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Original Article

In vitro Evaluation of Antioxidant Property, Chemo-profiling, Elemental Analysis of *Morinda umbellata* L.

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ABSTRACT

Background: *Morinda umbellata* L., commonly called as 'Manjanatti Kodi' in Tamilnadu, India, is a rare traditional medicinal plant. The study was aimed to investigate the antioxidant potentiality, phytochemical profiling and elemental mineral analysis of *M. umbellata* leaves. **Materials and Methods:** Antioxidant activity was estimated for hexane, chloroform, ethyl acetate, ethanol, methanol and aqueous leaf extracts by DPPH method; identification of phytocompounds by Gas chromatography–mass spectrometry (GC-MS) analysis and elemental mineral estimation by Atomic absorption spectroscopy (AAS) analysis. **Results:** Antioxidant activity was determined in all the extracts studied, where the maximum 50% inhibitory concentration was registered in ethanolic (17.92±0.04 µg/ml) and methanolic (19.35±0.60 µg/ml) extracts which was four times higher than standard ascorbic acid. About 19 phytocompounds were found in antioxidant rich ethanolic leaf extract by GC-MS analysis. Among the identified phytocompounds, 1,2-Benzenedicarboxylic acid, diethyl ester with a percent area of 94.98%, Guanosine (0.79%), Hexadecanic acid (0.72%), phytol (0.47%) were found to be predominant. The elemental mineral analysis is the first-hand report, that revealed essential elements such as Fe (5.65±0.58 ppm), Zn (0.74±0.05 ppm), Mn (0.46±0.04 ppm), and trace amounts of following non-essential elements (heavy metals) Cu (0.87±0.05 ppm), As (0.002±0.00 ppm), Cd (0.08±0.01 ppm), Pb (0.03±0.00 ppm), Hg (BDL). **Conclusion:** *M. umbellata* leaves were proved to possess strong antioxidant property due to an inclusive source of important bioactive components. It was also found to be non-toxic and the plant is a right choice for drug development. Hence, the present outcome provided a reliable scientific frame work, for the exploration of bioactive potentiality with pharmaceutical importance in future studies.

Keywords: Antioxidant activity, GC-MS, Elemental analysis, *Morinda umbellata,* Traditional medicinal plant.

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INTRODUCTION

Plants have been used as a potent source of medicine for various ailments and with the anticipation to maintain human health. Traditional medicine is the primary mode of health care for most of the population in India.[1] From time immemorial, the traditional Indian system of medicine relied on medicinal plants owing to their local availability, cost effective and no side-effects criteria. Universally, the therapeutically important medicinal plants were considered as 'herbal drugs' or 'natural medicine' which attracted the major group of population in the preparation of traditional medicines. About 80% of medicinal plants were used as traditional medicines in the form of pure extracts, of which 70% were authenticated as human medicine by World Health organization (WHO) survey report.[2] Recent research thirst for newer sources of natural antioxidants is spontaneously increasing due to the rising consumer needs for natural products. The use of synthetic drugs needs to be avoided since it leads to carcinogenic effects and side effects in the human body system. Plants are a significant source of natural antioxidants, that encompasses a massive quantity of secondary metabolites, mainly utilized for their defense mechanisms.[3] Antioxidant are important components of human health to safeguards the biological system as they counter against reactive oxygen species (ROS) or free radicals in the human body. These ROS are considered as toxic to body metabolism and is a major factor for cancer development. Antioxidants as dietary supplements or as medicine, should be considered as mandatory during intake, thus forming a balanced diet along with all other sources of nutrients such as vitamins, minerals, proteins, and iron. Reports on medicinal plant parts that acts as antimicrobial, anticancer, anti-inflammatory and other biological properties were because of phytochemicals like polyphenols, alkaloids, terpenoids, flavonoids as well other secondary metabolites.[4]

Heavy metal contamination in medicinal plants causing toxic ill effects during human consumption is one of the imperative constraints of quality, when considered as herbal drug. The basic plant metabolism requires macronutrients (O, H, C, K, P, N, S, Ca and Mg) and micronutrients (Cu, Ni, Zn, Co, Cl, Mn, B, Fe and Mo) and are known to be essential elements. Whereas, non-essential elements such as heavy metals (Ar, Cd, Pb, Hg) has no biological importance alter the water balance and nutrient assimilation and produce toxic effects in plants. They enter into human body via heavy metal contaminated food, drinking water, and air, causing severe effects in human biochemistry and is considered as great human health concern. Therefore, it is an important factor to profile the toxic effect of medicinal plants before the preparation of the drug to avoid the adverse effect in our biological system.[5] To screen out the healing agents (phytochemicals) from the medicinal plants, chemical profiling is another important factor for proper identification and purification of organic compounds from plants. Gas Chromatography and mass spectroscopy (GC-MS) is a well-known analytical technique that distinguishes the individual chemical component of a sample mixture used in the identification of various compounds especially phytocompounds.

Morinda umbellata L. is a rare traditional medicinal plant, a member of Rubiaceae family. The plant is a climbing shrub bearing umbel inflorescence with bright red colour fruits grows in high altitude regions in southern India, Sri Lanka, Deccan Peninsula to Burma, Southeast Asia, China, Philippines, Japan and Northern Australia. It is commonly known as 'Manjanatti Kodi' in Tamil and 'Pitadaru' in Sanskrit. According to the local medicinal practitioners and other literature survey implies that this medicinal plant has potent medicinal properties. Traditionally, the leaves are used to treat dysentery and diarrhea. Fruit exhibit laxative property and regularize the menstruation cycle in women.[6] The experimental plant *M. umbellata* possesses significant medicinal property and very fewer research work has been done so far. Hence, this plant is considered as a choice of selection for bioactive exploration with pharmaceutical importance, to study the antioxidant property, heavy metal components, and elucidate bioactive chemical constituents from *M. umbellata* a rare traditional medicinal plant.

MATERIALS AND METHODS

Collection and Preparation of Plant

The experimental plant *Morinda umbellata* L. was collected from Sirumalai hills, Dindigul district of Tamil Nadu, South India. The plant specimen (Voucher specimen No.2936) was authenticated by Rapinet herbarium, Department of Botany, St. Joseph's College (Autonomous), Tiruchirappalli and maintained in the laboratory. The fresh and healthy leaves were collected with the help of tribal people. The fresh leaf sample was washed and dried under shade for about ten days. The damaged and infected leaves were discarded and healthy dried leaves were grounded with the crusher. The crushed material was reground and the fine powder was stored in an airtight container for further experimental purpose. The hot continuous or Soxhlet (Franz von soxhlet) extraction procedure with sequential extraction of solvent gradient was carried out. Powdered leaf sample along with six solvents from low polarity to high polarity such as hexane<chloroform<ethyl acetate<ethanol<methanol<aqueous were used sequentially to get their respective extracts. The collected respective solvent extract was completely reduced to get the thick crude extract. The extract was stored in amber bottles and refrigerated for further analytical purposes.

In vitro Antioxidant Assay

The determination of antioxidant activity was performed by 2,2-diphenyl-1-picrylhydrazyl radical scavenging activity, where 2,2-diphenyl-1-picrylhydrazyl (DPPH) act as free radical. The free radical scavenging activity and 50% inhibitory concentration was determined at different concentrations (20, 40, 60, 80 and 100 µg/ml). The reaction mixture was read at 517nm in UV-Vis spectrometry and the percentage of antioxidant activity (AA%) was calculated using the formula, $(A_{blank} - A_{sample} / A_{blank}) \times 100$, where A_{blank} is the absorbance of control and A_{sample} is absorbance of plant extract. The maximum 50% inhibitory concentration (IC_{50}) was calculated using linear regression plots.[7]

Chromatographic Analysis

The bioactive compounds present *in M. umbellata* ethanolic leaf extract was predicted by Gas Chromatography combined with Mass Spectrometry method (PerkinElmer's TurboMass™ Version 5.2.0)[8] (Shimadzu Corporation, containing an AOC-20i autoinjector) fused silica column (Rtx 5Ms, 0.32 mm X 30 m X 0.50 µm), 5 µl was injected with helium gas at a constant flow rate of (1.73 ml /min). Injection temperature (270°C) and ion source temperature (200°C) was maintained. The oven temperature at 40°C (2 min), with an increase of 8°C/min, 150°C (10min), 250°C (15min), for 20 min at 280°C was set. Mass spectra were predicted at 70 Electron volt (eV) with scanning interval (0.5 sec) and the mass spectral fragments were obtained from 40 to 450 Da. Total GC running time was 51.25 min. Interpretation on Gas Chromatographic and Mass Spectrometry was made, via WILEY7.LIB. The nature of the bio active compounds of the plant *M. umbellata* were determined by comparison of compounds deposited in the NIST library.

Mineral and Heavy Metal Analysis

The mineral composition of *M. umbellata* was determined by Atomic absorption spectroscopy (AAS) analysis. The sample (100gm of plant powder) were ashed in muffle furnace and the ash obtained was digested with con. HCl. Then, it was filtered and made up to 100 ml.[9] The concentrations of minerals and heavy metals such as Copper, Zinc, Manganese, Iron and Arsenic, Cadmium, Lead, Mercury were quantified by flame AAS by PerkinElmer model Analyst 200 (USA). The absorbance was read at 470 nm in UV-Vis spectrophotometer. Mineral and heavy metal contents were expressed in ppm.[10]

Statistical Analysis

The experiment was carried out in triplicates. The results were calculated using SPSS (version 28.0.1.0 (142) IBM) and its group differences were analyses using one-way ANOVA. The data was expressed as mean±standard error and the significant differences of mean values were represented at $p<0.01$. The maximum 50% inhibitory concentration (IC_{50}) was calculated by the linear regression method.

RESULTS

DPPH Radical Scavenging Activity

The determination of antioxidant activity (AA) of *M. umbellata* leaves were carried out for hexane<chloroform<ethyl acetate<ethanol<methanol<aqueous solvent gradient system based on sequential polarity of low polar to high polar solvents. Among the six solvent extracts examined, the maximum 50% inhibitory concentration (IC_{50}) of 17.92±0.04 µg/ml and 19.35±0.60 µg/ml was recorded in ethanol and methanol extracts. Whereas other solvent extracts exhibited a moderate activity at 50% inhibitory concentration of 25.98±0.57 µg/ml in hexane extract, 33.94±0.58 µg/ml in chloroform extract, 46.55±0.63 µg/ml ethyl acetate extract and 51.54±0.60 µg/ml in aqueous extract. Corresponding to the extract concentration dependant manner, as and when the concentration of plant extract increases, the free radical (DPPH) scavenging activity also increased. The control used for AA determination was standard ascorbic acid. Among all other solvent extracts, the highest range of percentage scavenging activity was obtained in ethanol extract at 100µg/ml (17.92±0.04 µg/ml) which is four times higher than the control (66.16 ± 0.63 µg/ml). The results of antioxidant activity were tabulated in Table 1 and depicted in Figure 1 and 2.

Chemo-Profiling by GC-MS analysis

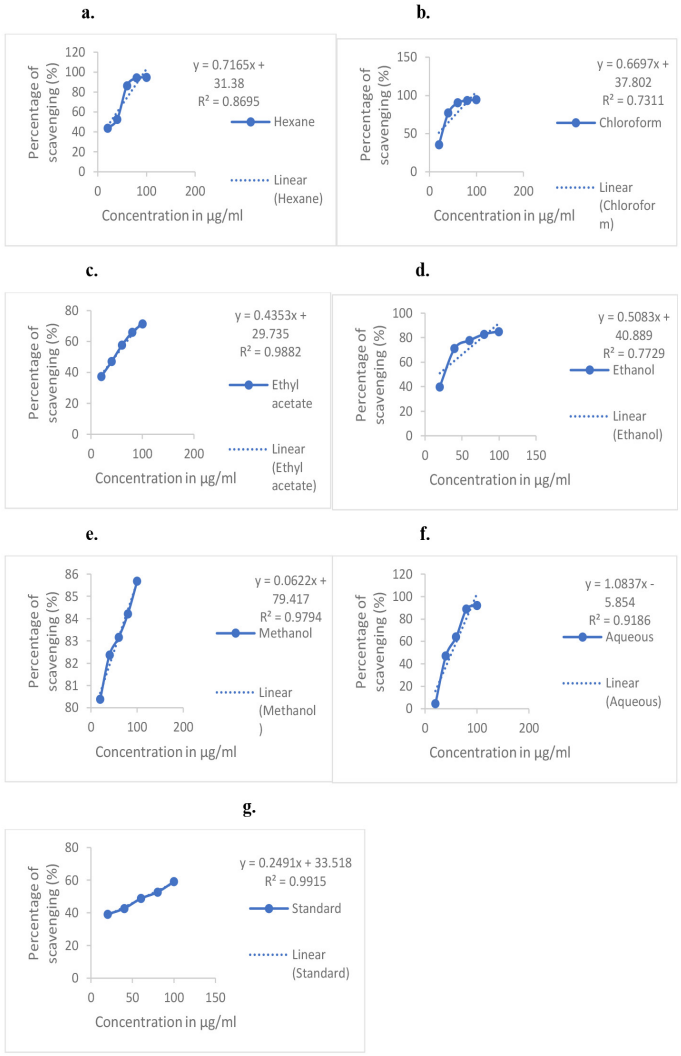
The Gas Chromatography and Mass Spectrometry (GC-MS) study was undertaken to elucidate bioactive compounds present in *M. umbellata* leaf. Based on the results of antioxidant activity on the solvent extracts studied, it was found that the ethanolic leaf extract expressed the maximum activity. Therefore, ethanolic extract was chosen for screening of chemo profiling. About 19 phytocompounds were identified and the spectrum of unknown compounds were compared with NIST library. All the compounds documented from *M. umbellata* exhibited some significant properties and some of the compounds revealed high antioxidant, anticancer properties. The predominant compound identified was 1,2-Benzenedicarboxylic acid, diethyl ester with percent area 94.98%, followed by 1-Undecene (0.03%), Guanosine (0.79%), 1-Undecanol (0.04%) Ethyl 1-hexyl-4-hydroxy-2(1H)-oxo-3-quinolinecarboxylate (0.01%), 1,3,4,5,-Tetrahydroxy-cyclohexanecarboxylic acid (0.78%),

Table 1: Antioxidant activity of different extracts of *M. umbellata* leaf by DPPH method

S. no	Different solvent extracts	Concentration of sample (µg/ml)	Percentage of Scavenging (%)	IC_{50} Value µg/ml
1.	Hexane	20	43.75±0.024	
		40	52.67±0.191	
		60	86.60±0.005	25.98±0.57
		80	94.19±0.003	
		100	94.64±0.003	
2.	Chloroform	20	35.48±0.022	
		40	76.78±0.021	
		60	90.17±0.004	33.94±0.58
		80	93.30±0.002	
		100	94.19±0.002	
3.	Ethyl acetate	20	37.43±0.034	
		40	46.94±0.079	
		60	57.49±0.003	46.55±0.63
		80	65.97±0.006	
		100	71.45±0.004	
4.	Ethanol	20	40.12±0.001	
		40	71.14±0.002	
		60	77.73±0.001	17.92±0.04
		80	82.83±0.003	
		100	85.10±0.002	
5.	Methanol	20	80.37±0.003	
		40	82.36±0.004	
		60	83.15±0.002	19.35±0.60
		80	84.21±0.002	
		100	85.67±0.001	
6.	Aqueous	20	4.46±0.003	
		40	46.87±0.003	
		60	63.83±0.004	51.54±0.60
		80	88.83±0.003	
		100	91.85±0.003	
7.	Standard	20	39.14±0.001	
		40	42.70±0.001	
		60	48.75±0.002	66.16±0.63
		80	52.66±0.001	
		100	59.07±0.001	

*Values are expressed as Mean ± Standard Error. IC_{50} values are the results of linear regression of % scavenging activities with different extracts of different concentration at 50% inhibition.

2,6,8-Trimethyl-4-nonanone (0.28%), 1,2-Benzenedicarboxilic acid, diethyl ester (0.14%), Pentadeconoic acid (Pentadecyclic acid (0.15%)), 1-Pentadecanol (0.03%), 2-Hexadecen-1-ol, 3,7,11,15-tetramethyl (0.30%) 1,2-Benzenedicarboxylic acid, diethyl ester (0.08%), 2-Hexadecen-1-ol,3,7,11,15-tetramethyl (0.05%), Cyclododecanol (0.13%), Hexadecanoic acid (0.72%), Phytol (0.47%), 9,12-Octadecadienoic acid (0.24%), 9,12,15-Octadecatrienoic acid, linolenate (0.71%), Docosanioic acid (0.09%). The identified active



a.Hexane extract b. Chloroform extract c. Ethyl acetate extract d. Ethanol extract e. Methanol extract f. Aqueous extract g. Standard (Ascorbic acid)

Figure 1: Inhibitory concentration (IC_{50}) of antioxidant property of *M. umbellata* leaf extracts.

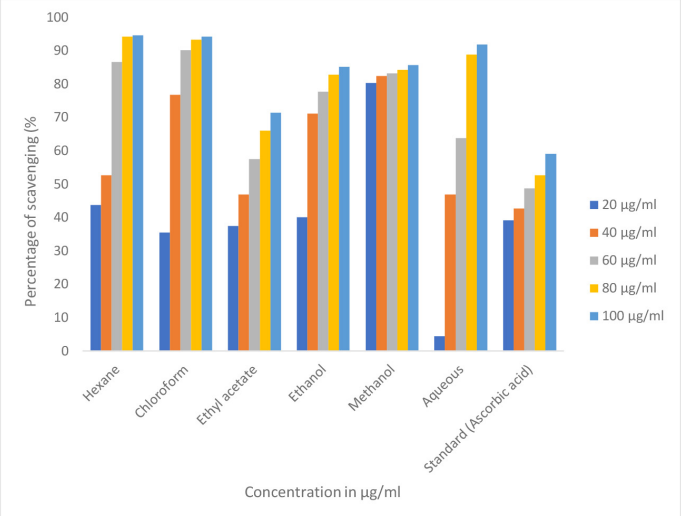


Figure 2: Percentage of scavenging activity of *M. umbellata* leaf extracts.

principles or bioactive compounds along with their retention time (RT) were tabulated in Table 2, and the chromatogram with spectral features of ethanolic leaf extract was depicted in Figure 3. Based on the literature survey, some of the identified compounds have been reported with biological activity and some of the other compounds have not yet been reported with any biological activity were presented in Table 2.

Determination of Mineral and Heavy Metal Contents

Following the present investigation on *M. umbellata*, it was found that the leaf possesses strong antioxidant properties due to the presence of highly active biological components. Keeping in this view, further elemental and heavy metal determination was undertaken to brighten the elements present in the experimental plant specimen. Since medicinal plants play a significant role in drug formulation, it is much important to

Table 2: GC-MS analysis of phytochemical components in ethanol leaf extract of *M. umbellata*.

Peak #	R. Time	Area%	Height%	Molecular weight (g/mol)	Molecular formula	Molecular name	Biological activity
1	7.223	0.03	0.10	154	$C_{11}H_{22}$	1-Undecene	Not yet Reported
2	8.209	0.79	0.46	283	$C_{10}H_{13}N_5O_5$	Guanosine	Not yet Reported
3	9.742	0.04	0.11	172	$C_{11}H_{24}O$	1-Undecanol	Not yet Reported
4	10.067	94.98	93.28	222	$C_{12}H_{14}O_4$	1,2-Benzenedicarboxylic acid, diethyl ester	Antifouling, Antimicrobial.[11]
5	10.417	0.01	0.03	355	$C_{22}H_{13}NO_4$	Ethyl 1-hexyl-4-hydroxy-2(1H)-oxo-3-quinolinecarboxylate	Not yet Reported
6	10.630	0.78	0.46	192	$C_7H_{12}O_6$	1,3,4,5-Tetrahydroxy-cyclohexanecarboxylic acid	Not yet Reported
7	10.775	0.28	0.29	184	$C_{12}H_{24}O$	2,6,8-Trimethyl-4-nonanone	Not yet Reported
8	11.086	0.14	0.33	222	$C_{12}H_{14}O_4$	1,2-Benzenedicarboxylic acid, diethyl ester	Not yet Reported
9	11.804	0.15	0.22	242	$C_{15}H_{30}O_2$	Pentadecanoic acid (Pentadecylic acid	Not yet Reported
10	12.087	0.03	0.09	228	$C_{15}H_{32}O$	1-Pentadecanol	Not yet Reported
11	12.630	0.30	0.68	296	$C_{20}H_{40}O$	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl	used in the fragrance industry, production of cosmetics, shampoos, toilet soaps, household cleaners, and detergents.[11]
12	12.807	0.08	0.15	222	$C_{12}H_{14}O_4$	1,2-Benzenedicarboxylic acid, diethyl ester	Not yet Reported
13	12.902	0.05	0.12	296	$C_{20}H_{40}O$	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl	Not yet Reported
14	13.113	0.13	0.25	184	$C_{12}H_{24}O$	Cyclododecanol	Not yet Reported
15	13.982	0.72	1.21	256	$C_{16}H_{32}O_2$	Hexadecanoic acid	Production of soaps, cosmetics, and release agents.[11]
16	15.559	0.47	0.96	296	$C_{20}H_{40}O$	Phytol	Antimicrobial, Anticancer, Cancer preventive, Diuretic, Antiinflammatory.[11]
17	15.833	0.24	0.34	280	$C_{18}H_{32}O_2$	9,12-Octadecadienoic acid	Antiinflammatory, hypocholesterolemic cancer preventive, hepatoprotective, nematicide, insectifuge, antihistaminic antieczemic, antiacne, 5-Alpha reductase inhibitor, antiandrogenic, antiarthritic, anticoronary, insectifuge.[11]
18	15.921	0.71	0.79	292	$C_{19}H_{32}O_2$	9,12,15-Octadecatrienoic acid, linolenate	Hypocholesterolemic, Nematicide Antiarthritic, Hepatoprotective, Anti androgenic, Nematicide, 5-Alpha reductase inhibitor, Antihistaminic, Anticoronary, Insectifuge, Antieczemic, Anticancer, Antibacterial, Antifungal.[11]
19	16.083	0.09	0.14	340	$C_{22}H_{44}O_2$	Docosanoic acid	Not yet Reported

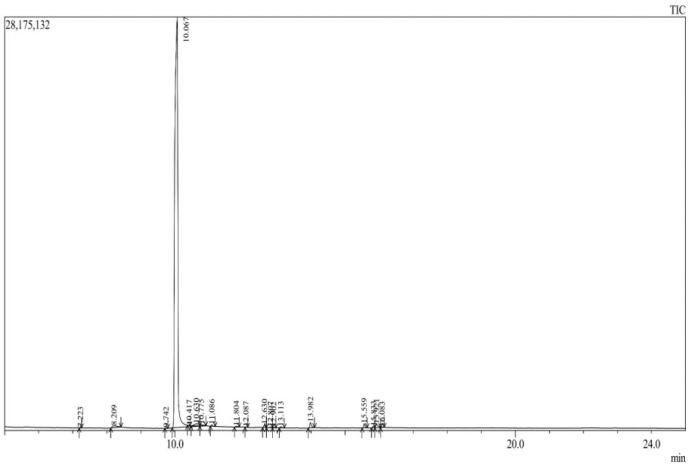


Figure 3: Gas chromatography and mass spectrum (GC-MS) of ethanolic leaf extract of *M. umbellata*.

Table 3: Mineral and heavy metals content of *M. umbellata* leaf

Sl. No.	Microlements and Heavy metals	Concentration (ppm)	*Permissible limit of FAO/WHO (ppm)
1.	Copper	0.87±0.05	10
2.	Zinc	0.74±0.05	5
3.	Manganese	0.46±0.04	0.5
4.	Iron	5.65±0.58	20
5.	Arsenic	0.002±0.00	0.05
6.	Cadmium	0.08±0.01	0.2
7.	Lead	0.03±0.00	2
8.	Mercury	BDL	0.5

*Data were expressed as mean ± standard error of triplicates (n=3). BDL refers to below detection limit.

*Source: WHO 1996.

screen whether the plant is contaminated by heavy metals. The present study revealed that the quantity of heavy metals and minerals in the *M. umbellata* leaf, were 0.87±0.05ppm of copper, 0.74±0.05ppm of zinc, 0.46±0.04ppm of manganese, 0.002±0.00ppm of arsenic 0.08±0.01ppm of cadmium, 0.03±0.00ppm of lead, mercury (BDL), and 5.65±0.58ppm of iron respectively. As per the present results, depicted in Table 3 it is indicated that the plant *M. umbellata* serves as a potent source of essential minerals and found to be less toxic as it consists of trace amounts of heavy metals (Figure 4). The minerals and heavy metals of *M. umbellata* were found to be below the recommended levels of FAO or WHO expert committee on food additives.[11]

DISCUSSION

The genus *Morinda,* has been widely known in Indian traditional system of medicine for its therapeutical potential against human ailments. Among various species, a rare traditional medicinal plant *M. umbellata* has been found to be a less studied member of the family Rubiaceae. Traditionally, the plant has been recognized as an important medicinal plant with various medicinal properties, but scientifically the plant needs a systematic examination. Thus, the present study was focused to explore the bioactive potentiality of *M. umbellata* leaves with pharmaceutical importance.

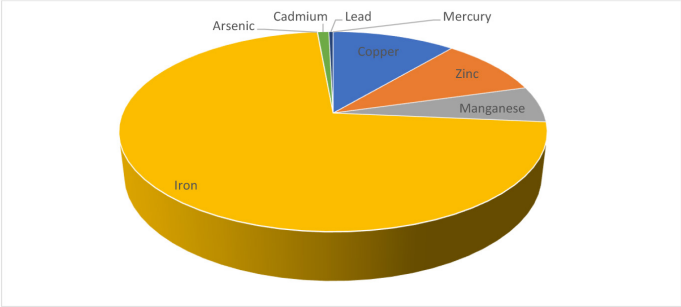


Figure 4: Concentration of minerals and heavy metals in *M. umbellata* leaf.

The antioxidant property of *M. umbellata* leaves was well established in all the solvent extracts studied. Especially the ethanol and methanolic leaf extracts revealed a higher antioxidant activity, which could be known as a potent antioxidant source used to treat and prevent various dreadly human diseases. The present results are in correlation with few reports where ethanolic extracts exhibited a maximum antioxidant activity in dried flowers of *Tithonia diversifolia* (205.80 µg/ml)[12] and ethanolic extract of *Carica papaya* leaf (151.36 µg/ml).[13] A maximum radical scavenging activity was reported in methanolic extracts[14] of *Rubus ellipticus* (11.01 µg/ml) and *R. niveus* (12.48 µg/ml). Biomolecules that inhibit the activity of free radicals and delay the oxidative damage of protein, lipids are known as antioxidants. These biomolecules might be responsible in the prevention of diseases like cancer, inflammation, diabetes, asthma, and degenerative eye disorder.[15,16] Phenolic compounds such as flavonoids and tannins are the major sponsors of the antioxidant capacity in plants.[17,18] Whereas, free radicals (ROS) impair DNA, proteins, lipids in human body system thereby causing serious illness due to oxidative stress such as cardiovascular diseases, cancers, neurological disorders. Usually, antioxidants degrade the ROS mediated oxidation process by prevention of free radical production through scavenging activity.[2] It was known that beneficial effects of both polyphenols and other essential oils were the major cause for the radical scavenging property.[3]

The antioxidant capacity of *M. umbellata* leaf might be due to presence of various bioactive constituents where profiling and identification of these components is much important. Among the 19 phytoconstituents identified, six compounds having high biological activity such as 1,2-Benzenedicarboxylic acid, diethyl ester (R*t*=10.067) constantly having antifouling and antimicrobial property, 2-Hexadecen-1-ol,3,7,11,15-tetramethyl (Rt=12.630) is the precursor for the manufacture of synthetic forms of Vitamin E and vitamin K1 used in the fragrance industry and preparations of cosmetics, shampoos and soaps.[19] Hexadecanoic acid (Rt=13.982) possesses palmitic acid is used to produce soaps and cosmetics.[20] Phytocompounds such as 9,12-Octadecadienoic acid (R*t*=15.833) possessing cancer-preventive, antiacne, hypocholesterolemic, anti-inflammatory, antieczemic, hepatoprotective, antiandrogenic, antihistaminic, antiarthritic, 5-Alpha reductase inhibitor, anticoronary and 9,12,15-Octadecatrienoic acid methyl ester (Rt=15.921) possessing hypocholestrolemic, antiarthritic, antieczemic, anticancer, antibacterial and antifungal properties have been reported.[21,22] Phytol (0.47%) possess anticancer property and has various applications in several industries, also it is used as a key ingredient in production of several fragrance and cosmetic products.[23] It was observed that *M. umbellata* possesses significant bioactive compounds which fortifies that the plant could be used in the exploration of effective drug candidate against various human ailments especially for various types of cancers.

Some of the essential elements in plants possess significant importance in the metabolic process of growth and development in human health but the quantity of those elements is of great concern when chosen for diet and medicine.[24] Copper is an important trace element for both vegetative and reproductive development of plants but might be toxic at excessive levels. It plays a vital role in cell metabolisms like physical growth, the progress of brain development, bone formation, and wound healing mechanisms. The dietary limit of zinc is 100ppm[25] similar study reported that spare amount of zinc can cause jaundice.[26] Iron deficiency is common nutritional issue in humans, caused due to improper diet practices.[24] Lead is known for its toxicological property considered as a universal pollutant detected in all biological and environmental systems.[27] The long-term exposure of low-level cadmium slowly affects the antioxidant enzymes[28-30] and this inhibition can lead to the amplified level of oxidative stress resulting in membrane damage and loss of enzymes like ATPases.[27] The presence of several bioactive compounds and elemental compositions subsidizes to the plant's healing properties.[31] Therefore, *M. umbellata* possesses significant medicinal property as it plays an eloquent role in human nutrition as a source of micronutrients, could serve as alternative drug for various human ailments and could be chosen as an effective toxic free drug candidate in the preparation of herbal formulations.

CONCLUSION

Hence, the current finding confirms the medicinal asset of *M. umbellata* and these properties might be due to various biologically active compounds and substantial secondary metabolites present in them. The exploration of bioactive compounds and their biological activity has paved the way to further investigations towards isolation, purification of active individual components present in *M. umbellata*. Therefore, considering all the obtained facts, *M. umbellata* leaf holds a strong antioxidant agent and could be considered as an imperative choice for pharmaceutical industries in the development of drugs against various dreadly diseases. The experimental plant is focused as a treasured source of therapeutic importance and serve as a potent therapeutant in the drug development process in further study. Thus, *M. umbellata* is a pharmaceutically important drug candidate which could serve a significant role in the betterment of healthy human lives.

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CONFLICT OF INTEREST

The authors declare that there was no conflict of interest.

ABBREVIATIONS

%: Percentage; **°C:** Degree Celcius; **e V:** Electron volt; **µg:** Microgram; **ml:** Millilitre; **AA:** Antioxidant Activity; **AAS:** Atomic Absorption Spectrometry; **ANOVA:** Analysis of Variance; **Ar:** Argon; **As:** Arsenic; **Ar:** Argon; **Ablank:** Absorbance of blank; **Asample:** Absorbance of sample; **B:** Boron; **BDL:** Below Detection Limit; **C:** Carbon; **Ca:** Calcium; **Cd:** Cadmium; **Cl:** Chlorine; **Co:** Cobalt; **Conc. HCl:** Concentrated Hydro Chloric Acid; **Cu:** Copper; **DPPH:** 2,2-diphenyl-1-picryl-hydrazyl-hydrate; **DNA:** Deoxyribonucleic acid; **FAO:** Food and Agriculture Organization; **Fe:** Iron; **GC-MS:** Gas Chromatography – Mass Spectrometry; **H:** Hydrogen; **Hg:** Mercury; **IC_{50}:** 50% inhibitory concentration; **K:** Potassium; **Mg:** Magnesium; **Min:** Minutes; **Mn:** Manganese; **Mo:** Molybdenum; **N:** Nitrogen; **Ni:** Nickel; **NIST:** National Institute of Standards and Technology; **O:** Oxygen; **P:** Phosphorus; **Pb:** Lead; **Ppm:** Parts per million; **R_t:** Retention time; **ROS:** Reactive Oxygen Species; **S:** Sulphur; **SPSS:** Statistical Package for Social Sciences; **USA:** United States of America; **UV-Vis:** Ultraviolet-visible; **WHO:** World Health Organization; **Zn:** Zinc;

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