## **RESEARCH NOTE/NOTA INVESTIGATIVA**

# NEMATODE COMMUNITYASSOCIATED WITH RECENTLY PLANTED BREADFRUIT, ARTOCARPUS ALTILIS, RHIZOSPHERE ON KAUA`I, HAWAI`I

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## ABSTRACT

Marahatta, S. P. 2018. Nematode community associated with recently planted breadfruit, *Artocarpus altilis*, rhizosphere on Kaua'i, Hawai'i. Nematropica 48:179-185.

Although, breadfruit, Artocarpus altilis (Parkinson) Fosberg, is a traditional staple food crop in the Pacific Islands, the nematode community associated with a recently planted breadfruit rhizosphere is not well documented. A field at the Kaua'i Community College was therefore prepared and divided into 4 equal blocks, and 64 breadfruit saplings were planted in December 2014. Soil samples were taken from four blocks before breadfruit transplanting (year 0) and the rhizospheres of half of the plants of each block at 1, 2, and 3 years after breadfruit tree planting. Nematodes were extracted from the soil using the Baermann funnel technique and identified to genus. Frequency of occurrence (%) of each genus was determined. Each genus was grouped into a respective trophic group. Compared to year 0, 1, and 2, the greatest herbivores numbers were found in year 3 (P < 0.01). Pooled data of years 1, 2, and 3, nematode genera were observed including bacterivores, fungivores, omnivores, and predators (beneficial nematodes), and herbivores (40.0%, 17.77%, 15.55%, 15.55%, and 11.11%, respectively). Beneficial nematodes constituted 40 genera. The most frequent genus was Cephalobus ( $95.83\% \pm 02.76\%$ ) followed by other Rhabditidae ( $82.29\% \pm 04.54\%$ ). Helicotylenchus, Paratylenchus, Axonchium, Pratylenchus, and *Xiphinema* were observed in 71.88%  $\pm$  08.27%, 29.17%  $\pm$ 12.01%, 14.58%  $\pm$  06.34%, 05.21%  $\pm$  01.04% and  $01.04\% \pm 01.04\%$  soil samples, respectively. A positive relationship existed between the total herbivores number in breadfruit rhizosphere and year after breadfruit planting. Current findings may serve as a reference for future researchers, extension workers, and farmers.

Key words: bacterivores, Cephalobus, fungivores, Helicotylenchus, herbivores, omnivores, predaceous, Rhabditidae

#### RESUMEN

Marahatta, S. P. 2018. La comunidad de nematodos asociada con la fruta de pan recientemente sembrada, *Artocarpus altilis*, rizosfera en Kaua`i, Hawai`i. Nematropica 48: 179-185.

Aunque la fruta del pan, Artocarpus altilis (Parkinson) Fosberg, es un cultivo alimenticio básico tradicional en las Islas del Pacífico, la comunidad de nematodos asociada con una rizosfera de la fruta del pan recientemente plantada no está bien documentada. Por lo tanto, se preparó un campo en el Colegio Comunitario de Kauai y se dividió en 4 bloques iguales, y se plantaron 64 plantones de fruta de pan en diciembre de 2014. Se tomaron muestras de suelo de cuatro bloques antes del trasplante de fruta de pan (año 0) y las rizósferas de la mitad de Plantas de cada bloque a 1, 2 y 3 años después de la siembra de árboles de pan. Los nematodos se extrajeron del suelo mediante la técnica del embudo de Baermann y se identificaron al género. Se determinó la frecuencia de aparición (%) de cada género. Cada género fue agrupado en un grupo trófico respectivo. En comparación con el año 0, 1 y 2, los mayores números de herbívoros se encontraron en el año 3 (P < 0.01). Los datos agrupados de los años 1, 2 y 3 se observaron géneros de nematodos, incluidos bacterívoros, fungívoros, omnívoros y predáceos (nematodos benéficos), y herbívoros (40.0%, 17.77%, 15.55%, 15.55% y 11.11%, respectivamente). Los nematodos benéficos constituyeron 40 géneros. El género más frecuente fue Cephalobus ( $95.83\% \pm 02.76\%$ ) seguido de otros Rhabditidae (82.29% ± 04.54%). Helicotylenchus, Paratylenchus, Axonchium, Pratylenchus y Xiphinema se observaron en 71.88%  $\pm$  08.27%, 29.17%  $\pm$  12.01%, 14.58%  $\pm$  06.34%, 05.21%  $\pm$  01.04% y 01.04%  $\pm$ 01.04% muestras de suelo, respectivamente. Existió una relación positiva entre el número total de herbívoros en la rizosfera de la fruta del pan y el año después de la siembra de la fruta del pan. Los hallazgos actuales pueden servir como referencia para futuros investigadores, trabajadores de extensión y agricultores.

Palabras clave: bacterívoros, Cephalobus, fungívoros, Helicotylenchus, herbívoros, omnívoros, depredadores, Rhabditidae

Breadfruit, Artocarpus altilis (Parkinson) Fosberg, is a traditional staple food crop grown for its starchy fruit throughout Oceania, which is popular throughout the tropics (Jones et al., 2011). Multiple plant-parasitic nematodes associated with breadfruit trees grown in other tropical areas, such as Jamaica, are documented (Coates-Beckford and Pereira, 1992). Moreover, several plant-parasitic nematodes such as Pratylenchus coffeae, Helicotylenchus erythrinae, H. multicinctus, and Meloidogyne incognita are reportedly associated with breadfruit tree decline symptoms in Jamaica (Coates-Beckford and Pereira, 1992). Breadfruittree-decline-associated characteristics such as premature fruit drop and leaf chlorosis (Coates-Beckford and Pereira, 1992) have been observed on breadfruits grown in Hawai'i (Helen A. Cox, personal communication). This could be due to the attack of multiple plant-parasitic nematodes on breadfruit and a factor in breadfruit tree decline in Hawai'i. Recently, Pratylenchus coffeae has been reported as a breadfruit associated plant-parasitic nematode in Hawai'i (Lau et al., 2018), and there might be multiple plant-parasitic nematodes associated with breadfruit trees in Hawai'i.

Plant-parasitic and beneficial nematodes are members of the soil community. Beneficial

nematodes such as bacterivores and fungivores are involved in soil nutrient cycling and help to create a healthier soil environment (Wang and McSorley, 2005). Additionally, beneficial predaceous nematodes can be used to control populations of plant-parasitic nematodes (Lambert and Bekal, 2002). However, a list of plant-parasitic and beneficial nematodes found on breadfruit plants grown in Hawai'i is lacking. Therefore, the objectives of research were to: (i) determine the plant-parasitic and beneficial nematodes in a breadfruit orchard on Kaua'i, Hawai'i, and (ii) evaluate the relationship between the plantparasitic nematodes and year of breadfruit planting.

An uncultivated field covered with Albizia trees and grasses at the University of Hawai'i, Kaua'i Community College was cleared, tilled, leveled, and 64 breadfruit, *A. altilis* 'Ma'afala', saplings were planted in December 2014. The orchard was named Ulutopia–'ulu Hawaiian for breadfruit and topia—a place with specified characteristics. The soil at the Ulutopia is a Puhi series silty clay loam (very-fine, ferruginous, isohyperthermic Humic Kandiustox) with a pH of 6.2. Ulutopia-grown breadfruit trees were irrigated immediately after transplanting, and field irrigation was continued for 9 months. Ten months after transplanting, field irrigation was discontinued.

Ulutopia was divided into 4 blocks with 16 breadfruit plants in each block. The day before breadfruit transplanting (year 0), 6 soil cores at 15cm deep were systematically collected from each block, composited into one sample/block, separately packed, and shipped to Earthfort Lab (Earthfort, Corvallis, OR) for biological analysis. The weight of each composite soil sample was ~250 g. After breadfruit transplanting, four soil cores at 15-cm deep were collected from four sides of breadfruit rhizospheres and composited into one sample. Eight composite soil samples from breadfruit plant rhizospheres of each block were taken separately at 1, 2, and 3 years after breadfruit transplanting. Annually (at year 1, 2, and 3), 32 soil samples were separately packed and shipped to Earthfort Lab for biological analysis.

Data for plant-parasitic and beneficial nematodes at years 0, 1, 2, and 3 were subjected to one-way analysis of variance (ANOVA) using the general linear model (GLM) procedure in Statistical Analysis System (SAS Institute, Cary, NC). Nematode abundance data were log(x + 1)-transformed. Only untransformed arithmetic means are presented. Means were separated by Waller-Duncan *k*-ratio (*k*=100) *t*-test wherever appropriate.

Frequency of reported nematodes was calculated from the pooled data of years 1, 2, and 3. Nematodes were grouped into trophic groups (Yeates *et al.*, 1993), and assigned colonizer-

persister (c-p) values (Bongers and Bongers, 1998).

The number (dependent variable) of plantparasitic nematodes (herbivores) was regressed on the year after breadfruit transplanting (independent variable) using the REG procedure (MODEL 1) in Statistical Analysis System (SAS Institute, Cary, NC). The estimated parameters, intercept and year, were used, and the straightline equation of the fitted model was presented.

Compared to year 0, greater trends of the herbivore numbers were recorded in years 1, 2, and 3, and more herbivores were found in year 3  $(P \le 0.01)$  (Table 1). This positive effect of host plant rhizosphere on herbivores over time is consistent with the findings of Toida (1991), who found consistently greater number of herbivorous nematode, Meloidogyne mali, in the plant rhizosphere in years 1, 2, and 3 after root inoculation of *M. mali* in mulberry in year 0. On the other hand, total number of beneficial nematodes recorded at all sampling times except year 1, were similar (P > 0.05) (Table 1), which might be due to the growth of similar flora such as grasses into the soil throughout the experiment period.

Out of the 20 nematode genera, 9 (45%) bacterivores (*Panagrolaimus*, Rhabditidae, *Rhabdolaimus*, Acrobeles, Cephalobus, Eucephalobus, Geomonhystera, Monhystrella, and Prismatolaimus), 5 (25%) fungivores (*Tylencholaimus*, Aphelenchoides, Aphelenchus, Ditylenchus, and Filenchus), 2 (10%) herbivores (*Pratylenchus* and Helicotylenchus), 3 (15%)

Kaua'i, Hawai'i. Nematode number/100 g soil ± SEM<sup>z</sup>

Table 1. Total herbivores and beneficial nematodes associated with soil from the breadfruit rhizoshere before (year 0) and after (year 1-3) breadfruit transplanting in

	Nematode number/100 g soil $\pm$ SEM <sup>z</sup>		
Year	Herbivores	Beneficial	
0	$26 \pm 3.67 \text{ B}$	$690 \pm 260.08 \text{ A}$	
1	$47 \pm 22.77 \text{ BC}$	294 ± 53.11 B	
2	$207 \pm 61.80 \text{ AB}$	1145 ± 179.75 A	
3	$198 \pm 34.90 \text{ A}$	505 ± 71.98 A	

<sup>z</sup> Means are average of 4 replications (blocks). Each replication had 1 sample in year 0, and 8 samples in years 1, 2, and 3. Standard error of means (SEMs) are calculated from the respective samples. Means in a column followed by the same letter(s) do not differ according to Waller-Duncan *K*-ratio (K=100) *t*-test.

Trophic Group <sup>x</sup>	Genera	c-p value <sup>y</sup>	Frequency of occurrence (%±SEM) <sup>z</sup>
Bacterivores	Cephalobus	2	95.83±02.76
	Rhabditidae	1	82.29±04.54
	Panagrolaimus	1	41.67±10.57
	Plectus	2	37.50±06.25
	Prismatolaimus	3	32.29±11.74
	Eucephalobus	2	20.83±08.14
	Monhystrella	2	14.58±02.76
	Acrobeles	2	12.50±01.80
	Wilsonema	2	12.50±01.80
	Diploscapter	1	$10.42 \pm 08.90$
	Cervidellus	2	$10.42 \pm 02.76$
	Geomonhystera	2	$07.29 \pm 04.17$
	Alaimus	4	04.17±02.76
	Prodesmodora	3	03.13±03.13
	Rhabdolaimus	3	03.13±01.80
	Discolaimium	4	03.13±01.80
	Diplogasteritus	1	01.04±01.04
	Zeldia	2	$01.04\pm01.04$
	Cephalobus	2	95.83±02.76
	Rhabditidae	1	82.29±04.54
	Khabuttade	1	02.27=04.34
Fungivores	Aphelenchoides	2	76.04±03.76
	Aphelenchus	2	61.46±02.76
	Filenchus	2	59.38±04.77
	Ditylenchus	2	56.25±03.61
	Diphtherophora	3	20.83±08.53
	Tylencholaimus	4	$07.29 \pm 05.80$
	Malenchus	3	$02.08 \pm 02.08$
	Psilenchus	2	01.04±01.04
Herbivores	Helicotylenchus	3	71.88±08.27
TICI UIVUICS	Paratylenchus	3	$29.17 \pm 12.01$
	Axonchium	5	$14.58 \pm 06.34$
	Pratylenchus Vinkin om a	3 5	05.21±01.04
	Xiphinema	5	01.04±01.04
Omnivores	Eudorylaimus	4	39.58±12.01
	Aporcelaimellus	5	18.75±06.25
	Épidorylaimus	4	09.38±03.13
	Microdorylaimus	4	08.33±02.76
	Thonus	4	$04.17 \pm 04.17$
	Mesodorylaimus	4	02.08±01.04
	Prodorylaimus	4	$01.04 \pm 01.04$
	Eudorylaimus	4	$39.58 \pm 12.01$
	Aporcelaimellus	5	18.75±06.25
	Epidorylaimus	4	09.38±03.13
	Microdorylaimus	4	08.33±02.76

Table 2. Nematodes associated with soil from the breadfruit rhizoshere in Kaua'i, Hawai'i with their trophic group and colonizer-persister (c-p) values.

Tuble 2: Continued.				
Trophic Group <sup>x</sup>	Genera	c-p value <sup>y</sup>	Frequency of occurrence (%±SEM) <sup>z</sup>	
	Ochicia	e-p value	(/0±5L1v1)	
Predaceous	Punguntes	4	$26.04 \pm 26.04$	
	Mylonchulus	4	19.79±07.29	
	Tobrilus	3	02.08±01.04	
	Clarkus	4	02.08±01.04	
	Thonus	4	$02.08 \pm 02.08$	
	Tripyla	3	$01.04 \pm 01.04$	
	Carcharolaimus	4	$01.04 \pm 01.04$	
	Punguntes	4	26.04±26.04	
	Mylonchulus	4	19.79±07.29	
	Tobrilus	3	02.08±01.04	
	Clarkus	4	02.08±01.04	

Table 2	Continued.
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<sup>x</sup>Nematode trophic groups are categorized as described by Yeates *et al.* (1993).

<sup>y</sup>Nematode c-p values are assigned as described by Bongers and Bongers (1998).

<sup>z</sup>Frequency of occurrence (%) is calculated from the soil analysis report of 96 soil samples taken in 2015, 2016, and 2017.

omnivores (Paraxonchium, Eudorylaimus, and Mesodorylaimus), and 1 (5%) predaceous (Clarkus) nematodes were observed before transplanting breadfruit trees in the soil. After transplanting breadfruit trees, 45 nematodes were observed in association with the breadfruit rhizosphere (Table 2). Bacterivores, fungivores, herbivores, omnivores, and predaceous genera accounted for 40.0% (N=18), 17.77% (N=8), 11.11% (N=5), 15.55% (N=7), and 15.55% (N=7), respectively. Out of the 18 observed bacterivorous nematodes, the most frequent genus was Cephalobus followed by other genera in the Rhabditidae. The most frequent fungivorous nematode was Aphelenchoides followed by Helicotylenchus, Eudorylaimus, Aphelenchus. and Punguntes were the most frequent herbivorous. omnivorous, and predaceous nematodes, respectively. All omnivorous and predaceous nematodes and 26 other nematodes were observed on <60% samples (Table 2). Occurrence of herbivororus nematodes, Helicotylenchus, Pratylenchus, and Xiphinema. and fungivorous nematodes, Aphelenchoides, Aphelenchus, and Ditylenchus, was consistent with the findings of Coates-Beckford and Pereira (1992), who had conducted a survey on breadfruit microorganisms, including nematodes, in Jamaica.

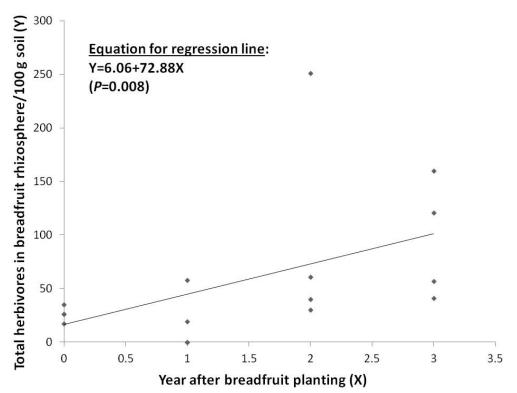
A positive relationship existed between the total herbivores number in breadfruit rhizosphere (Y) and year after breadfruit planting (X) (correlation coefficient (r) = 0.26). The equation for regression between Y and X was Y=

6.06+72.88 X (P < 0.01) (Fig. 1). This equation is applicable for predicting the total herbivores in breadfruit rhizosphere in Hawai'i over the time. In Jamaica. 319 herbivores (Helicotylenchus, Hoplolaimus, Longidorus, Meloidogyne, Pratylenchus, Rotylenchulus, Trichodorus, and Tylenchorhynchus) were recorded in the rhizosphere of declining breadfruits (Coates-Beckford and Pereira, 1992). Regression equation predicts that in 4.29 years, there will be an equal number of total herbivores in rhizospheres of Ulutopia grown breadfruits and the populations of nematodes reportedly associated with declining breadfruit trees in Jamaica (Coates-Beckford and Pereira, 1992).

The current research is the first report of the nematode community associated with breadfruit rhizosphere in Hawai'i, USA. This report could serve as a reference document for future researchers. Moreover, plant-parasitic nematodes, *Helicotylenchus, Paratylenchus, Axonchium, Pratylenchus,* and *Xiphinema* may develop as challenging pest(s) in the future because of the availability of perennial breadfruit host. Therefore, researchers and agriculture extension workers should consider conducting periodic soil nematode analysis of breadfruit rhizosphere and updating their findings.

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**Fig. 1**. Regression between total herbivores in breadfruit rhizosphere (Y) and year of breadfruit growing in Kaua'i, Hawai'i in 2014-2017 (X). This analysis is based on herbivore numbers on composite soil samples (100 g/sample) from four blocks (replications)/year. Each replication had 1 sample in year 0, and 8 samples in years 1, 2 and 3.

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