

## COMPARISON OF LEAF SIZE OF COMMON PLANTS BETWEEN FIELD MEASUREMENTS AND SCIENTIFIC FLORA RECORDS

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### Abstract

Plant trait data from scientific floras are frequently utilized in analyzing the geographical distribution and functional characteristics of plant species. Leaf area is an important functional trait of plants and can be estimated using the formula,  $Area = Length \times Width \times K$ . The length and width values are typically obtained from earlier records in scientific floras, and  $K$  represents the parameter. Since the data recorded in floras typically represents a range of values, estimating leaf area using this data requires making a reasonable choice. In this study, we concurrently collected data of accurately measured 1616 leaves from 39 common plants with entire or finely serrated leaves, including the leaf length, width, and area of each leaf. Furthermore, length and width of the studied plants was also extracted from the *Flora of China*. We conducted a comparative analysis of the two datasets and examined the potential use of the recorded information in floras for estimating leaf area. Additionally, we assessed the suitable value of parameter  $K$  to be employed in the formula. Our findings indicate that the data recorded in scientific floras are consistent with field measurements, and can be used to estimate leaf area. The present result also clears that the median values of leaf length and width from flora recordings provided the most accurate estimates of leaf area. Additionally, we determined that the appropriate value for the parameter  $K$  in the formula is  $\pi/4$  for leaves that are entire or finely serrated. This study provides insights into how data recorded in scientific floras can be applied in large-scale geographical studies of plant functional traits.

**Key words:** Flora; Estimate; Leaf size; Measure; Parameter  $K$

### Introduction

The variation of plant functional traits along large-scale geographic gradients have been studied increasingly in recent years (Violle *et al.*, 2014). The following studies have revealed several patterns of plant functional traits on macro geographical scales. For example, it was found that precipitation is a key factor affecting the height of trees at a global scale, with tropical trees reaching much greater height than those in temperate and cold zones (Moles *et al.*, 2009). Another pattern discovered was in the fruit types of plants, which displayed a distinct latitudinal distribution with a higher proportion of fleshy fruits in tropical regions and a higher proportion of dry fruits in colder zones (Tong *et al.*, 2021).

Leaf size refers to the one-sided projected surface area of a single leaf or an average leaf lamina, which exhibits a wide range of variation, spanning over 100,000-fold among plant species around the world (Cornelissen *et al.*, 2003; Wright *et al.*, 2017; Li & Wang, 2021). Leaf size is a critical functional trait for plants, with significant implications for leaf energy and water balance (Cornelissen *et al.*, 2003), as well as the photosynthesis, transpiration, and nutrient accumulation and transport of leaves (Xu *et al.*, 2009; Okajima *et al.*, 2012; Michaletz *et al.*, 2016; Wright *et al.*, 2017; Jin *et al.*, 2019; Lusk *et al.*, 2019; Conesa *et al.*, 2020; Li Y.Q. *et al.*, 2020a, Li Y.R. *et al.*, 2022). There are numerous methods to collect data of the leaf size for plants. The leaf size of one or a few plants can be investigated and measured in the field. Cristofori *et al.*, (2008) collected more than two thousand persimmon leaves in the central and southern regions of Italy and established a model to relate leaf area to leaf length and width for these leaves. The leaf size of plants can also be

measured using vast collections of leaves from specimens of plants stored in herbaria. Li *et al.*, (2020b) obtained valuable information on the geographic variation of functional traits of leaves for seven plant species in China by measuring the leaf length and width of over 6,000 specimens collected over the past 100 years.

Although researchers can conveniently access plant trait data from several online databases, such as the TRY database (Kattge *et al.*, 2011), GIFT database (Weigelt *et al.*, 2020), and Tundra Trait Team database (Bjorkman *et al.*, 2018), it can be very difficult to obtain leaf size data for all plants on large geographical scales (such as national, continental, or global scales) (Li *et al.*, 2020a). For some plants with very large leaves (such as palms), researchers can use the length of the rachis as a proxy for leaf area (Goldel *et al.*, 2015). For plants that have been recorded in the literature with a detailed morphological description, researchers often estimate leaf size by using the formula,  $Area = Length \times Width \times K$ , as the values of length and width could be obtained from these literature. This formula is derived from Montgomery's research on corn leaves, which demonstrated a positive proportionate relationship between leaf area and the product of leaf length and width (Montgomery, 1911). The parameter  $K$  typically varies in different studies, and has been reported to take on values such as 1/2, 2/3, 3/4, or  $\pi/4$  (Cooper, 1960; Wilf *et al.*, 1998; Wise *et al.*, 2000; McGlone *et al.*, 2009; Zhou *et al.*, 2019; Li *et al.*, 2020a; Cutts *et al.*, 2021; Luan *et al.*, 2021; Zhou *et al.*, 2022). For example, Zhou *et al.*, (2019) obtained information of functional traits on the vascular plants of Mount Kenya by referring to *Flora of Topical East Africa*. Li *et al.*, (2020a) obtained the length and width of plant leaves by referring to the *Flora of China* (FOC). They revealed that the changes in the average leaf size of the community were highly correlated with changes in climate and ecosystem primary productivity, and were not related to plant life forms (Li *et al.*, 2020a).

Most records of leaf length and width from scientific floras are reported as interval ranges, representing the minimum and maximum values of length or width. However, a number of recent studies have not taken into account these ranges and instead used rough approximations of the maximum (Zhou *et al.*, 2019; Li *et al.*, 2020a) or median (Cutts *et al.*, 2021) values. For instance, Zhou *et al.*, (2019) used the maximum values of leaf length and width values to calculate the leaf area of leaves. Cutts *et al.*, (2021), on the other hand, used the intermediate values of leaf length and width obtained from local floras to approximate the leaf area using a formula. These rough estimations may be somewhat different from the actual leaf area, which could lead to errors in testing ecological theories.

In this study, we collected the leaves of 39 common plants with entire or finely serrated margins of leaves and measured the length, width, and area of each leaf. In addition, we collected the range of leaf length and width for these plants from the FOC. We compared the differences in length, width, and area of leaves between the two data sets. Our goal was to determine whether the data recorded in FOC could be applied to estimate leaf area and to obtain a suitable parameter  $K$  for the estimation formula. Specifically, we aimed to address the following questions: (1) Are the leaf size data recorded in the floras consistent with actual field measurements? (2) Among the minimum, median, and maximum leaf area calculations, which value is closer to the actual measured area? (3) What is the appropriate value for the parameter  $K$  in the estimation formula?

## Materials and Methods

**Sample collection:** Healthy, mature leaves of common plants were collected from Meiling Mount in Nanchang, Jiangxi, China. The collected leaves had entire margins (e.g., *Cinnamomum camphora*) or serrate margins (e.g., *Euonymus centidens*), but were not shallowly or deeply lobed. For compound leaves (e.g., *Vitex negundo*), the largest leaflet was treated as a single leaf in our analysis (Zhou *et al.*, 2019). In total, 1616 leaves from 39 plants were collected and measured, with an average of approximately 40 leaves for each species.

**Measurement of leaf length, width and area:** Each leaf was scanned using a CanoScan LiDE 300 scanner (Canon, Tokyo, Japan) at a resolution of 300 dpi. The measurements for leaf length, width, and area were obtained from the scanned images of each leaf using Image-J software (Schneider *et al.*, 2012). The leaf length was defined as the distance from the base of the leaf blade to the tip, and the leaf width was defined as the maximum distance between two points on the leaf blade perpendicular to the line connecting the base and tip of the leaf.

**Leaf range data from floras:** We obtained the data of leaf length and width of each species were noted from FOC. The data of leaf length and width recorded in FOC were range data which shows the maximum and minimum values of leaf length and width, respectively. Additionally, we calculated the median value was calculated by using the maximum and minimum values of leaf length and width.

**Data analysis:** Pearson's correlation analysis was used to assess the consistency between the values of leaf length, width and area which collected in the field and the data recorded in FOC. The leaf length, width, and length  $\times$  width (representing area) recorded in FOC were divided into three groups based on their maximum, median, and minimum values, for comparative analysis. Linear regression and Wilcoxon test were used to explore the correlation and significance of differences between each group of data from our field measurements and FOC. All of these analyses were carried out using R 3.3.3 software (R Core Team, 2017).

## Results

**Comparison between the range of field-measured and flora-recorded data:** Our results revealed that the range of field-measured leaf length values for most plant species were not well-matched with the values recorded in FOC (Fig. 1). For some species, such as *Houttuynia cordata* and *Salix babylonica*, the field-measured leaf length values were clearly outside the recorded range in the floras (Fig. 1). Pearson correlation analysis showed a weak correlation between the range of field-measured leaf length and the recorded length range in FOC ( $cor = 0.37$ ,  $p < 0.05$ , Table 1). However, the ranges of field-measured leaf width and area values for most plants were consistent with those recorded in the floras (Figs. 2 and 3), especially for leaf area ( $cor = 0.70$ ,  $p < 0.001$ , Table 1).

**Correlation analysis and significance test between the mean values of field-measured and flora-recorded data:** The mean values of leaf length, width, and area obtained from field measurements were compared with the minimum, median, and maximum values of recorded data in FOC for each plant species. Linear regression analysis was conducted to examine the correlation between the median value of recorded data and the average value of field-measured data. The results indicated that the median value of recorded data had the strongest correlation with the average value of field-measured data, particularly for leaf width ( $r^2 = 0.75$ ,  $p < 0.001$ ) and area ( $r^2 = 0.68$ ,  $p < 0.001$ ) (Fig. 4). Significance tests revealed that only the median flora-recorded area ( $A_{mid}$ , calculated from median length  $\times$  median width) had no significant difference with field-measured leaf area ( $A_{field}$ ), whereas the minimum and maximum data did show significant differences ( $p = 0.26$ , Fig. 5).

**The determination of K:** We calculated the parameter  $K$  for each species using the formula,  $K = \text{Field-measured area}/\text{Flora-recorded leaf area}$ . The mean  $K$  values were then calculated for all the species, and we also compared the results using the minimum, median, and maximum values of length and width recorded in FOC. Our results showed that  $K$  calculated from the minimum values of records in FOC was 2.54, from the median values was 0.88, and from the maximum values was 0.49. Therefore,  $\pi/4$  (closer to 0.88 in our study) could be considered as the parameter  $K$  for an approximate calculation of leaf area from the values of length and width recorded in FOC.

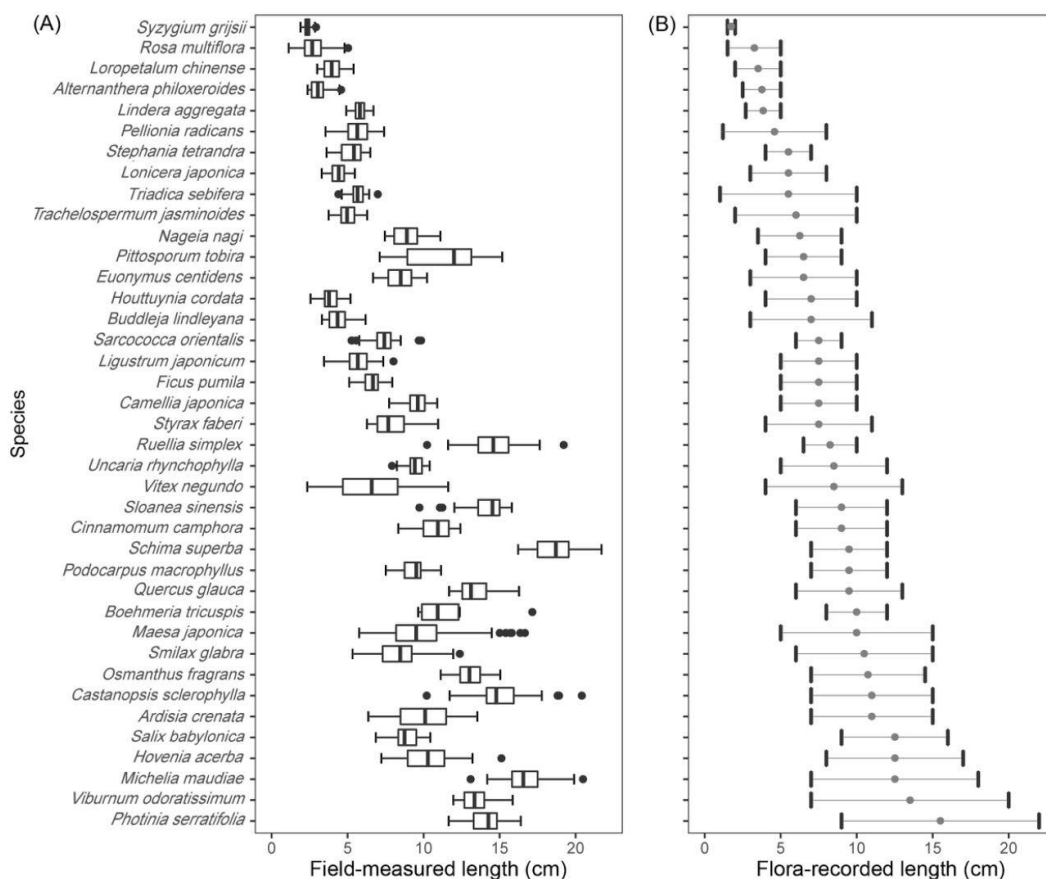


Fig. 1. The comparison between field-measured (A) and flora-recorded (B) leaf length ranges of 39 plants.

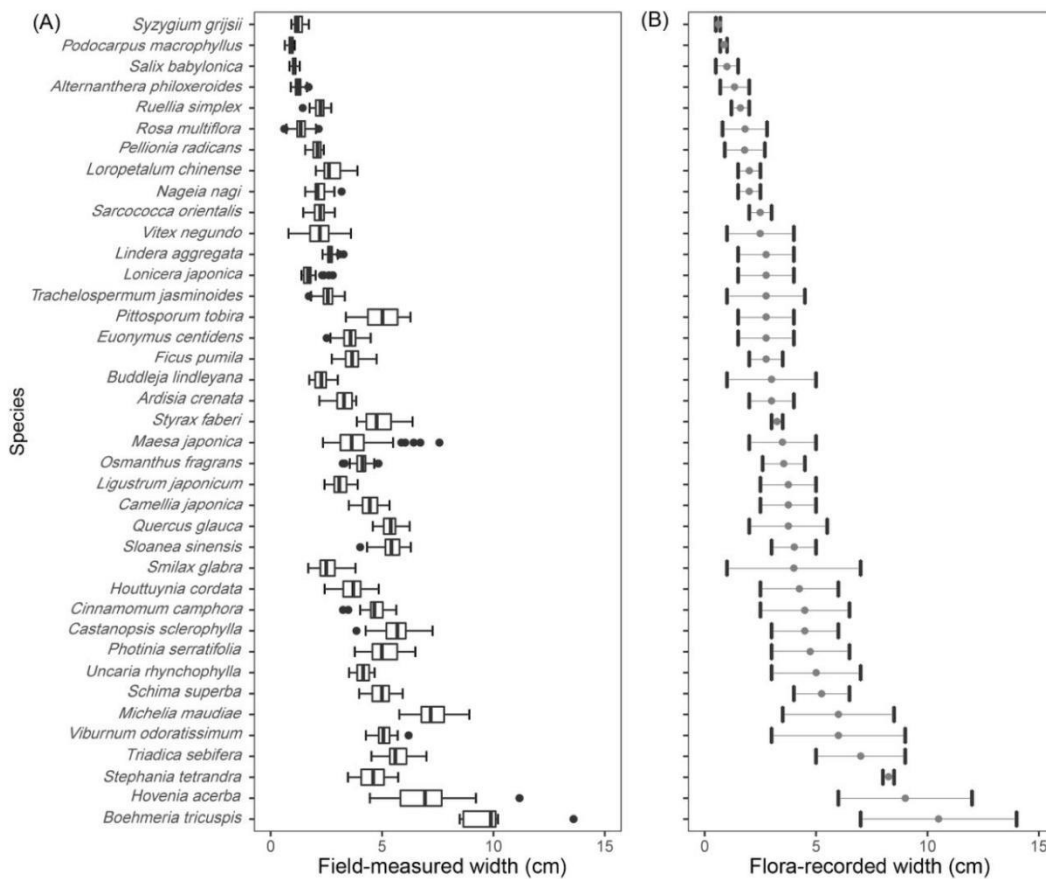


Fig. 2. The comparison between field-measured (A) and flora-recorded (B) leaf width ranges of 39 plants.

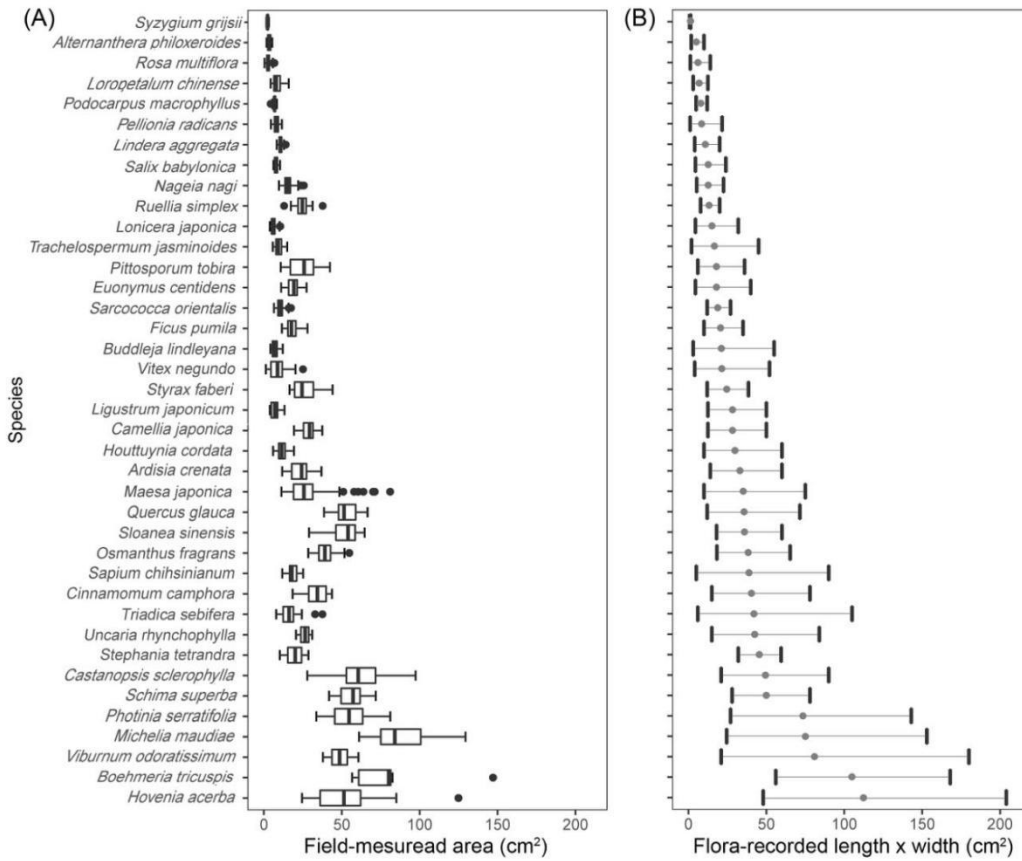


Fig. 3. The comparison between the field-measured leaf area (A) and flora-recorded leaf length × width (B) of 39 plants.

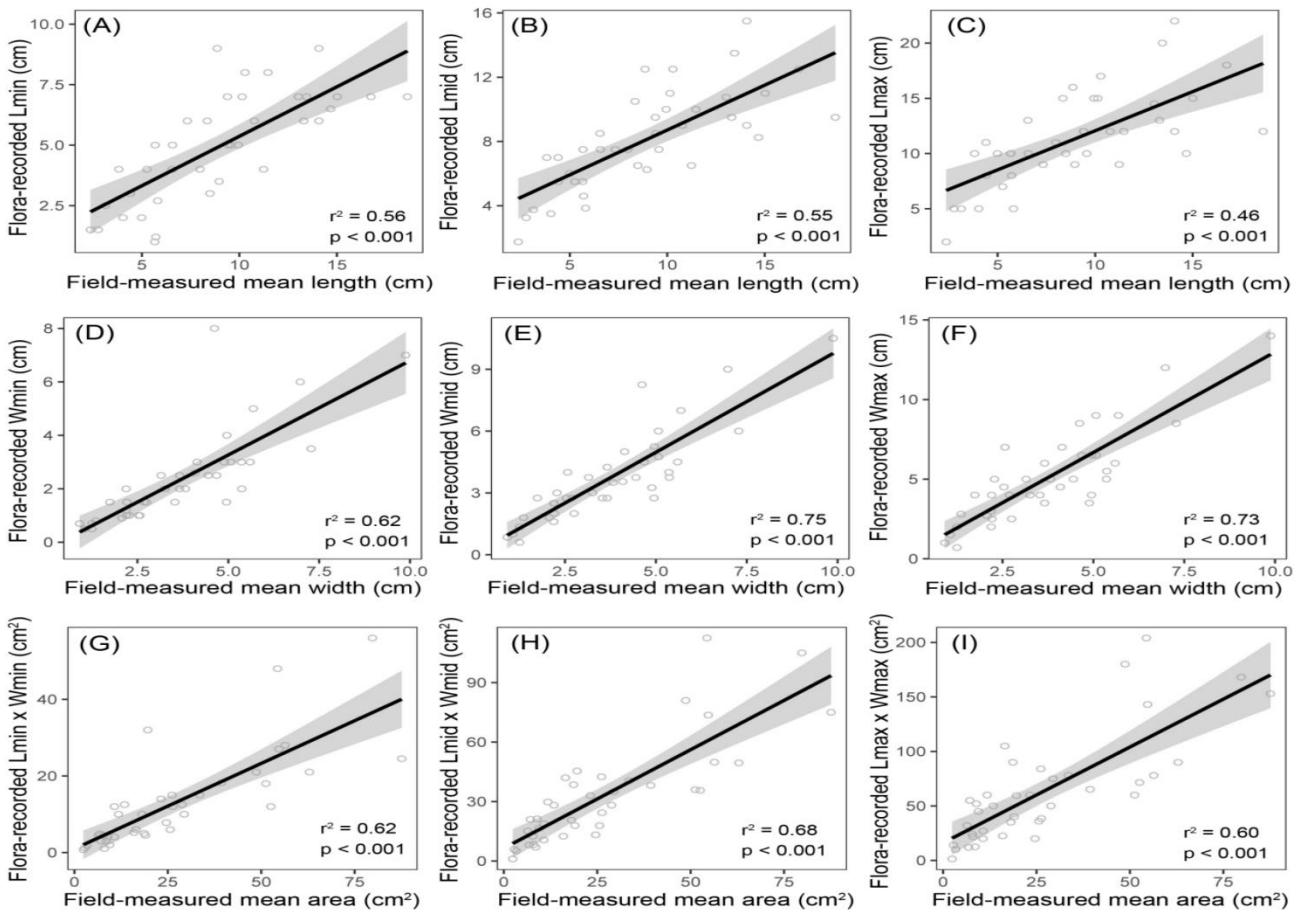


Fig. 4. Correlation analysis between the mean values of leaf length, width, and area of field-measured and flora-recorded data.

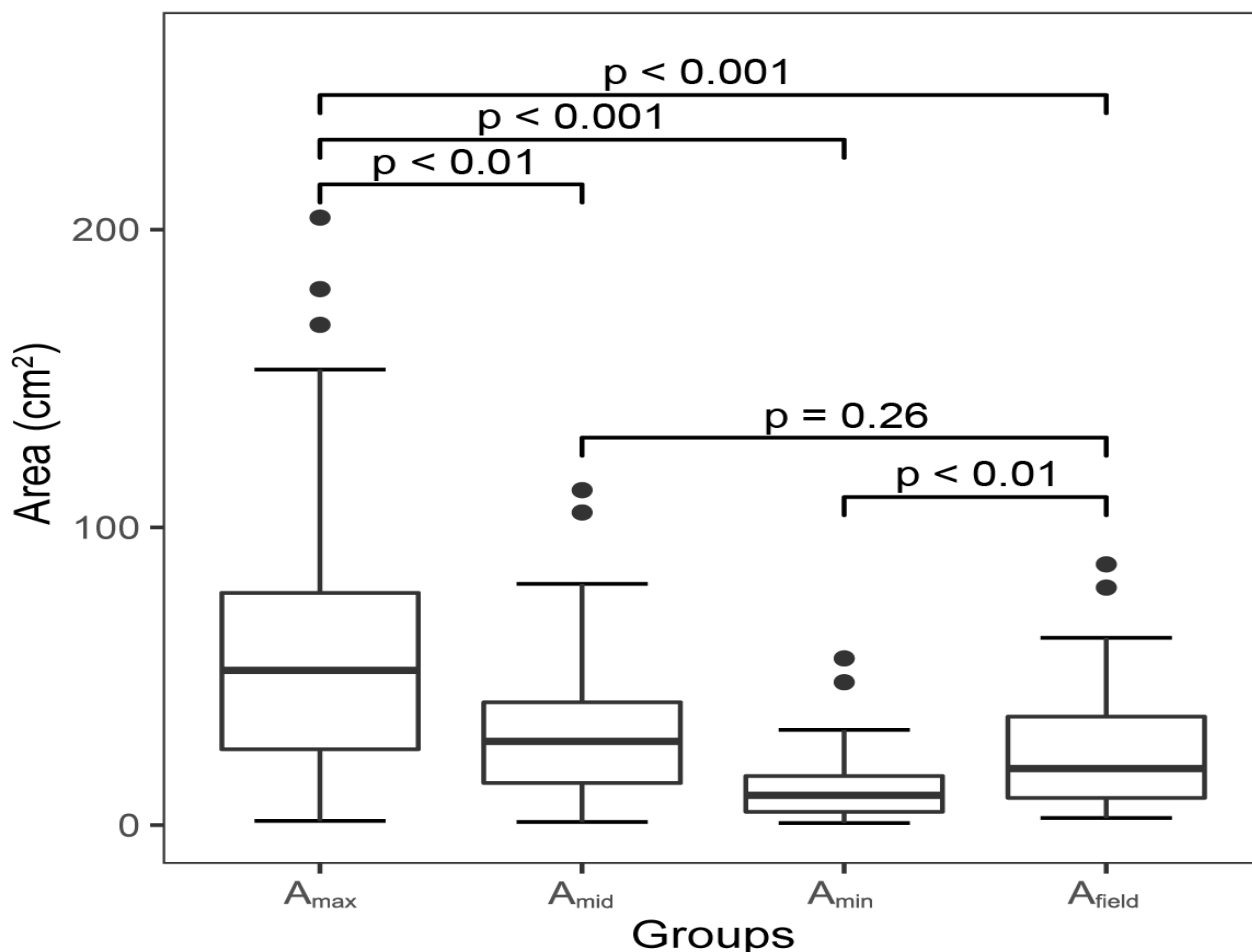


Fig. 5. Significance test of the difference between field-measured area and calculated area of flora-recorded data.  $A_{\max}$  represents the value calculated from the maximum leaf length times the maximum leaf width of flora-recorded data;  $A_{\text{mid}}$  represents the value calculated from the median leaf length times the median leaf width of flora-recorded data;  $A_{\min}$  represents the value calculated from the minimum leaf length times the minimum leaf width of flora-recorded data;  $A_{\text{field}}$  represents the field-measured leaf area. The  $p$ -value shows the significance between each two groups (Wilcoxon test).

**Table 1. Comparison between field-measured and flora-recorded ranges of leaf length, width and area.**

Models	t	df	cor	p-value
Field-measured leaf length range ~ Flora-recorded leaf length range	2.44	37	0.37	< 0.05
Field-measured leaf width range ~ Flora-recorded leaf width range	4.27	37	0.57	< 0.001
Field-measured leaf area range ~ Flora-recorded leaf length $\times$ width range	5.96	37	0.70	< 0.001

## Discussion

Scientific floras record the ranges of leaf length and width, which are essential information for morphological description of plants (Cutts *et al.*, 2021). These data are widely used to investigate the functional trait variation of leaves at a large scale (McGlone *et al.*, 2009; Zhou *et al.*, 2019; Li *et al.*, 2020a; Cutts *et al.*, 2021; Zhou *et al.*, 2022). We conducted field measurements of leaf length, width, and area of the 1616 leaves from 39 common plants, and compared these values with those recorded in FOC. Our results indicated that the variation ranges of most field-measured leaf range data are consistent with the ranges of the flora-recorded data, particularly for width and area. Furthermore, we found that there is no significant difference between the leaf area calculated by multiplying the median values of leaf length and width in the floras and the field-measured leaf area. We also determined that  $\pi/4$

is the most accurate parameter for estimating the leaf area of entire leaves based on data recorded in the floras.

The size of leaves significantly varies among interspecific and intraspecific plants (Michaletz *et al.*, 2016; Wright *et al.*, 2017; Li *et al.*, 2020b). Even leaves within the individual plant can exhibit considerable variation in size. By documenting the intraspecific variation in leaf size across a specific region, scientific floras can provide a rich source of data, while simultaneously reducing the impact of local-scale processes (Traiser *et al.*, 2005; Li *et al.*, 2016; Cutts *et al.*, 2021). Our findings indicate that there is a positive correlation between the variation in leaf length, width, and area obtained from field-measured data and the variation derived from data recorded in scientific floras, with the correlation being particularly strong for leaf width and area. This suggests that the accuracy of leaf width data recorded in scientific floras is higher than that of leaf length data. Previous studies also demonstrated that during the

evolution of broad-leaved plants, leaf width tends to be more conservative than leaf length, with variation in leaf area being primarily driven by changes in leaf length (Shi *et al.*, 2018, 2019; Su *et al.*, 2019). Furthermore, a comprehensive analysis of measurement data from numerous specimens of six plant species across China revealed that neither their leaf width nor their leaf area exhibited significant changes in response to variations in mean annual temperature (Li *et al.*, 2020b).

The availability of vast amounts of plant functional trait data is crucial for studying plant functional biogeography, particularly with respect to understanding the large-scale spatial distribution patterns and ecological drivers of key plant traits (Li & Wang, 2023). Our findings provide further confirmation that, following careful extraction, local scientific floras containing ample trait data are a reliable source for ecological analysis (Du *et al.*, 2020; Wang *et al.*, 2020; Cutts *et al.*, 2021). In fact, the functional leaf traits recorded in the floras were compiled by researchers who based their work on extensive investigations, specimen collections, and literature (Raven *et al.*, 2013). When utilizing the abundant trait data recorded in scientific floras, many studies fail to verify whether these data correspond well with actual field-measured data. Our results showed that the median value of leaf length times the median value of leaf width has the highest correlation with the actual field-measured leaf area, and the difference significance test also showed that there is no significant difference between the field-measured leaf area and the calculated leaf area value from median value of leaf length times the median value of width. These results indicate that, using the median value of leaf length and width recorded in the floras to estimate the leaf area of plants has high accuracy and certain reference value.

When compared with the destructive methods, non-destructive methods always rely on field-measured leaf length and width, and employ formula (e.g.,  $A = c \times L \times W$ , where  $A$  is leaf area,  $L$  is length,  $W$  is width, and  $c$  is the parameter) to calculate leaf area (Montgomery, 1911; Mokhtarpour *et al.*, 2010; Yu *et al.*, 2020; Schrader *et al.*, 2021). The correction factor  $c$  of the formula was reported to range from 0.39 for highly dissected, lobed leaves to 0.79 for oblate leaves (Schrader *et al.*, 2021). The similar formula ( $Area = Length \times Width \times K$ ) was also commonly employed to roughly estimate leaf area based on leaf length and width data recorded in the literature. Our study found that a value of  $\pi/4$  could be used as the  $K$  value in the leaf area estimation formula for entire (or finely serrated) leaves (Wise *et al.*, 2000; Luan *et al.*, 2021), instead of  $1/2$ ,  $2/3$ , or  $3/4$ , as used in previous studies (Cooper *et al.*, 1960; Wilf *et al.*, 1998; McGlone *et al.*, 2009; Zhou *et al.*, 2019; Li *et al.*, 2020a, b; Cutts *et al.*, 2021; Zhou *et al.*, 2022). This may be attributed to the blade of an entire leaf closely resembling an ellipse, and the area formula for an ellipse being  $S = (a/2) \times (b/2) \times \pi$ , which can be converted to  $S = a \times b \times \pi/4$ , where  $S$  is the area,  $a$  and  $b$  are length and width of the ellipse, respectively (Yu *et al.*, 2020; Schrader *et al.*, 2021).

In summary, we investigated the differences between field-measured and flora-recorded leaf size data and explored the feasibility of using data recorded in floras for estimating leaf area. We also evaluated the most reasonable parameter  $K$  in the formula for calculating leaf area. Our results demonstrate that the data recorded in scientific floras are consistent with actual field measurements and that it is possible to estimate leaf area using the flora

records. This study provides new insights for the application of plant functional trait data recorded in scientific floras for large-scale biogeographical studies. While our research only focused on the leaves with full-edge blade, and further investigation could be conducted on the leaf area parameter of leaves with other blade types, such as shallow-split and deep-split blades.

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