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OPILIONES BIODIVERSITY IN CUSUCO NATIONAL PARK, HONDURAS

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

By

BRITTANY NICOLE DAMRON
B.S., Wright State University, 2011

2014
Wright State University

WRIGHT STATE UNIVERSITY

GRADUATE SCHOOL

September 17, 2012

I HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER
MY SUPERVISION BY <u>Brittany Nicole Damron</u> ENTITLED <u>Opiliones Biodiversity in
Cusuco National Park, Honduras.</u> BE ACCEPTED IN PARTIAL FULFILLMENT OF
THE REQUIREMENTS FOR THE DEGREE OF <u>Master of Science</u>.

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ABSTRACT

Damron, Brittany Nicole. M.S. Department of Biological Sciences, Wright State University, 2014. Opiliones Biodiversity in Cusuco National Park, Honduras.

Biodiversity in the tropics is especially diverse and describing species found in the tropics is important. Opiliones species richness is higher in the tropics than the temperate zone, with diversity highest in low latitudes. This study aims to catalog the Opiliones community in Cusuco National Park, Honduras, while estimating species richness and describing their abundance, distribution and community structure. Fifty hours of sampling at 6 sites yielded 18 morphospecies and 264 individual Opiliones were collected. This study added one new described species to the Opiliones fauna of Honduras, and fourteen morphospecies may represent new species. An estimated 43 to 112 species could exist in Cusuco Park. The contribution of beta diversity to Cusuco's overall diversity is significant. There was no significant pattern in the distribution of Opiliones along elevation or moisture gradients. Increased spatial and temporal sampling is needed to better assess the diversity and richness of Opiliones within Cusuco National Park.

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I. INTRODUCTION

Biodiversity studies examine the variety of life that exists on Earth, and why species exist where they do in space and time. About 1.4 million organisms have been identified and named, most of them angiosperms and insects. Estimates for the actual number of species is much higher than this number, with some estimates as high as 100 million species on Earth (Ehrlich & Wilson, 1991). The majority of species are found in the tropics, with less found towards the poles (Gaston, 2000). Many possible mechanisms could explain why species richness decreases with increasing latitude, such as less energy (low productivity) available in higher latitudes, variable climate with changing seasons, and the geologically recent origin of temperate regions (Gaston, 2000). Understanding why species richness decreases with latitude will allow scientists to more fully describe the taxa on Earth. The taxa being studied can be very important when describing said taxa (Gaston, 2000, Ehrlich & Wilson, 1991), since some taxa are more diverse than others, and others still are poorly studied. Relatively little effort has been given to describing bacteria, fungi, and many invertebrate groups (Nematodes, Acari, etc.), leading to low species richness estimates for groups that may contain hundreds of thousands of species (Ehrlich & Wilson, 1991). Fully describing and understanding biodiversity patterns on Earth has many benefits. It can provide valuable knowledge about natural ecosystems and services they provide, such as aesthetic pleasures,

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entertainment, and maintaining the gas composition in the atmosphere (Ehrlich & Wilson, 1991).

Species diversity can vary within and among habitats. Robert H. Whittaker (1960) defined beta diversity (β-diversity) as the "extent of change in community composition" among samples, or sites. With this simple definition it is easy for ecologists to represent the change or dissimilarity in species composition within a similar region, or along a specific environmental gradient. In the tropics, environments are relatively stable. Many species are unable to tolerate changes in environmental conditions, so small environmental changes can act as significant barriers to dispersal or colonization (Janzen, 1967; Almeida-Neto, et al. 2006). This can lead to species in the tropics having restricted ranges and high rates of endemism. As a result, tropical communities exhibit high β-diversity and high regional diversity (Jankowski, et al. 2009).

Traditionally, a multiplicative approach has been used to calculate β-diversity in ecological studies. The multiplicative approach to β-diversity is typically defined as the total species diversity of a region, gamma (ϒ) diversity, divided by the mean species diversity of the sites within that region, or alpha (α) diversity (Whittaker, 1960). Recently, studies using additive diversity partitioning have become more prevalent, because this approach permits comparisons of diversity within and among samples in a given region (Veech, et al. 2002). Additive metrics beta diversity are not a new approach; MacArthur first introduced this idea when he defined the diversity between two bird communities as equal to the combined diversity of the two sites, minus the average within site diversity (MacArthur, et al. 1966), or in terms of Whittaker's beta

diversity, $\beta=\Upsilon-\alpha$. With this definition, beta diversity can be described regardless of spatial, temporal, or environmental gradients (Veech, et al. 2002). When using an additive diversity approach, alpha and beta diversity become commensurate, and are able to be compared across multiple spatial extents, and scales (Veech, et al. 2002). Diversity changes with spatial scale, and sometimes the diversity patterns observed at a small spatial scale are not the same as the diversity pattern observed at a larger spatial scale (Crist, et al. 2003). When diversity is additively partitioned, α-diversity, and β-diversity are measured in the same units. They therefore can be compared across multiple spatial extents in a single study (Crist, et al. 2003). The ability to compare across different spatial extents facilitates nested designs that accurately depict diversity patterns at multiple spatial scales. This allows researchers to look at the differences among replicates within a site, differences between sites within a location, and differences across locations within a region (Anderson, et al. 2010). One can then compare how the diversity changes as you increase sampling spatial extent, and how each nested scale may contribute to the overall diversity of a region. The use of a null model tells if observed patterns at each nested-level are significantly higher or lower than an expected value (Crist, et al. 2003).

In this study, I examined the distribution of Opiliones (Arachnida) morphospecies throughout Cusuco National Park, Honduras. There are approximately 6,000 described species of the order Opilione, and an estimated 10,000 undiscovered species (Pinto-da-Rocha, Machado, & Giribet, 2007). Opiliones are a nearly cosmopolitan taxon. No known extant species live in Antarctica (Pinto-da-Rocha, Machado, & Giribet, 2007).

Neotropical Opiliones have high diversity with 9 families being described in Central America; the most prevalent family is the Cosmetidae with 133 described species (Townsend, et al. 2010). Opiliones tend to prefer humid forests but can be found in many environmental conditions (Almeida-Neto, et al. 2006). Opiliones can be nocturnal or diurnal, though most in the tropics are nocturnal, leaving roosting sites at dusk to forage and look for mates in the leaf litter and on vegetation (Acosta, et al. 1995). Opiliones have been described as opportunistic predators and omnivores (Acosta, et al. 1995; Pinto-da-Rocha, Machado, & Giribet, 2007; Sabinao & Gnaspini, 1999; Townsend, et al. 2010).

In the Neotropics, diversity is high for many taxa (Jablonski, et al. 2006; Soininen, et al. 2007). Environmental gradients can play a significant role in tropical diversity, especially elevational gradients (Jankowski, et al. 2009). Maximum humidity can be found at higher elevations, which can lead to the formation of cloud forests (Grytnes and McCain, 2007). Cusuco National Park occurs in northwestern Honduras, in the Merendon Mountains, which is a Meso-American biodiversity hotspot. The elevation in Cusuco Park ranges from 400 meters above sea level (m a.s.l.), to over 2000m a.s.l. Cusuco is dominated by cloud forest, with major forests types ranging from moist, broad-leaved forest to arid pine stands (Cusuco Park report, 2011).   A large biodiversity inventory is going on in this park, as part of an effort to catalog species located in the region. Since a large elevational gradient exists within Cusuco Park, diversity should be high due to high habitat heterogeneity.

The objectives of this study were to catalog the Opiliones diversity within Cusuco Park and estimate the species richness of Opiliones for the entire park. To estimate the

number of species found in the park, a species-area relationship (SAR) curve was created. I also examined at which of the studies four hierarchal, or nested, levels β-diversity would be highest, and what locations supported the highest Opiliones diversity. Using the software program PARTITION, I was able to compare observed β-diversity of Opiliones to the β-diversity expected if there was no hierarchal diversity structure in my data. I hypothesized that the β-diversity at a larger spatial extent would contribute more to overall regional diversity. This hypothesis is based on the idea that as the spatial extent of sampling increased habitat heterogeneity would also increase. I also hypothesized that Opiliones would prefer the warmer wetter locations at lower elevations. I predicted that since Opiliones prefer humid forests, locations at higher elevations that were drier and colder would support fewer individuals and morphospecies than lower elevation locations.

II. BACKGROUND

Elevation and Biodiversity

Biodiversity can change along multiple environmental gradients in the environment, such as elevation.   In the past it was thought that biodiversity decreased with increasing altitude. Whittaker's research (1952, 1960, 1965) showed this pattern in vegetation communities in various mountain ranges. However, other patterns of richness can be observed with changes in elevation. Three often-observed patterns are decreasing richness with increasing elevation, a normal distribution in which mid-elevations have the highest richness, and a decreasing plateau in which richness remains constant until mid-elevation, after which richness decreases. Research examining the effect of elevation on species richness is increasingly revealing a normal distribution pattern. This may be partially due to endemic mountaintop species and lower elevation species having ranges that overlap at mid-elevations. However, this is not always seen across the entire biota since some taxa can exhibit more than one pattern depending on the biogoegraphical realms that are sampled. McCain (2005) found unique responses of bat richness to elevation depending on the region. In tropical regions bat richness decreased as elevation increased, but in temperate regions the bat richness was highest at mid-elevations (McCain, 2005).

Beta Diversity: Variation and Turnover

It is important to differentiate between spatial turnover and variation. Variation, in reference to beta diversity, is the change within a community at a defined spatial or

temporal extent. It can also be within a defined habitat or other specifically defined

experimental treatment (Anderson, et al. 2011). Beta diversity (or variation) and species

turnover are typically used interchangeably when measuring diversity. Species turnover

is always in reference to a specific environmental structure (such as an elevational or

latitudinal gradient). The comparisons must be pairwise among the sites along the

specific environmental gradient being examined (Vellend, 2001). Beta diversity

calculations, such as Whittaker's beta diversity, do not inherently measure change along a

gradient, but changes in composition between chosen pairs of samples (Vellend, 2001).

Whittaker's beta diversity index is calculated using the equation $\beta=\Upsilon/\alpha$. Υ is the total

number of species recorded, while α is the average diversity of each sample. This

equation can only be used along a gradient if each pairwise comparison is done along the

specific gradient. Variation in beta diversity is more applicable than spatial turnover

when asking questions such as how similar is species composition from one sample site

to another within a region (Anderson, et al. 2010). Spatial turnover, on the other hand,

answers questions such as how changes in elevation or latitudinal gradients influence the

observed diversity of taxa. This study looks at how species composition varies among

hierarchal spatial extents within Cusuco National Park, not how the species composition

changes along an environmental gradient. Therefore it focuses on beta diversity in terms

of spatial variation.

Opiliones Biogeography

     Harvestmen (Arachnida: Opiliones), are distributed worldwide and therefore are prime candidates for biogeographical studies. The order Opiliones contains approximately 48 families, but the phylogenetic relationships and number of subclades remain unresolved. Opiliones are found on all continents and major continental islands, with the exception of Antarctica where all species have gone extinct. Few studies have looked at the worldwide distribution of species, or families within this order (Pinto-da-Rocha, Machado, and Giribet, 2007). The suborder Cyphophthalmi arose 174-312 million years ago in the Gondwana portion of Pangea (Boyer, et al. 2007). Arachnids were some of the first organisms on land. The oldest Opilione fossil, believed to be an individual in the suborder Eupnoi, is approximately 400 million years old and was found in Devonian Rhynie Chert in Scotland (Dunlop, et al., 2004). It is believed that the Opiliones originated during the Silurian period some 420-440 million years ago, and at least in the Eupnoi, many modern day lineages maintain early morphological characteristics (Dunlop et al. 2004).

     Opiliones are most diverse in the tropics, with diversity decreasing towards the poles. Guatemala has an area over 100,000 km² and contains 51 described species of Opiliones, and likely many more undescribed species (Townsend et al. 2010). The United Kingdom, with a well-described fauna, has an area of over twice Guatemala's (over 200,000 km²), and contains only about 25 species (Savory, 1945). In the tropics, the highest diversity at a single site in the Atlantic Brazilian forest was 64 species. In temperate regions, species richness is rarely > 12 species at a single site (Proud et al.

2012). Species diversity and richness decreases with an increasing altitude (Almeida-Neto, et al. 2006).

Honduras Opiliones are poorly described, with only 9 species recorded. Belize has 30 described species, while Guatemala has 51 described species (Townsend, et al. 2010). The best described Central American Opiliones fauna is in Costa Rica, with 121 species recorded (Townsend, et al. 2010). Describing the Opiliones fauna is especially difficult since very little is known about their natural history, making collection of specimens biased (Townsend, et al. 2010).

The species collected in this study are members of families that are especially diverse in the Neotropics. The family Sclerosomatidae (Opiliones) is distributed throughout Indo-Malaysia and the Neotropics. The genus *Metopilio* (Family; Sclerosomatidae) is only found in the Neotropics, and ranges from the western United States to Costa Rica. The genus *Prionostemma* (Family; Sclerosomatidae) is distributed throughout the regions occupied by the family Sclerosomatidae (Pinto-da-Rocha, Machado, and Giribet, 2007). The family Cosmetidae, which belongs to the suborder Laniatores, is endemic to the New World, and peak diversity for this group is found California, Central America, and Northern South America. The Cosmetidae are the most prevalent family in Central American with over 133 species found there (Townsend et al. 2010). Cosmetidae are also the only Laniatorids found in the Northern hemisphere.

Opiliones Ecology

Opiliones occupy a variety of habitats, from soil and leaf litter, vegetation, and caves. Most Opiliones species prefer mesic environments, although some species occur in very arid conditions (Pinto-da-Rocha, Machado, and Giribet, 2007). Opiliones are thought to be prone to desiccation, and prefer microclimates that have high humidity and warmer temperatures (Edgar, 1971). During the day, Opiliones will seek out refuges to reduce the risk of desiccation. This makes them hard to observe in the field. Some tropical Opiliones will inhabit leaf cutter ant nests, fallen logs, palm fronds, and sometimes epiphytic bromeliads and ferns (Proud et al. 2012). Some Opiliones aggregate in large groups during the day, which is believed to regulate temperature and humidity (Machado & Vasconcelos, 1998; Grether & Donaldson, 2007), although the main purpose of this strategy is most likely defense. Decreases in relative humidity can increase desiccation risk, potentially decreasing the number of individuals observed during dry periods. Opiliones are typically negatively impacted by changes in their habitat, and some studies have concluded that both species richness and abundance are reduced by habitat degradation (Bragagnolo et al. 2007; Proud et al. 2012). Habitat degradation also plays a role in what species are seen and how abundant they are, mainly due to the changes in microclimate. Other studies have shown no change to occur when the habitat is modified (Corey & Stout, 1990).

III. METHODS

Study Area and Site Description

      I completed this study in Cusuco National Park (Cusuco), located in Cortes, Honduras. The park is located in the Merendon Mountains and is part of the Meso-American biodiversity hotspot (Cusuco Report, 2011). Cusuco contains a range of habitats, with elevations of just above sea level to 2,425 m a.s.l. (Cusuco Report, 2011). Some of the dominant habitat types found within the park are moist broadleaf forest, semi-arid pine forest, and Dwarf forest (bosque enaño).

      Cusuco contains 23,400 ha of protected forest with a 7,690 ha core zone and 15,750 ha buffer zone (Cusuco Park Report, 2011). There is some controlled deforestation in the buffer zone for residents, and a strict "no logging" policy in the core zone. Deforestation occurs in both zones, due to illegal coffee and other agricultural plantations (personal observation). Illegal deforestation is most prevalent on the Northwest portion of Cusuco, where elevation is lowest and the park is more accessible.

      Seven permanent research sites have been established within Cusuco, with each site containing 3-4 permanent transects with two to eight 20 x 20 m sampling plots along these transects. There are over 100 sampling plots established throughout the park. I sampled 9 plots at six different research sites. The plots encompassed a range of habitat types, including broad leaf forest and bosque enaño (dwarf forest), as well as a variety of understory vegetation consisting of palms, tree ferns, and bamboo. Plots were selected according to degradation level, and use by other invertebrate researchers. The plot was considered degraded if any deforestation had occurred within the few months prior to

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sampling. Degradation could be considered none, mild, or severe. No degradation meant that no recent logging had occurred, mild degradation meant that only small understory trees were removed and large trees were left standing, and severe degradation meant that the plot was completely logged and no trees remained standing. I selected sites that were either mildly or not degraded, and the only other invertebrate sampling occurring in the site was dung-baited pitfall traps.

Table 1: Elevation, temperature, relative humidity, date sampled, and time sampled for all sites and plots sampled in Cusuco National Park, Honduras.

Sites	Plots	Elevation (meters a.s.l.)	Temperature (C)	Relative Humidity Range	Days Sampling Occurred	Times Sampling Occurred
Base Camp	BC4_SS2	1623	16-19	80-99%	23 June, 2012	21:20-24:19
					24 June, 2012	20:04-22:44
	BC2_SS3	1362	18-21	79-88%	02 Aug., 2012	19:55-22:15
					04 Aug., 2012	19:45-21:35
Cantiles	CA5_SS2	1874	14-19	79-96%	28 June, 2012	20:10-21:39
					29 June, 2012	20:54-22:41
	CA3_SS1	2023	15-17	82-94%	30 June, 2012	18:55-21:26
					02 July, 2012	18:31-20:15
Guanales	GU1_SS2	1435	17-20	84-94%	05 July, 2012	18:22-19:48
					06 July, 2012	18:30-20:55
Cortecito	CO2_SS1	1407	17-20	90-97%	12 July, 2012	20:18-22:40
					13 July, 2012	20:22-23:11
	CO1_SS5	1216	19-20	89-93%	14 July, 2012	20:54-22:55
					15 July, 2012	20:10-23:00
Danto	DA0_SS2	1585	17-18	94-96%	20 July, 2012	20:15-22:45
					22 July, 2012	21:10-23:00
Santo Tomas	ST1B_SS4	669	22-25	82-97%	27 July, 2012	20:32-23:11
					28 July, 2012	19:57-22:54

Base camp plots can be characterized as broadleaf forest. The first plot I sampled at Base Camp (BC4_SS2) had a tree fern and palm dominated understory, as well as a sedge-like plant, and is located 1623 m a.s.l. In BC4_SS2 a large boulder was located in

the Southwest corner of the plot, while there was a large drop off in the Northeast side.

I recorded temperature and realtive humidity using an Amprobe psychrometer model

THWD-2 (Miramar, Florida). This information with dates and times is provided in

Table 1. The second plot sampled at Base Camp (BC2_SS3) was about 100 meters from

a river, and the understory was dominated by palm and tree ferns, with a thick leaf litter

layer, and had an altitude of 1362 m a.s.l. The plot has a 40˚ slope to the south, but the

site was easy to navigate using game trails.

Cantilles plots were typically broadleaf forest, or bosque enaño (dwarf forest).

The first Cantilles plot I sampled (CA5_SS2) was 1874 m a.s.l. It had an understory on

the western side dominated by ferns and bamboo, while the eastern side had been

deforested. The deforestation was recent, within the previous year or two. The eastern

side of the plot ended at a drop off, with non-degraded forest below. The second

Cantilles plot (CA3_SS1) was located at a higher elevation (2023 m a.s.l.) and had many

palm trees that were easily uprooted. The understory consisted of tree ferns and small

pines, and the soil was soft and easily disturbed. The plot had a steep slope from the

northeast corner to the southwest corner.

Only one plot was sampled at the Guanales site, due to inclement weather. The

plot (GU1_SS2) is located at 1435 m a.s.l., and is considered a broad leafed forest type.

It has an open understory, except in the southeast corner where bamboo was abundant. A

few palms were throughout the plot. Little leaf litter on the forest floor, and almost no

epigeic invertebrates were observed, except for an occasional Lycosidae and Sparassidae

spider.

The Cortecito site plots were located in broad leaf forest type. The first plot

sampled in the Cortecito site (CO2_SS1) is located 1408 m a.s.l. It had to be moved 10

m to the north of the original transect to avoid recent deforestation for a coffee plantation.

The plot contained palms of all sizes. During the second night of sampling there was an

ant colony movement, which was the most activity observed. The second plot

(CO1_SS5) is located 1216 m a.s.l., and is about 2 km from camp. CO1_SS5 contains

short spiky ferns and some tall palm-like trees. A steep drop off was located at the edge

of the plot. The plot is located about 0.5 km from two coffee plantations, and appears

undegraded.

Only one plot was sampled in the El Danto site due to consistent heavy rainfall

and researcher illness. The plot at El Danto (DA0_SS2) is located 1586 m a.s.l. It

contains broadleaf forest and a dense bamboo understory. Bamboo was so dense that

sampling at some points became extremely difficult, and navigation almost impossible.

The canopy is low in this site, with canopy trees < 10 m high. El Danto was especially

wet. It rained everyday, and most of the time the rain was torrential between 1600 and

1800.

The final site sampled was Santo Tomas. Only one plot was sampled. This plot is

the lowest elevation at 669 m a.s.l. The forest was broadleaf, and the understory

contained a palm-like plant and spiked palms. Boulders are in the middle of the plot,

creating a 5 m cliff. There is some water running under the plot and boulders, with a few

small sink holes. A river is located approximately 200 m from the plot. A drop off is

located at the northern edge of the plot, and a large slope comes into the plot from the south.

Sampling Procedures

I divided each of the 20 x 20 m sampling plots into sixteen 5 x 5 m subplots for data collection. Once I located the first corner tree, the site was oriented so that each side faced a cardinal direction. I then ran string across the plot from north to south and east to west to establish the 5 x 5m subplots. I completed this during the day (1300-1600) due to dangers associated with traversing an unfamiliar site in the dark. Unique site characteristics were recorded in a field notebook at this time, such as locations of boulders, drop offs, and vegetation. I recorded any notable weather in my field notebook during the day as well.

I assigned letters to each of the subplots, starting with the letter A in the northwest corner, and ending with P in the southeast corner. I then divided each of the subplots into 25 quadrats, and in the same manner that the subplots were assigned letters, each quadrat is assigned a number (1-25). I used a random number generator to select the quadrat to be sampled using the enclosed quadrat sampling method.

I started sampling at approximately dusk, and continued until all sampling was completed. Before I started sampling I recorded temperature, relative humidity, and that day's weather events. I also noted if there was anything of interest going on in the site at the time of sampling. I only sampled half of the plot each night, or eight quadrats one per

subplot, and then the next night the other half of the plot would be sampled. To avoid

sampling bias I flipped a coin to determine what side of the plot was sampled.

I first used enclosed quadrat sampling. I placed the quadrat at a randomly

selected location, and sampled for five minutes using an LED light. If a large obstacle

was in the way, such as a tree or boulder, I moved the quadrat to the one directly north of

the original quadrat. If that placed the quadrat out of the subplot or plot, then the quadrat

to the south was sampled. I searched vegetation up to my head height (1.5 m), and turned

over leaf litter to search it as well. I collected all the individuals I saw in a 5 minute

period, and placed them into a vial of 70% ethanol. I also placed a label in the vial with

the date, transect, plot, and quadrat sampled written in pencil. I sampled all quadrats in

the same manner, completing eight quadrats in one night. Each quadrat had its own vial

for specimens, which I combined while I identified morphospecies and recorded them in

my field notebook. I sketched and described any new morphospecies in a separate

notebook.

Next, manual ground and vegetation searches were used to sample. I walked the

perimeter of the plot for forty minutes (ten minutes per side) and collected all

Harvestmen seen on the ground or vegetation up to head height (1.5 m). I kept these

individuals in a separate vial containing 70% ethanol with a label like that placed in the

vials for quadrat sampling.

I completed leaf litter sampling at many of the plots (but not all), and individuals

from this sampling method have not been used in analysis. I sampled eight randomly

chosen sampled quadrats in each plot as well as eight corners of the site (four along the

outer edges and four along the site's midline running west to east). I sifted each quadrat through a leaf litter sorter for 5 minutes. I placed all the sampled quadrats leaf litter in one bag together while the eight unsampled quadrats were placed in another bag. I placed each of these bags in their own Winkler bags and allowed the samples to sit for two days, turning the leaf litter over after 24 hours.

The final sampling technique combined beat sheets and sweep netting. With each plot I would alternate between the sweep net or beat sheet as the first method. Starting in the southwest corner, I would sample along five transects parallel to the edges of the plot. Sweep net involved swinging a net from side to side hitting any vegetation at waist height and below. Beat sheet sampled all vegetation from about head height to waist height. I used a stick to beat or shake the leaves and branches on vegetation, while a small sheet was held beneath the vegetation. I sampled the first half of the transect with one method, and the second half with the other method. The following evening, I sampled five additional transects.

Species Sorting and Identification

The following morning after sampling, I would sort the specimens collected the night before according to morphospecies. I used a morphospecies guide created by Megan Lock and Dr. Stuart Longhorn the year prior and would add new morphospecies as needed in my notebook. I counted all the individuals for each morphospecies type, and then record the information in my field notebook. I started with individuals obtained from quadrat sampling. Using a sorting tray I separated them in to morphospecies. I

recorded any new morphospecies in a separate notebook (which included a written description as well as a sketch). Once I recorded those data, I placed the individuals into a container with a label and a piece of fabric on top of the specimen. I placed the specimens from different samples in new containers with one another to reduce space used.

I was unable to identify most of the specimens to species while I was in Cusuco Park. From February 27 to March 3, I worked with Dr. Victor Townsend (Virginia Weselyan College) to identify specimens to genus, and further to species when possible. We also prepared specimen genitalia for SEM and took pictures of individuals where preparation did not destroy the genitalia (see Appendix).

Data Analysis

The majority of the analyses of hierarchal diversity in Cusuco National Park was done with the software program PARTITION (Veech & Crist, 2009). In my study I used additive partitioning as a tool to partition Opiliones diversity into commensurate alpha and beta components. I partitioned Harvestmen diversity into three hierarchal, or nested, levels of spatial extent: 18 subplots, nine plots, and six sites. The total diversity is partitioned into the average within hierarchal level (α_i), and average among hierarchal levels (β_i), where *i* is the hierarchal scale. Across multiple spatial scales the calculation for total species richness is calculated by $\Upsilon = \alpha_1 + \alpha_2 + \alpha_3 + \beta_1$. Shannon diversity was also calculated for each spatial extent by PARTITION. This is done by partitioning the average diversity within a sample and the average number of species absent from that

sample, but does occurr in another sample. This is represented by $e^{H'}$, where $H' = -\sum p_i \ln p_i$, p_i is the proportion of individuals found in the ith species. I tested if the observed partitioned diversity was what would be found if all individuals of all species were randomly distributed throughout the subplots. This was done with the software PARTITION, using 10,000 randomizations.

I created a species area curve to estimate species richness for Cusuco National Park. I generated this curve by plotting the average species richness of an area. I then fit a linear relationship to this data to obtain an equation with a slope (z) and y-intercept (c). Very little area was sampled, and only a small number of species were collected. Therefore, a range of slopes (z) common to mainland diversity studies was evaluated to estimate the species richness.

I also compared pairwise site similarity in species composition. I calculated Jaccard similarity indices for each pair of sites by hand using the equation $C_j = a/a+b+c$; where "a" is the number of species shared by the compared sites, "b" is the species only found in one site and "c" is the number of species only found in the second site. Once I obtained these values, I tested for a correlation in community similarity among sites and differences in site elevation, using a Mantel test (vegan package in R)

I created scatter plots for morphospecies richness and abundance as related to elevation to determine if a pattern could be observed. The R^2 and P-value are reported for each.

RESULTS

I spent approximately 50 hours collecting a total of 264 individual Opiliones, representing 18 morphospecies belonging to two families: Sclerosomatidae and Cosmetidae. In the family Sclerosomatidae, I collected two genera, *Prionostemma* and *Metopilio*. In the family Cosmetidae, I collected four genera, *Cynorta*, *Eucynorta*, *Eucynortella* and *Paecileaema*. Of the seven known species of Honduras, I collected three in this study: *Cynorta bromeliacia*, *Paecileaema pectiginerum*, and *Cynorta dentipes*. I added four new genera to the Opiliones fauna of Honduras and one new species, *Paecileaema tolendense*.

The most abundant morphospecies collected was *Eucynorta* sp. "solid," with 85 individuals collected. *Eucynorta* sp. "solid" was the most widely distributed of the morphospecies, as it was collected in four of the six sites (Base camp, Cantilles, Cortecito, and El Danto). One morphospecies (Unknown Species 6) was represented by a single individual (Table 2).

The number of individuals, morphospecies, and unique morphospecies, found at each site is given in Table 3. Base camp's unique morphospecies was *Cynorta bromeliacea*. Cantille's unique morphospecies was *Prionostemma* sp "five". Of the morphospecies collected at Santo Tomas, three were *Cynorta dentipes*, *Eucynortella sp. legs*, and *Eucynorta sp. name*. Cortecito's unique morphospecies was Unknown species 6. There was only moderate overlap in species among sites. No pair of sites had a

21

Jaccard value of > 0.500 (Table 4). There was no significant correlation between Jaccard similarity and elevational difference (Mantel test, p = 0.17).

Beta Diversity

Alpha diversity (sampling effort within subplots, plots, and sites) was significantly less than what would be expected if individuals were randomly distributed (Fig. 1). Observed Shannon diversity for α_1 was 2.98, compared to an expected mean value of 5.02 (p < 0.0001). Observed Shannon diversity within plots (α_2) was 3.61, while the expected was value was 6.91 (p < 0.0001). Within-site Shannon diversity (α_3) was 3.98, while expected within site diversity was 7.51 (p < 0.0001). The observed Shannon diversity among sites (β_1) was almost twice that of the expected value of 1.34, with a value of 2.52 (p< 0.0001).

The percent contribution of additive diversity at each level is given in Fig. 2. Expected within additive diversity for all nested-levels is greater than the observed diversity (p < 0.0001). The most notable difference between the observed and expected data is the contribution of species richness among sites. Forty-five percent of species richness came from among sites in the observed data, while it was expected to be only 21%(p<0.0001).

Species Richness in Cusuco

A total of 18 morphospecies were collected for the park. A species accumulation curve (Fig. 3) had a slope (z) of 0.33 leading to an estimate of >400 species in Cusuco National Park. Table 5 contains species calculations for slopes ranging from 0.10 to 0.40. Slopes between 0.20 and 0.25 seem to produce the most reasonable estimate for the number of species that may be found in the park (43 and 112, respectively). The common distribution of z values range from 0.20 to 0.40 in log-log transformed species curves (Connor & McCoy, 1979).

The scatterplot created for the number of morphospecies collected at each site and their elevation did not show a relationship (Fig. 4), with a $r^2=0.00$. The scatterplot comparing abundances of Opiliones and elevation at each site also did not show any relationship (Fig. 5) with an $r^2=0.28$. Once Santo Tomas is removed and only the other five sites are compared the r^2 values increase to 0.87 and 0.77 respectively.

Table 2: Total abundance of each morphospecies in Cusuco National Park.

Morphospecies	Abundance
Paecileama pectigenerum	6
Prionostemma sp. 3	7
Prionostemma sp. 1	8
Cynorta dentipes	9
Metopilio sp. large	10
Prionostemma juvenile	11
Eucynortella sp. legs	12
Paecileaema tolendense (juveniles)	13
Eucynorta sp. solid	14
Sp13_Palp	15
Prionostemma sp. 4	16
Eucynorta sp. name	17
Unknown Species 6	18
Prionostemma sp. 5	19
Metopilio sp. small	20
Eucynorta sp. 1	21
Prionostemma juvenile 2	22
Cynorta bromeliacea	23
	24
Total number of individuals collected	25

Table 3: Number of morphospecies unique to each site, number of individuals, and

morphospecies collected at each site sampled.

Sites	morphospecies	no. individuals	Unique morphospecies
Base Camp	9	46	1
Cantilles	10	47	2
Guanales	11	48	3
Cortecito	12	49	4
El Danto	13	50	5
Santo Tomas	14	51	6

Table 4: Pairwise estimates of community similarity among sites, based on Jaccard index.

	Base Camp	Cantilles	Guanales	Cortecito	El Danto
Base Camp					
Cantilles	0.500				
Guanales	0.200	0.000			
Cortecito	0.154	0.182	0.250		
El Danto	0.300	0.250	0.167	0.222	
Santo Tomas	0.071	0.091	0.000	0.182	0.000

Percent Contribution to overall Diversity (Shannon Diversity)
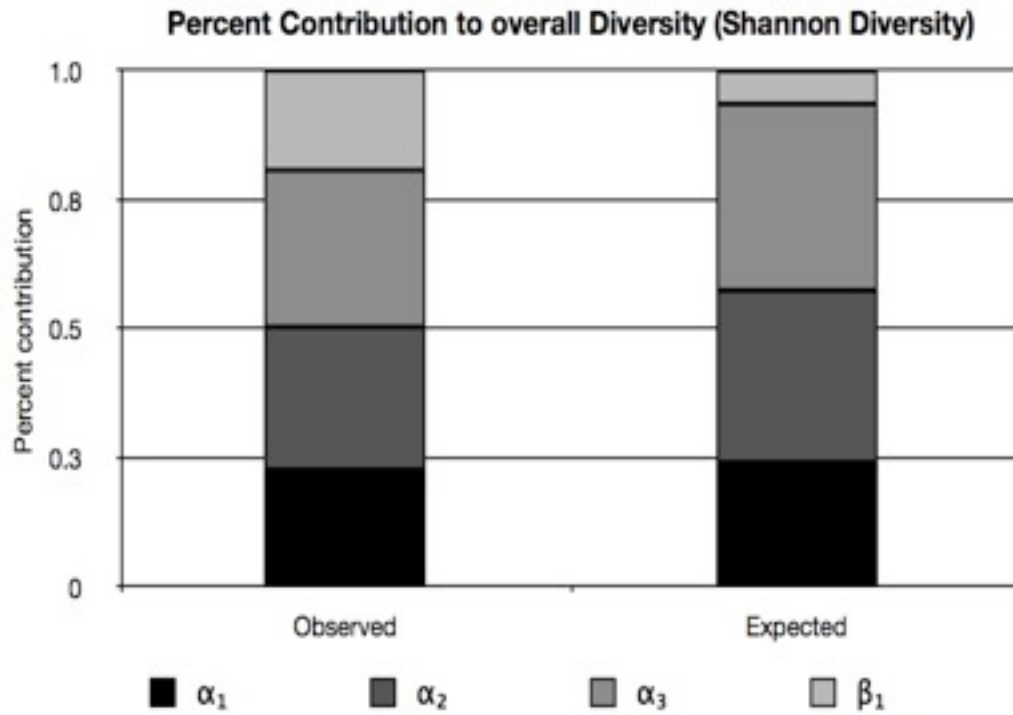
Fig. 1: Percent contribution of Shannon diversity, at four hierarchal levels, to overall diversity, observed and expected values (from PARTITION). α_1 is within subplots, α_2 is within plots, α_3 is within sites, and β_1 is among sites.
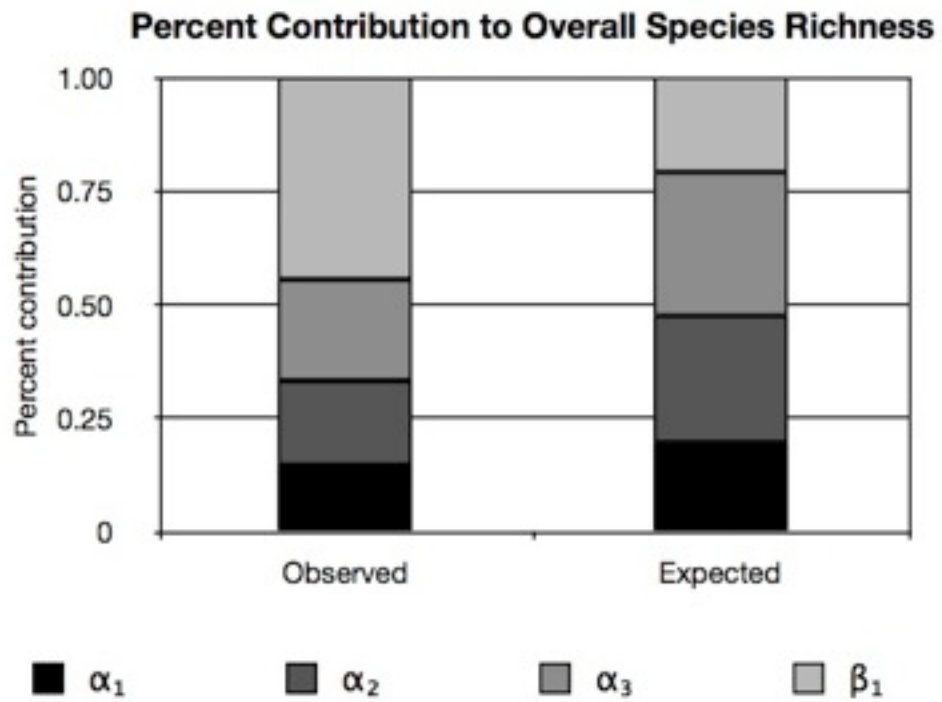
Fig. 2: Percent contribution of additive diversity (species richness) to overall species richness, at four hierarchal levels, observed and expected values (from PARTITION). α_1 is within subplots, α_2 is within plots, α_3 is within sites, and β_1 is among sites.

Species Richness and Area Relationship

$\log y = 0.3263 \log x - 0.044$

$r^2 = 0.8673$
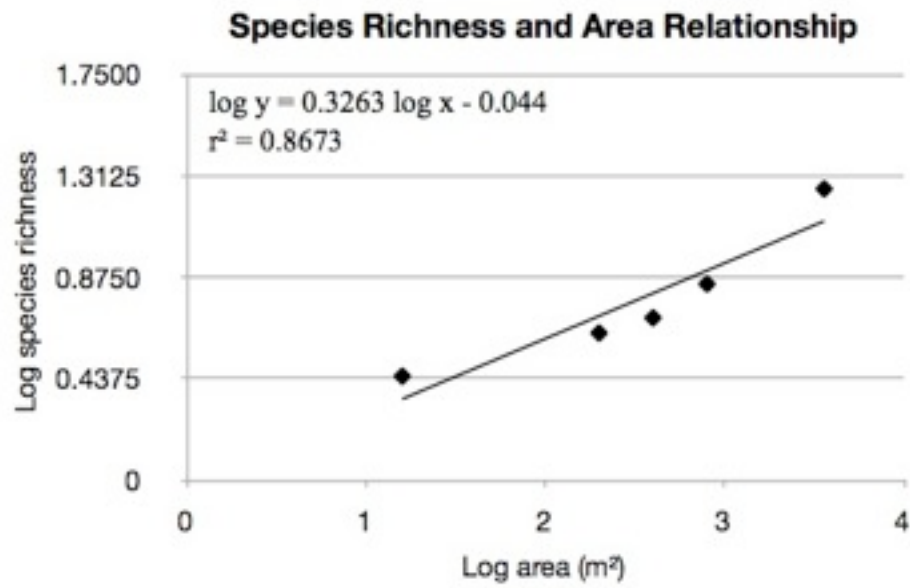
Fig. 3: Species -area curve; log (10) transformed species richness of Harvestmen, and log (10) transformed collection area.

Table 5: Estimated total number of species for Cusuco National Park, Honduras, for

different values of z.

z	S
0.35	768
0.30	293
0.25	112
0.20	43
0.15	16
0.10	6

Species Richness and Elevation of Sites
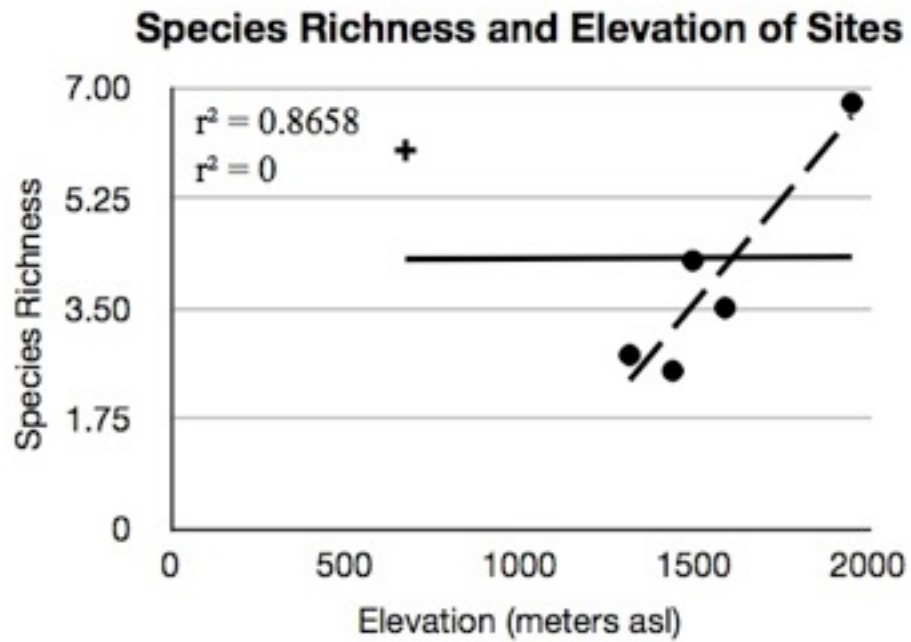
Fig. 4: The number of Morphospecies collected and elevation of each site, with and without the Santo Tomas (+) site inclued. The larger r² value describes the dashed line (with the Santo Tomas site removed). The smaller r² value describes the solid line (with the Santo Tomas site included).

Abundances and Elevation of Sites

$r^2 = 0.7728$

$r^2 = 0.2755$
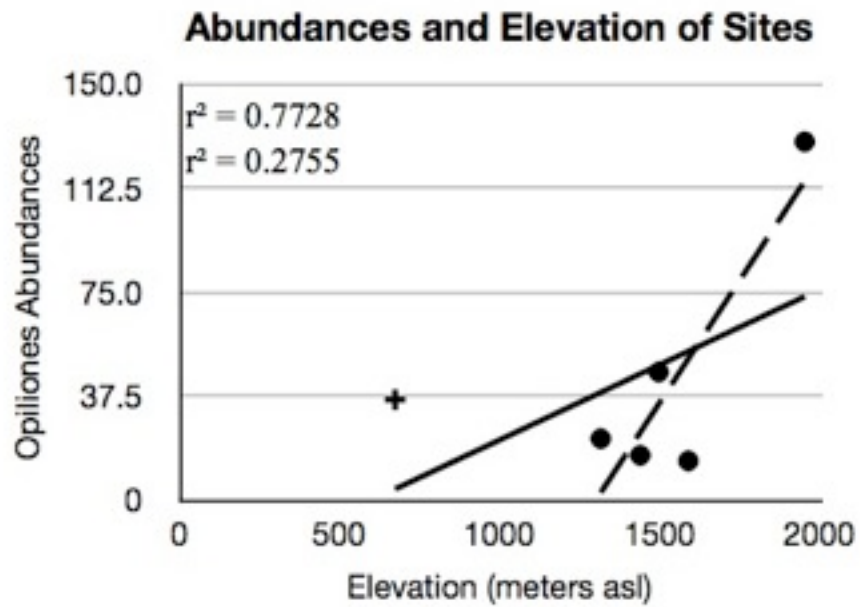
Fig. 5: The abundance of Opiliones at each site and their eleavtions, with and without the Santo Tomas (+) site inclued. The larger r^2 value describes the dashed line (with the Santo Tomas site removed). The smaller r^2 value describes the solid line (with the Santo Tomas site included).

DISCUSSION

The purpose of this study was to measure and catalog the Opiliones diversity within Cusuco National Park. This study also examined whether diversity was primarily distributed within or among sites. Both Shannon and additive diversity among sites was higher than expected, and lower than expected within sites. If I adjusted my species area curve to a slope of 0.20 and 0.25, I would expect to find 43 to 112 species within Cusuco. Slopes ranging from 0.20 to 0.40 are common log species/log area relationships (Connor, & McCoy, 1979). Terrestrial invertebrates tend to fall closer to 0.25, depending on the invertebrate being studied, and whether they are on an island or mainland (Gould, 1979; Sfenthourakis, 1996; Werner, & Burzko, 2005). In a study looking at European butterflies, the SAR slope was approximately 0.23 (Werner, & Burzko, 2005). This may be similar to what is seen in Opiliones, since the distribution of butterflies are dependent upon host plants and therefore may be restricted. Opiliones have low dispersal abilities and should have a similar, or potentially higher slope.

I did not expect the site with the most morphospecies and most individuals to be the highest elevation site. Base Camp and Cantilles had the highest number of morphospecies, and highest abundance respectively. Both of these sites are at or above 1400m asl, and Cantilles is especially dry. Santo Tomas did have the highest number of unique species, and had a similar abundance to Base Camp. Morphospecies richness and abundance does not appear to follow an elevation or moisture gradient. Pearson correlation values from the scatterplots including all sites were both low, indicating that

no pattern was evident according to elevation. Santo Tomas being removed did greatly

increase the correlation values. I believe this pattern may have been observed because

deforestation and human disturbance is lowest at Cantilles, and to a lesser degree, Base

Camp. Santo Tomas is easily accessible from outside the park, and deforestation is

especially high at this site (personal observation). Opiliones can be sensitive to changes

in microclimate caused by deforestation (Bragagnolo, et al. 2007), possibly explaining

the lower richness. This pattern may have also been an artifact of only one plot being

sampled at Santo Tomas, unlike in Cantilles and Base Camp.

My hypothesis that beta diversity would contribute more to overall diversity than

expected was supported by both Shannon diversity and Additive diversity analysis

conducted using PARTITION. As a greater spatial extent was sampled, habitat

heterogeneity increased due to the differences in elevation among sites. Cusuco National

Park has a wide range of elevations (Cusuco Park Report, 2011), which allows more

species to coexist within the park. Opiliones have not been used in nested-level diversity

studies before, but Crist et al. (2003) used beetle species found in tree canopies. They

looked at multiple levels of analysis, from samples within a tree, up to stands of trees.

The researchers found that their higher nested levels of sites and stands contributed more

to β-diversity than lower nested levels. The authors concluded that this was in part due to

rare species being found in only a single stand of trees or a single tree, while more

common species were more widely distributed. This is similar to the pattern I observed

in my data, with increased beta diversity as spatial extent increased.

Prior to my study, Honduras had only nine known species. I have added one species previously known from Belize, and an additional 14 undescribed species. This would bring the number of Opiliones species in Honduras to 24 species. This known fauna is now similar to those of the neighboring countries of Guatemala and Belize (51 and 30 species, respectively, Townsend, et al., 2010). The number of species found in Guatemala, Belize and Honduras should still be considered underestimations. From the modeled SAR slope of 0.25-0.30, the estimated species within Cusuco National Park alone would be 47 to 102, comparable to both Guatemala and Belize's overall diversity. Costa Rica is the best inventoried country in Central America, with 121 described species (Townsend, et al., 2010). Even this fauna is likely an underestimate. The most thoroughly described Neotropical Opiliones fauna is that of Brazil. Pinto-da-Rocha, et al., (2005) found that for one family (Gonyleptidae) in Atlantic Rainforest in Brazil there were a total of 363 species. Many studies of Opiliones focus on Brazil, and the number of known species there is approximately 1000 (Kury, 2003).

One of the biggest limitations of this study was the limited area sampled. Of the established plots found within Cusuco National Park, only about nine percent were sampled in my study. Sampling more plots at each site would increase the area sampled, improve species composition estimates of each site, and ultimately improve the richness estimate for the park derived from the species-area curve. More sampling may show that species that were rare in my standardized sampling are not rare at all. If so, the beta diversity structure reported in this study might break down. Coddington, et al., (2009) found in extensive tropical arthropod studies that singleton species accounts for an

average 32% of all species collected. The researchers used a data set from an arachnid survey to test if many hypotheses for singleton species were found to be true. They found that in the lognormal modeled communities that actual singleton species percentage was only about 4% (Coddington, et al., 2009). They reasoned that sampling should be increased spatially and temporally. If I increased my sampling spatially and temporally, I could better predict the number of species in Cusuco National Park.

Additional standardized sampling protocols could have been included in this study.   Intense leaf litter sampling could collect individuals that are less obvious when doing timed manual searches. Many Cyphophthalmid Harvestmen are only found in leaf litter and soil, and are especially small and easy to miss with my sampling protocols. Proud et al. (2012) found that Opiliones in Costa Rica are predominantly found on and in the leaf litter, while individuals were only rarely spotted on vegetation. By intensively sampling leaf litter at more sites in Cusuco, researchers should be able to increase the number of species known to occur within the park. Any collection of these individuals would increase the Opiliones fauna for Honduras. No standardized sampling was conducted above human head height. Sampling the subcanopy and canopy could contribute additional species discoveries. Very little is known about arthropod assemblages in tropical forest canopies (Erwin, 1983).

Fifteen new species have been added to the Opiliones fauna of Honduras. I found that sampling among sites contributed most to overall diversity. I was incorrect in hypothesizing that species richness and abundance would be greatest at low elevation sites, due to warmer temperatures and higher humidity. This study will be helpful to

future researchers examining zoogeography and community ecology of Opiliones. Future research should focus on specific factors that influence the Opiliones community structure. Most research thus far has looked at factors such as temperature and relative humidity, while few have looked at prey availability, predation rates, and parasitism, and their effects on community composition structure. More importantly, future research should focus on obtaining a more complete Opiliones fauna for Honduras. This study reveals that there are probably many new species to be found in Cusuco National Park. Doing timed searches at different times of year, should provide adults of species not normally found during a summer survey, and provide adults for the species of juveniles that were collected in this study. To establish a complete species list for Cusuco surveys should be conducted for a few years, or until new species are not found, to establish a complete species list. Once complete, this could serve for a model in other preserves throughout Honduras and elsewhere.

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APPENDIX A

Description of Morphospecies of Cusuco National Park, Honduras. All regular photography was done with a SONY NES5, with an 50-150 mm lens. SEM photography was completed with a Hitachi S-3400N, using an accelerating voltage of 10kV.

Paecileama pectigenerum (Sideburns_Lan MID00023) (Fig. A1; a & b, Fig. A2): This

species belongs to the Cosmetidae family, which is apparent from the palps being wider

on the tibial segment. Individuals in this species are red with yellow markings. The

markings are fine and on the sides of the ocularium, as well as some fine markings found

along the margin of the prosoma (cephalothorax). Males differ from females by having

enlarged chelicerae and being armed on the IV pari of legs. Scanning electron

microscopy (SEM) photography was completed for male genitalia (Fig. A2). Reference

specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "sp.3" (Cheeks_Palp, MID00107) (Fig. A3; a & b, Fig. A4): This species

belongs to the Sclerosomatidae family. The body is predominantly black, with yellow to

white markings in from of the ocularium. The II pair of legs are long, the longest legs on

an individual measured about 6cm. The legs are completely black except for the II pair

of legs which has a white spot on the second joint. SEM pictures were taken of female

and male genitalia. Male genitalia was not complete after preparations and the base was

all that was photographed. The female genitalia has densely packed hairs in clusters in

close proximity to the semical opening. Hairs along the ovipositor are similar in length to one another. The male genitalia base is spade shape. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "sp. 1" (Cyclops_Palp, MID00024) (Fig. A5; a & b, Fig. A6; a & b): This species belongs to the Sclerosomatidae family. The body is completely orange, while the ocularium and eyes are black. The legs are black as well as where the legs attach to the body. Between the III and IV pair of legs there is a white spot. The mouth parts are white. SEM photography was done for both males and females. Female ovipositor's last segment length is twice the width, and there are a row of setae on each side of the midline. Male genitalia has a straight stylus with elates coming off of the base. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Cynorta dentipes (Orangekneed_Lan MID00096) (Fig. A7; a & b, Fig. A8): This species belongs to the Cosmetidae family, and can be identified as *Cynorta dentipes* because of the eight tarsal segments on the III pair of legs. The body is light red to orange in color, and has yellow incomplete lines around the edges of the scutal areas. There are two spikes on the IV scutal area. The legs have yellow and black banding and males have a prominent spine on the either end of the femur of the IV pair of legs. SEM work was completed for a single male individual. The penis has paddle-shaped projections on the

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distal end, and has five setae along the edges. Reference specimen are kept at Oxford

Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Metopilio "sp. large" (Lightstripe, Stamped, and Spiked, MID00069, MID00082,

MID00030) (Fig. A9; a, b, c, d, e, & f, Fig. A10; a & b): Originally categorized as three

different species, but after doing SEM work found that they all belonged to the *Metopilio*

family group, and are all the same species. All are Palpitorids with short legs and large

bodies. On all types of the species there are two spikes located about halfway down the

length of the body. All have short legs, and have bumps along the edge of the

opisthosomal tergites. All legs are banded dark and light. The difference between the

types is the predominant markings found on the body. The Lightstripe variety had dark

areas around the spikes, and has yellow markings posterior to the spikes, and along the

midline in stripe. The Stamped variety looks much the same, but only has the yellow

"stamp" posterior of the spikes. The Spiked variety has similar markings to the

Lightstripe individuals, but has no noticeable yellowing in the patterns. SEM was

completed for both males and females. Female ovipositors are not typical for the

Metopilio group having a wider base than usual. There are a row of setae on each

segment, the distal hairs being longer than the posterior hairs, maintaining a specific

width down the length of the ovipositor. The male's penis is similar to other species in

Metopilio, but the stylus on the distal end is more corkscrew shaped than usual. There is

an unusual sulcus in the gland/stylus portion of the penis, and the penis is larger than

other *Metopilio*, maybe indicating that this species does not belong in this group.

Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "juvenile" (Mask_Palp MID00184) (Fig. A11; a & b): This species is from the Sclerosomatidae family. The individuals collected during this study were juveniles, and no SEM was able to be completed on them. Bodies are mostly dark, with a white "mask" in front of the ocularium. Palps have an additional appendage, not simple and straight. Legs are all dark except for light marking at the joints of the legs. These individuals do have similar bumps along the opisthosomal segments like the *Metopilio* sp. large morphospecies. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Eucynortella "sp. legs" (Legs_Lan, MID00138) (Fig. A12; a & b): This species belongs to the Cosmetidae family. SEM work was unable to be completed for a male specimen of this species, only females were collected. The individuals collected in this species has an orange body with yellow markings along the edges of the scutal areas. Each scutal area has dark coloration within it and no scutal area has any armature. The body is relatively flat which is common to the *Eucynortella* genus. The legs have alternating dark and light marks. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

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Paecileama tolendense (Ridge_Lan/Mayan Juvenile MID00185) (Fig. A13; a & b, Fig.

A14): This species belongs to the Cosmetidae family. The adult individuals are large

(more than 10mm in length). The body is dark red, with very fine yellow markings on

the body. No SEM work was completed because no adult males were available from the

sample. An adult picture is included, but no adults were collected in this study. Juveniles

are dark red, with long palps, and usually fluoresce red under UV light. Reference

specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Eucynorta "sp. solid" (Solid_Lan MID00032) (Fig. A15; a & b, Fig. A16): This species

belongs to the Cosmetidae family. SEM work was completed for male specimen

collected. The body seems to belong to the *Eucynorta* genus but not exactly typical for

the genus. The body is round, and dark red in color. The yellow markings have quite a

bit of variation. Some individuals will be completely colored yellow on the scutal areas,

while some other ones will only have the yellow markings around the outside of the

scutal areas. The penis structure is different in that the anterior end is more straight than

other Laniatorids. There are four setae along the edges of the penis. The two most distal

setae have a ring along the base, which is uncommon. A close up picture of this has been

taken. Reference specimen are kept at Oxford Natural History Museum, Parks Rd,

Oxford, Uk. OX1 3PW.

Species 13 (MID00186) (Fig. A17; a & b): This species was a juvenile, and it is to

difficult to tell what family, or genus it belongs to. Each instar can be very different from

the next even within a single individual. The whole body is very dark, underside as well.

The palps have appendages on it similar to the *Prionostemma* "juvenile". Reference

specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "sp. 4" (Species 14, MID00187) (Fig. A18; a & b, Fig. A19; a & b): This

species belongs to the Sclerosomatidae family. SEM work was completed for male and

female genitalia. The body is dark dorsally, and ventral side is light brown or orange,

with all black legs. The body has small bumps along the opisthosomal tergites. There are

no additional appendages on the palps. The ovipositor is more long and slender than

other *Prionostemma* species. The hair on the tuft areas on the distal end are short and

widely dispersed. Hairs on the shaft get shorter towards the posterior end of the

ovipositor. The penis is similar to other *Prionostemma* but the elytra on the sides are

more noticeable. Reference specimen are kept at Oxford Natural History Museum, Parks

Rd, Oxford, Uk. OX1 3PW.

Eucynorta "sp. name" (NoName_Lan MID00188) (Fig. A20; a & b): This species

belongs to the Cosmetidae family. No SEM work was able to be completed, since it

seemed that the penis was lost during preparation. The species is red in color with fin

yellow markings along the edge of the scutal areas. Males have large combs on the femur

of the III pair of legs, and enlarged chelicerae. There is armature in both males and

females on the IV scutal area, and the ends of the spines become lighter in color.

Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Unknown Species 6 (MID00089) (Fig. A21; a & b): This species was unable to be identified. It is a small Eupnoi, and is very dark in color. It may be similar to the *Prionostemma* collected in this study, since it has an additional appendage on the palps. The palps, and chelicerae are dark, but lighter than the rest of the body. The legs are long, and completely black. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "sp. 5" (Species 7 MID00189) (Fig. A22; a & b, Fig. A23): This species belongs to the Sclerosomatidae family. SEM was completed for only female genitalia. The body is predominantly dark with a lighter stripe down the midline of the body. Legs are dark except for a light patch at the joints of the legs. There are fine markings around the ocularium that are barely noticeable except under magnification. Mouthparts are simple and yellow in color. Female ovipositors have longer setae on the distal end, and the tufts at the end are less densely covered. There is a bulge just after the semical opening, and segmentation is very wide here. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Metopilio "sp. small" (Stripey_Palp MID00165) (Fig. A24; a & b, Fig. A25; a & b): This species belongs to the *Metopilio* group family. SEM work was completed for male

genitalia. The body on this species is orange in color, and has black markings/ stripe that starts at the ocularium, and continues along the midline. There are two back spikes about midway down the opithosoma. Legs are all black and long. SEM work showed that the penis for this species has a more cork screw-shaped stylus than other species within the *Metopilio* group. Elytra are noticeable, but not large. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Eucynorta "sp. 1" (Upsidedown_Y_Lan, MID00168) (Fig. A26; a & b): This species belongs to the Cosmetidae family. No SEM work was able to be completed, since it appears as if the penis was destroyed during preparation. The body is overall dark red, with yellow markings on each side of the ocularium, and down the midline of the body, producing a marking that looks like a "Y". Males have enlarged chelicerae, and a comb-like structure on the femur of the IV pair of legs. Reference specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Prionostemma "sp. 2" (Whitemouth_Palp MID00060) (Fig. A27; a & b, Fig. A28): This species belongs to the Sclerosomatidae family. SEM work was completed for both male and female genitalia, but males genitalia was damaged during the process. The body of this Palpitorid is all black with some iridescence on the body. Mouth parts are yellow, or white, and the legs are long and black. Body is especially small. The ovipositor has very narrow bands posterior to the semical opening. Setae on the distal end are much longer the setae on the base of the ovipositor. Base of the ovipositor is wider than the distal end,

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and each band has a single row of hairs on it. Reference specimen are kept at Oxford

Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Cynorta bromeliacia (Y_Lan, MID00037) (Fig. A29; a & b, Fig. A30): This is a known

species to Honduras in the Cosmetidae family. SEM work was completed on just male

genitalia. The body is dark red in coloration with yellow markings. The yellow markings

go along both sides of the body, and along the posterior end of the opithosoma. There is

armature (two spines) on approximately the IV scutal tergites. The males typically have

enlarged chelicerae, and have a comb-like structure on the femur of the IV pair of legs.

Using SEM photography, it can be seen that the male penis is almost trapezoid in shape

on the distal end. There are six hair like setae along the sides of the penis. Reference

specimen are kept at Oxford Natural History Museum, Parks Rd, Oxford, Uk. OX1 3PW.

Fig. A1 a. Photograph of *Paecileama pectigenerum*
b. Close up photograph of the body of *Paecileama pectigenerum*.

a.

b.
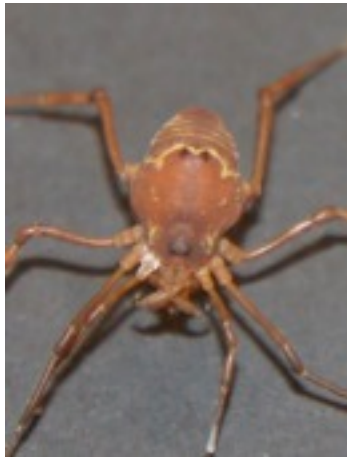
Fig. A2. Ventral view of *Paecileama pectigenerum* penis.

S3400 10.0kV 19.7mm x270 SE 3/1/2013   200um
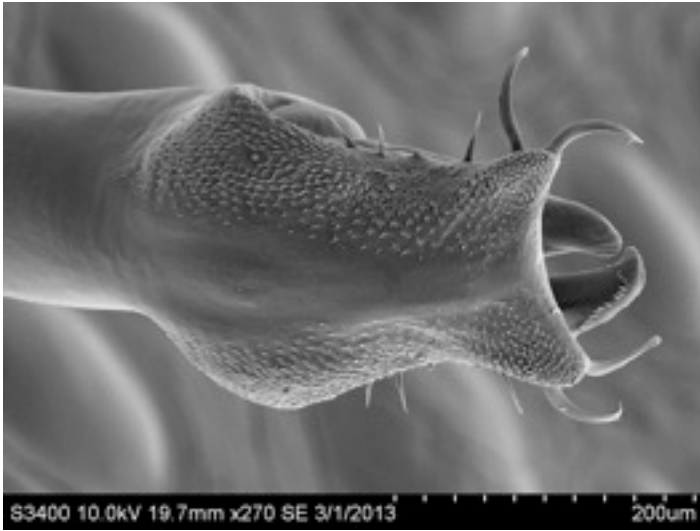
Fig. A3, a. Photograph of *Prionostemma* "sp. 3"
b. Close up photograph of *Prionostemma* "sp. 3".

a.

b.
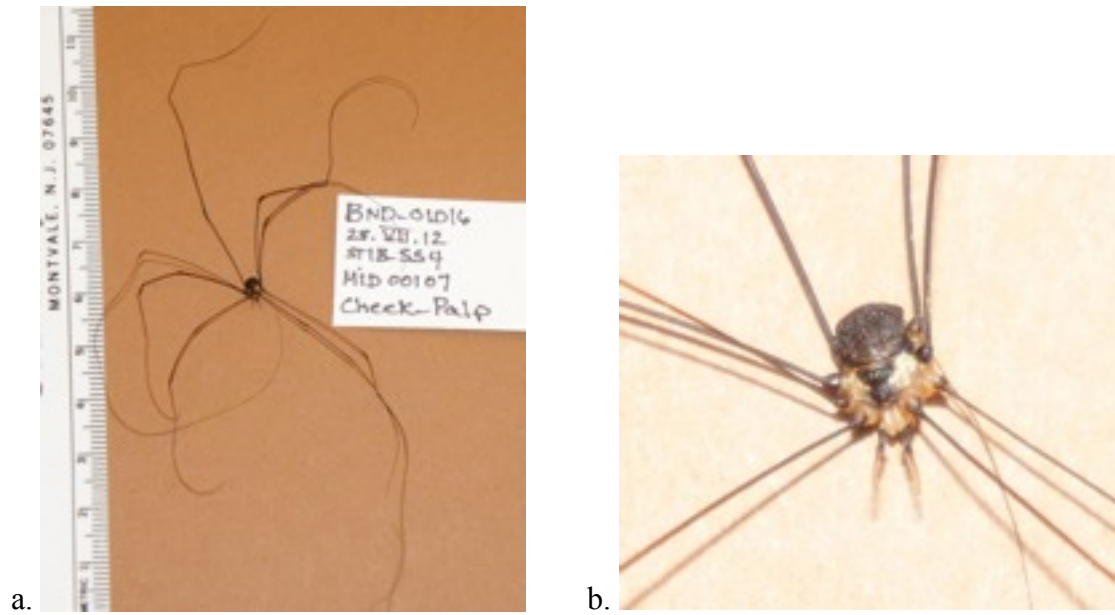
Fig. A4. Dorsal view of ovipositor of *Prionostemma* "sp. 3".
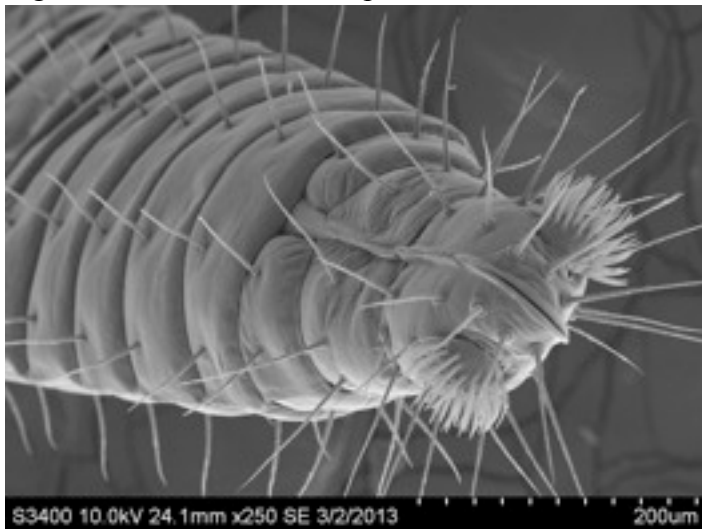
Fig. A5 a. Photograph of *Prionostemma* "sp. 1".
b. Close up photograph of *Prionostemma* "sp. 1".

a.

b.
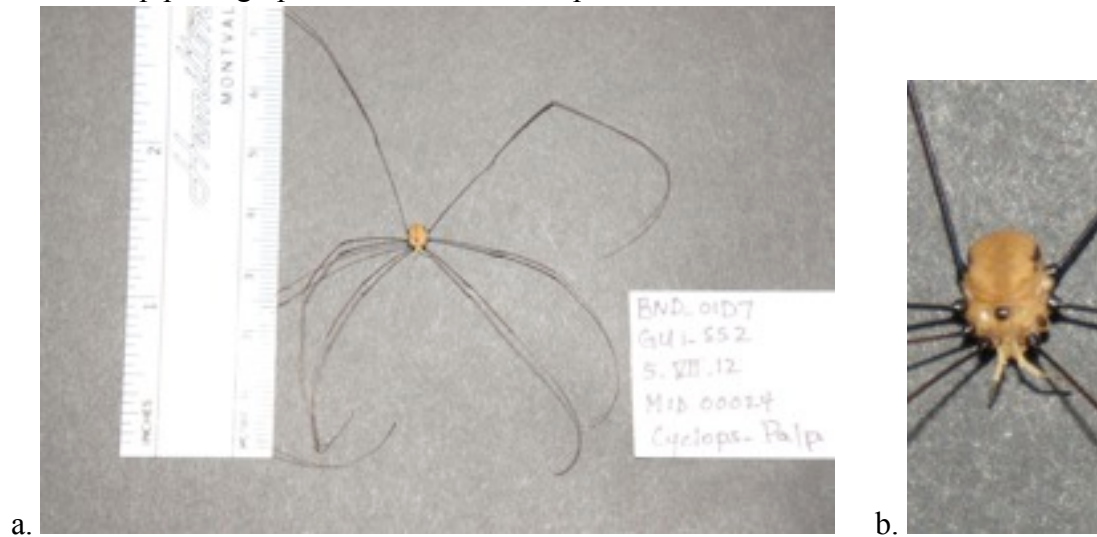
Fig. A6 a. *Prionostemma* "sp. 1" dorsal view of ovipositor
b. *Prionostemma* "sp. 1" dorsal view of penis

a.

b.
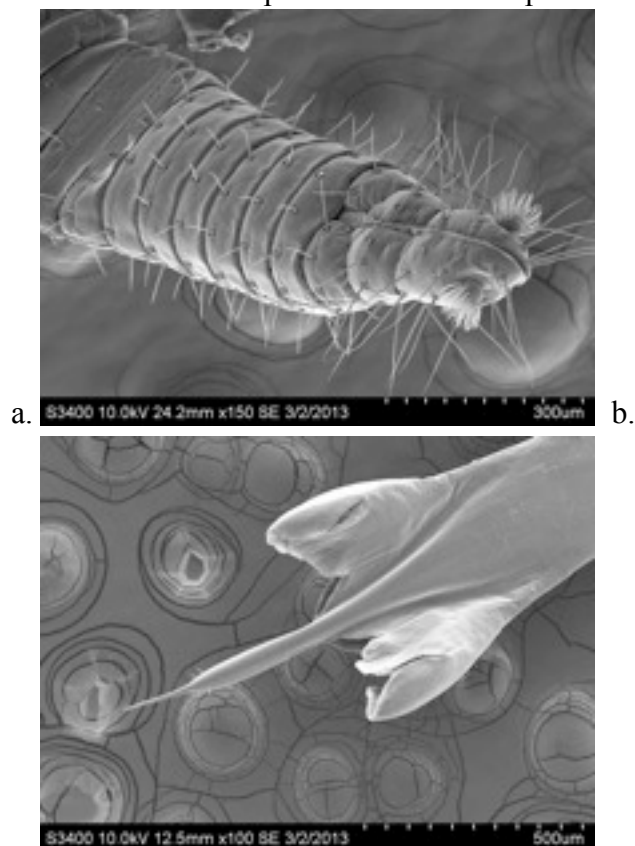
Fig. A7 a. Photograph of *Cynorta dentipes*
b. Photograph of the body of *Cynorta dentipes*

a.   b.
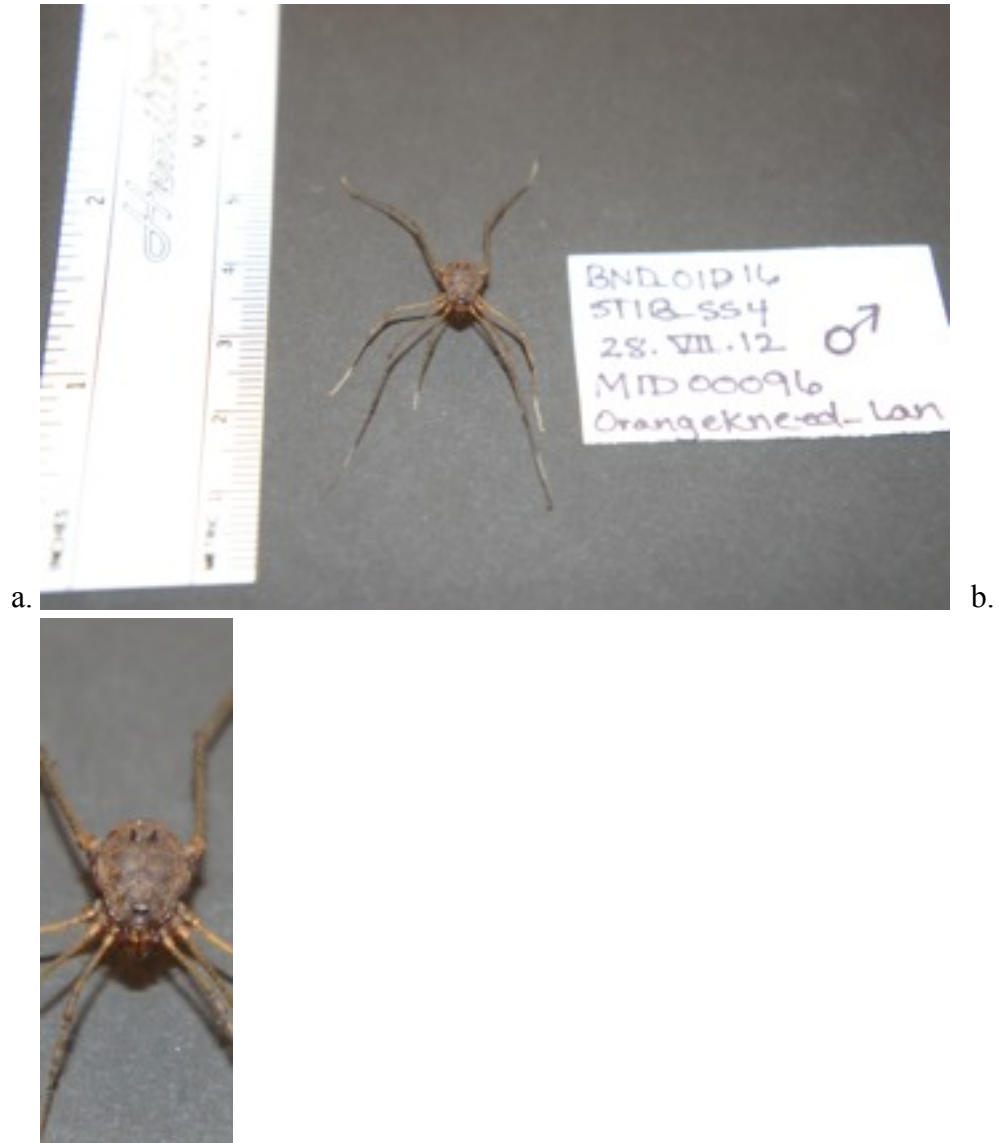
Fig. A8 Ventral view of *Cynorta dentipes* penis
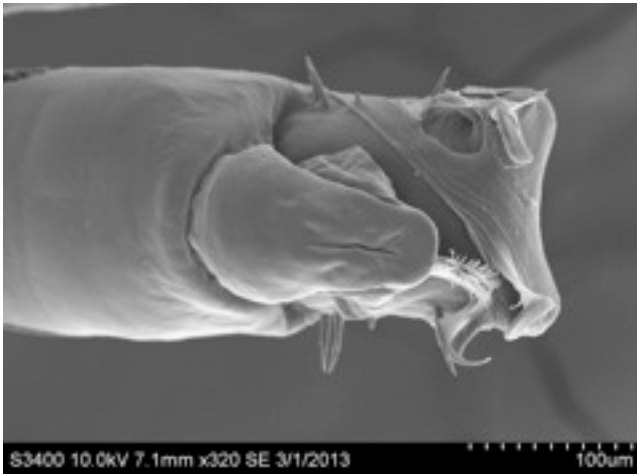
Fig. A9 a. Photograph of *Metopilio* "large", variety lightstripe.
b. Body of *Metopilio* "large" variety lightstripe.

a.

b.

c. Photograph of *Metopilio* "large" variety stamped
d. Body of *Metopilio* "large" variety stamped.

c.

d.

e. Photograph of *Metopilio* "large" variety spiked.
f. Body of *Metopilio* "large" variety spiked.

e.

f.

Fig. A10 a. Dorsal view of *Metopilio* "large" penis.
b. Dorsal view of *Metopilio* "large" ovipositor.
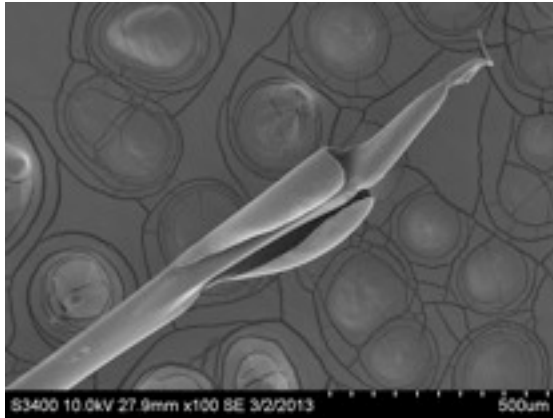
a.

b.
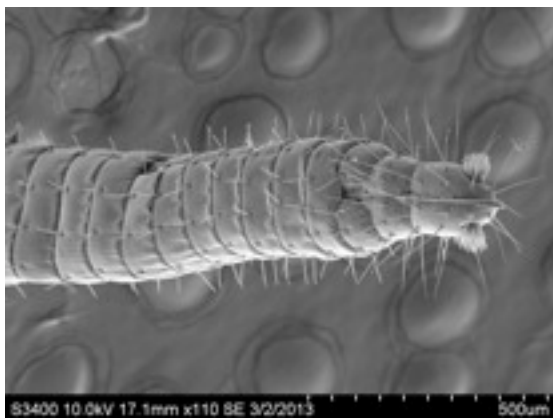
Fig. A11 a. Photograph of *Prionostemma* "juvenile"
b. Body of *Prionostemma* "juvenile"

a.

b.
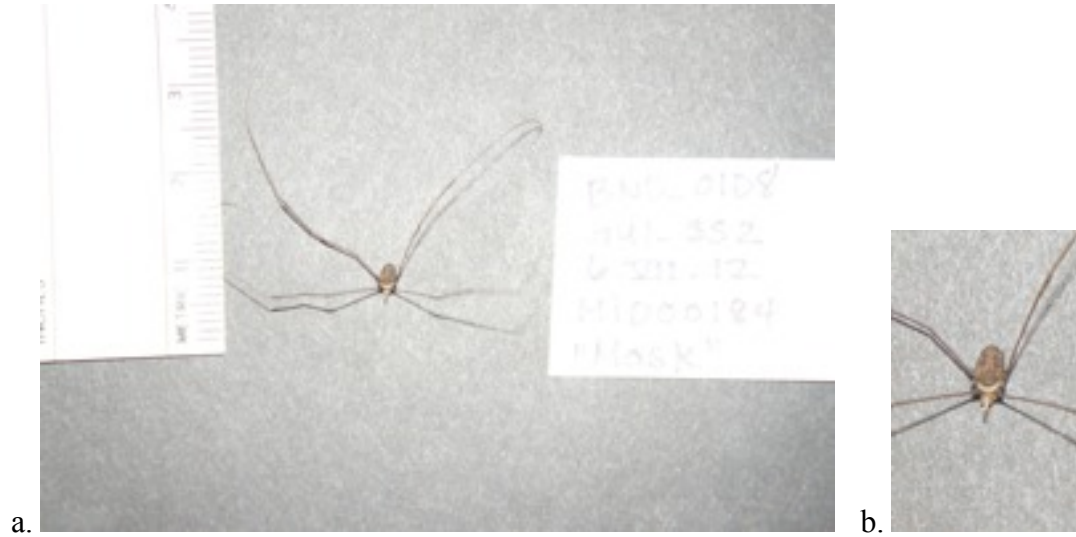
Fig. A12 a. Photograph of *Eucynortella* "sp. legs"
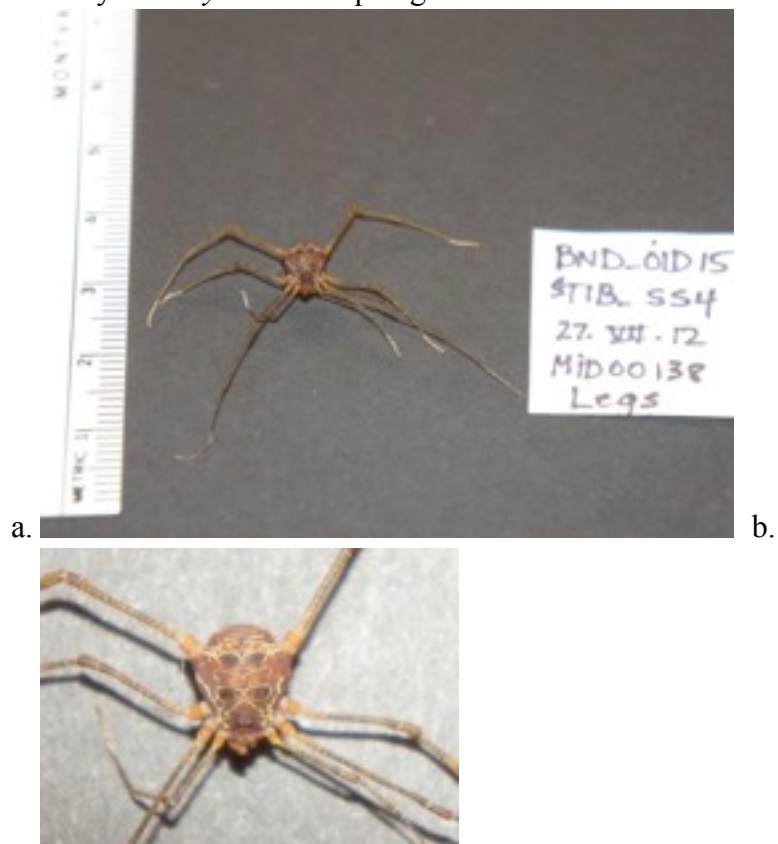b. Body of *Eucynortella* "sp. legs"

a.   b.

Fig. A13 a. Photograph of *Paecileama toledense* juvenile.
b. Body of *Paecileama tolendense* juvenile.

a.

b.

Fig. A14. Adult specimen of *Paecileama tolendense*. This individual was not collected durng standardized sampling, but was an opportunistic capture.

Fig. A15 a. Photograph of *Eucynorta* "sp. solid"
b. Body of *Eucynorta* "sp. solid"

a.

b.

Fig. A16. Ventral view of *Eucynorta* "sp.solid" penis.

Fig. A16 a. Photograph of Species 13, juvenile.
b. Body of Species 13, juvenile.
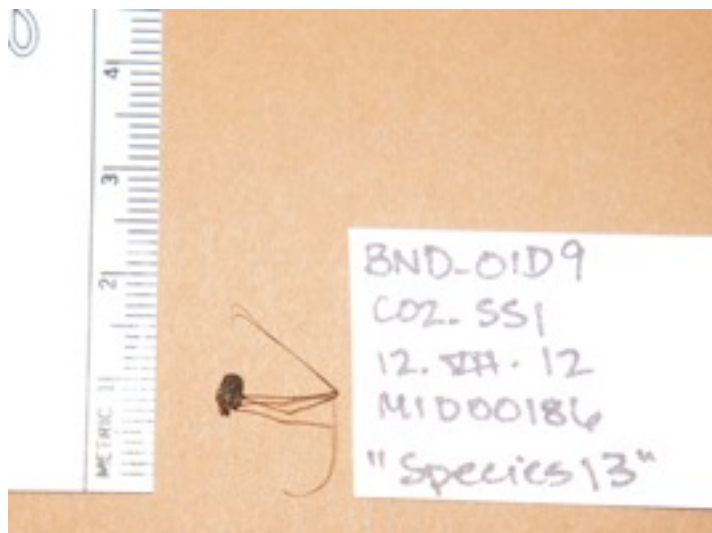
a.

b.

Fig. A18 a. Photograph of *Prionostemma* "sp. 4"
b. Body of *Prionostemma* "sp. 4"

a.   b.

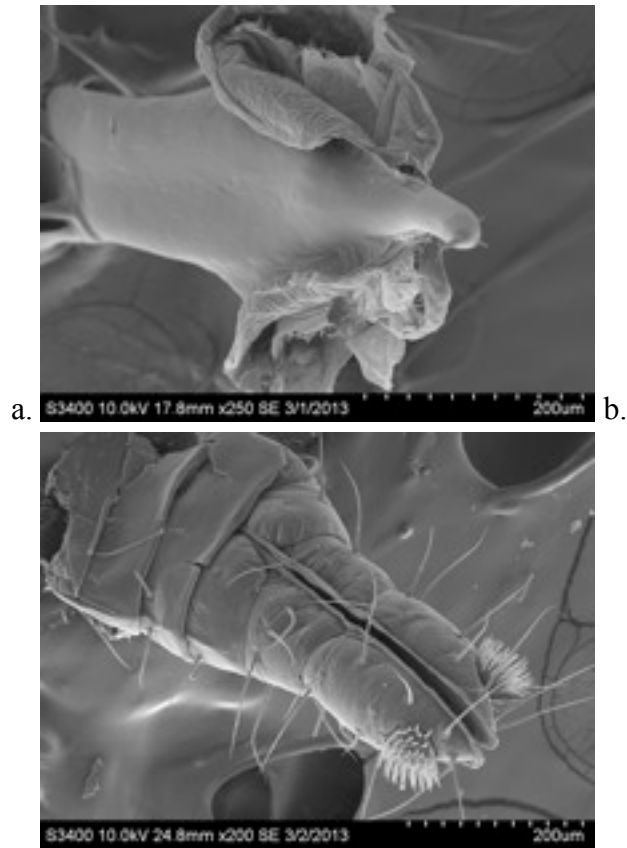Fig. A19 a. Dorsal view of *Prionostemma* "sp. 4" penis.
b. Dorsal view of *Prionostemma* "sp. 4" ovipositor.

a. b.

Fig. A20 a. Photograph of *Eucynorta* "sp. name"
b. Body of *Eucynorta* "sp. name"

a. 　　　　　　　　　　　　　　　　　　　　　　　b.

Fig. A21 a. Photograph of Unknown species 6.
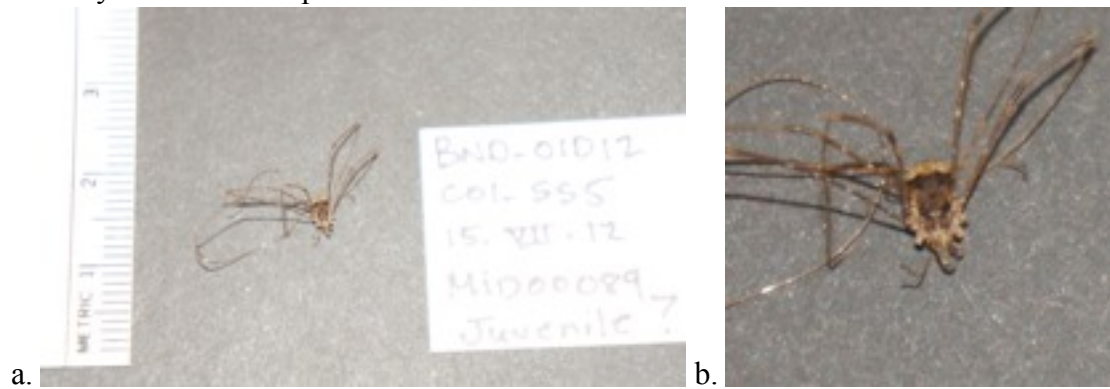b. Body of Unknown species 6.

a. 　　　　　　　　　　　　　　　　　　　　　　　b.

Fig. A22 a. Photograph of *Prionostemma* "sp.5".
b. Body of *Prionostemma* "sp.5".

a.

b.

Fig. A23. Dorsal view of *Prionostemma* "sp.5" ovipositor.

Fig. A24 a. Photograph of *Metopilio* "sp. small".
b. Body of *Metopilio* "sp. small".

a.

b.

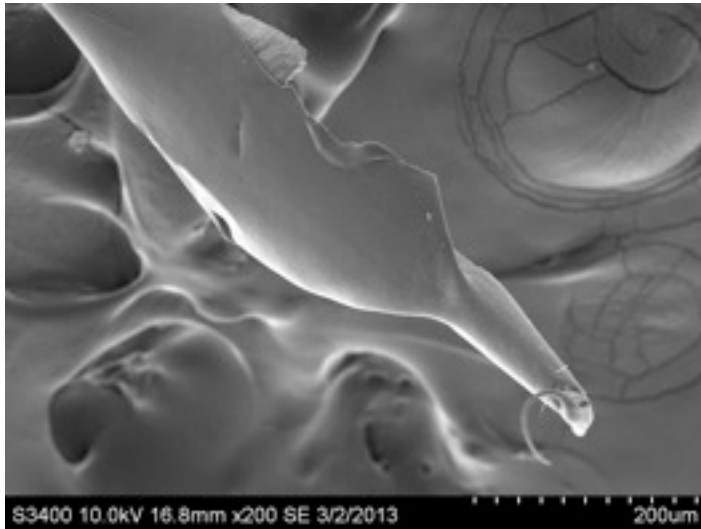Fig. A25 Dorsal view of *Metopilio* "sp. small" penis.

Fig. A26 a. Photograph of *Eucynorta* "sp.1".
b. Body of *Eucynorta* "sp.1".

a.

b.

Fig. A27 a. Photograph of *Prionostemma* "sp.2".
b.Body of *Prionostemma* "sp.2".

a.

b.

Fig. A28. Dorsal view of ovipositor of *Prionostemma* "sp.2".

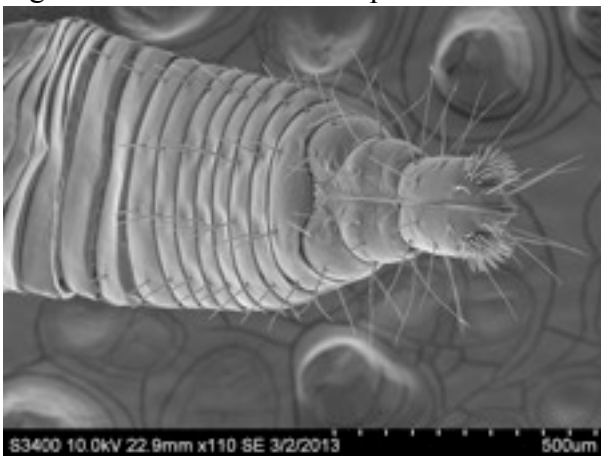Fig. A29 a. Photograph of *Cynorta bromeliacia*.
b. Body of *Cynorta bromeliacia*.

a.

b.

Fig. A30. Ventral view of *Cynorta bromeliacia* penis.