METHANOLIC EXTRACT OF *ACACIA NILOTICA* AND ANTIBACTERIAL ACTIVITY AGAINST HOSPITAL ISOLATES OF BENGALuru DISTRICT.  

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**Abstract** — Antibiotic resistance of bacteria now creating a great problem and the majority of the antibiotics are not effective against locally isolated bacteria. The clinical isolates from registered hospitals of Bengaluru district were analyzed or susceptibility against the methanolic extract of *Acacia nilotica*. Alkaloids exhibits moderate zone of inhibition, 6mm, 8mm, 5mm, 4mm, and 1mm. Where asflavonoids showed good zone of inhibition 7mm, 21mm, 19mm, 19mm and 21mm against *Staphylococcus aureus, Streptococcus faecalis, Pseudomonas aeruginosa, Bacillus subtilis* and *Escherichia coli* respectively.  

**Keywords**. Methanolic extract, Alkaloids, flavonoids, zone of inhibition.  

**Introduction**  
The plants that possess therapeutic properties or exert beneficial pharmacological effects on the animal body are generally designated as “Medicinal Plants”. It has now been established that the plants which naturally synthesis and accumulate some secondary metabolites, like alkaloids, glycosides, tannins, volatile oils and contain minerals and vitamins, possess medicinal properties. Herbal materials continue to be used as the primary source of medicines (Chitme et al., 2003). About 80% of the people in developing countries use traditional medicines for their primary health care (Kim, 2005). Antibiotics have undesirable side effects while the emergence of previously uncommon infections is also a serious medical problem (Marchese and Shito, 2001). Over 75% of the antibacterials in clinical use are of natural origin and most of them are obtained from fungal sources (Newman et al., 2003). *Acacia nilotica* is a tree 5-20 m high with a dense spherical crown, stems and branches usually dark to black colored, The plant has a wealth of medicinal uses. It is used for stomach upset and pain, the bark is chewed to protect against scurvy, an infusion is taken for dysentery and diarrhea. The pods are desirable as fodder for cattle, and the leaves, young shoots and young pods are thought to aid milk production.  

**Materials and methods**  
**Collection of Plants**  
The Mature fresh, disease free *Acacia nilotica* plant leaves sample was collected in locally from Siddarabetta, Tumkur district and Bangalore district. The collected samples were carefully stored in sterile Polythene bags without tightening and used for present study.  

**Sterilization and maintenance of plant materials**  
Mature fresh, disease free, Specimen of the plant has been deposited in Research lab; the leaves were washed thoroughly 2-3 times with running tap water, once with 70% ethanol and once with sterile water, shade-dried without any contamination. The dried leaves were then powdered by electric mill and stored in dry bottles for further use.  

**Preparation of crude extracts**  
The fine powder and the total mass were subjected to extraction by a hot percolation method with Methanol in soxhlet apparatus for 72 hrs. The temperature maintained was 40°C. Solvent extraction step was carried out for 16 hours and after extraction the extracts were concentrated by evaporation and stored at 4°C for further study. (Ashan M, et al., 1996, Kurian J C, 2003).  

**Collection and maintenance of the clinical bacterial cultures**  
Pathogenic bacteria used in the study were collected from different registered hospitals. The clinical samples like, upper respiratory tract infection, Urinogenital system and from blood samples. The bacteria selected in the present study were identified and certified by the registered hospitals. The collected bacterial samples were grown on nutrient agar media at 37°C and maintained at 4°C and till further user, The various extracts were tested against bacterial cultures: *Staphylococcus aureus, Streptococcus faecalis, Pseudomonas*
aeruginosa, Bacillus subtilis and Escherichia coli for antimicrobial activity.

**Antibacterial activity**

The antibacterial assay of methanolic extracts was performed by agar well diffusion method (Klastrup, 1975). The molten Mueller Hinton agar was inoculated with 100µl of the inoculums (1*10^6 CFU/ml) and poured into the petri plate. For agar well diffusion method, the agar plates were punched with wells (1mm) and the crude extracts (100µl throughout the study) were added to the wells. The plates were then incubated overnight at 37ºC. Microbial growth was determined by measuring the diameter of the zone of inhibition of each bacterial strain.

**Phytochemical analysis**

Phytochemical analysis of major Phyto-constituents of all the plant extracts were undertaken using standard qualitative methods as described by various authors. The plant extracts were screened for the presence of biologically active compounds like as steroids, terpenoids, alkaloids, flavonoids, coumarins, saponins, tannins, phenols, catechin, anthaquinione and quinine (C K Hindumathy, 2011).

**Result and discussion**

**Antibacterial activity of Acacia nilotica**

The antimicrobial activity of crude methanolic extracts of *Acacia nilotica*, was performed by the method described earlier and then analyzed for phytocompounds present in the (Table 1). Figure 2 shows the antibacterial activity in terms of zone of inhibition The methanolic extract of *Acacia nilotica*showed highest zone of inhibition against a gram + vecocci, *S. Aries*20 mm and least 16 mm against gram + vecocci *S. faecalis*, the other gram – vecocci *P. Aeruginosa*, *B. subtilis* and *E.coli*exhibits moderate zone of inhibition 18 mm, 17 mm and 19 mm respectively.

**Table 1: Antibacterial activity of Acacia nilotica**

<table>
<thead>
<tr>
<th>Organisms</th>
<th><em>S. aureus</em></th>
<th><em>St. faecalis</em></th>
<th><em>P. aeruginosa</em></th>
<th><em>B. subtilis</em></th>
<th><em>E. coli</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Zone of Inhibition (mm)</td>
<td>20</td>
<td>16</td>
<td>18</td>
<td>19</td>
<td>17</td>
</tr>
</tbody>
</table>

*S. aureus* = *Staphylococcus aureus*, *St. faecalis* = *Streptococcus faecalis*, *P. aeruginosa* = *Pseudomonas aeruginosa*, *B. subtilis* = *Bacillus subtilis*, *E. coli* = *Escherichia coli*

**Phytochemical analysis of Acacia nilotica**

The Phytochemical analysis of crude methanolic extracts of *acacia nilotica* performed by the method described earlier and then analyzed for phytocompounds like steroids, terpenoids, alkaloids, flavonoids, coumarins, saponins, tannins, phenols, catechin, anthaquinione and quinine preliminary analyzed and present in the (Table 2). Most of the secondary metabolites were identified in the methanolic extracts. Extracts of *acacia nilotica*indicates presence of terpenoids, alkaloids, saponins, tannins and phenolic compounds are noticed.

**Table 2: Phytochemical analysis of Acacia nilotica**

<table>
<thead>
<tr>
<th>PLANTS</th>
<th><em>Acacia nilotica</em></th>
<th>Conten ts (%)</th>
<th>0.181</th>
<th>13.7</th>
<th>16.7</th>
<th>0.1</th>
<th>11.8</th>
<th>0.201</th>
<th>0.0</th>
</tr>
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**Content of phytocompounds of Acacia nilotica**

In Table 3 shows the content of each phytocompound such as alkaloids, flavonoids, saponins, tannins, phenolic compounds, terpenoids and Anthraquinone. Figures 1 represent the presence of each phytocompound in methanolic extracts of *acacia nilotica*.

**Table 4: Antibacterial activity of different phytocompounds**

<table>
<thead>
<tr>
<th>Organisms</th>
<th><em>S. aureus</em></th>
<th><em>St. faecalis</em></th>
<th><em>P. aeruginosa</em></th>
<th><em>B. subtilis</em></th>
<th><em>E. coli</em></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Phytochemicals</strong></td>
<td>Alkaloids</td>
<td>Flavonoids</td>
<td>Saponins</td>
<td>Tannins</td>
<td>Phenolic compounds</td>
</tr>
<tr>
<td><em>S. aureus</em></td>
<td>06</td>
<td>19</td>
<td>00</td>
<td>00</td>
<td>00</td>
</tr>
<tr>
<td><em>St. faecalis</em></td>
<td>04</td>
<td>11</td>
<td>00</td>
<td>00</td>
<td>00</td>
</tr>
<tr>
<td><em>P. aeruginosa</em></td>
<td>03</td>
<td>19</td>
<td>03</td>
<td>00</td>
<td>00</td>
</tr>
<tr>
<td><em>E. coli</em></td>
<td>00</td>
<td>19</td>
<td>00</td>
<td>00</td>
<td>00</td>
</tr>
</tbody>
</table>
Figure 3: Antibacterial activity of different phytocompounds (methanol extract) of *Acacia nilotica*, against *S.aureus, St.faecalis, P.aeruginosa, B.subtlis, E.coli*

The separated bioactive compounds of methanolic extract of *acacia nilotica* Table 4 and figure 3 indicate the presence of Alkaloids, Flavonoids, saponins, Tannins, Terpenoids, Anthraquinone. The antibacterial activity of each bioactive compounds reveals that the alkaloids showed 06, 08, 05,04 and 01 mm zone of inhibition against *S.aureus, St.faecalis, P.aeruginosa B.subtlis and E.coli*. Flavonoids exhibit a very good antibacterial activity showed the highest 21 mm of zone against *E.coli* and *St.faecalis*, 19 mm against *S.aureus, P.aeruginosa* and *E.coli*. Saponins of *acacia niloticae* are not very effective and exhibit a little zone of inhibition against only in *P.aeruginosa*. The other bioactive compounds tannins, terpinoids and anthraquinone have not shown their existence.

**Conclusion**

Alkaloids and flavonoids by methanolic extract of *Acacia nilotica* showed zone of inhibition against hospital isolates. Flavonoids are promising bioactive compounds against *S.aureus, St.faecalis* and gram negative *P.aeruginosa, B.subtlis* and *E.coli*.

**References**

2. C.K. Hindumathy; Invitro Study of Antibacterial Activity of CymbopogonCitrus; *World Academy of Academy of Science, Engineering and Technology* 74 2011; 193-197.