# SOIL NEMATODE COMMUNITY SHIFTED TO BACTERIAL-BASED FOOD WEB IN RESPONSE TO THE TROPICAL AGRICULTURAL PLANTS

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## Abstract

# Soil Nematode Community Shifted to Bacterial-Based Food Web in Response to the Tropical Agricultural Plants

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Soil nematodes have significantly contributed to decomposition process of organic matter, and provide a valuable indicator of soil food web responsible for nutrient cycling. However, published data on nematode community dynamics due to plant changes in tropics are lacking even though plant species or plant type changes is a common practice in agriculture. This research examined nematode community response to plant changes, from forest tree to agricultural plants. We established microcosms of forest soil (*Agathis dammara*) and planted with *Oryza sativa*, *Amaranthus sp*, *Solanum lycopersicum*, *Arachis hypogaea*, and *Citrus reticulata* (105 pots). To further follow soil food web dynamics, we sampled the soil after 6, 15, and 23 weeks of plant growth for nematode enumeration. Ecological indices, food web indices, and canonical correspondence analysis (6900 data points) were used to measure the community. After six weeks of plant replacement, nematode abundance decreased to 40-86%, except for soil with *A. hypogaea* (2% increase). It required 15 weeks for nematode community to reach its original abundance in the soil of *C. reticulata* and all plants, 23 weeks for soil with *S. lycopersicum*. Although the ecological indices revealed similar values for nematode community across the selected plants (H':2.69-3.07, E: 0.46-0.68, Berger-Parker: 0.15-0.24), food web indices showed different patterns. The soil community changed from nematodes assembling fungal decomposition channel in the forest soil (CI: 73) to the community supporting the bacterial decomposition channel (BI: 43-78) when the plants altered. The community separated into three major nematode compositions according to the plants, i.e., the community of *S. lycopersicum*, the community of *Amaranthus sp-O. sativa- C. reticulata*, and the community of *A. hypogaea* ($R_{AX-1}$: 0.97, $R_{AX-2}$: 0.79, CV: 87%). Temporal changes of nematode genera diversity appeared not to affect their functional diversity but driving the community toward bacterivorous dominance with three separate compositions.

Keywords: soil food web, channel index, fungal-decomposition channel, temporal changes

## Introduction

Soil nematodes have significantly contributed to decomposition process of organic matter, and provide a valuable indicator of soil food web responsible for nutrient cycling.

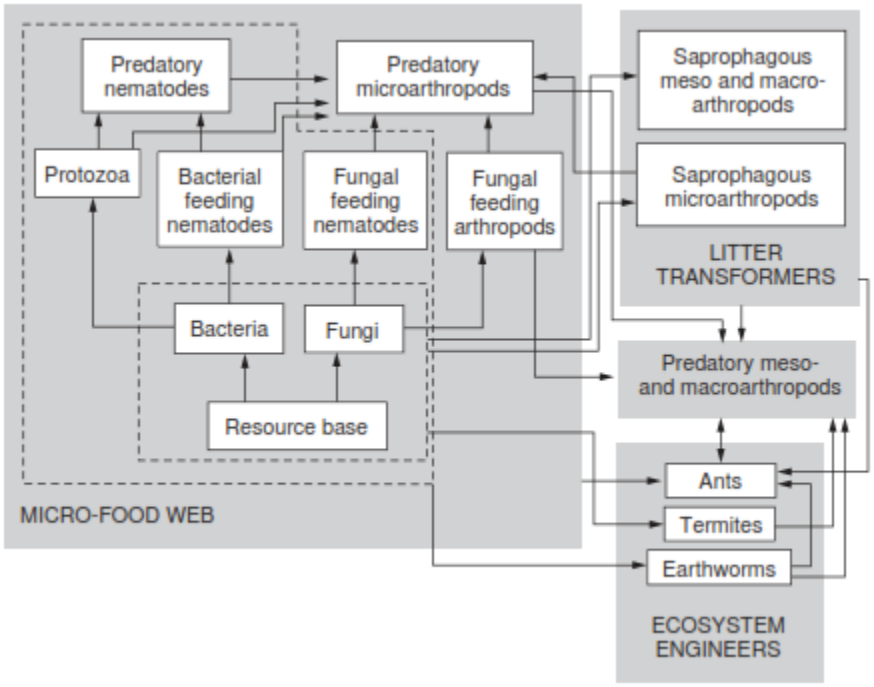



Figure 1. The soil food web organization (Wardle 2002, Lavelle et al., 1995)

However, published data on nematode community dynamics due to plant changes in tropics are lacking even though plant species or plant type changes is a common practice in agriculture. This research examined nematode community response to plant changes, from forest tree to agricultural plants.

## Materials and Methods

### *Microcosm Design*

The microcosms were set up in 105 pots of 15-cm diameter. Each pot was filled with soil for 20 cm depth. The soil was collected from a mature forest dominated by *Agathis dammara* trees located at Baturraden, Purwokerto, Indonesia. The soil texture was silt loam (60% silt, 22% sand, and 18% clay) with 9.81% Carbon, 0.71% Nitrogen, and pH of 6.85. The forest soil was selected to avoid effect of former agriculture plant and to investigate dominant plant changes. The microcosms were let to sit for six weeks before seed planting to allow recovery for the soil organisms after soil perturbation due to soil handling.

Five species representing types of tropical agricultural plants were selected, *Oryza sativa* (grass), *Amaranthus sp* (herb), *Solanum lycopersicum* (shrub), *Arachis hypogaea* (legume), and *Citrus reticulata* (tree). Each plant was grown from the seed in the microcosms, and made up the five treatments. Besides, soil with no plant and soil with all plants were assigned as the

controls. The microcosms were watered once a day to maintain the water availability for soil biota and plant growth. Wild plants were handpicked and returned to the pots after cutting them into small pieces. There was no external input, such as fertilizers and pesticides, applied to the soil during the experiments.

### *Soil Sampling*

Soil samples from five replicates of the seven treatments were obtained destructively from the microcosms (35 units) after seven, 15, and 23 weeks of plant growth. This sampling time was set to cover stages of the various plant growths and to follow nematode community after harvest time and plant death. Based on the standard practice in agriculture, *O. sativa* was commonly harvested after 12-14 weeks, *Amaranthus sp*. after about 4-5 weeks, *S. lycopersicum* after 8-10 weeks, *A. hypogaea* after approximately 14 weeks, and *C. reticulate* after years.

Nematodes as the main soil organism examined in the study were enumerated from each sample. Soil bacteria were measured as supporting variables to further describe the soil Biology in the microcosm soils. Soil pH, total Carbon, total Nitrogen, moisture, and temperature were, also, measured at each sampling time to describe the physico-chemical properties of the soil.

### *Nematode Enumeration*

We applied the modified Baermann funnel technique for extraction of nematode from the soil [4]. The soil sample (15-20 g) was wrapped with paper tissue and submerged in a funnel filled with water. After 24-48 hours, about 5 ml water at the tip of the funnel was collected in a conical tube and preserved with 4% formaldehyde, final solution. Nematodes from each sample were counted and identified at the genus level following Bonger [5], Tarjan [6], Ferris et al. [7], Freckman and Baldwin [8]. The nematodes were further assigned to functional guilds according to Bonger [9], Yeates et al [10] and Ferris et al [11] who separate nematodes into bacterivores (Ba), fungivores (Fu), predators (Pr), omnivores (Om) plant feeders (Pl), and place them into colonizer-persister (c-p) scale (1-5), and particular weight (0.8-5.0) to describe the nematode profiles.

### *Bacteria Enumeration and Plant Growth*

The bacteria abundance, extracted from each soil samples, were measured to describe the soil micro-food web. Bacteria are one of the essential basal resources in the micro-food web dominating agriculture soil. They were extracted from the soil and expressed in cfu.g-1 soil. The plant growth was measured following Hunt (1990) by calculating the Absolute Growth Rate (AGR), Relative Growth Rate (RGR), and simple ratio consisting of Leaf Area Ratio (LAR), Leaf Specific Area (SLA). The AGR was measured from the stem growth and number of leaves in two week intervals, $AGR = (L_2 - L_1)/(t_2 - t_1)$, where L: stem length or number of leaves. The RGR, LAR, and SLA calculations were based on three time sampling of plant dry weight. The growth was according to the formulae of $RGR = (\text{Log}_e W_2 - \text{Log}_e W_1)/(t_2 - t_1)$, $LAR = [(L_{A1}/W_1) + (L_{A2}/W_2)]/2$, $LSA = [(L_{A1}/L_{W1}) + (L_{A2}/L_{W2})]/2$, where W: total dry weight per plant, $L_A$: total leaf area per plant, $L_w$: total leaf dry weight per plant. The dry weight and organic content of above-ground and below-ground plant were also calculated to describe plant growth allocation.

### *Data Analysis*

Nematode community composition was used to calculate diversity, and food web condition among the plant types. The ecological indices included Shannon diversity (H’),

Evenness (E), and Berger-Parker Dominance ($D_{BP}$) [12]. The food web condition was based on Basal Index (BI), Enrichment Index (EI), Structured Index (SI), and Channel Index (CI) [11]. The indices were calculated as EI = (100)[e/(e+b)], SI = (100)[s/(s+b)], and CI = (100)[0.8$Fu_2$/(3.2$Ba_1$+0.8$Fu_2$)], where e = $\Sigma k_e n_e$, b = $\Sigma k_b n_b$, s = $\Sigma k_s n_s$, the $k_s$, $k_b$, $k_e$ = weight of structured, basal, and enriched indicator guilds, $n_s$, $n_b$, $n_e$ = abundance of nematodes in the respected guilds, F2 = fungivorous nematodes in c-p scale 2, and B1 = bacterivorous nematode in c-p scale 1 [11]. These food web indices were used to investigate the nematode food web development according to agricultural plant types.

To examine the plant growth patterns, we applied Principles Componen Analysis (PCA) to the plant data. Further, to investigate the genus composition of nematode community under influence of the plant types and how the community changed in relation to plant growth, we performed the Canonical Correspondence Analysis (CCA). We assigned plant types, bacterial abundance, plant growth, soil C, N and pH, soil moisture and temperature as the environmental factors, whereas nematode community as the species variables. The analysis was run in Canoco V.4.5 software.

## Results

### *Plant Characteristics*

The plants showed various patterns based on their 43 different variables consisting of growth indicators (AGR, RGR), simple ratio (SLA, LAR), and biomass alocation. *O. sativa*, *A. hypogaea,* and *C. reticulata* demonstrated distinct characteristics whereas *Amaranthus sp*, and *S. lycopersicum* did not show a clear patterns (**Figure 2**).

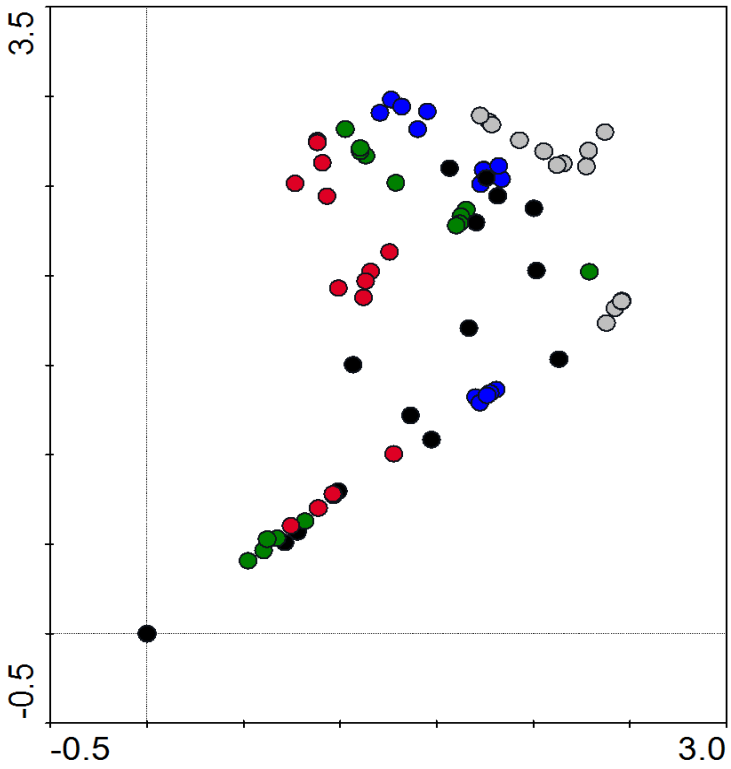


**Figure 2**. Plant species characteristics based on their AGR, RGR, SLA, and plant biomass alocation (DCA biplot, Eigenvalue Axis-1: 0.26, Axis-2:0.16, CV: 49.2%) (blue: *O. sativa*, black: *Amaranthus sp*, green: *S. lycopersicum,* grey: *A. hypogaea,* red: *C. reticulata*)

### *Nematode Community*

After six weeks of plant replacement, nematode abundance decreased to 40-86%, except for soil with *A. hypogaea* (2% increase). It required 15 weeks for nematode community to reach its original abundance in the soil of *C. reticulata* and all plants, 23 weeks for soil with *S. lycopersicum* (**Figure 3**).

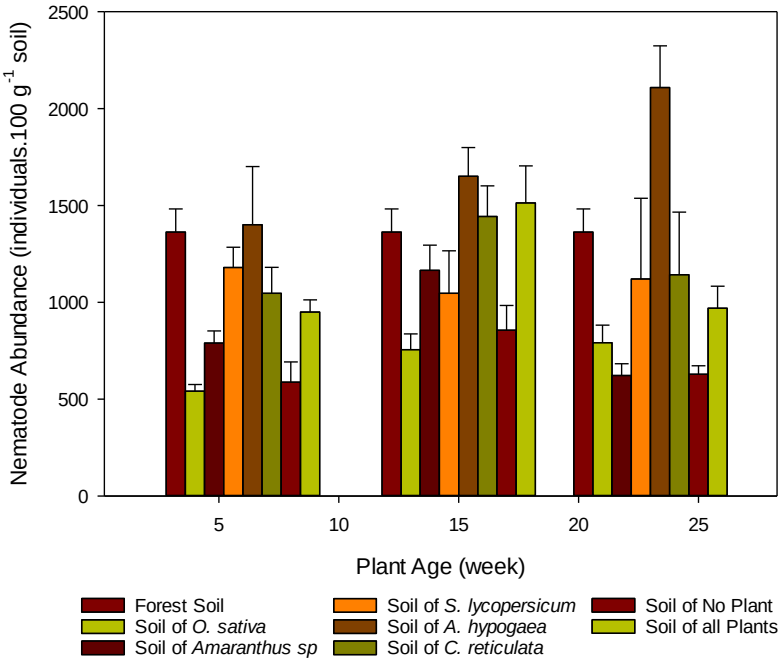



**Figure 3**. Nematode abundance in soil of different plant species and types over 23 weeks of growth (n: 5, n forest soil: 10, n $\pm$ sd)

Although the ecological genus indices revealed similar values for nematode community across the selected plants (H':2.69-3.07, E: 0.46-0.68, Berger-Parker: 0.15-0.24) (**Figue 4**), food web indices showed different patterns.

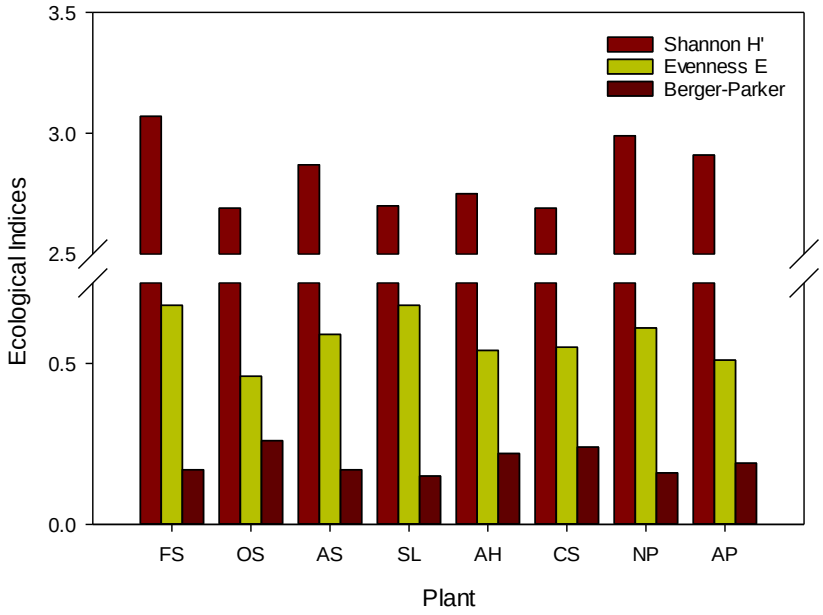
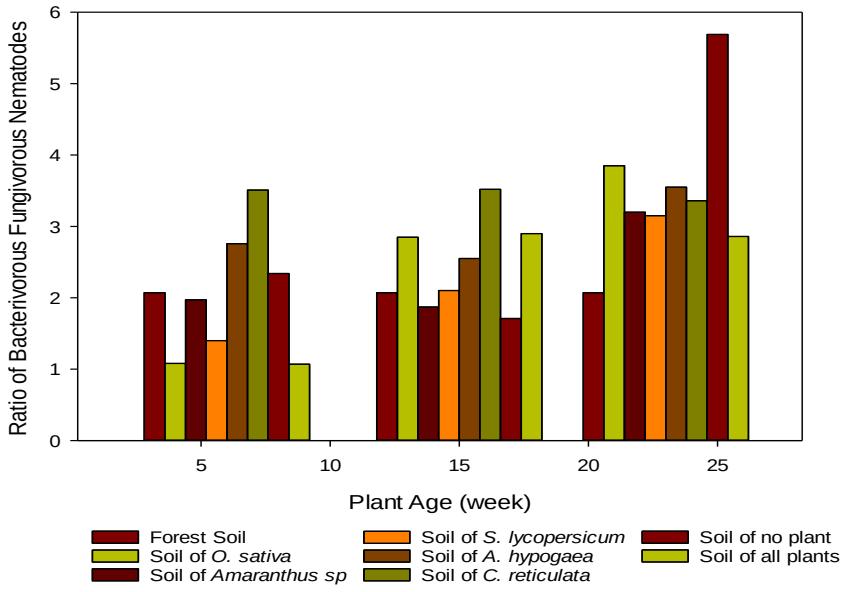
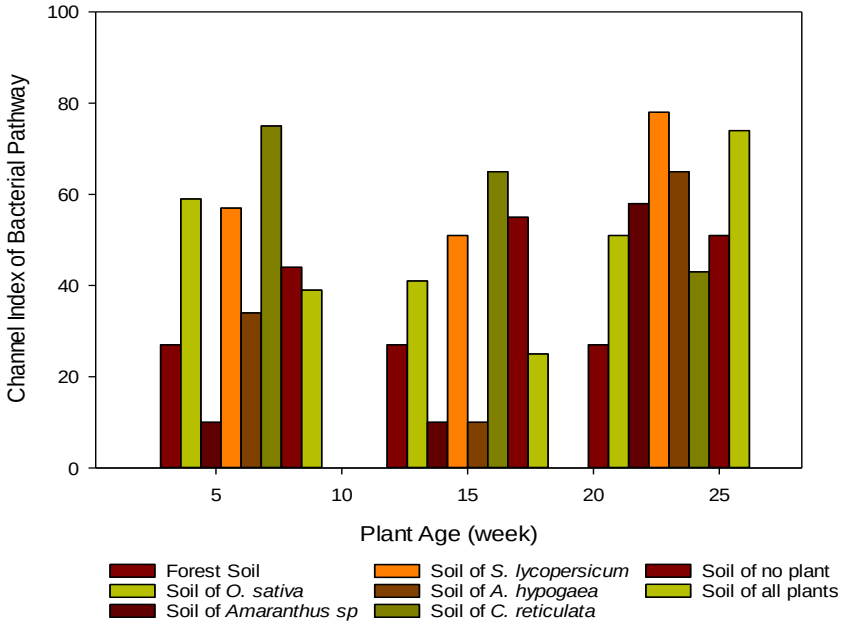



**Figure 4.** Nematode ecological indices based on genus level (FS: forest soil, OS: *O. sativa,* AS: *Amaranthus sp,* SL: *S. lycopersicum*, AH: *A. hypogaea*, CS: *C. reticulata*, NP: no plant, AP: all plants)

The soil community changed from nematodes assembling fungal decomposition channel in the forest soil (CI: 73) to the community supporting the bacterial decomposition channel (BI: 43-78) after 23 weeks when the plants altered (**Figure 5**).





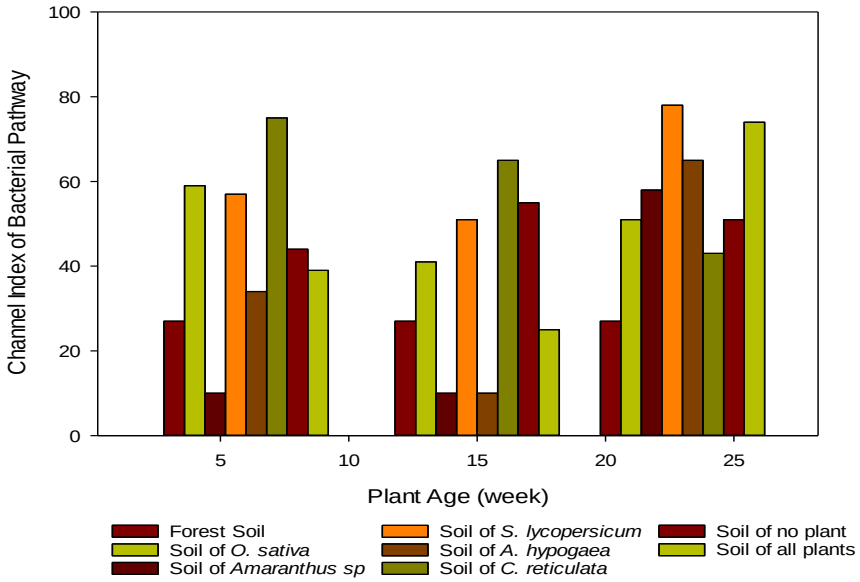
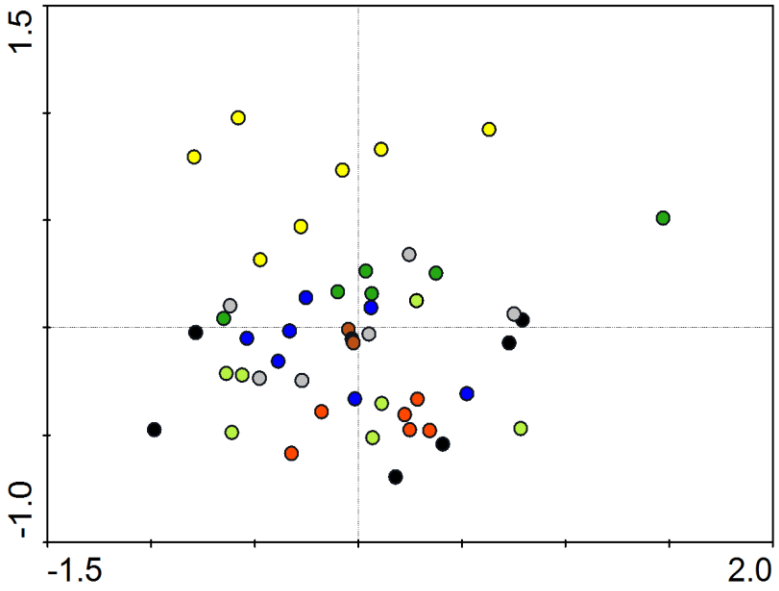
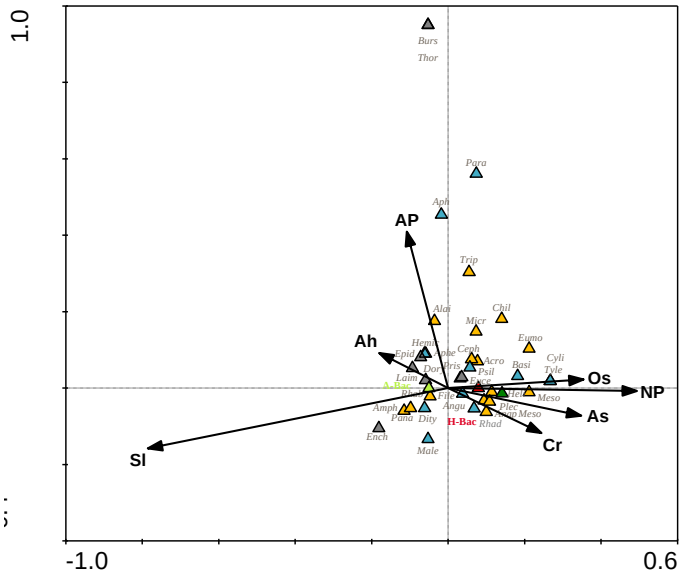




**Figure 5**. The ratio of bacterivorous to fungivorous nematode (top), Channel Index indicating fungal pathway (middle), and bacterial pathway (bottom) in soil of various plants over 23 weeks of age

The community separated into three major nematode compositions according to the plants, i.e., the community of *S. lycopersicum*, the community of *Amaranthus sp-O. sativa- C. reticulata*, and the community of *A. hypogaea* ($R_{AX-1}$: 0.97, $R_{AX-2}$: 0.79, CV: 87%).




## Conclusions

Temporal changes of nematode genera diversity appeared not to affect their functional diversity but driving the community toward bacterivorous dominance with three separate compositions.

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