Antioxidant Properties and Total phenolic content of herbs used in post partum diet therapy in Patna (Bihar), India

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Abstract: The traditional medicinal system of India offers a detailed diet therapy for post partum health care to rejuvenate women's health. The minority community in Patna (Bihar) uses selected herbs as traditional recipes for post partum health care. Current study was designed to investigate pharmacological studies to validate folk use of such herbal recipes. All plants were tested for DPPH, FRAP AND total phenolic content. The investigation showed significant therapeutic effect of folk medicinal recipes used as traditional post partum care.

Keywords: Patna, DPPH, FRAP, Phenols, Postpartum care.

I. Introduction

Antioxidant with free radical scavenging activities may have great relevance in the prevention and therapeutics of diseases in which oxidants or free radicals are implicated currently, the possible toxicity of synthetic antioxidants has been criticized. Thus interest in natural antioxidants, especially of plant origin, has greatly increased in recent years. Plants may contain a wide variety of free radical scavenging molecules, such as phenolic compounds, (e.g. phenolic acids, flavonoids quinones, coumarins, lignans, stilbenes, tanins), terpenoids (including carotenoids), and some other endogenous metabolites, which are rich in antioxidant activity. Epidemiological and invitro studies suggested that plants are major constituent in antioxidant based drugs/ formulations used for the prevention of complex diseases. The use of herbs in traditional way is becoming revitalized over the world. More over since modern medicine in becoming more wide spread but at a high cost. In rural area, it is useful in certain case like surgery. Traditional medicine still plays an important role and commonly used as primary form of health care for people especially in critical events such as in post partum care of women. In many south east Asian cultures post partum period is considered important from point of view of recovery, by offering a period of confinement ranging from 10 to 45 days. Many behavioral and dietary restrictions are followed during this period. Diet therapy includes preparations of medicinal herbs in the form of decoctions, infusions and cold extracts. The herbs given during this period are traditionally known to strengthen the body and mind and prevent disorder such as a post partum depression (PPD), insomnia, body aches indigestion, and oxidative stress. The diet is also helpful for growth and development of new born as bioactive molecules pass from mother to child through lactation. This study was then designed to test antioxidation and to determine total phenolic contents of selected herbs, used in diet therapy after parturition in postpartum care in Patna.

II. Material And Methods

II.1. Plant Material

The plant samples were obtained from the medicinal plant traders and identified in the laboratory based on local name and pharmacognosy.

II.2. Chemicals And Instruments

Folin-Ciocalteu's phenol reagent, gallic acid, anhydrous sodium carbonate, ethyl alcohol, methanol, 2,2-di phenyl-1-Picrylhydrazyl (DPPH) were of analytical grade and procured from local sources.

II.3. Sample Collection And Extraction

Ten species of herbs, commonly used in postpartum care were selected for the study. Each sample was collected from three outlets, which were pooled and considered as single sample. Each sample was extracted and analyzed in triplicates. One hundred gram of dry ground plant material was macerated in 70% ethyl alcohol shaken for five hours then kept at room temperature for 24 hours in closed containers. The extraction process was repeated three times. Then the extracts were filtered under vacuum and concentrated at reduced pressure using a rotary evaporator. The dried extracts were kept in the refrigerator at 4°C until use.
II.4. Determination Of Total Phenolic Content

The amount of total phenolics in extracts was determined with the Folin–Ciocalteu reagent. Gallic acid was used as a standard and the total phenolics were expressed as mg/g gallic acid equivalents (GAE)\(^4\). Concentration of 0.01, 0.02, 0.03, 0.04 and 0.05 mg/ml of gallic acid were prepared in methanol. Concentration of 0.1 and 1mg/ml of plant extract were also prepared in methanol and 0.5ml of each sample were introduced into test tubes and mixed with 2.5ml of a 10 fold dilute Folin–Ciocalteu reagent and 2ml of 7.5% sodium carbonate. The tubes were covered with parafilm and allowed to stand for 30 minutes at room temperature before the absorbance was at read at 760nm spectrophotometrically. All determination was performed in triplicate. The Folin-Ciocalteu reagent is sensitive to reducing compounds including polyphenols, thereby producing a blue colour upon reaction. This blue colour is measured spectrophotometrically. Thus total phenolic content can be determined\(^5\).

II.5. DPPH Radical-Scavenging Activity

DPPH radical-scavenging activity was determined according to Aoshima et al.\(^6\) This method is based on the ability of the antioxidant to scavenge the DPPH cation radical. Briefly, to 100 \(\mu\)L of sample extract or standard, 2.9 \(\mu\)L of DPPH reagent (0.1 mM in methanol) was added and vortexed vigorously. This was allowed to stand in dark for 30 min at room temperature, and the discoloration of DPPH was measured against a suitable blank at 517 nm. Percentage inhibition of the discoloration of DPPH by the sample extract was expressed as Trolox equivalents (mg/100 g).

II.6. FRAP Assay

Ferric reducing antioxidant power (FRAP) was determined according to Benzie and Strain\(^7\). In the presence of TPTZ, the Fe\(^{+2}\)-TPTZ complex exhibits blue color which is read at 593 nm. Briefly, 3.0 \(\mu\)L of working FRAP reagent was added to an appropriate volume/concentration of the sample extract, incubated for 6 min at room temperature, and the absorbance was measured at 593 nm against FeSO\(_4\) standard.

II.7. Statistical Analysis

All the data were recorded in triplicate (n=3) and results have been expressed as mean ± standard deviation. To correlate the results obtained with different methods, a regression analysis was performed and correlation coefficients were calculated.

III. Results And Discussion

The amount of total phenolics measured by folin-Ciocalteu method varied widely in herbal materials and ranged from 3.86 to 635.28 GAE/g dry weight (dw) Table (i). The highest level of phenolics was found in Quercus infectoria, while lowest was in Asparagus racemosus. Herbs like Butea monosperma, Terminalia chebula, showed a highest phenolic content of 416.56, 156 mg GAE/g of sample, respectively. Among the selected herbs Asparagus racemosus and Piper longum showed a very low phenolic content (3.86 and 4.11 mg GAE/g respectively). DPPH radical scavenging activities of herbs varied from 21.19 to 89.19 %. Quercus infectoria extract showed highest antioxidant capacity (89.19% of DPPH inhibition, followed by Trachyspermum ammi, Butea monosperma, Myrtisica fragrans and Zingiber officinalis, which all are also showed in 85% of DPPH inhibition. Among 10 sample 5 samples showed DPPH inhibition in the range of 80-90%. Ferric reducing antioxidant potential of plant extracts tested varied from 48 mM Fe\(^{+2}\)/g to 1225 mMFe\(^{+2}\)/g. Quercus infectoria (1225 mMFe\(^{+2}\)/g), Terminalia chebula (1119 mMFe\(^{+2}\)/g), Myrtisica fragrans (599 mMFe\(^{+2}\)/g) showed very strong feric ion reducing activities. In this assay, Piper longum (48 mMFe\(^{+2}\)/g) and Zingiber officinalis (49 mMFe\(^{+2}\)/g) showed the lowest feric reducing capacity. To correlate the results obtained with the different methods a regression analysis was performed (correlation coefficient, R). The results are shown in figures 1-3 and table 2. Correlation antioxidant potential were maximum between DPPH and FRAP (R=0.4345). The minimum correlation was found between DPPH and TPC (R=0.1015). It is noticed that the highest concentration of phenolic compound in the extracts were obtained using high polarity, the high content of phenolic compound and significant linear correlation between the values of the concentration and antioxidant activity indicated that these compounds contribute to the higher radical scavenging activity. Several herbs contain lower antioxidant activities in comparison to others. Folk use of these herbs may not necessarily be related to antioxidant activities, but for other purposes like stimulant and glactogogue properties. These herbs may also be consumed with different plant combination and act as bioenhancers.
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IV. Figures And Tables

Table 1: Antioxidant capacity and total phenolic content in 10 herbs used in diet therapy during postpartum healthcare.

<table>
<thead>
<tr>
<th>Herb Name</th>
<th>Local Name</th>
<th>Part Used</th>
<th>Total Phenolic content (mg GAE/gram)</th>
<th>DPPH% inhibition</th>
<th>FRAPmFe+2/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asparagus recemosus</td>
<td>Shatavari</td>
<td>Roots</td>
<td>3.86±0.32</td>
<td>25.26±0.61</td>
<td>70±1.16</td>
</tr>
<tr>
<td>Butea monosperma</td>
<td>Kamarkas</td>
<td>Gum</td>
<td>416.56 ±7.28</td>
<td>86.16±1.29</td>
<td>76.0±1.28</td>
</tr>
<tr>
<td>Curcuma longa</td>
<td>Haldi</td>
<td>Rhizome</td>
<td>15.56±0.32</td>
<td>75.35±1.25</td>
<td>71±1.56</td>
</tr>
<tr>
<td>Embelia ribes</td>
<td>Vaividang/</td>
<td>Fruits</td>
<td>13.16±0.32</td>
<td>69.57±0.71</td>
<td>39±2.16</td>
</tr>
<tr>
<td>Myristica fragrans</td>
<td>Jauphal</td>
<td>Seeds</td>
<td>11.0±0.03</td>
<td>85.89±1.58</td>
<td>599±3.50</td>
</tr>
<tr>
<td>Piper longum</td>
<td>Pipramul</td>
<td>Roots</td>
<td>4.11±0.21</td>
<td>43.47±0.10</td>
<td>48±0.86</td>
</tr>
<tr>
<td>Quercus infectoria</td>
<td>Majuphal</td>
<td>Galls</td>
<td>635.25±9.3</td>
<td>89.19±2.2</td>
<td>1225±9.39</td>
</tr>
<tr>
<td>Terminalia chebula</td>
<td>Harad/</td>
<td>Fruits</td>
<td>156.0±0.33</td>
<td>74.68±0.83</td>
<td>1119±8.87</td>
</tr>
<tr>
<td>Trachyspermum ammi</td>
<td>Ajwain</td>
<td>Seeds</td>
<td>13.56±0.98</td>
<td>86.29±1.01</td>
<td>951±7.42</td>
</tr>
<tr>
<td>Zingiber officinale</td>
<td>Saunth</td>
<td>Rhizome</td>
<td>10.3±0.39</td>
<td>85.12±0.51</td>
<td>49±0.69</td>
</tr>
</tbody>
</table>

Table 2. Correlation Coefficient (R) between assays.

<table>
<thead>
<tr>
<th>Assay</th>
<th>FRAP</th>
<th>DPPH</th>
</tr>
</thead>
<tbody>
<tr>
<td>DPPH</td>
<td>0.4345</td>
<td></td>
</tr>
<tr>
<td>TPC</td>
<td>0.3710</td>
<td>0.1015</td>
</tr>
</tbody>
</table>

Fig. 1: Correlation between DPPH and FRAP assays. Correlation coefficient R = 0.4345

Fig. 2: Correlation between TPC and DPPH assays. Correlation coefficient R = 0.1015
Conclusion

Results of our studies suggested that antioxidant properties of these herbs playing use in post natal recovery. The World Health Organization has also recognized the importance of traditional medicine and has created strategies and guide lines and standards for botanical medicine. Proven agroindustrial technologies need to be applied to the cultivation and processing of medicinal plants and the manufacture of herbal medicine. Based on this information it could be concluded that these herebs are natural sources of antioxidant substances of high importance. The investigation supports the folk use of these plants in post partum care.

Acknowledgement

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References