BIO Web of Conferences **154**, 01004 (2025) https://doi.org/10.1051/bioconf/202515401004
ICGRC & ICJAM 2024

Diversity and bioindicators of natural enemies in organic paddy fields implementing habitat modification

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Abstract. Indonesian rice farming relies heavily on intensive systems, adversely affecting soil quality, human health, and non-target organisms. Organic systems are a viable alternative characterized by organic fertilizers and biopesticides, avoiding genetically modified microorganisms and promoting biodiversity. This study investigated the impact of biopesticidal fertilizer (BF), and refugia plants on arthropod pests and natural enemy composition. Refugia plants, including Luffa acutangula, Elegans zinnia, Capsicum frutescens, Solanum licopersicum, Vigna unguiculata, and Cosmos caudatus, were planted in rice field bunds to promote habitat modification. A visual encounter survey (VES) was conducted in the morning, afternoon, and evening to examine arthropod composition. Results showed that habitat modification with BF balanced the visiting patterns of herbivorous, predatory, parasitoid, and pollinator insects. The importance value index of predatory insects was higher in plots using habitat modification (PV = 44.65%, and PG = 46.04%) compared to plots without habitat modification (KV = 37.71%, and KG = 38.54%). Changes in light intensity, air temperature, and humidity also influenced insect diversity on agricultural land. This study demonstrates the potential of habitat modification with biopesticidal fertilizers to promote balanced ecosystems in rice farming, reducing the reliance on intensive systems and their negative consequences.

1 Introduction

Intensive agriculture, the hallmark of farming practices in various parts of the world, including Indonesia, has led to severe environmental issues. The extensive use of external inputs such as synthetic fertilizers and pesticides has negatively impacted the balance of ecosystems, degraded soil quality, and reduced biodiversity [1]. These effects threaten the long-term productivity of agricultural land and raise concerns about the sustainability of the farming system itself [2]. As awareness of the importance of sustainability grows, an urgent

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need is to develop and implement more environmentally friendly and sustainable farming systems, such as organic agriculture [3].

Organic farming systems offer a more holistic and sustainable approach, emphasizing the prudent and eco-friendly management of natural resources [4]. A crucial strategy in organic agriculture is habitat modification, where habitat modifications are made to support biodiversity and enhance the presence of natural enemies that play a vital role in natural pest control [5-7]. Establishing refugia blocks, areas surrounding agricultural fields planted with local flora or plants that attract natural enemies, is one form of habitat modification that has proven effective in increasing the diversity and abundance of these beneficial organisms [3, 6, 7]. However, further research is required to determine the effectiveness of this practice across different ecosystem conditions and on a larger scale.

In addition to habitat modification, organic and environmentally friendly biopesticides are a crucial component of organic farming systems. Biopesticides from natural materials such as plants and microorganisms reduce negative environmental impacts and support agroecosystem sustainability [8]. However, the effectiveness of biopesticides in pest control is highly dependent on their integration with other strategies, such as habitat modification, which enhances the presence of natural enemies. Therefore, this research aims to explore the diversity of natural enemies and their role as bioindicators in organic rice fields that apply habitat modification and liquid biopesticidal fertilizers.

This research is crucial for enhancing our understanding of how organic farming practices, particularly those that combine habitat modification and biopesticidal fertilizers, can improve the sustainability of food production. By understanding the dynamics of natural enemy diversity and the function of its bioindicators, we can develop more effective and environmentally friendly land management strategies [8-11]. Furthermore, the findings from this study are expected to provide practical recommendations for farmers to adopt improved organic farming techniques that support productivity and maintain the balance of agricultural ecosystems.

2 Materials and methods

2.1 Study site

This field research was conducted from July 2021 to December 2022 in the organic red rice paddy fields located in Pagelaran District, Malang Regency (Fig. 1). This site was located at 8°11’57” and 112°36’09” in altitude. Meanwhile, insect identification and data analysis were carried out at the Ecology Laboratory and the Animal Diversity Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Brawijaya, Malang.

2.2 Equipment

The equipment used in this study is divided into three categories: field tools, laboratory instruments, and supporting tools. Field tools include raffia, wood, lux meter, thermo hygrometer, knife, notebook, smartphone, and pencil. Laboratory equipment consists of stereo microscope, urine laminary bottles, and Petri dishes. Meanwhile, supporting tools include identification books such as the work of Siwi (1991), Borror et al. (1996), and literature from the internet, as well as stationery and cameras.

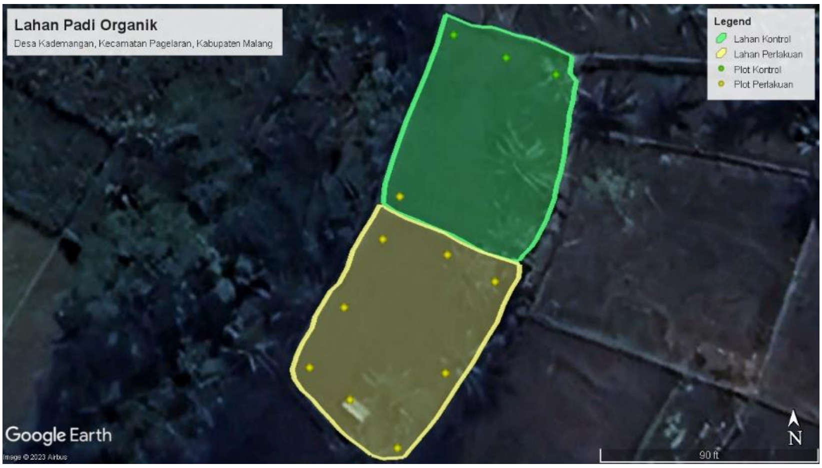

Fig. 1. Map of research location.

2.3 Environmental modification

Environmental engineering was implemented by preparing a 30-meter stretch of land along one side of the red rice paddy field's bund. This area was planted with refugia plants, specifically *Cosmos caudatus*, chili (*Capsicum frutescens*), tomato (*Solanum lycopersicum*), and long beans (*Vigna unguiculata*), which were planted before rice planting. These refugia plants were chosen because their flowers and leaves can attract natural enemies of pests. Observation points were set up along this refugia block. Meanwhile, liquid biopesticidal fertilizers were prepared through the fermentation of a combination of ginger rhizomes, turmeric, galangal, aromatic ginger, lesser galangal, *temulawak* (*Curcuma xanthorrhiza*), maja fruit, papaya leaves, coconut shell steam, and gadung tubers. These ingredients were crushed, mixed with coconut water and rice washing water, dissolved in water, and bioactivators were added before placing the mixture into a fermentation tank. The bioactivators were prepared from goat rumen, cow urine, rice bran, molasses, papaya, pineapple, and shrimp paste [12].

The application of biopesticidal fertilizer (BF) is conducted regularly every two weeks, with a concentration ratio of 1:15. In a 15-liter pesticide sprayer tank, 1 liter of BF is mixed with 14 liters of water. Spraying begins at the early planting stage and continues until harvest, covering all growth phases of the rice plant, including vegetative and generative stages. The spraying is evenly applied to the leaf surfaces, the underside of the leaves, and the soil area around the plant roots using a pesticide sprayer. This technique is designed to maximize the effectiveness of BF in controlling pests. Additionally, the spraying enriches the soil with organic residues, supporting soil microorganism activity and providing long-term ecological benefits to agricultural lands [12].

2.4 Sampling methods

Sampling and observations were conducted twice, during the red rice plants' vegetative phase (February) and the generative phase (June). Each observation phase lasted for five days. Observations were carried out using the Visual Encounter Survey (VES) technique in red rice fields, divided into 18 sites: 9 sites with habitat modification (P) and 9 sites without refugia blocks (K), with each site measuring 1 x 1 x 1 square meter. Observations were made during three time periods: T1 (08:00-10:00), T2 (12:00-14:00), and T3 (15:00-17:00). Arthropod identification was performed down to the family level, referencing Gibb et al. [13] and [14]. If a species could not be identified during the observation, the specimens were gathered and sent to the Animal Diversity Laboratory, Department of Biology, Universitas Brawijaya. Identification was carried out by Purnomo, M.Ling, an expert in insect taxonomy. The visual identification of the arthropods relied on references from Borror et al. (1996), Siwi (1991), and various online resources. All identified arthropods were categorized into guilds, and the number of individuals for each species was recorded. Additionally, abiotic factors measured included air humidity and temperature, which were recorded using a thermohygrometer, and light intensity measured with a lux meter [15].

2.5 Data analysis

The abundance and relative frequency of arthropod families and abiotic factors at all stations were compiled and used to determine the arthropod visitation patterns. Several indices were also calculated, including (1) the Shannon-Wiener Diversity Index (H') with the following equation:

$$H' = -\sum_{i=1}^{s} Pi \, ln \, Pi \tag{1}$$

Where H' represents the Shannon-Wiener diversity index, *S* is the total number of species, and Pi is the proportion of each species' abundance relative to the total abundance of all species. (2) Index of Evenness (*E*) with the following equation.

$$E = \frac{H\prime}{H\prime max} \tag{2}$$

(3) Dominance index of Simpson (*D*) with the following equation,

$$D = \frac{Ni(Ni-1)}{N(N-1)} \tag{3}$$

Where *Ni* is the number of individuals of the *i* species, and *N* is the total number of individuals found, and (4) Index of Taxa Richness (*R*) with the following equation.

$$R = \frac{S-1}{Ln(N)} \tag{4}$$

Additionally, two other indices are calculated: (5) the Indicator Value (IndVal) and (6) the Importance Value Index (IVI), which is determined by the following equation:

$$v = Fr \text{ x } Kr \tag{5}$$

$$INP = Fr + Kr \tag{6}$$

Where *v* is the indicator value, *Fr* represents the relative frequency, and *Kr* denotes the relative abundance.

3 Result and discussion

The environmental characteristics during each period of the engineered organic rice field varied. The average light intensity, humidity, and temperature fluctuated across different periods (Fig. 2). There was an increase in air temperature and light intensity from period 1

(T1) to period 2 (T2), followed by a decrease from T2 to period 3 (T3). Meanwhile, air humidity decreased from T1 to T2 and rose again in T3. Air humidity positively correlated with the number of arthropods visiting the field (Fig. 3), but a negative correlation with light intensity and air temperature. This study indicated that air temperatures ranged from 31.7°C to 35.2°C, which falls within the tolerance range for arthropods. The tolerance range for arthropod temperature is 15°C to 45°C, with an optimum temperature of 25°C [3].

Overall, the abundance of arthropods in the study area amounted to 11,576 individuals collected from twelve sites, consisting of 1,411 species in site K and 10,165 species in site P, across both the vegetative and generative phases. Fig. 3 shows that the average insect abundance was highest in T1 (35.21%), decreased in T2 (30.92%), and then increased again in T3 (33.88%). The insect abundance in T1 and T3 was nearly identical, likely due to abiotic factors, where air temperature, humidity, and light intensity supported insect activity and metabolism. This finding aligns with research by Setyadin et al. [15], which indicated that insects can optimally receive and respond to environmental conditions in the early morning. Rahardjo et al. [3] also noted that high temperatures and light intensity cause insects to visit less frequently, while higher humidity levels attract more insects. This behavior is due to their adaptation to avoid direct sunlight and high temperatures, helping to reduce dehydration, which would otherwise deplete a significant amount of water in their bodies [18-21].

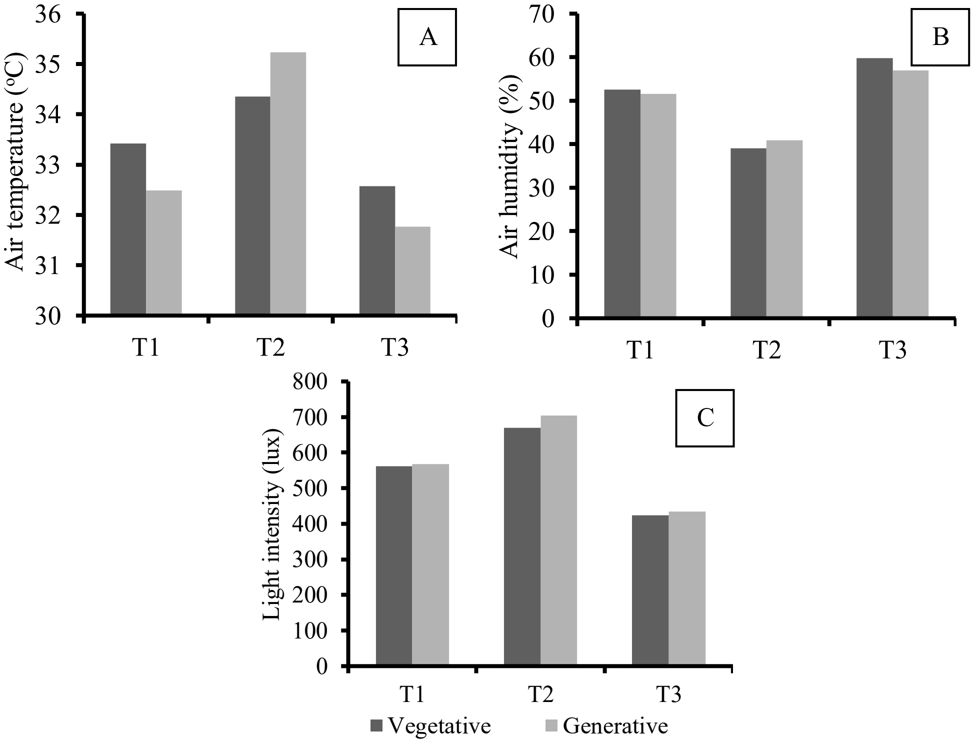

Fig. 2. Temporal variations of abiotic factors in the organic red rice field, (A) air temperature, (B) air humidity, (C) light intensity.

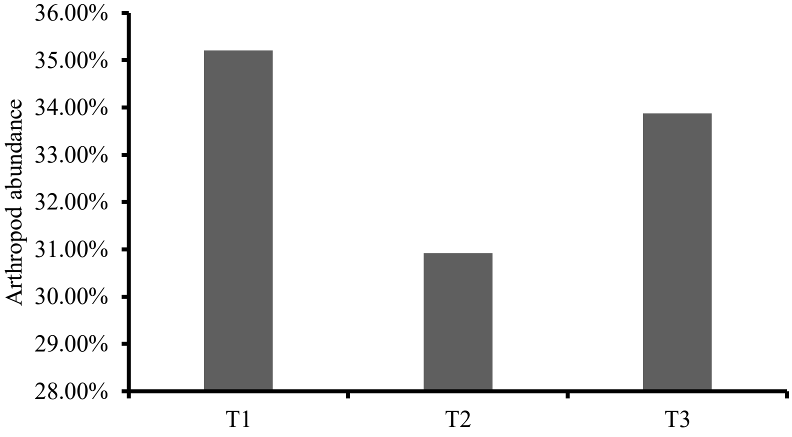

Fig. 3. Temporal variations of arthropod visits in the organic red rice field.

The research findings indicate that the relative abundance (KR) of predators in organic red rice fields, with the application of habitat modification, showed the highest percentages, particularly during the vegetative (PV) and generative (PG) phases, with 44.65% and 46.04%, respectively (Fig. 4). This dominance of predators signifies that the organic rice field environment provides sufficient nutrients and suitable habitat conditions to support natural predator populations. Conversely, the lowest KR was found among pollinating insects, which can be attributed to the scarcity of flowering plants in rice fields, which is typically a primary pollinator food source [20].

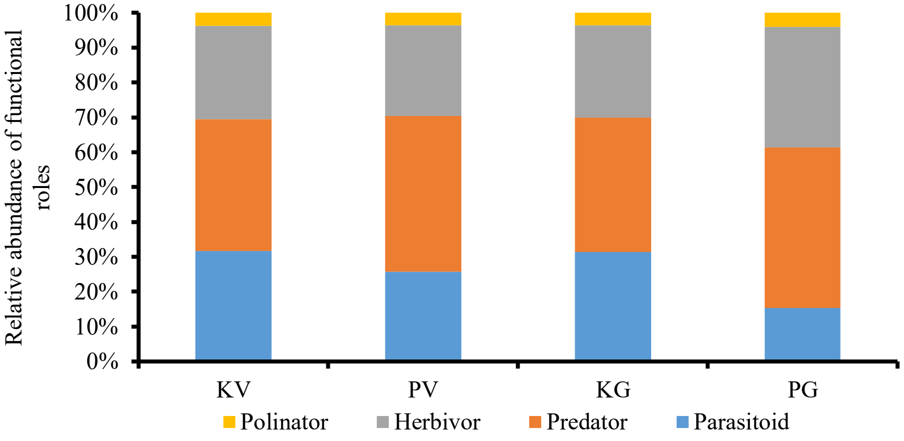

Fig. 4. The relative abundance of arthropods visiting organic red rice fields based on their functional roles.

These results align with the findings of Sorgog et al. [21], who revealed that applying organic farming and habitat modification, such as planting refugia plants, plays a crucial role in attracting natural enemies and enhancing their diversity. Refugia plants provide a complex habitat that serves not only as a food source but also as a shelter for predators and parasitoids, thereby increasing the effectiveness of natural pest control [3, 15] Additionally, the generative phase, characterized by the presence of flowers, attracts more pollinators, consistent with other studies highlighting the importance of plant biodiversity in enhancing the presence and effectiveness of pest-controlling insects [15]. Thus, habitat modification in organic rice fields increases the presence of natural enemies and creates a more balanced and sustainable agricultural ecosystem, as outlined in several studies by Alhmedi et al. [7] and Rahardjo et al. [3] on environmentally friendly farming practices.

Table 1. The Importance Value Index (IVI) of dominant arthropods visiting the organic red rice paddy fields.

Functional Role	**Family (Species)**	**KV (%)**	**PV (%)**	**KG (%)**	**PG (%)**
Predator	Formicidae (*Camponotus* sp.)	53.57	108.09	62.62	130.99
Parasitoid	Trichogrammatidae (*Trichogramma* sp.)	75.12	43.02	92.57	47.06
Herbivor	Chloropidae (*Chlorops oryzae*)	38.80	8.63	40.59	12.51
Herbivor	Chrysomelidae (*Aulacophora foveicollis*)	29.43	2.29	25.04	2.31
Predator	Syrphidae (*Episyrphus* sp.)	26.01	29.24	24.61	25.44
Parasitoid	Aphidiidae (*Aphidius* sp.)	7.03	47.30	2.36	53.15
Polinator	Muscidae (*Musca domesticai*)	4.44	26.66	0.00	25.12
Predator	Lycosidae (*Lycosa* sp.)	25.76	17.64	19.32	21.51
Polinator	Culicidae (*Aedes albopictus*)	10.57	15.74	17.67	22.17
Herbivor	Tephritidae (*Bactrocera* sp.)	23.78	2.28	28.35	4.55
Decomposer	Drosophilidae (*Drosophila* sp.)	0.00	15.25	2.36	23.03

The Importance Value Index (IVI) analysis provides a clear overview of the arthropod community structure in agricultural ecosystems. Based on the data in Table 1, it can be observed that each arthropod species plays an ecological role with varying contributions to the stability of the rice field ecosystem. In the predator category, Formicidae (*Camponotus* sp.) shows a very high IVI value, especially in the PG plot, with a value reaching 130.99%. This indicates that this species of ant plays a crucial role in controlling various pest species that potentially damage rice plants while maintaining the balance of other arthropod populations in the rice field. Although *Lycosa* sp. shows a lower IVI compared to Camponotus sp., they still contribute to pest control, particularly by consuming small insects that threaten rice crops [10].

In the parasitoid category, Trichogrammatidae (*Trichogramma* sp.) is the family with the highest IVI, showing a significant role in pest control through the parasitization of pest eggs. *Aphididus* sp. also contributes greatly to pest control, particularly for aphid populations that can damage rice crops. The presence of parasitoids such as *Trichogramma sp.* and *Aphididus* sp. help reduce the number of pest insects by decreasing their reproductive abilities, where the parasitoid lays its eggs on or inside the host, typically targeting the pest's eggs, larvae, or pupae. Once the parasitoid eggs hatch, the larvae feed on the host's internal tissues, utilizing the host's body as a resource until the host is consumed and eventually dies. This process effectively reduces pest populations naturally, providing a sustainable alternative to chemical pest control [24, 25].

In the herbivore category, Chloropidae (*Chlorops oryzae*), a major pest of rice plants, shows a negative impact on agriculture, with relatively high IVI values in the KV (38.80%) and KG (40.59%) plots. However, in the PV and PG plots (8.63% and 12.51%, respectively), the IVI is lower. Additionally, Chrysomelidae (*Aulacophora foveicollis*), another herbivore, shows a lower IVI, meaning its impact on plants is limited but still significant in the management of organic agricultural ecosystems.

Herbivores such as Chloropidae (*Chlorops oryzae*), Chrysomelidae (*Aulacophora foveicollis*), and Tephritidae (*Bactrocera* sp.) play significant roles in rice field ecosystems by attacking critical parts of rice plants, potentially reducing yield. *Chlorops oryzae* targets young shoots and stems of rice plants. Its larvae bore into the stems, damaging internal tissues and causing symptoms akin to stem borer attacks, which disrupt overall plant growth. *Aulacophora foveicollis* predominantly damages rice leaves. Adult feeding activity on leaf tissues diminishes the plant's photosynthetic capacity, directly affecting plant health and productivity. Meanwhile, *Bactrocera* sp. primarily attacks developing grains, with larvae tunneling into rice kernels, causing direct damage to maturing seeds and lowering harvest quality [23].

As pollinators, Muscidae (*Musca domestica*) and Culicidae (*Aedes albopictus*) contribute less to pollination, but their role remains important in maintaining biodiversity within agricultural ecosystems. Effective pollination is crucial for improving agricultural yields, especially for crops that rely on natural support for reproduction. Meanwhile, Drosophilidae (*Drosophila* sp.), as decomposers, have a high IVI in the PV and PG plots (15.25% and 23.03%, respectively), but are very low in the KV and KG plots. They play an essential role in decomposition and the nutrient cycle of the soil, supporting soil fertility and overall ecosystem health.

The use of refugia plants in the PV (vegetative phase) and PG (generative phase) plots has shown a significant positive impact on arthropod community structure. Refugia plants in these plots provide habitat-supporting natural enemies like *Camponotus sp.* and *Trichogramma sp.*, which play a key role in controlling rice pests. Conversely, in the KV and KG plots, which do not use refugia plants, pest control is more dependent on external inputs such as chemical pesticides or less eco-friendly pest management practices. Without refugia, natural enemy populations in these plots tend to be lower, leading to ecosystem imbalances [24]. This is evidenced by the low IVI values for predators and parasitoids, and the high IVI values for pests that can damage rice plants. Refugia plants play a crucial role in supporting integrated pest management. The IVI in the PV and PG plots shows a greater contribution from predator and parasitoid species, which is directly linked to the presence of refugia. In contrast, in the KV and KG plots, the IVI for predators and parasitoids is lower, reflecting the limited role of natural enemies in pest control.

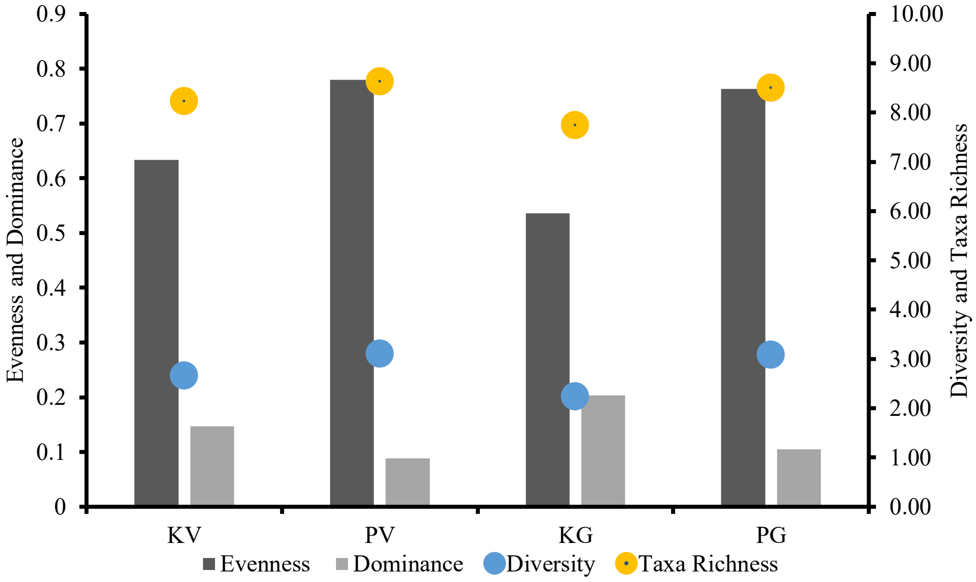

Fig. 5. Community structure indices of arthropod populations in organic red rice fields with and without refugia.

The Evenness Index (Fig. 5) illustrates the distribution of individuals among taxa within a community. Plots with refugia (PV: 0.7797 and PG: 0.7627) showed significantly higher uniformity compared to plots without refugia (KV: 0.6335 and KG: 0.5358). This uniformity indicates that the presence of refugia plants reduces the dominance of certain species, particularly herbivores like *Chlorops oryzae*, a major rice pest. Higher evenness reflects a more equitable distribution of resources within the ecosystem, such as food, shelter, and alternative habitats. Refugia provide a microenvironment supporting a diverse arthropod community, including predators (e.g., *Camponotus sp.*) and parasitoids (e.g., *Trichogramma sp.*). These findings align with Dronova [25], who demonstrated that increasing landscape heterogeneity through refugia blocks enhances species evenness.

The Dominance Index reflects the extent to which certain species dominate a community. Plots without refugia exhibited higher dominance values (KV: 0.1474 and KG: 0.2036) compared to plots with refugia (PV: 0.0888 and PG: 0.1051). Refugia facilitated the presence of natural enemies, such as parasitoids and predators, effectively suppressing dominant herbivore populations. For instance, *Chlorops oryzae* was more controlled in PV and PG plots due to refugia supporting natural enemy populations. Olsiviana et al. [24] found that refugia enhance biological control by reducing pest dominance, contributing to more balanced and environmentally friendly ecosystems.

Diversity Index values were higher in plots with refugia (PV: 3.10 and PG: 3.08) than in those without (KV: 2.65 and KG: 2.24). The presence of refugia blocks provided alternative habitats for various arthropods, including decomposers (e.g., *Drosophila sp.*) and pollinators (e.g., *Musca domestica*). This highlights refugia's role in creating more complex and diverse communities. High species diversity positively correlates with ecosystem stability, as diverse species with different ecological functions complement each other to maintain balance. Ortiz et al. [26] emphasized that high species diversity buffers environmental disturbances and supports sustainable agricultural systems.

Taxa Richness, representing the total number of species in a community, was also higher in plots with refugia (PV: 8.63 and PG: 8.50) compared to plots without refugia (KV: 8.22 and KG: 7.73). Refugia plants provide additional resources, such as nectar and pollen, that support arthropods with diverse ecological needs. Refugia blocks extend the lifecycle of predator and parasitoid species by offering protection during unfavorable periods in primary habitats. These findings align with Duff et al. [27], who reported that refugia increase taxa richness in agricultural landscapes by enhancing habitat and resource availability for species.

These four indices (Evenness, Dominance, Diversity, and Taxa Richness) collectively demonstrate how refugia blocks influence the structure of arthropod communities in organic red rice fields. Refugia plots exhibit higher evenness and diversity, lower dominance, and greater taxa richness compared to non-refugia plots. This data underscores the critical role of refugia in supporting natural pest control, enhancing ecosystem sustainability, and advancing organic agricultural production [28-30]. These findings corroborate prior studies by Gurr et al. [28] and Rahardjo et al. [3], which emphasized the importance of refugia in integrated pest management strategies for sustainable farming.

The results of the Indicator Value (IndVal) analysis show that Trichogrammatidae has the highest indicator value at 18.32% on plot KV and 18.22% on plot KG, while Formicidae recorded the highest IndVal on plot PV (36%) and PG (37.92%) (Fig. 6). The high indicator values reflect species associated explicitly with particular plot conditions. However, both families experienced a decrease in IndVal on plot P compared to plot K. This may be related to changes in habitat structure and resource distribution, which led to a decline in population or migration out of the plots. Conversely, the increased IndVal values for Pieridae and Aleyrodidae on plot PV can be linked to increased pest populations during the vegetative phase. Pieridae (butterflies) and Aleyrodidae (whiteflies) are herbivores typically associated with lush host plants during the vegetative phase. Their higher presence on plot P indicates that this plot provides favorable environmental conditions for these herbivores. The IndVal analysis indicates a strong relationship between specific arthropod species and the growth phase of red rice plants, as well as the effects of the implemented habitat modifications. IndVal values offer crucial information on arthropods' ecological adaptation and community function, particularly in how predator and parasitoid species function as biological control agents in organic farming systems. These findings align with previous research emphasizing the importance of environmental modification and plant diversity in supporting the presence of natural enemies in organic farming fields [3].

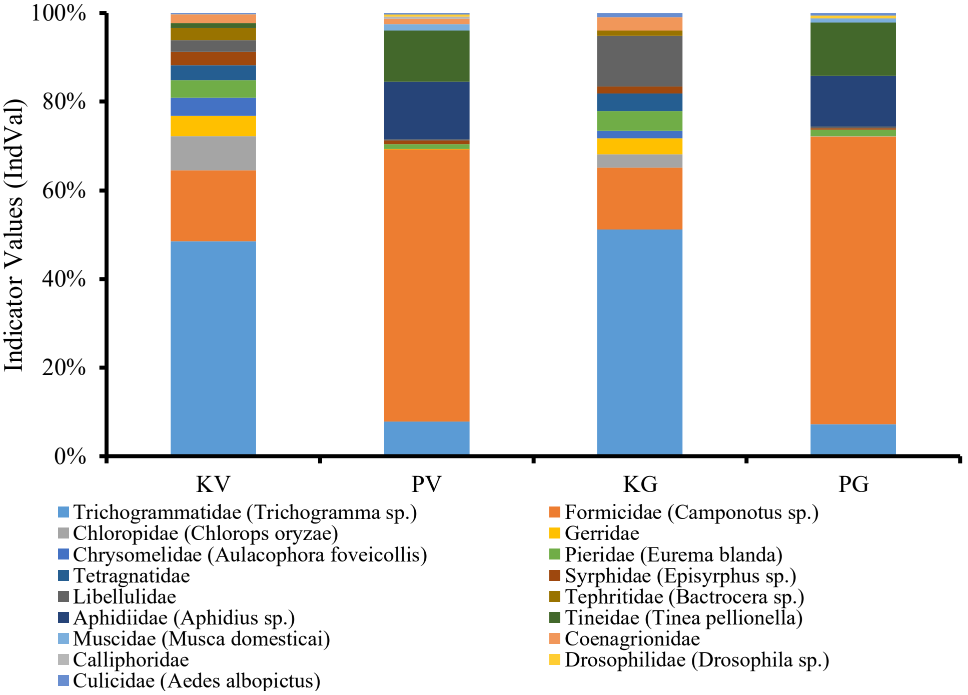

Fig. 6. The indicator Value (IndVal) of dominant arthropod communities visiting red rice paddy fields.

The high IndVal value for Formicidae indicates that Formicidae have a strong affinity for the organically managed red rice paddies, both during the vegetative and generative phases. This suggests that Formicidae are consistently present in organic paddy ecosystems in terms of frequent presence (high fidelity) and dominant abundance (high specificity). Their presence as biological indicators also signals that this group plays a crucial role in the ecosystem's structure, particularly regarding trophic interactions and natural pest control in organic red rice fields. Several Formicidae species are predators that can produce insecticidal compounds that kill agricultural pests, such as Plutellidae, Aleyrodidae, and others [31].

In the KG plot, Trichogrammatidae recorded the highest indicator value at 18.22%. Although this value is lower than that of Formicidae, the significant IndVal value for Trichogrammatidae indicates that this family plays a crucial role as natural parasitoids, particularly during the generative phase of rice plants. Trichogrammatidae is known as a group of natural enemy arthropods that are important for controlling pest populations in agricultural ecosystems, such as Plutellidae, Libellulidae, and Acrididae [32]. The lower IndVal compared to Formicidae may reflect their more specific habitat preferences or focus on biological control of certain pests, especially during the generative phase when their hosts are more available.

4 Conclusions

The conclusions of this study reveal that temporal fluctuations in abiotic variables, such as air humidity, temperature, and light intensity, significantly impact the patterns of insect visitation in organic rice fields. Observations identified four functional statuses of insects dominating the agricultural land: predators, parasitoids (natural enemies), pollinators, herbivores (pests), and decomposers. Data indicate that natural enemies are more abundant in fields with habitat modification (refugia) compared to fields without such modifications. Furthermore, fields implementing habitat modification display a more stable insect community structure characterized by various critical natural enemies as bioindicators of ecosystem stability. This underscores the role of habitat modification in enhancing diversity and bioindicator species of natural enemies and stabilizing the arthropod community, thereby supporting the success of biological control in organic farming systems.

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