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# Effects of Wastewater irrigation on nematode community structure in Agro-ecosystems Near Yamuna in Haryana, India

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**ABSTRACT**
*A survey of agricultural fields near Yamuna in Faridabad, Haryana was conducted to study the diversity and community structure of the soil inhabiting nematodes. A total of 32 genera belonging to 8 orders and 22 families were recorded. In terms of abundance, order Tylenchida was most abundant while in terms of number of genera, order Rhabditida was most frequent. In present study total number of nematodes significantly correlated with heavy metals positively. A low percentage of dorylaims in the crop fields (11% & 10%) clearly indicates that the soil is more disturbed. The lower values of MI in present study indicated a disturbed environment due to heavy metal contamination. The values of EI observed in the present study were very high at all the sites giving an idea of enriched ecosystem. The values for CI in present work were mostly low, lower at freshwater irrigated field and high at wastewater irrigated field.*
***Key words:*** *Nematode Communities Analysis, Maturity Index, Rhabditida, Wastewater, Yamuna.*

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## INTRODUCTION

Human society depends in many ways on services provided by soil ecosystems. The rapid development in the industry and other aspects of society are closely linked with the release of various waste materials into the environment, where their accumulation increases over time and negatively influences the soil ecosystem[1]. Introduction of heavy metals to the soil is more dangerous due to low degradability and high accumulation in soil horizons.

Within the last few decades, nematodes have gained increasing attention in freshwater ecotoxicology studies and their use in single-species toxicity tests [2,3,4,5], field studies [6,7,8], and in model ecosystems [9,10,11,12] is now well established. Their ubiquitous occurrence, ecological relevance, and universal applicability for variously complex ecotoxicological tools make nematodes excellent bioindicators [13].

Several studies have shown that nematode communities responded not only to the agricultural practices e.g. ploughing, crop rotation and water management [14], but also to the different organic and inorganic pollutants such as polycyclic aromatic hydrocarbons (PAH) or heavy metals [15,16,17].

The aim of the present study was to study the community structure of the soil inhabiting nematodes associated with crop fields near Yamuna river area in Faridabad, Haryana to assess the role of nematodes as indicators of soil condition.

## MATERIAL AND METHODS

Soil samples from agriculture fields near Yamuna river area in Faridabad, Haryana were collected. These fields havebeen irrigated with wastewaters and freshwaters formore than 30 years and vegetables like Tomato (*Solanum lycopersicum*), Okra (*Abelmoschus esculentus*), Cauliflower (*Brassica oleracea*), Carrot (*Daucus carota*), Mustard (*Brassica juncea*), Chilli (*Capsicum* spp.), Eggplant (*Solanum melongena*), Cabbage (*Brassica oleracea* var. *capitata*), Potato (*Solanum tuberosum*), Coriander (*Coriandrum sativum*) etc. are grown in these fields andsupplied to local market for consumption. From each field soil samples

were collected from a depth of 0-10 cm by using a hand spade. Samples were tagged, stored in sealed plastic bags and brought to laboratory for further processing.

Nematodes were extracted from 100 cc. of fresh weight of soil using Cobb's [18] sieving and decantation and modified Baerman's funnel techniques. All the nematodes from each extracted sample were counted and identified to genus level. Trophic groups were allocated according to Yeates *et al*. [19] and cp groups were assigned after Bongers [20]. Chemical analysis of the soil samples was done at soil testing laboratory, IARI, New Delhi. Nematode diversity was described using the Shanon's diversity index calculated at genus level (H'). Maturity index (MI) was calculated to estimate the relative state of two ecosystems studied. Trophic diversity was calculated by the trophic diversity index, (TDI) [21]. Structure index (SI) andenrichment index (EI) were calculated to determine therelative stability of the ecosystem studied.All indices were calculated by using MS Excel. Differences with $P < 0.05$ were considered significant and $P < 0.01$ as highly significant.

**Detailed Description of the Formulae Used are Given Below**

**Shannon's diversity (H′)** = −Σ (*pi* ln *pi*)

**Maturity Index (MI)**

$$MI = \sum_{i=1}^{n} V(i).f(i)$$

Where Vi= cp value of the ith taxon.

f(i) the frequency of that taxon in a sample

* Maturity index (MI) is calculated as the weighted mean of the individual c-p value.

**Plant Parasitic index (PPI)**

$$PPI = \sum PPiXi / \sum Xi$$

Where, Ppi = PP value assigned to taxon i according to Bongers (1990).

Xi = abundance of taxon i in the sample.

**Enrichment index (EI)** = (e/e+b) x100

**Structure index(SI)** = (s/s+b) x100

**Basal index (BI)** = (b/b+e+s) x 100

Where **e, b**&**s** are sum products of assigned weights and number of individuals of all genera.

**Trophic Diversity index (TDI) =** $1/\sum pi^2$

Where $pi^2$ is the proportional contribution of $i_{th}$ trophic group.

## RESULTS

Soil samples were analyzed for total organic carbon, total lead, total zinc and total copper. The soil chemical properties at wastewater and freshwater irrigated sites have been listed in table 1. It was observed that the concentration of all the three heavy metals were high at wastewater irrigated fields.

**Table 1. Ecological indices and other parameters for assessing the community dynamics**

| S.No. | Indices/Parameters | Values | |
|---|---|---|---|
| | | Freshwater irrigated field | Wastewater irrigated field |
| 1. | Maturity Index (MI) | 1.56 ± 0.23 (1.33 – 2.11) | 1.34 ± 0.24 (1.20 – 1.74) |
| 2. | Plant Parasitic Index (PPI) | 2.73 ± 0.36 (2.23 – 3.12) | 2.60 ± 0.22 (2.42 – 3.22) |
| 3. | Enrichment Index (EI) | 72.24 ± 11.23 (67.66 – 78.21) | 65.65 ± 10.35 (62.51 – 67.56) |
| 4. | Structure Index(SI) | 39.59 ± 11.22 (34.66 – 41.22) | 32.26 ± 11.12 (28.6 – 36.24) |
| 5. | Channel Index(CI) | 16.23 ± 12.42 (14.12 – 12.34) | 12.51 ± 8.51 (11.12 – 17.34) |
| 6. | Trophic Diversity Index (TDI) | 1.25 ± 0.22 (1.2 – 1.41) | 1.32 ± 0.35 (1.28 – 1.38) |
| 7. | Shannon's Diversity Index(H') | 1.58 ± 0.19 (1.2 – 1.82) | 1.80 ± 0.21 (1.52 – 2.31) |
| 8. | Total Organic Carbon (g/Kg) | 15.48 ± 1.24 (14.46 – 15.89) | 16.45 ± 1.12 (14.26 – 17.54) |
| 9. | Total Copper (mg/Kg) | 135 ± 11.34 (125.8 – 146.8) | 555.22 ± 18.26 (459.24 – 589.00) |
| 10. | Total Lead (mg/Kg) | 45.56 ± 10.8 (38.12 – 47.16) | 122.44 ± 11.72 (116.11 – 131.64) |
| 11. | Total Zinc (mg/Kg) | 432.22 ± 12.4 (286.3 – 490.9) | 1259.11 ± 55.75 (1112.2 – 1399.5) |

### Nematode Diversity

### In freshwater irrigated fields

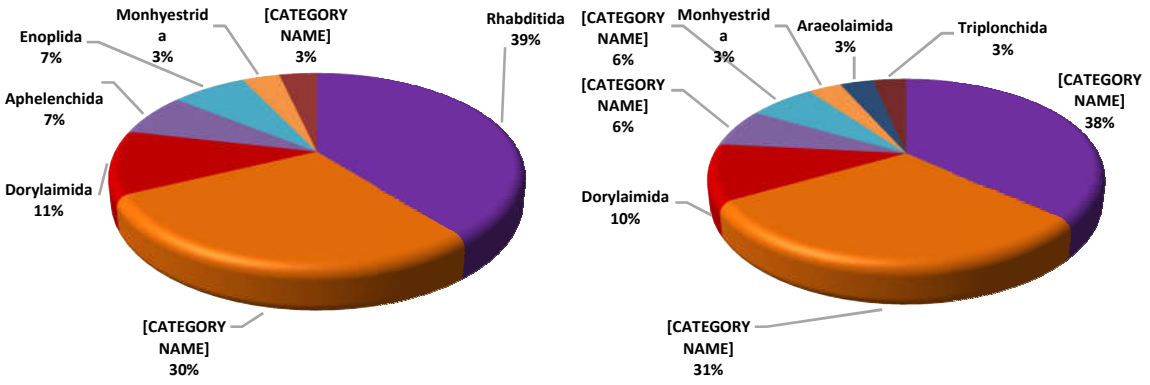
A total of 28 genera belonging to 7 orders and 19 families were recorded from the soil samples collected from crop fields irrigated with freshwater near Yamuna (Table 2). The number of genera varied from 3 to 18 per sample while in terms of abundance, the number varied from 142 to 1008 individuals per 100 cc of soil. *Meloidogyne* was the most abundant genus. In terms of number of genera (Fig. 1, A), the Order Rhabditida was most frequent (39%) with 11 genera under 5 families, followed by Tylenchida (30%)

with 8 genera under 7 families, Dorylaimida (11%) with 3 genera under 2 families, Aphelenchida and Enoplida (7%) each with 2 genera under 2 families, while Monhyestrida (3%) and Araeolaimida (3%) were represented by 1 genus each.

In terms of trophic diversity, the bacteriovores (43%) constituted the most dominant group (Fig. 2, A) followed by herbivores (32%), predators (11%), omnivores (7%) and fungivores (7%). Among bacteriovores, *Acrobeles* was the most dominant genus while *Meloidogyne*, *Aphelenchus*, *Mesodorylaimus* and *Mononchoides* were most dominant genera among herbivores, fungivores, omnivores and predators, respectively.

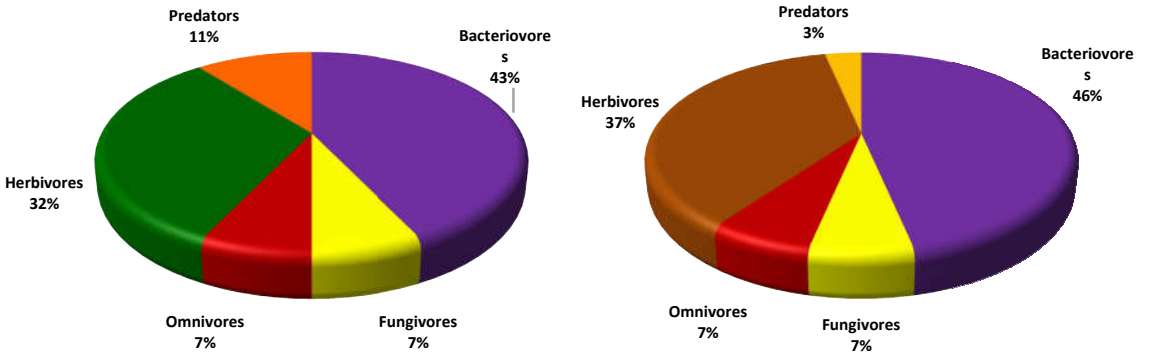
**Table 2: Population structure of soil inhabiting nematodes, their mean abundance per 100 cc soil ± SD (N = 30)**

| S.No. | Genera | c-p value | Order | N | Wastewater | N | Freshwater |
|---|---|---|---|---|---|---|---|
| | **Bacteriovores** | | | | | | |
| **1.** | *Bursilla* | 1 | Rhabditida | 6 | 3.24 ± 9.27 | 3 | 2.6± 2.22 |
| **2.** | *Mesorhabditis* | 1 | Rhabditida | 16 | 8.33 ± 10.35 | 7 | 4.4± 5.3 |
| **3.** | *Metarhabditis* | 1 | Rhabditida | 7 | 4.43 ± 8.56 | 2 | 1.2± 3.1 |
| **4.** | *Rhabditis* | 1 | Rhabditida | 1 | 0.30 ± 1.41 | 2 | 1.1± 1.4 |
| **5.** | *Acrobeles* | 2 | Rhabditida | 30 | 26.7± 21.5 | 30 | 28.2± 12.8 |
| **6.** | *Acrobeloides* | 2 | Rhabditida | 24 | 16.1 ± 12.10 | 20 | 11.7± 3.2 |
| **7.** | *Chiloplacus* | 2 | Rhabditida | 7 | 3.50 ± 7.87 | 3 | 2.1± 2.5 |
| **8.** | *Eucephalobus* | 2 | Rhabditida | 11 | 7.54± 13.70 | 3 | 2.8± 3.0 |
| **9.** | *Pseudacrobeles* | 2 | Rhabditida | 3 | 1.68 ± 5.98 | 0 | 0 ± 0 |
| **10.** | *Zeldia* | 2 | Rhabditida | 2 | 0.74± 3.78 | 2 | 0.4± 1.1 |
| **11.** | *Teratocephalus* | 2 | Rhabditida | 2 | 1.12 ± 2.87 | 3 | 2.8 ± 1.8 |
| **12.** | *Rhabdolaimus* | 2 | Araeolaimida | 2 | 1.34± 4.66 | 0 | 0 ± 0 |
| **13.** | *Chiloplectus* | 2 | Araeolaimida | 2 | 1.20 ± 3.88 | 3 | 1.3± 2.4 |
| **14.** | *Prismatolaimus* | 3 | Monhysterida | 12 | 5.54± 7.44 | 5 | 5.5± 2.1 |
| | **Fungivores** | | | | | | |
| **15.** | *Aphelenchoides* | 2 | Aphelenchida | 24 | 18.2± 19.40 | 20 | 11.8± 4.7 |
| **16.** | *Aphelenchus* | 2 | Aphelenchida | 26 | 18.9± 21.60 | 24 | 26.4 ± 3.6 |
| | **Omnivores** | | | | | | |
| **17.** | *Mesodorylaimus* | 4 | Dorylaimida | 3 | 2.1± 1.1 | 7 | 4.45± 9.66 |
| **18.** | *Minidorylaimus* | 4 | Dorylaimida | 3 | 1.7± 1.3 | 2 | 1.12 ±3.40 |
| | **Herbivores** | | | | | | |
| **19.** | *Xiphinema* | 5 | Dorylaimida | 2 | 0.4 ± 0.7 | 11 | 7.45 ± 9.10 |
| **20.** | *Pratylenchus* | 3 | Tylenchida | 25 | 22.4± 12.46 | 20 | 16.4± 6.5 |
| **21.** | *Psilenchus* | 2 | Tylenchida | 4 | 5.50 ± 17.42 | 3 | 3.8± 1.9 |
| **22.** | *Helicotylenchus* | 3 | Tylenchida | 23 | 18.2± 21.24 | 22 | 19.6± 5.8 |
| **23.** | *Hemicriconemoides* | 3 | Tylenchida | 2 | 1.22± 3.12 | 0 | 0 ± 0 |
| **24.** | *Hoplolaimus* | 3 | Tylenchida | 24 | 21.2± 18.22 | 23 | 21.6± 14.9 |
| **25.** | *Meloidogyne* | 3 | Tylenchida | 27 | 31.5 + 22.1 | 28 | 25.2± 11.8 |
| **26.** | *Rotylenchulus* | 3 | Tylenchida | 21 | 20.9± 17.45 | 15 | 12.9± 6.7 |
| **27.** | *Tylenchorhynchus* | 3 | Tylenchida | 24 | 23.15 ± 19.5 | 15 | 17.1± 11.4 |
| **28.** | *Trichodorus* | 4 | Triplonchida | 2 | 1.40 ± 1.20 | 0 | 0 ± 0 |
| **29.** | *Basiria* | 2 | Tylenchida | 29 | 38.21 ±23.45 | 2 | 1.2 ± 0.2 |
| | **Predators** | | | | | | |
| **30.** | *Tobrilus* | 3 | Enoplida | 3 | 3.23± 11.22 | 2 | 1.2 ± 0.4 |
| **31.** | *Mononchoides* | 1 | Rhabditida | 0 | 0 ± 0 | 3 | 1.2± 4.2 |
| **32.** | *Trypla* | 3 | Enoplida | 0 | 0 ± 0 | 3 | 2.1±1.1 |




**A** **B**

**Fig:1: Ordinal Diversity (Genera) of nematodes in Freshwater (A) and and wastewater irrigated fields (B).**




**A** **B**

**Fig:2: Trophic Diversity (Genera) of nematodes in Freshwater (A) and and wastewater irrigated fields (B).**

**In wastewater irrigated fields**

In case of wastewater irrigated crop fields, a total of 30 genera belonging to 8 orders and 21 families were recorded from the soil samples collected from crop fields irrigated with wastewater near Yamuna (Table 2). The number of genera varied from 5 to 14 per sample while in terms of abundance, the number varied from 224 to 1211 individuals per 100 cc of soil. *Basiria* was the most abundant genus. In terms of number of genera (Fig. 1, B), the Order Rhabditida was most frequent (38%) with 11 genera under 4 families, followed by Tylenchida (31%) with 9 genera under 8 families, Dorylaimida (10%) with 3 genera under 2 families, Aphelenchida (6%) and Enoplida (6%) each with 2 genera under 2 families, while Monhystrida (3%), Triplonchida (3%) and Araeolaimida (3%) were represented by 1 genus each.

Of the total 30 genera recorded, 14 were bacteriovores, 11 herbivores, 2 fungivores, 2 omnivore and 1 predator (Fig. 2, B). Among bacteriovores, *Acrobeles* was the most dominant genus while *Basiria*, *Aphelenchus*, *Mesodorylaimus* and *Tobrilus* were most dominant genera among herbivores, fungivores, omnivores and predators respectively.

It was found that *Mononchoides* and *Trypla* which are low cp value predators, were absent in wastewater irrigated fields where the concentration of the heavy metals was high. The presence or absence of these nematode genera at a particular site seems to be influenced by heavy metal concentration. The complete absence of mononchids at freshwater and wastewater irrigated fields suggest their utility as indicators of heavy metal pollution.

**Nematode Community Analysis**

Diversity and maturity indices were calculated to assess the diversity of nematodes at freshwater and wastewater irrigated fields (Table 1). The value of Shannon's diversity index was high at wastewater irrigated field ($p<0.05$), while it was low at freshwater irrigated field. The maturity index was high at freshwater irrigated field while it was low at wastewater irrigated field. Trophic diversity index was high at wastewater irrigated field while low at freshwater irrigated field. The high value of SI was observed at

freshwater irrigated field, while low value was found at wastewater irrigated field. The values of EI observed in the present study were very high at all the sites giving an idea of enriched ecosystem. The value for EI was high at wastewater irrigated field. The values for CI in present work were mostly low, lower at freshwater irrigated field and high at wastewater irrigated field.

## DISCUSSION

Soil Nematode communities and their structural changes were found to be one of the best biological tools for assessing soil processes and plant conditions in terrestrial ecosystems [22,23]. The soil environment significantly impacts on soil dwelling nematode communities. No single nematode index was universal in indicating the difference in soil health, but rather soil health requires a more indepth understanding of the nematode community composition, both trophic groups and life strategies [24]. Soil nematodes, as bioindicators of soil health, would not replace current soil chemical and physical tests, but would supplement information obtained and increase the understanding of the soil ecology and the effects of soil management. Nematodes respond differently to soil disturbance and therefore changes the nematode community composition [25,26].

The present work was aimed to study the effect of wastewater irrigation contaminated by heavy metals on community structure of the soil inhabiting nematodes associated with agricultural fields near Yamuna River in Faridabad, Haryana. Jagtap et al. [27] and Ahalavat & Chaubey [17] reported presence of various heavy metals in crop fields irrigated with wastewaters from Pune and Delhi, respectively that affects the community structure of the soil inhabiting nematodes. In the present study theconcentration of heavy metals were extremely high ascompared to earlier studies. It maybe due to the fact that nearby factories dumps about 95% of itswaste into the Yamuna. This water is used by farmers for irrigating their crop fields, thus paving a way for heavy metals to accumulate in agro-ecosystems.A low percentage of dorylaims in the crop fields (11% & 10%) clearly indicates that the soil is more disturbed as cropping always involves ploughing and/or tilling together with addition of fertilizers, organic matter and pesticides/weedicidesand. The dorylaims appear to be susceptible to these activities as also shown by Thomas [28] and Sohlenius and Wasilewska [29]. Hence, the sensitivity of the dorylaims is a good indicator of soil disturbance [30]. The results indicated that Cu and Zn had positive effects on the nematode communities and trophic structure and plant parasitic nematodes were most abundant at more polluted sites.

In present study total number of nematodes significantly correlated with heavy metals positively. Parmelee et al.[31] studied that certain trophic groups are more sensitive to copper than the total nematode population. This is in agreement in with present study where total abundance was higher at wastewater irrigated site with high heavy metal concentration but omnivores, predators and carnivores were lowest at this site. Shannon‘s diversity index (H‘) reflects diversity of nematodes in an ecosystem. Higher values of H‘show highly diverse ecosystem while low values show the contrary. The low values of Shannon’s diversity index reflect low diversity of nematodes at both sites. Smit et al. [32] also reported low values of H’ from Zn contaminated soils.

The MI has been used successfully as indicators for heavy metal pollution [33,34,35]. Various case studies [36] suggested that the MI is decreased by pollution (sewage waste, oil, heavy metals) but increases during the colonization process. The lower values of MI in present study indicated a disturbed environment due to heavy metal contamination. Ahalavat and Chaubey [17] also reported low values of MI in heavy metal treated soils. Enrichment index is a good indicator of enrichment in contaminated soils [37]. In present study almost no correlations were observed of EI with heavy metals. It is an agreement with Park et al. [38] and Ahalavat and Chaubey [17] who observed no significant differences in EI in heavy metal treated soils. Channel index (CI) indicated predominant decomposition channels in the soil food web, a high CI (>50 %) indicates fungal decomposition channels whereas low CI (<50 %) suggested bacterial decomposition channels. Low values of CI in present study correspond to bacterial decomposition channels. Similar effects of heavy metals pollution on fungivores and bacteriovores nematodes have been studied by Nagy et al. [39] and Ahalavat and Chaubey [17].

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