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Comparison of Biodiversity and Abundance of Earthworms in Maize Croplands, Irrigated with Sewage and Canal Water of District Faisalabad, Pakistan

COMPARISON OF BIODIVERSITY AND ABUNDANCE OF EARTHWORMS IN MAIZE CROPLANDS, IRRIGATED WITH SEWAGE AND CANAL WATER OF DISTRICT FAISALABAD, PAKISTAN

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ABSTRACT

The diversity and richness of earthworms that were irrigated with canal (Chak 200 RB Lathianwala) and sewage (Chokera) water in the district of Faisalabad were determined from August to November 2020. For collection of specimens, hand picking and digging method was used and then preserved in formalin. AAS (Atomic absorption spectrophotometer) was used to check the concentration of heavy metals in both water and soil samples. Total 10 species were identified from Chak 200 RB Lathianwala and showed Shannon-Weiner Diversity Index (H') =2.13, Evenness = 0.93 and Dominance = 0.07. The significant difference in abundance were determined ($t = 7.7115$, $p = 0.0001$ at $p < 0.05$). Water and soil samples of sewage water had an acidic pH (5.960, 6.213), while water and soil samples of canal water had a basic pH (7.748, 7.867). The levels of total dissolved solids, electrical conductance, dissolved oxygen, and alkalinity of sewage water were lower than those of canal water except for total suspended solids, turbidity, and metals, viz. Cr, Ca, Pb, Co, Cd, Zn, and Mg. In sewage water irrigated soil, their concentrations were also higher than canal water irrigated soil, except for Mg. A significantly positive correlation coefficient was observed among temperature and abundance at both localities. Results showed that acidic pH and higher concentrations of pollutants alter the diversity and abundance of earthworms.

Keywords: Earthworms, sewage water, canal water, soil, metals.

INTRODUCTION

Earthworms, belonging to the phylum Annelida, play a significant role within the community of invertebrates (Ghosh, 1993). Earthworms are regarded as engineers of ecosystems due to their physico-chemical and biological effects (Edwards, 2004). The factors that affect the diversification of earthworms are food availability, humidity, temperature, and physico-chemical factors in the soil (Curry, 2004). The research accessible on the taxonomies of species of earthworms was provided by Stephenson, (1923), who in his book, "Fauna of British India" explain taxonomical data on oligochaeta. They are specialized in maintaining the soil structure and physiological condition of the soil and utilized for the food chain, earthworms play a significant role in transforming waste energy from lower to higher tropic levels (Sivakumar et al., 2015). Earthworms have a tendency to migrate only over short distances and that's why they may be model sentinel terrestrial invertebrates for detecting anthropogenic organic contaminants in the food web (Bouche, 1992). Earthworms can aggregate organic and inorganic pollutants available in the land (Morrison et al. (2000); Phugare et al. (2012). The major eco-factor required for the life and growth of earthworms is water. The Bio-degradation procedure of earthworm is affected not only by quantity but also quality of water (Dayananda et al., 2008). Sewage water is also a source of enriched nutrients (Ullah et al., 2011). It enhances significant bio-nominal activities and very frequently alters the hydrology (Faruqui et al., 2004). Due to the availability of harmful substances and heavy metals in sewage water, it has adverse effects on the development of worms (Mahimairaja et al., 2000). In Pakistan, for irrigation purposes, almost all cities have a sewage water system (Qadir and Ghafoor, 1997) and International Water Management Institute showed that 32,500ha are instantly irrigated with waste water (Ensink et al., 2004). The farmers choose this water for irrigation because sewage nallahs, or drains have lower pumping cost and then boreholes, are inexpensive and more approachable to farmers with limited economic assets (Bakhsh and Hassan, 2005). Because of favorable soil and environmental conditions, maize is the grain crop,that gained third position in grown crops globally. In the manufacturing of maize, Pakistan is the largest country globally (Anonymous, 2006). The present study provided information on the fluctuation of earthworm diversity and abundance over the months on maize croplands of district Faisalabad.

MATERIALS AND METHODS

Sampling Sites

The current research was done to find out the impacts of environmental pollution on biodiversity and the relative abundance of earthworms in maize croplandsin designated areas of the region Faisalabad, Pakistan, for period of four months starting from August through November. After one month, sampling was done with three replicates, and for sampling, two sites were selected. One sampled site of maize crop was 'Chokera' which was being irrigated with sewage water, while the other site was 'Chak 200 RB Lathian wala' which was being irrigated with canal water.

Collection and Preservation of Earthworms

Up to 30cm depth, randomly selected samples were taken with a balanced quantity of soil per sample by excavating a quadrat of 50 cm^2 (Moreira et al., 2008). For earthworm assemblage, hand picking technique was utilized by Lewis and Taylor (1967). With water, all samples of earthworms were cleaned and transferred into a 5 % alcohol solution. Then specimens were moved into a dish and killed by adding 10 % solution of formalin. For hardening, all specimens were placed in this solution for one day. At last, they were stored in 5 % formalin. All samples were placed in plastic bottles and tagged with identification numbers related with site and month.

Identification

The samples were taken to the Biocontrol laboratory of the Department of Zoology, Wildlife, and Fisheries, University of Agriculture, Faisalabad, for counting and identification purposes. Different species of specimens were identified using stereomicroscope. Various structural attributes were utilized for this study modified from Stephenson (1923) and Bhatti (1962).

Collection and Digestion of Water Samples

From the feeders furrow, water samples were collected in every field. Preservation of the water sample was done by adding of HNO_3 (few drops) and placing it in freezer to stop microbial activity. After filtration of sample water, metals were analyzed as recommended by the AOAC (1990). The total suspended solids, total dissolved solids, pH, electrical conductivity and water alkalinity were determined using a digital meter (HANNA 8424).

Collection and Digestion of Soil Samples

Soil samples also collected from studied sites into polyethylene bags. Soil samples were first dried, then crushed and finally sieved with the help of a mesh (2 mm in size) to study physico-chemical parameters. The determination of metals was done by Atomic Absorption Spectrophotometer. Digestion with nitric-perchloric was done, by the operation suggested by the AOAC (1990). In a flask (250 ml), 0.5 g dried soil was settled. Nitric-perchloric acid (10 ml in 3:1 ratio) was added in a flask for 24-h. At 150 °C, the mixture was boiled until the solution color was altered from brown to yellow or transparent. The mixture was chilled and extracted by filter paper (Whatman No. 42) and transported by adding distilled water to 25 ml volumetric flask.

Determination of Physico-Chemical Parameters

Soil pH, EC and alkalinity were determined by soil water suspension 1:5, as described by the Rayment and Higginson (1992). 10gram (<2 mm) air dried soil was settled into a bottle; deionized water (50 ml) added and mechanically shaken for 10 minutes at 15 rpm. The electrode of the digital metre was immersed into the suspension of soil. The values were taken when chemical equilibrium was attained. Digital meter was calibrated as instructions by manufacturers. Mixture was stimulated with a stirrer during activity. Electrode was washed with deionized water between activities.

Statistical Analyses

Statistically data were analyzed to find out diversity and abundance, evenness and richness of species with Shannon-Weiner diversity index (Shannon, 1948). To check comparison t-test were used.

RESULTS AND DISCUSSION

Earthworms have a metameric body belonging to the phylum Annelida (Martin et al., 2008). They are desirable for ecological examination because they indicate soil pollution and can also be captured easily.

Table 1: Earthworm's diversity and relative abundance (%) in maize crop at Chak 200 RB Lathian wala.

Order	Family	Genus	Species	Total
Haplotaxida	Megascolecidae	Pheretima	*Pheretima posthuma*	87 (22.13 %)
			Pheretima hawayana	68 (17.30 %)
			Pheretima morrisi	46 (11.70 %)
			Pheretima houlleti	42 (10.68 %)
			Pheretima elongate	37 (9.41 %)
			Pheretima minima	33 (8.39 %)
			Pheretima lignicola	31 (7.88 %)
			Pheretima suctoria	19 (4.83 %)
			Pheretima bicincta	17 (4.32 %)
			Pheretima birmanica	13 (3.31 %)
Total			10	393 (100 %)

The goal of current research is to study the effect of pollutants on diversity and richness of earthworms in maize croplands at selected sites in Faisalabad. Samplings were done from August to November 2020. From the maize crop, 455 specimens were collected, out of which 393 specimens were taken from Chak 200 RB Lathian wala and 62 from the second site, which was Chokera. Order that idenified was Haplotaxida. A total of 10 species *P.*

suctoria, P. morrisi, P. houlleti, P. bisincta, P. elongate, P. hawayana, P. posthuma, P. minima, P. lignicola and *P. birmanica* were identified. From all other species, the most abundant was *Pheritima posthuma* with 87 specimens and an abundance of 22.13 %. Second most abundant species was *Pheritima hawayana* with 68 specimens and 17.30 % abundance. Least abundant species was *Pheritima birmanica* with 13 specimens was as 3.31 % (Table 1). From Chokera, all collected specimens were juveniles. Due to lack of morphometric characteristics, they could not be identified.

Table 2 represents the Shannon-Weiner diversity index and comparison (t-test) of earthworms at Chak 200 RB Lathian wala. At Chak 200 RB Lathian wala, the heterogeneity (H), evenness (E) and dominance (D) of the earthworm of were observed. Results of research showed H′= 2.13, E = 0.93 and D = 0.07. The t-value was 7.7115, and p-value was 0.0001. In both sites of district Faisalabad, the richness of worms was significantly different (Table 3). For comparison of abundance earthworm, t-test was used, which indicated a significant difference in selected sites, which were Chokera and Chak 200 RB Lathian wala (Table 4). At Chak 200 RB Lathian wala richness was high, while it was low at Chokera. According to previous research, high ammonia and integrated salts in animal waste may be toxic to earthworms (Curry, 2004).

Earthworm's diversity and abundance are largely affected when they are present in polluted soil containing metals (Spurgeon et al. 1994).

Table 2: Shannon-Weiner diversity index and comparison (t-test) of earthworms at Chak 200 RB Lathian wala.

Abundance	393
Total Number of Species (S)	10
Diversity (H′)	2.13
Dominance(D)	0.07
Evenness (E)	0.93
Mean ±SEM	39.30±7.35
T-value	5.34
d.f	9
p-value	0.0005
SD	23.25

The investigation of the parameters of water and soil was done. In water samples, no significant difference (p > 0.05) was exhibited by alkalinity (t = 1.528, p = 0.088), cobalt (t = 1.092, p = 0.158), chromium (t = 1.632, p = 0.076), cadmium (t = 1.649, p = 0.075), temperature (t=1, p= 0.379). while at α (p < 0.05), total suspended solid (t = 6.809, p = 0.0002), pH (t = 7.608, p = 0.0001), Zinc (t = 2.636, p = 0.019), magnesium (t = 0.624, p = 0.277), dissolved oxygen (t = 8.026, p = 0.0001), electrical conductivity (t = 4.086, p = 0.0032), total dissolved solids (t = 6.082, p=0.0004), turbidity (t = 3.176, p=0.009), lead (t = 4.213, p = 0.002) and calcium (t = 2.825, p = 0.015) exhibited highly significant results (Table 5).

Table 3: Abundance (%) of species of earthworms in maize at different depth and selected sites

Depth (cm)	Chokera	Chak 200RB Lathianwala	Total
0-10	34(7.47 %)	212(46.59 %)	246(54.06 %)
11-20	20(4.4 %)	128(28.13 %)	148(32.53 %)
20-30	8(1.75 %)	53(11.64 %)	61(13.39 %)
Total	62(13.6 %)	393(86.4 %)	455(100 %)

Table 4: Comparison of richness in selected sampling sites of Faisalabad.

	N	Mean ± SEM	SD	T-value	d.f	p
Chokera	4	15.50 ± 2.10	4.20	7.7115**	3	0.0001
Chak 200RB Lathianwala	4	98.3±10.5	21.0	-	-	-

N= sampling months, SD= standard deviation, d.f= degree of freedom, p=probability

Table 5: Physico-chemical parameters of sewage and canal water.

Parameters	Sewage water	Canal water	T-value	P-value
Temp.	32.00 ± 2.20	32.00 ± 2.20	1	0.379
pH	5.960 ± 0.145	7.748 ± 0.185	7.608*	0.0001
EC (ms/cm)	0.9125 ± 0.031	1.075 ± 0.024	4.086*	0.0032
DO (mg/L)	3.875 ± 0.272	6.4 ± 0.158	8.026*	0.0001
TDS (mg/L)	44.24 ± 4.56	86.15 ± 5.17	6.082*	0.0004
TSS (mg/L)	167.0 ± 15.6	55.63 ± 4.95	6.8091*	0.0002
Alkalinity (mg/L)	111.6 ± 13.5	134.45 ± 6.42	1.528	0.088
Turbidity (NTU)	218.0 ± 13.9	165.75 ± 8.80	3.176*	0.009
Co (mg/L)	1.138 ± 0.171	0.887 ± 0.152	1.092	0.1583
Cd (mg/L)	1.125 ± 0.161	0.735 ± 0.173	1.649	0.0751
Pb (mg/L)	2.800 ± 0.129	1.850 ± 0.185	4.213*	0.002
Zn (mg/L)	40.80 ± 2.79	26.50 ± 1.43	2.636*	0.01937
Cr (mg/L)	3.450 ± 0.140	3.250 ± 0.0645	1.6329	0.0767
Mg (mg/L)	228.70 ± 7.91	212.3 ± 25.1	0.6241	0.2777
Ca (mg/L)	914.8 ± 15.4	828.3 ± 26.4	2.825*	0.015

*Table values= Mean±SEM, *exhibit the significant difference at α= 0.05*

Table 6: Physico-chemical Parameters of soil irrigated with sewage and canal water.

Variables	Soil of Sewage water	Soil of Canal water	T value	P value
Temp.	30.00 ± 2.20	30.00 ± 2.20	0	0.5
pH	6.213±0.134	7.867±0.239	6.037*	0.0004
EC (ms/L)	2.2725± 0.0266	1.957± 0.0534	5.277*	0.0009
Alkalinity (mg/L)	2.875±0.161	4.900±0.272	6.4102*	0.0003
Co (mg/L)	4.025±0.105	3.775±0.103	1.6990	0.0701
Cd (mg/L)	0.3525 ± 0.0155	0.3275± 0.019	1.0101	0.1757
Pb (mg/L)	6.250±0.323	3.625±0.161	7.2746***	0.0001
Zn (mg/L)	35.42±1.57	24.30±1.38	5.3234*	0.0008
Cr (mg/L)	4.700 ± 0.258	3.900 ± 0.147	2.691*	0.017
Mg (mg/L)	277.8 ± 28.4	510.50 ± 6.29	7.993***	0.0001
Ca (mg/L)	1062.5±63.8	856.3 ± 25.5	3.001*	0.0119
Moisture	52.27 ± 1.58	60.63 ± 2.59	3.181*	0.0095
Texture of Soil	Sand	Clay		

Tables Value = Mean ± SEM
exhibit significant difference (p<0.05), * exhibit extremely significant difference (p<0.0001)*

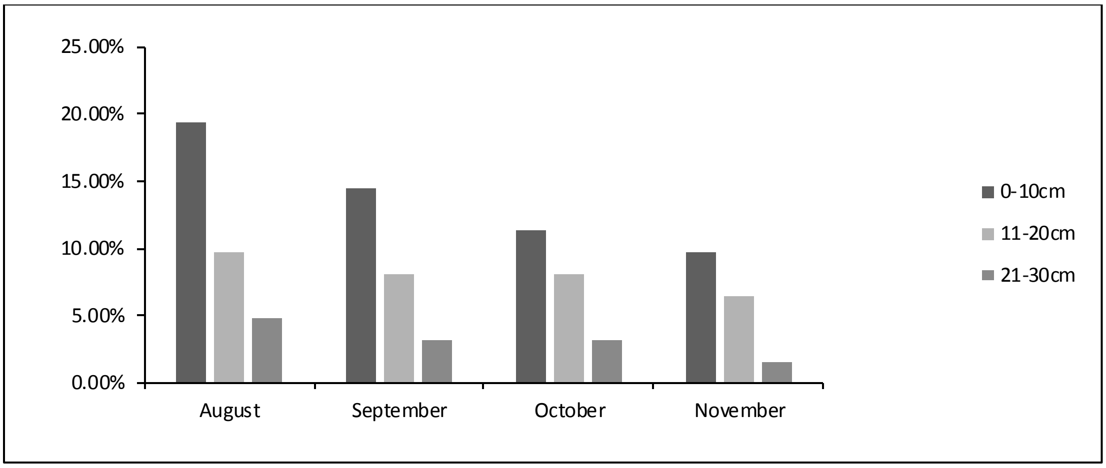

Figure 1: Monthly variation in relative abundance % of earthworms at Chokera.

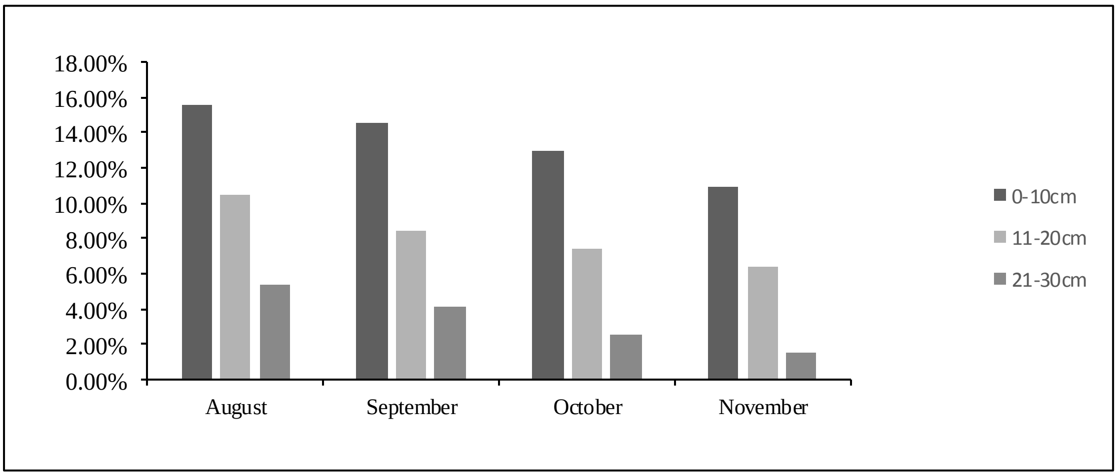

Figure 2: Monthly variation in relative abundance % of earthworms at Chak 200 RB Lathian wala.

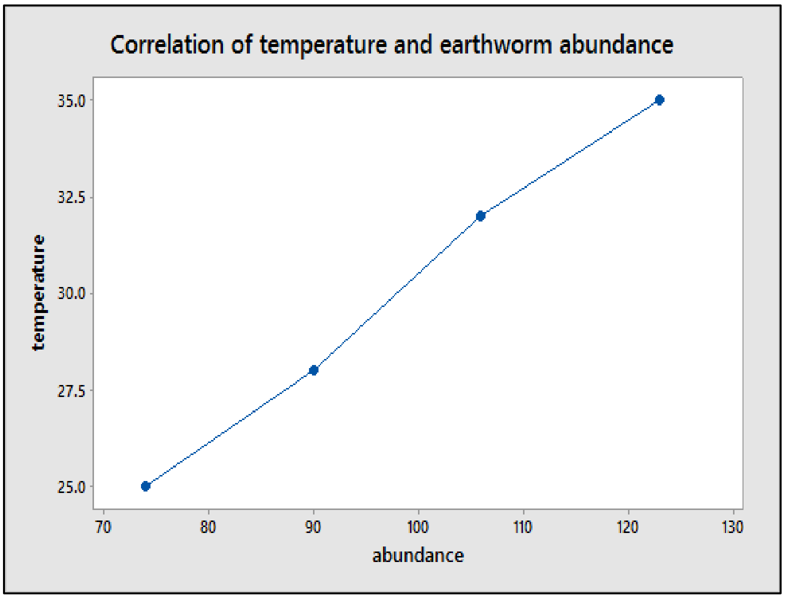

Figure 3: Association between temperature and earthworm abundance.

Heavy metallic contents were higher in samples of sewage water while lower in canal water. Overall, the pH was acidic in samples of sewage water and soil irrigated with sewage water, whereas the pH of canal water and soil irrigated with canal water was basic. Earthworms are present in somewhat acidic to alkaline pH (Chaudhuri and Bhattacharjee, 1999). State of relation among earthworm richness and physico-chemical factor were represented by Karmegam and Daniel (2007). At different sites, from a survey of three years in which they observed collected earthworms, there found a satisfactory relationship among the parameters and worms richness. They showed negative coefficients of pH and EC according to this argument, earthworms depend on these factors. For comparison, t-test of physico-chemical parameters were used. Figures 1 and 2 showed monthly variation in relative abundance % of earthworms at Chokera and Chak 200 RB Lathian wala. Table 6 exhibits means of physico-chemical parameters of soil that was being irrigated with sewage and canal water. The t-test exhibited slight difference ($p > 0.05$) of Cadmium ($t = 1.010$, $p = 0.175$) temperature ($t = 0$, $p = 0.5$) and Cobalt($t = 1.699$, $p = 0.070$) while electrical conductivity ($t = 5.277$, $p = 0.0009$), pH ($t = 6.037$, $p = 0.0004$), lead ($t = 7.274$, $p = 0.0001$), Zinc ($t = 5.323$, $p = 0.0008$), Chromium ($t = 2.691$, $p = 0.017$) and Magnesium ($t = 7.993$, $p = 0.0001$) and moisture ($t = 3.181$, $p = 0.009$) exhibited extremely significant difference. Similarly, intensity of heavy metals in soil irrigated with sewage water was higher and lower in canal water irrigated soil. In the current research the earthworms prefer soil of Chak 200RB Lathian wala and have high abundance than soil of Chokera. Similarly, Hendrix et al. (1992) observed spatial arrangement and population of earthworm according to changings of land. They found a strong correlation among abundance of earthworm and integrated substances of soil. Contents of moisture and use of land affect richness of earthworm (Edwards et al., 1995).

Result exhibited significantly positive correlation between both earthworm's richness and temperature (Figure 3). Regression line indicated that changes in temperature directly affect the specie abundance. The value of R^2 was 0.9928. In winter and summer, most of the species were absent as the species of earthworms are highly sensitive to the pressure of the ecological ingredient i.e. temperature, humidity (Pelosi et al., 2009). For a period of one year, monthly variations of earthworm fauna were represented by Goswani (2015).

CONCLUSION

The present research concluded that levels of pollutants affect the diversity and abundance of earthworms that are useful for soil fertility. Earthworms raise their abundance in loamy soil than sand-like soil. Different environmental factors, such as temperature, are very important for earthworms management. Acidic pH and a higher intensity of heavy metals have badly affected the diversity and abundance of earthworms.

AUTHOR'S CONTRIBUTION

SA* executed the research. NE supervisor and planned the research. FL and SA helped in manuscript write up. SA helped in conducting research in their laboratories.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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