Available online at:
http://journal.unila.ac.id/index.php/tropicalsoil
DOI: 10.5400/jts.2022.v27i1.37-47

Use of Biochar to Control Root-Feeding Soil Nematodes on Muna Local Tomatoes Variety

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Received 20 September, 2021; Revised 5 October, 2021; Accepted 15 December 2021

ABSTRACT

Energy pathways in soil nematode communities consist of energy pathways for roots, bacteria, and fungi. The dominance of the root energy pathway indicated an increase in nematode attacks on the roots that can be regulated through changes in food availability and the environment. This study aimed to (i) determines the effect of the biochar rate on soil nematode energy pathways on local tomato plants and (2) determine the biochar rate that can suppress the dominance of root-feeding nematodes of local varieties of Muna tomato plants. The treatment tested was the biochar rate expressed as a percentage of the biochar weight the soil weight, namely 0%, 5%, 10%, and 15%. Each was repeated three times, randomly placed in the experimental plot following the randomized block design procedure. The results showed that the addition of biochar to 10% of the soil weight decreased the abundance of the total nematodes and family Longidoridae, on the other hand, increased Aphelenchoididae, Spearman rho correlation. The abundance of root eaters decreased; on the other hand, fungivores increased with the biochar rate. Spearman rho indicated that fungivores were negatively correlated with root-feeders and omnivores while positively correlated with predators. It was concluded that applying biochar up to a rate of 10% of the soil weight before planting could suppress the abundance of root-feeding nematodes in the vegetative growth phase of Muna local tomatoes variety.

Keywords: Biochar rate, decreased, fungivorous, negatively, root-feeder

INTRODUCTION

Tomato is an economically significant fruit or vegetable crop second only to potatoes globally. The fruit contains antioxidants that prevent cancer, reduce the risk of developing heart disease and disorders, vitamins C and E, minerals, and carotene (Quinet *et al*. 2019). Recently, tomato fruit extract was proposed as an antiplatelet for patients infected with COVID-19 (O'Kennedy and Duttaroy 2021). Southeast Sulawesi, especially Muna island, has local tomatoes with curly fruit shapes. This local variety of Muna has been cultivated for generations by local farmers in Tongkuno District. Optimal production of local tomato plants is minimal by biotic stresses below the soil surface in the form of attack by root-eating nematodes that begin to occur in the early stages of flowering and then die during flowering (Alam *et al.* 2015). As a result, local farmers have difficulty developing local tomato cultivation.

The nematode community in the soil food web consists of root-feeders, fungivores, bacterivores, omnivores, and predators (Liang *et al.* 2020). Three energy pathways are identified in the soil nematode community from these feeding groups, namely the bacterial, fungal, and root-feeding pathways. Energy pathways of bacterivore nematodes and energy pathways of fungivores play a vital role in the decomposition process and mineralization of nutrients so that they become available in the soil to be taken up by roots for plant growth (Zhao and Neher 2014). The dominance of root-eating nematode energy pathways over other energy pathways indicates an increase in root nematode attack, implying that farmers' yield losses will be more significant (Tian *et al.* 2020).

For this reason, it is necessary to change the dominance of this soil nematode energy pathway.

The soil nematodes composition in the food web is sensitive to changes in soil environmental conditions (Zhang *et al*. 2019). Regulation of the dominance of each soil nematode energy pathway can be through bottom-up control by changing food availability and soil environmental conditions (Liu *et al*. 2016). Modifying food availability in the soil environment can use organic fertilizers (Xiong *et al*. 2018). The fertilizer application from animal manure increased the abundance of bacterivorous, omnivorous, and predatory groups. On the other hand, root-feeders decreased, while the response to fungivora was more varied (Yang *et al.* 2016). Due to the global pandemic of zoonotic diseases such as SARS-CoV, MERS-CoV, and Covid-19 which is currently underway (Di Marco *et al*. 2020), the use of animal manure from mammals needs to be reconsidered in implementing the concept of sustainable agriculture in the 21st century (Reganold dan Wachter 2016). For example, in guano fertilizer from bat droppings, Betacorona virus group C was detected (Wacharapluesadee *et al.* 2013). Viruses clustered into Betacoronavirus group C have caused an explosion of respiratory infections in Saudi Arabia (Woo *et al*. 2012). This fact indicates the importance of using other organic materials to change the dominant energy pathway of root-feeding nematodes to the composition of nematodes that prevent root-feeding attacks on the plant's roots.

Biochar from agricultural solid waste is synthesized through the pyrolysis process. Biochar is often used to ecologically engineer soil quality to increase agricultural productivity by modifying soil physical, chemical, and biological quality parameters (Hardy *et al*. 2019; Diatta *et al.* 2020). Ecological engineering of soil quality is directed at maximizing the capacity of soil biodiversity to increase sustainable agricultural productivity so that agricultural chemical inputs are minimized, including the use of nematicides to suppress the population explosion of root-feeding nematodes. Zhang *et al.* (2013) reported that the addition of biochar to the soil increased fungi-eating nematodes while suppressing the abundance and diversity of root-feeding nematodes in wheat orchards. A study conducted by Liao *et al.* (2016) found that the community structure of soil nematodes was sensitive to the addition of biochar into the soil. Liu *et al.* (2020a) found that applying high biochar doses increased plant productivity but decreased the abundance of bacterivorous, fungivorous, and root-feeding nematodes. It was further explained that increasing the amount of biochar shifted the composition of nematodes towards fungivorous dominance. The findings of Cole *et al*. (2021) that soil-applied in small amounts of biochar had a lower population of parasitic nematodes and higher predatory nematodes. The review of Domene *et al*. (2021) concluded that the change in dominance of the root-eating nematode group was determined by the dose of biochar in the agroecosystem. Almaroai and Eissa (2020) that the application of biochar at doses of 0, 5, and 10 Mg ha-1 showed the yield and quality of tomato increased in line with the increase in biochar. However, studies related to the impact of biochar application on soil nematode community structure in plants Muna local tomatoes are still being neglected.

This study aimed to determine the effect of soil nematode energy pathway and biochar dose on the growth of local tomato plants, as well as to determine the biochar dose that was able to suppress the dominance of root-eating nematode groups and increase the growth and yield of local varieties of Muna tomato plants.

MATERIALS AND METHODS

Site Description

This study was carried out in a farmer's garden located in Nanga-Nanga Village, Mokoau Village, Kambu District, Kendari City from June-September 2021, located at 122033"21' East Longitude and 402'44" South Latitude, and an altitude of 30 m above sea level. The topography is flat with a slope of 0-3%. Rainfall is 344.0667 mm month-1, air temperature is 25.53 ^{o}C, and humidity is 89%. The physicochemical characteristics of the soils are listed in Table 1.

Experimental Design

The treatments consisted of four levels of biochar rate (%, biochar weight/soil weight), namely 0% biochar (labeled B0), 5% biochar (labeled B1), 10% biochar (labeled B2), and 15% biochar (labeled B3). The experimental field plots were constructed according to the randomized complete block design with three replicates.

Experiment Plot Set-Up

All vegetation that grows above the soil surface is removed, then the land is plowed using a tractor equipped with a soil splitting knife, followed by a blade breaking up chunks of soil. In this area, twelve plots were made, each measuring 3 m in length, 2 m in width, and 15 cm in height. The distance between the plots in groups is away 60 cm, and between groups are away 80 cm.

Table 1. Soil Physico-chemical characteristics of the study site.

Parameter	Unit	Method	Value
Soil particle fraction:			
Sand	%	Pipette	14
Silt	%	Pipette	60
Clay	%	Pipette	26
Bulk density	G cm^{-3}		1.73
pH $_{(H2O, 1:5)}$	-	pH meter	5.0
pH $_{(KCl, 1:5)}$	-	pH meter	4.0
EC $_{(H2O, 1:5)}$	dS m^{-1}	Conductivity meter	0.101
Salinitas $_{(H2O, 1:5)}$	mg l^{-1}		50
C-organic	%	Walkley & Black	1.33
N-total	%	Kjeldahl	0.13
C/N ration	-	-	10
P-available	ppm	Bray 1	5.4
K-available	ppm	Morgan	76
Capacity Exchangeable Cation	cmol kg^{-1}	NH_4-acetate 1N, pH7	7.94

Preparation of Biochar

Organic solid waste in the form of wood is collected from farmers' gardens in Mokoau Village, Kendari. The dried pieces of wood are cut into pieces and put into a simple pyrolysis drum for making biochar. Biochar dried under the sun's heat is pulverized using a refining machine equipped with a sieve size < 2.00 mm per pore opening hole. Biochar powder is stored in containers and placed in a protected location from water attacks until used.

Application of Biochar

Remnants of coarse roots and other materials were removed from each experimental plot. The applied biochar has a water content of 10%. Each biochar treatment was spread evenly on the soil surface, then mixed with soil particles to a depth of 10 cm from the soil surface using a hoe.

Collection of Muna Local Tomato Variety

Muna local tomatoes with light-red and red ripeness levels (Alenazi *et al*. 2020), as presented in Figure 1, were collected from local farmers' gardens in Tongkuno District, Muna Regency, Southeast Sulawesi.

Tomato Seedling

The Muna local tomatoes are large and have reached the stage of maturity as mentioned above, cut in half vertically. The pulp (septum, placenta, and seeds) was separated from the pericarp using a spoon. The fruit's flesh is mixed with fine sand and kneaded until the seeds are separated from the placenta and funiculus in a plastic container, added with water, and cultured until the sand settles. Seeds are poured over a sieve carefully. Retained seeds were rewashed using 5.25% sodium hypochlorite until all the funiculus covering the seeds was released. Furthermore, the seeds were rinsed with water until the "bayclin" disappeared and air-dried. The seeds were germinated on a tray containing a mixture of topsoil, manure, and sand in a ratio of 1:2:1 and kept at room temperature.

Two-week-old tomato seedlings were transferred to polybags containing growth media composed of a mixture of sand, wood shavings compost, and biochar in a ratio of 1:1:1, which had been given a solution of NPK fertilizer. Seedlings were

Figure 1. Local curly tomatoes from Tongkuno, Muna Regency (Photo by PKM-RE Team 2021).

maintained in a nursery with a screen shade until they were transplanted into each experimental plot.

Cropping of Tomato Seedling

Each experimental plot made planting holes with a diameter of 7 cm to a depth of 10 cm with a distance of 40 cm in rows and 50 cm between rows. Tomato seedlings aged 48 days from the nursery were transplanted into each experimental plot. The polybag was removed then the soil and the roots of the seeds were inserted into the planting hole, then covered with soil.

Application of NPK Fertilizer

Fertilizers were given at a dose of 200 kg urea (46% N) ha^{-1}, 200 kg SP36 (36%P2O5) ha^{-1}, and 100 kg KCl (60% K2O) ha^{-1}. SP-36 fertilizer was given simultaneously with biochar application, while urea and KCl were given by making circular grooves 5 cm deep and as far as 8 cm from the base of the plant at the time of planting.

Crop Maintenance

One plant was maintained for each planting hole. Pests that visited the plant parts above the soil surface were taken and preserved in sample bottles containing 70% alcohol. Weeds growing above the soil surface in each plot were removed manually. Plant watering was carried out only when there was no rain, and soil moisture was < 35% measured with Lutron PMS-714.

Sampling, Extraction, and Identification of Soil Nematode

Soil samples were taken 14 days after planting, each plot was made a zig-zag sampling point, and soil from each point was taken to a depth of 10 cm using stainless steel cylinder (7.5 cm in diameter). The soil samples were composited, and 500 g was taken, then transported to the laboratory. A total of 250 g of each soil sub-sample was extracted using the Baerman Funnel technique, which was left for 24 hours (Cesarz *et al*. 2019). The nematodes were sieved using a 38 µm per pore opening (400 mesh). The nematode samples from each plot were oven-dried for 45 minutes at 70°C and cooled. The nematodes were filtered again on a 38 µm filter and flowed 70% alcohol into a sample bottle containing TAF (Triethanolamine Formalin) and stored for 24 hours in a refrigerated room (Ryss 2017). Nematodes were counted under a Boeco stereo microscope at 40x magnification by removing the nematodes one by one using a dropper. All nematodes in each sample were placed on a slide. Then, their morphology was identified up to the family level under a light microscope at 400x magnification following the identification guidelines from Freckman and Baldwin (1990), Panesar and Marshall (2005), and Abebe *et al*. (2006). Individuals from each family were counted. The family was allocated into trophic groups following the guidelines of Yeates *et al*. (1993).

Soil Nematode Diversity Index

The calculated diversity index includes:
(a) Shannon: $H' = -\sum P_i(1nP_i)$; (b) Hills N1:N1= $e^{H'} = \exp\left[-\sum P_i(1nP_i)\right]$; (c) Simpson: $\lambda = \sum (\frac{n_i}{N})^2$; (d) Hills N2 : N2= $\frac{1}{\lambda} = \frac{1}{\sum (\frac{n_i}{N})^2}$. Note Pi= the proportion of trophic group (family); i in the total nematode community; ni= the number of individuals in the trophic group (family)-i; N= total number of all individuals in the soil nematode community (Neher *et al.* 2004).

Soil Nematode Channel Index

The channel index (CI) of the nematodes was determined following the procedure described by Ferris *et al*. (2001) using the following formula:

$$CI = 100 \times \frac{Fu_2 W_2}{Ba_1 W_1 + Fu_2 * W_2}$$

which: Ba_1 is Bacterivore cp1; Fu_2 is Fungivore cp2; W_1 is constant of cp1 (3.2); W_2 is constant of cp2 (0.8). The cp value of each nematode family was taken from Bongers and Bongers (1998).

Statistical Analysis

The relative abundance of the nematode family and trophic group and the channel index of the nematode community was evaluated through analysis of variance (ANOVA), followed by the least significant distance (LSD) at the $p < 0.05$ level. The effect of biochar rate on the total nematode abundance was evaluated. The effect of biochar rate (B) assumed has a linear relationship with the abundance of each trophic group (Y), the regression is expressed as Y = aB + c (where a is the coefficient and b is the constant), and correlations to determine the relationship between trophic groups and biochar rate (Montgomery 2001). ANOVA, LSD test, and calculating each nematode community index were carried out using EXCELL assistance. Linear regression and correlation analysis were carried out using SPSS 16.0 software free download version (https://spssdownload.com/spss-16-download-free-full-version).

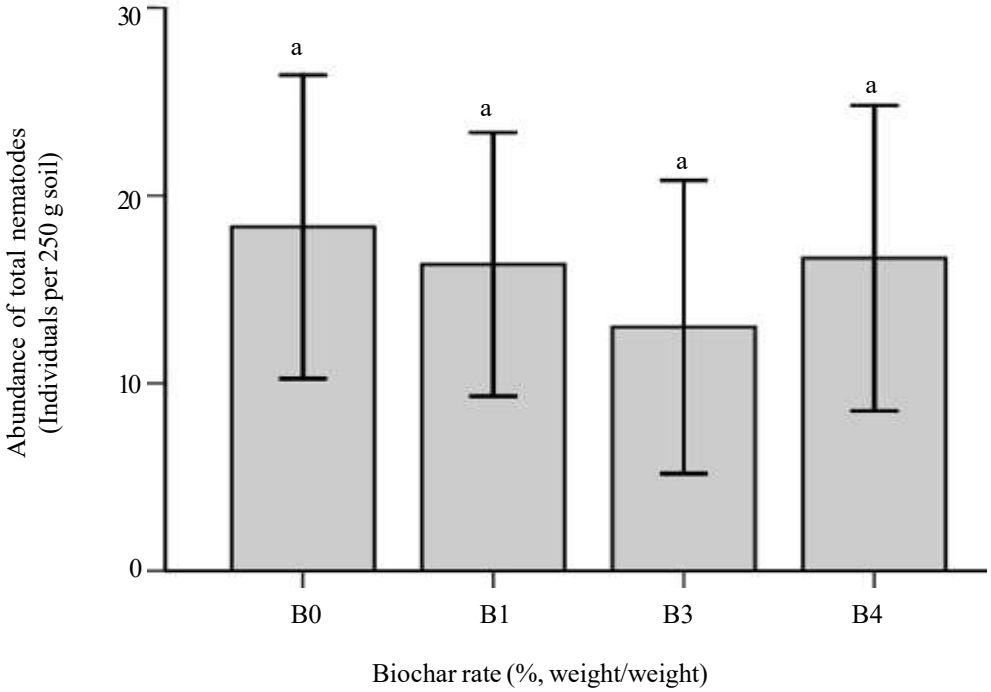

Figure 2. The total abundance of nematodes between different biochar rates. B_0 is 0% biochar, B_1 is 5% biochar, B_2 is 10% biochar, and B_3 is 15% biochar based on soil weight. The number above the bar followed by the same letter shows no significant difference according to the LSD test at the p> 0.05 level.

RESULTS AND DISCUSSION

The Abundance of Total Soil Nematodes

The results of the ANOVA showed that the dose of biochar had no significant effect (Fcalculate = 1.02 < Ftable 0.05 = 4.76, df = 3;6) on the total nematodes. The application of a 10% biochar rate (B2) decreased the abundance of total nematodes, although it was not significant at the level of p > 0.05. Figure 2 shows that the total nematodes in the B0 treatment tended to be higher than the soil that received the biochar treatment.

Relative Abundance of Nematode Families

A total of ten families of soil nematodes were found during the sampling period, namely Aphelenchoididae, Criconematidae, Desmodoridae, Longidoridae, Mononchidae, Nordiidae, Plectidae, Rhabditidae, Trichodoridae, and Tylenchidae. ANOVA showed that the effect of biochar dose on the relative abundance of Aphelenchoididae (Fcalculate = 15.85 > Ftable0.05 = 4.76, df = 3;6), and Longidoridae (Fcalculate = 13.97 > Ftable 0.05 = 4.76, df = 3;6) was significant, while the relative abundance of other families was not significant (Fcalculate< Ftable 0.05 = 4.76, df = 3;6). Table 2 shows that the highest relative abundance of Aphelenchoididae occurred in B2 treatment, which was significantly different from the others, while the difference in relative abundance between other families was not significant (LSD test at the p > 0.05 level). On the other hand, the lowest relative abundance of Longidoridae occurred in the B2 treatment, which was significantly different (LSD test at level p < 0.05) compared to other treatments.

Diversity of Soil Nematode

ANOVA showed that the effect of biochar dose on the size of soil nematode community diversity was not significant (Fcalculate< Ftable0.05 = 4.76, df = 3;6). Table 3 shows that the differences in family taxon Shannon index, Simpson diversity index, Hills N1 index, and Hills N2 index among the four treatments are not significant (LSD test at the p> 0.05 level).

Relative Abundance of Trophic Groups

A total of five trophic groups of soil nematodes were found, namely root-feeders, fungivores, predators, bacterivores, and omnivores (Table 4). ANOVA showed that biochar dose had a significant effect on the relative abundance of root-feeding nematodes (Fcalculate = 7.82 > Ftable0.05 = 4.76, df = 3;6) and fungivores (Fcalculate = 12.81 > Ftable0.05 =

Table 2. Relative abundance (mean±sd, n = 3) of soil nematode families with different biochar rates in tomatoes cropping of the Muna local variety.

Family	Trophic group	cp value	Relative abundance of the nematode family on different biochar rate			
			B_o	B_1	B_2	B_3
Aphelenchoididae	Fu	2	10.95 ± 2.18a	16.48 ± 15.06a	49.28 ± 11.58b	9.44 ± 5.78a
Criconematidae	Rf	4	6.60 ± 5.84a	3.92 ± 6.79a	3.03 ± 5.25a	4.31 ± 4.56a
Desmodoridae	Ba	3	10.14 ± 9.05a	5.66 ± 5.56a	7.41 ± 12.83a	3.85 ± 6.66a
Longidoridae	Rf	5	46.70 ± 8.91b	40.78 ± 7.25b	10.90 ± 1.71a	33.92 ± 6.15b
Mononchidae	Pr	4	16.10 ± 6.85a	13.93 ± 3.36a	18.10 ± 6.94a	23.19 ± 4.02a
Nordiidae	Om	4	0.00 ± 0.00a	1.45 ± 2.51a	0.00 ± 0.00a	4.31 ± 4.56a
Plectidae	Ba	2	3.70 ± 6.42a	0.00 ± 0.00a	1.52 ± 2.62a	5.59 ± 4.90a
Rhabditidae	Ba	1	2.90 ± 2.51a	6.60 ± 5.84a	3.03 ± 5.25a	7.69 ± 7.69a
Trichodoridae	Rf	3	1.45 ± 2.51a	4.86 ± 4.44a	5.22 ± 5.59a	3.85 ± 3.85a
Tylenchidae	Rf	2	1.45 ± 2.51	6.31 ± 6.53a	1.52 ± 2.62a	3.85 ± 6.66a

Remarks: Numbers followed by different letters in the same row showed significant differences according to the LSD test at the level of $p < 0.05$. B_0 is 0% biochar, B_1 is 5% biochar, B_2 is 10% biochar, and B_3 is 15% biochar based on soil weight. Rf = Root feeder, Fu = fungivore, Pr = predator, Ba = bacterivore, and Om = omnivore.The number following the label of each trophic group is the c-p value of the nematode family, according to Bongers and Bongers (1998).

Table 3. Diversity index (mean±sd, n = 3) soil nematodes on tomato cropping of the Muna local variety with different biochar rates.

Biochar rate	Diversity index				
	R	*H'*	N1	λ	N2
B_0	6.0 ± 1.00a	1.48 ± 0.19a	4.4 ± 0.86a	0.29 ± 0.07a	3.5 ± 0.83a
B_1	6.3 ± 1.15a	1.57 ± 0.11a	4.8 ± 0.51a	0.26 ± 0.02a	3.9 ± 0.32a
B_2	5.3 ± 2.52a	1.36 ± 0.43a	4.1 ± 1.65a	0.33 ± 0.12a	3.3 ± 1.11a
B_3	7.7 ± 1.53a	1.71 ± 0.21a	5.6 ± 1.26a	0.22 ± 0.04a	4.8 ± 1.09a

Remarks: B_0 is 0% biochar, B_1 is 5% biochar, B_2 is 10% biochar, and B_3 is 15% biochar based on soil weight. *R* is family richness, H' is Shannon index, λ is Diversity Simpson index, N1 is Hills N1 index, N2 is Hills N2 index. Numbers followed by the same letter in the same column show no significant difference according to the LSD test at the $p < 0.05$ level.

Table 4. Relative abundance (mean±sd, n = 3) of trophic groups on tomato cropping of the Muna local variety with different biochar rates.

Trophic groups	Relative abundance of the trophic group on the different biochar rate			
	B_o	B_1	B_2	B_3
Bacterivore	16.747 ± 5.34a	12.27 ± 8.75a	11.95 ± 11.21a	17.13 ± 11.04a
Fungivore	10.95 ± 2.18a	16.48 ± 15.06a	49.28 ± 11.58b	9.44 ± 5.78a
Omnivore	0.00 ± 0.00a	1.45 ± 2.51a	0.00 ± 0.00a	4.31 ± 4.56a
Root feeder	56.20 ± 4.38b	55.87 ± 19.67b	20.67 ± 7.51a	45.92 ± 0.40b
Predator	16.10 ± 6.85a	13.93 ± 3.36a	18.10 ± 6.94a	23.19 ± 4.02a

Remarks: B_o is 0% biochar, B_1 is 5% biochar, B_2 is 10% biochar, and B_3 is 15% biochar based on soil weight. Numbers followed by different letters in the same row are significantly different according to the LSD test at the $p < 0.05$ level.

4.76, df = 3;6) , while the relative abundance of other trophic groups was not significant (Fcalculate< Ftable 0.05 = 4.76, df = 3;6). Table 3 shows that the highest relative abundance of root-feeding nematodes occurred in B0 treatment, but compared to B1 and B3 treatments, there was no significant difference (according to the LSD test at the level of $p < 0.05$). The highest relative abundance of fungivorous nematodes occurred in B2 treatment, and compared to the others, it was significantly different (according to the LSD test at the level of $p < 0.05$). The difference in relative abundance between other trophic groups was not significant (according to the LSD test at level $p > 0.05$).

Channel Index

ANOVA showed that the biochar dose had no significant effect (Fhit< Ftab0.05 = 4.76, db = 3;6)

Table 5. Channel index (mean±sd, n = 3) of soil nematodes communities on tomato cropping of the local Muna variety with different biochar rates.

Biochar rate	Channel index (CI)
B_0	58.73 ± 36.06a
B_1	51.323 ± 45.045a
B_2	84.31 ± 51.32a
B_3	46.40 ± 45.04a

Remarks: B_0 is 0% biochar, B_1 is 5% biochar, B_2 is 10% biochar, and B_3 is 15% biochar based on soil weight. Numbers followed by different letters in the same row are significantly different according to the LSD test at the $p < 0.05$ level.

on the channel index in the soil nematode community in the food web. Table 5 shows that the difference in the channel index value between the four treatments was not significant (LSD test at level $p < 0.05$).

The Relationship of Biochar Rate with Trophic Groups and Between the Trophic Groups

The linear regression analysis of biochar rate and relative abundance of each trophic group nematode are shown in Figure 3. The results showed that relative abundances of a trophic group nematode were not significantly correlated with biochar rate. The relative abundances of bacterivorous (Figure 3A), fungivorous (Figure 3B), omnivore (Figure 3C), and predatory (Figure 3E) nematodes were increased with an increase of biochar rate. In contrast, the relative abundance of root-eating nematodes was decreased with an increase in biochar rate (Figure 3D). The maximum determination coefficient (R2) of biochar rate and relative abundance of predatory nematode is 0.251 (Figure 3E).

Analysis of Spearman rho correlation between biochar rate and nematode trophic groups and between trophic groups are listed in Table 6. The negative correlation between biochar rate and relative abundance of root-feeding nematodes was significant (Spearman rho = -0.519, sig. = 0.042), with predators positively correlated. (Spearman rho = -0.573, sig. = 0.026), while with other trophic groups, it was not significant (sig. > 0.05). The relative abundance of fungivorous nematodes was negatively correlated with root-feeders (Spearman rho = -0.753, sig. = 0.002) and to omnivores (Spearman rho = -0.661, sig. = 0.010).

Discussion

The addition of biochar to the soil aims to increase the organic matter content of the soil (Hua

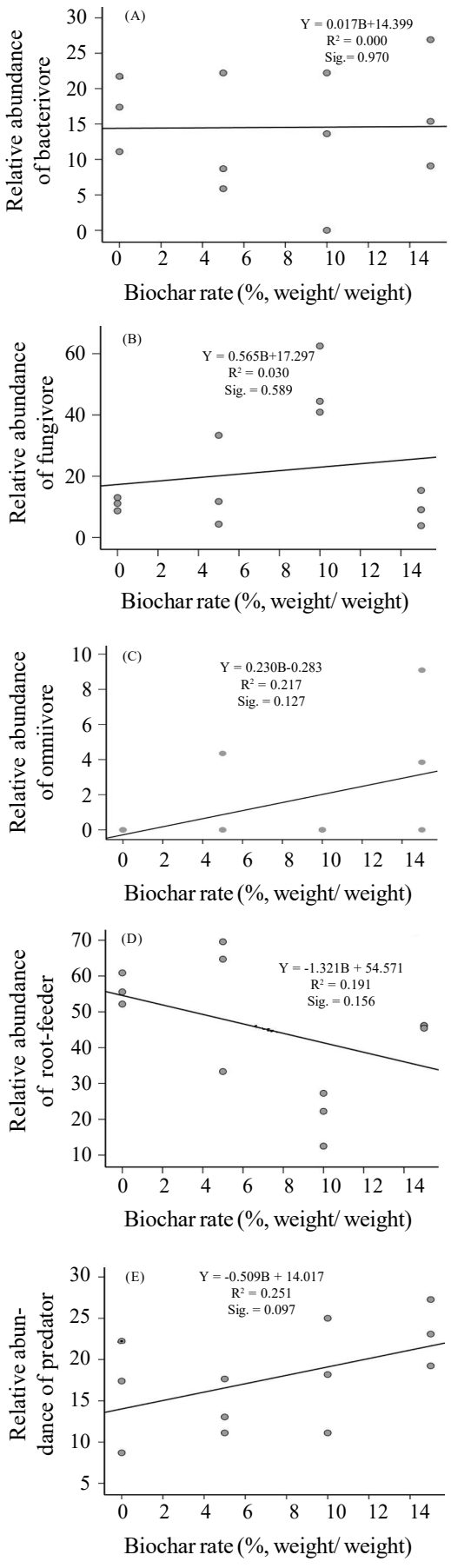

Figure 3. Linear regression between biochar rate and relative abundance of a trophic group.

et al. 2014; Joseph *et al*. 2019). Increasing soil organic matter content by adding biochar to the soil significantly reduced total soil nematodes (Liu *et al*. 2020a). Although not significant, the results of our study found that the total abundance of nematodes tended to decrease with biochar level. The decreasing trend occurred until the biochar rate was 10% of the soil weight, then above that rate, the abundance of the total nematodes increased again (Figure 1). In this study, the difference in total nematode abundance between biochar rates was insignificant. Our findings are consistent with previous studies that the abundance of total soil nematodes was not significantly different from the biochar rate (Domene *et al.* 2021). The phenomenon of an increase in the total abundance of nematodes at biochar levels above 10% is probably related to the increased abundance of nematode taxa that access their carbon needs through fungal energy pathways, and nematodes are tolerant to disturbance and reproduce quickly (Dai *et al.* 2021; Melakeberhan *et al.* 2021).

The response of soil nematode communities to adding biochar from wood can be demonstrated by changes in taxa or trophic groups (Cole *et al.* 2021). Our study found that the relative abundances of nematodes Aphelenchoididae and Longidoridae between biochar levels were significantly different, while between the other eight families were not significant. The relative abundance of Aphelenchoididae increased to 10% biochar level, then decreased above that level. In contrast, the relative abundance of Longidoridae decreased, then again increased above this rate (Table 2). The pattern of increasing abundance of the two soil nematode families with increasing levels of biochar compared to the findings of Zhang *et al.* (2013) on the nematodes Aphelenchoides, Hirschmanniella, and Tylenchus were similar. The abundance response of the two nematode families may be related to changes in soil character after application of biochar, life history, and reproductive strategies of nematodes (Quist et al. 2019; Nguyen *et al.* 2020). Adding biochar to acidic mineral soils increases soil pH and organic carbon (Adekiya et al. 2019). The abundance of soil nematodes from groups of fungivores c-p 2 and 4, as well as root-eating nematodes c-p5, were positively correlated with pH, organic C, and total soil N (Renco *et al.* 2020). Aphelenchoididae is categorized into fungivores and Longidoridae into root eaters/plant parasites based on their eating habits. Aphelenchoididae has a c-p value of 2, and Longidoridae has a c-p value of 5 (Table 2). The Aphelenchoididae can colonize and are resistant to pH levels, organic matter content, and total N created by adding biochar up to 10% of the soil weight.

On the other hand, Longidoridae can colonize and are resistant to soil environmental conditions created by adding biochar above the 10% level. The decrease in the relative abundance of Aphelenchoididae nematodes in biochar above the 10% level may be related to the increased availability of unstable organic C in soils more accessible to fungi (Xu *et al.* 2020). As a result, more fungal biomass is available for access by fungivorous nematodes, including Aphelenchoididae. This increase in the nematode population triggers the predation rate of fungivorous nematodes of the soil nematode communities in the food web (Mayrhofer *et al*. 2021). This decrease in the relative abundance of Aphelenchoididae opens up opportunities for increasing populations of Longidoridae (Krashevska *et al.* 2019).

Taxonomic group-based nematode community characteristics (including taxa richness and diversity index) have been recognized as an ecological tool for analyzing the effects of disturbance (including biochar addition) on soil nematode community structure and function (Urzelai et al. 2000). In our study, five measures of soil nematode community diversity were analyzed, namely family richness (R), Shannon index (H'), Simpson's diversity index (λ), Hills N1 index (N1), and Hills N2 index (N2). Our results found that the differences in family richness and the value of each diversity index between biochar doses were not significant (Table 3). This fact indicates that the addition of biochar has not been able to significantly change the composition of the soil nematode community, although the abundance of Aphelenchoididae and Longidoridae showed significant changes. These results may be related to the quality of available food that is accessible to nematodes between biochar levels, which are relatively similar (Eisenhauer *et al.* 2011), so that they have not been able to change the number of dominant families in the soil nematode community at each dose of biochar (Freckman and Ettema 1993; Kilowasid *et al.* 2013). This assumption is clarified by the results in Table 2, where the most dominant family was not seen in each biochar dose. The families and individuals in the soil nematode community are more distributed (Morin 2011).

The relative abundance of trophic groups is often used to measure the nematode community's response to changes in food availability and conditions in the agricultural soil environment (Lazarova et al. 2021). Our results found that the relative abundances of fungivorous and root-eating nematodes between biochar levels differed significantly, while the other

trophic groups were not significant (Table 4). These results reaffirm the previous study by Domene *et al.* (2021) that biochar levels promote bacterivorous and fungivorous nematodes while suppressing root eaters (herbivores). The linear regression in our results explained that the relative abundances of fungivores, omnivores, and predators tended to increase (Figures 3B, 3C, and 3E), whereas root feeders decreased with biochar levels. The increase of bacterivores was relatively small (Figure 3A) compared to other trophic groups. Our findings from the Spearmen rho correlation indicate a negative relationship between biochar levels and the relative abundance of root eaters, in contrast to a positive relationship with predators. Fungivores showed a negative relationship between trophic groups with root eaters and omnivores (Table 6). These facts indicate that the biochar level controlled the abundance of root-eating nematodes by increasing the population of fungivores. This correlation also illustrates that an increase in the relative abundance of omnivores prevents the dominance of fungivores.

The channel index (CI) describes the ratio of fungivorous nematodes to bacterivores, suggesting an energy pathway that contributes significantly to the decomposition and mineralization of hearts through soil nematode food webs (Kergunteuil *et al.* 2016). Although the CI values between biochar levels were not significantly different (Table 5), the table illustrates that the CI values in treatment B0, B1, and B2 were greater than 50, then decreased to <50 in treatment B3. This fact explains that the ratio of fungivorous nematodes to bacterivores in B0, B1, and B3 is higher than in B3. A CI value above 50 indicates that decomposers are predominated by fungi, while a CI value below 50 is dominated by bacteria (Liu *et al.* 2020b). The CI value in treatment B2 was the highest (CI = 84.31); this means that the availability of food for fungivorous nematodes in treatment B2 (10% biochar) was higher than in other treatments. The CI value in the B3 treatment was below 50 (CI = 46.40); this means that sufficient food resources are available for the decomposition process and nutrient mineralization through the bacterial energy pathway. The bacterial energy pathway explains that decomposition and nitrogen mineralization rate is faster than the fungal energy pathway (Holtkamp *et al.* 2011; Kilowasid *et al.* 2014). Increasing the nitrogen mineralization rate will have implications for increased nitrogen availability in the soil solution so that bacterial biomass increases and has a significant impact on fungal predation (Carrillo *et al.* 2016).

CONCLUSIONS

Application of biochar up to 10% rate of soil weight decreased the abundance of total nematodes, family Longidoridae (root eaters), on the other hand, increased Aphelenchoididae (fungivores). The biochar rate was positively correlated with the abundance of predatory nematodes. There was a negative relationship between the abundance of fungivorous nematodes with root-feeders and omnivores; on the contrary, a positive relationship with the abundance of predators.

ACKNOWLEDGMENTS

We would thank to Direktorat Pembelajaran dan Kemahasiswaan, Direktorat Jendral Pendidikan Tinggi, Kementrian Pendidikan, Kebudayaaan, Riset, dan Teknologi Republik Indonesia who funded this research through Program Kreativitas Mahasiswa 5 Bidang dan PKM GFK Tahun 2021, with contract number: 032/E2/PPK/SPPK/PKM/2021. Also to Mr. Zainuddin, secretary of Kelompok Tani Nanga-Nanga Makmur, and anyone who has helped in the field and laboratory.

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