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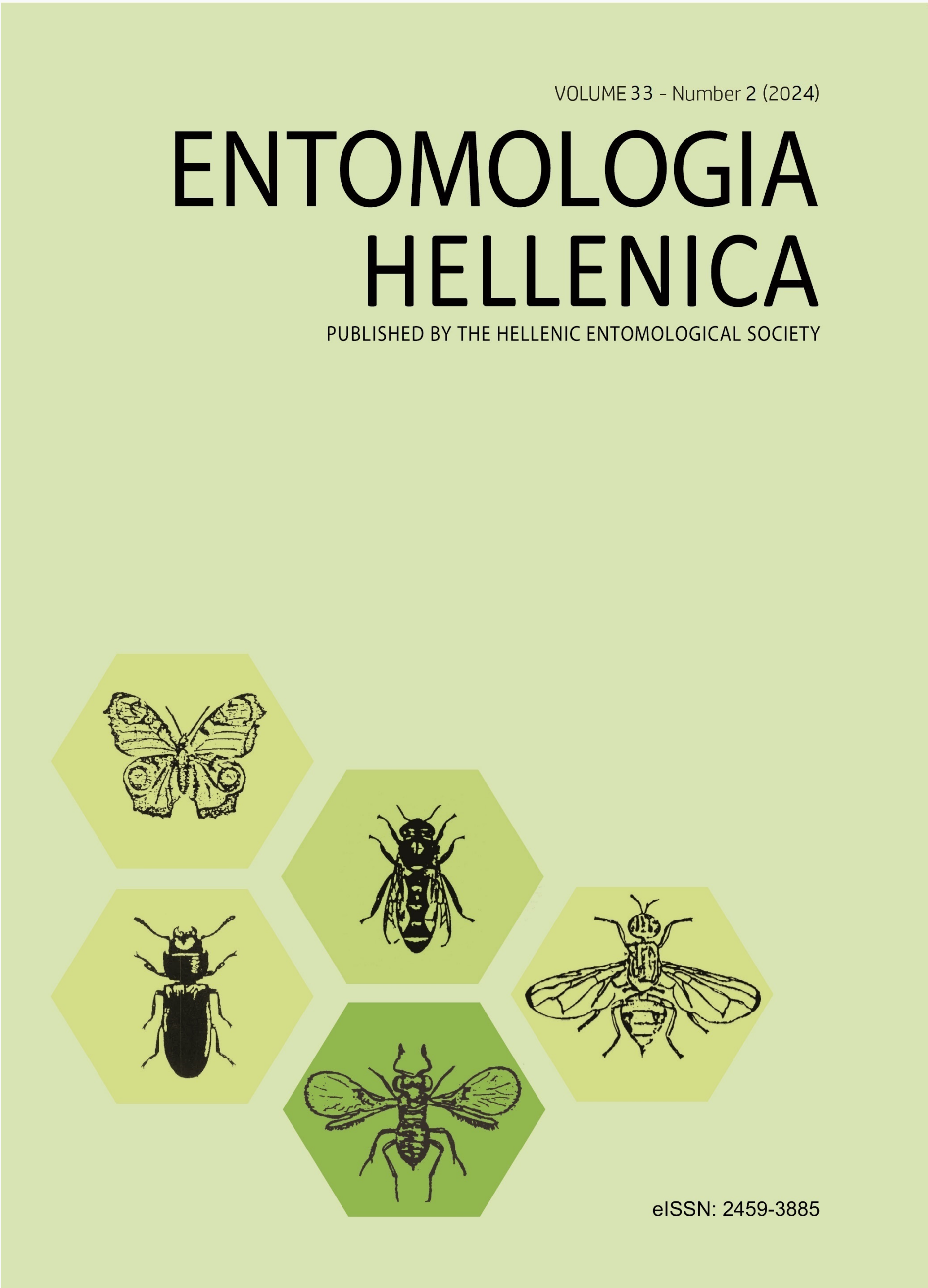

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Ecological Insights into Insect Diversity in Protected Area Networks of Kumaun Region, Western Himalaya

MANOJ KUMAR ARYA*, SURABHI BISHT* AND AMBIKA TIRUWA

Insect Biodiversity Laboratory, Department of Zoology

D.S.B. Campus, Kumaun University, Nainital- 263002, Uttarakhand, India

ABSTRACT

The Uttarakhand region of the Western Himalaya, known for its rich biodiversity, includes several protected areas ranging from tropical to temperate zones. This study explores insect diversity across the six protected areas within the Kumaun Himalayan region. Altogether, a total of 412 insect species from nine taxonomic orders and 70 families were documented. Order Lepidoptera was the most diverse with a maximum of 154 species, followed by Coleoptera (81 species), Hymenoptera (58 species), Orthoptera (33 species), Hemiptera (31 species), Odonata (28 species), Diptera (23 species), Isoptera and Neuroptera as the least dominant with two species each. Shannon's species diversity (Hs) ranged 3.99 to 4.95, with the highest diversity in Nandhaur Wildlife Sanctuary and the lowest in Naina Devi Himalayan Bird Conservation Reserve. Cluster analysis revealed two main diversity patterns, indicating significant beta diversity amongst the study areas. Species-wise occupancy and abundance analysis revealed that *Pieris brassicae*, *P. canidia*, and *Apis dorsata* had the highest relative abundance from all protected areas. Conversely, 91 insect species had a relative abundance of only 0.03% each. Furthermore, seven species demonstrated the highest normalized occupancy of 1.00, indicating their adaptability to diverse environmental conditions within the protected areas. These findings thus emphasize the importance of habitat diversity and targeted conservation strategies to maintain insect populations and ecosystem health in the Kumaun Himalaya.

KEY WORDS: Insects, Distribution, Protected area, Species richness, Kumaun Himalaya.

Introduction

Class Insecta (1,070,781 species) is the most successful group, and it alone accounts for over 80% of all arthropods (Zhang 2013). It is characterized by vast diversity and plays a crucial role in shaping terrestrial ecosystems (Steffan-Dewenter and Tscharntke, 2002; Samways, 2005). Being involved in various ecosystem processes such as pollination, decomposition, predation, serving as prey, bioindicators or influencing nutrient cycling, pest and parasite control (Nichols et al., 2008; Bonebrake et al., 2010; An and Choi, 2021), insects play multifaceted roles in ecosystem dynamics, plant reproduction, and trophic interactions. Even though insects have ecological roles, they are often overlooked in biodiversity research and conservation efforts, overshadowed by larger and more charismatic fauna.

Understanding the diversity and richness of insect assemblages across different protected areas is essential for effective conservation and management strategies. The Protected Area Networks (PANs) with national parks, wildlife sanctuaries and conservation reserves for biodiversity conservation are aimed at preserving the region's ecological integrity and safeguarding its unique biodiversity (Margules and Pressey, 2000). They are essential in the conservation of biodiversity and wildlife against further losses as a result

*Corresponding authors: dr.manojkumar19@rediffmail.com; surabhibisht17@gmail.com

of unparalleled anthropogenic impacts (Sharma et al., 2020). In Uttarakhand, there are 18 protected areas where numerous studies have been conducted to understand diversity and richness of various vertebrates and invertebrates, including insects. Published literature on insects from diverse protected areas of Uttarakhand includes Chaturvedi (1981), Baindur (1993), Arora (1994, 1995, 1997), Joshi et al. (1999; 2004), Kumar (2004), Uniyal (2004), Joshi and Arya (2007), Bhardwaj et al. (2008), Joshi et al. (2008), Kumar (2008), Bhargav et al. (2009), Singh (2009), Arya and Joshi (2011), Bhardwaj and Uniyal (2013), Tewari and Rawat (2013), Arya and Joshi (2014), Dayakrishna and Arya (2015), Dey et al. (2015), Singh and Sondhi (2016), Dayakrishna et al. (2016), Uniyal et al. (2016), Sanwal et al. (2017), Arya et al. (2018), Bandyopadhyay et al. (2019), Kumar et al. (2019), Arya and Dayakrishna (2020), Arya and Verma (2020), Arya et al. (2020 a,b), Arya et al. (2021), Chandra et al. (2023). Despite the ecological importance of insects and the pivotal role played by the PANs in biodiversity conservation, our understanding of insect diversity within the protected areas of the Kumaun region remains limited. The present study aims to provide comprehensive baseline data on insect diversity across six protected areas of Uttarakhand and a comprehensive overview of insect occupancy and abundance in these ecologically significant regions. By analyzing spatial heterogeneity, we aim to enhance conservation strategies and deepen our understanding of ecological dynamics in these important habitats.

Materials and Methods

Study area. The state of Uttarakhand lies in the central sector of Himalaya, an area of 53,483 sq. km accounting for 1.63% of India’s geographical area within 28°43' and 31°28' North Latitudes and 77°34' and 81°03' East Longitudes. The state’s major physiographic zones are the Upper Himalayas, the Shiwaliks and the Terai, which cover a range of diverse landscapes that support many endemic floral and faunal species. About 18.70% of the total area (9,885 sq. km) has been designated for the establishment and management of protected areas in the form of national parks, biosphere reserves and wildlife sanctuaries (Rodgers and Panwar, 1988). The state currently has seven wildlife sanctuaries, six national parks, four conservation reserves and one biosphere reserve. The following six protected area networks (PANs) situated at tropical, temperate and alpine zones from the Kumaun division were chosen for the present study:

Protected Area 1 (PA1) - Binsar Wildlife Sanctuary (BWLS)

Protected Area 2 (PA2) - Corbett Tiger Reserve (CTR)

Protected Area 3 (PA3) - Nandhaur Wildlife Sanctuary (NWLS)

Protected Area 4 (PA4) - Askot Wildlife Sanctuary (AWLS)

Protected Area 5 (PA5) - Nanda Devi Biosphere Reserve (NDBR)

Protected Area 6 (PA6) - Naina Devi Himalayan Bird Conservation Reserve (NDHBCR).

Figure 1 provides the location map of selected study sites within the state.

Data collection and identification of species. The study is a survey-based work conducted in six protected areas of Kumaun from July 2013 to June 2020, where insect sampling occurred on monthly basis along permanent linear transects (each measuring 300 m × 10 m) randomly distributed across each protected area. To ensure consistent sampling and comparison between sites, the study was divided into three distinct periods, each covering two years:

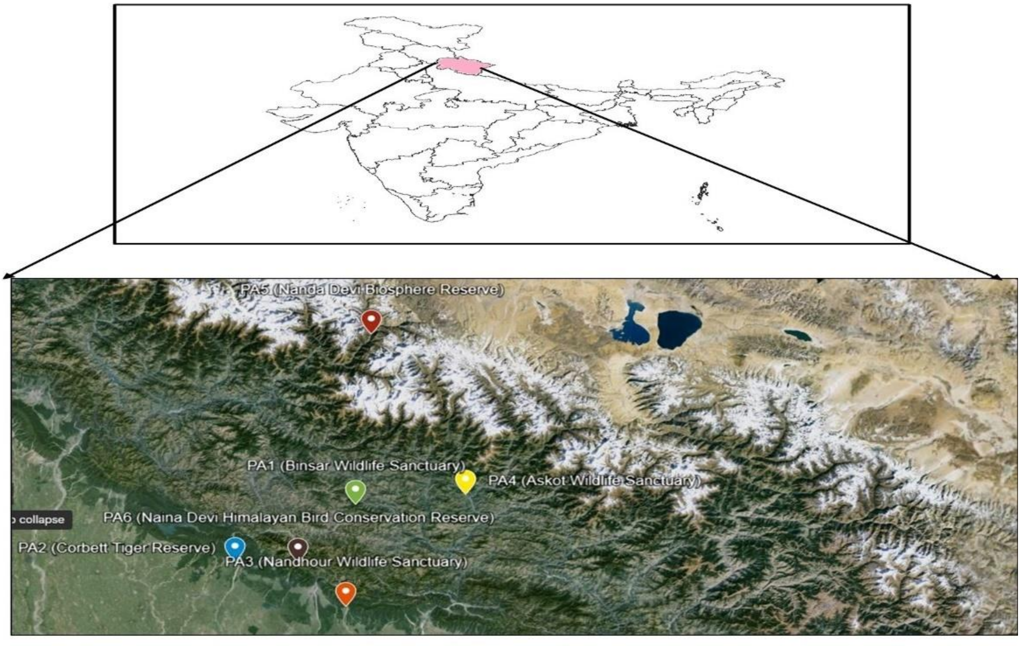

FIG. 1. Location map of selected study sites in study area. (Source: Google Earth)

- **2013-2014 and 2014-2015**: Askot Wildlife Sanctuary (AWLS) and Nanda Devi Biosphere Reserve (NDBR)
- **2015-2016 and 2016-2017**: Corbett Tiger Reserve (CTR) and Binsar Wildlife Sanctuary (BWLS)
- **2018-2019 and 2019-2020**: Nandhaur Wildlife Sanctuary (NWLS) and Naina Devi Himalayan Bird Conservation Reserve (NDHBCR)

Each site was sampled with equal intensity, ensuring consistency in the duration, number of transects and sampling effort across the respective two-year periods. This approach ensured that temporal and spatial biases were minimized, allowing a reliable comparison of insect abundance and diversity across the protected areas. Various techniques, such as modified Pollard walk, net sweeping, beating trays, baited pitfall traps, hand sorting, and light traps, were utilized from 8:00 am to 1:00 pm to estimate different taxonomic groups' abundances (Bhargav et al., 2009). Moth species were sampled between 7:30 pm and 9:30 pm using light traps with an 18 W incandescent lamp placed above a white entomological sheet. Species were identified using morphological descriptions from published literature and cross-checked with reference collections at the Insect Biodiversity Laboratory, Department of Zoology, D.S.B. Campus, Kumaun University, Nainital. Unidentifiable specimens were sent to the Northern Regional Station of the Zoological Survey of India in Dehradun and the Entomological Section at the Forest Research Institute in Dehradun for confirmation. Unknown species were categorized to the morphospecies level and recognized up to the genus level. Most butterfly species were visually identified in the field using published literature (Kehimkar, 2016; Sondhi and Kunte, 2018). Identified insects were then classified into different taxonomic groups to compile an inventory for the study sites.

Data analysis. The assemblage structure of insects was identified, using alpha diversity indices of insects, such as Shannon's Index (Hs) for species diversity, Margalef's Index (Hm) for species richness, and Simpson's Index (Ds) for species dominance using the program PAST 3.04 (Hammer et al., 2001). Bray-Curtis analysis was used to assess beta diversity by measuring the pairwise similarity of insect species abundance between the selected protected areas using the software Biodiversity Pro.

To compare occurrence of insect species in the proportion of protected areas, occupancy-abundance was calculated using Díaz et al. (2020) modified at a threshold of > 5 individuals. To standardize occupancy data, Normalized occupancy was then calculated and adjusted to a scale from 0 to 1. The thresholds were set based on the distribution of normalized occupancy values observed in preliminary analyses of the dataset. This measure helps allow for comparisons across different species (or their distribution patterns) or study areas.

Species Categorization. Species were categorized based on their normalized occupancy into three groups:

a) High Occupancy: Species with normalized occupancy > 0.7
b) Moderate Occupancy: Species with normalized occupancy between 0.5 and 0.7
c) Low Occupancy: Species with normalized occupancy < 0.5.

Results

Insect community structure across protected areas

During the study, a total of 412 insect species belonging to nine taxonomic orders and 70 families were identified throughout the study period. Order Lepidoptera was the richest in terms of relative number of species and individuals (37.37% species; 48.47% individuals), Coleoptera (19.66% species; 18.13% individuals), Hymenoptera (14.07% species; 17.32% individuals), Orthoptera (8% species; 5.36% individuals), Hemiptera (7.52% species; 2.56% individuals), Odonata (6.79% species; 4.8% individuals), Diptera (5.58% species; 2.17% individuals), Isoptera (0.48% species; 0.84% individuals) and Neuroptera (0.48% species; 0.31% individuals). Table 1 shows the distributional pattern of different insect species along six protected areas from the Kumaun division chosen for the study.

Of the reported total species, the maximum dominant were *Pieris brassicae* with a relative abundance of 5.15%, followed by *P. canidia* (4.73%), *Apis dorsata* (4.03%), *A. cerana* (3.33%), *Coccinella septumpunctata* (2.87%), *Aglais caschmirensis* (2.17%), *Apis florae*, *Catopsilia pomona* and *Coccinella septumpunctata vardivericata* (1.47% each), *Orthetrum sabina sabina* (1.36%), *Catopsilia pyranthe* (1.33%) and *Bombus haemorrhoidalis* (1.26%). Similarly, a total of 91 insect species were considered the least dominant with relative abundance of 0.03% each.

Diversity indices across different protected areas

Variations in species composition among different protected areas indicate Shannon’s species diversity (Hs) varied from 3.99 to 4.95. PA3 showed the highest species diversity (4.95), followed by PA2, PA1, PA4, PA5 and lowest in PA6 (3.99). Margalef’s species richness (Hm) was also found to be the highest in PA3 (27.76), followed by PA2 (21.73), PA4 (16.13), PA1 (14.74), PA6 (13.36) and the lowest in PA5 (12.5). Simpson’s dominance was also higher in PA3 (Ds = 0.98) and lower in PA6 (Ds = 0.97). Figure 2 shows the comparison of alpha diversity metrics across different protected areas.

The Bray-Curtis analysis approach for similarity, also revealed significant patterns of beta diversity among the studied sites.

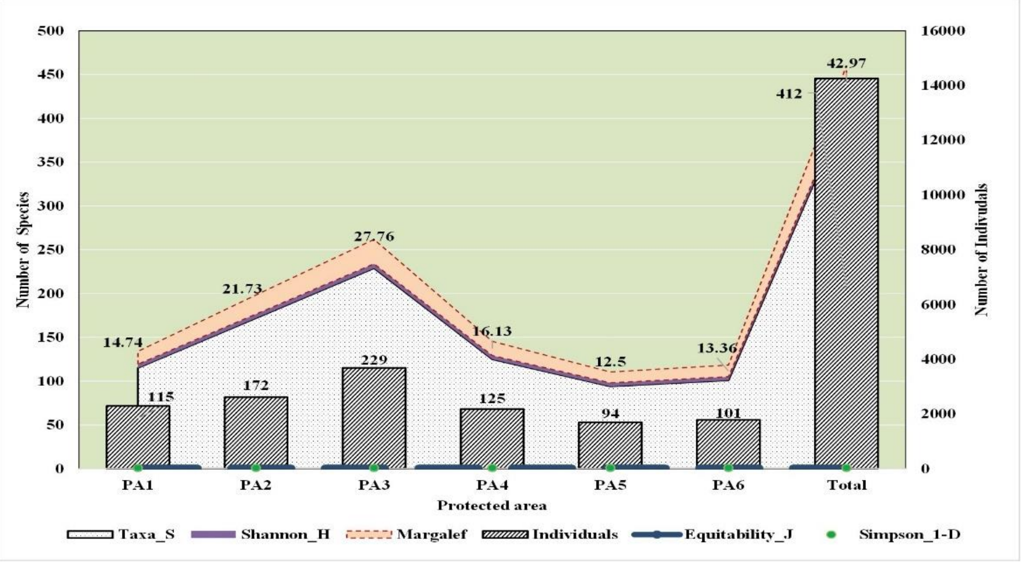

FIG. 2. Alpha diversity of insects in the PANs of Kumaun division of Uttarakhand.

The single linkage Bray- Curtis cluster analysis of species richness showed the % of similarity across the protected areas- showing two major clusters, the first cluster being PA3 and PA2, while the second cluster being PA1, PA4, PA5 and PA6. Single linkage cluster analysis depicted highest the beta diversity between PA5 and PA6 at 68.0% indicating similarities in environmental conditions or dispersal limitations for high compositional similarity of certain species, followed by PA2 and PA3 (58.56%), PA4 and PA5 (54.38%), PA1 and PA4 (50.87%) and PA1 and PA2 (39.11%). Figure 3 shows the Bray-Curtis similarity analysis of Beta diversity across protected areas.

Habitat specificity in species richness and status

PA1 (Binsar Wildlife Sanctuary) - In PA1, the Lepidoptera order was the highest with 43.47% contribution of species, followed by Coleoptera (20.0%), Hymenoptera (11.30%), Orthoptera (7.82%), Odonata (6.95%), Hemiptera and Diptera (5.21% each) orders.

PA2 (Corbett Tiger Reserve) - In the PA2, the Lepidoptera order was the highest with 38.95% contribution of species, followed by Coleoptera (15.11%), Hymenoptera and Orthoptera (11.04% each), Odonata (9.88%), Hemiptera (9.30%), Diptera (3.48%) and Neuroptera and Isoptera (0.58% each) orders.

PA3 (Nandhaur Wildlife Sanctuary) - In the PA3, the Lepidoptera order was the highest with 46.28% of species, followed by Coleoptera (18.34%), Hymenoptera (12.22%), Odonata (9.60%) Orthoptera (8.73%), Hemiptera (2.18%), Diptera (1.74% each) and Isoptera (0.87%).

PA4 (Askot Wildlife Sanctuary) - In the PA4, Lepidoptera was again the highest order with 38.4% contribution of species, followed by Coleoptera (18.4%), Hymenoptera (13.6%), Hemiptera and Diptera (8.0% each), Orthoptera (7.2% each), Odonata (5.6%), and Neuroptera (0.8% each).

PA5 (Nanda Devi Biosphere Reserve) and PA6 (Naina Devi Himalayan Bird Conservation Reserve) - in the PA5, the Lepidoptera order was the highest with

46.80 % contribution of species, followed by Coleoptera and Hymenoptera (17.0% each), Orthoptera and Diptera (5.31% each), Hemiptera and Odonata (4.25% each), whereas in the PA6, Lepidoptera was the highest order with 56.43% contribution of species, followed by Coleoptera (12.87%), Hymenoptera (10.89%), Orthoptera (6.93% each), Odonata and Hemiptera (4.95% each), Diptera (2.97%). The protected areas highlight no particular species confined within their boundaries. A comprehensive list of species recorded in each particular protected area has been listed in Table 2.

Species-wise occupancy and abundance patterns

Considering overall richness of species, common populations such as *Colias fieldii* Menetries, *Neptis sankara* (Kollar), *Coccinella septumpunctata* Linnaeus (Order: Coleoptera), *Crocothemis servilia servilia* (Drury), *Orthetrum glaucum* Brauer (Order: Odonata) were found to be present in all the chosen study sites (Table 1). However, in terms of species occupancy and abundance with >5 individuals, overall analysis of insect species across six protected areas in Uttarakhand revealed significant patterns:

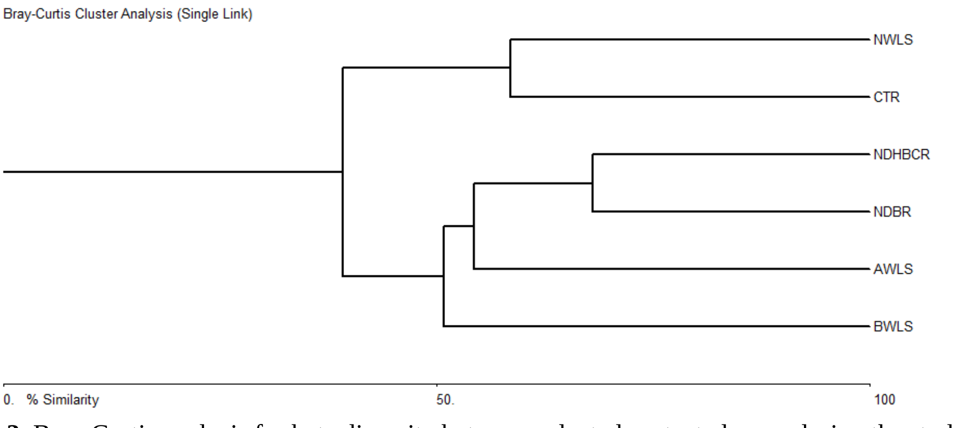

FIG. 3. Bray Curtis analysis for beta diversity between selected protected areas during the study period.

High Occupancy Species (Normalized Occupancy = 0.83)

Species with a normalized occupancy of 0.83 which were found in most, but not all, study sites. They are prevalent across most of the sites, indicating favorable habitat conditions but not universal adaptability. They include:

- Lepidoptera: *Aglais caschmirensis, Eurema brigitta, Macroglossum necteris, Vanessa cardui*
- Hymenoptera: *Apis dorsata*
- Orthoptera: *Paraconophyma scabra*.

Moderate Occupancy Species (Normalized Occupancy = 0.67)

Species with a normalized occupancy of 0.67 exhibited a less consistent distribution, suggesting potential specialization or sensitivity to specific environmental factors. These species include:

- Lepidoptera: *Aporia agathon*, *Dodona durga*, *Eurema hecabe*, *Gonepteryx nepalensis*, *Heliophorus sena*, *Melanitis leda*, *Papilio demoleus*, *P. bianor*, *P. polytes*, *Ypthima huebneri*
- Coleoptera: *Mylabris cichorii*

- Hymenoptera: *Apis cerana*
- Odonata: *Orthetrum sabina sabina*

These species may serve as indicators of specific habitat conditions or altitudinal zones.

Zero Occupancy Species

Certain species were absent from all sites where more than 5 individuals were recorded, indicating a normalized occupancy of 0. These species include:

- Lepidoptera: *Abisara bifasciata*, *A. fylla*, *Aeromachus stigmata*, *Agrius* sp., *Borbo bevani*, *Cepora nerissa*
- Coleoptera: *Adalia* sp., *Adelocera* sp., *Alcides* sp., *Anomala flavipes*, *Ateuchus* sp., among others.
- Hymenoptera: *Amegilla cingulata*, *Myzine dimidiata*, *Vespa* sp.
- Odonata: *Aristocypha quadrimaculata*, *Bayadera indica*, *Ischnura rubilio*
- Orthoptera: *Atractomorpha crenulata*, *Gastrimargus transversus*, *Heteropternis respondence*, and others.
- Hemiptera: *Anoplocnemis phasiana*, *Cletus punctulatus*, *Eurybrachus* sp., and others.
- Diptera: *Anthrrax* sp., *Bombylius* sp., *Lucilia* sp., and others.

Figure 4 illustrates species richness patterns across different study sites with respect to normalized occupancy levels (high, moderate and low). The number of species were higher in areas with low occupancy (L), with 234 species i.e. <0.5 normalized occupancy level, compared to medium (M) and high (H) occupancy levels, which have 40 and 13 species, respectively.

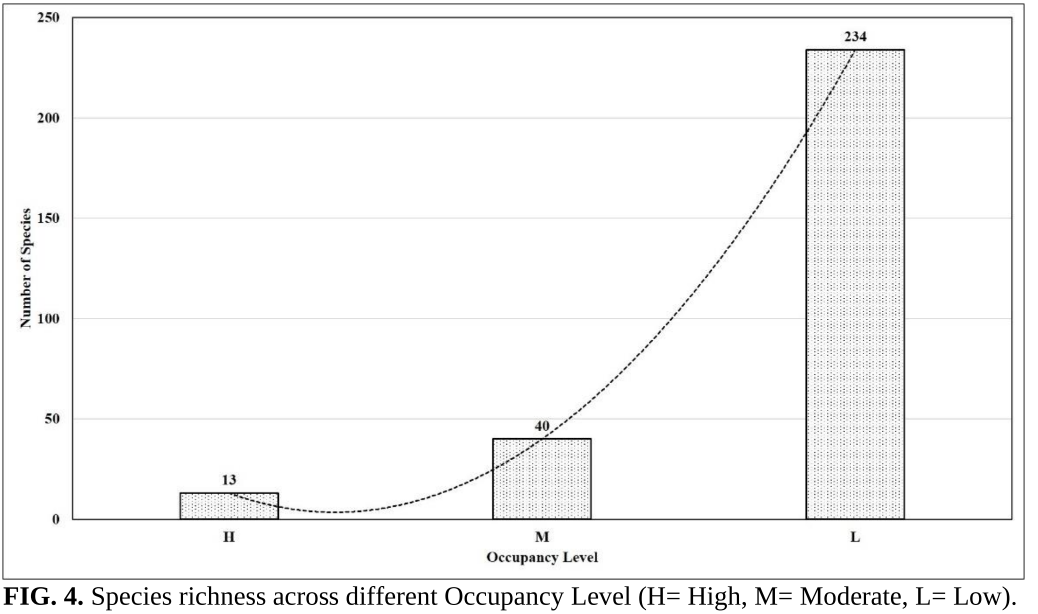

FIG. 4. Species richness across different Occupancy Level (H= High, M= Moderate, L= Low).

Discussion

Protected areas are crucial for understanding ecosystem health and management strategies. The present study contributes to the understanding of insect species richness and diversity within six protected areas. Similar to previous studies, (Joshi et al., 2008; Park et al., 2013; Verma and Arya, 2020), the order Lepidoptera exhibited the highest species richness, followed by the order Coleoptera.

The highest insect species diversity was observed in NWLS, whereas NDHBCR exhibited the lowest diversity. The Bray-Curtis similarity analysis revealed significant beta diversity, with environmental factors such as altitude and vegetation type influencing insect communities. The highest similarity was observed between PA5 and PA6 indicating similar environmental conditions or dispersal limitations. Comparative analysis of the recorded insect species richness of the selected protected areas from Kumaun division with the total insects record in the Indian Himalayan Region, represented about 1.66% of the total insect fauna (24,784 species) and 0.63% of the total insect fauna (65,047 species) in India (Chandra et al., 2018). This indicates that the protected areas studied hereby harbor a unique subset of the region's biodiversity. The observed pattern of species richness suggests a classic inverse relationship between species occupancy and abundance. The significant differences in species occupancy between sites underscore the importance of habitat heterogeneity in maintaining insect diversity. The majority of species are found at the low occupancy level, indicating that they are likely specialists with specific habitat requirements or limited distributions. These species, although numerous, are not widely distributed across the protected areas and may be dependent on particular ecological niches or microhabitats. This pattern is consistent with findings in other ecological studies where many species tend to be rare or have restricted ranges (Gaston, 1994). The moderate occupancy level (with 40 species), represents species that are somewhat more adaptable but still have specific habitat preferences that limit their distribution. These species may thrive in certain conditions that are not as widespread, suggesting a balance between habitat specialization and adaptability. In contrast, the high occupancy level, with only 13 species, includes those that are generalists and can thrive across a broad range of habitats. These species are highly adaptable and resilient to varying environmental conditions, which allows them to occupy multiple sites within the protected areas. Species like *Euploea core* and *Junonia iphita* were universally present, showcasing their adaptability. Conversely, species such as *Abisara bifasciata* and *Aeromachus stigmata* were absent from all sites, highlighting possible habitat or resource limitations. The relatively low number of such species underscores the ecological principle that generalist species are often fewer in number compared to specialists, as the latter have evolved to exploit specific ecological niches (Brown and Freitas, 2000).

These findings underline the need for targeted conservation strategies that address both specialist and generalist species. Protecting and restoring diverse habitats is crucial for sustaining insect populations and their ecological roles, such as pollination and nutrient cycling. The study highlights the importance of certain protected areas, like Nandhaur Wildlife Sanctuary, which show higher species richness, possibly due to better habitat quality, diversity of habitats, or more effective conservation practices. Conversely, areas with lower species richness may require targeted conservation efforts to enhance biodiversity. Long-term monitoring and research are thus essential for developing effective strategies to support insect diversity and ecosystem health.

TABLE 1. Species composition, distribution status and normalised occupancy of insects in the PANs of Kumaun division of Uttarakhand.

S. No.	Order	Family	Species name	PA1	PA2	PA3	PA4	PA5	PA6	R.A.	N.O.
1.	**Lepidoptera**	**Nymphalidae**	*Acraea issoria* (Hubner)	-	-	+	-	+	+	0.18	0.33
2.			*Aglais caschmirensis* (Kollar)	+	-	+	+	+	+	2.17	0.83
3.			*Argynnis childreni* Gray	+	-	-	-	+	-	0.17	0.33
4.			*Argynnis hyperbius* (Linnaeus)	+	-	-	-	-	-	0.07	0.17
5.			*Ariadne merione* (Cramer)	-	+	+	-	-	+	0.38	0.33
6.			*Athyma cama* Moore	-	-	+	+	-	+	0.24	0.33
7.			*Athyma perius* (Linnaeus)	-	-	+	+	-	-	0.14	0.17
8.			*Athyma zeroca* Moore	-	+	+	-	-	-	0.14	0.33
9.			*Aulocera swaha* Kollar	+	-	-	-	+	-	0.28	0.33
10.			*Aulocera padma* Kollar	+	-	-	-	-	+	0.10	0.17
11.			*Callerebia annada* (Moore)	+	-	-	-	+	+	0.17	0.17
12.			*Callerebia scanda* (Kollar)	+	-	-	-	+	+	0.21	0.33
13.			*Charaxes agrarius* Swinhoe	-	+	+	-	-	-	0.10	0.17
14.			*Charaxes bharata* Felder & Felder	-	+	+	-	-	-	0.21	0.33
15.			*Cyrestis thyodamas* Doyere	-	+	+	-	-	-	0.31	0.33
16.			*Danaus chrysippus* (Linnaeus)	+	+	+	+	-	+	0.52	0.33
17.			*Danaus genutia* (Cramer)	-	+	+	+	-	+	0.35	0.5
18.			*Euploea core* (Cramer)	+	+	+	+	+	+	0.77	1
19.			*Euploea midamus* (Linnaeus)	-	-	-	+	-	+	0.07	0
20.			*Euploea mulciber* (Cramer)	-	+	-	-	-	-	0.03	0
21.			*Euthalia aconthea* (Cramer)	-	+	+	-	+	-	0.10	0

22.			*Hestinalis nama* (Doubleday)	-	+	+	-	-	-	0.07	0
23.			*Hypolimnas bolina* (Linnaeus)	-	+	+	-	-	-	0.31	0.33
24.			*Junonia almana* (Linnaeus)	-	+	+	+	-	-	0.49	0.5
25.			*Junonia atlites* (Linnaeus)	-	+	+	-	-	-	0.14	0.33
26.			*Junonia iphita* (Cramer)	+	+	+	+	+	+	0.87	1
27.			*Junonia lemonias* (Linnaeus)	-	+	+	-	-	-	0.28	0.33
28.			*Junonia orithya* (Linnaeus)	-	+	+	+	-	+	0.31	5
29.			*Kallima inachus* (Doyere)	+	-	+	+	-	-	0.14	0.17
30.			*Kaniska canace* (Linnaeus)	+	-	+	-	-	-	0.07	0
31.			*Lasiommata schakra* (Kollar)	+	-	-	+	+	+	0.35	0.5
32.			*Lethe confusa* Aurivillius	-	-	+	+	-	+	0.17	0.17
33.			*Lethe verma* (Kollar)	+	-	-	-	-	-	0.14	0.17
34.			*Libythea lepita* Moore	-	-	+	-	-	-	0.03	0
35.			*Libythea* sp.	-	+	-	-	-	-	0.03	0
36.			*Melanitis leda* (Linnaeus)	-	+	+	+	-	+	0.49	0.67
37.			*Mycalesis* sp.	-	+	+	-	-	-	0.14	0.33
38.			*Neptis hylas* (Linnaeus)	-	+	+	-	+	-	0.10	0
39.			*Neptis sankara* (Kollar)	+	+	+	+	+	+	0.35	0.5
40.			*Neptis zaida* Doubleday	+	-	-	-	-	-	0.03	0
41.			*Parantica aglea* (Stoll)	-	+	+	+	-	+	0.14	0
42.			*Parantica sita* (Kollar)	-	-	-	+	-	-	0.03	0
43.			*Phalanta phalantha* (Drury)	+	+	+	+	+	+	0.87	1
44.			*Pseudergolis wedah* (Kollar)	+	-	-	-	+	+	0.42	5

45.			*Sephisa dichroa* (Kollar)	+	-	-	+	+	-	0.24	0.33
46.			*Symbrenthia lilaea* Hewitson	-	+	+	-	-	+	0.2	0.5
47.			*Tirumala limniace* (Cramer)	-	+	+	-	-	-	0.14	0.33
48.			*Vagrans egista* (Cramer)	-	-	+	-	-	-	0.07	0.17
49.			*Vanessa cardui* (Linnaeus)	+	-	+	+	+	+	0.70	0.83
50.			*Vanessa indica* (Herbst)	+	+	+	+	+	+	1.26	1
51.			*Ypthima huebneri* Kirby	-	+	+	-	+	+	0.28	0.67
52.			*Ypthima nareda nareda* (Kollar)	+	-	+	-	+	-	0.17	0.17
53.		**Lycaenidae**	*Arhopala amantes* (Hewitson)	-	-	+	-	-	-	0.07	0.17
54.			*Castalius rosimon* (Fabricius)	-	+	+	-	-	-	0.28	0.33
55.			*Euchrysops cnejus* (Fabricius)	-	+	-	-	-	-	0.07	0.17
56.			*Flos asoka* (de Niceville)	-	+	+	-	+	+	0.21	0.33
57.			*Freyeria trochylus* (Freyer)	-	+	-	-	-	-	0.10	0.17
58.			*Heliophorus sena* (Kollar)	+	+	+	+	+	+	0.73	0.67
59.			*Jamides celeno* (Cramer)	-	-	+	-	-	-	0.03	0
60.			*Lampides boeticus* (Linnaeus)	-	-	+	-	+	+	0.52	0.5
61.			*Loxura atymnus* (Stoll)	-	-	+	-	-	+	0.28	0.33
62.			*Lycaena pavana* (Westwood)	+	-	-	+	+	+	0.38	0.33
63.			*Lycaena phlaeas* (Linnaeus)	-	-	-	+	-	+	0.07	0
64.			*Megisba malaya* (Horsfield)	-	+	-	-	-	-	0.03	0
65.			*Pseudozizeeria maha* (Kollar)	-	+	+	-	-	-	0.07	0
66.			*Spindasis* sp.	-	+	-	-	-	-	0.03	0
67.			*Talicada nyseus* (Guerin-Meneville)	+	-	+	-	-	-	0.21	0.17
68.			*Tarucus indica* Evans	-	+	-	-	-	-	0.14	0.17

69.			*Tarucus nara* (Kollar)	-	-	+	-	-	-	0.03	0
70.			*Udara dilectus* Moore	+	-	-	-	-	+	0.07	0
71.			*Zizeeria karsandra* (Moore)	-	+	+	-	-	-	0.07	0
72.			*Zizina otis* (Fabricius)	-	+	+	-	-	-	0.07	0
73.			*Zizula hylax* (Fabricius)	-	-	+	-	-	-	0.03	0
74.		**Pieridae**	*Aporia agathon* (Gray)	+	-	-	+	+	+	1.05	0.67
75.			*Belenois aurota* (Fabricius)	+	-	-	-	+	-	0.59	0.5
76.			*Catopsilia pomona* (Fabricius)	+	+	+	-	-	-	1.47	0.33
77.			*Catopsilia pyranthe* (Linnaeus)	-	+	+	-	-	-	1.33	0.67
78.			*Cepora nerissa* (Fabricius)	-	+	+	-	-	-	0.07	0
79.			*Colias erate* (Esper)	-	+	+	-	+	-	0.10	0
80.			*Colias fieldii* Menetries	+	+	+	+	+	+	0.63	0.5
81.			*Delias eucharis* (Drury)	-	+	+	-	-	-	0.70	0.33
82.			*Eurema andersonii* (Moore)	-	+	+	-	+	-	0.21	5
83.			*Eurema blanda* (Boisduval)	-	+	+	-	+	-	0.42	5
84.			*Eurema brigitta* (Stoll)	+	+	+	+	-	+	0.35	0.83
85.			*Eurema hecabe* (Linnaeus)	+	+	+	+	-	+	0.77	0.67
86.			*Eurema laeta* (Boisduval)	+	+	+	-	-	-	0.10	0
87.			*Gonepteryx nepalensis* Doubleday	+	-	+	+	+	-	0.73	0.67
88.			*Leptosia nina* (Fabricius)	-	+	-	-	-	-	0.03	0
89.			*Pareronia hippia* (Fabricius)	-	+	+	-	-	-	0.07	0
90.			*Pieris brassicae* (Linnaeus)	+	+	+	+	+	+	5.15	1
91.			*Pieris canidia* (Linnaeus)	+	+	+	+	+	+	4.73	1
92.			*Pontia daplidice* (Linnaeus)	+	+	-	-	+	+	0.38	0.33

93.		**Hesperiidae**	*Aeromachus stigmata* (Moore)	-	-	+	-	-	-	0.03	0
94.			*Borbo bevani* (Moore)	-	-	+	-	-	-	0.03	0
95.			*Ochlodes brahma* Moore	+	-	-	-	-	-	0.03	0
96.			*Parnara guttatus* (Moore)	-	+	+	-	-	-	0.14	0.33
97.			*Potanthus dara* (Kollar)	-	-	+	-	-	+	0.14	0.33
98.			*Pseudocoladenia fatih* (Kollar)	-	-	+	-	-	-	0.07	0.17
99.			*Sarangesa dasahara* Moore	-	+	-	-	-	-	0.07	0.17
100.			*Sarangesa purendra* Moore	-	+	-	-	-	-	0.07	0.17
101.			*Spilalia galba* (Fabricius)	-	+	-	-	-	-	0.07	0.17
102.			*Tagiades cohaerens cynthia* Evans	+	-	-	-	+	-	0.07	0
103.			*Tagiades litigiosa* Moschler	-	-	+	-	-	+	0.10	0.17
104.			*Telicota bambusae* (Moore)	-	-	+	-	-	+	0.10	0.17
105.			*Telicota* sp.	-	+	-	-	-	+	0.07	0
106.			*Udaspes folus* (Cramer)	-	-	+	-	-	-	0.03	0
107.		**Papilionidae**	*Byasa polyeuctes letincius* (Fruhstorfer)	+	-	-	-	+	+	0.56	0.5
108.			*Graphium cloanthus* (Westwood)	-	-	-	+	-	-	0.31	0.17
109.			*Graphium nomius* (Esper)	-	+	+	-	-	-	0.45	0.33
110.			*Graphium sarpedon* (Linnaeus)	-	+	+	+	-	-	0.66	0.33
111.			*Papilio bianor* Cramer	+	-	+	+	+	+	0.45	0.67
112.			*Papilio clytia* (Linnaeus)	-	+	+	-	-	-	0.07	0
113.			*Papilio demoleus* (Linnaeus)	-	+	+	-	+	+	0.91	0.67
114.			*Papilio machaon* Linnaeus	-	-	-	+	-	+	0.10	0.17
115.			*Papilio polytes* (Linnaeus)	+	+	+	+	+	+	0.66	0.67

116.			*Papilio protenor* Cramer	-	-	+	+	+	-	0.35	0.33
117.		**Riodinidae**	*Abisara bifasciata* Moore	-	-	+	-	-	-	0.03	0
118.			*Abisara echerius* (Stoll)	-	+	-	-	-	+	0.10	0.17
119.			*Abisara fylla* (Westwood)	-	-	-	+	-	-	0.03	0
120.			*Dodona durga* (Kollar)	+	-	+	+	+	+	0.49	0.67
121.			*Dodona eugenes* Bates	+	-	-	-	+	+	0.10	0
122.			*Dodona ouida* Hewitson	+	-	-	-	-	-	0.03	0
123.			*Zemeros flegyas* (Cramer)	-	+	+	-	-	-	0.07	0
124.		**Erebidae**	*Calpe ophideroides* Guenee	+	-	-	-	+	+	0.14	0.17
125.			*Creatonotos transiens* (Walker)	-	-	+	+	-	-	0.10	0.17
126.			*Cyana bellissima* (Kollar)	-	-	+	-	-	+	0.10	0.17
127.			*Cyana detrita* Walker	-	-	+	-	-	+	0.10	0.17
128.			*Episteme adulatrix* (Kollar)	-	-	+	-	-	-	0.07	0.17
129.			*Erebus caprimulgus* (Fabricius)	-	-	+	-	-	-	0.03	0
130.			*Eressa confinis* (Walker)	-	-	+	-	-	-	0.03	0
131.			*Fodina pallula* Guenee	-	-	+	-	-	-	0.03	0
132.			*Lemyra* sp.	-	-	-	+	-	-	0.03	0
133.			*Machrobrochis prasena* (Moore)	-	-	+	-	-	+	0.10	0.17
134.			*Nyctemera adversata* (Schaller)	-	-	+	+	-	+	0.17	0.33
135.			*Nyctemera* sp.	-	-	-	+	-	+	0.10	0.17
136.			*Spirama retorta* Clerck	-	-	+	-	-	-	0.07	0.17
137.			*Syntomoides imaon* Cramer	+	-	+	+	+	-	0.38	0.33
138.			*Trigonodes hyppasia* Cramer	-	-	+	-	-	-	0.03	0
139.			*Vamuna remelana* (Moore)	-	-	+	-	-	-	0.03	0
140.		**Sphingidae**	*Agrius* sp.	-	-	-	+	-	-	0.03	0

141.			*Daphnis nerii* (Linnaeus)	-	-	+	-	-	-	0.03	0
142.			*Hemaris* sp.	-	-	-	+	-	-	0.03	0
143.			*Macroglossum necteris* Kollar	+	-	+	+	+	+	0.52	0.83
144.			*Sphinx* sp.	-	-	-	+	-	-	0.03	0
145.			*Theretra nessus* (Drury)	-	-	+	-	-	-	0.03	0
146.		**Crambidae**	*Bradina diagonalis* (Guenee)	-	-	+	-	-	-	0.07	0.17
147.			*Cnaphalocrocis medinalis* (Guenee)	-	-	+	-	-	-	0.07	0.17
148.			*Paliga damastesalis* (Walker)	+	-	-	-	-	-	0.07	0.17
149.			*Spoladea recurvalis* (Fabricius)	-	-	+	-	-	-	0.07	0.17
150.			*Tyspanodes linealis* (Moore)	-	-	+	-	-	-	0.07	0.17
151.		**Geometridae**	*Dysphania militaris* (Linnaeus)	+	-	-	-	-	-	0.07	0.17
152.			*Ourapteryx clara* (Butler)	-	-	+	-	-	-	0.07	0.17
153.		**Eupterotidae**	*Eupterote* sp.	-	-	+	-	-	-	0.07	0.17
154.		**Saturnidae**	*Actias selene* Hubner	-	-	+	-	-	-	0.07	0.17
155.	**Coleoptera**	**Scarabaeidae**	*Anomala antique* (Gyllental)	-	+	+	-	-	-	0.14	0.33
156.			*Anomala decipiens* (Arrow)	-	+	+	-	-	-	0.21	0.33
157.			*Anomala dimidiata* Hope	-	-	-	+	+	+	0.73	0.5
158.			*Anomala flavipes* Arrow	-	+	-	-	-	-	0.03	0
159.			*Anomala lineatopennis* Blanchard	+	-	-	+	+	-	0.52	0.5
160.			*Anomala* sp.	+	-	-	-	+	+	0.10	0
161.			*Ateuchus* sp.	-	+	-	-	-	-	0.03	0
162.			*Catharsius capucinus* (Fabricius)	-	-	+	-	-	-	0.03	0
163.			*Chiloba acuta* Wied	-	-	-	+	-	-	0.07	0.17

164.			*Clinteria spilota* Hope	-	-	-	+	-	-	0.07	0.17
165.			*Copris sacontala* Redtenbacher	-	+	+	-	-	-	0.14	0.33
166.			*Dsygnathus* sp.	-	-	-	+	-	-	0.07	0.17
167.			*Gymnopleurus subtilis* Walker	+	-	-	-	-	-	0.17	0.17
168.			*Gymnopleurus miliaris* (Fabricius)	-	+	+	-	-	+	0.77	0.5
169.			*Heliocopris bucephalus* (Fabricius)	-	-	+	-	-	-	0.03	0
170.			*Jumnos roylei* Hope	+	-	-	+	+	-	0.24	0.17
171.			*Lachnosterna cavifrons* Brenske	+	-	-	-	-	-	0.03	0
172.			*Lepidiota albistigma* Burmeister	-	+	+	-	-	-	0.14	0.33
173.			*Lytta limbata* Redtenbacher	+	-	-	-	-	-	0.07	0.17
174.			*Melolontha cuprescens* Blanchard	-	-	+	-	-	-	0.17	0.17
175.			*Onthophagus dama* (Fabricius)	-	-	+	+	-	-	0.42	0.33
176.			*Onthophagus gagates* Hope	+	-	-	-	-	-	0.14	0.17
177.			*Onitis falcatus* Wulfen	-	+	+	-	-	-	0.35	0.33
178.			*Oryctes nasicornis* (Linnaeus)	-	-	+	-	-	-	0.03	0
179.			*Oxycertonia versicolor* Fabricius	-	-	-	+	-	-	0.03	0
180.			*Protaetia pretiosa* Nonfried	+	-	+	-	-	-	0.24	0.33
181.			*Protaetia neglacta* Hope	+	-	-	-	+	+	0.24	0.33
182.			*Popilla cupricollis* Hope	-	-	-	+	-	-	0.03	0
183.			*Popillia* sp.	-	-	+	+	+	-	0.42	0.5
184.			*Pseudolucanus cantor* Hope	+	-	-	-	-	-	0.03	0
185.			*Sisyphus hirtus* Wied	-	-	-	+	-	-	0.03	0
186.			*Torynorrhina opalina* Hope	-	-	-	+	-	-	0.03	0
187.		**Chrysomelidae**	*Altica caerulescens* (Baly)	-	-	-	+	-	+	0.63	0.33

188.			*Altica himensis* Shukla	+	-	-	-	+	-	0.59	0.33
189.			*Charidotella* sp.	-	+	+	-	-	-	0.10	0.17
190.			*Colasposoma metallicum* (Clark)	-	+	+	-	-	-	0.10	0.17
191.			*Colasposoma splendidum* Fabricius	-	+	-	-	-	-	0.14	0.17
192.			*Corynodes peregrius* (Fuessly)	-	+	+	-	-	-	0.21	0.33
193.			*Gallerucida rutilans* Hope	+	-	-	-	-	-	0.03	0
194.			*Meristata sexmaculata* (Kollar & Redtenbacher)	+	-	-	-	+	-	0.28	0.33
195.			*Meristata trifasciata* Hope	+	-	+	-	+	+	0.28	0.67
196.			*Mimastra* sp.	+	-	-	+	+	-	0.21	0.5
197.			*Zygogramma bicolorata* Pallister	-	+	+	-	-	-	0.63	0.33
198.		**Coccinelidae**	*Adalia* sp.	-	-	+	-	-	-	0.03	0
199.			*Chilocorus infernalis* (Mulsant)	-	-	+	-	-	-	0.03	0
200.			*Coccinella septumpunctata vardivericata* Olivier	-	-	-	+	+	+	1.47	0.5
201.			*Coccinella septumpunctata* Linnaeus	+	+	+	+	+	+	2.87	1
202.			*Coccinella transversalis* (Fabricius)	-	-	+	-	-	-	0.07	0.17
203.			*Haluzia sanscrieta* Mulsant	+	-	-	-	+	+	0.31	0.5
204.			*Hippodamia varegata* Goeze	-	-	-	+	+	-	0.28	0.33
205.			*Leis dimidiata* (Fabricius)	-	+	+	-	-	-	0.07	0
206.			*Menochilus sexmaculatus* (Fabricius)	-	-	+	+	-	+	0.21	0.5
207.			*Oenopia kirbyi* (Mulsant)	-	-	-	+	-	-	0.03	0
208.			*Psyllora vigintiduopunctata* (Linnaeus)	-	-	+	-	-	-	0.03	0

209.		**Meloidae**	*Epicauta mannerheimi* (Maklin)	-	+	+	-	-	-	0.07	0
210.			*Epicauta* sp.	-	-	+	-	-	-	0.03	0
211.			*Hycleus* sp.	-	-	+	-	-	-	0.03	0
212.			*Mylabris cichorii* (Linnaeus)	+	+	+	+	-	-	0.52	0.67
213.			*Mylabris pustulata* (Thunberg)	-	-	+	-	+	-	0.42	0.33
214.			*Mylabris* sp.	+	-	-	-	-	+	0.28	0.33
215.		**Carabidae**	*Chlaenius* sp.	-	-	+	-	-	-	0.03	0
216.			*Ophonus indicus* Bates	-	+	-	-	-	-	0.03	0
217.			*Ophonus rufibarbis* Fabricius	-	-	+	-	-	-	0.03	0
218.			*Ophonus rubricollis* Hope	+	-	-	+	+	+	0.17	0.17
219.			*Pheropsophus* sp.	-	-	-	+	-	-	0.03	0
220.			*Scarites sulcatus* Olivier	+	-	+	-	-	-	0.07	0
221.			*Scarities* sp.	-	-	-	+	-	-	0.03	0
222.		**Cicindelidae**	*Calomera chloris* Hope	-	+	+	-	-	-	0.14	0.33
223.			*Cicindela flexuosa* (Fabricius)	-	-	+	-	-	-	0.21	0.17
224.			*Cicindela* sp.	-	+	-	-	-	-	0.21	0.17
225.			*Cosmodela intermedia* Chaudoir	-	+	+	-	-	-	0.28	0.33
226.		**Tenebrionidae**	*Cistelomorpha* sp.	+	-	-	-	-	+	0.21	0.33
227.			*Gonocephalum* sp.	-	+	+	-	-	-	0.42	0.33
228.		**Elateridae**	*Adelocera* sp.	-	+	+	-	-	-	0.07	0
229.			*Heteroderes macroderes* Candeze	-	+	+	-	-	-	0.07	0
230.		**Hydrophilidae**	*Hydrophilus* sp.	-	+	-	-	-	-	0.03	0
231.			*Hydrophilus triangularis* Say	-	-	+	-	-	-	0.03	0
232.		**Lagriidae**	*Cerogria nepalensis* Hope	+	-	-	-	-	-	0.03	0

233.		**Curculionidae**	*Alcides* sp.	-	+	-	-	-	-	0.03	0
234.		**Lucanidae**	*Metopodontus biplagiatus* (Westwood)	-	-	+	-	-	-	0.03	0
235.		**Cerambycidae**	*Dorysthenes huegelii* (Redtenbacher)	-	-	+	-	-	-	0.03	0
236.	**Hymenoptera**	**Apidae**	*Amegilla cingulata* (Fabricius)	-	+	+	-	-	-	0.07	0
237.			*Anthophora cofusa* Smith	+	-	-	-	+	-	0.14	0.33
238.			*Anthophora* sp.	-	-	-	+	-	-	0.07	0.17
239.			*Apis cerana* Fabricius	-	-	+	+	+	+	3.33	0.67
240.			*Apis dorsata* Fabricius	-	+	+	+	+	+	4.03	0.83
241.			*Apis florae* Fabricius	-	-	+	+	-	-	1.47	0.33
242.			*Apis laboriosa* Smith	+	-	+	+	+	+	0.35	0.17
243.			*Apis mellifera* Eschscholtz	-	-	+	+	-	-	0.73	0.33
244.			*Bombus haemorrhoidalis* Smith	-	-	+	-	+	+	1.26	0.5
245.			*Bombus* sp.	+	-	-	-	-	-	0.14	0.17
246.			*Bremus* sp.	-	-	-	+	+	-	0.14	0.33
247.			*Coelioxy* sp.	+	-	-	-	+	-	0.10	0.17
248.			*Crocisa ramosa* Lepeletier	+	-	+	-	-	+	0.42	0.5
249.			*Eriades decipiences* Spinola	-	+	-	-	-	-	0.07	0.17
250.		**Scoliidae**	*Campsomeriella collaris* (Fabricius)	-	-	-	+	-	-	0.07	0.17
251.			*Compsomeris asiatica himalaya* Bar.	+	-	-	-	+	-	0.10	0.17
252.			*Compsomeris prismatica* Smith	-	+	-	-	-	-	0.14	0.17
253.			*Myzine dimidiata* Guerin	-	+	-	-	-	-	0.03	0

254.			*Myzine petiolata* Smith	-	+	-	-	-	-	0.03	0
255.			*Phalerimeris* sp.	-	-	+	-	-	-	0.07	0.17
256.			*Scolia affinis* Guerin	-	+	+	-	+	-	0.38	0.5
257.			*Scolia* sp.	-	-	+	-	-	+	0.10	0.17
258.			*Scolia venusta* Smith	+	-	-	-	-	-	0.24	0.17
259.		**Sphecidae**	*Sceliphron caucasium* (Drury)	-	+	+	-	-	-	0.10	0.17
260.			*Sceliphron coromandelicum* Lepeletier	-	+	+	-	-	-	0.10	0.17
261.			*Sceliphron* sp.	-	-	+	-	-	-	0.07	0.17
262.			*Sphex umbrosus* Christ	-	+	-	-	-	-	0.07	0.17
263.			*Sphex* sp.	-	-	+	-	-	-	0.07	0.17
264.		**Vespidae**	*Delta dimidiatipennis* (Saussure)	-	-	-	+	-	-	0.07	0.17
265.			*Eumenes petiolata* (Fabricius)	-	+	-	-	-	-	0.07	0.17
266.			*Labus* sp.	-	+	+	-	-	-	0.10	0.17
267.			*Mandarinia* sp.	-	-	-	+	-	-	0.07	0.17
268.			*Polistes dorsalis* (Fabricius)	-	-	+	-	+	+	0.14	0.17
269.			*Polistes hebraeus* Fabricius	-	-	-	+	-	-	0.07	0.17
270.			*Polistes* sp.	-	-	-	+	-	-	0.07	0.17
271.			*Polistes stigma* (Fabricius)	-	+	+	-	-	-	0.10	0.17
272.			*Vespa cincta* Fabricius	-	+	-	-	-	-	0.07	0.17
273.			*Vespa* sp.	+	-	-	-	-	-	0.03	0
274.			*Vespa velutina* Lepeletier	-	-	+	-	+	-	0.10	0.17
275.			*Vespa velutina auraria* Smith	-	+	-	-	-	+	0.07	0
276.			*Vespula flaviceps* (Smith)	-	-	+	-	-	-	0.07	0.17

277.		**Formicidae**	*Campanotus* sp.	-	-	-	+	-	-	0.07	0.17
278.			*Camponotus compressus* (Fabricius)	-	+	+	-	-	-	0.14	0.33
279.			*Diacomma* sp.	-	-	-	+	-	-	0.07	0.17
280.			*Pachycondyla* sp.	-	-	-	+	-	-	0.07	0.17
281.			*Polyrhachis simplex* Mayr	-	+	+	-	-	-	0.10	0.17
282.			*Polyrhachis* sp.	-	-	-	+	-	-	0.07	0.17
283.		**Xylocopidae**	*Xylocopa auripennis* Lepeletier	-	+	+	-	-	+	0.17	0.33
284.			*Xylocopa fenestrata* Faber.	+	-	-	+	+	-	0.63	0.5
285.		**Halictidae**	*Halictus* sp.	-	-	+	-	-	+	0.10	0.17
286.			*Nomia curvipes* (Fabricius)	-	+	+	-	-	-	0.10	0.17
287.		**Pompilidae**	*Pepsis* sp.	-	-	+	-	-	-	0.07	0.17
288.			*Salius flavus* Fabricius	+	-	-	-	+	-	0.10	0.17
289.		**Ichneumonidae**	*Ichneumon* sp.	+	-	-	-	+	-	0.10	0.17
290.			*Ophion* sp.	+	-	-	-	+	-	0.10	0.17
291.		**Sphecidae**	*Ammophila atripes* Smith	-	-	+	-	-	-	0.07	0.17
292.			*Ammophila punctata* Smith	+	-	-	-	+	+	0.24	0.17
293.		**Andrenidae**	*Andrena cineraria* (Linnaeus)	-	-	+	-	-	-	0.07	0.17
294.	**Odonata**	**Libellullidae**	*Acisoma panorpoides panorpoides* Rambur	-	+	+	-	-	-	0.10	0.17
295.			*Aethriamanta brevipennis* (Rambur)	-	+	+	-	-	-	0.10	0.17
296.			*Brachythemis contaminata* (Fabricius)	-	+	+	-	-	-	0.10	0.17

297.			*Crocothemis servilia servilia* (Drury)	+	+	+	+	+	+	0.28	0.33
298.			*Orthetrum glaucum* Brauer	+	+	+	+	+	+	0.24	0.17
299.			*Orthetrum pruinosum* (Burmeister)	-	-	+	-	-	-	0.07	0.17
300.			*Orthetrum pruinosum neglectum* (Rambur)	+	+	-	+	+	+	0.77	0.5
301.			*Orthetrum taeniolatum* (Schneider)	+	+	+	-	-	-	0.14	0.17
302.			*Orthetrum sabina sabina* (Drury)	+	+	+	+	-	+	1.36	0.67
303.			*Orthetrum triangulare triangulare* (Selys)	-	+	+	-	-	-	0.14	0.33
304.			*Palpopleura sexmaculata sexmaculata* (Fabricius)	-	+	+	+	-	-	0.14	0.17
305.			*Pantala flavescens* (Fabricius)	-	+	+	-	-	-	0.10	0.17
306.			*Rhodothemis rufa* (Rambur)	-	-	+	-	-	-	0.07	0.17
307.			*Symptrum commixtum* Selys	-	-	-	+	-	+	0.07	0
308.			*Tholymis tillarga* (Fabricius)	-	+	-	-	-	-	0.03	0
309.			*Trithemis festiva* (Rambur)	-	+	+	-	-	-	0.10	0.17
310.			*Trithemis pallidinervis* (Kirby)	-	+	+	-	-	-	0.10	0.17
311.		**Coenagrionidae**	*Ceriagrion coromandelianum* (Fabricius)	-	+	+	-	-	-	0.10	0.17
312.			*Ischnura rubilio* Selys	-	-	+	-	-	-	0.03	0
313.			*Pseudagrion australasiae* Selys	-	-	+	-	-	-	0.07	0.17
314.			*Pseudagrion rubriceps rubriceps* Selys	-	+	+	-	-	-	0.10	0.17
315.		**Chlorocyphidae**	*Aristocypha fenestrella* Rambur	-	-	+	-	-	-	0.07	0.17

316.			*Aristocypha quadrimaculata* (Selys)	-	-	+	-	-	-	0.03	0
317.			*Paracypha unimaculata* (Selys)	-	-	+	-	-	-	0.07	0.17
318.		**Aeschnidae**	*Anaximma culiforns* Rambur	+	-	-	+	+	-	0.14	0.17
319.		**Euphaeidae**	*Bayadera indica* (Selys)	+	-	-	-	-	-	0.03	0
320.		**Synlestidae**	*Megalestes major* Selys	+	-	-	-	-	-	0.03	0
321.		**Calopterygidae**	*Neurobasis chinensis* (Linnaeus)	-	+	+	-	-	-	0.14	0.33
322.	**Orthoptera**	**Acrididae**	*Acrida exaltata* (Walker)	-	+	+	-	-	-	0.10	0.17
323.			*Aulacobothrus leutipus* Walker	-	-	+	+	+	+	0.35	0.33
324.			*Ceracris fasciata* (Brunner ven Wattenwyl)	-	+	+	-	-	-	0.10	0.17
325.			*Choroedocus illustris* (Walker)	-	+	+	-	-	-	0.10	0.17
326.			*Chorthippus almoranus* Uvarov	+	-	-	-	-	-	0.45	0.17
327.			*Cyrtacanthacris tatarica* (Linnaeus)	-	+	-	-	-	-	0.07	0.17
328.			*Diobolocantops innotabilis* (Walker)	-	+	+	-	-	-	0.10	0.17
329.			*Gastrimargus africanus africanus* (Saussure)	-	+	+	+	-	+	0.17	0.17
330.			*Gastrimargus transversus* Thunberg	+	-	-	-	-	-	0.03	0
331.			*Heteropternis respondence* (Walker)	+	-	-	-	-	-	0.03	0
332.			*Oedaleus* sp.	-	+	-	+	-	-	0.14	0.17
333.			*Oxya* sp.	-	-	+	-	-	-	0.07	0.17
334.			*Oxyrrheps obusta* (Haan)	-	+	-	-	-	-	0.03	0

335.			*Paraconophyma scabra* Walker	+	+	+	+	+	+	1.12	0.83
336.			*Patanga japonica* (Bolivar)	+	+	-	-	-	+	0.14	0.17
337.			*Phlaeoba antennata* Brunner	-	+	+	-	-	-	0.10	0.17
338.			*Phlaeoba infumata* Brunner	-	+	-	+	-	-	0.17	0.33
339.			*Phlaeoba panteli* Bolivar	-	-	+	-	-	-	0.07	0.17
340.			*Pternoscirta cinctifemur* Walker	+	+	-	-	-	-	0.10	0.17
341.			*Spathosternum prasiniferum prasiniferum* (Walker)	+	+	-	+	+	+	0.63	0.5
342.			*Sphingonotus longipennis* Saussure	-	+	+	-	-	-	0.10	0.17
343.			*Trilophidia annulata* Thunberg	-	+	-	-	-	-	0.03	0
344.			*Tyltropidius varicornis* (Walker)	-	-	+	-	-	-	0.07	0.17
345.			*Xenocatantops humilis humilis* (Serville)	-	-	+	-	-	+	0.10	0.17
346.			*Xenocatantops karnyi* Kirby	+	-	-	+	+	-	0.17	0.33
347.		**Tettigonidae**	*Elimaea* sp.	-	-	-	+	-	-	0.07	0.17
348.			*Letana linearis* (Walker)	+	-	-	-	+	-	0.10	0.17
349.			*Phaneroptera* sp.	-	-	+	-	-	-	0.07	0.17
350.		**Pyrgomorphidae**	*Atractomorpha crenulata* (Fabricius)	-	+	-	-	-	-	0.03	0
351.			*Aularches miliaris miliaris* (Linnaeus)	-	+	+	+	-	-	0.17	0.33
352.			*Chrotogonus trachypterus trachypterus* (Blanchard)	-	+	+	-	-	+	0.14	0.17
353.		**Gryllidae**	*Gryllus* sp.	-	-	+	-	-	-	0.07	0.17

354.			*Teleogryllus testaceus* (Walker)	-	-	+	-	-	-	0.07	0.17
355.	**Hemiptera**	**Coreidae**	*Anaplocne mispasina* Fabricius	-	-	-	+	-	-	0.07	0.17
356.			*Anoplocnemis phasiana* Fabricius	-	+	-	-	-	-	0.03	0
357.			*Cletus* sp.	-	-	-	+	-	-	0.07	0.17
358.			*Leptocorisa varicornis* Fabricius	-	+	-	-	-	-	0.03	0
359.			*Ochrochira albiditarsis* Westwood	+	-	-	-	-	-	0.03	0
360.			*Serinetha augur* (Fabricius)	-	+	+	-	-	-	0.10	0.17
361.		**Pentatomidae**	*Dalpada* sp.	+	+	-	+	+	+	0.31	0.33
362.			*Erthesina fullo* Thunberg	+	+	-	-	+	+	0.17	0.17
363.			*Murgantia histrionic* Hahn	-	-	-	+	-	-	0.03	0
364.			*Nezara viridula* Linnaeus	-	+	-	-	-	-	0.03	0
365.			*Sastragala* sp.	-	-	-	+	-	-	0.07	0.17
366.		**Lygaeidae**	*Lygaeus equestris* Linnaeus	+	-	-	+	-	-	0.10	0.17
367.			*Physopetata schlanbuschi* Brum	-	-	-	+	-	+	0.10	0.17
368.			*Physopetata gutta* Brum	+	-	-	-	+	+	0.35	0.33
369.			*Spilostethus hospes* Fabricius	-	+	-	-	-	-	0.03	0
370.		**Cicadellidae**	*Bothrogonia* sp.	-	-	+	-	-	-	0.07	0.17
371.			*Gaeanam maculata* (Fabricius)	-	-	-	+	-	-	0.07	0.17
372.			*Pycna repanda* Linnaeus	-	-	-	+	-	-	0.07	0.17
373.		**Cercopidae**	*Callitettix versicolor* (Fabricius)	-	+	+	-	-	-	0.10	0.17
374.			*Cosmoscarta septumpunctata* Walker	-	+	-	-	-	-	0.07	0.17

375.			*Cosmoscarta* sp.	-	-	+	-	-	-	0.07	0.17
376.		**Cicadidae**	*Haphsa nicomache* Walker	-	+	-	-	-	-	0.03	0
377.			*Oncotympana* sp.	-	+	-	-	-	-	0.07	0.17
378.			*Pomponia fusca* Olivier	-	+	-	-	-	-	0.03	0
379.		**Reduvidae**	*Euagoras plagiatus* Burmeister	-	+	-	-	-	-	0.03	0
380.			*Harpactor marginatus* Distant	-	+	-	-	-	-	0.03	0
381.			*Harpactor* sp.	-	-	+	-	-	-	0.07	0.17
382.		**Pyrrhocoridae**	*Dysdercus* sp.	-	-	-	+	+	+	0.14	0.17
383.		**Berytidae**	*Cletus punctulatus* Westwood	+	-	-	-	-	-	0.03	0
384.		**Eurybrachinadae**	*Eurybrachys* sp.	-	+	-	-	-	-	0.03	0
385.		**Ricaniidae**	*Ricania speculus* Walker	-	+	-	-	-	-	0.03	0
386.	**Diptera**	**Asilidae**	*Microstylum bicolor* Mcquart	-	-	-	+	-	-	0.07	0.17
387.			*Microstylum* sp.	-	+	-	-	-	-	0.03	0
388.			*Musca domestica* Linnaeus	-	-	-	+	-	-	0.07	0.17
389.			*Neoitamus* sp.	-	-	+	-	-	-	0.07	0.17
390.			*Philodious javanus* Wiedemann	+	-	-	-	-	-	0.03	0
391.			*Stenopagon oldroydi* Josephs and Pauri	+	-	-	+	+	+	0.38	0.33
392.		**Syrphidae**	*Episyrphus balteatus* (De Geer)	-	-	+	-	-	-	0.07	0.17
393.			*Eristalis tenax* (Linnaeus)	-	-	+	-	-	+	0.10	0.17
394.			*Syrphus confracter* Wiedemann	-	+	-	-	+	+	0.10	0
395.			*Syrphus fulvifacies* Brunetti	+	-	-	-	-	-	0.38	0.17
396.		**Tabanidae**	*Philoliche* sp.	+	-	-	+	+	-	0.14	0.17
397.			*Tabanus orientis* Walker	+	+	-	-	-	-	0.07	0
398.			*Tabanus* sp.	-	-	-	+	+	-	0.10	0.17

399.		**Bombyllidae**	*Anthrax georgicus* Macquart	-	-	-	+	-	-	0.07	0.17
400.			*Anthrax* sp.	-	+	-	-	-	-	0.03	0
401.			*Bombylius* sp.	-	-	+	-	-	-	0.03	0
402.		**Calliphoridae**	*Chrysomya* sp.	-	-	-	+	-	-	0.07	0.17
403.			*Lucilia* sp.	-	-	-	+	-	-	0.03	0
404.		**Tipulidae**	*Tipila himalayensis* Brunetti	+	-	-	-	+	-	0.07	0
405.			*Tipula* sp.	-	-	-	+	-	-	0.07	0.17
406.		**Bibionidae**	*Plecia* sp.	-	-	-	+	-	-	0.07	0.17
407.		**Muscidae**	*Ochromyia* sp.	-	+	-	-	-	-	0.03	0
408.		**Sarcophagidae**	*Sarcophaga annandalei* Senior-White	-	+	-	-	-	-	0.03	0
409.	**Neuroptera**	**Myrmeleontidae**	*Myrmeleon inanis* Gerstaecker	-	+	-	-	-	-	0.14	0.17
410.		**Chrysopidae**	*Chrysoperia carnea* (Rambur)	-	-	-	+	-	-	0.17	0.17
411.	**Isoptera**	**Termitidae**	*Microcerotermes championii* Snyder	-	-	+	-	-	-	0.35	0.17
412.			*Odonatatermes obesus* (Rambur)	-	+	+	-	-	-	0.49	0.33

Abbreviations used: PA1 (Protected Area 1): Binsar Wildlife Sanctuary; PA2 (Protected Area 2): Corbett Tiger Reserve; PA3 (Protected Area 3): Nandhaur Wildlife Sanctuary; PA4 (Protected Area 4): Askot Wildlife Sanctuary; PA5 (Protected Area 5): Nanda Devi Biosphere Reserve; PA6 (Protected Area 6): Naina Devi Himalayan Bird Conservation Reserve; RA: Relative abundance; NO: Normalised abundancy; (+): species present in particular protected area; (-): species not present in particular protected area.

TABLE 2. List of species of insects of different orders confined to respective Protective area Kumaun division of Uttarakhand.

PA/ Order	Lepidoptera	Coleoptera	Hymenoptera	Orthoptera	Odonata	Hemiptera	Diptera	Isoptera	Neuroptera
PA1	*Argynnis hyperbius, Dodona ouida, Dysphania militaris, Neptis zaida, Ochlodes brahma, Paliga damastesalis*	*Cerogria nepalensis, Gallerucida rutilans, Gymnopleurus subtilis, Lachnosterna cavifrons, Lytta limbata, Onthophagus gagates, Pseudolucanus cantor*	*Bombus* sp., *Vespa* sp., *Scolia venusta*	*Chorthippus almoranus, Gastrimargus transversus, Heteropternis respondence*	*Bayadera indica, Megalestes major*	*Cletus punctulatus, Ochrochira albiditarsis*	*Philodious javanus, Syrphus fulvifacies*	-	-
PA2	*Euchrysops cnejus, Euploea mulciber, Freyeria trochylus, Leptosia nina, Libythea* sp., *Megisba malaya,*	*Alcides* sp., *Anomala flavipes, Ateuchus* sp., *Cicindela* sp., *Colasposoma splendidum, Hydrophilus* sp., *Ophonus indicus*	*Eriades decipiences, Compsomeris prismatica, Sphex umbrosus, Eumenes petiolate, Vespa cincta*	*Cyrtacanthacris tatarica, Oxyrrheps obusta, Trilophidia annulata, Atractomorpha crenulata*	*Tholymis tillarg*	*Anoplocnemis phasiana, Leptocorisa varicornis, Nezara viridula, Spilostethus hospes, Cosmoscarta septumpunctata,*	*Philodious javanus, Syrphus fulvifacies*	-	-

	Sarangesa dasahara, *Sarangesa purendra*, *Spilalia galba*, *Spindasis* sp., *Tarucus indica*					*Haphsa nicomache*, *Oncotympana* sp., *Pomponia fusca*, *Euagoras plagiatus*, *Harpactor marginatus*, *Eurybrachys* sp., *Ricania speculus*			
PA3	*Abisara bifasciata*, *Actias selene*, *Aeromachus stigmata*, *Arhopala amantes*, *Bradina diagonalis*, *Borbo bevani*, *Cnaphalo-crocis medinalis*, *Daphnis nerii*,	*Adalia* sp., *Catharsius capucinus*, *Chilocorus infernalis*, *Chlaenius* sp., *Coccinella transversalis*, *Dorysthenes huegelii*, *Epicauta* sp., *Heliocopris bucephalus*, *Hycleus* sp., *Hydrophilus triangularis*	*Ammophila atripes*, *Andrena cineraria*, *Pepsis* sp., *Phalerimeris* sp., *Sceliphron* sp., *Sphex* sp., *Vespula flaviceps*	*Gryllus* sp., *Oxya* sp., *Phaneropte-ra* sp., *Phlaeoba panteli*, *Teleogryllus testaceus*, *Tyltropidius varicornis*	*Aristocypha fenestrella*, *Aristocypha quadrimacu-lata*, *Ischnura rubilio*, *Orthetrum pruinosum*, *Paracypha unimaculata*, *Pseudagrion australasiae*		*Bothrogonia* sp., *Cosmoscarta* sp., *Harpactor* sp. (Order: Hemiptera), *Bombylius* sp., *Episyrphus balteatus*, *Neoitamus* sp.	*Microcero-termes championii*	-

	Episteme adulatrix, *Erebus caprimulgus*, *Eressa confinis*, *Eupterote* sp., *Fodina pallula*, *Jamides celeno*, *Libythea* sp., *Ourapteryx clara*, *Pseudocoladenia faith*, *Spirama retorta*, *Spoladea recurvalis*, *Tarucus nara*, *Theretra nessus*, *Trigonodes hyppasia*, *Tyspanodes linealis*, *Udaspes folus*, *Vagrans egista*, *Vamuna remelana*, *Zizula hylax*	*Metopodontus biplagiatus*, *Ophonus rufibarbis*, *Oryctes nasicornis*, *Psyllora vigintiduopun-ctata*							

PA4	*Abisara fylla*, *Agrius* sp., *Graphium cloanthus*, *Hemaris* sp., *Lemyra* sp., *Parantica sita*, *Sphinx* sp.	*Chiloba acuta*, *Clinteria spilota*, *Dsygnathus* sp., *Oenopia kirbyi*, *Oxycertonia versicolor*, *Pheropsophus* sp., *Popilla cupricollis*, *Scarities* sp., *Sisyphus hirtus*, *Torynorrhina opalina*	*Anthophora* sp., *Campanotus* sp., *Campsomeriella collaris*, *Delta dimidiatipennis*, *Diacomma* sp., *Mandarinia* sp., *Pachycondyla* sp., *Polistes hebraeus*, *Polistes* sp.	*Elimaea* sp.		*Anaplocne mispasina*, *Cletus* sp., *Gaeanam maculata*,, *Murgantia histrionic*, *Pycna repanda*, *Sastragala* sp.	*Anthrax georgicus*, *Chrysomya* sp., *Chrysoperia carnea*, *Microstylum bicolor*, *Musca domestica*, *Plecia* sp., *Tipula* sp.	-	-
PA5	-	-	-	-	-	-	-	-	-
PA6	-	-	-	-	-	-	-	-	-

Abbreviations used: PA1 (Protected Area 1): Binsar Wildlife Sanctuary; PA2 (Protected Area 2): Corbett Tiger Reserve; PA3 (Protected Area 3): Nandhaur Wildlife Sanctuary; PA4 (Protected Area 4): Askot Wildlife Sanctuary; PA5 (Protected Area 5): Nanda Devi Biosphere Reserve; PA6 (Protected Area 6): Naina Devi Himalayan Bird Conservation Reserve; (-): species of no order confined to any protected area.

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Conflict of interests

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Οικολογικά Δεδομένα για την ποικιλομορφία των εντόμων σε δίκτυα προστατευόμενων περιοχών της περιοχής Kumaun, στα Δυτικά Ιμαλάια

MANOJ KUMAR ARYA*, SURABHI BISHT* AND AMBIKA TIRUWA

Insect Biodiversity Laboratory, Department of Zoology
D.S.B. Campus, Kumaun University, Nainital- 263002, Uttarakhand, India

ΠΕΡΙΛΗΨΗ

Η περιοχή Uttarakhand στα Δυτικά Ιμαλάια, γνωστή για την πλούσια βιοποικιλότητά της, περιλαμβάνει πολλές προστατευόμενες περιοχές που περιλαμβάνουν από τροπικές έως εύκρατες ζώνες. Η παρούσα εργασία διερευνά την ποικιλομορφία των εντομολογικών ειδών στις έξι προστατευόμενες περιοχές στην περιοχή Kumaun των Ιμαλαΐων. Συνολικά, τεκμηριώθηκαν συνολικά 412 είδη εντόμων από εννέα τάξεις και 70 οικογένειες. Η τάξη Lepidoptera ήταν η πιο ποικιλόμορφη με μέγιστο 154 είδη, ακολουθούμενη από τα Coleoptera (81 είδη), τα Hymenoptera (58 είδη), τα Orthoptera (33 είδη), τα Hemiptera (31 είδη), τα Odonata (28 είδη), τα Diptera (23 είδη) και Isoptera και Neuroptera ως τα λιγότερο κυρίαρχα με δύο είδη το καθένα. Η ποικιλότητα ειδών κατά Shannon (Hs) κυμάνθηκε από 3,99 έως 4,95, με την υψηλότερη ποικιλότητα να καταγράφεται στο καταφύγιο άγριας ζωής Nandhaur και τη χαμηλότερη στο καταφύγιο προστασίας πουλιών Naina Devi Himalayan. Η ανάλυση cluster αποκάλυψε δύο κύρια μοτίβα ποικιλομορφίας, υποδεικνύοντας σημαντική β-ποικιλομορφία μεταξύ των περιοχών μελέτης. Species-wise occupancy και η abundance analysis αποκάλυψε ότι τα είδη *Pieris brassicae*, *P. canidia* και *Apis dorsata* είχαν την υψηλότερη σχετική αφθονία από όλες τις προστατευόμενες περιοχές. Αντίθετα, 91 είδη εντόμων είχαν σχετική αφθονία με μόνο 0,03% το καθένα. Επιπλέον, επτά είδη παρουσίασαν την υψηλότερη κανονικοποιημένη πληρότητα 1,00, υποδεικνύοντας την προσαρμοστικότητά τους σε διαφορετικές περιβαλλοντικές συνθήκες εντός των προστατευόμενων περιοχών. Αυτά τα ευρήματα υπογραμμίζουν τη σημασία της ποικιλομορφίας των οικοτόπων και των στοχευμένων στρατηγικών διατήρησης για τη διατήρηση των πληθυσμών των εντόμων και της υγείας των οικοσυστημάτων στο Kumaun Himalaya.

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First record of the Zigzag elm sawfly *Aproceros leucopoda* (Hymenoptera: Argidae) in Greece

ANGELOS TSIKAS* AND PARASKEVI KARANIKOLA

Department of Forestry and Management of the Environment and Natural Resources, Laboratory of Forest Protection and Environmental Pollution, Democritus University of Thrace, School of Agricultural and Forestry Sciences, Ath. Pantazidou 193, 68 200, Orestiada, Greece

ABSTRACT

The Zigzag elm sawfly *Aproceros leucopoda* Takeuchi, 1939 (Hymenoptera, Argidae) is a defoliator of elm trees of Asian origin recently introduced into Europe. In this publication, *A. leucopoda* is recorded in Greece from Xanthi, constituting the first record of this alien species to the country. The presented locality is the southernmost point of its hitherto known distribution in Europe.

KEY WORDS: Alien species, Zigzag elm sawfly, defoliator pests, first record, *Ulmus* spp., Thrace.

Introduction

Introduction and spread of alien species during the last centuries have been facilitated by globalization and international trade, posing a significant worldwide threat to biodiversity and economy (Hulme 2009). Insects, being the most numerous terrestrial animal class (Stork 2018), have many species that spread outside their natural range. Not surprisingly, the total estimated number of alien insects is as high as a quarter of species (Liebhold et al. 2018). Some of these species can pose serious threats to various natural ecosystems, endangering the existence of many species, as well as the smooth functioning of ecosystems (Roques 2010; Smith et al. 2018; Olenici et al. 2022). In Greece, a total of 469 alien insect species have been identified (Demetriou et al. 2021).

Aproceros leucopoda Takeuchi, 1939 is a defoliator of elm trees (*Ulmus* spp.) of Asian origin, naturally occurring in East Asia – Japan (Takeuchi 1939; Naito 2004), China (Wu and Xin, 2006) and Russian Far East (Zhelochovtsev and Zinovjev 1995), introduced into Europe (probably in the early 2000) and North America (EPPO 2024). It is a thelytokous parthenogenetic species, and no males have ever been recorded. Females lay 7–60 eggs into the tips of consecutive indentations around the edges of leaves. Larvae hatch after 4–8 days, feeding exclusively on elms (Wu 2006; Blank et al., 2010; Yu et al. 2011; Martynov and Nikulina 2017). Feeding traces of early-stage larvae have a characteristic zigzag pattern. Later, the attacked leaf is completely consumed except for the thick middle vein. There are six larval instars, and development is completed within 9–18 days (Papp et al. 2018). Larvae make either a loosely spun cocoon with a net-like structure fixed to the lower surface of leaves, rarely on twigs or the ground, or a more compact, solid-walled cocoon with a grid of silk strands, found in the litter or soil in the field from at least as early as June (Wu 2006; Blank et al. 2010; Martynov and Nikulina, 2017). Pupation in the loosely spun cocoons occurs after 2–3 days, and adults emerge 4–7 days after. The

**Corresponding authors: atsikas@fmenr.duth.gr*

total period from oviposition to adult emergence takes 19–36 days (Wu 2006, Blank et al. 2010, Mol and Vonk 2015, Martynov and Nikulina 2017).

In this paper, the species *A. leucopoda* is recorded from Xanthi (Thrace) constituting the first record of this alien species in Greece.

Materials and Methods

On 13 May 2024, during ordinary hand-collecting samplings in a forest stand dominated by oaks near Mega Eumoiro, Xanthi [41.206ºN, 24.799ºE, alt. 305 m], we accidentally met a field elm (*Ulmus minor* Mill.) with the characteristic zigzag-shaped damage on a leaf (Blank et al. 2010; Doychev 2015; Martynov and Nikulina 2017) (Figure 1). By closer investigation, we located a larva feeding on the leaf. We sampled the leaf with the specimen and took it to the Laboratory of Forest Protection and Environmental Pollution for further investigation. The specimen was placed in a glass vial and preserved in 75% EtOH.

For the identification of *A. leucopoda*, the key of Blank et al. (2010) has been used, which includes detailed descriptions of larvae, adults and the characteristic damage caused by the feeding larvae on elm leaves. The images of the specimen included in this paper were taken using Nikon D90 camera and Olympus SZX7 stereomicroscope at magnifications between 80-140x, with a cold-light source equipped with two flexible cold light arms covered with a light diffusor, and a LED ring mounted on the stereomicroscope focus.

FIG. 1: Larva of *Aproceros leucopoda* Takeuchi, 1939 feeding on an elm leaf.

Results and Discussion

The larva was identified as *Aproceros leucopoda*. The diagnostic character used for distinguishing *A. leucopoda* from other Argidae species was the characteristic dark brown T-shaped marks on the 2nd and 3rd pair of thoracic legs (Figure 2).

FIG 2. *Aproceros leucopoda* larva.

This is the first reported occurrence of the zigzag elm sawfly in Greece, which comprises the first alien species of the family Argidae in the country. The species has been first recorded in Europe in 2003 from Hungary and Poland (Blank et al. 2010). Romania in 2005, Ukraine in 2006, Slovakia in 2007 (Blank et al. 2010), Moldova in 2008 (Timuş et al. 2008, misidentified as *Arge* sp.), Austria and Italy in 2009 (Zandiacomo et al. 2011), in Germany, the European part of Russia, Slovenia, Croatia the Czech Republic in 2011 (Kraus et al. 2011; Artokhin et al. 2012; De Groot et al. 2012; Matošević 2012; Jurásková et al. 2014), Serbia in 2012 (Glavendekić et al. 2013), in Belgium and the Netherlands in 2013 (Boevé, 2013; Mol and Vonk, 2015), in Bulgaria and Latvia in 2015 (Doychev 2015; Mihailova 2015), in France, Estonia, Bosnia and Herzegovina, the United Kingdom and Switzerland in 2017 (Legrand 2017; NPPO of Estonia 2017; Dautbašić et al. 2018; Forest Research 2018; Höllling 2018), in Luxembourg in 2018 (Burton et al. 2019) and in Lithuania in 2020 (Sinchuk et al. 2021) (Figure 3). In 2020, it was also documented occurring in North America (Martel et al. 2022). The presented locality is the southernmost point of hitherto known *A. leucopoda* distribution in Europe, but it is expected to be more widespread in Greece and spread over a significant part of the territory, due to the high occurrence of elms in green stands. The entrance route of *A. leucopoda* could have been the neighboring country of Bulgaria, where it has been first recorded in 2015, as a stowaway or spontaneous spread (Doychev 2015). Undoubtedly, its actual year of introduction may be much earlier. Low population levels along with insignificant damage to the hosts made its presence unnoticed so far.

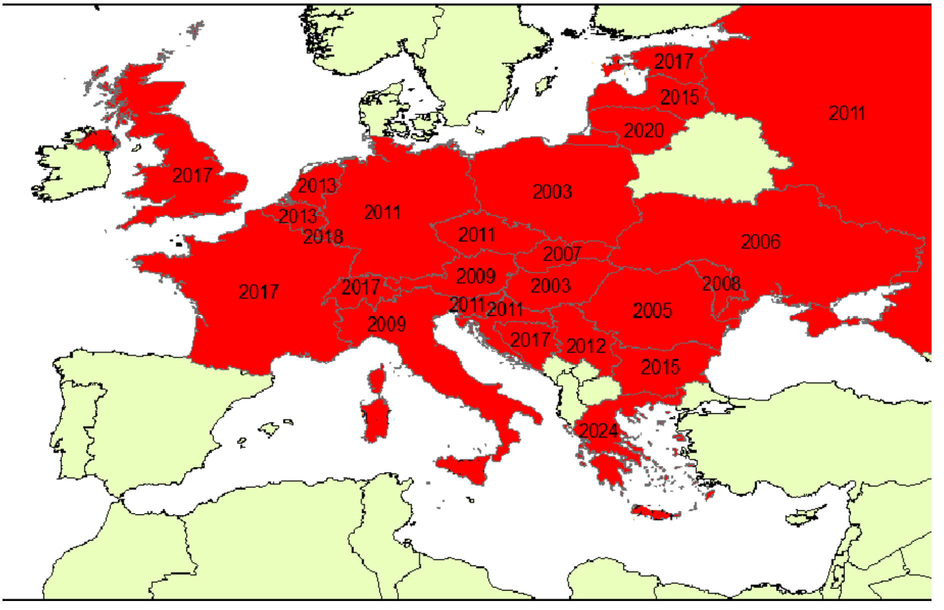

FIG. 3: Known distribution of *Aproceros leucopoda* Takeuchi, 1939 in Europe. Countries where the species has been previously reported are shaded red with the year of the first recording.

Considering the widespread presence of elms and the rapid reproduction enhanced by parthenogenesis (Blank et al. 2010), it is possible that *A. leucopoda* could quickly increase its abundance and distribution and dramatically increase the decline of elms already suffering from Dutch elm disease transmitted by bark beetles (*Scolytus* spp.) (Webber 1990) and elm yellows transmitted by leafhoppers (Pavan 2000; Carraro et al. 2004). Therefore, further research is needed for the actual and potential distribution of this species and its effects on the health of elm trees.

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Πρώτη καταγραφή του *Aproceros leucopoda* (Hymenoptera: Argidae) στην Ελλάδα

ΑΓΓΕΛΟΣ ΤΣΙΚΑΣ* ΚΑΙ ΠΑΡΑΣΚΕΥΗ ΚΑΡΑΝΙΚΟΛΑ

Τμήμα Δασολογίας και Διαχείρισης Περιβάλλοντος και Φυσικών Πόρων, Εργαστήριο Υλωρικής και Ρύπανσης Περιβάλλοντος, Δημοκρίτειο Πανεπιστήμιο Θράκης, Σχολή Επιστημών Γεωπονίας και Δασολογίας, Αθ. Πανταζίδου 193, 68 200, Ορεστιάδα, Ελλάδα

ΠΕΡΙΛΗΨΗ

Το *Aproceros leucopoda* Takeuchi, 1939 (Hymenoptera, Argidae) είναι φυλλοφάγο έντομο της φτελιάς ασιατικής προέλευσης που εισήχθη πρόσφατα στην Ευρώπη. Σε αυτή την εργασία, το *A. leucopoda* καταγράφεται για την Ελλάδα στην περιοχή της Ξάνθης, αποτελώντας την πρώτη αναφορά του ξενικού αυτού είδους για τη χώρα. Η θέση καταγραφής του είναι το νοτιότερο σημείο της μέχρι τώρα γνωστής εξάπλωσής του στην Ευρώπη.

Insecticidal activity of garden waste compost tea against *Aphis craccivora* (Koch) aphids: *In vitro* and *in silico* study

SALIM LEBBAL[*1], TOUFIK BENHIZIA[1], RODRIGO PÉREZ-SÁNCHEZ[2] & MARÍA REMEDIOS MORALES-CORTS[2]

[1]Abbas Laghrour University, Department of Agricultural Sciences, Laboratory of Management, Conservation and Valorization of Agricultural and Natural Resources (LMCVANR), Khenchela/Algeria

[2]University of Salamanca, Faculty of Agricultural and Environmental Sciences, Salamanca/ Spain

ABSTRACT

Aphids are important pests of many crops. However, the use of chemical insecticides has provoked ecological and health problems, thus the valuation of natural products becomes an interesting alternative. Compost teas are organic products that are viewed as substitutes for common pesticides. The present study aims to screen the potential insecticidal effect of garden waste compost tea, through *in vitro* and *in silico* approaches. Five concentrations of a compost tea (20, 40, 60, 80 and 100%) were tested against *Aphis craccivora*, in comparison to negative and positive controls. The repellency and toxicity tests were conducted under laboratory conditions. Moreover, an evaluation of the inhibitor capacity of some compost compounds against acetylcholinesterase was carried out using molecular docking. Results revealed that compost had a very weak insecticidal effect against *A. craccivora* (where the corrected mortality did not exceed 24%) compared to the tested chemical pesticide. Furthermore, the repellency test showed that the compost had some repellency effect in comparison to the tested chemical pesticide which had an attractant effect. Concerning the results of the molecular docking, pirimicarb (active molecule of pesticides) recorded a better S score than the three compost compounds.

KEY WORDS: Insect pests, repellency, toxicity, bioinsecticides, molecular docking.

Introduction

Biotic stress in plants caused by insect pests is one of the most significant problems, leading to yield losses (Tlak Gajger & Dar, 2021). These insect pests belong to different groups, including Aphididae. Among its important species, *Aphis craccivora* attacks about 50 crops in 19 different plant families (Blackman & Eastop, 2007). It causes damage by direct feeding as well as by transmission of plant viruses causing diseases such as broad bean yellow mosaic and bean leafroll (Weigand & Bishara, 1991).

Synthetic pesticides are being widely used for the management of pests to avoid losses both in field and post-harvest storage (Kumar et al., 2022). The estimated 30% losses from pests that would occur in the absence of pesticides, would spell economic and human disaster for many developing countries around the world (Saxena et al., 2014). However, global concern has been raised in recent times against the utility of synthetic insecticides in households and fields (Barua et al., 2016). Pesticides accumulate toxic residues on food grains used for human consumption and this may lead to health issues, in addition to the very high worldwide mortality rate due to

**Corresponding authors: salim-leb@hotmail.com*

pesticide poisoning (Singh & Kaur, 2018). Besides, pesticide residues may constitute a significant source of contamination of air, water and soil, and a large quantity of pesticides is released into the atmosphere during application, thereby inducing adverse climatic changes (Saxena et al., 2014). That's why for the future, it is necessary to develop a more environmentally friendly agriculture that will decrease inputs in chemicals and generate fewer harmful outputs such as pesticide residues (Wójcicka, 2010).

Consequently, identification and exploring for nature-originating pest control agents could become a possible substitute for the synthetic chemical pesticides (Kumar et al., 2022). Natural insecticides are chemical compounds or substances obtained from living organisms (Barua et al., 2016). Natural compounds known for their bioactivity against insects are considered safe, economical, biodegradable and easy to use (Singh and Kaur 2018). Among these, compost and compost teas have been shown, in previous studies, to possess control properties against a wide range of different plant pests and diseases (Bandara et al., 2010; Edwards et al., 2010; Alao et al., 2011; Shalaby et al., 2012; Pane et al., 2012; Martín, 2014; López-Martín et al., 2018; Morales-Corts et al., 2018; Suwandi et al., 2020; González-Hernández et al., 2021, 2022, among others). Concretely, compost teas are organic solutions obtained by the fermentation of compost in a liquid phase for a few days, with or without aeration (Al-Dahmani et al., 2003). The physical and biochemical quality of compost teas depends on the characteristics of the starting compost, as well as on other parameters that affect its production i.e., compost-to-water ratio, and aeration (Ingham, 1999; Scheuerell & Mahaffee, 2002; Martin et al., 2012).

On the other hand, the application of computational modeling guided the experimental procedure by elucidating the mechanisms of ligand-enzyme binding and greatly reducing the research time and costs (Badawy et al., 2022). Molecular docking is an *in silico* method which predicts the placement of small molecules or ligands within the active site of their target protein (Surabhi & Singh, 2018). The nature of the interaction between ligand and receptor depends on a balance in the chemical/physical forces between them and the forces between each of these molecules and the solvent or environment (Krumrine et al., 2003).

Thus, the current study's objective is to evaluate *in vitro* the effectiveness of compost tea solutions against *Aphis craccivora*, as well as to screen the insecticidal activity of its components, using molecular docking approach.

Materials and Methods

Compost tea preparation

The compost tea (CT) derived from composting gardening wastes (mainly a mixture of green and pruning debris, i.e., mainly leaves and branches of cypress (*Cupressus* sp.), willow (*Salix* sp.) and poplar trees (*Populus alba* L.), reaching a C/N ratio of 30), which were obtained from public gardens of the Province of Salamanca (Spain). This process was performed in aerated piles of 15 × 2 × 2 m for 180 days in a garden center located in Salamanca (Spain) (40◦57′23″ N; 5◦41′8″ W, 775 m a.s.l.). Piles were turned twice per week for eight weeks and once a week during the rest of the bioxidative process. Moreover, the moisture of the piles was controlled once a week, and when it dropped below 55%, water irrigation was applied up to maintain the moisture average around 60% during the whole process. The mature compost was obtained under ambient conditions in March. The main characteristics of the garden waste compost were: pH 7.8, C/N 11.4, 41.4% total organic matter, 2.1% total N, 10.90% humic acids and 0.33 dS/m Electrical conductivity

(Registration No. F0004957/2031, Viveros El Arca enterprise, Salamanca, Spain).

Compost was mixed with tap water in a ratio of 1:5 (v/v) in polyethylene non-degradable 1000 L containers at room temperature (20 ºC) for a brewing period of five days. Water had been previously aerated for 8 h to reduce chlorines concentration. The mixture was aerated for five hours every day by applying circular stirring and making fine bubbles of air using a pump (750 W-300 rpm). Then, it was filtered with a double-layered cheesecloth, and the aerated compost tea (CT) was stored in a dark container (50 L capacity at room temperature) until use.

Chemical and microbiological properties of the compost tea

The chemical properties of the CT were directly analyzed; pH, electrical conductivity (EC) and C:N ratio were determined as described by Morales-Corts et al. (2021). Furthermore, assimilable nutrient contents (NO^{3-}, PO_4^{3-}, K_2O, SO_4^{2-}, Ca^{2+} and Mg^{2+}) were analyzed with the nutrient analysis photometer HANNA HI 83225. Finally, humic acids were determined following the alkali-acid method described by Pant et al. (2012). Salicilic acid and indoleacetic acid (IAA) were quantified by mass spectrometry (HPLC).

Microbial analysis of the CT was estimated using the serial dilution spread plate method. To determine the microbial population, different selective culture media and CT dilutions were used for microorganism isolation: nutrient agar and 10^{-3} dilution for total aerobic bacteria, Ashby medium, and 10^{-2} dilution for N-fixing bacteria, ISP^{-2} medium and 10^{-1} dilution for actinobacteria and modified potato dextrose agar medium but no dilution for total fungi and *Trichoderma* ssp. quantification (Wickerham, 1951; Sanchis Solera, 1996; Stella & Suhaimi, 2010; Vargas-Gil et al., 2006). Then, plates were inoculated by depositing on the agar surface 0.1 mL of the CT dilution, which was spread on the media surface with sterile glass beads. Moreover, non-inoculated plates were included as a negative control. Petri dishes were incubated in the dark at 28 °C for 3 to 15 days, depending on the medium. After this time, colony-forming units (CFU) were counted to estimate the cultivable microorganism's population. This experiment was conducted using five replications.

In vitro assays

Five CT concentrations (20, 40, 60, 80 and 100%) were tested, in addition to the negative control (distilled water) and a positive control represented by a chemical aphicide 'Lazer' (produced by Ortiva, represented in Algeria by Phytoplus and registered under the homologation number 07 45 036) containing two active molecules (Lambda-cyhalothrine and Pirimicarb, 5%+10%, which are sodium channel modulators and acetylcholinesterase inhibitor, respectively (IRAC, 2022)).

Faba bean leaves, bearing *Aphis craccivora* aphids (of the same colony), were collected from a commercial field, that had not undergone any chemical treatment. The leaves along with wingless insects were left for few hours in the plastic box. Aphids were then collected by means of a small brush and used for the bioassay.

For screening the insecticidal activity under laboratory conditions (T = 25 °C; Relative Humidity= 36%), cut pieces of leaves were dipped in each concentration of the compost for 2-5 seconds and then the solution was drained out and pieces were kept for drying. Three replications of each concentration along with positive and negative controls were taken for bio-assay studies. Treated leaves were kept in the Petri dish with 10 aphids in each replicate. Mortality was observed after 24 h of treatment. Percent mortality was corrected through Abbots formula (1925):

Corrected mortality rate = [(Tmp - Cmp)/ (100 - Cmp)] × 100

where Tmp: mortality rate of the treatment and Cmp: mortality rate on the control.

Furthermore, ANOVA analysis followed by Student-Newman-Keuls test were performed by the means of SPSS software, to compare mean aphid mortalities.

Concerning the test of repellency *in vitro*, three replications for each concentration, in addition to the conventional pesticide, were considered. For each replicate, two leaf discs free of aphids were cut. After that, one leaf disc was dipped for 2-5 seconds in the corresponding solution. Each Petri dish was divided into two equal parts. Leaf discs dipped in treatment solution were placed in one side and untreated ones on the other side. 10 wingless aphids were placed at the center of the dish and left for 24 hours and then the number of aphids on each side was recorded. The repellency percentage was calculated according to the following equation:

PR = [(C – T) / (C + T)] × 100 (Pavela et al., 2009)

where: C = the number of aphids on control disc, and T = the number of aphids on treated disc.

In silico study

On the other hand, the evaluation of the insecticidal activity by molecular docking was carried out using the Molecular Operating Environment (MOE) software, version 2014.0901, designed by Chemical Computing Group Inc. (Canada). We studied *in silico* the ability of four ligands, including three compounds of the examined compost (salicylic acid, humic acid and indole-3-acetic acid), in addition to an active molecule of chemical pesticide (Pirimicarb), to inhibit a target enzyme of insects called acetylcholinesterase (AChE), having the PDB code 1QON.

Results

Compost tea properties

The studied characteristics (pH, EC, C-to-N ratio, assimilable macro and micronutrients, and humic acids) are shown in Table 1. Highlighted are the high NO3− and K2O concentrations (2240.4 and 2851.2 ppm, respectively) and the low C-to-N ratio of the extract, as well as the relevant humic acids amount.
Concerning the microbiological analysis of the compost tea, its microbiological composition was: total aerobic bacteria 2.7 × 107 cfu mL-1; N-fixing bacteria 2.7 × 107 cfu mL-1; actinobacteria 7.4 × 104 cfu mL-1; Trichoderma sp. and fungi were valued between 2.7 and 8.7 × 102 cfu mL-1.

In vitro assays

Results revealed that for the test of toxicity in vitro, there was a significant difference between the studied treatments. It seems that compost had very weak insecticidal effect against A. craccivora comparatively with the tested chemical pesticide. The corrected mortality did not exceed 24% for the different concentrations of compost tea (Table 2).
Results of the test of repellency in vitro showed that compost had some repellency effect against A. craccivora in comparison with the tested chemical pesticide which seems to have attractant effect (Table 3). All the tested concentrations of the compost were ranged in the class I of scale described by McDonald et al. (1970).

In silico study

Concerning the results of the molecular docking, the best S score was recorded for Pirimicarb (active molecule of pesticides) followed by indole-3-acetic acid (Table 4). This result confirms that obtained for in vitro test, i.e. the tested pesticide was more toxic than compost solutions.

*Corresponding authors: salim-leb@hotmail.com

TABLE 1: Chemical characteristics of the studied compost tea. *Results are indicated as mean ± standard deviation*

pH	EC (dS/m)	C/N	NO_3^- (ppm)	P_2O_5 (ppm)	K_2O (ppm)	SO_4^{2-} (ppm)	Ca^{2+} (ppm)	Mg^{2+} (ppm)	Humic acids (mg/L)	Salicilic acid (mg/L)	IAA (ng/L)
7.16±0.14	1.2±0.09	7.1	2240.4±186.6	149.7±18.3	2851.2±234.5	43±16	280±20	20±13	198±37	5.85±1.23	80±10

TABLE 2: Corrected mortality percentages of aphids on different solutions

Treatments	Corrected mortality (M ± SE)
20 %	0.00 ± 0.00 a
40 %	0.00 ± 0.00 a
60 %	23.70 ± 7.73 a
80 %	4.63 ± 3.34 a
100 %	9.72 ± 9.72 a
Chemical pesticide	100.00 ± 0.00 b
Signification	0,000 *

** Signification at P < 0.05*

TABLE 3: Percentages of repellency of aphids by different solutions

Treatments	Percentages of Repellency (M ± SE)
20 %	8.15 ± 39.34
40 %	17.04 ± 11.92
60 %	6.67 ± 48.07
80 %	18.52 ± 29.60
100 %	10.37 ± 5.78
Chemical pesticide	-14.07 ± 2.96

TABLE 4: S score of the studied ligands with the target protein 1QON

PubChem code	Ligands	Properties	S score (Kcal mol^{-1})
31645	Pirimicarb	Inhibitor of acetylcholinesterase	- 6.42
802	Indole-3-acetic acid	Compound of compost tea	- 5.23
90472028	Humic acid	Compound of compost tea	- 4.30
338	Salicylic acid	Compound of compost tea	- 4.64

Discussion

The extract's low C:N ratio, high NO^{3-} and K_2O concentrations, and high humic acid content were important characteristics of the studied compost tea. There are numerous factors that affect the quality of vermicompost tea, which in-turn affect its ability for disease and pest suppressiveness, among them are included compost grade, compost maturity, aeration, temperature, microbial inoculants, and compost to water ratio (Yatoo et al., 2021). For instance, Nur et al. (2023) indicated differences in the C:N ratio and humic acid concentration between aerated and non-aerated compost tea made from wild lotus. Orosz et al. (2021) reviewed many mechanisms of action of different compost extract components allowing control of plant diseases. There is a lot of published literature concerning studies regarding the effects of humic acids and different biostimulants on the growth, productivity, and protection of vegetable species is available, whereas literature concerning the study of the effects of compost teas on vegetable crops is more limited (Pilla et al., 2023).

On the other hand, the present investigation revealed that the compost had weak insecticidal effect against *A. craccivora*, in comparison to the chemical pesticide containing pirimicarb, as well as limited repellency activity. Similarly, Abdu-Allah (2012) indicated that the toxicity index of pirimicarb shows higher aphicidal activity than three petroleum ether plant extracts. Additionally, Cantelo (1985) found that compost treated with experimental insecticides affecting acetylcholinesterase (such as diazinon and dimethoate) were effective against the dipteran pest *Megaselia halterata*.

In contrast, Bandara et al. (2010) found that brinjal aphids, mealy bugs and thrips were significantly reduced in treated plants with different compost solutions, compared to the controls. Besides, other results have shown that organic compost liquid formulations acted as antifeedants for *Podagrica* sp., *Zonocerus variegatus* and *Bemisia tabaci,* while maize stover compost had a very good insect control (Alao et al., 2011). The compost extract containing amino acids showed a remarkable potential to develop into an effective biostimulant for protection from virus disease and its insect vector, *Aphis gossypii* (Suwandi et al., 2020).

Moreover, insect pests of the plant species *Telfairia occidentalis* that received foliar spray of compost extracts (from cassava peel and tithonia plant) were minimal compared with non-fertilized plants and those that received soil incorporated NPK fertilizer (Akanbi et al., 2007). Furthermore, Shalaby et al. (2012) mentioned that compost tea used as soil drench applied after sowing, followed by foliar application of compost tea proved to be the best treatment against insect infestations by *Pegomia mix*ta Vill, *Scrobipalpa ocellatella* Boyd and *Cassida vittata* Vill. Moreover, Edwards et al. (2010) indicated that all examined vermicompost extracts suppressed pest establishment on the plants, and their rates of reproduction for all three species of pests, significantly. The same researchers concluded that the most possible cause for the unacceptability of the plants to pests was the uptake of soluble phenolic materials from the vermicompost extracts into the plant tissues and these compounds are known to make plants unattractive to pests.

Likewise, previous studies confirmed the influence of compost as soil fertilizer on aphid populations (Ponti et al., 2007; Stafford et al., 2012).

On the other hand, the results of the molecular docking were consistent with the in vitro toxicity assay. The pirimicarb molecule proved to be the more effective in the inhibition of acetylcholinesterase, recording the best S score (-6.42 kcal mol^{-1}), compared to the compost compounds. Other *in silico* studies showed not very different S score results. For instance, Lebbal et al. (2021) noticed that α-Thujene ligand has the best score with 1QON target enzyme (-4.77), among seven compounds contained in *Thymus algeriensis* and *T. numidicus*, whereas, Badawy et al. (2022) indicated the docking energies in the AChE enzyme of -9.52, -9.38, and -8.52 kcal mol^{-1} for chlorpyrifos-methyl, diazinon, and malathion, respectively. Furthermore, Malak et al. (2022) found that among 38 studied bioactive phytochemicals, some compounds recorded docking scores of 9.11 to 7.14 against cattle tick *Rhipicephalus microplus* acetylcholinesterase protein.

Conclusion

In summary, the present study confirmed through in vitro and in silico screenings that the compost had low aphicidal effect. In addition, it has better repellent activity compared with the tested chemical aphicide. Nevertheless, its use as fertilizer should be combined with other control methods to reduce efficiently the negative impact of aphid populations.

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Εντομοκτόνος δράση του εκχυλίσματος κομπόστ απορριμμάτων κήπου κατά των αφίδων *Aphis craccivora* (Koch): *In vitro* και *in silico* μελέτη

SALIM LEBBAL*[1], TOUFIK BENHIZIA[1], RODRIGO PÉREZ-SÁNCHEZ[2] & MARIA REMEDIOS MORALES-CORTS[2]

[1]*Abbas Laghrour University, Department of Agricultural Sciences, Laboratory of Management, Conservation and Valorization of Agricultural and Natural Resources (LMCVANR), Khenchela/Algeria*

[2]*University of Salamanca, Faculty of Agricultural and Environmental Sciences, Salamanca/ Spain*

ΠΕΡΙΛΗΨΗ

Οι αφίδες αποτελούν σημαντικούς εχθρούς πολλών καλλιεργειών. Η χρήση χημικών εντομοκτόνων έχει προκαλέσει σημαντικά περιβαλλοντικά προβλήματα και προβλήματα στην ανθρώπινη υγεία. Έτσι, η αποτίμηση των φυσικών προϊόντων αποτελεί μια ενδιαφέρουσα εναλλακτική. Τα εκχυλίσματα κομπόστ (τσάγια κομποστοποίησης) είναι φυσικά προϊόντα που θεωρούνται υποκατάστατα της χρήσης κοινών φυτοφαρμάκων. Η παρούσα μελέτη στοχεύει να εξετάσει την πιθανή εντομοκτόνο δράση του εκχυλίσματος κομπόστ απορριμμάτων κήπου, μέσω προσεγγίσεων *in vitro* και *in silico*. Πέντε συγκεντρώσεις του εκχυλίσματος κομπόστ (20, 40, 60, 80 και 100%) δοκιμάστηκαν έναντι των αφίδων *Aphis craccivora*, σε σχέση με αρνητικούς και θετικούς μάρτυρες. Οι δοκιμές απώθησης και τοξικότητας πραγματοποιήθηκαν σε εργαστηριακές συνθήκες. Επιπλέον, πραγματοποιήθηκε αξιολόγηση της ικανότητας αναστολής ορισμένων ενώσεων κομποστοποίησης έναντι της ακετυλοχολινεστεράσης με χρήση μοριακής σύνδεσης. Τα αποτελέσματα αποκάλυψαν ότι το κομπόστ είχε πολύ ασθενή εντομοκτόνο δράση του *A. craccivora* (όπου η διορθωμένη θνησιμότητα δεν ξεπέρασε το 24%) σε σύγκριση με το χημικό εντομοκτόνο μάρτυρα. Επιπλέον, η δοκιμή απώθησης έδειξε ότι το κομπόστ είχε κάποια απωθητική δράση σε σύγκριση με το χημικό εντομοκτόνο μάρτυρα που φαίνεται να έχει ελκυστική δράση. Όσον αφορά τα αποτελέσματα της μοριακής δέσμευσης, το pirimicarb (δραστική ουσία του χημικού εντομοκτόνου) κατέγραψε καλύτερη βαθμολογία S από τις τρεις ενώσεις κομποστοποίησης.

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Feeding Categorisation of some Caeliferan and Ensiferan species (Insecta: Orthoptera) collected from selected grasslands in Sendai City, Japan

Abu ElEla Shahenda, A. A.[1, 3], Michio Kondoh[2], Masatoshi Hori[3], Atsuhiko Nagasawa[3], Koji Nakamura[4], Wael M. ElSayed[1]

[1]*Department of Entomology, Faculty of Science, Cairo University, Egypt 12613*

[2]*Graduate School of Life Sciences, Tohoku University, 6-3 Aoba, Aramaki, Aoba-ku, Sendai 980-8578, Japan*

[3]*Graduate School of Agricultural Science, Tohoku University, Sendai, Miyagi 980-8572, Japan*

[4]*Ishikawa Prefectural Natural History Museum, Kanazawa, Ishikawa 920-1147, Japan*

ABSTRACT

Although orthopteran species have often been regarded as polyphagous herbivores, most of them show variable degrees of diet selectivity and particular food preferences. Still some species possess peculiar feeding categories which need further investigation. An assemblage of 43 orthopteran species, 7 families, 12 subfamilies and 13 tribes was surveyed from five grasslands in Sendai City, Japan to investigate their feeding preferences and were classified into seven main feeding categories based on examination of morpho-mandibular characteristics and postmortem gut content analyses. The forbivorous category was the most dominant, whereas the herbivorous the least common.

KEY WORDS: Orthoptera, mandibular characteristics, gut contents analysis, feeding categories.

Introduction

Orthopterans have emerged as a crucial group of invertebrates for environmental monitoring and assessment. The Orthoptera order, comprising a diverse array of species, holds a prominent position amongst insect orders (Zhang 2011; Bidau, 2014). With approximately 28,000 identified species worldwide and ranked as the sixth largest order in Class Insecta, trailing only the Hemiptera order as declared by Cigliano et al. (2022). Species belonging to Orthoptera are present in every terrestrial environment and exhibit remarkable diversity (ElEla et al., 2010; Yadav and Kumar 2017).

Taxonomists classified Orthoptera into two distinct suborders: Ensifera, encompassing crickets, katydids, and their relatives, and Caelifera, which include grasshoppers and their allies (Song, 2018). Orthopteran species play considerably vital roles in terrestrial food webs, serving as a valuable protein source for various animals, including amphibians, birds, small reptilian species, as well as some mammalian species. Consequently, their scarcity could disrupt the delicate trophic structure within an ecosystem (Soliman et al., 2017). They play a central role in food webs, as they are mostly primary herbivores and constitute an abundant food resource for other groups of carnivorous organisms (Parr and Chown, 2003). The composition of orthopteran assemblage is considerably highly responsive to environmental changes, play a crucial role in the ecosystems function and can serve as a valuable environmental

**Corresponding author: shosho_ali76@yahoo.com*

indicator (O'Neill et al., 2003). Indeed, these species potentially serve as useful bioindicators for land disturbance (Saha et al. 2011) providing valuable insights into qualities of ecosystems and the effectiveness of ecological networks (Zhang et al., 2019).

Orthopteran species have attracted significant attention due to the extensive damage they inflict by their herbivory action on crops and various forms of green vegetation (Dakhel et al., 2020). Aspects of feeding strategies, including selection of food, preference of food, diverse feeding patterns and diet specialization have been studied by many researchers for diverse orthopteran species (e.g., Isely, 1944; Williams, 1954; Mulkern, 1967; Otte and Joern, 1976; Bernays and Chapman, 1978; Uvarov, 1977; Ohabuike, 1979, Joern; 1983, 1985; Bernays and Bright, 1993, ElSayed, 2005; ElShazly and ElSayed, 2006; ElEla et al., 2010, 2012; Kuřavová and Kočárek, 2016; ElSayed et al., 2020).

Adaptation to food intake has led to behavioral and morphological specialization of mouthparts (Snodgrass, 1935). Fry et al. (1978) found that caeliferan species, especially Acridomorpha, are often phytophagous, and the adaptations of their mandibular structures are associated with different types of food and different species of ecological groups of plants, grasses, forbs, flowers, and seeds (Isely, 1944; Patterson, 1983, 1984; Kang and Chen, 1994; Bernays, 1998; Gangwere et al. 1998; ElEla, 2011; ElEla et al. 2010, 2012; Di Russo et al. 2014; Kuřavová and Kočárek, 2016; ElSayed et al. 2020).

Considering the paucity of information on the assemblage of orthopteran species inhabiting the five selected grasslands in Sendai city, especially on the variation of mandibles in accordance with food preference, our team was stimulated to examine the morpho- mandibular variations of orthopteran species and evaluate the gut contents analysis as evidence supporting these morpho-adaptations. Moreover, there is a lack of information regarding the diet of tetrigid species which are relatively among the least-studied groups of Orthoptera (Flook and Rowell, 1997; Hochkirch et al. 2000 & 2006) especially those recorded in some parts of Asia (Song et al. 2015) and more specifically from Sendai City, Japan (ElEla et al. 2010).

Materials and Methods

Climate and topography. The study was carried out in Sendai City, Japan (lat. 38°16'05" N, long. 140°52'11" E). The area of the city is ca. 788.09 km^2 and stretches from the Pacific Ocean to the Ōu Mountains, which are the east and west borders of Miyagi Prefecture. As a result, the geography of the city is quite diverse. Eastern Sendai is a plain, the center of the city is hilly, and western areas are mountainous.

According to the Japan Meteorological Agency (http://www.data.jma.go.jp), meteorological records showed that Sendai City has a humid subtropical climate, which features warm and wet summers, and cool and dry winters. The summer season starts in June and ends at the end of September with significant seasonal variations in temperature and rainfall. Summers are warm, with an August average temperature of 24.1 °C and the majority of the annual precipitation is delivered during summer.

The city is rarely hit by typhoons, and experiences only 6 days with more than 10 centimeters of precipitation on average.

Monsoon season usually begins in late April to early October, which is later than in most cities in Japan. During this season, cold winds from the Okhotsk air mass, which are called "Yamase", blow in and depress daytime highs.

Extremes of temperature degrees range from −11.7 to 37.2 °C and Sendai City experiences fewer days of extreme temperatures (highs outside 0 – 30 °C at 19.6 days/year compared to Tokyo's average of 49 days/year.

Winters are cool and relatively dry, with the January temperature averaging at 1.5 °C. Concerning snow, snowfalls are much lower than at the cities on the Sea of Japan coast, such as Niigata and Tottori (Climate data for Sendai normals, extremes [1926–present], Japan Meteorological Agency, http://www.data.jma.go.jp).

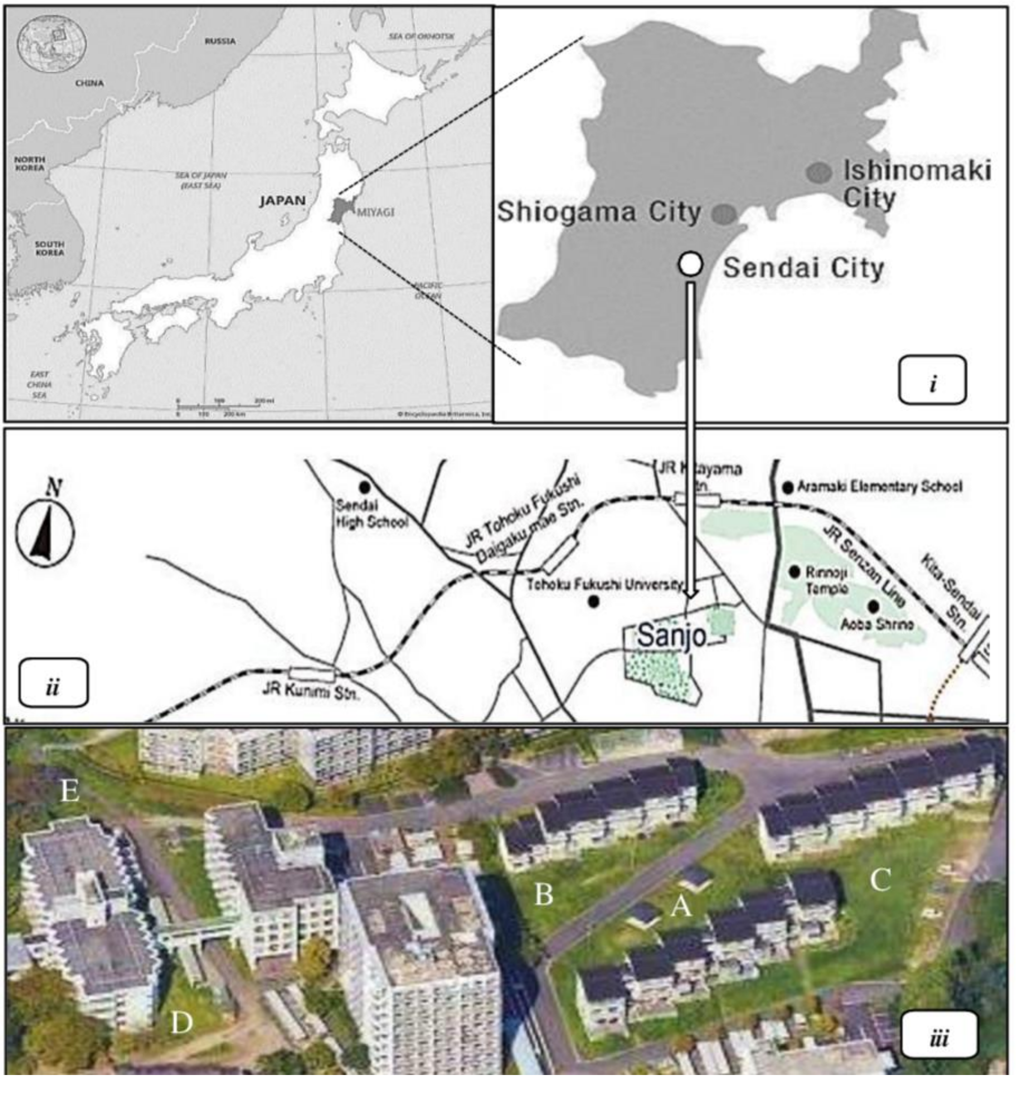

FIG. 1.**:** Map of the study site: (*i*) Map of Japan showing Miyagi Prefecture and Sendai City, Japan, (*ii*) Map of Sanjo-machi with the grassland plots (*iii*) aerial photograph showing the five grassland plots within Sanjo-machi (marked by white capital letters). (Source: Tohoku University https://www.tohoku.ac.jp/english/profile/campus/01/access/)

Study site. The study site comprised of five main plots of grasslands (Fig. 1). Grassland A and B were longitudinal, L-shaped transects, covered by short and long grasses, with a presence of several shrubs. Grassland C was adjacent to a parking lot. It was a longitudinal quadrate plot covered by short grasses, with long grasses on the edges. Grassland D was a T-shaped plot, with a canopy almost dominated by relatively short grasses. Grassland E was almost trapezoid with very few trees scattered all over (Fig. 1). The canopy was composed of mixed grasses and diverse flowering plant species. A small bond was located at the northern edge and was surrounded by long grasses.

In general, on analyzing the selected sampling sites for their floral composition, it was found comparatively rich with several, wildly grown, monocot and dicot species. The most dominant grass species was Bermuda grass, *Cynodon dactylon* (L.).

The five selected grasslands were almost intact, not subjected to any kind of land-use effects, reclaiming or being influenced by severe anthropogenic stresses. The grasslands were only mowed twice per year, the first mow being undertaken after the end of Spring (during the last week of April) and the second at the beginning of Autumn (during the first week of November) and before initiation of snow falls. Each mow lasted 2-3 days for all five grasslands. It should be noted that a negative effect on Orthoptera presence due to mowing (Humbert et al. 2010; Cizek et al. 2012; Chisté et al. 2016) was not possible since sampling was performed during a period not associated with the post-mowing period, as orthopterans start to appear at the end of June and completely disappear by the beginning of October.

Sampling protocol. Orthopteran assemblage was collected from the study sites, from the beginning of July until the end of September, over two consecutive years, 2022 and 2023, using standard sweep net. Concerning the time effort per unit, the sampling schedule was adjusted to be performed between 1000 and 1400 h. To minimize the bias in sampling, only one of the researchers was responsible for sampling the individuals of different orthopteran species. The collected individuals were promptly killed in the field and stored in proper sampling containers. Details of the sampling technique are given by ElEla (2011).

The collected specimens were brought to the laboratory for systematic identification using the key of Ichikawa et al. (2006). Individuals of different orthopteran species were counted, sorted and kept in individually marked clean glass vials for further analyses. Additional identified specimens were stored individually in secure vials which then freezed (- 25 ◦C).

Feeding category. Feeding categories of different orthopteran species were elucidated from mandibular morphological characteristics and compared with previous reports, when information data was available. Laboratory dissections were followed immediately after bringing the collected specimens to the lab. Mandibles of adult orthopterans were exposed by lifting the labrum and hauling the maxillae. The mandibles were lightly brushed with 80% ethyl alcohol and distilled water to remove adhering debris (ElSayed, 2005; Smith and Capinera, 2005). Mandibles were then carefully detached from the head capsule by loosening the abductor muscles connecting the mandibles to the head capsule by regular and gentle back and forth hauling of each mandible using forceps, under a stereomicroscope (Olympus SZX12, Hamburg, Germany). Mandibles were then examined at 20-40x magnification, under direct illumination and photographs were taken from both ventral and dorsal views of each mandible with a digital camera (Sony HDR-CX590V, Japan). Morpho- mandibular characteristics were carefully examined and recorded in attempts to predict the feeding category of each orthopteran species.

Due to the rather wide gap in the relative abundance amongst orthopteran species, five adult male individuals from each species were subjected to gut contents analysis (total n = 215). As in many instances, females of certain orthopteran species feed more often than males (Hochkirch, 1999), it is necessary to clarify that only males were processed in the analyses since food strategies often differ between sexes, where it has been shown that females could consume plant species richer in nutrients (Chapman, 1990) and fertilized females tend to consume more protein, so even in purely herbivorous species, arthropod remains in the gut could appear during this period for relatively higher demand in protein requirements for production of eggs (ElSayed, 2005). For species with more than five adult males, the five individuals were randomly selected among collected males.

Adult male individuals of the collected orthopteran species were dissected by dissecting scissors and the alimentary tract was exposed, then longitudinally opened and the contents of the gut were permanently mounted on a glass microscope slides (Mulkern and Anderson, 1959; Brusven and Mulkern, 1960; Ohabuike, 1979; Kang and Chen, 1994; LeGall et al., 1998, 2003; ElSayed, 2005; ElSayed et al., 2020). Drops of distilled water wereadded whenever required to minimize dryness of the collected gut contents. These slides were examined under a light binocular microscope (Olympus® CX41RF, Hamburg, Germany) at 40× magnification. Microscopic examinations of fragments including different plant species (monocots and dicots), arthropod body parts and/or other ingested matters including debris particles were performed (Kang and Chen, 1994; ElSayed, 2005; ElEla et al. 2010; ElSayed et al. 2020).

Qualitative records of the gut contents were made following ElEla et al. (2010) and ElSayed et al. (2020) to categorize each orthopteran species into a proper feeding category. The proposed seven feeding categories were:

1. Herbivorous feeders (H): The frequency of fragments of dicots species ≅ frequency of fragments of monocots species with absence of fragments from arthropod parts.
2. Herbivorous-mixed feeders (Hm): the same as herbivorous category with arthropod parts recorded in the gut.
3. Graminivorous feeders (G): Frequency of fragments of monocots species exceeding 75% of the gut contents and no arthropod matter detected.
4. Forbivorous feeders (F): Frequency of fragments of dicots species exceeding 75% of the gut contents with absence of fragments from arthropod parts.
5. Forbivorous-mixed feeders (Fm): resembles the forbivorous category with arthropod parts recorded in the gut.
6. Mixed-feeders (M): plant matters (including roots, tubers and subterranean parts) and arthropod and/or earthworm (Oligochaeta) parts encountered in almost equal proportions.
7. Detrito-bryophagous feeders (Db): detritus, mosses, algae and lichens were mainly encountered in the gut beside other plant matters.

Results

Taxonomic composition

The entire study area, for all five sites pooled, a total of 43 species (28 and 35 in 2022 and 2023, respectively), representing 25 genera, 7 families, 12 subfamilies and 13 tribes (Table 1) were recorded over the two consecutive years. The total number of collected individuals was 988 and 1324 during 2022 and 2023, respectively. The most dominant orthopteran species was *Atractomorpha lata* (Mochulsky, 1866). Overall, five orthopteran species were dominating the assemblage. These species were *A. lata* (Mochulsky, 1866), *Conocephalus maculatus* (Le Guillou, 1841), *Loxoblemmus equestris* (Saussure, 1877), *Mecopoda niponensis* (Haan, 1843), *Acrida cinerea* (Thunberg, 1815) (Table 1). These species constituted 59.72% of the total assemblage during 2022 and 52.34% of the total assemblage during 2023 (Table 1). The sex ratio varied considerably among the different species and even between years (Table 1). Moreover, an absence of males or females of relatively rare species, e.g. *Locusta migratoria* (Linnaeus, 1758) and *Loxoblemmus tsushimensis* (Ichikawa, 2001) was recorded, as well as the absence of both sexes of other species e.g. *Euconocephalus varius* (Walker, 1869) during a specific year, 2022, was also recorded (Table 1).

Feeding category

Based on the observations of the variability of mandibular characteristics and analysis of contents of the alimentary canal, seven different feeding categories were identified (Table 1). Out of 43 recorded species sampled over the study period, 12 species (ca. 27.91%) were mixed-feeders (M) (Table 1).

Gryllidae and Trigonididae showed mandibles with comparatively sharp incisors and relatively long knife-shape terebral ridge. These mandibular characteristics could delineate a predacious feeding behavior. Based on postmortem gut content analyses, guts of dissected species showed fragmentary parts of plant materials including roots, occasionally debris matters and subterranean arthropod body parts. Consequently, the feeding category of these eight species could be positioned in the mixed-feeding category (M).

On the second rank of our recorded feeding categories, nine detrito-bryophagous feeders (Db), which comprised approximately 20.93% of the categories, were recorded in all tetrigid species (Table 1). Dissected guts of these species showed peculiar contents in which their guts were mainly loaded with detritus, algal parts, mosses and fungal hyphae.

Graminivorous (G) and forbivorous-mixed feeders (Fm) were positioned in the third rank (ca. 16.28% for each category). The main graminivorous feeders frequently showed mandibular abrasions (Fig. 2). Both incisor and molar areas were subjected to obvious mechanical wear as a result of feeding habit, as is the case for *Acrida cinerea* (Thunberg, 1815) (Fig. 2).

On the other hand, species which are members in Conocephalini and Mecopodini were forbivorours-mixed feeders (Fm) where the incisor area was prominent and robust comprising a comparatively larger portion of the mandible and was associated with sharp terebral ridge (Fig. 2). In comparison, the molar area was small with two short and parallel molar slats.

Three species were forbivorous (F) in which the mandibular characteristic features were associated with the processing of forb resources (Table 1). The incisor area is equipped with robust and sharp teeth. The molar area consists of a small molar ridge forming a trituration area with molar slats for grinding forbs thoroughly (Fig. 2). The gut content analyses of these species showed that the contents were loaded with irregular fragments of forbs.

The three Phaneropterini species, *Hexacentrus japonicus* (Karny, 1907), *Phaneroptera nigroantennata* (Brunner von Wattenwyl, 1878) and *P. falcate* (Poda, 1761) were herbivourous-mixed feeders (Hm), with prominent and sharp incisors, without prominent wearing, and a comparatively smaller molar area (Fig. 2).

TABLE 1. Orthopteran species collected from the study sites in Sendai, Japan, with their families, subfamilies, tribes, number of individuals and their feeding category.

Family	Subfamily	Tribe	Orthopteran species	No. of individuals (♂:♀) 2022	No. of individuals (♂:♀) 2023	Feeding category*
Acrididae	Acridinae	Acridini	*Acrida cinerea* (Thunberg, 1815)	54 (23: 31)	47 (19:28)	G
			A. conica (Fabricius, 1781)	33 (18:15)	39 (24:15)	G
	Melanopilinae	Podismini	*Parapodisma mikado* (Bolivar, 1890)	0	11 (8:3)	F
	Oedipodinae	Aiolopini	*Aiolopus thalassinus tamulus* (Fabricius, 1798)	23 (10:13)	10 (6:4)	H
			Oedaleus infernalis (Saussure, 1884)	10 (3:7)	19 (12:7)	G
		Locustini	*Locusta migratoria* (Linnaeus, 1758)	0	7 (5:2)	G
			Gastrimargus marmoratus (Thunberg, 1815)	0	9 (5:4)	G
	Oxyinae	Oxyini	*Oxya japonica* (Thunberg, 1815)	12 (4:8)	22 (15:7)	G
			O. yezoensis (Shiraki, 1910)	0	8 (5:3)	G
Pyrgomorphidae	Pyrgomorphinae	Atractomorphini	*Atractomorpha lata* (Mochulsky, 1866)	308 (131:177)	458 (168:290)	F
			A. sinensis (Bolivar, 1905)	0	14 (8:6)	F
Tetrigidae	Tetriginae	Tetrigini	*Euparatettix tricarinatus* (Bolívar, 1887)	13 (8:5)	0	Db
			Formosatettix niigataensis (Storozhenko & Ichikawa, 1993)	0	15 (7:8)	Db
			Tetrix akagiensis (Uchida & Ichikawa, 1991)	13 (9:4)	0	Db
			T. japonica (Bolivar, 1887)	21 (15:6)	29 (13:16)	Db
			T. kantoensis (Uchida & Ichikawa, 1991)	29 (18:11)	28 (14:14)	Db
			T. larvatus (Bei-Bienko & Mishchenko,1951)	16 (6:10)	37 (16:21)	Db
			T. macilenta (Ichikawa, 1993)	18 (6:12)	22 (14:8)	Db
			T. minor (Ichikawa, 1993)	7 (2:5)	0	Db
			T. silvicultrix ichikawa (Ichikawa, 1993)	20 (6:14)	21 (12:9)	Db
Eneopteridae	Oecanthinae	Oecanthini	*Oecanthus similator ichikawa* (Ichikawa, 2001)	12 (6:6)	0	F
			Loxoblemmus aomoriensis (Shiraki, 1930)	23 (9:14)	48 (22:26)	M

Gryllidae	Gryllinae	Gryllini	*L. equestris* (Saussure, 1877)	68 (39:29)	69 (45:24)	M
			L. tsushimensis (Ichikawa, 2001)	0	12 (7: 5)	M
			L. sylvestris (Matsuura, 1988)	6 (5:1)	0	M
			Teleogryllus emma (Ohmachi & Matsuura, 1951)	0	12 (5:7)	M
			T. infernalis (Saussure, 1877)	8 (2:6)	20 (6:14)	M
			Trigonidium pallipes (Stål, 1861)	7 (7: 0)	0	M
			Velarifictorus aspersus (Walker, 1869)	0	7 (5:2)	M
			V. mikado (Saussure, 1877)	43 (17:26)	90 (36: 54)	M
			V. ornatus (Shiraki, 1911)	14 (8:6)	0	M
Tettigoniidae	Conocephalinae	Conocephalini	*Conocephalus maculatus* (Le Guillou, 1841)	75 (43:32)	94 (59:35)	Fm
			C. melaenus (Haan, 1843)	21 (14:7)	14 (5:9)	Fm
			C. japonicus (Redtenbacher, 1891)	0	9 (6:3)	Fm
			Euconocephalus varius (Walker, 1869)	0	11 (8:3)	Fm
			Ruspolia dubia (Redtenbacher, 1891)	0	12 (10:2)	Fm
			R. lineosa (Walker, 1869)	38 (22:16)	59 (27:32)	Fm
	Mecopodinae	Mecopodini	*Mecopoda niponensis* (Haan, 1843)	85 (39:46)	25 (7:18)	Fm
	Phaneropterinae	Phaneropterini	*Hexacentrus japonicus* (Karny, 1907)	0	11 (7:4)	Hm
			Phaneroptera falcate (Poda, 1761)	0	20 (15:5)	Hm
			P. nigroantennata (Brunner von Wattenwyl, 1878)	0	8 (6:2)	Hm
Trigonididae	Nemobiinae	Pteronemobiini	*Dianemobius furumagiensis* (Ohmachi & Furukawa, 1929)	0	7 (5:2)	M
			Pteronemobius fascipes (Walker, 1869)	11 (6:5)	0	M
		Total number of individuals (♂:♀)		**988 (476:512)**	**1324 (632:692)**	

*Feeding category: Db: Detrito-bryophagous feeder, F: Forbivorous feeder, Fm: Forbivorous-mixed feeder, G: Graminivorous feeder, H: Herbivorous feeder, Hm: Herbivorous-mixed feeder M: Mixed feeder.

Table (1) showed that only one acridid species, *Aiolopus thalassinus tamulus* (Fabricius, 1798), possessed a purely herbivorous feeding category (H) which comprised approximately the least proportions of the total categories (ca. 2.33%). The incisor and molar areas were almost of uniform proportions. The incisors were parallel and sharp with no clear abrasions either in molar or incisor areas of the mandible (Fig. 2).

It was clear that the major differences in mandibular structure were related to differences in feeding behavior rather than the taxonomic position of the species as family Acridinae and Tettigoniidae possessed more than feeding category (Table 1).

Discussion

In the majority of phytophagous insect orders, Chapman (1982) noted that oligophagy or monophagy was observed in more than 50% of the species whereas the Orthoptera, stand apart from the other orders, more than 60% of the species have been classified as polyphagous and 25% as graminivorous (Picaud et al. 2003).

Orthopteran species are considered as one of the more diverse taxa with thousands of extant described species possessing prominent diversity in forms and habitats (Grimaldi and Engel, 2005; Bidau, 2014; Eades et al. 2014; Song et al. 2015) and representing crucial links in food chains (Badenhausser, 2012). Orthopteran species are widespread and important herbivores in natural grassland ecosystems and agro-ecosystems as they consume a considerable portions of grasses and forbs (Köhler et al. 1987, Blumer and Diemer 1996; ElSayed, 2005; Bharamal and Koli 2014) and regulate plant community structure (Zhang et al. 2011). Indeed, Order Orthoptera plays an influential and functional role in the recycling and equilibrium of natural ecosystems by enhancing the conversion of biomass through their easily transformable excrement (Samways, 1994; Hao et al. 2015; Ngoute et al. 2021). All the previous aspects triggered the urge to study the variations in feeding categories of different orthopteran species through mandibular morpho- characteristic variations and gut contents analysis rather than relying on field or laboratory observations or food choice tests.

The aspects of food strategies including selection of food, feeding patterns, food preferences, specialization in dietary requirements have been studied by many authors in a diverse orthopteran species (e.g., Williams, 1954; Gangwere, 1961; Mulkern, 1967; Otte and Joern, 1976; Bernays and Chapman, 1978; Uvarov, 1977; Joern, 1983, 1985; Bernays and Bright, 1993). Still the food strategy based on the structure of the mouthparts, habitat preferences, or feeding behavior was observed in only a few species (ElSayed et al. 2020).

Morphological variations in mouthparts, including mandibles, were subjected to further thorough research and itemized investigations have been directed by numerous authors in various regions, significant among them were Snodgrass (1928), Gangwere (1965, 1966), Gangwere et al. (1976, 1998) and Patterson (1984), Smith and Capinera (2005) in North America, Liebermann (1968), Gangwere and Ronderos (1975) in South America, Williams (1954); Kaufmann (1965), Gangwere and Morales (1973) in Europe, Gangwere and Spiller (1995); Gangwere et al. (1998) in the Mediterranean islands, Gapud (1968), Feroz and Chaudhry (1975), Kang et al. (1999), ElEla et al. (2010, 2012), ElSayed et al. (2020) in Asia, Chapman (1964), Le Gall et al. (1998, 2003) and ElSayed (2005) in Africa.

The strong relationship with diet makes morphological characteristics of mouthparts an important trait for insect evolutionary biologists (Snodgrass, 1928; Brues, 1939) and systematists (Mulkern, 1967). One of the first who coped with this topic was Isely (1944) who studied the de-

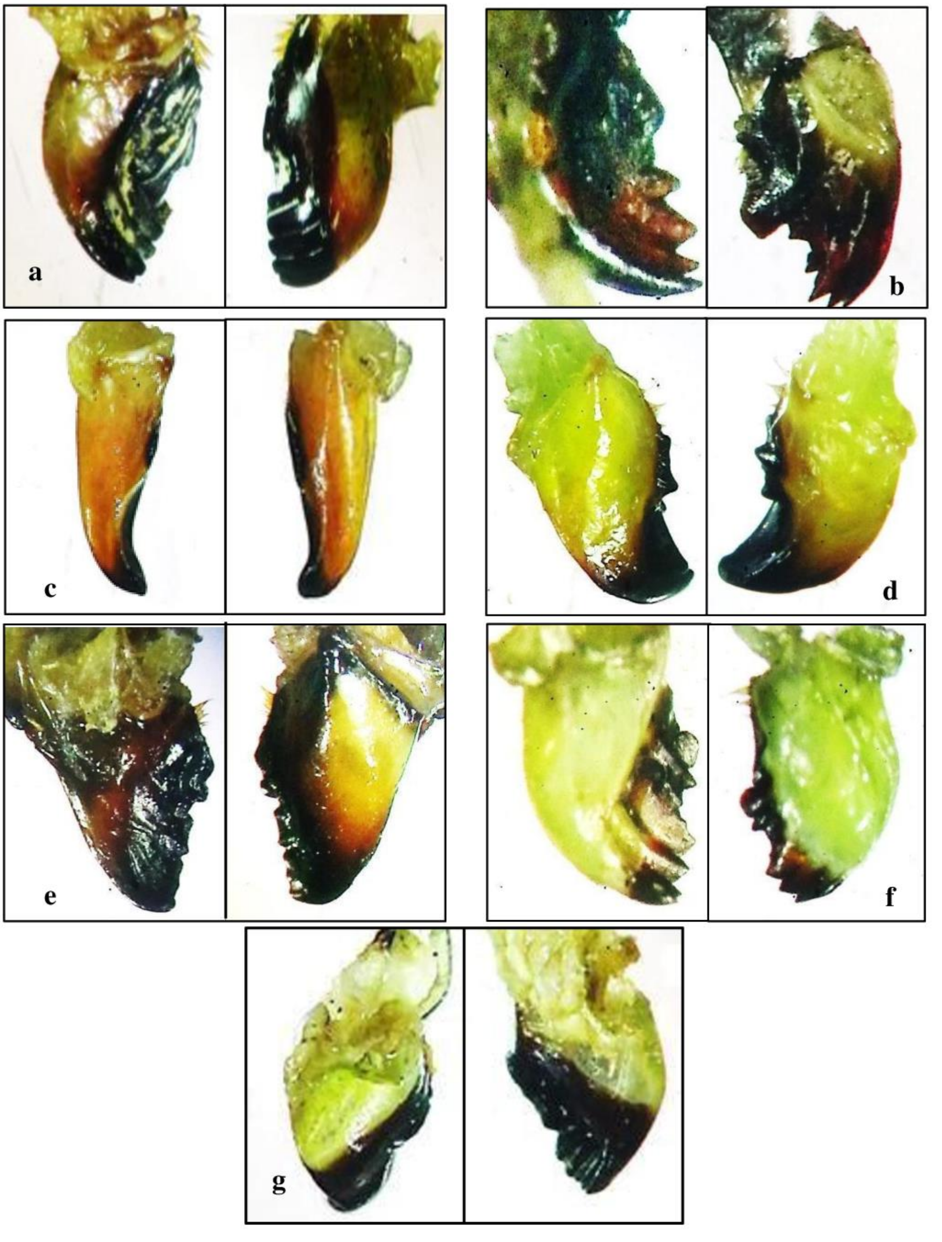

FIG. 2.**:** Images of morpho-mandibular variability representing different feeding categories. a. *Tetrix japonica* (Bolivar, 1887), a representative of Forbivorous species (F); b. *Hexacentrus japonicus* (Karny, 1907), a representative of Herbivourous-mixed species (Hm); c. *Conocephalus melaenus* (Haan, 1843), a representative of Forbivorous-mixed species (Fm); d. *Velarifictorus mikado* (Saussure, 1877), a representative of Mixed-feeder species (M); e. *Aiolopus thalassinus tamulus* (Fabricius, 1798), a representative of Herbivorous species (H); f. *Acrida cinerea* (Thunberg, 1815), a representative of Graminivorous species (G).

tailed morphological and structural features of mouthparts and correlated these characteristics with various feeding habits. Initial results and information offered by Isely (1944) on categorizing mandibles according to mandibular groups have since been shown to be prevalent, especially, in grasshoppers and other entomological taxa.

Isely (1944), on his research on 89 species, proclaimed two extreme forms of mandibles, i.e. graminivorous and forbivorous, with a series of intermediate forms associated with a mixed diet. Generally, Isely (1944) defined three groups of mandibles according to the overall structure and distinctive diet: (i) graminivorous (grass feeding type) with molar area adapted for grinding and incisors typically merged into a scythe like edge, (ii) forbivorous(forb or broad leaf plant feeding type) having a molar area composed of a depression encircled by elevated teeth and a strong interlocking incisor, (iii) herbivorous (mixed- feeding type) that have features of both of the previously mentioned groups.

On the other hand, Joern (1979) proposed that orthopteran species possess a broad spectrum in diversity of diet and consequently feeding behavior ranged from strict specialist to extreme generalist.

In accordance, several researchers (Gangwere, 1965; Chapman, 1990; Smith and Capinera, 2005) agreed with Isely (1944) and they had merely classified three main categories based on the plant categories. These categories were graminivorous, forbivorous and herbivorous (mixed-feeding). However, due to variations in mandibular morpho-characteristics, other studies provided broader classification of feeding categories (Kuřavová and Kočárek, 2016; ElEla et al. 2010; ElSayed et al. 2020).

Comparisons were performed by Patterson (1983) concerning the morphological structure of the mandibles with their niche variations. Hypothesis of niche variations predicts greater overall mandibular characteristic variability in orthopteran species having relatively the broadest trophic niches. In this study, mandibular characteristics of orthopteran species vary in different groups and these variations are therefore believed to meet the different feeding habits and in correspondence specifying a definite feeding category. These were in accordance with findings of several studies (Gangwere, 1965; ElEla, 2011; ElSayed et al. 2020).

Apparent observed similarities among members belonging to the same subfamily and striking differences between subfamilies assume a strong phylogenetic component to feeding adaptations as suggested by Otte and Joern (1977) and Joern (1979).

Ensiferan species, on the other hand, feeding categories ranged from forbivorous category which was observed in a single eneopterid species to strictly mixed feeding category. Ensiferans tended to feed on taxonomically diverse plant species (Palmer et al. 2024) beside frequent proportions of arthropod parts which could suggest predaceous or cannibalistic habits where some ensiferan species specialized on rather unusual food sources (ElEla, 2010; ElSayed et al. 2020).

The consumption of a specific diet is commonly associated with the shape and structure of the mandibles. Graziella et al. (2015) showed significant differences in the shapes of the mandibles among the variant forms of grasshopper species which may be due to the role of trophic diversification in the morphological differentiation of insect. The apparent diverse variations among mandibular characteristic features of different orthopteran species were remarkable to consider. In our research, more types of mandibles were suggested in association with analyses of gut contents in an attempt to position each collected species in the proper feeding category.

Although orthopteran species have often been regarded as polyphagous herbivores (ElSayed et al. 2020), most of these species show degree of dietary specialization (Mulkern, 1967; Uvarov, 1977; ElSayed et al. 2020). Orthopteran species possessed a dietary specialization was reflected in patterns of mandibular variations, and yet this diversification in morpho-mandibular characteristics could be utilized to catalogue orthopteran species in proper feeding category.

In this study, caeliferan species were strictly not engaged in cannibalistic behavior as confirmed by the absence of arthropod parts in their guts. However, other studies have recorded cannibalistic behavior in some caeliferan species only under crowded conditions (Bomar and Lockwood, 1994; ElEla, 2010).

It was interesting to spot the light on the peculiar detrito-bryophagy which was restricted to the tetrigid species with mosses, algal hyphae and debris matters were frequently encountered in their guts (Kuřavová and Kočárek, 2016).

Studies performed on tetrigids are considerably rare and restricted to a few species (Paranjape and Bhalerao, 1985; Blackith and Blackith, 1987; Reynolds et al. 1988; Hochkirch et al. 2000, Gröning et al. 2007; Kuřavová and Kočárek 2015; Kuřavová and Kočárek, 2016; Musiolek and Kočárek 2016), and even fewer and fragmentary in Japan (ElEla et al. 2010; ElEla, 2011; Tan et al. 2017; ElSayed et al. 2020).

The feeding habit of Tetrigidae has been described, ranging from observational to quantitative data, in only a few species (Tan et al. 2017). However, the detrito-bryophagy has been observed in almost all species (Verdcourt, 1947; Paranjape and Bhalerao, 1985; Hochkirch et al., 2000; Bidau, 2014; Kuřavová and Kočárek, 2015).

In a related species, *Tetrix undulata* (Sowerby, 1806), Verdcourt (1947) performed the faecal analysis and found a variety of mosses species in 80% of the fecal pellets. Lock (1996) analyzed crop content of three specimens of *T. subulata* (Linnaeus, 1758) and observed the presence of algae, detritus and sand grains with no traces of mosses or higher plants. Dietary habit of *T. undulata* (Sowerby, 1806) was studied by Hodgson (1963) and he observed feeding of the species on grasses (monocots), mosses (variety of species), lichens and algae, and on humus. Besides that, he observed also feeding on the dead bush cricket *Pholidoptera griseoaptera* (De Geer, 1773) in insectarium. Further studies were conducted by Hochkirch et al. (2000) who found that males of *T. undulata* (Sowerby, 1806) were exclusively feeding on mosses and algae, while females also included grasses and forbs in their diet. On other related tetrigid species, Reynolds et al. (1988) studied the diet of two tropical tetrigids in Sulawesi by analyzing their crop contents and they observed the presence of mosses or vascular plants in 82% of specimens of *Scelimena celebica* (Bolivar, 1887) and in 100% of specimens of *Diotarus pupus* (Bolivar, 1887). Indeed, it is far from precise to extend the detrito-bryophagy among all members of Tetrigidae since more documentation of dietary preference for each species is required in different spots and among different sexes.

Feeding category could be considered as an additional important ecological trait used to analyze the structure of orthopteran fauna. The collected orthopteran species were relatively dominated by forbivorous (F) and mixed (M) feeding species. However, mixed-feeding habit also comprised a relatively major percentage in the feeding categories of the assemblage.

Although Min and Min (2008), in their taxonomic studies, mentioned that *Aiolopus thalassinus* is an important graminivorous pest on different plants and grasses; our results from mandibular structure and gut contents analysis revealed a forbivorous feeding category for this species.

Comparisons in our research have shown that orthopteran species of the family Acrididae exhibited the broadest trophic niches with three feeding categories which support diverse patterns of mandibular characteristics and dietary preferences. Herbivorous and forbivorous species with mixed feeding habits (Hm and Fm) have more complex mandibular characteristics and relatively more heterogeneous food intake. These associations, in regard of morphological characteristics and dietary preference in each group, support Patterson's niche variation hypothesis Patterson (1983).

Possibly, species with mixed feeding categories (M, Fm and Hm) circumvent the defense systems by feeding only small portions of many plant species and other organic parts thus keeping the toxic levels of secondary metabolites (allelochemicals) below some critical level (Freeland and Janzen, 1974). However, since this is a per se mechanism and is species-specific, more investigations are required.

Although little information is known about the chemical properties of most grasses in the study sites, grasses accumulate relatively high levels of silica in their tissue which could be an alternative tactic, with potential benefits, including antiherbivore defense (Vicari and Bazely 1993; ElSayed et al. 2020; Quigley et al. 2020).

Graminivorous species differ in their dietary choices when feeding on graminoids (monocots) in natural grassland vegetation when compared to forbivorous or herbivorous species and this considered as species-specific (Joern, 1985). Bemays et al. (1989) have also reported that tannins are often considered a major class of allelochemical defense in dicotyledonous plant species (dicots) and they have also been considered as deterrents to graminivorous orthopteran species (Bemays and Chapman, 1977) which could partly explain the selection of grasses by graminivorous species.

Mandibular abrasion in both incisor and molar areas in graminivorous species (G) could be attributed to the fact that these species feed regularly on silica-rich grasses. These silica particles could act as a sand paper that wear the mandibles of these graminivorous species (ElSayed et al. 2020). It was reported that silica causes wear to insect mandibles which could potentially impact on the performance of herbivory (Baker et al., 1959; Vicari and Bazely, 1993; Massey and Hartley, 2009; ElSayed et al. 2020).

Our analysis of the gut contents of orthopteran species with mixed-feeding category (M) or even species of mixed feeding habit beside main category (i. e. Hm and Fm) could suggest that arthropod parts and other detritus of plant matters are not incidentally consumed but they are main part of their dietary preference. These species could intentionally feed on arthropod parts which were predominant in the gut of the entire dissected individuals. However, more detailed investigations and analyses on this aspect are required.

Conclusion

The gathered data on the comparatively small assemblage give only a hint concerning the whole story of evolution of functional food strategies among orthopeteran species. A general scheme for explicating the diet of a given orthopteran species, or other taxa group, could be started with detailed inspections of their mandibular morphological characteristics.

Although most species with forb feeding mandibles could feed on mixture of grasses and forbs, one of the confirmation avenue is associating mandibular characteristics with observations and data gathered from analyzing the gut contents.

It is hoped that more researchers will find that the study of morpho-mandibular characteristics and gut contents analysis is rewarding will contribute to the advancement of our knowledge concerning feeding strategies in Orthoptera. This research tries to open the door for more studies to analyze more mandibles from more taxa (Patterson, 1983).

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Conflict of interest

The authors declare no conflict of interest associated with this article and Prof. Dr. Ames Cheryl Lynn (International Integrative Research and Instruction Unit, Tohoku University, Japan) for her persistent cooperation and continuous encouragement.

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Διατροφική Κατηγοριοποίηση ορισμένων ειδών Caelifera και Ensifera (Insecta: Orthoptera) από επιλεγμένους αγρούς στην πόλη Sendai της Ιαπωνίας

Abu ElEla Shahenda, A. A.[1, 3], Michio Kondoh[2], Masatoshi Hori[3], Atsuhiko Nagasawa[3], Koji Nakamura[4], Wael M. ElSayed[1]

[1]*Department of Entomology, Faculty of Science, Cairo University, Egypt 12613*

[2]*Graduate School of Life Sciences, Tohoku University, 6-3 Aoba, Aramaki, Aoba-ku, Sendai 980-8578, Japan*

[3]*Graduate School of Agricultural Science, Tohoku University, Sendai, Miyagi 980-8572, Japan. Email: masatoshi.hori.a3@tohoku.ac.jp*

[4]*Ishikawa Prefectural Natural History Museum, Kanazawa, Ishikawa 920-1147, Japan*

ΠΕΡΙΛΗΨΗ

Αν και τα είδη των ορθοπτέρων έχουν συχνά θεωρηθεί ως πολυφάγα φυτοφάγα, τα περισσότερα από αυτά παρουσιάζουν ποικίλους βαθμούς επιλεκτικότητας και ιδιαίτερες προτιμήσεις ενδιαιτήματος. Επιπλέον, ορισμένα είδη παρουσιάζουν ιδιόμορφες προτιμήσεις διατροφής που χρήζουν περαιτέρω διερεύνησης. Δείγματα από ένα σύνολο 43 ειδών ορθοπτέρων, από 7 οικογένειες, 12 υποοικογένειες και 13 φυλές, από πέντε αγρούς στην πόλη Σεντάι της Ιαπωνίας, συλλέχθηκαν για να διερευνηθούν οι διατροφικές τους προτιμήσεις, και ταξινομήθηκαν σε επτά κύριες διατροφικές κατηγορίες με βάση την εξέταση των μορφολογικών χαρακτηριστικών της γνάθου και αναλύσεις του μεταθανάτιου περιεχομένου του εντέρου.

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Indications of Understorey Management Practices impact on Vascular Plant and Arthropod Diversity in Olive Groves on Lesvos, Greece

N. SCHEEPENS, G. STAVRIANAKIS*, R.T. KOMBA, E. SENTAS AND T. KIZOS

Laboratory of Rural Geography & Precision Farming Systems, Department of Geography, University of the Aegean, Mytilene, Greece

ABSTRACT

This study investigates the impact of three understorey management practices – herbicide application, understorey clearing, and undisturbed understorey – on the biodiversity of plant and arthropods in olive groves in the Gera region on Lesvos, Greece. The study found that herbicide application had a negative effect on plant diversity, but less pronounced effects on arthropods. The rapid recovery of arthropod biodiversity is likely due to the high structural complexity in the Gera region. Abandoned olive groves displayed the lowest arthropod abundance and vegetation, leading to gradual impoverishment of plant biodiversity and negative impacts on arthropod diversity. The proportion of annual species in the plant cover was positively associated with arthropod abundance. The study proposes a new eco-scheme that supports farmers for maintaining understorey plant cover, with periodical clearing through ruminant grazing, to enhance plant and arthropod biodiversity in olive grove systems.

KEY WORDS: Mediterranean ecosystems, olive cultivation, agrobiodiversity, plant diversity, arthropod diversity, understorey management practices.

Introduction

A crop that has traditionally been extensively managed in structurally complex and stable agro-forestry systems, supporting high levels of biodiversity, is the olive tree (*Olea europaea* L.) (Sobreiro et al. 2023; Stattegger et al. 2023; Vasconcelos et al. 2022). The importance of olive groves globally is evident, with the world's cultivation area being approximately 10.3 Mha in 2021, yielding around 23 million tons of olives worth 23.891 million US dollars (FAOSTAT 2022; Jiménez 2023). Of the total production, 95% is originated in the Mediterranean region, signifying the importance of olive groves for the region (Fraga et al. 2020; FAOSTAT 2022).

During the recent years, olive grove management has become more intensive (Carpio et al. 2019), involving increased use of synthetic fertilizers and pesticides and irrigation, changes in soil management techniques, and a shift from traditional low-density (50-200 trees ha^{-1}) agroforestry systems to intensive (401 to 1500 trees ha^{-1}) or super-intensive (1501-2500 trees ha^{-1}) monoculture cropping systems (Guzmán et al. 2022; Jiménez et al. 2023; Jiménez-Alfaro et al. 2020; Sobreiro et al. 2023). On the other hand, there is an increasing trend of agricultural abandonment of marginal olive groves (Van der Sluis et al. 2014). This trend is caused by a combination of agricultural policies and a transition of the economy towards the service sector, causing the marginalisation of farming and an increasing trend of people abandoning

*Corresponding author: gmstavrianakis@geo.aegean.gr

rural areas (rural exodus) (Carmona-Torres et al. 2023).

The intensification of olive cultivation has negative impacts on biodiversity and agroecosystems, leading to the simplification of landscapes and removal of natural vegetation. Sustainably managed olive groves have the capacity to support high levels of biodiversity, which provide a range of ecosystem services (Bateni et al. 2021; Berg et al. 2018). In olive groves, functional diversity plays a pivotal role in crop protection, biological control of pests and overall productivity (Castro et al. 2021; Gkisakis et al. 2016). Plant life provides balance to any ecosystem, as it contributes to the moderation of climate, regulation of water flow and reduction of soil erosion, reducing the risk of runoff and nutrient loss (Solomou & Sfougaris 2021).

This paper presents the first results of this research. The study investigates the impact of three different olive grove understorey management practices (agrochemical use, undisturbed understorey, mechanical clearing of understorey) on plant and arthropod species richness and diversity. The aim of this study is to add to the knowledge of agrobiodiversity in olive grove systems through investigating the effects of selected management practices on richness and abundance of plant and arthropod species.

Materials and Methods

The studied olive groves are situated in the Gera region, located in the south-eastern part of Lesvos. The region spans an area of 86.4 km^2 and its landscape is hilly and characterized by continuous olive groves arranged in terraces, reaching elevations up to 550 metres above sea level. There is minimal cultivation of land for other agricultural activities, and some olive plantations have been abandoned over the past few decades (Dimopoulos et al. 2023). The island of Lesvos has a typically Mediterranean climate, characterized by short, mild winters and hot, dry summers, with significant variations in climatic conditions resulting from the influence of regional mountains and atmospheric circulation patterns (Douma et al. 2016; Stattegger et al. 2023).

Three of the sampling plots are organically managed with undisturbed plant cover, three of the sampling plots are organically managed with cleared understorey, and three of the sampling plots are conventionally managed and occasionally sprayed with herbicides, as is typical in some traditionally managed fields. The cleared sampling plots are grazed by sheep during the summer months and with electrical hand mowers and chainsaws where necessary during the harvest period (October – January). The sprayed sampling plots are sprayed with herbicide, with last spraying having occurred in May 2023. To reduce the influence of surrounding landscape complexity, it was ensured that each sampling plot was surrounded by other plots with similar management practices.

The diversity of the plant ground cover in each sampling plot during the months of March and April was estimated using three linear transect walks of 25m in length of SW-NE direction (Chalmers & Parker 1989; Pieper 1978). Soil arthropod populations were monitored with pitfall traps, while flying insect populations were sampled with yellow sticky traps. Of both trap types, three were positioned within each sampling plot. After a period of seven days, the samples were collected from the traps. Moreover, three sensors (iButtons) were installed in each field to monitor hourly measurements of temperature (ºC) and humidity (%RH).

The data was analysed using the statistical programme IBM SPSS Statistics (Version 29.01.0 2021). The summary statistics of the collected plant and arthropod data were quantitatively described to summarize the plant and arthropod richness and abundance observed across the sampling plots representing the different understorey regimes. and presented with a frequency table. For the collected plant data, several indices of α-diversity, referring to the species richness within a functional community on a local scale, and evenness, a measure of the relative abundance of the different species in an area, were calculated. An analysis of variance test (ANOVA) was conducted to analyse whether the total arthropod abundance and the abundance of certain arthropod taxa differ significantly across the olive groves with different understorey treatments. Following the ANOVA tests, post-hoc tests (Fisher's Least Significant Difference (LSD)) were conducted to further investigate and compare specific groups within the data to determine which pairs differ significantly from each other. To demonstrate the changes in temperature and relative humidity across the sampling periods, the mean, minimum and maximum hourly temperature and humidity were calculated. Besides the comparisons of plant and arthropod richness and abundance across the whole period, it was also analysed whether there were variations in the average and total number of collected specimens for each understorey treatment and across both trap types. For this, summary statistics were obtained and an ANOVA test, followed by a post-hoc test (LSD) were conducted. A linear regression model was carried out to research the relationship between arthropod abundance (dependent factor) and the mean temperature, mean humidity, and presence of annual plant species (as percentage of the total plant cover in March and April, and as percentage of the total floristic composition in May).

Results

From Fig. 1, it can be observed that the sprayed fields have the highest percentage of bare ground (56.7% in April and 38.7% in April, followed by fields with cleared understorey (4.7% in March and 6.7% in April) and undisturbed understorey (1.3% in March and 0.7% in April). The presence of grasses was most abundant in the olive groves with cleared understorey across both months (58% in March and 56% in April), although the percentage of grass cover increased for both undisturbed (33,3% in March and 58% in April) and sprayed fields (16,7% in March and 31,3% in April). The perennial plant coverage was relatively similar across all understorey management regimes. The annual plant coverage was found to be highest in the cleared fields (15,3% in March and 9,3% in April), followed by the undisturbed fields (6% in March and 0,7% in April), and the sprayed fields (0,7% in March and 1,3% in April). The presence of shrubs was predominantly found in the undisturbed fields (18,7% in March and 7,3% in April), but a minimal shrub cover was also detected in the sprayed fields in March (1,3%).

A total of 95 plant taxa were found across the nine sampling plots. These plant taxa belong to 15 orders, 25 families, 60 genera and 78 species. More in detail, 45 plant taxa were observed in the sampling plots with cleared understorey, corresponding to 12 orders, 15 families, 34 genera, and 34 species. In the sprayed fields, 37 taxa were observed, corresponding to 12 orders, 15 families, 26 genera and 32 species. 46 plant taxa were observed in the sampling plots with undisturbed understorey, corresponding to 13 orders, 19 families, 31 genera and 35 species. The species-richest families observed across all nine sampling plots are Poaceae (20,7%), Asteraceae (18,5%), and Fabaceae (13,0%), together comprising more than half of all observed taxa (Table 1).

In terms of α-diversity values, calculated using the Shannon, Menhinick, Margalef, and Simpson indices, it can be derived that α-diversity is lower in the sampling plots with sprayed understorey compared to the cleared and undisturbed sampling plots (Table 2). While α-diversity values for the cleared sampling plots were higher using the Shannon and Menhinick indices, the α-diversity values for the undisturbed sampling plots were higher using the Margalef and Simpson indices. The Simpson's Evenness index was highest in the undisturbed olive groves, narrowly followed by the cleared olive groves, and lastly the sprayed olive groves.

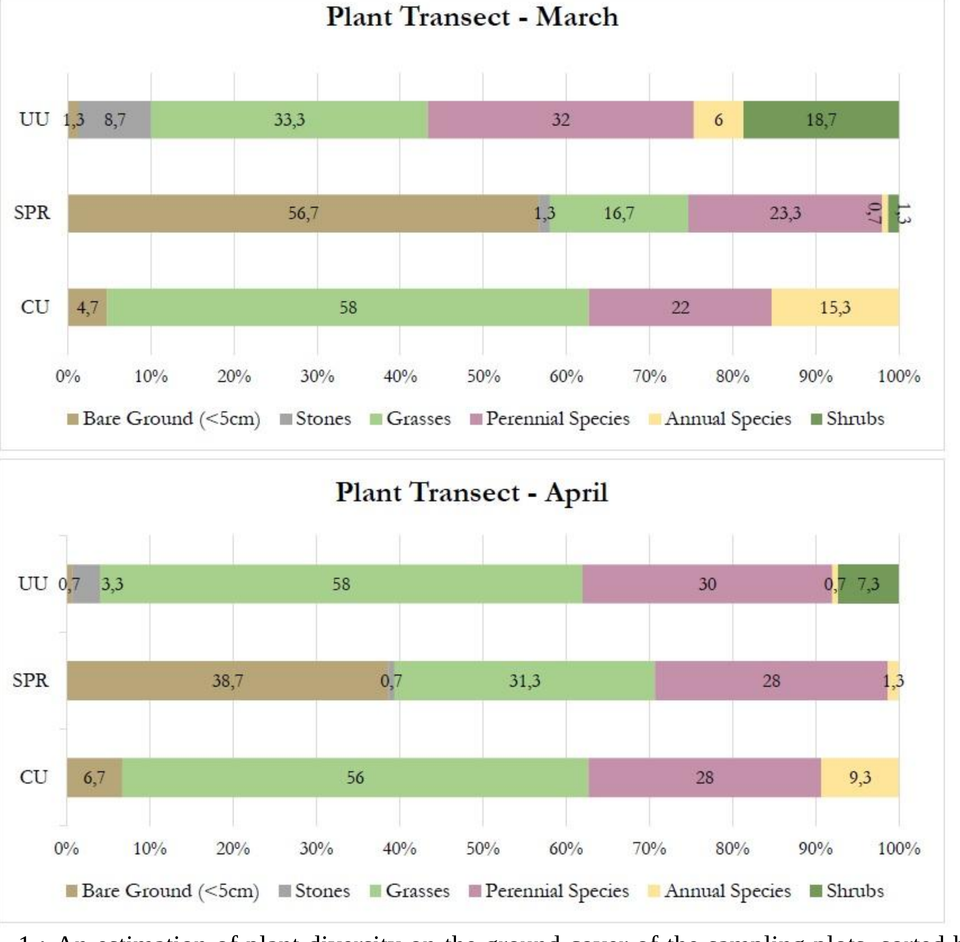

FIG. 1.: An estimation of plant diversity on the ground cover of the sampling plots, sorted by understorey management practices. CU = cleared understorey, SPR = sprayed understorey, UU = undisturbed understorey.

A total of 18.403 arthropods were captured, classified into 23 orders, as well as another 29 families, 19 genera and 3 species, found in all three researched understorey management practices (Table 3). The most dominant order in the whole sampling period were Diptera, accounting for 51,69% of the total catches, followed by Hymenoptera (18,21%), Hemiptera

*Corresponding author: gmstavrianakis@geo.aegean.gr

(13,53%), Coleoptera (5,93%) and Psocoptera (4,78%).

Of the total, 17.009 arthropods were captured via the yellow sticky traps, while 1.394 arthropods were captured via the pitfall traps. In the pitfall traps, arthropods belonging to 17 orders, 12 families, 5 genera and 2 species were found. In the yellow sticky traps, arthropods belonging to 3 classes, 11 orders, 10 families, 5 genera and 1 species were found.

TABLE 1. The most common vascular plant families observed in the nine sampling plots.

Poaceae	20,7%
Asteraceae	18,5%
Fabaceae	13,0%
Apiaceae	5,4%
Caryopyllaceae	4,4%
Gerniaceae	4,4%
Plantaginaceae	4,4%
Other families	29,2%
Total	100,0%

TABLE 2. Values of α-diversity (α) and evenness indices for sampling plots with cleared, sprayed and undisturbed understorey.

	Index	Cleared	Sprayed	Undisturbed
a	Shannon	2,243	1,619	2,144
	Menhinick	1,107	0,822	0,992
	Margalef	6,484	6,296	7,188
	Simpson	0,925	0,876	0,928
Evenness	Simpson's Evenness Index	13,259	8,037	13,925

Values of arthropod catches fluctuated across the different understorey management practices. In the fields with cleared understorey, a total of 6.564 arthropod specimens were collected, while in the sprayed fields and the undisturbed fields 6.253 and 5.586 arthropod specimens were collected respectively. The mean abundance in the cleared fields was the highest, with an average of 20,84 arthropod specimens found. This was followed by the sprayed fields, with a slightly lower average of 20,64 arthropod specimens found, and lastly the undisturbed fields, with an average of 18,94 specimens found (Fig. 2). However, these variations in relative abundance are not statistically significant (Table 4).

The relative abundance of specific taxa observed across the different understorey treatments was compared (Table 5). While the abundance of specific taxa differed across understorey management regimes, these differences were mostly not significant. Overall, a significantly higher soil arthropod abundance was observed in the sampling plots with sprayed understorey. When it comes to specific orders, the abundance of leafhoppers ($p = ,004$) and true bugs (Hemiptera) ($p = ,005$) was significantly higher in sampling plots with undisturbed understorey. The abundance of Hymenoptera, on the other hand, was significantly higher in the sampling plots with sprayed understorey ($p = ,031$). Lastly, the abundance of Psocoptera was significantly higher in the sampling plots with cleared understorey ($p = ,014$).

To investigate the effects of the recorded temperature, relative humidity, and annual plants (as percentage of total plant cover) on total arthropod abundance, a linear regression model was performed (Table 6). When looking at the effects of the individual variables, the results suggest that the percentage of annual plant species has a significant (p = ,005) and relatively strong positive effect on arthropod abundance (unstandardized coefficient B: 7,726). The percentage of annual plant species also has a relatively stronger effect compared to mean temperature (% annuals standardized coefficient B: 0,544; mean temperature standardized coefficient B: 0,045), the effect of which also does not display statistical significance (p = 0,789). The collinearity statistics suggest that there is no multicollinearity between the two explanatory variables of this model based on the tolerance (0,939) and VIF values (1,065).

TABLE 3. Arthropod taxa observed throughout the three rounds of field work conducted in March, April and May 2024. In total 18,403 arthropods were collected.

Order	Understorey Management		
	Cleared	**Sprayed**	**Undisturbed**
Acari	0	2	1
Aranae	109	254	192
Chilopoda	0	0	1
Clitelatta	0	2	0
Coleoptera	442	429	220
Collembola	0	0	1
Diplopoda	34	3	1
Diptera	3.480	3.250	2.783
Embioptera	0	0	1
Gastropoda	10	1	0
Hemiptera	836	443	1.211
Hymenoptera	1.106	1.413	833
Isopoda	0	1	1
Isoptera	6	52	9
Lepidoptera	36	44	46
Neuroptera	2	0	4
Opiliones	1	2	2
Opisthopora	0	2	0
Pseudoscorpiones	1	0	0
Psocoptera	413	302	164
Raphidioptera	0	4	1
Siphonaptera	1	0	0
Sterrnorrhyncha	14	1	4
Thysanoptera	73	48	110
Total	**6.564**	**6.253**	**5.585**

When looking at the effects of the explanatory variable (% annual plant species) across the different understorey management regimes, it can be observed that the only significant model was for the sampling plots with cleared understorey (CU: p = ,037; SPR: p = ,178; UU: p = ,763) (Table 7). For the CU model, the percentage of annual plant species has a strong positive effect on arthropod abundance (unstandardized coefficient (B): 10,411; standardized coefficient B: 0,601) that is statistically significant ($p < ,05$).

TABLE 4. Results of the post-hoc test (LSD) analysing the relationship between understorey management type and arthropod abundance.

Type of Management (I)	Type of Management (J)	Mean Difference (I-J)	Significance
Cleared	Sprayed	,201	,953
	Undisturbed	1,903	,580
Sprayed	Cleared	-,201	,953
	Undisturbed	1,701	,624
Undisturbed	Cleared	-1,903	,580
	Sprayed	-1,701	,624

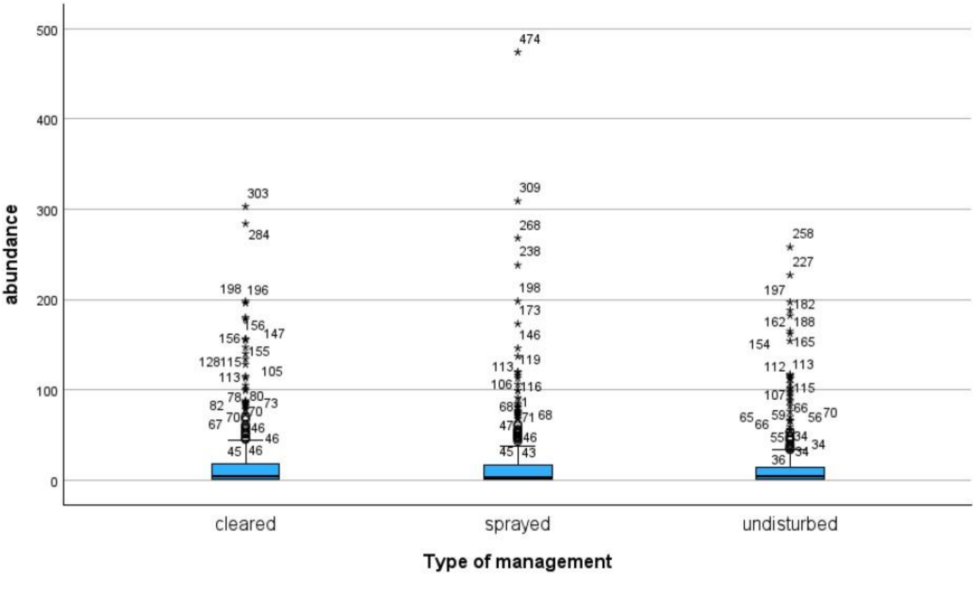

FIG. 2.**:** Boxplot of mean arthropod abundance across the sampling plots representing the different understorey management regimes.

Discussion

The aim of this study was to perform a preliminary investigation on whether plant and arthropod diversity patterns in olive groves located in the Gera region on Lesvos, Greece, differ significantly across the three different analysed management practices (i.e. spraying of herbicides, clearing of the understorey, and undisturbed understorey).

Our study provides valuable insights into arthropod richness and abundance in olive groves under different treatments, during the peak activity period in Lesvos region (March-May). Its short duration limits the long-term study of arthropod communities in this agroecosystem, which

TABLE 5. One-way ANOVA test between overall arthropod richness, abundance, and the olive grove's understorey treatment.

	Understorey Management					
	Cleared		Sprayed		Undisturbed	
	Average	St. Dev.	Average	St. Dev.	Average	St. Dev.
Soil arthropod abundance	4,57	8,05	6,07	11,15	2,70	3,06
Flying insect abundance	30,98	49,96	28,59	56,45	24,08	42,55
Acari	-	-	1,00	,00	1,00	-
Araneae	3,89	5,21	7,94	11,60	7,11	13,26
Carabidae	2,00	1,41	1,00	,00	1,33	,52
Chilopoda						
Cicadellidae	26,88	30,20	13,07	18,35	42,63	43,52
Clitelatta	-	-	2,00	-	-	-
Coleoptera	5,59	9,95	6,22	11,47	3,10	3,71
Collembola	-	-	-	-	1,00	-
Diplopoda	3,40	3,37	1,00	,00	1,00	-
Diptera	82,86	80,66	83,33	101,27	81,85	71,07
Formicidae	4,94	5,63	9,64	14,06	4,05	4,63
Gastropoda	2,00	1,00	1,00	-	1,83	,98
Glaphyridae	30,00	39,60	17,63	27,11	1,67	1,15
Hemiptera	22,00	27,06	10,78	16,18	32,73	40,57
Hymenoptera	22,57	24,45	29,44	33,38	15,72	18,20
Malacostraca	-	-	1,00	-	1,00	-
Isoptera	1,50	1,00	13,00	17,09	1,50	,84
Lepidoptera	2,25	1,81	2,93	2,52	2,88	2,92
Neuroptera	1,00	,00	1,00	,00	1,00	,00
Opiliones	1,00	-	1,00	,00	1,00	,00
Opisthopora	-	-	2,00	-	-	-
Phasmatodea	-	-	-	-	1,00	-
Pseudoscorpiones	1,00	-	-	-	-	-
Psocoptera	17,96	13,85	11,62	15,09	7,13	4,39
Raphidioptera	-	-	2,00	1,41	1,00	-
Siphonaptera	1,00	-	-	-	-	-
Sterrnorrhyncha	2,80	2,49	1,00	-	4,00	-
Stylommatophora	4,00	-	-	-	-	-
Thysanoptera	6,46	4,70	3,43	3,03	8,46	7,66

*Statistically significant differences (p<,005) are marked with gray shade

are known to exhibit fluctuations, both seasonally and annually. Weather conditions, crop phenology, and pest outbreaks influence arthropod populations (Stavrianakis et al. 2024), and a more extended investigation would allow us to assess the consistency of our findings over time. Despite these limitations, our results contribute to the growing body of knowledge on arthropod communities in olive groves and provide a foundation for future research aimed at understanding the factors, such as farming practices, driving their diversity and abundance.

Following the results from this research, it seems that olive grove abandonment and understorey management through clearing have similarly positive effects on the diversity of plant species. However, it is likely that the highest taxonomic diversity being observed in the abandoned fields with undisturbed understorey is due to the different successional stages of abandonment observed in these sampling plots. In the early stages of land abandonment (< 20 years), plant diversity increases, with herbaceous plants and woody shrubs coexisting. However, these higher biodiversity levels tend to decrease as plant succession progresses (De Paz et al. 2022). The abandonment of traditional olive groves causes a gradual decrease in plant diversity mainly through a lower proportion of annual species and the prevention of the establishment and growth of shade-intolerant perennial herbs that are characteristic for traditional olive groves (Maccherini et al. 2013; Kakampoura & Panitsa 2022). The biodiversity impacts of abandonment are especially high because, unlike other more intensively managed agricultural systems, traditionally cultivated olive orchards support a high level of biodiversity (Loumou & Giourga 2003). Besides long-term negative biodiversity impacts, abandonment of olive groves also has a number of other negative environmental impacts, such as increased risk of fires and soil erosion (Jiménez et al. 2023).

TABLE 6. ANOVA for the linear regression model with arthropod abundance as the dependent variable and mean temperature and % of annual plant species as the predictors. And contribution of each variable to the model.

Model		Sum of Squares	df	Mean Square	F	Sig.	R^2 (adjusted)
1	Regression	505703,21	2	252851,61	5,394	,012*	25,3%
	Residual	1125039,31	24	46876,64			
	Total	1630742,52	26				

a. Dependent variable arthropod abundance
b. Predictors: (Constant): mean temperature, % annuals

Model	Understandarized coefficient (B)	Standarized coefficient (B)	95% Confidence Interval		Sig.	Collinearity	
			Lower Bound	Upper Bound		Tolerance	VIF
(Constant)	503,523		-7,41	1014,45	,053		
% annuals	7,726	,544	2,60	12,85	,005*	,939	1,065
Mean temperature	3,903	,045	-27,18	34,99	,789	,939	1,065

* p<0.05

The discrepancy between the number of species observed in the organically managed sampling plots versus the conventionally managed sampling plots treated with herbicides were not as pronounced as in prior studies (Solomou & Sfougaris 2011; 2013; 2021), likely due to the limited and sporadic application of herbicides in the sprayed sampling plots. Still, the organically managed sampling plots with understorey periodically cleared by sheep and mechanical hand mowers, displayed higher plant biomass and diversity. Interestingly, a higher abundance of annual plant species could be observed in the sampling plots with cleared understorey, which is confirmed by previous studies (Solomou & Sfougaris 2011; Kakampoura & Panitsa 2022; Stavrianakis et al. 2024). These annual species support important plant-insect interactions, which in turn provide pollination services to nearby agricultural areas (Kakampoura & Panitsa 2022).

In line with findings from previous research, the highest arthropod abundance was observed in the olive groves with cleared understorey (6564 specimens), while the lowest arthropod abundance was observed in the olive groves with undisturbed understorey (5586 specimens). The variation between arthropod abundance between olive groves with cleared and sprayed understorey is, however, minimal, with 6253 arthropod specimens collected in the sprayed sampling plots. The similar values of abundance observed in cleared and sprayed sampling plots might be explained by the high landscape complexity of the study area, with a complex mosaic of olive orchards typically under traditional, low-intensive management with limited input of synthetic agrochemicals. This is in line with the intermediate landscape complexity hypothesis, which states that in both simple landscapes with <1% of non-crop habitat and complex landscapes with >20% of non-crop habitat, only minimal positive effects of local management practices aimed at conserving biodiversity (such as organic farming) can be expected because of poor species pools in cleared landscapes and high immigration from semi-natural habitats in complex landscapes (Tscharntke et al. 2005; 2012). Instead, such local management practices are more effective in simple landscapes with a high proportion of non-crop habitat (>20%) (Tscharntke et al. 2005; 2012).

TABLE 7. ANOVA for the linear regression models with arthropod abundance as the dependent variable and mean temperature and % of annual species as the explanatory variables, separated by understorey management regime. And contribution of each explanatory variable to the model presented.

Model	F	df	Sig.	R^2 (adjusted)
CU	6,028	2	,037*	55,7%
SPR	2,337	2	,178	25%
UU	,283	2	,763	-21,8%

Model		Understandarized coefficient (B)	Standarized coefficient (B)	95% Confidence Interval		Sig.	Collinearity	
				Lower Bound	Upper Bound		Tolerance	VIF
CU	(Constant)	-251,137		-1318,232	815,958	,586		
	% annuals	10,411	,601	-,004	20,826	,05*	,919	1,089
	Mean temperature	47,000	,409	-22,119	116,120	,147	,919	1,089
SPR	(Constant)	1217,619		217,851	2217,386	,025*		
	% annuals	8,465	,731	-1,412	18,343	,081	,771	1,298
	Mean temperature	-36,965	-,511	-98,704	24,773	,193	,771	1,298
UU	(Constant)	494,155		-647,291	1635,601	,330		
	% annuals	4,083	,268	-10,804	18,971	,527	,951	1,051
	Mean temperature	5,039	,074	-61,639	71,718	,859	,951	1,051

* $p<0.05$

The lowest abundance of arthropods was observed in the abandoned olive groves with undisturbed understorey (5.586 specimens), which is likely due to the increased dominance of woody shrubs over the different successional stages of abandonment, thereby reducing the number of annual flowering species and perennial herbaceous species, eventually resulting in reduced arthropod diversity levels (De Paz et al. 2022). Since the floral diversity in the abandoned groves studied in this thesis is still relatively diverse due to the short time span since abandonment, it is expected that the arthropod diversity in these groves will only decrease. Some groups may, however, benefit from the structurally more complex vegetation of abandoned olive groves. A significantly higher abundance of leafhoppers (Hemiptera Cicadellidae) was observed in the sampling plots with undisturbed understorey. This might be due to the herbivorous nature of this group, benefiting from the abundance of well-developed shrubs in abandoned olive groves. However, leafhoppers can also act as pests due to the direct damage to leaves or vectors of diseases, having potential negative effects on ecosystem health (Carpio et al. 2020; Dalmaso et al. 2023). Mean temperature proved to not be a significant explanatory variable for arthropod abundance in this study (p = 0,789). This can likely be explained by the limited sampling period of this study, with measurements only being taken during the Spring season. While temperature (and relative humidity) have been proven to have strong effects on arthropods (Chown et al. 2011).

The observed reduction in plant species richness and coverage following herbicide application and understorey clearing was expected due to its direct impact on plant communities. However, the link between plant diversity and arthropod abundance is more complex (Stavrianakis et al. 2023). While a direct correlation might be anticipated, factors such as habitat structure, plant community composition, and the timing of interventions can influence arthropod diversity.

Olive groves with diverse understorey plant cover provide such a complex environment that supports a wider range of arthropod species. Herbicide application and understorey clearing can simplify this structure, limiting suitable habitats for arthropods. Additionally, certain plant species may provide specific resources or attract particular arthropod groups (Schaffers et al. 2008). The timing of these interventions can also influence the impact on arthropod populations. Understanding these ecological mechanisms is crucial for conserving arthropod biodiversity in olive grove systems.

As a result, the study proposes a new eco-scheme that supports farmers for maintaining understorey plant cover, with periodical clearing through ruminant grazing, to enhance plant and arthropod biodiversity in olive grove systems. This eco-scheme represents a novel approach to biodiversity management in olive groves. By incentivizing farmers to maintain understorey plant cover while implementing periodic clearing through ruminant grazing, the scheme aims to strike a balance between agricultural productivity and ecological conservation. This approach differs from traditional management practices that often involve intensive herbicide use or complete understorey removal, which can have detrimental effects on biodiversity.

The innovation of the eco-scheme lies in its recognition that a diverse understorey plant community plays a crucial role in supporting arthropod biodiversity. By promoting a mosaic of habitats through periodic clearing, the scheme helps to create a more structurally complex environment that can accommodate a wider range of species. Additionally, the use of ruminant grazing offers a sustainable and environmentally friendly method for managing understorey vegetation, reducing the need for chemical inputs.

In conclusion, our initial findings highlight the significant role that landscape structure plays in supporting biodiversity within olive grove ecosystems. These results suggest that even under low-intensity management practices, olive groves can maintain a healthy level of plant and arthropod diversity when situated within landscapes that promote ecological connectivity. Further research in this direction is important to refine Integrated Pest Management (IPM) protocols and inform policy decisions within the Common Agricultural Policy (CAP) to balance agricultural productivity with long-term biodiversity conservation within olive-growing regions.

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Ενδείξεις για την επίδραση των πρακτικών διαχείρισης του υποορόφου στην ποικιλότητα των αγγειακών φυτών και των αρθρόποδων σε ελαιώνες στη Λέσβο, Ελλάδα

N. SCHEEPENS[1], Γ. ΣΤΑΥΡΙΑΝΑΚΗΣ[1]*, R.T. KOMBA[1], Ε. ΣΕΝΤΑΣ[1] ΚΑΙ Θ. ΚΙΖΟΣ[1]

[1]Εργαστήριο Γεωγραφίας της Υπαίθρου και Συστημάτων Γεωργίας Ακριβείας, Τμήμα Γεωγραφίας, Πανεπιστήμιο Αιγαίου, Μυτιλήνη, Ελλάδα

ΠΕΡΙΛΗΨΗ

Η παρούσα μελέτη διερευνά την επίδραση τριών πρακτικών διαχείρισης του υποορόφου - εφαρμογή ζιζανιοκτόνων, εκκαθάριση του υποορόφου και αδιατάρακτος υποορόφος - στη βιοποικιλότητα των φυτών και των αρθρόποδων σε ελαιώνες στην περιοχή της Γέρας στη Λέσβο, Ελλάδα. Η μελέτη διαπίστωσε ότι η εφαρμογή ζιζανιοκτόνων είχε αρνητική επίδραση στην ποικιλότητα των φυτών, αλλά λιγότερο έντονες επιπτώσεις στα αρθρόποδα. Η ταχεία ανάκαμψη της βιοποικιλότητας των αρθρόποδων οφείλεται πιθανότατα στην υψηλή δομική πολυπλοκότητα στην περιοχή της Γέρας. Οι εγκαταλελειμμένοι ελαιώνες εμφάνισαν τη χαμηλότερη αφθονία αρθροπόδων και βλάστησης, οδηγώντας σε σταδιακή φτωχοποίηση της φυτικής βιοποικιλότητας και αρνητικές επιπτώσεις στην ποικιλότητα των αρθροπόδων. Το ποσοστό των ετήσιων ειδών στη φυτοκάλυψη συσχετίστηκε θετικά με την αφθονία αρθρόποδων. Η μελέτη προτείνει ένα νέο οικολογικό σχήμα που υποστηρίζει τους γεωργούς για τη διατήρηση της φυτοκάλυψης του υποορόφου, με περιοδικό καθαρισμό μέσω της βόσκησης μηρυκαστικών, για την ενίσχυση της βιοποικιλότητας των φυτών και των αρθρόποδων στους ελαιώνες.

