## INTERNATIONAL JOURNAL OF ADVANCES IN PHARMACY, BIOLOGY AND CHEMISTRY

**Research Article** 

# A model quantitative ANNOVA test using spss software to find out the five morphological leaf traits of five different genera

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### ABSTRACT

Cost-effective alternative easy, accurate, inexpensive and non-destructive methods to determine individual leaf area of plants are a useful tool in agronomical and physiological studies. The morphological characterization of five different types of wild plants from Nirmala College for Women Campus of Coimbatore, Tamilnadu, southern India was carried out. This paper introduces to determine an individual five different morphological leaf traits, including leaf length, leaflet length, petiole length, number of leaflet in each leaf and number of branches in each leaf was measured in 200 plants. The ANOVA test was performed among the populations of each species as well as among species showed significant difference (p<0.05) for all quantitative characters used. Leaf morphological dissimilarities are possibly attributed to the genetic variations, developed as a result of adaptation to diverse environmental conditions. The important characteristics of this cost-efficient technique are its easiness and suitability for precise, non-destructive estimates. This model can be applied to other vegetative and reproductive any quantitative characters.

Keywords: Leaflet length, ANOVA test, wild plants.

#### **INTRODUCTION**

The crown architecture of plants is highly diverse, but it remains unclear how this diversity affects light interception and growth across species. Generally speaking, plant crowns are defined by the number, size, shape, three-dimensional distribution and orientation of their leaves. Together, these variables determine the size, the phyllotaxy (arrangement of leaves) ,length of leaflet and petiole when viewed from a given direction. Hence, leaves are important organs for photosynthesis and play an important role in survival and growth of a plant. As leaves adapt to a light gradient inside the canopy, not only the inclination angles but also the morphology, anatomy, size and mass of their petioles and laminas are modified.

Recent studies in plant traits have found that some relationships between specific leaf traits are globally repeated despite large variations in the values of the traits across individual species with very diverse phylogenetic, bio geographical and environmental affinities<sup>1,2</sup>. Therefore, such kinds of studies can be very useful in species with wide geographical ranges, for which the little information is available.

Flowering plants can have either simple or compound leaves. Leaflets originate on compound

leaves in a manner very similar to that of leaf lobes initiation in simple leaves<sup>3</sup>. Our main study objectives were to determine total and effective leaf and leaf clumping, and to evaluate the significance of variations in petiole length, leaf size and leaflet on stand

Light-harvesting efficiency in dense poplar plantations. Specifically, we hypothesize that, in stands with similar leaf, with longer petioles and larger leaves intercept light more efficiently than leave with shorter petioles and smaller leaves.

Leaf area can be measured either by destructive or non-destructive measurements. A common approach for non-destructive leaf area estimation is to develop ratios and regression estimators by using easily measured leaf parameters such as length and width<sup>4</sup>.

As the primary site for photosynthesis, leaves are of fundamental and functional important to green plants. Differences in morphological, anatomical and physiological characteristics of leaves along environmental gradients can be found across species<sup>5</sup>. In this study, we compared ecophysiological traits of leaves of five different *Justicia adhatoda L. Annona squamosa, Polyalthia longifolia, Punica granatum* and *Psidium guajava* distributed in different geographic regions with different climate pattern. Although recently identification of medicinal plants, especially *Justicia adhatoda L, Punica granatum* has attracted attention, in general there is limited information about medicinal plant populations and the studies performed mainly concern the identification of species by using morphological characters and numerical analysis has been performed to identify the intra and inter populations variation. This study attempt to evaluate genetic variation by morphological study of among and within five different plants were carried out.

### MATERIALS AND METHODS Study sites

The experiments were carried out in and around College Campus of Coimbatore, Tamilnadu, India. Nirmala College For Women Campus is situated Latitude: 11°1′6″N Longitude: 76°58′21″E. Five different leaf morphological traits, including leaf length, leaflet length, petiole length, number of leaves in each leaflet and number of branches in each leaflet of *Justicia adhatoda L. Annona squamosa, Polyalthia longifolia, Punica granatum* and *Psidium guajava* were selected.

Fifteen trees were selected randomly. The following data based on the five morphological trait levels: leaf length, leaflet length, petiole length, number of leaves in each leaflet and number of branches in each leaflet. Each experiment was replicated twenty times with one tree for each replicate.

### PLANT MATERIALS

- 1. Annona squamosa Linn, commonly known as Sugar apple, belonging to the family Annonaceae, is said to show varied medicinal effects, including insecticide, antiovulatory and abortifacient.
- 2. Punica granatum Linn, commonly known as the pomegranate is a popular fruit of tropical and subtropical regions, belonging to the family punicaceae. The potential include applications antioxidant, anticarcinogenic, and anti-inflammatory properties of pomegranate constituents have been published, focusing on treatment and prevention of cancer, cardiovascular disease, diabetes, dental conditions, erectile dysfunction, bacterial infections and antibiotic resistance, and ultraviolet radiation-induced skin damage
- 3. *Justicia adhatoda* Linn ,commonly known as *Adhatoda vasica* Nees,is a medicinal plant belonging to the family acanthcaeae .Vasicine and vasicinone is alkaloid strong

respiratory stimulant activity, hypotensive activity and cardiac-depressant effect.

- 4. Polyalthia longifolia Linn,commonly known as Ashoka tree (Saraca indica) is belonging to the family Annonaceae has showed many medicinal effects such as antimicrobial, hepatoprotective / antiinflammatory, antifungal, cytotoxic, anti-ulcer, antibacterial and hypotensive
- 5. *Psidium guajava* is commonly known as Guava a tropical fruit produced by the guava tree. The fruit, leaves, and juice are also used as medicine. People use guava for colic, diarrhea, diabetes, cough, cataracts, high cholesterol, heart disease, and cancer.

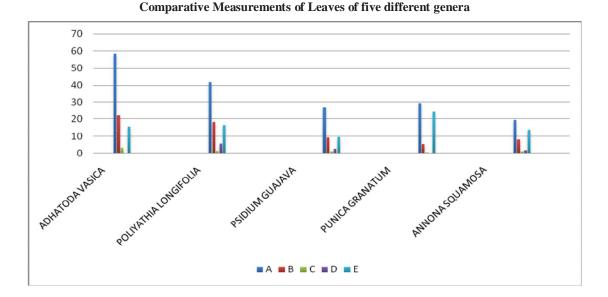
#### The Following Parameters Were Determined

- 1. Length of the Newly Developed leaflet (cm): Twenty leaflet of one year old in the four directions points (East- West- North and South) were tagged for measuring newly developed shoots length.
- 2. Number of Leaves per Shoot (cm): Leaves developed on the new shoots were also counted.
- **3.** Number of Leaflets per Shoot (cm) : Leaflet developed were counted on the new shoots.
- 4. Length of the New Developed petiole (cm): The length of the petiole was measured.
- 5. Length of the New Developed leaf (cm): The lengths of each leaf developed on the new shoots were counted.

### STATISTICAL ANALYSIS

In order to determine significant difference for quantitative morphological characters among populations and species studied, analysis of variance (ANOVA) followed by Least Significant Test (LSD) was performed. Species or populations considered as fixed factors without transformation. The statistical analysis of the data was carried out using the SPSS version 16.0 and the Stat Graphics Plus version 5.1 statistical packages. Analysis of variance (ANOVA) was used to test for differences between the different plant types and morphological parameters in relation to the leaf length were used as the variate. We used linear and non-linear regression analyses in the form y = a + bbx and y = a + b1x + b2x2 to test for statistical relationships between leaf morphological variables and different plant types .Regression fits and associated r2 and P values were given in each panel. All regressions were considered significant at P < 0.05.

#### **RESULTS AND DISCUSSIONS**



**Comparative Measurements of Leaves of five different genera** 

	Adhatoda vasica	Poliyathia longifolia	Psidium guajava	Punica granatum	Annona squamosa
А	$58.43\pm8.86^a$	$41.78 \pm 4.58^{a}$	$27.07 \pm 3.13^{a}$	$29.40 \pm 7.42^{a}$	$19.33 \pm 3.42^{a}$
В	$22.13 \pm 2.85^{b}$	$18.26 \pm 3.02^{b}$	$9.16\pm0.58^{\text{b}}$	$5.38\pm0.83^{\text{b}}$	$7.98 \pm 1.18^{\circ}$
С	$3.34 \pm 0.53^{d}$	$1.35 \pm 0.49^{d}$	$1.15\pm0.24^{\rm c}$	$0.50\pm0.00$	$1.30\pm0.18$
D	-	$5.75 \pm 0.79^{\circ}$	$2.65 \pm 0.49^{\circ}$	-	$1.65 \pm 0.49$
Е	$15.55 \pm 3.50^{\circ}$	$16.50 \pm 1.70^{b}$	$9.65 \pm 2.08^{b}$	$24.30\pm4.31^{a}$	$13.70 \pm 2.05^{b}$
CD (p<0.05)	6.15	3.84	2.49	5.23	2.35

A= Length of the Newly Developed leaflet; B= Length of the Newly Developed leaf; C= Length of the Newly Developed petiole; D= Number of Leaflets per Shoot; E= Number of Leaves per Shoot.

#### MORPHOLOGICAL TRAITS

Differences amongst natural populations of Justicia adhatoda L. Annona squamosa, Polyalthia longifolia, Punica granatum and Psidium guajava for all measured traits were remarkable. Adhatoda *vasica* has the largest leaf length (58.43cm), length of Leaf(22.13 cm) length of petiole(3.34 cm) and number of leaves(15.55 cm), while Annona squamosa showed the smallest leaf length, length of Leaf, number of leaflet (19.33 cm, 7.98cm, 1.65 cm respectively), but there was no leaflets shoot belonged to Adhatoda vasica and Punica granatum (Figure 1). This result can be explained by the stronger relationship between length of newly developed petiole and number of leaflets per shoots than between Length of leaflet and number of leaves (Table 2).

Adhatoda vasica and Punica granatum showed the highest and lowest values for Length of the New Developed leaf, that were  $22.13 \pm 2.85$  and  $5.38 \pm 0.83$  respectively (Figure 3f). Punica granatum and Psidium guajava had the maximum and

minimum values for Number of Leaves per Shoot of  $24.30 \pm 4.31$  and  $9.65 \pm 2.08$ , respectively (Figure 1).

## CORRELATIONS BETWEEN MORPHOLOGICAL TRAITS

Table 2 shows the correlations between morphological traits. As shown in this table, length of leaf was positively correlated with leaflet length and length of petiole (p < 0.05). However the relationship of leaf length and length of petiole with *Adhatoda vasica* was stronger than with *Annona squamosa* (Table 2). There was a high positive correlation between leaf length and petiole length (p < 0.05). Increasing the investment in petioles needs to synthesize more xylogens, and longer petioles will lead the leaf to bend<sup>6</sup>.

#### DISCUSSION

We found a high level of morphological variability among populations of different types of plants in Nirmala College for Women campus (Figure 3). Principal component analysis also indicates that there are several multivariate directions of variation in the morphological traits among the investigated populations.

The observed leaves differences may be attributed to different genetic architectures developed as a result of adaptation to diverse environmental conditions exiting in distributional area of of Justicia adhatoda L. Annona squamosa, Polyalthia longifolia, Punica granatum and Psidium guajava.Our sampling covered a narrow Latitude: 11°1'6"N Longitude: 76°58'21"E. ranges (Table 1), and the mean  $\pm$  SD didn't differ noticeably among the studied locations. Therefore, it can be said that the dissimilarities in leaves morphological traits of different plant types were unrelated to the latitude, longitude of the studied locations. Therefore, it seems that the dissimilarities in the amount and distribution of the precipitation in the studied locations are likely to influence transportable water by plants, which has to be transferred from stem to leaves and from leaves to atmosphere in the growing season, and induce variations in leaf characteristics<sup>7,8</sup>. Elongation of the leaf and petiole also shows a trade-off between resource acquisition and resource-conservative stress tolerance<sup>9</sup>.

In our study, we found a high level of morphological variability among populations of *Justicia adhatoda, Annona squamosa, Polyalthia longifolia, Punica granatum* and *Psidium guajava.* However, multisite common garden experiments would be needed in order to completely separate environmental and genetic factors explaining the observed level of natural variability.

#### CONCLUSIONS

Morphological leaf traits noticeably differ among the five natural populations of Justicia adhatoda L. Annona squamosa, Polyalthia longifolia, Punica granatum and Psidium guajava. Nirmala College For Women Campus of Coimbatore, Tamilnadu, India. The observed leaf differences may be attributed to different genetic architectures developed as a result of adaptation to diverse environmental condition. In general both measurable and categorical characters studied can differentiate plant species from each other and may be used in taxonomical study. In this paper, this method described was used to estimate individual leaf length, petiole length, leaflet length, number of leaves and leatlets of five different plant species. Therefore method can estimate precisely and in large quantities leaf area of plants in many

experimental comparisons without the use of costly instruments. However, we recognize that the limited geographical and phylogenetic scope in our research allows only a preliminary assessment of this expectation

### ACKNOWLEDGEMENTS

The authors are grateful to the Department of Botany, Nirmala College for Women, Affiliated to Bharathiar University, Coimbatore, Tamilnadu, for providing the co-operation and support. The authors would like to thank the euro informatics for the guidance in statistical analysis.

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