Characterization of Earthworm Taxonomic Structure of Coffee (*Coffea arabica*) Plantations in Puerto Rico

by

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A thesis submitted in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

in

BIOLOGY

UNIVERSITY OF PUERTO RICO

MAYAGÜEZ CAMPUS

2010

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Abstract

The taxonomic structure of earthworms of coffee plantations at three different localities in Puerto Rico (in the municipalities of Las Marías, Lares and Jayuya), representing different soil types, was assessed. Organisms were manually sorted from 25 x 25 cm microplots. Eight earthworm species were identified: *Onychochaeta borincana*, *Pontoscolex corethrurus*, *P. melissae*, *P. spiralis* and *Pontoscolex* sp., which belong to the Glossoscolecidae family, and *Amynthas gracilis*, *A. rodericensis*, and a pheretimoid species, which belong to the Megascolecidae family. No clear patterns between soil properties and the taxonomic structure and density of earthworms at these coffee plantations were detected. Higher densitys of exotic species were found in the most disturbed areas, as expected. *P. corethrurus* was found in all the localities having shaded and sun coffee. Lares, the area of the second highest elevation, had the highest density of species. On the other hand, Jayuya had the highest species number. *P. melissae*, a rare species that had been reported from only three locations in Puerto Rico when it was described in 1991, was found only in Jayuya.

Resumen

Se estudió la estructura taxonómica de las lombrices de tierra de las plantaciones de café en tres localidades (en los municipios de Las Marías, Lares y Jayuya), representando tres tipos de suelo distintos de Puerto Rico. Los organismos se separaron manualmente de micromuestras de 25 x 25 cm. Se identificaron ocho especies de lombrices de tierra: *Onychochaeta borincana*, *Pontoscolex corethrurus*, *P. melissae*, *P. spiralis* y *Pontoscolex* sp. pertenecientes a la Familia Glossoscolecidae, y *Amynthas gracilis*, *A. rodericensis* y una especie de feretimoide, pertenecientes a la Familia Megascolecidae. No emergió un patrón claro entre las propiedades del suelo y la estructura taxonómica y la abundancia de lombrices de tierra en estas plantaciones de café. Como esperado, se encontró una abundancia más alta de especies exóticas en las áreas más perturbadas. *P. corethrurus* se encontró en todas las plantaciones investigadas. Lares, el área con la segunda elevación más alta, tiene la densidad de especies más alta. Por otro lado, Jayuya, el área más aislada, tiene el número más alto de especies. *P. melissae*, una especie rara que sólo se había reportado de tres localidades cuando se describió en 1991, se encontró solamente en Jayuya.

Por mis hijos: Andrés y Eva

Acknowledgements

I would like to thank the Biology Department of the University of Puerto Rico at Mayagüez for providing me the opportunity and support I needed to continue my studies.

My heart-felt gratitude to the professors who contributed, encouraged and helped to achieve this goal:

Dr. José A. Amador of the University of Rhode Island for his help not only during the field visits but also for providing the soil analyses needed as part of this research.

Dr. Sonia Borges of the University of Puerto Rico, for corroborating the identification of species and for her countless readings and revisions of the manuscript.

The members of my Graduate Committee, doctors Jaime Acosta of the Biology Department and David Sotomayor-Ramírez from the Department of Crops and Agroenvironmental Sciences for their constant and sound advice and direction during the completion of this work.

I would also like to thank those who in one way or another helped me at different stages of this investigation, especially, Dr. Raúl E. Macchiavelli from the Department of Crops and

Agroenvironmental Sciences for his collaboration in the management and statistical analysis of data.

Also, I wish to thank Dr. Mónica Alfaro, president of my Graduate Committee, for not only accepting me as her student but for her constant guidance and supervision. Most of all, I want to thank her for her invaluable moral and academic support during this investigation and for her motivation and sound academic recommendations and personal advice. More than my mentor, Mónica is my friend.

Finally, I want to thank my family. To my aunt Vivian, who encouraged me to keep studying and was always available to lend a hand with my children. To my parents for their countless hours of babysitting and for their moral and academic support when I thought I couldn’t finish. Most importantly, I want to thank my husband who not only stayed with the kids but dug plenty of holes during field visits and helped me keep my eyes on the prize. Gracias, mi amor.

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Introduction

The alpha taxonomy of the earthworms of Puerto Rico has been very well studied and documented (Borges, 2004). Studies have also been undertaken on the microbiology and ecology of these annelids in the island. Some of the most recent ecological studies have dealt with forests with varying degrees of disturbance (Alfaro and Borges, 1996; Borges and Alfaro, 1997; González and Zou, 1999 a and b; González et al., 1999; Hendrix et al., 1999; Hubers at al., 2003; Lachnicht et al., 2002; Liu and Zou, 2002) and abandoned croplands and pastures (González et al., 1996; Sánchez-de León et al., 2003; Zou and González, 1997; Borges et al., 2006). However, the earthworm populations of commercial agroecosystems have not been reported. This thesis will focus on surveying the earthworm populations of commercially active sun and shaded coffee agroecosystems in Puerto Rico, one of the most important agroecosystems in the mountainous regions of the island (Alvarado and Monroig-Inglés, 2007). Also, their presence and density will be related to the soil quality of the plantations.

Undertaking the study of earthworm populations in commercially active sun and shaded coffee agroecosystems and their relationship to soil quality is of great importance because the knowledge obtained can be applied to improve sustainability and proper management of coffee and other agroecosystems. It may also provide more evidence for the use of earthworms as bioindicators of soil quality and fertility.

Literature Review

The earthworms of Puerto Rico are the best studied in the West Indies (Fragoso et al., 1995). Two peregrine species, *Pheretima rodericensis* (Grube, 1879) (now *Amynthas*) and *Pheretima biserialis* (Perrier, 1875) (now *Polypheretima elongata* Perrier, 1872), was the first group reported for the island (Michelsen, 1902). Until 1962, when Gates described *Trigaster rufa* (now *Neotrigaster rufa*), all the species known for Puerto Rico were exotic (Borges, 2004). By 1988 the known species included seven exotic and three native species and a new genus for the island and the Antilles, namely *Estherella*. Publications by Borges (1994), Borges and Moreno (1989, 1990a, 1990b, 1991, 1994) and James (1991) have added to the list and presently 29 species have been reported for Puerto Rico.

According to Borges (2004), the majority of earthworms in Puerto Rico are megascolecids and glossoscolecids, representing 35% and 41% of species, respectively. The author states that 38% of the reported species are exotic and that 64% or 18 of the native species belong to genus *Estherella*. Borges (2004) adds that the alpha taxonomy for the earthworms of Puerto Rico is nearly complete and that in the last fifteen years more than 20 studies, dealing with various aspects of these annelids in

Puerto Rico, have been published. Of course, this is not to say that the taxonomic work on the earthworms of Puerto Rico is finished. In fact, she mentions that there are regions of the island and the other islands of the Puerto Rican archipelago that have been only slightly studied or not studied at all.

There have been various ecological studies of the earthworms of Puerto Rico. According to Borges (2004), these studies have focused on the earthworms of forests with varying degrees of disturbance (Alfaro and Borges, 1996; Borges and Alfaro, 1997; González and Zou, 1999 a and b; González et al., 1999; Hendrix et al., 1999; Lachnicht et al., 2002; Liu and Zou, 2002; Hubers at al., 2003; Borges et al., 2006) or in abandoned pastures and croplands (González et al., 1996; Zou and González, 1997; Sánchez-de León et al., 2003). Other works have suggested that earthworms constitute the most important animal fractions of the soils in El Verde, Luquillo Experimental Forest (LEF) (Moore and Burns, 1970) and that their contribution to the litter and soil fauna of El Verde (LEF) is comparable to tropical forests in Nigeria and Mexico (Pfeiffer, 2006).

Although there are few ecological works, some suggestions have been proposed about the relationships between native and exotic species. These suggestions are that: (1) earthworm communities in natural and disturbed areas are composed of both native and exotic species and that native species prevail in natural ecosystems while exotic species predominate in disturbed areas; (2) natural ecosystems have earthworm communities with lower density and biomass than disturbed areas and (3) native earthworm survival in disturbed areas, depends on the time the site has been disturbed and the intensity of the destructive practices (Borges, 2004). Most of these studies, however, have focused on disturbed forests and abandoned pastures and croplands.

The earthworms of active agroecosystems have gone largely ignored in Puerto Rico and most of the tropics. Some works have focused on the effect of earthworm populations on nutrient cycling, crop production or soil structure, but most of these works either involved the inoculation of earthworms in the agroecosystem or were performed outside of the tropics (Lavelle et al., 1999). Sánchez-de León (2006) studied earthworm populations and soil microbial biomass and their relationship to coffee production in Costa Rica, however the analysis was conducted in experimental coffee plots not in commercially active coffee plantations.

Coffee was introduced in Puerto Rico at around 1736 and coffee plantations occupied some 77,000 hectares at the beginning of the 20th Century. At present it is estimated that

22,000 hectares are dedicated to this crop (Alvarado and Monroig-Inglés, 2007) but it still is one of the most important crops in the island producing over $41,000,000 at the farm level in sales or 8.11% of total agricultural sales in Puerto Rico. It is surpassed only by horticulture specialties and plantains in sales and more land (38,535 hectares) is dedicated to this crop than any other in Puerto Rico (USDA, 2007). The coffee producing region of Puerto Rico includes the municipalities of Adjuntas, Ciales, Jayuya, Lares, Maricao, Las Marías, San Sebastian and Utuado. Plus, it also includes parts of the municipalities of Añasco, Guayanilla, Juana Díaz, Mayagüez, Moca, Orocovis, Peñuelas, Sabana Grande, San Germán, Villalba and Yauco (USDA, 2007).

The Conjunto Tecnológico Para la Producción de Café (1999) has stated that the coffee growing region of Puerto Rico has some predominant ecologic factors and characteristics. Its temperature fluctuates between 20°C and 27°C with an annual average of 24°C, rainfall is between 1900 to 2400 mm with a short dry season from December to early April, relative humidity is usually between 70 to 85% and the region´s altitude is between 150 to 190 meters. The soils of the region are usually acidic (pH between 4.0 and 5.5) with low natural fertility and have clayey texture. The soils belong to three Orders: Ultisol, Inceptisol and Oxisol. This, plus the fact that they are deep soils with organic matter content between 3 and 6%, have good drainage and are moderately heavy make these soils appropriate for the production of coffee (Conjunto Tecnológico Para la Producción de Café, 1999).

Objectives

This thesis had four objectives:

1. Identify the earthworms of sun and shaded coffee agroecosystems in Puerto Rico.
2. Relate their distribution with the physicochemical characteristics of the soil.
3. Compare the oligochaetofauna of the two systems of coffee agriculture, sun and shaded, employed in Puerto Rico.
4. Establish new collection records.

Materials and Methods

Three study sites within the municipalities of Las Marías, Lares and Jayuya were established in October 2006 by López (2008). Selection of the sites was based on soil type, vegetation and climactic factors. The sites were chosen for having soil types representative of those of the coffee growing regions of Puerto Rico. The sites are located in the Wendolí and Serrallés farms of Barrio La Pica in Jayuya, in the Buena Vista farms of Barrio Bartolo in Lares and the Las Juanitas farms and "predio" Miguel Avilés in the Barrio Furnias in Las Marías. According to López (2008), Jayuya's soil is an Oxisol from the series Los Guineos (*very fine, kaolinick, isothermic, Humic Hapludox)*, the soil in Lares is an Inceptisol from the series Anones (*fine, parasequic, isohyperthermic, Humic Drystrudeps*) and in Las Marías the soil is an Ultisol from the series Humatas (*very fine, parasequic, isohyperthermic, Typic Haplohumults*). Each farm has fields including the two agroecosystems, coffee under full sun and coffee under shade, and a secondary forest that served as a control area. There were three replicates in each agroecosystem and control, each with plot sizes of approximately 400 m^2. Table 1 shows the climactic characteristics and the location of the study sites.

These plantations are between 8 and 15 years old and have an approximate density of 3,500 plants per hectare in sun coffee and 2,500 plants per hectare in shaded coffee (López, 2008). According to López (2008), the Caturra, Borbón and Limaní varieties of *Coffea arabica* species are the predominant crops in these plantations. They were managed according to the program set forth by the owners and administrators of the plantations.

Table 1: Location and climatologic characteristics of the study sites. Reproduced from López (2008)

		Characteristics				
Municipality	**Ecosystem**	Latitude (North)	Longitude (West)	Elevation (m)	Average annual precipitation (mm)	Average annual temperature (°C)
Jayuya	Sun	18°10’50”	66°37’50”	765	1935	23°
	Shade	18°09’41”	66°38’46”	785		
	Forest	18°09’44”	66°38’46”	817		
Lares	Sun	18°11’43”	66°50’55”	575	2290	24°
	Shade	18°11’59”	66°50’49”	636		
	Forest	18°11’46”	66°50’55”	605		
Las Marías	Sun	18°14’44”	66°00’25”	297	1870	26°
	Shade	18°14’39”	66°00’08”	288		
	Forest	18°14’43”	66°00’26”	285		

López (2008) also adds that the areas of secondary forest belong to the Very Wet Subtropical Forest Life Zone using the ecologic classification system created by Holdridge (1969). This life zone is characterized by forests with a canopy with more than twenty meters in height and a great diversity of tree species (Ewel and Whitmore, 1973). The area of study, on the other hand, is dominated by trees that are thirty years old or younger, as stated by López (2008).

Earthworm sampling in Las Marías and Jayuya was performed on November 14, 2007, while sampling in Lares was performed on December 7, 2007. Sampling in all three sites followed the same method. Earthworms were extracted by handsorting twenty-seven 25 cm x 25 cm (0.0625 m^2) haphazardly selected samples. The detection limit for this size sample is 16 ind/m^2. Worms were removed from the litter as well as from soil to a depth of 30 cm and preserved in a 4% formalin solution. This solution was replaced as needed until it remained clear, and the specimens were weighed a month after collection. Specimen identification was done using the taxonomic key by Borges (1996).

Soil texture was determined using the rapid method proposed by Kettler et al. (2001). Subsets were dried for other analyses. Percentage of moisture, organic matter, total C, total N and pH were obtained from Sotomayor-Ramírez (2008).

A $\log_{10}$ transformation was applied to the density data in order to equalize variances and normalize distributions. The two-way analysis of variance (ANOVA) tests with replication was applied to the density data of earthworms to test for differences between soil types and between the two coffee agroecosystems and/or interaction effects. Any significant differences ($P < 0.05$) were further compared using Tukey Test Range (HSD) multiple mean comparison test. All data was analyzed using the statistical package from Infostat Software Version 6.12, 2005.

Results

The physicochemical characteristics of the soil have been reported by Sotomayor-Ramírez (2008). The complete results of the statistical analyses are included in Appendix A. Appendix B shows the earthworm species and their density and biomass at each location.

Physicochemical characteristics of the soil

Soil texture. The soils from Las Marías (Table 2) are mainly composed of silt. The highest average percentage (74.00%) of this component was found in the shaded agroecosystem. As Table 2 illustrates, the soils of Jayuya are mostly composed of silt; with the highest average percentage (68.57%) of this component found in the coffee-shade agroecosystem. The secondary forest of Jayuya is the site where the highest percentage of sand was recorded at all locations. Lares (Table 2) has soils that are also mostly composed of silt and its highest average percentage (57.47%) was recorded in the coffee-shade agroecosystem.

The results from the analysis of variance (ANOVA) indicate that the location significantly affects the percentage of silt ($p \leq 0.0058$), sand ($p \leq 0.0432$) and clay ($p \leq 0.0093$) (Table 3). This analysis also implies that the treatment significantly affects the percentage of silt ($p \leq 0.0234$), sand ($p \leq 0.0078$) and clay ($p \leq 0.0086$) (Table 3).

Table 2: Average physicochemical characteristics of the soil at the three localities. Percentage of moisture, organic matter, total C, total N and pH obtained from Sotomayor-Ramírez (2008). OM: Organic matter

Characteristic	*Las Marías*			*Jayuya*			*Lares*		
	Sun	Shaded	Forest	Sun	Shaded	Forest	Sun	Shaded	Forest
Sand (%)	15.80	16.83	25.33	27.13	18.37	40.67	14.40	23.23	29.00
Silt (%)	65.30	74.00	61.23	44.96	68.57	51.31	51.80	57.47	51.80
Clay (%)	18.80	9.12	13.43	27.91	13.06	7.93	33.70	19.27	19.20
Moisture (%)	48.70	44.73	52.14	43.70	60.77	73.88	43.90	46.44	51.97
OM (%)	6.73	6.73	6.43	7.17	6.87	7.47	5.40	4.43	6.17
Total C (%)	3.13	4.22	4.20	3.34	4.01	6.87	2.60	4.18	5.10
Total N (%)	0.32	0.44	0.43	0.34	0.43	0.68	0.30	0.42	0.50
pH	4.23	4.23	5.73	4.73	4.00	4.57	4.30	4.00	4.30

Table 3: Significance levels from ANOVA for the physicochemical characteristics of the soil. OM: Organic matter

	% Sand p-value	% Silt p-value	% Clay p-value	% Moisture p-value	% OM p-value	% Total C p-value	% Total N p-value	pH p-value
Model	0.0289	0.0076	0.0126	0.0005	0.7062	0.0009	0.6085	0.3882
Location (Loc)	0.0432	0.0058	0.0093	0.0008	0.8195	0.1525	0.6262	0.6350
Treatment (Trt)	0.0234	0.0078	0.0086	0.0093	0.5080	0.0001	0.3555	0.0634
Loc*Trt	0.1779	0.5096	0.5283	0.0095	0.5892	0.1150	0.6785	0.8852

Soil moisture. The highest average soil moisture (73.88%) was recorded for the forest of Jayuya and the lowest average percentage of moisture (43.70%) was in the coffee-sun agroecosystem of Jayuya (Table 2). As seen in table 3, the location ($p \leq 0.0008$) and the treatment ($p \leq 0.0093$) significantly affect the moisture of the soil. Also, the variance analysis indicates that there is an interaction between the location and the treatment ($p \leq 0.0095$) (Table 3).

Percentage of organic matter. The soil with highest average organic matter (7.47%) content was found at the forest of Jayuya, while the lowest average percentage (4.43%) was recorded in the coffee-shade agroecosystem of Lares (Table 2). The analysis of variance (ANOVA) does not indicate any relationship between the percentage of organic matter and the location ($p \leq 0.8195$) and the treatment ($p \leq 0.5080$) or that there is an interaction ($p \leq 0.5892$) between location and treatment.

Percentage of total organic carbon. The highest average percentage of total organic carbon (6.87%) in soil was recorded in the forest of Jayuya and the lowest (2.60%) in the coffee-sun agroecosystem of Lares (Table 2). The analysis of variance indicates a significant relationship between the percentage of total organic carbon and the treatment ($p \leq 0.0001$) (Table 3). However, the analysis of variance (ANOVA) does not indicate a significant relationship between the percentage of total organic carbon and the location ($p \leq 0.1525$) or an interaction ($p \leq 0.1150$) between the location and the treatment (Table 3).

Percentage of total nitrogen. The forest of Jayuya had the highest average percentage of total nitrogen content (0.68%) and the lowest average percentage (0.30%) was recorded in the sun-coffee agroecosystem of Lares (Table 2). Nevertheless, the analysis of variance (ANOVA) does not indicate a significant relationship between the percentage of total nitrogen and the location ($p \leq 0.6262$) and treatment ($p \leq 0.3555$) or an interaction ($p \leq 0.6785$) between the location and the treatment (Table 3).

pH. The most acidic soils (pH = 4.0) were recorded for the coffee-shade agroecosystems of Lares and Jayuya while the most basic soil (pH = 5.7) was found in the forest of Las Marías (Table 2). Still, the analysis of variance (ANOVA) does not indicate a significant relationship between the pH and the location ($p \leq 0.6350$) and treatment ($p \leq 0.0634$) or an interaction ($p \leq 0.8852$) between the location and the treatment (Table 3).

Earthworms

The results of the taxonomic earthworm structure at sampling stations are summarized in Figure 1.

Species. Eight earthworm species were identified: *Onychochaeta borincana* Borges, 1994, *Pontoscolex corethrurus* (Müller, 1857), *Pontoscolex melissae* Borges and Moreno, 1991, *Pontoscolex spiralis* Borges and Moreno, 1991 and *Pontoscolex* sp., which belong to the Glossoscolecidae family and *Amynthas* *gracilis* (Kinberg, 1867), *Amynthas rodericensis* (Grube, 1879) and a pheretimoid sp., which belong to the Megascolecidae family. *Pontoscolex* sp. and the pheretimoid could only be identified to the genus level because they were immature or macerated and, because of this, could not be classified as native or exotic. All the megascolecids are exotic. Among the glossoscolecids, *O. borincana*, *P. melissae* and *P. spiralis* are native and *P. corethrurus* is considered a peregrine species.

The earthworm species distributions at the sampling sites are shown in Figure 1. Only three of the eight species were found in the samples from Las Marías. *Pontoscolex corethrurus* was the most abundant species found in these samples. *A. rodericensis* was found only in the coffee-sun ecosystem and *P. spiralis* only in the coffee-shade ecosystems. No earthworms were found in any of the samples from the secondary forest in Las Marías. The samples from Jayuya had the most species, with five of the eight species found being reported from these sites. P*ontoscolex corethrurus* was reported in all the ecosystems in Jayuya and also was the most abundant species. Of the eight species reported only three were found in the samples from Lares.

Four species were found in the samples from the coffee-sun ecosystem: *P. corethrurus, A. rodericensis,* and a pheretimoid and *Pontoscolex* sp. *Pontoscolex corethrurus* was found in all the samples from the coffee-sun ecosystem in Las Marías, Jayuya and Lares and is the most abundant species of this ecosystem. *A. rodericensis*, the pheretimoid sp. and *Pontoscolex* sp. were also reported in the coffee-sun ecosystem in the samples from Las Marías, Jayuya and Lares, respectively (Figure 1).

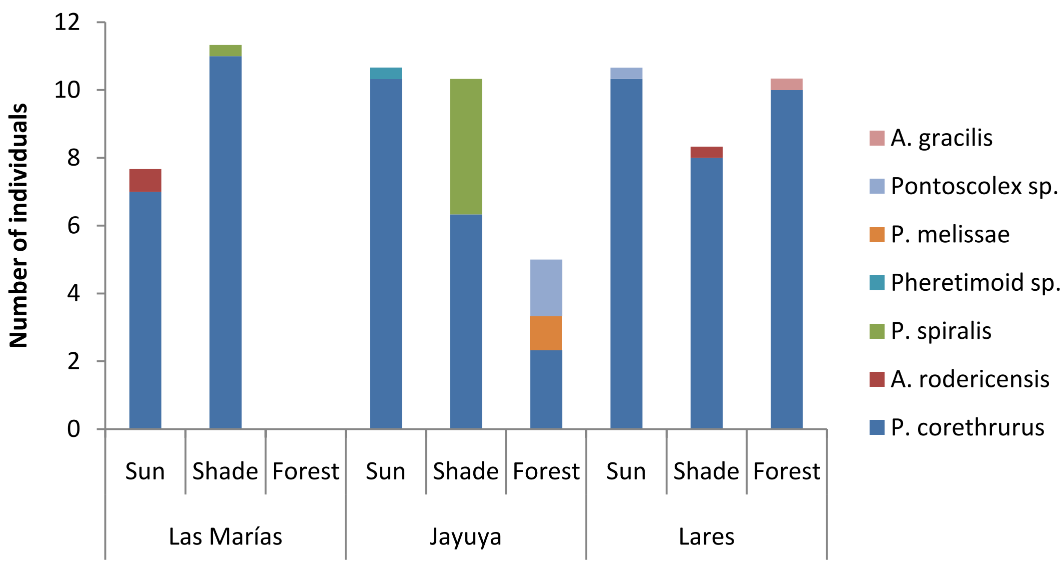

Figure 1: Average number of individuals and species distribution

As evident from Figure 1, only three species were found in the coffee-shade agroecosystem: *P. corethrurus*, *P. spiralis* and *A. rodericensis*. *P. corethrurus* was found in all the samples and was, by far, the most abundant species from this agroecosystem. *P. spiralis* was found in the coffee-shade agroecosystem from Jayuya and Las Marías, while *A. rodericensis* was reported only from the coffee-shade agroecosystem from Lares (Figure 1).

On the other hand, four species were reported from the secondary forest: *P. corethrurus*, *P. melissae*, *Pontoscolex* sp. and *A. gracilis*. No earthworms were found in the forest from Las Marías. *P. corethrurus* was found in all the samples and was the most abundant species from this agroecosystem, as well. *P. melissae* and *Pontoscolex* sp. were also reported from the forest of Jayuya and *A. gracilis* was reported only from the forest in Lares.

Density. The highest average density per location was recorded for Lares (156.44 ind/m^2) (Figure 2). The highest average density of earthworms per treatment (181.33 ind/m^2) was recorded in the coffee-shade ecosystem of Las Marías, while the lowest average density (0 ind/m^2) was in the forest of Las Marías (Figure 3). Statistical analysis showed a significant relationship between the density and the treatment ($p \leq 0.0211$). However, there is no relationship between the location ($p \leq 0.4458$) and the density and all these factors have no interaction ($p \leq 0.4060$) (Table 4). Sun-coffee and shade-coffee agroecosystems had similar earthworm density and biomass while secondary forests had lower values. Shade-coffee agroecosystems had the most species.

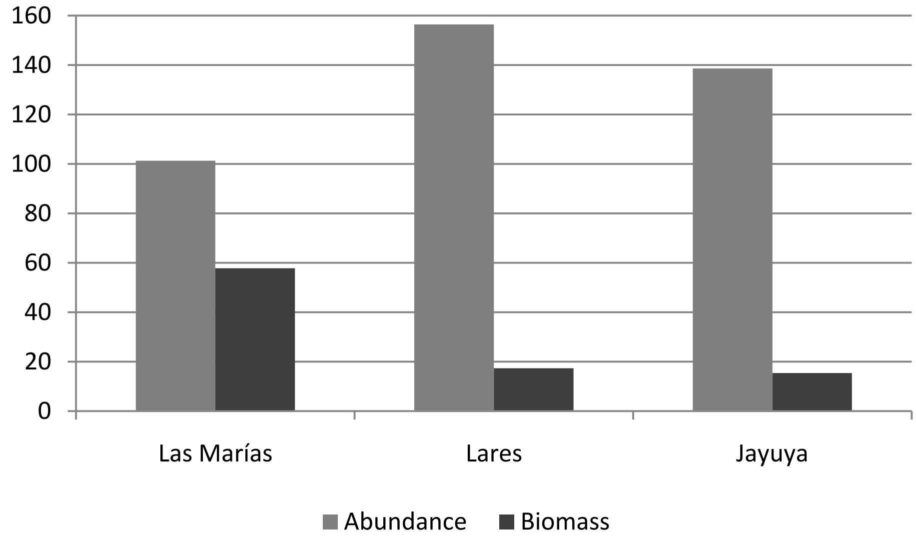

Figure 2: Average earthworm density (ind/m^2) and biomass (g/m^2) per location

The highest average density (181.33 ind/m^2) in Las Marías was recorded in the coffee-shade agroecosystem and the lowest average density (0 ind/m^2) (Figure 3). In Jayuya, the highest average density (170.67 ind/m^2) was reported in the coffee-sun agroecosystem, while lowest average density (80 ind/m^2) was recorded in the secondary forest (Figure 3). Lares' highest average density (170.67 ind/m^2) was recorded in the coffee-sun agroecosystem (Figure 3), while the lowest average density (120.75 ind/m^2) was reported from the secondary forest (Figure 3).

Of all the treatments the one with the highest average density was the coffee-shade agroecosystem (159.99 ind/m^2). The coffee-shade agroecosystem from Las Marías had the highest average density (181.33 ind./m^2) and the one from Lares had the lowest (133.33 ind./m^2). The second highest average density was recorded for the coffee-sun agroecosystem (154.67 ind./m^2). The highest average density (170.67 ind/m^2) for this treatment was recorded in Jayuya and Lares and the lowest average density (122.67 ind/m^2) in Las Marías (Figure 3). The lowest average density (66.91 ind/m^2) was recorded in the secondary forest (Figure 3). Of this treatment the highest average density (120.75 ind./m^2) was recorded in Lares and the lowest average density (0 ind./m^2) in Las Marías.

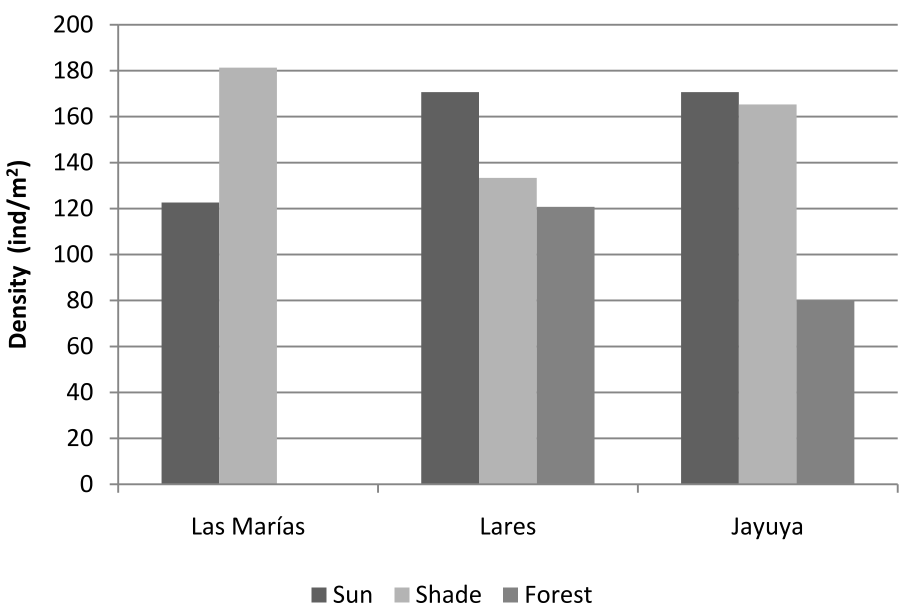

Figure 3: Average density (ind/m^2) of earthworms per location and treatment

Table 4: Significance levels from ANOVA for density and biomass

	Density	Biomass
Model	0.1337	0.0001
Location (Loc)	0.4458	0.0001
Treatment (Trt)	0.0211	0.0101
Loc*Trt	0.4060	0.0024

Biomass. The highest average biomass per location was recorded for Las Marías (57.78 g/m^2) (Figure 2). The highest average biomass per treatment (111.89 g/m^2) was recorded in the shade-coffee agroecosystem of Las Marías. The lowest total biomass (0g/m^2) was recorded in the forest of Las Marías (Figure 4). As seen in table 4, the statistical analyses suggest a significant relationship between the biomass and the location ($p \leq 0.0001$) and treatment ($p \leq 0.0101$). Also, there is an interaction between these factors ($p \leq 0.0024$) (Table 3).

The highest average biomass (111.89 g/m^2) in Las Marías was recorded in the coffee-shade agroecosystem and the lowest average biomass was recorded in the secondary forest (0 g/m^2) (Figure 4). In Jayuya, the highest average biomass (82.59 g/m^2) was reported in the coffee-sun agroecosystem and the lowest average biomass (25.65 g/m^2) was recorded in the secondary forest (Figure 4). Lares' highest average biomass (75.18 g/m^2) was recorded in the coffee-sun agroecosystem and the lowest average biomass (34.37 g/m^2) from the coffee-shade agroecosystem (Figure 4).

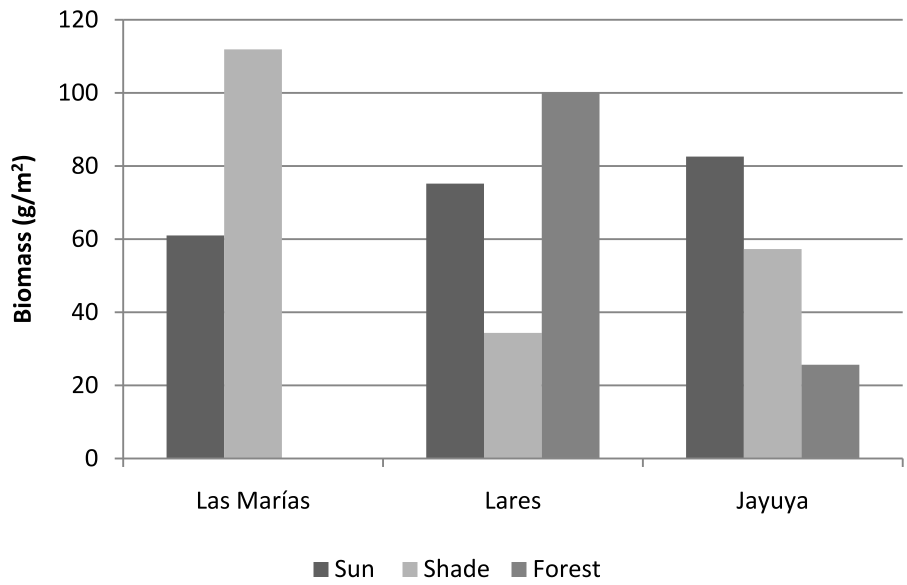

Figure 4: Average biomass (g/m^2) of earthworms per location and treatment

The treatment with the highest average biomass was the coffee-sun agroecosystem (72.92 g/m^2). The highest average biomass for the sun agroecosystem (82.59 g/m^2) was recorded in Jayuya and the lowest (61 g/m^2) in Las Marías (Figure 4). The treatment with second highest average biomass (67.84 g/m^2) was the coffee-shade agroecosystem. The coffee-shade agroecosystem from Las Marías had the highest biomass (111.89 g/m^2), while the lowest biomass was recorded in Lares (34.37 g/m^2). On the other hand, the lowest average biomass (41.91 g/m^2) was recorded in the secondary forest (Figure 4). Of these ecosystems, the forest from Lares had the highest biomass (120.75 g/m^2) and the lowest was recorded from Las Marías (0 g/m^2).

Discussion

Earthworms are especially sensitive to moisture and organic matter content of the soil they inhabit. However, preferences for these and other soil physical and chemical characteristics have been determined for very few species (Edwards and Bohlen, 1996). Not all species have the same requirements, for example some tolerate soil moisture as low as 10% while other require higher values. Several scientists state that most earthworm species favor a neutral pH but species have been reported from soils from a pH 2.0 to 9.0 (Edwards and Bohlen, 1996). Organic matter content of the soil influences the distribution of earthworms. In general terms, worms are with poor organic matter do not hold many earthworms. Soil physical and chemical characteristics found in this study seem to be within the tolerance range for many earthworm species. *Pontoscolex corethrurus*, the most numerous species found here, has been reported from a variety of conditions; pH 3.8-8.2, organic matter 0.9-12.6, sand 3-91%, clay 6-87% (Fragoso et al., 1999a).

Most agroecosystems, especially crop sites, are characterized by low earthworm density and species number (Fragoso et al., 1999a). The number of species found at each location here compares well with other plantations. A total of three species were found in the sun and four in the shaded coffee plantations. González et al. (1996) only reported two species in mahogany and pine plantations. Zou (1993) found two species, *P. corethrurus* and *A. gracilis*, in tree plantations in Hawaii. In coffee plantations in Costa Rica, Sánchez de León et al. (2006) only found a total of two species. However, Feijoo and associates (2007) found 11 species of earthworms in mixed coffee culture and 12 species in coffee monoculture.

The density and biomass are higher in coffee plantations than in secondary forests. This agrees with Fragoso et al. (1995) and Borges (2004) who state that earthworm communities in disturbed areas have a higher density and biomass than those in natural ecosystems. The average earthworm density and biomass for coffee-sun (154.67 ind/m^2) and coffee-shade (159.99 ind/m^2) agroecosystems were higher than those reported in other plantations: pine and mahogany (González et al., 1996), eucalyptus (Zou, 1993) and flower bulbs and vegetables (Didden, 2006). These values are also higher than those reported in other coffee plantations (Hairah et al., 2006). In experimental plots in Costa Rica, Sánchez-de León et al. (2006) found lower earthworm densities in sun managed systems (both with conventional and medium conventional treatments), in *Terminalia* shaded coffee plantations (in high conventional, medium conventional and organic treatments) and in *Erythrina* shaded plantations (with high conventional treatments). In Costa Rica, earthworm density and fresh weight were higher in the *Erythrina* shaded agroecosystems with medium conventional (186 ind/m^2) and organic treatments (265 ind/m^2).

As expected, earthworm density and biomass in these coffee plantations were considerably lower than those reported for active pastures in Puerto Rico (Zou and González, 1997; Zou et al., 2006) and grasslands in the Netherlands (Didden, 2006). Fragoso et al. (1995) stated that active pastures are characterized by their high earthworm density and biomass.

Earthworm density and biomass may vary according to the type of culture and whether they are mixed or not. For example, Zou (1993) reported higher densities in plantations with pure *Albizia* and those with a mixture of *Eucalyptus* and *Albizia* than in plantations of pure *Eucalyptus*. Lower density was informed in diversified cultures compared with non-diversified cultures (Huerta et al., 2006). In this investigation, earthworm density and biomass were very similar in both coffee-shade (159.6 ind/m^2, 73.96 g/m^2) and coffee-sun (155 ind/m^2, 76.06 g/m^2) agroecosystems. This contrasts with the findings of Hairah and collaborators (2006) who found differences between shaded multistrata coffee (with fruit and timber trees), shaded coffee and sun monoculture coffee. Multistrata coffee had the highest density (149 ind/m^2) and biomass (18 g/m^2). The shaded coffee and sun coffee had considerably lower but similar values (83 ind/m^2, 7 g/m^2 and 88 ind/m^2, 12 g/m^2, respectively). Sánchez-de León et al. (2006) also found differences in earthworm densities between sun and shaded treatments. Furthermore, they found that earthworm density was higher in *Terminalia* shaded coffee than in *Erythrina* shaded coffee.

These differences could be caused by diverse agricultural practices or soil characteristics. In this investigation the relation between earthworm density and the soil properties did not show a clear trend towards any variable. The different kinds of treatments at each location act independently of each other, and no real variables were identified to explain the distribution patterns.

Earthworm density and biomass of the secondary forests in this research were lower than those found in both coffee plantations. This was also reported by González and associates (1996) when comparing mahogany and pine plantation with adjacent secondary forests. Although lower than the coffee plantations, the earthworm density in our secondary forests is dramatically higher than those informed in Bosque del Pueblo in Adjuntas, Puerto Rico, a region severely disturbed by human intervention

(Borges et al., 2006). However, it was similar to the one reported in a *Spathodea campanulata* Beauv. forest in the north coast of Puerto Rico (Lugo et al., 2006), which, in turn, was more similar to those of mature forests than to secondary forests or pastures (Zou and González, 1997). The absence of earthworms from Las Marías' forest may be a consequence of the random sampling employed and the aggregate distribution of these organisms.

The eight species found in this investigation had already been reported for Puerto Rico (Borges 2004) but do constitute new records for these areas that had never been studied. All the species belonging to the Family Megascolecidae, *Amynthas gracilis*, *Amynthas rodericensis* and the pheretimoid are exotic, while the glossoscolecids *Onychochaeta borincana*, *Pontoscolex melissae*, *Pontoscolex spiralis*, are native species for the region (Borges 2004). *Pontoscolex corethrurus* is considered an exotic peregrine species. *O. borincana*, *P. melissae* have only been collected from Puerto Rico. *P. spiralis* has also been reported from Martinique (Fragoso et al., 1999a). *O. borincana* has been reported from areas used for agriculture and is fairly common, especially in disturbed areas (Borges 2004). *P. melissae*, on the other hand, has rarely been collected, only in Laguna Tortuguero, Hormigueros and Ponce, and this constitutes the only other time it has been reported since it was described in 1990 (Borges and Moreno, 1990a). *P. melissae*, *P. spiralis* and *P. corethrurus* have been found in disturbed areas (Fragoso, et al., 1995).

The color of *Amynthas rodericencis* can vary from gray, red, or brown and it has a white, red, gray, or red to pinkish clitellum. Its length runs from 67 to 154 mm and weighs between 0.71 to 2.2 g. It has been reported from various sites in Puerto Rico, including Guajataca Forest, Canóvanas, Guayama, Cidra, Guaynabo and Mayagüez, among others. Its distribution does not seem to be limited by elevation, the composition of the soil or by the amount of leaf litter present. It has been found with *Onychochaeta borincana*, *Pontoscolex corethrurus*, *Estherella aguayoi* Borges and Moreno, 1991, *Polypheretima elongata* (Perrier, 1872), *Pontoscolex cynthiae* Borges and Moreno, 1991, *Eudrilus eugeniae* (Kindberg, 1867), *Amynthas corticis* (Kindberg, 1967), *Pontoscolex melissae*, *Ocnerodrilus occidentalis* Eisen, 1878, *Pontoscolex spiralis*, *Borgesia sedecimsetae* (Borges and Moreno, 1991) and *Drawida barwelli* (Beddard, 1886) (Borges, 1988). More recently, it has been reported in *Dacryodes excelsa* and *Heliconia caribea* communities (González and Zou, 1999) and a wet tropical forest (Zou et al., 2006) both in the Luquillo Mountains of Puerto Rico. Borges (Pers. Comm.) believes that *A.*

rodericensis is an epigeic species. It a common exotic earthworm that has been found in several tropical natural ecosystems and agroecosystems such as crops, pastures and tree plantations (Fragoso et al., 1999a).

Amynthas gracilis is an epigeic species (Feijoo et al., 2007) of grayish color with a similarly pigmented clitellum. It measures 117 to 118 mm long and weighs between 1.04 and 1.14 g. Until now, this species has only been reported in Guayama in a soil with a high percentage of clay and in a region belonging to Subtropical Wet Forest Life Zone (Borges, 1988). Borges (1988) reported that *A. gracilis* has been collected accompanied by the following species: *Polypheretima elongata*, *Onychochaeta borincana* and *Pontoscolex corethrurus*. Fragoso et al. (1999a) mention that *A. gracilis* has been found in tropical agroecosystems such as crops, pastures, tree plantations and fallows. They include this species among the exotics with high range of edaphic (or environmental) plasticity that are also tolerant to very low concentrations of organic matter, nutrients and nitrogen.

Onychochaeta borincana is a species that lacks pigmentation, weighs between 1.1 to 2.13 g and measures between 80 to 160 mm in length. This species has been found in various areas in Puerto Rico, including Laguna Cartagena, Tropical Dry

Forest and Very wet Lower Mountain Forest. It has also been reported in a wide range of elevations, from 90 to 1097 m, and in different conditions and types of soils (dry or humid, with plenty or few roots, sandy). It’s one of the most collected species in Puerto Rico and Borges (1988) reported it has been found along with *P. corethrurus* in 17 samples and *P. elongata* in 5 occasions. In addition, it was reported from Bosque del Pueblo in Adjuntas (Borges et al., 2006) and *Spathodea campanulata* forests in the north coast of Puerto Rico (Lugo et al., 2006). Borges (Pers. Comm.) believes *O. borincana* is an endogeic species.

Pontoscolex melissae lacks pigmentation as well, but when preserved in formalin it turns beige with a red clitellum. It measures from 36 to 56 mm long and weighs between 0.13 and 0.31 g. Like *P. corethrurus* and *P. vandersleeni*, *P. melissae* has the setae in the posterior segments of the body in a quincunx pattern (Borges, 1988). Borges and Moreno (1990a) reported this species from Tortuguero Lagoon, Hormigueros and Mayagüez and it has been found with *Polypheretima elongata*, *Drawida barwelli*, *Onychochaeta borincana*, *Pontoscolex corethrurus*, *Amynthas corticis*, *Amynthas rodericensis* and *Pontoscolex cynthiae*. Borges (Pers. Comm.) believes *P. melissae* is an endogeic species.

Pontoscolex spiralis is red but after treatment with formalin it turns beige with a red clitellum. It measures from 91 to 124 mm long and weighs between 0.34 and 0.63 g. Like *Pontoscolex eudoxiae*, *Pontoscolex nogueirae* and *Pontoscolex maracaensis*, *P. spiralis* has regular setal rows (Borges, 1988). This species has been found in Humacao, Maunabo, Bosque de Guavate, Mayagüez, Río Abajo Forest, Maricao, Toro Negro Forest, Guaynabo, Fajardo and El Verde in the Forest of Luquillo. According to Borges (1988), it has been collected in different life zones, elevations (15 to 792 m) and types of soils and with the following species: *Pontoscolex corethrurus*, *Polypheretima elongata*, *Onychochaeta borincana*, *Amynthas rodericensis* and *Eudrilus eugeniae*. Lugo et al. (2006) reported *P. spiralis* as the dominant species in secondary *Spathodea campanulata* forests in the north coast of Puerto Rico and González and Zou (1999b) found *P. spiralis* in *Dacryodes excelsa* Vahl communities in the Luquillo Long-Term Ecological Research site in Luquillo, Puerto Rico. Fragoso and collaborators (1999a) state that *P. spiralis* belongs to the endogeic mesohumic ecological category. They state that this is one of the native species found in agroecosystems, specifically in crops, pastures and tree plantations in Martinique.

Pontoscolex corethrurus is also an endogeic mesohumic species (Fragoso et al., 1999a) that lacks pigmentation but when conserved in formalin it turns beige while the clitellum remains red. It measures between 48 and 113 mm in length and weighs between 0.32 and 0.78 g. It is considered the most widely distributed earthworm species due to human interaction, having been transported since the 1500's from its place of origin somewhere in the American tropics (Gates, 1972). Of all the Puerto Rican species reported by Borges (1988) it was the one most commonly collected and was found in a wide variety of conditions: clay soils and sandy soils, dry soils and saturated soils, and from greenhouses to landfills and forest reserves. It was mostly collected in the Subtropical Wet Forest Life Zone. *P. corethrurus*, the most common and abundant species in this study, is also the most common earthworm species in Puerto Rico and has been collected in areas subjected to human impact, different types of soils, even those with low organic matter content and in soils that are dry and soils that are saturated with water (Borges, 2004). As cited by Huerta et al. (2006), Fragoso (2001) has described this species as an exotic cosmopolitan species that inhabits agroecosystems in tropical areas. *P. corethrurus* was the dominant species in this study, as has occurred in coffee plantations in Costa Rica (Sánchez-de

León et al., 2006), in four different land use systems in Brazil (Nunes et al., 2006), in Bosque del Pueblo, Puerto Rico (Borges et al., 2006), in agroecosystems under intense cultivation practices and coffee mixed with citrus in the La Vieja River Basin in Colombia, (Feijoo et al., 2007) and in all agroecosystems studied in central and southeastern Tabasco, Mexico (Huerta et al., 2006). It has also been reported from Baño de Oro in the Luquillo Experimental Forest in Puerto Rico (Borges and Alfaro, 1997), *Spathodea campanulata* forests in northern Puerto Rico (Lugo et al., 2006), in *Dacryodes excelsa* and *Heliconia caribea* communities in the Luquillo Long-Term Ecological Research site in the Luquillo Experimental Forest, Puerto Rico (González and Zou, 1999b) and in an active pasture and a primary native wet forest in the northeastern Luquillo Mountains of Puerto Rico (Zou et al., 2006). *P. corethrurus* has been collected from natural ecosystems in the tropics and is also the most common exotic species found in several tropical agroecosystems like crops, pastures, tree plantations and fallows (Fragoso et al., 1999a). This species is included among the exotics with high range of edaphic (or environmental) plasticity that are also tolerant to very low concentrations of organic matter, nutrients and nitrogen (Fragoso et al., 1999a).

Fragoso et al. (1999b) discuss that the predominance of a particular ecological category of earthworms depends on environmental factors. For example, epigeics predominate in colder regions while engogeic species dominate in the tropics. They state that tropical endogeics can shift to epigeic communities if soil nutrients and rainfall are low. Many changes occur in the earthworm communities of natural ecosystems that are converted to agroecosystems, among them the ecological categories. There is a trend towards an endogeic dominance in agroecosystems (Fragoso et al., 1999b). The two megascolecids species found in this study are epigeic while the four glossoscolecids species are endogeic. Endogeics and epi-endogeics seem to be the most important categories in agroecosystems because they are able to transform the edaphic profile with their burrows and cast production (Fragoso et al., 1999b).

Exotic species are common in disturbed tropical ecosystems. Some exhibit very wide ranges of environmental plasticity as was informed for *P. corethrurus* and *A. gracilis* by Fragoso et al. (1999a). These scientists studied 20 exotic and 27 native earthworm species and determined that the degree of tolerance or environmental plasticity was higher in exotics. This characteristic defines them as most important in maintaining soil fertility in agricultural lands (Lee, 1987). Most of the native species are not so tolerant and are usually restricted to natural ecosystems except in countries with low-input technologies (India, for example) or low annual precipitation. Very few native species, such as *Onychochaeta elegans* in Cuba, are common in tropical agroecosystems (Fragoso et al., 1999a).

Earthworm communities in both natural and disturbed ecosystems are composed of native and exotic species but the number of native species is higher than the exotic ones in natural ecosystems (Borges, 2004). This seems to be the pattern observed in this study where both exotic and native species were found in all locations and treatments, except the coffee-sun agroecosystem were only exotics were collected. Because statistical analyses showed no significant differences in soil properties this could be due to collection methods or to agricultural practices. Typically, “the intensity of agricultural practices is negatively correlated with the amount of native species and the total density and biomass of earthworms” (Fragoso et al., 1999b, page 28). Coffee-sun treatments have a much higher number of coffee plants (3,500/hectare) than shaded treatments (2,500/hectare). Thus, coffee-sun agroecosystems would require more agricultural activities which, in turn, may affect the establishment of native earthworm species that are more susceptible to the amount and duration of disturbances (Fragoso et al., 1995). Disturbances such as surface clearing for planting, tillage, and the use of fertilizers and insecticides in these areas may favor the successful colonization of exotic species that are adapted to a wider range of environmental variability.

González et al. (1996) studied earthworms in pine and mahogany plantations comparing them with contiguous secondary forests and found that earthworm density, fresh weight and number of native species was higher in the secondary forest. Zou and González (1997) did not find native species in young secondary succession communities. Sánchez-de León, et al. (2003) found native species in mature secondary forests with more than 77 years of abandonment but only exotic species in active pastures and young secondary forests (25-40 years after abandonment). The number of native species in impacted ecosystems depends on the time end intensity which with an area has been subjected to stress (Fragoso et al., 1995). The fact that only one native species was found in the secondary forests in this study may be a consequence of the collection design or to the number of years these forests have been abandoned perhaps a mere thirty years. In this case, the only forest with native species *P. melissae* was Jayuya, which also happens to be the most isolated location of all.

Conclusions

The analyses of the results of the study of the earthworms of these coffee plantations in Puerto Rico allow the following conclusions:

- The density and biomass is similar to those reported from other areas, even though the study areas have undergone anthropogenic disturbances.
- A total of eight earthworm species were identified. As in other earthworm communities a combination of exotic (4) and native (3) species were found.
    - Sun coffee had only exotic species
    - Shaded coffee and secondary forests had exotic and native earthworm species
    - Shaded coffee had the highest number of species
- *Pontoscolex corethrurus* was found in all sites and was the dominant species in all systems as well.
- As in other tropical agroecosystems, most of the earthworm species are endogeic.
- Sun coffee (monoculture) and shaded coffee (mixed culture) had similar earthworm density.
- Secondary forests had lower earthworm density and biomass than coffee plantations.

- Relationship between earthworm density and biomass and the soil properties did not show a clear trend for all variables. This does not mean that no relationship exits, just that it was not detected. The disturbance of the study areas could produce such a complex pattern that no variable stands out to explain this distribution.

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Appendix A

Results of Statistical Analyses

Análisis de la varianza (ABUNDANCIA)

Variable	N	R^2	R^2 Aj	CV
Abun. (ind/m2)	28	0.43	0.20	58.27

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	80052.21	8	10006.53	1.83	0.1337
Lugar	9231.46	2	4615.73	0.84	0.4458
Trt	52064.40	2	26032.20	4.76	0.0211
Lugar*Trt	23078.84	4	5769.71	1.05	0.4060
Error	103990.75	19	5473.20		
Total	184042.96	27			

Test:Tukey Alfa=0.05 DMS=87.06233

Error: 5473.1974 gl: 19

Lugar	Medias	n	
Las Marías	101.33	9	A
Jayuya	138.67	9	A
Lares	141.58	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=87.06233

Error: 5473.1974 gl: 19

Trt	Medias	n		
Bosque	66.92	10	A	
Sol	154.67	9		B
Sombra	160.00	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=206.98407

Error: 5473.1974 gl: 19

Lugar	Trt	Medias	n	
Las Marías	Bosque	0.00	3	A
Jayuya	Bosque	80.00	3	A
Lares	Bosque	120.75	4	A
Las Marías	Sol	122.67	3	A
Lares	Sombra	133.33	3	A
Jayuya	Sombra	165.33	3	A
Lares	Sol	170.67	3	A

Jayuya	Sol	170.67	3	A
Las Marías	Sombra	181.33	3	A

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (BIOMASA)

Variable	N	R^2	R^2 Aj	CV
Biomasa (g/m2)	28	0.78	0.69	108.87

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	9353516.35	8	1169189.54	8.49	0.0001
Lugar	4520915.87	2	2260457.94	16.41	0.0001
Trt	1629951.76	2	814975.88	5.92	0.0101
Lugar*Trt	3376514.36	4	844128.59	6.13	0.0024
Error	2617572.65	19	137766.98		
Total	11971089.00	27			

Test:Tukey Alfa=0.05 DMS=436.79965

Error: 137766.9814 gl: 19

Lugar	Medias	n		
Jayuya	55.17	9	A	
Lares	69.88	10	A	
Las Marías	924.47	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=436.79965

Error: 137766.9814 gl: 19

Trt	Medias	n		
Bosque	41.91	10	A	
Sol	380.30	9	A	B
Sombra	627.32	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=1038.45803

Error: 137766.9814 gl: 19

Lugar	Trt	Medias	n		
Las Marías	Bosque	0.00	3	A	
Jayuya	Bosque	25.65	3	A	
Lares	Sombra	34.37	3	A	
Jayuya	Sombra	57.28	3	A	
Lares	Sol	75.18	3	A	
Jayuya	Sol	82.59	3	A	
Lares	Bosque	100.09	4	A	
Las Marías	Sol	983.13	3	A	B
Las Marías	Sombra	1790.29	3		B

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (% ARENA)

Variable	N	R^2	R^2 Aj	CV
% Arena	28	0.55	0.35	36.58

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	1561.58	8	195.20	2.85	0.0289
Lugar	509.06	2	254.53	3.72	0.0432
Trt	629.44	2	314.72	4.60	0.0234
Lugar*Trt	482.30	4	120.57	1.76	0.1779
Error	1299.09	19	68.37		
Total	2860.67	27			

Test:Tukey Alfa=0.05 DMS=9.73090

Error: 68.3732 gl: 19

Lugar	Medias	n	
Las Marías	19.35	9	A
Lares	19.83	10	A
Jayuya	28.73	9	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=9.73090

Error: 68.3732 gl: 19

Trt	Medias	n		
Sol	19.16	9	A	
Sombra	19.48	9	A	
Bosque	29.27	10		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=23.13447

Error: 68.3732 gl: 19

Lugar	Trt	Medias	n		
Lares	Sol	14.48	3	A	
Las Marías	Sol	15.88	3	A	
Las Marías	Sombra	16.83	3	A	
Jayuya	Sombra	18.35	3	A	B
Lares	Bosque	21.74	4	A	B
Lares	Sombra	23.26	3	A	B
Las Marías	Bosque	25.34	3	A	B
Jayuya	Sol	27.13	3	A	B
Jayuya	Bosque	40.72	3		B

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (% LIMO)

Variable	N	R^2	R^2 Aj	CV
% Limo	28	0.62	0.46	16.01

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	2600.49	8	325.06	3.84	0.0076
Lugar	1159.09	2	579.55	6.84	0.0058
Trt	1071.67	2	535.83	6.33	0.0078
Lugar*Trt	288.82	4	72.20	0.85	0.5096
Error	1608.91	19	84.68		
Total	4209.39	27			

Test:Tukey Alfa=0.05 DMS=10.82925

Error: 84.6793 gl: 19

Lugar	Medias	n		
Lares	51.86	10	A	
Jayuya	54.95	9	A	
Las Marías	66.87	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=10.82925

Error: 84.6793 gl: 19

Trt	Medias	n		
Bosque	52.95	10	A	
Sol	54.03	9	A	
Sombra	66.70	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=25.74571

Error: 84.6793 gl: 19

Lugar	Trt	Medias	n		
Jayuya	Sol	44.96	3	A	
Lares	Bosque	46.31	4	A	
Jayuya	Bosque	51.31	3	A	B
Lares	Sol	51.80	3	A	B
Lares	Sombra	57.49	3	A	B
Las Marías	Bosque	61.23	3	A	B
Las Marías	Sol	65.34	3	A	B
Jayuya	Sombra	68.58	3	A	B
Las Marías	Sombra	74.03	3		B

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (% ARCILLA)

Variable	N	R^2	R^2 Aj	CV
% Arcilla	28	0.59	0.42	44.10

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	1970.33	8	246.29	3.46	0.0126
Lugar	861.41	2	430.70	6.05	0.0093
Trt	878.99	2	439.49	6.17	0.0086
Lugar*Trt	233.62	4	58.41	0.82	0.5283
Error	1353.07	19	71.21		
Total	3323.39	27			

Test:Tukey Alfa=0.05 DMS=9.93099

Error: 71.2141 gl: 19

Lugar	Medias	n		
Las Marías	13.78	9	A	
Jayuya	16.32	9	A	
Lares	26.50	10		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=9.93099

Error: 71.2141 gl: 19

Trt	Medias	n		
Sombra	13.82	9	A	
Bosque	15.97	10	A	
Sol	26.81	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=23.61018

Error: 71.2141 gl: 19

Lugar	Trt	Medias	n		
Jayuya	Bosque	7.96	3	A	
Las Marías	Sombra	9.14	3	A	
Jayuya	Sombra	13.07	3	A	B
Las Marías	Bosque	13.43	3	A	B
Las Marías	Sol	18.78	3	A	B
Lares	Sombra	19.25	3	A	B
Lares	Bosque	26.51	4	A	B
Jayuya	Sol	27.91	3	A	B
Lares	Sol	33.73	3		B

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (MOISTURE %)

Variable	N	R^2	R^2 Aj	CV
moisture (%)	28	0.73	0.61	13.99

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	2537.76	8	317.22	6.30	0.0005
Lugar	1059.03	2	529.52	10.51	0.0008
Trt	608.63	2	304.31	6.04	0.0093
Lugar*Trt	918.79	4	229.70	4.56	0.0095
Error	957.07	19	50.37		
Total	3494.83	27			

Test:Tukey Alfa=0.05 DMS=8.35228

Error: 50.3722 gl: 19

Lugar	Medias	n		
Lares	44.92	10	A	
Las Marías	48.54	9	A	
Jayuya	59.45	9		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=8.35228

Error: 50.3722 gl: 19

Trt	Medias	n		
Sol	45.46	9	A	
Sombra	50.65	9	A	B
Bosque	56.81	10		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=19.85692

Error: 50.3722 gl: 19

Lugar	Trt	Medias	n		
Jayuya	Sol	43.71	3	A	
Lares	Sol	43.92	3	A	
Lares	Bosque	44.41	4	A	
Las Marías	Sombra	44.73	3	A	
Lares	Sombra	46.44	3	A	
Las Marías	Sol	48.75	3	A	
Las Marías	Bosque	52.14	3	A	
Jayuya	Sombra	60.77	3	A	B
Jayuya	Bosque	73.88	3		B

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (OM %)

Variable	N	R^2	R^2 Aj	CV
OM (%)	28	0.22	0.00	115.30

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	463.18	8	57.90	0.68	0.7062
Lugar	34.43	2	17.21	0.20	0.8195
Trt	120.10	2	60.05	0.70	0.5080
Lugar*Trt	246.14	4	61.54	0.72	0.5892
Error	1625.40	19	85.55		
Total	2088.58	27			

Test:Tukey Alfa=0.05 DMS=10.88461

Error: 85.5473 gl: 19

Lugar	Medias	n	
Las Marías	6.63	9	A
Jayuya	7.17	9	A
Lares	9.18	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=10.88461

Error: 85.5473 gl: 19

Trt	Medias	n	
Sombra	6.01	9	A
Sol	6.42	9	A
Bosque	10.55	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=25.87733

Error: 85.5473 gl: 19

Lugar	Trt	Medias	n	
Lares	Sombra	4.43	3	A
Lares	Sol	5.37	3	A
Las Marías	Bosque	6.43	3	A
Las Marías	Sol	6.73	3	A
Las Marías	Sombra	6.73	3	A
Jayuya	Sombra	6.87	3	A
Jayuya	Sol	7.17	3	A
Jayuya	Bosque	7.47	3	A
Lares	Bosque	17.75	4	A

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (pH)

Variable	N	R^2	R^2 Aj	CV
pH	28	0.32	0.04	26.44

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	13.33	8	1.67	1.13	0.3882
Lugar	1.37	2	0.69	0.47	0.6350
Trt	9.44	2	4.72	3.20	0.0634
Lugar*Trt	1.67	4	0.42	0.28	0.8852
Error	28.03	19	1.48		
Total	41.36	27			

Test:Tukey Alfa=0.05 DMS=1.42931

Error: 1.4751 gl: 19

Lugar	Medias	n	
Jayuya	4.23	9	A
Lares	4.68	10	A
Las Marías	4.73	9	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=1.42931

Error: 1.4751 gl: 19

Trt	Medias	n	
Sombra	4.08	9	A
Sol	4.21	9	A
Bosque	5.36	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=3.39807

Error: 1.4751 gl: 19

Lugar	Trt	Medias	n	
Lares	Sombra	4.00	3	A
Jayuya	Sombra	4.00	3	A
Jayuya	Sol	4.13	3	A
Las Marías	Sombra	4.23	3	A
Las Marías	Sol	4.23	3	A
Lares	Sol	4.27	3	A
Jayuya	Bosque	4.57	3	A
Las Marías	Bosque	5.73	3	A
Lares	Bosque	5.78	4	A

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (Total C)

Variable	N	R^2	R^2 Aj	CV
Total C (%)	28	0.71	0.58	22.35

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	41.93	8	5.24	5.72	0.0009
Lugar	3.81	2	1.90	2.08	0.1525
Trt	29.99	2	14.99	16.37	0.0001
Lugar*Trt	7.85	4	1.96	2.14	0.1150
Error	17.40	19	0.92		
Total	59.33	27			

Test:Tukey Alfa=0.05 DMS=1.12631

Error: 0.9160 gl: 19

Lugar	Medias	n	
Las Marías	3.85	9	A
Lares	4.11	10	A
Jayuya	4.74	9	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=1.12631

Error: 0.9160 gl: 19

Trt	Medias	n		
Sol	3.02	9	A	
Sombra	4.14	9	A	
Bosque	5.54	10		B

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=2.67772

Error: 0.9160 gl: 19

Lugar	Trt	Medias	n			
Lares	Sol	2.60	3	A		
Las Marías	Sol	3.13	3	A	B	
Jayuya	Sol	3.34	3	A	B	
Jayuya	Sombra	4.01	3	A	B	
Lares	Sombra	4.18	3	A	B	
Las Marías	Bosque	4.20	3	A	B	C
Las Marías	Sombra	4.22	3	A	B	C
Lares	Bosque	5.55	4		B	C
Jayuya	Bosque	6.87	3			C

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (Total N)

Variable	N	R^2	R^2 Aj	CV
Total N (%)	28	0.25	0.00	167.31

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	6.73	8	0.84	0.80	0.6085
Lugar	1.01	2	0.50	0.48	0.6262
Trt	2.29	2	1.15	1.09	0.3555
Lugar*Trt	2.45	4	0.61	0.58	0.6785
Error	19.93	19	1.05		
Total	26.66	27			

Test:Tukey Alfa=0.05 DMS=1.20531

Error: 1.0490 gl: 19

Lugar	Medias	n	
Las Marías	0.39	9	A
Jayuya	0.48	9	A
Lares	0.83	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=1.20531

Error: 1.0490 gl: 19

Trt	Medias	n	
Sol	0.31	9	A
Sombra	0.43	9	A
Bosque	0.96	10	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=2.86552

Error: 1.0490 gl: 19

Lugar	Trt	Medias	n	
Lares	Sol	0.28	3	A
Las Marías	Sol	0.32	3	A
Jayuya	Sol	0.34	3	A
Lares	Sombra	0.42	3	A
Las Marías	Bosque	0.43	3	A
Jayuya	Sombra	0.43	3	A
Las Marías	Sombra	0.44	3	A
Jayuya	Bosque	0.68	3	A
Lares	Bosque	1.79	4	A

Letras distintas indican diferencias significativas(p<= 0.05)

Análisis de la varianza (C/N)

Variable	N	R^2	R^2 Aj	CV
C/N	28	0.17	0.00	20.81

Cuadro de Análisis de la Varianza (SC tipo III)

F.V.	SC	gl	CM	F	p-valor
Modelo	14.96	8	1.87	0.48	0.8520
Lugar	3.75	2	1.87	0.49	0.6225
Trt	0.87	2	0.43	0.11	0.8940
Lugar*Trt	8.50	4	2.12	0.55	0.7009
Error	73.29	19	3.86		
Total	88.25	27			

Test:Tukey Alfa=0.05 DMS=2.31135

Error: 3.8576 gl: 19

Lugar	Medias	n	
Lares	8.99	10	A
Jayuya	9.74	9	A
Las Marías	9.77	9	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=2.31135

Error: 3.8576 gl: 19

Trt	Medias	n	
Bosque	9.25	10	A
Sombra	9.60	9	A
Sol	9.64	9	A

Letras distintas indican diferencias significativas(p<= 0.05)

Test:Tukey Alfa=0.05 DMS=5.49506

Error: 3.8576 gl: 19

Lugar	Trt	Medias	n	
Lares	Bosque	7.76	4	A
Jayuya	Sombra	9.27	3	A
Lares	Sol	9.30	3	A
Las Marías	Sombra	9.63	3	A
Las Marías	Sol	9.77	3	A
Jayuya	Sol	9.87	3	A
Las Marías	Bosque	9.90	3	A
Lares	Sombra	9.90	3	A
Jayuya	Bosque	10.10	3	A

Letras distintas indican diferencias significativas(p<= 0.05)

Appendix B

Earthworm species, density and biomass

Earthworm species, density and biomass in Las Marías

Species	Sun			Shaded			Forest		
	1	2	3	1	2	3	1	2	3
Pontoscolex corethrurus	4	5	12	16	11	6	0	0	0
Amynthas rodericensis	0	0	2	0	0	0	0	0	0
Pontoscolex spiralis	0	0	0	0	1	0	0	0	0
Onychochaeta borincana	0	0	0	0	0	0	0	0	0
Pheretimoid sp.	0	0	0	0	0	0	0	0	0
Pontoscolex melissae	0	0	0	0	0	0	0	0	0
Pontoscolex sp.	0	0	0	0	0	0	0	0	0
Amynthas gracilis	0	0	0	0	0	0	0	0	0
Density (ind/m^2)	64	80	224	256	192	96	0	0	0
Biomass (g/m^2)	24.4	26.9	132.9	149.6	103.6	82.4	0	0	0

Earthworm species, density and biomass in Jayuya

Species	Sun			Shaded			Forest		
	1	2	3	1	2	3	1	2	3
Pontoscolex corethrurus	13	10	8	9	8	2	7	0	0
Amynthas rodericensis	0	0	0	0	0	0	0	0	0
Pontoscolex spiralis	0	0	0	0	9	3	0	0	0
Onychochaeta borincana	0	0	0	0	P	0	0	0	0
Pheretimoid sp.	0	0	1	0	0	0	0	0	0
Pontoscolex melissae	0	0	0	0	0	0	0	3	0
Pontoscolex sp.	0	0	0	0	0	0	0	0	5
Amynthas gracilis	0	0	0	0	0	0	0	0	0
Density (ind/m^2)	208	160	144	144	272	80	112	48	80
Biomass (g/m^2)	92.6	76.9	78.2	43.1	103.5	25.3	22.8	8.8	45.3

P: present

Earthwormspecies, density and biomass in Lares

Species	Sun			Shaded			Forest		
	1	**2**	**3**	**1**	**2**	**3**	**1**	**2**	**3**
Pontoscolex corethrurus	12	11	8	10	10	4	10	3	17
Amynthas rodericensis	0	0	0	1	0	0	0	0	0
Pontoscolex spiralis	0	0	0	0	0	0	0	0	0
Onychochaeta borincana	0	0	0	0	0	0	0	0	0
Pheretimoid sp.	0	0	0	0	0	0	0	0	0
Pontoscolex melissae	0	0	0	0	0	0	0	0	0
Pontoscolex sp.	0	0	0	0	0	0	0	0	0
Amynthas gracilis	0	0	1	0	0	0	0	0	0
Density (ind/m^2)	192	176	144	176	160	64	160	48	272
Biomass (g/m^2)	80.2	61.8	83.5	59.9	69.7	17.4	35.5	29.5	63.3