Original Research Article

Study on Antibacterial and Antifungal Activities of Sterculia lychnophora Extracts

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ABSTRACT

The study investigated the crude extracts obtained from the seeds of the plant Sterculia lychnophora for antibacterial activity and antifungal activity. The antimicrobial properties were determined by Agar cup method using Escherichia coli, Staphylococcus aureus, Streptococcus pyogenes, Candida albicans, Klebsiella pneumoniae and Salmonella typhi as test organisms. The diameter of zone is measured and recorded in mm. Results obtained in the present study revealed that the four extracts of plant Sterculia lychnophora posses potential antibacterial activity against E. coli, S. aureus and S. typhi and antifungal activity against C. albicans. Whereas S. pyogenes and K. pneumoniae were found to be resistant against all extracts under study.

Keywords
Sterculia lychnophora, Antibacterial and antifungal activity

Introduction

Natural products are believed to be important source of new chemical substances which have potential therapeutic effects. Medicinal plants are extensively investigated both in vitro and in vivo to examine for their potential activities (Pancharee, 2010). The past three decades have seen a dramatic increase in microbial resistance to antimicrobial agents that lead to repeated use of antibiotics and insufficient control of the disease (Prashanth Kumar et al., 2006). Many secondary metabolites of plant

Chemicals are derived biosynthetically from plant primary metabolites. The secondary metabolites can be classified into several groups on the basis of their chemical classes (Angela et al., 2008). Plants contain many active compounds such as alkaloids, steroids, tannins, glycosides, volatile oils, fixed oils, resins, phenols and flavonoids which are deposited in their specific parts such as leaves, flowers, bark, seeds, fruits, root, etc (Prashanth Kumar et al., 2006).

Sterculia species have been extensively used in traditional medicine in the various countries where they are available. In the Solomon Islands, S. lepidoto-stellata is used to treat common cold. S. macrophilla is used
as an aphrodisiac in Java. In Australia, *S. quadrifolia* is used as peanut tree. In Guiana, *S. rubigenosa* used as fruit diet. *S. scaphigera*, in Thailand, is used to treat fever (Angela et al., 2008) and also shows the analgesic, anti-inflammatory, antioxidant and antiulcer activity (Dhage et al., 2013). *S. shillinglawii* is also said to treat fever and action is tonic (Angela et al., 2008). *S. tragacantha* is used in the treatment of boils, diarrhea, dyspepsia, fever, gonorrhea, snake bite, syphilis, tapeworm (Irvin, 1961), used to treat malaria (Denis et al., 2002) and Antibacterial activity (Orisakeye and Olugbade, 2012). *S. quinqueloba* shows anti-mycobacterial activity in *vitro* (Wilson et al., 2015).

For a long history Chinese people used the dried and ripe seed of *Sterculia lychnophora* is a well-known Chinese medicine. In Cambodia it is used as tea. This traditional drug is known for its remedy and prevention against pharyngitis. Since ancient times in China it has also been used for the treatment of tussis and constipation (Coles, 1981) and in India used to treat heavy menopausal bleeding (Sharma et al., 2012). The presence of flavonoids, terpenoids, phenolics, polysaccharides and histamines in other species of genus *Sterculia* were reported (Ru-Feng Wanga et al., 2003). Cerebrosides compounds showed neuroprotective effect which is present in *Sterculia lychnophora* (Ru-Feng Wang et al., 2013). In *Sterculia lychnophora* species showed secondary metabolites like tannins, saponins, flavonoids, phenols, terpenoids, alkaloids and lipids (Palve Anuradha et al., 2015). The *Sterculia lychnophora* seed has been claimed to treat cough, pain and clear phlegm and used as antipyretics (Pancharee, 2010). *In-silico* docking study showed that ligand 1 (soyacerebrosids 1) and ligand 2 (1-O-\(-\beta-D-\) glucopyranosyl-(2S,3R,4E,8Z)-2-((2-hydroxyoctadecanoyl) amido)-4,8-octadecadiene-1,3-diol) was having high E total score against 3IFN protein but better result was shown by the ligand 1 since it was having highest negative score compared to ligand 2 (Shetty et al., 2014). Thus the present study was aimed to examine the *Sterculia lychnophora* extract to show antibacterial and antifungal activity.

**Materials and Methods**

**Plant material collection**

The plant material Malva nut (*Sterculia lychnophora*) was bought from APMC market, Vashi, Navi Mumbai in May 2013. The plant was authenticated by Dr. Shankaran Potti comparing herbarium specimen at Department of Botany, Sathaye College, Mumbai. The damage less seeds were dried in hot air oven at 60°C-65°C for 12hrs and blended into powder, this powder of *Sterculia lychnophora* were used for aqueous and methanol extraction and phytochemical analysis.

**Extraction**

Extraction method used:

a. Hot Extraction (Soxhlet)

b. Cold Extraction

**Hot extraction**

The powder of *Sterculia lychnophora* which is ground using blender is used for the preparation of hot extracts using soxhlet apparatus. The condenser is supported by rubber-padded clamps attached to a 4 inch iron rod. Then condenser fits to thimble chamber. The thimble was prepared using Whatman’s filter paper no.1 where 20g *Sterculia lychnophora* (Malva nut) powder is placed into it. Then these thimbles were inserted into extraction tube of soxhlet apparatus which are connected other extraction tube with tube. The round bottom
flask with solvent is boiled using heating mantle and maintained the temperature at 100°C for water and 65°C for methanol. This process is carried out until the color of solvent overflowing from thimble chamber turns to colorless. Later the extract was allowed to cool and then evaporated on water bath to reduce the volume 20ml which is stored in borosil bottle and refrigerated at to -4°C. The hot extract used for antimicrobial assay and HPTLC analysis (Pimpliskar, 2004).

**Cold extraction**

For cold extraction 10g of seeds (*Sterculia lychnophora*) were weighed and crushed in mortar and pestle by adding 8ml of solvent (distilled water and methanol) and kept for 45mins. Then the extract was filtered using Whatman’s filter paper no.1 and in similar way again 6-8ml of solvent added and filtered. At the end volume was made to 20ml by adding solvent and stored in an air tight bottle and then refrigerated at -4°C and then this extract was then used for phytochemical tests and antimicrobial assay (Pimpliskar, 2004).

**Antimicrobial activity**

**Preparation of cotton swabs**

A supply of cotton wool swabs on wooden applicator sticks was prepared. They were sterilized in culture tubes in the autoclave.

**Experimental procedure**

The sterilized nutrient medium (20 ml) was poured in sterilized Petri dishes under aseptic condition and allowed to solidify on a plane surface.

All the extracts were first brought to room temperature and then used for bioassay test against the microorganisms by Agar cup method. The sterile agar plates were inoculated with test cultures (Table 1) of *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus pyogenes*, *Candida albicans*, *Klebsiella pneumoniae* and *Salmonella typhi* by dipping a sterile swab into inoculums. Excess inoculums were removed by pressing and rotating the swab firmly against the side of the tube, above the level of the liquid. The swab was streaked all over the surface of the medium three times, rotating the plate through an angle of 60° after each application. Finally the swab was passed round the edge of the agar surface. The inoculation was dried for a few minutes, at room temperature, with the lid closed. After drying four wells were made with the help of flame sterilized cork borer of 8mm diameter made of stainless steel. Four wells were then loaded with 80µl of four extracts with the help of micropipette and were kept for diffusion in refrigerator at 4°C for 45mins to 1hr. The plates were then placed for incubation at required temperature i.e. *Escherichia coli*, *Streptococcus pyogenes*, and *Salmonella typhi* was kept at 37°C while *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Candida albicans* at room temperature for 24hrs to 48hrs. The diameter of zone is measured and recorded in mm (Thamaraiselvi et al., 2012).

**Results and Discussion**

The antibacterial activity of *Sterculia lychnophora* against test organisms is shown in table 2. Results obtained in the present study revealed that the four extracts of plant *Sterculia lychnophora* posses potential antibacterial activity against *E. coli* (Fig. 1) and *S. aureus* (Fig. 2) *S. typhi* and (Fig. 3), antifungal activity against *C. albicans* (Fig. 4). Whereas *S. pyogenes* and *K. pneumoniae* were found to be resistant against all extracts under study. When tested by the
Agar cup method, the cold aqueous extract showed highest antibacterial activity against *S. typhi* more than 12mm, a significant activity was recorded against *S. aureus* and *E. coli* around 8–12mm, there was the lowest activity against *C. albicans* and no activity was showed against *S. pyogenes* and *K. pneumoniae*.

**Table.1** Microorganisms with strain number and suitable media at suitable incubation temperature were studied

<table>
<thead>
<tr>
<th>Sr.No.</th>
<th>Name of the cultures</th>
<th>NCIM</th>
<th>Growth media</th>
<th>Incubation Temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Escherichia coli</em></td>
<td>ATCC10536</td>
<td>Nutrient agar</td>
<td>37°C</td>
</tr>
<tr>
<td>2</td>
<td><em>Salmonella typhimurium</em></td>
<td>ATCC 23564</td>
<td>Nutrient agar</td>
<td>37°C</td>
</tr>
<tr>
<td>3</td>
<td><em>Staphylococcus aureus</em></td>
<td>2602</td>
<td>Nutrient agar</td>
<td>37°C</td>
</tr>
<tr>
<td>4</td>
<td><em>Streptococcus pyogenes</em></td>
<td>2608</td>
<td>Nutrient agar</td>
<td>37°C</td>
</tr>
<tr>
<td>5</td>
<td><em>Candida albicans</em></td>
<td>Clinical isolates</td>
<td>Sabouraud agar</td>
<td>Room temp.</td>
</tr>
<tr>
<td>6</td>
<td><em>Klebsiella pneumoniae</em></td>
<td>Clinical isolates</td>
<td>Nutrient agar</td>
<td>37°C</td>
</tr>
</tbody>
</table>

**Table.2** Antimicrobial effect of Sterculia lychnophora extracts (Zone of inhibition in mm)

<table>
<thead>
<tr>
<th>Extract</th>
<th><em>S.aureus</em></th>
<th><em>E.coli</em></th>
<th><em>S.typhi</em></th>
<th><em>S.pyogenes</em></th>
<th><em>K.pneumoniae</em></th>
<th><em>C.albicans</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Cold Water</td>
<td>++</td>
<td>++</td>
<td>+++</td>
<td>_</td>
<td>_</td>
<td>+</td>
</tr>
<tr>
<td>Hot water</td>
<td>_</td>
<td>+</td>
<td>_</td>
<td>_</td>
<td>_</td>
<td>+</td>
</tr>
<tr>
<td>Cold Methanol</td>
<td>++</td>
<td>++</td>
<td>_</td>
<td>_</td>
<td>_</td>
<td>+</td>
</tr>
<tr>
<td>Hot Methanol</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>_</td>
<td>_</td>
<td>+</td>
</tr>
</tbody>
</table>

**Key:**
- No zone of inhibition
- Zone of inhibition 8mm-12mm
- Zone of inhibition more than 12 mm

**Fig.1** Zone of inhibition of bioassay of cold and hot methanol and cold and hot aqueous extract of *Sterculia lychnophora* on *E.coli*

**Fig.2** Zone of inhibition of bioassay of cold and hot methanol and cold and hot aqueous extract of *Sterculia lychnophora* on *S.aureus*
Hot aqueous extract showed the activity against *E. coli* and *C. albicans* and there was no inhibition activity against *S. aureus*, *S. typhi*, *S. pyogenes* and *K. pneumoniae*. Cold methanol extract showed a significant growth against *S. aureus* and *E. coli* around 8-12mm, the lowest activity was against *C. albicans* of around 8mm and there was no growth against *S. typhi*, *S. pyogenes* and *K. pneumoniae*. Hot methanol extract showed a significant activity against *S. aureus* around 8-12mm, lowest activity was against *E. coli*, *S. typhi* and *C. albicans* around 8mm and no activity was showed against *S. pyogenes* and *K. pneumoniae*.

In conclusion, the study showed antimicrobial activity of extracts of seeds of *Sterculia lychnophora* against *S. aureus*, *E. coli* and *S. typhi* and the highest zone of inhibition was of cold water extract against *S. typhi* whereas *S. pyogenes* and *K. pneumoniae* were resistant against all extracts. Thus, the seeds of *Sterculia lychnophora* possess in vitro antibacterial and antifungal activity which can be used in treatment of infectious diseases.

**Reference**


Coles, M. 1981. Study of medicinal plants in Gloucester village, Ph.D. Thesis, Dept. of Botany, University of Sierra Leone, Sierra Leone.


