COMPARATIVE PHYTOCHEMICAL ANALYSIS OF DIFFERENT PARTS OF STHIRAA- Desmodium gangeticum (L.) DC.

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ABSTRACT

Desmodium gangeticum (L.) DC. (Family: Fabaceae) commonly known as Sthira in Sanskrit, is an important medicinal plants of Ayurveda. Root of this plant is one of the main ingredients of many Ayurvedic formulations used in various disorders mentioned in classical texts. The widespread uses of this important medicinal plant by different pharmaceutical industries along with the current appraisal of attention in herbal medicine have led to an ever-increasing demand for this species. Because of this reason, to face the huge demand, the market samples of this plant contain whole plant instead of root alone. Presently the various pharmaceutical companies use the whole plant instead of root. Hence this becomes a contradictory issue to what Acharyas have advised. The situation become even more adverse as there is no detailed comparative data available on different useful parts. On this background present study was undertaken to compare the phytoconstituents of root, shoot and whole plant of Sthira through HPTLC analysis. Study showed the root sample contains more number of phytoconstituents which supports the scientific logic of ancient Acharyas for advising the root as useful part of Sthira. Other two samples shoot and whole plant part contains equal quantity of phytoconstituents. Study suggests for further analysis on utilising the shoot part instead of uprooting whole plant which may prevent the complete destruction of this important plant.
KEYWORDS: Sthiraa, root, shoot, whole plant comparative HPTLC.

INTRODUCTION

Desmodium gangeticum (L.) DC. commonly known as Sthiraa is a well-known and important medicinal plant in Ayurveda. In the Ayurvedic literature it has included under Dashamoola (group of ten roots) and it is one of the main ingredients of numbers of Ayurvedic formulations used in various disorders.[1,2] The plant is an erect, diffusely branched perennial under shrub of Fabaceae family, commonly found almost throughout India. Leaves are simple, ovate lanceolate.[3] Flowers are white or purple or lilac in elongate lax terminal or auxiliary racemes. Fruits glabrescent, pods sparsely pubescent with hooked hairs, joints separate when ripe into indehiscent one seeded segments, seeds compressed and reniform.[4]

In Kerala and Tamil Nadu the roots of this plant are used as prisniparni. Another species Desmodium latifolium DC. is often used as a substitute for Desmodium gangeticum in parts of Travancore and Cochin.[5-7] It contains various classes of bioactive principles such as flavonoid glycosides, pterocarpanoids, lipids, glycolipids, lactones[8, 9] and alkaloids[9,10]. Pterocarpan-gangetin, gangetin and desmodin has been isolated from the whole plant and root.[11-14] The plant is found to possess various biological activities such as anti-inflammatory, analgesic and antipyretic activity,[15,16] Anti-ulcer, cytoprotective and anti-secretary activity[17] hypocholesterolemic[18], anti-diabetic activity[19], anti-leishmanial activity[20] and anti-implantation activity[21] along with it is effective as anti-writhing and central nervous system (CNS) depressant activity[22], in improving memory[23] and in healing different types of wounds.[24,25] The extensive uses of this plant by different pharmaceutical industries coupled with the recent revival of interest in herbal medicine have led to an ever-increasing demand of this species.[26] Hence most of the market samples of this plant contain whole plant instead of only root. Therefore, Ayurvedic formulations with Sthira contain whole part of this plant which becomes contradictory of Ayurvedic classics as they have opined the root as useful part. It has therefore become essential to find out the comparative analysis of root and other part of plant to ensure the quality of the raw drug. On this background, present study was undertaken to compare the phytoconstituents of root, shoot and whole plant of Sthiraa through HPTLC analysis.

MATERIALS AND METHODS

Plant materials: Roots, Shoot (aerial part) and whole part of Desmodium gangeticum (L.) DC. were collected during May 2015, from the campus of VPSV Ayurveda College,
Kottakkal. Materials were authenticated at Centre for Medicinal Plant Research (CMPR), AVS, Kottakkal. Collected materials were washed thoroughly using running tap water, rinsed in distilled water and shade dried in open air.

**Preparation of aqueous extract:** The shade dried three different samples of *Sthiraa* were taken separately. The samples were pulverized, finely sieved and soaked 500g of each sample powder in 2 lit of distilled water separately for 24 h and after that materials were filtered. The filtrates were evaporated in a rotator evaporator and used for the experimentation.

**HPTLC analysis:** HPTLC of water extract of root, shoot and whole plant samples of *Sthiraa* were compared in solvent system-Toluene: Ethyl acetate: formic acid (5: 5: 1). Standard procedure was followed for the analysis as per method mentioned in API.

**Procedure:** Samples were applied on the plate using Camag automatic TLC sampler 4 attached to camag HPTLC system. The samples (2µl) each were spotted on aluminum backed pre-coated silica gel plate 60F-254 plate (5×10 cm) in the form of bands with width 8 mm by using Hamilton syringe (100µl). Then the plates were developed in different solvent systems in a twin trough chamber to a distance of 9 cm.

**Visualization:** The plates were dried in air and examined under UV 254 nm and under UV 366 nm. *R*<sub>f</sub> values and colors of the resolved bands were recorded. Photographs of the plates were captured using camag TLC visualizer.

**Derivatization:** Derivatizations of the plates were done by using iodine solution. Photographs of the plates were captured using camag TLC visualizer.

**Scanning:** Densitometric scanning of the plates was done by using camag TLC scanner 3 at 254 and 366 nm.

**OBSERVATION AND RESULT**

HPTLC profile of water extract of whole plant and shoot of *D. gangeticum*, showed 4 spots with *R*<sub>f</sub> values of 0.17, 0.31, 0.40, 0.62 under UV 254 nm (Fig-1,2a, Table-1). But samples of root showed 6 spot with *R*<sub>f</sub> values of 0.12, 0.31, 0.32, 0.40, 0.46, 0.68 (Fig-1, 2a,Table-1). When the plates were viewed at UV 366 nm, the whole plant and shoot sample showed 3 spots with *R*<sub>f</sub> values of 0.25, 0.39, 0.46 (Fig-1,2b,Table-2). But at 366 nm sample of root showed 8 spots with *R*<sub>f</sub> values of 0.09, 0.12, 0.16, 0.32, 0.39, 0.46, 0.52, 0.60 (Fig-
1,2b,Table-2). After derivatization of the plates by using iodine solution sample of root showed 4 spots with \( R_f \) values of 0.12, 0.15, 0.32, 0.61(Fig-1,Table-3) where as no spot was found in sample of whole plant and shoot.

Table 1: HPTLC comparison of different useful part of *Sthiraa* under UV 254nm

<table>
<thead>
<tr>
<th>Whole Plant</th>
<th>Shoot</th>
<th>Root</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>-</td>
<td>0.12 (L Brown)</td>
</tr>
<tr>
<td>0.17(D Brown)</td>
<td>0.17(D Brown)</td>
<td>-</td>
</tr>
<tr>
<td>0.31(D Brown)</td>
<td>0.31(D Brown)</td>
<td>0.31(D Brown)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.32 (L Brown)</td>
</tr>
<tr>
<td>0.40(D Brown)</td>
<td>0.40(D Brown)</td>
<td>0.40(D Brown)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.46(D Brown)</td>
</tr>
<tr>
<td>0.62(D Brown)</td>
<td>0.62(D Brown)</td>
<td>0.62(D Brown)</td>
</tr>
</tbody>
</table>

*D- Dark, L-Light

Table 2: HPTLC comparison of different useful part of *Sthiraa* under UV 366nm

<table>
<thead>
<tr>
<th>Whole Plant</th>
<th>Shoot</th>
<th>Root</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>-</td>
<td>0.09(FL Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.12(FL Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.16(FL Blue)</td>
</tr>
<tr>
<td>0.25 (FL Blue)</td>
<td>0.25(FL Blue)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.32 (L Blue)</td>
</tr>
<tr>
<td>0.39(FL Blue)</td>
<td>0.39(FL Blue)</td>
<td>0.39 (D Blue)</td>
</tr>
<tr>
<td>0.46(FL Blue)</td>
<td>0.46(FL Blue)</td>
<td>0.46(D Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.52(D Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.60(D Blue)</td>
</tr>
</tbody>
</table>

*D- Dark, L-Light, FL-Florescent

Table 3: HPTLC comparison of different useful part of *Sthiraa* after derivatization:

<table>
<thead>
<tr>
<th>Whole Plant</th>
<th>Shoot</th>
<th>Root</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>-</td>
<td>0.12 (L Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.15(L Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.32 (L Blue)</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>0.61 (L Blue)</td>
</tr>
</tbody>
</table>

*L-Light
(Track-1: whole plant, Track-2: shoot part, Track-3: root part of *D. gangeticum*)

Solvent-Toluene : Ethyl acetate:formic acid (5 : 5: 1)

Figure 1. HTLC photo documentation of water extract of different useful part of *Sthiraa*

**DISCUSSION**

Ayurvedic classical text has documented the root as the useful part of *Desmodium gangeticum* (L.) DC. which is the botanical source of *Sthiraa*. Conversely due to high demand, less availability and economical issue whole plant is used in place of root. Hence the present study was undertaken for comparative phytochemical analysis of root, shoot and whole plant of *Desmodium gangeticum* (L.) DC. Here water extract of 3 different plant
samples were analysed. Water extract was specially selected as in most of the Ayurvedic formulations drugs are used in decoction or other water dissolved form. Study showed the samples of whole plant and shoot of the plants contain 4 similar colour of band in HPTLC under UV 254 nm but sample of root showed 6 spot (Fig-1, 2a, Table-1). That indicates the root of Stthira contain more number of chemical compounds than whole plant and shoot part. It may be due to the presence of more quantity of stem and leaf portion than root in whole plant. Spot with R_f values of 0.31, 0.40, 0.62 were found in all the three samples, that denotes 3 similar type of chemical compound may present in all three samples. Extra bands were found in root at R_f values of 0.12, 0.32, 0.46 but band at R_f value 0.17 (Fig-1, 2a, Table-1). which was found in both whole plant and shoot samples, was missing in root sample. That denotes one similar compound of whole plant and shoot is absent in root. At UV 366nm also root sample showed more compounds 8 dark spot (Fig-1,2b,Table-2) where as other two sample showed only 3 spot with similar colour and R_f value (Fig-1,2b,Table-2). Spot with R_f values of 0.39, 0.46 (Fig-1,2b,Table-2) were found in all the three samples, that denotes 2 similar type of chemical compound may present in all three samples. Extra band was found in root at R_f values of 0.09, 0.12, 0.16, 0.32, 0.52, 0.60 (Fig-1, 2b,Table-2) but band at R_f value 0.25 (Fig-1, 2b,Table-2) which was found in both whole plant and shoot samples, was missing in root sample. That denotes one similar compound which is present in whole plant and shoot, is absent in root. After derivatization of the plates by using iodine solution, sample of root showed 4 spots with R_f values of 0.12, 0.15, 0.32, 0.61 (Fig-1,Table-3) where as no spot was found in sample of whole plant and shoot. That denotes the 4 chemical compounds present in root are soluble in Iodine. 2 spots with R_f values of 0.12, 0.32 (Table-1, Table-2, Table-3) was found in root sample under UV 254nm, 366nm and also after derivatization of the plates by using iodine solution.

CONCLUSION

Result of the present study indicates that root of the Desmodium gangeticum (L.) DC. contain more number of active compounds compared to whole plant and shoot part of the plant. These data supports the logic behind the documentation of root of Stthira as the main useful part by Ayurvedic classics and also categorizing this plant under Dashamoolaa (group of ten roots). Study also revealed that shoot part and whole plant (with root) contains similar pattern of chemical compounds. Hence shoot part can be recommended instead of whole plant which can save the plant from complete destruction. Further scientific study should be done on this aspect.
ACKNOWLEDGEMENT
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REFERENCES


