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Phylogenetic Relationships of Grasshoppers (Orthoptera) in Rice Field Ecosystems of Parigi Moutong Regency Based on Morphological Characters

Mia Pinasti Azizah, I Made Budiarsa*, Manap Trianto, I Nengah Kundera, Abdul Ashari, Zulaikhah Dwi Jayanti
Department of Biology Education, Faculty of Teacher Training and Education, Tadulako University.
Jl. Soekarno Hatta No KM 9, 94148, Central Sulawesi, Tel./Fax. (0451) 422611, Indonesia.

Corresponding author
budiarsa_imade@yahoo.com

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Abstract

Grasshoppers are insects belonging to the order Orthoptera, which comprises a highly diverse group of species distributed across various ecosystems, including agricultural habitats such as rice fields. This study aimed to analyze the phylogenetic relationships among Orthopteran grasshoppers inhabiting rice field ecosystems in Parigi Moutong Regency based on morphological characters and to describe the clustering patterns of the species identified in the area. Sampling was conducted in rice fields using an exploratory survey method with free collection techniques. Species identification was carried out through the examination of morphological characteristics using biodiversity references and taxonomic identification keys. Phylogenetic relationships were analyzed using the Unweighted Pair-Group Method with Arithmetic Average (UPGMA) implemented in the PAleontological STatistics software (PAST 4.03), while Principal Component Analysis (PCA) was performed and visualized in a biplot to support the clustering results. A total of nine grasshopper species were identified: *Phaneroptera falcata*, *Oxytauchira brachyptera*, *Scudderia texensis*, *Atractomorpha crenulata*, *Tettigonia viridissima*, *Conocephalus fuscus*, *Phlaeoba infumata*, *Oxya serville*, and *Phlaeoba fumosa*. The phylogenetic analysis grouped the species into two major clusters with three levels of relationship. A very close relationship was observed between *Phlaeoba infumata* and *Phlaeoba fumosa* (similarity index [SI] = 91%). A close relationship was found between the subclusters consisting of *Atractomorpha crenulata*, *Phlaeoba infumata*, and *Phlaeoba fumosa*, and those consisting of *Oxytauchira brachyptera* and *Oxya serville* (SI = 62%). In contrast, a more distant relationship was observed between *Conocephalus fuscus* and the group comprising *Scudderia texensis*, *Phaneroptera falcata*, and *Tettigonia viridissima* (SI = 47%). These phylogenetic patterns were supported by the PCA biplot, in which *P. fumosa*, *P. infumata*, *O. serville*, and *O. brachyptera* were positioned within the same component, whereas *S. texensis*, *P. falcata*, and *T. viridissima* occupied different regions, and *C. fuscus* was clearly separated from the other species. This study provides scientific information on morphology-based phylogenetic relationships among grasshopper species in rice field ecosystems, which may serve as a foundation for taxonomic studies and as a reference for biology education concerning grasshopper phylogeny.

Keywords: Grasshoppers; phylogenetic relationships; morphological characters; UPGMA.

INTRODUCTION

Grasshoppers are insects belonging to the order Orthoptera, which comprises a highly diverse group of species widely distributed across various ecosystems, including agricultural habitats such as rice fields (Faiza et al., 2024). The presence of grasshoppers in rice field ecosystems not only reflects high levels of biodiversity but also indicates environmental conditions capable of supporting a wide range of organisms (Zuliani et al., 2024). Within food webs, grasshoppers function primarily as herbivores that utilize plants as their main food source. In addition, they can serve as bioindicators of environmental change, including ecosystem disturbances and pollution (Prakoso, 2022). Beyond their ecological significance, grasshoppers exhibit considerable morphological diversity among species. Differences in body size, wing morphology, body coloration, antennal structure, and hind leg morphology are among the characteristics commonly used for species identification (Pelu et al., 2025). Such morphological variation forms the basis of taxonomic classification and can be utilized to investigate phylogenetic relationships among species and genera (Fitriani et al., 2025).

Insect identification is generally conducted through the examination of external morphological characters because each species possesses a unique combination of traits that can be used for differentiation (Yasmin et al., 2024). The identification process typically relies on taxonomic keys, scientific literature, and comparisons with reference specimens to ensure accurate species determination (Indriati & Hidayat, 2023). Morphological characters obtained during identification are subsequently compiled into character matrices, which serve as the foundation for assessing similarities among taxa (Ramadhani & Purnomo, 2023). Comparisons of shared and distinct characteristics allow researchers to infer phylogenetic relationships, with species exhibiting greater morphological similarity generally considered more closely related (Alami et al., 2025).

Numerous studies have investigated insect phylogeny using morphological approaches combined with phylogenetic analyses. Astuti and Ruslan (2019) demonstrated that species within the order Orthoptera can be grouped into several phylogenetic clusters based on morphological similarities, with the highest similarity value reaching 92.3% among closely related species and the lowest approximately 33.87% among distantly related taxa. Furthermore, Cai et al. (2019) reported that phylogenetic analyses effectively separate insect species into clusters according to character similarity, thereby illustrating evolutionary relationships among species. These findings suggest that phylogenetic analysis is an effective approach for understanding species relationships based on shared morphological traits.

In modern taxonomy, phylogenetic analyses increasingly employ morphological datasets processed through quantitative methods such as cluster analysis and Principal Component Analysis (PCA) to provide more objective assessments of species grouping patterns (Palupi et al., 2023; Artha et al., 2025). The results are commonly visualized using dendrograms and biplots, which facilitate systematic interpretation of phylogenetic relationships. The degree of relatedness among species is represented by similarity values expressed as percentages, allowing clearer comparisons of taxonomic affinities (Agustina & Hasanuddin, 2021; Hamidah et al., 2024). Compared with purely descriptive approaches, these quantitative techniques offer greater accuracy in identifying patterns of similarity and variation among taxa.

Parigi Moutong Regency possesses considerable potential as a habitat for diverse grasshopper species due to its extensive agricultural landscape covering approximately 30,403.20 ha and its predominantly flat to gently undulating topography. These environmental conditions support agricultural activities while simultaneously providing suitable habitats for various insect species. Preliminary observations indicated that rice field ecosystems in several villages harbor relatively high grasshopper diversity. However, scientific information regarding phylogenetic relationships among grasshopper species in this region remains limited and has not been specifically documented. Previous studies have primarily focused on species inventories and diversity assessments without examining the phylogenetic relationships among the recorded species. Understanding such relationships is important for evaluating morphological similarities, clarifying taxonomic affinities among species, and supporting the development of local insect taxonomy studies. This study aimed to analyze the phylogenetic relationships of Orthopteran grasshoppers inhabiting rice field ecosystems in Parigi Moutong Regency based on morphological characters and to describe the clustering patterns of the identified species.

MATERIALS AND METHODS

Study Area

This study was conducted in December 2025. The research was carried out in three rice field locations in Parigi Moutong Regency, Central Sulawesi, Indonesia, namely Olaya Village, Parigi District; Boyantongo Village, West Parigi District; and Kayuboko Village, South Parigi District (Figure 1). These locations were selected because they represent important rice-growing areas and provide suitable habitats for various grasshopper species (Orthoptera) associated with agricultural ecosystems.

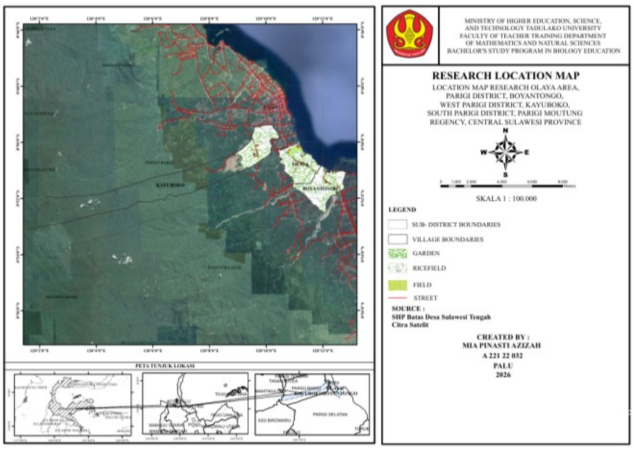

Figure 1. Map of the research location in Parigi Moutong Regency, Central Sulawesi, Indonesia.

Procedures

Sample Collection

Grasshopper sampling was conducted using an exploratory survey method with a free-collection technique. Sampling was carried out twice daily, in the morning from approximately 07:00–10:00 WITA and in the afternoon from 14:00–17:00 WITA. Specimens were captured using an insect net. To ensure adequate representation, a minimum of three individuals of each species encountered at a sampling location were collected whenever possible. Captured grasshoppers were carefully placed in insect collection boxes and labeled according to the sampling location and collection time to facilitate subsequent identification. In addition, environmental parameters, including temperature, relative humidity, and light intensity, were measured during each sampling session.

Identification of Grasshopper Morphological Characters
Species identification was performed through the examination of morphological characteristics using biodiversity references and taxonomic identification keys. Each specimen was subsequently classified taxonomically from the kingdom level to the species level. A total of 36 morphological characters were observed, including head shape, leg morphology, body coloration, body form, and antennal structure. The observed characters were recorded in a binary character matrix, in which a value of "1" indicated the presence of a particular character and a value of "0" indicated its absence. This coding system facilitated subsequent statistical and phylogenetic analyses. During the identification process, all specimens were also photographed using a digital camera to provide visual documentation and support the accuracy of species determination.

Data Analysis
The morphological data, compiled in the form of a binary character matrix, were analyzed using the software PAST (PAleontological STatistics) through cluster analysis employing the UPGMA (Unweighted Pair-Group Method with Arithmetic Average) algorithm. This method calculates the degree of similarity or distance among taxonomic units and progressively groups the most similar pairs into larger clusters until a complete hierarchical classification is obtained. The results were presented as a dendrogram illustrating the phylogenetic relationships among grasshopper species based on shared morphological characteristics collected from the three study locations. To complement the cluster analysis, Principal Component Analysis (PCA) was performed and visualized as a biplot to illustrate species grouping patterns. PCA reduces multidimensional morphological data into a smaller number of principal components, allowing relationships among species to be interpreted based on the proximity of their positions in the ordination space. Species sharing similar morphological characteristics are expected to cluster closely together, whereas morphologically distinct species are positioned farther apart. Therefore, the PCA results were used to support and validate the clustering patterns generated by the UPGMA analysis. In addition, the Similarity Index (SI) was calculated to quantify the degree of morphological similarity among species. The resulting similarity values were categorized into four levels of phylogenetic relationship: very close (≥75%), close (51–74%), distant (26–50%), and very distant (≤25%). These categories were used to facilitate the interpretation of species relationships and to provide a clearer understanding of morphological affinities among the grasshopper species recorded in the rice field ecosystems of Parigi Moutong Regency.

RESULTS AND DISCUSSION

Grasshopper Species Recorded
The results of the study revealed the presence of nine grasshopper species belonging to eight genera and three families within the study area (Figure 2). The recorded species were *Phaneroptera falcata*, *Oxytauchira brachyptera*, *Scudderia texensis*, *Atractomorpha crenulata*, *Tettigonia viridissima*, *Conocephalus fuscus*, *Phlaeoba infumata*, *Oxya serville*, and *Phlaeoba fumosa*. The occurrence of these taxa indicates a relatively diverse grasshopper assemblage within the rice field ecosystem of Parigi Moutong Regency. The taxonomic diversity observed suggests that the species composition recorded in the study area is sufficiently representative of the Orthopteran community associated with rice field habitats. The presence of multiple genera and families reflects the ecological heterogeneity of the agricultural landscape, which provides a variety of microhabitats and food resources that support different grasshopper species. Such diversity highlights the ecological importance of rice field ecosystems as habitats for Orthopteran insects and provides a valuable basis for subsequent analyses of morphological variation and phylogenetic relationships among the recorded species.

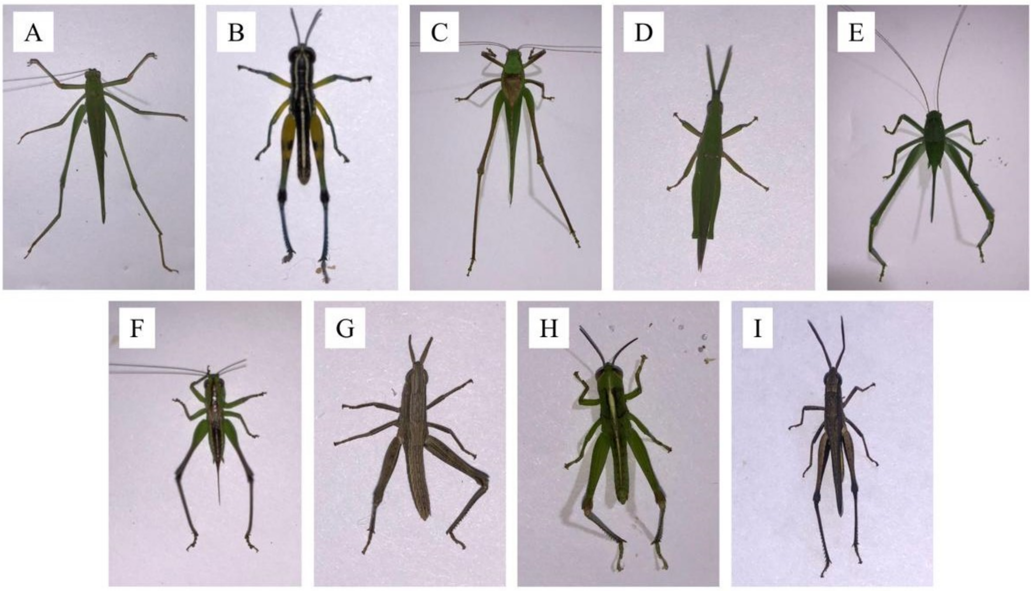

Figure 2. Grasshopper species: A. *Phaneroptera falcata,* B. *Oxytauchira brachyptera,* C. *Scudderia texensis,* D. *Atractomorpha crenulata,* E. *Tettigonia viridissima,* F. *Conocephalus fuscus,* G. *Phlaeoba infumata,* H. *Oxya serville,* and I. *Phlaeoba fumosa.* Scale: 1 cm.

The identified species were subsequently subjected to a systematic examination of morphological characters to obtain standardized and measurable data. A total of 36 morphological characters were recorded for each species, including traits related to body shape, coloration, antennae, wings, legs, and head morphology. These characters served as the basis for assessing morphological similarities and differences among species and were later used in the phylogenetic analyses. The collected morphological data were compiled, coded, and summarized in a binary character matrix to facilitate statistical analysis. A complete description of the observed morphological characters for all identified grasshopper species is presented in Table 1.

Table 1. Morphological character distribution among grasshopper species.

No	**Characteristics**	**Species**								
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
1	Hind femur larger than fore femur	1	1	1	1	1	1	1	1	1
2	Hind femur smaller than fore femur	0	0	0	0	0	0	0	0	0
3	Antennae equal to or longer than the entire body length	1	0	1	0	1	0	0	0	0
4	Antennae approximately half the body length or shorter	0	1	0	1	0	1	1	1	1
5	Tarsi consisting of three segments	0	1	0	1	0	0	1	1	1
6	Tarsi consisting of four segments	1	0	1	0	1	1	0	0	0
7	Ovipositor long and needle-shaped	0	0	0	0	1	1	0	0	0
8	Ovipositor long and sword-shaped	0	0	1	0	0	0	0	0	0
9	Ovipositor short	1	1	0	1	0	0	1	1	1
10	Fore tibia enlarged	0	0	0	0	0	0	0	0	0
11	Fore tibia slender (reduced in width)	1	1	1	1	1	1	1	1	1
12	Body large and elongated	1	0	1	0	1	0	0	0	0
13	Body small and elongated	0	1	0	1	0	1	1	1	1
14	Forelegs adapted for grasping prey	0	0	0	0	1	1	0	0	0
15	Forelegs not adapted for grasping prey	1	1	1	1	0	0	1	1	1
16	Femur serrated with spine-like projections	0	0	0	0	0	0	0	0	0
17	Femur smooth, without serrations	1	1	1	1	1	1	1	1	1
18	Body flattened and elongated, resembling a leaf	1	0	1	0	1	1	0	0	0
19	Body flattened and elongated, resembling a twig	0	0	0	1	0	0	0	0	0
20	Body oval, robust, and heavily sclerotized	0	1	0	0	0	0	1	1	1
21	Body and wing coloration similar	1	0	1	1	1	0	1	0	1
22	Body and wing coloration different	0	1	0	0	0	1	0	1	0
23	Body predominantly dull brown	0	0	0	0	0	0	1	0	1
24	Body predominantly glossy brown	0	1	0	0	0	0	0	0	0
25	Body predominantly green	1	0	1	1	1	1	0	1	0
26	Head oval-shaped	1	0	1	0	1	0	0	0	1
27	Head rounded	0	0	0	0	0	1	0	0	0

No	Characteristics	Species								
		1	2	3	4	5	6	7	8	9
28	Head triangular	0	1	0	0	0	0	1	1	1
29	Head pointed	0	0	0	1	0	0	0	0	0
30	Head concealed beneath the pronotum	0	0	0	0	0	0	0	0	0
31	Pronotum extending posteriorly	0	0	0	0	0	0	0	0	0
32	Pronotum not extending posteriorly	1	1	1	1	1	1	1	1	1
33	Antennae with setae (hair-like structures)	1	0	1	0	1	1	0	0	0
34	Antennae without setae	0	1	0	1	0	0	1	1	1
35	Prothorax elongated	0	0	0	0	0	0	0	0	0
36	Prothorax short or absent	1	1	1	1	1	1	1	1	1

Information: 1. *Phaneroptera falcata,* 2. *Oxytauchira brachyptera,* 3. *Scudderia texensis,* 4. *Atractomorpha crenulata,* 5. *Tettigonia viridissima,* 6. *Conocephalus fuscus,* 7. *Phlaeoba infumata,* 8. *Oxya serville,* and 9. *Phlaeoba fumosa.*

UPGMA Analysis

The phylogenetic analysis of the nine grasshopper species based on 36 morphological characters revealed a clustering pattern illustrated by the UPGMA dendrogram generated using the Jaccard similarity index (Figure 3). The results demonstrated varying levels of morphological similarity among species, as indicated by the Similarity Index (SI) values at each cluster branching point. These similarity values reflect the degree of shared morphological characteristics among the analyzed taxa and provide insight into their phylogenetic relationships. The highest similarity value was recorded between *Phlaeoba infumata* and *Phlaeoba fumosa*, with an SI of 91%, indicating a very close phylogenetic relationship and a high degree of morphological resemblance. The close association of these two species is reflected by their placement within the same nearest branch of the dendrogram. At a similarity level of 62%, a larger cluster was formed through the grouping of *Atractomorpha crenulata*, *Phlaeoba infumata*, *Phlaeoba fumosa*, *Oxytauchira brachyptera*, and *Oxya serville*. This clustering pattern suggests that these species share a considerable number of morphological characteristics and maintain relatively close phylogenetic relationships, although their similarity is lower than that observed between *P. infumata* and *P. fumosa*. At a lower similarity level of 47%, *Conocephalus fuscus* clustered with *Scudderia texensis*, *Phaneroptera falcata*, and *Tettigonia viridissima*. This grouping indicates a more distant phylogenetic relationship characterized by greater morphological divergence compared with the previously described clusters. The lower similarity value suggests that these species possess fewer shared morphological traits and exhibit more distinct structural characteristics. Overall, the dendrogram demonstrates a hierarchical pattern of relationships among the nine grasshopper species, highlighting varying degrees of morphological similarity that reflect their taxonomic affinities and evolutionary relatedness.

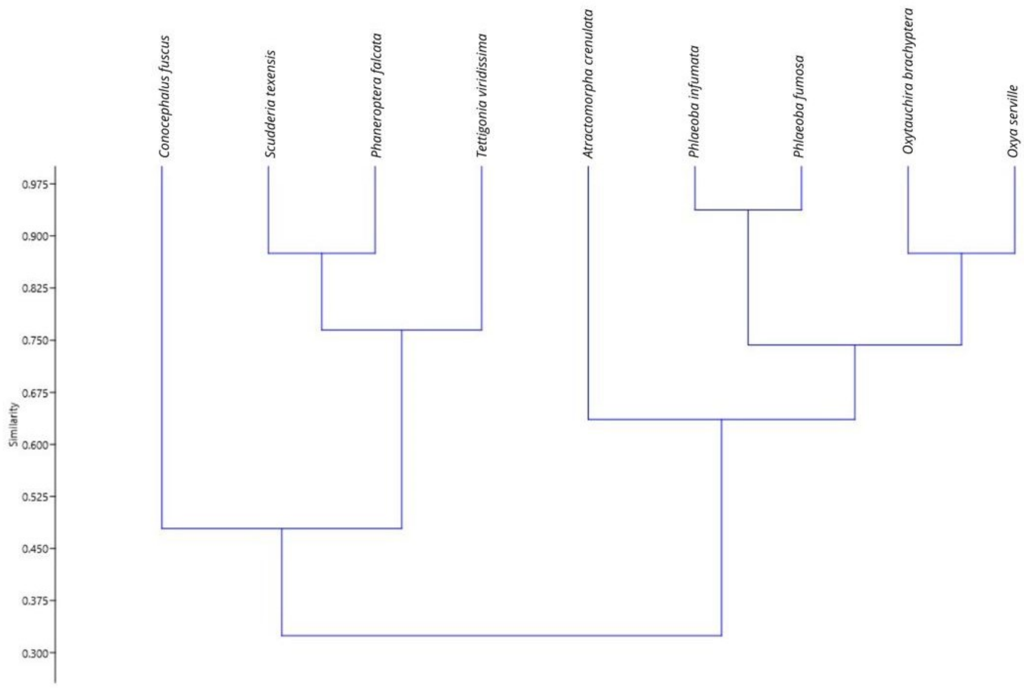

Figure 3. UPGMA dendrogram of nine grasshopper species based on 36 morphological characters.

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PCA Biplot Analysis

The morphological variation among the nine grasshopper species was visualized using a PCA biplot based on the first two principal components (PC1 and PC2). The spatial distribution of species on the biplot provides a graphical representation of morphological similarities and differences among the analyzed taxa. Species positioned close to one another on the plot exhibit a high degree of similarity in their morphological characteristics, whereas greater distances between species indicate more pronounced morphological divergence (Figure 4). The PCA biplot facilitates the interpretation of species grouping patterns by reducing complex multidimensional morphological data into two principal axes that explain the largest proportion of variation. Consequently, the ordination pattern enables the identification of clusters of morphologically similar species as well as species that possess distinctive character combinations. The distribution of species within the PCA space therefore complements the results of the UPGMA cluster analysis and provides additional evidence for the phylogenetic relationships inferred from morphological characters.

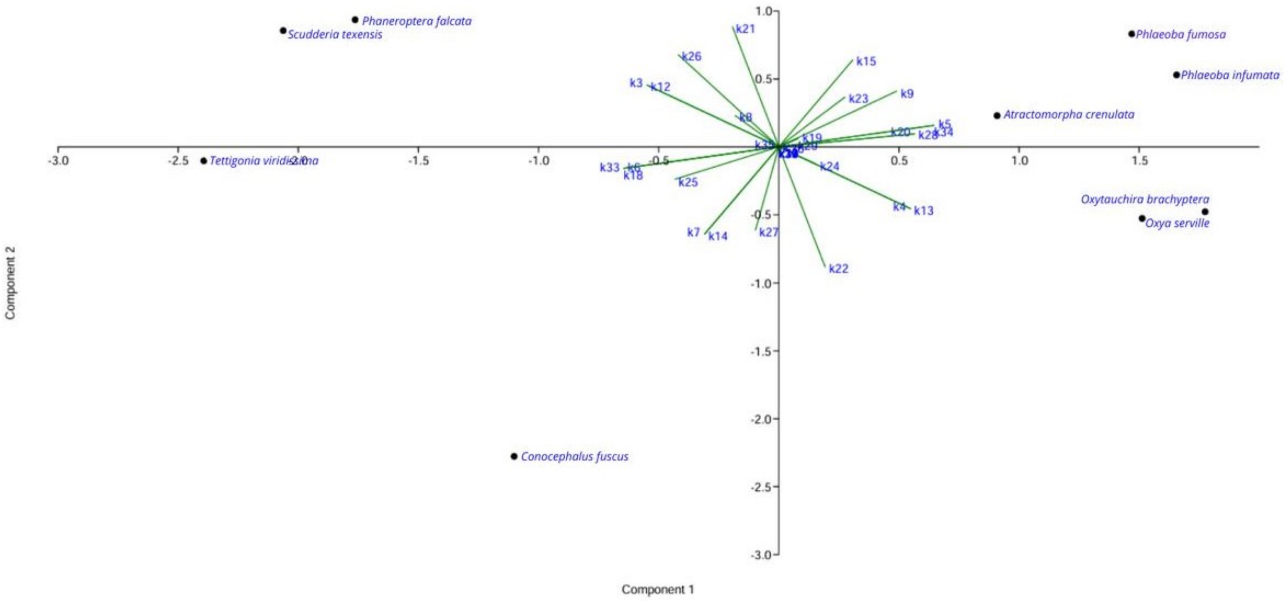

Figure 4. Grafik biplot distribusi spesies berdasarkan dua komponen utama (PC1 dan PC2).

Discussion

The results of this study revealed the presence of nine grasshopper species within the rice field ecosystem of Parigi Moutong Regency, namely *Phaneroptera falcata*, *Oxytauchira brachyptera*, *Scudderia texensis*, *Atractomorpha crenulata*, *Tettigonia viridissima*, *Conocephalus fuscus*, *Phlaeoba infumata*, *Oxya serville*, and *Phlaeoba fumosa*. The occurrence of these species indicates that the study area provides suitable environmental conditions for sustaining grasshopper populations. Measurements of environmental parameters showed an average relative humidity of 64.35%, which falls within a moderate range, an average daily air temperature of 32.7°C, and a light intensity of 4,580 lux, indicating adequate illumination. These abiotic factors play essential roles in supporting grasshopper physiological processes and behavioral activities, including metabolism, movement, foraging, and reproduction. The findings suggest that rice field ecosystems offer favorable habitats for grasshopper species belonging to different taxonomic groups. Such ecosystems are generally characterized by abundant vegetation and suitable microclimatic conditions that support herbivorous insects such as grasshoppers (Putri & Nelly, 2025). The availability of vegetation as a primary food resource, combined with relatively stable environmental conditions, is considered a key factor influencing grasshopper diversity within a habitat (Abrori et al., 2021).

These findings are consistent with those reported by Saroni and Gustina (2021), who found that air temperatures ranging from 32°C to 35°C are suitable for insect survival and contribute to grasshopper diversity, while relative humidity levels between 76% and 92% support breeding, growth, and activity. Furthermore, Irwanto and Gusnia (2021) reported that the effective temperature range for insect activity is approximately 15–25°C, with a maximum tolerance limit reaching 45°C. Therefore, the temperature conditions recorded in the rice field ecosystem remain within the tolerance range of grasshoppers and are capable of supporting their growth and development. In addition to temperature and humidity, insect activity is strongly influenced by light intensity. Previous studies have shown that light intensities around 4,664 lux can facilitate feeding behavior, molting processes, and reproductive activities in insects (Irwanto & Gusnia, 2021). Consequently, the environmental conditions observed in the present study appear favorable for maintaining diverse grasshopper communities.

The phylogenetic analysis based on morphological characters grouped the studied grasshopper species into two major clusters with an overall similarity level of 32% (Figure 3). The first cluster consisted of *Conocephalus fuscus*, *Scudderia texensis*, *Phaneroptera falcata*, and *Tettigonia viridissima*, whereas the second cluster comprised *Atractomorpha crenulata*, *Phlaeoba infumata*, *Phlaeoba fumosa*, *Oxytauchira brachyptera*, and *Oxya serville*, which were united at a similarity level of 62%. Within the first cluster, the closest phylogenetic relationship was observed between *S. texensis* and *P. falcata*, which shared a high similarity value of 89%. This pair subsequently clustered with *T. viridissima* at a similarity level of 76%. In contrast, *C. fuscus* exhibited a lower degree of similarity to the other members of the group, joining the cluster at a similarity level of 47%, indicating greater morphological divergence.

Within the second cluster, the strongest phylogenetic relationship was observed between *Phlaeoba infumata* and *Phlaeoba fumosa*, which shared a similarity value of 91%. This exceptionally high similarity suggests that both species possess very similar morphological characteristics and likely belong to closely related taxonomic lineages. Similarly, *Oxytauchira brachyptera* and *Oxya serville* exhibited a close relationship with a similarity value of 88%, indicating substantial overlap in morphological traits. These clustering patterns demonstrate that species sharing similar morphological features tend to group together, reflecting their taxonomic affinities and evolutionary relatedness. The observed relationships further support the usefulness of morphological characters as reliable indicators for assessing phylogenetic relationships among grasshopper species in agricultural ecosystems.

The findings of this study are consistent with those reported by Astuti and Ruslan (2019), who analyzed similarity levels among Orthopteran species using morphological characters and visualized the results through a dendrogram. Their study demonstrated that certain species pairs exhibited very high similarity values, reaching up to 92.3%, whereas other species showed more distant phylogenetic relationships. High similarity index values indicate a substantial degree of morphological resemblance among species (Simbolon & Aji, 2021). Such similarities arise because closely related species share numerous morphological traits, including body shape, antennal structure, and hind leg morphology. The abundance of shared characteristics reflects a close phenetic relationship, which is based on the overall similarity of observable morphological traits (Danong et al., 2023). In addition, morphological resemblance may also be influenced by similarities in habitat conditions and food resources, which can lead species to develop comparable adaptive characteristics. Conversely, species with lower similarity values possess greater morphological differences and consequently form separate clusters within the dendrogram (Fathonah & Barokah, 2025). These differences may result from variations in body shape, the size of particular body parts, or other distinguishing morphological structures that separate one species from another (Pangestu et al., 2024).

The proximity of species within the ordination space reflects the degree of morphological similarity. For example, *P. infumata* and *P. fumosa* were positioned very close to one another in the upper-right quadrant, while *O. brachyptera* and *O. serville* clustered together in the lower-right quadrant. Likewise, *P. falcata* and *S. texensis* were located close together on the left side of the plot. These spatial relationships indicate high morphological similarity among the respective species pairs. The direction and length of the character vectors further illustrate the contribution of individual morphological traits to the formation of the principal components. Characters oriented toward the right side of the biplot primarily influenced the grouping of *O. brachyptera*, *A. crenulata*, *P. infumata*, *O. serville*, and *P. fumosa*, whereas characters oriented toward the left contributed to the clustering of *P. falcata*, *S. texensis*, and *T. viridissima*. Meanwhile, characters directed downward played an important role in separating *C. fuscus* from the remaining species. Overall, the PCA results confirmed the existence of two major species groups and identified *C. fuscus* as the most morphologically distinct taxon. This grouping pattern was consistent with the UPGMA analysis, which also produced two principal clusters and placed *C. fuscus* as the most divergent species.

The application of PCA in the present study is in agreement with the findings of Ramadhani and Purnomo (2023), who reported that PCA effectively identifies morphological characters contributing to species grouping and clarifies relationship patterns among samples. The PCA results strengthened the clustering pattern obtained from the dendrogram analysis. Visualization through a biplot based on the first two principal components (PC1 and PC2) revealed species distribution patterns according to variation in morphological characters (Maulana et al., 2022). Species located close together within the ordination space exhibited a high degree of morphological similarity, whereas species positioned farther apart displayed more substantial morphological differences (Pangestu et al., 2024). The first principal component (PC1) accounted for the largest proportion of morphological variation among species and therefore played the dominant role in explaining species separation. The second principal component (PC2) provided additional discriminatory power by distinguishing species groups based on specific morphological characteristics. Through the application of relatively simple but robust analytical methods, this study generated valid and practical scientific findings for investigating grasshopper phylogenetic relationships at the local scale. The research integrated ecological, taxonomic, and statistical approaches, beginning with the assessment of environmental conditions supporting grasshopper communities and continuing with phylogenetic analyses based on 36 morphological characters. The resulting relationships were visualized through dendrogram and PCA biplot analyses, providing a comprehensive understanding of species grouping patterns and morphological affinities among grasshoppers inhabiting rice field ecosystems in Parigi Moutong Regency (Sajidah et al., 2025).

CONCLUSIONS

The phylogenetic analysis showed that *Phlaeoba fumosa* and *P. infumata* had the closest relationship (91% similarity) and clustered with *Atractomorpha crenulata*, *Oxya serville*, and *Oxytauchira brachyptera* (62% similarity). In contrast, *Scudderia texensis*, *Phaneroptera falcata*, and *Tettigonia viridissima* formed a separate group (47% similarity), while *Conocephalus fuscus* was the most distinct species. The PCA biplot supported the UPGMA results, grouping *P. fumosa*, *P. infumata*, *O. serville*, and *O. brachyptera* together, whereas *S. texensis*, *P. falcata*, and *T. viridissima* occupied a different region, and *C. fuscus* remained clearly separated. These findings confirm the usefulness of morphological characters for assessing grasshopper phylogenetic relationships in rice field ecosystems.

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Competing Interests: The authors declare that there are no competing interests.

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