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DIVERSITY AND ABUNDANCE OF THE BEETLES IN DIFFERENT CROPS OF DISTRICT LAYYAH

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ABSTRACT

Insects are the most dominant and diverse creatures on the earth. The Coleoptera order, to which beetles belong, is the most distinct order among insects, with 400,000 species described in 170 families. Not only do they prey on crop-damaging pests such as jassids, aphids and whiteflies, but they are also excellent decomposers. The coccinellidae beetles are recognized as being of great economic significance in agroecosystems. The present study was executed to estimate the diversity and abundance of beetles in different crops of district Layyah. Samples were collected fortnightly by using pit fall traps, sweep netting, optionally by hand-picking in the morning and afternoon from seasonal crops (wheat, maize and mustard) in covered glass vial jar then preserved in ethanol and brought to laboratory. The species richness of seven *Coccinella septempunctata, E. indica, S. quadrillum, C. transversalis, H. axyridis, C. sexmaculata and P. vigintiduopunctata* was measured. The diversity and abundance of coccinellidae species was recorded maximum in maize crop while at wheat crop was observed minimum. The diversity index of coccinellids species was recorded as (H^0=1.7633), Evenness (0.904) and Dominance (0.8006) among the three crops of district Layyah. *C. septempuntata* was the most abundant species, accounting for 181 (34.54%) and least abundant species was *S. quadrillum* with 30 (5.72%) specimens. The obtained data and results were analyzed statistically by one-way ANOVA. Then p-value of all the f-ratio of six months for three crops was less than 0.001 which shows the highly significant results.

INTRODUCTION

Maize (*Zea mays* L), commonly known as the Queen of Cereals and one of the most important cash crop, maize provides the primary source of calories and minerals (Nasar *et al*., 2023). According to the Pakistan Economic Survey, the production of maize accounts for 2.9% of the total output of agriculture and 0.6% of GDP. Wheat (*Triticum aestivum*) is the primary cereal crop and main source of food in Pakistan, an important source of calories and proteins contains 18–20%. It was grown on a total of 8734 thousand hectares in 2018 and it contributed 1.7% to the nation's GDP and 9.1% to the value added in agriculture. (Elahi *et al*., 2022). Mustard (*Brassica juncea*) is grown in Rabi season, the mustard seeds have a high nutritional value and contain 30 to 45% protein and 40 to 42% oil. Mustard cake is made from the residual meal after extracting the oil from the mustard seed. 25 to 30% crude protein, 1.8 to 2.0% phosphorus, 5% nitrogen and 1.0 to 1.2% present in the oil cake. (kumar *et al*., 2022).

Insects are the most dominant and diverse creatures on earth, ubiquitous and are usually distinct members of the phylum Arthropoda under the Insecta. Coleoptera, to which beetles belong, with 400,000 species described in 170 families. Over 6000 species in 300 genera have been recognized throughout the world. Additionally, more than 300 species from the Indo-Pak subcontinent and more than 71 species from Pakistan were reported (McKenna *et al*., 2019). Beetles can be utilized to identify alterations of all kinds in the environment. (Ghanne*et al*., 2018). Insects often jump using spring-loaded mechanism that store elastic energy and transform muscle effort into fast movement at appropriate moment. These systems, sometimes known as "catapult mechanisms" (Nadein and Betz, 2016).

Diversity refers to an extensive variety of variations between individuals; biological diversity consequently refers to diversity within the world of living things (Vanclay, 1992).The family Coccinillidea belongs to order Coleoptera is the most species-rich group of organisms at present that bears a large group of predators. Herbivorous ladybirds are herbivorous and crop-eating, and therefore agricultural pests. In a ladybird beetle called *Harmonia axyridis*, a gene that controls wing color pattern polymorphism has been found. The leg joints of ladybird beetles usually discharge a toxic yellow haemolymph which includes alkaloids and volatile organic substances that predators find repulsive when they attack. (Niimi and Ando, 2021). For instance, several insecticides, particularly thiamethoxam, tolfenpyrad, imidacloprid, pyriproxifen, deltamethrin and heptenophos, could cause toxic effects on *C. septempunctata* that are both acute and sublethal. (Hori *et al*., 2011). Various pesticides, especially imidacloprid, chlorantraniliprole and thiamethoxam, may have physiological impacts on an insect's orientation, predation, longevity, and reproduction. However, two fungicides (Tebuconazole and Myclobutanil) very resistant to *H. axyridis*. (Zhu*et al*., 2023).

Rove beetles are a highly diverse group of insects belonging to the Staphylinidae family (Xiao *et al.,* 2017). Rove beetles are abundant and dominant in agro ecosystems, accounting for 53% and 60% of soil surface adult and larval abundance, respectively. Ground beetles were previously identified as potential indicators of ecological disturbances in Palearctic agro ecosystems (Bohac and Jahnova, 2015).The most important and abundant groups of aquatic insects are water beetles around the world, with more than 13000 known species. Diptera is the only insect order which contains more aquatic taxa than other order coleoptera. The largest families of water beetles includes Hydrophilidae and Dytiscidae (Short, 2018).

Natural enemies of aphids that live in agriculture environments are often exposed to pesticides during pursuing hosts or prey. The application of neonicotinoid pesticides, which preferably affect the nicotinic acetylcholine receptors of insects, increased significantly in the 1980s. The acute toxic effects of various insecticides at field rates and mechanisms of resistance to *C. septempunctata* have been observed. Imidacloprid may have non-lethal consequences on *C. septempunctata*; however these effects and any potential population-level modifications have not yet been identified. The sublethal effects of imidacloprid are also reported at three low concentrations on the growth, demographic characteristics and fecundity of *C. septempunctata* in this environment (Xiao *et al.*, 2016).

Agriculture landscapes are a popular system for studying wildlife ecosystem services because of human dependence on proper functioning and a high level of disturbance. However, dung accumulation promotes the spread of pests and diseases. Dung beetle plays a crucial role in dissolving of manure in both tropical and temperate agro ecosystems, improving livestock production efficiency (Herrero and Thornton, 2013).

Adalia bipunctata L. (Coleoptera: Coccinellidae) is most widely used predatory ladybird beetle in Europe, yet there is no scientific evidence to support the impact of its discharge on lime trees. The aphid *Dysaphis plantaginea* which attacks on apple trees was effectively managed by released *A. bipunctata* in one of few instances of ladybirds' successful augmentative management in different systems in the open air. (McKenna *et al.,*2019). Carabid beetles may be assembled in organized way using pitfalls and are found in a wide range of biotopes. They are frequently regarded as reliable bioindicators and helpful assessment tools for the purposes of nature conservation. (Desender *et al*., 2010).

Particularly in highly industrialized areas, biodiversity is quickly declining due to the introduction of harmful species, fragmentation of biotopes, eutrophication and changes in the climate. For the control of aphids, various methods have been proposed. These include host plant resistance, cultural, physical, mechanical, biological and chemical barriers. (khan *et al*., 2011). Low yield can be caused by a number of abiotic and biotic stresses including unfavorable weather, improper fertilizer application, a lack of high-yielding varieties, low soil fertility, the presence of diseases, insect pests and most importantly a lack of IPM techniques to control insect pests. (Akhtar *et al.,*2004).

METHODOLOGY

Study area

Layyah is a district of province Punjab, Pakistan and its total area is about 6291 km^2. Geographically it is located in south west part of province of Punjab (30° 57˙55 North and 70° 56’38 East). The district Layyah is divided into 3 tehsils namely Layyah, Choubara and Karor Lal Esan. The climate of this area is dry and rainfall is rare. *(Yasmeen et al., 2023).Its soil is good for the production of both Rabi and Kharif crops due to their environmental and climate conditions. An extensive survey was carried out in the selected crops (Maize, mustard and wheat) from different localities. An area of 1 acre for each crop was selected for sampling, accordingly.*

Sampling

Sampling was done fortnightly for the period of about six month in the morning and afternoon. Sampling of Coleopteron species at different developmental stages was taken from selected crops at dawn when maximum foraging is seen. Insect fauna was taken in December to May. Samples were collected fortnightly by using pit fall traps, sweep netting, optionally by hand-picking from seasonal crops (wheat, maize and mustard) in covered glass vial jar.

Hand picking

This method was used to catch insects that are safe for people to handle when they are caught with their bare hands. The majority of medium-sized to larger insects can be handled by this method. Hand gloves are necessary for hand picking method.

Sweep net method

Sweep net are used to collect insect at random locations. When using a sweep net, one can either run while pulling the net through the grass or quickly sweep the net back and forth in the grass.On top of the foliage, a sweep net may collect moderately dispersed insect species.

Pitfall trap method

A pitfall trap method is used to catch insects that are moving on the surface surface of the ground. Pitfall traps typically consist of a beaker that has been buried so that its lip is level to the

earth's surface. The beaker's open top was covered to keep out rain and small creatures while allowing insects to enter inside.

Preservation

The collected specimens was taken in covered glass vials and immediately preserved in 70% ethanol, 10% formaldehyde aqueous solution with one or two drops of glycerin and brought to pest control laboratory. Each collection was labeled with respective information.

Identification

The collected specimens were identified morphologically up to species level by taxonomic keys provided by Naz *et al.,* (2012) and Hayat (2013). The identification of collected specimen was also being done with the help of literature keys by Rafi *et al*. (2005) and Patil *et al*. (2019).

Statistical analysis

To check the diversity and distribution of beetle’s fauna in different crops of Layyah, Shannon-Weiner diversity index was used. It tells us about the abundance of beetle’s species in different sites. To determine the importance of geographical and temporal variation in diversities, F statistical test is used and then single factorial ANOVA was also used. Shannon-Wiener Index is used to check the diversity of beetles and given as: $H=\sum [(pi) \times \ln(pi)]$

Where

Pi is a measure of the proportion of a species, and it is computed as "ni/N," where "ni" indicates the

number of individuals in the species and "N" for all the sample's individuals.

- S = richness of species
- Hmax=ln(S) = Maximum diversity possible
- E =H/Hmax=Evenness

RESULTS

The current field study was conducted to evaluate the diversity and abundance of coccinellidae beetles (Coleoptera) in different crops regarding maize, mustard and wheat in district Layyah. A total of 524 adults from seven species of coccinellid were collected from three different crops.An area of one acre was selected from each of three crops. As a result of extensive survey and sampling on weekly basis, following 7 species was identified.

C. Septempunctata
C. Transversalis
H. Axyridis
P. vigintiduopunctata
C. Sexmaculata
E. Indica
S. quadrillum

The most abundant species of Coccinellidae that was sampled was *Coccinella septempunctata* which is about 181 specimens (34.54%). *Scymnus quadrillum* was lowest during sampling about 30 (5.72%) specimens.

Sr. No.	Family	Sub-Family	Genus	Species	Site 1: Maize						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	-	11	20	23	13	67
2				*C. transversalis*	-	-	8	14	16	4	42
3				*H. axyridis*	-	-	4	8	9	5	26
4			Psyllobora	*P. vigintiduopunctata*	-	-	3	6	8	2	19
5			Cheilomenes	*C. sexmaculata*	-	-	4	3	7	3	17
6		Epilachninae	Epilachna	*E. indica*	-	-	1	5	8	1	15
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	3	2	1	5	11
Total No.	**1**	**3**	**5**	**7**	**0**	**0**	**34**	**58**	**72**	**33**	**197**

Table 1: Diversity and abundance of beetles (Coleoptera) in maize crop in district Layyah.

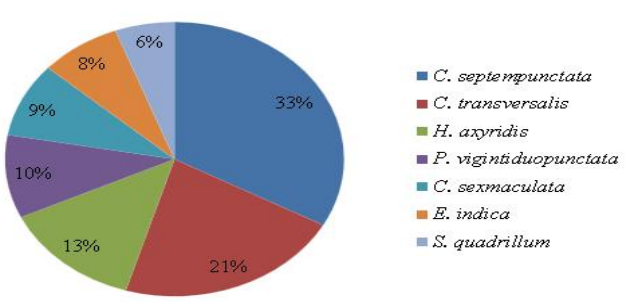

Figure 2: Relative abundance of beetles in maize crop in district Layyah.

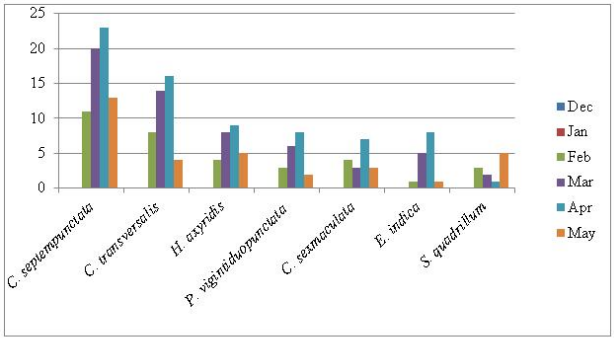

Figure 1: Diversity and abundance of Beetles (Coleoptera) in Maize crop in district L

Sr. No.	Family	Sub-Family	Genus	Species	Site 1: Maize						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	-	5.58	10.17	11.69	6.59	34.03
2				*C. transversalis*	-	-	4.06	7.12	8.12	2.03	21.33
3				*H. axyridis*	-	-	2.03	4.06	4.56	2.55	13.2
4			Psyllobora	*P. vigintiduopunctata*	-	-	1.52	3.04	4.06	1.01	9.63
5			Cheilomenes	*C. sexmaculata*	-	-	2.03	1.52	3.57	1.52	8.64
6		Epilachninae	Epilachna	*E. indica*	-	-	0.5	2.53	4.06	0.5	7.59
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	1.54	1.01	0.5	2.53	5.58
Total No.	**1**	**3**	**5**	**7**	**0**	**0**	**17.26**	**29.45**	**36.56**	**16.73**	**100**

Table 2: Relative abundance of beetles (Coleoptera) in maize crop of district Layyah.

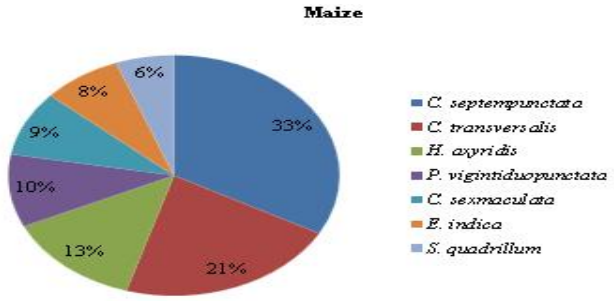

Figure 2: Relative abundance of beetles in maize crop in district Layyah.

Source	DF	SS	MS	F	p-value
Between	5	617.83	123.567	6.17	0.0003
Within	36	721.14	20.032		
Total	41	1338.98			

Table 3: Analysis of variance in maize crop regarding diversity and abundance of beetles in district Layyah.

P<0.001 = Highly significant; P>0.05 = Non-significant; P<0.05 = Significant.

The analysis of variance (ANOVA) to compare the mean difference of maize crop regarding the abundance of beetles. F-ratio is greater than 0.05, which means that there is less variation among the group mean. The p-value is less than 0.001, indicating that habitat is highly significant (Table 4.3).

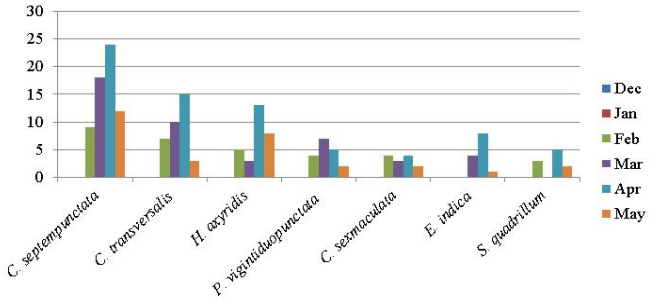

Figure 3: Diversity and abundance of Beetles (Coleoptera) in Mustard crop in district Layyah.

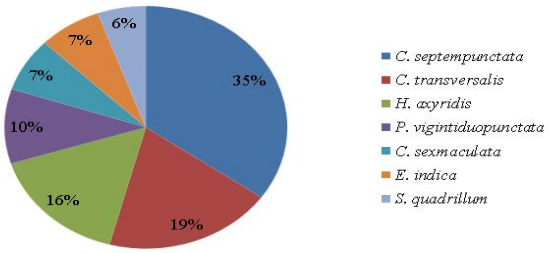

Figure 4: Relative abundance of Beetles (Coleoptera) in mustard crop in district Layyah.

The analysis of variance (ANOVA) to compare the mean difference of mustard crop regarding the abundance of beetles. F-ratio is greater than 0.05, which means that there is less variation among the group mean. The p- value is less than 0.001, indicating that habitat is highly significant (Table 4.6).

Sr. No.	Family	Sub-Family	Genus	Species	Site 2: Mustard						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	-	4.97	9.92	13.52	6.62	35.03
2				*C. transversalis*	-	-	3.86	5.52	8.28	1.65	19.31
3				*H. axyridis*	-	-	2.76	1.65	7.18	4.41	16
4			Psyllobora	*P. vigintiduopunctata*	-	-	2.2	3.86	2.76	1.1	9.92
5			Cheilomenes	*C. sexmaculata*	-	-	2.2	1.66	2.2	1.1	7.16
6		Epilachninae	Epilachna	*E. indica*	-	-	-	2.2	4.41	0.55	7.16
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	1.65	-	2.76	1.1	5.51
Total No.	**1**	**3**	**5**	7	**0**	**0**	**17.64**	**24.81**	**41.02**	**16.53**	**100**

Table 5: Relative abundance of beetles (Coleoptera) in mustard crops of district Layyah.

Sr. No.	Family	Sub-Family	Genus	Species	Site 3: wheat						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	4	11	22	14	-	51
2				*C. transversalis*	-	3	6	11	5	-	25
3				*H. axyridis*	-	-	2	9	8	-	19
4			Psyllobora	*P. vigintiduopunctata*	-	3	1	8	5	-	17
5			Cheilomenes	*C. sexmaculata*	-	2	3	5	4	-	14
6		Epilachninae	Epilachna	*E. indica*	-	-	1	3	7	-	11
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	3	5	1	-	9
Total No.	**1**	**3**	**5**	7	**0**	**12**	**27**	**63**	**44**	**0**	**146**

Table 6: Diversity and abundance of beetles (Coleoptera) in wheat crops of district Layyah.

Sr. No.	Family	Sub-Family	Genus	Species	Site 3: wheat						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	2.75	7.53	15.06	9.58	-	34.93
2				*C. transversalis*	-	2.05	4.12	7.55	3.44	-	17.13
3				*H. axyridis*	-	-	1.39	6.2	5.47	-	13.01
4			Psyllobora	*P. vigintiduopunctata*	-	2.03	0.68	5.47	3.44	-	11.65
5			Cheilomenes	*C. sexmaculata*	-	1.39	2.05	3.47	2.73	-	9.58
6		Epilachninae	Epilachna	*E. indica*	-	-	0.68	2.05	4.79	-	7.54
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	2.05	3.43	0.68	-	6.16
Total No.	**1**	**3**	**5**	**7**	**0**	**8.22**	**18.46**	**43.23**	**30.09**	**0**	**100**

Table 7: Relative abundance of beetles (Coleoptera) in wheat crops of district Layyah.

Source	DF	SS	MS	F	p-value
Between	5	460.762	92.1524	7.61	0.0001
Within	36	435.714	12.1032		
Total	41	896.476			

Table 8: Analysis of variance in wheat crop regarding diversity and abundance of beetles in district Layyah.

P<0.001 = Highly significant;P>0.05 = Non-significant; P<0.05 = Significant.

The analysis of variance (ANOVA) to compare the mean difference of wheat crop regarding the abundance of beetles. F-ratio is greater than 0.05, which means that there is less variation among the group mean. The p- value is less than 0.001, indicating that habitat is highly significant (Table 4.9).

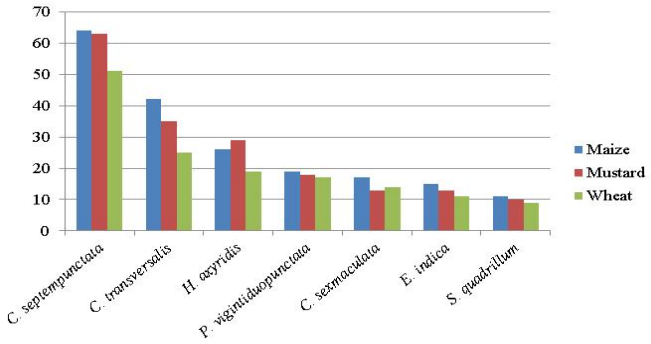

Figure 6: Total diversity and abundance of beetles (Coleoptera) in selected crops of district Layyah.

Source	DF	SS	MS	F	p-value
Between	1	4609.5	4609.5	30.7	0.0000
Within	40	6005	150.12		
Total	41	10614.5			

Table 10: Analysis of variance in all selected crop regarding diversity and abundance of beetles in district Layyah.

P<0.001 = Highly significant; P>0.05 = Non-significant; P<0.05 = Significant.

The analysis of variance (ANOVA) to compare the mean difference of three crops regarding the abundance of beetles. F-ratio is 30.7, which means that there is less variation among the group mean. The p- value is less than 0.001 which indicate highly significant result (Table 4.12).

Sr. No.	Family	Sub-Family	Genus	Species	Site 2: Mustard						Total
					Dec	Jan	Feb	Mar	Apr	May	
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	-	-	9	18	24	12	63
2				*C. transversalis*	-	-	7	10	15	3	35
3				*H. axyridis*	-	-	5	3	13	8	29
4			Psyllobora	*P. vigintiduopunctata*	-	-	4	7	5	2	18
5			Cheilomenes	*C. sexmaculata*	-	-	4	3	4	2	13
6		Epilachninae	Epilachna	*E. indica*	-	-	-	4	8	1	13
7		Scymniinae	Scymnus	*S. quadrillum*	-	-	3	-	5	2	10
Total No.	**1**	**3**	**5**	**7**	**0**	**0**	**32**	**45**	**74**	**30**	**181**

Table 11: Diversity and abundance of beetles (Coleoptera) in mustard crops of district Layyah.

Sr. No.	Family	Sub-Family	Genus	Species	Layyah			
					Maize	Mustard	Wheat	Total
1	Coccinellidae	Coccinellinae	Coccinella	*C. septempunctata*	12.78	12.02	9.73	34.53
2				*C. transversalis*	8.01	6.67	4.78	19.48
3				*H. axyridis*	4.98	5.56	3.62	14.12
4			Psyllobora	*P. vigintiduopunctata*	3.62	3.43	3.24	10.3
5			Cheilomenes	*C. sexmaculata*	3.24	2.48	2.68	8.41
6		Epilachninae	Epilachna	*E. indica*	2.88	2.48	2.09	7.44
7		Scymniinae	Scymnus	*S. quadrillum*	2.09	1.92	1.71	5.72
Total No.	**1**	**3**	**5**	**7**	**37.6**	**34.55**	**27.85**	**100**

Table 12: Relative abundance of beetles (Coleoptera) in selected crops in district Layyah.

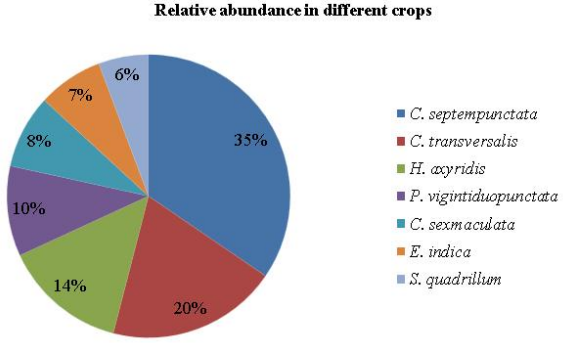

Figure 7: Relative abundance of Beetles (Coleoptera) in different crops in district Layyah.

Sites	S	N	Shannon Diversity index(H)	Diminance	Evenness
Maize	7	197	1.76	0.7999	0.903
Mustard	7	181	1.75	0.7970	0.897
Wheat	7	146	1.78	0.8051	0.913
Total	7	524	1.7633	0.8006	0.904

Table 13: Diversity index regarding different species of Beetles (Coleoptera) in different crops of district Layyah.

Shannon diversity index, dominance and evenness of all species in different crops given in (Table 4.13).It shows the total Shannon index (H) is 1.7633, Dominance is 0.8006 and Evenness is about 0.904.

DISCUSSION

Beetles diversity is highly important as it act as biological control agent. One of the most prominent groups of insect predators is coccinellid beetle, commonly referred to as ladybird beetle (Coleoptera: Coccinellidae). In all regions of the world, coccinellids play a vital role in natural control of insect pests such as mealy bugs, aphid, mites, thrips and scale insects (Khan *et al.*, 2007). According to (Bailon *et al.*, 2022), predatory coccinellids, additionally referred to as lady beetles, are among the most essential beneficial insects for agro ecosystems and forests. During sampling, 7 species were recorded from the selected crops of district Layyah. However, the abundance is much higher in maize (*Zea mays*) crop. During this exploration, seven species of Lady Bird beetle under five genera and three subfamilies (Subfamily Coccinellinae, Subfamily Epilachninae and Subfamily Scymniinae) were reported. Subfamily Epilachninae and Scymniinae represent only one genus. Another genus is Genus Coccinella which is represented by three species namely *C. septempunctata*, *C. transversalis* and *H. axyridis*. The species *C. septempuctata* is cosmopolitan and adopted to almost all habitats of agricultural crops, range lands and forests. *H. axyridis*, a predatory insect, contributes substantially to natural pest control by reducing the population's density.

Coccinella septempunctata was most abundant, accounting for 181 (34.53%) of total in selected crops. In these three crops, the maize crop has much more abundance which is about 197 (337.60%) of total specimens. On the other hand, wheat crop has less abundance of beetles accounting for 146 (27.85%). The findings of present study are in accordance with (Inayatullah *et al.*, 2005), reported that 204 speciemens of *C. septempunctata*in six month, which is more abundant in Azad Jammu and Kashmir.

The second most abundant specie of beetles is *C. transversalis*. The abundance of *S. quadrillum* is about 30 (5.72%) in all three crops. While in wheat crop, its abundance is less than other two crops which is about 9 (1.71%). The abundance of *C. sexmaculata*was accounting for about 44 (8.41%). As compared to other crop, this was more abundant in maize crop. Its abundance in maize crop was about 17 (3.24%). In mustard and wheat crop, it was about 2.48% and 2.68% respectively. (Sharma and Joshi, 2010) reported this species. The abundance of

H. axyridis and *P. vigintiduopunctata*were 14.12% and 10.30% respectively of overall sampling of six months. *P. vigintiduopunctata*was also most abundant in maize crop about 3.62%. The maximum abundance of beetles was founded in maize crop about 37.60%. The outcomes of present research showed an agreement with the (Ahmed *et al*., 2017) and (Bodla *et al*., 2021). (Bhatnagar, 2016), reported that among the coccinellids species, the most abundant specie is *C. sexmaculata*in maize crop.

To check the diversity, *Shannon-Wiener diversity Index was used.* Table 13 reveals the Shannon diversity index. In maize crop, the diversity index (H) =1.76, Dominance is 0.7999 and evenness was 0.903. In wheat crop, the diversity index (H) =1.78, Dominance is 0.8051 and evenness was 0.913. In mustard crop, the diversity index (H) =1.75, Dominance is 0.7970 and evenness was 0.897. The overall diversity index is (H) = 1.7633of all three crops.

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