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## Survey of a Neotropical anuran assemblage (Pacaya-Samiria Reserve, Peru)

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SURVEY OF A NEOTROPICAL ANURAN ASSEMBLAGE  
(PACAYA-SAMIRIA RESERVE, PERU)

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

By

ERIC WOEBBE  
B.S., Wright State University, 2010

Wright State University  
2017

WRIGHT STATE UNIVERSITY

GRADUATE SCHOOL

April 18, 2017

I HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER MY  
SUPERVISION BY Eric Woebbe ENTITLED Survey of a Neotropical anuran assemblage (Pacaya-Samiria Reserve, Peru) BE ACCEPTED IN PARTIAL FULFILLMENT OF THE  
REQUIREMENTS FOR THE DEGREE OF Master of Science.

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## **ABSTRACT**

Woebbe, Eric. M.S., Department of Biological Sciences, Wright State University, 2017. Survey of a Neotropical anuran assemblage (Pacaya-Samiria Reserve, Peru).

The Pacaya-Samiria National Reserve of Peru is one of the largest protected areas of the Amazon rainforest, yet it has hosted only a few studies of frogs and toads (anurans). The primary goals of my study are: 1) To conduct an inventory of the local anuran species, 2) to quantify richness among habitats, 3) to compare differences in beta diversity across a river versus a continuous landscape, and 4) to compare difference in body size between nocturnal and diurnal individuals. After eight weeks of daytime and nighttime surveys during the dry season, a total of 601 individuals were identified across 28 species and 6 families. Of these 601 individuals, 50% were found on aquatic vegetation, 29% were found in the forest during the nighttime, and 21% were found in the forest during the daytime. Beyond describing the community composition and species richness, I also quantified the overall diversity and found a difference of body size between diurnal and nocturnal anurans. The overall diversity of the region seems to be mostly from local alpha diversity, but beta diversity was slightly higher than expected when considering populations on opposite sides of the river. This might indicate that the wide rivers of this region are weak barriers of dispersal for anurans. Furthermore, a Wilcoxon rank-sum test showed with great significance that there is a difference of body size between diurnal and nocturnal anurans in this region. This might support theories that anuran species are driven apart by interspecific competition and intraguild predation, or that only larger species can tolerate the cooler temperatures during the nighttime. In conclusion, it would help future studies to spend a greater length of time in the region, to be performed during the wet season as well as the dry season, and to collect data more relevant to explaining any underlying forces influencing the community.

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## INTRODUCTION

The Pacaya-Samiria National Reserve is part of an equatorial wet broadleaf evergreen rainforest in Peru (Fig. 1). With a size of approximately 20,800km<sup>2</sup>, this region is the second-largest protected area in Peru, and one of the largest in the Amazon River basin. The reserve contains one of the largest protected areas of flooded forest in the Amazon. Pacaya-Samiria was established in the 1940s for the protection of the paiche (a large Osteoglossomorph fish in the genus *Arapaima*). At least 100,000 indigenous people live within the boundaries of the reserve.

This reserve is named for two rivers that flow diagonally through its middle. The northern and southern boundaries of the reserve are bordered by the Marañón and Ucayali Rivers, respectively. These two major rivers merge at the northeastern corner of Pacaya-Samiria to form the headwaters of the Amazon River. The climate in this region is typical of a humid lowland tropical forest; there is a high annual precipitation averaging about 2500 mm, and the mean annual temperature is 24-26°C. The dry season occurs June through August. The wettest months are December through March, and much of the region is flooded during this time.

There are few studies from this reserve that focus on frogs and toads (collectively known as anurans). To fill this knowledge gap, I inventoried the anuran community along riverine habitats in Pacaya-Samiria to describe species composition and richness. Species composition and richness are properties of ecological communities, and they provide a foundation for advancing biological conservation (Bawa et al. 2004, Gotelli and McCabe 2002, Ricklefs 2004). Conducting a compositional inventory of a region is often the first step to protecting it (Tangley 1990). Species richness is a count of the number of species present relative to the number of individuals, and is often presented in the form of an index. In addition to examining species composition and richness, I also explored patterns of beta diversity, or the extent of species replacement or biotic change along an environmental gradient (Wilson

and Shmida 1984). I further explored my data by quantifying the difference between body sizes of anurans found during the daytime and the nighttime.

Here, I analyze a Neotropical anuran assemblage with the following objectives: 1) To conduct an inventory of the local species, 2) to quantify richness among habitats, 3) to compare differences in beta diversity across a river versus a continuous landscape, and 4) to compare difference in body size between nocturnal and diurnal individuals. To do this, I first sampled anurans in two habitat types in the Pacaya-Samiria Reserve. I used these data to describe species composition and richness. I then analyzed beta diversity among sampling plots, and tested the hypothesis that rivers act as dispersal barriers for anurans. Finally, I quantify differences between the body sizes of nocturnal and diurnal anurans, and tested the hypothesis that nocturnal anurans will be larger in size than diurnal anurans.

## METHODS

Data were collected in the Pacaya-Samiria National Reserve. For eight weeks during the dry season (starting on June 17, 2012), our research crew had living quarters aboard a ferry, which was docked adjacent to a security outpost. Two major study sites were accessed by traveling along the Samiria River on the ferry, but transects and remote sites were accessed by motorized dugout canoes that were docked at the outpost.

My research took place in two primary types of habitat: aquatic vegetation and várzea (seasonally flooded forest). The várzea sites were located in untouched, primary forest. Equal numbers of transects were completed on both sides of the river at each site. Most of these transects were freshly cut by native guides, but four had been established earlier in the year. Patches of aquatic vegetation were surveyed along riverbanks in smaller channels of water where the current was slow enough to keep disturbance to a minimum (Fig. 2). These patches of vegetation consisted almost entirely of grasses, free-floating water lettuce (*Pistia* spp.), and water hyacinth (*Eichhornia crassipes*). Patches of



these plants are known to drift downriver when they ever break free from a larger group (Upton, et al. 2014; Fig. 3).



**Figure 1 - A map of Northern South America, where Pacaya-Samiria Reserve is indicated in blue (ArcGIS Pro 2017).**

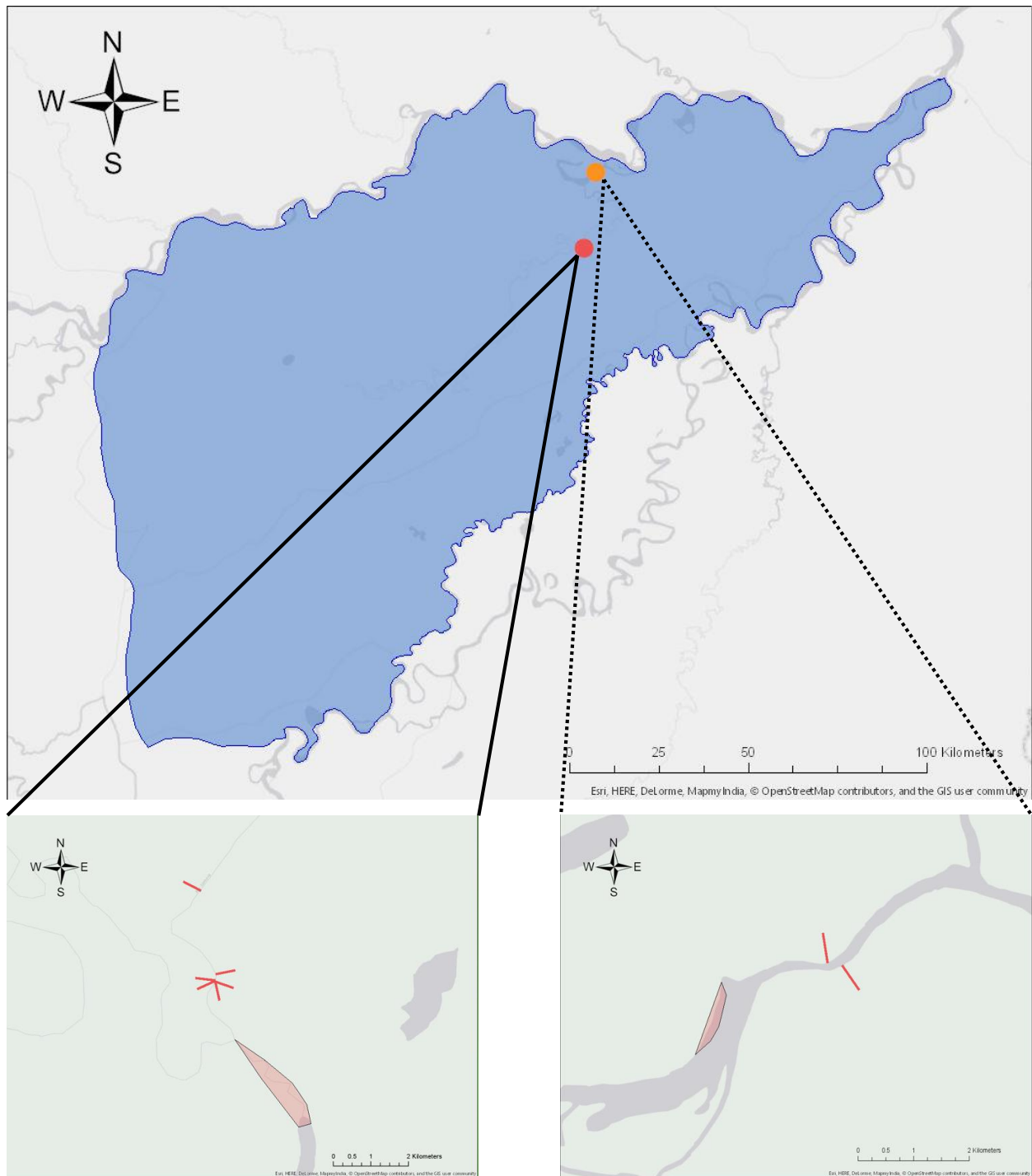


**Figure 2 - Typical patches of aquatic vegetation (AV) along the left side of this channel located at site 1, indicated by the red arrow. These patches consisted almost entirely of grasses, water lettuce, and water hyacinth.**



**Figure 3 – This figure shows a large patch of floating vegetation that was drifting downriver. This might be a suitable raft that helps with dispersal of amphibians.**

Sampling sites were located among two major regions along the Samiria River, which are approximately 21.8 km apart (Fig. 4). The width of the river at site 1 was approximately 91 m, and the width of the river at site 2 was approximately 188 m. Six weeks were spent surveying site 1, with six terrestrial transects and one general zone of aquatic vegetation. Two weeks were spent surveying site 2, with two terrestrial transects and one general zone of aquatic vegetation. Each terrestrial transect had a length of 500 meters and a width of approximately 3 meters, and vegetation patches were visited on a site-by-site basis within each zone. A typical aquatic vegetation patch was roughly 5 x 5 m in size and consisted mainly of grasses (*Poaceae* spp.) and water hyacinth. Surveys were conducted both night and day, during the times when amphibians are most active (Rodriguez and Duellman 1994). Diurnal transects occurred somewhere between 7:00-14:00 and nocturnal transects between 19:00-23:00. There were 72 transects completed (37 daytime, 35 nighttime) and 25 aquatic surveys. The average time required to sample each terrestrial transect and aquatic survey was roughly 1.5 hours. An aquatic survey consisted of fifteen minutes spent at each of four vegetation patches, typically the time that search effort was exhausted and no new specimens could be found. The length of time spent searching terrestrial transects varied slightly, but minimized the possibility of overlooking specimens.



**Figure 4 – A view of Pacaya-Samiria Reserve highlighted then blown up to show both study sites along rivers and nearby lakes (ArcGIS Pro 2017). Six weeks were spent at site 1, which consists of 6 terrestrial transects and 1 general zone of aquatic vegetation, indicated in red. Two weeks were spent at site 2, which consists of 2 terrestrial transects and 1 general zone of aquatic vegetation. The transect layouts are rough estimates of GPS data.**

Several assistants always participated in the surveys. I also had at least one native guide who was experienced with the local forest and wildlife. To keep my data collection consistent, I was the only person who would measure, weigh, and photograph the specimens. During the surveys all accessible microhabitats were searched, both visually and audibly, including leaf litter, tree trunks, tree branches, decayed logs, and fallen palm leaves. Due to the cryptic nature of anurans, disturbance of the vegetation using a probing stick was the most systematic method of detection. The probing sticks were approximately 1.5 m in length and were cut from locally-abundant palm plants. The probing was performed up to at least 1 meter within both sides of the transects. Unlike the diurnal surveys, night transects were performed with headlamps and flashlights without the use of probing sticks. Probing sticks were unnecessary at night because the vast majority of nocturnal anurans are found above the leaf litter, and the few ground-dwelling species are much larger than the diurnal species.

When a specimen was found it was identified then carefully captured and handled for measurement and photographic documentation. It was then promptly released exactly where it was found. Individuals handling a frog were prohibited from using insect repellent, especially those that contain DEET. Gloves were not used when handling specimens because the chytrid fungus (*Batrachochytrium dendrobatidis*) in Peru appears to only be a threat to species at higher elevations in the Andes Mountains (Catenazzi et al. 2010). Chytrid fungus currently grows at higher elevations where temperature and moisture are more ideal for its growth. Furthermore, the species name and time of capture were recorded, as well as the snout-to-vent length (SVL) and file numbers of any photos taken. The SVL was measured to the nearest mm with a set of calipers. When identification of an individual was difficult in the field, photographs and recorded observations were used at a later time with the help of field guides and other resources. Despite efforts, some specimens escaped before they were identified or measured, in which case they have been omitted from this study. To distinguish between species there were four illustrated field guides that were used (Bartlett and Bartlett 2003, Gagliardi-Urrutia

2010, Guerrero et al. 2011, Rodriguez and Duellman 1994). For the purposes of my study, species and family names follow the nomenclature of Gagliardi-Urrutia (2010) and Guerrero et al. (2011). These two sources are the most recent and include up-to-date terms for genus and species names that have recently been changed by the scientific community.

I first divided my samples into three types of habitat: the forest during the daytime (diurnal forest, DF), the forest during the nighttime (nocturnal forest, NF), and the aquatic vegetation along river banks (aquatic vegetation, AV). I then quantified the difference in the anuran community between these habitat types by partitioning species using a Bray-Curtis dissimilarity index (Weinstein 1976, Bloom 1981). Similar to the Sorensen index, the Bray-Curtis dissimilarity equation is meant to show how different sites are from one another, as opposed to how similar. Dissimilarity was calculated using the following equation:

$$D = (\sum |x_{1j} - x_{2j}|) / (\sum(x_{1j} + x_{2j}))$$

where  $x_{ij}$  = frequency of species  $j$  in sample  $i$  (Weinstein 1976).  $D$  ranges between 0 and 1, where 0 means the two sites have the same species composition and relative abundances, and 1 means the two sites do not share any species in common. Since only two sites can be compared simultaneously, I compared all three combinations of habitats in a pairwise manner (i.e. DF vs. NF, DF vs. AV, NF vs. AV).

Next I analyzed the species richness of the region by generating rarefaction curves of each habitat type. Again, this analysis was completed using the species richness measured in diurnal forest, nocturnal forest, and aquatic vegetation habitats. I used EstimateS to create rarefaction curves to show the likely number of species present based on the number of individuals located, with 95% confidence intervals. I set the program to run individual-based analyses in order to show the estimated richness across all sample sites of each habitat (Colwell et al. 2012).

Beta diversity can be quantified as an additive term, and is useful in different ways than computing beta diversity as a multiplicative term (Lande 1996, Veech et al. 2002). In this case, total species diversity ( $\gamma$ ) across a landscape is determined by two things: the mean species diversity at the habitat level ( $\alpha$ ) and the differentiation of species among habitats ( $\beta$ ), where  $\gamma=\alpha+\beta$ . This allows beta diversity to be quantified across spatial and temporal scales whether or not it lacks a direct gradient, or even if it includes diurnal and nocturnal species or the addition of multiple habitat types.

To analyze  $\beta$ -diversity, I created a matrix that contained the number of species found among sampling locations, with sites represented as rows and species counts as columns. I combined both day and night surveys for each terrestrial transect, considering a river segregates populations in space, not in time. I had also combined the three transects on each side of the river at site 1 so they were comparable to the two single transects found at site 2. With four total sites: east and west site 1, east and west site 2, I entered these data into the program PARTITION 3.0 (Veech and Crist 2009) in order to calculate within-sample diversity ( $\alpha$ ) and among-sample diversity ( $\beta$ ) (Appendix C). PARTITION reports the results in the form of additive beta diversity, including the expected and observed averages for both  $\alpha$  and  $\beta$ . In this case,  $\alpha$ -diversity would show the ratio of species found at every site, while  $\beta$ -diversity would show the ratio of species found on a single side of the river.

Finally, I compared the size structure of different habitats using snout-to-vent length (SVL) measurements. Of the 601 anurans included in my study, only 499 were successfully measured after capture. To compare the difference of body size between anurans, I used the Wilcoxon rank-sum test to quantify the difference in SVL between habitats. I only used the samples from DF and NF because these two populations occupy the same space over time. The Wilcoxon rank-sum test is a non-parametric test for two independent samples and does not assume a normal distribution.

## RESULTS

I located a total of 685 individual anurans, 84 of which had escaped or were indistinguishable and were omitted from this study. The remaining 601 specimens represent a total of 28 species and 6 families (Appendix A). Each family and the percent contribution to the total are as follows: 64% Hylidae, 14% Leptodactylidae, 7% Microhylidae, 7% Strabomantidae, 4% Bufonidae, 4% Pipidae. Of the total individuals found, 21% were in diurnal forest, 29% in nocturnal forest and 50% in aquatic vegetation (Appendix A, B). Overall, 21% were diurnal and 79% nocturnal, since nearly all species found in aquatic vegetation were nocturnal (96.2%). The few specimens found in aquatic vegetation during the daytime all seemed to be inactive and hiding. On average we encountered 5.5 frogs per hour, 4.2 per terrestrial transect and 12 per aquatic survey. Almost all diurnal forest specimens were ground-dwellers (93%) and the majority of nocturnal forest specimens were arboreal (68%). The two most common species of each habitat type can be found in Table 1.

**Table 1 – The two most common species found in each habitat type, number of individuals encountered (n), and the percentage of individuals in that habitat represented by that species (%).**

Habitat	Species	n	%
<b>Floating Vegetation</b>	<i>Hypsiboas punctatus</i>	116	38.9
	<i>Dendropsophus triangulum</i>	98	32.7
<b>Diurnal Forest</b>	<i>Scarthyla goinorum</i>	119	92.2
	<i>Scinax pedromedinae</i>	6	3.1
<b>Nocturnal Forest</b>	<i>Osteocephalus taurinus</i>	54	31.4
	<i>Osteocephalus lepreurii</i>	20	11.6

Community composition differed among habitat types. There was little overlap in composition and abundance between DF and NF habitats (Bray-Curtis dissimilarity= 0.92), DF and AV habitats (0.98), and NF and AV habitats (0.97). Species richness differed among habitat type, which increases in order of diurnal forest (5 species, 129 individuals), aquatic vegetation (12 species, 300 individuals), and nocturnal forest (22 species, 172 individuals; Figure 5).



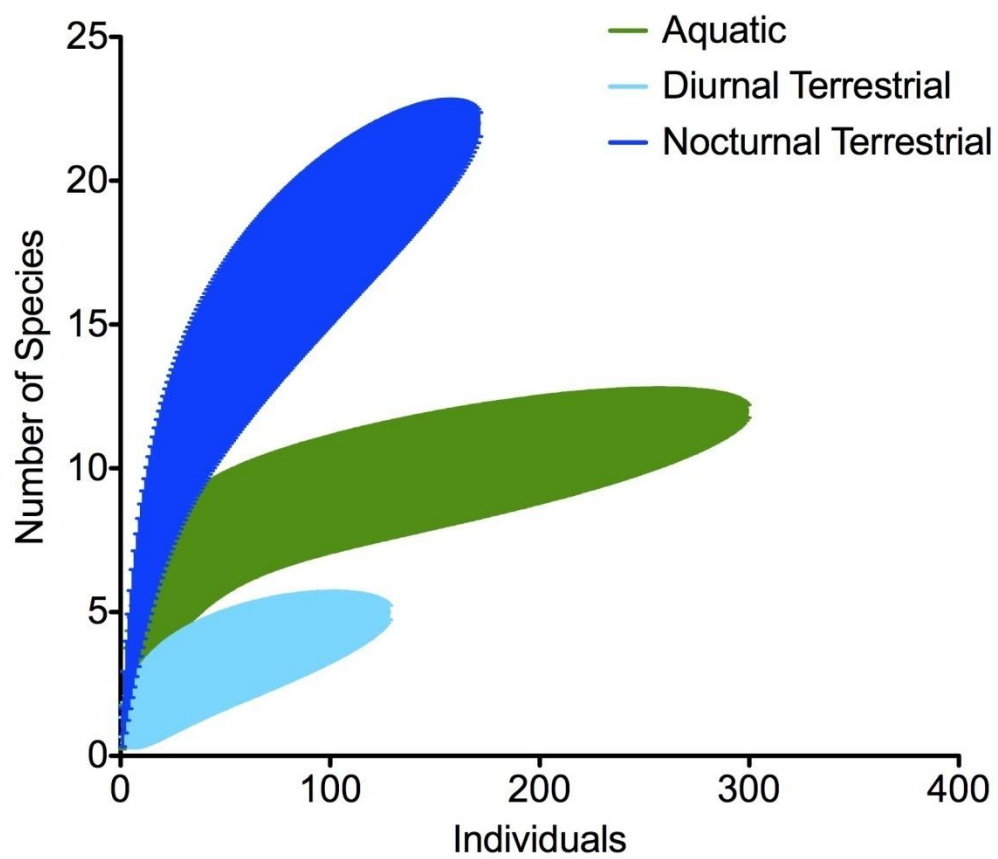


Figure 5 – These rarefaction curves depict the number of species found among the number of individuals located, and 95% confidence intervals for each habitat type.

Additive partitioning of diversity revealed that on opposite sides of the river,  $\beta$ -diversity was significantly higher (28%) than expected (23%) based on a null model ( $p = 0.003$ ; Figure 6). Considering the observed  $\alpha$ -diversity, it is expected that 70% or more of my species count would be found at every site included in the analysis. Less than 30% of my species would be found on a single side of the river, although this could be due to the lack of sampling as seen in Fig. 5.

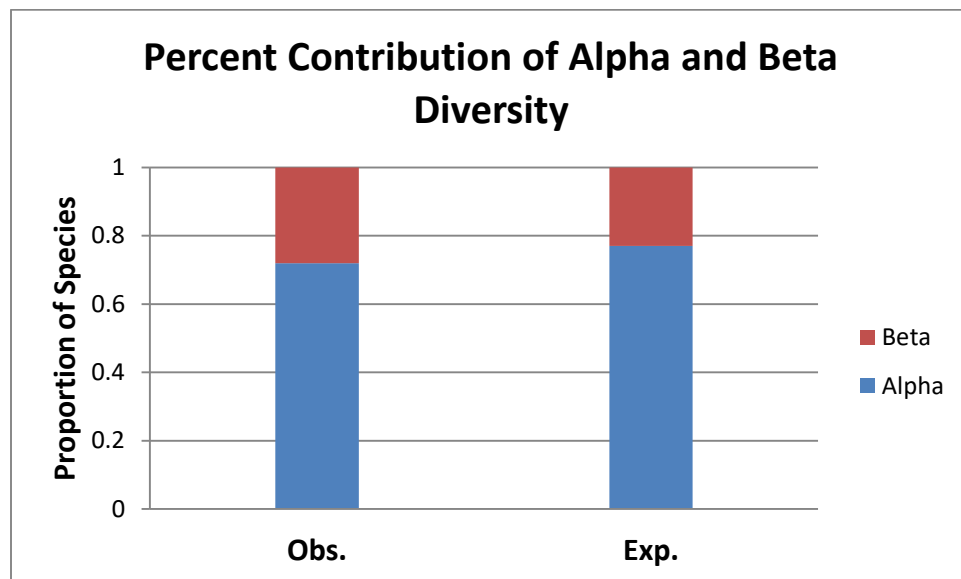


Figure 6 - Results from PARTITION indicate no significant difference in  $\alpha$ -diversity ( $p = 0.967$ ) and a significant difference in  $\beta$ -diversity ( $p = 0.003$ ). This chart excludes anurans in aquatic vegetation, and includes a total of 23 species across only the terrestrial transects on East and West sides of the Samiria River at both study sites.

Species in the nocturnal forest habitat had by far the largest average SVL ( $n = 158$ ; 57.4 mm). Floating vegetation had the second largest average SVL ( $n = 223$ ; 26.6 mm). Diurnal forest had the lowest mean SVL ( $n = 118$ ; 19.0 mm). Nocturnal species had greater SVL than diurnal species in the same location (Wilcoxon rank-sum test;  $z = -11.7$ ;  $p < 0.001$ ).

## DISCUSSION

I identified 601 frogs across 28 species in eight weeks of surveys. This accounts for 19% of anuran species that are known to occur in the more completely surveyed Manu National Park in southern Peru (Catenazzi et al. 2013). This region harbors some of the greatest anuran diversity in the world, but it also contains many unique types of habitat. Catenazzi et al. (2013) compiled all known amphibians across 10 different habitat types in Manu National Park, but my study only includes 2 out of the 10 habitats they surveyed (várzea and aquatic vegetation). There were extensive surveys also done by Upton et al. (2014) in the Pacaya-Samiria National Reserve that took place only in aquatic vegetation. For this study I had found 63% of the species they included, but their surveys were spread across 8 months of the year compared to my 2 months. Upton et al.'s (2014) inventory included all of aquatic vegetation anurans I found.

Rarefaction analysis depicts both the overall richness of a habitat and the likelihood that not all species were observed (Colwell et al. 2012). Two of my species accumulation curves (DF and AV) are nearing a plateau, but they may not yet have reached an asymptotic phase (Pineda and Halffter 2003). Species sampled from these habitats are therefore probably representative. However, the curve for the NF habitat is far from asymptotic, and indicates that a greater sampling effort is needed to adequately characterize the anurans in this habitat. For example, my study only includes 24% of the species estimated to be in the region near the city Iquitos, as well as in the entire Loreto region (Rodriguez and Duellman 1994, Gagliardi-Urrutia 2010). Also, my surveys did not include at least 8 other habitat types that could be sampled in the lowland rainforests of Peru (Catenazzi 2013). To my knowledge, my survey yielded no species new to science.

I predicted beta diversity of anurans would be structured in part by rivers acting as dispersal barriers. Diversity partitioning revealed  $\alpha$ -diversity has the greatest contribution to  $\gamma$ -diversity. However,

the  $\beta$ -diversity was significantly higher than expected despite its minor role in structuring overall diversity. This suggests that the diversity of the anuran assemblage is weakly structured regionally at the scale of this analysis, suggesting rivers might act as a weak dispersal barrier. Studies of diversity partitioning often include many more sites than used in this study, so the results should be interpreted with caution.

Beta diversity of taxa typically increases as larger areas are sampled even of a homogenous region (Caley and Schluter 1997, Dahl et al. 2009, Garcia et al. 2007). This is due to the sampling effects and the species-area effect in tropical rainforests, as spatial subsets of the data become increasingly distinct as the species pool increases. A study done by Dahl et al. (2009) compared the beta diversity of frogs in three distant regions of the world: New Guinea, Amazonia, and Europe. They hypothesized that the relatively uniform lowland forests would be characterized by low beta diversity regardless of the region. The region of Amazonia included many types of habitat (terra firma, flooded forest, swamps, river shores, etc.). They found a total of 70 species in the Amazon, 44 in New Guinea, and 12 in Europe. Beta diversity in Amazonia was high, with an average of 36 of 70 species being found at single sites. Beta diversity was highest in Amazonia, intermediate in New Guinea, and lowest in Europe. Gascon (1996) hypothesized that the Juruá River in lowland Brazil is a geographic barrier for amphibian litter fauna. He quantified species richness, abundance, and Jaccard similarity, and concluded that the river was not a significant barrier for dispersal. A later study done by Gascon et al. (2000) tested the same hypothesis with frogs and small mammals. They measured species richness on opposing river banks and genetic samples. They found no major difference between populations on opposite sides of the Juruá River, suggesting that this wide river was not an effective barrier of dispersal.

There were significant differences of body size among species in the different habitat types. Diurnal and nocturnal terrestrial species live within the same space, but are not active at the same time.

Considering their body sizes are so drastically different, it might indicate some mechanism is selecting for this difference (Reid et al. unpublished manuscript). Body size differences among frog species reduces competition for food (Pfennig and Murphy 2003, Vignoli et al. 2009). However, no published research I am aware of compares the difference of body size between diurnal and nocturnal anurans, so this may be a novel pattern. Studies in other regions could help determine if differences in body size between diurnal and nocturnal anurans form a general pattern. I did not conduct any experiments to test mechanisms that could cause a diel divergence in body size.

In the case of anurans, specifically, a difference of body size based on diel activity could imply not only interspecific competition, but intraguild predation. Many anurans are opportunistic feeders that readily hunt other anurans (Toft 1985). It is also thought that diurnal and nocturnal species may prefer different types of prey, whether insectivores or carnivores. Depending on the prey available during the time of day, a larger body size may be necessary (Reid et al. unpublished manuscript).

There are certain limitations of my study. Major limitations include: a small number of sampling locations, a lack of distance between sampling locations, and a limited sampling duration of only eight weeks. For a proper inventory of the anuran community in the entire Pacaya-Samiria Reserve, future research will need to include a greater number of transects in sampling regions, and include sampling locations along a variety of rivers. Considering the enormous size of the Pacaya-Samiria reserve, more spatially-extensive and less time-sensitive surveys should be conducted. Also, weather and other seasonal factors should be measured considering their influence on anuran detectability (Gascon et al. 2000). Most importantly, research should also focus on sampling anurans at different times of the year, both when the rivers are flooded and when they are not. Considering the seasonally flooded riparian zones along rivers such as the Samiria, habitat type would need to be defined for each sampling location in order to standardize the comparisons of species composition across the river (Gascon 1996). Finally, I also suggest that future researchers wear gloves or take similar precautions when handling anurans to

help prevent the spread of chytrid fungus into lowland areas with the progression of global climate change (Catenazzi et al. 2010).

In conclusion, I identified 28 species of anurans in the várzea and riverine aquatic vegetation of Pacaya-Samiria Reserve. I found very little overlap in species composition among várzea and aquatic vegetation, and within várzea when sampled during the day and night. Greater sampling effort is needed to characterize the nocturnal composition of várzea habitats. The Samiria River might be a barrier limiting dispersal of some anuran species. Significant size differences between diurnal and nocturnal anurans within várzea could be driven by an ecological or evolutionary mechanism, but it is unknown whether this size difference is unique to this study or is a more general pattern.

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## APPENDIX A

Total individuals of species found in each habitat type, where DF = diurnal forest, NF = nocturnal forest and AV = aquatic vegetation. Including the 601 individuals listed here, a total of 685 specimens were located. The remaining 84 had escaped prior to identification or were not found during an official survey, and thus were eliminated from my study.

Species	Family	DF	NF	AV	Total
<i>Rhinella marina (Bufo marinus)</i>	Bufonidae	0	1	4	5
<i>Dendropsophus brevifrons</i>	Hylidae	0	2	0	2
<i>Dendropsophus haraldschultzi</i>	Hylidae	0	1	25	26
<i>Dendropsophus parviceps</i>	Hylidae	2	5	0	7
<i>Dendropsophus triangulum</i>	Hylidae	0	1	98	99
<i>Hypsiboas geographicus</i>	Hylidae	0	13	0	13
<i>Hypsiboas lanciformis</i>	Hylidae	0	0	18	18
<i>Hypsiboas punctatus</i>	Hylidae	0	0	116	116
<i>Osteocephalus deridens</i>	Hylidae	0	4	0	4
<i>Osteocephalus leprieurii</i>	Hylidae	0	20	0	20
<i>Osteocephalus planiceps</i>	Hylidae	0	13	0	13
<i>Osteocephalus taurinus</i>	Hylidae	0	54	0	54
<i>Scarthyla goinorum</i>	Hylidae	119	13	1	133
<i>Scinax cruentommus</i>	Hylidae	0	3	0	3
<i>Scinax Garbei</i>	Hylidae	1	0	11	12
<i>Scinax pedromedinae</i>	Hylidae	6	1	0	7
<i>Sphaenorhynchus carneus</i>	Hylidae	0	0	15	15
<i>Sphaenorhynchus dorisae</i>	Hylidae	0	0	1	1
<i>Sphaenorhynchus lacteus</i>	Hylidae	0	1	9	10
<i>Leptodactylus discodactylus</i>	Leptodactylidae	1	1	0	2
<i>Leptodactylus leptodactyloides</i>	Leptodactylidae	0	13	1	14
<i>Leptodactylus pentadactylus</i>	Leptodactylidae	0	13	0	13
<i>Leptodactylus petersii</i>	Leptodactylidae	0	2	0	2
<i>Chiasmocleis ventrimaculata</i>	Microhylidae	0	1	0	1
<i>Ctenophryne geayi</i>	Microhylidae	0	1	0	1
<i>Pipa pipa</i>	Pipidae	0	0	1	1
<i>Pristimantis altamazonicus</i>	Strabomantidae	0	7	0	7
<i>Pristimantis delius</i>	Strabomantidae	0	2	0	2
<b>Total (28 species)</b>	<b>6 families</b>	<b>129</b>	<b>172</b>	<b>300</b>	<b>601</b>

## APPENDIX B

Reference photographs for the identification of each species.

For the purposes of my study, species and family names follow the nomenclature of

Gagliardi-Urrutia (2010) and Guerrero et al. (2011).

All photos were taken by Eric Woebbe unless otherwise indicated:

<sup>1</sup>Gagliardi-Urrutia (2010), <sup>2</sup>Guerrero et al. (2011), <sup>3</sup>Bartlett and Bartlett (2003)



**1. *Rhinella marina***  
**(*Bufo marinus*)<sup>2</sup>**  
Bufonidae



**2. *Dendropsophus brevifrons*<sup>1</sup>**  
Hylidae



**3. *Dendropsophus haraldschultzi*<sup>1</sup>**  
Hylidae



**4. *Dendropsophus parviceps***  
Hylidae



**5. *Dendropsophus triangulum*<sup>1</sup>**  
Hylidae



**6. *Hypsiboas geographicus*<sup>1</sup>**  
Hylidae



**7. *Hypsiboas lanciformis*<sup>1</sup>**  
Hylidae



**8. *Hypsiboas punctatus***  
Hylidae



**9. *Osteocephalus deridens*<sup>1</sup>**  
Hylidae



**10. *Osteocephalus leprieurii*<sup>3</sup>**  
Hylidae



**11. *Osteocephalus planiceps***  
Hylidae



**12. *Osteocephalus taurinus***  
Hylidae



**13. *Scarthyla goinorum***  
Hylidae



**14. *Scinax cruentommus***  
Hylidae



**15. *Scinax Garbei***  
Hylidae



**16. *Scinax pedromedinae***  
Hylidae



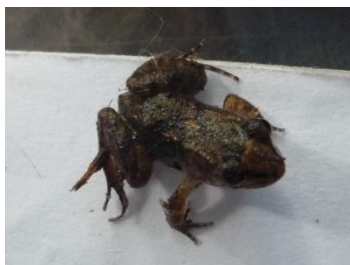
**17. *Sphaenorhynchus carneus*<sup>1</sup>**  
Hylidae



**18. *Sphaenorhynchus dorisae*<sup>1</sup>**  
Hylidae



**19. *Sphaenorhynchus lacteus*<sup>1</sup>**  
Hylidae



**20. *Leptodactylus discodactylus***  
Leptodactylidae



**21. *Leptodactylus leptodactyloides*<sup>1</sup>**  
Leptodactylidae



**22. *Leptodactylus pentadactylus***  
Leptodactylidae



**23. *Leptodactylus petersii*<sup>1</sup>**  
Leptodactylidae



**24. *Chiasmocleis ventrimaculata***  
Microhylidae





**25. *Ctenophryne geayi***  
Microhylidae



**26. *Pipa pipa***  
Pipidae



**27. *Pristimantis altamazonicus*<sup>1</sup>**  
Strabomantidae



**28. *Pristimantis delius*<sup>1</sup>**  
Strabomantidae

## **Appendix C**

### Software

#### **EstimateS**

Version 9

#### **PARTITION**

Version 3.0

##### **River Barriers Hypothesis**

- Number of Levels of Data: 1
- Lowest Level of Analysis: 1
- Number of Samples at Each Level: 1 = 2
- Sampling Design: Balanced
- Randomization Method: Individual-based
- Sample weighting: No
- Q-diversity metrics: None
- Randomizations: 10000

#### **ArcGIS Pro**

Version 1.4.0

##### **Site Maps**

- Light Gray Canvas Basemap