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Research Article

BIODIVERSITY OF EARTHWORM (OLIGOCHAETA- ANNELIDA) IN KUSHINAGAR DISTRICT OF UTTAR PRADESH, INDIA

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ABSTRACT

Earthworm samples were collected in different fields at intervals of 25 days at 768 randomly selected locations (8 blocks x 4 villages x 4 fields x 6 sites) in the Kushinagar district of northeast Uttar Pradesh, India. A total of 18893 individuals were collected, representing 9 species, 7 genera, and 3 families. Among them, the most dominant species was *Lampito mauritii* (15.68%) followed by *Metaphire posthuma* (14.44%), *Eisenia fetida* (11.01%), *Perionyx excavatus* (10.22%), *Ramiella bishambari* (10.17%), *Eutyphoeus waltoni* (9.87%), *Eutyphoeus incommodus* (9.82%), *Amynthas morrisi* (9.69%), and *Dichogaster bolaui* (9.10%). In Kushinagar district, Megascolecidae and Acanthodrilidae accounted for the largest families, each representing 50.03 % and 38.96% distribution in the fields in different blocks. Lumbricidae family represents only 11.01% of distribution in the fields. Different ecological indices were calculated at all the locations and date intervals and its consequences were discussed.

Keywords: Earthworms, Fields, Earthworms biodiversity, Ecological Indices.

INTRODUCTION

India is a vast country with an extensive diversity of fauna and flora, along with tropical and subtropical climates. Because soil organisms enhance the soil fertility, soil faunal population is particularly important in many agroecosystems (Lee, 1985; Werner and Dindal, 1989). Earthworms, centipedes, millipedes, insects, mites and microbes are among the most common soil organisms. Earthworms are one of the first eucoelomate multicellular invertebrates, belonging to the phylum Annelida and class Oligochaeta. The maximum diversity of earthworms is found in Western Ghats and western coast plains of India (Narayanan *et al*., 2020). According to Blouin *et al*., (2013), earthworms are functionally extremely significant, varied, and wide-ranging, making them possibly valuable for managing biodiversity and ecological services. The earthworm's diversity has significance for research because they manage organic wastes, change the physical, chemical, and biological characteristics of soil, and enhance soil fertility and structure (Doan *et al*., 2013; Singh *et al*., 2016, Singh *et al*., 2021; Siddiqui, *et al*., 2022; Singh and Singh, 2023a, b, c; Fatima *et al*., 2023).

Michaelsen (1907), Stephenson (1914, 1923,1924), Gates (1940) and Julka (1976) were the pioneer workers in the field of earthworm diversity and density research in India, and numerous other Indian scientists have subsequently added to the earthworm diversity data. According to Julka and Paliwal (2005), 89% of the earthworm species and around 71% of the genera are endemic to India, one of the countries with the highest diversity of earthworms. Presently, there are approximately 7000 species recorded globally (Lavelle and Lapied, 2003), of which 3000 to 3500 are valid, and 505 species and subspecies belonging to 67 genera and 10 families have been reported from India (Julka, 2014; Ahmed and Julka, 2017; Kathireswari, 2016; Narayanan *et al*., 2017). Prakash (2017) had reported around 50 species of earthworms comprising 28 genera and 6 families in different areas of Uttar Pradesh. Six earthworm species were identified by Kumar and Singh (2013) in the several blocks of Gorakhpur district, an area in northeast Uttar Pradesh. The ecological or functional groups of earthworms are epigeic, endogeic, and anecic. Epigeic earthworms are strongly pigmented species that reside on the earth's surface. Endogeic species are usually

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light pigmented or non-pigmented, and they are capable of dig complex horizontal burrows in the soil. Anecic species generally large and have consistent pigmentation on their anterior and posterior ends. They feed on surface residue along with soil after pulling it into their vertical burrows. The aim of this study was to examine the diversity, number, and distribution of earthworms in different blocks of Kushinagar district.

MATERIALS AND METHODS

Site of Investigation

The present study was conducted in Kushinagar (Figure1) district of Uttar Pradesh, situated between 26°44'23.5680.37" N and 83°53' 13.0956" E**.** The present investigation was carried out for a period of four months July to October 2021. Sampling was conducted at 25 days interval in the following randomly selected 768 sites of 8 blocks of district Kushinagar: 1. Dhudhai (26° 47' 51.83" N, 84° 8' 8.57" E, 84 m asl), 2. Seorahi (*26° 43' 24.0960" N, 84° 14' 1.1400",* 80 m asl), 3. Sukrauli (*27° 32' 24" N, 83° 37' 48",* 80 m asl), 4. Ramkola (26° 54' 09" N, 83° 50' 31" E, 89 m asl), 5. Padrauna (26° 53' 44" n, 83° 58' 21" E, 89 m asl), 6. Hata (26° 44' 28.32" N, 83° 44' 42.94" E, 85 m asl), 7. Fazilnagar (*26° 40' 57.7380" N, 84° 2' 59.2152",* 78 m asl), 8. Kasia (*26° 44' 44" N, 83° 55' 10",* 89 m asl). Numerous rivers and riverines are heavily inundated these areas. The period from July to October when crops are harvested accounts for more than 80% of the entire yearly rainfall.

Sampling Methods

Earthworms were collected by hand sorting and digging from multiple fields, following the method recommended by Julka (1988) and Kumar and Singh (2013). Earthworm samples were taken from 8 blocks, 4 villages in each block, and 4 fields in each village. In each village 6 sites at different locations were selected randomly for the sampling. A hoe was used to dig up to 20 cm depth of soil (40 x 40 x 20 cm) and sorted it for the earthworms after a 60-100-minute interval. The specimens were anaesthetized in 70% alcohol for 20-60 seconds, washed with tap water and preserved in 5% formalin solution for further taxonomical study. The earthworms that were collected from each location were counted and identified following the keys provided by Gates (1959, 1972), Julka (1988), and Blakemore (2007a, 2012). After identification, all the specimens are held in the Zoology Laboratory of DDU University in Gorakhpur, Uttar Pradesh.

Data analysis for diversity indices

The Shannon-Wiener diversity index (H), which incorporates species richness and evenness and is suited to a simple statistical analysis, was used in this study because it is sensitive to changes in the abundance of rare species in a community. To compare the earthworm communities, Simpson index (λ), Margalef richness index (R), and Pielou's evenness index (E) were also calculated. The Simpson index (λ) is sensitive to changes in a community's most abundant species. Below are details of each index.

a. Shannon-Wiener Diversity Index (H)

The Shannon-Wiener diversity index has traditionally been employed to measure the impact of habitat quality, including the effects of polluted effluents. Because it excludes habitat-specific requirements for certain species, this index has recently fallen out of favor. The Shannon-Wiener index findings should be utilized with caution, but it still offers a useful learning tool for contrasting two different habitats. The species richness (the number of species within the community) and the species equitability (how evenly distributed the numbers of distinct species are) are two quantitative measurements that are combined in this measure. It is computed using the equation below: According to Solow (1993), $H = -\Sigma p_i \ln p_i$, where p_i = is the observed proportion of a certain species. Indicating that all of the sample's species are the same would be a value close to zero. This index's evident flaw that values in the middle are ambiguous means that use of it must be done with caution.

b. Simpson Index (λ) and Simpson's Diversity Index (D)

The biodiversity of a habitat is frequently measured using Simpson's diversity index, also called as species diversity index. It considers both the total number of species and the relative abundance of each species. The probability of the two randomly chosen individuals in the environment do not belong to the same species is represented by the Simpson index. It is calculated using the equation below: According to Solow (1993), $\lambda = \Sigma n_i (n_i - 1)/[N (N-1)]$, where n_i is the number of individuals in species i and $N = \Sigma n_i$. λ can have a value between 0 and 1. In this index, 0 denotes indefinite diversity and 1 denotes the absence of diversity. That is, the bigger the value of λ, the lower the diversity. The Simpson's index of diversity, $D = 1 - \lambda$, is frequently subtracted from 1 to solve this difficulty since it is neither intuitive nor logical. Similar to the previous index, the value of this one ranges from 0 to 1, but now the higher the value, the greater the sample diversity. This is a better explanation. The index in this instance indicates the likelihood that two individuals picked at random from a sample will be of different species.

c. Margalef Richness Index (R)

This index offers a measure of species richness that is generally sample size normalized without utilizing more complex rarefaction procedures. Using the equation below, it is calculated: R = N -1/In (n), where N is the total number of species in a community and n is the total number of individuals observed. Since N and R are simple and easy to calculate, but sensitive to sample size (Magurran, 1988), the Margalef's index of species richness reduces the effect of sample size bias (Odum, 1971). Studies pertaining to earthworms have used this index successfully.

d. Pielou's Evenness Index (E)

Pielou's evenness is a diversity index, a biodiversity measure that measures how numerically equal the community is. The Pielou's Evenness Index may be utilized to evaluate how even a community is: E = H/Hmax, where Hmax = - S [1/S.ln S] = ln S, where S is the total number of species, and H is the number determined from the Shannon diversity index. Therefore, E = H/ln S, where E is embarrassed between 0 and 1. The less variation in communities between the species, the higher the value of E.

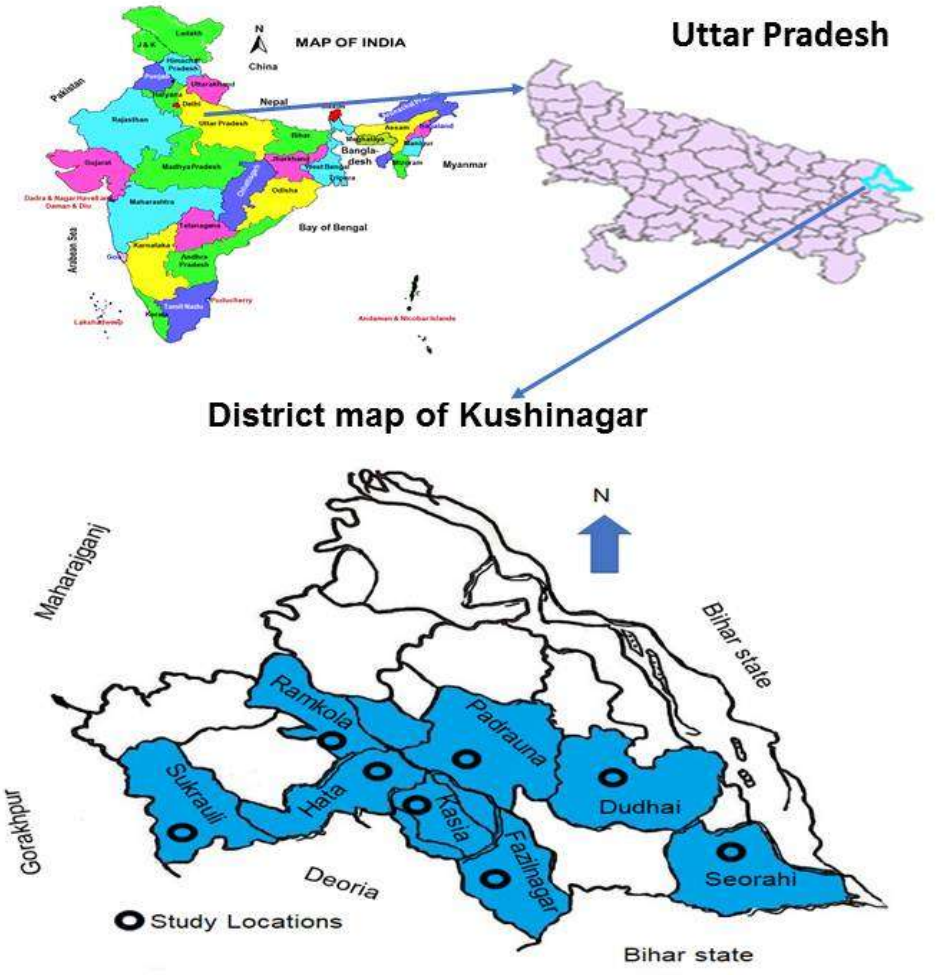

Figure 1. Map of the study site (1-8 blocks showing the collection places) in district Kushinagar of northeast U.P.

RESULTS AND DISCUSSION

The earthworms list collected from the fields in several blocks of Kushinagar district northeast of Uttar Pradesh and their relative abundance is displayed in Table 1 while the earthworm distributions at different days (data of all locations were pooled) and at different locations (data of days of sampling were pooled) are shown in Tables 2 and 3, respectively. A total of 18893 individuals belonging to 9 species, 7 genera and 3 families were collected during the study period, July to October 2021 (Table 1; Figure 2). Of the total 9 species, *Lampito mauritii* were most dominant (Accounted for 15.68% of the total species) followed by *Metaphire posthuma* (14.44%), *Eisenia fetida* (11.01%), *Perionyx excavates* (10.22%), *Ramiellabishambari* (10.17%), *Eutyphoeus waltoni* (9.87%), *Eutyphoeusin commodus* (9.82%), *Amynthas morrisi* (9.69%), and *Dichogaster bolaui* (9.10%). Megascolecidae and Acanthodrilidae were the largest families, each representing 50.03 % and 38.96% distribution in the fields in different blocks of Kushinagar district. Lumbricidae family represents only 11.01 % of the distribution in fields. In the overall data the most dominant species of earthworm were *Lampito mauritii* and *Metaphire posthuma*.

Table 1. List of earthworms collected from the fields in different blocks of Kushinagar district of northeast Uttar Pradesh and their Relative Abundance.

Family	Species	No.of Individuals	Diversity in percentage
Acanthodrilidae	*Ramiella bishambari*	1921	10.17
Acanthodrilidae	*Eutyphoeus incommodus*	1855	9.82
Acanthodrilidae	*Eutyphoeus waltoni*	1865	9.87
Acanthodrilidae	*Dichogaster bolaui*	1719	9.10
Lumbricidae	*Eisenia fetida*	2080	11.01
Megascolecidae	*Lampito mauritii*	2963	15.68
Megascolecidae	*Metaphire posthuma*	2729	14.44
Megascolecidae	*Amynthas morrisi*	1831	9.69
Megascolecidae	*Perionyx excavatus*	1930	10.22
Total		18893	

Table 2. Number of earthworms collected from several fields in different blocks of Kushinagar district in northeast Uttar Pradesh at different days (data of all locations pooled).

Earthworm's species	Days of sampling				Total
	Day 1	Day 2	Day 3	Day 4	
Lampito mauritii	800	773	708	682	2963
Metaphire posthuma	718	723	656	632	2729
Eisenia fetida	561	538	487	494	2080
Amynthas morrisi	526	503	434	368	1831
Ramiellabishambari	521	498	450	452	1921
Eutyphoeusin commodus	500	513	428	414	1855
Eutyphoeus waltoni	527	516	401	421	1865
Perionyx excavatus	512	523	449	446	1930
Dichogaster bolaui	468	474	409	368	1719
Total	5133	5061	4422	4277	18893

Table 2 demonstrates that the variation in the distribution of different species of earthworms significantly differ between different collection dates ($F = 71.96$, $P < 0.001$, $n_1 = 3$, $n_2 = 24$) as well as species ($F = 162.88$, $P < 0.001$, $n_1 = 8$, $n_2 = 24$).

Summary of computation of 2-way analysis of variance

Source of variation	Sum of squares	D.O.F.	Variance	F value	Significance
Variation between dates	63576.52	3	21192.17	71.96	$P < 0.001$
Variation between species	383767.72	8	47970.96	162.88	$P < 0.001$
Residual variation	7068.27	24	294.51		
Total variation	454412.51	35	12983.21		

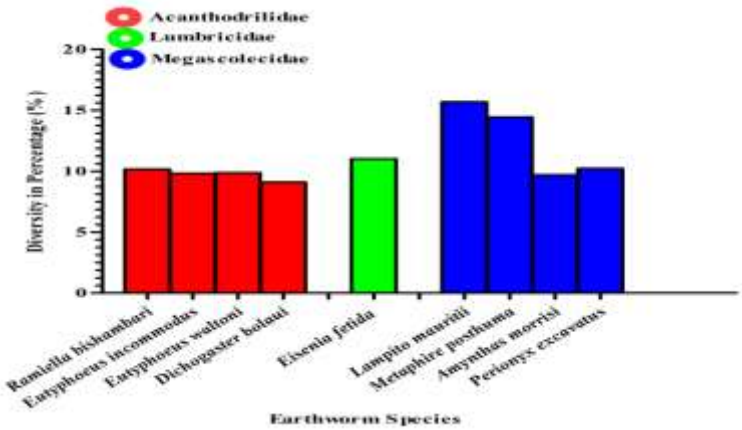

Figure 2. Percentage diversity of acanthodrilid, lumbricid and megascolecid earthworms in several areas in Kushinagar district at various locations.

Table 3. Number of Individuals of collected earthworms from different fields in different blocks of Kushinagar district of northeast Uttar Pradesh at different locations (1. Dudhai, 2. Seorahi, 3. Sukrauli, 4. Ramkola, 5. Padrauna, 6. Hata, 7. Fazilnagar, 8. Kasia (data of days of sampling in fields pooled)**.**

Species	Locations								Total
	1	2	3	4	5	6	7	8	
Lampito mauritii	385	379	388	384	356	359	359	353	2963
Metaphire posthuma	364	352	360	356	321	329	331	316	2729
Eisenia fetida	273	274	282	262	265	240	219	265	2080
Amynthas morrisi	226	242	235	226	232	232	209	229	1831
Ramiella bishambari	264	260	240	227	235	241	214	240	1921
Eutyphoeusin commodus	243	229	221	221	249	226	227	239	1855
Eutyphoeus waltoni	242	217	229	216	251	264	201	245	1865
Perionyx excavatus	252	247	253	246	243	242	210	237	1930
Dichogaster bolaui	197	198	211	189	228	232	232	232	1719
Total	2446	2398	2419	2327	2380	2365	2202	2356	18893

Summary of computation of 2-way analysis of variance

Source of variation	Sum of squares	D.O.F.	Variance	F value	Significance
Variation between locations	4310.43	7	615.78	2.71	Not significant
Variation between species	191814.69	8	23976.84	105.46	$P < 0.001$
Residual variation	12732.19	56	227.36		
Total variation	208857.32	71	2941.65		

Table 3 shows that the variation in the distribution of different earthworm's species differ significantly between different species ($F = 105.46$, $P < 0.001$, $n_1 = 8$, $n_2 = 56$) but not between locations ($F = 2.71$, $P > 0.05$, $n_1 = 7$, $n_2 = 56$). It implies that the distribution of all species of earthworms is homogenous at all places of sampling (Figure 3).

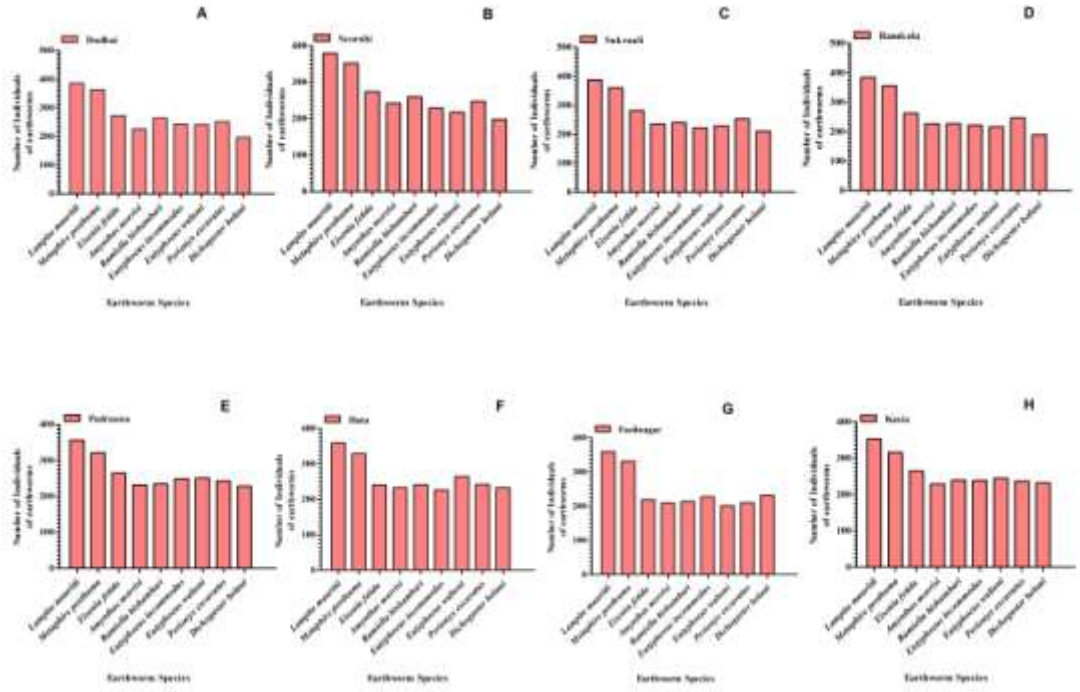

Figure 3. Earthworm's species collected from the fields in different blocks of Kushinagar district of northeast Uttar Pradesh.

From the data displayed in Tables 1 and 2, 4 ecological indices, viz., Shannon-Weiner diversity index (H), Simpson diversity index (D), Margalef richness index (R) and Pielou evenness index (E) were calculated to observe the diversity, richness and evenness of the species of earthworm in the target area in the fields of different blocks of Kushinagar district in northeast Uttar Pradesh (Tables 4 and 5). Shannon-Wiener index provides a good learning tool for comparing two distinct habitats. It combines two quantifiable measures: the species richness (the number of species within the community) and the species equitability (how even are the numbers of individual species). A value near 0 indicates no diversity in the species in the samples while a value near 4.6 indicates that the number of individuals is evenly distributed between all the species. Table 4 demonstrates the values of Shannon-Weiner diversity indices of earthworm collected at different 768 locations (8 blocks x 4 villages x 4 fields x 6 sites) at different sampling days. Almost all values ranged between 2.174 to 2.182 which demonstrated that the distribution of every species in the sample is almost the same, and indicated that the number of individuals is somewhat evenly distributed between all the species. Results showed a non-significant variation in the values of Shannon-Weiner diversity indices caused by either due to date of sampling or locations.

Simpson's diversity index (D) is calculated by subtracting Simpson index (λ) from 1, i.e., D = 1-λ, and is usually used to quantify the biodiversity of a habitat. It includes both the total number of species and the relative abundance of each species. D can have a value between 0 and 1. With this index, 1 denotes infinite variety and 0 denotes no diversity, meaning that the higher the value, greater the sample diversity. In this instance, the index denotes the likelihood that two individuals selected at random from a sample will be of different species. Table 5 displays the values of Simpson diversity indices for all locations and all days of sampling. Its values ranged between 0.883 to 0.887. The 2-way ANOVA did not yield any significant variation in the indices when the samples were taken from different locations or different days of samplings. It showed that the diversity of earthworm species with reference to their abundance is almost the same in the field of different blocks of Kushinagar district of northeast Uttar Pradesh in each sample, the probability that two individuals selected randomly will belong to different species is very high.

Margalef richness index (R) provides a measure of species richness that is roughly normalized for sample size without using more complex rarefaction techniques. Table 5 displays the variations in the R in the samples taken from different sites and sampling days. All of the data ranged between 1.03 to 1.04. The data analysis showed that the impact of different locations and collection days on R was insignificant. It proved that in northeast Uttar Pradesh, the earthworm's species richness did not vary by both collecting sites and time of collection in northeast Uttar Pradesh.

Table 4. Parameters and Indices of earthworm community observed in different fields and days of sampling (data of all locations pooled) (N: Number of Individuals, S: Number of Species, H: Shannon Index, λ: Simpson Index, D: Simpson's Index of Diversity, R: Margalef Richness Index, E: Pielou's Evenness Index.

Days of sampling	N	S	H	λ	D	R	E
1st day	5133	9	2.181	0.115	0.885	0.936	0.993
2nd day	5061	9	2.182	0.115	0.885	0.937	0.993
3rd day	4422	9	2.175	0.116	0.884	0.953	0.990
4th day	4277	9	2.174	0.116	0.884	0.956	0.990
Total/Mean	18893	9	2.178	0.115	0.884	0.945	0.991

Table 5. Parameters and Indices of earthworm community observed in different field at different locations in Kushinagar district of northeast Uttar Pradesh (data of days of sampling pooled). (N: Number of Individuals, S: Number of Species, H: Shannon Index, λ: Simpson Index, D: Simpson's Index of Diversity, R: Margalef Richness Index, E: Pielou's Evenness Index**.**

Locations	N	S	H	λ	D	R	E
1	2447	9	2.175	0.116	0.884	1.03	0.990
2	2398	9	2.175	0.116	0.884	1.03	0.990
3	2419	9	2.174	0.116	0.884	1.03	0.989
4	2327	9	2.170	0.117	0.883	1.03	0.987
5	2380	9	2.185	0.114	0.886	1.03	0.995
6	2365	9	2.183	0.114	0.886	1.03	0.994
7	2202	9	2.174	0.116	0.884	1.04	0.989
8	2356	9	2.181	0.113	0.887	1.03	0.992
Total	18893	9	2.177	0.115	0.885	1.03	0.991

A measure of biodiversity known as the Pileou evenness index (E) assesses how numerically equal the community is. It had a value range of 0 to 1. Higher E values indicate higher evenness or less variety in the communities of between the species. Table 5 displays the variations in the E in the samples taken from different sites and date of sampling which ranged between 0.989 to 0.994. Two-way ANOVA showed insignificant effect of locations and did not yield any significant effect of date of sampling. It demonstrated that the evenness of earthworm communities not varied with collection sites of different blocks of Kushinagar district in northeast Uttar Pradesh. In the current survey three earthworm families and nine earthworm species recorded from different fields of Kushinagar district. According to data analysis, the Padrauna block had the most species diversity measured by the Shannon-Wiener index and the Simpsons index (H' = 2.184 and D = 0.886), while the Ramkola block had the lowest diversity (H' = 2.170 and D = 0.883). Higher values of these indices represent more species diversity, while lower values represent less diversity. The variety of earthworms ranged from 1 to 15 species, with the majority of earthworm communities having between 3-6 species, according to Edwards and Bohlen (1996). According to Singh (1997), 7 to 11 species were discovered in cultivated, non-cultivated, grassland, garden, and sewage soils. According to Fragoso *et al*. (1999), the number of species present in a particular earthworm community, which can range from 3 to 17 in tropical and temperate habitats, is the simplest approach to assess species diversity. In tropical rainforests, 4 to 14 species were often found. According to Goswami (2015) observations earthworm communities in the ecosystems of the Indian Botanic Garden, Howrah, India, varied from 6 to 10 species and had an identical variety. In this regard, the various fields in the Kushinagar district in northeast Uttar Pradesh are represented, and the earthworm communities there, which contain nine earthworm species, show similar diversity.

CONCLUSION

From the current study, we observed several earthworm species in fields of various blocks in the Kushinagar district of the northeast Uttar Pradesh, including *Lampito mauritii*, *Metaphire posthuma*, *Eisenia fetida*, *Amynthas morrisi*, *Ramiella bishambari*, *Eutyphoeus incommodus*, *Eutyphoeus waltoni*, *Perionyx excavates* and *Dichogaster bolaui.* The distribution of all species of earthworms is homogenous at all places of sampling.

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