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GC-MS ANALYSIS AND ANTIMICROBIAL ACTIVITY OF METHANOLIC LEAF EXTRACT OF NYCTANTHES ARBORTRISTIS AND MURRAYA KOENIGII

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Abstract: In this modern era, medicinal plants are important for the drug industries for the production of therapeutic compounds. Hence the present study was aimed at determining phytochemical analysis of leaves of locally grown Nyctanthes arbortristis, Murraya koenigii plants for their antibacterial activity against selected bacterial pathogens. For the present study, herbal extracts of leaves of Nyctanthes arbortristis and Murraya koenigii were prepared by Cold maceration method by using methanol as solvent. The antibacterial activity was determined by the agar well diffusion technique and the prepared extract was evaluated for physicochemical characters. The bioactive compounds present in the methanol extract a fraction of Nyctanthes arbortristis and Murraya koenigii leaf were identified by GCMS analysis. The maximum antibacterial activity was observed for a methanolic extract of Nyctanthes arbortristis than Murraya koenigii. Both the methanolic extracts exhibited maximum activity against the organism Staphylococcus aureus. The physicochemical characters showed the presence of carbohydrates, alkaloids, tannins, cardiac glycosides, fatty acids, and oils. The GCMS analysis showed the presence of 45 peaks indicating the presence of 45 compounds in both the methanolic extract of Nyctanthes arbortristis and Murraya koenigii. The Constituents obtained from Nyctanthes arbortristis extraction with high quantity were 2-(4-Methoxyphenyl)ethanol(10.46), Neophytadiene(10.33), n-Hexadecanoic acid(8.95) and in Murraya koenigii extraction 1-Methyl-pyrrolidine-2-carboxylic acid(41.80), Phytol(14.66), and Caryophyllene(6.50) were present in high amounts. It can be concluded that leaves of Nyctanthes arbortristis and Murraya koenigii can be used for therapeutic purposes.

Keywords - GCMS, antimicrobial activity, Nyctanthes arbortristis, Murraya koenigii, Staphylococcus aureus, Caryophyllene.

INTRODUCTION

Herbs have always been the main form of medicine since the traditions. Nowadays it is becoming a popular form of medicine all over the world. Herbal medicines also offer promising and highly efficient novel bioactive molecules. Medicinal plants act as a reservoir for various chemical compounds which serve as drugs. It is a potential source of new lead molecules and clues for modern drug design through synthesis. *Nyctanthes arbortristis* commonly known as Parijataka or Night jasmine. It belongs to the family Oleaceae. It is now considered as a valuable source for several medicines against various diseases. It is also used for the development of some industrial products. Every part of *Nyctanthes arbortristis* is used for medicinal purposes due to its health-benefiting properties. The popular medicinal uses of *Nyctanthes arbortristis* are antihelminthic, anti-pyretic, laxative, for rheumatism, skin ailments and as a sedative. In addition it has analgesic, anti-inflammatory, antifungal and antibacterial activity, which makes this medicinal plant suitable for its use

as a therapeutic agent. [1] Murraya koenigii, commonly known as curry leaf or Kari patta in Indian dialects. It belongs to the family Rutaceae. It includes 150 genera and 1600 species. Murraya koenigii is distributed from South and East Asia to Australia. Murraya koenigii is a highly valued plant for its characteristic aroma and medicinal value. Murraya koenigii has been extensively used in Indian cooking for centuries. It has been used in developing countries for the primary and traditional healthcare system. In several ancient systems of medicine, Murrava koenigii, has wide therapeutic applications such as in bronchial abnormalities, piles, vomiting, skin infections, etc. The medicinal values have been observed especially for leaf, stem, bark, and oil. It has several pharmacological activities like anticancer activity, antioxidant activity, anti-inflammatory activity, anthelmintic activity, antidiabetic anti-ulcer. It also has cholesterol-reducing activities, and antimicrobial activity. [2] Drug resistance against human pathogenic bacteria has recently been reported worldwide. Medicinal plants are an expensive gift from nature as they provide important therapeutic aids for alleviating human ailments. Gas Chromatography-Mass Spectroscopy (GC-MS) is the best technique to identify the bioactive constituents. Such as long-chain hydrocarbons, alcohols, acids, esters, alkaloids, steroids, amino acids, flavonoids, tannins, and nitro compounds. [3][4] Therefore, characterization of medicinal plant extracts is necessary due to their numerous benefits to science and society. However, Nyctanthes arbortristis and Murraya koenigii have received notably less research attention. Considering the therapeutic properties of these plants, the present study was designed to evaluate the antibacterial activity of Nyctanthes arbortristis and Murraya koenigii. For this, the leaf extracts of Nyctanthes arbortristis and Murraya koenigii were used. The present study also deals with the GC-MS analysis of the methanolic leaf extract of Murraya koenigii and *Nyctanthes arbortristis* to identify unknown bioactive compounds.

MATERIALS AND METHODS:

Plant material:

Nyctanthes arbortristis and *Murraya koenigii* leaves were taken from the botanical garden of Dayanand College, Solapur. The leaves were identified in the Department of Botany, D.B.F. Dayanand College of Arts and Science, Solapur [Maharashtra].



Figure 1: Nyctanthes arbortristis



Figure 2: Murraya koenigii

Plant extract preparation and Phytochemical analysis study:

Preparation of plant extract:

Fresh leaves were washed under the running tap water and dried under shade at room temperature. Dried leaves were powdered in the electronic grinder. The cold extract was prepared by taking the 10 g powder in 50 ml of solvent (100% methanol) and kept at room temperature for 48 hours. Stirring of the solution was done after each 4 to 5 hours. After that solution was filtered using Whatman filter paper. Finally, the filtrate was transferred to rota vapour to evaporate the solvent and to get a solid extract. The extract was kept in a refrigerator at -40 C, to be used for further study.[5]

Preliminary Phytochemical analysis of both extracts:

A preliminary phytochemical analysis was done to find out the active chemical principle of the particular plant.

Physical characteristics of plant extract:

Physical characteristics of the plant extracts like colour, odour, and consistency were studied.

The percentage yield of plant extract:

The percentage yield of the plant extracts in methanol was determined in terms of the total quantity of powder in grams taken for the preparation of the extract.

Detection tests of plant extracts:

Detection of Alkaloids:

50 mg of Solvent-free extract was mixed with a few ml of dilute HCL and filtered. The filtrate was used for various tests as follows.

Wagner's test - To a small aliquot of filtrate in a test tube, a few drops of Wagner's reagent were added. The development of a reddish-brown precipitate indicated a positive test.

Detection of Carbohydrates:

Benedict's test - To 0.5 ml of filtrate, 0.5 ml of Benedict's reagent was added. The mixture was heated for 2 min in a boiling water bath. A characteristic coloured filtrate indicated the presence of sugar.

Detection of Amino acids and proteins:

100 mg extract was dissolved in 10 ml distilled water and filtered through Whatman no.1 filter paper. The filtrate was used to test the presence of proteins and amino acids.

Biuret test - One drop of 2% copper sulphate solution was added to 2 ml of filtrate. To this, 1ml of ethanol was added followed by the addition of excess potassium hydroxide pellets. The development of pink colour in the ethanol layer indicated the presence of proteins.

Detection of Saponins:

Foam test - 50 mg of extract was dissolved in 20 ml of distilled water. The suspension was shaken in a graduated cylinder for 15 min. The development of two cm layer of foam indicated the presence of Saponins. **Detection of Tannins:**

Ferric chloride test - 50 mg of extract was dissolved in 5 ml of distilled water and then a few drops of 5% Ferric chloride were added. The development of dark green colour indicated the presence of tannins.

Detection of flavonoids:

Magnesium and hydrochloric acid reduction test - 50 mg of the extract was dissolved in 5 ml of alcohol and a few fragments of magnesium ribbon and concentrated hydrochloric acid were added dropwise. The development of pink to crimson colour indicated the presence of flavonoids.

Detection of anthraquinones:

50 mg of extract was dissolved in distilled water. 1 ml dilute ammonia solution was added to 2 ml of extract and shaken vigorously. The development of pink colour in ammonia layer indicated the presence of anthraquinones.

Detection of Cardiac glycosides:

Killer kiliani test - 50 mg of the extract was dissolved in distilled water and filtered. Then 1 ml of glacial acetic acid and a drop of Ferric chloride and a drop of concentrated sulfuric acid were added to 2 ml of filtrate. The development of green blue colour to the upper layer and reddish-brown colour at the junction of the two layers indicated the presence of cardiac glycosides.

Detection of fixed oils and fat:

Spot test- A small aliquot of the extract was pressed between two filter papers. The development of oil stains on the paper indicated the presence of fixed oils.[6][7]

Antimicrobial Activity:

Test bacterial strains:

In this study, the test microorganisms used for antibacterial sensitivity testing included two Gram-positive bacteria *Staphylococcus aureus* and *Bacillus cereus*, and two Gram-negative bacteria *Escherichia coli*, and *Pseudomonas aeruginosa*. Pure cultures were obtained from Dr. Vaishampayan Memorial Govt. Medical College, Solapur. All strains were maintained on nutrient agar slant at 4°C and activated on Mueller Hinton Agar plates 24 hr prior to any antimicrobial test. The fresh culture of bacterial strains were grown on Mueller Hinton Broth (MHB) and further used for antibacterial assays.

Antibacterial activity of the extracts:

The antibacterial activity of the extract was determined by the agar diffusion method. For this, the fresh culture of *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli*, and *Pseudomonas aeruginosa* were suspended in sterile saline to get turbidity of 0.5 McFarland standards. 0.1 ml amount of this suspension was spread aseptically on a sterile Muller Hinton agar medium [Hi media]. Then wells [6 mm diameter] were bored by a sterile cork borer. 0.2 ml amount of each extract [100 mg /ml in 10% DMSO] was added to the wells. It was allowed to diffuse by keeping the plates in freeze for 20 min. 10 % DMSO in one of the wells served as a negative control. Antibiotic, Gentamycin [300mcg, Hi Media] disc was used as a standard positive control.

After diffusion of extracts, the plates were incubated at 37 °C for 24 hrs. The diameter of the zone of inhibition was then measured in mm.[8]

GS-MS Analysis of extracts:

The methanol leaf extracts obtained from samples were subjected to Gas Chromatography and Mass Spectroscopy for the determination of bioactive volatile compounds. Some of the important features are summarized below. GC-MS analysis of the sample was carried out using Shimadzu Make QP-2020 with nonpolar 60 M RTX 5MS Column. Helium was used as the carrier gas and the temperature programming was set with the initial oven temperature at 50°C and held for 1 min and the final temperature of the oven was 250° C with the rate at 10°C [min. sup. -1]. 2 µL sample was injected with splitless mode. An electron ionization detector was used in the instrument with an operating mass range of 20- 550. The total running time for a sample was 26 min. Quantitative determinations were made by relating respective peak areas to TIC areas from the GC-MS.

Identification of phytoconstituents:

Interpretation on the mass spectrum of GC-MS was done using the database of the National Institute Standard and Technology (NIST) having more than 139,498 compounds (111,768 in the EI library), covering both polar and non-polar columns. The mass spectrum of the unknown component was compared with the spectrum of the known component stored in the NIST library. Quantitative determinations were made by relating respective peak areas to TIC areas from the GC-MS. The principle name, molecular weight, retention time, and peak area percentage of the test materials were ascertained.[9]

RESULTS AND DISCUSSIONS:

Physical characteristics of leaves extract of Nyctanthes arbortristis and Murraya koenigii:

Name of plant ovtragt	Solventused	Physical characteristics			
Name of plant extract	Solvent useu	Colour	Consistency	Odour	
Nyctanthes arbortristis	Methanol	Dark green	Soli <mark>d sticky</mark>	Aromatic	
Murraya koenigii	Methanol	Dark green	Solid oily	Aromatic	

Table 1: Physical characteristics of extract of Nyctanthes arbortristis and Murraya koenigii:

The physical characteristics of the methanol extract of both plant leaves are depicted in Table 1. The methanol extract of *Nyctanthes arbortristis* and *Murraya koenigii* are both dark green in colour, with an aromatic odour while the consistency of *Nyctanthes arbortristis* was solid sticky whereas the consistency of *Murraya koenigii* was solid oily.

Preliminary phytochemical analysis of Nyctanthes arbortristis leaves extract:

Sr. No.	Phytochemicals	Methanol extract		
1	Alkaloids	+		
2	Carbohydrates	+		
3	Saponin	-		
4	Protein	-		
5	Amino acids	-		
6	Anthraquinones	-		
7	Tannin	+		
8	Flavonoids	-		
9	Fatty acid and oil +			
10	Cardiac glycosides	+		

Table 2: Preliminary Phytochemical analysis of Nyctanthes arbortristis.



Note: + indicates presence of phytochemicals, - indicates absence of phytochemicals

Figure 3: Phytochemical Test for Nyctanthes arbortristis

Table 2 shows the phytochemical analysis of the leaf extract of *Nyctanthes arbortristis*. The preliminary phytochemical analysis revealed extract is rich with bioactive compounds like alkaloids, carbohydrates, tannin, cardiac glycosides, fatty acids, and oil in methanol extract of *Nyctanthes arbortristis*. Alkaloids and tannins have the potential for anti-hyperglycaemic and anti-inflammatory activities; cardiac glycosides are fruitful in developing potential drugs as they have the ability to exert a beneficial stimulation on diseased heart.

Preliminary phytochemical analysis of Murraya koenigii leaves extract:

Table 3: Pr	eliminary Phytochemical	analysis of Murraya koe	enigii.
Sr. No.	Phytochemicals	Methanol extract	
1	Alkaloids	+	
2	Carbohydrates	+	
3	Saponin		
4	Protein	-	
5	Amino acids	-	
6	Anthraquinones	-	
7	Tannin	+	
8	Flavonoids	-	
9	Fatty acid and oil	+	
10	Cardiac glycosides	+	

Alkaloids Carbohydrates Cardiac glycosides Flavonoids Proteins Amino acids

Note: + indicates presence of phytochemicals, - indicates absence of phytochemicals

In Table 3 the results of phytochemical analysis of *Murraya koenigii* leaf extract are mentioned. Through various chemical tests, it was found that the leaf extract of Murraya koenigii is rich in alkaloids, carbohydrates, tannin, fatty acid and oil, and cardiac glycosides. The alkaloids present have known antimicrobial activity and also for the tannin. The study conducted by Sushmita Choudhury et.al.(2013) also found the presence of the same phytochemicals except for tannin. [10] In the present study, the presence of Tannin is due to the geographical area.

Figure 4: Phytochemical Test for Murraya koenigii

Antibacterial activity of Nyctanthes arbortristis & Murraya koenigii leaves extract:

Table 4: Antibacterial activity of *Nyctanthes arbortristis* leaf extract:

Sr. No.	Name of organism	Solvent used	Diameter of zone in mm
		DMSO	
1	Staphylococcus	Methanol	15
1	aureus	Gentamycin	30
		Plant extract	21
		DMSO	
2	Du sillur su ssiss	Methanol	15
	Bucilius species	Gentamycin	22
		Plant extract	26



Bacillus against Nyctanthes arbortristis



Staphylococcus against Nyctanthes arbortristis

Figure 5: Antibacterial activity of Nyctanthes arbortristis

Table 5: Antibacterial	activity	of	Murraya	koenigii	leaf
extract:					

Sr. No.	Name of organism	Solvent used	Diameter of zone in mm
1	Staphylococcus aureus	DMSO Methanol Gentamycin Plant extract	15 27 32
2	Bacillus species	DMSO Methanol Gentamycin Plant extract	15 30 25
3	E. Coli	DMSO Methanol Gentamycin Plant extract	 15 29 28
4	Pseudomonas aeruginosa	DMSO Methanol Gentamycin Plant extract	 18 29 30



E. Coli against

Murraya koenigii

Figure 6: Antibacterial activity of Murraya koenigii

Note: -- indicates no zone of inhibition

Antibacterial activities of both plant leaf extracts are mentioned in table 4 and 5. In the present study, the antibacterial activities of leaf extract of *Nyctanthes arbortristis* and *Murraya koenigii* significantly inhibited the test organisms. The methanolic leaf extract of *Nyctanthes arbortristis* has greater antibacterial activity than leaves extract of *Murraya koenigii*. Maximum inhibition was recorded against *Staphylococcus aureus* for both plant leaves extract of *Nyctanthes arbortristis* and *Murraya koenigii* with the zone of diameter 21mm and 32 mm respectively. The results clearly showed that both extracts have shown antibacterial activity equivalent to that of standard antibiotics.

46

GCMS analysis

GCMS analysis of Nyctanthes arbortristis:

In the methanol extract of the leaf of *Nyctanthes arbortristis* total of 45 bioactive compounds were detected and identified. Out of 45, the major compound was 2-(4-Methoxyphenyl)ethanol (10.46%) followed by Neophytadiene (10.33%), n-Hexadecanoic acid (8.95%). A study conducted by Bibek Sigdel et al.,2021 found a total of 13 compounds after GCMS analysis of the leaf extract of *Nyctanthes arbortristis*. [11] In the present study, the leaf extract shows the presence of 45 compounds this is due to the geographical area. Neophytadiene and n-Hexadecanoic acid has known antimicrobial activity. The antimicrobial activity shown by the *Nyctanthes arbortristis* might be due to these compounds. The bioactive compounds isolated and detected are mentioned in Table 6. Figure 7 shows the chromatogram of GCMS analysis of *Nyctanthes arbortristis*. The chromatogram shows different 45 peaks indicating 45 compounds present in it.



Figure 7: Chromatogram of Nyctanthes arbortristis

Peak <mark>#</mark>	R.Time	Area	Area%	Name	Similarity
1	5.882	1170068	6.74	2-Pentanone, 4-hydroxy-4-methyl-	96
2	7.299	357482	2.06	Dihydroxyacetone	87
3	9.646	197616	1.14	2-Hydroxy-gamma-butyrolactone	89
4	11.784	116666	0.67	Pentanal	83
5	13.870	1259157	7.25	Benzofuran, 2,3-dihydro-	91
6	15.339	202137	1.16	2-Methoxy-4-vinylphenol	94
7	16.160	1816641	10.46	2-(4-Methoxyphenyl)ethanol	97
8	16.588	267251	1.54	1'-Hydroxy-4,3'-dimethyl-bicyclohexyl-3,3'-dien-2- one	82
9	16.875	288244	1.66	Benzeneethanol, 4-hydroxy-	94
10	17.014	96791	0.56	3-Nitrobenzyl iodide	76
11	17.720	720369	4.15	Pyrazolidine-3,5-dione, 4-phenyl-	84

Table 6: Bioactive compounds present in Nyctanthes arbortristis

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	1				
12	18.369	111505	0.64	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a- trimethyl-, (R)-	96
13	18.501	-8878	-0.05	Methyl trans-4-methylcinnamate	78
14	18.610	180180	1.04	Phenol, 4-ethenyl-2,6-dimethoxy-	93
15	18.944	96705	0.56	Diethyl Phthalate	84
16	19.071	393754	2.27	Cyclopenta[c]pyran-4-carboxylic acid, 7-methyl-, methyl ester	89
17	20.675	400203	2.30	4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol	85
18	21.712	1794099	10.33	Neophytadiene	95
19	21.765	77765	0.45	2-Pentadecanone, 6,10,14-trimethyl-	88
20	21.967	253896	1.46	Neophytadiene	91
21	22.098	47969	0.28		61
22	22.161	405713	2.34	Neophytadiene	92
23	22.393	84234	0.49	2(1H)-Naphthalenone, 4a,5,6,7,8,8a-hexahydro- 7.alphaisopropyl-4a.beta.,8a.betadimethyl-	59
24	22.605	508507	2.93	Hexadecanoic acid, methyl ester	95
25	22.671	202120	1.16	1-(2-Hydroxy-ethyl)-2-methyl-1H-benzoimidazole- 5-carboxylic acid methyl ester	78
26	22.963	1553331	8.95	n-Hexadecanoic acid	93
27	23.301	119430	0.69	1-Heptacosanol	89
28	23.557	211815	1.22	(1R,6S)-6-Hydroxy-6-methyl-4-oxocyclohex-2-en- 1-yl benzoate	87
29	24.085	65853	0.38	(1R,6S)-6-Hydroxy-6-methyl-4-oxocyclohex-2-en- 1-yl benzoate	82
30	24.252	121930	0.70	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	92
31	24.310	969727	5.59	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	92
32	24.416	1317805	7.59	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl-, acetate, [R-[R*,R*-(E)]]-	92
33	24.604	223521	1.29	9,12-Octadecadienoic acid (Z,Z)-	93
34	24.659	460046	2.65	7-Tetradecenal, (Z)-	87
35	24.859	244315	1.41	Octadecanoic acid	91
36	25.174	52192	0.30	1-Heptacosanol	88
37	25.654	164743	0.95	Glutaric acid, tridec-2-yn-1-yl geranyl ester	83
38	27.279	52472	0.30	Benzenecarbothioic acid, S-ethyl ester	86

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39	27.368	55348	0.32	Benzoic acid, (5,5-dimethyl-4-oxo-2-cyclohexenyl) ester	84
40	27.745	59924	0.35	2-Methylhexacosane	76
41	27.825	-120739	-0.70		28
42	27.839	219616	1.26	Hexadecanoic acid, 2-hydroxy-1- (hydroxymethyl)ethyl ester	93
43	28.007	76503	0.44	Nonacosanal	88
44	29.456	375024	2.16	7-Hexadecenal, (Z)-	86
45	30.684	99551	0.57	2,6,10,15,19,23-Pentamethyl-2,6,18,22- tetracosatetraen-10,15-diol	92
		17362601	100.00		

GCMS analysis of *Murraya koenigii*:

In the methanol extract of the leaf of *Murraya koenigii*, 45 compounds were identified by GCMS analysis. 1-Methyl-pyrrolidine-2-carboxylic acid (41.80%) was the major compound followed by Phytol (14.66%), Caryophyllene (6.50%). The compound identified and detected in the methanol extract is shown in Table 7. Figure 8 shows the GCMS chromatogram of the methanol extract of *Murraya koenigii*. GCMS analysis in the present study was carried out to explore the potential of bioactive compounds found in *Murraya koenigii*. The study conducted by Mohammad Taghi Ghaneian et.al. (2015) concluded that Phytol has antimicrobial activity. [12] β -caryophyllene have strong antimicrobial activity against periodontopathogens.[13]



Figure 8: Chromatogram of Murraya koenigii

Peak#	R.Time	Area	Area%	Name	Similarity
1	5.882	2701378	3.24	2-Pentanone, 4-hydroxy-4-methyl-	97
2	12.703	1985404	2.38	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6- methyl-	95
3	13.881	34881442	41.80	1-Methyl-pyrrolidine-2-carboxylic acid	88
4	16.987	5426116	6.50	Caryophyllene	95
5	17.079	378055	0.45	transalphaBergamotene	94
6	17.241	860591	1.03		85
7	17.455	884045	1.06	1,4,7,-Cycloundecatriene, 1,5,9,9-tetramethyl-, Z,Z,Z-	95
8	17.890	192153	0.23	0.23 Naphthalene, decahydro-4a-methyl-1- methylene-7-(1-methylethenyl)-, [4aR(4a.alpha.,7.alpha.,8a.beta.)]- 91	91
9	17.971	982538	1.18	1.18 Naphthalene, 1,2,3,4,4a,5,6,8a octahydro4a,8dimethyl-2-(1-methylethenyl [2R(2.alpha.,4a.alpha.,8a.beta.)]- 94	94
10	18.194	118827	0.14	Naphthalene, 1,2,3,5,6,8a-hexahydro-4,7- dimethyl-1-(1-methylethyl)-, (1S-cis)-	89
11	18.625	187629	0.22	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-, (E)-	86
12	18.942	756868	0.91	Diethyl Phthalate	95
13	19.068	2025524	2.43	Caryophyllene oxide	91
14	19.394	2052366	2.46	Decanal	78
15	19.697	1677918	2.01	10,10-Dimethyl-2,6- dimethylenebicyclo[7.2.0]undecan-5.betaol	93
16	19.944	1077928	1.29	Neointermedeol	93
17	20.048	420055	0.50	(1R,7S,E)-7-Isopropyl-4,10- dimethylenecyclodec-5-enol	83
18	20.699	272759	0.33	(E)-4-(3-Hydroxyprop-1-en-1-yl)-2- methoxyphenol	88
19	20.801	606716	0.73	2-Amino-3-hydroxypyridine	80
20	21.115	470159	0.56	6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a- tetrahydrobenzofuran-2(4H)-one	93
21	21.715	1132313	1.36	Neophytadiene	93
22	21.767	330398	0.40	2-Pentadecanone, 6,10,14-trimethyl-	92
23	21.972	202118	0.24	Bis(3,7-dimethyloct-6-enyl) phthalate	77
24	22.167	273944	0.33	Neophytadiene	92

Table 7: Bloactive compounds present in <i>Murraya koeni</i>
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IJCRT2310506 International Journal of Creative Research Thoughts (IJCRT) www.ijcrt.org e537

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25	22.609	560780	0.67	Hexadecanoic acid, methyl ester	95
26	22.973	3458253	4.14	n-Hexadecanoic acid	93
27	23.627	286690	0.34	1,6,10,14,18,22-Tetracosahexaen-3-ol, 2,6,10,15,19,23-hexamethyl-, (all-E)-	87
28	24.256	195573	0.23	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	91
29	24.314	711041	0.85	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	93
30	24.425	12236367	14.66	Phytol	93
31	24.612	302023	0.36	9,12-Octadecadienoic acid (Z,Z)-	91
32	24.672	1446133	1.73	Dichloroacetic acid, tridec-2-ynyl ester	86
33	24.772	50558	0.06	Phytyl stearate	91
34	24.864	291990	0.35	Octadecanoic acid	88
35	25.954	235795	0.28	3-Cyclopentylpropionic acid, 2- dimethylaminoethyl ester	93
36	27.842	179911	0.22	Hexadecanoic acid, 2-hydroxy-1- (hydroxymethyl)ethyl ester	91
37	28.588	218201	0.26	6,11-Dihydro-8-methoxy-5H-benzo[a]carbazole	83
38	28.941	548395	0.66	3-Meth <mark>yl-9</mark> H-carbazole-2,7-diol	93
39	29.466	220665	0.26	0.26 2,4,6(1H,3H,5H)-Pyrimidinetrione, 5-[[[2- (1-cyclohexen-1-yl)ethyl]amino]methylene]-1- (2-fluorophenyl)- 65	65
40	30.430	170890	0.20	Mahanimbine	92
41	30.507	526604	0.63	O-Methylmurrayamine A	89
42	30.735	8280 <mark>3</mark> 5	0.99	Koenimbin	94
43	31.087	340296	0.41	.alphaTocospiro A	91
44	31.404	498900	0.60	Mahanimbine	65
45	31.701	236523	0.28	Mahanimbine	68
		83440867	100.00		

*R. Time – Retention time

CONCLUSION:

From the present study it was concluded that the antibacterial activity was observed for both plant extracts against selected organisms. Among these *Nyctanthes arbortristis* exhibited maximum activity than *Murraya koenigii*. The *Staphylococcus aureus* shows maximum inhibition for both the plant extracts where DMSO serves as negative control and antibiotic as a positive control. Both the plant extract showed presence of alkaloids, tannins, cardiac glycosides, carbohydrates, fatty acids and oils and absence of Saponin, Proteins, Amino acids, Anthraquinones, flavonoids. GCMS analysis revealed total 45 bioactive compounds in both the extracts. Out of which major 3 phytochemicals present in *Nyctanthes arbortristis* are 2-(4-Methoxyphenyl) ethanol(10.46%), Neophytadiene (10.33%), n-Hexadecanoic acid (8.95%) have known antibacterial activity as

mentioned in some literatures. The most abundant compound found in leaf extract of *Murraya koenigii* was 1-Methyl-pyrrolidine-2-carboxylic acid (41.80%) followed by Phytol (14.66%), and Caryophyllene (6.50%). Such phytochemical constituent have been reported to possess antibacterial, antioxidant values. Thus, the phytochemical analysis, GCMS studies and antibacterial activity studies of plant extracts provides scientific evidence for the traditional clam and use of *Nyctanthes arbortristis* and *Murraya koenigii* plant extract for therapeutic purposes and due to their utilization as potential natural antibacterial agents could be of high economic value.

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