

**CHEMOTAXONOMIC INVESTIGATION OF THE
GENUS *JUSTICIA* IN KERALA (UGC sanction order
No. MRP(S)-0745/13-14/KLCA023/UGC-SWRO
dated 28th March, 2014)**



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EXECUTIVE SUMMARY

Nature has been a source of medicinal agents for thousands of years and a large number of modern drugs have been derived from natural sources. Many of these drugs were isolated from plants based on the leads from their use in traditional or tribal systems of medicine. Traditional medicine, mostly based on plants derived products, has served as a source of alternative medicine, new pharmaceuticals and healthcare products. There is hundreds of medicinal plant species recognized to possess therapeutic value. Recently the search for components with novel biological properties has gained increasing importance due to growing concern about the alarming increase in the rate of deadly diseases like AIDS, cancer, diabetes, tuberculosis etc. Medicinal plants are natural products and their chemical composition varies depending on several factors, such as botanical species, chemotypes, the plant part used (root, bark, heart wood, stem, leaves, seed, flower, root, fruit, etc.), time of harvest, geographic area and also processing, storage conditions, etc. This variability can result in significant differences in pharmacological activity. Conventional pharmaceutical products, herbal medicinal products may vary in composition and properties, and increasing reports of adverse reactions has drawn the attention of many regulatory agencies for the standardization of herbal formulations. In this context, correct identification and quality assurance is an essential prerequisite to ensure reproducible quality of herbal medicine, which contributes to its safety and efficacy. Considering the importance of quality standardization and bio-prospecting of medicinal plants, the present study was conducted to compare different spp. of *Justicia* based on morphological, anatomical and phytochemical characters.

In the present study considerable differences were observed between species in their morphological, anatomical and phytochemical characters.

Morphological comparison revealed substantial variation in floral and vegetative features of different spp. of *Justicia*. Anatomical comparison of the petiole of *J. adhatoda* and *J. beddomei*, showed that the vascular bundle is 'V' shaped in *J. adhatoda* and 'U' shaped in *J. beddomei*. In both the species the epidermis is single layered and embedded with cells containing cystolith of calcium carbonate. Upper epidermal cells are narrow and rectangular in *J. beddomei* and Spongy cells are rounded in *J. beddomei* and tangentially elongated in *J. adhatoda*. Adaxial side of the midrib in *J. adhatoda* is more or less flat and wide. In *J. beddomei* the adaxial side is concave or notched. Epidermal cells of stem of *J. beddomei* contain cystoliths.

Anatomical comparison of the petiole revealed that T.S of petiole of *J. betonica*, and *J. gendarussa*, showed more or less similar characters in shape of midrib and arrangement of vascular bundles. The adaxial side of the petiole possess a central notch, which is narrow and deep in *J. betonica* and wide and shallow in *J. gendarussa*. The abaxial side is convex and it is deep convex in *J. gendarussa*. The vascular bundle, which is placed in the centre is 'U' shaped in with slight variation.

Adaxial side of the midrib in *J. betonica* is in the form of two convex, collenchymatous projections. In *J. gendarussa* the midrib is raised and convex in nature towards the adaxial side. The palisade cells are extended into the midrib and continuous in *J. gendarussa*. But in *J. betonica* the palisade cells do not form a continuous layer in the midrib region, but extend only upto the middle leaving a gap in the centre.

Anatomical comparison of roots of *J. betonica* and *J. gendarussa* show similarity due to the absence of a continuous ring of pericyclic fibres which is present in some other species of *Justicia* and the presence of air spaces in cortical region. But in the pericyclic region a few solitary fibres are present in *J. betonica* which is totally absent in *J. gendarussa*. Cortex of the stem is well differentiated into outer collenchymatous, middle chlorenchymatous and inner parenchymatous regions in *J. betonica* and *J. gendarussa*.

Anatomical comparison of the petiole of *Justiciasantapau*, *Justiciatrinervia* and *Justiciawynaadensis* revealed that many of the characters observed were unique to each taxon. The vascular bundle, which is placed in the centre is 'U' shaped in all the species with slight variations, i.e., the vascular bundle is wide 'U' in *J. santapau* with three bundles.

T.S of midrib portion of the leaf shows much variation in the shape especially on the adaxial side and in the arrangement of vascular bundles and palisade cells. In *J. santapau* the adaxial side of the midrib is projected but the top portion is flattened. The adaxial side of the midrib of *J. wynaadensis* shows similarity with that of *J. betonica*, but here the two projections are angular and appear like two horns. Comparative study of the T.S of the lamina of the selected species of *Justicia* leaf showed similarity in the anatomical characters with a few variations. In all the species both the epidermis are single layered and embedded with cells containing cystolith of calcium carbonate.

Anatomical comparison of roots of *Justicia* showed a wide range of variation with some exceptions of similarity among some of the species. *J. santapau* shows the presence of a thick and continuous ring of stone cells in the cortical region. *J. wynaadensis* stands apart

from other species due to the presence of wide cortex and scattered nature of solitary stone cells in the cortex. *J. trinervia* shows typical characters of a root.

Excellent separation of the components was achieved on high-performance precoated TLC plates by using optimized mobile phase consisting of toluene: ethyl acetate: methanol (8:2:1). Species specific and part specific marker compounds at specific R_f were identified in *J. adhatoda* and *J. beddomei*. Comparison of total phenolic and total flavonoid content between different parts of *J. adhatoda* and *J. beddomei* revealed that maximum TPC and TFC were in Leaf. Among the samples studied, highest TPC was observed for the leaf of *J. adhatoda* (38.75 mg GAE) followed by the leaf of *J. beddomei* (35.20 mg GAE). Highest TFC was equal in the leaves of *J. adhatoda* and *J. beddomei* (35.20 mg QE).

Excellent separation of the components was achieved on high-performance precoated TLC plates by using optimized mobile phase consisting of toluene: ethyl acetate: methanol (8:2:1). Detailed analysis and densitometric scanning performed under UV254 and 366 nm and after derivatisation using spray reagent revealed the presence of several unique bands.

In conclusion, the present study was successful in developing morphological, anatomical and phytochemical markers for identifying different spp. of *Justicia* even in the absence of flowers, fruits or any other conventional taxonomic identification characters. This will be of immense help in identifying genuine drug even in dried form. This can be used in the Ayurvedic drug industry to identify spurious drugs to improve the quality and efficacy of traditional healthcare system. Phytochemical similarity between different spp. of *Justicia* established based on HPTLC, TPC and TFC can be used as lead for substituting each other in the traditional health care system to reduce the harvesting pressure on a single raw drug.

OUTCOMES EMANATED FROM THE PROJECT

- Standardised morphological characters for the identification of different spp. of *Justicia* such as *J. adhatoda*, *J. beddomei*, *J. betonica*, *J. gendarussa*, *J. santapau*, *J. trinervia* and *J. wynaadensis*.
- Standardised Anatomical characters for the identification of different spp. of *Justicia* such as *J. adhatoda*, *J. beddomei*, *J. betonica*, *J. gendarussa*, *J. santapau*, *J. trinervia* and *J. wynaadensis*.
- Standardised Phytochemical characters for the identification of different spp. of *Justicia* such as *J. adhatoda*, *J. beddomei*, *J. betonica*, *J. gendarussa*, *J. santapau*, *J. trinervia* and *J. wynaadensis*.
- Analysed total phenolic and flavonoid contents in different spp. of *Justicia* such as *J. adhatoda*, *J. beddomei*, *J. betonica*, *J. gendarussa*, *J. santapau*, *J. trinervia* and *J. wynaadensis*.