Introduction

Mona Island is a Natural Reserve under the management of the Department of Natural Environment Resources (DNER). Mona Island iguana (*Cyclura cornuta stejnegeri*), is an endemic reptile species of this region which is considered endangered. A headstarting program has been implemented since 1999 by the DNER for Mona Island iguana.

As part of the evaluation of this conservation strategy, several animals were tracked after release using radiotelemetry, in order to determine movement and dispersal range. Two release sites were used: Corral Wiewandt (CW, highly intervened by humans) and Carabinero (CAR, better preserved). Each iguana was followed for a certain period of time, and observations were not equally spaced. For each observation, UTM coordinates were obtained.

The objective of this study is determine whether behavior of releases iguanas differ between the two release sites.

### TABLE 1: Morphometrics, sex, release date, release site Corral Wiewandt (CW) and Carabinero (CAR), number of recapture locations (n)

<table>
<thead>
<tr>
<th>Iguanas</th>
<th>Sex</th>
<th>Release date</th>
<th>Release site</th>
<th>Release BM (kg)</th>
<th>N</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>F</td>
<td>Apr – 02</td>
<td>CW</td>
<td>1.12</td>
<td>69</td>
</tr>
<tr>
<td>2</td>
<td>M</td>
<td>Apr – 02</td>
<td>CW</td>
<td>1.05</td>
<td>102</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>Apr – 02</td>
<td>CW</td>
<td>1.04</td>
<td>43</td>
</tr>
<tr>
<td>4</td>
<td>M</td>
<td>Apr – 02</td>
<td>CW</td>
<td>1.06</td>
<td>96</td>
</tr>
<tr>
<td>5</td>
<td>F</td>
<td>Apr – 02</td>
<td>CW</td>
<td>0.95</td>
<td>4</td>
</tr>
<tr>
<td>6</td>
<td>M</td>
<td>Aug – 02</td>
<td>CAR</td>
<td>1.12</td>
<td>33</td>
</tr>
<tr>
<td>7</td>
<td>F</td>
<td>Aug – 02</td>
<td>CAR</td>
<td>1.05</td>
<td>17</td>
</tr>
<tr>
<td>8</td>
<td>M</td>
<td>Aug – 02</td>
<td>CAR</td>
<td>1.04</td>
<td>13</td>
</tr>
<tr>
<td>9</td>
<td>M</td>
<td>Aug – 02</td>
<td>CAR</td>
<td>1.01</td>
<td>41</td>
</tr>
</tbody>
</table>

Based on expert opinion, an informative U(0,10) prior was selected for $\alpha_y$. Vague normal priors were assigned to the coefficients of the log-linear models.

![FIGURE 1: Trajectories for the headstarted Mona Island iguanas](image)

For modeling trajectories, we will suppose that north-south movement and east-west movement are independent and have the same variability.

### Results and Discussion

#### TABLE 2: Effective sample size and DIC values for models fitted

<table>
<thead>
<tr>
<th>Model</th>
<th>DIC</th>
<th>DIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Equal variance</td>
<td>520.840</td>
<td>7543.330</td>
</tr>
<tr>
<td>Sex</td>
<td>569.619</td>
<td>6076.720</td>
</tr>
<tr>
<td>Body Mass</td>
<td>569.926</td>
<td>5193.560</td>
</tr>
<tr>
<td>Release Place</td>
<td>602.846</td>
<td>6056.880</td>
</tr>
<tr>
<td>Sex × Body Mass</td>
<td>583.806</td>
<td>5209.010</td>
</tr>
<tr>
<td>Sex × Release Place</td>
<td>609.044</td>
<td>5798.740</td>
</tr>
<tr>
<td>Body Mass + Release Place</td>
<td>573.619</td>
<td>5066.340</td>
</tr>
<tr>
<td>BM + RP + Sex</td>
<td>653.342</td>
<td>6101.700</td>
</tr>
</tbody>
</table>

The DIC value for the model including only sex is substantially lower than DIC values for all models. According to this, no evidence of differences between release sites or influence of the body mass was found. It can be supposed that the same effort is required for getting food and resources in Carabinero and Corral Wiewandt, and no modifications seem to be required in release strategy.

### Reference


**BOA software (Smith 2007)** was used for convergence diagnostics. DIC criterion (Spiegelhalter et al 2002) was used for model selection.

### TABLE 3: Posterior quantities for variances, model including sex.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Sex</th>
<th>Mean</th>
<th>Sd</th>
<th>2.5%</th>
<th>97.5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\alpha_y$ (F)</td>
<td>F</td>
<td>23.96</td>
<td>1.339</td>
<td>21.47</td>
<td>26.72</td>
</tr>
<tr>
<td>$\alpha_y$ (M)</td>
<td>M</td>
<td>33.6</td>
<td>0.9584</td>
<td>31.77</td>
<td>35.52</td>
</tr>
<tr>
<td>$\sigma_y$</td>
<td></td>
<td>2.841</td>
<td>2.009</td>
<td>0.1106</td>
<td>7.385</td>
</tr>
</tbody>
</table>

Posterior quantities for the selected model show that males move longer distances than females. The precision of position measurement is consistent with the values expected by experts (2-3 mts).

**Funded by National Science Foundation, HRD #0734826, PR-LSAMP Program HDR #0601843 and University of Puerto Rico, Central Administration and Río Piedras Campus**
Dormancy in Caladenia: A Bayesian approach to evaluating latency

Dormancy is common in many terrestrial orchids in southern Australia and other temperate environments. The difficulty for conservation and management when considering dormancy is ascertaining whether non-emergent plants are dormant or dead. Here we use a multi-state capture–recapture method, undertaken over several seasons, to determine the likelihood of a plant becoming dormant or dying following its annual emergent period and evaluate the frequency of the length of dormancy. We assess the transition probabilities from time series of varying lengths for the following nine terrestrial orchids in the genus Caladenia: C. amoena, C. argocalla, C. clavigera, C. elegans, C. granitica, C. macroclavia, C. oenochila, C. rosella and C. valida from Victoria, South Australia and Western Australia. We used a Bayesian approach for estimating survivorship, dormancy and the likelihood of death from capture–recapture data. Considering all species together, the probability of surviving from one year to the next was ~86%, whereas the likelihood of observing an individual above ground in two consecutive years was ~74%. All species showed dormancy of predominantly 1 year, whereas dormancy of three or more years was extremely rare (<2%). The results have practical implications for conservation, in that (1) population sizes of Caladenia species are more easily estimated by being able to distinguish the likelihood of an unseen individual being dormant or dead, (2) population dynamics of individuals can be evaluated by using a 1–3-year prediction vs. reality: Can a PVA model predict population persistence 13 years later? The challenge of conservation biology is to make models that predict population dynamics and have a high probability of accurately tracking population change (increase, decrease, constancy). In this study we modeled 6 small populations of an epiphytic orchid using a Leefkovich type analysis to predict population growth pattern based on monthly surveys for approximately 1.5 years. In addition, sensitivity and elasticity analyses were used to identify life stages with high sensitivity or elasticity that have the largest influence on population growth rate. We re-censused the populations 13 years after the first study and compared the structure of the populations to predictions based on the earlier census data. One objective was to determine if populations had achieved a stable size distribution over the 13 years period. Population growth rate models suggested that all populations should have persisted. Effective population growth rates were similar to those expected except for one where the population went extinct. The prediction slightly (but not significantly) overestimated the actual population growth rates of some populations. Elasticity analysis revealed that the adult stage is critical in the life cycle. The observed stage distributions of the populations were not stable at the beginning of the survey and neither were they after 13 years. We suggest that this might be caused by external perturbations that result in unequal mortality between life stages and stochastic recruitment events. The ability of the matrices to predict population size approximately eight generations in the future is encouraging and warrants the continued use of these approaches for PVA.

Circular distribution of an epiphytic herb on trees in a subtropical rain forest.

The distribution of the endangered epiphytic orchid Lepanthes eltoroensis on the bole of trees was investigated in the Yunque National Forest, Puerto Rico. Using circular statistics, we evaluated if there was preference for cardinal position on the bole of trees along two trails (Tradewinds and El Toro). In addition we tested if larger trees had larger population, and if the distribution of the orchids varied between the two walking trails. Orchids were preferentially distributed on the northwestern side of the bole of trees. Moreover, we found no evidence that the size of trees affected the number of individual orchids. This survey suggests that there is a preference for specific cardinal position on trees for the orchid and thus relocation or establishment of new populations should consider this information to maximize survivorship of this rare orchid.

2008-2009 Publications only


Pavel Kindlmann, Elvia J Meléndez-Ackerman & RL Tremblay (Submitted 06-04-2009). Disobedient orchids: colonization and extinction rates in orchid metapopulations contradict theoretical predictions based on patch connectivity. Ecography


Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus

Raymond L. Tremblay, Dept. of Biology University of Puerto Rico Humacao/Río Piedras Campuses / raymond@hpcf.upr.edu

Examples from recent publications

Circular frequency distribution of orchids on trees

Lepanthes rubripetala

<table>
<thead>
<tr>
<th>Direction</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>North</td>
<td>358</td>
</tr>
<tr>
<td>East</td>
<td>341</td>
</tr>
<tr>
<td>South</td>
<td>418</td>
</tr>
<tr>
<td>West</td>
<td>0</td>
</tr>
</tbody>
</table>

Contributions of Sewage Effluents to Nutrients, Discharge, and DOC in Streams in Puerto Rico

INTRODUCTION

Some waste water treatment plants (WWTP) in Puerto Rico continuously discharge their effluents directly to streams. With urbanization, generation of sewage effluents increases and an increase of nutrients is expected to increase above background levels. Excess nitrogen and phosphorus are of particular concern because eutrophication of inland reservoirs is limited by phosphorus inputs, and coral reefs are damaged by excessive nitrogen inputs.

Effluent discharges are regulated based on organic load contributed to the receiving water body. Stream communities rely on organic matter and the type of organic matter in the stream can be altered by changes in the landscape or human activities.

Economic development on the heavily populated island of Puerto Rico has resulted in increasing wastewater generation. Therefore, it is imperative to document the impacts of WWTP effluents on stream quality in order to devise better decision-making tools for stream management and restoration.

Research objective: Our goal was to examine the contribution of WWTP to stream nutrients, discharge and dissolved organic carbon (DOC).

METHODS

Four WWTP were sampled to determine the relative contributions to stream nutrients, river discharge, and river DOC concentrations during base flow conditions during 2007-2008. Two of these four WWTP were sampled intensively and the other two were used as synoptic sites. Three stations were established at each site: upstream from the sewage effluent, at the WWTP effluent, and downstream from the effluent. At each station, water samples were collected to analyze the effluent contribution to stream phosphate, nitrate, ammonium, and DOC flux. Export of nutrients for the upstream and effluent station was calculated as the product of nutrient concentration and instantaneous stream flow and summed to obtain the downstream daily export. Analysis of the biodegradability of organic matter (BOD) and the specific UV absorbance (SUVA) were conducted to observe any contribution from the sewage effluent to the quality of organic matter in the stream.

RESULTS

Nitrates, phosphates, ammonium and DOC effluent concentrations highly increased background concentrations with a median of 5.0 mg L⁻¹, 0.4 mg L⁻¹, 0.04 mg L⁻¹, and 4.4 mg L⁻¹, respectively (Figure 1). At the downstream station nutrient concentrations were lower than the sewage effluent concentrations but higher than background levels indicating the change in stream water chemistry by the WWTP.

Incubations for BOD samples showed higher oxygen consumption by bacteria on the effluent in all sites compared to those samples collected in the upstream station (Figure 2a). Mean SUVA values were elevated at the upstream station indicating that the DOC in the stream contains high percentage of aromaticity (refractory organic matter) (Figure 2b). Low SUVA values were observed in the sewage effluent suggesting labile organic matter. According to our BOD and SUVA findings, sewage effluents are contributing labile organic matter to the stream changing the type of organic matter in downstream communities.

Seaweed effluents contributed a substantial amount of the daily downstream export of phosphate and nitrate at the streams with loads up to ten times higher than before the effluent (Figure 3a and b). The daily downstream flux of ammonium and DOC at Río Fajardo was mainly contributed by the stream, however, at Río Bairoa the sewage effluent contributed all the ammonium and DOC (data not shown). The reason for this is due to the differences in discharge between the effluent and stream at both sites. Most of the discharge at Río Fajardo is contributed from the stream (61 to 94%) but the sewage effluent at Río Bairoa contributes about ten times higher than before the effluent. (Figure 3a and b). The daily discharge at both sites is shown in Figure 3c.

ACKNOWLEDGMENTS

Thanks to Víctor Figueroa, José J. Carrion, Rafael Benitez, Marlene Aquino, Jody Potter and Maylen Pérez for their help with field work. We thank Jeff Merriam and Jody Potter from the University of New Hampshire for analyzing water chemistry samples. Thanks to the Limnological Laboratory and the Institute for Tropical Ecosystem Studies at the University of Puerto Rico (UPR), Rio Piedras Campus for laboratory and field equipment. Special thanks to Dr. Gustavo Martinez laboratory group at the Agricultural Experimental Station at the Botanical Gardens UPR Mayaguez Campus for their assistant in the field and laboratory.

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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
ANTIBIOTIC RESISTANCE GENES IN FERAL GOATS OF MONA ISLAND

ABSTRACT

Background: Domesticated animals are exposed to antibiotics used in veterinary medicine or as feed additives. Over 4 centuries ago, goats were brought by the Spaniards to the Caribbean island of Mona, and have lived since then with very low human impact. The aim of this work was to determine the presence of antibiotic resistance genes in fecal bacteria from feral goats from Mona Island.

Methods: We sampled feces from 5 Mona Island feral goats, and from 5 domestic goats from a farm in mainland Puerto Rico. Fecal DNA was isolated using the MoBio Powersoil kit. PCR was used to amplify tetracycline resistance genes tet(M), tet(O), tet(Q), tet(S) and tet(W).

Results: All feral and domestic goats harbored intestinal bacteria with tet(O) and tet(W) and none had tet(M) or tet(S). Domestic goats harbored in addition tet(Q).

Conclusion: These results confirm that even animals with no or very low exposure to antibiotics, harbor intestinal bacteria with antibiotic resistance genes. In goats, domestication practices increase the number of tetracycline antibiotic resistance genes in the intestinal bacteria.

INTRODUCTION

- Antibiotics are used in veterinary medicine and as feed additives (Aminov et al. 2001).
- Wild and feral animals have had less or no exposure to antibiotics
- The Spaniards brought goats to the Caribbean island of Mona (Figs 1 and 2) 400 years ago, and have lived since then with very low human impact (Ponenberg, 1992).
- Tetracycline was discovered in the 1950's (Roberts 2005). It is a broad spectrum and common antibiotic.
- The three described mechanisms of tetracycline are ribosome protection, efflux pumps and enzymatic inactivation of tetracycline (Roberts 2005).
- tet(M), tet(O) and tet(W) are examples of tetracycline resistance genes that have been isolated from the mouth and fecal matter of diverse animals (Gueimonde et al. 2006).
- The aim of this work was to determine the presence of antibiotic resistance genes in fecal bacteria from feral goats from Mona Island.

METHODOLOGICAL OVERVIEW

DNA Extraction using the MoBio Power Soil Kit

PCR Amplification

Bacterial 16S rDNA Amplification

tetM, tetO, tetQ, tetS and tetW amplification

Agarose Gel Electrophoresis

RESULTS

<table>
<thead>
<tr>
<th>Sample</th>
<th>tet(M)</th>
<th>tet(O)</th>
<th>tet(Q)</th>
<th>tet(S)</th>
<th>tet(W)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feral Goats</td>
<td>0</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>Domestic</td>
<td>0</td>
<td>5</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

- Intestinal bacteria in feral animals had tet(O) and tet(W) resistance genes
- Domestic animals harbor in addition tet(Q), absent in feral animals

CONCLUSION

- Animals with very low or no exposure to antibiotics, such as feral goats, already harbor intestinal bacteria with antibiotic resistance genes.
- Domestication practices increase the number of tetracycline antibiotic resistance genes in goat intestinal bacteria.

FUTURE WORK

- Compare the structure of the intestinal bacterial community of feral vs domestic goats, using using 16S rDNA Microarray technology (G2-Phylomips)

REFERENCES


Michelle JOVANNE RIVERA-RIVERA and Maria Gloria DOMINGUEZ-BELLO
Department of Biology, University of Puerto Rico, Río Piedras, P.R.
Star coral (Montastraea annularis spp. complex) population collapse: An unequivocal sign of climate change impacts in Caribbean coral reefs

Abstract

A catastrophic warming event occurred during 2005 throughout the northeastern Caribbean Sea that caused a mass coral bleaching event in Puerto Rico that was followed by an unprecedented mass mortality of the Montastraea annularis species complex, the most significant reef-building coral in the Atlantic. It resulted in a severe net physiological fragmentation of large coral colonies. Permanent photo-stations were established in 4-6 m deep reef terraces dominated by M. annularis. at four sites in Culebra Island, Puerto Rico. Digital photography was used to document changes in benthic community structure before (2005) and after (2007-2009) this event.

Mass coral mortality caused a 70 to 99% decline in % living tissue cover. There was a significant difference in % living tissue cover loss through time, among sites, particularly at a control site outside a no-take MPA. Abundant physiological tissue fragments were formed in each colony, typically ranging from just below 1 to 105 cm², but mostly in mean sizes below 10 cm², which showed higher mortality trends. There was no significant difference in mean fragment size distribution among sites. Fragment density was significantly higher (p < 0.0001) at Cayo Luis Peña (97/m²) in comparison to other sites (24-67/m²). Fragment density declined from 15 to 31% between 2007 and 2009.

There are not known precedents through the Caribbean of catastrophic events of this magnitude. The synergistic consequences of climate change and variable local anthropogenic impacts in major reef engineer taxa still remain largely unknown. However, a single warming event was unequivocally capable of causing an acute coral mortality event that resulted in a major ecological collapse.

Methods

Data collection and analysis: From left to right: (A) M. annularis; (B) A 2.5 x 2 m permanent photo-station quadrant grid established in triplicate locations within each study site; (C) Detail of partial colony mortality in M. annularis within one subquadrat.

Study sites:
- Luis Peña Channel No-Take Natural Reserve
- PFL= Península Flamenco
- PCR= Playa Carlos Rosario
- CLP= Cayo Luis Peña
- Control site
- PSO= Punta Soldado

Results


From top left to right: (A-B) Bleached colonies of M. annularis (2005); (C) Partial colony mortality in partially bleached colonies (2006); (D) Post-bleaching fragmentation of colonies (2007); (E) Development of algal turfs on partially killed colonies; (F) Mortality of colony fragments (2009).

PERMANOVA % tissue loss

![Table showing PERMANOVA results](image)

Mean fragment abundance per size category.

Conclusions

There was a dramatic coral mortality event after prolonged massive coral bleaching following unprecedented sea surface warming (31.8°C, 14.3 Degree Heating Weeks). There are not known precedents through the Caribbean of events of this magnitude. This will have deleterious long-term ecological implications for coral reef construction and resilience under increasing sea surface warming and ocean acidification trends. Coupled with major recurrent failures in sexual reproduction in the near future, this may permanently compromise coral reef ecosystem resilience at least at a regional scale.

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus

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University of Puerto Rico, Dept. Biology, CATEC, Coral Reef Research Group, coral_giac@yahoo.com
Ecophysiological evaluation of the invasive capacity of an exotic plant (Spathodea campanulata-tulipán africano) in a secondary forest of Puerto Rico

Abstract

The purpose of this research is to evaluate the potential invasibility of Spathodea campanulata (African tulip) in a secondary forest at the University of Puerto Rico at Humacao. We intended to assess ecophysiological characteristics that will allow S. campanulata to be successful in the forest and how it interacts with Guarea guidonia, a native specie in Puerto Rico. We predicted that S. campanulata has the ability to survive and grow more rapidly than the native species G. guidonia, and, therefore, successfully invades disturbed habitats. We calculated the specific leaf area (SLA) of 33 seedlings of S. campanulata and 33 seedlings of G. guidonia distributed in open areas with little vegetation. The results show that seedlings of S. campanulata have higher values of SLA than G. guidonia. High values of SLA have a relationship with growth and may indicate more efficiency in photosynthetic capacity, pointing out the invasion capacity of S. campanulata.

Introduction

Is Spathodea campanulata an invasive specie in the secondary forest in Humacao municipality of Puerto Rico? The purpose of this research is to evaluate the potential invasibility of S. campanulata (African tulip) in a secondary forest at the University of Puerto Rico at Humacao. Exotic plants have a capacity to acclimate to a variety of habitats. These species possess characteristics like higher values of specific leaf area and light-saturated photosynthesis than native species that allow them to acclimate to different environmental conditions, enabling them to expand their distribution (Baruch, Z. & G. Goldstein, 1999). This causes exotic species to invade habitats and compete with native species. We intended to assess ecophysiological characteristics that may allow S. campanulata to be successful in the forest and how it interacts with Guarea guidonia, a native species in Puerto Rico. We predicted that S. campanulata has the ability to survive and grow rapidly because of their physiological characteristic that allow them to compete with other native species like G. guidonia, and successfully invade disturbed habitats.

Methodology

The study is being conducted in a secondary forest at the University of Puerto Rico at Humacao. We selected 33 seedlings of S. campanulata and 33 seedlings of G. guidonia distributed in open areas with sparse vegetation. Each leaflet lamina was scanned to obtain a digital image to measure the area using ImageJ software. Then, the leaves were dried at 80°C for 24 hours, to measure the dry weight. The specific leaf area of each leaf was calculated by dividing the area of the leaf lamina by its dry weight (Vile, D., 2005). Gas exchange will be measured using a LI-6400 portable photosynthesis system, in fully expanded blades of leaflets in seedlings of S. campanulata and G. guidonia. In addition, we will be calculating chlorophyll concentration, distribution and stomatal density.

The statistical analysis consisted of Two-Sample T Test to determine if exist significant differences in specific leaf area (SLA) between S. campanulata and G. guidonia, and a regression analysis to test the hypothesis that vegetation cover influence SLA, (Statistix 8, Analytical Software).

Results

The Two-Sample T Test shows that exist significant differences in SLA value between both species. S. campanulata has highest SLA values than G. guidonia (Table 1). Figure 1 show that S. campanulata has a trend for a positive relationship between SLA and forest cover density. Meanwhile, G. guidonia has a non-significant but negative tendency between SLA and forest cover density (Figure 1).

Conclusions

In conclusion, seedlings of S. campanulata have higher values of specific leaf area (SLA) than G. guidonia as we expected. High values of SLA have a relationship with growth and may indicate more efficiency in photosynthetic capacity. The seedlings of S. campanulata have the ability to survive and grow rapidly because of their physiological characteristic that allow them to compete with other native species like G. guidonia, and successfully invade disturbed habitats. This characteristic has to be associated with photosynthetic parameters to better evaluate the invasive capacity of S. campanulata.

References


Acknowledgements

I thank to Xavier Jaime, for his help in the laboratory, to Arëlis Hernandez, Maria Morales and Marios Del Paula for their help in field work. Also, I thank to Bio-MINDS and MARC-Minority Access to Research Careers Programs for financial support.

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This research was funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus.
INTRODUCTION

Seed rain and seed bank dynamics are a critical component for the establishment, development and regeneration of native plant communities. On Mona Island, Puerto Rico, the invasive African grass, *Megathyrsus maximus* (Figure 1) has invaded many areas. Grasses often become invasive and may have direct and indirect negative effects on native biodiversity and ecosystems (D’Antonio et al. 1998).

To better understand the ecological role of *M. maximus* on Mona Island, we are collecting and documenting information on the native seeds that reach the soil. With this information, our goal is to determine the effects of *M. maximus* on the distribution patterns and availability of Cactaceae seeds in the soil. We hypothesize that *M. maximus* interferes with seed rain deposition and we predict there will be a reduction in diversity and abundance of Cactaceae seeds in areas where the grass has invaded.

METHODS

Our research project is being conducted in the Mona Island Reserve, Puerto Rico (Figure 2). Samples were collected from four sites, three of which are dominated by *M. maximus*. Soil samples were collected with a 5 x 5 cm soil core sampler, stored in mesh bags and allowed to dry for laboratory analysis (Figure 3).

Due to the large amount of species with small seeds in this community, we further process the remains from each sieve with the modified flotation method. First, we prepare a 0.5g/ml flotation solution of K2CO3. We divide each soil sample into four 50 ml centrifuge vials and fill the top of the vials with the solution. We manually shake each vial until the mixture is homogenized and centrifuge samples at 5,000 RPM for 20 minutes. We collect the floating organic material from the top of the vials and vacuum filter it using #3 Whatman filter paper. Again, we retain the mineral debris in the centrifuge vials for future verification. We place the filter paper with the organic material in the oven at 60°C for about 3 hours. After the sample is completely dry, we proceed to identify, count and photograph seeds under the dissecting scope.

RESULTS

As of this point, we have recovered seeds from over 27 species. Samples from the non-invaded site have different seed bank composition than invaded sites although some species occur in all sites. Seeds from three species including *M. maximus* are found in both invaded and non-invaded sites (Table 1). Invaded sites did not show lower seed diversity so far.

<table>
<thead>
<tr>
<th>Invaded (n=4)</th>
<th>Non-invaded (n=3)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>M. maximus</em></td>
<td><em>M. maximus</em></td>
</tr>
<tr>
<td><em>M. intortus</em></td>
<td><em>E. ciliaris</em></td>
</tr>
<tr>
<td><em>S. peruvianus</em></td>
<td><em>S. peruvianus</em></td>
</tr>
<tr>
<td><em>R. reticulata</em></td>
<td><em>C. hirsutus</em></td>
</tr>
<tr>
<td><em>S. nodiflora</em></td>
<td><em>B. brillosa</em></td>
</tr>
<tr>
<td>Unknown species (11)</td>
<td>Unknown species (8)</td>
</tr>
</tbody>
</table>

Table 1 – Species found in seed bank at invaded and non-invaded sites.

*M. maximus* sites are associated with an overall reduction in soil seed abundance. In total, fewer native seeds were recovered from invaded sites. We have recovered very few cacti seeds from any site up to this point (Table 2). Non-invaded sites are dominated by *C. hirsutus* and *S. peruvianus*.

<table>
<thead>
<tr>
<th>Invadid</th>
<th>Non-invaded</th>
</tr>
</thead>
<tbody>
<tr>
<td>26</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>127</td>
</tr>
<tr>
<td>21</td>
<td>46</td>
</tr>
</tbody>
</table>

Table 2 – Number of seeds found in grass invaded and non-invaded sites.

CONCLUSIONS

Although several samples remain to be analyzed, preliminary results indicate that *M. maximus* can reduce considerably the abundance of native seeds in the soil. Too few cacti seeds have been recovered to draw significant conclusions about the effects of *M. maximus* on cacti species but ongoing research should clarify the effects.

LITERATURE CITED


INTRODUCTION

The last decades have experienced dramatic changes in the overall climate (UNFCCC 2002, EEA-2006, Chambers 2006). In the Caribbean, the annual average temperature has increased about 0.5°C with an expected trend for drier periods based on the convection zones (Neelin et al., 2006) and the study of rain patterns over the last 15 years (Heartsill-Scalley et al., 2007).

Such climate variation may affect the phenology of epiphytic plants in tropical environments (Parmesan & Yohe 2003). If so, these epiphytic plants may be useful tools to study the response of plant populations to atmospheric and/or climatic changes. This study focused on the demographic responses of Lepanthes rupestris a highly studied epiphytic and lithophytic orchid (Fig 1 and 2) endemic to montane forests of Puerto Rico which exhibits metapopulation structures.

OBJECTIVES

- To evaluate and describe long-term precipitation and temperature patterns at the El Verde Field Station of the Luquillo Experimental Forest.
- To determine the relationships between the changes in population size and demographic dynamics, the colonization and extinction rates of the subpopulations of Lepanthes rupestris with seasonal changes within and between the years in precipitation and temperature.

STUDY SITE

The Quebrada Sonadora is a tributary of first order of the Espíritu Santo River. This tributary is 1000 m long. It crosses an area of mature secondary forest, dominated by Dacryodes excelsa. This area is located at the Experimental Station El Verde. The station is located in the Northwest corner of the Experimental Forest of Luquillo (18th 10’N, 65th 30’W). This area’s vegetation is of the Humid Tropical Forest type. The monthly average temperature varies between 21°C in January and 25°C in September (Brown et al., 1983). The annual average precipitation is 371.95 cm, with a standard error of 79.47 (McDowell & Estrada-Pinto 1988).

RESULTS

The mean annual precipitation showed different patterns of variability and intensity at different times during the period of 33 years. Major changes occurred starting in 1991. The first major change was a dry period between 1991 and 1995. Then starting from 1992, there was a higher frequency of events, where the intensity of the mean annual precipitation for the wet season was below the average calculated for the 33 yr period (1975-1991: 4 / 6 events vs. 1992 – 2007: 11/17 events), (Fig. 1b). Also the mean number of dry days per year was lower between the period of 1997-2007 than between 1975 and 1996 (Fig 2a).

CONCLUSIONS

There have been measurable increases in temperature (Max and Min) and decrease at the number of dry periods at the El Verde Field Station. None of the variation in demographic variables at the subpopulation and metapopulation scale was associated with temperature variation. However, there were associations between variables related to precipitation variability (number of dry days and length of periods without rain). Specifically, subpopulation size and growth rate (lambdas) was negatively correlated with the number of days without rain and the length of periods without rain although not consistently between substrate types (rocks vs. trees).

Precipitation was positively related to extinction rates but this association had one year lag time. The length of intervals with dry days was also positively related with colonization rates and with extinction rates but the lag period for colonization rates was shorter. We hypothesize that if observed trends of reductions in the number of dry days per year for El Verde persist, these climate changes may have positive impacts on L. rupestris in terms of its abundance and persistence.

REFERENCES


Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
Introduction

Plant breeding systems and the factors that regulate sexual systems in natural plant populations may influence the frequency and quality of reproductive events (Richards 1997). Data on the reproductive biology and breeding systems are very important to establish effective management and recovery programs for rare and endangered plant species (Liu and Koptur, 2003, Kwak and Bekker 2006), by helping identify the actual factors threatening the persistence of endangered plant populations (Kwak and Bekker 2006).

Objectives

• Determine the breeding system of Guaiacum sanctum (Lignum Vitae, Fig. 1) on Mona Island Reserve (Fig. 2).

• Hypothesis: Based on the generality that outcrossing mating systems are the norm among tropical tree species and observed flower morphology features in G. sanctum, this species is likely to exhibit a breeding system that promotes outcrossing (animal pollination, protogyny or protandry and/or self-incompatibility).

Methods

We combined data on natural pollination and hand-pollination experiments to determine the breeding system of (G. sanctum) following Kearns and Inouye (1993) protocols. I selected 10 plants of G. sanctum on Mona and applied five different pollination treatments with five replicate flowers on each as indicated below:

1) Hand-pollination using self pollen to test for self compatibility.
2) Hand-pollination using a pollen mixture from at least two or three individuals at least 10 m away to test for outcrossing ability (Richards 1997, Barrett and Harder 1996).
3) The bagging of mature buds to test for automatic self-pollination (autogamy, Fig. 5).
4) The bagging andemasculating (anthers removed) of mature buds to test for apomixis (i.e. agamosperm).
5) Open pollination (no manipulation) to see natural levels of pollination.

Treatment number four produced no fruits, therefore this treatment was excluded from the analysis. Differences in fruit set were analyzed using paired t-tests between the following treatment pairs: self vs. cross and autogamy vs. open. We use the Ruiz-Zapata & Katling-Arroyo (1978) self-incompatibility index (ISI) calculated by dividing the average seed set produced via hand self by the average seed set produced via hand cross pollination.

Results and Discussions

Pollination treatments differed in the number of fruits produced. Artificial cross-pollinations produced twice the fruits and twice the seeds produced by the artificial self-pollination treatment (Fig. 3a; t = 3.17, p = 0.0113; Fig. 4a; t = 4.231, p = 0.0022). The bagged pollination treatment produced almost no fruits or seeds relative to the open (control) pollination treatment (Fig. 3b; t = 11.5, p = <0.001; Fig. 4b; t = 9.87, p = <0.0001) suggesting that G. sanctum requires animal visitations for pollination to take place and that this species is not autogamous. The apomixis treatment produced no fruit indicating that no seeds are produced from unfertilized ovules in G. sanctum. Percentage-wise, open natural pollinations produced slightly less fruits than artificial cross pollinations but these treatments were equivalent in terms of seed set. The combined results may suggest that pollination may be somewhat pollination limited (fruit set differences) but that natural pollinations are likely to be generated predominantly by outcrossing (equivalent fruit sets between open and outcross-pollinations). The value of the ISI index was 0.63.

Conclusions

G. sanctum showed a partial self-compatible breeding system (Ruiz-Zapata & Katling-Arroyo, 1978), but produced more fruits and seeds via outcross pollination and had a limited capability to produce seed throughout autogamous self-pollination. This species depends on animal pollination for reproduction/ Thus, its management and recovery actions across different sites would likely needs to consider, the health of its pollinator community as well.

Literature Cited

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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus.
Evaluation of potential demographic differences in the subpopulations of the Mona island iguana Cyclura cornuta stejnegeri across different forest types in Mona island, Puerto Rico

Abstract: Timely research has been directed to endangered species which typically exhibit limited dispersal capabilities, low population numbers and dependency for special habitats for breeding or foraging purposes. This is the case of Rock iguanas which exhibit high degrees of endemism. In Mona Island Reserve, depression forest types have been identified as critical habitats for the endemic, Cyclura cornuta stejnegeri. Here we describe a proposal to address, the importance of these sites in regard to habitat suitability and population dynamics of this species.

Introduction

Cyclura is a genus of lizards from the family Iguanidae and only occur on islands in the West Indies. Rock iguanas have a high degree of endemism (Dayhuff 2006) and there are nine described species. Every species and subspecies within the genus Cyclura is endangered (Alberts et al. 200). These iguanas are vital to their native ecosystems as seed dispersers for native vegetation and their loss may have serious consequences (Alberts et al. 2004). Wild populations of these lizards are directly and indirectly impacted by land development, overgrazing by domestic and feral livestock and predation by humans and feral mammals (Dayhuff 2006). Characteristics such as body size, weight, longevity and clutch size vary between species and could have an effect on the life history strategies of these organisms. Timely research has been directed to endangered species which typically exhibit limited dispersal capabilities, low population numbers and dependency for special habitats for breeding or foraging purposes (Perotto-Baldivieso et al. 2008), this is the case of the Mona rock iguana.

Depression forest sites have been referred to as critical habitats for the Mona Island Rock Iguana (Haneke 1995), yet quantitative evidence is lacking. Variable sizes and spatial variation of this forest type could influence habitat use by these animals to the extent that these variables relate to resource availability and landscape connectivity. With this study, we want to address, then, the importance of these sites in regard to habitat suitability (breeding, dispersal) and population dynamics of Mona rock iguana.

Objectives

- Assess the reproductive biology of the Mona island iguana by looking at differences between the depression forests and the platform in terms of the reproductive success of the Cyclura individuals.
- Assess differences in the movement patterns for the different demographic stages of the Mona island iguana between depression forests and the platform area.
- Assess the differences in habitat utilization by the Mona iguana at depression and platform forests.

Methodology

Study Site

Mona Island is located in the middle of the Mona channel, between the coasts of Dominican Republic and Puerto Rico. It is an oceanic island with a subtropical dry forest climate. The largest (93%) habitat type of the island is the limestone rocky “plateau” (Pérez-Buitrago 2007). Another unique habitat type (1%) are the forest depressions located in the plateau. The vegetation within these forest patches is denser and taller than other surrounding vegetation types (Perotto-Baldivieso et al. 2008). The plateau environment has received little human impact and for iguanas, the coastal plain, but particularly the areas that run parallel and close to the beach, offer most of the area (74%, Haneke 1995) available on the island for nesting; although to a lower extent the plateau depressions are also used for this purpose.

Methods will include:

- Visual censuses will be carried out to determine the distribution of the iguanas in the depression and platform forests.
- Spatial analysis will be carried out to develop distribution maps by season (nesting and courtship), microhabitat and habitat use zones.
- Head start and wild caught iguanas will be used to determine movement patterns of the three life stages in both forest types with GPS radio collars.

References

Dayhuff, Becky. 2006. Rock Iguanas of the Caribbean. All at Sea Magazine.

Our thanks to Miguel A. García for his technical advice, Néstor Pérez, Alberto Álvarez and Frederick Abbott for their assistance in the field and unvaluable advice and the personnel from the DNER for logistical support.

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Ground Water Dynamics of the Littoral Coastal Zone of the Guanica Dry Forest, Puerto Rico

Abstract

Climatic change may have detrimental effects on the coastal habitats of Puerto Rico due to reductions in rainfall, increase in sea levels, and changes in the hydrology of coastal wetlands. Due to these concerns, a ground water monitoring system was established in October 2008 to study the Guanica Dry Forest ground water dynamics. This study aims to understand the relationship between groundwater levels and tidal fluctuations, rainfall, ground water salinity, and temperature. The risk of salinization of freshwater coastal ephemeral ponds in the Guanica Dry Forest (where the threatened Puertorican Crested Toad reproduces) will also be evaluated. Furthermore, this investigation will establish the basis of a long-term study of groundwater dynamics in the littoral coastal zone of the Guanica Dry Forest that could allow the assessment of the impacts of climate change in these sensitive habitats.

Site Description

Due to its scientific and natural richness, in 1981 the United Nations Educational, Scientific and Cultural Organization (UNESCO), recognized the Guanica Dry Forest (GDF) as a Biosphere Reserve. The GDF is located on the southwest coast of Puerto Rico. It extends from the municipality of Guanica to the municipality of Ponce and measures approximately 4,400 ha. This includes approximately 8 nautical miles and 21 km. of coast. The GDF is the most arid zone in the Island. The rainy season occurs from August to November. Its yearly precipitation is about 30 inches. Temperature fluctuates from 27-38 °C. Relative humidity varies from 65-80%. These characteristics form an environment that can be described as hostile. Species living in this environment possess special adaptations to survive under extremely dry and harsh conditions. The predominant geology is primary and secondary sedimentary rocks, mostly karst. The flora has adapted to these extreme conditions. Strong winds, salt water and scarce water limit vegetation growth. Even with these limitations, its flora is one of the most diverse in Puerto Rico. There are approximately 550 species of plants in the GDF. From these plant species, 45 are threatened or in danger of extinction. The GDF also hosts diverse fauna including threatened and endangered species. Within these species, the Puertorican Crested Toad, Pelthophryne lemur, is one of most studied.

Monitoring Station at Tamarindo’s Ephemeral Pond

This station is located at the Tamarindo pond. It has a metric scale that measures in parts per thousand (ppt). Every sample is then analyzed using a salt refractometer that measures in parts per thousand (ppt). A sample of ground water of each well is obtained with a U20 HOBO® water level data loggers installed. One is placed at soil level to read hydrostatic pressure and temperatures of the pond’s waters after rain events. The second one is placed high enough that even in rain events, it doesn’t get submerged. Its purpose is to measure atmospheric pressure. This pressure is later used with data recorded from the data loggers of the four wells to correct hydrostatic pressure for variations in atmospheric pressure.

Salinity Samples

A sample of ground water of each well is obtained with a ground water bailer every 0.5-meter from the top of the well’s casing. Beginning close to where the water level was measured prior and ending at the bottom of the well. Every sample is then analyzed using a salt refractometer that measures in parts per thousand (ppt).

Rainfall and Sea Level Data

Rainfall data is obtained from measurements taken by the Department of Natural Resources in Guánica. Data of sea level is obtained from the web page of the National Oceanic and Atmospheric Administration (NOAA) at Maguayes Island.

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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus
Spatial analysis of foraging activity of the Polygynous Red Imported Fire Ant Solenopsis invicta (Hymenoptera: Formicidae).

I. Abstract: Densities of polygyne Solenopsis invicta are higher in the introduced range than the monogyne social form and this might be because the lack of intraspecific aggression, but the mechanisms of how polygyne colonies persist at these high densities are still unknown. Lack of aggression might lead colonies to compete by exploitative mechanisms in which some ants remove food before others would find it. For instance: do they share resources? If so, to what extent? I will answer this question by marking foragers and recruits externally with ultraviolet fluorescent ink. Recapture will allow me to map complete polygyne foraging areas and also observe resource sharing of colonies at baits. Some crucial and basic questions about polygyne foraging organization will be answered: 1) How is the mound volume of a polygyne colony related to its foraging area? 2) How far do polygyne ants forage? 3) Does a colony monopolizes a single bait or does it share with others? 4) Can bait distance and/or colony biomass determine food acquisition success? 5) Do polygyne foraging areas overlap? 6) If so, to what extent?

II. Methods: 
1) Gp-9 Protein Method
   - Highly Correlated with Gp-9 allele (a pheromone binding protein)
   - **BB** Monogyne **Bb** Polygyne
   - 10 ants/colony
   - One heterocygote determines polygyny

2) Mark-Recapture Foragers
   - Ether/Paint
   - Anesthetized Ant

3) Estimate Forager Population, Determine Maximum Foraging Distance and Overlap Area

III. Preliminary Results:
- Polygyne Fire Ants were found at Hatillo P.R. at a cattle area.
- 73.1% of 155 baits around a focal mound contained only S. invicta.
- 8% of baits enclosed another ant species among fire ants.
- Maximum foraging distance was 5m away from mound. This is further than reported by Weeks, (2004) (4m).
- Due to low recapture percents, (<10%), maximum foraging distance might be underestimated.
- Only 3 more ant species occurred at baits and were not abundant.
- Three different vectors of ants were seen while capturing from sausages. Six vectors are the maximum reported. (Zakarov & Thompson, 1998).

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Funded by National Science Foundation, HRD #0734826, USDA-CSREES 2008-38422-19211 and University of Puerto Rico, Rio Piedras Campus
Effect of exotic grass, *Megathyrsus maximus*, on native biodiversity in Mona Island Reserve

**Introduction**

- Species invasions are considered one of the leading causes of global biodiversity loss (Wonham, 2005) and are the second cause of species endangerment in the United States (Brooks and Pyke, 2002).

- The African grass, *Megathyrsus maximus*, has established extensive stand on Mona Island and there is no information about its effect on its natural communities.

- When grasses get to colonize a space they may influence species composition and ecosystem stability” (D’Antonio and Vitousek, 1992).

**Question**

Is the presence of *Megathyrsus maximus* related to differences in abundance and diversity of above ground vegetation and soil fauna in Mona Island Reserve?

**Hypothesis**

Grass dominated areas will show lower abundance and richness of vegetation and soil mesofauna.

**Study Site**

**METHODS**

**Results**

**III. Species composition and relative abundance**

**Mesofauna**

- Grass invaded areas consistently showed less vegetation and soil fauna species richness at the sample level. At the plot level (larger scale), invaded areas did show less plant species but not necessarily less mesofauna species.

- We found no convergence in overall plant composition between invaded plots or non-invaded plots.

- Soil mesofauna was less abundant in grass invaded environments.

- Collembolans and Acari dominate soil fauna at sampled sites but the relative ratio of Collembolans and Acari related to grass presence.

- Collembolans were less abundant on grass-invaded sites.

**Conclusions**

- Grass invaded areas consistently showed less vegetation and soil fauna species richness at the sample level. At the plot level (larger scale), invaded areas did show less plant species but not necessarily less mesofauna species.

- We found no convergence in overall plant composition between invaded plots or non-invaded plots.

- Soil mesofauna was less abundant in grass invaded environments.

- Collembolans and Acari dominate soil fauna at sampled sites but the relative ratio of Collembolans and Acari related to grass presence.

- Collembolans were less abundant on grass-invaded sites.

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Funded by National Science Foundation, HRD #0734826, NSF-UMEB DBI 0602642 and University of Puerto Rico, Central Administration and Rio Piedras Campus
Effect of exotic grass *Megathyrsus maximus* on soil mites fauna in Mona Island Reserve.

**Introduction**

- Biological invasions alter ecosystem processes in invaded areas, thereby causing functional as well as compositional change (D’Antonio and Vitousek, 1992).
- In dominated sites, grasses can alter ecosystem processes from nutrient recycling to regional microclimate.
- Exotic grasses compete effectively with native species in a wide range of ecosystems. (Figure 2)

**Hypothesis**

1. The presence of the invasive grass, by creating a difference in humidity and vegetative composition, creates a difference in the soil mite composition.
2. Prostigmata mites will exhibit higher frequencies regardless of vegetation environment by having better adaptations to changes in the environment.

**Methods**

**Study site**

-Mona Island Natural Reserve is located between Puerto Rico and Hispaniola

-Xerophytic vegetation (average temp = 25 °C, Aver. annual precipitation 800 mm)

-The project was conducted in the Plateau Shrub Forest (North east of the island) in areas where *Megathyrsus maximus*, an African grass has established since 1940’s.

**Results**

We recovered 1014 mites in total in all pitfall traps combined. (Figure 3)

We found a higher relative abundance of Prostigmata mites than the other two suborders (data not shown).

**Discussion**

Soil mite fauna was higher in grass invaded areas but this tendency was not strong in one of the blocks. This could be an effect of invasion time (St. John et.al. 2001) as a factor of the effects of the grass to the community.

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**Figure 1:** Dynamics in Soil Ecology (Neher, 1999)

**Figure 2:** Invaded area with *Megathyrsus Maximum* in Mona Island

**Figure 3:** Mites found in soil samples: Prostigmata, Oribatida, Mesostigmata (in that order)

**Figure 4:** Working in Mona Island and working with the soil mite fauna in Mona Island Reserve.

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**Classification of Mites:**

- Using Lactophenol, we clean the mites
- Identified with ASP keys (Fig. 4)
- Contact experts to verify identifications.

**Questions**

Is there an association between the presence of invasive grass *Megathyrsus maximum* and the soil mites composition in Mona Island?

Are there differences in the relative composition of mites with different functional roles (i.e. Predators, Mycrophytophagus, Parasitoids) as a function of the presence or absence of the invasive grass *Megathyrsus maximum*?

What is the biodiversity of soil mites in the lowland dry limestone grassland and shrubland of Mona Island?

---

**Table 1:** Some Families found in the samples.

<table>
<thead>
<tr>
<th>Order</th>
<th>Family</th>
<th>Functional Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oribatida</td>
<td>Neoluidae</td>
<td>Saprophagus</td>
</tr>
<tr>
<td>Oribatida</td>
<td>Cymberenaemaiidae</td>
<td>Saprophagus</td>
</tr>
<tr>
<td>Oribatida</td>
<td>Galummidae</td>
<td>Saprophagus</td>
</tr>
<tr>
<td>Oribatida</td>
<td>Scheloribatidae</td>
<td>Saprophagus</td>
</tr>
<tr>
<td>Oribatida</td>
<td>Phthiracaridae</td>
<td>Saprophagus</td>
</tr>
<tr>
<td>Prostigmata</td>
<td>Bdellidae</td>
<td>Predator</td>
</tr>
<tr>
<td>Prostigmata</td>
<td>Tetranychidae</td>
<td>Parasitic</td>
</tr>
<tr>
<td>Prostigmata</td>
<td>Erythraeidae</td>
<td>Predators</td>
</tr>
<tr>
<td>Mesostigmata</td>
<td>Dermassynidae</td>
<td>Predator</td>
</tr>
</tbody>
</table>

**Figure 4:** Working in Mona Island and working with the identifications, respectively.

---

**References**

- Lourdes B Lastra-Díaz , Dra. Elvia Melendez-Ackerman, Lorna Moreno, University of Puerto Rico Río Piedras Campus / lou_lastra@hotmail.com

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INTRODUCTION

The hoatzin is the only folivorous bird known to have foregut fermentation. Since changes in plant availability must influence the dietary choices and affect the structure of the digestive microbiota in herbivores, we hypothesized that hoatzins from different habitats would have different crop microbial communities.

To test this hypothesis we characterized the crop microbiota of three birds in two locations in Venezuela, the rivers Orinoco and Cojedes.

We compared plant availability and foraging behavior in the two locations, and used a 16S rDNA microarray, to characterize crop bacteria.

RESULTS

• Plant composition but not richness, were different between the 2 locations (Table 1), (13 of the 18 plant families exclusive to one location),
• Coccoloba, was present and dominant in both habitats (Table 1).
• Only two common plant families (Polygonaceae and Fabaceae) were consumed at the two locations (Table 1).
• 38 bacterial phyla with ~1,600 bacterial OTUs were found, with no differences in richness in the 2 locations (Table 2, Figure 1).
• Had Higher richness in Orinoco crop communities than at Cojedes (Figure 2).
• Location explained as much as 40% of sample variance (Figure 3).
• There were only 60 significantly different OTUs, 25 abundant in Cojedes and 35 in Orinoco crops (Figure 4).

CONCLUSION: The bacterial differences in the crop is likely to respond to the selective effect of the dietary substrates particular to each habitat, and might imply differences in crop bacterial function.
INTRODUCTION

The hoatzin (Opisthocomus hoazin) is a South American folivorous bird, with a crop microbial ecosystem that ferments dietary plant material.

• Juvenile birds progressively become independent from the chicks fed by adults regurgitated crop liquid contents.

We hypothesized that the crop bacterial ecosystem develops in chicks soon after hatching, continuing until they acquire the mature crop community through mechanisms of microbial succession, similar to that reported in the intestine of mammals.

• The aim of this work was to compare the crop bacterial community structure in hoatzins from three age groups: newly hatched chicks, juveniles and adults, by performing 16S rDNA libraries and hybridizing 16S amplicons onto the G2 PhyloChip.

METHODS

DNA Extraction of Crop Contents

16S rRNA genes amplified cloned and sequenced

Sequences aligned with the NAST aligner. Taxonomic placement of the clones, tree building and distance matrices done in ARB.

Rarefaction curves done in EstimateS. Community analyses in UniFrac.

RESULTS

• No significant differences in richness between the age groups (p>0.005) (Table 1).

• Undersampled (Fig 1).

• Firmicutes and Bacterioidetes dominated the crop bacterial community (Fig 2). The microarray detected 10x more bacteria but 91% of the cloned bacteria were novel.

• Crop community changes occur with age (Fig. 3,4,5)

Table 1. Clone library-based estimates of diversity in birds of different age groups.

<table>
<thead>
<tr>
<th>Metric</th>
<th>N sequences</th>
<th>Good's Dominance Index</th>
<th>Observed Richness (OTUs)</th>
<th>Richness (SIM)</th>
<th>Diversity</th>
<th>Evenness</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chicks</td>
<td>750</td>
<td>0.99</td>
<td>127 (0.53)</td>
<td>165 (0.50)</td>
<td>4.60</td>
<td>0.03</td>
</tr>
<tr>
<td>Juveniles</td>
<td>659</td>
<td>0.89</td>
<td>149 (0.51)</td>
<td>191 (0.51)</td>
<td>4.70</td>
<td>0.02</td>
</tr>
<tr>
<td>Adults</td>
<td>624</td>
<td>0.90</td>
<td>120 (0.52)</td>
<td>161 (0.53)</td>
<td>4.90</td>
<td>0.03</td>
</tr>
<tr>
<td>Preadult sample</td>
<td>2,112</td>
<td>0.91</td>
<td>294 (0.54)</td>
<td>324 (0.54)</td>
<td>4.6</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Fig 1. Observed (solid lines) and estimated (dotted lines) clone library-based rarefaction curves for chicks, juvenile and adult birds.

Fig 2. Neighbor-joining tree of the 40 phyla detected by the DNA microarray.

CONCLUSIONS

• Despite most of the richness is already in the crop of chicks, the structure of the bacterial community changes with age.

• Age changes likely to respond to the opening of new niches through the bird’s dietary changes.
Glyphosate is a post-emergent, systemic and non-selective herbicide used in both agricultural and non-agricultural areas. Glyphosate-based herbicides are among the first herbicides used worldwide, and their use is not restricted. Trade names include Armada, Dardo, Fuste, Kleenup, Landmester, MON-0573, Poloéd (sesquisodium), Quotometer, Ranger, Rodeo, Roundup (isopropyl ammonium), Spasor, Squadron, Sting, Stirrup, Touchdown (trimesium), Tumbleweed, and Wellop.

**URBAN ENVIRONMENTAL IMPACTS**

- Pesticides are used prevalently in agricultural zones but in the last years many studies have shown the impact of urban uses which is important at the local scale due to application on surfaces like roads, sidewalks, and lawns.
- Used worldwide, glyphosate can be a major pollutant of rivers and surface waters. It can also contaminate organisms including humans, but also food, feed, and ecosystems.
- The glyphosate is very sensitive to surface runoff. This is due to its high solubility in water ($10.1 \text{ g L}^{-1}$ at $20 \text{ C}$) and high sorption ($>1.000 \text{ L kg}^{-1}$) to soil particles.
- Have been found in groundwater from small private water supply systems.
- Rainfall events of high intensity increase glyphosate transfer by runoff on hard surfaces.
- Once in water, glyphosate is not readily broken down by water or sunlight. Dissipation from water is usually due to sorption to sediment or suspended particles or uptake by plants and biodegradation.
- In 2008, 1,405,279 kg of herbicides were imported to Puerto Rico from the United States. Uses are not specified. Since the product is not restricted, its urban use is conceivable.
- Little data exists on the environmental occurrence of glyphosate and its metabolite, AMPA, derived from the urban use of glyphosate and there is none for Puerto Rico.
- A better understanding of the potential contribution of glyphosate and AMPA to streams derived from the urban use of this herbicide is needed.

**References:**
Puerto Rico Planning Board, Program of Economic and Social Planning, Subprogram of Economic Analysis.
http://www.landbrugsinfo.dk/Planteavl/Plantevaern/Bekaempelsesmidler/Sider/plk06_99_1_W_Bruesch.pdf

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### TOXICITY TO AQUATIC ORGANISMS

<table>
<thead>
<tr>
<th>Organism Group</th>
<th>Toxic Effects on Organism Groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphibians</td>
<td>Biochemistry, Development, Genetics, Growth, Morphology, Mortality</td>
</tr>
<tr>
<td>Aquatic Plants</td>
<td>Accumulation, Biochemistry, Development, Growth, Morphology, Mortality, Physiology, Population</td>
</tr>
<tr>
<td>Crustaceans</td>
<td>Mortality</td>
</tr>
<tr>
<td>Echinoderms</td>
<td>Development</td>
</tr>
<tr>
<td>Fish</td>
<td>Accumulation, Avoidance, Biochemistry, Enzymes, Growth, Histology, Immunological, Intoxication, Mortality</td>
</tr>
<tr>
<td>Insects</td>
<td>Intoxication, Population</td>
</tr>
<tr>
<td>Mollusks</td>
<td>Behavior, Development, Enzyme(s), Genetics, Growth, Mortality, Physiology</td>
</tr>
<tr>
<td>Nematodes and Flatworms</td>
<td>Population</td>
</tr>
<tr>
<td>Phytoplankton</td>
<td>Biochemistry, Development, Physiology, Population</td>
</tr>
<tr>
<td>Zooplankton</td>
<td>Accumulation, Behavior, Enzyme(s), Genetics, Intoxication, Mortality, Population, Reproduction</td>
</tr>
<tr>
<td>Duck</td>
<td>Morphophysiology of the male genital system</td>
</tr>
</tbody>
</table>

### TOXICITY TO HUMANS

<table>
<thead>
<tr>
<th>Effect</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skin toxicity</td>
<td>Amerio et al 2004. <em>Clinical Toxicology</em>; 42(3):17-319</td>
</tr>
</tbody>
</table>

Sheila M. Soler-Llavina and Jorge Ortiz-Zayas, University of Puerto Rico-Rio Piedras / jorgeortiz_ites@yahoo.com

Funded by National Science Foundation, HRD #0734826
University of Puerto Rico, Rio Piedras Campus
Detecting vertical transfer of antibiotic resistance genes in the absence of antibiotics

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Department of Biology, University of Puerto Rico, Río Piedras Campus/ amalicea09@hotmail.com

INTRODUCTION

• Bacteria can develop resistance to antibiotics.
• Hosts can carry a reservoir of resistant bacteria in their microbiome.
• Tetracycline has been used for over 50 years and has enterohepatic circulation, so intestinal microbes are exposed via bilis to the injected antibiotic.
• Our hypothesis is that antibiotic resistance can be vertically transmitted between generations, in the absence of antibiotics. Mothers readily colonize their offspring with bacteria that have antibiotic resistance genes.
• To test this hypothesis, we aimed to detect resistance genes in the offspring of mothers carrying resistance.

METHODOLOGY

• Stool samples were collected from 6 female FVB mice, pre-tetracycline treatment and post-tetracycline treatment.
• Females were crossed with males.
• Intestines from offspring mice were collected at day 1 and 9 after being born (Figure 1).
• DNA was extracted from stools and intestines using the MoBio Power Soil Kit, followed by PCR amplification of 16SrDNA genes and five tetracycline resistance genes: tet(O), tet(Q), tet(W), tet(S) and tet(M) (Pei et al. 2004; Villedieu et al. 2003).

PRELIMINARY RESULTS

• All mothers (treated and controls) showed presence of three resistance genes: tet(O), tet(Q), tet(W). We were unable to amplify tet(M). One post-treat mother from the control group amplified for tet(S) gene (Table 1).
• Concerning the offspring at day 1, all mice amplified for tet(O). Only four mice (three control and one experimental) amplified for tet(S) (Table 2).
• At day 9, all mice amplified for tet(O) and tet(S) (Table 2).

Table 1. Number of mothers with tetracycline resistance genes of ribosomal protection.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Animal Group</th>
<th>tet(O)</th>
<th>tet(Q)</th>
<th>tet(W)</th>
<th>tet(S)</th>
<th>tet(M)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tetracycline</td>
<td>Mothers pre-treatment (n=3)</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>To be done</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Mothers post-treatment (n=3)</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Control</td>
<td>Mothers pre-treatment (n=3)</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>To be done</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Mothers post-treatment (n=3)</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 2. Number of offspring with tetracycline resistance genes of ribosomal protection.

<table>
<thead>
<tr>
<th>Mice descendants of mothers</th>
<th>Age (days)</th>
<th>tet(O)</th>
<th>tet(Q)</th>
<th>tet(W)</th>
<th>tet(S)</th>
<th>tet(M)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treated (N=3)</td>
<td>1</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>Controls (N=3)</td>
<td>1</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>0</td>
</tr>
</tbody>
</table>

CONCLUSIONS

• Mice show intrinsic resistance to tetracycline.
• There is vertical transfer of tetracycline resistance genes from mother to offspring at early stages in life, without the selective pressure of antibiotics.

REFERENCES

Diversity of fungi in the Hoatzin crop: an 18S rDNA-based approach

INTRODUCTION

The Hoatzin is a unique South American herbivorous bird and the smallest vertebrate to have a fermentative crop, similar to the cow rumen. Like bacteria, fungi play an important role in foresgut fermentative ecosystems by producing volatile fatty acids that are absorbed by the animal, thus contributing to the nutritional economy of the host.

AIM: Characterize the fungal diversity in the crops of three adult birds by amplifying fungal 18S ribosomal genes followed by library construction and sequence analysis.

METHODOLOGY

Table 1. Fungal Diversity estimates.

<table>
<thead>
<tr>
<th>Diversity Estimates</th>
<th>Fungal clones</th>
</tr>
</thead>
<tbody>
<tr>
<td>N clones</td>
<td>201</td>
</tr>
<tr>
<td>Collected OTUs</td>
<td>122</td>
</tr>
<tr>
<td>Good's coverage index</td>
<td>63%</td>
</tr>
<tr>
<td>Shannon index</td>
<td>4.5</td>
</tr>
</tbody>
</table>

Table 2. Fungal Diversity found in the crop of the hoatzin

<table>
<thead>
<tr>
<th>Fungal Habitats</th>
<th>N. Clones</th>
<th>Unique Clones *</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nectar</td>
<td>12</td>
<td>11/12 (92%)</td>
</tr>
<tr>
<td>Soil, Rocks, Air, Decaying Org.</td>
<td>44</td>
<td>21/44 (48%)</td>
</tr>
<tr>
<td>matter, Wood, Marble, Monuments</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plant epiphytes and fungi in</td>
<td>20</td>
<td>9/20 (45%)</td>
</tr>
<tr>
<td>Leaves</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td>1</td>
<td>100%</td>
</tr>
<tr>
<td>Plant parasites</td>
<td>27</td>
<td>1/27 (3.7%)</td>
</tr>
<tr>
<td>Plant pathogens</td>
<td>51</td>
<td>23/51 (45%)</td>
</tr>
<tr>
<td>Molds</td>
<td>3</td>
<td>100%</td>
</tr>
<tr>
<td>Grains and Fruits</td>
<td>1</td>
<td>100%</td>
</tr>
<tr>
<td>Tree endophyte/ marline gut</td>
<td>4</td>
<td>3/4 (75%)</td>
</tr>
<tr>
<td>Tree epiphyte</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>Agaricale and other mushroom-like</td>
<td>3</td>
<td>2/3 (67%)</td>
</tr>
<tr>
<td>fungi</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cryptococcus sp.</td>
<td>27</td>
<td>4/27 (15%)</td>
</tr>
<tr>
<td>Psilocybe Antarctica soil</td>
<td>1</td>
<td>100%</td>
</tr>
<tr>
<td>Lichens</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Magnoliophy plants</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>TOTAL</td>
<td>201</td>
<td>41%</td>
</tr>
</tbody>
</table>

Conclusions

• The majority of the fungi found in the hoatzin crop were associated with plants (diet-related).
• Interestingly, one Species-like OTU seems to be indigenous to the crop: Cryptococcus sp.

Table 2. Fungal Diversity found in the crop of the hoatzin

Funded by National Science Foundation, NSF: IOS 0716911, DDIG 0709840 and HRD #0734826 and University of Puerto Rico, Río Piedras Campus

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*Presenting author e-mail: miguelrivera427@hotmail.com
*Corresponding author
INTRODUCTION

The Hoatzin is one of the world's few obligate folivorous birds inhabiting the riverine forests of northern South America. It is the smallest vertebrate to have an enlarged foregut, in which plant structural carbohydrates are converted into simple sugars through microbial fermentation.

We hypothesize that this ecosystem is likely to harbor a diverse fermentative microflora.

Our aim was to characterize bacterial cultures isolated from hoatzin crop contents by amplifying and sequencing 16S rRNA genes.

METHODOLOGY

Crop contents were used as inocula to Medium M10 (1). Pure cultures were stored freeze-dried with glycerol (Figure 3).

Bacterial isolates were subjected to 16S rRNA PCR amplification

Agarose gel electrophoresis

Purification of PCR products using Qiagen Purification Kit

Sequencing of 16S amplicons

BLAST using the Greengenes Database

RESULTS

• A total of 41 pure cultures preserved in glycerol were amplified for the 16S rRNA gene but only 27 were sequenced.

• The majority of the bacterial cultures were from phylum Firmicutes (Table 1) as had been previously found by cloning/sequencing (2).

• We found most bacteria to be facultative anaerobes and one specific culture is related to rumen fiber degraders (Clostridium lituseburense).

• Unclassified sequences (related to clones in the database) and sequences less than 97% ID to any bacteria, will be further sequenced aiming to a complete 16S gene (~1,500bp) for accurate identification.

<table>
<thead>
<tr>
<th>Phylum</th>
<th>Family</th>
<th>Genus</th>
<th>Species</th>
<th>N cultures sequenced</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteobacteria</td>
<td>Enterobacteriaceae</td>
<td>Escherichia</td>
<td>Escherichia coli</td>
<td>2</td>
</tr>
<tr>
<td>Firmicutes</td>
<td>Bacillaceae</td>
<td>Bacillus</td>
<td>Bacillus licheniformis</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>Clostridiaceae</td>
<td>Clostridium</td>
<td>Clostridium lituseburense</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Streptococcaceae</td>
<td>Streptococcus</td>
<td>Streptococcus galgaliticus</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>Enterococcaceae</td>
<td>Enterococcus</td>
<td>Enterococcus casseliflavus</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>UNCLASSIFIED</td>
<td>1</td>
</tr>
</tbody>
</table>

TOTAL CULTURES 27

CONCLUSION

The bacteria isolated from the hoatzin crop mostly resemble those found in the human gut and the cow rumen pointing to the importance of molecular culture-independent ecology studies for assessing bacterial diversity in such a unique fermentative ecosystem.

REFERENCES


Aspergillus flavus: genetic variation, patterns of distribution and substrate specificity

Abstract

Aspergillus flavus is an opportunistic pathogen of humans, animals and plants, characterized by aflatoxin production and tolerance to salinity and high temperature. Recently we have found that A. flavus is common in sea fans (Gorgonia sp.) in waters around Puerto Rico. However the source of inoculum is unclear. Also, it is unclear whether A. flavus strains demonstrate substrate specificity. Few phylogeographic studies have focused on marine fungi. In this study, we compared genetic similarity between A. flavus strains isolated from several substrates and sites to determine if there is an association between substrate and phylogeny, and genetic distance vs. geographic distance. DNA was extracted and parts of three genes were sequenced. Ten Amplified Fragment Length Polymorphisms (AFLP) combinations were used to fingerprint some of the isolates from sea fans. Phylogenetic relationships were estimated with maximum parsimony (for sequences) and neighbor-joining (for AFLPs). Results showed no correlation between substrate specificity and phylogenetic distribution. A. flavus strains appear to be generalists. No clade was noted to have a specific phylogeographic distribution, so there was no evidence of endemism in A. flavus strains. Improved resolution of genetic identity of A. flavus strains can help us understand the dynamics of opportunistic pathogens and their role in coral reefs.

Methods

Around 160 A. flavus strains were isolated from different substrates and sites. Strains were identified by morphology and by sequencing the nuclear ribosomal ITS region using universal fungal primers ITS1F & ITS4. Regions of two protein-coding loci, o-methyltransferase and β-tubulin were also sequenced. BLAST searches in GenBank showed that the most similar sequences were A. flavus for all strains. AFLPs (Amplified Fragment Length Polymorphisms) were used to estimate relatedness among strains of A. flavus. 75 strains were used, a subset of those in the genealogical tree. Peak Detection Threshold Intensity was >100.

Results and Discussion

AFLPs. Peaks on the AFLP chromatogram show three alleles (columns) for three strains of A. flavus (rows). Only polymorphic peaks (black boxes) are informative. (Program: GeneMapper 4.0).

Fig. 1. A) Gorgonia ventailina colony with necrotic aspergillosis lesion (arrow). B) Aspergillus flavus. 1) colonies on PDA, 2&3) conidiophores.

Fig. 2. AFLPs. Peaks on the AFLP chromatogram show three alleles (columns) for three strains of A. flavus (rows). Only polymorphic peaks (black boxes) are informative. (Program: GeneMapper 4.0).

Sampling Sites

<table>
<thead>
<tr>
<th>Outgroup</th>
<th>India</th>
<th>Puerto Rico</th>
<th>Estados Unidos</th>
<th>Panamá</th>
<th>Africa</th>
</tr>
</thead>
<tbody>
<tr>
<td>India</td>
<td>Puerto Rico</td>
<td>Estados Unidos</td>
<td>Panamá</td>
<td>Africa</td>
<td></td>
</tr>
<tr>
<td>Puerto Rico</td>
<td>India</td>
<td>Estados Unidos</td>
<td>Panamá</td>
<td>Africa</td>
<td></td>
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<tr>
<td>Estados Unidos</td>
<td>Puerto Rico</td>
<td>India</td>
<td>Panamá</td>
<td>Africa</td>
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<td>India</td>
<td>Africa</td>
<td></td>
</tr>
<tr>
<td>Africa</td>
<td>Puerto Rico</td>
<td>Estados Unidos</td>
<td>Panamá</td>
<td>India</td>
<td></td>
</tr>
</tbody>
</table>

The parsimony tree based on partial sequences two genes revealed no pattern of association among A. flavus strains isolated from different sites (Fig. 3). The neighbor-joining tree of strains from sea fan tissue revealed no pattern of relatedness among A. flavus strains isolated from healthy vs. diseased tissues (Fig. 4). High genetic similarity between strains from healthy and diseased tissue suggests that every strain present in the tissue is a potential pathogen, as long as it encounters a susceptible host. Strains from a single site were often related, especially in Jobos (Fig. 4). This pattern contradicts a common assumption for microbial biogeography. ‘Everything is everywhere.’ However, no association between substrate and phylogeny was seen on a global scale (Fig. 5).

Fig. 3. Do A. flavus populations show global phylogeographic structure? Maximum parsimony tree using two protein-encoding loci from different strains of A. flavus. Each clade with bootstrap support is represented by a circle; size of the circle is proportional to the number of strains. No. parsimony-informative characters = 42; CI = 0.80.

Fig. 4. Do A. flavus populations show phylogeographic structure within Puerto Rico? Neighbor-joining tree based on AFLP of A. flavus strains. Colors show sampling sites. Icons show strains from diseased (♭) and healthy (♮) tissue. Bootstrap values are based on 2000 reps. Consistency index (CI) = 0.42.

Fig. 5. Are A. flavus populations substrate-specific? Neighbor-joining tree based on AFLPs for 75 A. flavus strains. Terminal nodes are collapsed for ease of viewing. Bootstrap values based on 2000 reps. 526 parsimony-informative characters; CI = 0.13.


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Funded by National Science Foundation, HRD #0734826 NOAA Sea Grant, NIH-SCoRE and UPR, Central Administration and Río Piedras Campus
Status of viruses causing symptoms in cucurbits in Puerto Rico

Introduction

Cucurbits provide important basic ingredients for the Caribbean diet. Pumpkins are the second most important vegetable crop in terms of revenue generated in Puerto Rico. Virus and severe virus vector outbreaks are a frequent and major cause of low yields and phytosanitary limitations to growing cucurbits in Puerto Rico. Continuous growing throughout the year and overlapping of susceptible crops makes Puerto Rico, a highly diverse island, an excellent and dynamic environment for plant viruses to evolve (Figure 1). Those aspects are also a major challenge for the development of control strategies. A survey was conducted to assess the types and prevalence of viruses infecting cucurbits. The virus isolates were mechanically transmitted, serological assays were conducted to identify the various viruses and RT-PCR followed by sequencing was done for the potyviruses, which were the most frequently encountered viruses in this survey.

Materials and Methods

Virus Samples - A total of ninety-nine cucurbits plants (mainly pumpkin and watermelon) showing virus-like symptoms were sampled in ten municipalities of Puerto Rico (Adjuntas, Coamo, Corozal, Isabela, Puerto Rico (Adjuntas, Coamo, Corozal, Isabela, Santa Isabel, Villalba) (Figure 2). Mechanical transmission - Assays were conducted at the Rio Piedras Experimental Station under greenhouse and laboratory conditions. All samples were inoculated to Cucurbita moschata ‘Walthan’ using phosphate buffer and carbonarum. Inoculated plants were observed for 21 days for development of symptoms. Infected tissue were freeze-dried for preservation. Serological assays - ELISA and ‘immunosnips’ assays (Agdia Inc.) were conducted to identify the occurrence and predominance of the following virus-taxon: potyvirus, PRSV, ZYMV, CMV, and SmpMV. Inoculated plants were observed for 21 days for development of symptoms. Infected tissue were freeze-dried for preservation. Serological assays - ELISA and ‘immunosnips’ assays (Agdia Inc.) were conducted to identify the occurrence and predominance of the following virus-taxon: potyvirus, PRSV, ZYMV, CMV, and SmpMV. Inoculated plants were observed for 21 days for development of symptoms. Infected tissue were freeze-dried for preservation. Serological assays - ELISA and ‘immunosnips’ assays (Agdia Inc.) were conducted to identify the occurrence and predominance of the following virus-taxon: potyvirus, PRSV, ZYMV, CMV, and SmpMV.

RT-PCR and sequencing - A core portion of the coat protein subunit gene was amplified in a PCR reaction using the degenerate primers M1 (5’- ATGGTHTGGTGYATHGARAAYGG-3’) and M2 (5’-TGCTGCKGCYTTCATYTG-3’) (1). Bands were cut from the gel, cleaned, and sequenced with ABI Dye Terminator kit (2 times per sample) (4). Sequences were edited using Vector NTI software (Invitrogen) then aligned using CLC Workbench 3. Phylogenetic trees were produced by Unweighted Pair Group Method with Arithmetic mean (UPGMA) with Bootstrap methods. Sequences of known potyvirus isolates deposited at Genbank were used as control for taxonomic information.

Results and Discussion

One hundred percent of the samples were positive for at least one of the tested viruses. Serologic tests showed more than 90 percent of the samples to be positive for potyvirus, with 45% positive for PRSV and 40% positive for ZYMV. Ten percent of the samples were from an unknown potyvirus. WMV, SmpMV and CMV were reported in less than 2 percent of the samples. Occurrence of co-infection with 2 or more viruses was common.

Conclusions

Potyviruses were the most common group of viruses causing symptoms in cucurbits in Puerto Rico. Often multiple viruses were found co-infecting the same plant. Papaya Ringspot Virus and Zucchini Yellow Mosaic Virus were the most frequent species reported. The different virus isolates induced a broad range of symptoms in Cucurbita moschata, indicating their high variability. The variability of isolates were confirmed by the sequences of the coat protein gene fragment amplicon.

Literature cited

José Carlos V. Rodrigues / Linda Wessel-Beaver. Crops & Agroecosystems Department, University of Puerto Rico - Mayaguez, jose_carlos@mac.com

Figure 1 - Cucurbits are grown commercially (A and B) in lower flat coastal areas and in the mountains. They can also be found naturally growing in the central region (C) of the island. There are a great diversity of virus-like symptoms associated with cucurbits in Puerto Rico (D, E, and F).

Table 1. Twelve potyvirus isolates sequenced from Puerto Rico.

<table>
<thead>
<tr>
<th>Code</th>
<th>Location</th>
<th>Plant host</th>
<th>Collectd</th>
</tr>
</thead>
<tbody>
<tr>
<td>Potty 1</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Oct/2007</td>
</tr>
<tr>
<td>Potty 3</td>
<td>Santa Isabel</td>
<td>Cucurbita lanata</td>
<td>Feb/2008</td>
</tr>
<tr>
<td>Potty 4</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Mar/2008</td>
</tr>
<tr>
<td>Potty 5</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 6</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 7</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 8</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 9</td>
<td>Juana Diaz</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 10</td>
<td>Isabela</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 11</td>
<td>Isabela</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
<tr>
<td>Potty 12</td>
<td>Isabela</td>
<td>Cucurbita lanata</td>
<td>Apr/2008</td>
</tr>
</tbody>
</table>

Figure 2 - Collecting sites for cucurbits in Puerto Rico from 2005 to 2008. The numbers inside the symbols represent the number of samples from the area. (Relief map from US Geological Survey)

Figure 3 - Results of mechanical inoculation, serology and RT PCR were used to characterize the isolates from Puerto Rico. (A) Different symptoms observed in Cucurbita moschata ‘Walthan’ 21 days after the inoculation. (B) ELISA plate results, and (C) A RT-PCR coat protein gene fragment (327bp) was obtained from the virus isolates using potyvirus primers. (D) Chromatogram of a sequence of a Puerto Rican potyvirus isolate.

Figure 4 - A phylogenetic tree produced by Unweighted Pair Group Method with Arithmetic mean (UPGMA) with Bootstrap on nodes. Coat protein gene fragments of from Genbank accessions PRSV AB127935= Papaya Ringspot Virus and ZYMV AJ420019 = Zycchini Yellow Mosaic Virus were used as references.

Figure 5 - Frequency of detection by serological means of different viruses in cucurbit in Puerto Rico between 2006 to 2009. Mechanical transmission to Cucurbita moschata ‘Walthan’ was conducted and expression of symptoms was observed 2 weeks after inoculation (Figure 3). RNA was extracted from some of the field samples and inoculated plants (Table 1). RT-PCR was conducted with potyvirus degenerate primers and the amplicons were sequenced to identify more specifically the potyviruses. A phylogenetic tree produced by Unweighted Pair Group Method with Arithmetic mean (UPGMA) with Bootstrap methods showed a diversity of virus isolates (Figure 4). The information has been used to select virus strains to challenge resistant pumpkin lines in a current breeding program designated to introduce resistance to viruses in pumpkins.
**Methods:** We used standard molecular protocols to sequence nuclear and mitochondrial loci from nearly 500 individuals representing all available *Anelosimus* species, emphasizing 8 focal S. American species. We analyzed the phylogenetic data using parsimony and Bayesian methods, and used standard and statistical parsimony to reconstruct haplotype networks. We calculated intraspecific sequence divergence using the dnadist Package of Phylip. We simulated lineage diversification in Mesquite to test for restricted diversification rates in social spiders.

**Results:** Phylogeny - The species level phylogeny reveals multiple origins of social behavior in cobweb spiders. Sociality is phylogenetically scattered and significantly reduces lineage diversification.

Genetic variability – The four social species have significantly less genetic variability than the four subsocial species, and these differences are most dramatic between social-subsocial sister species. *Anelosimus eximius* colony separation – We found 25 haplotypes among the 135 individuals sampled from 38 colonies. Haplotypes cluster neatly into colonies, most of which contain a single haplotype, or two closely related haplotypes. The haplotype phylogeny does not reflect the geographic distribution of colonies. We found no evidence for colony mixing, or dispersal between colonies.

**Discussion:** Phylogenetics suggest that almost every social *Anelosimus* species evolved sociality independently, Simulations show that social clades are less diverse than expected. Hence sociality can be characterized as an evolutionary dead end in spiders.

**Conclusion:** The evolution of sociality in spiders leads to lineage isolation and inbreeding, reducing genetic variability at the species level, and thereby restricting diversification of social lineages.

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Work done in collaboration with W. P. Maddison and L. Aviles
Microsatellite and Mitochondrial Markers Show Low Genetic Structure in the Elkhorn Coral of Puerto Rico

ABSTRACT

Fine scale genetic structure of the coral Acropora palmata was investigated by sampling 9 reefs in Puerto Rico and 1 in the Bahamas as an out-group. Of a total of 276 samples, 201 had unique genotypes and the remaining 75 were clones. 48 of these unique samples representing 4 reefs were also profiled for mitochondrial haplotypes, resulting in 12 unique mitochondrial haplotypes. The analyzed samples originated from one panmictic population, rejecting the hypothesis of eastern and western populations of A. palmata. AMOVA analyses also indicated that <1% of the variance in allelic frequencies was explained by differences between sampling localities. Genetic diversity was surprisingly high considering the observed demographic declines.

INTRODUCTION

● Principally as a result of White Band disease infections (Fig. 1), A. palmata, one of the main Caribbean reef-builder coral (Fig.2), has been reduced by 80-95% since the 1970’s.  
● Although the species shows encouraging signs of recovery and resistance to WBD, it is of utmost importance to assess its genetic diversity and genetic structure.

Using five highly polymorphic markers, Baums et al. (2005) showed that A. palmata is divided into two populations (eastern and western), with Puerto Rico proposed as a hybrid zone1. Only western reefs of Puerto Rico were sampled.

OBJECTIVE: Because of the apparent hybrid characteristics of elkhorn corals from Puerto Rico, we wanted to determine if finer local genetic structure existed around the island.

MAIN HYPOTHESIS: Population differentiation exists between reefs categorized as West and East populations.

Microsatellites

<table>
<thead>
<tr>
<th>Region</th>
<th>Reef</th>
<th>Samples Unique genets</th>
<th>N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mona</td>
<td>South/West</td>
<td>47</td>
<td>38</td>
</tr>
<tr>
<td>Culebra</td>
<td>South/West</td>
<td>19</td>
<td>16</td>
</tr>
<tr>
<td>Vieques</td>
<td>South/West</td>
<td>21</td>
<td>20</td>
</tr>
<tr>
<td>Mona</td>
<td>East</td>
<td>11</td>
<td>9</td>
</tr>
<tr>
<td>Vieques</td>
<td>East</td>
<td>21</td>
<td>20</td>
</tr>
<tr>
<td>Bahamas</td>
<td>East</td>
<td>22</td>
<td>20</td>
</tr>
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<td>Bahamas</td>
<td>West</td>
<td>15</td>
<td>18</td>
</tr>
<tr>
<td>Bahamas</td>
<td>West</td>
<td>16</td>
<td>22</td>
</tr>
</tbody>
</table>

Table 1. Number of samples and genets sampled from 10 locations. A genet is a coral genotype shared by natural clone coral colonies, a frequent event in A. palmata.

● 27.2% of our samples were clones, i.e. they had a genotype identical to another sampled colony within the same reef.

● Analyses in STRUCTURE (pK<0.05: 3.8365.5 ~ 0.2) and AMOVA (0.72% of variation explained among populations, Fst=0.00718, non-significant) indicated that our samples belonged to a panmictic population within the same reef.

● Consideration of recent decline of elkhorn corals.

● Previous work by Gardner et al. 2003 showed that the species shows encouraging signs of recovery and resistance to WBD, it is of utmost importance to assess its genetic diversity and genetic structure.

METHODS

Sampling

To reach our objective, we sampled 9 reefs evenly spread out around Puerto Rico and used samples from the Bahamas as an out-group. We selected the same five microsatellite markers described by Baums et al. (2005), with an additional mitochondrial marker.

DNA extraction

PCR of the mt control region

sequencing

PCR of 5 microsatellite markers

genotyping

Mitochondrial control region

Table 2. Analyses of Molecular Variation (n=201) including the portion of any variation in allele frequencies attributable to the reefs of origin of the samples in each haplotype.

CONCLUSIONS

● 27.2% of the samples were natural clones as indicated by our microsatellite markers.

● Genetic diversity at the studied loci was surprisingly high considering the recent decline of elkhorn corals.

● Population differentiation was not detected, even between reefs that were previously reported in either western or eastern regions.

● Future work will include samples from both eastern and western regions (Jamaica, the US Virgin Islands and Guadeloupe) to confirm these results.

● Rather than special conservation efforts to protect genetically unique reefs in Puerto Rico, global protection may be a better strategy to preserve elkhorn coral around the island.

Literature cited


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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus
Phylogeography and Conservation Genetics of Tabebuia heterophylla (DC.) Britton (Bignoniaceae)

Introduction

Tabebuia heterophylla (DC.) Britton (Bignoniaceae) is a charismatic neotropical tree species known for its attractive flowers and economically valuable wood. The species is considered to have an extremely wide distribution throughout the Caribbean (Figure 1), where it thrives in a diverse range of habitats, soil conditions, and altitudes (Gentry, 1992). In Puerto Rico, where it is very common, the species is found from dry to wet forest habitats (Figure 2), from sea level to high altitudes, and is morphologically highly variable (e.g., leaf size and leaflet number variation). However, it is not known whether the variation is due to phenotypic plasticity or whether there are genetically distinct populations and evolutionary lineages. The purpose of the project is to study the phylogeography and genetic diversity of natural populations of Tabebuia heterophylla through the use of DNA sequence and microsatellite data.

Chloroplast Phylogeny

In order to understand the phylogeography of the species, 10 previously tested chloroplast regions were used for a preliminary analysis. The geographic sampling included individuals sampled from populations in Mona, Guánica, Isabela, Quebradillas, and San Juan (Figure 4a). A total of approximately 5 kilobases of chloroplast sequence data were obtained for each sample. As seen on the chloroplast DNA gene tree (Figure 4b), preliminary results indicate a strong genetic differentiation between Mona Island populations and Puerto Rico populations.

Future Directions – Although the chloroplast DNA gene tree have shown very interesting results, additional sampling is needed from different populations throughout Puerto Rico, including the islands of Vieques and Culebra. Samples from Dominican Republic are also needed to see how they compare to the Mona Island populations which are geographically intermediate between Puerto Rico and the Dominican Republic.

Microsatellite Data

A microsatellite library was developed using a modified protocol by Tomas Hrbek et al. (UPR-Rio Piedras) and Glenn and Schable (2005). 21 out of 96 sequenced clones contained microsatellite regions. Primer pairs were designed for these potential regions and PCR testing showed 90% success. Testing for genotyping suitability led to the identification of 5 loci with potential polymorphism suitable for population analyses (Figure 5).

Future Directions - Further development of microsatellite sequences will be required to achieve a minimum of 10 polymorphic loci that will be used for the population analyses of the species.

Significance

The project will provide extremely important information about the evolutionary history and genetic diversity of an economically important native tree species commonly used for horticultural purposes and reforestation. The information may be crucial since Tabebuia heterophylla has in recent years suffered greatly from the predation of Holothrips tabebuia, a new gall-inducing species that cause severe leaf deformation (Cabrera and Segarra, 2008). Information about the genetic diversity of different populations and habitat types, ecological characteristics, and the possible correlations to severity of impact and resistance to infestation, may be critical for conservation purposes. In addition, genetic diversity information is important to determine the best seed source for reforestation projects.

Literature Cited

Genetic Characterization of Mona Island Feral Goats and Pigs using Microsatellite Markers

Introduction

Feral goats (Capra hircus) and pigs (Sus scrofa) were introduced more than four centuries ago by Spanish settlers to Mona Island. They were set loose to reproduce and provide a source of fresh meat for travelers to Mona and other the Caribbean islands. Pigs and goats can be found in a wide range of habitats ranging from wet to dry ecosystems. Both species can also survive in harsh environments due to various physiological and behavioral adaptations. Because of their ability to adapt to different environments (Moran-Fehr et al., 2004) the goat, as well as the pigs are expected to show genetic responses to the range of environmental conditions they experience (Galal 2005).

The use of microsatellite markers is one of the most powerful means to study genetic diversity and differentiation among populations. Microsatellite markers are highly polymorphic, randomly distributed throughout the genome and neutral with respect to selection (Agha et al., 2008). The characterization and of the genetic diversity of animals from Mona Island and other islands in the Caribbean will give us a better understanding on the ecological and evolutionary processes affecting the long term survival of the regionally adapted land races. The genetic characterization also provides us with data that can be used for more efficient management.

Objectives

Specific Questions:
1) What are the patterns of neutral genetic diversity in feral Mona goats and pigs, and how do these compare with other feral populations in the Caribbean, Europe and Africa?
2) Do feral goats and pigs of Mona Island represent unique landraces that may be useful for stock improvement of agricultural breeds?
3) What is the evolutionary and demographic history of the feral Mona goats and pigs of Mona Island?

Materials and Methods

Sample Collection

• 50 goat and 20 pig samples have been collected during hunting season in Mona (Figure 1).
• Samples consist of tissues collected during DNR sponsored culls and from fecal samples.
• Whole genomic DNA has been extracted using Qiagen DNA easy kits for all pigs and goats samples.

Microsatellite primers

• A standardized set of 50 pig and 30 goat microsatellite markers proposed by the European-funded EconoGene project (Russell et al. 2003, SanCristóbal et al. 2006) are being used.
• The amplified loci are being genotyped on the ABI 3130 and analyzed in GeneMapper Software Version 4 (Figure 2).

Preliminary Results

• 30 goat microsatellite primers have been tested. 23 primers have successfully amplified and an additional 7 are currently being optimized.
• 9 out of the 23 successfully amplified goat loci showed no variation, in a samples of 20 individuals tested (Table 1 **).
• 14 out of the 23 successfully amplified goat loci are polymorphic with 7 of these having 3 or more alleles per locus.
• The entire sample of Mona Island goats is currently being characterized.
• Pig loci amplifications are in progress.

Literature Cited

Development of Microsatellite Markers for Population Genetic Studies of Harrisia portoricensis and Related Species

Introduction

Harrisia portoricensis (fig.1-2) is a columnar cactus endemic to Puerto Rico. It was first collected and described from southern Puerto Rico, but urban and agricultural development extirpated these populations. At present, the species is restricted to the islands of Mona, Monito and Desecheo. In addition to its limited distribution, H. portoricensis is threatened by the activity of feral goats and pigs on two of these islands. These and other considerations motivated its inclusion in the U.S. Fish and Wildlife Service threatened and endangered species list (U.S. Fish and Wildlife Service 1990).

Microsatellites or Simple Sequence Repeat (SSR) markers are being widely used in plants for a variety of applications including the assessment of the genetic diversity of natural populations. Microsatellites are typically highly polymorphic and robust, and also have a high level of transferability to related species (Varshney et al. 2005). The use of these markers could prove very useful for the study of genetic structure, gene flow and the degree of inbreeding in populations of Harrisia portoricensis. These analyses could in turn provide insights into the factors and processes that affect the survival of the species, information that will be valuable for formulation of effective management and conservation strategies.

Objectives

1) To develop microsatellite markers potentially useful to assess various population genetics parameters in H. portoricensis.

2) To test the developed primers in other species of Harrisia in order to examine their transferability.

Materials and Methods

- Seven primer pairs successfully amplified microsatellite loci in all samples of H. portoricensis and the nine additional species.
- Six loci were monomorphic and just one polymorphic (with two alleles) for H. portoricensis (Fig. 3).
- Allelic variation was found among the different species of Harrisia (Fig. 4). Upto four loci were polymorphic for some of these species.

Results

The microsatellite markers developed in this study are the first markers for the genus Harrisia. Their transferability seems high since they easily amplify across a wide range of species. The markers appear to be much less variable in H. portoricensis than in other Harrisia species. More extensive sampling from different subpopulations and populations of H. portoricensis as well as the other species is needed in order to accurately characterize these markers and their conservation genetic value.

Conclusions and Future directions

We thank Alan Frank from the University of South Florida, and Rosalina Berazain from the National Botanic Garden in Cuba, for sending samples of the additional species of Harrisia.

Acknowledgements

We thank Alan Frank from the University of South Florida, and Rosalina Berazain from the National Botanic Garden in Cuba, for sending samples of the additional species of Harrisia.

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Department of Biology, University of Puerto Rico, Rio Piedras Campus

Funded by National Science Foundation, HRD #0734826 and the University of Puerto Rico, Rio Piedras Campus
The UPR Herbarium is the center of the scientific activity of the Botanical Garden. The herbarium preserves approximately 40,000 plant specimens from the Flora of Puerto Rico. Our plant specimen collection includes Type specimens and historical collections from the early 20th Century. The herbarium is a training and research facility for students and scientists studying themes like taxonomy, systematics, evolution, biogeography, conservation, and ethnobotany. Through workshops and talks the training extends to the community and citizens interested in science or simply nature. The UPR herbarium is a partner of local and international herbaria; it is in constant collaboration with local and US agencies dedicated to plant species conservation. Our collection is continually growing. Accordingly, we are evolving to make space for the future and to make our data available to the scientific community.

**Who are we?**

The UPR herbarium attracts students with different interests in biology (botany, conservation, ecology, horticulture and agriculture) and students from other backgrounds like art and marketing who are interested in nature and/or the museum aspects carried in the Flora of Puerto Rico. Our plant specimen collection includes Type specimens and historical collections from the early 20th Century. The herbarium is a training and research facility for students and scientists studying themes like taxonomy, systematics, evolution, biogeography, conservation, and ethnobotany. Through workshops and talks the training extends to the community and citizens interested in science or simply nature. The UPR herbarium is a partner of local and international herbaria; it is in constant collaboration with local and US agencies dedicated to plant species conservation. Our collection is continually growing. Accordingly, we are evolving to make space for the future and to make our data available to the scientific community.

**Training and Outreach**

The UPR herbarium attracts students with different interests in biology (botany, conservation, ecology, horticulture and agriculture) and students from other backgrounds like art and marketing who are interested in nature and/or the museum aspects carried in the Flora of Puerto Rico. Our plant specimen collection includes Type specimens and historical collections from the early 20th Century. The herbarium is a training and research facility for students and scientists studying themes like taxonomy, systematics, evolution, biogeography, conservation, and ethnobotany. Through workshops and talks the training extends to the community and citizens interested in science or simply nature. The UPR herbarium is a partner of local and international herbaria; it is in constant collaboration with local and US agencies dedicated to plant species conservation. Our collection is continually growing. Accordingly, we are evolving to make space for the future and to make our data available to the scientific community.

**Research**

- **History of Lichenology in Puerto Rico and Checklist of Lichens of Puerto Rico;** Joel Mercado and Dr. Eugenio Santiago-Valentin.
- **Phylogenetics, Diversification Patterns and Biogeography of the Caribbean Tabebuia Gomes ex D.C. (Bignoniaceae);** Nizka M. Martínez
- **Phenology of Plants from Hacienda La Esperanza Natural Reserve in Manati;** Dr. Eugenio Santiago and volunteers.
- **Patterns of endemicity of the Flora of Puerto Rico;** Dr. Eugenio Santiago
- **Early Floristic Accounts of Puerto Rico by Domingo Bello y Espinosa.** Dr. Eugenio Santiago in collaboration with Dr. Javier Francisco Ortega (FIU-FTBG) and Museo de la Naturaleza (Cabilde de Tenerife).
- **Aerobiology in the Tropics and the Importance of Aeroallergens in the Respiratory Health (asthma and rhinitis);** Elson Viruet, Dr. Benjamín Bolaños (UPR-RCM)

**Projects**

- **Databasing, Digitizing and Georeferencing of Specimen Collections.** These tasks are part of the NSF sponsored project BRCII that involves the merging and publication of the data from the three herbaria present in Río Piedras: UPR, UPRRP and SJ.
- **LAPI (Latin American Plants Initiative).** This international project involves the partnership of herbaria to build a database and images of Type specimens from Latin America.
- **Pollen Reference Collection;** Elson Viruet
- **Propagation of Rare and Endangered Plants of Puerto Rico.** This is an ongoing project that studies the germination and early development of rare plants. Species like *Pleodendron macranthum* (Bail.) Tiegh., *Harrisia portoricensis* Britton, *Eugenia woodburyana* Alain and *Juglans jamaicensis* C. DC. are successfully breed.

**Collaboration and Support**

- **Departamento de Recursos Naturales y Ambientales**
- **United States Fish and Wildlife Service**
- **Conservation Trust of Puerto Rico**
- **Florida International University**
- **Faichild Tropical Botanical Garden**
- **Universidad Autónoma de Santo Domingo**
- **New York Botanical Garden**
- **Museo de la Naturaleza (Cabilde de Tenerife)**
- **University of Washington – Seattle**
- **Universidad de Puerto Rico-Recinto de Ciencias Médicas (UPR-RCM)**

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**Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus**
**Introduction**

*Tabebuia* Gomes ex DC. (Bignoniaceae) is an extremely diverse Neotropical genus of woody species with great economical value and wide distribution from Mexico to Argentina. Although ca 108 taxa have been described (Gentry, 1992), approximately 61 are distributed throughout the Caribbean with 88% endemicity to the region (Figure 1). The genus also exhibits great versatility in morphological variation (e.g., flower color, unifoliolate to multi-foliolate), as well as adaptations to a wide range of ecological conditions (e.g., from cloud forest to arid zones, diverse geology, and high to low altitudes). However, the evolutionary relationships of the Caribbean species has still not been elucidated. The purpose of the study is to test the monophyly of the Caribbean group and evaluate the evolutionary relationships. In addition, the gathered information will help evaluate patterns of morphological evolution, adaptive-ecological diversification, as well as biogeographical patterns.

**Methods**

**Taxonomic sampling.** The preliminary analysis includes 19 species from the genus *Tabebuia* and outgroups representing different genera in the Bignoniaceae family.

DNA extraction, amplification, and sequencing. Genomic DNA was extracted from leaves using Dneasy Plant Mini Kit (Qiagen). PCR was used to amplify the nrITS region using the set of primers ITS-5ang, specific for angiosperm plants, and universal primer ITS-4. PCR products were cleaned and sequenced. Sequencher 4.9 (Gene Codes) was used for contig assembly and sequence editing.

**Phylogenetic analysis.** MEGA 4.0 (Tamura, Dudley, Nei, and Kumar, 2007) was used for the ClustalW multiple sequence alignment feature and phylogenetic analysis. The evolutionary history was inferred under Maximum Parsimony optimality criterion. The resulting bootstrap consensus tree inferred from 1000 replicates is taken to represent the evolutionary history of the taxa analyzed. Branches corresponding to partitions reproduced in less than 50% bootstrap replicates were collapsed.

**Phylogram**

Figure 1. Distribution of *Tabebuia* in the Greater Antilles and the Bahamas (number of species/number of endemic species).

Figure 2. Maximum Parsimony consensus tree. Branches with bootstrap support greater than 50% are shown above the branches. Branches with less than 50% are collapsed in a polytomy.

**Preliminary Results**

**Overview**

The preliminary reconstructed phylogeny (Figure 2) suggests that the genus *Tabebuia* is paraphyletic, congruent with chloroplast DNA sequence data (Grose and Olmstead, 2007). In addition, the data shown suggest a clade that consist of 3 continental species (*T. ochracea*, *T. guayacan*, and *T. impeditiglossa*). Another clade contains the sampled Caribbean taxa in close relationship with a pair of continental taxa (*T. rosea* and *T. aurea*).

**Cuba/Bahamas-Hispaniola relationship**

The two species sampled that occur in Cuba and the Bahamas (*T. bahamensis* and *T. lepidota*) are related to the sampled Hispaniolan taxa, with the exception of *T. ophiolitica*. However, Cuba/Bahamas-Hispaniola grouping has a low bootstrap support.

**Puerto Rico and eastern Caribbean taxa**

There is no robustness for grouping Puerto Rican species with the exception of the two endemic species that are distributed in the cloud forest ecological life zone (*T. rigida* and *T. schumanniana*). They differ by only one base pair position on the analyzed sequences. *T. heterophylla*, considered to be a highly diverse and widely distributed species throughout the Caribbean, including Puerto Rico, seems to have a close relationship with *T. pallida*, a species considered to be distributed in the eastern Caribbean and often confused with *T. heterophylla*.

**Caribbean group relationship with continental *T. rosea* and *T. aurea**

Two continental species come out in the Caribbean clade. *T. rosea* is imbedded in the clade (bootstrap support 100%). Although *T. aurea* also comes out as related to the Caribbean group, bootstrap support is weak. These patterns for *T. aurea* and *T. rosea* with the Caribbean taxa coincide with results from chloroplast sequence data (Grose and Olmstead, 2007).

**Future Directions**

- To continue the taxonomic sampling of the genus in additional geographical regions.
- Incorporate other nuclear and chloroplast regions to add resolution to elucidate the evolutionary relationships.
- Carry out ecological and morphological (e.g., leaves and flower) assessments of the taxa.

**Literature Cited**


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2- Botanical Garden Herbarium, University of Puerto Rico

3- Biology Department, Autonomous University of Santo Domingo, Dominican Republic

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Rio Piedras Campus
Introduction
Pollen is produced by the male cone or flower organ (stamen), and represents the microgametophytic generation in spermatophytes. Pollen morphology has proved to be of significant taxonomic utility in different research areas, such as plant systematics, paleobotany, ethnobotany, forensics, and aerobiology, among others. Today, there is no reference collection for pollen identification for Puerto Rico. This initiative aims to develop a pollen reference collection of the Island. At present, this collection supports two projects.

Pollen Research Projects

1. Diversification of Economically Important Caribbean Trees: This project is studying the evolution, diversification, and biogeography of the Neotropical genus Tabebuia by comparing morphological, ecological and molecular data. We are developing morphological descriptions of the pollen of species of Tabebuia as well as outgroups, and are developing a digital image bank to support pollen identification.

2. Aerobiology in the Tropics and the Importance of Aeroallergens in the Respiratory Health (asthma and rhinitis): Pollen and fungi spores are been considered possible allergens present in the Puerto Rico environment. We are working on an initiative to develop a pollen reference collection of pollen for common plant species in Puerto Rico. This reference collection will be a tool to identify pollen particles.

Methods

I. Sampling
Pollen is collected from wild plants, cultivated plants and herbarium specimens. Voucher specimens are deposited in the Herbarium of the UPR Botanical Garden (Index Herbarium: UPR).

II. Identification
Pollen is set on a microscope slide with gelatin stain (Calberla). The morphology is assessed under a Nikon Eclipse E200 microscope. Digital images are obtained with a Nikon DS-fi1 digital camera.

III. Pollen Reference Collection
Pollen descriptions and images are gathered and organized by family to develop a reference catalog.

IV. Aerobiology
Particles collected from the air, with the Burkard Automatic Volumetric Spore Trap, are analyzed for pollen presence. Pollen identification will be possible for the Department of Microbiology (UPR-RCM) with the Pollen Reference Collection.

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus
Sensing climate change in an ephemeral coastal pond: implications to the conservation of biodiversity in a subtropical dry forest

Abstract

There is concern that climate change may reduce rainfall in the Caribbean and increase sea levels thus altering groundwater supplies and coastal habitats. The potential effects of rainfall reduction and sea level changes will likely be more evident in dry lowland subtropical forests which are more susceptible to fires and to sea level changes. In order to improve management decisions, a systematic monitoring program of surface and groundwater levels was established in 2005 at an ephemeral coastal pond located in the Guánica Biospheric Reserve in Southwestern Puerto Rico. The program has provided invaluable data in support of conservation of endangered wildlife. The data suggest the ephemeral coastal ponds are critical habitats highly susceptible to climate change and to storm surges drastically altering habitat conditions and posing stresses to freshwater endangered fauna.

Hydrology of the Tamarindo Pond

The Guánica Biosphere Reserve (GBR) is located in southwestern Puerto Rico (Figure 1). Rain shadows, creates a subtropical dry forests with low rainfall (930 mm/yr) and high evapotranspiration (730 mm/yr). Despite low rainfall, episodic storms can create ephemeral ponds that become important wildlife habitats. The Tamarindo Pond is the largest freshwater pond of the GBR (full volume = 513 m³). In 2005, a water level monitoring program started at the Tamarindo Pond. Figure 3 shows a water budget developed based on the data gathered at the site. Water infiltration is considered negligible due to a thick clay layer located below the pond. In 2008, the monitoring program was expanded to monitor ground water level and salinity. Ground water data revealed that below the pond, the ground water is highly sensitive to tidal changes (Figure 4). However, the impermeable clay layer maintain freshwater conditions in this pond.

Coastal Sensitivity

In the Caribbean, climate change is expected to increase sea levels, reduce rainfall, and increase the severity of tropical storms and hurricanes. Freshwater coastal habitats, such as the Tamarindo Pond, are at high risk of increasing their salinity given their proximity to the ocean, their dependency on rain events, and vulnerability to storm surge events. We have documented the vulnerability of the Tamarindo Pond to coastal disturbances. In 2007, two hurricanes passed south of the site and generated higher storm surges that impacted the southwest coast of Puerto Rico (Figures 5 and 6). Figure 7 shows the effect of these disturbances on the water level dynamics of the Tamarindo Pond for similar calendar periods of different years. Note the cyclic pattern that appeared in water levels 2007 after the passage of the storms. This pattern was associated with tidal changes in the ocean (Figure 8). The fact that in 2008, water levels were back to normal (Figure 7), reveals the resilience of the Tamarindo Pond to coastal disturbances. However, as climate continues to change, is long-term monitoring of coastal habitats will be necessary to fully understand their resilience.

Climate change and the Puerto Rican Crested Toad (PRCT)

The occasional intense rain events that occur at the GBR fill the Tamarindo Pond and provoke massive breeding events of the PRCT (Peteophryne kerri). This species was thought to be extinct until 1984, when a breeding population was discovered at the Tamarindo Pond. Currently, the species is listed as threat under the Federal Endangered Species Act. The Tamarindo population is the only natural breeding population of the PRCT and has been carefully monitored since 1984. Successful metamorphosis occur under low salinity (<10ppt) and prolonged ponding (>28 days) (Figure 9). By analyzing historic rainfall records and tadpole population data, we have been able to link climate and the PRCT population in Guánica (Figure 10). Mass breeding of adults (>200) occurs during seasonal heavy rainfall events (>14 cm). Several events per year contribute to a greater mass breeding. Tadpoles successfully develop during summer or early fall before the pond gets too cold (<27°C), and when long ponding (20-30 days) provides suitable habitats. Record mass breeding takes place during over-the-average annual rainfall (>1.2 times). The predicted decrease in rainfall, sea level rise, and increase hurricane frequency for the Caribbean increase the risk of extinction of the PRCT population at the Tamarindo Pond. For this reason, several conservation entities are creating new habitats and reintroducing tadpoles reproduced in captivity (Figure 11). Through our applied limnological research, we have contributed to the habitat management and recovery of this species.

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Funded by National Science Foundation through the Center for Applied Tropical Ecology and Conservation of the University of Puerto Rico (CATEC)
Mycorrhizal specificity of the invasive orchid 

Oeceoclades maculata in Puerto Rico

Introduction

The terrestrial orchid Oeceoclades maculata is found throughout Puerto Rico. Originally from Africa, it has been called the most widespread orchid around the world. This success has been attributed to its tendency for self-pollination, but may also be due to its relationship with mycorrhizal fungi. Mycorrhizae contribute to plant success by absorbing nutrients and water from the soil. The fungi invade the cells of the root and make globular structures known as pelotons which are digested by the plant. To study this relationship and determine their level of specificity we focus on two techniques: identification of the mycorrhizal fungi present in mature roots by DNA sequencing, and measuring success of seed germination with different fungi. We asked: does O. maculata associate with a wide variety of mycorrhizal fungi or does it specialize? Does specificity vary within stages in development? Low specificity would help the orchid colonize new areas because it would not depend on a single group of fungi. High specificity could contribute to invasiveness if the fungal partner is very widely distributed.

Methods

Fungal isolation. Individual pelotons were removed from plants cells. Samples were examined with light microscopy and Rhizoctonia-like fungi were transferred to culture in Potato Dextrose Agar (PDA). In addition, some mycorrhizal roots were used for DNA extraction directly.

DNA sequencing for identification of fungi. DNA was extracted from pure culture and mycorrhizal roots and the ITS region were amplified by PCR with specific orchid fungal primers ITS OF1 and ITS OF4. The PCR product was purified and sequenced. To identify fungi, BLAST searches were used to find the most similar sequences in GenBank. Direct amplifications from roots allow detection of mycorrhizal fungi that do not grow in culture.

Symbiotic seed germination experiment. Seeds of O. maculata were sown on plates of cellulose agar previously inoculated with different mycorrhizal fungi. Control plates had seeds but no fungi. Each treatment was replicated six times. Plates were observed for seed germination every month for 10 months.

Results

<table>
<thead>
<tr>
<th>Closest relative in GenBank</th>
<th>Site</th>
<th>DNA Extraction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Psathyrella candolleana</td>
<td>El Yunque</td>
<td>Root</td>
</tr>
<tr>
<td>P. candolleana</td>
<td>Naguabo</td>
<td>Root</td>
</tr>
<tr>
<td>P. candolleana</td>
<td>Humacao</td>
<td>Root</td>
</tr>
<tr>
<td>P. candolleana</td>
<td>Rio Piedras</td>
<td>Root</td>
</tr>
<tr>
<td>P. candolleana</td>
<td>Mayaguez</td>
<td>Root</td>
</tr>
<tr>
<td>P. candolleana</td>
<td>Mayaguez</td>
<td>Culture</td>
</tr>
<tr>
<td>Ceratobasidium</td>
<td>El Yunque</td>
<td>Root</td>
</tr>
<tr>
<td>Ceratobasidium</td>
<td>San German</td>
<td>Root</td>
</tr>
<tr>
<td>Ceratobasidium</td>
<td>El Yunque</td>
<td>Root</td>
</tr>
<tr>
<td>Sparassia</td>
<td>Naguabo</td>
<td>Culture</td>
</tr>
<tr>
<td>Fomitopsis</td>
<td>Naguabo</td>
<td>Culture</td>
</tr>
<tr>
<td>Fomes mellea</td>
<td>Guanica</td>
<td>Culture</td>
</tr>
<tr>
<td>Tulasiella</td>
<td>Naguabo</td>
<td>Root</td>
</tr>
</tbody>
</table>

10-month old seedlings of Oeceoclades maculata from seeds inoculated with Psathyrella candolleana. Seeds inoculated with other fungi, and control seeds without fungi, did not germinate. From left: Camille Timossini, Carla Saladini, Rocio Garriga.

Conclusions

• Oeceoclades maculata is able to use a broad range of fungi as mycorrhizal associates during its adulthood; however, for its germination it needs association with a specific fungus, P. candolleana. P. candolleana is common worldwide, which increases the number of sites in which Oeceoclades seeds can germinate.

• O. maculata’s invasive spread in Puerto Rico may be partly attributed to its low specificity during its adult stage.

Acknowledgement

We’d like to thank CREST- CATEC, UPR and Nilibeth Hurtado.

Figure 1. Oeceoclades maculata. (Hart & Watts, 1822).

Figure 2. Cross-section of mature root of O. maculata. Many mycorrhizal pelotons are visible in the cortex.

Figure 3. Rhizoctonia fungi isolated from Oeceoclades maculata roots.

Figure 4. Specificity in seed germination. 10-month-old seedlings of Oeceoclades maculata from seeds inoculated with Psathyrella candolleana. Seeds inoculated with other fungi, and control seeds without fungi, did not germinate. From left: Camille Timossini, Carla Saladini, Rocio Garriga.

Figure 3. Phylogenetic tree of Basidiomycota. Positions of mycorrhizal fungi from O. maculata are shown at right.

Acknowledgement

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Crime scene investigation of recreational marine fisheries: The impact of shore-based recreational fishing of Queen Conch (Strombus gigas) in Culebra Island, PR

Abstract

Queen conch (Strombus gigas) is one of the most significant edible benthic invertebrate species through the Caribbean, but its populations have largely declined through time. Its recreational fishery impact still remains poorly documented. Crime Scene Investigation (CSI) approaches were used to examine shell dumps (“concheros”) along the shoreline in Culebra Island, PR, to test four hypotheses: (1) Conch is illegally captured within the Luis Peña Channel No-take Natural Reserve (LPCNR); (2) Illegal-sized shells constitute most of the fishery; (3) There is shell harvesting within the seasonal closure; and (4) There is no difference in shell parameters within and outside the no-take reserve.

Six replicate 50 x 4 m (200 m²) belt transects along or close to the shoreline were used to count and measure all recent empty shells along the shoreline at eight shores in Culebra Island (4 within LPCNR, 4 control sites outside) between 2007 and 2009. There was significant poaching of shells within the no-take reserve. Mean shell size was 12.1 cm within LPCNR, and 16.4 cm outside the reserve, and mean lip width was below 1.06 mm. More than 99% of the shells inspected failed to meet minimum size limits (Minimum shell length: 9” (22.9 cm); Minimum lip width: 3/8” (9 mm)). Evidence of recent illegal captures within the seasonal closure was also found, even within the no-take reserve. There was no difference in shell abundance among sites, even within the no-take reserve.

Shells were largely aggregated in specific remote locations, usually under tree shades. Crime scene locations are regular spots of difficult access, frequently used by poachers. Our study confirms that Conch is being continuously poached by recreational and/or artisanal fishers, even within the no-take reserve, suggesting the need of more aggressive education, patrolling and enforcement.

Methods

Six replicate 50 x 4 m belt transects were haphazardly established along the shoreline. Two treatments were tested: Luis Peña Channel No-Take Natural Reserve, and Control sites outside. Maximum shell length (±0.5 cm) and maximum lip width (0.01 mm) were measured on each sample. Data were log10-transformed and analyzed by means of a one-way ANOVA.

Results

One way ANOVA of total shell length and lip width

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Treatment</th>
<th>LPCNR</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shell Length</td>
<td>n= 368</td>
<td>132</td>
<td>132</td>
</tr>
<tr>
<td>Lip Width</td>
<td>n= 368</td>
<td>132</td>
<td>132</td>
</tr>
</tbody>
</table>

Site parameters are regular spots of difficult access, usually under tree shades. Crime scene investigation of recreational marine fisheries:

Conclusions

CSI evaluation of “concheros” showed that there was illegal conch harvesting: (1) of juvenile individuals; (2) during the seasonal closure; and (3) within the no-take reserve. There was also a fairly similar intensity of shore-based recreational fishing both within and outside the no-take reserve. Shells were significantly smaller within the no-take reserve, suggesting possibly major long-term impacts of poaching. More than 99% of the harvested shells were juvenile or sub-adult individuals. All shells found within the no-take reserve failed to meet minimum legal size. Presence of hidden “concheros” suggested that recreational fishing occurs at predictable sites that should be frequently patrolled. Lack of patrolling, enforcement and education are major issues affecting the conservation of Queen Conch stocks in Culebra Island, as well as in PR. There is a need to establish a long-term monitoring program to address ecological change in seagrass communities and conch populations. Future legal seasonal closure periods need to be reviewed.
Abstract

Mona Island, located nearly halfway between La Española and PR, is considered a unique and pristine natural wonder in the Caribbean. Its coral reef ecosystems are paramount for maintaining the meta-population connectivity of most commercially-important fish species in the region. But an unprecedented sea surface warming event occurred during 2005 throughout the northeastern Caribbean Sea that caused a mass regional coral bleaching event. It was followed by significant coral mortality. This study is the result of an exploratory expedition carried out in June 2006 by Sociedad Ambiente Marino and the UPR-Coral Reef Research Group to conduct a rapid ecological assessment to address what was the impact of the bleaching and coral mortality event in Mona Island. Digital video-imaging was used to document the status of benthic community structure at six locations.

Coral reefs were characterized by a dramatic phase shift favoring macroalgal-cyanobacterial dominance. Percent coral cover ranged from 3 to 14%, when estimates conducted between 1981 and 2000 showed % coral cover values ranging from 10 to 35%. Percent macroalgal cover ranged from 42 to 85%, often dominated by unpalatable brown algae, Dicyota spp. Percent cyanobacterial cover ranged from 0.3 to 13%. Most large reef-building coral species were showing significant signs of mortality. There was also a significant difference ($p=0.0002$) in community structure among sites, which were clustered in four different patterns according to degree of mortality and post-mortality trajectory. Differences were actually attributed to the high frequency of other benthic categories such as recently dead corals (RDC), crustose coralline algae (CCA) and rubble (SPR). This is the first known mass coral mortality event reported in Mona Island and represents an unequivocal sign of climate change impacts.

Methods

Data collection and analysis: A total of 73 digital video-transects were randomly filmed at 6 different locations in Mona Island during June 2006. Five replicate digital images were randomly selected from each transect and analyzed for benthic component cover (i.e., coral, algal functional groups, sponges, others) using CPCE v3.4. Data was analyzed using multivariate statistical approaches.

Results

There was a significant difference ($p=0.0002$) in community structure among sites, which were clustered in four different patterns according to degree of mortality and post-mortality trajectory. Differences were actually attributed to the high frequency of other benthic categories such as recently dead corals (RDC), crustose coralline algae (CCA) and rubble (SPR). This is the first known mass coral mortality event reported in Mona Island and represents an unequivocal sign of climate change impacts.

Conclusions

There was a significant coral mortality event in Mona Island following prolonged mass coral bleaching as a result of unprecedented sea surface warming during 2005-2006. Benthic community structure shifted from coral to macroalgal-cyanobacterial dominance. Most colonies of large reef-building species suffered significant tissue mortality, thus dangerously compromising their reproductive output. Under the forecasted trend of sea surface warming and ocean acidification, particularly under overfished conditions, this may compromise net reef accretion rates. If these types of events become recurrent in the near future, coral reef ecosystem resilience might be significantly impaired and coral reefs might be about to suffer an entire ecosystem collapse with potential negative consequences to regional fisheries due to a connectivity loss.
Strengthening Educational Capacities in Geospatial Science and Technology for Agricultural and Natural Resources Management

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Funded by USDA CSREES, # 2008-38422-19211 and University of Puerto Rico, Río Piedras Campus

OBJECTIVES

“Enhance educational capacity in geospatial science in agriculture and natural resources, via infrastructure and curriculum development.”

- To establish a state-of-the-art GeoSpatial Laboratory at UPR-RP library for instruction and research.
- To reshape a course on Geospatial Analysis in Natural Resource Management and Conservation, offered to undergraduate/graduate students in Environmental Science and Biology at UPR-RP.

ACTIVITIES

- Development of Systematic Educational approach Via Cooperation
  - Curricula development;
  - Advanced training program at TAMU;
  - Student Summer Internship; and
  - Workshops for faculty to disseminate training materials and extend our model to other Hispanic-Serving Institutions.

- GeoSpatial Laboratory
  - Computer network including one high-speed server and 18 computer workstations;
  - Geospatial software licenses;
  - Standard software;
  - High-resolution plotter; and
  - Hand-held GPS.

EXPECTED IMPACTS

The project will impact at least 50 UPR-RP students and faculty, improving the quality of their educational experience, and building the institution’s capacity to attract and retain outstanding underrepresented students.

Specifically, a public geospatial lab in UPR-library will be established and benefit students/faculty accessible to UPR-library. A curriculum on Geospatial Analysis in Natural Resource Management and Conservation will be reshaped and offered each year. Training program will be offered to 10 students and 2 faculty each year at TAMU. Five students will have summer internships at both USDA Forest Service and UPR-RP. One graduate student will be supported via research assistantship.

OBJECTIVES (con.)

- Increase outstanding opportunities for student experiential learning and faculty preparation via advanced training program (twenty students and four faculty) at TAMU and student summer internships (five interns) at IITF of USDA Forest Service and UPR-RP”.
- “Diversify student populations entering and succeeding in agriculture and natural resources fields by recruiting and retaining Hispanic students, and preparing them for successful careers in these fields.”
- “Develop long-term cooperation among Hispanic-Servicing Institution (UPR-RP), top tier institution in agriculture (TAMU), and USDA (Forest Service) beyond the project, educating tomorrow’s Hispanic leaders in USDA and academia.”

EVALUATION

- The extent to which the scientific instrumentation and education capacity for geospatial analysis is enhanced at UPR-RP through course development and implementation of laboratory facility that operate at an optimal level;
- The intensiveness and effectiveness of use of new instrumentation by researchers and students for studies and research projects in key areas to USDA priorities;
- Satisfaction from student and faculty survey on project activities; and
- Extent to which the participation enhances motivation and involvement in education and research in the area of natural resource management and conservation.
Introduction

Organic matter is classified in groups, and total organic carbon (TOC) is the sum of dissolved organic carbon (DOC) and particulate organic carbon (POC). DOC composes most of the TOC and therefore it is our main focus.

Puerto Rico has been showing an increase in urbanization, especially in the San Juan Metropolitan Area, which is decreasing the extensions of riparian zones. This in turn reduces the natural inputs of allochthonous organic matter to rivers and are being replaced by organic loads from wastewater and urban runoff.

The purpose of this project is to characterize the quality of organic carbon in an urban tropical river. Studies have shown that organic carbon originating from sewage and physical processes decrease its lability.

Study Site

The project took place in the Río Piedras watershed in the San Juan Metropolitan Area of Puerto Rico. This is an example of an urban tropical river with an urban coverage of 63%.

Methodology

A. Field methods:
- Sampling dates: November 2008, January and April 2009
- Location: Five different sites (Figure 1)
- Sample collection for analysis of: BOD (triplicates)
  DOC (data not shown)
  COD (data not shown)

B. Data analysis

Thomas graphing method: Subtract daily DO from Day 0; This subtraction is used in equation: (t/BODt)^(1/3) and plot.

From linear regression use slope (b) and y intercept (a) in equation: k=6(b/a)

Results

Table 1. Average biodegradability constants (k, n=3) calculated for each of the sampling sites. Standard deviations for the means are shown in parenthesis.

<table>
<thead>
<tr>
<th>Site</th>
<th>Order</th>
<th>Elevation, ft amsl</th>
<th>Biodegradability constant, d⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>RP Sector Mocelo</td>
<td>1</td>
<td>719</td>
<td>0.517 (0.036), 0.220 (0.015), 0.270 (0.060)</td>
</tr>
<tr>
<td>RP Chiclana</td>
<td>1</td>
<td>356</td>
<td>0.564 (0.014), 0.154 (0.013), 0.095 (0.049)</td>
</tr>
<tr>
<td>RP Barbara Ann</td>
<td>1</td>
<td>166</td>
<td>0.135 (0.003), 1.508 (0.208)</td>
</tr>
<tr>
<td>RP UMET</td>
<td>3</td>
<td>46</td>
<td>0.461 (0.014), 0.180 (.004), 0.290 (.005)</td>
</tr>
<tr>
<td>RP Cuartel General</td>
<td>4</td>
<td>10</td>
<td>0.266 (0.003), 0.669 (.028), 0.507 (.143)</td>
</tr>
</tbody>
</table>

Discussion

During low flows, the biodegradability of organic carbon was significantly higher (Walsh t-test; P < 0.05) in the upstream stations than further downstream (Figure 3). During high flows, the biodegradability significantly increased downstream (Walsh t-test; P < 0.05) as seen in Figure 4. Regardless of flow, the mean biodegradability constants increased downstream but not significant (Figure 5). Also as riparian vegetation decreases, biodegradability increases as demonstrated in figure 6. The range of the biodegradability constants of organic carbon along the Rio Piedras compares to that of polluted/treated waters and raw sewage (Thomas 1950).

Conclusions

The observed low biodegradability downstream during low flows could be associated to allochthonous inputs of organic carbon generated upstream which biodegrades as it travels downstream becoming more recalcitrant. During high flows, surface runoff and combined sewer flows discharge organic carbon into the river, increasing the biodegradability downstream. However, in tropical urban watersheds, these patterns can be altered by random and sudden organic carbon (of varying biodegradability) inputs from any point source as occurred in the April sampling (Table 1). Thus, the organic matter fluxes in the urban Rio Piedras are highly dynamic and sensitive to flow conditions. This projects has shown that with some modifications, the Thomas graphing method, which was developed originally to characterize wastewaters, can be useful in characterizing the lability of organic matter in natural bodies of water.

Table 2. Typical values for the biodegradability constant (k) according to Thomas (1950).

<table>
<thead>
<tr>
<th>Sample</th>
<th>K (20°C) (d⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw Sewage</td>
<td>0.35-0.70</td>
</tr>
<tr>
<td>Well-treated sewage</td>
<td>0.12-0.23</td>
</tr>
</tbody>
</table>

Bianca Rodriguez Cardona and Jorge R. Ortiz-Zayas, University of Puerto Rico, Río Piedras Campus

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus
Plant-Insect Interactions: Do herbivores restrict the reproductive output of *Spathoglottis plicata*?

Introduction

Many exotic plants have naturalized in Puerto Rico, some of which may pose a threat to native species. One such exotic is *Spathoglottis plicata*, an orchid native to Southeast Asia. Popular as an ornamental, this orchid has thrived in disturbed areas in many parts of wet tropics. In Puerto Rico, *S. plicata* likely escaped from gardens and nurseries, and now thrives in two flower color variations, white and magenta.

Exotic species often lack natural predators, often giving the plants an advantage over the native flora. However, we found that beetles feast on the flowers and fruits of *Spathoglottis plicata*, which affects reproductive success of the orchid. Ant—plant interactions involving native species often are mutualistic: energy for protection. We have occasionally observed ants attacking the beetles on the inflorescence. Here we ask whether the beetles significantly alter reproductive success of *S. plicata* and if the ants, when present, are effective beetle deterrents.

Objectives

- Assess beetle damage and monitor the production of fruits and flowers of *S. plicata*.
- Evaluate the effects of each insect group on the reproductive success of *S. plicata*.

Hypothesis

We expect fruit set to be the following: Total exclusion = Beetle exclusion > Ant exclusion = No exclusion.

Experimental Design

We established 4 treatments groups (see table on the right). Each group will have a minimum of five plants, each with a developing inflorescence. These will be monitored weekly. To evaluate the individual effects of each insect group, ants will be excluded applying Tangle Trap paste to the base of each inflorescence. Beetles will be excluded placing a fine-mesh net bag over the inflorescence (Inf.).

<table>
<thead>
<tr>
<th>Figure 3</th>
<th>Name</th>
<th>Excluded Factor</th>
<th>Type of Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>No-Exclusion</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td>B</td>
<td>Ant-Exclusion</td>
<td>Ants</td>
<td>Tangle Trap paste</td>
</tr>
<tr>
<td>C</td>
<td>Beetle-Exclusion</td>
<td>Beetle</td>
<td>Net</td>
</tr>
<tr>
<td>D</td>
<td>Total Exclusion</td>
<td>Ants &amp; Beetles</td>
<td>Tangle Trap paste &amp; Net</td>
</tr>
</tbody>
</table>

- Dependent variables will be flower & fruit damage, and fruit set
- Data will be analyzed using ANOVAs

Study Site

The study site is in north-central Puerto Rico, along side Route 10, km 72.9 and 73.3. The area is known as the “Mogotes”, characterized by limestone “haystack” hills. Highest altitude is 300 m above sea level, with an annual temperature of 25°C.

Reference


Acknowledgements

Many thanks to Nadia Paola Flores Saldana for her help in producing this poster. Special thanks to my undergraduate companions Alvaro Bravo and Wilnelia Recart for field assistance during the initial set-up of the experiments.

Carlos J. Vega, James D. Ackerman & Ana A. Cuevas Padro.
University of Puerto Rico, Rio Piedras Campus, Department of Biology

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
Combined impacts of sea surface warming, local runoff pulses, and meso-scale Caribbean gyres on coral reef rehabilitation success: Lessons learned to cope with climate change

Abstract

Low-tech coral aquaculture methods have been successfully used in Culebra Island, PR, since year 2003 to propagate threatened staghorn coral (Acropora cervicornis). Harvested corals are being used through the Culebra Island Community-Based Coral Aquaculture and Reef Rehabilitation Program to restock their populations and rehabilitate bomb-cratered coral reefs. But an unprecedented sea surface warming event occurred during 2005 that caused extensive mortalities in Acroporid corals. It was followed by mass coral bleaching, runoff pulses, meso-scale gyres, and further mortality. This study documented what was the response of transplanted corals to these events.

Low-tech coral transplanting was a successful tool to propagate warm water-resistant genetic strains of corals. Most colonies survived the warming event and exhibited growth rates several orders of magnitude faster than wild populations. But they were susceptible to post-bleaching mortality due to White Band Disease Type II-like outbreaks which were susceptible to post-bleaching mortality due tougged strains of corals. Growth rates were markedly 10-fold from background 0.1-0.3 mg/m3. Right: Runoff and recurrent increases in meso-scale nutrient pulses and meso-scale gyres, and further mortality. This study documented what was the response of transplanted corals to these events.

Methods

A total of 105 warm-water resistant fragments of A. cervicornis were transplanted to Punta Soldado coral reef using masonry nails and plastic ties as part of a major coral farming and reef rehabilitation project. Colonies survival and growth rates were monitored through time before and after a major sea surface warming and bleaching event. Another cohort was also transplanted and monitored before and after the impact of a major meso-scale gyre during 2009. Impacts of these events were evaluated.

Meso-scale gyres and major local runoff pulses bring significant nutrient pulses to coral reefs. Left: Meso-scale gyre from an Amazon River water plume (April 23, 2009) that lasted over a month. Chlorophyll a (Chl-a) concentrations increased up to 10-fold from background 0.1-0.3 mg/m³. Right: Runoff pulse event associated to major rainfall (Nov. 23, 2003) that produced a 2 to 5-fold increase in Chl-a concentrations.

Results

Coral mortality events have been largely associated to rainfall (= recurrent runoff pulses) and sea surface temperature anomalies in Culebra Island since 2003.

Conclusions

Low-tech coral farming is a paramount tool to restock depleted coral populations, for habitat enhancement and restoration, to rehabilitate reef ecological redundancy, and for landscape reconstruction. However, the interaction of sea surface warming, sediment- and nutrient-laden runoff pulses, and recurrent meso-scale gyres have resulted in significant coral mortality. Runoff pulses have often ephemeral impacts, but poor land use and deforestation have enhanced their frequency and severity. Gyres can substantially increase dissolved nutrient concentrations up to 5 to 10-fold over periods of one month or more with a link to coral mortality. Overfishing has resulted in indirect increases of coral predator densities, further enhancing mortality. Adaptive responses in coral farming and reef restoration will be critical to keep up with climate change stress in the near future.

Edwin A. Hernández-Delgado, Raisa Hernández-Pacheco, Tadgí Ruíz-Morales, University of Puerto Rico, Dept. Biology, CATEC, Coral Reef Research Group, coral_giac@yahoo.com/ Jaime Fonseca, Iván Olivo, Samuel Suleimán, Sociedad Ambiente Marino/Mary Ann Lucking, Coralations
A comparison of fine root density of plants in the Coastal Plateau of the Guánica Dry Forest Reserve

Introduction

Root production is an important process in the dynamic carbon and nutrient cycle of terrestrial ecosystems. Root productivity of plant community can be affected by temperature, rainfall and availability of nutrients. In dry forests, root density within the soil system depends on seasonal resources availability, age and soil properties (Pavon and Briones, 1998).

In tropical dry forests experience seasonal drought. This seasonality is more pronounced in regard to dry and wet periods than to seasonal fluctuations in temperature (Castellanos et al., 1991). Due to the lack of information on how biomass of these forests respond to the seasonality of rainfall (Murphy and Lugo, 1986), our study focuses on understanding of root production by plants in the Guánica Dry Forest as a function of water availability. The Guánica Dry Forest is located in south western Puerto Rico (Fig 1). The rainfall recorded here over the period of 70 years varied from 500 mm to 1500 mm. The soils of the Guánica Dry Forest normally exhibit a deficit of water, ten out of twelve months of the year and seasonal climatic conditions indicate run-off only during the months of September and October (Lugo et al., 1978).

Considering the great variation in water availability we hypothesized that the fine root density (<2mm) of plants in the Guánica Dry Forest will reflect the seasonal patterns of rainfall, with an increase in root density during the rainy season.

Methodology

- In February, March, May and July (dry season) and September and October 2008 (wet season) samples of fine roots were collected from 3 trees of 5 species (n=15). (Coccoloba microstachya, Eriotheca fruticosa, Ficus citrifolia, Pisonia Albida and Tabebuia heterophylla).
- Surface soil within a 100 cm² quadrat was collected down to bedrock, under the canopy of the dwarf trees. The soil depth was very shallow and irregular with maximum depths of 10 cm in crevices.
- All root material was sorted and separated by sieves and hand sorting.
- The root samples were oven-dried at 60 C for 72 hours, weighed and root density (g/cm³) calculated.
- We transformed total root density data and ran a two way ANOVA [SigmaStat Version 3].

Results and Discussion

Table 1: Total Root Density (g/cm³) for each species during the sampling months

<table>
<thead>
<tr>
<th>Species</th>
<th>Feb</th>
<th>Mar</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>Aug</th>
<th>Sep</th>
<th>Oct</th>
<th>Average</th>
<th>SD</th>
<th>SOE</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. microstachya</td>
<td>0.042</td>
<td>0.015</td>
<td>0.021</td>
<td>0.023</td>
<td>0.003</td>
<td>0.005</td>
<td>0.018</td>
<td>0.014</td>
<td></td>
<td></td>
<td></td>
<td>3.914</td>
<td>0.000</td>
</tr>
<tr>
<td>E. fruticosa</td>
<td>0.008</td>
<td>0.007</td>
<td>0.026</td>
<td>0.011</td>
<td>0.004</td>
<td>0.009</td>
<td>0.011</td>
<td>0.008</td>
<td></td>
<td></td>
<td></td>
<td>3.914</td>
<td>0.000</td>
</tr>
<tr>
<td>F. citrifolia</td>
<td>0.010</td>
<td>0.017</td>
<td>0.014</td>
<td>0.026</td>
<td>0.013</td>
<td>0.008</td>
<td>0.013</td>
<td>0.007</td>
<td></td>
<td></td>
<td></td>
<td>3.914</td>
<td>0.000</td>
</tr>
<tr>
<td>P. albida</td>
<td>0.017</td>
<td>0.014</td>
<td>0.017</td>
<td>0.007</td>
<td>0.007</td>
<td>0.006</td>
<td>0.012</td>
<td>0.005</td>
<td></td>
<td></td>
<td></td>
<td>3.914</td>
<td>0.000</td>
</tr>
<tr>
<td>T. heterophylla</td>
<td>0.012</td>
<td>0.012</td>
<td>0.013</td>
<td>0.012</td>
<td>0.008</td>
<td>0.006</td>
<td>0.011</td>
<td>0.003</td>
<td></td>
<td></td>
<td></td>
<td>3.914</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Although there was variation in total root density (Table 1), we found no significant differences among species (Fig. 2.)

We found significant changes in root density for all species during the sampling period (F = 3.914, df = 5, p ≤ 0.012). There was an increase in root density for all species in May and a decrease of root density for all species in September. Although other studies found an initial increase of root density after rainfall (Pavon and Briones, 1998), our study found a decrease in root biomass when highest rainfall was recorded (414 mm) in the Guánica Dry Forest (Fig. 3 & 4). We suspect that the timing of our sampling did not coincide with the flushing of root production but rather with the increase in decomposition of dead roots which resulted in lower root biomass for the rainy season.

Conclusions

Plants in the Guánica Dry Forest respond to changes in rainfall. Other environmental factors such as nutrient availability and timing of decomposition must be considered for future work. Our results are part of a broader study, expected to be carried out in the next 5 years. The data generated in this study is essential to understand nutrient dynamics and productivity of the ecosystem in order to predict how species respond to climate change.

Reference Cited


Acknowledgements

We gratefully acknowledge the Department of Natural and Environmental Resources, DNER, of the Government of Puerto Rico, especially Mr. Miguel Canals, Forest Manager of the Guánica Dry Forest Reserve.

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Figure 1: A Puerto Rico. B) Map of Dry Forest of Guánica.

Figure 2: Total root density among five species in the Guánica Dry Forest

Figure 3: Temporal changes in total root density for five species collected.

Figure 4: Rainfall measured in Guánica during sampling year.
It is important to understand and study the different adaptations and responses of plants changes in climate, especially, those living in arid environments where high temperatures, low precipitation, soil depth, soil-moisture availability, among others determines forest productivity, growth characteristics, water loss and physiognomy (1).

Under conditions when plants are not stressed plants preferentially fix 12C, and the fixed CO2 becomes 13C depleted. Under conditions where the plant is stressed it increases 13C fixation as a result of stomatal closure and enzymatic fractionation (2). Therefore, we hypothesize that δ13C of leaf tissue will reflect month to month changes in water availability as a function of the precipitation.

In this research we studied mangroves in the Guánica Dry Forest Reserve to understand the seasonal changes of water use efficiency by plants in relation to water availability. The natural abundance of isotopic carbon (δ13C) of leaf tissue was used as a measure of long term water use efficiency (3).

METHODOLOGY

• Three trees of each species of mangrove (Laguncularia racemosa, Avicennia germinans and Conocarpus erectus) were studied at the Guánica Dry Forest from December 2007 to January 2009.

• Field measurements required collecting samples of leaf, stems, soil samples, groundwater and precipitation, once per month.

• In the laboratory we oven-dried leaves at 60°C, and ground them to a fine powder. The soil, water and stem samples were stored at a temperature below 4°C.

• Water (δD and δ18O) and carbon (δ13C) isotope analyses were done using the standard protocols of the Stable Isotope Laboratory in the Department of Biology in the University of Miami (Florida).

RESULTS AND DISCUSSION

Precipitation patterns are characteristic of this forest with dry seasons bringing high amounts of precipitation during the wet months. There were significant differences in δ13C of adult leaves of A. germinans from L. racemosa and C. erectus, indicating that Avicennia leaves were more water-stressed than the leaves of the other two species. The proportion of groundwater used by the three species of mangroves reflect a difference in patterns of water uptake (Fig. III). A. germinans used more groundwater than C. erectus but had higher δ13C values in their leaves indicating other factors such as biochemical content, leaf expansion, and salinity of water used may affect δ13C in leaves (5). L. racemosa shifts water uptake from ground water in dry season to surface water during the rainy season. This suggests that both A. germinans and L. racemosa are deeper rooted while C. erectus has superficial roots and therefore depends more on precipitation.

Seasonal values of δ13C in adult leaves differed for each species. The decrease of δ13C in all species during rainy season reflect the decrease in water stress. When comparing precipitation patterns with seasonal patterns of δ13C (Fig. IV), we observed that δ13C decreased from June 20th to July 29th for both A. germinans and L. racemosa when precipitation decreased, however C. erectus had no leaves in this sampling period. C. erectus had higher values of δ13C in the sampling period, July 30 to August 21st, than the other two species because this species started to produce new leaves which explains the higher δ13C values (6).

CONCLUSION

We conclude that the three species display an adjustment in δ13C in relation to water stress in the Guánica Dry Forest.

Acknowledgements

We gratefully acknowledge the Department of Natural and Environmental Resources, DNER, of the Government of Puerto Rico, especially Mr. Miguel Canals, Forest Manager of the Guánica Dry Forest Reserve.

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Where is the water coming from in Puerto Rico?
Temporal and regional dynamics of precipitation in Puerto Rico as determined by isotopic signatures of δ¹⁸O and δD.

INTRODUCTION

- The annual amount, pattern, frequency and intensity of precipitation determine ecosystem dynamics and ground water recharge. Global climate change and regional climate variability are influencing precipitation dynamics in our region. To what extent are these factors determining the eco-hydrology of the island needs to be assessed. The location of Puerto Rico in the upper northeastern portion of the Caribbean and the Central Mountain range that divides the island results in the island being affected by both northeasterly trades in the northern part and southeasterly trades in the south. Therefore the origin of precipitation events can vary depending on geographic location, time of the year and strong meteorological events. In general the natural abundance of δ¹⁸O and δD in precipitation fluctuate with latitude and altitude, seasons and by unusual meteorological events such as monsoons, hurricanes and tropical storms. Therefore we can utilize the variations of the natural abundance of δ¹⁸O and δD in precipitation to trace the origins of precipitation and groundwater recharge.

- We analyzed the sources of precipitation on two contrasting sites in Puerto Rico: Northeastern (Rio Piedras) and southwestern region (Guánica Dry Forest). As they represent the moist region (1800 mm annual precipitation) and the semiarid region (700 mm annual precipitation) in the island (Fig. 1). Rio Piedras and Guánica have a bimodal pattern, usual for the Caribbean region, with precipitation peak in May-June and relative minimum in December-March and July-August. The Guánica Dry Forest, however has an unpredictable year annual and monthly precipitation pattern.

- We hypothesized that the isotopic signatures (δ¹⁸O and δD) of precipitation in the Guánica Dry Forest (GDF) (southwest) and Rio Piedras (RP) (northeast) will reflect the sources and amount of precipitation. We also tested the hypothesis that the isotopic signature of groundwater in the Guánica Dry Forest will be similar to the isotopic signature of precipitation because there will be very little evaporation prior to infiltration as Guánica Dry Forest is found on highly permeable limestone bedrock with little soil to retain precipitation.

METHODS

- We collected precipitation and groundwater samples monthly in the GDF from December 2007 to January 2009 and precipitation samples for RP from April 2008 to January 2009.
- Samples were sent to the University of Miami for analysis of δ¹⁸O and δD.

RESULTS AND DISCUSSION

- Temporal changes of δ¹⁸O in GDF are significantly variable suggesting that air masses carrying moisture to this area travel under unpredictable conditions throughout the year, while the non-significant differences among sampling periods for RP suggest similar conditions of humidity in the air masses, except under unusual meteorological events such as tropical storms that pass the island (Fig. 3).

- Nested periods of high cumulative precipitation recharge the groundwater in GDF as is reflected by the similar isotopic signature between the two. (Fig. 4).

CONCLUSIONS

- Precipitation in Puerto Rico originates from the mixing of different air masses under conditions of varying sea surface temperatures, relative humidity and winds except under extreme meteoric conditions such as tropical storms and strong fronts.
- Origin of precipitation is different between the northeastern site and the southwestern site of the island. The northeastern winds influence the precipitation of the northeast coast. The Central mountain range creates a rain shadow effect that limits the amount of precipitation originating in the north to reach the southwestern part of the island.
- The south western part of the island depends on the southeastern winds bringing precipitation to the area. Short, small precipitation events have a more positive isotopic signature indicating higher temperature and evaporation conditions in the Caribbean Ocean.
- The dominance of southeasterly rains in the Guánica Dry Forest establishes a strong limitation of water availability for ecosystem dynamics and ground water recharge.
- Ground water recharge in the Guánica Dry Forest take place only when precipitation is above 90 mm which corresponds to Tropical Storms or strong fronts conditions.
- The dependency of ground water dynamics on extreme weather phenomena increases the vulnerability of dry forests to the predicted drying of the Caribbean.

References Cited


Acknowledgements

We would like to acknowledge the support of the Department of Natural and Environmental Resources of Puerto Rico, especially the staff of the Guánica Dry Forest Biosphere Reserve.

Govender Y1,2, Cuevas, E 1,2, Sternberg, L., & Canals, M. Center for Applied Tropical Ecology and Conservation, University of Puerto Rico P.O. Box 23360, San Juan, Puerto Rico, 00931-3360 Department of Ecology, University of Puerto Rico, P.O. Box 23360, San Juan, Puerto Rico, 00931-3360 Department of Biology, University of Miami, Coral Gables, Florida, 33124

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
Introduction:
A research project is being proposed to study the relationship between land use and the hydrology of the Southern Aquifer between Santa Isabel and Salinas, Puerto Rico. Land use change in the area will be studied from 1930 to present. The water cycle will be carefully studied in there areas of different land use in the Santa Isabel area: forest, urban, and active cropland. The proposed research will built on the existing hydrologic knowledge generated by the United States Geological Survey (USGS). Because socio-economical factors typically drive water and land use decisions, a socio-ecological approach will be applied to assess community-perceptions on natural resources valuation.

Problem:
Excessive water extraction and an increased in urban areas have contributed to the hydrologic alteration of the Southern Aquifer (Rodriguez and Gomez-Gomez 2009; Figures 1 and 2). The high and constant water extractions are linked to aquifer salinization and to the deterioration of water quality which has historically meet the water demands of the local communities in the Santa Isabel and Salinas area. Groundwater salinization has also led to the closure of agricultural wells. For example, interviews conducted at Martex Farms, a grower of tropical fruits in the Santa Isabel area, revealed the closure of some of their wells due to aquifer salinization.

Objectives:
1. To study the effect of land use change on the water fluxes in the recharge areas of the Southern Aquifer.
2. To generate a model of the water cycle in the Santa Isabel and Salinas areas showing the effects of external stressors.
3. To assess community-perceptions on the value of groundwater resources in the Santa Isabel-Salinas area.
4. To develop educational material to inform the community of the short and long term effects of excessive water extractions on human health and on the local economy.

Potential methodology:
1. Develop a study of land use change based on aerial photographs to analyze land cover changes in the Santa Isabel and Salinas area.
2. Analyze historical trends in the aquifer water quality based on published data from wells in the area.
3. Develop a synoptic groundwater monitoring program on existing wells located in the Salinas and Santa Isabel areas and quarterly measure water quality parameters such as: pH, specific conductance, water table levels, salinity, nutrients and turbidity.
4. Set up an intensive water cycle monitoring program over one year in three areas in Santa Isabel of different land use. The monitoring program will provide the necessary information to understand the effects of land cover on aquifer hydrology.
5. Using the data gathered, develop a groundwater model of the Southern Aquifer area. The model will be calibrated to assess salinization potential under varying water extraction and recharge scenarios.
6. Develop an age-stratified community survey based on interviews to assess perceptions on the value of the aquifer resource and the problems associated with excessive exploitation and climate change.
7. Integrate the “Plan de Uso de Terrenos” prepared by the Puerto Rico Planning Board to evaluate land use plans for the aquifer recharge areas.
8. Develop maps to present the existent problem, and to show flux models and the water capacity of the area.
9. Develop an educational manual with activities for teachers and students based on the ground water resources of Puerto Rico.

Bibliography:
Introduction

- In rivers, the microbial loop explains how microbes oxidize DOC to obtain energy making carbon available to higher trophic levels in the river foodweb.
- DOC biodegradability, however, is variable depending on its source. Its concentrations and quality can be affected by allochthonous sources, but can also be affected by anthropogenic sources such as wastewaters and runoff from urban or agricultural areas. Anthropogenic carbon inputs to rivers can increase the concentration of labile DOC (Balakrishna, 2006).

Hypotheses

Rivers located in the upper forested regions of the watersheds have the highest concentration of labile DOC, which will diminish through the altitudinal gradient. Also, if anthropogenic activity on the watershed increases loads of organic carbon to the river, urbanized watersheds will have higher DOC (potentially labile) and POC levels than forested watersheds.

- The objective of this investigation is to determine the change in the quantity and quality of dissolved organic carbon along altitudinal and land use gradients in three tropical watersheds in Northern Puerto Rico.

Study Site

Figure 1. Detailed view of the studied watersheds: Río Mameyes (top left), Río Canóvanas (top right), and Río Piedras (bottom). Stars illustrate the location of the USGS gauging stations.

Methods

We compared the quality of the DOC along an altitudinal gradient in three different watersheds with different urban cover. We collected water samples in ten sites from each watershed during three sampling campaigns. Regression analysis were applied to explore altitudinal patterns of BOD5 and organic carbon quantity. This poster shows the gathered data and analysis.

To evaluate temporal changes in DOC quality, water samples were collected once during the wet season of 2007 and during the dry season of 2008 and 2009. Table 1 summarizes the sampling dates for each of the watersheds and the flow conditions associated to each sampling event.

Table 1. Flow conditions during the sampling events in each of three watersheds.

<table>
<thead>
<tr>
<th>River and gauging station</th>
<th>Date</th>
<th>Mean daily flow (m³/s)</th>
<th>Exceedance probability (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Río Canóvanas at Campo Rico</td>
<td>8/11/07</td>
<td>0.34</td>
<td>53</td>
</tr>
<tr>
<td>50061800</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Río Piedras at Hato Rey 50049100</td>
<td>8/9/07</td>
<td>0.65</td>
<td>50</td>
</tr>
<tr>
<td>Río Mameyes NR Sabana 50065500</td>
<td>8/2/07</td>
<td>0.28</td>
<td>99</td>
</tr>
<tr>
<td>Río Canóvanas at Campo Rico</td>
<td>3/25/08</td>
<td>0.24</td>
<td>70</td>
</tr>
<tr>
<td>50061800</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Río Piedras at Hato Rey 50049100</td>
<td>3/24/08</td>
<td>0.37</td>
<td>80</td>
</tr>
<tr>
<td>Río Mameyes NR Sabana 50065500</td>
<td>3/27/08</td>
<td>0.71</td>
<td>66</td>
</tr>
<tr>
<td>Río Canóvanas at Campo Rico</td>
<td>4/11/2009</td>
<td>0.37</td>
<td>47</td>
</tr>
<tr>
<td>50061800</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Río Piedras at Hato Rey 50049100</td>
<td>4/24/2009</td>
<td>0.42</td>
<td>77</td>
</tr>
<tr>
<td>Río Mameyes NR Sabana 50065500</td>
<td>4/8/2009</td>
<td>0.31</td>
<td>98</td>
</tr>
</tbody>
</table>

Discussion

The regression between the DOC and the BOD5 of the three watersheds (Figure 2) revealed that the quantity of BOD5 increases exponentially as the DOC concentration increases. This correlation can be explained by the level of anthropogenic activity in each watershed. In a forested watershed such as the Río Mameyes, the levels of DOC are lower than in the transitional (Río Canóvanas) and the forested basin (Río Piedras) because there are no anthropogenic inputs of organic matter; only natural inputs.

In Figure 3, we can observe the relationship between BOD5, DOC and the SUVA with stream order. In the SUVA analysis, the sampled rivers did not show any significant differences among stream orders. Large rivers also show significantly high BOD5 values compared to low order sites (Single Factor ANOVA; P = 0.03). Similarly, the high stream order sites showed significantly higher DOC concentrations than low order streams (Single Factor ANOVA; P = 0.004).

Conclusions

Our results suggest that in the studied tropical watersheds, headwater streams show relatively low DOC concentrations of low biodegradability compared to downstream river segments. Human activities, which are concentrated in the coastal plains, are associated to high organic inputs to rivers of high organic content and relatively high lability compared to upstream forested sites. Based on our results, BOD5 concentrations remained fairly similar in both wet and dry seasons suggesting that DOC quality may be insensitive to flows.

Acknowledgements:

I want to thank Prof. Alonso Ramirez for his mentorship during the UMEB Program and Rafael J. Benítez Joubert for assistance with field and lab work.

Alexandra S. Marcano Rivas and Jorge R. Ortiz Zayas, Institute for Tropical Ecosystem Studies, University of Puerto Rico

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Changes in structural characteristics of leaves as related to water availability from trees growing in the Costal Plateau of Guánica Dry Forest

Introduction

Trees in the Coastal Plateau of Guánica Dry Forest are growing under a combination of environmental stressors such as water, nutrient availability, salt spray, and wind that determine their capacity to grow and maintain their photosynthetic structures. Leaf structural characteristics such as weight, area and Specific leaf area (SLA, cm²/g dry weight) provide a measure of how plants use nutrients and water from their environment. We compare leaf’s characteristics in relation to water availability. SLA is a method that is easily measured and relates to how species acquire and use water resources.

SLA is defined as the measurement of the leaf’s thickness that is calculated dividing leaf’s area by dry weight of the leaf (Garnier, et al. 2001). This measurement demonstrates the ratio between area and dry weight which establishes the physiological cost of production on the leaf (Medina et al. 1990, Dingkuhn, et al., 2001). We hypothesize specific leaf area is directly proportional to water availability. Therefore, the rainfall conditions through out the year would reflect changes in SLA.

Study Site

The Guánica Dry Forest is localized in the southwest coast of Puerto Rico. This forest has an extension of approximately 11,000 acres of land which include 8 marine nautical miles and 21 km of coast (Fig. 1). It has an annual average rainfall of 762 mm but may vary from 500mm to 1600mm during unusually wet years. The rainy period is from August to November and the drought season is from December to April (Lugo & Murphy, 1986). During this period, the rainfall is only 10% the annual mean rainfall. The temperatures fluctuate from 24°C to 28°C, and a maximum temperature of 39°C, the variability. Other determinants such as nutrient availability, (e.g. phosphorus) may be because of the influence by genetics (Pereyra-Irujo, et al., 2001). This measurement demonstrates the ratio between area and dry weight which establishes the physiological cost of production on the leaf (Medina et al. 1990, Dingkuhn, et al., 2001).

Methodology

From November 2007 to October 2008 we collected fifteen to twenty adult leaves each month from five different species: Pisonia albida, Coccoloba microstachya, Ficus citrifolia, Enhalis fruticosa and Tabebuia heterophylla. A Tukey Test was performed to determine differences in SLA among species. Significant differences were found among species (F= 502.7, df = 4, p < 0.0001). P. albida and F. citrifolia presented similar patterns in their structural changes. Both species reflect monthly changes in available water for leaf area (cm²), weight (g) and SLA (g/cm³) while the other three species maintain a similar pattern throughout the year except in the rainy season (fig. 3). Both P. albida and F. citrifolia respond to the lower rainfall recorded in February by reducing their SLA in the following months. P. albida, an obligative deciduous, dropped all its leaves by end of March and the newly expanded leaves collected in the April have significantly lower leaf area and weight (fig 3.). F. citrifolia is a facultative deciduous plant while the other three species are leaf exchangers and only reflect extreme changes in water availability (fig 3.).

Table 1. Differences in leaf characteristics among adult leaves of five tree species. Letters in columns indicates statistically significant differences among species (Tukey test, p ≤ 0.00)

<table>
<thead>
<tr>
<th>Species</th>
<th>N</th>
<th>Area (cm²)</th>
<th>Weight (g)</th>
<th>SLA (cm²/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. microstachya</td>
<td>436</td>
<td>7.50 ± 3.37 c</td>
<td>0.13 ± 0.06 c</td>
<td>58.46 ± 11.96 d</td>
</tr>
<tr>
<td>E. fruticosa</td>
<td>560</td>
<td>8.67 ± 2.77 b</td>
<td>0.19 ± 0.07 b</td>
<td>46.80 ± 9.91 e</td>
</tr>
<tr>
<td>F. citrifolia</td>
<td>377</td>
<td>21.86 ± 12.2 0 a</td>
<td>0.21 ± 0.11 a</td>
<td>95.42 ± 20.13 b</td>
</tr>
<tr>
<td>P. albida</td>
<td>320</td>
<td>20.96 ± 9.94 a</td>
<td>0.22 ± 0.11 a</td>
<td>97.81 ± 18.11 a</td>
</tr>
<tr>
<td>T. heterophylla</td>
<td>884</td>
<td>4.05 ± 1.92 d</td>
<td>0.05 ± 0.03 d</td>
<td>77.29 ± 13.56 c</td>
</tr>
</tbody>
</table>

Results and Discussion

All species respond to changes in water availability in the Guánica Dry Forest. The rainfall experienced during the year caused different structural changes for all species (Table 1). Significant differences were found among species (P= 0.057, df = 4, p < 0.0001). P. albida and F. citrifolia presented similar patterns in their structural changes. Both species reflect monthly changes in available water for leaf area (cm²), weight (g) and SLA (g/cm³) while the other three species maintain a similar pattern throughout the year except in the rainy season (fig. 3). Both P. albida and F. citrifolia respond to the lower rainfall recorded in February by reducing their SLA in the following months. P. albida, an obligative deciduous, dropped all its leaves by end of March and the newly expanded leaves collected in the April have significantly lower leaf area and weight (fig 3.). F. citrifolia is a facultative deciduous plant while the other three species are leaf exchangers and only reflect extreme changes in water availability (fig 3.).

Conclusion

The obtained pattern confirms that the environmental conditions affect leaf structural characteristics; however the range of change during the leaf expansion is species specific because of the influence by genetics (Pereyra-Irujo, et al., 2008). Plants in the Guánica Dry Forest respond to changes in water availability, especially during leaf expansion. Specific Leaf Area increased with rainfall however cumulative rainfall only explains around 30 % of the variability. Other determinants such as nutrient availability, (e.g. phosphorus) may be driving this parameter.

Bibliography


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Bayesian analysis for the growth of Mona Island iguana in headstart facility (Cyclura cornuta stejnegeri)

Introduction
Mona Island is a Natural Reserve under the management of the Department of Natural Environment Resources (DNER). Mona Island iguana (Cyclura cornuta stejnegeri), is an endemic reptile species of this region which is considered endangered. A headstarting program has been implemented since 1999 by the DNER for Mona Island iguana. It is desired to predict how long it will take for an animal to achieve the minimum size required for release. In early stages a linear growth equation can be a good approximation. Hierarchical linear models are used for analyzing the growth of iguanas in the headstart facility. These models incorporate explanatory variables such as sex and cohort (1999-2000) or (2004-2005).

Model formulation

\[ S_i(t) = S_{0i} + r_it + \varepsilon_i \]

Size of the i-th individual at time t
Size of the i-th iguana at birth
Rate of growth for the i-th iguana
Random error \( \varepsilon_i \sim N(0, \sigma_e^2) \)

Prior distributions

\[ \beta \sim \text{HalfNormal}(0,1) \]
\[ \sigma^2 \sim U(0,1000) \]

Models considered in the analysis

Table: Summary of individuals in the data set

<table>
<thead>
<tr>
<th>Year</th>
<th>Female</th>
<th>Male</th>
</tr>
</thead>
<tbody>
<tr>
<td>1999</td>
<td>26</td>
<td>14</td>
</tr>
<tr>
<td>2000</td>
<td>13</td>
<td>10</td>
</tr>
<tr>
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<td>2005</td>
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<td>5</td>
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Model comparison for SVL

<table>
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<th>DIC</th>
<th>∆DIC</th>
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<tr>
<td>Equal growth</td>
<td>2.98</td>
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<td>392.06</td>
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<tr>
<td>Growth rate depends on sex</td>
<td>4.02</td>
<td>1857.23</td>
<td>378.94</td>
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<td>Growth rate depends on cohort</td>
<td>4.16</td>
<td>1888.73</td>
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<td>Growth rate depends on sex and cohort</td>
<td>6.06</td>
<td>1855.55</td>
<td>377.16</td>
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Model comparison for mass

<table>
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<tr>
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<th>DIC</th>
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<tr>
<td>Equal growth</td>
<td>2.16</td>
<td>6386.23</td>
<td>495.89</td>
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<tr>
<td>Growth rate depends on sex</td>
<td>3.17</td>
<td>6385.58</td>
<td>405.24</td>
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<tr>
<td>Growth rate depends on cohort</td>
<td>3.23</td>
<td>6249.90</td>
<td>269.56</td>
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<tr>
<td>Growth rate depends on sex and cohort</td>
<td>5.30</td>
<td>6342.61</td>
<td>262.27</td>
</tr>
</tbody>
</table>

Models were fitted using WinBUGS software (Spiegelhalter et al 2003).

Predicted SVL

\( pD = \text{Measure of model complexity (effective number of parameters)} \)
\( \text{DIC} = \text{Measure of goodness of fit. Lower DIC implies better fit.} \)

According to DIC values hierarchical models have better fit than nonhierarchical models.

For SVL, DIC values seem to support the model with common mean and the model with cohort differences. Nevertheless, the common mean model was selected by biological reasons.

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Funded by National Science Foundation, HRD #0734826, and University of Puerto Rico, Central Administration and Río Piedras Campus
Introduction

Feeding is an important component in consumer-resource interactions of population-community ecology and in energy and nutrient transfers of ecosystems ecology (Bengtsson 1998). Food webs are used to describe the complexity of feeding interactions. The level of omnivory of the members of an ecosystem can influence food web dynamics, interspecific interactions, and trophic structure (Morin and Lawler 1996, Polis and Strong 1996). Omnivory is defined as feeding on more than one trophic level (Pimm and Lawton 1978).

Anolis lizards are the most abundant and conspicuous components of the diurnal community in the Caribbean islands, (Williams 1969). Although considered strict insectivorous, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, considered strict insectivorous, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002). Given the uncertainty about the trophic ecology of anole lizards, some species add fruits to their diet (Herrel et al. 2002).

We present field observations and preliminary data about omnivory/frugivory of Puerto Rican anole lizards. This data is part of the research being developed to determine their degree of omnivory in Puerto Rico and their possible role as seed dispersers in forests on karst with different ecophysiological dynamics.

Omnivory

The analysis of stable isotopes (13C and 15N) in organisms can be used to indicate the average diet and actual energy links in food webs (Post 2002). The ratio of carbon isotopes (δ13C) changes little as carbon moves through the food web, indicating that carbon isotopes reflect the δ13C value of a consumer tissues (Kelly 2000). In contrast, the nitrogen isotopes (δ15N) can indicate trophic position and level of omnivory of anole lizards. A more extensive sampling of the isotopic variation of other lizards (Fig. 1).

I examined preliminary the trophic position of A. cristatellus using isotopic analysis (13C 15N) and found an omnivorous diet when compared with other lizards (Fig. 1). A more extensive sampling of the isotopic variation of Anolis lizards is currently being undertaken and will allow us to determine the trophic position and level of omnivory of anole lizards.

Anoles as potential seed dispersers

There is no published data about seed dispersal by anoles lizards, an important phenomenon related to feeding habit that depends, but not only, on the degree of frugivory of a species (Cáceres 2002). Extensive use of fruits, diversity and viability of seeds are three aspects to consider in the evaluation of lizards as a seed disperser. To assess the diversity of fruits and the viability of seeds consumed by the giant anole, A. cuvieri, we trapped 15 individuals in the moist karst forest in Dorado, PR. We collected 30 fecal pellets of the trapped animals and determined the ingested items in each pellet. In addition to arthropods and snails, 327 seeds corresponding to 4 species of fleshy fruits from a vine, a shrub and trees were found in the fecal pellets (Fig. 3). All seeds were germinated in Petri dishes with absorbent paper.

Conclusion

The preliminary data suggest that Anolis lizards are omnivorous and potential seed dispersers. Detailed studies will be carried to determine the level of omnivory, trophic position, and role as seed dispersers of Anolis lizards. The information gathered in those studies will allow to determine the role of anoles lizards in food webs as well as in the function and dynamics of insular tropical ecosystems.

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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
PRELIMINARY STUDY OF THE SOIL MICROBIOLOGY ASSOCIATED TO ISOLATED DWARF TREE SPECIES IN THE COASTAL PLATEAU OF GUÁNICA DRY FOREST

ABSTRACT

Soil function and productivity depends on microbial communities for nutrient cycling, antagonistic effects and decomposition of organic matter among others. Mass and diversity of soil microbial communities depend on substrate characteristics determined by the resource input of the dominant plant species. Plant species effects and microbial can also be detected at much more localized spatial scales within habitats. The Coastal Plateau of the Guánica Dry Forest in Southwestern Puerto Rico is ideal for this type of study as dwarfed trees of different species are growing on cracks in the calcareous rocks isolated from each other. The aim of this preliminary study was to optimize the fumigation extraction method for dry land soils and to determine the soil microbial biomass associated to tree species representing different litter quality. Soil samples were obtained underneath the leaf litter of 4 isolated tree species. The substrate was shallow and predominantly humic. Microbial biomass was calculated following the fumigation extraction method described by Vance et al. (1987). Our results confirm that the fumigation extraction method is appropriate for the determination of soil microbial biomass in our substrates and that microbial carbon is correlated to the percentage of water content.

INTRODUCTION

Soil microbiota is defined as the part of the organic matter in soil that constitutes living microorganisms smaller than 5-10µm. It serves in many vital process including nutrient cycling and storage, organic matter decomposition, antagonism, plant microbe interactions among others. Both biotic and abiotic factors influence the microbiota present in soils. For example the quality and quantity of organic residues, litter, soil organic matter and roots of a tree can affect the soil microbiota. Studying the soil microbiota is important in terms of understanding the fertility of a given soil ecosystem and which is the contribution of microbes in term of nutrient input and output. Given that plant species vary in their effects on soil organism it can be expected that the processes regulated by organisms will also be responsive to plant species effects. The Coastal Plateau of the Guánica Dry Forest in Southwestern Puerto Rico is ideal to study plant species effects on microbiota as dwarfed trees of different species are growing on cracks in the calcareous rocks isolated from each other. The aim of this preliminary study was to optimize the fumigation extraction method with soil samples taken from under the leaf litter of three isolated tree species at the Guánica Dry Forest, in order to determine the microbial organic carbon and microbial biomass present in these areas.

METHODS

We sampled three isolated trees from four species: Tabebuia heterophylla, Pisonia albida, Conocarpus erectus and Erithalis fruticosa. The leaf litter layer was separated and the humic soil was sampled down to rock surface. The samples were stored at 4°C until analysis. The soils from each tree were analyzed separately. The samples were processed and analyzed following the fumigation extraction method described by Vance et al. (1987) as described in Anderson and Ingram (1993).

RESULTS

<table>
<thead>
<tr>
<th>Tree Species</th>
<th>Soil Water Content (%)</th>
<th>Soil Microbial Carbon (µg/g soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tabebuia heterophylla</td>
<td>57.6 0.87 a</td>
<td>1900 40 a</td>
</tr>
<tr>
<td>Conocarpus erectus</td>
<td>57.5 0.55 a</td>
<td>1350 30 b</td>
</tr>
<tr>
<td>Erithalis fruticosa</td>
<td>47.9 0.36 b</td>
<td>990 20 c</td>
</tr>
<tr>
<td>Pisonia albida</td>
<td>31.0 0.57 c</td>
<td>580 40 d</td>
</tr>
</tbody>
</table>

There were significant differences in water content and microbial biomass carbon among soils from the different tree species. The exceptions were Conocarpus and Tabebuia that had similar water content but different microbial biomass values (Table 1). Our results are within the range found by Galicia and García-Oliva (2008) in a dry tropical deciduous forest in México.

There was a significant linear correlation between water content and microbial biomass carbon indicating that water is a limiting factor for microbial biomass dynamics ($C_{mic} = -0.16344 + 0.0024\%WC; R=0.85$).

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There was a significant linear correlation between water content and microbial biomass carbon indicating that water is a limiting factor for microbial biomass dynamics ($C_{mic} = -0.16344 + 0.0024\%WC; R=0.85$).

Soil quality and microbial activity will be measured by enzyme activity assays and nutrient analyses.

Microbial community structure will be assayed by using the Phylochip microarray technology, and FAME in order to determine which are the microbial functional groups present and if microbial communities differ among plant species and soil water availability.

CONCLUSIONS

The extraction fumigation method described by Vance et al. 1987 is suitable for soils obtained from the Guánica Dry Coastal Forest.

Our preliminary results indicate that water availability is a determinant factor for microbial biomass in the system.

Water content in the soil among species could be a result of the water retention capacity of the substrate and the density of the canopy architecture of the tree species.

FUTURE WORK

Seasonal sampling and analysis will take place in order to compare microbial biomass dynamics throughout the year.

Soil quality and microbial activity will be measured by enzyme activity assays and nutrient analyses.

Microbial community structure will be assayed by using the Phylochip microarray technology, and FAME in order to determine which are the microbial functional groups present and if microbial communities differ among plant species and soil water availability.

REFERENCES


Leaf litter decomposition in tropical and temperate rainforests: teaching climate-driven ecosystem processes outside the science classroom.

Introduction
One of my goals as a future teacher is to motivate students to better understand the environment and the natural processes that regulate the ecosystem functionality. One way to accomplish this is by teaching ecosystem processes, conducting field research to improve the science lessons.

- Leaf litter decomposition is an important ecosystem process. It refers to the process that converts dead organic matter into smaller and simple compounds.

- Litter decomposition is mainly a biological process, its rates can vary with abiotic or biotic factors like:
  - leaf species - treatment (location on soil)
  - temperature - arthropods (abundance)

- As a future teacher I have seen the necessity of creating innovative ways to communicate the importance of ecological processes to my students. With this same purpose projects like the Long Term ecological research network (LTER) exist:
  - LTER Network (Figure 2) - create science research for teachers and their students
  - Ecoplots - resource for science teachers on the web.

![Figure 3: The LTER Network](image)

**Question**
How does the rate of litter decomposition differ between tropical and temperate rainforests?

**Objectives**
Improve science lessons related to ecosystem function and climate.

Develop an experimental lesson plan based on field experiences to assist teachers in conveying the concept of climate-driven ecosystem processes to students, the comparison of leaf litter decomposition rates between two environments with different climate will be used to help understand this concept.

Exchange and compare leaf litter decomposition data between Luquillo Experimental Forest and HJ Andrews.

Determine the role of arthropods in controlling leaf decomposition by comparing rates above and below the soil surface.

![Figure 4: Selected plant species](image)

**Methodology**

**Results**

- Comparison of leaf litter decomposition of five species, three from a tropical rainforest and two from a temperate rainforest. Leaf litter bags were placed above and under the soil surface of the LDF. In the tropical rainforest (El Yunque), the three tropical species showed similar decomposition rates above and below surface, whereas the two temperate species showed drastic differences. Alnus glutinosa had the highest decomposition rate of all species, especially below surface.

![Figure 5: Comparison of leaf litter decomposition of five species, three from a tropical rainforest and two from a temperate rainforest.](image)

**Conclusions**

- Participation in authentic scientific research gives teachers confidence and motivation. At the same time, these experiences can be implemented as part of their class work.

- With this research we can develop a lesson plan that we can integrate to the curriculum. By doing this we can understand climate driven ecosystem processes and motivate students to do scientific research.

- In the experiment performed here, location (tropical vs. temperate) was more important than arthropods (above vs. below ground) in determining leaf litter decomposition rates. We propose that high humidity and temperature in tropical forest are the main drivers explaining this difference.

- Lack of arthropod effects could be the result of the duration of the experiment. Incubation over longer periods of time could potentially increase arthropod participation of this process.

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Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Rio Piedras Campus.
Introduction
Aquatic habitats play an important role in the reproduction of adult amphibians and in the development of their larvae. In these habitats, the interactions of biotic and abiotic factors influence tadpole ecology. Tadpole growth rates, duration of larval period, size at metamorphosis and survival to metamorphosis can be influenced by the physical environment (Alford 1999). In the case of tadpoles, abiotic parameters like desiccation risk, water temperature, dissolved oxygen concentration, pH, and salinity are a major concern for the success of a breeding event (Ultsch et al. 1999).

Salinity is an abiotic factor that has raised concern for the management group in charge of ensuring the conservation of the Puerto Rican Crested Toad (Peltophryne lemur), the only endemic toad in Puerto Rico, currently listed as a threatened species. P. lemur breeds at the Tamarindo Pond, the main natural breeding pond in Puerto Rico. This pond has been influenced in the past by salt intrusion events and it is believed that this may affect negatively the development of the tadpoles.

Objective
To identify the effects of increasing salinity on the survival and growth of tadpoles of P. lemur.

Experimental design
The laboratory experiment was set up in a randomized block design. After being reared, tadpoles (Gosner stage 24 – 26) were housed individually in plastic containers. Water temperature was maintained 27 – 28°C in 12h:12h light-dark periods. Water in the containers was replaced daily and their placement on the shelves was rotated between shelves. Tadpoles were fed once a day fish flakes and a spinach leaf. There were six salt water treatments: control group (0 ppt), 2, 4, 6, 8, 10 ppt, ten replicates per treatment. Aquarium commercially available salt was used to do the salt water mixtures. Pictures and tadpole weights were taken every other day from the beginning of the experiment till the tadpoles metamorphosed.

Preliminary results
As the salt concentration in the water increased, the weight of the tadpoles decreased. The weights of the tadpoles at metamorphosing, three times longer than tadpoles raised at 0, 2, 4 and 6ppt. Mortality increased in higher than 10ppt the toads will not breed (C. Pacheco, FWS, Canals, DRNA, personal communication). We have also reported that if the salt concentration in the natural pond were tadpoles have tolerated levels up to 8ppt for various days and can metamorphose at 5ppt successfully (M. Canals, DRNA, personal communication). We also have reported that if the salt concentration in higher than 10ppt the toads will not breed (C. Pacheco, FWS, personal communication). Monitoring salt concentration in the natural breeding pond are important in managing the only breeding pond available for the toads.

Discussion
Some of the negative effects of increased salinity for tadpoles include reduced growth rate, delayed metamorphosis, physical abnormalities, and increased mortality (Chinathamby et al. 2006, Rios-López 2008). We did not report any type of deformity or malformation. However, two of the most important results are the higher mortality rate at 10ppt and the increased time to metamorphosis in the 8ppt treatment. Seven out of ten tadpoles raised at a 10ppt died within 72 hours of exposure. Tadpoles raised at 8ppt lived for about 80 days without metamorphosing, three times longer than tadpoles raised at 0, 2, 4 and 6ppt. As the salt concentration in the water increases, the weight of the tadpoles was reduced. These preliminary laboratory results have relevance compared to what we have seen in the natural pond were tadpoles have tolerated levels up to 8ppt for various days and can metamorphose at 5ppt successfully (M. Canals, DRNA, personal communication). We have also reported that if the salt concentration in higher than 10ppt the toads will not breed (C. Pacheco, FWS, personal communication). Monitoring salt concentration in the natural breeding pond are important in managing the only breeding pond available for the toads.

References


Acknowledgments: Travel and housing were provided by CATEC (NSF-HRD: 0734826) and materials were provided by PRLSAM # HRD-0832961. Special thanks go to personnel at Fort Worth Zoo for their guidance, support and for continuing the experiment. I thank Dr. Neftalí Ríos and Dr. Dallas Alston for reviewing the initial proposal.
Nitrogen Dynamics in a Tropical Cave Stream

Introduction

Nitrogen is a major limiting nutrient, controlling biodiversity, structure and productivity of many aquatic ecosystems. Inputs to aquatic environments have increased due socioeconomic development and changes in land-use. Excessive loads can eutrophy freshwater systems and constitute a major threat to estuaries and coastal waters.

Cave streams play a major role in the integration of nutrient cycles and ecological processes in many watersheds. The impact of excessive N₂ loads on the cave stream ecosystem and its receiving surface and coastal waters will primarily depend on the dynamics of N₂ transportation and transformations that occur within the cave environment.

Objectives

1. Document in-stream relative composition of nitrogen forms over one water year.
2. Determine and evaluate nitrogen retention (nutrient retentive streams have short uptake lengths).

Rationale

1. If under base flow conditions, nitrogen cycling along a tropical cave stream is sustained at a near equilibrium condition, then, relative composition and concentration of nitrogen forms, population size of N-dependent chemoautotrophs and in-stream proportion of nitrogen inputs to outputs will not change significantly with time.
2. In a tropical cave stream, with high allochthonous nitrogen inputs, nitrogen export is favored over retention.
3. Energy-yielding transformations are favored in tropical cave streams, compared to tropical surface streams.

Methods

The investigation is currently conducted at the El Convento Cave-Spring System, located at the southwestern coast of Puerto Rico.

In-stream concentration of nitrogen species (bical/organic nitrogen, ammonium and nitrate) were measured at selected stations, during one water year. Daily average discharge was computed from a long-term water-level monitoring station installed at the surface reach.

Results

The relative stability between nitrate inputs and outputs suggest that N₂ export is favored over retention.

Nitrifying bacteria fluctuated spatially and with time. A downstream decreasing trend is observed.

The data suggest an efficient nitrification process within the cave stream, compared to the surface reach.

Denitrification data suggest a significant immobilization of N₂ in the cave stream bottom sediments.

Enumeration of nitrifiers by the “most probable number” method was assessed periodically. Experimental assays for measuring potential nitrification and denitrification (acetylene block) rates were accomplished. Uptake length determinations will be conducted at representative cave stream stations.

Concentration of nitrate increases along the cave stream drainage system and showed a similar fluctuation pattern with time.

Potential Nitrification Rates at El Convento Cave Stream and Surface Reach Stations

Potential Denitrification Rates at El Convento Cave Stream and Surface Reach Stations

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Funded by the National Science Foundation (HRD # 0734826) and the University of Puerto Rico, Río Piedras Campus
CHEMICAL COMPOSITION OF ATMOSPHERIC PARTICLES IN THE GUANICA’S DRY FOREST: CARBONACEOUS AEROSOLS IN AFRICAN DUST

Abstract

The Caribbean region is influenced by natural and anthropogenically derived aerosols. Guánica’s Dry Forest (GDF) is located in the semi-arid southwestern Puerto Rico and is an excellent example of dry forest life zone. The fundamental question that our research addresses is How does seasonal variation in aerosol physical and chemical properties from African dust and anthropogenic-derived particles influence the Guánica’s Dry Forest (GDF)? On a short-term basis we want to respond to this question by providing a better understanding of the chemical composition, seasonal variation, and impact on microclimate of aerosols focusing on African dust and anthropogenic-derived particles in the GDF.

Introduction

- Guánica’s Dry Forest (GDF) is one of the most intact mature dry forests in the Caribbean and a UNESCO Man and Biosphere Reserve.
- African dust blows from the west coast of Africa, moves across the Atlantic and, after few days, impacts the Caribbean, particularly during the summer months.
- Anthropogenic influences at the GDF might be due to the impact of the emissions from a thermoelectrical plant, Costa Sur, located in Guayanilla.
- During nighttime, the predominant wind direction at GDF is northeast, but in daytime is southeast.

Impact of Aerosols

<table>
<thead>
<tr>
<th>Impacts</th>
<th>General Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Health</td>
<td>Transporting toxic substances, can be inhaled through the respiratory tract</td>
</tr>
<tr>
<td>Climate</td>
<td>Scattering and absorption of solar radiation. Altering cloud properties through their ability to act as cloud condensation nuclei.</td>
</tr>
<tr>
<td>Ecosystem</td>
<td>Reduces sunlight, which could have effects in terrestrial and marine biological productivity</td>
</tr>
<tr>
<td>Visibility</td>
<td>Reduction in visibility due to African Dust</td>
</tr>
</tbody>
</table>

Figures 1-2: African dust affects the visibility and the quality of the air. These are views of the Guánigan Island, from our sampling station in GDF, in days without (left picture) and with African Dust (right picture) influence.

Experimental

Sampling Site
- Marine site located in Guanica’s Dry Forest, PR., 17.961 N, 66.842 W. (Figures 3-4)

Methods
- Stacked Filter Units (SFUs) - collects fine (Dp < 1.7 µm) and coarse (Dp > 1.7 µm) particles.
- Davis weather station – basic meteorological parameters.
- EC/OC analyzer (Sunset Lab) - determines the mass concentrations of total carbon (TC), elemental carbon (EC), and organic carbon (OC).

Preliminary Results

Graph 1-2: Night-time samples have higher concentrations of carbonaceous aerosols than daytime. The positive artifact is greater at nighttime than at daytime.

Conclusions

- During nighttime, the predominant wind direction at GDF is northeast, but in daytime is southeast.
- The average concentration for TC was 0.1652 µg m⁻³, EC 0.0066 µg m⁻³ and OC 0.1587 µg m⁻³.
- The average of positive artifact for nighttime was 26.15% and daytime was 5.09%.

Future Outlook

- Gravimetric analysis.
- Chemical characterization of the water-soluble ions and the water-soluble organic carbon and nitrogen species.
- Determination of the contribution of those species to the total aerosol mass and determination of their influence to the GDF’s microclimate.

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(1) Institute for Tropical Ecosystem Studies, University of Puerto Rico, San Juan, PR. (2) Department of Chemistry, University of Puerto Rico, San Juan, PR.
Statistical analysis of the relationship between size and clutch size for Mona Island iguana (*Cyclura cornuta stejnegeri*)

*Cyclura cornuta stejnegeri* is an endangered species

- Low recruitment of juveniles into the adult population.
- Modifications in its habitat
  - Incorporation of *Casuarina equisetifolia* (Australian pine)
  - Predation by exotic mammals
- Cats & pigs

**Goal**

To analyze data relating the size of a female (snout-vent length – SVL, tail length and body mass) and the number of eggs laid in each clutch.

**Method**

For the analysis of data Generalized Linear Models based on the Poisson distribution were used. All analyses were performed with the statistical software R.

**Variables**

- **Response variable:** number of eggs laid in the nest (clutch size)
- **Explanatory variables:** Snout-Vent Length (SVL) and weight

Several iguanas had more than one nesting period, hence the average of the measurements were used.

**Results**

- **SVL**
  - Let $q(svl)$ be the mean clutch size for an iguana with $SVL = svl$. Then, the fitted model is
    $$\log(q(svl)) = 0.507 + 0.044svl$$
    (p-val = 2.65x10^{-5}, statistically significant)

- **Weight**
  - Let $q(w)$ be the mean clutch size for an iguana with $Weight = w$. Then, the fitted model is
    $$\log(q(w)) = 1.874 + 0.151w$$
    (p-val 7.8x10^{-6}, statistically significant)

**Discussion**

- Clutch size seems to be associated with both SVL and Weight.
- Observation 17 is a potential outlier. It should be carefully analyzed in order to determine if it can be eliminated.

**Conclusion and Future Work**

- The clutch size of Mona Island iguana depends on the size of the female, either measured by SVL or by Weight.
- In subsequent work, SVL will be used, because it does not depend on temporal availability of resources (it is more stable than weight).
- Future work will involve relating the relationship found in this work with growth curves.

**References**


**Acknowledgments**

- RISE Program
- NIH
- NSF
- Department of Mathematics, UPR Río Piedras
- Department of Natural Resources of Puerto Rico
- PR-LSAMP

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INTRODUCTION

Germination, establishment, and survival of young plants appear to be the most critical phases in the life cycle of cacti (Godínez-Alvarez et al. 2003). Thus, understanding factors affecting processes of germination and establishment in cactus species is important to develop successful conservation and management strategies.

Harrisia portoricensis is an endangered cactus endemic to Puerto Rico and geographically restricted to three small Caribbean Islands located to the west of Puerto Rico: Mona, Monito and Desecheo (USFW 1990).

In the south eastern side of the island, the exotic grass Megathyrsus maximus occurs ubiquitously and seems to be spreading. This exotic grass was apparently introduced to Mona Island around 1960’s to support cattle activities.

RESULTS

• The proportion of germinated seeds in control treatment was higher than the proportion of germinated seeds placed in shade treatment in the field. None of the seeds sown in open sites with direct sunlight exposure germinated (Fig. 1).
• The survival of transplanted seedlings was low. At the end of the experiment only 7 individuals were alive at shaded sites in the field. All seedlings transplanted to open sites died by the second month (Fig. 2).
• Seedling survival in shade sites was not higher when seedlings were excluded from potential predators.
• By the end of 15 month in the field, a higher proportion of seedlings survived beneath the canopy of the native shrubs, specifically Croton discolor (40%) and R. uncinata (30%) than beneath the shade of M. maximus (15%) (Fig. 3).
• Mean and maximum temperatures and PAR values were much higher under the canopy of M. maximus than under the canopies of native shrubs.
• The average diameters of seedlings growing under C. discolor and R. uncinata were twice the average diameter of seedlings growing under M. maximus (Fig. 4).

DISCUSSION

• Field experiments demonstrated that suitable conditions for germination and establishment of H. portoricensis seedling occur in shaded areas beneath the canopy of shrubs.
• We also detected that the identity of the shade providers have a significant effect on the process of germination and establishment of M. maximus may added vulnerabilities to this cactus species by reducing the amount of optimal sites for germination and establishment.
• Our data suggested that the modification by native shrubs species of microenvironmental conditions beneath their canopies may play a primary role in the germination, establishments, and growth of H. portoricensis.

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus
INTRODUCCION
Information on the breeding system of endangered plants is often useful piece of information for conservation and management of wild populations. This is so as the foraging behavior of the pollinators and the plant breeding system are factors that may also affect fruit-set and therefore plant reproductive success (Boch & Waser 2001).

Harrisia portoricensis (Fig. 1) is an endangered cactus endemic to Puerto Rico and geographically restricted to three small Caribbean Islands located to the west of Puerto Rico: Mona, Monito and Desecheo (USFW 1990). The population at Mona Island is the largest population identified to date.

METHODS

A. Floral Traits
Twenty mature flowers were collected from different individuals. For each flower, we performed morphological measures of floral structures to assess floral similarity to other cactus species (Fig. 3).

B. Floral Visitors
Thirty flowers from 20 different plants were video-recorded from flower opening to flower closing. A total of 210 h of video-recording was examined, taking detailed notes on the number and time of flower visits to assess their potential as pollinators. In addition, we performed 112 h of direct flower observations.

DISCUSSION
Given its flower morphology, H. portoricensis could be pollinated by moths or bats. However visits to flowers are uncommon events.

Controlled pollinations demonstrated that H. portoricensis has partially self-compatible breeding system (based on ISI). Nevertheless, this species is not capable of autofertilization and a pollen vector is required for sexual reproduction.

Our combined results suggest that for this species an endogamous breeding system should be favored by natural selection: (1) levels of inbreeding depression detected should be enough for selfing to be favored by selection, (2) selfing should not be high suggesting a high capacity for seed production.

RESULTS

Little is known about the breeding system of this cactus species but we do know that Harrisia portoricensis appears to be genetically uniform throughout its distribution range based on studies of allelic variation in allozymes (Santiago-Velez 2000). This is somewhat surprising given that most of the columnar cacti studied so far in the tropics have been characterized as self-incompatible hermaphrodite plants (Valiente-Banuet et al. 1997, Fleming 2002).

Based on this general observation one would have expected H. portoricensis individuals to be more variable. One possibility for this lack of variation may relate to the presence of breeding systems conducive to inbreeding.

Fig 1. Predated fruit (left) and flower buds

Here we present data on the floral traits, floral visitor, and breeding system of H. portoricensis to address the extent by which this cactus breeding system promotes self-fertilization events.

STUDY SITE
The work presented here was performed in Mona Island Reserve (Fig. 2). The island is located between Puerto Rico and Hispaniola and covers and area of 5517 ha. Annual mean precipitation is 810 mm and mean annual temperature is 25°C. Harrisia portoricensis is an endemic columnar cactus with threatened species status since 1990.

Fig 2. Mona Island Reserve

Table 1. Floral traits of Harrisia portoricensis. Means SE, N=20 flowers

<table>
<thead>
<tr>
<th>Trait</th>
<th>External Length</th>
<th>Internal Length</th>
<th>Perianth Width</th>
<th>Anthers Height</th>
<th>Stigma Height</th>
<th>Number of Stamens</th>
<th>Number of Ovules</th>
</tr>
</thead>
<tbody>
<tr>
<td>Length (cm)</td>
<td>(21.16)</td>
<td>(18.85)</td>
<td>(5.25)</td>
<td>(12.76)</td>
<td>(17.18)</td>
<td>(774)</td>
<td>(2413)</td>
</tr>
<tr>
<td>Width (cm)</td>
<td>(0.98)</td>
<td>(0.82)</td>
<td>(0.27)</td>
<td>(0.97)</td>
<td>(0.61)</td>
<td>(3.94)</td>
<td>(21.09)</td>
</tr>
</tbody>
</table>

Table 2. Fruit : flower ratio from each pollination treatment.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Flowers</th>
<th>Fruits</th>
<th>Fruit:flowers ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cross-pollination</td>
<td>17</td>
<td>11</td>
<td>0.65</td>
</tr>
<tr>
<td>Self-pollination</td>
<td>17</td>
<td>8</td>
<td>0.47</td>
</tr>
<tr>
<td>Natural</td>
<td>17</td>
<td>15</td>
<td>0.88</td>
</tr>
<tr>
<td>Autogamy</td>
<td>17</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Agamospermy (emasculated + covered)</td>
<td>17</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Fig 3. Harrisia flower (A) At anthesis. (B) Morphological measurements: ECL, external corolla length; ICL, internal corolla length; PD, perianth diameter; AH, anther height; SH, stigma height.

Fig 4. Means for (A) seed length, (B) seed width, (C) seed mass, and (D) germination rate for H. portoricensis in three pollination treatments. Different letters indicates significant differences between pollination treatments (P < 0.001).

Fig 5. Multiplicative fitness (w) estimates for each pollination-treatment performed in H. portoricensis that set fruits. Different letters indicates significant differences between pollination treatments (P < 0.001).
Introduction

Today about half of the island has been naturally reforested with native and introduced species (Lugo and Helmer 2004). These new forests have been criticized around the world, but studies showing the real influence of introduced species upon the ecosystems are relatively few. We studied the litterfall dynamics of a secondary forest established in valleys at El Tallonal, in the karst region of Arecibo, Puerto Rico. This forest is dominated by Castilla elastica (Moraceae), an introduced tree species from Central America and the North of South America. It’s common in secondary forests (Cokeley et al. 2000) and is a deciduous tree species.

Material and Methods

We established two plots, Tallonal 1 (N’18 24 424 W’66 44 080) and Tallonal 2 (N’18 24 469 W’66 43 779), and ten baskets (0.25 m²) were installed at each one to collect the litter fall. The baskets were empty biweekly, next the samples were dried, sorted and weighted. The reported data was collected from March 1st 2008 to January 15th 2009. Tallonal 2 data started from April 10th.

Results and Discussion

The leaf fall was similar between the two plots (Figure 4. a and b). There was a peak (5-6 g m⁻² d⁻¹) at dry season in both sites, from March-April, followed by low rates until January. Because C. elastica is a deciduous species, it loses almost all leaves during dry season, which explains the huge amount of C. elastica leaf fall during this period. There are differences between C. elastica leaf fall and other species leaf fall along the year. At Tallonal 2 C. elastica leaf fall was higher than Other species. The opposite occurred in Tallonal 1, which possibly is because the Tallonal 2 has earlier stage of regeneration.

Conclusions

Castilla elastica has an important role in the amount of organic matter released to the ecosystem, and its importance varies with its dominance and among seasons.

Both sites differ in the proportions of C. elastica and Other species leaf fall, showing different dynamics specially from August to January.

References


Cokeley, W., GD. Paye, C. Roberts and D. Beardsall. 2000. Fruit Dispersal of Castilla elastica (Moraceae), an introduced tree species from Central America and the North of South America. It’s common in secondary forests (Cokeley et al. 2000) and is a deciduous tree species.

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How microbes and hosts interact?

Ongoing Research Projects

1- The microbiome of animals and humans:
   Changes associated with lifestyles (humans, animals)
   Host response to GI microbes
   Demogeography of H. pylori

2- Antibiotic resistance in isolated animals and humans

Recent Publications


Dominguez-Bello MG, Blaser MJ. Do you have a probiotics in your future? Microbes and Infection 10, 1072, 2008


Collaborators

Microbiomes
Ruth Ley, Cornell University, USA
Martin Blaser, New York University, USA
Jeffrey Gordon, Washington University School of Medicine, USA
Phil Hugenholtz, DOE, Berkeley, USA
Fabian Michelangeli, Maria Alexandra Garcia, Monica Contreras, Magda Magris and Glida Hidalgo, Venezuela

Genetics and Bioinformatics
Rob Knight, University of Colorado, Boulder, USA
Josep Bassagania, Virginia Bioinformatics Institute, Virginia Tech., USA
Humberto Ortiz-Zuazaga, High Performance Computing Facility (HPCF) UPR

Mathematics and Statistics
Luis Pericchi, Dept Mathematics, University of Puerto Rico, USA
Maria E Perez, Dept of Mathematics, University of Puerto Rico, USA

Anthropology
Elisabetta Marini, University of Cagliari, Italy
Francisco Salzano, UFRGS, Brasil
Maria Catira Bortolini, UFRGS, Brasil

Students trained: Jan 2004–Oct 2009
Undergraduate students= 20 (currently 3)
Graduate students= 2 (currently 1 with thesis)
PhD theses finalized since 2004: 2
Arthropod diversity in caves with moderate and high anthropogenic presence.

INTRODUCTION

• Extensive diversity of organisms are found in subterranean habitats varying in degrees of permanence and residence (Culver 2009).

• Cave organisms are classified as troglobionts, those that are accidental and temporal subterranean visitors, and as trogloxenes (Fig.1), permanent residents of subterranean caves (Culver 1982).

• Scattered taxonomical literature presume tropics (Fig.2) have low subterranean species diversity (Fig.3) and most cave surface dwelling species are undescribed (Mitchell 1969 and Trajano 2001).

OBJECTIVES

Study the potential association between arthropod diversity and the abiotic environment (thermal, luminar and humidity) in caves within the Northern Karstic region of Puerto Rico (Fig.5).

Identify and compare arthropods diversity (Figs. 4 and 6) in caves with moderate and high human impact.

HYPOTHESES

• Cave arthropod diversity and composition will relate to light availability, relative humidity, and temperature.

• More diversity will be present in the zone near the entrance of the cave.

• Arthropods diversity will be lower in caves with higher human presence.

METHODOLOGY

Study Site (Fig.1)

• The North Karst Belt of Puerto Rico (Fig.7) is located along 160 km in the north coast of Puerto Rico between the municipalities of Loíza and Aguadilla. (Miller 2009)

• Four caves (Fig.7) with moderate and high human presence located in the municipalities:
  - Cueva Balcones, Morovis,
  - Cueva Infierno, Florida,
  - Cueva #41 y #50 en Utuado.

RESULTS TO DATE

• We placed and collected the 60 pitfall traps.

• Relative humidity, temperature, and light availability were measured in all the caves.

• 12 Pitfall of Cueva Infierno have been processed to date.

• We have counted morphospecies Hymenoptera, Blattaria, and Coleoptera orders

LABORATORY METHODS

• Classification of morpho species and orders with taxonomical keys in the laboratory.

• Heptane flotation

• Digital photography to create digital reference collection

• Data analysis will be conducted in the laboratory with taxonomical keys

• Statistical analysis of diversity and composition across environments and caves

DISCUSSION

Preliminary results show Hymenoptera order is more abundant within three zones in Infierno cave. After collecting all the pitfalls, observations suggest that diversity for Coleoptera and Hymenoptera is highest at the most luminic zone (next to entrance). This research will contribute to update the records for cave organisms in Puerto Rico and set more rigorous baseline studies for future ecological research in biospeleology.

ACKNOWLEDGEMENTS

This research is possible to many friends help me constantly in the subterranean world. Thanks: Waldemar Alcobas, Fernando Castro, Lourdes Lastra, Larisa Martinz, Johanna Colón, Ginna Malley, Zevio Schnitzer, and specially to José Luis Gómez. Also my mentor Elvia Meléndez-Ackerman for the patience and guidance in the surface.

LITERATURE CITED


Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Rio Piedras Campus.
There goes the neighborhood: Reproductive success of *Bletia patula* when *Spathoglottis plicata* moves in

**Abstract**
Naturalized exotic species are homogenizing the earth’s biota and are increasing densities of pests in newly colonized areas. When exotics change ecosystems and affect native species, then they are invasive. Do high density populations of *Spathoglottis plicata* (a naturalized orchid) affect populations of *Bletia patula* (a native orchid) through a common florivorous, orchid specialist weevil, *Stethobars polita*? We will determine the effect of *S. plicata* on the reproductive success of *B. patula* when *S. plicata* is present at variable densities. In addition, we will determine the florivory preference of weevils between *S. plicata* and *B. patula*. Finally, we will create a distribution model map for both orchid species, to predict the localities where both species may be interacting.

**Introduction**
*Spathoglottis plicata* a naturalized exotic orchid dominates a vast area where native orchids such as *Bletia patula* live. The two species are attacked by a common florivorous weevil, *Stethobars polita*, a native beetle that specializes on orchids. Herbivory is often density-dependent and in some cases may affect reproductive success directly or indirectly. The purpose of this study is to determine if *Spathoglottis plicata* densities affect the reproductive success of *Bletia patula* through the presence of *Stethobars polita*. We will also assess the current and potential distributions of the two orchid species.

**Objectives**
- Assess the effects of high density populations of *S. plicata* on the reproductive success of *B. patula*.
- Create maps of distribution models for the orchids in study.

**Methods**
**Study Site**

Puerto Rico is part of the West Indies and our study site for creating the distribution models. Data is being gathered at Río Abajo and Guajataca Forest Reserves (Figure 2). Both areas are part of the north karstic region of the island. Río Abajo study site is located along Rt. 10 and Guajataca study site is along Rt. 4446.

**Determining the reproductive success of *B. patula* when *S. plicata* is sympatric**

The reproductive success of *B. patula* (n = 60) is determined in the absence and presence of *S. plicata*. Our variables are the number of buds, flowers, weevils, ants and the damage done by *S. polita* to the fruits, flowers and buds of the studied plants. We also are measuring the distance of the studied plants to their three nearest neighbors of *B. patula* and *S. plicata*.

**Choice experiments**

We are determining the weevils’ flower preferences by exposing 3 weevils to *B. patula* and *S. plicata* (white or magenta variants) (Figure 3). We are measuring flower damage every 24 hours for two days. Each choice experiment is replicated. The choice experiment will be done monthly.

**Potential distribution of *B. patula* and *S. plicata* in Puerto Rico**

Potential distribution will be done using the Maxent algorithm, with at least 30 occurrence points for each species. Maxent will predict places that share similar environmental factors as the occurrence points (Figure 4). Thus, we will determine the possible places where the two species could be present and we should reveal the potential zones of overlap.

**Preliminary Results**

**Figure 5.** Bletia varieties in the presence and absence of *S. plicata*. Average number of weevils (Z = -1.948, p = 0.05), fruits (Z = 1.851, p = 0.06), and flowers (Z = 2.156, p = 0.03).

**Figure 6.** Choice experiments. Weevils of *S. plicata* magenta with flowers of *B. patula* and *S. plicata* magenta. The damage scale: 0 = no damage and 10 = completely damaged.

**Figure 7.** Choice experiments. Done to weevils of *S. plicata* white with flowers of *B. patula* and *S. plicata* white. The damage scale: 0 = no damage, and 10 = completely damaged.

**Acknowledgments**
For all the help you have given us in the field work: Ricardo Arriaga, Alvaro Bravo, Paul CaraDonna, Isamailsh Espino, Wildelina Gonzalez and Carlos Vega.

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and University of Puerto Rico, Central Administration
and Río Piedras Campus
Abstract

Coral reefs are under increasing threats that have undermined their resistance ability to disturbance, their ecological functions and their ecosystem resilience. A combination of long-term impacts by natural factors (i.e., hurricanes, meso-scale gyres), a sort of local human factors (i.e., water quality degradation, sediment- and nutrient-loaded runoff, overﬁshing), and climate-related factors (i.e., sea surface warming, bleaching) have resulted in net coral decline trends along the wider Caribbean region. This study is aimed at addressing long-term ecological change in coral reef benthic communities across different coral reefs in Puerto Rico (PR). This presentation is focused in Playa Carlos Rosario, Culebra Island, with 12 years of data.

Coral species richness declined by a mean factor of 54% during the period of 1997 to 2009. Percent living coral cover plummeted by 81%, or an annual mean of 6.8% loss. Coral cover loss following the 2005 unprecedented sea surface warming and mass bleaching event was 66%. Total algal cover has increased by a factor of 91%, but macroalgae cover has increased by 1,064%, or a mean annual increment of 89%. This suggest unequivocal impacts by recurrent sediment- and nutrient-loaded runoff pulses, in combination with recurrent impacts from meso-scale gyres that can increase background chlorophyll a concentrations by a 5-10 fold factor. Coral:macroalgal ratios have declined by 97%. Mortality trends in Montastraea annularis spp. complex has averaged 70% during the same period and 48% since 2005. These trends are widespread through different locations in PR, suggesting that coral reefs are in the peril of an ecosystem collapse. There is a need to develop mathematical models to predict what would be the future of Caribbean-wide coral reefs in the face of climate change impacts.

Methods

Data collection and analysis: A long-term coral reef monitoring program was launched in 1997. Data for this presentation comes specifically from one of the oldest monitoring sites located at Playa Carlos Rosario, Culebra Island. A total of twelve permanent transects are distributed in four replicates along three depth zones (<4 m, 4-8 m, and 8-12 m). Data were analyzed using a combination of univariate and multivariate statistical approaches to test for spatial and temporal patterns of change in benthic community composition. Only partial data was analyzed for this presentation.

Results

Long-term trends in benthic coral reef communities at one of the representative sites (Playa Carlos Rosario, Culebra Island): A) Coral species richness; B) % coral cover; C) % total algal cover; D) % macroalgal cover; and E) Coral:macroalgal cover. Dramatic coral decline has resulted in a permanent phase shift in benthic community structure.

Conclusions

Coral reef decline in PR has been significant during the last two decades. This have resulted in a major ecological phase shift in benthic community structure, from coral to macroalgal dominance. Local human factors, mostly water quality degradation, in combination with climate-related impacts associated to increased sea surface temperatures (SSTs), and mass coral bleaching and mortality. Preliminary mathematical modeling approaches suggest that M. annularis spp. complex may undergo extinction of there is a significant increase in SSTs. Current efforts include developing further modeling to forecast long-term climate change impacts at the coral reef community level.
Abstract

- The San Juan Bay Estuary (SJBE) is an important natural coastal resource which comprises several types of habitats, five lagoons, and the largest mangrove forest in Puerto Rico: the Piñones Commonwealth Forest. During the last one hundred years, humans have altered the SJBE extensively by dredging, filling, and by the discharge of domestic and industrial wastes. The hydraulic characteristic of the Laguna San José (LSJ), its dynamics and nutrient loads are not completely understood. A preliminary simple box model (according to Land-Ocean Interaction in the Coastal Zone-LOICZ guidelines) is proposed to calculate the water budget for the LSJ. Our overall goal is to develop a N budget for the LSJ. This research is relevant to the management and restoration efforts of the SJBE.

Introduction

- The San Juan Bay Estuary (SJBE) in Northern Puerto Rico (Figures 1 and 2) is comprised of five lagoons interconnected by several canals. It represents a unique tropical estuarine ecosystem enclosed within the highly populated San Juan Metropolitan Area.
- Because nitrogen is considered a limiting nutrient in coastal waters (Howarth et al. 1996), hydrologic characterization and analysis of nitrogen fluxes to the SJBE should provide invaluable information to direct pollution control efforts. In addition, since the magnitude and fate of anthropogenic nitrogen inputs to the SJBE are unknown, it is, therefore, of both national and global interest to understand human-related nitrogen fluxes in the tropical SJBE.
- The LOICZ-(Gordon et al., 1994) study of the International Geosphere-Biosphere Programme includes guidelines to estimate water, salt and nitrogen budgets in semi-enclosed coastal areas.
- The objective of this work is to present the relative water contribution of the watersheds that drain into the LSJ, water budget and a conceptual nitrogen budget for the LSJ using a steady-state box model approach.

Materials & Methods

- Direct discharge measurements were made biweekly at permanent cross sections at each watersheds (fig. 1).
- A preliminary water balance budget according to the LOICZ approach (Gordon et al., 1996) was applied to estimate the water residence time of the LSJ. The conceptual premise under the LOICZ approach is to establish a compensatory residual flow ($V_R$) to balance the fresh water volume entering the system, such as runoff ($V_Q$), precipitation ($V_P$), ground water ($V_G$), other flows ($V_O$), and the evaporative losses ($V_E$) (Sylaios 2003).

The water balance for LSJ was calculated using the following equation:

$$ VR = VP - VO - VG + VE $$

The data presented in this poster were summarized from the following reports: Ellis and Gómez-Gómez (1975), Ellis and Gómez-Gómez (1976), and Gómez-Gómez and Ellis (1983), Cerco et al., 2000, and Ortiz-Zayas et al. 2006 and preliminary data obtained from the present dissertation. Meteorological data were gathered based on observations made by the National Weather Service in San Juan, Puerto Rico.

Results, Discussion & Conclusions

- Figure 3 presents the annual water budget for the LSJ. The water exchange (residence time) of LSJ was calculated at 55.6 days.
- A water residual outflow ($V_R$) occurs as a result of the water discharge from the Quebradas Juan Méndez, San Antón, Baldorioty de Castro Pump Station, precipitation ($V_P$) and evaporation ($V_E$). Since the N budget for tropical estuarine ecosystems is not well understood, one of our overall goals is to develop a N budget for LSJ (Figure 4).

References

**Mission**

- Training of human resources at the graduate, undergraduate and post-doctoral levels
- State-of-the-art research in conservation biology and environmental issues
- Infrastructure improvement
- Production of relevant results for policy and conservation management

**Objectives**

- To promote research programs that synthesize multiple levels of biological organization.
- To produce high quality research and students (graduate and undergraduate) in areas of applied ecology and conservation.

**Goals**

- Increase the research productivity of our faculty in the field in applied ecology and conservation.
- Strengthen the participation of our undergraduate and graduate students in research activities.
- Expand the research infrastructure of the University.
- Foster long-term research collaborations among scientists within Puerto Rico and with national and international government and academic institutions.

CATEC research focuses on the impact of climate change and climate variability on fauna, flora, and ecosystem functioning in the Caribbean. We chose the research activities for their potential applications to the Caribbean Basin or the tropics in general.

As a biodiversity hotspot, the Caribbean is home to about 12,000 plant species and 1,500 animal species, with nearly 50 percent endemic to the region. CATEC is studying how past and present climate change affects biodiversity and ecosystem function in the Caribbean and the threats to endangered and economically important native species.

CATEC, an intra campus, and inter-campus Center, has established collaborations with national and international universities and institutions such as, Cornell University, University of Miami, Florida International University, Florida Institute of Technology, San Diego Zoo, Toledo Zoo, Earhart College in Indiana, Universidad Nacional Autónoma de México at Iztacala, Fairchild Botanical Garden, The Leibniz Center for Tropical Marine Ecology Bremen, Germany, New York Botanical Garden, Washington University, Texas A&M College Station, Universidad de Antioquia (Colombia), Universidad Nacional de Colombia at Palmita, Universidad del Zulia and Universidad Simón Bolívar (Venezuela), National Botanical Garden Rafael Moscoso (Dominican Republic), St. Eustatius National Parks (The Netherlands), Universidade Federal do Amazonas (Brazil) among others. We continue to build on the collaborative ties with the Department of Natural Resources and Environment of the Government of Puerto Rico and the US Fish and Wildlife Service.
Spatial Distribution and Performance of Native and Invasive Ardisia (Myrsinaceae) Species in Puerto Rico: The anatomy of an invasion

INTRODUCTION

Establishing and spread of invasive species into new habitats is considered the second most significant threat to biodiversity worldwide1, this is in part due to the displacement and extinction of native species. However, the mechanisms behind the success of exotic invasions are not well understood.

Field comparison between native and non-native or invasive congener pairs, has been considered as an effective method for identifying characteristics that support invasiveness2. This is because native and invasive congeners should share many phenotypic traits and ecological similarity3. Those traits for which an invasive exhibits superior ecological performance are likely to contribute to its invasiveness.

We studied the tree Ardisia elliptica, an invasive species considered among 100 of the “World’s Worst” invaders by the Invasive Species Specialist Group (SSG), and A. obovata a native congener that shares some of the same habitats as the invasive.

HYPOTHESIS AND PREDICTIONS

Invasive species have life history traits for rapid colonization and population growth, and should share many features with the native congeners species in the area where they co-occur. If exotic species perform better than native species, then we expect:

1) Invasive species to have a higher number of individuals in all demographic classes than the native, and spatial patterns should be aggregated and overlap between native and invasive in the study area.

2) Seedling growth should be greater in the invasive species than in the native.

3) The rate of herbivory should be greater for the native species than the invasive.

4) and germination should be greater in the invasive species.

METHODS

1. We established 520 plots (1 plot = 4.9 m2) along the entire Monagas Park trail system. From each plot we counted: early juveniles (<0.7 cm), juveniles (0.7-0.9 cm) and adults of both species. In each plot we took geographical coordinate (GPS).

2. We transplanted 42 seedlings of the invasive species (A. elliptica) pairing each one with the native species (A. obovata) in an area where both species coexist. We also measured 43 randomly selected seedlings of each species, damage-free and approximately the same size as the transplants, in the same habitat. After 6 months we estimated growth (Growth = initial biomass – final biomass).

3. We also determined % herbivory for each leaf and an average for each plant, prior to drying the plants collected from the field.

4. We randomly established thirty plots (60 cm2) along a trail where both species occurred. Each plot had 20 seeds per species; we put a row of A. elliptica followed with a row of A. obovata.

RESULTS

Table 1. Spatial distribution of the exotic A. elliptica and native A. obovata, in each demographic category in the study area. Is: index of aggregation, if the value of Is > 1 is aggregated, Is = 1 is regular; and Is < 1 is random distribution. The p means the significance of the pattern. We used the SSAI program (Spatial Analysis by Distance index) to calculate the Is and X (the saddle mean of spatial association for two sets of data), to determine the probability level (p) 0.025 = significant association or > 0.05 significant disassociation.

<table>
<thead>
<tr>
<th>Species</th>
<th>Category</th>
<th>Is</th>
<th>p</th>
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<td>0.0002</td>
<td>aggregated</td>
</tr>
<tr>
<td></td>
<td>adult</td>
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<td>0.0002</td>
<td>aggregated</td>
</tr>
<tr>
<td></td>
<td>juveniles</td>
<td>4.194</td>
<td>0.0002</td>
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<td>seedling</td>
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<td>0.0002</td>
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<td>seedling</td>
<td>3.825</td>
<td>0.0002</td>
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Fig. 1. Fruit and seeds of A. elliptica (top) and A. obovata (below).  
Fig. 2. The study area, Julio Enrique Monagas Urban Park, located in the metropolitan area of Puerto Rico (32°05'39.5" N, 66°32'23.8" W).  
Fig. 3. Comparison of growth in the exotic Ardisia elliptica and the native A. obovata (Myrsinaceae). Transplanted-annual individuals (a), Field control (b). Average effect medians, as well as 25th and 75th percentiles, whiskers extend to 10th and 90th percentiles.

CONCLUSIONS

• Aggregated spatial pattern is the prevailing pattern in tropical forests and usually is associated with nutrient-rich patches, topography and dispersion capacity.

• The lack of total distribution overlap between the two species may be the result of different time for their establishment and possibly, small differences in preference for microhabitat.

• Contrary to many studies (e.g. Leicht-Young 2007), we found greater growth in the seedling stage by the native species. However, many native plants can grow more and take advantage than exotic species just in particular environments, as greater light conditions, more nutrients available and absence of competitors.

• Comparison between A. elliptica and A. obovata, indicated that herbivores and pathogens showed no preference for any particular species in the forest; percent leaf loss was the same between the two. Either there are no specialist herbivores that attack only the native species, or the same specialist herbivores of the native species also attack the exotic congener.

• The invasive species has higher germinability than native species. Furthermore, more seeds were lost by the native species. Either native seeds are more prone to rot and disappear or they are more susceptible to seed predation.

• The invasive species, A. elliptica, has the faster time of germination and the higher percentage of germination in the field. These two traits contribute to its aggressiveness in the study area.

REFERENCES


ACKNOWLEDGEMENTS

We thank NSF-CREST-CATEC and DEGI for funding, and field assistants.

FOR FURTHER INFORMATION CONTACT

Marcia Carolina Muñoz  
Email: marcarm@gmail.com

Funded by National Science Foundation, HRD #0734826  
and University of Puerto Rico, Central Administration  
and Rio Piedras Campus
SPM Participants

SPM Diversity

Publications and Presentations

Table 1. Presentations by SPM

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<td>3/8 (37%)</td>
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Abstract

Obtaining environmental data may require to access a database. But, what if we have only images available and not a database? We developed an algorithm, using Python™ programming language, and we could access public web images and obtain information from them. Our interest was to access NEXRAD radar images to obtain precipitation data from Isla de Mona and Humacao, Puerto Rico, but this algorithm is also applicable to any other image that represents an environmental data. Eventually, we will be comparing our data results to precipitation measurements, directly obtained from a pluviometer, and to historical data from the NEXRAD radar database.

Methodology

1. Access radar image from NEXRAD website.
2. Save image and localize pixels corresponding to Isla de Mona and Humacao.
3. Save, in a text file, precipitation ranges obtained from the pixels corresponding to Isla de Mona and Humacao.
4. Repeat these first three steps every 350 seconds.
5. After saving all the data in text files (one file per access), merge all the files corresponding to the same date into a single file.
6. Access these single files to obtain each day average of precipitation.
7. Graph averages of precipitation (inches) of each day.

Future Work

1. Compare obtained data to historical data from NEXRAD website database.
2. Create an interface to select precipitation data from specific locations, using information obtained from radar images or from historical data in NEXRAD database.
Seed rain in a removal experiment in a cactus-shrub area invaded by an exotic grass in Mona Island

Abstract
As part of an experiment to test the effects of removal of an invasive species (Megathyrsus maximus) in cactus populations in the Eastern coastal area of Mona Island, we collected seeds and plant parts in tray traps, located beside the experimental plots. Monthly samples were separated (using sieves) and classified in the laboratory. With the exception of seeds that were counted and weighted, other plant parts were oven-dried (60 °C) for biomass measurement. Our results show the seasonal variation of seed diversity and production, and biomass variation of plant parts (leaves, stems, and reproductive components) and their relationship to precipitation and other environmental and biotic factors.

Introduction
The invasion of exotic species in a small island may produce a rapid loss of biota (through competitive exclusion, habitat change) and changes in ecosystem functions. We are investigating the effect of an invasive grass species (Megathyrsus maximus) in a population of cactus in a shrub and dwarf trees community in Mona Island. We are specifically interested in the ecological process of seed dispersal in areas where a removal experiment has been conducted for almost two years. We expect significant changes in the seed rain amount and composition, as well as in the dead plant material collected beside the experimental plots. The differences will reflect the effects of vegetation structure and composition of the experimental plots.

Methodology
The study site is located in the East part of Mona Island, in an area invaded by M. maximus. Figure 2 shows the distribution of the six experimental areas, each of them with three treatment plots (5 x 10 m). Each experimental plot has four collecting trays (50 x 70 cm) at each side. Every month since January 2008 the material collected in each tray is collected and processed in the Laboratory of Terrestrial Ecology and Ecophysiology, at UPR-Humacao. The material from each side is first separated in the following categories: seeds, leaves, reproductive material, stems, grass material (vegetative), and others. All the materials, except seeds, are oven dried at 60°C and their biomasses are determined. The seeds are separated according to their phenotype, and photographed for identification. Whenever possible, seeds are planted to obtain seedlings, and take pictures for later identification.

Figure 1. Plant communities in the East part of Mona Island. (a) Cactus populations with shrubs and dwarf trees. (b) Areas invaded by Megathyrsus maximus.

Table I. Number of seed morphotypes (average and standard error) found in sampling trays located in a non-invaded area and treatment plots in the grass invaded area (see Figure 2.)

<table>
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<th>Sampling Date</th>
<th>No Grass Area</th>
<th>Positive Control</th>
<th>Partial Trimming</th>
<th>Complete Removal</th>
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<tr>
<td>February 2009</td>
<td>2.08 (0.47)</td>
<td>1.86 (0.46)</td>
<td>2.76 (0.46)</td>
<td>1.10 (0.23)</td>
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<td>June 2009</td>
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<td>2.56 (0.34)</td>
<td>2.82 (0.58)</td>
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</table>

Figure 2. Field experimental design.

Figure 3. (a) Average (+s.e.) number of seeds collected in the sampling trays located next to the treatment plots (PC, PT, PR). (b) Average (+s.e.) vegetative biomass (g) of grasses; location and treatments as in (a). Results of ANOVA with repetitions are shown.

Figure 4. Example of seedlings obtained in the laboratory from seeds collected in the sampling trays in the control and experimental locations in Mona Island.

Figure 5. Example of seedlings obtained in the laboratory from seeds collected in the sampling trays in the control and experimental locations in Mona Island.

Leisha Claudio1, Noelia Cruz1, Leidy González1, Xavier Jaime1, Keishla Negron1, Alichy Ubiles1 and Denny S. Fernandez1,
1Biology Department, University of Puerto Rico at Humacao; 2Florida Gulf Coast University

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration, and Río Piedras Campus
Additional support from matching funds from UPR at Humacao
Goals

To serve our community of undergraduate students, providing opportunities to participate in high quality research projects in the field of ecology.

To develop interdisciplinary research and educational projects with members of the UPR system and other local and national universities.

To train undergraduate students on basic and specialized skills applicable to the field of ecology.

To develop an ecoinformatic laboratory to localize, catalog, analyze, synthesize, and disseminate public ecological and environmental data linked mainly to South-East Puerto Rico, and to make it available for undergraduate courses and research projects, and to the local communities.

Undergraduate Education Projects

• Incorporation of mathematical and computational approaches and methods in the ecological curriculum: the LabTEE is the practicing environment for undergraduate students to work in interdisciplinary research projects that requires mathematical and computational tools. Students are offered to take the course Computational and Mathematical Methods Applied to Biological Systems and conduct research at the LabTEE.

• Using large public databases in the ecology and environmental undergraduate curriculum: I am participating in a joint project between the Ecological Society of America, the National Ecological Observatories Network, and the National Center for Ecological Analysis and Synthesis, to develop activities that require the use of large, continental scale datasets in the undergraduate curriculum.

• Extended Summer undergraduate research experience: in collaboration with Case Western Reserve University next year we will start a project that includes the exchange of students between both institutions, and the matching of research mentors to extend the research experience after the Summer, in the participants own institutions. The LabTEE will be the local base for two CWRU students next Summer.

Current Students in the LabTEE

Noelia Cruz – Microbiology
Xavier Jaime – General Biology and Wildlife Management
Alichy Ubies – Wildlife Management
Leidy González – General Biology
Esther Morales – Wildlife Management (MARC student)
Elizabeth Rivera – Mathematics and Biology
Mara Castro – Marine Biology
Emily Pérez – General Biology
plus two graduate students using LabTEE facilities: María Vega (UPR-Mayaguez) and Eveneida Rodríguez (Turabo University)

Recent Publications


Denny S. Fernandez, Biology Department, University of Puerto Rico at Humacao
Introduction

Tropical Estuaries

Site Río Mameyes Estuary

Study Sites

Atlantic Ocean

Legend

Flower River

Figure 1. This figure shows the two watersheds under study: Río Mameyes and Río Sabana.

Table 2.

<table>
<thead>
<tr>
<th>Date</th>
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Conclusions

Estuaries are highly productive coastal ecosystems.

- Influenced by tidal, wave, and wind actions that control water mixing.
- Estuaries recipients of allochthonous organic matter (OM).
- OM is processed and exported to ocean under high-flow conditions (Borges, 2005).
- Estuaries regulate the quantity and quality of OM that reaches the ocean
(McCallister, et al., 2006).

Anthropogenic activities in the coastal region are changing the ecological integrity of the coastal system (Rabouille et al., 2001).

Estuaries are highly productive coastal ecosystems.

- Temperature
- Nutrients
- Organic matter

Objectives

- Study how anthropogenic activities affect the processing and export of carbon and whole community metabolism in tropical estuaries.

Methods

- For six months, each estuary was visited weekly.
- Water column profiling was conducted prior to collect water samples.
- Dissolved oxygen and temperature were measured with a YSI
- Water samples were analyzed with a TOC-Analzyer (Teledyne Tekmar Apollo 9000) and the absorbance at 254nm was measured for each sample for the SUVA₂₄₅₉ determination (Weishaar et al., 2003) on all collected water samples.
- USGS gauging stations daily discharge data was used for the export and mass balance analysis.

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Conclusions (cont.)

The Mameyes Estuary have a higher NPP but also have a higher CR₂₄ than the Sabana Estuary. Could it be possible that there activities in the fields near the estuary are causing nutrient enrichment of the estuary (Table 1.2)?

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The Sabana Estuary have a higher NPP but also have a higher CR₂₄ than the Sabana Estuary. Could it be possible that there activities in the fields near the estuary are causing nutrient enrichment of the estuary (Table 1.2)?
Objective
To compare the exotic freshwater fish community and determine similarity patterns among several reservoirs.

Methods

Sampling
Boat mounted electrofishing
Twice per year (Jan-Jun / Jul-Dec) in five reservoirs.

Eleven fish species
Largemouth bass (Micropterus salmoides)
Peacock bass (Cichla ocellaris)
Armored catfish (Pterygoplichthys spp.)
Redear sunfish (Lepomis microlophus)
Bluegill sunfish (Lepomis macrochirus)
Mozambique tilapia (Oreochromis mossambicus)

Analysis
We used Bray-Curtis ordination to determine the similarity of the exotic freshwater fish communities among several reservoirs. Ordination is a multivariate approach used principally for exploratory analyses in community ecology. Bray Curtis in particular, is a distance based technique that arranges samples using a distance matrix. Thus, similarity equals proximity. Three years of data were analyzed.

Study Sites
Five reservoirs sampled per year (9 distinct reservoirs in total, since some were repeated).

Results

Conclusion
* Each reservoir harbors a particular assemblage of exotic freshwater fish. Knowing these patterns is important since in Puerto Rico the freshwater sport fishery relies almost exclusively on introduced (exotic) fishes like the largemouth bass and butterfly peacock bass.

* Among the recent invasive species coming from the aquarium trade and aquaculture, a few such as the red devil (Amphilophus spp.) and the jaguar guapote (Parachromis managuensis) are slowly being accepted by recreational anglers as legitimate sport fishes.

* This information on the similarity of the exotic fish communities inhabiting Puerto Rico reservoirs can be used to help optimize reservoir management strategies, including focused educational campaigns to avoid additional unplanned species introductions.

* The full impact of these unplanned introductions on the reservoir ecosystems remains to be determined.

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1Puerto Rico Department of Natural and Environmental Resources, 2Center for Applied Tropical Ecology and Conservation

Funded by National Science Foundation, HRD #0734826 and University of Puerto Rico, Central Administration and Río Piedras Campus
Locating Drone Congregation Areas (DCA) or honey bee mating congregations.

Abstract: Honey bee mating at 30 meters above ground and 12 miles/hour makes studies on conservation, hybridization, and mating behavior difficult. Establishing a reliable set of criteria that could aid in finding DCAs is important for conservation, for assessing genetic diversity, and to study bee behavior. Based on published descriptions of few DCAs we made extensive searches in areas that are up to 2 km from known bee colonies. In 36 locations we found 8 DCAs. Through global positioning of the locations and landscape analyses we determined extensive characteristics of each location at 100m, 200m, 400m, and 800m diameter around the geographic center. We used these data in recursive partitioning analyses to identify characteristics associated with presence of a DCA.

1. Introduction

Honey bees are important both for conservation and agriculture as generalist pollinators. Partially because of this importance, we know more on honey bees than almost any other insect. Recent losses of domestic colonies across the world (Giray et al. 2009) show we need to focus on what bees do outside the colony. Understanding navigation and cues that bees utilize could help us understand impact of stressors that may have subtle influences on orientation of bees. Understanding mating behavior can help us determine strategies to preserve genetic variation found in native habitats of the bee. This variation is thought to be an important resource for a resilient agricultural bee population. In Puerto Rico anolf combined with Africanized bees with European introgression and reduced defenses are found (Rivera-Marchand et al. 2008). The hybridization process that occurred in Puerto Rico could be better understood by sampling mating locations or DCAs.

2. Methods and Results

2.a. Preliminary search criteria:

The sites chosen were:

- near known honey bee colonies (1-2km)
- protected from wind by a land projection, etc.
- In open areas (grassland, agricultural, artificial)
- Included potential visual markers (tree line, road)

2.b. DCA searches:

A helium-filled meteorological balloon was fitted with a black cork, (roughly size of a queen, and impregnated with synthetic queen pheromone), and this balloon was raised along transects at each candidate loci, moving further away from known bee colonies, in areas fitting the preliminary search criteria. Once a DCA were found the perimeter was determined by moving away from the drone congregation towards peripheries until drones stopped following the bait. GPS coordinates were recorded.

2.c. Environmental characteristics of DCAs:

We analyzed the environmental characteristics describing areas with DCAs (n = 8) and ones without (n = 28), using Geographic Information Systems (ArcGIS 9.3) analysis (ESRI 2003). For each site we evaluated the role of landscape characteristics such as landcover types (i.e. pasture, urban, crops), climatic factors (i.e. temperature and precipitation), number of apiaries in the vicinity, and river and trail densities within diameters of 100, 200, 400 and 800 meters. To statistically analyze the data we performed a Multi Response Permutation Procedure (PROC-ORD 4.27; McCune et al. 2002) and a Recursive Partitioning Analysis (JMP 5; Quinn and Keough 2002). At 100 and 800m it was not possible to distinguish the areas where DCAs were present or absent. In contrast, at 200 and 400m diameter areas we found characteristics correlated with presence of DCAs.

3. Conclusion and future directions

The most important finding of this study is the presence of a limited set of characteristics that correlate with presence of drone congregation areas. The DCA can not be on a steep terrain, hills or other land projections should have a Southern aspect (open towards south), open areas are extensive, yet urban or forest visual markers are present at about 10% of the cover. Our analysis will include the river, roads, magnetic field anomalies of the DCAs. We will build predictive models to search for DCAs in other locations to test our findings.

Literature Cited:


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Roadside Ménage à Trois: A Native Pest on an Exotic Orchid Weed Defended by Ants?

Abstract  Populations susceptible to herbivory are more susceptible to attack under high density conditions. Naturalized species occasionally produce high density populations because of the absence of predators in their new habitats. Spathoglottis plicata is a naturalized orchid that produces locally dense populations in the Rio Abajo Reserve of Puerto Rico and is attacked by a native florivorous weevil, Stethobaris polita, an orchid specialist. The orchid produces extrafloral nectaries on the floral bracts, buds and developing fruits and has been seen behaving aggressively towards the beetles. The purpose of this project is to determine whether or not the weevil works as a natural biological control, the ants defend the orchid against the beetle, and if either activity is density dependent.

Hypothesis
• If the beetle Stethobaris polita produces significant damage to reproductive organs of the Spathoglottis plicata, then the beetle could be considered as a natural population control agent of this naturalized species.
• Spathoglottis plicata at high population densities will have higher rates of flower damage than at low densities because beetles respond to higher resource availability.

Materials and Methods
Study System Spathoglottis plicata is a naturalized exotic orchid species native to Southeast Asia. It possesses extrafloral nectaries that are frequently visited by ants. The plant shows a high fruit set suggesting autogamous pollination.

Study Sites We have two study sites in Puerto Rico: the Rio Abajo Biological Reserve and the Julio Enrique Monagas Urban Park. Rio Abajo Reserve is in north-central Puerto Rico, near the border of the Municipalities of Utuado and Arecibo, Route 10, km 72.9 and 73.3 in a moist limestone region at 18°19.00’N, 66°43.00’W. Julio Enrique Monagas Park is in Bayamón, forming part of the San Juan metropolitan area in the northeast. Both study sites are within the mogotes, a region of limestone haystack hills. The Rio Abajo population of S. plicata is attacked by Stethobaris polita whereas plants in Monagas Park are beetle-free. Spathoglottis plicata in both study sites are visited by several different species of ants.

Data collection We selected at least 50 target plants of white and also pink Spathoglottis plicata, each with developing inflorescences from August 2008 to February 2009 in areas with diverse population densities. We performed monthly censuses on the target plant as well as its five nearest S. plicata neighbors. We recorded the number of flowers, flower damage, fruit production, and the number of ants and weevils on the inflorescence. We also measured the length of the longest leaf, number of leaves and the height of the inflorescence of the target plant. We tested for differences among the two morphs of S. plicata: white and magenta using t-tests. We evaluated the relationship between weevil and ant densities to target plant fruit production and the relationship between the density of flowers and inflorescences to the number of weevils and ants in the area (JMP Version 4.0.4, SAS Institute).

Preliminary Results and Discussion
General characteristics of the two morph of S. plicata and the presence of weevils and ants.

We found a low correlation between number of weevils and magenta flower damage. Also ant density was significant in the fruit production for the white morph. More data on low density populations is required to fully understand the orchid/weevil/ant interaction.

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