ESTIMATION OF TOTAL PHENOLIC AND FLAVONOID CONTENT OF POLYHERBAL FORMULATION DASHANG LEPA

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ABSTRACT

**Background:** Indian system of medicine mainly based on polyherbal compound to treat various types of diseases. *Dashang lepa* is one of the prestigious formulation of ten indigenous drugs which are having predominantly anti-inflammatory properties and clinically used in inflammatory swelling like boil, furunculosis, cellulitis, erysipelas etc. As several reports are on various individual phytochemical, phenolic and flavonoid content but none is having study of polyherbal compound *Dashang lepa*. **Objectives:** Total phenolic and total flavonoid content were studied of alcoholic extract of *Dashang lepa*. **Method:** Methanolic extraction was done by hot percolation method through soxhlet apparatus. Phenolic content was estimated using Follin ciocalteau reagent, flavonoid using aluminum chloride (2%) reagent as quercetin equivalent. **Results:** Estimation of total phenolic and total flavonoid content showed that *Dashang lepa* extract was having phenolic content (15.8 ± 1.14) μg Gallic acid equivalents (GAE) and flavonoid content (22.4 ± 2.54) in μg of quercetin equivalents (QE). **KEYWORDS:** Polyherbal compound, *Dashang lepa*, Phenolic, Flavonoid, Follin ciocalteau reagent, quercetin.

INTRODUCTION

*Dashang lepa* is mentioned in several *Ayurvedic* literatures and clinically used as anti-inflammatory in various diseases [¹⁵]. It is used by making a paste with lukewarm cow’s pure butter (Ghrita) as external application. *Dashang lepa* is used in different superficial inflammatory condition as local applicant by the Department of Shalya Tantra, Faculty of Ayurveda, IMS, BHU, Varanasi, since last one decade. Clinically it is very effective after local application against inflammatory condition over such diseases as cellulitis, boil, furunculosis, erysipelas, unripe abscess, lymphadenitis, dermatitis and also in arthritis. *Dashang lepa* contains Shirisha [Albizia lebbeck (L.) Benth.], Madhuyashti [Glycyrrhiza glabra Linn.], Tagara [Valeriana wallichii Dc.], Raktachandnam [Pterocarpus santalinus Linn.F.], Ela [Elettaria cardamomum (L.) Maton], Jatamansi [Nardostachys jatamansi (D.Don) Dc.], Haridra [Curcuma longa Linn.], Daruharidra [Berberis aristata DC.], Kushta [Saussurea lappa C.B.Clarke], Hriffera [Pavonia odorata Willd]. A preliminary phytochemical analysis, thin layer chromatography, loss on drying (moisture content), Ash value and pH value of *Dashang lepa* have been analysed by the authors[⁶]. But evaluation of total flavonoid and phenolic content was necessary to find out anti-inflammatory action of *Dashang lepa* so this study was conducted. As all these Plants are able to synthesize a multitude of organic molecules/phytochemicals, referred to as “secondary metabolites”[⁷,⁸]. Phenols are associated with diverse functions, including nutrient uptake,
protein synthesis, enzyme activity, photosynthesis, structural components and allelopathy\[9-11\]. Phenolics show an array of health promoting benefits in human health. They are of current interest due to their important biological and pharmacological properties, especially the anti-inflammatory\[12\], antioxidant\[13\], antimutagenic and anticarcinogenic activities\[14, 15\].

Flavonoids are universal within the plant kingdom. More than 5000 flavonoids have been identified in nature\[16\]. They functions as stress protectants in plant cells by scavenging reactive oxygen species produced by the photosynthetic electron transport system\[17\]. Due to UV-absorbing properties, flavonoids protect plants from the UV radiation of the sun and scavenge UV-generated reactive oxygen species\[18\].

So this study was planned to evaluate total phenolic and flavonoid content of the drug *Dashang lepa*.

**MATERIAL AND METHODS**

**Plant material and preparation of extract**

The plants of *Dashang lepa* were collected from local market of Varanasi (India). The identification of the plants was done by Department of Dravyaguna, BHU, Varanasi. Air shade dried and pulverized plants parts were extracted with hydroalcoholic solvent separately by hot percolation method through soxhlet apparatus. Thereafter extract was dried using rotary evaporator and dried extract was put to the process of standardization. This extract was used to investigate the total content of phenols and flavonoids.

**Estimation of total phenolic content**

Total Phenolic concentration of *Dashang lepa* alcoholic extract was measured by Folin-Ciocalteu assay \[19\]. Briefly, 5ml of distilled water, 0.5-1.0ml of sample, and 1.0ml of Folin-Ciocalteu reagent was added to a 25ml flask. The content was mixed and allowed to stand for 5-8min at room temperature. Next 10ml of 7% sodium carbonate solution was added followed by distilled water. Solution were mixed and allowed to stand at room temperature for 15min, and then absorbance was recorded at 750 nm. Total phenolic content was standardized against Gallic acid and expressed as milligram per liter of Gallic acid equivalents (GAE). The linearity range for this assay was determined as 0.5-5.0mg/l GAE (R²=0.999), giving an absorbance range of 0.050-0.555 absorbance units. Experiments were performed in triplicates and results were recorded as mean ± SEM (Standard Error Mean).

**Estimation of total flavonoid content**

Total flavonoid content was measured by using aluminum chloride (2%)\[20,21\] in which it was mixed with solution of test samples. Absorbance reading at 415nm (Elico SL 177) were taken after 10 min against a blank sample consisting of 5ml of sample solution and 5ml of methanol without aluminum chloride. The total Flavonoid content was determined using a standard curve of quercetin at 0-50μg/ml. The average of three readings were used and then expressed in μg quercetin equivalent flavones per mg extract. Experiments were performed in triplicates and results were recorded as mean ± SEM (Standard Error Mean).

**RESULTS**

Estimation of total phenolic and total flavonoid content showed that *Dashang lepa* extract was having phenolic content (15.8 ± 1.14) μg Gallic acid equivalents (GAE) and flavonoid content (22.4 ± 2.54) in μg of quercetin equivalents (QE). The amount of total phenolic and flavonoids for the test samples are summarized in table no. 1.

<table>
<thead>
<tr>
<th>Table 1: Phenolic and flavonoid content <em>Dashang lepa</em></th>
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<tr>
<td>Extract</td>
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<tr>
<td>1. Total phenolic content Gallic acid equivalents (GAE)</td>
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<tr>
<td>2. Total flavonoid content Quercetin equivalents (QE)</td>
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**CONCLUSION**

This study indicates that polyherbal compound *Dashang lepa* has showed presence of high amount of phenolic and flavonoid content suggesting the use of *Dashang lepa* for treatment of various inflammatory conditions.
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PHOTOGRAPHS

Shirisha  Madhuyashti  Tagara
Raktachandnam  Ela  Jatamansi
Haridra  Daruharidra  Kushta
Hrivera  Dashang Lepa
<table>
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<th>Total phenolic Content (µg of GAE/mg)</th>
<th>Total Flavone content (µg of QE/mg)</th>
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<td>Total phenolic/flavonoid content</td>
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**Total phenolic and flavonoid Content of *Dashang lepa***