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Assessment of butterfly biodiversity across four different habitats of Kota, Rajasthan, India

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Abstract

Pollinating insects have been undergoing a decline in occurrence, abundance and diversity in many parts of the world (Ollerton et al., 2014; Potts et al., 2016) [23, 31]. Out of all the pollinating insects butterflies are the most important ones acting as "bioindicators". As the butterflies are performing the critical ecosystem service so the decline in their diversity is a matter of concern. Study area Kolipura with low level of anthropogenic activities showed the highest diversity and richness of butterfly species. The study was carried out in the year 2022-2023 and 2023-2024. For the sampling of butterflies "Line Transect Method" was used. In both the year's maximum abundance (1186, 1204) and species richness (38) was reported at site 4 (Kolipura). At site 4 the highest value of Shannon-Weiner Index and Simpson diversity index was reported in both the years followed by site 1 (Chatra Villas Garden), followed by site 3 (Bhopatpura) and lowest value of Shannon-Weiner Index and Simpson diversity index was reported at site 2 (Abheda) in both the years. The highest species richness was observed at site 4 followed by site 1, site 3 and site 2 in the year 2022-2023. The order of species richness among the sites changed in the following year (site4>site1>site2>site3). This change happened because if S is high, but N is even higher, it causes the Margalef's index to decrease. In the year 2022-2023, the highest species evenness was seen at site 4 followed by site 3, site 2 and site 1. While in the next year, the order changed to (site4>site3>site1>site2) this. Site 1, moved from 4th to 3rd indicates improved evenness overtaking site 2, this shows positive ecological change and reduced dominance of few species. Site 2, dropped from 3rd to 4th indicates that despite an increase in its evenness value, other sites (especially site 1) improved more, resulting in a relative drop in rank indicating slower or less effective improvement. Nymphalidae family was the most dominant family at all the four sites. The least dominant family at site 1 and site 4 was Hesperiidae family while this family is altogether absent at site 2 and site 3. At site 1 in the first years the most abundant species reported were Eurema laeta and Danaus chrysippus while in the year 2023-2024 the most abundant species was Eurema hecabe. At site 2 Pseudozizeeria maha was the most abundant species reported in the year 2022-23, whereas in the year 2023-24 at site 2, Danaus genutia was the most abundant species reported. At site 4, Danaus chrysippus and Eurema hecabe was the most abundant species in the year 2022-23 and 2023-24 respectively. This shift in abundance indicates a possible change in habitat conditions, resources availability, or climatic factor that favored at site 2 Danaus genutia and Eurema hecabe at site 4 in the following year. It highlights the dynamic nature of butterfly populations and their sensitivity to environmental changes. The decline in host and nectar plants for Danaus genutia causing its population to decrease. At site 3, Danaus genutia was the most abundant species reported in both the years.

Keywords: Diversity indices, species richness, species evenness, relative abundance, relative dominance, butterfly, Kota

Introduction

Butterflies are the most studied insect group in the world. But a lot of changes are being seen in the diversity of butterflies and there are many reasons of it. Factors which are negatively impacting the diversity of butterflies are Urbanization and Pesticides used in the agriculture practices. The diversity of wild flora and many cultivated crops is maintained by the essential ecosystem service called pollination. The decline in pollinators is alarming as it raises questions regarding food security and stability of ecosystem function (Potts et al., 2010) [30]. Several anthropogenic drivers are threatening the abundance, diversity and health of wild and managed pollinators, and the pollination service they provide to wild plants and crops (Vanbergen, & Insect Pollinators Initiative, 2013). Intensive agriculture activities,

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indiscriminate use of herbicides and insecticides, ground burning and monoculture farming is adversely affecting the diversity of butterflies. The drivers for the decline in insect pollinators worldwide include habitat transformation or fragmentation (Kennedy et al., 2013) [11], loss of diversity and abundance of floral resources (Kremen et al., 2007) [15], inappropriate use of pesticides (Pettis et al., 2013) [28] and climate change (Schweiger et al., 2010) [38]. Around 95% individuals of butterflies are dying due to parasitic infestation, predators and parasitoids attack, habitat loss and pesticidesinsecticides poisoning in agriculture ecosystems before attaining their adulthood. The natural area without or less anthropogenic activities is providing the best habitat for butterflies to adapt, whereas in areas with significant human interference butterflies are generally unable to adapt to the surrounding environment. Hence, assessing the biodiversity of butterfly populations greatly aids in monitoring and measuring environmental changes.

Plan of restoration of biodiversity and development of management strategies can be done in future with the help of present investigation in order to ensure the diversity of butterflies and the benefits provided by them. So the results of present study will uncover the health of ecosystem and provide the baseline data which is focusing on the need to conserve the ecosystem for the Lepidopterans biodiversity.

Materials Methods: Study area: Site 1 Chatra vilas garden is located at Nayapura Kota just adjacent to Kishore Sagar Talab. The geographical coordinates lies between latitude 25.2012° north and longitude 75.8566° east. Area covered by C.V garden is 62.1 acres. The Chatra villas garden is enriched with unique flora and fauna. Different plant species present in Chatra villas garden are Leucophyllum frutescens, Lantana camara, Nuphar advena, Hamelia patens, Phlox, Crinum asiaticum, Langerstroemia indica, Parthenium hysterophorus, Azadirachta indica, Murraya paniculata, Tridex Procumbens, Mangifera indica, Nelumbo nucifera, Acacia arabica, Calotropis procera, Hibiscus rosa-sinensis, Psidium guajava etc.

Site 2 is Abheda, which is situated near the Chambal river of Kota, Rajasthan. The geographical coordinates of Abheda Mahal lies between latitude 25.2000° North and longitude 75.7905° east. Abheda Mahal is located about 8 Kms from the main Kota city. This area is covered by grass and various plant species like Allamanda cathartica, Bougainvillea glabra, Calotropis procera, Hibiscus rosa-sinensis, Lantana camara, Acacia catechu, Eicchornia crassipes, Ficus benghalensis, Parthenium hysterophorus and Azadirachta indica etc.

Site 3 is Bhopatpura Village which is a small Village/hamlet in Talera Tehsil in Bundi District of Rajasthan State, India. It comes under Bhopatpura Panchayath. The geographical coordinates of Bhopatpura Village lies between latitude 25.314676° North and longitude 75.735832° east. In this village Rosa rubiginosa and Tagetes erecta flowers are cultivated. Other crops are also cultivated in this village like Triticum aestivum, Cicer arietinum and different types of vegetables (Spinacia oleracea, Raphanus sativus, Pisum sativum and capsicum frutescense).

Site 4 is Kolipura Village which is located in Ladpura tehsil of Kota district in Rajasthan, India. It is situated 38 km away from sub-district headquarter Ladpura (tehsildar office) and 38 km away from district headquarter Kota. It is located near the Mukundra Hills Tiger Reserve which is a protected area

that includes the Mukundra Hills National Park, Darrah Sanctuary, Jawahar Sagar Wildlife Sanctuary, and part of the National Gharial Sanctuary. The geographical coordinates of Kolipura Village lies between latitude 25.2021° North and longitude 75.8567° east. This area is endowed with lush green vegetation. Various plant species observed in this area are as follows- Different plant species present in Chatra villas garden are Leucophyllum frutescens, Lantana camara, Nuphar advena, Hamelia patens, Phlox, Crinum asiaticum, Langerstroemia indica, Parthenium hysterophorus, Murraya paniculata, Tridex Procumbens, Mangifera indica, Nelumbo nucifera, Acacia arabica, Calotropis procera, Hibiscus rosasinensis. Psidium guajava Allamanda cathartica. Bougainvillea glabra, Calotropis procera, Acacia catechu, Eicchornia crassipes, Ficus benghalensis, Parthenium hysterophorus and Azadirachta indica etc.

Methods of sampling and identification of Butterflies

The sampling was done by "Line Transect Method". All the sites were visited at regular intervals. Butterflies were observed between 7 am to 11 am and in evening 4 pm to 6 pm. To identify the butterflies, they were caught using aerial net, photographed and then safely released back into the environment. Insects were photographed using Nikon Z611. The insects were identified based on their wings color, pattern, shapes, sizes, with the help of entomological experts, and available literature.

Statistical analysis: Data of identified species was analyzed for richness and abundance by using various diversity indices.

Shannon-Wiener diversity index (H')

It is alpha diversity index and it depends upon species richness and species evenness. Shannon-Wiener diversity index (Shannon, 1949) was calculated

 $H'=-\Sigma (p_i * ln(p_i))$

Where pi = S / N

S = number of individuals of one species

N = total number of all individuals in the sample

ln = logarithm to base e

2: Simpson's Index (D)

Simpson's index denotes the alpha diversity of the selected area. It depends on both species richness and evenness. The Simpson's Index (Simpson, 1949) was calculated as-

 $D = \Sigma ni (ni-1) / N (N-1)$

Where

N = total number of individuals

ni = number of individuals of ith species

Simpson Index of Diversity (1 - D)

The Simpson Index of Diversity (1 - D) is a measure of diversity that takes into account both richness (the number of species) and evenness (the relative abundance of species). It reflects the probability that two individuals randomly selected from a sample will belong to different species.

 $1-D=1-\Sigma ni (ni-1)/N (N-1)$

- ni = number of individuals of species i
- N = total number of individuals of all species

- D = Simpson's Index (dominance index; probability two individuals are the same species)
- 1–D = Simpson's Diversity Index (probability two individuals are different species)

4 Margalef's Index (D_{Mg})

This index is also an alpha diversity index and used as a simple measure of species richness (Magurran, 1988).

$$D_{Mg} = (S - 1) / \ln N$$

S = total number of species

N = total number of individuals in the sample

ln = natural logarithm

5 Pielou's Evenness Index (J')

Pielou's index is an alpha diversity index that measures how evenly species are distributed in a community ((Pielou, 1969).

$$J' = H' / ln(S)$$

H' = Shannon - Wiener diversity index

S = total number of species in the sample

6 Relative abundance of butterfly families

The relative dominance of butterfly families was calculated by using dominance index.

Relative abundance (family wise) = $ni \times 100/N$

Where ni = number of butterflies in the 'i' th family, and N = the total number of butterflies in all the families collected in each habitat

7. Jaccard's Index (J)

The Jaccard Similarity Index measures the proportion of shared species between two communities relative to the total number of species found in both.

$$J = S_c \! / S_a \! + \! S_b \! + \! S_c$$

Where S_a and S_b are the number of species unique to Site a and Site b respectively, and S_c is the number of species common to both the sites.

Results and Discussion

Butterfly species distribution among families and species composition: In the year 2022-2023, total of 1,612 individuals and in the year 2023-2024 total of 1,692 individuals of 38 species belonging to 5 families of superfamily Papilionoidea were recorded. In both the years similar number of species were reported at site 4 (Kolipura-38 species), site 2 (Abheda- 11 species) and site 3 (Bhopatpura- 8 species). Whereas at site 1 (C.V garden) 19 species were reported in the year 2022-2023 and 25 species were reported in the year 2023-2024.

In terms of species the least dominant family in both the years is Hesperiidae at site 1 and site 4. At site 2 (Abheda) the least dominant family in terms of species was Lycaenidae (18.18%) followed by Pieridae (27.27%) in both the years. While at site 3 in both the years, Lycaenidae (25%) and Pieridae (25%) showed the similar percentage in terms of species composition.

The most dominant family in terms of species composition at all the four sites in both the years was Nymphalidae family.

Table 1: Family wise percentage composition of species in the year 2022-2023

S. No.	Families	C.V garden	Abheda	Bhopatpura	Kolipura
1	Papilionidae	15.78%	-	-	10.52%
2	Pieridae	21%	27.27%	25%	23.68%
3	Lycaenidae	21%	18.18%	25%	18.42%
4	Nymphalidae	36.8%	54.54%	50%	47.36%
5	Hesperiidae	5.26%	-	=	5.26%

Table 2: Family wise percentage composition of species in the year 2023-2024

S.No.	Families	C.V garden	Abheda	Bhopatpura	Kolipura
1	Papilionidae	16%	-	-	10.52%
2	Pieridae	16%	27.27%	25%	23.68%
3	Lycaenidae	24%	18.18%	25%	18.42%
4	Nymphalidae	40%	54.54%	50%	47.36%
5	Hesperiidae	4%	-	-	5.26%

Table 3: Comparison between site 1, 2, 3 and 4 with respect to families, species and individuals in the year 2022-2023, 2023-2024

Sites	Number of Families		Number of species		Number of individuals	
Sites	2022-2023	2023-2024	2022-2023	2023-2024	2022-2023	2023-2024
1	5	5	19	25	205	365
2	3	3	11	11	191	86
3	3	3	8	8	30	37
4	5	5	38	38	1186	1204

At site 1 in the first years the most abundant species reported were *Eurema laeta and Danaus chrysippus*. While in the year 2023-2024, *Eurema hecabe* was the most abundant species. At site 2 was *Pseudozizeeria maha* (50) was the most abundant species reported in the year 2022-23, whereas in the year 2023-24 at site 2, *Danaus genutia* (20) was the most abundant species reported. Due to indiscriminate cutting of plants at site 2, plant composition in the area changed it lead to the decline of host plants preferred by *Pesudozizeeria maha*

and favored those plants used by *Danaus genutia*. At site 3, *Danaus genutia* was the most abundant species reported in both the years. At site 4, *Danaus chrysippus* (64) and *Eurema hecabe* (66) was the most abundant species in the year 2022-23 and 2023-24 respectively, so due to change in plant composition the dominant species shifted. It highlights the dynamic nature of butterfly populations and their sensitivity to environmental changes.

Species diversity, richness and abundance

Alpha diversity: Alpha diversity refers to the number and variety of species (or other taxonomic units) in a specific, relatively small, and uniform habitat.

In both the years the highest Relative abundance was showed by the Nymphalidae family followed by family Pieridae, Lycaenidae, Papilionidae and Hesperiidae. The abundance of Nymphalidae can be due to polyphagous habit that helped them to live in all habitats (Sreekumar and Balakrishnan, 2001b) [43]. The higher number of Pieridae and Lycaenidae is supported by studies of Bernard who reported that that these two families can be seen almost everywhere. But at the site 2 in the year 2022-2023 the second highest Relative dominance was showed by the Lycaenidae family followed by Pieridae (Table 5a).

Table 5a: Relative abundance of various families at four different sites in the year 2022-2023

S.No.	Families	C.V garden	Abheda	Bhopatpura	Kolipura
1	Papilionidae	3.41%	-	-	4.72%
2	Pieridae	37%	25.65%	30%	32.20%
3	Lycaenidae	8.29%	27.74%	20%	15.59%
4	Nymphalidae	50.73%	46.59%	50%	46.62%
5	Hesperiidae	0.48%	-	-	0.84%

Table 5b: Relative abundance of various families at four different sites in the year 2023-2024

S.No.	Families	C.V garden	Abheda	Bhopatpura	Kolipura
1	Papilionidae	3.56%	-	-	3.23%
2	Pieridae	32.05%	31.39%	29.72%	34.05%
3	Lycaenidae	17.26%	22.09%	18.91%	17.77%
4	Nymphalidae	46.57%	46.51%	51.35%	43.10%
5	Hesperiidae	0.54%	-	-	1.82%

Family Papilionidae and Hesperiidae were altogether absent at site 2 (Abheda) and site 3 (Bhopatpura village) in both the years. This result favors the statement of Ombugadu *et al.*, (2021) ^[24]. The reason for the lowest species richness and abundance showed by Hesperiidae family may be their flight period (early morning hours at dawn and dusk, Kehimkar, 2008) ^[10] and narrow host plant range and absence of suitable habitat.

Margalef's index is a measure of species richness, considering the number of species and total individuals of a community. Margalef's index of site 1 (3.381) indicates the moderate species richness which can be improved further with conservation practices. At site 2 and site 3 low Margalef's index (Table-6) indicates a more limited community which have slightly low diversity. Low diversity at these two sites is due to the habitat disturbances and habitat loss. Highest Margalef's index of site 4 reflects the highest species richness and suggests that the ecosystem is much diverse as compared to all the other sites of present study. So the site 4 is ecologically stable and well balanced habitat making it a key conservation area of all the study sites.

During the study period increase in Margalef's index (Table-6) at site 1 indicates a notable improvement in the species richness, this is due to the habitat restoration and improved condition in the habitat. In the two successive years increase in Margalef's index at site 2 shows that there is slight recovery in species richness, though it is relatively low.

At site 2 increase in Margalef's index value is due to decrease in the total number of individuals in the community so it indicates the health of the ecosystem is declining. This is due to indiscriminate cutting of plants from the ecosystem hence disturbing the ecological balance of the ecosystem.

At site 3 there is no remarkable change in the Margalef's index, so there is no notable change in the species richness at site 3.

At site 4 species richness is still the highest and stable, which is indicated by minimal change in the Margalef's index value of site 4 in the two successive years. So making the habitat an ecological hotspot for biodiversity. Positive trend at site 1

suggests that there is improvement in species richness indicating ecological recovery in the habitat.

In the present study highest Shannon-Weiner index and Simpson index of diversity at site 4 indicates a well-balanced ecosystem with high number of species and even distribution of species in both the years. The findings of this study further supported by Hill *et al.*; Brown, Bonebrake *et al.* and Akwashiki *et al.* ^[7] who reported great abundance of butterfly species in less disturbed habitats. In the Undisturbed site the higher diversity and abundance was observed, which may be due to the availability of nectar and host plants of butterflies. This result favors the statement of Sreekumar and Balakrishnan (2001a) ^[42] they suggest that prevalence of butterfly species at a particular habitat depends on a wide range of factors, of which the availability of the food is the most important.

Second highest Shannon-Weiner index was observed at site 1 (2.288 and 2.924) in both the years, indicating moderate diversity and reasonably diverse community having rich species distribution.

Relatively low Shannon-Weiner index and Simpson index of diversity was reported at site 2 and site 3 which indicates low diversity with fewer species being dominant. In the present study high Shannon-Weiner index of site 4 and site 1 indicates that these two sites are having stable and healthy ecosystem with good resource availability. Lower Shannon-Weiner index and Simpson index of diversity of site 2 and site 3 in the present investigation indicates the environmental disturbances.

In the two successive years there is increase in Shannon-Weiner and Simpson index of diversity at site 1 which indicates the higher species diversity compared to last year, which indicates improved habitat conditions, better resource availability for the butterflies at site 1.

At site 2 and site 3 there is slight increase in Shannon-Weiner index and Simpson index of diversity (Table-6) suggests that there is minor improvement in species diversity but still relatively low as compared to site 1 and site 4. During the study over the last two years there is slight drop in Shannon-

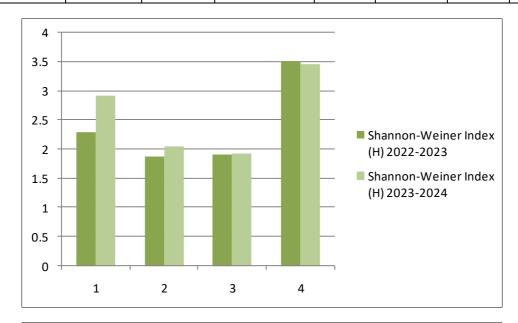
Weiner index and Simpson index of diversity at site 4 which suggests that high diversity is still maintained at site 4 but experiencing small changes in species distribution. Other possible cause for these minor changes is the increasing anthropogenic activity in the study area. So the monitoring of this natural area should be done to ensure the ecosystem stability.

Pielou's index: During the present investigation at site 4 had the highest evenness (Table-6), which indicates that the species are distributed more equally. Therefore no single species is dominating in the ecosystem. Thus ecosystem at

site 4 is stable and healthy ecosystem with minimal competition issues. Similarly site 3 also has high evenness indicating good species balance in the ecosystem though slightly lower than site 4. However site 1 and site 2 have moderate evenness with some species being more dominant at these sites. This in turn reduces the evenness of the ecosystem. Such results are due to the habitat disturbances like indiscriminate cutting of plants in the ecosystem. Lower evenness in the ecosystem contributes to the lower biodiversity as dominant species outcompete the other species.

Table 6: Comparison of butterfly diversity indices in site 1, site 2, site 3 and site 4

Sites		Shannon-Weiner Index (H')		Simpson index of diversity (1-D)		Pielou's Index (J')		Margalef's Index (D _{Mg})	
	Sites	2022-2023	2023-2024	2022-2023	2023-2024	2022-2023	2023-2024	2022-2023	2023-2024
	1	2.288	2.924	0.865	0.938	0.777	0.908	3.381	4.068
	2	1.883	2.056	0.824	0.858	0.785	0.857	1.904	2.245
	3	1.905	1.938	0.863	0.862	0.916	0.932	2.058	1.939
	4	3.506	3.458	0.968	0.966	0.963	0.950	5.227	5.216



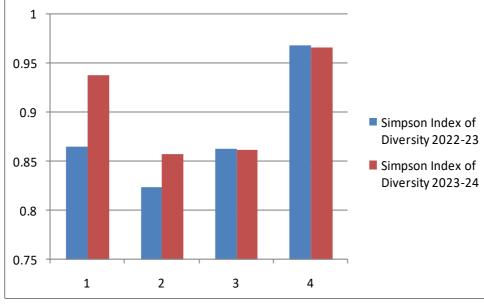
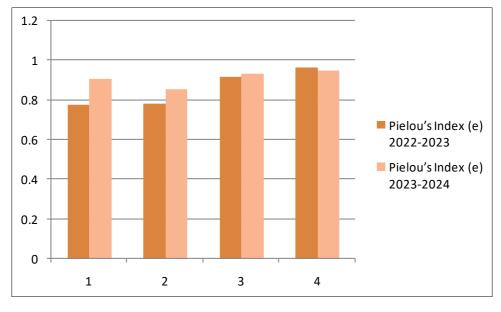


Fig 1a & 1b: Comparison of Simpson's index of diversity and Shannon-Weiner index of site 1, site 2, site 3 and site 4.



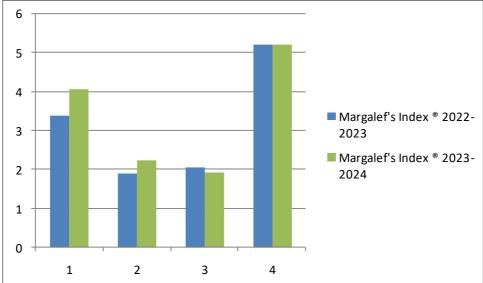


Fig 2a & 2b: Comparison of Pielou's index (species evenness) and Margalef's index (species richness) of site 1, site 2, site 3 and site 4

2. Beta diversity

It is a measure of changes in species composition between different ecosystems. It reflects how the biodiversity varies across the spatial scales.

Beta diversity in the year 2022-2023, site 2 and site 3 show relatively high similarity as indicated by the Jaccard index value of site 2 and site 3 (0.30) suggesting that 30% of the species are common between these two sites. The Jaccard index value (0.27) of site 1 and site 2 indicates moderate similarity with nearly half of the species shared between the two sites. Similarly Jaccard index value of site 1 and site 4 (0.25) also indicates moderate similarity showing that 25% of the species are shared between these two sites.

Jaccard index value (0.23) of site 1 and site 3 shows a lower similarity with approximately 23% of species in common. Low Jaccard index value of S2-S4 and S3-S4 suggests minimal overlap in species composition between these two pairs of sites. Overall site 2 and site 3 share the most species, while site 3 and site 4 have least in common so beta diversity is highest in S3-S4 as these two sites show significant difference in species composition.

Beta diversity is lowest at S2-S3 sites, as these two sites share

similar species indicating the homogeneity in the ecosystem. Beta diversity in the 2023-2024 at S2-S3 sites show high similarity, which is indicated by the highest Jaccard's index value (0.30). As 30% of the species are common in between these two sites. In the year 2023-2024 Jaccard's index value of S1-S4 reached 0.28, indicating the high similarity with 28% of the species shared between these two sites. S1-S2 sites showed lower similarity with comparatively low Jaccard index value (0.23), with approximately 23% of the species in common.

S1-S3 sites overlap with 19% of species shared between these two sites, which is indicated by their Jaccard index value (0.19). Lowest value of Jaccard index of S2-S4 (0.18) and S3-S4 (0.15) indicates that there is very little overlap in species composition between these two pairs of sites. Overall, we can say that S2-S3 site share the most species while S3-S4 have the least common species in both the years.

Beta diversity is highest at S3-S4 as these two sites show significant difference in species composition in both the years. Beta diversity is lowest at S2-S3 sites as these two sites share similar species in both the years indicating the homogeneity in the ecosystem.

Table 7: Jaccard's Index of Butterflies (2022-23, 2023-2024)

Sites	2022-2023	2023-2024
S1-S2	0.27	0.23
S1-S3	0.23	0.19
S1-S4	0.25	0.28
S2-S3	0.30	0.30
S2-S4	0.18	0.18
S3-S4	0.15	0.15

3. Gamma Diversity: It describes the overall species diversity across communities within a large geographical area.

Table 8: Gamma Diversity in the year 2022-2023 at site 1, 2, 3 & 4

Sites	Number of families of butterflies	Number of species of butterflies	Number of individuals of butterflies
1	5	19	205
2	3	11	191
3	3	8	30
4	5	38	1186
Gamma diversity	5	38	1,612

Table 9: Gamma Diversity in the year 2023-2024 at site 1, 2, 3 & 4

Sites	Number of families of butterflies	Number of species of butterflies	Number of individuals of butterflies
1	5	25	365
2	3	11	86
3	3	8	37
4	5	38	1204
Gamma diversity	5	38	1,692

Conclusion

Based on the work carried out during this study, we can conclude that the diversity of butterfly species was found to be higher in natural sites due to minimal human interference. This indicates that the conservation of natural and less-disturbed areas is essential for preserving biodiversity.

The study also found that the number of butterflies was relatively low in agriculture lands. The main reason for this is the excessive and indiscriminate use of pesticides and chemical substances by farmers, often without proper knowledge. These chemicals are not only harmful to pests but also to beneficial pollinators like butterflies. Since butterflies play an important role in crop pollination, their declining numbers are a matter of concern for the agricultural system as well. There is a need for farmers training to promote integrated pest & pollinator management (IPPM) strategies to reduce over reliance on chemical pesticides in the management of crop pests (Meissle et al., 2010). The second possible reason for the decline in butterfly numbers may also attributed to the practice of monoculture farming, which reduces habitat heterogeneity and limits the availability of diverse nectar and host plants.

During the study, we also observed that at Abheda site, in the following year, there was a noticeable decline in butterfly diversity due to excessive and insensitive cutting of plants. This indicates that if we continue to exploit natural resources indiscriminately, it will not only disturb the ecological balance but also endanger the existence of those species that are integral to the ecosystem. Protecting nature is not just about conserving the environment, but also about safeguarding all the living beings that depend on it.

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