COMPARATIVE STUDY OF BANANA PEEL AND BLACK TEA EXTRACTS AGAINST PERIODONTITIS CAUSING Staphylococcus aureus

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Abstract: Black tea and banana (Musa paradisiaca) is to contribute natural remedies and useful in curing bacterial infections. Banana plants (Musa paradisiaca) are grown in worldwide and all parts of it have medicinal application. The extract of banana peels and black tea was detected from phytochemical analysis. These can also be applied for dental caries and are of great significance in therapeutic treatments. This study aimed to compare the activity of banana peel and black tea against periodontitis causing S.aureus.

Keywords – Black tea and banana peel extract, Fluoride, Phytochemical, Antibacterial, Agar well diffusion.

INTRODUCTION
Various plants and their products have been in use for their medicinal value since ancient times (Meckes et al.,1995 ).Natural products have been an integral part of the ancient traditional medicine system (Sarkar et al.,2007). Use of herbal remedies for prevention and cure of ailments is of increasing interest due to the superiority and efficiency of activity provided by phytoconstituents in herbs and undesirable effects of modern medicine. Plants are still major sources of the foods worldwide and phytochemicals can play a significant role in modern food and pharmaceutical industries (Asif Ahmed Kibria et al.,2019).

Periodontitis, the multifactorial diseases are mainly derived by interaction between invasions of causative bacteria and host immune response of varied degrees (Jenabian et al.,2012). Usually gram negative bacteria play a central role and gram positive bacteria are rare. Among, Staphylococcus aureus plays a rare role in causing Periodontitis as compared to gram negative organism.
The unripe peel of banana and of the leaves are taken in dysentery and diarrhea and used for treating malignant ulcers (Girish et al., 2008). The waste material of banana peel has medicinal properties and various bioactive compounds flavonoids, tannins, alkaloids, glycosides and terpenoids are present in banana peel which exerts a pharmacological effect, especially antioxidant, antibiotic, anti-inflammatory (Imam et al., 2011 and Chabuck et al., 2013).

Black tea is more oxidised than all other types of teas. It contains antioxidants and other substances that might help protect the heart and blood vessels (Diane et al., 2007). Tea is one of the most popular and liked beverages worldwide. Tea is known to possess various biological properties (Daljit Singh Arora et al., 2007). The biological properties of tea and coffee are accredited mainly to their polyphenolic contents (flavonoids, catechins, tannins). The ability of these components to scavenge reactive oxygen species impart them potent antioxidant activities in vitro and in vivo. In the oral tract, S. aureus has been associated with dentoalveolar infections, and oral mucosal lesions (Ga-Yeon Kim et al., 2015).

MATERIALS AND METHODS

BACTERIAL CULTURE

The bacterial culture Staphylococcus aureus were obtained from Microbiological Laboratory of Kovai Medical Center and Hospital, Coimbatore. Those bacterial cultures were maintained by regular subculturing on nutrient agar slants and stored at 4°C.

SAMPLE COLLECTION AND PREPARATION

The banana peels used were collected from banana (Musa paradisiacal) brought from market. They were dried in hot air oven at 120°C for 3h 40min and ground into powder with mechanical blender and sieved. The obtained powdered samples were soaked in 70% methanol, ethanol, hexane and aqueous are placed on the shaker for 72hrs at room temperature. The sample and extracts used for the extraction is 1:3(ratio) respectively.

The black tea is collected from market and blended into fine powder. Then soaked in 70% methanol, ethanol, hexane and aqueous and placed on the shaker for 72hrs or three days at room temperature. The sample and solvent was used for the extraction is 1:3 ratio respectively.

PHYTOCHEMICAL ANALYSIS OF EXTRACTS

Qualitative phytochemical analysis of the crude extracts of the banana peel and black tea collected were determined as follows

1) **Alkaloids**: To 2ml of extract add few ml of dil. Hcl and filtered and add few drops of Mayer’s reagent (saturated aqueous solution of picric acid). Yellow precipitate confirms presence of alkaloids.

2) **Flavonoids**: To the few ml of extracts, few drops of NaOH and few ml of dil. Hcl was added. Yellow color is obtained when NaOH added become colorless after adding dil. Hcl which confirms the presence of flavonoids.

3) **Saponins**: To 2 ml of dis H2O add 5ml of extract. If foam persists for 10 minutes it indicates the presence of saponins.

4) **Tannin**: Few drop of FeCl3 (10%) solution was added with 2-3ml of extract and greenish grey or dark blue color indicates positive.
5) Phenols: 2ml of extract mixed to 5% FeCl₃ solution and deep blue color indicates presence of phenols.

ANTIBACTERIAL ACTIVITY
The banana peel and black tea extracts were used to test the inhibitory activity against the Periodontitis causing *Staphylococcus aureus*. The formation of clear zone was used to determine the efficacy of banana peel and black tea extracts. Bacterial inoculums were grown in nutrient broth for overnight or 24hrs.

SCREENING OF ANTIBACTERIAL ACTIVITY USING WELL DIFFUSION METHOD
The antimicrobial activity of *Musa paradisiaca* (Banana) peel and black tea extracted with different solvent (methanol, ethanol, hexane, aqueous) were tested against Periodontitis causing *Staphylococcus aureus* was measured using the well diffusion method. Muller Hinton Agar plates were prepared. Log phase test specimen *Staphylococcus aureus* were swabbed on the Muller Hinton Agar plate surface using the sterile cotton swab. Wells were made with the sterile gel puncture (cork borer) on Muller Hinton Agar plate and the 550mg/ml of the *Musa paradisiaca* and 50mg/ml of black tea solvent extracts were loaded on to the wells with MIC. The plates were incubated at 37°C for 24hrs and zone of inhibition around the well was measured in millimeters (mm).

MINIMUM INHIBITORY CONCENTRATION
Minimum inhibitory concentration (MIC) is defined as the lowest concentration of antimicrobial that will inhibit the visible growth of microorganism after overnight incubation. Bacterial inoculums were grown in nutrient broth for overnight. The extract of *Musa paradisiaca* (banana) peel and black tea of different solvent were subjected into the determination of MIC using the tube dilution technique. The ethanolic extract of banana peel and methanolic extract of black tea were taken. A double fold serial dilution was made using Muller Hinton Broth (MHB). The concentration obtained for Black tea was 50mg/ml, 25mg/ml, 10mg/ml, 7mg/ml, 3.5mg/ml, 3mg/ml, 2.5mg/ml, 2mg/ml, 1.6mg/ml and the concentration obtained for banana peel was 550mg/ml, 520mg/ml, 500mg/ml, 480mg/ml, 250mg/ml, 120mg/ml, 60mg/ml, 32mg/ml and 8mg/ml. Equal volume of extract and Muller Hinton broth (2ml) was dispensed into sterilized test tubes. A quantity (0.1ml) of standardized inoculums (0.9×10⁸ cfu/ml) was added to each of the test tubes which were incubated aerobically at 37°C for each 24hrs. The lowest concentration of the extracts which inhibited microbial growth (no turbidity) was recorded as the (MIC) minimum inhibitory concentration.
RESULT AND DISCUSSION

PHYTOCHEMICAL ANALYSIS
The phytochemical active compounds were qualitatively analyzed from the four types of extracts such as methanolic, ethanolic, hexane and the aqueous of both *Musa paradisiaca* (banana) peel and black tea.

**TABLE: 1 Extracts of *Musa paradisiaca* and Black tea**

<table>
<thead>
<tr>
<th>Phytochemical</th>
<th>Methanolic Extract</th>
<th>Ethanol Extract</th>
<th>Hexane Extract</th>
<th>Aqueous Extract</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Banana peel</td>
<td>Black tea</td>
<td>Banana peel</td>
<td>Black tea</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Phenol</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

ANTIBACTERIAL ASSAY
The antibacterial assay for the eluted fractions was measured using well diffusion method. The zone of inhibition was measured and tabulated.

**TABLE: 2**

<table>
<thead>
<tr>
<th>Solvents</th>
<th><em>Musa paradisiaca</em> (banana peel)</th>
<th>Black tea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methanolic extract</td>
<td>3.66±1.24</td>
<td>5.66±1.24</td>
</tr>
<tr>
<td>Ethanol extract</td>
<td>3.66±1.24</td>
<td>3.66±0.471</td>
</tr>
<tr>
<td>Hexane extract</td>
<td>3.66±0.471</td>
<td></td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>3.66±0.471</td>
<td></td>
</tr>
<tr>
<td>DMSO (Negative control)</td>
<td>3.66±0.471</td>
<td></td>
</tr>
<tr>
<td>Streptomycin (Positive control)</td>
<td>7±0.816</td>
<td>6.33±1.247</td>
</tr>
</tbody>
</table>

The ethanolic extract of banana peel (*Musa paradisiaca*) and methanolic extract of black tea showed effective inhibition against *Staphylococcus aureus*. 
MINIMUM INHIBITORY CONCENTRATION
The minimum inhibitory concentration is the lowest concentration of extract to kill or inhibit the growth of microorganisms. The MIC was performed for Staphylococcus aureus on ethanolic extract of Musa paradisiaca (banana) peel and methanolic extract of black tea. The turbidity is checked to identify the MIC. The minimal inhibitory concentration of Ethanol extract of banana peel is 500mg/ml and methanolic extract of black tea is 7mg/ml.

TABLE: 3

<table>
<thead>
<tr>
<th>Extracts</th>
<th>MIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Banana peel(Ethanol extract)</td>
<td>500mg/ml</td>
</tr>
<tr>
<td>Black tea(Methanolic extract)</td>
<td>7mg/ml</td>
</tr>
</tbody>
</table>
The preliminary phytochemical screening showed *Musa paradisiaca* (banana) peel and black tea contain some secondary metabolites such as alkaloids, phenols, flavonoids, Saponins and tannins. Secondary metabolites present in plants have been reported by Rabe (2000) to be responsible for the antimicrobial activity associated with some ethnomedicinal plants.

Black tea and banana (*Musa paradisiaca*) is to contribute natural remedies and useful in curing bacterial infections. Where, the black tea contributes fluoride equal to the adequate amount as 0.05mg F/kg body weight/day. Where fluoride in tea could be beneficial for the prevention of dental caries but excessive intake is not by children at the age of risk to dental caries should be avoided. The black tea is only effective when it is not added with milk, sugar and it has active compounds only when it is taken alone without any additives.

The antibacterial efficacy of both banana peel and black tea extracts were detected with measuring the zone of inhibition. The positive control used is Streptomycin and DMSO as negative control.

**CONCLUSIONS**

The study showed the difference that the ethanolic extract of banana peel (*Musa paradisiaca*) was effective than its methanolic extract where the methanolic extract of black tea was effective than its ethanolic extract. The ethanolic extract of banana peel shown highest MIC of 7mg/ml and the MIC of black tea methanolic extract against *S.aureus* was 500mg/ml. In this present study, banana peel (*Musa paradisiaca*) and black tea extract to screen the efficacy on periodontitis causing *S.aureus* as a comparative study and revealed both samples are effective on *S.aureus* but of different solvents.

**ACKNOWLEDGMENT**

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**CONFLICTS OF INTEREST**

The authors have no conflicts of interest to publish this research article in this journal.
REFERENCE


