Anticataract Activity of Piper Betle on goat eye lenses

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ABSTRACT
The present work was aimed to determine the efficacy of different ethanolic extract of piper betle leaves for anticataract activity by using glucose induced goat eye lenses as in-vitro models. In this study goat lenses were subjected to induce cataract by using glucose. It was confirmed that phytoconstituents of Piper Betel basically having antioxidant properties and it was responsible for effective anticataract activity. Photographic examination of the eyes showed that action with different extracts of piper betle leaves retarded the development of lens opacification.

KEYWORDS: Anticataract, Piper Betle, Antioxidant.

1. INTRODUCTION
Cataract, a visual destruction causing disorder in lens transparency occurs mainly due to opacification or optical dysfunction of lens. It reduces the amount of incoming light and results in deterioration of the vision ¹¹. ²². Cataract is mainly responsible for almost 80% of blindness cases in India ³³. There is sufficient evidence that oxidative stress plays a role in the mechanisms of cataract genesis, there is an increasing interest in developing suitable antioxidant nutrients, plant origin that could be effective in delaying or preventing the formation of cataract ⁴⁴. P. betle leaves have been used in preparation of medicine due to its antioxidant and antimicrobial properties. The ethanolic extract of leaves has antimicrobial activity against human pathogens including both gram-positive and gram-negative bacteria. By phytochemical screening it was found that carbohydrate, protein, polyphenolic compounds, flavonoid and alkaloids were present in its leaves ⁵⁵, ⁶⁶.

1.1 Plant Summary: A green leafy plant growing as a ground cover or small climber, very similar in growth habits to pepper. The plant growing environment prefers warm, humid conditions, but can tolerate some drought. The betel leaf is used in a number of traditional remedies for the treatment of stomach ailments, infections, and as a general tonic. It is often chewed in combination with the betel nut (Areca catechu), as a stimulatory. Some evidence suggests that betel leaves have immune boosting properties as well as anti-cancer properties.¹⁵
Synonyms: Chavica Beta. Artanthe Hixagona
Kingdom: Plantae
Order: Piperales
Family: Piperaceae
Genus: Piper  
Species: P.Petle  
Test: Pungent tasting and warming.  
Division: Magnoliphyta  

2. MATERIALS & METHODS:  
Piper betle Leaves collection & Authentication: Leaves were collected and washed with several times with distilled water to remove the traces of foreign material. The collected leaves were air dried at room temperature kept to shed dry until constant weight was attained.  
Drugs: Ascorbic Acid received from Fine lab Chemicals, Mumbai  
Ethanolic extract of piper betle Leaves:  
About 200 gm dried leaves powdered and soaked with 250 ml ethanol to obtain extract. Extract kept evaporating to dry.  
Goat eye lenses: Goat eye balls were obtained from the slaughter house and transferred to the laboratory in ice box. Lenses removed from eyeballs by intracapsular extraction method and stored at cool condition.  
Preparation of Lens Culture: Removed lenses were incubated in artificial aqueous humor (NaCl - 140 mM, KCl - 5 mM, MgCl2 - 2 mM, NaHCO3 - 0.5 mM, NaH(PO4)2 - 0.5 mM, CaCl2 - 0.4 mM and Glucose 5.5 mM) at room temperature and pH 7.8 for 72 h. Cetrimide were added as to the culture media to prevent bacterial contamination. Glucose in a concentration of 55 mM was used to make cataract [7].  

3. RESULTS & DISCUSSION:  
3.1 Phytochemical investigation  
Phytochemical investigation tests shown that main chemical compounds found in piper beetle extracts are alkaloids. Current studies on results showed that these compounds possess anticataract activity. 

Table 1. Evaluation of phytochemicals results

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Chemical constituents</th>
<th>Test for Ethanolic extract</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>Mayers Test , Hagers Test</td>
<td>+++</td>
</tr>
<tr>
<td>2</td>
<td>Glycosides</td>
<td>General test for glycosides</td>
<td>++</td>
</tr>
<tr>
<td>3</td>
<td>Cardiac Glycoside</td>
<td>Legal’s test , Baljet Test</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>Terpenoids</td>
<td>Salwosky Test</td>
<td>+++</td>
</tr>
<tr>
<td>5</td>
<td>Tannins</td>
<td>FecI3 test</td>
<td>++</td>
</tr>
<tr>
<td>6</td>
<td>Saponins</td>
<td>Frothing Test</td>
<td>++</td>
</tr>
</tbody>
</table>

+ Presence +++ intense Presence –absent
3.2 Evaluation of Ethanolic Extract of Piper Betle leaves

Table 2. Evaluation of Ethanolic Extract of Piper Betle leaves

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Parameter</th>
<th>Observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Organoleptic</td>
<td>Light brown solution</td>
</tr>
<tr>
<td>2</td>
<td>pH</td>
<td>7.4</td>
</tr>
<tr>
<td>3</td>
<td>Viscosity</td>
<td>40 cps</td>
</tr>
<tr>
<td>4</td>
<td>Clarity</td>
<td>Clear brownish solution</td>
</tr>
</tbody>
</table>

3.3 Experimental design

Dried Ethanolic extracts of p. beetle was prepared in concentration of 500 µg/ml by using distilled water and used for different groups as,

Table 3. Experimental Study Groups

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Study Groups</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Normal Control (F1)</td>
<td>Lens +Glucose (5.5 mM)</td>
</tr>
<tr>
<td>2</td>
<td>Negative Control (F2)</td>
<td>Lens + Glucose (55mM)</td>
</tr>
<tr>
<td>3</td>
<td>Ascorbic acid (F5)</td>
<td>Lens + Glucose (55mM) + Ascorbic acid</td>
</tr>
<tr>
<td>4</td>
<td>Ethanolic Leaf Extract (F4)</td>
<td>Lens + Glucose (55mM) + Ethanolic Leaf Extract</td>
</tr>
</tbody>
</table>

3.4 Photographic Evaluation

Figure 1. Photographic evaluation
Evaluation done by placing the experimental lenses on wired mesh with their back region touches to mesh and observed for opacification. From the results it was observed, cataract induced lenses having opaque nature. Ethanolic extracts shows better anticataract activity as compared to ascorbic acid. Photographic observation shows that piper beetle leaves ethanolic extracts shows anticataract activity invitro in goat lenses.

4. CONCLUSION:
The present work it was concluded that, piper beetle leaves containing phytoconstituents used as antioxidants. These antioxidants shows anticataract activity and it was verified by using invitro goat lenses.

5. ACKNOWLEDGEMENT: The authors would like to thank Sudhakarrao Naik Institute of Pharmacy, Pusad. For providing the facilities required in preparation of this research article.

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