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Antibacterial activity of Putranjiva Roxburghii wall: A medicinal plant

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Abstract---Medicinal plants can be defined as plants that possess therapeutic properties or exert a beneficial pharmacological effect on the human or animal body. *P. roxburghii* is a good moderate sized evergreen medicinal tree with pendent branches and the leaves are simple and alternately arranged. The aim of this study was to screen and determine significant antibacterial activity of dry seeds of *P. roxburghii*. The ethanol extract of *P. roxburghii* seed showed highest zone of inhibition in all the bacterial strains tested *Staphylococcus aureus* (G+) (16mm). *Streptococcus mutans* (G+) (19mm) *Bacillus subtilis* (G+) (20mm). *E.coli* (G-) (21mm). *Klebsiella pneumonia* (G-) (14mm) *Proteus vulgaris* (G-) (16mm). The acetone extract of *P. roxburghii* seeds showed a minimum zone of inhibition against the bacterial pathogens. They are *Staphylococcus aureus* (G+) (11mm). *Streptococcus mutans* (G+) (8mm) *Bacillus subtilis* (G+) (14mm). *E.coli* (G-) (15mm). *Klebsiella pneumonia* (G-) (13mm) *Proteus vulgaris* (G-) (15mm). The aqueous extract of *P. roxburghii* seeds showed the lowest zone of inhibition against the bacterial strains. They are *Staphylococcus aureus* (G+) (13mm). *Streptococcus mutans* (G+) (9mm). *E.coli* (G-) (14mm).

Keywords---Putranjiva roxburghii wall, antibacterial activity.

Introduction

P. roxburghii belongs to the family *Putranjivaceae* is a vulnerable species. Lucky bean tree, child's amulet tree, child life tree are some of its common names in English. Initially *P. roxburghii* plant existed in the *euphorbiaceae* family and it is distributed mainly in the tropics of various types of vegetation and habitats being one of the largest, most complex and diverse families of angiosperms. It has recently been divided into four families, according to classification systems based on molecular phylogeny *euphorbiaceae sensu stricto*, *phyllanthaceae*, *putranjivaceae* and *picrodendraceae*. (Wurdack, *et al.*, 2005). The famous botanist Roxburgh recognized this plant and accordingly the plant is named as *P. roxburghii*.

P. roxburghii seeds extract exhibited significant antibacterial activities as well as it provided much hypoglycemic properties. (Varma, *et al.*, 2011). *P. roxburghii* Wall. has been claimed to possess antibacterial properties by many investigators. Antimicrobial resistance is seriously threatened human public health and new antimicrobial agents should be needed. (Minj, *et al.*, 2016). Ayurvedic classics have highlighted the importance of *P. roxburghii* plant as one which helps in procreation, burning sensation, elephantiasis and to treat eye diseases (Unnikrishnan, *et al.*, 2015). *P. roxburghii* is used to anti-inflammatory and antioxidant, antinociceptive, anti-inflammatory properties (Reanmongkol, *et al.*, 2009). The famous botanist Roxburgh recognized this plant and accordingly the plant is named as *P. roxburghii*. It is utilized as an active healing plant and also used to separate some organic chemicals as well as this plant is used in pharmacognosy and therapeutic actions.



Fig 1. Morphology and Dried seeds of *P. roxburghii*

Material and Methods

Sample collection and extract preparation

The seeds of *P. roxburghii* Wall. (Figure 1) were collected from Lalbag Botanical Garden Bengaluru, India and authenticated (TBGT 91090) in JNTBGRI

Thiruvananthapuram Palode, Kerala, India. The collected seeds were shade dried and ground in to fine powder and used for the preparation of ethanol, acetone and aqueous extracts.

Bacterial strains

Different bacterial strains viz *S. aureus*, *S. mutans*, *B. subtilis*, *K. pneumoniae*, *P. vulgaris*, and *E.coli*, were purchased from Microbial Type Culture Collection and Gene Bank (MTCC) Chandigarh. The bacterial strains were maintained on nutrient agar and used for the antibacterial activity.

Nutrient broth preparation

Pure culture from the plate were inoculated into nutrient agar plate and sub cultured at 37°C for 24 h. Inoculum was prepared by aseptically adding the fresh culture into 2 ml of sterile 0.145 mol/L saline tube and the cell density was adjusted to 0.5 McFarland turbidity standard to yield a bacterial suspension of 1.5×10^8 cfu/ml. Standardized inoculum used for antibacterial test.

Antibacterial activity

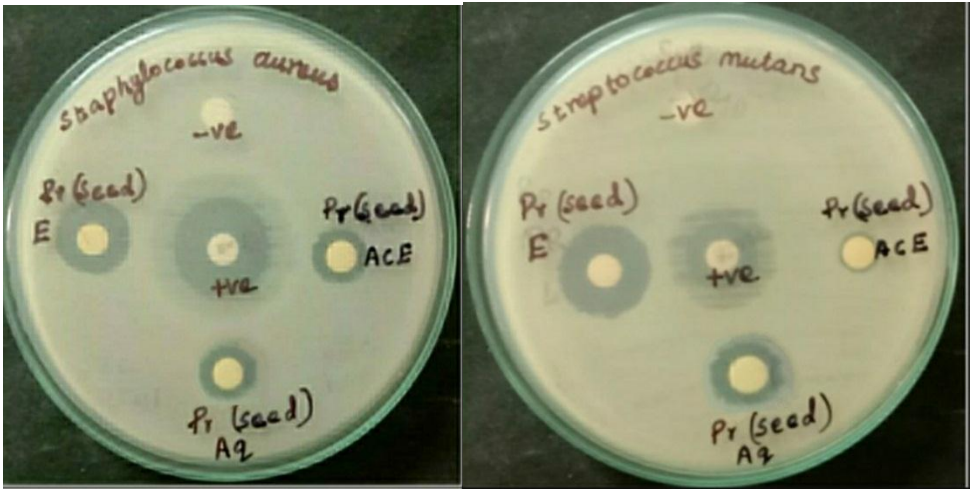
The antibacterial activity was carried out using disc diffusion method. The medium was prepared by dissolving 38 g of muller hinton agar medium (Hi Media) in 1000 ml of distilled water. The dissolved medium was autoclaved at 15 Lbs pressure at 121°C for 15 min (pH 7.3). The autoclaved medium was cooled, mixed well and poured petriplates (25 ml/plate) the plates were swabbed with *S. aureus*, *S. mutans*, *B. subtilis*, *K. pneumoniae*, *P. vulgaris*, and *E.coli*, Finally, about 10 µL of sample *P. roxburghii* seed extracts was loaded onto the disc then placed on the surface of muller-hinton medium and the plates were kept for incubation at 37°C for 24 hours. At the end of incubation, inhibition zones were examined around the disc and measured with transparent ruler in millimetres. The results were compared with the standard drug, Streptomycin. The absence of zone inhibition was interpreted as the absence of activity (Kohner, *et al.*, 1994; Mathabe, *et al.*, 2006). The activities are expressed as resistant, if the zone of inhibition was less than 7 mm, intermediate (8-10 mm) and sensitive if more than 11 mm (Assam, *et al.*, 2010).

Table 1
Antibacterial activity of ethanol, acetone, aqueous extracts of *P. roxburghii*

Name of the bacterial strains	Zone of inhibition (mm in diameter)				
	ethanol extract	acetone extract	aqueous extract	<i>Streptomycin</i>	Negative control
<i>S. aureus</i> (G+)	16	11	13	16	-
<i>S. mutans</i> (G+)	19	8	9	15	-
<i>B. Subtilis</i> (G+)	20	14	-	15	-
<i>E.coli</i> (G-)	21	15	14	14	-
<i>K.pneumonia</i> (G-)	14	13	-	15	-

<i>P. vulgaris</i> (G-)	16	15	-	14	-
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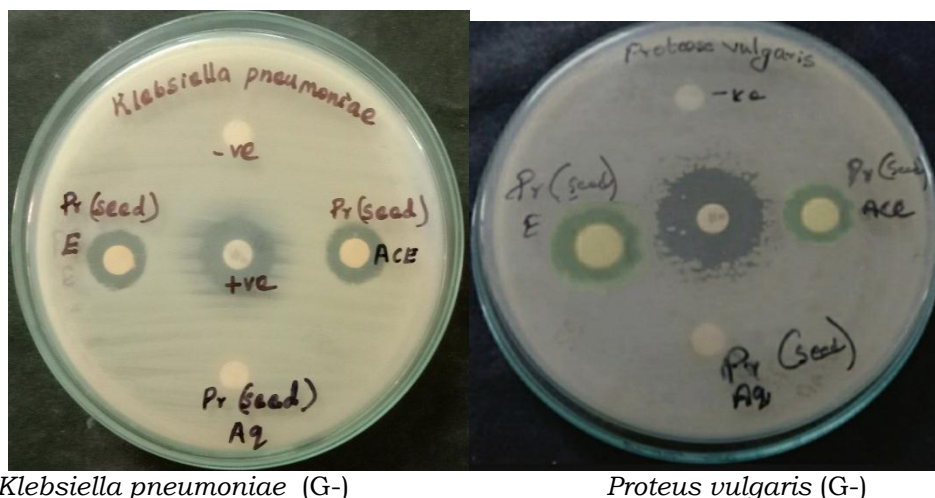
Key words : PC Positive control (*Streptomycin*), NC Negative control, “-“ No Zone, mm (Millimetre), G+ (Gram Positive Organism), G- (Gram Negative Organism).



Staphylococcus aureus (G+) *Streptococcus mutans* (G+)
Fig 2. Antibacterial activity of *P. roxburghii* seed against bacterial pathogens



Bacillus subtilis (G+) *E.coli* (G-)



Klebsiella pneumoniae (G-)

Proteus vulgaris (G-)

Result

The result on the antibacterial activity of different extracts of *P. roxburghii* seed is given in (Table 1). The ethanol extract of *P. roxburghii* seeds showed a highest zone of inhibition against the pathogens. *E.coli* (G-) (25mm). *B.subtilis* (G+) (20mm). *S. mutans* (G+) (19mm). *S. aureus* (G+) (13mm). *P. vulgaris* (G-) (16mm). *K. pneumoniae* (G-) (14mm). The acetone extract of *P. roxburghii* seed showed a minimum zone of inhibition against the pathogens. *E.coli* (G-) (18 mm). *B. Subtilis* (G+) (16 mm). *P. vulgaris* (15mm) *K. pneumoniae* (G-) (13mm). *S. aureus* (G+) (11mm) *S. mutans* (G+)(8 mm). The aqueous extract of *P.roxburghii* seed showed a lowest zone of inhibition against the pathogens. *E.coli* (G-) (14 mm). *S. aureus*(G+) (13 mm). *S.mutans* (G+)(9 mm).

Conclusion

In the present study plant extracts with different compounds showed antibacterial activity and used extracts was successfully evaluated. The antibacterial activity was based on the standard disc diffusion method. The process of extraction for a particular compound is dependent on the solubility of the component in the solvent. Plant extracts contained a very complex structure with the active ingredients present in the form of natural organic compounds. This study revealed the medicinal plant *P. roxburghii* seeds extracts could be a potent source of antibacterial activity. So, they could serve as potential candidates for future drug development approaches for curing antibacterial effects.

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