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Antioxidant Content of Selected Medicinal Plants Used by Kaani Tribes of Kanyakumari District in Tamilnadu India

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Abstract **Objectives:** Despite the great advances observed in modern medicine in recent decades, plants still make an important contribution to health care. The objectives of this study were to determine the moisture content and antioxidant capacity of the selected medicinal plants. **Methods and Materials:** The medicinal plants (20) were collected from Kanyakumari district of Tamilnadu. The moisture content of the medicinal plants was analyzed using Infra red moisture analyzer. For the analysis of antioxidants by DPPH (2,2-diphenyl-1-picryl hydrazyl assay) & FRAP (Ferric Reducing Antioxidant Power), phenol by Folin-ciocalteau method and flavonoids by aluminium chloride method the leaves was shade dried, powdered and stored in brown bottle containers. The aqueous and ethanol extracts of the medicinal plants were evaluated for their antioxidant capacity All recorded values of Moisture and Antioxidant analysis are mean of triplicates. **Results and Discussion:** The moisture content of the selected medicinal plants ranged from 36.00 to 89.86%. Blepharis Maderaspatens had highest DPPH activity in both aqueous (8.870) and ethanolic extracts (71.82) expressed in mg of GAE/g dry weight. The antioxidant activity AOA was 5.6 times higher in ethanolic extracts. The Mean DPPH activity of the selected medicinal plants was found to be 3.402±2.22 in aqueous extract and 19.114±17.01 in ethanolic extract. In FRAP, Blepharis Maderaspatens (633.76) had the highest AOA in ethanol extracts and Valarai (300.70) in water extracts expressed in µM of (Ascorbic Acid Equivalence Capacity) AAEC/g. The Mean FRAP value of the selected medicinal plants was found to be 145.44±129.53 in ethanolic extract and 117.96±79.17 in aqueous extracts. Blepharis Maderaspatens had the highest phenolic content in both aqueous (12.8) and ethanolic extracts (35.6). **Conclusion:** The study concluded that there was a wide difference in antioxidant content among the medicinal plants. The ethanolic extracts revealed the presence of high concentrations of AOA, Total Phenol Content (TPC), and Total Flavanoid Content (TFC) in the medicinal plants than water. Further investigation in quantification of individual antioxidant components in these medicinal plants possessing good free radical scavenging ability is needed.

Keywords: *medicinal plants, antioxidant activity, phenols, flavanoids, Kaani tribes, aqueous and ethanolic extract*

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1. Introduction

Plants play an important role in the health of millions of people's life in many villages ranging from 75– 80% of the world population, mainly targeting primary health care in the developing countries because of better cultural acceptability, compatibility with human body and lesser side effects. Currently, there is a drastic increase in the usage of herbal medicine in the developed countries [1].

Traditional medicinal practices form an integral part of complementary or alternative medicine. Although their efficacy and mechanism of action have not been scientifically tested in most cases, these simple medicinal preparations often mediate beneficial responses due to their chemical constituents [2]. Herbal plants have many pharmacologically active compounds like flavonoids, alkaloids, tannin, steroids, glycosides, phenols which are stored in specific parts like leaves, bark, flowers, seeds, fruits, root etc [3].

Despite the great advances observed in modern medicine in recent decades, plants still make an important contribution to health care. Much interest, in medicinal plants however, emanates from their long use in folk medicines as well as their prophylactic properties, especially in developing countries. Large number of medicinal plants has been investigated for their antioxidant properties. Natural antioxidants either in the form of raw extracts or their chemical constituents are very effective in preventing the destructive processes caused by oxidative stress [4].

2. Materials and Methods

The medicinal plants were collected from Kanyakumari district of Tamilnadu. Kanyakumari district situated in the

southern most part of Western Ghats region of the Indian peninsula which is located between 77° 15' and 77° 36' of east of longitude and 8° 03' and 8° 35' north of Latitude.

The samples were mostly collected in the villages namely Mothiramalai, Thottamalai, Chappangupparai, Koovaikadu, Kothaiyar, Alamparai, Koruvakuzhi, Puravilaim Valayamthanki, Vannachipparai, of Thiruvattar and Thovallai of Kanyakumari District (Pechiparai hills, Perunchani hills, Arrukaani hills, Kothaiyaru, Keeriparai and Pathukaani hills), of Kanyakumari District in Tamil Nadu, India which are inhabited by *Kaani* tribals disbursed in the deep forest areas.

2.1. Sample Collection and Storage

The plant materials were fresh selected, healthy and free from contamination. The collected plants were thoroughly washed in water allowed to drain, packed in zip lock plastic covers and transported overnight from Kanyakumari District to Coimbatore District in Tamil Nadu, India for analysis. They were also checked at regular intervals for any physical, chemical or biological damage according to the guidelines given by WHO [5]. For the chemical analysis of Antioxidants the leaves were shade dried, powdered and stored in brown bottle containers at 4°C for further use.

Database on medicinal plants given by the National Medicinal Plants Board, Ministry of Health and Family Welfare, Government of India [6] was used for the identification of the plants collected from the study area. Digital record of the collected plants was maintained and the local name was obtained from the tribes to enable accurate identification. A botanist was also consulted to make sure that the name of the plant in the local language (Tamil) and its botanical name was correct. Table 1 gives the list of selected plants.

2.2. Procurement of Chemicals

2,2-diphenyl-1-picryl hydrazyl (DPPH), TPTZ (2,4,6-tripyridyltriazine), Folin-Ciocalteu reagent, Gallic acid, Aluminium chloride, Catechol, Ethanol, Deionised Water, Glacial acetic acid etc., were obtained from local agents. The standard for the analysis of Antioxidant (AO) parameters was obtained from Sigma-Aldrich Chemical Private Limited, Germany.

2.3. Preparation of Standards

The results of DPPH and Total phenolic assays were expressed in Gallic Acid Equivalence (GAE), Ferric Reducing Antioxidant Power (FRAP) in Ascorbic acid equivalence Capacity (AAEC) and Flavonoids in catechols.

DPPH: 5mg of gallic acid dissolved in 100ml distilled water (50µg/ml)

FRAP: Ascorbic Acid (M.W.176.13) 1000µM/ml

Total Phenols: 5mg of gallic acid dissolved in 100ml distilled water (50µg/ml)

Flavonoids: Stock standard solution was prepared by dissolving 100mg of catechol in 100ml of 80% ethanol. Working standard solution was prepared by making up 10ml of stock solution to 100ml by 80% ethanol.

2.4. Sample Extraction

The dried and powdered samples were extracted using 80% ethanol (v/v) and deionized water separately. Each sample of 1 g was weighed into conical flask that was wrapped with aluminium foil and 50 mL 80% ethanol solution or 50 mL of deionized water was added for extraction. Then it was well ground in a mortar and pestle followed by centrifugation (at 3200 rpm for 15 min.), and filtration with Whatman No.1 filter paper to obtain a clear solution. The supernatant solution was suitably stored in brown bottle containers at 4°C for the analyses. During the estimation, if the absorbance was higher than the standard, further dilution was made with respective solvents for the assay.

Table 1. List of selected Plants

S.No.	Name in Local Language(Tamil)	Family Name	Botanical Name
1	Vallarai	Apiaceae	*Centella Asiatica*
2	Murivuporunthi	Acanthaceae	*Blepharis Maderaspatens*
3	Thuththi	Malvaceae	*Abutilon Indicum*
4	Thumbai	Lamiaceae	*Leucas Aspera*
5	Krishna Thulsi	Lamiaceae	*Ocimum Sanctum*
6	Nochi	Verbenaceae	*Vitex Negundo*
7	Kuppameni	Euphorbiaceae	*Acalypha Indica*
8	Shatavari	Liliaceae	*Asparagus Racemosus Wild*
9	Vilvam	Rutaceae	*Aegle Marmelos*
10	Thazhuthamai	Verbanaceae	*Clerodendrum Phlomides*
11	Nayuruvi	Amaranthaceae	*Achyranthes Aspera*
12	Brahmi	Plantaginaceae	*Bacopa Monnieri*
13	Uzhinja or Valliuzhinja	Sapindaceae	*Cardiyospermum Halikakabam*
14	Adathoda	Acanthaceae	*Adhatoda Vasica Nees*
15	Sivanaarvembu	Fabaceae	*Indigofera Aspalathiodes*
16	Ponnavarai	Fabaceae	*Cassia Auriculata*
17	Sirukanpeelai	Amaranthaceae	*Aerva Lanata*
18	Kurunthotti	Malvaceae	*Sida Cordifolin*
19	Amirthavalli	Menispermaceae	*Tinospora Cordifolia*
20	Nilavembu	Acanthaceae	*Andrographis Paniculata*

2.5. Moisture

Moisture content of all the fresh leaves of the selected medicinal plants was estimated using Sartorius Infra-red Moisture Analyzer. Infra-red moisture analyzer (IMA) uses thermo gravimeter which extracts moisture from a sample by heating (thermo) it and reading the resulting loss of weight (gravimetry) using a balance or scale [7].

2.6. DPPH Assay

A simple method that has been developed to determine the AOA of foods utilizes the stable α,α-diphenyl-β-picryl hydrazyl, characterized as a stable free radical by virtue of the delocalization of the spare electron over the molecule as a whole, so that the molecules do not dimerise as would be the case with other molecule. The delocalization gives rise to change in colour from deep violet to yellow, characterized by the absorption band in solution centered at about 517nm. When a solution of DPPH is mixed with that of a substance that can donate a hydrogen atom, then this gives rise to the reduced form. The molar absorption of the DPPH radical at 517nm reduces from 9660 to 1640 when the odd electrons of DPPH radical becomes paired with hydrogen from a free radical scavenging antioxidant to form the reduced DPPH-H with the loss of violet colour [8].

2.7. FRAP

FRAP actually measures only the reducing capability based upon the ferric ion, which is not relevant to antioxidant activity mechanistically and physiologically. However, in contrast to other tests of total antioxidant power, the FRAP assay with slight modifications is simple, speedy, inexpensive, and robust and does not require specialized equipment [9]. An amount of 200 µL extracted samples were mixed with 3 mL FRAP reagent in test tubes and vortexed. Blank samples were prepared for both methanol and deionized water extracted samples. Both samples and blank were incubated in water bath for 30 minutes at 37°C and the absorbance of the samples was determined against blank at 593 nm. Series of stock solution at 200, 400, 800, 1200 and 1600 µM were prepared (r^2 = 0.9944) using aqueous solution of $FeSO_4$.$7H_2$ O as standard curve. The values obtained were expressed as µM of ferrous equivalent Fe (II) per gram of freeze dried sample.

2.8. Total Phenols

Total phenol contents were estimated using Folin Ciocalteu reagent method [10,11]. A dilute extract of each plant extract (0.5 ml of 1:10 g/mL) or gallic acid used as standard was mixed with Folin Ciocalteu reagent (5 mL, 1:10 diluted with distilled water) and aqueous Na_2CO_3 (4 mL, 1M). A blue colour was developed in each tube because the phenols undergo a complex redox reaction with Phosphomolibdic acid in Folin-ciocalteau reagent in alkaline medium. The mixture was allowed to stand for 10 min and the absorbance was measured by colorimetry at 765 nm. The standard curve was prepared using 0, 50, 100, 150, 200, 250 mg/L solutions of Gallic acid in methanol: water (50:50, v/v). Total phenol contents were expressed in terms of Gallic acid equivalent (mg/g of dry mass), which was used as the reference compound, [12].

2.9. Flavonoids

Flavonoids present in the extract formed, a charge transfer complex with several heavy metals to give a characteristic colour. In this reaction, the high electropositive nature of aluminium (Al^{3+}, Aluminium Chloride) attracts the atomic nuclei of the aromatic rings in the flavanoids through the µ-electrons and creates a charge-transfer resonance hybrid. This hybrid is highly stable in the aqueous medium, which then interacted with the sodium nitrite in an alkaline medium to form a pink coloured complex that is spectrophotometrically measured at 510nm [7]. 1mL of test sample and 4 ml of water were added to a volumetric flask (10 ml volume). Added 0.3mL of 5 % Sodium nitrite, and after 5 min added 0.3 mL of 10% Aluminium chloride. After 6 min incubation at room temperature, 2 mL of 1 M Sodium hydroxide was added to the reaction mixture. Immediately the final volume was made up to 10 mL with distilled water. The absorbance of the reaction mixture was measured at 510 nm against a blank spectrophotometrically (Shimadzu UV-1609, Japan). Results are expressed as catechine quivalents (mg catechin/g dried sample).

All the tests were conducted in triplicates. The data of all the parameters were statistically analyzed and expressed as mean ± S.D with the aid of SPSS 17.0 Windows version.

3. Results and Discussion

All human cultures have a history of herbal medicines, usually making use of the plants found closest to home. Even today in the times of advanced technology and medical science certain communities still depend on plants for their healing. Herbal medicines – the original human health care products – are still fully present and available to our lives. Common herbs and spices – including ginger, turmeric and garlic, cloves, and cinnamon as well as fenugreek seeds and leaves, artichoke leaf extract, and holy basil are commonly available and used in the food preparations. Thyme tincture can outperform conventional acne treatments. Even today, plant diversity is still indispensable for human well being and provides all or a significant number of the remedies required in health care.

The results in Table 2 indicate a wide range of DPPH values from 0.937 to 71.821 which is expressed in mg of GAE/g. The values of DPPH activity were higher in ethanolic extracts than water extracts. The AOA in our study was 5.6 times higher in ethanolic extracts; this is in accordance with the earlier findings that reported AOA was 2.1 times higher in ethanolic extracts [13].

Among all the samples, *Blepharis Maderaspatens* had the highest DPPH activity in both aqueous (8.870) and ethanolic extracts (71.82).

In our study, the leaves were found to have varying levels of antioxidant activity. Ethanolic extracts had highest DPPH activity in *Abutilon Indicum, Asparagus Racemosus Wild, Centella Asiatica*. Water extracts had

highest AOA in *Blepharis Maderaspatens*, *Centella Asiatica and Cardiyospermum Halikakabam*. Ethanolic extracts had least DPPH activity in *Abutilon Indicum*, *Aerva Lanata* and *Cardiyospermum Halikakabam* and water extracts in *Aerva Lanata*, *Bacopa Monnieri* and *Tinospora cordifolia*.

The results in Table 2 indicate a wide range of FRAP values from 32.03 to 633.76 µM of AAEC/g. FRAP power of the medicinal plants were higher in ethanolic extracts and medium in water extracts. It was evident that ethanolic extracts of the medicinal plants had high reducing power. Among all the plants, *Blepharis Maderaspatens* had the highest AOA in ethanol extracts and *Centella Asiatica* the highest AOA in water extracts. Water extracts of *Centella Asiatica*, *Asparagus Racemosus Wild* and *Leucas Aspera* had the highest AOA, while ethanol extracts of *Blepharis Maderaspatens*, *Ocimum Sanctum* and *Leucas Aspera* had the highest AOA. Water extracts of *Cardiyospermum Halikakabam*, *Aerva Lanata* and *Sida Cordifolin* had least AOA. And ethanolic extracts had least AOA in *Asparagus Racemosus Wild*, *Cardiyospermum Halikakabam*, *Achyranthes Aspera*.

The Mean DPPH activity of the selected medicinal plants was found to be 3.402±2.22 in aqueous extract and 19.114±17.01 in ethanolic extract. Generally, the antioxidant properties of extracts were found to be concentration dependent [14]. Based on the reported results, the ethanol extracts which are more polar solvent, were more effective in extracting antioxidants compared to water extract in DPPH assay. Hence, to assess the antioxidant capacity of sample, a variety of methods must be used in parallel, because different methods often give different results [15]. DPPH radical scavenging activities of the antioxidants are considered to be due to their hydrogen donating abilities. This method is a widely used method to evaluate antioxidant activities in relatively a short time compared to other methods. [16,17]

Higher plants have been used as a source of drugs by mankind for several thousand years. In fact, ancient man was totally dependent on green plants for his day-to-day needs of medicaments. With the development of modern medicine, synthetic drugs and antibiotics, the importance of plants as raw material for drugs decreased considerably. However, plants were used as a basis of some of the most important drugs, even in the modern system of medicine.

Plants can provide biologically active molecules and lead structures for the development of modified derivatives with enhanced activity and reduced toxicity. The small fraction of flowering plants that have so far been investigated have yielded about 120 therapeutic agents of known structure from about 90 species of plants. Some of the useful plant drugs include vinblastine, vincristine, taxol, podophyllotoxin, camptothecin, digitoxigenin, gitoxigenin, digoxigenin, tubocurarine, morphine, codeine, aspirin, atropine, pilocarpine, capscicine, allicin, curcumin, artemesinin and ephedrine among others. In some cases, the crude extract of medicinal plants may be used as medicaments. Even at the dawn of 21st century, 11% of the 252 drugs considered as basic and essential by the WHO were exclusively of flowering plant origin, [18].

Table 2. DPPH and FRAP values (on Dry weight basis) in the Ethanolic and Aqueous Extract of selected Medicinal plants

Plant Name	Moisture Content (%)	DPPH (mg of GAE/g) Aqueous)	DPPH (mg of GAE/g) (Ethanol)	FRAP µM of AAEC/g (Aqueous)	FRAP µM of AAEC/g (Ethanol)
Centella Asiatica	88.61	8.770	37.894	300.70	125.43
Blepharis Maderaspatens	89.86	8.870	71.821	203.16	633.76
Abutilon Indicum	50.92	2.016	5.083	55.82	96.01
Leucas Aspera	74.62	3.141	14.220	236.12	224.37
Ocimum Sanctum	87.88	4.033	28.363	134.96	282.47
Vitex Negundo	68.10	3.510	7.109	145.07	188.05
Acalypha Indica	78.25	5.130	14.788	102.21	146.74
Asparagus Racemosus Wild	69.67	2.640	48.635	271.36	32.03
Aegle Marmelos	75.45	3.756	8.780	34.75	153.17
Clerodendrum Phlomides	67.47	3.690	31.766	152.94	105.41
Achyranthes Aspera	63.04	1.621	12.648	92.59	57.72
Bacopa Monnieri	61.06	1.028	17.319	82.05	104.65
Cardiyospermum Halikakabam	72.67	5.494	6.153	41.83	51.31
Adhatoda Vasica Nees	65.23	2.240	17.666	95.16	106.63
Indigofera Aspalathiodes	69.11	2.922	12.557	111.23	84.80
Cassia Auriculata	75.31	2.834	14.089	69.35	129.75
Aerva Lanata	36.00	0.937	5.250	40.17	66.26
Sida Cordifolin	61.00	1.740	10.153	43.92	83.64
Tinospora Cordifolia	63.00	1.351	8.756	46.29	96.78
Andrographis Paniculata	65.61	2.325	9.244	99.59	139.94
Mean ± SD	**69.14±12.63**	**3.402± 2.22**	**19.114± 17.01**	**117.96±79.17**	**145.44 ±129.53**

The Mean FRAP value of the selected medicinal plants was found to be 145.44±129.53 in ethanolic extract and 117.96±79.17 in aqueous extracts. In an earlier study the FRAP ranged from 0.06 to 25 mM/L. There was significant linear correlation between total phenolic content and FRAP. According to their antioxidant capacity, 70 medicinal plant extracts could be divided in five groups: (a) very low FRAP (<1 mM/L) n = 9; (b) low FRAP (1–5 mM/L), n = 37; (c) good FRAP (5–10 mM/L), n = 15; (d) high FRAP (10–20 mM/L), n = 8; and (e) very high FRAP (>20 mM/L), n = 1 medicinal plant extract [19].

FRAP assay has been used to determine antioxidant activity as it is simple and quick. Besides that, the reaction is reproducible and linearly related to molar concentration of the antioxidants [20]. However, some disadvantage was found in this method as FRAP assay does not react fast with some antioxidants such as gluthathione [21].

Plant phenolics are generally involved in defences against ultraviolet radiation or aggression by pathogens, parasites, and predators, as well as contributing to plants colour. For eg., phenolics are the most important compounds affecting flavour and colour difference among white, pink and red wines; they react with oxygen and are critical to the preservation, maturation and aging of the wine, [22]. Because of the heterogeneity of natural phenolics and the possible interference from other readily oxidized substances in the plant materials, it is not surprising that several methods have been used for determination of total phenolics and none are perfect. Among such methods are the Folin-Denis method (FD), Folin-Ciocalteu method (F-C), permanganate titration, colorimetry with iron salts, and ultraviolet absorbance. In most cases, F-C has been found preferable as compared to the other methods, [10].

It is known that different phenolic compounds have different responses in the Folin-Ciocalteu method. Similarly the molecular antioxidant response of phenolic compounds varies remarkably, depending on their chemical structure. In addition, there may be some interference rising from other chemical components present in the extract, such as sugars or ascorbic acid [23].

Despite their wide distribution, the health effects of dietary polyphenols have come to the attention of nutritionists only in recent years. Researchers and food manufacturers have become more interested in polyphenols due to their potent antioxidant properties, their abundance in the diet, and their credible effects in the prevention of various oxidative stress associated diseases.

In the present study, TPC and flavonoids of the selected samples was estimated by using Folin-Ciocalteau method and aluminium chloride method respectively and presented in Table 3.

Blepharis Maderaspatens had the highest phenolic content in both aqueous and ethanolic extracts. The other plants were found to have varying levels of phenols. The quantity of phenolics ranging from 1.6 to 35.6 expressed as mg of GAE/mg. Ethanolic extracts recorded the highest TPC in *Abutilon Indicum*, *Asparagus Racemosus Wild*, *Centella Asiatica* and *Ocimum Sanctum*. Water extracts of *Blepharis Maderaspatens*, *Centella Asiatica* and *Ocimum Sanctum* had highest TPC.

Table 3. TPC and Flavonoid content (on dry weight basis) in the Ethanolic and Aqueous Extract of selected Medicinal plants

Plant Name	Phenols mg of GAE/g (Aqueous)	Phenols mg of GAE/g (Ethanol)	Flavonoids mg of catechol / g (Aqueous)	Flavonoids mg of catechol /g (Ethanol)
Centella Asiatica	10.2	17.1	28.2	29.6
Blepharis Maderaspatens	12.8	35.6	32.5	35.4
Abutilon Indicum	3.4	2.0	11.3	11.9
Leucas Aspera	3.4	7.4	12.6	13.1
Ocimum Sanctum	9.4	10.16	15.2	15.6
Vitex Negundo	6.1	6.9	9.8	9.4
Acalypha Indica	5.5	5.8	7.5	7.7
Asparagus Racemosus Wild	1.6	16.63	11.3	11.0
Aegle Marmelos	5.9	5.0	15.3	14.6
Clerodendrum Phlomides	3.4	8.3	13.6	12.2
Achyranthes Aspera	4.1	2.3	7.9	8.3
Bacopa Monnieri	3.9	11.2	12.1	13.8
Cardiyospermum Halikakabam	4.0	3.2	6.3	14.6
Adhatoda Vasica Nees	3.5	8.3	2.5	5.9
Indigofera Aspalathiodes	4.2	3.7	8.2	5.7
Cassia Auriculata	5.1	5.5	9.4	10.9
Aerva Lanata	1.7	1.6	14.4	15.3
Sida Cordifolin	2.0	2.1	8.5	9.2
Tinospora cordifolia	3.3	3.8	4.9	4.7
Andrographis paniculata	4.4	3.5	5.6	6.3
Mean ± SD	**4.8 ± 2.8**	**8.0 ± 7.8**	**11.8 ± 7.2**	**12.7 ± 7.6**

Ethanolic extracts recorded lowest phenolic content in *Abutilon Indicum*, *Aerva Lanata* and *Sida Cordifolin*. Water extracts had the least phenolic content in *Asparagus Racemosus Wild*, *Aerva Lanata*, *Sida Cordifolin*. In our study, we found that the TPC was 1.7 times higher in ethanolic extracts. The mean phenolic content of the selected medicinal plants were found to be 4.8±2.8 in aqueous extracts and 8.0±7.8 in ethanolic extracts.

Many polyphenols contribute significantly to the antioxidant activity and act as highly effective free radical scavengers which is mainly due to their redox properties, which can play an important role in adsorbing and neutralizing free Radicals, quenching singlet and triplet oxygen or decomposing peroxides [24].

Blepharis Maderaspatens recorded the highest flavonoid content in both ethanolic and water extracts and the other samples were found to have varying levels of flavonoid contents ranging from 2.5 to 35.4 mg of catechol/g. Aqueous and ethanolic extracts had the highest TFC in *Blepharis Maderaspatens* and *Centella Asiatica* followed by *Ocimum Sanctum* in ethanol extract and *Aegle Marmelos* in water extract.

Most of the samples had high Flavonoid content in the ethanol than in the water extract. Ethanolic extracts recorded lowest flavonoid content in *Tinospora cordifolia*, *Indigofera Aspalathiodes* and *Adhatoda Vasica Nees*. Water extracts had least flavonoid content in *Adhatoda Vasica Nees*, *Tinospora cordifolia* and *Andrographis paniculata*. The mean flavonoid content of the selected medicinal plants was found to be 11.8±7.2 in aqueous extracts and 12.7±7.6 in ethanolic extracts.

The concentration of flavonoids in various plant extracts of the species *M. peregrinum* was determined using spectrophotometric method with aluminum chloride. The content of flavonoids was expressed in terms of rutin equivalent (the standard curve equation: $y = 17.231x - 0.0591$, $r^2 = 0.999$), mg of RU/g of extract. The concentration of flavonoids in plant extracts from *M. peregrinum* ranged from 18.72 to 54.77 mg/g. Methanolic, acetone and ethyl acetate extracts contained the highest flavonoid concentration. The concentration of flavonoids in methanol extract was 54.77 mg RU/g, which was very similar to the value of acetone extract concentration. The lowest flavonoid concentration was measured in petroleum ether and water extract. The concentration of flavonoids in plant extracts depends on the polarity of solvents used in the extract preparation [25]. The extract of *C. longa*, which contained highest amount of flavonoid and phenolic compounds, exhibited the greatest antioxidant activity and among non-conventional species *C. Caecia* species contained the highest phenol and Flavonoid which can be explored for new drug preparation [26].

The curative properties of medicinal plants are perhaps due to the presence of various secondary metabolites such as alkaloids, flavonoids, glycosides, phenols, saponins, sterols etc. The leaves of the plants investigated were rich in alkaloids, flavonoids, reducing sugars, phenols and also showed presence of amino acids. They were known to show medicinal potential and physiological activities [27].

In our study on the AOA of 20 medicinal plants we opine that *Blepharis maderaspatensis* has the highest AO content. It is also reported that *Blepharis maderaspatensis L. Roth (Acanthaceae*) often used in India by hyperlipidaemic subjects as an alternative therapeutic tool to treat hyperlipidaemia is being used in folk medicine as a diuretic and wound healing agent. It is also used to treat the disorders such as boils, bone fracture, diarrhoea and lactation [28].

4. Conclusion

There was a wide difference in antioxidant content among the selected medicinal plants. Some of the plants had highest TPC, TFC, and AOA. The ethanolic extracts revealed higher concentrations of AOA, TPC, and TFC in the medicinal plants than in the water extract. Based on these results, it may be concluded that the selected home remedy plant materials are good sources of antioxidants. Further studies on medicinal plant can be carried out to investigate the potential health benefits of phytochemicals in disease prevention. Randomized clinical trials are needed to establish the value of herbal drugs developed from medicinal plants.

The effectiveness of herbal medicine is not always corroborated by scientific evidences but it is continually used by almost 80 percent of people. In some parts of the world specifically the United States herbal remedies are classified as dietary supplement because it can neither be classified as food nor drugs. This is because laws pertaining to dietary supplements are relatively lax comparing to drugs or foods. Hence much research is required to strengthen this field of alternate medicine.

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Statement of Competing Interests

There is no competing interest between the authors.

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