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Biodiversity of Earthworm (Oligochaeta-Annelida) in Gorakhpur District of Northeast Uttar Pradesh, India

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Abstract: Earthworm samples were taken from different fields at 25 days interval at randomly selected 768 (8 blocks x 4 villages x 4 fields x 6 sites) locations in district Gorakhpur of northeast Uttar Pradesh, India. A total of 19613 individuals belonging to 9 species, 6 genera and 2 families were collected. The most dominant species was *Lampito mauritii* (15.19%) followed by *Metaphire posthuma* (14.25%), *Perionyx excavatus* (11.45%), *Eutyphoeus nicholsoni* (10.61%), *Eutyphoeus waltoni* (10.46%), *Dichogaster bolaui* (10.18%), *Eutyphoeus orientalis* (10.12%), *Eutyphoeus incommodus* (9.65%), and *Amynthas morrisi* (8.10%). Both the families, Acanthodrilidae and Megascolecidae accounted almost equal distribution in the fields in different blocks of Gorakhpur, each representing 51% and 49%, respectively. Different ecological indices were calculated at all the locations at different intervals and its consequences are discussed.

Keywords: Fields, Earthworms, Ecological indices, Biodiversity, Shannon-Weiner Diversity Indices, Simpson Diversity Index, Margalef Richness Index

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Introduction

Earthworms (Annelida: Oligochaeta) have long been accepted as the farmer's friend, natural ploughmen, and earth's intestines. The earthworms, well-known terrestrial segmented worms, are the first eucoelomate multicellular invertebrates (Kale and Karmegam 2010). More than 7,000 species have been recorded to date (Lavelle and Lapied, 2003). In India, 505 species and subspecies of earthworms comprising 67 genera and 10 families have been discovered (Kathireswari, 2016; Ahmed and Julka, 2017). The Western Ghats and western coast plains of India have the highest diversity of earthworms (Narayanan *et al*., 2020). Earthworm diversity is crucial to research because various ecological categories of earthworms perform critical roles in

soil processes, ecosystem functions and managing organic wastes (Singh *et al*., 2021; Singh and Singh, 2023a, b). Julka (1988) found that native earthworm species account for approximately 89% of overall earthworm diversity in India. They are commonly referred to as ecosystem engineers because of their important role in modifying the physical, chemical, and biological aspects of soil hence, enhancing soil fertility and structure (Doan *et al*., 2013; Singh *et al*., 2016, Siddiqui *et al.,* 2022; Singh and Singh, 2023a, b, c; Fatima *et al*., 2023). Earthworms boost microbial activity, mix and consolidate soil, increase water penetration rate, soil moisture content, and water retention capacity, among other things. They also improve breakdown of litter, soil organic matter dynamics, nutrients cycle, plant development, and prevent several soil-borne diseases (Chandran *et al*., 2012). Earthworms are functionally highly important and varied, which makes them potentially valuable for biodiversity and ecosystem service management (Blouin *et al*., 2013). Prakash (2017) has described 50 species of earthworms belonging to 28 genera and 6 families from different regions of the Uttar Pradesh. Kumar and Singh (2013) studied earthworm diversity in the different blocks of Gorakhpur, a northeast district of Uttar Pradesh and reported 6 earthworm species. Earthworms are divided into three ecological or functional groups based on their dietary habits on different food resources: epigeic, endogeic, and anecic (Bouche, 1972). Epigeic earthworms are those that reside on the earth's surface and are highly coloured. Endogeic species generally include light coloured or non-pigmented animals that dig complicated horizontal tunnels in the soil. Anecic species are large and have homogeneous pigmentation at their anterior and posterior ends. They feed on surface waste as well as dirt after pushing it into their vertical burrows.

The primary goal of the present research study was to collect existing information on earthworm biodiversity in the Gorakhpur district (Northeast Uttar Pradesh, India).

Materials and Methods

Site of Investigation:

Present study was conducted at Gorakhpur district (Fig. 1) of Uttar Pradesh, situated between 26°45'49.37" N and 83°24' 14.8" E. The total geographical area of the district is about 3483 Sq. km which constitutes about 1.44 % of the total area of the state. The present investigation was carried out for a period of four months. Sampling was conducted at the following randomly selected 768 sites of eight blocks of Gorkhpur: (1) Caimpierganj (27° 05' 60" N, 83° 16' 60" E, 84 m asl), (2) Bhathat (26° 89' 80" N, 83° 48' 86" E, 85 m asl), (3) Bansgaon (26° 32' 56.51" N, 83° 20' 48.43" E, 82.37 m asl), (4) Khajani (26° 66' 52" N, 83° 24' 92" E, 83 m asl), (5) Piprauli (26° 73' 07" N, 83° 24' 81" E, 83 m asl), (6) Sahjanwa (26° 43' 59" N, 83° 20' 03" E, 80 m asl), (7) Uruwa (26° 44' 845" N, 83° 28'49 " E, 80 m asl), and (8) Pali (26° 59' 238" N, 83° 31' 523" E, 80 m asl). Gorakhpur district is well inundated by several rivers and riverine. More than 80% of the total annual rainfall is received during the crop season, July to October.

Sampling Methods:

Earthworms were collected by digging and hand sorting method from different fields following the technique as described by Julka (1988) and Kumar and Singh (2013). Sampling of earthworm was done in 8 blocks, 4 villages and 4 fields. In each village in each field, 6 sites at different locations were selected for sampling. To exploit the maximum expulsion of earthworms, different solutions such as aqueous solution of potassium permagnet, formalin and simple water were sprayed on the soil. After an interval ranging from 60-100 min, soil (40 x 40 cm) was dug by a hoe and up to 20 cm and was sorted out for the earthworms. After anesthetizing in 70% alcohol for 20-60 sec, the specimens were washed with tap water and kept in 5% formalin solution for further studies. From each place, the collected earthworms were counted and identified. The preserved specimens were identified following the

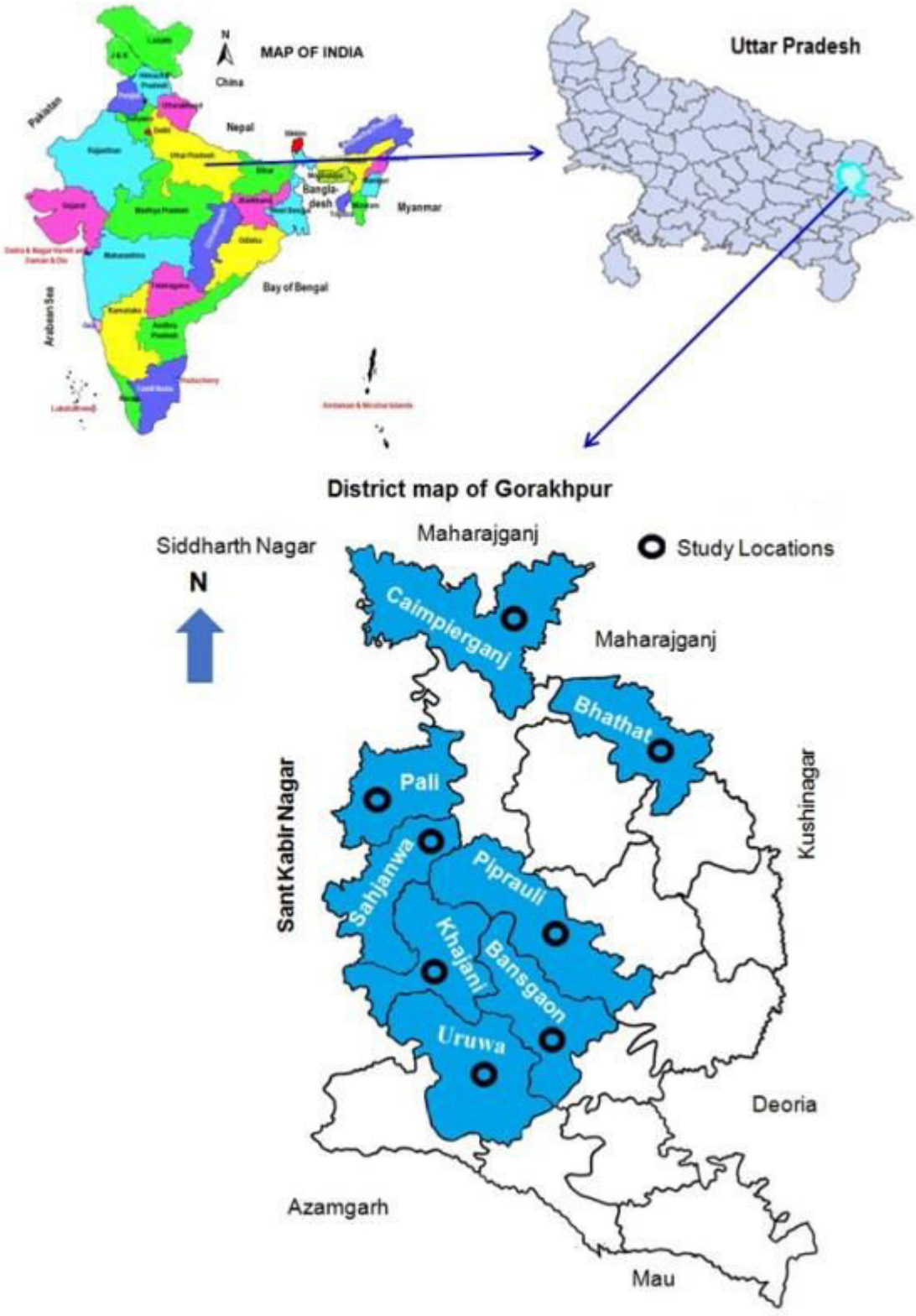

Fig. 1: Map of the study site (1-8 blocks showing the places of collection) in Gorakhpur district of northeast Uttar Pradesh.

monographs by Gates (1959, 1972); Julka (1988) and Blakemore (2007, 2012). After identification, all specimens are stored in the laboratory of Zoology in D.D.U. Gorakhpur University, Gorakhpur, Uttar Pradesh, India.

Data analysis for diversity indices:

This study employed the Shannon-Wiener diversity index (H), which integrates species richness and evenness and is easily adaptable to a statistical analysis. It is sensitive to changes in the number of typical species in a population. To further aid in comparison, the Simpson index (λ), which is sensitive to changes in a community's most abundant species, the Margalef richness index (R), and Pielou's evenness index (E) of earthworm communities was also calculated. Below are listed the traits of each index:

(a) *Shannon-Wiener Diversity Index* (*H*):

The Shannon-Wiener diversity index has traditionally been adopted to assess the impact of habitat quality, including the effects of polluted effluents. Because it excludes habitat-specific requirements for certain species, this index has recently lost popularity. The Shannon-Wiener index is a useful learning tool for contrasting two different habitats, despite the necessity to utilize the data with caution. 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 calculated using the equation (Solow, 1993):

$$H = -\Sigma p_i \ln p_i$$

where pi is the observed proportion of a certain species. If the value is close to 0, all of the species in the sample are the same. Because of this index's evident flaw values in the centre are ambiguous care must be exercised while utilizing it.

(b) *Simpson Index and Simpson's Diversity Index*:

The biodiversity of a habitat is frequently measured using Simpson's diversity index (D), also called the species diversity index. It considers both the total number of species and the relative abundance of each species. The probability that two randomly chosen individuals in the environment do not belong to the same species is represented by the Simpson index (λ). It is calculated using the equation (Solow, 1993) below:

$$\lambda = \Sigma n_i (n_i - 1)/[N (N-1)]$$

where n_i = the number of individuals of species i, and $N = \Sigma n_i$. λ can have a value between 0 and 1. In this index, 0 denotes infinite diversity and 1 denotes the absence of diversity. That is, the variety decreases as λ's value increases. In order to solve this problem because it is neither intuitive nor logical. λ is sometimes subtracted from 1 to get Simpson's index of diversity D = 1 - λ. This index's value similarly runs from 0 to 1, but now the higher the number, the more diverse the sample. This is more sense. In this instance, the index denotes the probability that two individuals drawn 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 normalised without utilising more intricate rarefaction techniques. 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 influence 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 indicator 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

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

Family	Species	No. of Individuals	Diversity in percentage
Acanthodrilidae	*Dichogaster bolaui*	1996	10.18
Acanthodrilidae	*Eutyphoeus incommodus*	1892	9.65
Acanthodrilidae	*Eutyphoeus nicholsoni*	2081	10.61
Acanthodrilidae	*Eutyphoeus orientalis*	1984	10.12
Acanthodrilidae	*Eutyphoeus waltoni*	2052	10.46
Megascolecidae	*Amynthas morrisi*	1588	8.10
Megascolecidae	*Lampito mauritii*	2979	15.19
Megascolecidae	*Metaphire posthuma*	2795	14.25
Megascolecidae	*Perionyx excavatus*	2246	11.45
Total		19613	

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

Species of earthworms	Days of sampling				Total
	Day 1	Day 2	Day 3	Day 4	
Lampito mauritii	825	798	689	667	2979
Metaphire posthuma	739	740	684	632	2795
Eutyphoeus nicholsoni	547	559	507	468	2081
Eutyphoeus incommodus	559	499	426	408	1892
Eutyphoeus waltoni	601	509	516	426	2052
Perionyx excavatus	609	622	516	499	2246
Eutyphoeus orientalis	539	526	474	445	1984
Dichogaster bolaui	557	538	474	427	1996
Amynthas morrisi	488	424	337	339	1588
Total	5464	5215	4623	4311	19613

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	93436.53	3	31145.51	60.53	P < 0.001
Variation between species	388210.39	8	48526.30	94.31	P < 0.001
Residual variation	12348.72	24	514.53		
Total variation	493995.64	35	14114.16		

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 lower the variance in communities across species, the greater the value of E.

Results

Species Composition:

The list of earthworms collected from the fields in different blocks of Gorakhpur district of northeast Uttar Pradesh and their relative abundance is displayed in Table 1 while the distributions of the earthworms 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 19613 individuals belonging to 9 species, 6 genera and 2 families were collected during the study period, July to October 2021 (Table 1, Fig. 2). Of the total 9 species, *Lampito mauritii* species was most dominant (accounted for 15.19% of the total species), followed by *Metaphire posthuma*

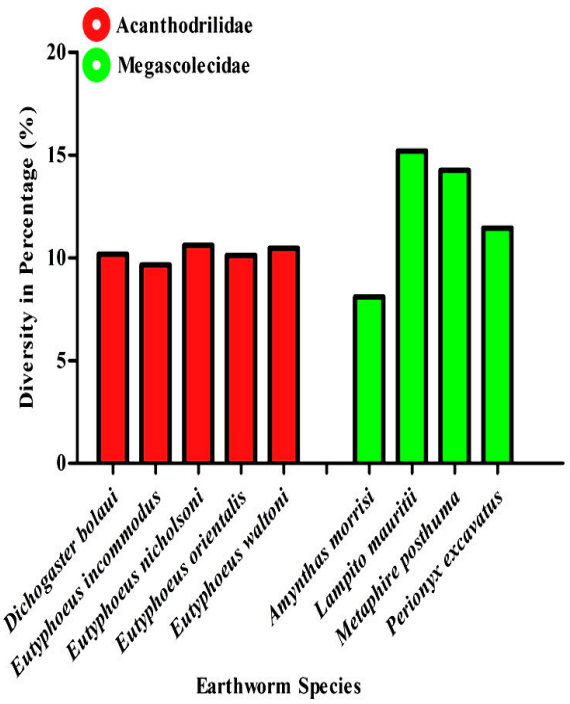

Fig. 2: Percentage diversity of acanthodrilid and megascolecid earthworms in several areas in Gorakhpur district at various locations.

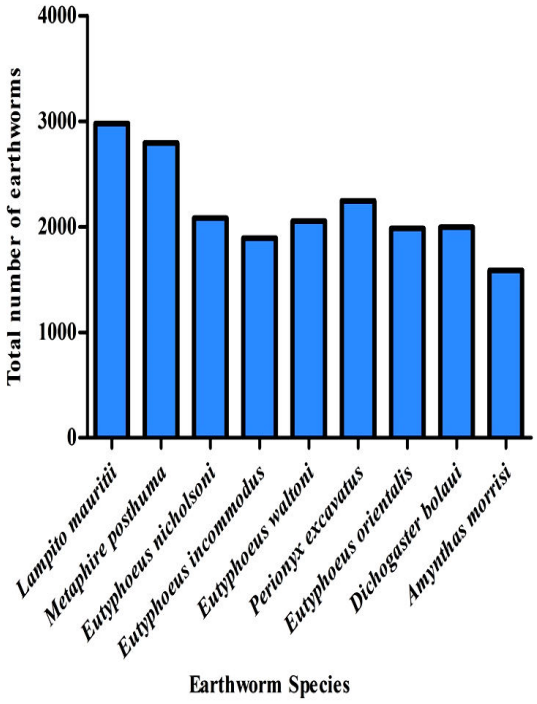

Fig. 3: The number of earthworms counted from various fields in several blocks of northeast Uttar Pradesh, Gorakhpur district.

Table 3: Number of individuals of earthworms collected from different fields from Northeast Uttar Pradesh at different locations (1. Campierganj, 2. Bhathat, 3. Bansgaon, 4. Khajani, 5. Piprauli, 6. Sahjanwa, 7. Uruwa and 8. Pali) (data of days of sampling in fields pooled)

Species	**Locations**								**Total**
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	
Lampito mauritii	372	368	369	376	359	373	363	399	2979
Metaphire posthuma	332	345	362	366	329	361	336	364	2795
Eutyphoeus nicholsoni	261	247	254	257	270	260	275	257	2081
Eutyphoeus incommodus	237	240	236	227	241	232	246	233	1892
Eutyphoeus waltoni	257	262	266	255	255	255	259	243	2052
Perionyx excavatus	307	314	297	308	280	276	240	224	2246
Eutyphoeus orientalis	243	257	255	257	251	250	248	223	1984
Dichogaster bolaui	255	253	254	241	255	247	251	240	1996
Amynthas morrisi	189	196	185	185	200	193	209	231	1588
Total	2453	2482	2478	2472	2440	2447	2427	2414	19613

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	468.21	7	156.07	0.27	Not significant
Variation between species	194105.19	8	24263.15	42.00	$P < 0.001$
Residual variation	13865.92	56	577.75		
Total variation	208439.32	71	5955.41		

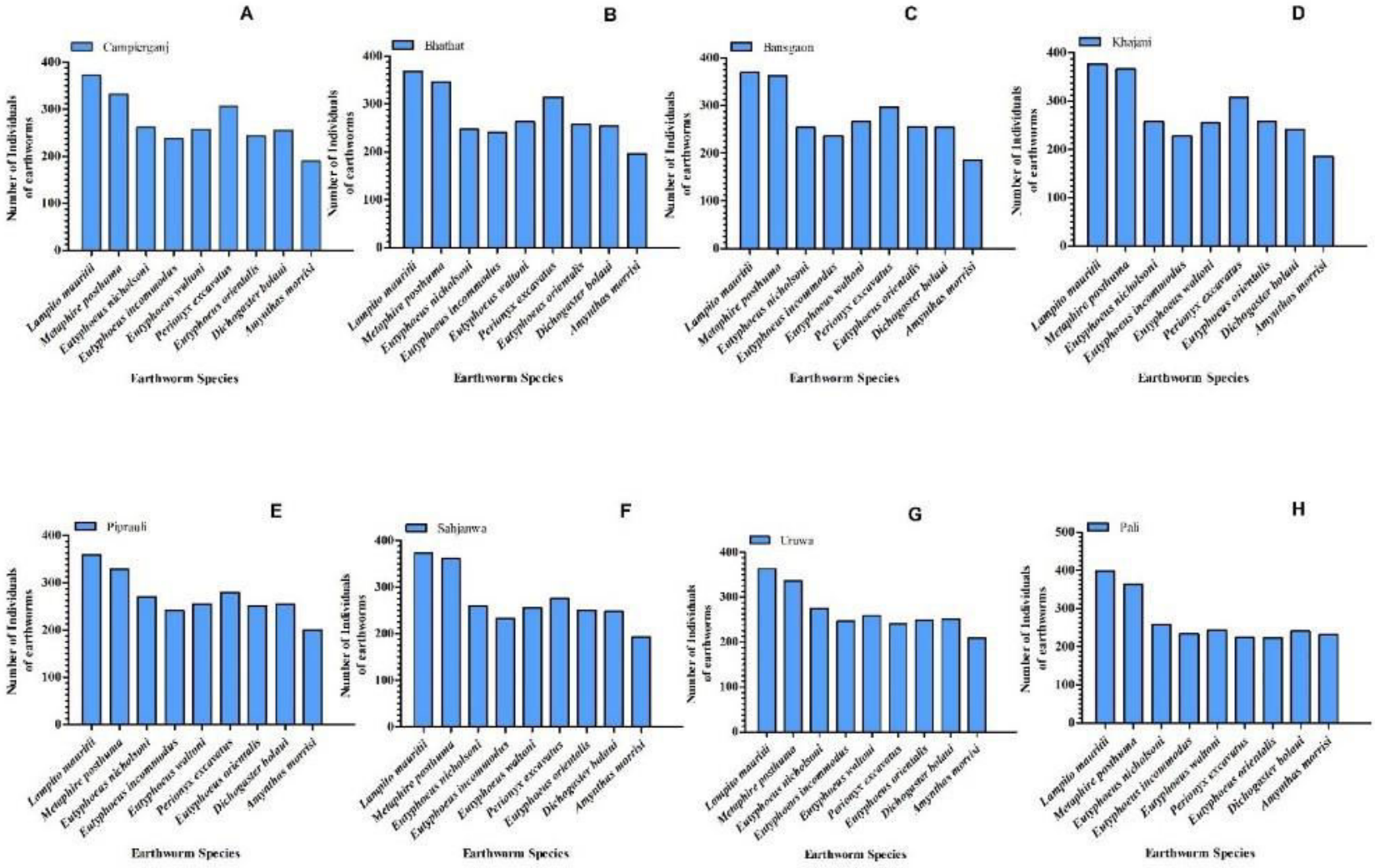

Fig. 4: Earthworms species collected from the fields in different blocks of Gorakhpur district of northeast Uttar Pradesh.

Table 4: Parameters and Indices of earthworm community observed in fields different 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
Day 1	5464	**9**	2.184	0.114	0.886	0.930	0.994
Day 2	5215	**9**	2.124	0.115	0.885	0.935	0.967
Day 3	4623	**9**	1.982	0.116	0.884	0.948	0.902
Day 4	4311	**9**	1.904	0.116	0.884	0.956	0.867
Total/Mean	**19613**	**9**	**2.049**	**0.115**	**0.885**	**0.942**	**0.933**

(14.25%), *Perionyx excavatus* (11.45%), *Eutyphoeus nicholsoni* (10.61%), *Eutyphoeus waltoni* (10.46%), *Dichogaster bolaui* (10.18%), *Eutyphoeus orientalis* (10.12%), *Eutyphoeus incommodus* (9.65%), and *Amynthas morrisi* (8.10%). The most dominant species of overall data were *Lampito mauritii* and *Metaphire posthuma*.

Table 2 demonstrates that the variation in the distribution of different species of earthworms differ significantly between different dates of collection ($F = 60.53$, $P < 0.001$, $n_1 = 3$, $n_2 = 24$) as well as species ($F = 94.31$, $P < 0.001$, $n_1 = 8$, $n_2 = 24$) (Fig. 3).

Table 3 shows that the variation in the distribution of different species of earthworms differ significantly between different species ($F = 42.00$, $P < 0.001$, $n_1 = 8$, $n_2 = 24$) but not between locations ($F = 0.27$, $P > 0.05$, $n_1 = 7$, $n_2 = 24$). It implies that the distribution of all species of earthworms is homogenous at all places of sampling (Fig. 4).

Diversity, Evenness and Richness Indices:

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 Gorakhpur (Tables 4, 5).

(a) *Shannon-Wiener Diversity Index* (*H*):

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 zero indicate no diversity in the species in the samples while a value near 4.6 indicate 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 days of sampling. Almost all values ranged between 1.90 to 2.18 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.

(b) *Simpson Diversity Index* (*D*):

Simpson's diversity index (D) is calculated by substracting Simpson index (λ) from 1, i.e., $D = 1 - \lambda$, and is usually used to quantify the biodiversity of a habitat. It takes into account the number of species present, as well as the relative abundance of each species. The value of D ranges between 0 and 1. With this index, 1 represents infinite diversity and 0, no diversity, i.e., the greater the value, the greater the sample diversity. In this case, the index represents the probability that two

Table 5: Parameters and Indices of earthworm community observed in different field at different locations in 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	2453	9	2.179	0.115	0.885	1.025	0.992
2	2482	9	2.180	0.115	0.885	1.023	0.992
3	2478	9	2.177	0.115	0.885	1.024	0.991
4	2472	9	2.174	0.116	0.884	1.024	0.989
5	2440	9	2.184	0.114	0.886	1.026	0.994
6	2447	9	2.177	0.115	0.885	1.025	0.991
7	2427	9	2.183	0.114	0.886	1.026	0.994
8	2414	9	2.173	0.117	0.883	1.027	0.989
Total/Mean	**19613**	**9**	**2.178**	**0.115**	**0.885**	**1.025**	**0.992**

individuals randomly selected from a sample will belong to 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 0.886. The 2-way ANOVA did not yield any significant variation in the indices when the samples were taken from different locations or different days samplings. It showed that the diversity of earthworm species with reference to their abundance is almost the same in the fields of different blocks of Gorakhpur district of northeast Uttar Pradesh in each sample. The probability that two individuals selected randomly will belong to different species is very high.

(c) *Margalef Richness Index* (*R*):

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 date of sampling. All of the data ranged between 1.023 to 1.027. The analysis of data revealed insignificant effect on R of different localities and days of collection. It demonstrated that the species richness of the earthworm did not vary with collection sites and time of collection in northeast Uttar Pradesh.

(d) *Pielou Evenness Index* (*E*):

Pileou evenness index (E) is a measure of biodiversity which quantifies how equal the community is numerically. Its values ranged between 0 and 1. The higher value of E refers to the high evenness or less variation in communities 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 revealed 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 in northeast Uttar Pradesh.

Discussion

Nine earthworm species and two earthworm families have been identified from various fields of blocks in the current survey. About one-sixth of the families reported from this country reside in the Gorakhpur area of northeast Uttar Pradesh. According to data analysis, the Piprauli 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 Pali block had the lowest diversity (H' = 2.173 and D = 0.883). Greater species diversity is indicated by higher values of these indices, while less diversity is indicated by lower values. According to Edwards and Bohlen (1996), the diversity of earthworms ranged from 1 to 15 species, with the majority of earthworm communities including between 3-6 species. 7 to 11 species were found in cultivated, non-cultivated, grassland, garden, and sewage soils, as reported by Singh (1997). According to

Fragoso *et al*. (1999), the easiest way to evaluate species diversity is the number of species present in a specific earthworm community, which can range from 3 to 17 in tropical and temperate habitats. Usually, 4 to 14 species were present in tropical rainforests. Goswami (2015) observed that 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 field environments in the Gorakhpur district in northeast Uttar Pradesh are represented, and the earthworm communities there, which contain nine earthworm species, show similar diversity.

Conclusion

From the present study we have observed the different type of earthworm species i.e., *Lampito mauritii*, *Metaphire posthuma*, *Perionyx excavatus*, *Eutyphoeus waltoni, Eutyphoeus nicholsoni, Dichogaster bolaui, Eutyphoeus orientalis*, *Eutyphoeus incommodus* and *Amynthas morrisi* in fields of different blocks of Gorakhpur district of northeast Uttar Pradesh.

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