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Antibacterial activity of the fruits of Careya arborea Roxb. (Lecythidaceae)

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ABSTRACT

Plan: The present study was planned to investigate the antibacterial activity of ethyl acetate, ethanol and hexane extracts of the fruits of CareyaarboreaRoxb.

Methodology: Agar diffusion assay was carried out using the extracts.

Outcome: All the tested bacterial strains viz., Escherichia coli, Salmonella typhimurium, Listeria monocytogenes, Staphylococcus aureus and Staphylococcus epidermidis) were found to be sensitive to all the 3 concentrations of ethyl acetate and ethanolic extracts of fresh and dry fruit, in ascending order.

Keywords: Careya arborea, antibacterial activity, Lecythidaceae

1. INTRODUCTION

Careya arborea is a medicinal plant used in Oriental medical traditions. The bark, leaves and fruits

are used in Ayurveda in the treatment of ulcers, haemorrhoids, tumors, etc¹. In Chinese medicine the tree is known as Ka Li Yu Rui². The tree is also commonly found in the North East of Thailand, where people traditionally eat the shoots, young leaves and fresh flowers. Known as Kradonbok in Thai language, its leaf is used in the treatment of wounds and flowers are used for relieving cough³. In India, the tree grows in deciduous forests and grasslands.



Figure 1. Fruits of Careya arborea Roxb.

Known as *Katabhi* in Sanskrit, it is a medium sized deciduous tree growing up to a height of 15 metres, with thick, dark grey bark having shallow cracks. Fruits of Careya arboreaare large, globose, green, glabrous berries, crowned with persistent calyx and style .Persistent calyx is a distinguishing feature of Lecythidaceae.

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There are some reports on the antibacterial activity of leaf and stem bark of *Careyaarborea*⁴⁻⁶. The antibacterial activity of the fruits of *Careya arborea* was studied, considering the paucity of information on this subject.

2. MATERIALS AND METHODS

Fresh fruits of *Careya arborea* (Figure 1) were plucked from a tree in the campus of CARe Keralam Ltd, authenticated by the botanist at Kerala Agricultural University, Mannuthy, and assigned herbarium accession number (Care K 101/herb/13). They were cut into small pieces. One portion was dried under shade and the other portion was refluxed at 35-40°C, for twenty minutes after boiling. Ethyl acetate, ethanol and hexane were used as solvents⁷. The extracts were filtered, concentrated by evaporating the solvents and used for antibacterial study.

2.1 Agar Diffusion assay

Brain heart infusion broth (HIMEDIA) 10 ml was inoculated with the test cultures, incubated at 37°C for 18 hours^{8,9}. Muller Hinton agar (HIMEDIA) was prepared, autoclaved and about 15-20ml poured into Petri dishes. The agar was allowed to set and harden. Pure cultures of all experimental bacterial cultures including *Escherichia coli* (433), *Salmonella typhimurium* (3231), *Listeria monocytogenes* (657), *Staphylococcus aureus* (9886) and *Staphylococcus epidermidis* (3086) were obtained from the Microbial Type Culture Collection (MTCC), Institute of Microbial Technology (IMTECH), Chandigarh¹⁰. The cultures were swabbed on the agar surface using sterile cotton swab. 8mm cork borer was used to make wells. The extracts were added into the wells at 100μl, 150μl, and 200 μl concentrations. Gentamicin 10mcg concentration, a standard antibacterial drug was used as positive control. The plates were left at room temperature for 2 hours to allowdiffusion of extract and incubated overnight at 37°C with face upwards¹¹. The diameter of the zones of inhibition was measured with measuring scale. Inhibition of the bacterial growth was measured in mm⁸.

3. RESULT AND DISCUSSION

Three different concentrations of ethyl acetate (EA) extract, ethanolic extract and hexane extract of fresh and dry fruit of *Careya arborea* were tested and compared for their antibacterial effect against three Gram negative and two Gram positive organisms. The results showed that all the tested bacterial strains were found to be sensitive to all the 3 concentrations of EA and ethanolic extracts of fresh and dry fruit, in ascending order (Tables 1 and 2). EA extract of fresh fruit was shown to have potent antibacterial action than EA extract of dry fruit. But, ethanolic extract of both fresh and dry fruit were found to have somewhat similar antibacterial potential.

In case of hexane extract of fresh fruit, 150 and 200 μ l showed inhibitory activity against *S.typhimurium* in ascending order. *S. aureus* was found to be sensitive to 200 μ l of hexane extract of fresh fruit and the remaining 2 concentrations were inactive. But in the case of hexane extract of dry fruit, *E.coli* was found to be sensitive to 200 μ l concentrations only.

S.typhimurium had only intermediate action against all the 3 concentrations of hexane dry fruit extract and 200µl of the same was inhibitory in action on L. monocytogenes and S. epidermidis. Hexane dry fruit extract had no action on S. aureus. Both EA and ethanolic extracts of fresh and dry fruit of C. arborea were observed to have high antibacterial activity than their hexane extract. From Tables 1 and 2 it is evident that the standard antibacterial drug Gentamicin has either the same or less activity in inhibiting the growth of the test organisms. The EA and ethanolic extracts have considerably higher activity when compared to Gentamicin.

Three reports are available on the antibacterial activity of various parts of *Careyaarborea*. Daduanget al⁵ reported the antibacterial activity of leaves of *Careya sphaerica* (Synonym: *Careya arborea*) and a similar study was reported by Behera et al⁶. Another study on stem bark was reported by Kumar et al., (2006). According to the study of Kumar et al⁴most of the bacterial and fungal species were inhibited by the extract. Methanolic extract of stem bark showed considerable broad spectrum antibacterial activity on all bacterial strains tested at 25-200 µg/disc.

Behera et al⁶ explain that the leaf extract in the mixture of solvents contained polyphenols which exhibited significantly lower zones against *S. aureus* and *E.coli*. The activity against *E. coli* and *S. aureus* may be due to interaction of more lipophilic flavonoids with extracellular and soluble proteins to complex with bacterial cell wall for disruption.

These antibacterial effects are obviously brought about by well-defined chemical entities present in the leaf, stem bark or fruit of *Careya arborea*. Several characteristic compounds like careyagenolide, careaborin, hydroquinone, resorcinol, syringic acid, vanillic acid, 4-hydroxy-3-(p-hydroxyphenyl)-5,7-dimethoxy-coumarin, 3-(o-hydroxyphenyl) coumarin, 3', 4, 4', 7-tetra methoxy-, trans-2, 3, cis-2, 4-(+)-3-flavanol and 2-methoxydibenzofuran have been isolated from leaves and rootof this tree¹³⁻¹⁵.

Search of literature reveals that very few studies have been carried out on the chemical entities in *Careya arborea* fruits. The present study shows that the fruit of *Careyaarborea* contains compounds with strong antibacterial activity. Therefore, there is an urgent need to isolate the antibacterial compounds from this fruit.

Table 1: The antibacterial activity of fresh fruit of Careya arborea (Zone size in mm)

Organism	Ethyl acetate (EA)						Ethanol (E)						Hexane (H)									#Gentamicin (GEN)	
	100 μl	*In	150 µl	In	200 µl	In	EA	100 µl	In	150 µl	In	200 µl	In	Е	100 μl	In	150 µl	In	200 µl	In	Н	10 mcg	In
E.coli	24	*S	28	S	29	S	-	25	S	28	S	29	S	-	-	*R	-	R	-	R	-	22	S
S.typhimurium	24	S	26	S	28	S	-	19	S	20	S	22	S	-	-	R	15	R	18	R	-	24	S
L.mono cytogenes	22	S	23	S	24	S	-	27	S	28	S	28	S	-	-	R	-	R	-	R	-	24	S
S.aureus	30	S	33	S	36	S	-	16	S	17	S	22	S	-	-	R	-	R	18	R	-	27	S
S.epidermidis	23	S	25	S	26	S	-	22	S	23	S	25	S	-	-	*I	17	S	19	S	-	25	S

^{*}In - Inference, *S - Sensitive, *R - Resistant, *I - Intermediate # Gentamicin zone interpretation 12- Resistant (R), 13-14 - Intermediate (I), 15 - Sensitive (S)¹²

Table 2: Antibacterial activity of dry fruit of Careya arborea (Zone size in mm)

Organism	Ethyl acetate (EA)							$Ethanol\left(E ight)$								Hexane (H)							#Gentamicin (GEN)	
	100 µl	*In	150 μl	In	200 µl	In	E A	100 μl	In	150 μl	In	200 μl	In	Е	100 μl	In	150 μl	In	200 µl	In	Н	10 mcg	In	
E.coli	15	*S	18	S	31	S	-	24	S	26	S	27	S	-	-	*R	-	R	17	S	-	23	S	
S. typhimu rium	15	S	17	S	19	S	-	19	S	20	S	28	S	-	12	R	13	R	14	*I	-	25	S	
L. mono cytogenes	12	R	14	I	15	S	-	20	S	22	S	23	S	-	-	R	13	I	15	S	-	23	S	
S. aureus	15	R	20	S	24	S	-	25	S	30	S	33	S	-	-	R	-	R	-	R	-	31	S	
S. epidermidis	-	R	14	I	16	S	-	22	S	24	S	26	S	-	-	R	-	R	17	R	-	30	S	

 $[*]In-Inference, *S-Sensitive, *R-Resistant, *I-Intermediate , \#Gentamicin zone interpretation, 12-Resistant (R), 13-14-Intermediate (I), 15-Sensitive (S)^{12}$

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