SUCCESSFUL EVALUATION OF ANTIMICROBIAL ACTIVITY OF GREEN TEA AGAINST VARIOUS PATHOGENIC ORGANISMS

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ABSTRACT
Green tea, a beverage consumed around the world, has progressed from casual beverage to medicinal powerhouse through the centuries. Green tea, made from the leaves of the \textit{Camellia sinensis} plant. In this investigation attempt was made to check antimicrobial activity of green tea against eleven different pathogenic bacteria; \textit{E.coli}, \textit{S.aureus}, \textit{Shigella}, \textit{S.paratyphi A}, \textit{S.paratyphi B}, \textit{S.pyogen}, \textit{S.typhimurium}, \textit{P.mirabilis}, \textit{P.vulgaris}, \textit{C.diptheria}, \textit{K.pneumoniae} etc. The method used to check antimicrobial activity was Kirby bauer method. Two types of extracts were used to assessed antimicrobial activity that is Boiled and overnight mixed extract. Green tea is showing good antimicrobial activity against all pathogens.

Index Terms: Antimicrobial activity, Kirby bauer method, Green tea.

I. INTRODUCTION
Tea is a very popular drink world-wide. It is produced from the plant \textit{Camellia sinensis}, which is grown in at least 30 countries, and grows best in certain tropical and subtropical regions Green tea, a beverage consumed around the world, has progressed from casual beverage to medicinal powerhouse through the centuries. There are several different types of tea available on the market, including green, black, white, herbal, and oolong. Green tea, made from the leaves of the \textit{Camellia sinensis} plant, is unfermented; the freshly plucked tea-leaf is steam blasted in perforated drums or cooked in iron pans, denaturing its oxidizing enzymes.

Green tea has been shown to have antimicrobial effects against a variety of gram positive and gram negative bacteria. These antimicrobial effects will be discussed in more detail later in this paper.
II. MATERIAL AND METHODS

A. Sample
Green tea were purchased from local market, Ratnagiri, Maharashtra, India. Unprocessed green tea were selected for study as it may contain natural ingredients.

B. Storage
Sample were stored in dry place and away from sunlight, so as to avoid physicochemical properties of green tea.

C. Test sample preparation
Test samples were prepared for analysis by two methods. 1 gm of Green tea were added to 10 ml of sterile saline. This mixture were kept for overnight incubation and second sample were prepared by boiling the same mixture. Both the supernatant were used as sample for further analysis.

D. Cultures used
Total 11 samples were selected for study. Pure cultures were obtained from culture collection laboratory of Biological Sciences Department, Gogate Jogalekar College, Ratangiri, Maharashtra, India. While selecting cultures care was taken that pathogens which causes infection to all physiological tracts of human body were selected. Namely, E.coli, S.aureus, Shigella, S.paratyphi A, S.paratyphi B, S.pyogen, S.typhimurium, P.mirabilis, P.vulgaris, C.diptheria, K.pneumoniae were selected.

E. Testing for antimicrobial activity by agar well diffusion method
Petriplates containing 20ml Muller Hinton medium were seeded with 24hr culture of bacterial strains. (E.coli, S.aureus, Shigella, S.paratyphi A, S.paratyphi B, S.pyogen, S.typhimurium, P.mirabilis, P.vulgaris, C.diptheria, K.pneumoniae). Wells were cut and 20 µl of the both the samples were added. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well.

F. Minimum Inhibitory Concentration (MIC)
Various concentrations of Green tea samples were Prepared using suitable stock and diluents (as mentioned in the following tables). Then 0.1 ml of respective cultures were added to each tube and all tubes were incubated at appropriate temperature (37°C).
Stock | St. Nutrient Broth containing 20% green tea (both overnight and boiled samples were prepared separately)
---|---
Diluent | St. Nutrient Broth
Range | 2-20% green tea at intervals of 2%
Total volume | 5 ml
Culture | 24 hrs old culture of *E.coli*, *S.aureus*, *Shigella*, *S.paratyphi A*, *S.paratyphi B*, *S.pyogen*, *S.typhimurium*, *P.mirabilis*, *P.vulgaris*, *C.diptheria*, *K.pneumoniae* (Optical density adjusted to 0.1 at 530nm)
Incubation period and time | 37°C /24 hrs

After performing MIC, each respective sample were plated to Sterile Nutrient agar plate. And further analysed for growth of organisms. CFU (colony forming Unit) were counted for each concentration.

### III. RESULT AND DISCUSSION

#### A. Results of agar well diffusion method

<table>
<thead>
<tr>
<th>CULTURES USED</th>
<th>SET 1</th>
<th>SET 2</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>BOILED GREEN TEA (100MG/ML)</strong></td>
<td><strong>OVERNIGHT GREEN TEA (100MG/ML)</strong></td>
<td><strong>BOILED GREEN TEA (100MG/ML)</strong></td>
</tr>
<tr>
<td><em>E.coli</em></td>
<td>23mm</td>
<td>20mm</td>
</tr>
<tr>
<td><em>S.aureus</em></td>
<td>30mm</td>
<td>-</td>
</tr>
<tr>
<td><em>Shigella</em></td>
<td>20mm</td>
<td>15mm</td>
</tr>
<tr>
<td><em>S.paratyphi A</em></td>
<td>15mm</td>
<td>-</td>
</tr>
<tr>
<td><em>S.pyogen</em></td>
<td>18mm</td>
<td>15mm</td>
</tr>
<tr>
<td><em>S.typhimurium</em></td>
<td>-</td>
<td>20mm</td>
</tr>
<tr>
<td><em>P.mirabilis</em></td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>P.vulgaris</em></td>
<td>26mm</td>
<td>22mm</td>
</tr>
<tr>
<td><em>C.diptheria</em></td>
<td>24mm</td>
<td>20mm</td>
</tr>
<tr>
<td><em>K.pneumoniae</em></td>
<td>-</td>
<td>20mm</td>
</tr>
<tr>
<td><em>S.paratyphi B</em></td>
<td>-</td>
<td>17mm</td>
</tr>
</tbody>
</table>
Fig. 1: Petriplate showing zone of inhibition with *E.coli*.

Fig. 2: Petriplate showing zone of inhibition with *S.aureus*.

Fig. 3: Petriplate showing zone of inhibition with *Shigella*.

Fig. 4: Petriplate showing zone of inhibition with *S.paratyphi A*. 
Fig. 5: Petriplate showing zone of inhibition with *S.pyogen*.

Fig. 6: Petriplate showing zone of inhibition with *S.typhimurium*.

Fig. 7: Petriplate showing zone of inhibition with *P.mirabilis*.

Fig. 8: Petriplate showing zone of inhibition with *P.vulgaris*.
IV. DISCUSSION

In conclusion, this experiment has shown that green tea in general shows antimicrobial properties. Almost all of the organisms tested showing susceptibility for green tea. All these bacterial species selected comprises all physiological tracts as per the infections are concerned. Hence, we can make conclusion that green tea is effective for overall health of an individual.
V. REFERENCES


