

# ENTOMOFAUNA ASSOCIATED WITH AGROFORESTRY SYSTEMS IMMERSSED IN THE MESOPHILIC MOUNTAIN FOREST OF ATZALAN, PUEBLA, MEXICO

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## ABSTRACT

Agroforestry systems contribute to entomological conservation by allowing the development of interactions that maintain the stability of the agroecosystem. To determine the diversity of insect families and their trophic guilds associated with agroforestry systems immersed in the mountain mesophyll forest, two agricultural systems (agroforestry and agrosilvopastoral) were studied in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico. Insect collections were conducted at 10 sites from November 2021 to October 2022. The frequencies of observation (FO), the richness estimator (Jackknife test), and the indices of relative abundance (RAI), diversity (Shannon-Wiener), equity, and evenness (Jaccard) were determined. Kruskal-Wallis,  $\chi^2$  and cluster analysis tests were performed. The frequencies of observation showed high values for the families Drosophilidae (34.15 %), Tachinidae (9.45 %), and Sciaridae (4.95 %). The RAI had higher values for Drosophilidae (0.68), Tachinidae (0.18), and Sciaridae (0.09). The Jackknife test presented average values of 70.1, 91.7, and 94.77. The Shannon-Wiener index exhibited an average diversity of  $H' = 3.78$ , 4.12, and 4.18. The Jaccard index displayed values of 45, 53, and 55 %, respectively. The Kruskal-Wallis test exhibited significant differences for abundance, richness, and diversity. The  $\chi^2$  test showed that abundance and richness for the agrosilvicultural condition were not as expected by the model. Cluster analysis showed the formation of two, two, and three clusters for abundance and five, two, and three, respectively, for richness. The evaluated systems harbor a high diversity of insect families that apply the differential use of the available trophic resource and allow the ecological balance of the analyzed agroecosystems. It is recommended to broaden the structural complexity of agroforestry systems in order to contribute to the conservation of entomological resources.

**Keywords:** agroforestry, agrosilvopastoral, bioindicators, biological control, diversity, monophagy.

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## INTRODUCTION

Globally, the entomological group represents a taxon of great evolutionary success, with wide diversity, abundance, and distribution. Despite not having a real figure, it is estimated that there are about 5.5 million species, pointing out that around 80 % of the total remain to be described (Stork, 2018). This diversity places these organisms as an omnipresent resource that makes use of the different ecological niches of the planet (Scudder, 2009). However, anthropogenic factors such as overexploitation of resources, expansion of the agricultural frontier, and irrational use of chemical inputs have brought different taxonomic groups to the brink of extinction, and many others have depleted their populations (Raven and Wagner, 2020). To date, it is estimated that between 250 000 and 500 000 insect species have become extinct, representing between 5 and 10 % of the entomological diversity of the planet (Cardoso *et al.*, 2020). In Mexico, knowledge about the current diversity and problems faced by the entomofauna in terms of conservation status is scarce (Castillo *et al.*, 2018). Almost 48 000 species of insects have been recorded, representing 66 % of the known fauna in the country (SEMARNAT, 2012). However, due to human factors such as increased agricultural production, mining companies, the exploitation of fossil fuels, and urbanization, an exponential reduction in biological diversity has been registered (Raven and Wagner, 2020).

Efforts to document the loss of diversity have focused on the study and conservation of vertebrates and large, charismatic organisms. Therefore, there is little work on smaller groups, such as insects (Falcón-Brindis *et al.*, 2021). As an example, Morón and Terrón (1984) evaluated the diversity of necrophilous insects in tropical and mesophilic forests with different degrees of disturbance, showing that, despite the impact on these ecosystems, high levels of diversity and abundance of this entomological group are still maintained.

Agroforestry systems represent a sustainable alternative that contributes to mitigating the problems caused by environmental deterioration and ensuring food security (Villanueva-López *et al.*, 2019). These systems are integrated through the deliberate combination of herbaceous, shrub, and tree species in association with animals, allowing for better productive development. At the same time, a broad vertical-horizontal multi-layer structure is generated, providing optimal light, temperature, and moisture conditions and creating differential habitats that allow the development of a wide variety of organisms (Bentrup *et al.*, 2019). The latter favors the connection between vegetation remnants (patches) and allows the movement and dispersal of flora and fauna (Villanueva-López *et al.*, 2019).

In this context, insects, being evolutionarily and ecologically versatile organisms, make differential use of a wide range of niches, contributing to the ecological stability of agroecosystems (Jankielsohn, 2018). Likewise, entomofauna (particularly the orders Hymenoptera and Hemiptera) play an important role in the pollination of productive components and provide a diverse range of products (honey, wax, royal jelly, propolis,

silk, and dyes, among others) that contribute to the productive economy and constitute an element of the cultural identity of the communities.

The presence of parasites and predators contributes to biological pest control and reduces the use of chemical products, while decomposers favor the recycling of nutrients and soil fertility. The presence of ants and beetles improves the quality and structure of the edaphic component, in addition to being a source of food for various taxonomic groups (particularly birds, bats, and different mammals) and bioindicators of the health and sustainability of agroforestry systems (Ramos *et al.*, 2020). However, despite the importance of these organisms, there are few studies that consider the diversity of entomofauna within these systems (Kaur *et al.*, 2023).

In the town of Atzalan, which belongs to the municipality of Xochiapulco in the northern highlands of Puebla, Mexico, agriculture is practiced for subsistence purposes and, in most cases, through a multi-layered arrangement with an agroforestry system structure, immersed in the mesophilic mountain forest. Given the structure and floristic composition of these agroecosystems, it is inferred that they could harbor a considerable number of insects that could play an important role in the maintenance and ecological stability of these bioproductive environments. Therefore, the objective of this study was to determine the diversity of insect families and their trophic guild within agroforestry systems in this region.

## MATERIALS AND METHODS

The study area is located at 19° 53' 49'' N and 97° 37' 17'' W, at an altitude of 1565 m. The natural vegetation is defined by mountain mesophilic forest (MMF), with pine-oak dominance, an average annual rainfall of 599 mm, average temperatures of 22 °C, and humidity of 62 %. The predominant soil types are Andosol and Luvisol. Ten study units (SU) were established, with each point representing a system (three agroforestry and seven agrosilvopastoral systems). These points were selected by systematic sampling, guaranteeing the independence of data to prevent insects recorded at a certain point from being recorded again in another SU (León-Burgos *et al.*, 2019).

The agricultural components that integrated the evaluated systems (agroforestry and agrosilvopastoral) were maize fields (maize, beans, squash, and chili), fruit trees (avocado, peach, plum, coffee, apple, orange, lemon, banana, guava, capulin, custard apple, tangerine, and pomegranate, among others), and medicinal, aromatic, ornamental, and forage plants (grasses and legumes). The livestock component of the agrosilvopastoral system (free grazing and stabled) consisted of poultry (chickens, turkeys, and ducks), rabbits, sheep, cattle, pigs, and horses. The variables were monitored on a monthly basis from November 2021 to October 2022.

The collection of insects was carried out with a beating net on herbaceous, shrubby, and arboreal plants. For the latter stratum, only insects recorded at a height of less than 2 m were considered; in parallel, insects were collected with Malaise traps (Sánchez-Flores *et al.*, 2019; Soca-Flores *et al.*, 2022). These methodologies were applied and conducted

in the first eight days of each month. Taxonomic keys were used to identify insects at the family level as proposed by Borror *et al.* (2005). The trophic guild of insects was determined through the insect guide proposed by Zumbado and Azofeifa (2018). The frequency of observation (FO), relative abundance (RAI), richness (Jackknife test), and diversity (Shannon-Wiener) were determined, and a dissimilarity analysis (Jaccard index) was performed. To detect possible differences in the abundance, richness, and diversity of the insect families in different samplings, Kruskal-Wallis analyses were used. To determine whether the proportion of individuals recorded was adequate, a  $\chi^2$  analysis was applied. Both analyses were performed in JMP IN v.8.0.2 (SAS Institute Inc., Cary, NC, USA). For all cases,  $\alpha = 0.05$  was used. Finally, to visualize the similarity between the abundance and richness of recorded families, cluster analyses were developed using the XLSTAT statistical software version 2018.7.5 (XLSTAT, 2018).

## RESULTS AND DISCUSSION

A total of 15 363 insects were collected, and 11 orders and 92 families were determined. Of the total number of specimens, 10 995 were linked to the agrosilvopastoral system and 4368 to the agroforestry system. Moreover, two families were found to be exclusive to the agroforestry system, and 24 to the agrosilvopastoral system (Table 1).

**Table 1.** Frequency of insects per family recorded and their taxonomic classification based on Borror *et al.* (2005), in the agricultural systems studied in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

Order	Suborder	Superfamily	Family	Recorded frequency		
				AS	ASP	General
Blattodea	NA	Blaberoidea	Ectobiidae	1	5	6
Coleoptera	Polyphaga	Curculionoidea	Attelabidae	2	3	5
			Curculionidae	13	83	96
		Buprestoidea	Buprestidae**	0	1	1
		Elateroidea	Cantharidae**	0	4	4
		Caraboidea	Carabidae	10	12	22
		Cerambycoidea	Cerambycidae**	0	21	21
		Chrysomeloidea	Chrysomelidae	80	243	323
		Coccinelloidea	Coccinellidae	10	19	29
		Elateroidea	Elateridae	17	109	126
		Tenebrionoidea	Oedemeridae	2	2	4
			Meloidae	1	17	18
			Tenebrionidae	10	20	30
		Cucujoidea	Erotylidae	1	10	11
		Elateroidea	Lampyridae	7	14	21
		Elateroidea	Lycidae	6	11	17
		Cucujoidea	Nitidulidae**	0	2	2
		Scarabaeoidea	Scarabaeidae	7	83	90

**Table 1.** Continue.

Order	Suborder	Superfamily	Family	Recorded frequency		
				AS	ASP	General
		Cucujoidea	Silvanidae**	0	2	2
		Staphylinoidea	Staphylinidae	4	27	31
		Forficuloidea	Forficulidae	25	26	51
Dermaptera	Neodermaptera					
Diptera	Nematocera	Tipuloidea	Tipulidae	112	129	241
		Sciaroidea	Sciaridae	260	502	762
	Brachycera		Bibionidae**	0	1	1
		Muscoidea	Anthomyiidae**	0	1	1
		Asiloidea	Asilidae**	0	4	4
		Oestroidea	Calliphoridae	110	326	436
		Empidoidea	Dolichopodidae	3	4	7
		Ephydroidea	Drosophilidae	1616	3631	5247
		Empidoidea	Empididae**	0	3	3
		Lauxanioidea	Lauxaniidae*	1	0	1
		Muscoidea	Muscidae	1	18	19
		Tephritoidea	Ulidiidae	4	2	6
		Oestroidea	Sarcophagidae	8	1	9
			Stratiomyidae	1	1	2
			Syrphidae	33	76	109
		Oestroidea	Tachinidae	615	837	1452
		Tephritoidea	Tephritidae	3	6	9
		Asiloidea	Therevidae	1	7	8
Hemiptera	Auchenorrhyncha	Membracoidea	Membracidae	9	267	276
			Cicadellidae	40	119	159
	Heteroptera	Cercopoidea	Cercopidae**	0	2	2
		Coreoidea	Coreidae	47	418	465
		Pentatomoidea	Cydnidae	1	1	2
			Scutelleridae	4	2	6
		Lygaeoidea	Lygaeidae	3	10	13
		Coreoidea	Alydidae**	0	5	5
		Miroidea	Miridae	81	290	371
		Notonectoidea	Notonectidae**	0	1	1
		Pentatomoidea	Pentatomidae	17	84	101
		Pyrrhocoroidea	Pyrrhocoridae**	0	3	3
		Reduvioidea	Reduviidae	2	17	19
Hymenoptera	Apocrita	Apoidea	Andrenidae**	0	2	2
			Apidae	172	537	709
			Sphecidae	9	32	41
		Ichneumonoidea	Braconidae**	0	1	1
		Formicoidea	Formicidae	3	2	5
		Ichneumonoidea	Ichneumonidae	127	349	476
		Proctotrupoidea	Pelecinidae**	0	4	4
		Pompiloidea	Pompilidae	1	1	2
		Scolioidea	Scoliidae	12	51	63
		Vespoidea	Vespidae	160	421	581
Lepidoptera	NA	Bombycoidea	Apatelodidae**	0	1	1
		Pyraloidea	Crambidae**	0	7	7



**Table 1.** Continue.

Order	Suborder	Superfamily	Family	Recorded frequency		
				AS	ASP	General
		Noctuoidea	Erebidae	1	15	16
		Gelechioidea	Gelechiidae	1	8	9
		Geometroidea	Geometridae	46	154	200
		Papilionoidea	Hesperiidae	12	37	49
		Lasiocampoidea	Lasiocampidae	2	2	4
		Zygaenoidea	Limacodidae*	2	0	2
		Papilionoidea	Lycaenidae	7	6	13
		Noctuoidea	Noctuidae	68	139	207
		Noctuoidea	Nolidae	1	3	4
		Noctuoidea	Notodontidae**	0	5	5
		Papilionoidea	Nymphalidae	17	53	70
		Papilionoidea	Papilionidae**	0	5	5
		Papilionoidea	Pieridae	56	187	243
		Pyraloidea	Pyralidae	142	411	553
		Bombycoidea	Saturniidae	2	6	8
			Sphingidae	1	2	3
		Tortricoidea	Tortricidae	56	325	381
Neuroptera	Hemerobiiformia		Chrysopidae**	0	1	1
			Hemerobiidae	10	48	58
Odonata	Anisoptera	Aeshnoidea	Aeshnidae	3	4	7
		Libelluloidea	Macromiidae**	0	6	6
Orthoptera	Caelifera	Acridoidea	Acrididae	196	556	752
		Pyrgomorphoidea	Pyrgomorphidae	59	72	131
		Acridoidea	Romaleidae	12	27	39
	Ensifera	Grylloidea	Gryllidae	20	23	43
		Stenopelmatoidea	Stenopelmatidae**	0	1	1
		Tettigonioidae	Tettigoniidae	2	8	10
Thysanoptera	Terebrantia		Adihetero- thripidae**	0	1	1

AS: agroforestry system; ASP: agrosilvopastoral system; NA: not available. \*Families exclusive to the agroforestry system; \*\*families exclusive to the agrosilvopastoral system.

The frequency of observation (FO) analysis presented high values for the families Drosophilidae (34.15 %), Tachinidae (9.45 %), Sciaridae (4.95 %), Acrididae (4.89 %), and Apidae (4.61 %) (Table 2). The relative abundance index (RAI) showed higher values for the families Drosophilidae (0.68), Tachinidae (0.18), and Sciaridae (0.09) (Table 2). Therefore, it is evident that these insect families represent the greatest dominance of individuals present in the systems analyzed. The population structure of the family Drosophilidae, a major pest commonly known as fruit flies, is determined by biological control through parasitism by insects of the family Tachinidae (an

**Table 2.** Observation frequency index and relative abundance of the insect families registered in the agricultural systems evaluated in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

Families	Frequency of observation (FO) index			Relative abundance index (RAI)		
	Agroforestry	Agrosilvopastoral	General	Agroforestry	Agrosilvopastoral	General
Acrididae	4.487	<b>5.057</b>	4.895	0.090	<b>0.101</b>	0.098
Adiheterothripidae	0.000	0.009	0.007	0.000	0.000	0.000
Aeshnidae	0.069	0.036	0.046	0.001	0.001	0.001
Alydidae	0.000	0.045	0.033	0.000	0.001	0.001
Andrenidae	0.000	0.018	0.013	0.000	0.000	0.000
Anthomyiidae	0.000	0.009	0.007	0.000	0.000	0.000
Apatelodidae	0.000	0.009	0.007	0.000	0.000	0.000
Apidae	3.938	4.884	4.615	0.079	0.098	0.092
Asilidae	0.000	0.036	0.026	0.000	0.001	0.001
Attelabidae	0.046	0.027	0.033	0.001	0.001	0.001
Bibionidae	0.000	0.009	0.007	0.000	0.000	0.000
Braconidae	0.000	0.009	0.007	0.000	0.000	0.000
Buprestidae	0.000	0.009	0.007	0.000	0.000	0.000
Calliphoridae	2.518	2.965	2.838	0.050	0.059	0.057
Cantharidae	0.000	0.036	0.026	0.000	0.001	0.001
Carabidae	0.229	0.109	0.143	0.005	0.002	0.003
Cerambycidae	0.000	0.191	0.137	0.000	0.004	0.003
Cercopidae	0.000	0.018	0.013	0.000	0.000	0.000
Chrysomelidae	1.832	2.210	2.102	0.037	0.044	0.042
Chrysopidae	0.000	0.009	0.007	0.000	0.000	0.000
Cicadellidae	0.916	1.082	1.035	0.018	0.022	0.021
Coccinellidae	0.229	0.173	0.189	0.005	0.003	0.004
Coreidae	1.076	3.802	3.027	0.022	0.076	0.061
Crambidae	0.000	0.064	0.046	0.000	0.001	0.001
Curculionidae	0.298	0.755	0.625	0.006	0.015	0.012
Cydnidae	0.023	0.009	0.013	0.000	0.000	0.000
Dolichopodidae	0.069	0.036	0.046	0.001	0.001	0.001
Drosophilidae	<b>36.996</b>	<b>33.024</b>	<b>34.153</b>	<b>0.740</b>	<b>0.660</b>	<b>0.683</b>
Ectobiidae	0.023	0.045	0.039	0.000	0.001	0.001
Elateridae	0.389	0.991	0.820	0.008	0.020	0.016
Empididae	0.000	0.027	0.020	0.000	0.001	0.000
Erebidae	0.023	0.136	0.104	0.000	0.003	0.002
Erotylidae	0.023	0.091	0.072	0.000	0.002	0.001
Forficulidae	0.572	0.236	0.332	0.011	0.005	0.007
Formicidae	0.069	0.018	0.033	0.001	0.000	0.001
Gelechiidae	0.023	0.073	0.059	0.000	0.001	0.001
Geometridae	1.053	1.401	1.302	0.021	0.028	0.026
Gryllidae	0.458	0.209	0.280	0.009	0.004	0.006
Hemerobiidae	0.229	0.437	0.378	0.005	0.009	0.008
Hesperiidae	0.275	0.337	0.319	0.005	0.007	0.006
Ichneumonidae	2.908	3.174	3.098	0.058	0.063	0.062
Lampyridae	0.160	0.127	0.137	0.003	0.003	0.003
Lasiocampidae	0.046	0.018	0.026	0.001	0.000	0.001
Lauxaniidae	0.023	0.000	0.007	0.000	0.000	0.000
Limacodidae	0.046	0.000	0.013	0.001	0.000	0.000

**Table 2.** Continue.

Families	Frequency of observation (FO) index			Relative abundance index (RAI)		
	Agroforestry	Agrosilvopastoral	General	Agroforestry	Agrosilvopastoral	General
Lycaenidae	0.160	0.055	0.085	0.003	0.001	0.002
Lycidae	0.137	0.100	0.111	0.003	0.002	0.002
Lygaeidae	0.069	0.091	0.085	0.001	0.002	0.002
Macromiidae	0.000	0.055	0.039	0.000	0.001	0.001
Meloidae	0.023	0.155	0.117	0.000	0.003	0.002
Membracidae	0.206	2.428	1.797	0.004	0.049	0.036
Miridae	1.854	2.638	2.415	0.037	0.053	0.048
Muscidae	0.023	0.164	0.124	0.000	0.003	0.002
Nitidulidae	0.000	0.018	0.013	0.000	0.000	0.000
Noctuidae	1.557	1.264	1.347	0.031	0.025	0.027
Nolidae	0.023	0.027	0.026	0.000	0.001	0.001
Notodontidae	0.000	0.045	0.033	0.000	0.001	0.001
Notonectidae	0.000	0.009	0.007	0.000	0.000	0.000
Nymphalidae	0.389	0.482	0.456	0.008	0.010	0.009
Oedemeridae	0.046	0.018	0.026	0.001	0.000	0.001
Papilionidae	0.000	0.045	0.033	0.000	0.001	0.001
Pelecniidae	0.000	0.036	0.026	0.000	0.001	0.001
Pentatomidae	0.389	0.764	0.657	0.008	0.015	0.013
Pieridae	1.282	1.701	1.582	0.026	0.034	0.032
Pompilidae	0.023	0.009	0.013	0.000	0.000	0.000
Pyrilidae	3.251	3.738	3.600	0.065	0.075	0.072
Pyrgomorphidae	1.351	0.655	0.853	0.027	0.013	0.017
Pyrhocoridae	0.000	0.027	0.020	0.000	0.001	0.000
Reduviidae	0.046	0.155	0.124	0.001	0.003	0.002
Romaleidae	0.275	0.246	0.254	0.005	0.005	0.005
Sarcophagidae	0.183	0.009	0.059	0.004	0.000	0.001
Saturniidae	0.046	0.055	0.052	0.001	0.001	0.001
Scarabaeidae	0.160	0.755	0.586	0.003	0.015	0.012
Sciaridae	<b>5.952</b>	4.566	<b>4.960</b>	<b>0.119</b>	0.091	<b>0.099</b>
Scoliidae	0.275	0.464	0.410	0.005	0.009	0.008
Scutelleridae	0.092	0.018	0.039	0.002	0.000	0.001
Silvanidae	0.000	0.018	0.013	0.000	0.000	0.000
Sphecidae	0.206	0.291	0.267	0.004	0.006	0.005
Sphingidae	0.023	0.018	0.020	0.000	0.000	0.000
Staphylinidae	0.092	0.246	0.202	0.002	0.005	0.004
Stenopelmatidae	0.000	0.009	0.007	0.000	0.000	0.000
Stratiomyidae	0.023	0.009	0.013	0.000	0.000	0.000
Syrphidae	0.755	0.691	0.709	0.015	0.014	0.014
Tachinidae	<b>14.080</b>	<b>7.613</b>	<b>9.451</b>	<b>0.282</b>	<b>0.152</b>	<b>0.189</b>
Tenebrionidae	0.229	0.182	0.195	0.005	0.004	0.004
Tephritidae	0.069	0.055	0.059	0.001	0.001	0.001
Tettigoniidae	0.046	0.073	0.065	0.001	0.001	0.001
Therevidae	0.023	0.064	0.052	0.000	0.001	0.001
Tipulidae	2.564	1.173	1.569	0.051	0.023	0.031
Tortricidae	1.282	2.956	2.480	0.026	0.059	0.050
Uliidae	0.092	0.018	0.039	0.002	0.000	0.001
Vespidae	3.663	3.829	3.782	0.073	0.077	0.076



entomological group mostly recognized for its role in biological pest control), which also controls the population dynamics of the family Sciaridae, which contains species that could become pests in agricultural systems. On the other hand, individuals of the Sciaridae family play an important role in nutrient recycling, since, being saprophytic organisms, they contribute to degrade and reincorporate nutrients into the soil. This trend is in agreement with Armijos-Vásquez *et al.* (2020) and Huang *et al.* (2021), who indicate that the most representative specimens in agroforestry systems and natural forest areas correspond to the orders Diptera and Hymenoptera. These authors attribute these values to factors such as the physiological age of the vegetation, the productive stage of the crop, and the agro-climatic characteristics, which define the food availability and the reproductive period of the insects. In this context, Ambele *et al.* (2023) describe how agroforestry plantations present a mixed tree cover structure, similar to natural forests, providing differential habitats for a wide diversity of insects that contribute to key agroecosystem processes (pollination, soil fertilization, organic matter decomposition, nutrient cycling, and biological pest control). Of the 92 families described, 17 trophic guilds were recorded, divided into monophagous, oligophagous, and polyphagous groups. Additionally, members of certain families include pest insects and biological controllers, seed dispersers, organic matter decomposers, and indicators of environmental quality (Table 3). The differential use of the trophic resource shows sympatric coexistence and highlights the ecological importance of insect diversity (at the family level) contributing to the ecological stability of the agroforestry systems analyzed.

**Table 3.** Feeding habits of insect families recorded in the agricultural systems evaluated in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

Families	Phy	Sap	Par	Fun	Pre	Pol	Nec	Gran	Rhz	Xyl	Frug	Car	Hem	Necro	Suck	Omn	Myce	Mono	Oli	Poly	BiolC	Pest	EnQI	OorMD	Seedis
Acrididae	x															x			x	x					
Adiheterothripidae	x				x															x					
Aeshnidae					x															x					
Alydidae	x				x											x				x					
Andrenidae						x	x												x	x					
Anthomyiidae	x	x	x													x				x					
Apatelodidae	x																			x					
Apidae						x	x											x	x						
Asilidae			x		x															x					
Attelabidae	x	x													x	x		x							
Bibionidae	x	x				x	x									x									
Braconidae			x			x	x											x			x				
Buprestidae						x	x			x								x	x						
Calliphoridae		x	x		x		x			x				x		x				x					
Cantharidae	x				x	x	x									x				x	x				

**Table 3.** Continue.

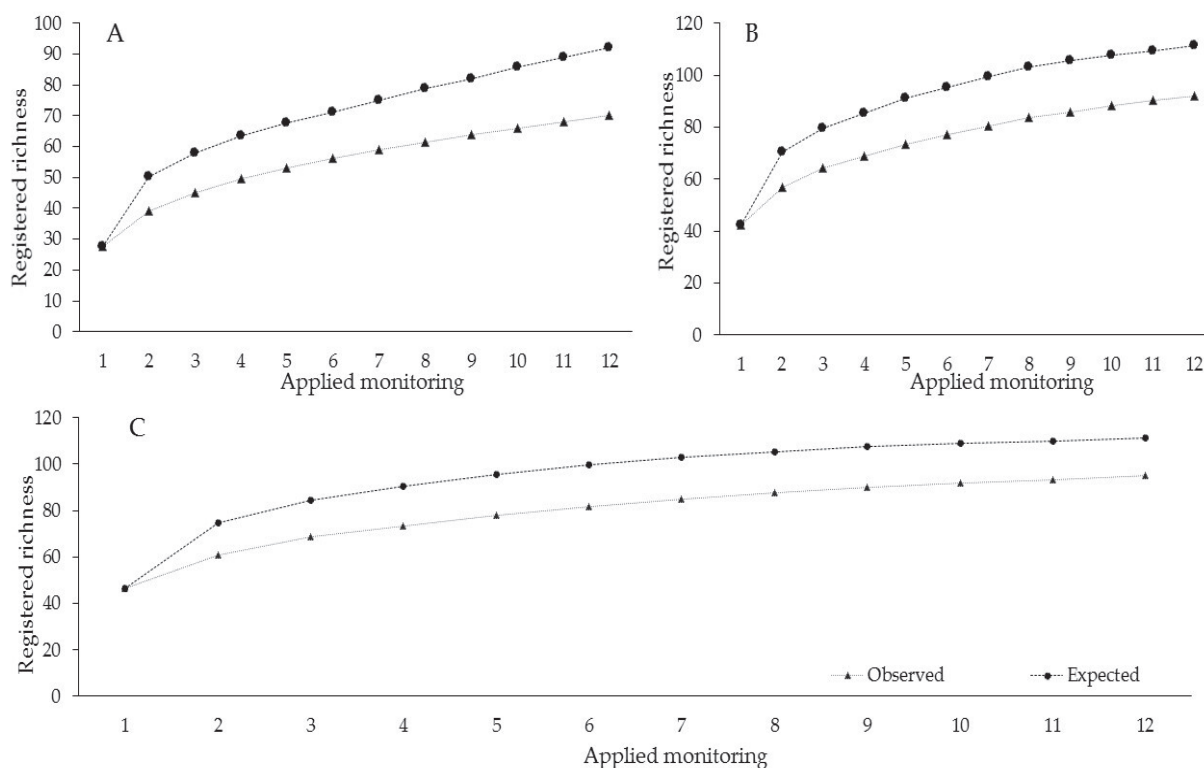
Families	Phy	Sap	Par	Fun	Pre	Pol	Nec	Gran	Rhz	Xyl	Frug	Car	Hem	Necro	Suck	Omn	Myce	Mono	Oli	Poly	BiolC	Pest	EnQI	OorMD	Seedis
Carabidae		x			x									x					x						
Cerambycidae	x									x					x					x		x			
Cercopidae	x									x					x					x		x			
Chrysomelidae	x					x			x	x						x				x					
Chrysopidae					x	x	x									x			x		x				
Cicadellidae	x								x						x					x		x			
Coccinellidae				x	x	x	x												x		x				
Coreidae	x														x					x		x			
Crambidae	x					x	x	x	x											x		x			
Curculionidae	x			x				x	x	x						x		x		x		x			
Cydnidae	x								x						x					x		x			
Dolichopodidae					x											x				x	x				
Drosophilidae		x		x			x				x				x					x		x			
Ectobiidae		x												x		x				x		x			
Elateridae		x			x			x	x		x					x				x		x			
Empididae					x		x												x		x				
Erebidae	X																			X					
Erotylidae																									
Forficulidae		x			X									x	X				X	x					
Formicidae	x				x									x	x				x						x
Gelechiidae	x																		x			x			
Geometridae	x						x													x		x			
Gryllidae	x								x							x				x		x			
Hemerobiidae					x	x	x									x			x		x				
Hesperiidae	x						x											x	x				x		
Ichneumonidae			x				x												x		x		x		
Lampyridae					x	x	x												x		x		x		
Lasiocampidae	x																		x						
Lauxaniidae	x	x					x									x				x					
Limacodidae	x						x													x					
Lycaenidae	x						x											x							
Lycidae					x							x						x			x				
Lygaeidae	x				x										x					x		x			
Macromiidae					x						x								x				x		
Meloidae	x		x		x	x	x												x						
Membracidae	x																			x		x			
Miridae	x				x										x					x		x			
Muscidae		x			x								x							x	x				
Nitidulidae		x		x				x		x	x			x			x			x					
Noctuidae	x						x				x								x			x			
Nolidae	x																	x		x					
Notodontidae	x																			x					
Notonectidae					x														x		x				
Nymphalidae	x							x											x		x				
Oedemeridae	x	x				x	x													x					
Papilionidae	x						x											x	x				x		
Pelecinidae	x		x																x		x				

**Table 3.** Continue.

Families	Phy	Sap	Par	Fun	Pre	Pol	Nec	Gran	Rhz	Xyl	Frug	Car	Hem	Necro	Suck	Omn	Myce	Mono	Oli	Poly	BiolC	Pest	EnQI	OorMD	Seedis
Pentatomidae	x				x						x									x	x	x			
Pieridae	x						x												x			x			
Pompilidae			x		x		x											x			x				
Pyralidae	x								x		x								x			x			
Pyrgomorphidae	x																		x						
Pyrrhocoridae	x				x			x								x				x					
Reduviidae					x								x						x	x					
Romaleidae	x																	x		x					
Sarcophagidae		x	x		x		x					x		x										x	
Saturniidae	x																	x		x					
Scarabaeidae	x	x							x										x			x			
Sciaridae		x		x					x													x			
Scoliidae			x				x				x								x		x				
Scutelleridae	x														x			x		x					
Silvanidae								x											x			x		x	
Sphecidae					x		x											x		x	x				
Sphingidae	x						x				x								x						
Staphylinidae			x		x											x				x	x				
Stenopelmatidae		x												x					x					x	
Stratiomyidae		x																x						x	
Syrphidae					x															x	x				
Tachinidae			x																	x	x				
Tenebrionidae	x	x							x	x				x		x				x				x	
Tephritidae	x						x	x			x									x		x			
Tettigoniidae	x				x															x					
Therevidae					x														x		x				
Tipulidae		x			x								x						x					x	
Tortricidae	x						x	x			x									x					
Ulidiidae	x	x									x			x						x		x		x	
Vespidae					x		x												x		x				

Phy: phytophage; Sap: saprophage; Par: parasitoid; Fun: fungivore; Pre: predator; Pol: pollenophage; Nec: nectarivore; Gran: granivore; Rhz: rhizophage; Xyl: xylophage; Frug: frugivore; Car: carnivore; Hem: hematophage; Necro: necrophage; Suck: sucker; Omn: omnivore; Myce: mycetophagous; Mono: monophagous; Oli: oligophagous; Poly: polyphagous; BiolC: biological control; EnQI: environmental quality indicator; OorMD: organic matter decomposer; Seedis: seed disperser.

The mean observed values of insect family richness were 54.97, 75.3, and 79.33 for agroforestry, agroforestry, and general, respectively. The expected results using the Jackknife estimator showed means of 70.1, 91.7, and 94.77 families, respectively. Therefore, so far, with the sampling effort applied, 76, 82.69, and 85.2 % of the insect families theoretically present in the evaluated agroecosystems are known (Figure 1).



**Figure 1.** Richness (Jackknife test) of families recorded in the agroforestry systems analyzed. A: agroforestry; B: agrosilvopastoral; C: general overview of systems.

The Shannon-Wiener index obtained minimum, maximum, and average values of  $H' = 3.3$ ,  $3.92$ , and  $3.78$  for the agroforestry system;  $H' = 3.74$ ,  $4.23$ , and  $4.12$  for the agrosilvopastoral system; and  $H' = 3.83$ ,  $4.29$ , and  $4.18$  for the general condition. These results represent the high value of the entomological diversity recorded, showing how the systems evaluated allow a differential use of the available trophic resources that allow ecological stability to be maintained. Furthermore, the systems analyzed represent a sustainable production alternative that contributes to the conservation of entomofauna at the local level.

The richness and diversity of insects with higher value in the agrosilvopastoral system was the result of a higher sampling effort (seven sites). It was also associated with ecological habitat variables that are influenced by the presence of the animal component. In these systems, the impact of livestock modifies the structural arrangement of the agricultural, forestry, and herbaceous resources (particularly in pastures). This creates greater habitat heterogeneity and offers a wide range of niches and microhabitats that can be occupied by diverse insect families. The structural diversity of physical-ecological and vegetation resources favors a greater complexity of ecological interactions, such as pollination, biological pest control, competition, and

predator-prey relationships. The presence of livestock introduces new elements to the ecosystem, such as dung, which attracts decomposing insects such as dung beetles (Rigueiro-Rodríguez *et al.*, 2010).

Martins *et al.* (2020) point out that agroforestry systems, unlike conventional crops, conserve a greater diversity of insects that maintain ecological stability in equilibrium. These authors point out that the heterogeneous structure of agroforestry systems determines the movement of insect herbivores, their duration, abundance, and mortality. This promotes ecological interactions (host-parasitoid-plant) that provide niches for feeding, nesting, shelter, and development, which have a wide range of microclimatic spaces that buffer against extremes of temperature and rainfall (Centeno-Alvarado *et al.*, 2023).

Agroforestry systems are represented by the incorporation of woody plants (trees and shrubs) associated with crops and/or livestock in the same space, which promotes the conservation of different taxonomic groups, particularly insects (Bentrup *et al.*, 2019). Silva-Neto *et al.* (2023) point out that the woody plant component provides alternative food for insects, particularly pollinators, by offering a food resource when agricultural crops are not flowering. Kingazi *et al.* (2024) also showed that agroforestry systems encourage pollinator presence because the woody plant cover increases floral availability, which draws a wider variety of insects.

Centeno-Alvarado *et al.* (2023) indicate that the vegetation structure of agroforestry systems, particularly trees, provides protection against extreme environmental events and agricultural disturbances. Under the same approach, Varah *et al.* (2020) have shown that floristic diversity in agroforestry systems increases insect diversity, particularly pollinators, which is attributed to the asynchronous floral diversity found in these agroecosystems. This phenomenon was corroborated in the present study. Throughout the monitoring period, pollinating insects (particularly of the order Hymenoptera) were recorded, which developed a differentiated spatio-temporal use of the available resource throughout the annual cycle.

According to the percentage values derived from Jaccard's index for the similarity of richness documented during the various surveys, agroforestry accounted for 45 %, agroforestry for 53 %, and general for 55 %. There is a great dissimilarity in the wealth of families recorded. This difference was determined by the annual seasonality of the plant component and the landscape-level structure that defines the physical-ecological ensemble of utilization niches, which determines the population dynamics of insects and other taxonomic groups over time. In contrast, Galbraith *et al.* (2020) and Tarigan *et al.* (2021) showed no significant differences in the richness and abundance of insects present in agroforestry systems. However, in some cases, insect abundance was lower compared to conventional production systems. However, Kingazi *et al.* (2024) mention that there is still very little work on this subject, so there is still a lack of research to generate conclusions and propose strategies for improvement at the agroforestry level. The Kruskal-Wallis analysis showed statistically significant differences between the



medians analyzed for abundance, richness, and diversity recorded in each of the agroforestry systems evaluated (Table 4).

**Table 4.** Kruskal-Wallis results for abundance, richness, and diversity recorded in the agroforestry systems evaluated in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

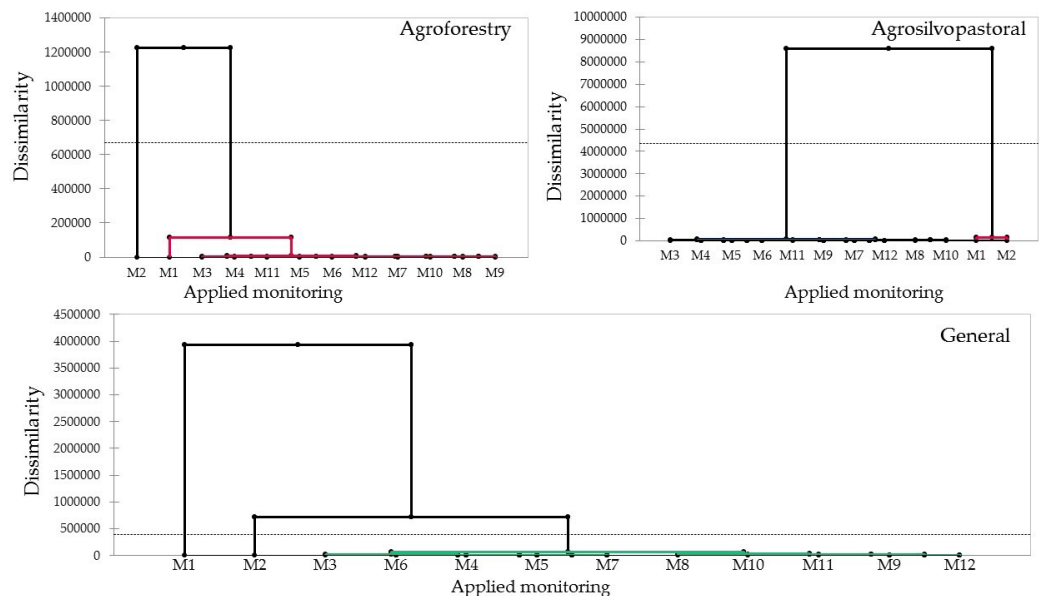
Chi-square	Degrees of freedom	Prob > Chi-square
21.64	Agroforestry (abundance) 11	0.02
22.44	Agrosilvopastoral (abundance) 11	0.02
20.65	General (abundance) 11	0.03
22.17	Agroforestry (richness) 11	0.02
21.07	Agrosilvopastoral (richness) 11	0.03
20.06	General (richness) 11	0.04
20.07	Diversity 2	<0.00

The  $\chi^2$  results demonstrate that the abundance in the agroforestry system and the recorded richness of families do not reflect what may exist in the agroecosystems evaluated. In turn, the diversity recorded corresponds to what could occur under the conditions studied (Table 5).

**Table 5.**  $\chi^2$  results for abundance, richness and diversity recorded in the agroforestry systems evaluated in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

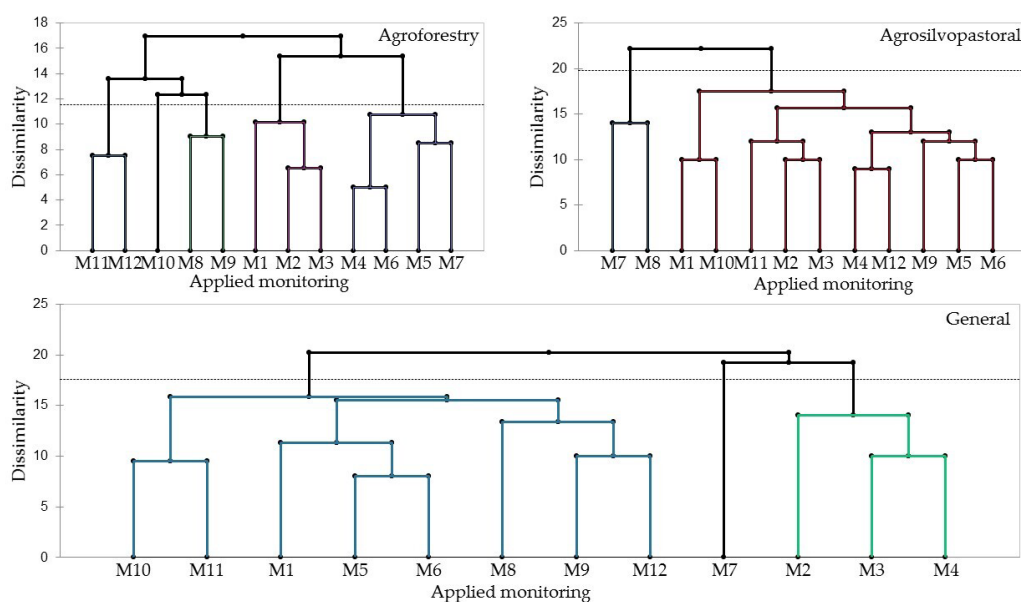
N	Degrees of freedom	Chi-square	Prob > Chi-square
840	Agroforestry (abundance) 484	543.86	0.03
1104	Agrosilvopastoral (abundance) 836	822.79	0.62
1140	General (abundance) 1056	1053.36	0.51
840	Agroforestry (richness) 11	22.19	0.02
1104	Agrosilvopastoral (richness) 11	21.09	0.03
1140	General (richness) 11	20.08	0.04
36	Diversity 58	63.00	0.30

The cluster analysis revealed the formation of several clusters. For abundance, agroforestry = two, agrosilvopastoral = two, and general = three (Figure 2), and for richness: agroforestry = five, agrosilvopastoral = two, and general = three (Figure 3). Different percentages of dissimilarity were shown in the record of both parameters (abundance and richness) (Table 6). These results are attributed to the annual seasonality that defines the food availability and reproductive cycle of the insects, associated with the plant structure and physical-ecological factors present throughout the year.



**Figure 2.** Cluster analysis of the abundance of insect families recorded in the different surveys. The horizontal line (Euclidean line) defines the number of clusters for each condition assessed; the colors show the grouping of the monitoring sites with the highest similarity in the abundance of families recorded.

This study provides information on agroforestry systems immersed in mesophilic mountain forests in relation to the incidence of insects; however, in order to propose better agroforestry management practices, it is necessary to develop specific works that allow visualizing the effect of these systems and the structure of the habitat on the population parameters of insects as a resource indicator of environmental quality, contributing to the conservation of insects and various taxonomic groups for this region in particular.



**Figure 3.** Cluster analysis of the richness of insect families recorded in each monitoring. The horizontal line (Euclidean line) defines the number of clusters for each condition assessed; the colors show the grouping of the monitoring sites with the highest similarity in richness of families recorded.

**Table 6.** Absolute and percentage value results of the clusters formed with the abundance and richness estimators of insect families recorded in the agroforestry systems evaluated in Atzalan, in the municipality of Xochiapulco, Puebla, Mexico.

Variance decomposition		Absolute	Percentage
Agroforestry abundance			
Intraclass		136 098.72	10.01 %
Intraclass		1 223 275.27	89.99 %
Agrosilvopastoral abundance			
Intraclass		372956.30	4.16 %
Intraclass		8581899.45	95.84 %
General abundance			
Intraclass		181 959.00	3.77 %
Intraclass		4 645 049.58	96.23 %
Agroforestry richness			
Intraclass		57.41	49.64 %
Intraclass		58.25	50.36 %
Agrosilvopastoral richness			
Intraclass		123.10	84.75 %
Intraclass		22.15	15.25 %
General richness			
Intraclass		107.50	73.17 %
Intraclass		39.41	26.83 %

## CONCLUSIONS

The heterogeneous structure of agroforestry systems contributes to insect incidence by providing differential trophic niches. It is recommended to increase the diversity of plant strata (vertical-horizontal) to conserve insect provision and maintain balanced population dynamics and positive ecological interactions as indicators of environmental quality. The evaluated agroforestry systems conserve a wide diversity of insect families and represent an option in the face of current scenarios of habitat loss, degradation, and fragmentation in natural forests.

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