, steroids, glycosides, aliphatics, and diterpenoid lactones (Upadhyay et al., 2010). The miraculous cure for the covid-19 epidemic, Giloy, was recently thought to be the solution. However, owing to the small number of clinical research conducted on Tinospora, there is no conclusive proof of an anti-disease impact, which is why it is not prescribed as a medication. The Ministry of Health and Family Welfare of India has recommended the use of Guduchi (Giloy) Ghana Vati for the treatment of mild COVID19 symptoms (Tinospora, 2020). Such helpful plants must be investigated further, which is why we chose Tinospora cordifolia for this study in order to assess its antibacterial efficacy against three distinct kinds of bacterial strains.

It is the goal of the today's society to discover a natural antibiotic. The use of plant species with therapeutic potential in herbal treatment is used to treat a wide range of infections and infections-related diseases. These stems and leaves of the plant Tinospora cordifolia were studied for their antibacterial activity, and the results of this study were published in this article. Using Nutrient agar, three bacterial strains were obtained in lyophilized form from the Microbial type culture collection centre in Chandigarh, India, and then reactivated in

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the laboratory. Ethanol has been used to extract bioactive compounds from a variety of plant species. Extracts from both plant parts were shown to have antibacterial action against all bacterial strains tested, but there was a substantial difference in their antibacterial efficacy between the two plant sections.

MATERIALS AND METHODS

Sources of Test Bacterial strains
Microbial Type Culture Collection and Gene Bank, Chandigarh, provided a total of three bacterial cultures, all of which were preserved in liquid form. Using the temperature range indicated in Table 1, they were revived by incubating them in nutritional broth.

Sources of plant sample
Dr. Inam Mohammad, Taxonomist at DAV University in Jalandhar, Punjab, India, recognized the plant sample, which was obtained from the Saranpur town in the city of Jalandhar.

Preparation of Plant extracts
In order to evaluate the antibacterial effectiveness of the plant, it was split into two parts, including the stem and leaves, and both were shade-dried for 25 days to determine the antibacterial activity. For the aim of extraction, ethanol was employed as a solvent. One gram of each plant material was dissolved in ten milliliters of the solvent and allowed to settle for four consecutive days. The antibacterial activity of the chosen plant material was assessed after 24 hours, 48 hours, and 96 hours of exposure to the plant material. After being gathered, the plant samples were filtered using whatman number 1 filter paper and stored in the refrigerator at -81 degrees Celsius until being utilized (Alanis et al., 2005).

Phytochemical screening
Several biochemical assays were carried out in order to get a preliminary characterization of the phytochemicals contained in the plant extract, according to the methodology provided by Rao et al. (Rao and Kaladhar, 2014).

Antibacterial activity using well diffusion test
In order to assess the antibacterial activity of plant extracts, the well diffusion test was carried out (Magaldi et al., 2004). In order to cultivate the bacterial isolates, nutrient agar was employed as the medium. The microbial inoculum was injected onto the agar plate by distributing 100 μL of volume across the plate. With the assistance of an aseptic borer with a 6 mm diameter, another hole was created in the culture plate for the next step. Approximately 10 μL of the gathered plant samples were put into each well and incubated at 37 °C for 18 to 24 hours, depending on the species. A clear ruler was used to measure the zone of inhibition, which was measured in millimeters.

Statistical analysis
Students’ t-test was used to evaluate the results of all the trials, which were represented as the mean standard deviation of five separate experiments.

RESULTS AND DISCUSSION

_Tinospora cordifolia_ stem and leaves were obtained from Jalandhar, Punjab, and tested for antibacterial efficacy against three distinct bacterial strains in this study. Initial phytochemical screening was accomplished via the use of biochemical assays, the results of which are shown in Table 2. Analysis of _T. cordifolia_’s leaf extract revealed the presence of alkaloids, terpenes (a type of terpene), carbohydrates (a type of carbohydrate), flavonoids (a type of flavonoids), amino acids (a type of amino acid), glycosidases (a type of glycosidases), phenols (a type of tannin), and tannins (a type of tannin). A well diffusion test was also used to evaluate the antibacterial effectiveness of the plant extracts. Effectiveness of _Tinospora cordifolia_ leaves collected from Jalandhar, Punjab, against three bacterial species is shown in Figure 1, and the efficacy of _Tinospora cordifolia_ stem obtained from Jalandhar, Punjab, against three bacterial species is depicted in Figure 2, respectively. When the plant was exposed to the solvent for a longer period of time, the antibacterial activity of the plant was shown to be much higher. A difference between stem and leaf extracts was discovered, according to the results of the study. _Tinospora_ leaves and stems have been shown to have antibacterial activity against _Pseudomonas fluorescens_, _E. coli_, and _Staphylococcus aureus_, although the efficacy of the medicinal plant has been shown to vary depending on the bacterial strain tested in each study. Three bacterial species were tested against _Tinospora cordifolia_ stems and leaves collected from Jalandhar, Punjab, and the comparison results are given in figures 3-5. More strong bactericidal activity against _Pseudomonas fluorescens_ and _E. coli_ was observed in the stems of _Tinospora cordifolia_ than the leaves. _Staphylococcus aureus_, on the other hand, had the inverse behavior. In previous studies, the efficacy of the plant stem has been confirmed. For example, Jeyachandran and his colleagues investigated _Tinospora cordifolia_’s antibacterial activity against gram-positive and gram-negative bacteria, stating that the greatest activity was observed in plant stem extracts prepared in ethanol as opposed to extracts prepared in the other solvents (Jeyachandran et al., 2003). This study's results indicate that _Tinospora cordifolia_’s leaves and stem have antibacterial action, which varies depending on the bacterial strains tested and the amount of time allowed for the extract to interact with the solvent.
Figure 1: Antibacterial efficacy of *Tinospora cordifolia* leaves against three bacterial species.

Figure 2: Antibacterial efficacy of *Tinospora cordifolia* stem against three bacterial species.

Figure 3: Comparison between the inhibition zones of stem and leaves against *Pseudomonas fluorescens*. 
Figure-4: Comparison between the inhibition zones of stem and leaves against *E. coli*.

Figure-5: Comparison between the inhibition zones of stem and leaves against *Staphylococcus aureus*.

Table 1. Temperature and incubation time required for the growth of bacterial cultures.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Bacterial culture</th>
<th>Temperature required</th>
<th>Incubation time</th>
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</thead>
<tbody>
<tr>
<td>1.</td>
<td>MTCC-118 (<em>E. coli</em>)</td>
<td>37 °C</td>
<td>48 hours</td>
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<tr>
<td>2.</td>
<td>MTCC-103 (<em>Pseudomonas fluorescens</em>)</td>
<td>25 °C</td>
<td>48 hours</td>
</tr>
<tr>
<td>3.</td>
<td>MTCC-96 (<em>Staphylococcus aureus</em>)</td>
<td>37 °C</td>
<td>48 hours</td>
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</tbody>
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Table 2. Biochemical tests performed for the preliminary phytochemical compounds

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Biochemical test</th>
<th>Leaves</th>
<th>Stem</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.)</td>
<td>Alkaloids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>2.)</td>
<td>Terpenoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>3.)</td>
<td>Carbohydrates</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>4.)</td>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>5.)</td>
<td>Amino acids</td>
<td>+</td>
<td>-</td>
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</tbody>
</table>
CONCLUSION

According to the findings of the present research, the ethanol extracts of *Tinospora cordifolia* have antibacterial activity that varies somewhat depending on the bacterial strains. Based on greatest antibacterial activity shown by both the plant parts, it can be concluded that the both the sections of the medicinal plant may provide considerable protection against a wide range of bacterial infections. As a result, the research recommends that while developing antibacterial medicines from plants, the different parts to be taken into account at all times.

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REFERENCES


