BIODIVERSITAS ISSN: 1412-033X
Volume 25, Number 11, November 2024 E-ISSN: 2085-4722
Pages: 4244-4252 DOI: 10.13057/biodiv/d251124

Leaflet morphometric variation of *Elatostema integrifolium*, an Anoa forage plant, in Abdul Latief Forest Park, South Sulawesi, Indonesia

ISNA RASDIANAH AZIZ[1,♥], ADE MUSPA[2], CUT MUTHIADIN[1], HAJRAH[1], DEVI ARMITA[1], MASRIANY[1], ZULKARNAIN[1], SYARIF HIDAYAT AMRULLAH[1], ASWAR RUSTAM[1], DIRHAMZAH[1], MUHAMMAD KHALIFAH MUSTAMI[3], DADAN SUPARDAN[4], DAVID SEGURA-SEQUEIRA[5,♥]

[1]Department of Biology, Faculty of Science and Technology, Universitas Islam Negeri Alauddin Makassar. Jl. H.M. Yasin Limpo No. 36, Romangpolong, Gowa 92113, South Sulawesi, Indonesia. Tel./fax.: +62-411-1500363, ♥email: isna-rasdianah@uin-alauddin.ac.id

[2]Department of Biology, Faculty of Science and Technology, Universitas Sipatokkong Mambo. Jl. dr. Wahidin Sudirohusodo No. 75, Bone 92733, South Sulawesi, Indonesia

[3]Department of Biology Education, Faculty of Tarbiyah and Teacher Training, Universitas Islam Negeri Alauddin Makassar. Jl. H.M. Yasin Limpo No. 36, Romangpolong, Gowa 92118, South Sulawesi, Indonesia

[4]Department of Madrasah Ibtidaiyah Teacher Education, Faculty of Tarbiyah, Institut Agama Islam Negeri Curup. Jl. Dr. AK Gani No. 01, North Curup, Rejang Lebong 39119, Bengkulu, Indonesia

[5]Las Arrieras Nature Reserve. Heredia Province, Sarapiqui, Horquetas 3000, Costa Rica, Central America

Manuscript received: 29 June 2024. Revision accepted: 16 November 2024.

Abstract. *Aziz IR, Muspa A, Muthiadin C, Hajrah, Armita D, Masriany, Zulkarnain, Amrullah SH, Rustam A, Dirhamzah, Mustami MK, Supardan D, Segura-Sequeira D. 2024. Leaflet morphometric variation of* Elatostema integrifolium*, an Anoa forage plant, in Abdul Latief Forest Park, South Sulawesi, Indonesia. Biodiversitas 25: 4244-4252.* Accurately identifying plant taxa is crucial for conservation efforts, and leaf characteristics play a vital role in this process. *Elatostema integrifolium* (D.Don) Wedd., a forage plant and primary food source for Anoa (*Bubalus* spp.), is a potential candidate for differentiation based on leaflet morphology. This study aimed to quantify the variation in morphological characteristics of *E. integrifolium* leaflets, investigate the distribution patterns of these traits, and assess the degree of morphological similarity between different locations in Abdul Latief Forest Park, South Sulawesi, Indonesia. The study employed a systematic sampling across three designated stations (Station I, Station II, and Station III) representing varying management functions within the park. Forty plants were collected from each station (n = 3), and 11 morphometric traits of their leaflets were analyzed. Univariate analysis was performed using SPSS ver. 29, with whisker-box plots generated for data visualization. Multivariate analysis, implemented in MATLAB® ver 9.14, incorporated a two-pronged approach. Principal Component Analysis (PCA) was initially conducted using a subset of seven key morphological traits identified during exploratory analysis. Results revealed that variation within stations was significant for all the scored leaflet morphological traits. The distribution of morphological profiles of *E. integrifolium* across the investigated sites exhibits minimal segregation. Hierarchical cluster analysis based on morphological traits revealed a closer relationship between the leaflet characters observed in Station I and Station II, compared to their similarity with Station III. It is projected to enhance the precision of plant identification and taxonomy, deepen our comprehension of plant adaptation, and ultimately guide the formulation of focused conservation strategies for *E. integrifolium* populations.

Keywords: Cluster analysis, *Katiloporo*, morphology character, principal component analysis, understory forage

INTRODUCTION

Sulawesi, an Indonesian island within the Wallacea biodiversity hotspot, serves as a critical ecological transition zone between the Asian and Australian continents, harboring a wealth of endemic flora and fauna (BBKSDA 2023; Pusparini et al. 2023). *Elatostema integrifolium* (D.Don) Wedd. (local name: *Katiloporo*), a perennial herbaceous or sub-shrub species within the family Urticaceae characterized by its entire leaves (Triyutthachai et al. 2022; Xin et al. 2023), is a forage species and vital food source for the Anoa (*Bubalus* spp.), a Sulawesi-endemic ungulate classified as Endangered under the IUCN Red List (Burton et al. 2016; BBKSDA 2023). Mingkid et al. (2023) reported that Genus *Elatostema* is an abundant natural forage for Anoa in North Sulawesi and has a dominant presence in Central Sulawesi (Saleh and Hartana 2017). Similarly, this species is documented as the dominant food source for Anoa in Southeast Sulawesi (Broto 2015) and South Sulawesi (Aziz et al. 2023). This plant thrives in tropical-subtropical regions, primarily in Asia, in habitats with warm temperatures, high humidity, and shady forest floors beneath closed canopies, typically grows in acidic soil, and is often found near riverbanks (Aziz et al. 2023; Mingkid et al. 2023). The presence of *Elatostema* within forest ecosystems would contribute to the development of a healthy understory layer, thereby enhancing the ecological functionality of the habitat (Sakio 2020; Mingkid et al. 2023).

Beyond their role in plant sustenance, leaves possess crucial characteristics for accurate plant taxon identification. Quantitative leaf characteristics, such as leaf area, petiole length, and vein patterns, alongside qualitative traits like leaf shape, margin, and texture, play a significant role in refining and strengthening plant identification methods (Larese et al. 2014; Manivannan et al. 2016;

Temam et al. 2021; Du et al. 2022). However, field-based species identification using leaves can be challenging due to several factors. Morphological variations within a single species, caused by genetic diversity (Coates et al. 2018) and environmental pressures (Scales and Butler 2016), can lead to overlapping characteristics between individuals (Forsman and Wennersten 2016). Additionally, similarities between characters or dual natures in different species can further complicate identification, especially for closely related taxa (Gabaldón et al. 2017; Korshunova et al. 2019). Finally, the use of incomplete leaf samples due to limitations in field collection (Carranza-Rojas et al. 2017), feature extraction, or damage (Lee et al. 2017) can significantly limit the available information for accurate identification. Furthermore, variations in leaflet characteristics across diverse geographic locations underscore the phenomenon of local adaptation and phenotypic plasticity (Fan et al. 2020; Yao et al. 2021), leading to the development of traits that enhance their survival and fitness within their specific environments.

Despite prior studies by Fu et al. (2014, 2019, 2021), Yin et al. (2021) in China, Rajapaksha et al. (2019) in Sri Lanka, Rodda and Monro (2018) in Malaysia, Lin et al. (2014) in Vietnam, and Triyutthachai et al. (2022) in Thailand of morphological features of *Elatostema* sp. in tropical regions, research on morphometric variations using leaflets is currently lacking. Studies have primarily utilized principal component analysis and cluster analysis in *Elatostema* sp. for specific purposes, such as assessing forest floor vegetation composition (Tsai et al. 2018), investigating metal accumulation and phytoextraction potential (Yang et al. 2014), and documenting their use as food sources by indigenous communities (Upadhyay et al. 2021).

Abdul Latif Forest Park, a conservation area spanning approximately 720 hectares in Sinjai District, South Sulawesi Province, faces the dual challenge of maintaining ecological integrity while serving as a public resource for education and tourism. This delicate balance is particularly critical for the endangered Anoa, whose survival is intertwined with the availability of its primary food source, *E. integrifolium*. The park's heterogeneous topography and diverse habitat mosaic likely contribute to quantitative and qualitative variations in *E. integrifolium* leaflet traits, potentially leading to localized adaptations within specific ecological niches. These variations may have significant implications for Anoa foraging behavior and, ultimately, their survival within the forest park. Limited knowledge regarding the morphological characteristics of *E. integrifolium* within the Abdul Latief Forest Park represents a gap that necessitates further research. The objective of this study was to determine the diversity of morphological characters in *E. integrifolium* leaflets, analyze their distribution patterns, and assess the proximity of these characters across three stations within the Abdul Latif Forest Park. We anticipate that the findings will contribute to improved accuracy in plant identification and taxonomy, enhance our understanding of plant adaptation, and ultimately inform the development of targeted conservation strategies for *E. integrifolium* populations.

MATERIALS AND METHODS

Study area

The present research divided the investigation area within Abdul Latief Forest Park, South Sulawesi, Indonesia, a known hotspot for Anoa (*Bubalus* spp.) visitation, into three distinct stations. The three observation stations were strategically chosen to represent distinct management blocks within the Forest Park (Figure 1). Station I is situated in the protection block, Station II is located in the utilization block, and Station III is situated in the collection block. The block designation is to align with the guidelines outlined in Government Regulation Number 34 of 2002 (BPHN 2002).

Plant sampling

In each designated study area, 40 individuals of *E. integrifolium* were sampled from a total of 120 individuals, yielding a total of 300 leaflets for subsequent analysis, which was chosen based on stringent criteria, including full leaf development, absence of insect infestation, and disease. For herbarium preparation, leaves were individually pressed between sheets of newspaper. This press was then exposed to sunlight for desiccation over six days, with daily newspaper changes to ensure proper drying (Soladoye et al. 2010). The resulting herbarium specimens were labelled comprehensively for taxonomic purposes. Morphological characterization focused on the right leaflet within the first pair of central leaves (Soladoye et al. 2010; Brus et al. 2011) on each chosen *E. integrifolium*. A total of 11 quantifiable morphological characteristics were identified and systematically analyzed using phenetic methods. ImageJ ver. 2.0 was utilized to measure leaf area, as depicted in Figure 2.

A suite of environmental variables was measured in situ across the three study stations to understand the ecological niches of *E. integrifolium*. These data, presented in Table 1, offer valuable insights into the potential environmental factors influencing the growth and distribution of this species.

Table 1. Characteristic of three study stations in the Abdul Latief Forest Park, South Sulawesi, Indonesia

Characters of habitat	Population name		
	Station I protection block	Station II utilization block	Station III collection block
Latitude	05°18' S	05°18' S	05°18' S
Longitude	120°00' E	120°00' E	120°00' E
Altitude (masl)	1,576	1,430	1,544
Temperature (°C)	17.3	22	18.2
Noise (dB)	47.02	41.89	39.21
Soil pH	5.9	6.2	6.1
Soil moisture (cm Hg)	5.93	6.2	7.16
Relative humidity (%)	88.1	73.7	78.4
Wind velocity (ms^{-1})	0.3	0.1	0.6

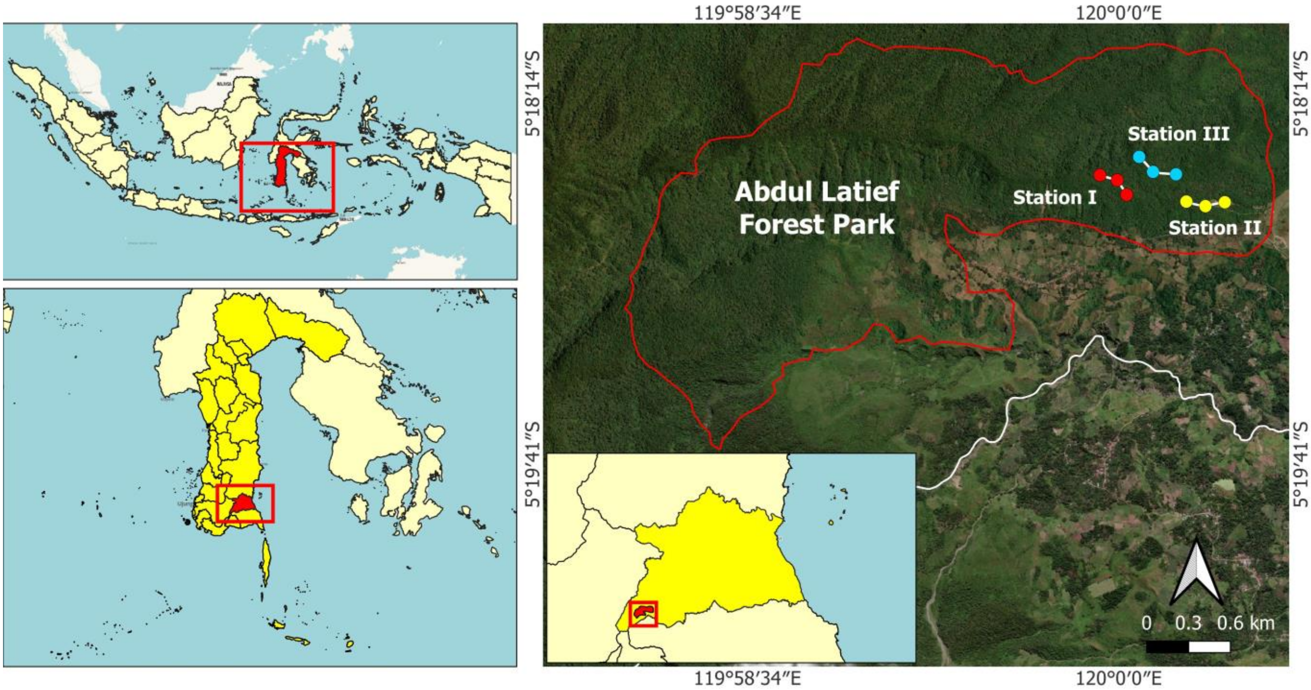

Figure 1. Map of study location in Abdul Latief Forest Park, South Sulawesi, Indonesia (5°18'30.0"S 120°00'00"E) and three sampling stations: Station I in protection block (red dot); Station II in utilization block (yellow dot); Station III collection in block (blue dot)

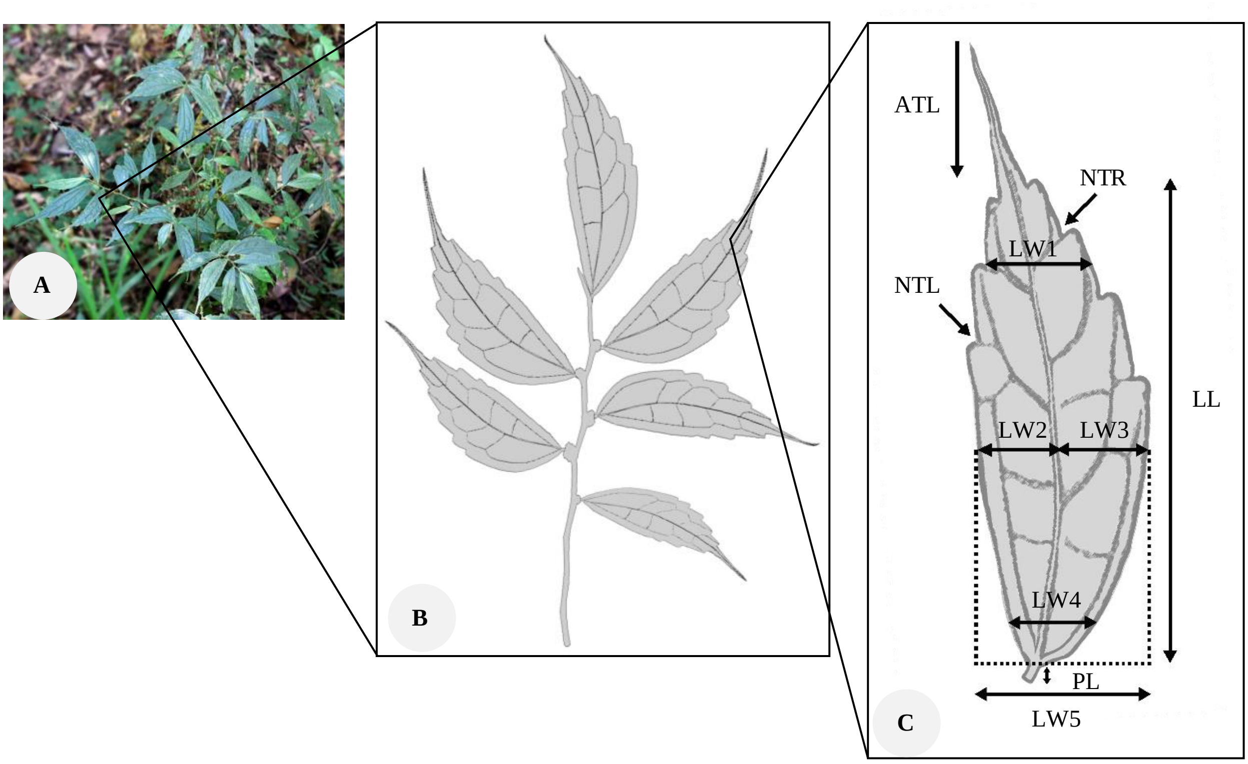

Figure 2. *Elatostema integrifolium* selected in Abdul Latief Forest Park, South Sulawesi, Indonesia: A. Morphological features; B. Leaf with regularly arranged leaflets; C. Scored morphological traits for leaflets including ATL (Apex Tile-Like; tail-like projection at the apex), LL (Leaflet Length), LW1 (Leaflet Width measured 1 cm beneath the tail-like projection at the apex), LW2 (Leaflet Width measured at half of the leaflet length from central to the left leaf margin), LW3 (Leaflet Width measured at half of the leaflet length from central to the right leaf margin), LW4 (Leaflet Width measured 0.5 cm above the leaflet base), LW5 (Leaflet Width or LW2+LW3), NTL (total of teeth on the left leaflet side), NTR (total of teeth on the right leaflet side), LL/LW5 (Leaflet Length/Leaflet Width), and PL (Petiolate Leaflet length measured beneath at the base of the leaflet)

Data analysis

Phenotypic analysis was conducted using both univariate (Soladoye et al. 2010; Sharma and Pandit 2011) and multivariate statistical methods (Brus et al. 2011). For the univariate analysis, quantitative measurements were analyzed using descriptive statistics calculated in SPSS ver. 29, including arithmetic means ($\bar{x}$), Standard Deviations (SD), and Coefficients of Variation (CV). To visualize the range of variation for each scored morphological character, whisker-box plots were generated. Statistically significant differences were evaluated using Tukey's Honestly Significant Difference (HSD) test. The multivariate analysis utilized a sequential approach implemented in MATLAB® ver 9.14. All 11 morphological traits from the specimens were included in Principal Component Analysis (PCA). Notably, each specimen collected from the three stations was treated as an Operational Taxonomic Unit (OTU) for Euclidean distance analyses.

RESULTS AND DISCUSSION

Marked variations were observed across the 11 measured morphological characters in *E. integrifolium* leaflets collected from the three stations. Table 2 shows that the Coefficient of Variation (CV) for LW1 exhibited the greatest degree of variation, ranging from 26.89% to a remarkable 184.42% within the Station II (utilization block). Conversely, leaflet length displayed the least variation across all three stations (protection: 9.88%, collection: 10.71%, utilization: 10.88%). The three lowest average values (<1 cm) were also observed for LW1, PL, and LW4 across all three stations.

Station III consistently exhibited the highest median values and the greatest variation in the majority of leaflet morphological character measurements (LL, NTL, NTR, LW1, LW3, LW4, ATL, PL). This observation suggests that leaflets from Station III tend to be larger and more variable in size compared to those from the other two stations. Interestingly, the character traits of Station II often displayed intermediate values between those of Station I and Station III. This pattern suggests that Station II may represent a transitional zone between the two extremes (Figure 3).

Following the measurement of 11 quantitative morphological characters (Table 2), a data filtering process was implemented to identify and retain only those characters exhibiting statistically significant values. This refined dataset, containing seven traits, was then subjected to correlation matrix analysis (Table 3). Principal Component Analysis (PCA) was conducted on this dataset, resulting in the extraction of three principal components (Comp. 1, Comp. 2, and Comp. 3) that collectively explained 76.81% of the total variance. These components effectively differentiated the characters across the three investigated blocks. The first Principal Component (PC1) accounted for 35.09% of the explained variance and highlighted two particularly important taxonomic characters: LW3 and LW4. These characters exhibited high loadings of 0.861 and 0.812, respectively. PC2, explaining 26.35% of the variance, emphasized the significance of ATL and PL for taxonomic differentiation. Finally, PC3, contributing the least variance (15.36%), identified a single significant taxonomic character leaflet width measured at half of the leaflet length from central to the left leaf margin.

Table 3. Loadings of principal components analysis for seven quantitative traits of *Elatostema integrifolium*

Quantitative traits	Loadings of PCA		
	Comp.1	**Comp.2**	**Comp.3**
ATL	0.181	0.843	0.188
LW2	-0.473	0.076	0.811
LW3	0.861	0.137	0.310
LW4	0.812	-0.166	0.401
NTL	-0.609	0.521	0.048
NTR	0.651	0.432	-0.339
PL	-0.080	0.790	-0.093
Initial eigenvalues	2.457	1.845	1.075
Proportion of variance	35.09	26.35	15.36
Cumulative proportion	35.09	61.45	76.81

Table 2. Statistical parameters (mean, SD, CV) of 11 quantitative leaflet traits for *Elatostema integrifolium*

Quantitative traits	Station I Protection block			Station II Utilization block			Station III Collection block		
	$\bar{x}$	SD	CV	$\bar{x}$	SD	CV	$\bar{x}$	SD	CV
ATL	2.19	0.24	10.93	2.29	0.32	13.84	2.40	0.24	10.39
LL	10.68	1.06	9.88	11.09	1.21	10.88	11.70	1.25	10.71
LW1	0.23	0.09	37.76	0.41	0.75	184.42	0.26	0.07	26.89
LW2	1.46	0.20	13.87	1.50	0.19	12.81	1.59	0.35	22.08
LW3	1.39	0.21	15.32	1.44	0.22	15.31	1.54	0.19	12.32
LW4	0.94	0.18	18.62	0.91	0.10	11.23	0.99	0.16	16.11
LW5	2.85	0.29	10.33	2.94	0.23	8.08	3.13	0.37	11.67
NTL	3.88	0.81	3.81	4.00	0.88	21.93	4.50	0.85	18.89
NTR	3.81	0.91	23.88	4.00	0.88	21.93	4.20	1.14	27.03
LL/LW5	3.77	0.40	10.71	3.78	0.39	10.40	3.78	0.51	13.52
PL	0.369	0.11	29.23	0.36	0.12	33.37	0.40	0.08	20.41

Note: $\bar{x}$: mean (cm); SD: Standard Deviation (cm); CV: Coefficient of Variation (%)

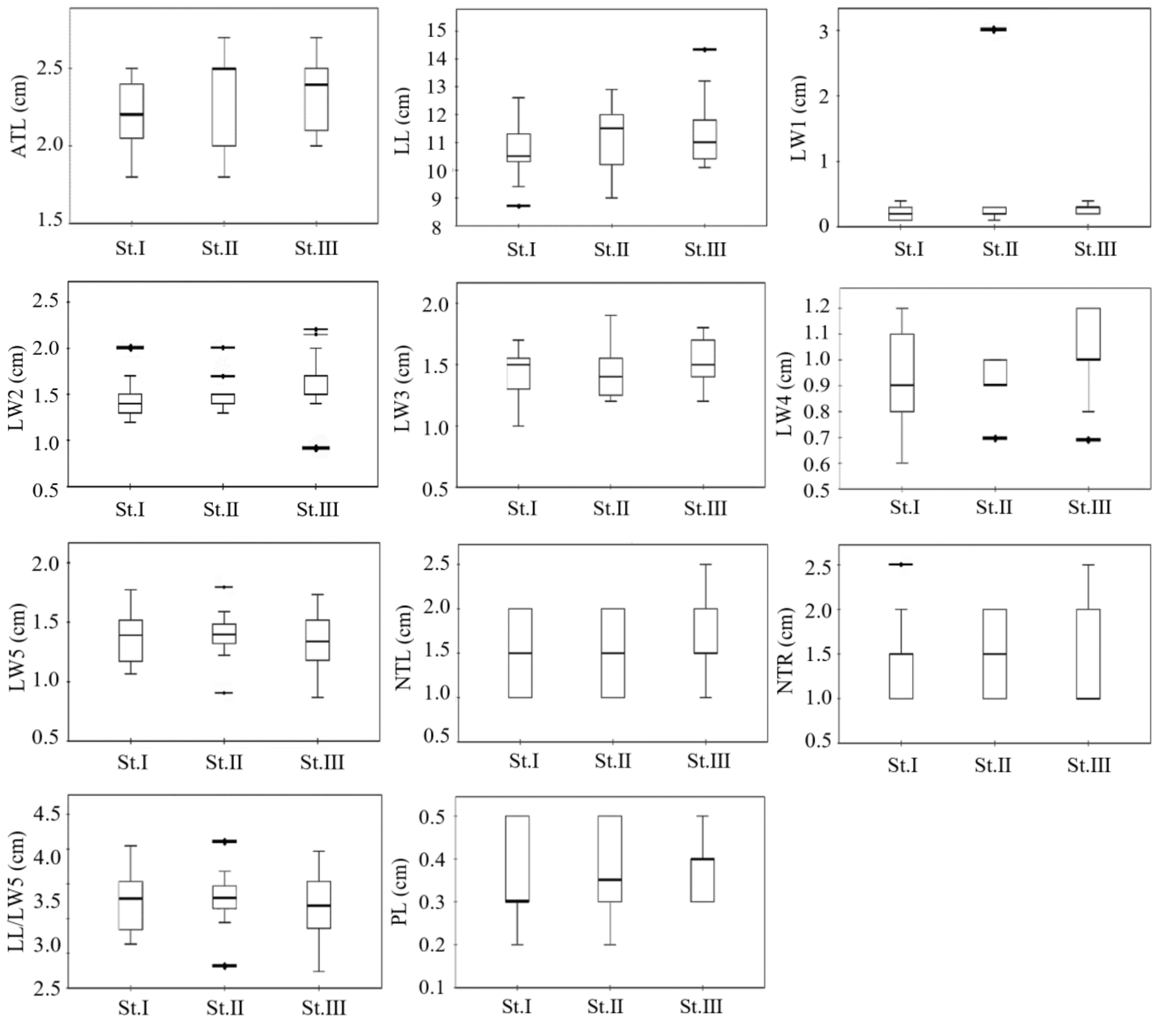

Figure 3. Range variation of *Elatostema integrifolium* leaflet represented by box plots of 11 traits included in PCA: box = interquartile range; thick horizontal line within box = median; line upper box = uppermost value; line lower box = lowermost value; circle with a thin line outside the box = outlier; circle with a thick line outside the box = more than two outliers; St. = Station

Figure 4 depicts the outcomes of PCA, visualizing the distribution of individuals from each station (Station I, Station II, and Station III) based on key morphological characters (NTL, NTR, LW2, LW3, LW4, ATL, and PL). The PCA scatterplot reveals a high degree of overlap among the stations in terms of these characters.

Hierarchical cluster analysis based on eleven quantitative morphological traits revealed a closer relationship between the leaflet characters observed in Station I and Station II compared to their similarity with Station III. This clustering pattern is evident from the dendrogram, where the branch lengths connecting Station I and Station II are shorter than the branch lengths connecting them to Station III (Figure 5).

Discussion

The environmental heterogeneity observed across the three study stations within Abdul Latif Forest Park offers a foundation for ecological investigations (Table 1). Located at lower altitude, Station II experiences notably higher average temperatures compared to Station I and Station III; these temperatures differential suggests that *E. integrifolium* exhibiting a broader thermal tolerance spectrum may be more likely to thrive across all three sites. These findings align with previous research by Fauzan et al. (2018) and Chen et al. (2023), which reported the successful growth of *Elatostema* in regions with average annual temperatures ranging from 16.4℃ to 23.8℃. Moreover, soil pH and moisture levels may influence nutrient availability, ultimately affecting the growth and survival of *E. integrifolium*. The proximity of Station I and Station II to nearby rivers, in contrast to Station III, likely contributes to similar soil conditions at these two locations. Despite its distance from a direct water source, substantial tree canopy at Station III contributes to soil moisture retention. The dense vegetation provides shade, thereby reducing evaporation and enhancing moisture within the soil (Yang et al. 2015; da Silva et al. 2021). The relative humidity levels recorded at the three stations, ranging from 73.7% to 88.1%, are slightly higher than those reported by Broto (2015) in Southeast Sulawesi. However, these conditions are generally conducive to the growth of *E. integrifolium*, underscoring the importance of our research in understanding plant ecology.

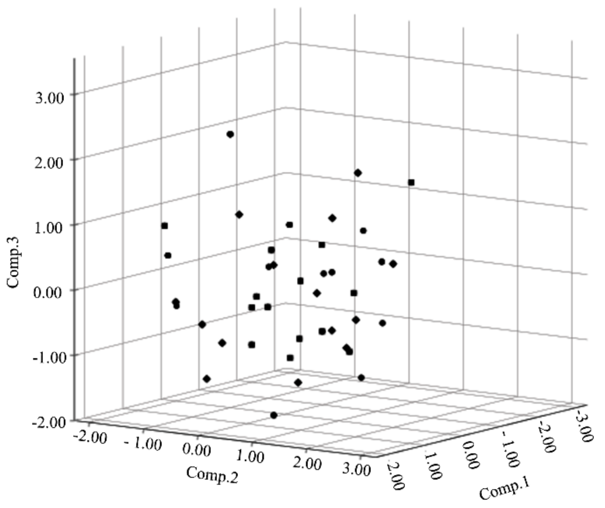

Figure 4. A Scatterplot of three components of PCA was formed, representing three populations of *Elatostema integrifolium*: Station I (triangle), Station II (square), Station III (circle)

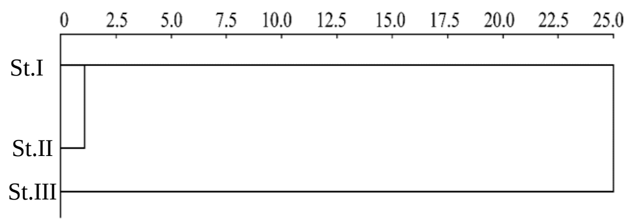

Figure 5. Cluster analysis of three stations represented by phenogram using eleven quantitative traits. St. = Station

The *E. integrifolium* population within the park exhibits robust growth, with average leaflet dimensions exceeding 11.70 cm in length and 3.13 cm in width, which qualifies them as macro-leaflets. The leaflets display an oval morphology with a slight asymmetry and a glabrous surface. The venation pattern is predominantly rectilinear and parallel, with the presence of both secondary and tertiary veins exhibiting a subtle slant towards the leaflet margins, characteristic of pinnate-parallel venation. Additionally, the tail-like projection at the apex demonstrates variation in length, with the average maximum size reaching approximately 2.4 cm, corresponding to roughly 20% of the total leaflet length (Figure 2, Table 2).

Statistical analyses demonstrate significant variations across 11 quantitative traits of *E. integrifolium* specimens collected from the three defined stations (Table 2). These findings suggest a strong influence of environmental heterogeneity on leaflet growth and morphology within the population. However, it is important to consider that these variations may not be solely attributable to factors such as light availability and temperature. As Station II experiences higher average temperatures and potentially increased light absorption, noise levels may be indirectly correlated with these factors due to increased human activity in this area. This is supported by the observation that Station II functions as a utilization block with restricted public access for ecotourism and educational purposes. Therefore, noise levels could serve as an indicator of human-induced environmental disturbance, potentially influencing *E. integrifolium* growth and morphology and indirectly affecting Anoa foraging behavior. However, it is crucial to acknowledge that establishing a direct causal link between noise and leaflet morphology might be challenging, and noise should be considered alongside other influential factors such as light availability, temperature, and competition. LW1, a measurement taken 1 cm below the apical tail-like projection, exhibited the greatest degree of variation among the examined traits. This variation appears to be largely attributable to differences in the width of serrations on the leaflet margins. Discrepancies in the distances between consecutive teeth on either side of the leaflet directly impact overall leaflet width. This observed pattern suggests that resource allocation within the plant prioritizes both maximizing leaf surface area and protecting the serrated margins, likely in response to factors such as light availability and temperature. Elevated temperatures recorded at Station II are hypothesized to influence the intensity of light absorption by the leaves, consequently promoting the development of wider teeth. This enhanced light capture may explain the observed wider leaflets with more prominent teeth at this location. These findings support observations by Baumgartner et al. (2020) who documented substantial variations in tooth morphology within *Vitis aestivalis* Michx. populations subjected to differing temperature and rainfall regimes. The observed morphological shift towards wider leaves with protruding teeth represents a potential adaptation, particularly shade-tolerant understory forage. The broader leaf surface area facilitates the interception of a greater quantity of photons, enhancing the potential for light capture (Niinemets et al. 2015; Onoda et al. 2017). Additionally, the presence of protruding teeth may function as light traps, scattering and redirecting incident light toward the leaf surface (Berry and Goldsmith 2020), thereby maximizing the utilization of even obliquely angled light for photosynthesis (Maslova et al. 2021). These morphological features, in synergy, could demonstrably enhance the plant's capacity for light capture (Schrader et al. 2021), potentially leading to increased photosynthetic efficiency (Li et al. 2022). As a consequence, this adaptation may promote the growth and survival of shade-adapted understory plants by facilitating a more robust energy production through photosynthesis in environments characterized by limited sunlight availability (Rossatto et al. 2018). Conversely, Leaflet Length (LL) displayed the least variation across the three stations, with a consistent coefficient of variation of approximately 10%. The consistently low coefficient of variation observed for leaflet length across the three study stations suggests a degree of phenotypic uniformity in this trait. Given the relative isolation of these populations compared to external regions, the prevalence of herbivorous pressures within the study area is likely elevated. As an adaptive strategy to deter herbivore attacks, including those from Anoa, *E. integrifolium* may have evolved to produce uniformly sized leaflet lengths. Within the context of this study, leaflet width exhibits no correlation with leaflet length.

Specimens from Station III exhibited greater morphological variation compared to those from the other two stations. Notably, Station III functions as a collection block, where biophysical conditions, for instance, the type and density of plants in this area, the nutrient content, and the wide area, are managed to support both natural plant populations and those artificially planted for research and local plant cultivation. This co-existence of natural and cultivated plants within the same habitat likely contributes to the observed morphological diversity in Station III. The emergence of such variations is likely a complex phenomenon influenced by various factors, potentially including plant physiological adaptations that manifest as changes in leaf morphology (Liu et al. 2019). Furthermore, the presence of a river stream traversing Station III provides a vital source for pollinating insects and birds. The small, inconspicuous greenish or yellowish-green flowers arranged in cymose inflorescences are likely more susceptible to cross-pollination by this fauna. This facilitation of cross-pollination could promote genetic exchange between natural and artificially grown plants through gene flow (Kuo et al. 2021), potentially leading to offspring with novel trait combinations (Capó et al. 2023). By contrast, Station II appears to occupy an intermediate ecological niche between Station I and Station III. This is evident from the observed morphological characteristics of *E. integrifolium* at each station, as well as the measured environmental parameters. Notably, Station II, a utilization block with restricted public access for ecotourism and educational purposes, exhibits values that fall between those observed at the other two stations. These observed discrepancies in environmental conditions across the stations are likely to be the driving force behind the distinct phenotypic shifts observed in the plant populations at each location.

Our study in PCA provided valuable insights into the sources of morphological variation among *E. integrifolium* specimens collected from the three stations (Table 3). The analysis revealed that leaflet width measurements (LW3 and LW4), along with ATL and PL characteristics, were the key factors differentiating the specimens across the study sites. While PC3 explained a smaller proportion of the variance, LW2 also emerged as a potential contributor to morphological differentiation. These findings suggest that leaflet width may be a particularly important morphological trait for *E. integrifolium* in this region.

It is also noteworthy that *E. integrifolium* leaflets are a food source for Anoa (Aziz et al. 2023), which exhibit feeding behavior adjustments based on encountered leaves. Wider leaves may necessitate more effort and specific techniques for Anoa to handle, while narrower leaves might be easier to manage. Anoa utilize their long, flexible tongues to reach and grasp *E. integrifolium* leaflet petioles, wrapping their tongue around the leaves before pulling them into their mouths. Leaf width likely influences the ease of grasping and the force required for Anoa to pluck the leaflet. Additionally, while Anoa's role in pollination is minimal, they can incidentally ingest and disperse this species' seeds through their feces (Mustari 2020), potentially contributing to the establishment of new individuals in different locations. Variations in leaflet width may also represent an adaptation and defence mechanism against herbivory in general (Caldwell et al. 2016), alongside environmental factors like water availability, light, temperature, and soil pH (Viani et al. 2014; Apgaua et al. 2015; Wang et al. 2019, Tserej and Feeley 2021). Smaller and narrower leaflets could indicate a higher abundance of herbivores in three stations, including Anoa, within a particular habitat. While the distribution of individuals across the three stations was analyzed to identify potential differentiating morphological characteristics, the employed scatter plot visualization did not reveal distinct separation between the stations. The set of characters chosen for analysis may have needed to be sufficiently discriminatory, potentially leading to a pattern where these characters tend to exhibit intermediate values across all stations rather than concentrating in specific locations. Furthermore, limitations in the number of samples collected in the field might have contributed to the need for more resolution in the scatter plots (Figure 4). Reduced sample size can hinder the detection of subtle morphological variations between individuals, potentially causing them to appear intermingled in the visualizations.

Euclidean distance analysis provided further insights into the relationships between stations. Station I and Station II exhibited the closest relationship (distance = 2.274), while Station I and Station III (distance = 4.418) and Station II and Station III (distance = 4.161) were more distant. This suggests that the morphological traits in Station II possess transitional traits, with a greater similarity to Station I compared to Station III. Variations and modifications observed in *E. integrifolium* leaflet likely arise from a complex interplay of genetic, environmental, and evolutionary factors. This combined influence fosters remarkable diversity and ongoing phenotypic change within plant populations. Supporting this notion, McKown et al. (2014) demonstrated a strong correlation between *Populus trichocarpa* Torr. & A.Gray ex Hook. traits, population structure, and geographic climate, which influence the species' genotypic and phenotypic diversity. The remarkable diversity of leaf shapes arises not only from their developmental context but also from the dynamic modulation of genetic regulatory activity (Chitwood and Sinha 2016; Maugarny-Calès and Laufs 2018). This modulation can occur throughout development, in response to environmental pressures (Bar and Ori 2014), or even during the evolutionary process across species (Vlad et al. 2014).

To ensure the long-term sustainability of *E. integrifolium* populations, accurate identification is crucial. While leaflet morphology can be helpful, its limitations necessitate a comprehensive approach that includes other identification criteria and molecular techniques. This is especially important given the increasing challenges posed by global climate change. Rising temperatures in the species' habitat can induce drought conditions, which are detrimental to its preferred moist and acidic soil environments. Monitoring changes in leaf morphology can serve as an early warning system for climate change or habitat disturbances like deforestation. This data can be used to develop effective conservation strategies for protecting local plant populations within these forests. Effective forest management requires collaboration among stakeholders such as managers, relevant agencies, researchers, and the local community. This collaborative approach is critical for ensuring the protection and preservation of this species as a valuable element of South Sulawesi's Forest biodiversity. We conclude that the observed patterns of morphometric variation in *E. integrifolium* leaflets, particularly the distinct differences in selected morphological characters, highlight the potential for further molecular research. This knowledge can significantly contribute to ongoing efforts to conserve local genetic resources.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the guidance and collaboration provided by the management division of the Abdul Latif Forest Park and the Environment and Forestry Service of Sinjai District, South Sulawesi, Indonesia. Universitas Islam Negeri Alauddin, Makassar, Indonesia supported this research.

REFERENCES

Apgaua DMG, Ishida FY, Tng DYP, Laidlaw MJ, Santos RM, Rumman R, Eamus D, Holtum JAM, Laurance SGW. 2015. Functional traits and water transport strategies in lowland tropical rainforest trees. PLoS One 10 (6): e0130799. DOI: 10.1371/journal.pone.0130799.

Aziz IR, Hasyimuddin H, Muspa A, Masriany M, Zulkarnain Z, Mustami MK. 2023. Identification, abundance and diversity of forage plants for Anoa (*Bubalus* spp.) in Abdul Latief Forest Park. Acta Ecol Sin 43 (1): 202-208. DOI: 10.1016/j.chnaes.2022.09.008.

Bar M, Ori N. 2014. Leaf development and morphogenesis. Development 141 (22): 4219-4230. DOI: 10.1242/dev.106195.

Baumgartner A, Donahoo M, Chitwood DH, Peppe DJ. 2020. The influences of environmental change and development on leaf shape in *Vitis*. Am J Bot 107 (4): 676-688. DOI: 10.1002/ajb2.1460.

BBKSDA. 2023. A Breath of Fresh Air in the Rescue Efforts of the Mountain Anoa in South Sulawesi. Natural Resources Conservation Center (BBKSDA) South Sulawesi, Ministry of Environmental and Forestry, Makassar, Indonesia. https://ksdasulsel.menlhk.go.id/. [Indonesian]

Berry ZC, Goldsmith GR. 2020. Diffuse light and wetting differentially affect tropical tree leaf photosynthesis. New Phytol 225 (1): 143-153. DOI: 10.1111/nph.16121.

BPHN. 2002. Government regulation of the Republic of Indonesia (PP) number 34 year 2002 (34/2002). National Legal Development Agency (BPHN), Ministry of Law and Human Rights, Jakarta, Indonesia. https://bphn.go.id/. [Indonesian]

Broto BW. 2015. The vegetation structure and composition of Anoa (*Bubalus* spp.) habitat in Mekongga Mountains Protected Forest, Kolaka, Southeast Sulawesi. Pros Sem Nas Masy Biodiv Indon 1 (3): 615-620. DOI: 10.13057/psnmbi/m010339. [Indonesian]

Brus R, Ballian D, Bogunić F, Bobinac M, IdžOjtić M. 2011. Leaflet morphometric variation of service tree (*Sorbus domestica* L.) in the Balkan Peninsula. Plant Biosyst 145 (2): 278-285. DOI: 10.1080/11263504.2010.549660.

Burton J, Wheeler P, Mustari A. 2016. *Bubalus quarlesi*: The IUCN Red List of Threatened Species 2016: e. T3128A46364433. https://www.iucnredlist.org/fr/species/3128/46364433.

Caldwell E, Read J, Sanson GD. 2016. Which leaf mechanical traits correlate with insect herbivory among feeding guilds? Ann Bot 117 (2): 349-361. DOI: 10.1093/aob/mcv178.

Capó M, Borràs J, Perelló-Suau S, Rita J, Cursach J. 2023. Phenotype–fitness relationships and pollen-transfer efficiency of five orchid species with different pollination strategies. Am J Bot 110 (6): e16198. DOI: 10.1002/ajb2.16198.

Carranza-Rojas J, Goeau H, Bonnet P, Mata-Montero E, Joly A. 2017. Going deeper in the automated identification of Herbarium specimens. BMC Evol Biol 17 (1): 181. DOI: 10.1186/s12862-017-1014-z.

Chen M, Mo F, Zheng L, Bin G, Zou Z, Chen P, Xue Y. 2023. Correlation and community stability analysis of herbaceous plants in Dashiwei Tiankeng Group, China. Forests 14 (6): 1244. DOI: 10.3390/f14061244.

Chitwood DH, Sinha NR. 2016. Evolutionary and environmental forces sculpting leaf development. Curr Biol 26 (7): R297-R306. DOI: 10.1016/j.cub.2016.02.033.

Coates DJ, Byrne M, Moritz C. 2018. Genetic diversity and conservation units: Dealing with the species-population continuum in the age of genomics. Front Ecol Evol 6: 165. DOI: 10.3389/fevo.2018.00165.

da Silva IAG, Dubeux Jr. JCB, Santos MVF, de Mello ACL, Cunha MV, Apolinário VX, de Freitas EV. 2021. Tree canopy management affects dynamics of herbaceous vegetation and soil moisture in silvopasture systems using arboreal legumes. Agronomy 11 (8): 1509. DOI: 10.3390/agronomy11081509.

Du J, Li B, Lu X, Yang X, Guo X, Zhao C. 2022. Quantitative phenotyping and evaluation for lettuce leaves of multiple semantic components. Plant Methods 18: 54. DOI: 10.1186/s13007-022-00890-2.

Fan X, Yan X, Qian C, Bachir DG, Yin X, Sun P, Ma X-F. 2020. Leaf size variations in a dominant desert shrub, *Reaumuria soongarica*, adapted to heterogeneous environments. Ecol Evol 10 (18): 10076-10094. DOI: 10.1002/ece3.6668.

Fauzan DM, Suleman S, Pitopang R. 2018. Autecology of *Impatiens mamasensis* Utami & Wiriad in the Tongoa Resort Area of Lore Lindu National Park, Central Sulawesi. Biocelebes 12 (2): 1-16. [Indonesian]

Forsman A, Wennersten L. 2016. Inter-individual variation promotes ecological success of populations and species: Evidence from experimental and comparative studies. Ecography 39 (7): 630-648. DOI: 10.1111/ecog.01357.

Fu L-F, Do VT, Wen F, He C-X. 2014. *Elatostema arcuatobracteatum* (Urticaceae), a new species from Vietnam. Phytotaxa 174 (2): 111-115. DOI: 10.11646/phytotaxa.174.2.6.

Fu L-F, Monro AK, Yang T, Wen F, Pan B, Xin Z-B, Zhang Z-X, Wei Y-G. 2021. *Elatostema qinzhouense* (Urticaceae), a new species from limestone karst in Guangxi, China. PeerJ 9: e11148. DOI: 10.7717/peerj.11148.

Fu L-F, Xin Z-B, Monro AK, Wen F, Li S, Wei Y-G. 2019. Supplementary descriptions of inflorescence and achene of two species of *Elatostema* (Urticaceae) from China. Taiwania 64 (2): 198-201. DOI: 10.6165/tai.2019.64.198.

Gabaldón C, Fontaneto D, Carmona MJ, Montero-Pau J, Serra M. 2017. Ecological differentiation in cryptic rotifer species: What we can learn from the *Brachionus plicatilis* complex. Hydrobiologia 796: 7-18. DOI: 10.1007/s10750-016-2723-9.

Korshunova T, Picton B, Furfaro G, Mariottini P, Pontes M, Prkić J, Fletcher K, Malmberg K, Lundin K, Martynov A. 2019. Multilevel fine-scale diversity challenges the 'cryptic species' concept. Sci Rep 9 (1): 6732. DOI: 10.1038/s41598-019-42297-5.

Kuo B-J, Jhong Y-S, Yiu T-J, Su Y-C, Lin W-S. 2021. Bootstrap simulations for evaluating the model estimation of the extent of cross-pollination in maize at the field-scale level. PLoS One 16 (5): e0249700. DOI: 10.1371/journal.pone.0249700.

Larese MG, Namías R, Craviotto RM, Arango MR, Gallo C, Granitto PM. 2014. Automatic classification of legumes using leaf vein image features. Pattern Recognit 47 (1): 158-168. DOI: 10.1016/j.patcog.2013.06.012.

Lee SH, Chan CS, Mayo SJ, Remagnino P. 2017. How deep learning extracts and learns leaf features for plant classification. Pattern Recognit 71: 1-13. DOI: 10.1016/j.patcog.2017.05.015.

Li Q, Wen J, Zhao C-Z, Zhao L-C, Ke D. 2022. The relationship between the main leaf traits and photosynthetic physiological characteristics of Phragmites australis under different habitats of a salt marsh in Qinwangchuan, China. AoB Plants 14 (6): plac054. DOI: 10.1093/aobpla/plac054.

Lin Y, Duan LD, Bi HY. 2014. A new variety and two new records of *Elatostema* J.R. Forster & G. Forster (Urticaceae) from Vietnam. Bangladesh J Plant Taxon 21: 27-32. DOI: 10.3329/bjpt.v21i1.19254.

Liu C, Li Y, Xu L, Chen Z, He N. 2019. Variation in leaf morphological, stomatal, and anatomical traits and their relationships in temperate and subtropical forests. Sci Rep 9 (1): 5803. DOI: 10.1038/s41598-019-42335-2.

Manivannan A, Anandakumar CR, Ushakumari R, Dahiya GS. 2016. Characterization of Indian clusterbean (*Cyamopsis tetragonoloba* (L.) Taub.) genotypes using qualitative morphological traits. Genet Resour Crop Evol 63: 483-493. DOI: 10.1007/s10722-015-0266-y.

Maslova NP, Karasev EV, Xu S-L, Spicer RA, Liu X-Y, Kodrul TM, Spicer TEV, Jin J-H. 2021. Variations in morphological and epidermal features of shade and sun leaves of two species: *Quercus bambusifolia* and *Q. myrsinifolia*. Am J Bot 108 (8): 1441-1463. DOI: 10.1002/ajb2.1706.

Maugarny-Calès A, Laufs P. 2018. Getting leaves into shape: A molecular, cellular, environmental and evolutionary view. Development 145 (13): dev161646. DOI: 10.1242/dev.161646.

McKown AD, Klápště J, Guy RD, Geraldes A, Porth I, Hannemann J, Friedmann M, Muchero W, Tuskan GA, Ehlting J, Cronk QCB, El-Kassaby YA, Mansfield SD, Douglas CJ. 2014. Genome-wide association implicates numerous genes underlying ecological trait variation in natural populations of *Populus trichocarpa*. New Phytol 203 (2): 535-553. DOI: 10.1111/nph.12815.

Mingkid J, Siahaan R, Katili D, Tampemawa R. 2023. Distribution of *Elatostema* J.R. Forster & G. Foster of Mopilu River Riparian, South Minahasa-North Sulawesi. J Biotechnol Conserv Wallacea 3 (1): 15-19. DOI: 10.35799/jbcw.v3i1.47231. [Indonesian]

Mustari AH. 2020. Ecology, Behavior, and Conservation of Anoa. IPB Press, Bogor. [Indonesian]

Niinemets Ü, Keenan TF, Hallik L. 2015. A worldwide analysis of within-canopy variations in leaf structural, chemical and physiological traits across plant functional types. New Phytol 205 (3): 973-993. DOI: 10.1111/nph.13096.

Onoda Y, Wright IJ, Evans JR, Hikosaka K, Kitajima K, Niinemets Ü, Poorter H, Tosens T, Westoby M. 2017. Physiological and structural tradeoffs underlying the leaf economics spectrum. New Phytol 214 (4): 1447-1463. DOI: 10.1111/nph.14496.

Pusparini W, Cahyana A, Grantham HS, Maxwell S, Soto-Navarro C, Macdonald DW. 2023. A bolder conservation future for Indonesia by prioritising biodiversity, carbon and unique ecosystems in Sulawesi. Sci Rep 13 (1): 842. DOI: 10.1038/s41598-022-21536-2.

Rajapaksha R, Gunawardena N, Garasin U, Pushpakumara G, Premakantha T, Wijesundara S, Averyanov L. 2019. *Elatostema rigidiusculum* (Urticaceae), a new species endemic to Sri Lanka. Phytotaxa 404 (2): 85-90. DOI: 10.11646/phytotaxa.404.2.4.

Rodda M, Monro AK. 2018. *Elatostema muluense* (Urticaceae), a new species from the extraordinary caves of Gunung Mulu National Park, Malaysia. Kew Bull 73: 41. DOI: 10.1007/s12225-018-9768-z.

Rossatto DR, de Araújo PE, da Silva BHP, Franco AC. 2018. Photosynthetic responses of understory savanna plants: Implications for plant persistence in savannas under tree encroachment. Flora 240: 34-39. DOI: 10.1016/j.flora.2017.12.009.

Sakio H. 2020. Long-Term Ecosystem Changes in Riparian Forests. Springer Nature, London. DOI: 10.1007/978-981-15-3009-8.

Saleh MFRM, Hartana A. 2017. Plant species diversity of Pangi Binangga Nature Reserve, Central Sulawesi. Media konservasi 22 (3): 286-292. DOI: 10.29244/medkon.22.3.286-292. [Indonesian]

Scales JA, Butler MA. 2016. Adaptive evolution in locomotor performance: How selective pressures and functional relationships produce diversity. Evolution 70 (1): 48-61. DOI: 10.1111/evo.12825.

Schrader J, Westoby M, Wright IJ, Kreft H. 2021. Disentangling direct and indirect effects of island area on plant functional trait distributions. J Biogeogr 48 (8): 2098-2110. DOI: 10.1111/jbi.14138.

Sharma SK, Pandit MK. 2011. A morphometric analysis and taxonomic study of *Panax bipinnatifidus* Seem. (Araliaceae) species complex from Sikkim Himalaya, India. Plant Syst Evol 297 (1-2): 87-98. DOI: 10.1007/s00606-011-0501-8.

Soladoye MO, Onakoya MA, Chukwuma EC, Sonibare MA. 2010. Morphometric study of the genus *Senna* (Mill) in South-Western Nigeria. Afr J Plant Sci 4 (3): 44-52. DOI: 10.5897/AJPS.9000175.

Temam N, Mohammed W, Aklilu S. 2021. Variability assessment of okra (*Abelmoschus esculentus* (L.) Moench) genotypes based on their qualitative traits. Intl J Agron 2021 (1): 6678561. DOI: 10.1155/2021/6678561.

Triyutthachai N, Fu L-F, Triboun P, Wei Y-G, Pornpongrungrueng P. 2022. Three new species of *Elatostema* (Urticaceae) from Thailand. PhytoKeys 215: 37-50. DOI: 10.3897/phytokeys.215.94591.

Tsai H-C, Chiang J-M, McEwan RW, Lin T-C. 2018. Decadal effects of thinning on understory light environments and plant community structure in a subtropical forest. Ecosphere 9 (10): e02464. DOI: 10.1002/ecs2.2464.

Tserej O, Feeley KJ. 2021. Variation in leaf temperatures of tropical and subtropical trees are related to leaf thermoregulatory traits and not geographic distributions. Biotropica 53 (3): 868-878. DOI: 10.1111/btp.12919.

Upadhyay AK, Gogoi R, Mitra PK. 2021. Ethnobotany of the genus *Elatostema* J.R. Forster & G. Forster. Ethnobot Res Appl 21: 1-24. DOI: 10.32859/era.27.01.1-24.

Viani RAG, Rodrigues RR, Dawson TE, Lambers H, Oliveira RS. 2014. Soil pH accounts for differences in species distribution and leaf nutrient concentrations of Brazilian woodland savannah and seasonally dry forest species. Perspect Plant Ecol Evol Syst 16 (2): 64-74. DOI: 10.1016/j.ppees.2014.02.001.

Vlad D, Kierzkowski D, Rast MI, Vuolo F, Ioio RD, Galinha C, Gan X, Hajheidari M, Hay A, Smith RS, Huijser P, Bailey CD, Tsiantis M. 2014. Leaf shape evolution through duplication, regulatory diversification, and loss of a homeobox gene. Science 343 (6172): 780-783. DOI: 10.1126/science.1248384.

Wang C, He J, Zhao T-H, Cao Y, Wang G, Sun B, Yan X, Guo W, Li M-H. 2019. The smaller the leaf is, the faster the leaf water loses in a temperate forest. Front Plant Sci 10: 58. DOI: 10.3389/fpls.2019.00058.

Xin Z-B, Monro AK, Wang R-F, Fu L-F. 2023. An integrative approach to species delimitation sinks three Chinese limestone karst *Elatostema* (Urticaceae) species. PhytoKeys 236: 83-96. DOI: 10.3897/phytokeys.236.114837.

Yang L, Chen L, Wei W. 2015. Effects of vegetation restoration on the spatial distribution of soil moisture at the hillslope scale in semi-arid regions. Catena 124: 138-146. DOI: 10.1016/j.catena.2014.09.014.

Yang W, Li H, Zhang T, Sen L, Ni W. 2014. Classification and identification of metal-accumulating plant species by cluster analysis. Environ Sci Pollut Res Intl 21 (18): 10626-10637. DOI: 10.1007/s11356-014-3102-6.

Yao L, Wang D, Wang D, Li S, Chen Y, Guo Y. 2021. Phenotypic plasticity and local adaptation of leaf cuticular waxes favor perennial alpine herbs under climate change. Plants 11 (1): 120. DOI: 10.3390/plants11010120.

Yin D-H, Huang T-F, Lu Z, Duan L-D. 2021. A new species and a new series of *Elatostema* (Urticaceae) from south-western China. PhytoKeys 180: 65-71. DOI: 10.3897/phytokeys.180.65813.