Antioxidant potential of five Ksheerapaka’s and Kashaya’s, Ayurvedic decoctions

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Five milk decoctions and water decoctions prepared by using coriander; ginger, pepper, tulsi and turmeric were evaluated for in-vitro antioxidant activity using standard DPPH, ABTS and hydrogen peroxide methods. The milk decoctions exhibited potent antioxidant activity when compared to their corresponding water decoctions. The total phenol content of the milk decoctions was also found to be high supporting the antioxidant activity. The study provides a scientific validation of the common preference of milk decoctions over water decoctions in Ayurveda for a few plants.

Keywords: Ayurveda, Ksheerapaka, Kashaya, Free radicals, Radical scavenging activity, Antioxidant activity


Ayurveda is not only a living tradition, but also a healthy tradition. In Ayurveda, preserving the life is the first step and promoting its health is the next. In Ayurveda, for several plants milk decoctions (Ksheerapaka’s) are preferred over water decoctions, (Kashaya’s) for the prevention and treatment of certain diseases. These preparations are used as a cure for cold and cough and taken as prophylactic during rainy & winter seasons. Coriander, ginger, pepper, tulsi, and turmeric are some of the commonly used crude drugs in such preparations. Ethnomedicinal uses of these drugs include cardiac diseases, stimulant, tonic, carminative, liver disorders, anaemia, cough, asthma, etc. Many of these uses are related to antioxidant activity. These plants also contain phenolic substances, which when present are responsible for the antioxidant nature. There is an increasing evidence to support the involvement of free radical reactions in several human diseases. Many plants and their decoctions are rich sources of antioxidants, which can prevent the occurrence of a large number of diseases like arteriosclerosis, hypertension, cancer and inflammatory diseases. In the study, comparative antioxidant potential of five Ksheerapaka’s over Kashaya’s of five commonly used plants have been evaluated (Table 1). Invitro antioxidant activity was carried out using scavenging of DPPH, ABTS and hydrogen peroxide methods. Total phenol content was estimated using Folin Ciocalteu reagent.

Methodology

Dried rhizomes of turmeric (Curcuma longa Linn., Family- Zingiberaceae) and ginger (Zingiber officinale Rosc., Family– Zingiberaceae), seeds of pepper (Piper nigrum Linn., Family– Piperaceae), coriander (Coriandrum sativum Linn., Family – Umbelliferae) and leaves of tulsi (Ocimum sanctum Linn., Family- Labiatae) were collected from the Municipal market, Ootacamund, and authenticated by Medicinal Plants Survey and Collection Unit, Government Arts College, Ootacamund. The plant materials were powdered separately and used for extraction. Each plant powder (1 gm) was extracted separately with fresh milk (100 ml) under reflux for 1hr in a RB flask. The mixture was cooled, filtered through a muslin cloth and centrifuged. The supernatant was collected, made up the volume to 100 ml with fresh milk and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above. The final volume was made up to 100 ml with distilled water and used. Similarly, each plant material was extracted separately with distilled water (100 ml) under reflux for 1hr and processed as above.
To determine the percentage inhibition of DPPH free radical, the observed potent antioxidant activity by DPPH method was compared to their corresponding water decoctions. The percentage inhibition observed by ginger, pepper and turmeric milk decoctions was found to be 2.8, 1.75 and 1.3 times more than that observed for the corresponding water decoctions. In ABTS method, the milk decoctions of ginger, tulsi and turmeric showed more activity compared to their water decoctions. The percentage inhibitions of these three milk decoctions were found to be 2.3, 14 and 3 times more than that observed for the corresponding water decoctions. Both the decoctions of coriander showed almost similar activity in the ABTS and DPPH methods, respectively. Similarly, in the scavenging of H₂O₂, milk decoctions of coriander, ginger, pepper and turmeric showed more activity compared to their water decoctions. The percentage inhibition observed by ginger, pepper and turmeric milk decoctions was found to be 2.8, 1.75 and 1.3 times more than that observed for the corresponding water decoctions. The increase in the total phenol estimation, all the milk decoctions exhibited more total phenol content compared to their corresponding water decoctions. The decrease in the total phenol content for coriander, ginger, pepper, tulsi and turmeric milk decoctions was found to be 4, 4.2, 2.68, 1.8 and 3.2 times more than their corresponding water decoctions, respectively.

**Discussion**

*Ksheerapaka’s* are commonly used in household remedies since ancient times in India and are preferred over *Kashayas*. In the study, a comparative evaluation of antioxidant activity of *Ksheerapaka’s* of five commonly used plants was carried out. In most of the methods, all the milk decoctions showed better antioxidant activity than the corresponding water decoctions. The total phenol content of milk decoctions was also found to be high. Since, total phenol content is directly related to antioxidant, the observed potent

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**Table 1—*In-vitro* antioxidant activity and total phenol content of five milk and water decoctions**

<table>
<thead>
<tr>
<th>Sample</th>
<th>Percentage Inhibition* by method</th>
<th>Total phenol content* (mg/gm of gallic acid equivalent)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DPPH</td>
<td>ABTS</td>
</tr>
<tr>
<td></td>
<td>Milk extract</td>
<td>Water extract</td>
</tr>
<tr>
<td>Coriander</td>
<td>49.04 ± 3.05</td>
<td>45.83 ± 11.59</td>
</tr>
<tr>
<td>Ginger</td>
<td>43.99 ± 5.60</td>
<td>15.64 ± 1.75</td>
</tr>
<tr>
<td>Pepper</td>
<td>57.22 ± 2.50</td>
<td>32.63 ± 1.10</td>
</tr>
<tr>
<td>Tulsi</td>
<td>47.42 ± 6.30</td>
<td>51.33 ± 1.40</td>
</tr>
<tr>
<td>Turmeric</td>
<td>57.72 ± 0.89</td>
<td>44.25 ± 2.00</td>
</tr>
</tbody>
</table>

* Average of three determinations, (-) means no inhibition

490 nm against the corresponding test blanks. Percentage inhibition of DPPH free radical was calculated using the following formula:

\[
\text{% Inhibition} = \left(\frac{\text{Control} - \text{sample}}{\text{Control}}\right) \times 100
\]

This involves a reaction between ABTS and potassium persulphate to produce the ABTS radical cation, a blue green chromogen. In the presence of antioxidant-reductant, the coloured radical is converted back to colourless ABTS, the absorbance of which is measured at 734 nm. To 0.2 ml of each sample, 1 ml of distilled DMSO and 0.16 ml of ABTS solution were added and incubated for 20 min. Absorbance was measured at 734 nm. Percentage inhibition of ABTS radical cation was calculated.

A solution of hydrogen peroxide (20 mM) was prepared in phosphate buffered saline (PBS, pH 7.4). Each sample (1 ml) was added to 2 ml of hydrogen peroxide solution in PBS and after 10 min, the absorbance was measured at 405 nm. The percentage inhibition was calculated. The total phenol was determined by using Folin Ciocalteu reagent. To 0.4 ml of each sample, 2 ml of Folin Ciocalteu reagent and 1.6 ml sodium carbonate (0.7 M) were added. The solutions were kept at room temperature for 2 hrs and the absorbance was measured at 750 nm using a Shimadzu UV–160 spectrophotometer. Using Gallic acid monohydrate, a standard curve was prepared and linearity was obtained in the range of 1–10 μg/ml. Using the standard curve, the total phenol content was calculated and expressed as the gallic acid equivalent in mg/g of each sample.

**Results**

Among the five milk decoctions, coriander, ginger, pepper and turmeric decoctions showed potent antioxidant activity by DPPH method when compared...
antioxidant activity of the milk decoctions may be due to their high total phenol content potentials\(^2\),\(^3\). Milk is both a non-polar and a polar solvent due to its high fat and water contents. Hence, both these types of active constituents are extracted in milk. This may be the reason for its preference over water for extraction in Ayurveda. Apart from being most essential for life activity, the other advantages of milk are its natural source, presence of most of the vitamins & minerals and its use as a general tonic. Hence, along with taking more quantities of phytoconstituents as drugs, the recipient of Ksheerapaka's is having the benefit of these nutrients. In an earlier report, milk decoction of pippali (long pepper) exhibited 27 times higher brine shrimp lethality than the water decoction. The amount of phytoconstituents extracted by milk was found to be more when studied by HPTLC\(^8\). The study supports the same in terms of antioxidant potentials. The study provides a proof and scientific validation for the preference of Ksheerapaka's over Kashaya's in Ayurveda. However, further phytochemical and in vivo antioxidant studies are needed to confirm the same.

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References