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Biological Evaluation of *Artocarpus altilis* (Parkinson) Fosberg

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ABSTARCT: An emerging infectious disease is an infectious disease whose incidence has increased in the past 20 years and could increase in the near future. Search for therapeutic drugs against these diseases is progressing and is the need of the hour. Therefore, the present study was conducted to evaluate the bioefficacy of *Artocarpus altilis* fruits against various human pathogens such as *Klebsiella pneumonia, Streptococcus mutans, Lactobacillus* sps, *Bacillus subtilis* and *Enterococcus faecalis* and also to determine the presence of active compounds in the fruit through GC MS analysis. Furthermore, molecular docking was also conducted to study the interaction of molecules. *Klebsiella pneumonia* and *Streptococcus mutans* were found more sensitive towards fruit extract with a zone of inhibition of 19mm. The major peak area percentage reported through GC MS analysis was 12.75 and the corresponding compound was found to be Tris (tert-butyl dimethylsilyloxy).

Keywords: *Artocarpus altilis*, Bioefficacy, GC MS.

INTRODUCTION

Artocarpus altilis (Parkinson) Fosberg (Breadfruit) is a species of flowering tree in the mulberry and jackfruit family (Moraceae). With a better understanding of breadfruits morphology, its potential as a global solution to food security has been gaining popularity (Kervyn *et al*., 2023). Its name is derived from the texture of the moderately ripe fruit when cooked, similar to freshly baked bread and having a potato-like flavor. Breadfruit trees grow to a height of 26m (85ft). The large and thick leaves are deeply cut into pinnate lobes (Julia and Morton 2018). The fruit is nutritious and a valuable staple food in most pacific islands. The trees are monoecious, with male and female flowering growing on the same tree. The male flowers emerge first, followed by the female flowers. All parts of the tree yield latex, which is useful for boat caulking. *Artocarpus altilis* is very famous for its nutritional therapeutics, and medicinal properties. The medicinal use of *Artocarpus altilis* is abundant and unlimited. There are many traditional declarations of this plant that is yet to be proven. Thus, the aim of this study is to analyse the pharmacological properties of *Artocarpus altilis* fruit extracts.

MATERIALS AND METHODS

A. Determination of antibacterial activity of methanolic fruit of Artocarpus altilis (Parkinson) Fosberg

Antibacterial activity testing was done by Kirby Bauer disc diffusion assay (Bauer *et al.*, 1966) using 10ul of methanolic fruit extract of *Artocarpus altilis* selected after initial phytochemical screening, at a concentration of 150 mg/ml (Jiyauddin *et al.*, 2014) against human pathogens such as *Klebsiella pneumonia* (MTCC 530), *Streptococcus mutans, Bacillus subtilis* (MTCC 739), *Enterococcus faecalis* (MTCC 2912) and *Lactobacillus* (MTCC 1026). Inoculums were prepared individually for all the test organisms by aseptically transferring the cultures into 2 ml of sterile 0.145 mg/l saline tube and the cell density was adjusted to 0.5 McFarland turbidity standard to yield a microbial suspension of 1.5×10^8 cfu/ml. Standard 6 mm antibiotic discs of Streptomycin (25 µg) was used as positive control against bacterial cultures where as plane disc impregnated with 10 µl of DMSO was used as negative control. The seeded agar plates were dried for 15 minutes and incubated at 37ºC for 24 hours. The antibacterial activity was assessed by

measuring the diameter of inhibition zone in mm using transparent ruler and recorded in triplicates.

B. GC-MS analysis of methanolic fruit extract of Artocarpus altilis (Parkinson) Fosberg

The methanolic fruit extract of *Artocarpus altilis* was analysed on a Gas chromatography-Mass spectrometer (GC clarus 500 Perkin Elmer) equipped with a Elite-5 MS (5% Di-phenyl / Di-methyl poly siloxane) column (30*0.25mm df), 0.25µm film thickness, Mass detector Turbomass gold-Perkin Elmer. The oven temperature was programmed as isothermal at 110°C for 2 minutes, then raised to 280°C at the rate of 5°C/min and held at this temperature for 9 minutes. Helium was used as the carrier gas at the rate of 1ml/minute. Effluent of the GC column was introduced directly into the source of the MS via a transfer line (200°C). Ionization voltage was 70ev and ion source temperature was 200°C. Scan range was 45-450amu. Compounds were tentatively identified by comparison of mass spectra of each peak with those of authentic samples in the NIST MS library.

RESULTS AND DISCUSSION

A. Determination of antibacterial activity of methanolic fruit extract of Artocarpus altilis (Parkinson) Fosberg

The results revealed that methanolic fruit extract of *Artocarpus altilis* was found more effective against *Klebsiella pneumonia* and *Streptococcus mutans* with an inhibition zone of 19mm followed by *lactobacillus* sps (15mm) and *Bacillus subtilis* (10mm). *Enterococcus faecalis* was found resistant with negligible zone of inhibition. All the pathogens such as *Streptococcus mutans, Klebsiella pneumonia, Lactobacillus* sps., *Bacillus subtilis* and *Enterococcus fecalis* were found sensitive towards the standard antibiotic Streptomycin with an inhibition zone of 20mm (Assam *et al.,* 2010). Negative control showed negligible activity against all the five test organism (Table 1). Jiyauddin *et al.* (2014) tested the antimicrobial activity of methanolic fruit extract of *Artocarpus altilis* against *S. aureus* with an inhibition zone of 15 mm was revealed thus suggesting the efficiency of fruit against gram positive infections. Similarly Jalal (2015) investigated the bioactivity of methanolic fruit extracts of *Artocarpus altilis* against *Klebsiella pneumonia, E. coli, Staphylococcus aureus, Candida albicans etc* was also reported indicating the efficiency of *Artocarpus altilis* extract against both Gram –negative and Gram – positive pathogens.

Table 1: Antibacterial activity of methanolic fruit extract of *Artocarpus altilis* (Parkinson) Fosberg.

Group	*Klebsiella pneumonia* (mm)	*Streptococcus mutans* (mm)	*Lactobacillus* sps (mm)	*Bacillus subtilis* (mm)	*Enterococcus faecalis* (mm)
Methanol extract	19	19	15	10	NA
PC	20	20	20	20	20
NC	NA	NA	NA	NA	NA

Each value is an average of three values. PC – Positive control; NC – Negative control

B. Gas Chromatography - Mass Spectrometry (GC-MS)

The methanolic fruit extract of *Artocarpus altilis* analysed on a Gas chromatography-Mass spectrometer (GC clarus 500 Perkin Elmer) revealed the presence of 20 peaks (Table 2 and Fig. 1). Identification of mass spectrum of GC MS was conducted using the database of National Institute Standarad and Technique (NIST-Version-2005) having 62,000 patterns. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. The spectrum of unknown component was compared with the spectrum of the known component stored in the NIST data library. The name, molecular weight and molecular formula of the components of the test materials were determined. The major peak area percentage reported was 12.75 and the corresponding compound was found to be Tris (tert-butyl dimethylsilyloxy).

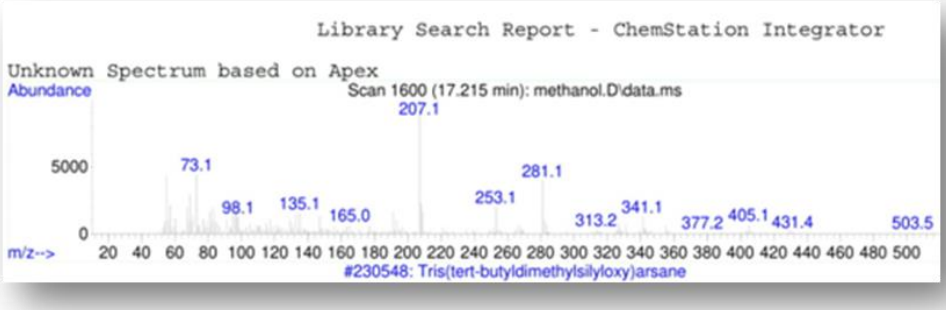

Fig. 1. GC MS chromatogram of Tris (tert-butyl dimethylsilyloxy).

Table 2: GC MS analysis of *Artocarpus altilis* (Parkinson) Fosberg.

Sr. No.	RT	Name of the Compound	Molecular Formula	Molecular Weight	Peak Area %	Biological Activity
1.	15.012	Octasiloxane 1,1,3,3,5,5,7,7,9,…	$C_{16}H_{48}O_7Si_8$	577.2	3.85	Antimicrobial activity
2.	15.986	Benzo(h)quinoline, 2,4-dimethyl-	$C_{15}H_{13}N$	207.27	4.99	Wound healing, antibacterial, Antioxidant activity
3.	16.336	2-Ethylacridine	$C_{15}H_{13}N$	207.27	2.92	Antimicrobial and anticancer activity
4.	16.421	N,N-dimethyl 1-4-nitroso-3-(trimet	$C_8H_{10}N_2O$	150.18	2.73	Not yet identified
5.	16.506	1H-Indole,5-methyl-2-phenyl-	$C_{15}H_{13}N$	207.27	2.86	Antimicrobial and anticancer activity
6.	16.686	1,2-Bis (trimethylsilyl) benzene	$C_{12}H_{22}Si_2$	222.47	5.58	Antibacterial and antioxidant activity
7.	16.780	1,4-Bis (trimethylsilyl) benzene	$C_{12}H_{22}Si_2$	222.47	4.04	Antibacterial and anti inflammatory activity
8.	16.828	Cyclotrisiloxane, hexamethyl-	$C_6H_{18}O_3Si_3$	222.46	1.533	Antimicrobial, anticancer and antioxidant activity
9.	16.913	Cyclotrisiloxane, hexamethyl-	$C_6H_{18}O_3Si_3$	222.46	5.95	Antimicrobial, anticancer and antioxidant activity
10.	16.960	2-Ethylacridine	$C_{15}H_{13}N$	207.27	6.43	Antimicrobial and anticancer activity
11.	17.215	Tris (tert-butyl dimethylsilyloxy)	$C_{18}H_{45}AsO_3Si_3$	468.7	12.75	Not yet identified
12.	17.319	1,2-Bis (tri methylsilyl)benzene	$C_{12}H_{22}Si_2$	222.47	6.07	Antibacterial and antioxidant activity
13.	17.357	1,2-Benzisothiazol-3-amine tbdms	$C_7H_6N_2S$	150.2	1.45	Antibacterial activity
14.	17.452	Octasiloxane,1,1,3,3,5,5,7,7,9,…	$C_{16}H_{48}O_7Si_8$	577.2	4.26	Antimicrobial activity
15.	17.508	Silane, chloro diethyl heptyloxy-	$C_{11}H_{25}ClOSi$	236.85	2.05	Antimicrobial activity
16.	17.660	1,4-Bis (trimethylsilyl) benzene	$C_{12}H_{22}Si_2$	222.47	4.11	Antibacterial and anti inflammatory activity
17.	17.726	Silicic acid, diethyl bis (trimet…	$C_{10}H_{28}O_4Si_3$	296.58	1.15	Antimicrobial and antioxidant activity
18.	19.503	Anthracene,9,10-dihydro-9,9,10,…	$C_{14}H_{12}O_2$	212.24	6.04	Anti-inflammatory and anticancer activity
19.	20.657	2,4-Cyclohexadien-1-one,3,5-bis…	$C_{14}H_{21}NO$	219.32	12.29	Antimicrobial, antioxidant and anticancer activity
20.	21.574	1,2-Bis (trimethylsilyl) benzene	$C_{12}H_{22}Si_2$	222.47	8.96	Antibacterial and antioxidant activity

Udaya *et al.* (2017) performed GC-MS for the methanolic leaf extracts of *Artocarpus altilis* and it revealed the presence of 12 different compounds, i.e. 2H-1,4-Benzodiazepin-2-one, 7-chloro-1,3- dihydro-5-phenyl-1-(trimethylsilyl)-3-[(trimethylsilyl)oxy]; Cyclodecasiloxane, eicosamethyl-; 2,15-Heptadecadiene, 9-(ethoxymethyl); Pentadecanoic acid, 14-methyl-,methyl ester; 10-Octadecenoic acid, methyl ester; Ethanol, 2-(9-Octadecenyloxy)-, (Z)-; 16-Octadecenoic acid, methyl ester; Docosanedioic acid, dimethyl ester; Stigmast-5-en-3-ol, oleate; a-Sitosterol trimethylsilyl ether; Ergosteryl acetate and 4,6,8(14)-Cholestatriene which are of either nutraceutical or pharmaceutical importance. Arul and Karpagam (2018) analyzed and characterized the phytoconstituents of ethanolic extract of the aerial parts of an edible plant *Alternanthera philoxeroides* (Mart.) using GC-MS analysis. The GC-MS analysis showed different peaks with low and high molecular weight, determining the presence of twelve bioactive compounds including Tris

(tert-butyl dimethylsilyloxy) which is the major compound detected in the present study. Biological activity of this major component was not yet determined and therefore need to be assessed in future. Aladesanmi *et al*. (2022) reported the presence of eleven major compounds with antiplasmodial activity in breadfruit through GC MS analysis. Yet a number of compounds with reported bioactivity such as antimicrobial, antioxidant, anticancer, anti- plasmodial, anti-infammatory activity was also identified in the present work which highlights the biological potential of breadfruits.

CONCLUSIONS

The present study revealed the bioefficacy of *Artocarpus altilis* (Parkinson) Fosberg fruits in terms of antimicrobial activity brought about by the presence of various compounds detected by GC MS.

FUTURE SCOPE

The identified compounds can be considered further for bioassay guided isolation followed by structural elucidation, molecular docking and drug designing.

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Conflict of Interest. None.

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