

Original Research Article

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Banana (*Musa* AAA cv Williams) Response to Chemical and Biological Nematicides

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ABSTRACT

The effect of the rotation of chemical and biological nematicide cycles per year on banana (*Musa* AAA cv. Williams) root weight, root nematode control and crop yield were compared in a commercial banana plantation in Costa Rica testing six treatments in a randomized complete block design with six replicates. Treatments consisted of the rotation of one, two or three chemicals or even the rotation of four biological nematicide cycles per year or the rotation of alternating chemical and biological nematicides in the year plus the untreated control. Averaging the 27 root nematode samplings after treatments application, the rotation of three chemical nematicide cycles by year reduced *R. similis* (P= 0.0005) by 44.5%, *Helicotylenchus* spp. (P = 0.0005) by 49.4% and total nematodes (P< 0.0001) by 45.3%. In contrast, the rotation of four biological nematicide cycles by year only drops not significantly *R. similis* by 6.9%, *Helicotylenchus* spp. by 9% and total nematode by 7.3%. Then, it seems that effective biological nematicides are not yet available for bananas, despite a substantial, positive literature on this topic. The number of uprooting plants was higher (P< 0.0001) in the untreated plots, with 101 during the experimental period, which would be equivalent between 161 and 188 uprooted plants by hectare that would end in a lost between 303 and 354 boxes by hectare by year. Even though in the second ratoon crop cycle, the rotation of three chemical nematicide cycles by year gave 308 more boxes per hectare (5.5 mt) than the untreated plots, the difference was not large enough to be significant (P= 0.2177). Difference in yield started at the third (P= 0.0405), and was increased at the fourth (P= 0.0009) ratoon crop, and final harvest (P= 0.0047), where the rotation of three chemical nematicide cycles by year improved yield by 506 (9.1 mt), 840 (15.2 mt) and 791 (14.3 mt) boxes by hectare by year, which resulted in a net profit (deducted the treatment cost and the packing cost of the additional boxes) of US \$2,131; \$3,743 and \$3510 ha⁻¹ year⁻¹, respectively. Such improvement in yield, over the untreated plots, should be higher, because it did not consider the yield lost by uprooted plants.

Keywords

Banana, chemical nematicides, *Helicotylenchus* spp, *Radopholus similis*

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Introduction

Abiotic (soil texture, wind, radiation, temperature, rain) and biotic (black Sigatoka, nematodes, black weevil) factors affect banana (*Musa* AAA) production. Within the root pests, nematodes (*Radopholus similis*, *Helicotylenchus* spp., *Meloidogyne* spp. and *Pratylenchus* spp.) are the main problem. Araya and Vargas (2018) in an analysis done of the root samples collected during the years 2000 to 2008, found high populations of *R. similis* and *Helicotylenchus* spp. in all the years, all months of the year, and in all counties where the banana is grown in the country. Nematodes delay foliar emission, lengthen the crop's vegetative cycle, reduce bunch weight and yield (Quénéhervé *et al.*, 1991a, 1991b; Jaramillo *et al.*, 2019; Chávez *et al.*, 2020).

Up to date, chemical nematicides are still feasible and economic option for the control of nematodes in bananas. Its applications are carried out when nematode analysis indicate populations above the established economic threshold. The molecules approved for use in bananas are alternated according to their physical-chemical characteristics, considering the climatic condition to prevent their biodegradation. However, certifications, supermarkets and consumers are looking for a final consumer fruit obtained with a low use of pesticides, especially of the toxicological profile IA and IB. The nematicides available to control nematodes in bananas belong mostly to these toxicological bands, which limits and restricts their application.

On the market there are fungal and bacterial nematicides (Abb-Elgawad and Hassan, 2018; Ruii, 2018). Within these, *Trichoderma* species (Cumagun and Moosavi, 2015; Hernández *et al.*, 2016; Poveda *et al.*, 2020; D'Errico *et al.*, 2020) and different species of *Bacillus* (De Araujo and Pletto, 2009; Cumagun and Moosavi, 2015; Gao *et al.*, 2016; Villarreal-Delgado *et al.*, 2017; Radhakrishnan *et al.*, 2017) are applied to various crops to control nematodes. In the case of fungi, it is known that they reduce nematode populations since

they infect eggs and females of sedentary endoparasitic nematodes such as *Meloidogyne*, *Heterodera*, and *Globodera* (Manzanilla *et al.*, 2013). However, they have been also evaluated in migratory endoparasites such as *R. similis* (Vergara *et al.*, 2012) and their application had promoted plant growth (Hernández *et al.*, 2016). In the case of *Bacillus* spp., it is reported that they secrete several metabolites that trigger plant growth and prevent pathogen infection, that induced physiological changes in plants as an adaptation to abiotic and biotic stresses, and degrading substances from *Bacillus* spp. damage pathogenic bacteria, fungi, nematodes, viruses, and pests (Radhakrishnan *et al.*, 2017; Li *et al.*, 2018). According to Borriss (2020) today there is strong evidence that plant associated *Bacillus amyloliquefaciens* FZB42 trigger pathways of induced systemic resistance, which protect plants against attacks of pathogenic microbes, viruses, and nematodes.

In addition, there are on the market, seaweed (*Ascophyllum nodosum*) extracts (Wu *et al.*, 1998; Whapham *et al.*, 1994; Tarjan, 1977) that are applied to various crops to control sedentary nematodes (Manzanilla *et al.*, 2013) such as *Meloidogyne*, *Heterodera* and *Globodera* and in some cases for migratory endoparasites such as *Pratylenchus* (Tarjan, 1977). Wu *et al.*, (1997) attribute the nematode control to the betaine content. Additionally, Seenivasan and Senthilnathan (2018); El-Sherif *et al.*, (2015) and Yass *et al.*, (2020) had reported nematode reduction and stimulation of vegetative growth variables with the application of humic acid extracts. Chitwood (2002) and Seenivasan and Senthilnathan (2018) mentioned that the nematicidal effect could be due to active principles present in humic acid such as carboxyl, phenolic, alcoholic, hydroxyl, and carbonyl groups.

This motivated to evaluate this kind of biological nematicides available on the market. Therefore, the objective of the present experiment was to compare the effect of biological and chemical nematicides on nematode control, root system recovery and banana (*Musa* AAA) yield.

Materials and Methods

Experimental site location, characteristics, and cultural practices

The field experiment was carried out for the parent plant and four consecutive ratoon crop cycles within a nematode infected long-term ratoon commercial banana (*Musa* AAA cv. Williams) plantation that was replanted in 2016 after 48 years of being cultivated with bananas and infested with nematodes, located in Guácimo county, province of Limón, Costa Rica. The soil was taxonomically classified as an Inceptisol and it had a sandy clay loam texture (47% sand, 21% silt and 32% clay) with a pH of 5.3 and 1.58% organic matter. The following concentrations of extractable cations were found, using Mehlich 3 (Mehlich, 1984) as the extractant: Ca 6.6, Mg 1.4, and K 1.11 cmol L⁻¹, and P 22.0, Zn 1.8, B 0.13, Cu 2.2, Fe 97.0 and Mn 22.0 µg ml⁻¹. The block or cable where the experiment was established had an annual yield of 3300 boxes of 18.14 kg per hectare for 2019, with a plant density of 1680 plants by hectare.

Desuckering was carried out every 10 weeks, leaving each production unit with a bearing mother plant, a large daughter sucker (follower) and a small grand-daughter (peeper) when possible. Shooting plants were propped with double polypropylene twine to the bottom of two well-developed adjacent plants. The follower sucker of each production unit was fertilized with 45 g every 15 days and when lime or nematicide was applied, the fertilization was done 30 days after with 90 g which resulted in a rate of 75 or 150 kg ha⁻¹, respectively, with a formula adapted to the soil and crop requirements, completing 350 kg N, 12 kg P₂O₅, 513 kg K₂O, 54 kg S and 54 kg of MgO in the year. Lime (Magprill 25% Ca and 9% Mg) was applied twice a year in front of the follower suckers with 80 g equivalent to 135 kg ha⁻¹.

All water requirements were supplied by rainfall, where the annual precipitation was of 2,686 and 3,342 mm, distributed throughout the year, for 2020,

and 2021, respectively. The driest month in 2020 was March with 46.8 mm and for 2021 was February with 144.6 mm. The accumulated precipitation in the first 9 months of 2022 was 1950.3 mm where the driest month was January with 57.9 mm. A complex system of primary, secondary, and tertiary drains was provided to disperse excess rainfall and prevent water logging during heavy rains. At establishment of the experiment, those drains were re-excavated in the area to deepen the water table level. Mean daily maximum/minimum temperatures were 36.3/17.6, 34.9/17.3, and 34.4/17.8°C, for 2020, 2021 and 2022, respectively.

Leaf fungi, especially black Sigatoka (*Pseudocercospora fijiensis*), was managed by defolating weekly to reduce the pressure of black Sigatoka inoculum and by aerial spraying of alternate fungicides which resulted in 56 sprayings each year at 6 days intervals. Fungicides were applied in emulsion with miscible oil and water in a spray solution of 20 L ha⁻¹. Weeds were controlled spraying every 5-8 weeks Reglone® 20SL 1.5 L ha⁻¹ or glyphosate® 36% SL 1.2 L ha⁻¹ in 160 L of water. Before setting the experiment, nematodes were controlled every year by 2-3 chemical nematicide applications (Counter® 15GR-AMVAC, Rugby® 10GR-FMC, Vydate® 24SL-DuPont, Verango® 50SC-Bayer, Mocap® 15GR-AMVAC) per year, based on the nematode population.

Treatments evaluated

Six treatments were evaluated; 1: rotation of three chemical nematicide cycles per year every four months (Mocap®, Counter®, Solvigo®, Verango®, Vydate®, Nematicur®, Mocap®), 2: rotation of one chemical nematicide with the rotation of one biological nematicide per year every six months (Mocap®, Cronox®, Counter®, Galvanize™ soil, Nematicur®), 3: rotation of four biological nematicide cycles per year every three months (Nematus® - Cronox® + Rhizomagic® - Galvanize® Soil - Dalgin® H15 - Cronox® - Galvanize® Soil - ACF Dryland soil kit - Cronox® + Rhizomagic® - Cronox®), 4: rotation of one

chemical nematicide by year with the rotation of three biological nematicide cycles by year every three months (Counter® - Nematus® - Galvanize® Soil - Cronox® - Solvigo® - Galvanize® Soil - Nematus® - Cronox® + Rhizomagic® - Mocap®), 5: rotation of two chemical nematicide cycles per year alternating each cycle with the rotation of two biological nematicides by year every three months (Counter® - Cronox® - Solvigo® - Galvanize® Soil - Mocap® - Galvanize® Soil - Nemaicur® - Cronox® + Rhizomagic® - Counter®), and 6: the untreated control (Table 1).

The applied chemical nematicides were those available including Counter® 15GR biodac-terbufos-AMVAC, Verango® fluopyram-Bayer, Vydate® oxamyl-DuPont, Mocap® biodac-ethoprophos-AMVAC, Nemaicur® biodac-phenamiphos-AMVAC and Solvigo® 36% abamectin + 72% thiamethoxan-Syngenta (Table 1). The rates used per follower sucker were the recommended by the manufacturer in the product label of 3 g a.i. for Counter®, Mocap®, and Nemaicur®, 2.4 g a.i. for Vydate®, 0.3 g a.i. for Verango®, and 1.19 ml of the commercial product for Solvigo®. Verango® and Solvigo® were applied in a water solution spreading 100 ml onto the soil surface with the manual knapsack hand sprayer (Protecno).

The biological nematicides applied were: Nematus® (*Ascophylum nodosum*, *Bacillus amyloliquefaciens*, *Bacillus licheniformis*, *Bacillus subtilis*, *Bacillus* sp., *Bacillus* spp. total microbial content 1×10^{10} spores ml^{-1} - Santa Clara Agrociencia) 2 L ha^{-1} , Galvanize® Soil (*Bacillus subtilis*, *Enterococcus faecium* y *Bacillus licheniformis* total microbial content 3.75×10^8 ufc g^{-1} -Alltech) 8 kg ha^{-1} , Cronox® (*Trichoderma asperellun* 1×10^9 ufc g^{-1} -Biotor Labs) 3 kg ha^{-1} , Rhizomagic® (*Ascophylum nodosum* extract 46%, N 4.3%, P_2O_5 3.8%, K_2O 2.6%, B 800 ppm, Cu 100 ppm, Fe 300 ppm, Mn 850 ppm, Mo 50 ppm, Zn 800 ppm – FMC) 1 L ha^{-1} , Dalgin H-15 (total humic extract 15.5%, humic acids 7.5%, fulvic acids 8%, potassium oxide 3.7%, free amino acids 1.8%, seaweed extract 6.7% - Sustainable Agro Solutions, S.A.) 5 L ha^{-1} , ACF

Dryland soil kit (*Bacillus amyloliquefaciens* 0.5×10^6 ufc ml^{-1} , *Bacillus subtilis* 0.5×10^6 ufc ml^{-1} , *Bacillus licheniformis* 0.5×10^6 ufc ml^{-1} , humic acids 3.6%, fulvic acids 2.9% - Blueplanet Labs) 30 L ha^{-1} .

The rate per hectare of each biological nematicide was divided by 1680, that was the plant density by hectare. Since the biological nematicides were applied with a manual knapsack hand sprayer (Protecno 20 L) calibrated to discharge 100 ml of solution in two pumpings, it was filled with 10 L of water, the amount of nematicide corresponding to 170 plants was added and after shaken it was gauged to 17 L, then re-shaked again and thereafter 100 ml of the solution were applied in front of each follower sucker.

The rectangular dome plots for each treatment consisted of 150-175 production units. Plots were arranged in a randomized complete block design with six replicates. The application of the chemical or biological nematicides was made by spreading the product in a banded arc with a radius of approximately 0.40 meter around each follower sucker pseudostem, sprouting from the base of the sucker, using the Swissmex backpack equipment specific for Counter®, Nemaicur®, and Mocap®, the spotgun for Vydate® and the Protecno-20 L manual knapsack hand sprayer for Verango®, Solvigo® and the biological nematicides. Plant debris was removed from the soil surface prior to distributing the chemical and biological nematicides onto moist soil as directed by the product label. During the development of the experiment, no rooting or organic matter was applied in the experimental area.

Nematode sampling and extraction procedure

One day before the nematicide application, and then every 30 days up to the 27 months that the experiment lasted, root samples were collected in each repetition. Each sample consisted of the roots of three follower suckers between 1.5-2.5 m height from recently flowered plants or prompt to bearing. In front of each follower sucker, a hole of 15 cm

length, 15 cm wide and 30 cm depth (soil volume of 6.75 L) was dug at the plant base using a shovel. All the roots found were collected and placed in labelled (treatment and repetition) plastic bags and delivered to CORBANA nematology laboratory in coolers.

In the laboratory, the root samples were registered and processed as soon as possible, and when it was necessary, stored in a refrigerator adjusted to 6-8 °C until being processed. The roots were rinsed free of soil, separated in functional living roots (white or cream-colored roots with light symptoms of nematode damage) and non-functional roots (roots with necrosis, root decay, rotten roots by excess water, snapping), left to dry off the surface moisture and weighed (Cas computing scale precision 5 kg ± 1 g). During the root separation process, in some roots, it was necessary to cut some damaged parts, which were classified accordingly. The total root weight corresponds to the sum of functional and non-functional roots.

Functional roots were cut into 1-2 cm length pieces and after homogenization, 25 g were selected at random or the amount available was used. These roots were macerated (Taylor and Loegering 1953, Araya 2002) in a kitchen blender (Osterizer; Sunbeam-Oster) for two periods of 10 seconds at low and 10 sec at high speed and nematode recovered in 0.025 mm (No 500) sieve. The nematodes were identified at the genus and species level, when possible, based on the morphological characteristics under a light microscope, following the key of Siddiqi (2000). The population densities of all plant-parasitic root nematodes present were recorded, and the values were converted to numbers per 100 g of roots. Total nematodes correspond to the sum of the phytoparasitic nematode species detected. Toppled plants in each experimental unit were recorded during all the experimental period and included as nematode control response variable.

Yield variables evaluated

When starting the experiment, in each repetition, 20 production units selected randomly, excluding those from plot edges, edge drains, cable edges, dumpings,

replanting plants or with double ratoon suckers, were progressively harvested which corresponded to the parent plant. The stem of each parent plant harvested was labelled with a code number (treatment, repetition, plant number 1 to 20; Example: T-1, R-1, P-1), and date of harvest, bunch weight, number of hands by bunch were recorded separately for each production unit. Then the code number and date of harvest of each parent plant was passed to its follower sucker and at its harvest (first ratoon crop), date of harvest, bunch weight, number of hands by bunch were recorded separately for each plant, and so on for the four consecutive ratoon crop cycles.

Harvesting of the parent plants and the selected ratoon suckers was done by calibration, starting when bunches reached 10 weeks of age. When in the second hand, the central fruit of the outer whorl had a diameter of at least 35 mm-diameter the bunch was harvested. If in week 12, it did not reach the minimum 35 mm-diameter required, they were harvested with the grade they had. Bunch weight (Tru-Test electronic scale XR3000 kg ± 1g), and number of hands by bunch were recorded.

The ratio, which is the number of boxes of 18.14 kg given by each bunch, was calculated considering a reduction of 21,5%, because was the average of the farm during the experimental time, which includes 11% of bunch stalk and 10.5 % of non-marketable fruit.

With the data of the number of bunches harvested in 2019 in the cable where the experiment was located, and the number of plants in that area, the initial ratooning was calculated in 1.69. Since the date of bunch harvest was registered for the parent plant and the 4 ratoon crop cycles in each production unit, the ratooning in each ratoon crop cycle was calculated dividing the 365 days of the year by the number of days between bunch harvest in each production unit for each of the following crop cycles.

The first harvest corresponds to the mother-parent plant and the others are from ratoon crop cycles. The harvest of those parent plants started on April 28,

2020 and finished on June 30, 2020. Harvest of the ratoon crops were between October 10, 2020 and February 23, 2021, between March 9, 2021 and December 10, 2021, between September 3, 2021 and July 7, 2022, and between March 4, 2022 and September 30, 2022 for the first, second, third and fourth ratoon crop, respectively.

The harvest of the fourth ratoon crop cycle was not completed and was ended when at least 10 bunches were harvest in each repetition on September 30, 2022. An additional harvest (final harvest) was done in each repetition, in 20 different production units that were also selected randomly, excluding those from plot edges, edge drains, cable edges, dumpings, replanting plants or with double ratoon suckers, that were progressively harvested from June 21, 2022 until September 16, 2022. The ratio was calculated again as before, and to calculate the yield (97.5% bunch recovery 1638 * ratio * ratooning), the ratooning found for the fourth crop cycle was used.

Data analysis

Root and nematode data were averaged by experimental plot across the 27 months excluding the first evaluation pre-treatment application. The composition of the nematode population was determined before treatment application and then for the average of the 27 root samplings.

Data of root weights before treatment application, and thereafter for the average of the 27 root samplings, were subjected to ANOVA by Proc GLM of SAS and mean separation by LSD-test. The number of nematodes was analyzed with generalized linear models, using the log transformation as link function and negative binomial distribution of the errors for the first nematode sampling alone, and thereafter for the average of the 27 nematode samplings together after the application.

Bunch weight, ratio, ratooning, and number of boxes of 18.14 kg per hectare per year (97.5% bunch recovery; 1, 638 bunches * ratio * ratooning) were averaged for each repetition and ratoon crop and subjected to ANOVA in PC-SAS® version 9.4.

Results and Discussion

Root content and nematode populations

In the root sampling carried out before treatment application, no difference was found in the root weight of total roots ($P= 0.9704$), nor functional roots ($P= 0.9354$). The root weight varied between 28 and 32.8 g for total roots and between 21.2 and 26.5 g for functional roots per follower sucker among treatments (Figure 1A-B). Similarly, in this sampling, no difference was detected in the population per 100 g of roots per sucker for *R. similis* ($P= 0.3083$), *Helicotylenchus* spp. ($P= 0.7168$) and total nematodes ($P= 0.4413$) among the suckers submitted to the different treatments (Figure 2A-C). Nematode populations among suckers applied with the treatments fluctuated for *R. similis* between 4,800 and 15,600, for *Helicotylenchus* spp. between 1,933 and 5,733 and for total nematodes between 8,667 and 19,533 individuals per 100 g of roots. The composition of the nematode population before treatments application was: 70% of *R. similis*, 29.8% of *Helicotylenchus* spp. with a negligible amount (0.2%) of *Meloidogyne* spp. (data not shown).

Root content and nematode populations through the 28 samplings are presented in Figure 1A-B and Figure 2A-C. Across the different samplings, with few exceptions, the root content and nematode populations followed a similar pattern in all treatments. After treatments application, when comparing the average of the 27 samplings, no differences were found among treatments in total root weight ($P= 0.9816$), nor in functional root weight ($P= 0.8569$), which ranged between 25.8 and 29.1 g and between 21.7 and 23.5 g per follower sucker, respectively (Figure 3A-B).

The highest nematode population per 100 g of roots by follower sucker of *R. similis* ($P= 0.0005$), *Helicotylenchus* spp. ($P= 0.0005$) and total nematodes ($P< 0.0001$) was found in the untreated plants (Figure 2A-C and Figure 4A-C). Compared to the untreated plants, the treatment with the rotation

of three chemical nematicide cycles by year reduced *R. similis* by 44.5%, *Helicotylenchus* spp. by 49.4% and the total nematode population by 45.3%, while in the plants treated with the rotation of four biological nematicide cycles by year diminished *R. similis* by 6.9%, *Helicotylenchus* spp. by 9% and the total nematode population by 7.3% (Figure 4A-C). The second treatment in nematode reduction was that with the rotation of two chemical nematicide cycles by year alternated with the rotation of two biological nematicide cycles which drops *R. similis* by 27.2%, *Helicotylenchus* spp. by 32.6% and total nematode population by 28.2%. Treatments with one chemical nematicide by year reduced *R. similis* by 22.5 and 27.2%, *Helicotylenchus* spp. by 32.6 and 38.1% and total nematode populations by 25.5 and 28.2%.

Averaging the 27 samplings taken after treatments application, the nematode population composition maintains the same pattern, but changing the proportions, where *R. similis* increased to 81.8%, *Helicotylenchus* spp. was reduced to 18.0%, and *Meloidogyne* spp. remain negligible with 0.2% (data not shown).

Uprooting plants

The number of uprooting plants during the experimental period was higher ($P < 0.0001$) in the untreated plots with 101 falling over plants followed by those applied with four biological nematicide cycles by year with 66 plants and in a third statistical group the other treatments which varied between 15 and 33 uprooted plants (Table 2).

Yield variables

In bunch weight, no difference among treatments was found for the parent plant ($P = 0.1430$) and for the first ($P = 0.8281$) and second ($P = 0.0506$) ratoon crop cycles (Table 3). Bunch weight varied between 26.4 and 29.5 kg for the parent plant, between 25.9 and 27.2 kg for the first ratoon crop cycle and between 25.3 and 28.6 kg for the second ratoon crop cycle. With exception of the number of hands per

bunch, at the second ratoon crop ($P = 0.0306$) which oscillated between 6.9 and 7.5 per bunch, in the other yield variables; ratio ($P > 0.0510$), ratooning ($P > 0.1666$) and boxes per hectare per year ($P > 0.1429$) no differences were observed among treatments for the parent plant, and the first and second ratoon crop cycle. Although in the second ratoon crop cycle, the treatment with the rotation of three chemical nematicide cycles per year gave 308 more boxes per hectare than the untreated plants (Table 3), the difference was not large enough to be significant ($P = 0.2177$).

Differences in yield ($\text{boxes ha}^{-1} \text{ year}^{-1}$) were found at the third ($P = 0.0405$) and fourth ($P = 0.0009$) ratoon crop cycle, and the final harvest ($P = 0.0047$). In both ratoons, and the final harvest, the highest yield was observed in the plants treated with the rotation of three chemical nematicide cycles by year, with 506, 840 and 791 more boxes per hectare per year than the untreated plants, respectively (Table 3). Even though, plants treated with the rotation of four biological nematicide cycles per year improved yield in 50, 331 and 280 more boxes $\text{ha}^{-1} \text{ year}^{-1}$ over the untreated plants, such increased was 456, 509 and 511 boxes, respectively, lower than that observed with the commercial (rotation of three chemical nematicide cycles by year) treatment.

The increase in yield at the third and fourth ratoon crop and final harvest came mainly from differences in bunch weight, ratio, and ratooning. At the third and fourth ratoon crop, and final harvest, bunch weight in the untreated plants drops from 29.5 kg per bunch, when the experiment was established (parent plant) to 23.6, 25.5, and 26.3 kg per bunch, respectively. In contrast, in the plants treated with the rotation of three chemical nematicide cycles by year, bunch weight was stable or improved a little, with 27.2 when the experiment was set up, and 27.0, 29.1 and 29.5 kg per bunch, at the third and fourth ratoon crop cycle, and final harvest, respectively. In parallel, the ratio, which is the number of banana fruit boxes (18.14 kg) obtained from each bunch, was higher in the plants treated with the rotation of the three chemical nematicide cycles per year with

1.17 and 1.26 boxes per bunch at the third and fourth ratoon crop, and 1.28 at the final harvest, compared with the untreated plants, where the ratio was 1.02 and 1.10, and 1.14 boxes per bunch, respectively.

Although no difference ($P= 0.2101$) in ratooning was found in the third ratoon crop, the highest ratooning was reported in the plants applied with the rotation of the three chemical nematicide cycles by year with 1.72 bunches by production unit by year. The same response was observed in the fourth ratoon crop, where the highest ($P= 0.0016$) ratooning was observed again in the plants treated with the rotation of the three chemical nematicide cycles by year with 1.90, while the lowest was found in the untreated plants with 1.71 bunches by production unit by year.

In the sampling done before product application, no differences among treatments were found in root contents and nematode populations. This means that any difference that was found after applying the treatments, should be attributed to its effect. The three nematode genera detected are well known pathogens in banana roots (Gowen *et al.*, 2005; Quénéhervé, 2008; Dubois and Coyne, 2011; Volcy, 2011; Guzmán-Piedrahita, 2011a, 2011b; Sikora *et al.*, 2018), and agreed with those reported in Costa Rica (Vargas *et al.*, 2006; Vargas *et al.*, 2015; Araya and Vargas, 2018). Also, are in parallel with those found in the main banana producing exporting countries like Ecuador (Chávez and Araya, 2010; Aguirre *et al.*, 2016a, 2016b; Jaramillo *et al.*, 2019), Colombia (Castillo *et al.*, 2010), and the Philippines (Arceo, 2007).

When the experiment started, the nematode population consisted mainly of *R. similis* (70%), and *Helicotylenchus* spp. (29.8%), increasing the proportion of *R. similis* at the end of the experiment to 81.8% and reducing *Helicotylenchus* spp. to 18% of the phytoparasitic nematode community, while *Meloidogyne* spp. remain like the initial proportion with 0.2%. This agrees with that observed in Cavendish banana plantations with the presence of

the three nematode genera found here, where greater proportion of *R. similis* has been found when nematode control is done frequently, as reported earlier in Costa Rica by Moens *et al.*, (2004) and Calvo and Araya (2005).

The high population of *R. similis* and *Helicotylenchus* spp. was favored by the banana production system, that even though banana is an annual crop, its production is in perennial monoculture. *Radopholus similis* is a migratory endoparasite that causes necrotic lesions along the entire root; in the epidermis, cortical parenchyma, and vascular cylinder (Blake, 1966; Orton and Siddqi, 1973; Jackson *et al.*, 2003; Volcy, 2011; Guzmán-Piedrahita 2011a; Sikora *et al.*, 2018). In contrast, *Helicotylenchus* spp. is an ecto-endoparasite (Blake 1966; Orion and Bar-Eyal, 1995; Guzmán-Piedrahita, 2011b; Sikora *et al.*, 2018) that induces necrotic lesions on the surface of the roots.

The reduction found in nematode population with the rotation of three chemical nematicide cycles by year was of 44% for *R. similis*, 49% for *Helicotylenchus* spp. and 45% for total nematodes which agreed with results of Araya and Cheves (1997a, 1997b) in Costa Rica, who found reductions of 22 to 63% for *R. similis* and 25 to 89% for *Helicotylenchus* spp. and Moens *et al.*, (2004), also in Costa Rica, who recorded drops between 18 and 59% for the total phytoparasitic nematode population. These percentage decreases in nematode population were also in parallel with Jaramillo *et al.*, (2019) in Ecuador, who reported reductions between 20 and 49% for *R. similis*, between 31 and 50% for *Helicotylenchus* spp. and between 29 and 49% for total nematodes, and with Chávez *et al.*, (2020), also in Ecuador, who found drops between 22 and 49% for *R. similis*, between 23 and 40% for *Helicotylenchus* spp. and between 25 and 45% for total nematodes.

In Ivory Coast, Quénéhervé *et al.*, (1991a; 1991b; 1991c) indicated reductions of *R. similis* between 22.7 and 90.7% and between 32.5 and 100% for

Helicotylenchus spp., and Castillo *et al.*, (2010) in Colombia found drops between 24 and 37% for *R. similis*, between 38 and 60% for *Helicotylenchus* spp., and between 25 and 33% for total nematodes. In Belize, Salguero *et al.*, (2016), found decreases between 33 and 47% for *R. similis*, between 36 and 65% for *Helicotylenchus* spp. and between 35 and 59% for total nematodes.

The no significant reduction observed with the rotation of four biological nematicide cycles by year of 6.9% for *R. similis*, 9% for *Helicotylenchus* spp. and 7.3% for the total phytoparasitic nematodes disagreed with Bruce da Silva *et al.*, (2022) who mentioned control of *R. similis* with *Trichoderma*.

Also, these results differed with Castellanos (2016) who reported significant reduction of *R. similis*, *H. multicinctus* and *M. incognita* in three commercial banana (*Musa* AAA) plantations of the cultivars of the Cavendish subgroup (Grande Naine, Bonifacio, and Williams) treated with *Trichoderma asperellum*. Likewise, differed with results in other crops, where Hernández (2014) and Hernández *et al.*, (2015) found that *T. asperellum* induced 90% of *M. incognita* mortality at 24 h *in vitro* conditions and in semi-controlled conditions it reduced the number of eggs by female in tomato plants and with Castro (2018), who did not find *Meloidogyne* spp. on soil cultivated with *Cucumis melo* when three applications of the mixture of *T. asperellum* and *Pochonia chlamydosporia* were done at planting and 30 and 50 days after sowing.

In the case of *Bacillus* spp., diverged with Mendoza and Sikora (2009), who found 63.7% control of *R. similis* with the application of *Bacillus firmus*, with Peixoto *et al.*, (2015) who found control of *R. similis* with *Bacillus* spp. and with Araújo *et al.*, (2018) who reported reduction of *R. similis*, *Meloidogyne* spp., *Pratylenchus* spp., and *Helicotylenchus* spp. with the application of *Bacillus subtilis*, all those studies in banana pot experiments. On other crops, De Araujo and Pletto (2009) found that the application of the isolate PRBS-1 of *Bacillus subtilis* reduced the fresh root mass and the egg masses of

Meloidogyne in tomato roots. With the application of *Ascophyllum nodosum*, these results contradict Tarjan (1977), who found reduction of *Pratylenchus coffeae* populations in citrus nursery plants treated with *A. nodosum*, also disagreed with Radwan *et al.*, (2012), who reported 86.9% reduction in the number of root galling induced by *M. incognita* in tomato, with Whapham *et al.*, (1994) who reported reductions in the fecundity of *M. javanica* in tomato, and with Wu *et al.*, (1998) who found that decreased the fertility of *M. javanica* in *Arabidopsis thaliana*, in all cases when were treated with extracts of the algae.

As well differed with Wu *et al.*, (1997) who reported reduction of *M. javanica* and *M. incognita* populations in tomato when treated with extracts of the algae.

Furthermore, this results also dissent from Seenivasan and Senthilnathan (2018) who reported in a banana pot experiment testing concentrations of 0.04%, 0.08%, 0.2% and 0.4% of humic acid reductions of *M. incognita* soil density between 53.5 and 56.7%, root infection between 61.9 and 63.8%, egg population between 61.9 and 63.8% and reproduction rate between 55.7 and 56.6%. Then, it seems that effective biological nematicides are not yet available for bananas, despite a substantial, positive literature on this topic.

The no differences found in root weights more likely was due to the plant sampled and conditions of the experimental area. Root sampling was done on follower suckers between 1.5 and 2.5 m height which means that they were in active growth with young roots that if were infected with nematodes, the time maybe was not enough to develop clear symptoms. Additionally, the block or cable where the experiment was carried out, drainage channels were re-excavated when it was set up to lower the water table level and prevent water logging which promotes root health. So, snapping roots either by excess soil humidity or by the presence of number of pathogens (fungi-bacteria) in the nematode-induced lesions which hastens the destructions of roots, was

prevented. Furthermore, the classification of functional roots is subjective (visual) and depends on root symptoms. Roots infected by *R. similis* show reddish-brown lesions on the outer part of the roots penetrating throughout the cortex and then turns necrotic and *Helicotylenchus* spp. feeds on the outer cells of the root cortex, it produces a small-dashes reddish-brown to necrotic lesions. However, if the banana roots are still white and cream, it does not mean that they are free of nematodes. As indicated by Ayoub (1980); Mai (1985); McKenry and Roberts (1985) extensive loss of yield can occur when one or more nematode species may be feeding on a given plant, without showing obvious or specific plant symptoms. Here, maybe the nematode population, lower of 5000 per 100 g of roots, in some samplings, was not enough to develop root symptoms but it reduced ratooning and yield. It is known that in white-cream roots infected with nematodes histological and physiological cell alterations occurs (Blake, 1966; Wyss, 2002; Grunewald *et al.*, 2009; Haegeman *et al.*, 2010; Jones *et al.*, 2016) which restrict water and nutrients uptake (Agrios, 2005; Haegeman *et al.*, 2010; Sikora *et al.*, 2018).

In the untreated plots, there were 101 falling over plants during all the experimental period of four ratoon crop cycles. This number of uprooting plants partially agreed with the review of Gowen (1993) who reported between 13 and 29 and between 9 and 56 toppled plants for the first and second ratoon crop, respectively, in untreated plots. Toppling is one of the main ways of crop losses and occurs usually on fruit bearing stems that suffered destruction of the primary roots by nematodes resulting in poor anchorage that are exposed to winds or heavy rainfall. Generally, when these plants fall, they knock down neighboring plants that are also flowered or about to flowering. This means that the current bunch is lost, where the follower sucker may have taken at least 7 months to produce it and the re-planting new plant will take at least 9-10 months to produce a new bunch. The experimental plots were of 150 to 175 production units and there were six repetitions, so from 900 to

1050 plants, 101 fell and if we consider the plantation density that was of 1680 plants per hectare, then between 161 and 188 plants would fall by hectare. Compared to the rotation of three chemical nematicide cycles by year, where 19 plants fell that corresponds between 30 to 35 plants by hectare, which results in a lost between 72 to 84 boxes by hectare by year ($30 * \text{ratio } 1.26 * \text{ratooning } 1.90$ in the fourth ratoon crop = 72 or $35 * 1.26 * 1.90 = 84$) in the untreated plots the lost was between 303 and 354 boxes by hectare by year ($161 * 1.10 * 1.71 = 303$ or $188 * 1.10 * 1.71 = 354$).

In the production variables evaluated, no differences were found for the parent plant. This is reasonable, since this harvest started when the experiment was set up (April 2020) and finished on June 30, 2020, which means that the bunches harvested came from stems with the commercial nematode control done (2 to 3 chemical nematicide cycles by year) before the treatment application, because nematicides are always applied in from of the follower suckers. The same was observed for the first ratoon crop, where no differences in yield variables were found which was expected, because those bunches came again from plants without the effect of the treatments applied.

In the second ratoon crop cycle, the rotation of three chemical nematicide cycles per year improved yield by 308 more boxes per hectare compared to the untreated plants, but such difference was not large enough to be significant, more likely due to many bunches still came from plants without having the complete treatment effect, since this harvest started 11 months after the experiment was set up and finished December 10, 2021. In addition, at the harvest of the parent plant, when the experiment was set up, the untreated plots yielded 277 more boxes than the rotation of three chemical nematicide cycles by year. So, the chemical control treatment leveled the production with the control and then surpassed it.

Differences in yield appeared at the third (+ 506 boxes) and was increased at the fourth (+ 840 boxes)

ratoon crop cycle, which was confirmed with the final harvest, where the difference was 791 boxes. In both ratoons, and the final harvest, the improvement was induced by the rotation of three chemical nematicide cycles by year where ratooning was higher, and plants gave heavier bunches and accordingly higher ratio.

In the third ratoon crop, the difference (rotation of three chemical nematicide cycles by year vs untreated plants) of 3.4 kg that corresponds to 0.15 units in ratio ended in 422 more boxes (97.5% bunch recovery $1638 * 0.15$ ratio units * 1.72 ratooning) per hectare and the other 84 boxes to complete the 506 more boxes, came from the increase of 0.05 units in ratooning. At the fourth ratoon crop, bunch weight was increased in 3.6 kg over the untreated plants which resulted in 0.16 units more in ratio that ended in 498 more boxes by hectare by year ($1638 * 0.16$ ratio * 1.9 ratooning = 498 boxes), and the other 342 boxes to complete the 840 boxes increased, came from the 0.19 units augmented in ratooning.

In the final harvest, the difference of 3,2 kg with respect to the untreated control ended in 0.14 units more of ratio which gave 435 ($1638 * 0.14$ ratio * 1.90 ratooning) more boxes by hectare by year. The improvement in ratooning of 0.19 units means that the interval between harvest was reduced in 21 days, which agreed with Quénéhervé *et al.*, (1991b), who found a cumulative reduction in time to harvest according to the cycle of 28 days in the first, 57 days in the second and 128 days in the third harvested cycle in plants treated with nematicide.

Similarly, Quénéhervé *et al.*, (1991c) and Gowen (1995) reported an increase in the harvest period from 13 to 30 and from 22 to 73 days, respectively, in plants infected with nematodes that were not treated compared with those applied with nematicide. In congruence with this extension in the period to harvest, Roderick *et al.*, (2012) reported an increase of 13.6 more days to harvest in Mbwarzirume banana plants to which they added nematodes compared to plants without the addition

of nematodes. The highest number of boxes per hectare per year was due to the rotation of three chemical nematicide cycles per year that resulted in a significant reduction of nematodes, which favored water and nutrients up take, allowing faster growth of the plants, which led to a higher ratooning and heavier bunches. In the third and fourth ratoon crop cycle, and final harvest, the application of such chemical nematicides improved yield in 506 (9.1 mt), 840 (15.2 mt) and 791 (14.3 mt) more boxes per hectare per year than plants of the untreated plots, respectively. Such improvement in yield, over the untreated plots, should be higher, because it did not consider the yield lost by uprooted plants.

The absence of increase in yield for the parent plant, first and second ratoon crop was due to the nematode control done in the farm before the experiment was established of 2 to 3 chemical nematicide cycles per year. Since in a commercial banana plantation different phenological stages (peepers, suckers in different vegetative growth, flowering, and fruiting plants) are present at the same time, which allows fruit to harvest all year around, in the parent plant, first and second ratoon crop, there were still harvested plants with the nematode control effect in the untreated plots, while in the third and fourth ratoon crop, all harvested plants were free of nematode control.

Additionally, in the untreated plots a higher number of boxes per hectare per year was observed at the parent plant, which was a little higher than the average of the cable where the experiment was run, which yielded 3300 boxes per hectare in 2019.

These results confirm that banana nematodes are serious threat to banana production in Costa Rica as has been reported by Moens *et al.*, (2004); Araya and Lakhi (2004); Vargas *et al.*, (2006). Similarly, in Ecuador, Jaramillo *et al.*, (2019) and Chávez *et al.*, (2020) found that those nematodes reduced banana yield. Then, this agreed with Dita *et al.*, (2013) thoughts, that nematodes continue to be a serious threat to banana production in Latin America and the Caribbean.

Table.1 Description of the treatments evaluated with the sequence of the nematicides and month of application

Treatment	Nematicide and months of evaluation																								
	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
1. 3c / year	Mo				Co				So				Ve				Vy				Ne				Mo
2. 1c 1b / year	Mo						C						Co						Ga						Ne
3. 4 b / year	Nt			CR			Ga			Da			C			Ga			Dr			CR			C
4. 1c 3b / year	Co			Nt			Ga			C			So			Ga			Nt			CR			Mo
5. 2c 2b / year	Co			C			So			Ga			Mo			Ga			Ne			CR			Co
6. Untreated																									

Note: 0= April 2020 when the experiment was established and 24= April 2022 when the last application was done. Treatment 1: 3 c / year = rotation of 3 chemical nematicides / year, Treatment 2: 1 c 1 b / year= rotation of 1 chemical nematicide by year and rotation of 1 biological nematicide by year, Treatment 3: 4 b / year= rotation of 4 biological nematicides by year, Treatment 4: 1 c 3 b / year= rotation of 1 chemical nematicide by year and rotation of 3 biological nematicides by year, Treatment 5: 2 c 2 b / year= rotation of 2 chemical nematicides by year and rotation of 2 biological nematicides by year. Chemical nematicides; Mo= Mocap® 15GR ethoprofos 3 g a.i. AMVAC, Co= Counter® 15GR terbufos 3 g a.i. -AMVAC, Ne= Nematicur® 15GR Phenamiphos 3 g a.i. – AMVAC, Ve= Verango® fluopyram 0.3 g a.i. -Bayer, Vy= Vydate® 24SL oxamyl 2.4 g a.i. -DuPont, So= Solvigo® abamectina + thiametoxan 1.19 ml of the commercial product – Syngenta, all rates of chemical nematicides by follower sucker; biological nematicides: Nt= Nematus® 2 L ha⁻¹, C= Cronox® 3 kg ha⁻¹, Ga= Galvanize® Soil 8 kg ha⁻¹, Da= Dalgin® H15 5 L ha⁻¹, CR= Cronox® 3 kg ha⁻¹ + Rhizomagic®1 Lha⁻¹; Dr= ACF Dryland soil kit 30 L ha⁻¹. The rate of Verango, Solvigo and all biological nematicides were applied in a water solution of 100 ml by follower sucker.

Table.2 Number of falling over plants by treatment during the experimental period from April 2020 to September 2022.

Treatment	Number of falling over plants
3 chemical nematicide cycles by year	19
2 nematicide cycles by year; 1 chemical 1 biological	33
4 biological nematicide cycles by year	66
4 nematicide cycles by year; 1 chemical 3 biological	31
4 nematicide cycles by year; 2 chemical 2 biological	15
Untreated control	101
Probability	P< 0.0001

Fig.1 A-B Root content (g) by follower sucker in banana plants (*Musa* AAA cv Williams) treated with different number of chemical and biological nematicides cycles per year. Each point is the average of six repetitions. In each repetition, three follower suckers from 1.5-2.5 m height were excavated at its base and in front of it, making a hole of 15 cm long by 15 cm wide and 30 cm depth from where all the roots were collected.

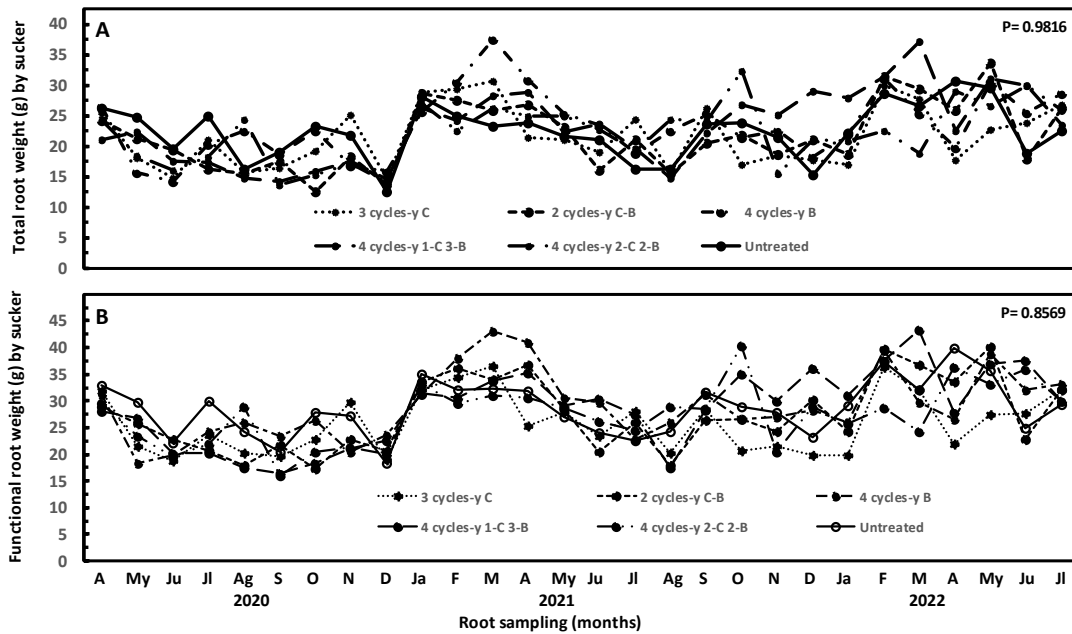


Table.3 Banana (*Musa* AAA cv. Williams) yield parameters according to the nematode management per year and cost benefit relationship at the third and fourth ratoon crop and final harvest. Sell price of each box of 18.14 kg was US \$8.30.

Treatment	Bunch weight Kg	Number of hands / bunch	Ratio	Ratooning	Boxes ha ⁻¹ year ⁻¹	Difference in boxes with untreated	Additional income US \$	Treatment cost US \$	Additional packing cost US \$	Net income US \$	Net profit by dollar
Parent plant at experiment establishment											
3 cycles/year-C	27.2	7.3	1.18	1.69	3266	-277		246			
2 cycles/year-C-B	27.4	7.4	1.19	1.69	3294	-249		176			
4 cycles/year-B	26.4	7.2	1.14	1.69	3156	-387		375			
4 cycles/year-1C-3B	25.8	7.1	1.12	1.69	3100	-443		393			
4 cycles/year 2C-2B	27.7	7.3	1.20	1.69	3322	-221		354			
Untreated	29.5	7.6	1.28	1.69	3543	0					
Probability	P= 0.1430	P= 0.3967	P= 0.1424		P= 0.1429						
First ratoon crop											
3 cycles/year-C	27.1	7.5	1.17	1.86	3565	0		223			
2 cycles/year-C-B	26.7	7.5	1.16	1.82	3458	-107		163			
4 cycles/year-B	26.0	7.3	1.13	1.83	3387	-178		346			
4 cycles/year-1C-3B	25.9	7.3	1.12	1.79	3284	-281		371			
4 cycles/year-2C-2B	27.2	7.5	1.18	1.81	3498	-67		331			
Untreated	27.1	7.5	1.17	1.86	3565	0					
Probability	P= 0.8281	P= 0.4862	P= 0.8329	P= 0.5809	P= 0.8055						
Second ratoon crop											
3 cycles/year-C	28.0	7.3	1.21	1.81	3587	+ 308	+ 2556	259	1093	1204	4.65
2 cycles/year-C-B	25.3	6.9	1.10	1.75	3153	- 126		166			
4 cycles/year-B	25.5	7.0	1.10	1.78	3207	- 72		333			
4 cycles/year-1C-3B	25.6	7.0	1.11	1.69	3073	-206		376			
4 cycles/year-2C-2B	28.6	7.5	1.24	1.76	3575	+ 296	+ 2457	357	1051	1049	2.94
Untreated	25.3	7.0	1.10	1.82	3279	0					
Probability	P=	P=	P=	P=	P=						

	0.0506	0.0306	0.0510	0.1666	0.2177						
Third ratoon crop											
3 cycles/year-C	27.0	7.5	1.17	1.72	3296	+ 506	+ 4200	273	1796	2131	7.80
2 cycles/year-C-B	23.2	7.0	1.01	1.62	2680	-110		179			
4 cycles/year-B	23.6	7.1	1.02	1.70	2840	+ 50	+ 415	349	178	- 112	
4 cycles/year-1C-3B	23.7	7.1	1.03	1.65	2784	-6		385			
4 cycles/year-2C-2B	24.8	7.3	1.07	1.66	2909	+ 119	+ 998	379	422	186	0.49
Untreated	23.6	7.2	1.02	1.67	2790	0					
Probability	P= 0.0137	P= 0.1953	P= 0.0138	P= 0.2101	P= 0.0405						
Fourth ratoon crop											
3 cycles/year-C	29.1	7.6	1.26	1.90	3921	+ 840	+ 6972	247	2982	3743	15.15
2 cycles/year-C-B	25.8	7.3	1.12	1.83	3357	+ 276	+ 2291	158	980	1153	7.30
4 cycles/year-B	25.8	7.3	1.12	1.86	3412	+ 331	+ 2747	319	1175	1253	3.93
4 cycles/year-1C-3B	26.2	7.3	1.13	1.87	3461	+ 380	+ 3154	340	1349	1465	4.31
4 cycles/year-2C-2B	26.2	7.3	1.13	1.88	3480	+ 399	+ 3312	335	1416	1560	4.66
Untreated	25.5	7.3	1.10	1.71	3081	0					
Probability	P= 0.0123	P= 0.3348	P= 0.0115	P= 0.0016	P= 0.0009						
Final harvest of 20 bunches by repetition from June 21, 2022 until September 16, 2022											
3 cycles/year-C	29.5	7.7	1.28	1.90	3984	+ 791	+ 6565	247	2808	3510	14.2
2 cycles/year-C-B	26.5	7.4	1.15	1.83	3447	+ 254	+ 2108	158	902	1049	6.6
4 cycles/year-B	26.3	7.4	1.14	1.86	3473	+ 280	+ 2324	319	994	1011	3.2
4 cycles/year-1C-3B	25.7	7.3	1.11	1.87	3400	+ 216	+ 1718	340	735	643	1.9
4 cycles/year-2C-2B	26.9	7.4	1.16	1.88	3572	+ 379	+ 3146	335	1345	1465	4.4
Untreated	26.3	7.4	1.14	1.71	3193						
Probability	P= 0.0045	P= 0.0279	P= 0.0044	P= 0.0016	P= 0.0047						

Ratio= number of boxes of 18.14 kg per bunch [78.5% of the bunch weight was packet (21.5% rejection that includes 11% bunch stalk and 10.5% rejected bananas) / 18.14 kg by box]. 1,680 plants per hectare from which 97.5% of the bunches were processed (1,638 bunches), ratooning= number of bunches harvested by each banana stool by year, boxes per hectare per year= (1,638 bunches * ratio * ratoon). Each value is the mean of six replicates and in each replicate from 12-20 bunches were harvested. C: chemical nematicide, B: biological nematicide. Net profit= (additional income – treatment control cost – banana box packing cost of US \$3.55 each). Product cost by hectare: Counter® 15GR \$96, Verango® 50SC \$170 / ha, Vydate® 24SL \$120, Nematicur® 15G \$128 / ha, Mocap® 15GR \$128 / ha, Solvigo \$140, Cronox \$135, Galvanize Soil \$160, Dalgin \$60, Nematius \$200, Dryland Soil \$85, Rhizomagic \$10, application cost by cycle \$17. The product cost by year was divided by the ratooning in each treatment.

Fig.2 A-C. Number of nematodes per 100 g of banana (*Musa AAA cv. Williams*) roots per follower sucker treated with different number of chemical and biological nematicides cycles per year. Each point is the average of six repetitions. In each repetition, three follower suckers of 1.5-2.5 m height were dug in their base and in front, making a hole of 15 cm long by 15 cm wide and 30 cm depth from where all roots were collected.

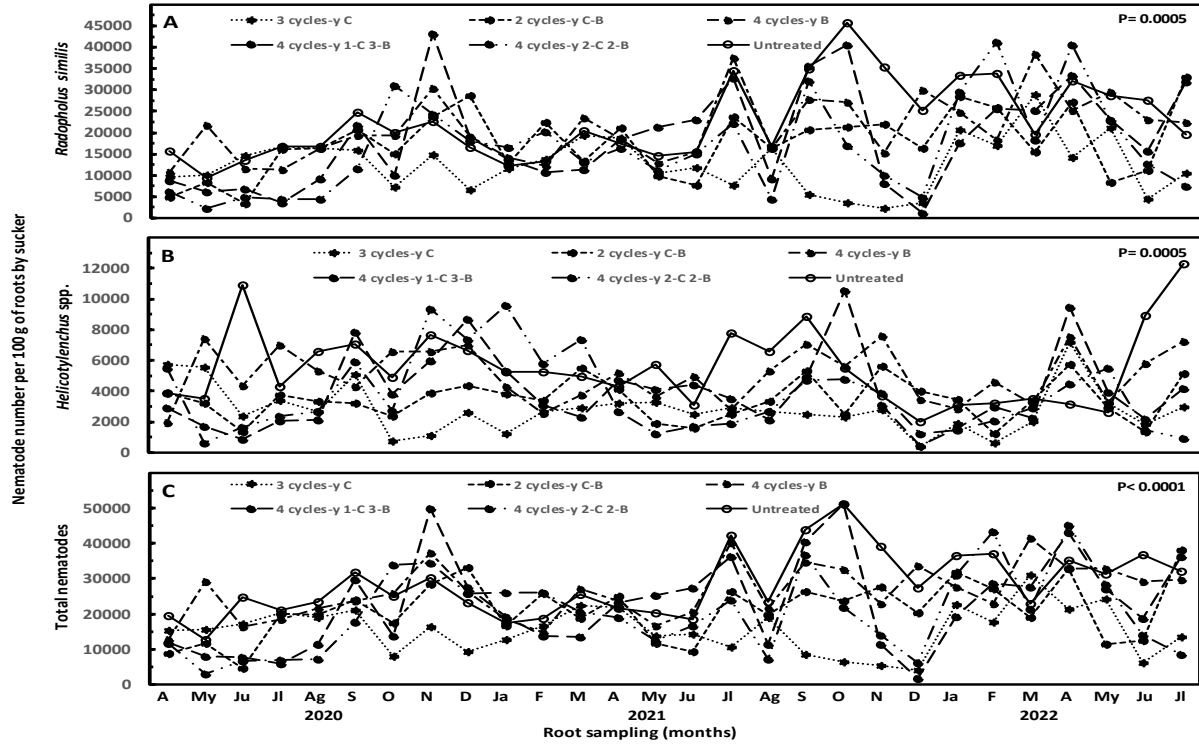


Fig.3 A-B. Average root content (g) per follower sucker in banana plants (*Musa AAA cv Williams*) treated with different number of chemical and biological nematicides cycles per year. Each bar is the average of 168 observations (28 samples * six repetitions) and in each repetition the value is the average of three follower suckers. In each follower sucker, a hole 15 cm long by 15 cm wide and 30 cm depth was excavated at the base, and all roots were collected.

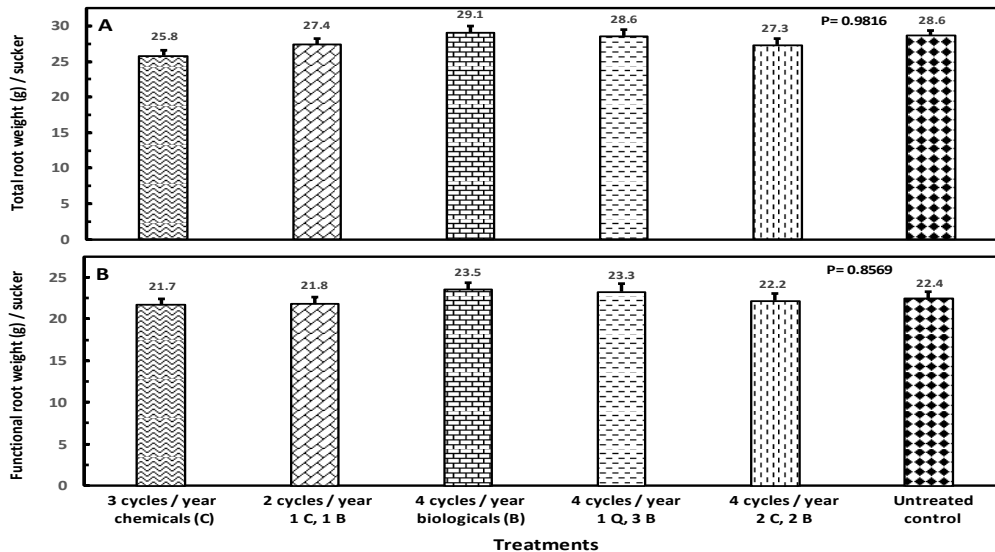
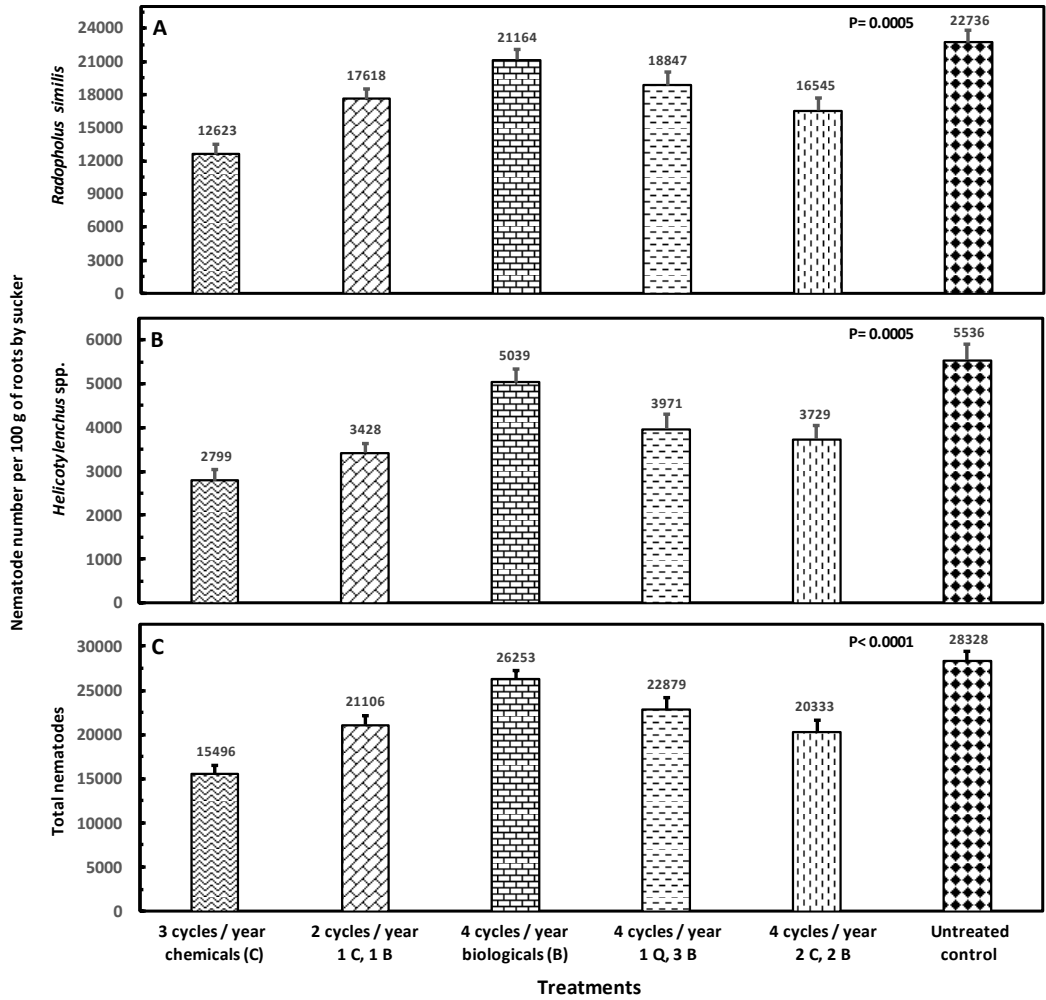


Fig.4 A-C. Number of nematodes per 100 g of banana roots (*Musa AAA cv Williams*) by follower sucker treated with different number of chemical and biological nematicides cycles per year. Each bar is the mean \pm standard error of 168 observations (28 samplings * six repetitions) and in each repetition the value is the average of three follower suckers of 1.5-2.5 m high. A hole of 15 cm long x 15 cm wide and 30 cm depth was dug in front of each follower sucker and all roots were collected.



The percentage of yield increase was of 18% and 27% for the third and fourth ratoon crop, respectively, and 24.6% for the final harvest, which were agreed with some of the percentages compiled by Gowen and Quénéhervé (1990), who mentioned increases from 14-263% and Gowen (1995), who cited increases from 5 to 275% and were lower than that reported by Stanton and Pattison (2000) of 46%. The increased in production found were in line with that reported by Quénéhervé *et al.*, (1991a), who indicated increases in production between 463 to 1,328 boxes (8.4-24.1 tm), with Pattison *et al.*,

(1999) who reported increases between 655 to 953 boxes of 13 kg (8.5-12.3 tm), with Salguero *et al.*, (2016), who found increases between 545 and 832 boxes of 18.14 kg (9.9-15.1 tm), with Jaramillo *et al.*, (2019), who mentioned increases between 545 and 1158 boxes of 18.14 kg (9.9 - 21 mt), with Chávez *et al.*, (2020), who reported grows between 226 and 730 boxes of 18.14 kg (4.0 to 13.2 tm) and was lower than that reported by Araya and Lakhi (2004), who cited increases of 1,245 boxes of 18.14 kg (22,6 tm) per hectare per year, controlling nematodes through the application of chemical

nematicides. The highest yield (number of boxes per hectare per year) was observed in plants treated with the rotation of three chemical nematicide cycles per year in parallel with that reported by Jaramillo *et al.*, (2019) and Chávez *et al.*, (2020) in Ecuador and Araya (2003) in Costa Rica, who registered higher yields as the number of nematicide cycles per year increased in banana plantations infected with nematodes. This increased in production because of nematodes control is agreed with Guerout (1972); Charles *et al.*, (1985); Quénéhervé *et al.*, (1991a, 1991b), and Salguero *et al.*, (2016), who cited negative and significant linear correlations between the populations of *R. similis*, *Helicotylenchus* spp. and total nematodes with bunch weight in bananas.

The high population of *R. similis* and the increased achieved in yield with the application of chemical nematicide indicated that their parasitism reduces growth, development, and production in accordance with observations by Gowen and Quénéhervé (1990); Gowen (1993, 1995); Araya (2004); Gowen *et al.*, (2005); Quénéhervé (2008); Guzmán-Piedrahita (2011a); Roderick *et al.*, (2012); Coyne *et al.*, (2013); Sikora *et al.*, (2018). In the case of *Helicotylenchus*, McSorley and Parrado (1986); Gowen and Quénéhervé (1990); Chau *et al.*, (1997); Barekye *et al.*, (1998, 2000); Gowen (2000); Ssango *et al.*, (2004); Guzmán-Piedrahita (2011b); Coyne *et al.*, (2013); Salguero *et al.*, (2016) reported that *H. multicinctus* and *H. dihystra* damaged the banana root system and reduced yield between 19% (Speijer and Fogain, 1999) and 34% (Reddy 1994). Additionally, Sijmons *et al.*, (1994) indicated that the induction and maintenance of feeding sites of *Helicotylenchus* spp. causes physiological changes in the structure of cells.

The presence of nematodes with different parasitic habits; *R. similis* migratory endoparasite and *Helicotylenchus* spp. an ecto-endoparasite most likely exacerbates root damage since lesions can develop at feeding sites and through root tissue. In addition, plants often activate post-infection resistance mechanisms, even in cases where the population of nematodes increases over time, and

the nematode-plant interaction is compatible. Therefore, together these processes can represent a high energy expenditure for plants that can interfere with the filling and development of the bunch.

Given that both nematode genera cause damage to the crop, for the implementation of options for their management, the population of all the phytoparasitic nematodes present should be considered, as has been suggested by Araya (2004); Ramclam and Araya (2006); Salguero *et al.*, (2016) and Aguirre *et al.*, (2016a; 2016b).

During the development of the experiment, the market price of a box of 18.14 kg of bananas was US \$8.30 and that of a nematicide application cycle including the application cost was Counter® 15FC \$113, Verango® \$187, Vydate® 24SL \$137, Mocap® 15GR \$145, Nemacur® 15GR \$145, Solvigo® \$157, Cronox® \$152, Galvanize Soil® \$177, Dalgin® \$77, Nematus® \$217, Dryland Soil® \$102, and Rhizomagic® \$10 per hectare. The cost of the fertilizer, control of black Sigatoka and weeds, and other tasks was the same for the control plots and those treated with chemical or biological nematicide, since the increase recorded was for bunch weight, ratio, and ratooning.

The additional net income from the increase in yield, deducted the packing cost of \$3.55 for each additional box and the cost of the product and its application was of US \$1204, \$2131, \$3743 and \$3510 for the second, third and fourth ratoon crop, and final harvest, respectively, by hectare per year with the rotation of three chemical nematicide cycles by year. This net gain agrees with that indicated by Jaramillo *et al.*, (2019) who found amounts between \$2,550 and \$5,759, with Chávez *et al.*, (2020) who indicated amounts between \$1050 and \$3432 and Pattison *et al.*, (1999) who reported amounts between \$2,494 and \$5,910 per hectare per year. This means, that for every dollar invested in nematode control, the net profit was of US \$4.65, \$7.80, \$15.15 and \$14.2 for the second, third and fourth ratoon crop cycle, and final harvest, respectively.

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