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Comparative Analysis of Antioxidant Activity in Various Plant Species

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ABSTRACT

Poly-phenolic compounds are secondary metabolites which protect the plants against the pathogenic attack and harmful effects of UV radiations coming from the sun. There has been a lot of interest generated in the last few years about the health benefit accruing out of the dietary polyphenols. Most plants contain poly-phenolic or other antioxidant compounds at different concentrations. These compounds occur in all parts of the plant such as wood, bark, stems, leaves, fruit, roots, flowers, pollen or even seeds at different concentrations. Long term use of diets rich in plant polyphenols may offer protection against cancers, neurodegenerative disorders and other diseases of cardiovascular system. In the current study nine plants (Neem, Tulsi, Papaya, Mint, Wheat, Lemon Grass, Curry, Mustard and Bajra) were used. The methanolic extracts from various edible and non-edible parts of the plant (viz. roots, stem, leaf, fruit, flower, bark etc.) were used for comparative analysis of the occurrence of poly-phenolic compounds. The content of total phenolics in the extracts was determined spectrometrically according to the Folin−Ciocalteu procedure and calculated as tannic acid equivalents. Remarkably high polyphenolic content was found in the leaves of Neem (*Azadirachta*spp.), Curry (*Murrayakoengii)* and Tulsi (*Ocimum sanctum*). Significantly high activity was also seen in cotton flowers, wheat leaves, *Ocimum*roots and Papaya leaves. This systematic investigation of a number of plants proved important for the search of natural sources for potent antioxidants and thus their medicinal importance.

Keywords: Polyphenols, FolinCiocalteu, antioxidant, Total Phenolic content (TPC), Tannic acid equivalents (TAE)

INTRODUCTION

Plants have been widely used by mankind since antiquity as food, fodder, medicines and as a source of industrial raw materials. Phyto-metabolites have attracted interest of R&D over many years (1). These compounds provide environmental adaptation to the plants and used as pharmaceuticals by the mankind (2). Antioxidants provide protection against the oxidative stress in the cells and are therefore, useful in the treatment of many human diseases, such as cancer, cardiovascular diseases and inflammatory disorders (3).

Poly-phenolic secondary metabolites present in different parts of the plants provide protection against the pathogens and also protect the plants from UV radiations. In the last few years, lot of interest in the health benefits arising out of the dietary plant poly-phenolic compounds acting as antioxidants has arisen. Natural antioxidants in the food products can come from endogenous components present in the food or as a result of food processing or they can also be added to the food products externally as additives. Most plants contain compounds with antioxidant activity which may be present in all parts of the plant such as root, stem, leaf, bark and other parts. Epidemiological studies and associated meta-analyses strongly suggest that consumption of diets rich in plant polyphenols for long periods offer protection against development of cancers, cardiovascular diseases, and diabetes, osteoporosis and neurodegenerative diseases (4)(5). The natural compounds are preferred over the synthetic drugs and therefore, the antioxidants of plant origin are preferred by the health professionals (6)(7). Krishnaiah et al., (3) in their review of the antioxidant potential of medicinal plants have reported the potential side-effect free use of such compounds in the food processing industry and in preventive medicines. Farrukh et al. (8) have reported the antioxidant and free radical scavenging activity of twelve traditionally used medicinal plants from their methanolic crude extracts. The antioxidant capacity and phenolic content of anti-inflammatory and medicinal plants in Colombia has also been reviewed (9). A significant correlation was established between the ORAC and the TBARS tests, confirming that the antioxidant capacity is partly due to transfer mechanisms involving hydrogen atoms. These findings suggest the potential of these plants as valuable source of natural bioactive molecules. Consumption of food items rich in antioxidants is recommended by many studies so as to augment an array of free radical scavengers inside the body. The concern about the safety of synthetic antioxidants has resulted in to their replacement by natural compounds. The traditional medicinal systems describe plants as a rich source of dietary antioxidants. Thus, the present study attempts to investigate and search novel plant materials as a rich source of antioxidants. The study comprehensively investigates the total phenolic content from extracts obtained from different parts of 9 plant materials.

METHODOLOGY

A) Plant Samples

Different parts, namely- leaves, roots, stems, barks, flowers, buds, petioles of 9 different plant *Triticumaestivum, Menthaspp, Azadiractaindica, Ocimumspp, CymbopogonSpp, Murrayakoengii, Sinapis alba, Carica papaya, Pennisetumglaucum*were collected from Botanical Garden of Sri Venkateswara College and others parts of National Capital Region (NCR) of India.

B) Processing of Plant extracts

All the collected parts of plants' were rinsed thrice with distilled water and excess water was dripped off and subsequently removed with blotting paper. Then the plants samples were weighed and their methanolic extracts prepared.

C) Preparation of Plant extracts

All the plants' sample were extracted in 1ml of 0.3 N Methanolic-HCl and centrifuged for 10 min at 300 rpm at room temperature. The process was repeated with the residue and supernatant was kept in water bath at 60^0 to 100^0 C till a dry residue was obtained.

D) Assays for Total Phenol Content (TPC)

The total phenolic compounds in the extract were determined by Folin&Ciocalteu's method (10) with slight modification. 1 ml of dried residue (supernatant) of plant extract was dissolved in 5 ml distilled water. To 1 ml of this extract, additional 6 ml of distilled water was added. This was mixed with 0.5 ml of 50%Folin&Ciocalteu's phenol reagents (FCPR) (v/v) and 1 ml of 35% Na_2CO_3(w/v). The mixture was incubated for one hour at room temperature and absorbance was read using spectrophotometer at 630 nm. Tannic acid was taken as standard and a calibration curve was prepared using various aliquots. TPC was calculated by the following formula/;

$$P= c*V/e$$

Where, P-total content of phenolic compounds expressed in mg/g plant extract, in Tannic Acid Equivalents (TAE); c- the concentration of tannic acid ascertained from the calibration curve, mg/ml; V- the volume of the extract , ml; e- the weight of pure plant extract, g.

E) Statistical Analysis

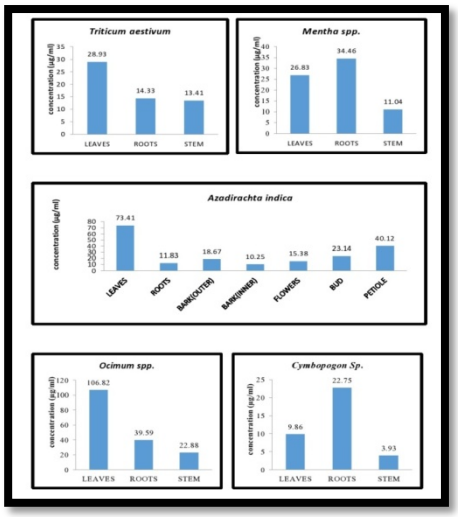
Analyses were run in triplicates and the results were expressed as mean values with standard error. The total content of phenolic compounds in µg/ml was determined from the regression equation of the Calibration curve ($y=0.0076x-0.0139$; $R^2=0.9965$) calculated using MS Excel software.

RESULTS

Plants produce a large number of primary and secondary metabolites of which Phenolic compounds are the most important group of secondary metabolites. The TPC (mg/g), determined from regression equation of calibration curve ($y=0.0076x-0.0139$; $R^2=0.9965$) and expressed in TAE (Tannic acid equivalents), varied between 0.904 and 36.47 (Table I). Highest TP Content was noted in the leaves of *Murrayakoengii* whereas the lowest content was found in the stems of *Sinapis alba* and *Cymbopogon*sp. Figure I and Figure II show the comparative analysis of the poly-phenolic contents in the different parts of the plants viz roots, stems, leaves, flowers, bark , etc.Remarkably high polyphenolic content was found in the leaves of Neem (*Azadirachta*spp.), Curry (*Murrayakoengii)* and Tulsi (*Ocimum sanctum*). Significantly high activity was also seen in cotton flowers, wheat leaves, *Ocimum*roots and Papaya leaves. These results throw light to the fact that plants can be excellent sources of polyphenols. More exhaustive studies in this area could provide an impetus to the use of plant based materials as a source of antioxidants.

Table I- Total Phenolic Content from Plant Samples

S.NO	PLANT SAMPLE	PLANT PART	TPC (mg/g TAE)
1	*Azadirachta idica* (NEEM)	LEAVES ROOTS BARK(OUTER) BARK (INNER) FLOWERS BUD PETIOLE	22.022 3.549 5.601 3.075 4.614 6.943 12.036
2	*Cymbopogon sp.* (LEMON GRASS)	LEAVES ROOTS STEM	2.957 6.825 1.180
3	*Carica papaya* (PAPAYA)	LEAVES ROOTS STEM	10.457 6.430 3.628
4	*Ocimum sp* (TULSI)	LEAVES ROOTS STEM	32.045 23.755 13.729
5	*Mentha longifolia* (MINT)	LEAVES ROOTS STEM	8.049 10.338 3.312
6	*Triticum aestivum* (WHEAT)	LEAVES ROOTS STEM	8.680 4.299 4.180
7	*Pennisetum glaucum* (BAJRA)	LEAVES ROOTS STEM	5.838 4.220 4.180
8	*Murraya koengii* (CURRY)	LEAVES ROOTS STEM	36.470 3.549 3.746
9	*Sinapis alba* (MUSTARD)	LEAVES ROOTS STEM FLOWER	4.851 1.338 0.904 2.522



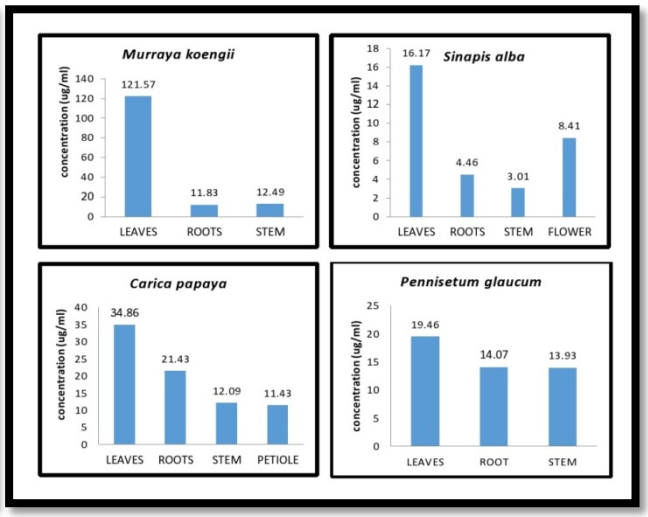


Figure I Figure II

Figure I- Comparative Analysis of the TPC content in Wheat, Mint, Neem, Holy Basil and Lemon Grass

Figure II – Comparative Analysis of the TPC content in different parts of Curry, Papaya, Mustard and Bajra

DISCUSSION

Oxidative stress adversely affects the human health and under the stress conditions our bodies produce more reactive oxygen. The use of dietary antioxidants and their positive health effects are well known. More than 80 % of the inhabitants on the earth use traditional medicines for their primary health care needs which comes from plant based sources (11). The Enzymatic antioxidants (e.g., superoxide dismutase, glutathione peroxidase, and catalase) and non-enzymatic antioxidants (e.g., ascorbic acid ,tocopherol,

glutathione, carotenoids, and flavonoids) cannot cope with the excessive ROS and hence dietary supplements are needed to combat the imbalance (12)(13)(14)(15)(16). Antioxidants have the capacity to quench the reactive free radicals, in the absence of which a large number of diseases may manifest. One possible way of increasing the capability of a person to fight diseases is to supplement the diet with antioxidant compounds that are obtained from plant sources (Knekt et al., 1996). The antioxidant capacity, total phenolic content (TPC) and total flavonoid content (TFC) of different extracts prepared from various parts of *Moringaoleifera* Lam. was studied (17). It was found that *M. oleifera* exhibits strong antioxidant activity and could serve as prospective source of natural antioxidants to food and health industries. These natural plant antioxidants can therefore serve as a type of preventive medicine. Numerous studies are going on globally to identify pharmacologically potent antioxidants.

CONCLUSION

Diversity of active and therapeutically useful compounds from plants are provided by the traditional Indian medical systems. 9 such plants were studied and their Total Phenolic Content was evaluated in this study. The overall results of the present study certainly provides positive baseline information to ascertain the potency of the crude extracts obtained from various parts of *Triticumaestivum, Mentha*spp, *Azadirachtaindica, Cymbopogon*spp., *Murrayakoengii, Brassica* sp., *Carica papaya, Gossypiumherbaceum, Pennisetumglaucum.* However, further investigations are suggested to identify and isolate the efficacy of the individual components present in the crude extracts. Such studies would be useful in establishing their applications in pharmaceutical and food industries.

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