

## Agroforestry systems and diurnal butterflies in areas with border effect in mesophilic forest

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### Abstract

In 2016, day butterflies were tracked in order to establish the importance of 33 edge effects (EB) formed by three traditional agroforestry systems immersed in a fragmented mesophilic forest in Xochitla, Mixtla de Altamirano, Veracruz. Systematic sampling and counting in points with fixed radius were established: the analyzes were carried out by: A) families with EB; B) subfamilies with EB; C) families by sampling; and D) subfamilies by sampling. Wealth and relative abundance were assessed using Jackknife1 and diversity with Shannon-Wiener. Kruskal-Wallis for the three indices, Cluster (wealth and abundance) and  $\chi^2$  (abundance). Jackknife1 showed values: 70.62; 47.6; 96.66, 80.27%. There were no differences:  $F=16$ ,  $p=0.724$ ;  $F=9$ ,  $p=0.4966$ ;  $F=20$ ,  $p=0.6584$ ;  $F=13$ ,  $p=0.5953$ . Cluster exhibited 3 to 5 amalgamations. The IAR presented minimum-maximum average values of 0.001-0.578. There were no discrepancies:  $F=26$ ,  $p=0.5642$ ;  $F=20$ ,  $p=0.6271$ ;  $F=22$ ,  $p=0.8414$ ;  $F=30$ ,  $p=0.4376$ . Cluster exhibited 3 to 5 amalgamations.  $\chi^2$  manifested values:  $p=0.5396$ ,  $p=0.7577$ ,  $p=0.5708$ ,  $p=0.3083$ . Shannon-Wiener exhibited values:  $H'=1.38$ ;  $H'=2.4$ ;  $H'=1.49$ ;  $H'=2.49$ . In two cases there were no differences:  $F=3$ ,  $p=0.2839$ ;  $F=3$ ,  $p=0.2839$ ; however, in two if:  $F=5$ ,  $p<0.0003$ ;  $F=11$ ,  $p<0.0015$ . Results are shown, respectively for A, B, C, D. The parameters under study of diurnal butterflies in the considered EB were estimated-compared, this lays the basis for future research of Lepidoptero fauna in agroforestry and EB systems for this particular region from Veracruz.

**Keywords:** biodiversity, lepidoptera, monitoring, Zongolica mountain range.

Reception date: January 2020

Acceptance date: March 2020

## Introduction

Among the most threatened ecosystems in Mexico is the mountain mesophilic forest (BMM; CONABIO, 2010). This type of habitat has a restricted distribution nationwide (1%), housing approximately 10% of the territory's flora and fauna (Ochoa-Ochoa and Mejía-Domínguez, 2014), highlighting the presence of 459 butterfly taxa from Papilionidae, Pieridae and Nymphalidae families (Monteagudo *et al.*, 2014).

A mesophilic forest of ecological interest is located in the Zongolica mountain range, located within the Huautla-Zongolica subregion, which covers the states of Veracruz, Oaxaca and Puebla, this area is listed as a critical priority for conservation, management and restoration of such an ecosystem, since agricultural activity and deforestation have caused drastic changes in land use patterns and biodiversity, which have not been evaluated (CONABIO, 2010).

Lepidoptera, in addition to playing a preponderant role within trophic chains and pollination (Munyuli, 2011), can be used for habitat monitoring. Day butterflies are one of the most reliable biological groups as bioindicators, because of their sensitivity to environmental modifications and their ease of observation and identification (Villarreal *et al.*, 2006) additionally, they have a potential use in environmental education and bio-commerce programs.

Despite its ecological, economic and social importance, in the state of Veracruz this group of insects has been studied more from a taxonomic point of view and in conserved habitats of mesophilic mountain forest and tropical evergreen forest (Hernández *et al.*, 2010) without conducting research within agroforestry systems and even less in areas with edge effect, the lepidopterofauna of the Zongolica mountain range being one of the least known (UNIBIO, 2013). This is due to the inadequate instrumentation of modern conservation policy, which has focused on the protection of pristine areas, until recently ignoring the potential of agricultural areas, especially agroforestry systems, where crops are associated with plants woody, for the conservation of biodiversity (Perfecto *et al.*, 2003).

Depending on their degree of structural complexity, these productive systems can sustain greater biodiversity than monocultures, in addition to other environmental services (carbon capture), contributing to the conservation and sustainable use of butterflies in fragmented agricultural landscapes, and contributing to the development of rural communities (Jose, 2009; Francesconi *et al.*, 2013). Therefore, in this investigation we evaluated the edge effect in three traditional agroforestry systems (coffee, corn, backyard) and its potential relationship in the maintenance of the diversity of diurnal butterflies in a fragmented mountain mesophilic forest in Xochitla, Mixtla from Altamirano, Veracruz.

## Materials and methods

### Study area

It is located at coordinates 18° 36 75.7', 18° 37.097' north latitude and 96° 57.635', 96° 57.989' west longitude, between the heights of 1154-1505 masl; it has an area of 21.7 ha, which are located within Xochitla, Mixtla de Altamirano, Veracruz, Mexico (INEGI, 2009). In this area

a polygon composed of 24 plots was delimited, using the edge effect criterion as a confluence between each of them (EB).

### **Sampling and data collection**

44 monitoring points were selected through systematic sampling at convenience, determining 33 types of EB: 25 associated with traditional agroforestry systems (coffee, corn, backyard) and eight with reforested areas, secondary vegetation and primary vegetation. The location of each point was established considering at least 20 m between each point.

On average, the butterflies were tracked each month, between June-October 2016, with five samples lasting 4-5 days each, during clear days (without rain), from 8:00 to 17:00 h, 20 min of collection was used at each monitoring point, with a total of 73.3 h of effective sampling effort. Throughout the study, the visiting hours for the monitoring points considered were rotated. In each EB, the method of counting in points with fixed radius proposed by Ralph *et al.* (1996); however, it was modified using the center as a monitoring point and the specimens around it were collected within a radius of 15-20 m.

The capture of adults (imagos) was by means of aerial entomological networks, the trapped specimens were immobilized by exerting pressure on their thorax, subsequently introducing them to a lethal bottle with ethyl acetate; the sacrificed organisms were placed in properly labeled paper and cellophane envelopes and were dried at room temperature for later identification (Beutelspacher 1983). The study of the samples was carried out at the Autonomous University Chapingo (UACH), using the procedure suggested by Andrade *et al.* (2013) for wetting specimens; however, the assembly was carried out following the methodology proposed by De la Maza (1993).

The mounted organisms were put to dry at room temperature for a period of two weeks, and at the end of this one they proceeded to their taxonomic determination using as reference the works of De la Maza (1993); Glassberg (2007); Hernández *et al.* (2010), as well as virtual lists illustrated by Warren *et al.* (2017); Garwood (2016).

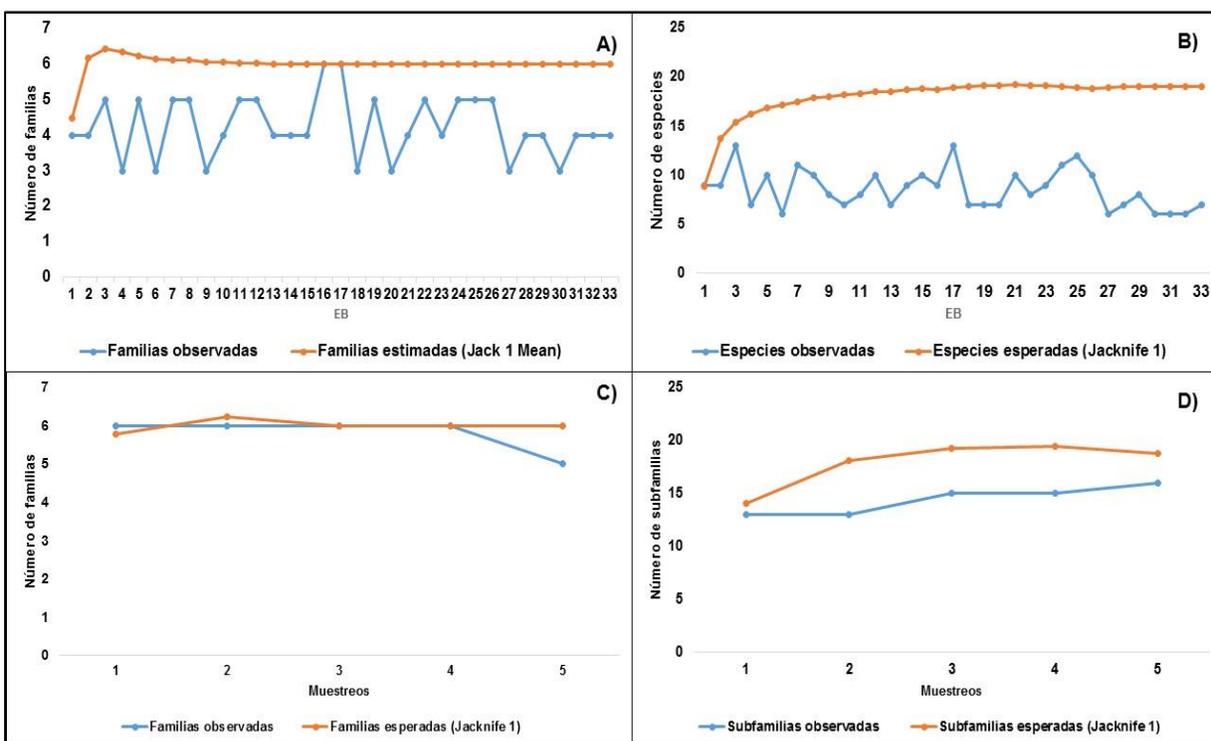
### **Analysis of data**

To determine the species wealth of the EB, presence-absence data were used for the data of: A) families by EB; b) subfamilies by EB; C) families by sampling; and D) subfamilies by sampling registered butterflies; same that were analyzed by Jackknife1 (Bojorges *et al.*, 2006), said analysis was carried out in Estimates 9.1.0 (Colwell, 2013). To estimate abundance, the relative abundance index was applied (Moreno *et al.*, 2011). The diversity of species was obtained through Shannon-Wiener, this estimate was developed in Estimates 9.1.0. (Colwell, 2013). In order to infer possible differences in wealth, abundance and diversity, the Kruskal-Wallis test was applied (McDonald, 2014). To graphically show possible differences in wealth and abundance, Cluster analysis was developed (Briceño *et al.*, 2016). In order to know if the record is similar to what was expected,  $\chi^2$  was applied, the latter analyzes were performed in JMP IN v. 12.2.0 (SAS Institute Inc., 2015).

## Results

The occurrence in the study area of 147 (12.5%) of the 1173 species-subspecies of diurnal butterflies registered for the state of Veracruz, which belong to 18 of the 21 subfamilies of diurnal lepidoptera of such entity (Luis *et al.*, 2011). The presence of an unreported species for the state, *Adelpha seriphia*, belonging to the Nymphalidae family, subfamily Biblidinae, was recorded.

The wealth results, obtained through the Jackknife1 estimator of the presence-absence data for: A) families per EB; B) subfamilies by EB; C) families by sampling; and D) subfamilies by sampling, show that up to now 70.62 is known with the sampling effort implemented; 47.60; 96.66 and 80.27%, respectively, of the species theoretically predicted by the models (Figure 1).



**Figure 1. Rarefaction curves estimated by Jackknife1 from the estimated data in the evaluated EB.**

The results of clusters for the wealth (number of species) of the previously indicated data; evidenced the conformation of 4; 5; 3; 3 Clusters (amalgamations), respectively, which shows a differential graphic trend in wealth for each of them (Figure 2).

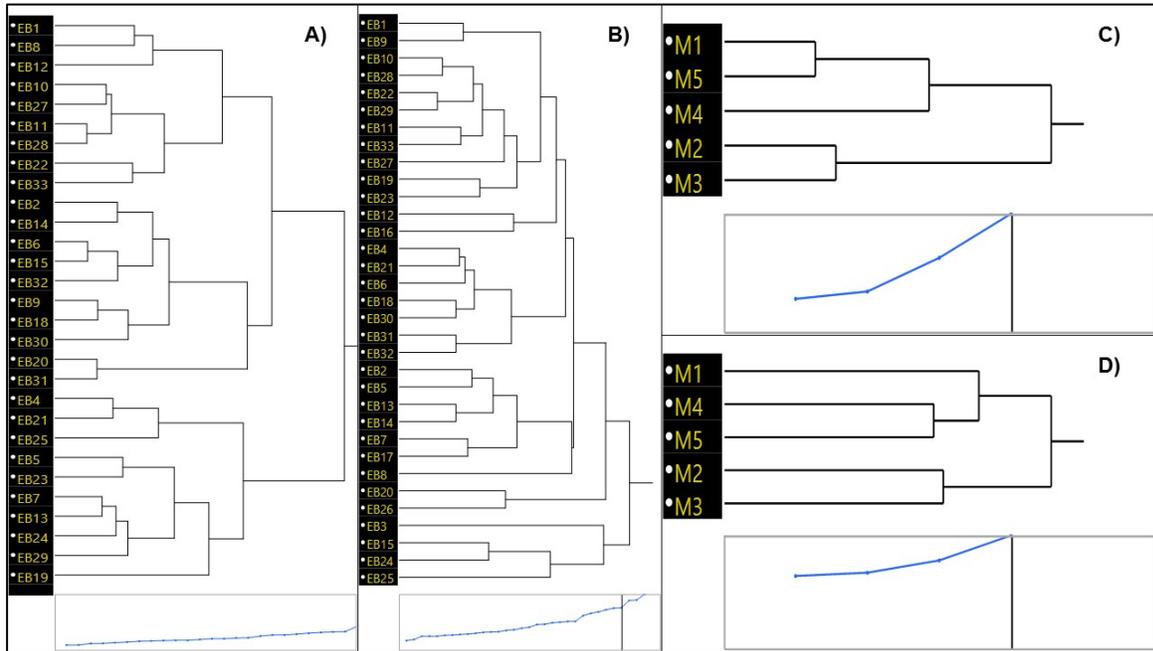


Figure 2. Wealth clusters for the data recorded in the assessed EB.

The results obtained through the relative abundance estimator made with the frequency data for: A) families per EB; B) subfamilies by EB; C) families by sampling; and D) subfamilies by sampling, show the following average values (Figure 3).

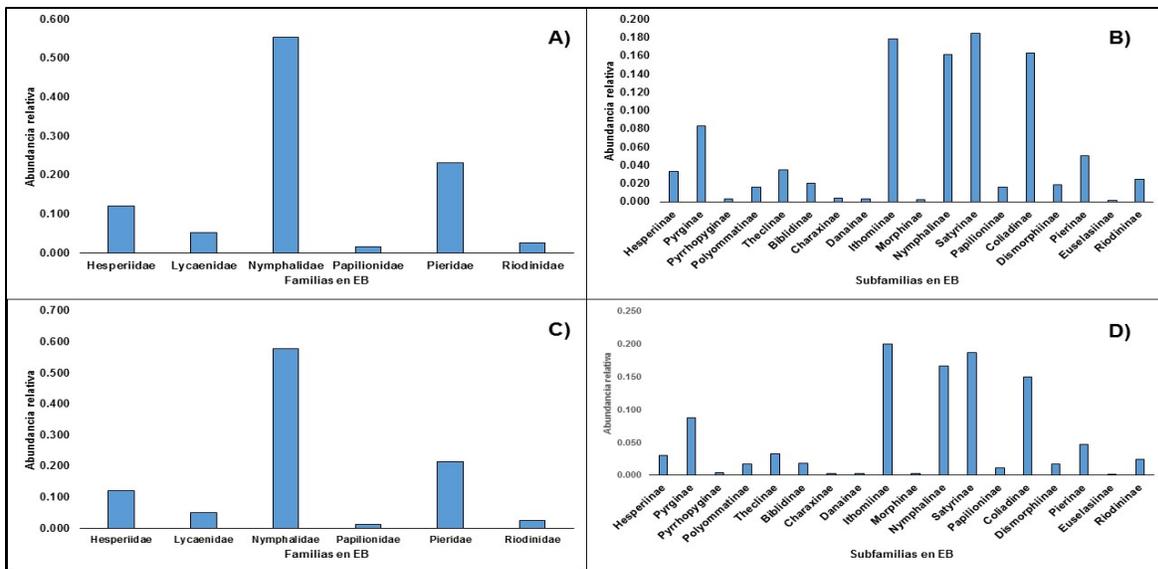
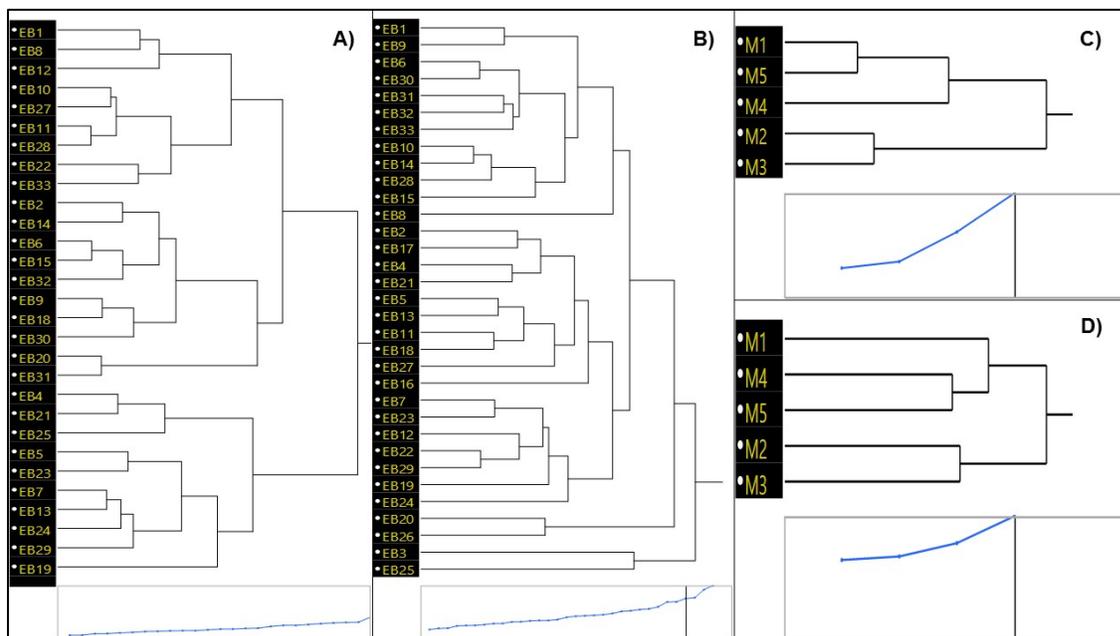


Figure 3. Relative abundance of number of individuals of registered day butterflies.

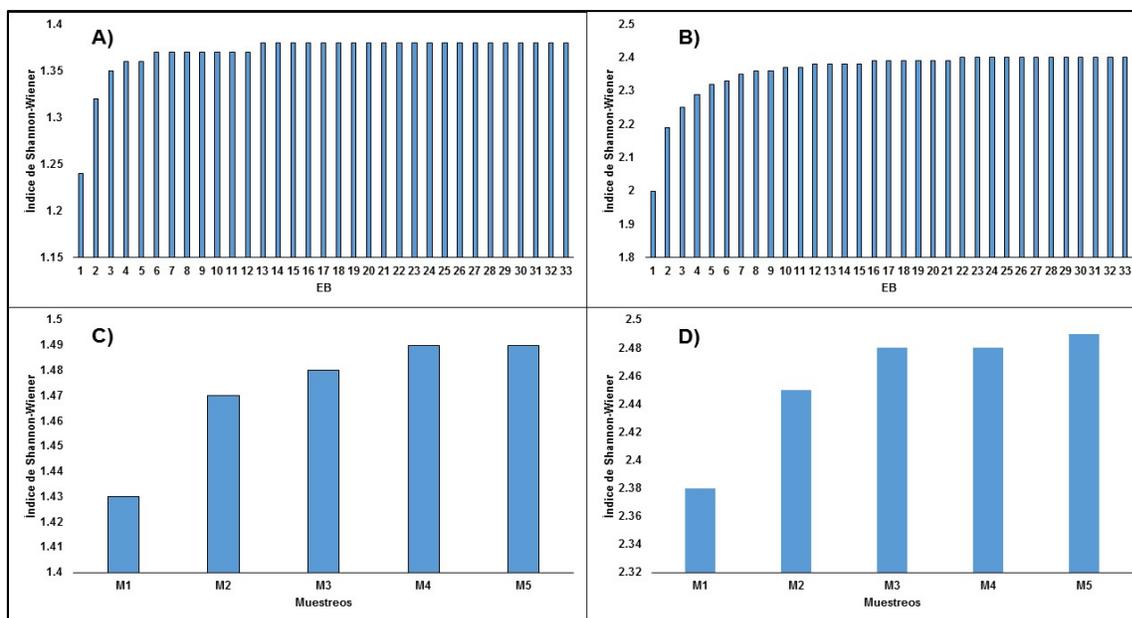
The abundance cluster results for the data: A) number of individuals per family; B) number of individuals per subfamily; C) number of individuals per family in each sampling; and D) number of individuals per subfamily in each sampling, evidenced the conformation of 5; 5; 3 and 3 frequency groups or Clusters (amalgamations), respectively, which shows a differential graphic trend in abundance for each of them (Figure 4).



**Figure 4. Cluster of abundance of the data recorded in the evaluated EB.**

The results of  $\chi^2$  for the aforementioned data; show that the proportion in abundance is statistically similar in each of them, showing values of  $p= 0.5396$ ;  $p= 0.7577$ ;  $p= 0.5708$  and  $p= 0.3083$ , respectively.

The estimated diversity results; through Shannon-Wiener of presence-absence data for: A) families by EB; B) subfamilies by EB; C) families by sampling; and D) subfamilies by sampling; evidenced values of  $H' = 1.38$ ;  $H' = 2.4$ ;  $H' = 1.49$  and;  $H' = 2.49$ , respectively (Figure 5).



**Figure 5. Diversity results obtained by Shannon-Wiener in the evaluated EB.**

Kruskal-Wallis results of wealth, abundance and diversity for the data: A) families per EB; B) subfamilies by EB; C) families by sampling; and D) subfamilies by sampling, showed that there are no differences between them, except for diversity (families and subfamilies of the EB; Table 1).

**Table 1. Kruskal-Wallis results for wealth, abundance and diversity.**

	Degrees of freedom	Chi square
Wealth		
A) Families by EB	16	0.724
B) Subfamilies by EB	9	0.4966
C) Families by sampling	20	0.6584
D) Subfamilies by sampling	13	0.5953
Abundance		
A) Families by EB	26	0.5642
B) Subfamilies by EB	20	0.6271
C) Families by sampling	22	0.8414
D) Subfamilies by sampling	30	0.4376
Diversity		
A) Families by EB	5	0.0003
B) Subfamilies by EB	11	0.0015
C) Families by sampling	3	0.2839
D) Subfamilies by sampling	3	0.2839

## Discussion

The wealth patterns found in families coincide with the studies reported by Monteagudo *et al.* (2014); Mas and Dietsch (2003); Perfecto *et al.* (2003); Connell (1978) who also registered some of the families evaluated; however, they did not consider the EB as a determining variable in the wealth of butterflies, so their presence in EB in agroforestry systems was still unknown, which assumes basic information of this group of lepidoptera under compatible production conditions with the environment.

The records for species wealth by sampling coincide with that reported by Hernandez *et al.* (2016) who applied a study for the central zone of the state of Veracruz, observing that the highest registration of species occurs from the end of May to the month of August, which agrees with the results of this research, because in this period observed the highest peak of recorded butterflies.

In contrast Francesconi *et al.* (2013) examined the potential of agroforestry and other agricultural practices for the conservation of butterflies in Sao Paulo, Brazil, determining that the coffee-shaded agroforestry system has greater potential for the conservation of butterflies than cassava and sugarcane monocultures, which differs from this by registering the highest number of butterflies in backyard associated with a home garden; however, EB was not considered a determining factor in species wealth, except for a site with fragmented mesophilic forest.

In the opposite way, in an investigation carried out with frugivorous butterflies in fragments of secondary forest and mature forest in Central Sulawesi, Indonesia; Veddeker *et al.* (2005) determined that sites with secondary vegetation harbor less species wealth compared to mature forests. However, although the butterfly records in the present study, under these conditions, are lower compared to other conditions evaluated, these authors did not link the EB as a determining condition, much less the analysis of a site with agroforestry areas immersed in a fragmented mesophilic forest, contemplating not only frugivorous species but also the lepidoptera fauna in general.

The abundance obtained in subfamilies of registered butterflies coincides with Sanchez *et al.* (2013) who, in a coffee agroforestry system in Costa Rica, identified Satyrinae and Ithomiinae as the subfamilies with the largest number of individuals, especially of the *Hermeuptychia hermes* species. However, the general trend for the registration of this species in the present study, was not only significant for the area with coffee presence conditions, but also showed the highest frequency in the registration of subfamilies for all the EBs contemplated.

In addition, the tree vegetation associated with traditional agroforestry systems shows values of wealth and abundance close to those of preserved habitats, this due to the inclusion of fruit, ornamental and other multi-purpose trees, favoring the occurrence of lepidoptera, which take advantage of food resources offered by such elements of the vegetation throughout its phenological cycle since the largest number of individuals was registered on the site, mostly made up of ornamental and fruit plants from the backyard-home garden coinciding with that reported by (Marín *et al.* , 2011). However, despite agreeing with these authors, the importance of the present is that despite considering edge effects and submerged sites in a mesophilic forest, the study was applied in different agroforestry systems, with different structures.

In a similar study, Robinson and Sutherland (2002) evaluated the abundance and wealth of diurnal butterflies on uncultivated edges of agricultural fields in the United Kingdom, applying different management treatments, identified that the planting of grass and wild flowers increases the abundance of butterflies, which is consistent with this because the highest butterfly records were observed at a point where ornamental plants were a main feature of the habitat.

For their part, Moguel and Toledo (1999) confirm that coffee agroforestry systems have a greater abundance of butterflies, representing a viable system for the protection of biodiversity. However, in the present investigation, coffee cultivation, as a traditional system compared to the other conditions evaluated, did not show a high abundance index, its importance lies in being one of the first records to evaluate day butterflies in agroforestry systems of a mesophilic forest having the edge effect as a priority.

The diversity obtained is consistent with Drewniak *et al.* (2016) who applied a study around the lepidoptera fauna associated to an urban nature reserve (General San Martín, Córdoba, Argentina), report similar values to that taxonomic group to those registered here. However, the current study was applied at sites close to agroforestry systems (disturbed areas) taking into account the edge effect between conserved sites and such systems.

In this way, the first records on the diversity of butterflies submerged in a mesophilic forest are disclosed, applying direct capture of the specimens and contemplating the EB as a precise assessment factor in particular places in Mexico contemplating different agroforestry systems for said study.

## Conclusions

The wealth, abundance, diversity of species of diurnal butterflies in the EB considered were estimated and compared. With this, it was possible to establish the relationship and importance between the three traditional agroforestry systems and an important part of the diversity of diurnal butterflies, specific to this type of EB in Xochitla, Mixtla de Altamirano, Veracruz.

The first records on the knowledge of Lepidoptero fauna of this type of systems in the area are reported, so it will prove to be the basis for future studies in which it is suggested to implement this scientific-methodological design within the plots that make up said production systems, which make up the EB under study, increasing the sampling effort on a space-time scale, in order to obtain a more complete and accurate list of the diversity of butterflies that make use of these agroecosystems, immersed in this Mesophilic forest of fragmented mountain, for this particular region of Mexico.

## Acknowledgments

To the National Council of Science and Technology (CONACYT). To the Master of Science in Agroforestry for Sustainable Development, of the Department of Soils. To Mr. Alfonso Pérez Amayo and Mr. Santos Xochiquiquisqui Xochiquiquisqui, for their invaluable support during the collections. To C. Agustín Rodríguez Fuentes for his support in the taxonomic identification of entomological material.

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