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Phytochemical, FTIR and GCMS Analysis and Antibacterial Activity of Methanol Extract of Leaf and Bark of *Madhuca longifolia*

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Abstract: *Madhuca longifolia* is a traditional medicinal plant known for its ethno-medicinal applications. In the present study, the phytochemical, Fourier Transform Infrared Spectroscopy (FTIR) and Gas chromatography-Mass spectrometry (GC-MS) analysis were carried out with the methanol mediated leaf and bark extracts of *Madhuca longifolia* to identify the important functional groups and phytochemical constituents. In addition, the aqueous, ethanol and methanol crude extracts of *Madhuca longifolia* were tested against *Escherichia coli*, *Klebsiella pneumonia*, *Proteus mirabilis, Bacillus cereus* and *Enterococcus faecalis.* The qualitative phytochemical analysis of *Madhuca longifolia* reveals the presence of various bioactive compounds such as tannins, alkaloids, flavonoids, saponins, glycosides, etc., and the methanol extract shows remarkable antibacterial activity with 100 µg/ml concentration against the tested organisms followed by the ethanol mediated leaf and bark extracts. The FTIR spectra observed with leaf and bark extracts of *Madhuca longifolia* revealed the occurrence of functional characteristic peaks of alcohols, primary and secondary amines, alkenes, alkynes and alkyl halides. The GC-MS analysis of methanol extracts from the leaf and bark of *Madhuca longifolia* detected the presence of eight phytochemical compounds. 1-Hexadeconal and 10-Octadecanoic acid observed in the leaf and bark methanol extract were responsible for the bactericidal efficacy observed in the present investigation. The results obtained in the present investigations will create an avenue for the identification and isolation of pharmaceutical compounds with different modes of action.

Keywords: *Madhuca longifolia*, Gas chromatography-Mass spectrometry, Fourier Transform Infrared Spectroscopy, Bactericidal, Methanol extract

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Introduction

Traditional herbal medicines were considered as the base for the invention of modern medicines. The global public health concern in the treatment of microbial infectious diseases is due to the progressive increase of drug resistance among the organisms (Shakya *et al*., 2013). The development of antibiotic resistance among bacterial strains is one of the major challenges faced by the health care sector around the world. More than 25% of the annual mortality in developing countries was

due to infectious diseases (Mahady, 2005). Persistent increase in the antibiotic resistance and emergence of the new strains of microbes as well as the debilitating side effects by the synthetic antibiotics increase the thrust for the invention of new nontoxic natural compounds to treat the drug-resistant bacterial infections (Costa *et al.*, 2016; Mehani *et al*., 2016). Plants were considered the major source for the synthesis of new molecules for the treatment of drug-resistant pathogens (Harvey, 2007). In recent periods, the quest for the isolation and identification of natural biological compounds with antimicrobial activities has increased. Natural bioactive compounds isolated from the traditional plants were capable of inhibiting the growth of drug-resistant bacteria (El-Shahaby *et al*., 2019). Different parts of the medicinal plants were subjected to solvent extraction to obtain crude drugs with potential medicinal values (Singh *et al*., 2012). The bioactive secondary metabolites of the plants such as flavonoids, alkaloids, phenols, glycosides and tannins will play a major role in the biological activity of the plant extract over the microorganisms.

Madhuca longifolia (common name Mahua; family Sapotaceae), is a fast-growing tree (Chakma and Patel, 2011). *Madhuca longifolia* possesses hepatoprotective, anti-inflammatory, antioxidant and antitumor activities (Baume *et al*., 2014). *Madhuca longifolia* is a large deciduous tree valued for its medicinal properties. The flowers are used for the preparation of expectorants which help to treat bronchitis and are also helpful in increased secretion of breast milk (Kalaivani and Jagadesan, 2013). The flower extracts possess antibacterial potential (Akshatha *et al*., 2013). Methanol extract of *Madhuca indica* possesses potential antioxidant activity (Bulbul and Begum, 2014). The anticancer potential of *Madhuca longifolia* leaf extracts was reported (Chinnadhurai *et al*., 2019). Leaves were used to treat chronic bronchitis and cancer (Bhamik *et al*., 2014). The present study aims to determine the antibacterial efficacy of leaf and bark extracts of *Madhuca longifolia* and to evaluate the bioactive compounds responsible for the bactericidal activities.

Materials and Methods

Plant collection and identification:

Fresh specimens of leaf and bark of *Madhuca longifolia* were collected from the Presidency College, Chennai, India and the characterization and identification of the plant was done with the research department of Plant Biology and Biotechnology, Presidency College, Chennai, India. The leaves and bark collected from the plants were shade dried and coarsely powdered using a mixer blender. The powdered materials were refrigerated until further use. Water, methanol and ethanol mediated *Madhuca longifolia* leaf and bark were extracted using soxhlet apparatus and the extracts were concentrated using a vacuum evaporator and the concentrated crude extracts were stored at 4 C until further analysis.

Phytochemical analysis:

Preliminary qualitative analysis of the presence of various bioactive compounds that exist in the leaf and bark extracts of *Madhuca longifolia* was estimated using a standard protocol (Harborne, 1973; Thooyavan and Karthikeyan, 2016).

Fourier Transform Infrared Spectroscopy (FTIR):

The methanol-mediated leaf extracts were analysed using an FT-IR spectrometer with a range of 450-4000cm^{-1} versus the percentage of transmittance. The presence of various functional groups in the methanol-mediated leaf and bark extracts was analyzed using Fourier transform infrared (FT-IR) spectroscopy. The spectrum was recorded with a wavelength ranging from 400 to 4000 cm^{-1} with a resolution of 4 cm^{-1}. The functional groups present in the methanol-mediated extract of *Madhuca longifolia* were obtained using the method of Nandiyanto *et al*. (2019).

Gas chromatography-mass spectrometry (GC-MS) analysis:

The methanol leaf extracts were subjected to GC-MS analysis for the determination of the molecular composition of active fractions using GC-MS JOEL-GC mate equipped with the elite 1 capillary column. The unknown compounds were identified using the NIST database with the active fraction observed (Tyagi and Agarwal, 2011).

Antibacterial activity:

Antibacterial efficacy of aqueous, methanol and ethanol mediated leaf and bark extracts of *Madhuca longifolia* was evaluated against gram-negative bacteria *Escherichia coli*, *Klebsiella pneumonia*, *Proteus mirabilis* and gram-positive bacteria *Bacillus cereus* and *Enterococcus faecalis* using the disc diffusion method. The sterile Petri plates were loaded with nutrient agar and the bacterial culture suspension was spread evenly on the surface of the agar plates. The sterile filter paper disc with a diameter of 5 mm was used in the present study. The sterile disc was loaded with solvent-mediated extract of 25 µg/ml, 50 µg/ml, 75 µg/ml or 100 µg/ml concentrations on the Petri plate along with the standard amoxicillin disc as positive control. The plates were incubated overnight at 37 C. The zone of inhibition was identified by measuring the diameter of the clear zone around the disc.

Results

The medicinal properties of the plant extracts depend on the presence of various secondary metabolites and their concentration. The phytochemical screening of crude aqueous, ethanol and methanol extracts of leaf and bark of *Madhuca longifolia* revealed the presence of major bioactive compounds and the results are presented in Table 1. The presence of tannins, cardiac glycosides, flavonoids, phenols, alkaloids and coumarins were recorded with the aqueous, methanol and ethanol solvent-mediated leaf extracts of *Madhuca longifolia.* However, the concentration of the specific active compounds varied with reference to the solvent used for the extraction. The bark extract of *Madhuca longifolia* showed the presence of tannins, cardiac glycosides, phenols, saponins, alkaloids, steroids and leucoanthocyanin. Flavonoids and coumarins were found in leaf extract and not in the bark extracts. The presence of saponin and leucoanthocyanin were observed in bark extract and these metabolites were not present in the leaf extracts. The observed results suggest that the variation and presence and absence of certain bioactive compounds in the specific extracts depend on the availability as well as the solubility of the specific metabolites in the solvent and the biological activities of the extracts may vary depending on the nature of the plant metabolites that exist in the specific extracts.

The crude leaf and bark extracts of *Madhuca longifolia* were tested against Gram-negative (*Escherichia coli, Klebsiella pneumonia, Proteus mirabilis)*, and Gram-positive bacteria (*Bacillus cereus and Enterococcus faecalis)* to evaluate the bactericidal efficacy of *Madhuca longifolia.* The results presented in Tables 2 and 3 reveal the potential antibacterial activity of the tested extracts. The alcoholic extracts showed potential antibacterial activity against all the five tested organisms with disc diffusion assay. A dose-dependent increase in the zone of inhibition was observed with leaf as well as bark extracts. The zone of inhibition ranged from 13.42 mm to 15.38 mm with reference to ethanol mediated leaf extract while the inhibition zone ranged from 13.5 mm to 15.4 mm with ethanol mediated bark extract. The zone of inhibition ranged from 13.5 mm to 15.5 mm in methanol mediated leaf extract group while it ranged from 13.6 mm to 16.1 mm on exposure to methanol-mediated bark extract. The ethanol leaf extract of *Madhuca longifolia* showed a marked zone of inhibition in the growth of *Escherichia coli*, *Klebsiella pneumoniae, Proteus mirabilis, Bacillus cereus* and *Enterococcus faecalis* with a diameter ranging from 13.64 mm to 15.38 mm with 100 µg/ml, while the methanol extract showed a potentially broad-spectrum antibacterial activity

Table 1: Phytochemical analysis of different solvent mediated extracts of leaf and bark of *Madhuca longifolia*

S. No.	Phytochemical Compounds	Leaf			Bark		
		Aqueous	Ethanol	Methanol	Aqueous	Ethanol	Methanol
1	Tannins	++	++	++	++	+++	++
2	Terpenoids	+	-	++	+	-	+
3	Cardiac glycosides	+++	+	+	+	+++	+
4	Flavonoids	++	++	++	+	-	-
5	Phenols	+	+	+	+	++	+
6	Carbohydrates	++	++	++	++	++	+
7	Saponins	++	-	-	+	+++	+++
8	Alkaloids	+	++	+	+	++	++
9	Proteins	+	-	-	+	-	-
10	Emodins	-	-	-	+	+++	+
11	Coumarins	++	++	++	+	-	-
12	Steroids	-	+	+	++	++	++
13	Anthocyanins	-	-	-	-	+	-
14	Leucoanthocyanins	-	-	-	++	+	+

- Not present; + Traces; ++ Moderate Presence; +++ Strong Presence

against all the tested organisms and the zone of inhibition ranged from 13.5 mm to 15.5 mm in diameter (Table 2).

The results of crude bark extracts of *Madhuca longifolia* are shown in Table 3. The aqueous extracts of bark showed inhibition of 12.15 mm, 10.7 mm, 11.3 mm, 11.8 mm and 12.9 mm in diameter with reference to *Escherichia coli*, *Klebsiella pneumoniae, Proteus mirabilis, Bacillus cereus* and *Enterococcus faecalis,* respectively. However, the ethanol and methanol-mediated bark extracts showed moderate bactericidal activity. The potential antibactericidal activity was observed from the methanolic crude extract against the tested organisms and the zone of inhibition varied from 13.6 to 16.1 mm in diameter. In addition, the ethanolic extract showed a moderate inhibition zone ranging from 13.5 mm to 15.4 mm with reference to *Proteus mirabilis* and *Enterococcus faecalis.* The observed results suggest that the ethanol and methanol mediated leaf and bark extracts possessed potential antibacterial activity against Gram-positive and Gram-negative bacteria tested in the present study.

The observed results on bactericidal activity are in correlation with the presence of secondary metabolites and their concentration. The functional groups of the extracts were identified with Fourier Transform Infrared Spectroscopy and the results are presented in Tables 4 and 5. The presented results reveal the presence of various functional groups in the methanol-mediated leaf and bark extracts of *Madhuca longifolia.* The data illustrated in Figures 1 and 2 on the peak values represent the functional groups that exist in the methanol extract. FTIR spectrum is used to analyse and identify the functional

Table 2: Bactericidal Efficacy of aqueous, ethanol and methanol mediated Leaf Extracts of *Madhuca longifolia*

Extracts	Bacterial Strains	Zone of Inhibition (mm)				
		25 µg/ml	50 µg/ml	75 µg/ml	100 µg/ml	Amoxicillin
Aqueous	*Escherichia coli*	6.72 ± 0.21	8.2 ± 0.27	9.3 ± 0.10	10.5 ± 0.16	13.5 ± 0.21
	Klebsiella pneumoniae	7.18 ± 0.08	9.08 ± 0.06	10.06 ± 0.14	13.6 ± 0.13	12.08 ± 0.17
	Proteus mirabilis	6.64 ± 0.08	8.06 ± 0.14	8.32 ± 0.08	11.0 ± 0.12	12.02 ± 0.32
	Bacillus Cereus	6.62 ± 0.13	7.58 ± 0.14	11.74 ± 0.28	12.76 ±0.25	12.56 ± 0.22
	Enterococcus faecalis	7.28 ± 0.06	8.44 ± 0.06	11.44 ± 0.55	12.28 ± 0.15	11.6 ± 0.19
Ethanol	*Escherichia coli*	8.74 ± 0.16	9.72 ± 0.11	11.6 ± 0.15	13.64 ± 0.10	13.62 ± 0.28
	Klebsiella pneumonia	11.04 ±0.33	12.94 ± 0.15	13.5 ± 0.14	15.38 ± 0.15	11.84 ± 0.28
	Proteus mirabilis	10.66 ±0.22	12.46 ± 0.21	13.44 ± 0.10	15.28 ± 0.13	12.36 ± 0.20
	Bacillus cereus	8.02 ± 0.10	9.28 ±0.06	12.76 ± 0.23	13.74 ± 0.11	13.18 ± 0.27
	Enterococcus faecalis	9.04 ± 0.12	11.28 ± 0.33	12.58 ± 0.18	13.42 ± 0.09	11.78 ± 0.15
Methanol	*Escherichia coli*	11.0 ± 0.22	11.7 ± 0.25	12.0 ± 0.22	13.5 ± 0.16	12.2 ± 0.12
	Klebsiella pneumonia	9.62 ± 0.18	11.9 ± 0.29	13.4 ± 0.12	15.1 ± 0.18	11.8 ± 0.25
	Proteus mirabilis	10.5 ± 0.15	13.0 ± 0.22	14.4 ± 0.18	15.5 ± 0.15	13.1 ± 0.29
	Bacillus cereus	9.7 ± 0.25	12.3 ± 0.34	13.7 ± 0.12	14.8 ± 0.3	13.7 ± 0.25
	Enterococcus faecalis	11.2 ± 0.3	12.7 ± 0.25	14.7 ± 0.25	15.2 ± 0.20	12.3 ± 0.46

groups by applying the spectrum and the peak values observed.

The FTIR analysis of methanol leaf extract showed the presence of alcohols, alkenes, alkyne, primary amines, aliphatic amines and alkyl halides. The characteristic stretching frequencies for O-H, C-H, C=C, represent wave numbers 3426cm^{-1}, 2920 cm^{-1}and 2077 cm^{-1} respectively. Asymmetric stretching observed with 1623cm^{-1} 1054cm^{-1} and 721cm^{-1} represented the presence of aliphatic amines (N-H). The peaks observed at 1353cm^{-1} and 581cm^{-1} are assigned to CH and C-Br representing phenols and alkyl halides. Aliphatic amines are the primary functional group present in the methanol leaf extract followed by alcohols (OH). The FTIR spectrum presented in Figure 2 represents the functional groups of methanol bark extracts of *Madhuca longifolia* and the presence of medium peaks at 2933 cm^{-1}and 1450 cm^{-1} represents the alkenes (C-H). A strong peak

Table 3: Bactericidal efficacy of aqueous, ethanol and methanol mediated Bark Extracts of *Madhuca longifolia*

Extracts	Bacterial Strains	Zone of Inhibition (mm)				
		25 µg/ml	50 µg/ml	75 µg/ml	100 µg/ml	Amoxicillin
Aqueous	*Escherichia coli*	8.7 ± 0.25	10.4 ± 0.33	11.38 ± 0.25	12.5 ± 0.35	12.6 ± 0.36
	Klebsiella pneumoniae	7.9 ± 0.18	9.1 ± 0.29	10.8 ± 0.37	10.7 ± 0.68	13.2 ± 0.25
	Proteus mirabilis	8.1 ±	8.8 ± 0.25	11.0 ± 0.22	11.3 ± 0.25	12.5 ± 0.27
	Bacillus Cereus	8.3 ± 0.33	10.1± 0.35	10.4 ± 0.18	11.8 ± 0.25	12.3 ± 0.25
	Enterococcus faecalis	9.4 ± 0.18	11.1 ± 0.29	12.0 ± 0.35	12.9 ± 0.36	11.7 ± 0.33
Ethanol	*Escherichia coli*	10.32 ± 0.25	11.9 ± 0.18	13.5 ± 0.15	14.5 ± 0.15	11.8 ± 0.25
	Klebsiella pneumonia	11.7 ± 0.22	12.7 ± 0.25	13.6 ± 0.18	14.4 ± 0.18	12.4 ± 0.18
	Proteus mirabilis	10.7 ± 0.24	11.9 ± 0.18	13.5 ± 0.21	13.5 ± 0.47	12.4 ± 0.4
	Bacillus cereus	11.1 ± 0.29	12.3 ± 0.33	13.8 ± 0.23	14.3 ± 0.25	13.8 ± 0.20
	Enterococcus faecalis	12.1 ± 0.18	13.6 ± 0.29	14.4 ± 0.18	15.4 ± 0.18	14.7 ± 0.20
Methanol	*Escherichia coli*	11.5 ± 0.35	11.9 ± 0.29	13.0 ± 0.27	13.6 ± 0.33	10.6 ± 0.18
	Klebsiella pneumonia	11.3 ± 0.25	13.0 ± 0.15	13.7 ± 0.25	15.3 ± 0.21	12.9 ± 0.18
	Proteus mirabilis	10.7 ± 0.53	11.9 ± 0.18	14.7 ± 0.25	15.6 ± 0.18	12.2 ± 0.33
	Bacillus cereus	11.0 ± 0.22	12.3 ±0.25	13.9 ± 0.18	14.6 ± 0.18	11.9 ± 0.18
	Enterococcus faecalis	11.6 ± 0.12	13.6 ± 0.29	14.1 ± 0.29	16.1 ± 0.18	12.6 ± 0.18

Table 4: FTIR peak values of methanol mediated leaf extract and the functional groups assigned

FTIR Peak Value (cm^{-1})	Group	Functional Group Assigned
3426	O-H Stretching (S)	Alcohol
2920	C-H Stretching (M)	Alkene
2077	C=C Stretching (W)	Alkynes
1623	N-H Bending (M)	Primary amine
1353	C-H Bending (W)	Phenol
1054	CH Stretching (M)	Aliphatic amines
721	NH Bending (W)	Primary/Secondary Amine
581	C-Br-stretching (W)	Alkyl halide

Table 5: FTIR peak values of methanol mediated leaf extract and the functional groups assigned

FTIR Peak Value (cm^{-1})	Group	Functional Group Assigned
3390	O-H Stretching (S)	Alcohol
2933	C-H Stretching (M)	Alkene
1617	N-H Bending (M)	Primary amine
1519	C-C Stretching (W)	Aromatics
1450	C-H Bending (M)	Alkene
1384	N-O Stretching (M)	Nitro compounds
1259	C-H Stretching (M)	Alky halides
1111	C-N Stretching (M)	Aliphatic amines
772	N-H Bending (W)	Primary amines

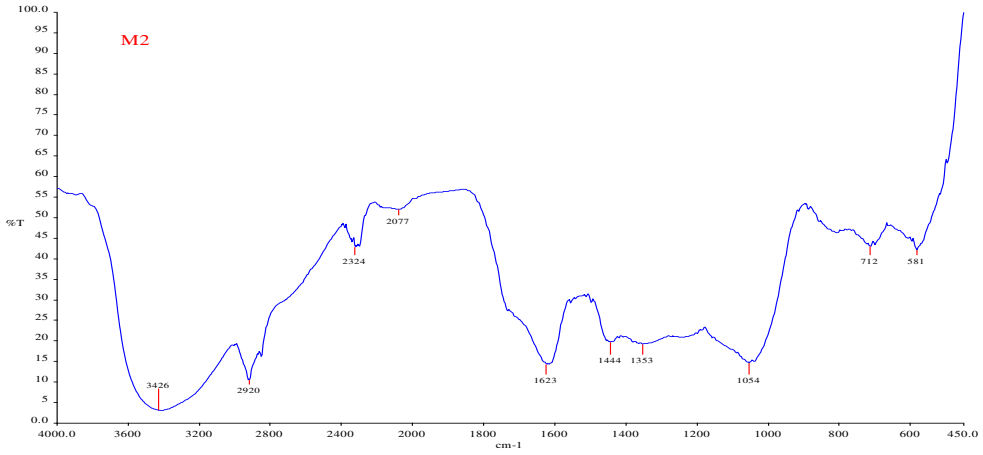

Fig. 1: FTIR Spectrum of methanol mediated leaf extract of *Madhuca longifolia.*

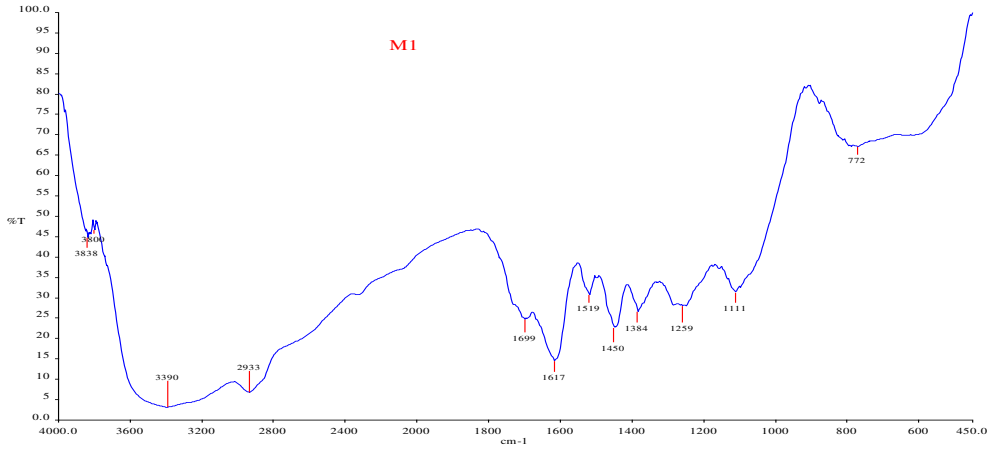

Fig. 2: FTIR Spectrum of methanol mediated bark extract of *Madhuca longifolia.*

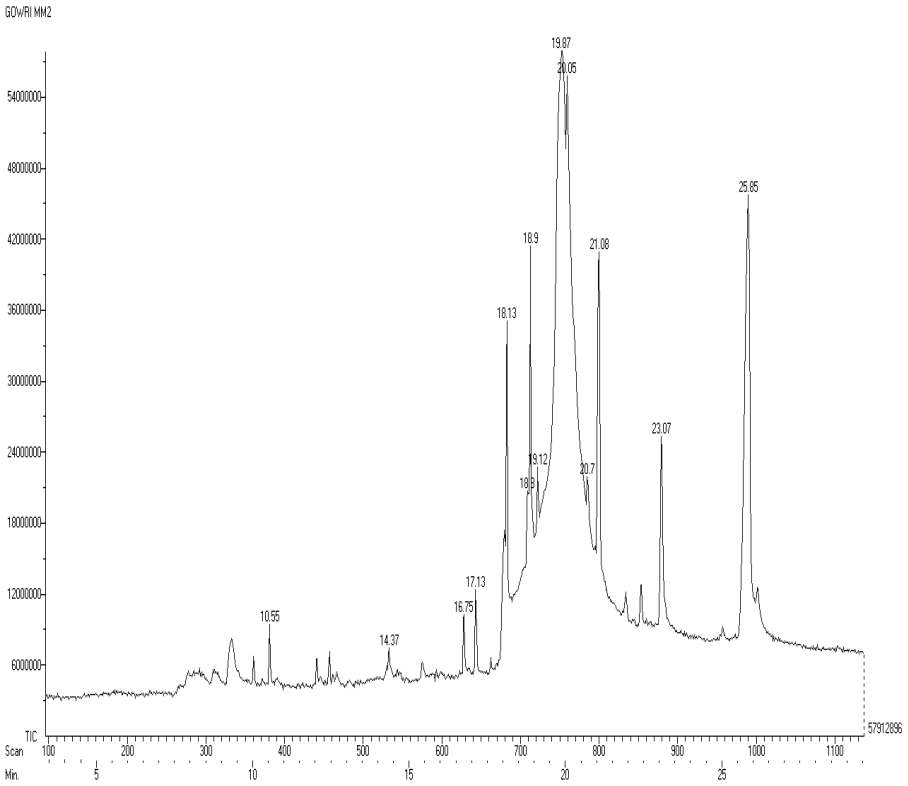

Fig. 3: GC-MS spectrum of methanol mediated leaf extract of *Madhuca longifolia.*

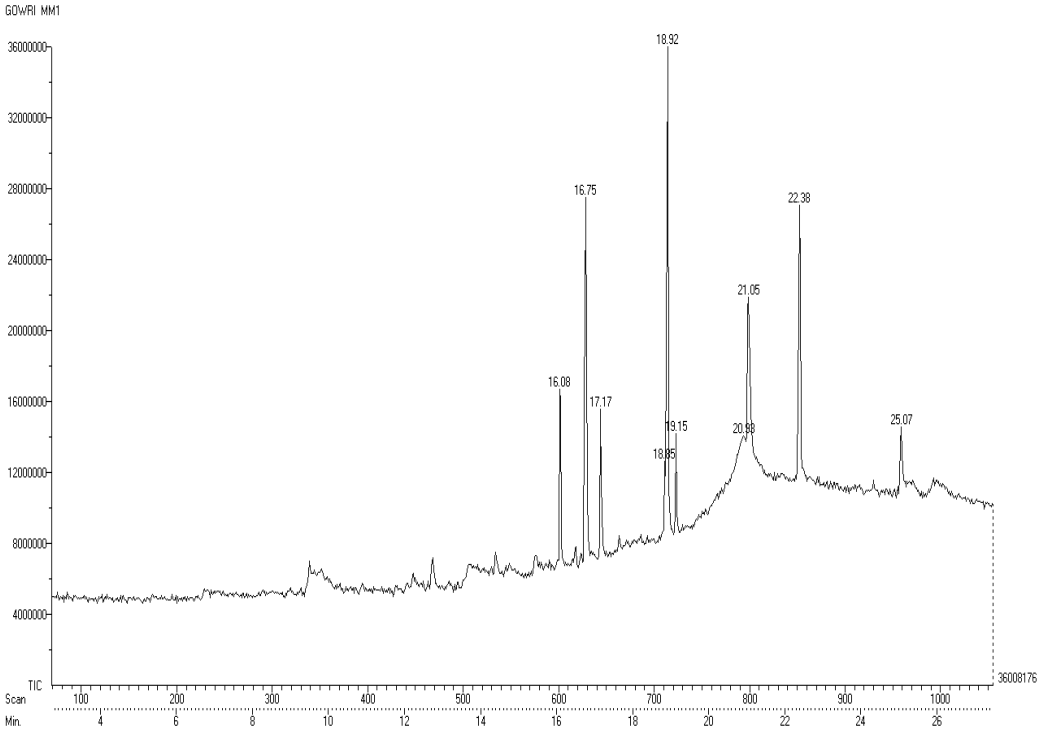

Fig. 4: GC-MS spectrum of methanol mediated bark extract of *Madhuca longifolia.*

Table 6: Phytocomponents identified in the methanol mediated leaf extract of *Madhuca longifolia*

S. No.	RT	Name of the compound	Molecular formula	Molecular weight
1	10.55	4-tert-butylcyclohexyl acetate	$C_{12}H_{22}O_2$	198.30
2	16.75	1- Nonadecanol	$CH_3(CH_2)_{18}OH$	284.52
3	17.13	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270.45
4	18.13	Isopropyl palmitate	$C_{19}H_{38}O_2$	298.50
5	18.9	10-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$	296.48
6	19.87	6- Octadecenoic acid (z)	$C_{18}H_{34}O_2$	282.46
7	20.05	9-Octadecenoic acid (E)	$C_{18}H_{34}O_2$	282.46
8	23.07	Hexadecanoic acid, 2- hydroxyl 1-(hydroxyl methyl) ethyl ester	$C_{19}H_{38}O_4$	330.50

Table 7: Phytocomponents identified in the methanol mediated bark extract of *Madhuca longifolia*

S. No.	RT	Name of the compound	Molecular formula	Molecular weight
1	16.08	n-butyl myristate	$C_{18}H_{36}O_2$	284.47
2	16.75	1- Hexadecanol	$CH_3(CH_2)_{15}OH$	242.44
3	17.17	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270.45
4	18.92	10- Octadecanoic acid, methyl ester	$C_{19}H_{36}O_2$	296.48
5	19.15	Heptadecanoic acid, 16 methyl, methyl ester	$C_{19}H_{38}O_2$	298.50
6	21.05	2- Prepenoic acid, 3 –(4- methoxy phenyl, 2- ethylhexy ester	$C_{18}H_{26}O_3$	290.39
7	22.38	Hexanoic acid, 2- ethyl, hexadecyl ester	$C_{24}H_{48}O_2$	368.72
8	25.1	1,3- Dioxolane-4 methanol, 2 pentadecyl, acetate, trans	$C_{21}H_{40}0_4$	367.53

observed at 3390cm^{-1} represents the OH-Stretching. Peaks observed at 1617cm^{-1} and 772cm^{-1} confirm the presence of primary amines (NH), in addition to the stretch observed at 1519cm^{-1} representing the aromatic amines, small stretch at 1384cm^{-1}represents the presence of nitro-compound. The presence of aliphatic amines (CN)were also recorded with the stretch at 1259cm^{-1}.

The chemical profiling of the methanol-mediated leaf and bark extracts of *Madhuca longifolia* by GC-MS analysis revealed the presence of various phytoconstituents. The NIST database analysis of the chromatogram identified in the methanol leaf extract is presented in Table 6 and Figure 3. The presence of 4-tert-butyl cyclohexyl acetate, Hexadecanol, Hexadecenoic acid methyl ester, Isopropyl palmitate, 10-octa decanoic acid, etc., are responsible for various pharmacological activities such as antioxidant, antibacterial, anti-inflammatory and anticancer activities along with hemolytic pesticidal properties. The presence of 1-Hexadecenol, Hexadecenoic acid, 10-Octadecanoic acid was recorded with methanol leaf and bark extracts of *Madhuca longifolia* (Tables 6, 7; Figs. 3, 4). The results obtained in the present investigation are correlated with the presence of these compounds and the antibacterial activity.

Discussion

Madhuca longifolia is an indigenous medicinal plant traditionally used for the treatment of various diseases such as skin diseases, rheumatism, headache and is also used to treat snakebite (Singh *et al*., 2018). The present study evaluates the antibacterial activity of different leaf and bark extracts of the tested plant. WHO has recognized that the resistance developed among the microorganisms against the drugs is a global

threat (Vanduin and Doi, 2017). The increase in the multidrug resistance among microbes compromises the available antimicrobials in developing and underdeveloped countries. It is essential to find the medicinal plants, the prospective source for the development of novel pharmaceutical compounds which can control the impact caused by drug-resistant bacteria. The bioactive compounds derived from the medicinal plants can be used as a source of alternative treatment of infections caused by microorganisms (Bakal *et al*., 2017). The multidrug-resistant bacteria *Klebsiella pneumoniae* was found to be most sensitive to plant extracts (Kebede *et al*., 2021). The present study corroborates with the above findings as ethanol and methanol mediated leaf and bark extracts showed potential inhibition zone with reference to *K. pneumoniae* and *E. coli* which are considered to be the most unresponsive strains for the antibacterial agents (Dholaria and Desai, 2018). However, in the present study, a moderate bactericidal activity was recorded with the leaf and bark extracts. The bioactive compounds endowed with the medicinal plants can inhibit the growth of *E. coli* (Valli et al., 2012). Presence of tannins, steroids, saponins, flavonoids and glycosides in the seed extracts of *Madhuca longifolia* has been reported (Pavan Kumar *et al*., 2011). The phytocompounds possess to have antibacterial potential and the alkaloids derived from the medicinal plants show antimicrobial activity (Omar *et al*., 1992; Benbott *et al*., 2012). In addition, the tannins possess antimicrobial properties. The present investigation is in agreement with the above findings as the leaf and bark extracts confirm the presence of tannins, alkaloids and saponins, which are responsible for the antibacterial activity recorded. Tannins present in the plant extracts form irreversible complexes with the proteins resulting in the inhibition of cellular protein synthesis (Akinpelu and Onakoya, 2006). It is confirmed that the presence of bioactive secondary metabolites of the leaf and bark of *Madhuca longifolia* are responsible for the bactericidal activity observed and the potential of the leaf and bactericidal extracts vary depending on the presence of various phytoconstituents and suggests that the bactericidal efficacy of the extracts were due to the combined action of various bioactive compounds.

Results obtained in the bactericidal activity of the leaf and bark extracts of the plant suggest that the *Madhuca longifolia* is a potential source for the isolation of prospective drug to control the microbial pathogens. The preliminary findings of the present study showed the presence of many bioactive compounds in the methanol extracts of the leaf and bark of the plant. The FTIR analysis and the GC-MS analysis were performed to analyse the presence of various functional groups and compounds in the methanol leaf and bark extracts.

In the FTIR spectrum, the functional groups of the methanol extracts of leaf and bark were separated based on the peak ratio. The FTIR analysis of leaf extract revealed the presence of alcohols, alkenes, alkynes, aliphatic amines and primary and secondary amines, while the methanol bark extract possessed the functional groups such as alcohols, primary amines, alkenes, aliphatic amines along with nitro compounds and alkyl halides.

Alkyl halides, alkanes are present in the plant extracts containing the highest number of functional groups which are found to have antibacterial efficacy (Janakiraman *et al*., 2011). The presence of alkyl halides and alkanes were recorded and the potential antibacterial efficacy observed in the present study may be due to the presence of biologically active functional groups in the tested extracts. The alkaloid structure includes primary amines, alcohols, alkenes and aromatics as their functional groups (Nagalakshmi and Anuradha, 2017). The GC-MS spectrum presented in Figure 3 revealed the presence of eight biologically active compounds in the methanol leaf and bark extracts of *Madhuca longifolia*. 1-hexadecanol (RT16.75) and 10-Octadecanoic acids (RT18.9) are found to be present both in leaf

as well as bark methanol extracts. 1-Hexadecenol and 10-Octadecanoic acid are considered bactericidal agents (Yasa *et al*., 2009; Asghar and Choudhry, 2011). In addition, the compounds identified with the GC-MS analysis have antioxidant and anticancer activities. The results obtained in the present study confirms that the constituents of the tested plant extracts were proven to be the reservoir of bioactive compounds with medicinal properties and may be further evaluated for the isolation of novel pharmaceutical compounds used for therapeutic applications.

Conclusion

In the present study the methanol leaf and bark extracts of *Madhuca longifolia* showed potential antibacterial activity against the tested organisms, the bark extract showed maximum efficacy against both Gram-positive and Gram-negative bacteria in comparison with the methanol leaf extracts of the tested plant and the difference in the bactericidal activity may be due to the presence of a higher concentration of bioactive compounds in the bark. The FTIR and GC-MS findings confirmed the presence of major bactericidal compounds and also confirmed the potential of methanol solvent-mediated bark extract against infectious organisms.

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