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Preliminary phytochemical analysis, GC-MS studies and Antioxidant activity of *Majidea zangueberica* J. Kirk leaf extracts

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Abstract

The study paves an insight into the medicinal properties of the ornamental plant *Majidea zangueberica* belonging to the family Sapindaceae. The phytochemical screening of chloroform (CE) and hexane extract (HE) of leaves revealed the presence of various compounds such as alkaloids, flavonoids, glycosides, saponins, steroids, tri-terpenoids, cardiac glycosides and protein. The total soluble carbohydrate, proteins and chlorophyll content present in the leaves of *M. zangueberica* was 40 ± 1.07 mg/gram, 58.33 ± 2.22 mg/gram and 0.5483 ± 0.017 mg/fresh weight respectively. Higher total phenolic content were present in chloroform extract of *M. zangueberica* leaves than hexane extract (3.83 ± 0.12). The GCMS analysis in CE confirmed the presence of 40 compounds with various peak areas and biological activity. The leaves showed higher antioxidant property (inhibition-69.75%) in its CE than HE. Further study may provide a good source of medicinally important drugs in near future.

Keywords: *Majidea zangueberica*, phytochemical screening, qualitative, quantitative, GC-MS, antioxidant

1. Introduction

Nature has been a supply of medicinal resource for thousands of years and high numbers of recent medication are isolated from nature^[1]. Phytochemicals are natural bioactive compounds found in plants which work together with nutrients and act as a defence mechanism against disease^[2]. Phytochemical analysis involves extraction, screening and identification of bioactive components present in plant parts. Quantitative evaluation of phytocompounds is a key parameter in setting a standard for crude drugs. Carbohydrates are macromolecules that comprise elements of water and carbon^[3]. The source of energy and proteins are derived partly from carbohydrates through the synthesis of amino acids. They occur throughout the plant cells, in conjugated forms^[4]. Most of the medicinal plants are enriched with phenol which reveals biological and pharmacological properties like antimicrobial, antiviral, antioxidant and anti-inflammatory and validates the use of the plant in ethno medicine^[5]. Leaf pigment analysis is a crucial parameter that's often measured as an indicator of chloroplast content, photosynthetic mechanism and plant metabolism^[6].

In the recent years, GCMS is considered as a helpful tool to identify the phytochemicals from the medicinal plants and various natural products; because it is a non-destructive, direct and fast analytical method for identification of terpenoids, fatty acids and other phytochemicals and is the best technique to identify the bioactive constituents of long chain hydrocarbons, alcohols, acids, esters, ethers etc.^[7]. Antioxidants are phytochemical compounds which protect cells against damage caused by molecules known as free radicals^[8]. DPPH radical scavenging activity is a simple and wide used technique for testing *in-vitro* inhibitor activity of natural compounds or plant extracts.

Majidea zangueberica J. Kirk, commonly known mgambo tree, native to East Africa is a small ornamental tree belongs to the family Sapindaceae. Till date, limited data is available on the medicinal properties of this species and are therefore considered as an area yet to be explored. The medicinal property of a plant is mainly due to presence of certain phytoconstituents, especially the phenol and flavonoids have anticancer and antioxidant property. The present investigation is designed to evaluate the leaf extracts of *Majidea zangueberica* for phytochemical constituents (qualitative and quantitative), GCMS studies and antioxidant activity.

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2. Materials and Method

2.1. Collection and Preparation of extract

Fresh plant material was collected from PSG College of Arts and Science (Autonomous), Coimbatore-641014, Tamil Nadu, India and were taxonomically identified by the Department of Botany, PSG College of Arts and Science. The leaf was washed under running tap water to remove the dust and debris, air dried and powdered



Plate 1: *M. zangueberica* habit and fruit

30 grams of powdered leaf were extracted with 250ml of organic solvent (hexane and chloroform) by using Soxhlet apparatus and the solvent was evaporated to make the final volume one fourth of the original volume.

2.2. Preliminary phytochemical analysis

The extracts were subjected to preliminary phytochemical tests to determine the group of secondary metabolites present in the plant material using standard procedure

2.2.1. Test for alkaloids

To the 1ml of extract in two separate test tubes, 2-3 drops of Dragendorff's and Mayer's reagents were separately added. An orange red precipitate turbidity with Dragendorff's reagent or white precipitate with Mayer's reagent would indicate the presence of alkaloids [9].

2.2.2. Test for flavonoids

To the 1 ml of extract 1ml of ferric chloride is added. The formation of brown colour indicates the presence of flavonoids [10].

2.2.3. Test for glycosides

Keller-Kiliani test: To the 2 ml of extract 1ml of glacial acetic acid with ferric chloride and concentrated H_2SO_4 was added. The formation of blue colour indicates the presence of glycosides [11].

2.2.4. Test for saponins

One ml of sample extracts was taken and 5 ml of distilled water was added and vigorously shaken. Formation persistent froth that lasted for at least 5 minutes indicates the presence of saponins [12].

2.2.5. Test for tannins

Two ml of the extracts were diluted with distilled water in separate test tubes; 2 – 3 drop of 5% ferric chloride solution was added. A green-black or blue colouration indicates the presence of tannins [9].

2.2.6. Test for steroids

Two ml of the extract were taken in separate test tubes and

evaporated to dryness. The residues were dissolved in acetic anhydride and then chloroform was added. Conc. H_2SO_4 was added. Formation of brown ring at the interphase of the two liquids and the appearance of the violet colour in the supernatant layer indicate the presence of steroids [9].

2.2.7. Test for terpenoids

5 ml of extract were mixed with 2ml of chloroform and con. H_2SO_4 to form a layer. Formation of reddish brown colour at the interface shows the presence of terpenoids [13].

2.2.8. Test for triterpenoids

Chloroform test: To 1ml of extract, few drops of chloroform were added and it was treated with concentrated sulphuric acid solution. Appearance of reddish brown colour shows the presence of triterpenoids [14].

2.2.9. Test for phenols

5 ml of concentrated extracts were taken and 2ml of neutral ferric chloride solution was added. Appearance of violet colour indicates presence of phenol [15].

2.2.10. Test for quinone

Addition of 1 ml of extract with 5ml of conc. HCl will result yellow colour precipitate indicating the presence of quinone [14].

2.2.11. Test for Cardiac glycosides

Keller-Kiliani test: To 1ml of extract 2 ml of glacial acetic acid and few drops of 5% ferric chloride solution was added. Then it was treated with few drops of concentrated sulphuric acid. Appearance of reddish brown layer indicates the presence of cardiac glycosides [14].

2.2.12. Test for proteins

Biuret test: To 1ml of the test solution, few drops of 0.7% copper sulphate solution and 1ml of 10% NaOH were added and mixed thoroughly. Formation of purple or violet colour confirms proteins [14].

2.2.13. Test for carbohydrates

Benedict's test: To 1ml of extract, 2ml of Benedict's reagent was added and heated in boiling water bath for 5 minutes. Formation of brick red colour indicates the presence of carbohydrates [14].

2.3. Quantitative Analysis

2.3.1. Total soluble carbohydrate

Estimation of total carbohydrate content was determined by using slightly modified method of Hedge and Hofreiter [16]. To 100 mg of plant material 5 ml of 2.5N HCl was added and boiled for three hours in water bath. The hydrolysed sample mixture was cooled to room temperature and neutralized it with Na_2CO_3 . The reaction was completed when effervescence formation was stopped. The volume of plant extract was made to 100 ml and centrifuged at 3000 rpm for 20 min. Supernatant was taken for analysis by a spectrophotometric method using anthrone reagent at 630nm. Total carbohydrate content was determined from the standard curve plot with glucose.

2.3.2. Total proteins content

The total protein content in extracts was determined based on Lowry's method [17]. 100mg plant material was extracted by using 10 ml 0.2M phosphate buffer and centrifuged at

3000rpm for 10minutes. Supernatant was collected and made up to 10 ml with distilled water and 1ml of the supernatant was diluted to 10ml with distilled water and used for estimation. About 5 ml of alkaline copper reagent and 0.5 ml of folin-ciocalteau reagent were added to each test tube, including the blank. The reaction mixture was incubated for 30 min at room temperature and absorbance was taken at 700 nm by using spectrophotometer. The total protein content was determined from the standard curve plot with BSA.

2.3.3. Total phenolic content

The total phenolic content of the leaf extract was determined by using slightly modified method of Ainsworth [18]. About 0.5 mL of the each extract was taken and volume adjusted to 1 mL distilled water. About 0.5 ml of folin-ciocalteau and 2.5ml of 5% sodium carbonate were added (including the blank). The reaction mixture was incubated in the dark room at room temperature for an hour and the absorbance was measured at 725 nm using spectrophotometer. The total phenolic content was determined from the standard curve plot with gallic acid.

2.3.4. Chlorophyll Analysis

One gram of leaf sample was taken and ground to fine paste with 20ml of 80% acetone and centrifuged at 5000 rpm for 5 minutes. The supernatant was transferred to 100ml volumetric flask and the procedure was repeated till the residue becomes colourless. Absorbance of the solution was estimated at 645 and 663nm against the blank (80% Acetone) [19]. The concentrations of chlorophyll a, chlorophyll b and total chlorophyll were calculated by using the following equation:
 Total Chlorophyll: $[20.2(A_{645}) + 8.02(A_{663})] \times V / (1000 \times W)$
 Chlorophyll a: $[12.7(A_{663}) - 2.69(A_{645})] \times V / (1000 \times W)$
 Chlorophyll b: $[22.9(A_{645}) - 4.68(A_{663})] \times V / (1000 \times W)$

Where,

A= absorbance at specific wavelengths,
 V= final volume of chlorophyll extract and
 W= weight of tissue.

2.4. GC-MS Analysis

The GC – MS analysis was carried out using a shimadzu GCMS QP2010 Plus make with 30m x 0.25 µm DF of capillary column with carrier gas Helium. The instrument was set to an initial temperature of 100°C, and maintained at this temperature for 2 min. At the end of this period the oven temperature rose up to 280°C, at the rate of an increase of 5°C /min, and maintained for 9 min. Injection port temperature was ensured as 280 °C and Helium flow rate as one ml/min. The ionization voltage was 70eV. The samples were injected in split mode as 10:1 and 2 µl of the sample was injected. Mass spectral scan range was set at 35-800 (m/z). The scan time was 1-53 min and the speed was 1666. Using computer searches on a NIST Version –Year 2011 were used MS data library and comparing the spectrum obtained through GC – MS compounds present in the plants sample were identified.

2.4.1. Identification of components

Interpretation on mass spectrum of GC-MS was done using the database of National Institute of Standards and Technology (NIST) and pubchem. The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library and pubchem. The name, molecular weight, structure and biological activity of the components of the test materials were tabulated.

2.5. Antioxidant activity

2.5.1. DPPH radical scavenging activity

The antioxidant activity of the hexane and chloroform extract of *Majidea zangueberica* leaf extract was measured on the basis Blois [20]. Sample extracts at 500µL concentration were taken and the volume was adjusted to 1 mL with methanol. About 3 mL of a 0.1 mM methanol solution of DPPH was added to both the samples and standards (BHT) and shaken vigorously. Negative control was prepared by adding 1 mL of methanol in 3 ml of 0.1 mM methanol solution DPPH. The tubes were allowed to stand for 20 min at 27°C. The absorbance of the sample was measured at 517 nm against the blank (methanol). The inhibition percentage was calculated by using the following formula.

$$\text{Inhibition \%} = \frac{A_c - A_s}{A_c} \times 100$$

Where, A_c is the absorbance of the control

A_s is the absorbance of the sample

3. Result and Discussion

3.1. Preliminary phytochemical analysis

Plants are very important source of potentially useful bioactive principle for the development of new chemotherapeutic agents [21]. The biological and pharmacological properties of many plants are still unknown. Phytochemical constitutes of plants serves as defense mechanism against by many microorganisms. The therapeutic properties of medicinal plants are possibly due to the presence of various secondary metabolites [22]. Thus the preliminary screening test may be useful in the detection of the bioactive principles and subsequently may lead to the drug discovery and improvement.

The phytochemical analysis of hexane and chloroform leaf extract of *M. zangueberica* was analysed for the compounds such as alkaloids, cardiac glycosides, flavonoids, glycosides, saponins, steroids, tannins, etc. The preliminary phytochemical analysis of both sample revealed the presence of nine compounds i.e. alkaloids, flavonoids, glycosides, saponins, steroids, tri-terpenoids, cardiac glycosides and protein and absence of quinone (Table. 1). The chloroform extracts showed positive result towards tannin, terpenoids, carbohydrate and phenolic compound. The chloroform extract showed the presence of most of the phytochemicals analysed.

Table 1: Preliminary phytochemical analysis of *Majidea zangueberica* leaves.

S No	Phytoconstituents	Hexane	Chloroform
1	Alkaloids Wagner's test	+	+
	Dragendoff's test	+	+
2	Flavonoids	+	+
3	Glycosides	+	+
4	Saponins	+	+
5	Tannins	-	+
6	Steroids	+	+
7	Terpenoids	-	+
8	Triterpenoids	+	+
9	Phenolic compounds	-	+
10	Quinone	-	-
11	Cardiac glycosides	+	+
12	Protein	+	+
13	Carbohydrates	ND	+

+ Present; – Absent; **ND** Not detected

3.2. Quantitative analysis

The present study provides the estimated amounts of the primary metabolites (total soluble carbohydrates, total

proteins and chlorophyll) and secondary metabolites (total phenols) present in the leaf. Further the quantitative phytochemical screening may aid in the detection of the bioactive elements that can be further evaluated.

The total soluble carbohydrate and proteins present in leaves sample of *M. zangueberica* are 40 ± 1.07 and 58.33 ± 2.22 respectively. The amount of chlorophyll a, chlorophyll b and total chlorophyll content of the leaves of *Majidea zangueberica* were 0.3827, 0.1656 and 0.5483 ± 0.017 respectively.

Carbohydrates and Proteins are one of the main components of living things. Plant sugars can be used as artificial sweetener and they can even help in diabetes by supporting the body in its rebuilding [23]. The presence of higher protein level in the plant points towards their possible increase food value or that a protein base bioactive compound could also be isolated in future [24]. Greens are vital sources of protective food which are highly beneficial for the maintenance of good

health and prevention of diseases. Chlorophyll or its derivatives can be utilized as a photodynamic agent in tumor or cancer therapy [25].

3.2.1. Total phenolic content

The phenolic compounds are one of the largest and most universal groups of plant metabolites. They possess biological properties such as antioxidant, anti-inflammation, antiageing, anticarcinogen [26]. Hence total phenolic content is most important for the purpose of evaluation of crude drugs for its pharmacological activity.

Comparisons on total phenolic content of hexane and chloroform extract of leaves are shown in table 2: Total phenolic content was found to be higher in chloroform than hexane extract when compared with a standard of gallic acid.

Table 2: Total phenolic content of different extracts of *Majidea zangueberica* leaves.

S. No	Sample	Phenols mg/g dry tissue
1	Hexane	2.46 ± 0.11
2	chloroform	3.83 ± 0.12

3.3. GC-MS analysis

GC-MS analysis of chloroform leaves extract of *M. zangueberica* showed the presence of 40 different compounds with various percentages is shown in Figure-1. The name, molecular weight, structure and biological activity of the components of the test materials were tabulated (Table 3). Out of these forty compounds, Naphthalene, decahydro-1,4-dimethyl-7-(1-methylethyl)- was found to be maximum (13.6%) with retention time of 22.410. The other important constituents such as the 9,12,15-octadecatrienoic acid, methyl ester (7.05), Hexadecanoic acid (6.86), 7-hexadecyn-1-ol (6.84), Eicosanoic acid (6.68) were found to be present.

The spectrum of compounds showing strong antibacterial, antioxidant, and anti-inflammatory activities was revealed by the GC-MS analysis such as, Naphthalene, decahydro-1, 4-dimethyl-7-(1-methylethyl) –was reported to having antiseptic

and carcinogenic activity. 9,12,15-octadecatrienoic acid, methyl ester- is an unsaturated fatty acid, reported to having anti-inflammatory, anti-arthritic, hypocholesterolemic, hepato-protective, anti-cancer, antihistaminic, anti-acne, nematocide, insectifuge and antieczemic properties. Hexadecanoic acid (palmitic acid) and Eicosanoic acid are saturated fatty acid, having anti-inflammatory, pesticide, hypocholesterolemic, flavouring agent, nematocide, lubricant, antioxidant and anti-androgenic [27], 3, 7, 11, 15-tetramethyl-2-hexadecen-1-ol -(phytol) is an important diterpene component of all plants such as chlorophyll and vitamin E and K, which used in cosmetics, shampoos, toilet soaps, household cleaners and it shows anesthetics, anticancer, antidiuretic activity and high antimicrobial against the food borne pathogens [28].

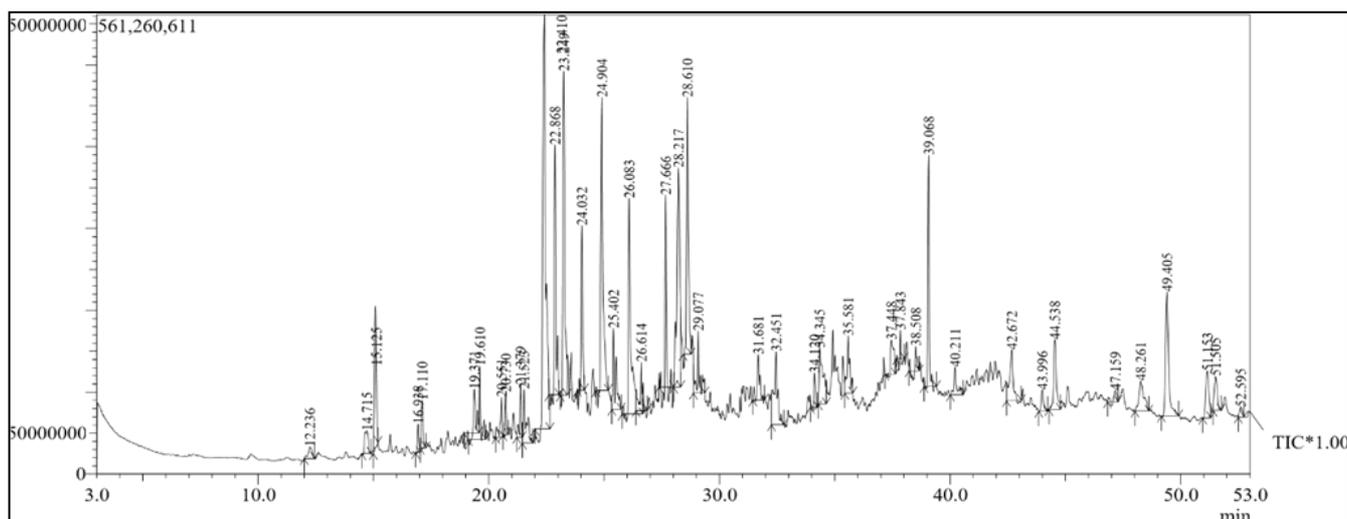
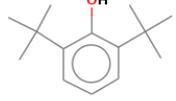
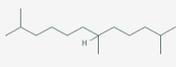
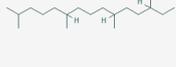
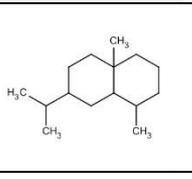
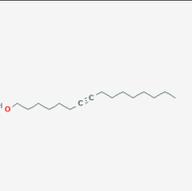
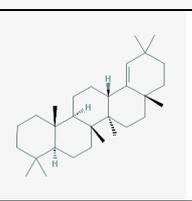
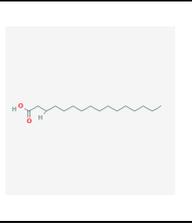
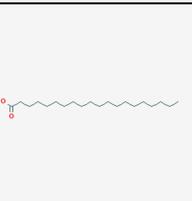
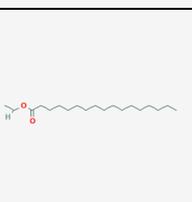
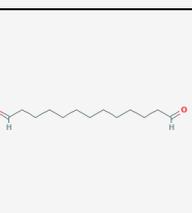
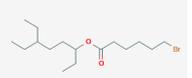
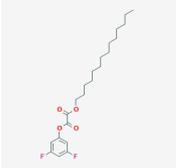
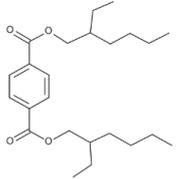
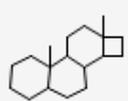


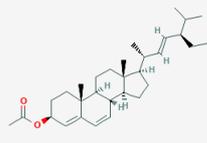
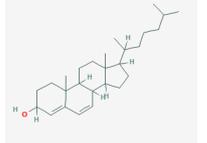
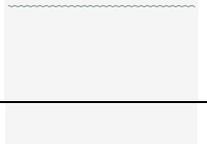
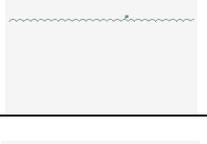
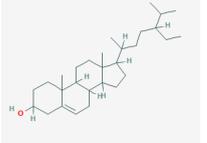
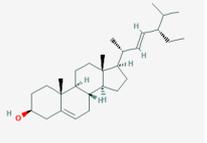
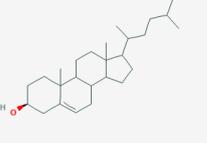
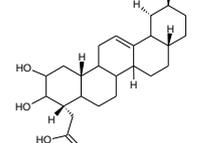
Fig 1: GC-MS analysis of phytochemicals identified from chloroform extracts of *Majidea zangueberica* leaves

Table 3: GC-MS analysis of phytochemicals identified from chloroform extracts of *Majidea zangueberica* leaves.

S No	Rt.	Name of th compound	Molecular formula	Molecular weight g/mol	Peak value	Structure	Biological activity
1	12.236	Heptad cane	C ₁₇ H ₃₆	240.475	0.51		Anti-inflammatory, antimicrobial, antioxidant. antifungal agent
2	14.715	Pentadecanal	C ₁₅ H ₃₂	212.421	0.84		Antimicrobial and antioxidant activity, Flavouring agents. Sugar-phosphatase inhibitor, Chymosin inhibitor.
3	15.125	Phenol, 2,6-bis(1,1-dimethylethyl)	C ₁₄ H ₂₂ O	206.3239	2.28		Antioxidant, antibacterial activity
4	16.938	9-eicosene, (E)-	C ₂₀ H ₄₀	280.54	0.48		Antimicrobial, cytotoxic.
5	17.110	2,6,11-Trimethyldodecane	C ₁₅ H ₃₂	212.421	0.87		Antimicrobial.
6	19.371	Tetrad cane	C ₁₄ H ₃₀	198.394	0.53		Antifungal, diuretic, anti-tuberculosis, nematocidal, antibacterial property
7	19.610	2,6,10,13-tetramethylpentadecane	C ₁₉ H ₄₀	268.529	2.64		Plasmacytomagenic activity.
8	20.551	Eicosane	C ₂₀ H ₄₂	282.556	0.84		Antioxidant, antifungal, antibacterial, anti-tumour, a precursor for the manufacture of synthetic forms of vitamin E and vitamin K1 and cytotoxic effect.
9	20.730	4-Methyldocosane	C ₂₃ H ₄₈	324.637	0.72		Anti-inflammatory, antimicrobial, antioxidant, diuretic activity, anticancer.
10	21.379	1-nonadecene	C ₁₉ H ₃₈	266.513	0.82		Antifungal, antituberculosis, anticancer activity.

11	21.523	Octadecane	$C_{18}H_{38}$	254.502	1.62		Antibacterial, antifungal.
12	22.410	Naphthalene, decahydro-1,4a-dimethyl-7-(1-methylethyl)-	$C_{15}H_{28}$	208.3828	13.06		Used as insecticide, cytotoxic activity.
13	22.868	3,7,11,15-tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	296.5310	4.55		Anti-inflammatory, antimicrobial, antioxidant, diuretic activity, anti-cancer.
14	23.249	7-hexadecyn-1-ol	$C_{16}H_{30}O$	238.415	6.84		No activity reported.
15	24.032	Olean-18-ene	$C_{30}H_{50}$	410.73	2.42		Cytotoxic activity, antitumor activity, HIV inhibition.
16	24.904	Hexadecanoic acid	$C_{16}H_{32}O_2$	256.43	6.86		Antibacterial, major part of human breast milk, food additive, anti-inflammatory, antioxidant, hypocholesterolemic, nematocide, pesticide, antiandrogenic, hemolytic, 5-alpha reductase inhibitor. Antidiarrheal activities, antitumor.
17	25.402	Nonadecyl trifluoroacetate	$C_{21}H_{39}F_3O_2$	380.536	2.11		No activity reported.
18	26.083	Eicosanoic acid	$C_{20}H_{40}O_2$	312.538	6.68		Antimicrobial activity.
19	26.614	Heptadecanoic acid, ethyl ester	$C_{19}H_{38}O_2$	298.5038	1.20		Antioxidant and antimicrobial activity.
20	27.666	Tridecanedial	$C_{13}H_{24}O_2$	212.333	2.07		No activity reported

21	28.217	9,12,15-octadecatrienoic acid, methyl ester	$C_{19}H_{32}O_2$	292.46	7.05		Antibacterial activity. Anti-inflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematocide, insectifuge antihistaminic, antiarthritic, anticoronary, antieczemic antiacne, 5-alpha reductase inhibitor antiandrogenic.
22	28.610	1,2,4-trioxolane-2-octanoic acid,5-octyl-,methyl ester	$C_{19}H_{36}O_5$	344	5.08		No activity reported.
23	29.077	1-eicosanol	$C_{20}H_{42}O$	298.555	1.58		Antitumor, antimalarial, antifungal, antioxidant.
24	31.681	6-bromoheptanoic acid, 5-ethyl-3-octyl ester	$C_{16}H_{31}BrO_2$	35.326 334	1.19		No activity reported
25	32.451	Nonadecyl heptafluorobutyrate	$C_{23}H_{39}F_7O_2$	480.552	2.89		No activity reported.
26	34.120	Oxalic acid, 3,5-difluorophenyl tetradecyl ester	$C_{22}H_{32}F_2O_4$	398.491	0.52		No activity reported
27	34.345	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{19}H_{38}O_4$	330.5026	2.42		Antioxidant, anti-inflammatory and Anthelmintic activities.
28	35.581	Behenic alcohol	$C_{22}H_{46}O$	326.6000	1.61		Antimicrobial), antiviral activity against HSV.
29	37.448	Octadecanoic acid, 2,3-dihydroxypropyl ester	$C_{21}H_{42}O_4$	358.5558	1.16		Anticancer, antimicrobial.
30	37.843	Terephthalic acid, di(2-ethylhexyl) ester	$C_{24}H_{38}O_4$	390.56	0.01		Retinoidal activity.
31	38.508	1-octacosanol	$C_{28}H_{58}O$	410.771	0.27		Antinociceptive, anti-inflammatory, antibacterial, antioxidant.
32	39.068	d-Norandrostane (5.alpha., 14.alpha.)	$C_{18}H_{30}$	246	3.50		Antibacterial activity.

33	42.672	(22E)-stigmasta-4,6,22-trien-3-yl acetate	C ₃₁ H ₄₈ O ₂	452.723	1.90		Anticancer, antimicrobial, anti-inflammatory, antiarthritic, antiasthma, diuretic.
34	43.996	Cholesta-4,6-dien-3-ol	C ₂₇ H ₄₄ O	384.648	0.52		Antimicrobial.
35	44.538	Hexacontane	C ₆₀ H ₁₂₂	843.636	2.13		No activity reported.
36	47.159	Tetrapentacontane	C ₅₄ H ₁₁₀	759.474	0.29		Anti-inflammatory, anti-oxidant.
37	48.261	Stigmast-5-en-3-ol	C ₂₉ H ₅₀ O	414.707	1.65		Antimicrobial activity, anti-tumour activity, analgesic, anti-inflammatory, anti-mutagenic, antipyretic, anthelmintic, apoptotic, chemo protective, hypocholesterolemic, angiogenic, genotoxic, anti-oxidant, anti-diabetic, antimicrobial antifungal infections.
38	49.405	Stigmasterol	C ₂₉ H ₄₈ O	412.702	4.30		Anti-osteoarthritic effect, hypoglycaemic effect, antioxidant, anti-microbial, anti-mutagenic activity, anti-inflammatory activity., inhibit tumor promotion, anti HIV reverse transcriptase.
39	51.505	Ergost-5-en-3-ol, (3.beta.,24R)	C ₂₈ H ₄₈ O	400.691	1.12		Antioxidant, hypocholesterolemic.
40	52.595	Methyl commate D	C ₃₁ H ₅₀ O ₄	486	0.30		Antimicrobial, anti-inflammatory.

3.4. Antioxidant activity

Comparisons on antioxidant property of hexane and chloroform extract of leaves are shown in table 4: chloroform extract have higher antioxidant ability than hexane extract when compared with a standard BHT. The antioxidant inhibition percentage of chloroform and hexane were 69.75±0.086 and 41.53±0.073 respectively.

Table 4: The antioxidant property of chloroform and hexane extract of *Majidea zangueberica* leaves.

Sample	Concentration	% Inhibition
Chloroform (MZ-C)	500µl	69.75±0.086
Hexane (MZ-H)	500µl	41.53±0.073
BHT	500µl	83.839±2.32

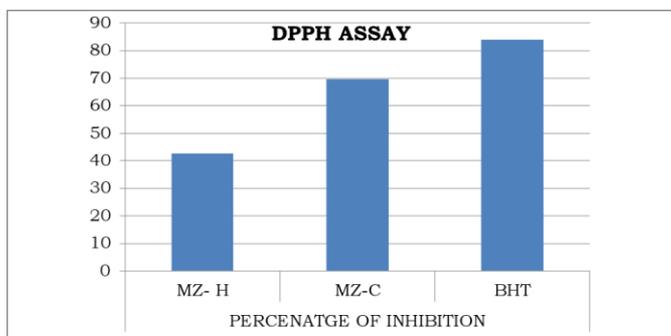


Fig 2: Antioxidant activity of chloroform and hexane extracts of *Majidea zangueberica* leaves.

The Graphical representation of comparison of total phenol and antioxidant scavenging activity percentage showing the chloroform extract of *M.zangueberica* leaves showed higher than the hexane extract. That is there was a positive linear correlation between the antioxidant activity and total phenolic content.

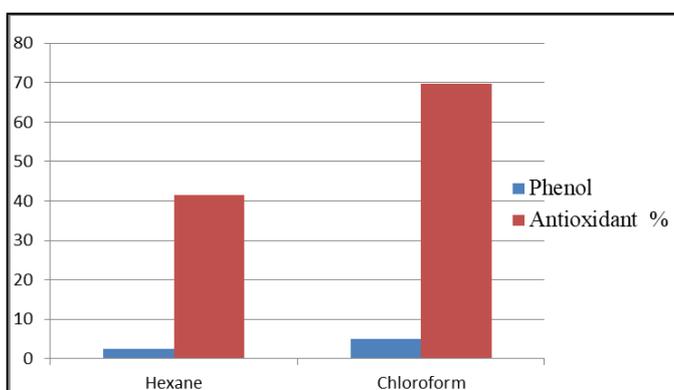


Fig 3: comparison of total phenol and antioxidant inhibition % of *M. zangueberica* in different extract.

4. Conclusion

In the present study the phytochemical screening (qualitative and quantitative), GCMS, and antioxidant activity of *M. zangueberica* leaf extracts composed of various phytoconstituents having antibacterial, antioxidant, anti-inflammatory activities and other several medicinal properties. Thus these types of analyses are the primary step towards understanding the nature of active principles in this ornamental plant which will be helpful for further research.

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