Studies on Phenolic Compounds and Anti-Oxidation Property Present in Medicinal Plants of Genus *Ficus*

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ABSTRACT

In recent years, the use of natural antioxidants present in traditional medicinal plants has become of special interest in the scientific world due to their presumed safety and nutritional and therapeutic value. In this present study, investigate phenolic compounds of genus Ficus medicinal plant. The medical plants *Ficus Benghalnesis*, *Ficus Racemosa*, and *Ficus Carica* were analysed for their total phenolics content by using Folin-Ciocalteu assay. The total phenolic contains in species *Ficus Benghalnesis* is 3.18±1.499 mg/ml and Antioxidant activity is 0.84±0.395 µg/ml. while *Ficus Carica* total phenolic contain is 2.46±1.018 mg/ml and antioxidant activity is 0.27±0.127 µg/ml and in *Ficus Racemosa* total phenolic contain is 3.01±1.461 mg/ml and antioxidant activity is 0.03±0.042 µg/ml. The Bark parts of the *Ficus Benghalnesis* showed higher antioxidant activity that catching in scavenging DPPH free radicals, which indicates that the Extract has good potential as a source for natural antioxidants to prevent Free radical mediated oxidative damage.

KEYWORDS: Antioxidants, Folin-Ciocalteu assay, DPPH free radicals, *Ficus Benghalnesis*, *Ficus Racemosa*, *Ficus Carica*
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Introduction

Presently demands of plant derived antioxidants, especially, the phenolic have gained importance due to their therapeutic importance including antioxidant potential. Epidemiological studies have shown that consumption of plant foods containing antioxidants is beneficial to health because it down-regulates many degenerative processes and can effectively lower the incidence of cancer and cardio-vascular diseases (Arabshahi & Urooj, 2007). High recovery of antioxidant compounds from plant materials can be achieved through different extraction techniques, after studying their chemistry and distribution in the plant matrix. For example, phenolic compounds are present in higher concentrations in the outer tissues (epidermal and sub-epidermal layers) of fruits and grains than in the inner tissues (mesocarp and pulp) (Antolovich et al, 2000). Most frequently used technique for isolation of plant antioxidant compounds is solvent extraction technique. Yield of plant extract and antioxidant activities are strongly dependent on the nature of solvent used for extraction, due to the presence of different antioxidant compounds with varied chemical properties and polarities. These compounds may or may not be soluble in a particular solvent. Frequently polar solvents are used for the recovery of polyphenols from a plant materials. The most suitable solvents are aqueous mixtures containing ethanol, methanol, acetone, and ethyl acetate (Pesche et al, 2006). Organic polar solvents methanol and ethanol have been widely used to extract antioxidant compounds from different plants and plant-based foods (Abdille et al, 2005) (fruits, vegetables etc.) such as plum, strawberry, pomegranate, broccoli, rosemary, sage, sumac, rice bran, wheat bran and bran, mango seed kernel, citrus peel, and many other fruit peels (Pesche et al, 2006). Efficacy of ethyl acetate to extract phenolic compounds from onion and citrus peel have been also reported (Zia-ur-Rehman, 2006). It was reported, mixtures of ethanol and acetone used to extract that maximum phenolic compounds from barley flour (Bonoli et al, 2004). Similarly, aqueous methanol was used to extracting large amounts of phenolic compounds from rice bran (Chatha et al, 2006) and from Moringaoleifera leaves (Siddhuraju & Becker, 2003). Extraction of antioxidant compounds from various plant materials including rice bran, wheat bran, oat groats and hull, coffee beans, citrus peel and guava leaves using aqueous 80% methanol (methanol: water, 80:20 v/v) (Anwar et al, 2006).
The medicinal plants selected for the present investigation, which includes *Ficus Carica*, *Ficus Racemosa*, *Ficus Benghalensis* have long been used in the folk medicine due to their potential health promoting and pharmacological attributes, which are mainly ascribed to the presence of antioxidant constituents such as phenolic acids and flavonoids. It is important to establish appropriate means to evaluate and quantify effective antioxidant principles of medicinally or economically viable plant materials. The present study therefore was conducted with the main objective of investigating the most potent antioxidant compounds, especially phenolic compounds from Bark of *Ficus Carica*, *Ficus Racemosa*, *Ficus Benghalensis* medicinal plants.

**Material and method**

**Plant materials:**

The targeted plant species for the present experiment is *Ficus Benghalensis* (banyan tree), *Ficus Racemosa* (Audumbar tree), and *Ficus Carica* (figs tree). Plants were collected from different areas of Maulana Azad college campus, Aurangabad. The extract was then prepared by macerating the plant material in three solvent methanol, ethanol and Aceton in ratio (1:10) with plant material and mixture kept at room temperature for 14 days. Then, the extract was filtered; the filtrate was concentrated by air dry. Filtrate stored at -20 ºc for further experiment.

**Total phenolic assay:**

Total phenolic contents were estimated by using Folin-Ciocalteu method with slight modification. The methanolic extracts and Gallic acid (standard phenolic compound) were mixed with Folin-Ciocalteu reagent (0.5 ml) and incubate at room temperature for 3 min followed addition of 2% Na2CO3 and incubated for 1 min in boiling water bath. The sample under tap water and optical density recorded at 650nm. The amount of phenolic was determined by plotting standard graph and concentration expressed in Gallic acid equivalents (GAE mg/ml).

**Antioxidant assay:**

DPPH radical scavenging activity of the different extracts was estimated using a slight modification of the protocol reported earlier. (Yamaguchi T., et al., 1998). For a typical reaction, 2ml of 100um DPPH solution in ethanol/acetone/methanol was mixed with increasing phenolics concentration of extract. The ascorbic acid was used as standard reference antioxidant. The reaction mixture was recorded at 517nm against the blanks. For the control, DPPH solution in ethanol/acetone/methanol was taken without plant extracts and the optical density was carried out in triplicate. The decrease in optical density of DPPH on
addition of test samples in relation to the control was used to calculate the antioxidant activity, as percentage inhibition of DPPH radical scavenging calculated using the following equation.

Effect of scavenging (\%) = \left(1 - \frac{A_{\text{sample}} (517 \text{nm})}{A_{\text{control}} (517 \text{nm})}\right) \times 100.

Statistical Analysis:
The experimental results were expressed as the mean\(+\) (-) standard deviation (SD) of replicates. Where applicable, the data were subject to one way analysis of variance (ANOVA) and differences between samples were determined by Bio stat 2,0,1,5,8,4 program. Values of p<0.005 were regarded as significant.

Results and discussion:
The interest in the phenolics has increased outstandingly due to their prominent free radical scavenging activity. Phenolic compounds could be classified as simple phenols, a single aromatic ring bearing at least one hydroxyl group, and polyphenols with at least two phenol subunits like flavonoids or three and more phenol subunits called tannin. The phenolics content in different solvent extracts prepared from barks of \textit{Ficus Carica}, \textit{Ficus Racemosa}, \textit{Ficus Benghalnensis} respectively, were estimated by using a standard method of Folin-ciocalteu. Ethanol has been proven as effective solvent to extract phenolic compounds (table 1). The maximum phenolics 3.18±1.499 mg/ml GAE was estimated in Ethanol followed by Acetone and Methanol of \textit{Ficus Benghalnensis}. The presence of minimum amount of phenolics among all three species 1.14±0.537 mg/ml was observed in \textit{Ficus Carica} species (table 1).

The antioxidant potential was determined by using DPPH (Diphenylpicrylhydrazyl). The increasing concentrations of phenolics were treated with DPPH and their effects were observed like dose dependents Antioxidative potential (figure 1, 2, 3). The maximum IC50 value 0.28 ± 0.395µg/ml was observed in \textit{Ficus Benghalnensis} species and the minimum IC50 was observed in \textit{Ficus Racemosa} species (table 2). The antioxidant potentials in \textit{Ficus Benghalnensis} and \textit{Ficus Carica} were similar to standard antioxidant i.e. ascorbic acid (0.50). The IC50 value of \textit{Ficus Racemosa} was minimum and hence it was confirmed that the phenolics in \textit{Ficus Benghalnensis} have an enormous Antioxidative potential than two other species. The phenolics in \textit{Ficus Carica} also have an excellent Antioxidative potential. DPPH is a stable free radical . When antioxidant reacts with this stable radical, the electron becomes paired off and bleaching of the colour stochiometrically depends on the number of electrons taken up.

Graph 1: Graph of three target species total phenolics content obtained in target species compared With Standered Gallic acid
Fig 2: DPPH scavenging activity of targeted species compared with standard Ascorbic acid

Table 1: Total phenolic content and Antioxidative potential of *Ficus* species

<table>
<thead>
<tr>
<th>Sr.no</th>
<th>Medicinal plants</th>
<th>Total Phenolics contain GAE in mg/ml (n=3)</th>
<th>IC50 µ/ml (n=3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td><em>Ficus Benghalnesis</em></td>
<td>3.18±1.499</td>
<td>0.84±0.395</td>
</tr>
<tr>
<td>2.</td>
<td><em>Ficus Carica</em></td>
<td>2.46±1.018</td>
<td>0.27±0.127</td>
</tr>
<tr>
<td>3.</td>
<td><em>Ficus Racemosa</em></td>
<td>3.01±1.461</td>
<td>0.03±0.042</td>
</tr>
</tbody>
</table>

**Conclusion:**
The result of Ethanolic extract of *Ficus Carica, Ficus Racemosa, and Ficus Benghalnesis* showed strong antioxidant and free radical scavenging activity. It has been recognized that the total phenolic content enhance the free radical scavenging activity due to the presence of hydroxyl groups. The Bark parts of the *Ficus Benghalnesis* showed higher antioxidant activity that catching in scavenging DPPH free radicals, which indicates that the Extract has good potential as a source for natural antioxidants to prevent Free radical mediated oxidative damage. A little differences of antioxidant activity also were observed in all three species.
taken for experiment plants. The total phenolic contains in species *Ficus Benghalensis* is 3.18±1.499 mg/ml and Antioxidant activity is 0.84±0.395µg/ml. In *Ficus Carica* total phenolic contain is 2.46±1.018mg/ml and antioxidant activity is 0.27±0.127µg/ml. And in *Ficus Racemosa* total phenolic contain is 3.01±1.461 and antioxidant activity is 0.03±0.042µg/ml.
REFERENCES


