



Assessment of phenol and flavonoid content in the plant materials

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ABSTRACT

Entire phenolic and flavonoid content are the basis for evolution of the preventive role of plant material as compensating factor to oxidative damage caused by free radicals which is a critical etiological aspect responsible for several chronic human ailments, the maximum content of phenol was found in *Capsicum annuum* and the richest source of flavonoid in *Lactuca sativa*. However the highest flavonoid/ phenolic ratio have been estimated in *Lactuca sativa*. As a role it is proved by result that leaves are rich source of phenolics and flavonoid content.

Key Words: Phenolic, Flavonoid, *Capsicum annuum*, *Lactuca sativa*

INTRODUCTION

Indians have been using plants as a source of medicine due to their abundant chemical properties since time immemorial. Crude extracts of herbs and other plant material rich in phenol content and flavonoids are of boosting interest in food industry and medical field. Phenolic compounds have an aromatic ring that contains various attached substituent group such as hydroxyl (-OH), carboxyl (-COOH) and methoxy (-OCH₃) group and often other non-aromatic ring structures.

Phenolics hinder oxidative degradation of lipids and thereby enhance the excellence and nutritional value of food. Phenolics possess a wide spectrum of biochemical activities such as antioxidant, antimutagenic, anticarcinogenic as well as ability of modifying gene expression (Nakamura *et al.*, 2003). More than 400 flavonoids have been identified in different higher and lower plant species (Tapiero *et al.*, 2002).

Flavonoids are a group of polyphenolic compounds with known properties which include free radical scavenging, inhibition of hydrolytic and oxidative enzymes and anti-inflammatory action (Frankel, 1995). Due to its antioxidant properties it

can interfere with the oxidative process by reacting with free radicals chelating, catalytic metals and also by acting as oxygen scavengers (Dreosti, 2002; Harborne and Turner, 1984).

Phenolics and flavonoids of fruits and vegetables are source of natural antioxidants which is important to combat various human diseases including atherosclerosis, arthritis, ischemia and reperfusion injury of many tissues, central nervous system injury, gastritis, cancer and AIDS (Kumpulainen and Salonen, 1999).

The endeavour of present study is focused on determination of total phenolics and flavonoids content in various plant parts on state of Uttar Pradesh in India

METHODS AND MATERIALS

All plant parts of onion (*Allium cepa*), sugar apple (*Annona squamosa*), bastard teak (*Butea monosperma*), green pepper (*Capsicum annuum*), amaltas (*Cassia fistula*), Madagascar periwinkle (*Catharanthus roseus*), orange (*Citrus spp.*), carrot (*Daucus carota*), fig (*Ficus carica*), pipal (*Ficus religiosa*), ladyfinger (*Hibiscus esculentus*), salad (*Lactuca sativa*), mango (*Mangifera indica*), apple (*Malus pumila*), oleander (*Nerium oleander*), pinkhead smartweed (*Polygonum capitatum*), radish

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(*Raphanus sativus*), tomato (*Solanum lycopersicum*), east Indian rosebay (*Tabernaemontana divaricata*) were collected from the vicinity of Lucknow in state of Uttar Pradesh in India. The leaves were dried at room temperatures and further crushed in a pestle mortar. 5 gm of dried leaf powder was extracted in 50 ml of methanol by maceration (24 h). The solvent was concentrated at 35°C temperature and the resulting extracts were used for determination of flavonoids and phenolics content.

In case of fruits (sugar apple, fig, mango, ladyfinger, apple, tomato) and root samples (carrot and radish) had been collected from different regions in similar stage of ripeness, fruits and roots were cut into small pieces and frozen in liquid nitrogen, the samples were freeze-dried after one week, lyophilization was applied for maintaining in uniform storage condition.

Total Phenolic Assay

The total phenolic content of fruits and vegetables were determined by using the Folin-Ciocalteu assay (Singleton, *et al.*, 1965) An aliquot (1 ml) of extracts or standard solution of gallic acid (20, 40, 60, 80 and 100 mg/l) was added to 25 ml volumetric flask, containing 9 ml of distilled deionised water (dd H₂O). A reagent blank using dd H₂O was prepared. One millilitre of Folin-Ciocalteu's phenol reagent was added to the mixture and shaken. After 5 min, 10 ml of 7 % Na₂CO₃ solution was added to the mixture. The solution was diluted to volume (25 ml) with dd H₂O and mixed. After incubation for 90 min at room temperature, the absorbance against prepared reagent blank was determined at 750 nm with an UV-Vis Spectrophotometer Lambda 5. Total phenolic content of plant was expressed as mg Gallic acid equivalents (GAE)/100 g fresh weight. All samples were analyzed in triplicates

Total flavonoid assay

Total flavonoid content was measured by the aluminum chloride colorimetric assay (Zhishen *et al.*, 1999). An aliquot (1ml) of extracts or standard solution of catechin (20, 40, 60, 80 and 100 mg/l) was added to 10 ml volumetric flask containing 4 ml of dd H₂O. To the flask was added 0.3ml 5%NaNO₂. After 5 min, 0.3 ml 10% AlCl₃ was added. At 6th min, 2 ml 1 M NaOH was added and the total volume was

made up to 10 ml with dd H₂O. The solution was mixed well and the absorbance was measured against prepared reagent blank at 510 nm. Total flavonoid content of fruits and vegetables was expressed as mg catechin equivalents (CE)/100 g fresh mass. All samples were analyzed in triplicates.

RESULT AND DISCUSSION

The result for total phenolic and flavonoid content and their ratio in the studied plant material are demonstrated in table 1.

All plant parts (leaf, stem, and root) have been selected due to its medicinal value. In the case of leaf material data clearly indicates that the richest phenolic content was found in *Butea monosperma* (210.59 mg GAE/100g) which is followed by *Catharanthus roseus* (163.02 mg GAE/100g).Comparatively highest total flavonoid content was found in *Lactuca sativa* (76.80 mg CE/100g) which is followed by root of *Raphanus sativus* (43.5 mg/ CE/100 g). Both of these are eaten as vegetables. *Nerium oleander* show particularly low content of entire phenolic and entire flavonoids (30.10 mg GAE/100g and 1.04 mg CE/100 g respectively.

In case of fruit, *Annona squamosa* exhibits highest phenolic (183.01 GAE mg /100 g) but the green pepper which is used as spice has highest phenolic content (245.1 mg GAE /100g).

Table1. Phenolics and flavonoids acid content in the plant materials

Plants species	Total Phenolics mg GAE/100 g fresh mass	Total Flavonoids mg CE/100 g fresh mass	Flavonoids / Phenolics	Plants parts
<i>Allium cepa</i> (Onion)	152.90	18.50	0.12	Stem
<i>Annona squamosa</i> (Suger apple)	183.01	19.50	0.11	Fruit
<i>Butea monosperma</i> (Bastard teak)	210.59	30.02	0.14	Leaf
<i>Capsicum annum</i> (Green pepper)	245.10	27.50	0.11	Fruit
<i>Capsicum annum</i> (Red pepper)	172.90	12.80	0.07	Fruit
<i>Cassia fistula</i> (Amaltas)	159.23	23.90	0.15	Leaf
<i>Catharanthus roseus</i> (Madagascar periwinkle)	163.02	32.60	0.19	Leaf
<i>Citrus spp.</i> (Orange)	151.10	12.10	0.08	Fruit
<i>Daucus carota</i> (Carrot)	98.00	26.9	0.27	Root
<i>Ficus carica</i> (Fig)	59.80	28.20	0.47	Fruit
<i>Ficus religiosa</i> (Pipal)	60.05	20.01	0.33	Leaf
<i>Hibiscus esculentus</i> (Ladyfinger)	159.7	48.20	0.31	Fruit
<i>Lactuca sativa</i> (Salad)	118.20	76.80	0.65	Leaf
<i>Mangifera indica</i> (Mango)	138.0	19.40	0.14	Fruit
<i>Malus pumila</i> (Apple)	126.20	48.50	0.38	Fruit
<i>Nerium oleander</i> (Oleander)	30.10	1.04	0.034	Leaf
<i>Polygonum capitatum</i> (Pinkhead smartweed)	82.01	21.6	0.26	Leaf
<i>Raphanus sativus</i> (Radish)	160.00	43.5	0.27	Root
<i>Solanum lycopersicum</i> (Tomato)	76.90	12.8	0.17	Fruit
<i>Tabernaemontana divaricata</i> (East Indian rosebay)	129.00	28.00	0.22	Leaf

Results are presented as mean value of triplicates.

The presented data also reveals that leaves of *Nerium oleander* and *Lactuca sativa* have lowest as well as highest content of phenolics and flavonoids respectively. The variation of phenolics in fruits depends on many factors, it is known that the different stages in the process of fruits development e.g., in red pepper, it increases during the ripening stage due to maximum deposition of anthocyanin and flavonoids. The given result shows particularly only the phenolic and flavonoid contents but also their distribution in different parts of the studied fruits and vegetables, the maximum flavonoid / phenolic ratio

Was found in *Lactuca sativa* which is a leaf vegetable (0.65).

The given data for total phenolic and total flavonoid content is a pedestal for assessment of the preventive role of not only edible fruit and vegetables but also those individuals which are not eaten generally e.g. *Butea monosperma* and *Cassia fistula*, but these individuals have medicinal value which is a cornerstone for further research. They are indispensable tools towards in depth studies on the specimen of strong biological activity of phenolic compounds.

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