Final Report
Sea Grant March 3, 2009

Executive Summary
Project Title: Developing a protocol to use remote sensing as a cost effective tool to monitor contamination of mangrove wetlands
Date: 3 march 2009
Project Number: R-21-1-06.
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Dates covered: 22 May 2006 to 1 March 2008 and extension until March 2009

A. Executive Summary

Summary of Impacts and Contributions

1. Objectives: To apply the remote sensing techniques of mineral exploration to the monitoring of mangrove wetlands for the presence of metal contamination. Similar as in mineral exploration, a cost effective technique that does not require costly field studies, will make an excellent tool for government agencies in charge of monitoring the health of wetlands and the possible contamination of mangrove forests.

   The research involved:

   1) The analyses of substrate and leaves in the top of the canopy of mangroves. The selection of a non-contaminated control area (e.g. Guanica, Punta Ballena, and compare the chemical results, the reflectance spectra of the leaves, the Normalized Difference Vegetation Index (NDVI), and the pH of the seawater, with other possible contaminated sites. For the latter the following sites were selected Joyuda Lagoon with possible Ni, Co contamination next to a Ni-Co laterite, Arecibo Lagoon in the watershed of the porphyry copper deposits and with various industrial sites, and Guayanilla Bay with reported mercury contamination.
   2) Processing of satellite images using differences in reflectance.
   3) Plot data in a Geographical Information System (GIS) for use by agencies in charge of pollution control.

   Objective 1) was partially established. The analyses of some of the heavy metals (e.g. As, Hg) needed chemicals that required an explosive license as ordered by Homeland Security. None of the PIs had this license and efforts to find such a person willing to help us were unsuccessful. Discussions with the Purchasing Department seemed to work, but still the chemicals were never purchased. Objective 2) a pilot project using AVIRIS near Punta Ballena did not provide the desired results, due to discrepancies between ground-truthing and the image data.

Objective 3) was never started. The original idea was that the recruited graduate student would carry out this part.

2. Advancement of the Field
   The following discoveries were made and will help future work
   a. Transport of metals from the substrate to the top leaves and the transport of metals within the tree is not the same for all metals.
   b. Transport of metals into, and within the red mangrove (*Rhizophora mangle*) is good to excellent for Cu, Co, Pb, and Cr, whereas the transport of Ni is little and the transport for Cd is none existent. Black mangrove (*Avicennia germinans*) seems to concentrate Cu, Co, and Cd.
   c. In summary red mangrove can be used to monitor for Cu, Co and possibly Pb and Cr, but not for Ni and Cd. Black mangrove can be used to monitor for Cu, Co and Cd.
3. Problems encountered
   A major problem involved the chemical analyses, due to the requirement of having an explosive license to purchase the needed chemicals. We tried unsuccessfully to find a person who could do the purchasing for us, subsequently we explained the Purchasing Dept. we only needed little quantities and they promised to process the order, but no chemicals were ever purchased. We are now discussing with Dr. Massol if we can solve the problem.
4. Research Impacts
   The discovery of the behavior of metals in the various mangroves.
5. Other important impacts or products
   a. List of students supported and otherwise involved

      Graduate Students employed:
      Ms Marianela Mercado Burgos Aug 2007 – Dec 2008
      nela.guayaba@gmail.com
      Ms. Mercado was recruited as graduate student to work in the project. She was supported for one semester. After that she decided to change her research project.

      Undergraduate students employed:
      <u>Mr. Augustine Rodríguez-Román.</u> Aug 2006 - May 2007
      ride_with_stylee@hotmail.com
      Mr. Rodriguez had carried out two undergraduate research projects in preparation of the proposal: Rodríguez-Román, A. (2005 a b)
      Mr. Rodriguez became the most experienced undergraduate student participating in all the steps of the sampling of soil and leaves, determining GPS locations, obtaining pH of water data, and analyzing soil and leaves. He co-authored an internal research report with Ms. Delíz: Delíz and Rodriguez (2007)

Ms. Belyneth Delíz-López Aug 2006 - May 2007
belydel@hotmail.com
Ms. Delíz was recruited as undergraduate research assistant and participated in all the steps of the sampling of soil and leaves, determining GPS locations, obtaining pH of water data, and analyzing soil and leaves. She co-authored an internal research report with Ms. Delíz: Delíz and Rodriguez (2007)

Ms. Angela Perez Jan - May 2007 and Aug – Dec 2007
isabela117@gmail.com
Ms. Perez was recruited as an undergraduate research assistant and carried out sampling of leaves and soils and determined the reflectance of the leaves.

Other undergraduate students involved
Ms. Almaris Martínez-Colón
Ms. Martinez participated as an undergraduate research student in the required course GEOL 4055. She compared the reflection of different levels of red mangrove leaves in Joyuda and Guanica. She produced an undergraduate research report: Martinez-Colon (2006).

Ms. Yomayra Román-Colón
yomayra.roman@gmail.com
Ms. Roman participated as an undergraduate research student in the required class GEOL 4049. She compared reflectance spectra of mangroves in the Punta Ballena area with reflectance data obtained from AVIRIS images. She produced the undergraduate research report: Roman-Colon (2006)

Graduate student research report
Mercado-Burgos and Viguilla, 2007
Undergraduate Research Reports (see full title in bibliography)
Delíz-López and Rodríguez -Román, 2006
Martínez-Colón, 2006
Rodríguez-Román, 2006.
Román-Colón, 2006

6. Sources of matching funds
   No matching funds were obtained. However the Department of Geology provided transportation during the field work.
7. New extra mural funds
   No extra mural funds were obtained
8. Benefits:
   No additional impacts or contributions beyond those described above

B. Final Report Narrative

Brief statement of the problem

Vegetation may take up the metals in their roots, stems, and leaves, and serve as sensors of contamination that integrate pollution over longer time periods, less dependant on daily or seasonal fluctuations. The characteristic that vegetation reacts to the geochemical conditions of the substrate has found a use in the remote sensing techniques applied to mineral exploration, where large areas can be efficiently surveyed without expensive field studies (Goetz et al., 1983). Labovitz et al. (1983) demonstrated that the metal content in the soil changed the leaf reflectance, especially in those parts of the spectrum used for chlorophyll content and water absorption, and that the variation in trace element content was associated with leaf reflectance. Schellekens et al. (2005) studied the application of remote sensing for metal content in the substrate in a diverse tropical forest.

The proposed study tries to apply the remote sensing techniques of mineral exploration to the monitoring of mangrove wetlands for the presence of metal contamination. Mangrove wetlands are specially well-suited for this technique, because the vegetation in mangrove wetlands is not very diverse, four species are known to occur in Puerto Rico, with the red mangrove (*Rhizophora mangle*), the white mangrove (*Laguncularia racemosa*), and the black mangrove (*Avicennia germinans*) as the most abundant. Similar as in mineral exploration, a cost effective technique that does not require costly field studies, will make an excellent tool for government agencies in charge of monitoring the health wetlands and the possible contamination of mangrove forests.

Similar as in mineral exploration, a cost effective technique that does not require costly field studies, will make an excellent tool for government agencies in charge of monitoring the health wetlands and the possible contamination of mangrove forests.

Methods used

In order to correlate the reflectance pattern with contamination or non-contamination of the substrate, chemical analyses for heavy metals were carried out for sediments in the root system and leaves in the top of the canopy. These high leaves are selected because these are observed by remote sensing techniques. However first the assumption that the metals are transported from the substrate to the highest leaves had to be tested. Therefore as a first experiment in addition the analyses of substrate and top leaves, leaves of the lowest braches and the intermediate branches were analyzed, both for metal content as well as reflectance. The analyses were carried out on an Atomic Absorption Spectrometer, following procedure described by Massol-Deya et al (2005). The reflectance spectra of the leaves were determined in the field using the GER 1500 spectroradiometer. The pH and temperature of the seawater were determined using a Thermo Orion pH meter model 230A.

To obtain the desired results the following pilot project was carried out:

1. Comparison of known heavy metal contaminated mangroves with non-contaminated mangroves
    a. Joyuda lagoon next to the nickel-cobalt laterite (Acevedo et al., 2000)

b. Guayanilla reported mercury contamination (Lopez and Teas, 1978; Stary and Lopez, 1979)
c. Arecibo lagoon in watershed with porphyry copper deposits (Bawiec, 2000)
d. Guanica and Punta Ballena, pristine environments next to the Guanica subtropical dry forest having no run-off. No metal contamination discovered by Massol-Deya et al., 2005)

2. Study of the transport of metals from the substrate to higher levels in the mangrove, and study of the transport of metal within the mangrove tree.
   a. Red mangrove (*Rhizophora mangle)* in Guayanilla and Joyuda.
   b. Black mangrove (*Avicennia germinans*) in Punta Ballena.
3. The use of AVIRIS images in Punta Ballena. Comparison of reflectance patterns in mangroves with reflectance patterns of pixels in the AVIRIS image.

Results and findings

1. Site descriptions
   a. The Joyuda lagoon is situated next to the nickel-cobalt laterite deposit bordering the lagoon on the landward side (Heydenreich and Reynolds, 1959; Cram, 1972). The laterite forms a high and water draining from the laterite end up in the lagoon. As a result Ni and Co are expected to occur in the lagonal sediments.
   b. Guayanilla Bay is a known area of mercury contamination (Lopez and Teas, 1978). Guayanilla Bay is located in the Yauco River Valley surrounded by Ponce Limestone, mudstone from the Juana Diaz Formation and alluvial sediments. The Yauco River also brings additional sediments from Upper Cretaceous volcanic derived rocks. Along the Guayanilla Bay, many industries can be observed (Rodriguez, 2006) Stary and Lopez (1979) tried to establish a base-line for mercury concentrations in probably non-contaminated mangrove swamps, using leaves, wood, roots and propagules of mangroves to compare these to mercury levels in Guayanilla Bay. Mercury levels in Guayanilla Bay when compared to other locations expressed values 10 times higher than of other coastal areas such as the Joyuda Lagoon, Punta Ostiones, Guanica Bay, and the Phosphorescent Bay (Stary and Lopez, 1979).
   c. Arecibo and is located in the northern part of the island and has a substrate that is derived from limestone, and carbonate sediments. The mangroves in the Arecibo area thrive in an enclosed lagoon. In this site was the AAA Sewage Treatment Plant, the lagoon was near an urbanization, and the rice plantation *D'Aqui* was located near this vast wetland environment.
   d. The Guanica and Punta Ballenas sites are located along the shore of the Guanica Subtropical Dry Forest. Substrate of these mangroves consists of limestone fragments derived from the on-land exposures of the limestone in the Guanica area and the carbonates produced by corals and other marine organisms. No surface drainage occurs from the land into the marine environment. In addition Massol-Deya et al. (2005) analyzed the sediments in the bay and did not encounter metal contamination.

Analyses for metal concentrations in the substrate and the leaves of different mangroves are given in Appendix 1 tables 1 to 5

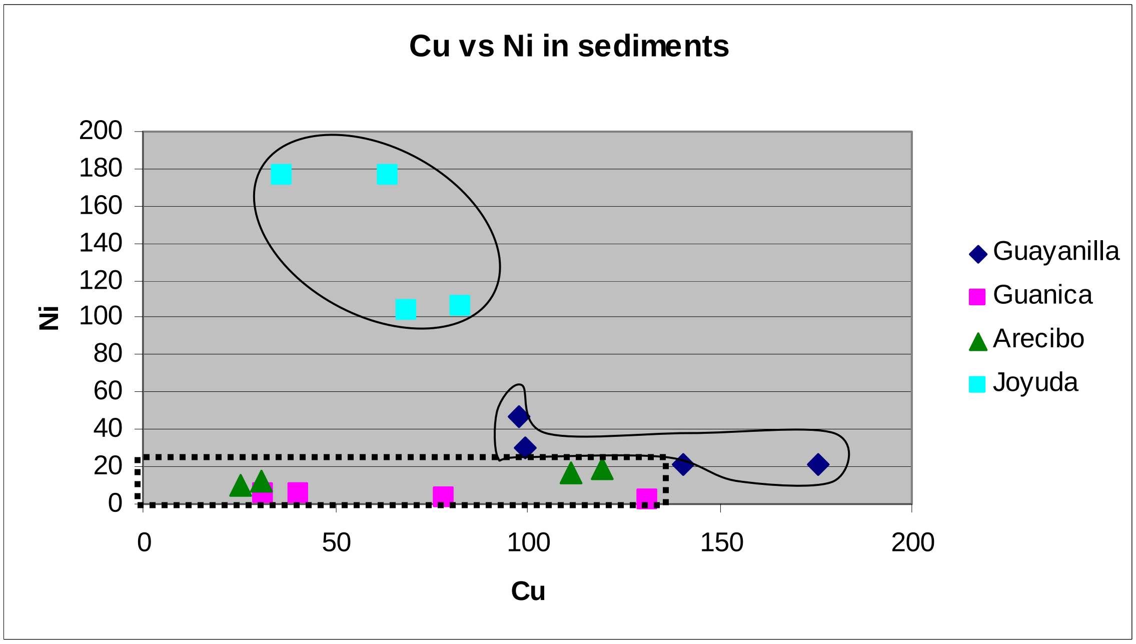


Figure 1 Cu vs Ni in sediments. Guanica is considered the control as a non-contaminated site. The results are found in the dashed lined box in the figure. Notice the high Ni content in sediments of Joyuda lagoon next to the Ni-Co laterite. Guayanilla has slightly higher Cu and Ni than the control.

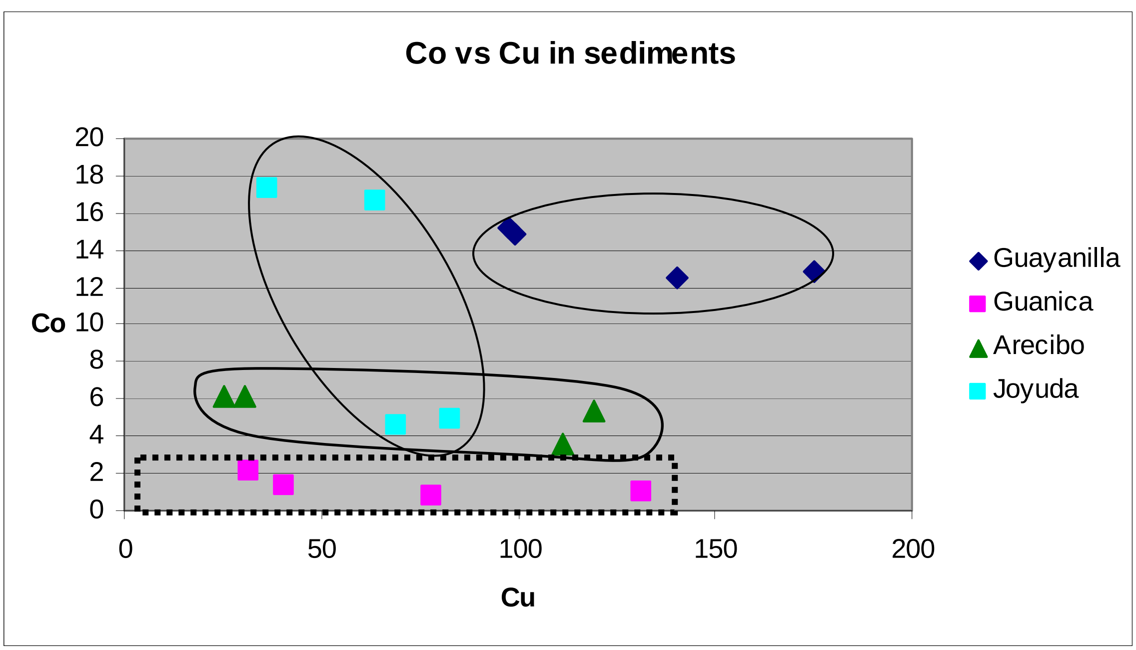


Figure 2 Co vs Cu in sediments. Guanica is considered the control as a non-contaminated site. The results are found in the dashed lined box in the figure. Guayanilla and Joyuda show distinctly higher cobalt contents, whereas Arecibo is less contaminated

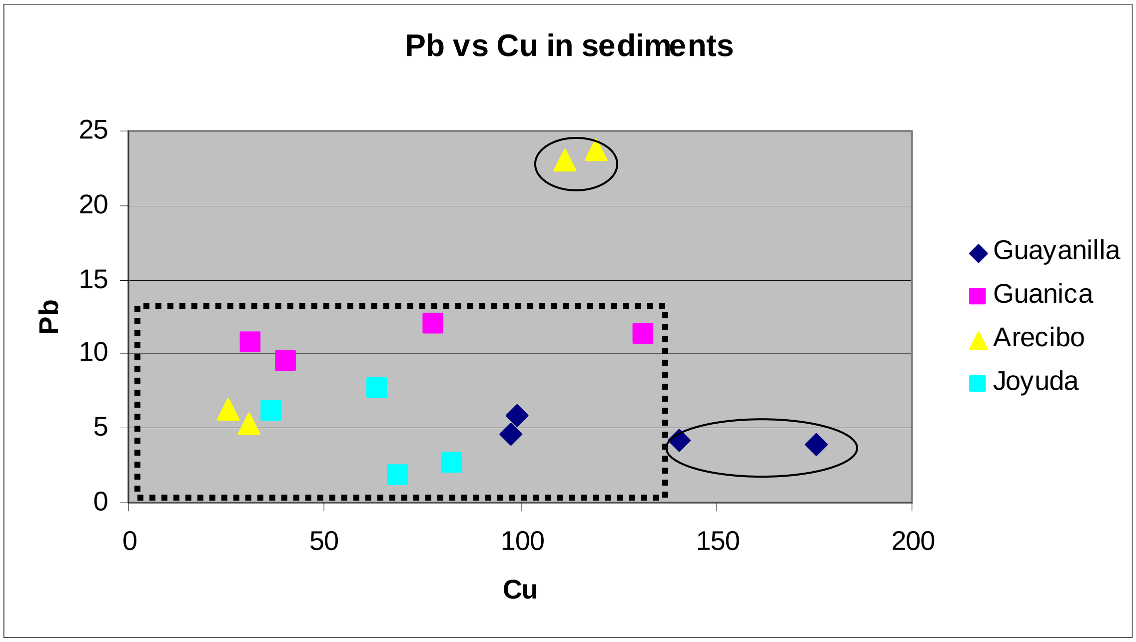


Figure 3 Pb vs Cu in sediments. Guanica is considered the control as a non-contaminated site. The results are found in the dashed lined box in the figure. The Pb concentration of the non-contaminated Guanica site is higher than possible contaminated sites. Two stations in Arecibo show distinct lead contamination.

2. Metal transport from substrate to top of canopy and within the tree.

In order to assess the uptake of metals in the mangrove tree and its transport to the top leaves of the canopy, an enrichment ratio was defined where the metal content of the top leaves was divided by the metal content of the substrate. An example is given in figure 4 for a red mangrove from Guayanilla. In this case the enrichment ratio came out as 0.41 suggesting good transport from substrate to top leaves.

Average substrate	Average bottom leaves	Average middle leaves	Average top leaves	Ratio Top/substrate
98.4	31.2	33.7	40.1	0.41

Figure 4. Example of enrichment ratio for red mangrove (*Rhyzophora mangle*) in Guayanilla.

- **Summary of top/substrate ratios for red mangrove (*Rhizophora mangle)***

– **Cu**		
• Guayanilla	0.4	observable transport
• Joyuda	1.2	metal concentration in tree
• Punta Ballena	0.2	observable
– **Ni**		
• Guayanilla	0.01	very little uptake
• Joyuda	0.02	very little uptake
– **Co**		
• Guayanilla	0.2	observable

In order to assess the transport within the tree an enrichment ratio was defined where the metal content of the top leaves where divided by the metal content of the lower leaves. An example is given for the same red mangrove in Guayanilla. Notice here that the ratio is >1, indicating that the tree concentrates Cu extracted from the soil.

Average substrate	Average bottom leaves	Average middle leaves	Average top leaves	Ratio Top/bottom
98.4	31.2	33.7	40.1	1.3

Figure 5. Example of enrichment ratio within the tree for red mangrove (*Rhyzophora mangle*) in Guayanilla. Notice concentration of Cu in top leaves ratio>1

- **Summary of top/bottom ratios for red mangrove (*Rhizophora mangle)***

– **Cu**	Guayanilla	1.3: good transport
	• Joyuda	1.4: good transport
– **Ni**	Guayanilla	0.3: little transport
	• Joyuda	0.4: little transport
– **Co**	Guayanilla	2.5: very good transport
– **Cd**	Guayanilla	0: no transport
– **Pb**	Guayanilla	0.9: good transport
– **Cr**	Guayanilla	1.3: good transport

- **Summary of top/substrate ratios for black mangrove (*Avicennia germinans)***

–	**Cu**	Pta Ballena	6.5: concentration
–	**Co**	Pta Ballena	concentration
–	**Cd**	Pta Ballena	concentration

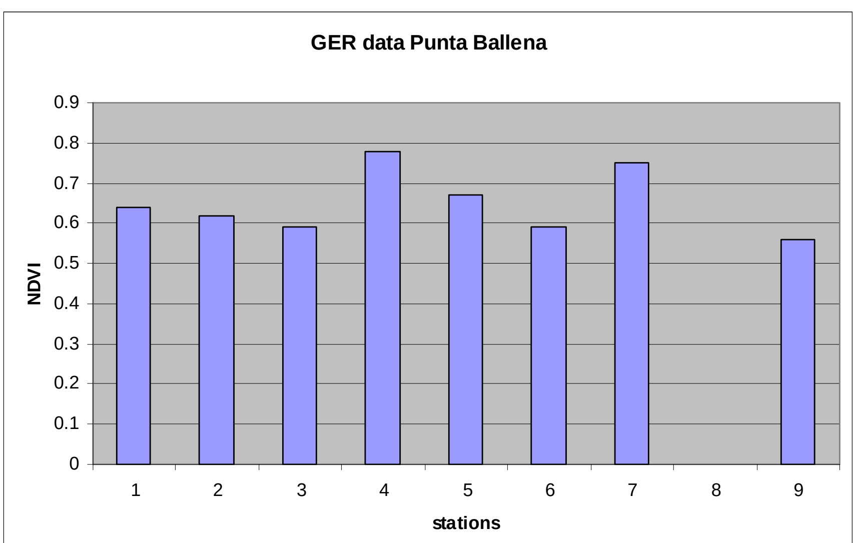
Co and Cd are below detection in the substrate but Co and Cd have an average of 7.6 ppm and 0.3 ppm respectively in the top leaves

In order to use mangrove as a monitor plant using remote sensing, the tree has to take up the metal from the substrate and transport it to the top of the canopy. On the basis of the present data it is suggested that red mangrove (*Rhizophora mangle*) can be used to monitor for Cu, Co, and possibly Pb and Cr, but not for Ni and Cd. The black mangrove (Avicennia germinans) can be used to monitor for Cu, Co, and Cd.

3. Use of AVIRIS (Advanced Visible and InfraRed Imaging Spectrometer)

The AVIRIS has the capability to determine a reflectance spectrum for each pixel and to calculate the Normalized Difference Vegetation Index (NDVI) per pixel. In the Punta Ballena area a pilot project was carried out where the reflectance spectra was measured for mangrove trees using the GER-1500 spectroradiometer. The station was located using a GPS. NDVIs for the tree determined on the ground were compared to NDVIs determined by AVIRIS. Stations 1 to 6 were averaged, and the average NDVI compared to the NDVI determined by AVIRIS for this area. Similarly the average of station 7 and 9 was compared to the NDVI determined by AVIRIS for this area. In both cases the NDVI determined by AVIRIS was too high. This was due to the fact that single trees were smaller than 1 pixel and reflectance from other components of the pixel, like sand, was added to the reflectance of the mangrove.

A



B

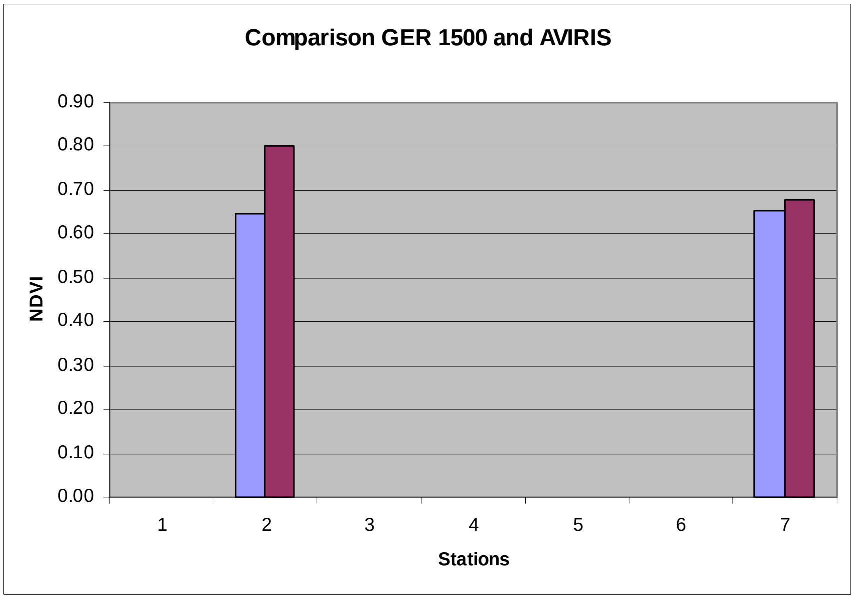


Figure 6. A) NDVIs measured with the handheld spectroradiometer GER 1500 shows a wide spread of reflectance data. B) Comparison of average NDVI of station 1-6 and station 7 to 9 (both blue) with their respective AVIRIS data (red).

Objectives accomplished or not accomplished

Chemical analyses of the substrate and leaves were to be carried out of As, Cd, Cr, Pb, Hg, Ni, Co. This objective was only partly met. Not all elements could be analyzed all the time and the elements As and Hg required chemicals for which we needed a ‘explosive license’. Nor the PI or the Co-Pi had such a license and attempts to use a license in the Biology department failed. The Co-PI explained the purchasing department that only a small amount was needed for the analyses. The purchasing department agreed to buy the chemicals but never did. A number of analyses were carried out (see Appendix 1).

Chemical analyses and reflectance spectra from leaves in the top of the canopy. Chemical analyses of the leaves encountered the same problem as the analyses of the substrate. The reflectance spectra were determined from most of the leaves (bottom, middle, and top). However in the last year the radiospectrometer had to be returned to the factory for servicing and calibration. No spectra could be measured in that time.

Compare the reflectance data for contaminated and non-contaminated sites. NDVIs were calculated for red, black, and white mangroves in contaminated and non-contaminated sites. However no statistically valid difference between the two could be discerned.

Process satellite images using the differences in reflectance. Except for a pilot project using AVIRIS this objective was not met. The main reason being that no robust chemical data existed for the various areas and we waited with the purchase of the images until we were certain in which area we could detect contamination.

Discussion of project impacts and products

a. List of students supported and otherwise involved

Graduate Students employed:

Ms Marianela Mercado Burgos Aug 2007 - Dec2008
nela.guayaba@gmail.com
Ms. Mercado was recruited a graduate student to work in the project. She was supported as a graduate student for one semester. She co-authored a report in a Remote sensing class that redid a previous undergraduate research project without contributing new data. (Mercado-Burgos and Veguilla, 2007). After one semester she decided she wanted to change her research project.

Undergraduate students employed:

Mr. Augustine Rodríguez-Román. Aug 2006 - May 2007
ride_with_stylee@hotmail.com
Mr. Rodriguez had carried out two undergraduate research projects in preparation of the proposal:

Augustine Rodríguez-Román: Leaf reflectance of possibly heavy metal contaminated mangroves compared to non-contaminated mangroves: A possible tool to discern heavy metal contamination using remote sensing. [2005]

Augustine Rodríguez-Román: Geo-biological study of heavy metal contamination in coastal areas: Remote sensing techniques applied for mineral exploration [2005]

Mr. Rodriguez became the most experienced undergraduate student, taking samples of both soil and leaves, determining GPS location of sample sites, carrying out measurements of reflectance of leaves, and determining the pH of the water. Mr. Rodriguez also carried out the chemical analyses of the leaves and soil samples. Mr. Rodriguez was an undergraduate research assistant during a large time of the project. He co-authored an internal research report with Ms. Delíz: Delíz and Rodríguez (2007)

Ms. Belyneth Deliz-López Aug 2006 - May 2007
belydel@hotmail.com
Ms. Delíz was recruited as undergraduate research assistant and participated in the collection of samples of both soil and leaves, determining GPS location of sample sites, carrying out measurements of reflectance of leaves, and determining the pH of the water. Ms. Delíz also carried out the chemical analyses of the leaves and soil samples. Together

with Mr. Rodriguez she co-authored an internal report: Delíz and Rodríguez (2007)

Ms. Angela Perez Jan - May 2007 and Aug – Dec 2007
isabela117@gmail.com
Ms. Perez was recruited as an undergraduate research assistant. She carried out sampling of leaves and soils and determined the reflectance of the leaves. Unfortunately during here participation in the project no chemical analyses were carried due to the problems purchasing chemicals, and the GER radiospectrometer had to be send away for servicing.

Other undergraduate students involved
Ms. Almaris Martínez-Colón
Ms. Martinez participated as an undergraduate research student in the required course GEOL 4055. She compared the reflection of different levels of red mangrove leaves in Joyuda and Guanica. She produced an undergraduate research report: Martinez-Colon, Almaris, (2006)

Ms. Yomayra Román-Colón
yomayra.roman@gmail.com
Ms. Roman participated as an undergraduate research student in the required class GEOL 4049. She compared reflectance spectra of mangroves in the Punta Ballena area with reflectance data obtained from AVIRIS images. She produced the undergraduate research report: Román-Colón, Yomayra A. (2006)
http://gers.uprm.edu/pdfs/topico_yomayra1.pdf

Graduate student Research Reports

Mercado Burgos, Marianela and Veguilla, Ricardo, 2007, Relación entre NDVI y contenido de metales en Rhizophora mangle en el suroeste de Puerto Rico: unpublished research paper in GEOL 6225 Advanced geological remote sensing. http://gers.uprm.edu/geol6225/pdfs/mercado_veguilla_2007.pdf

Undergraduate Research Reports

Deliz-López, Belyneth and Rodríguez -Román, Augustine, 2006, Monitoring metal contamination of mangroves using remote sensing techniques: Guayanilla: Internal Research report Dept. of Geology, UPRM

Martínez-Colón, Almaris, 2006, Distribution of metals and leave reflectance in red mangrove (Rhyzophora mangle). Comparison between Joyuda and Guanica mangrove areas: Unpublished Undergraduate Research report Dept. of Geology, UPRM

Rodríguez-Román. Augustine, 2006, Remote sensing techniques for mineral exploration used to monitor metal contamination of

mangroves: The Guayanilla and Arecibo mangroves: Unpublished Undergraduate Research report Dept Geology UPRM, 25p.

Román-Colon, Yomayra A., 2006, The use of AVIRIS to monitor the contamination in mangrove wetlands, Unpublished report undergraduate research Dept Geology UPRM, 15p. http://gers.uprm.edu/pdfs/topico_yomayra1.pdf

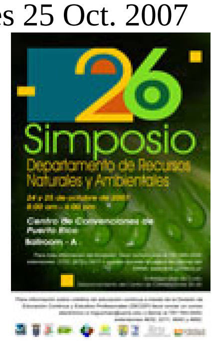
Presentations:

"Usando Percepción remota como una herramienta para monitorear contaminación de mangles" Sociedad Horticultura del Oeste, San German, PR, 1 October 2006.

"Developing a protocol to use remote sensing as a cost effective tool to monitor contamination of mangrove wetlands" 2nd Annual Symposium for Coastal and Marine Research, UPR Sea Grant College program Mayagüez, PR, 5 October 2006.

"Can we use remote sensing to monitor contamination in mangrove wetlands?" Sociedad Geologica Estudantil, UPRM, Mayagüez, PR, 1 March 2007

"Developing a protocol to use remote sensing as a cost effective tool to monitor contamination of mangrove wetlands: Initial results" XXVI Simposio del Departamento de Recursos Naturales y Ambientales 25 Oct. 2007



Abstracts published (students in bold)

Rodríguez-Román, Augustine and J.H. Schellekens, 2006, Leaf reflectance comparison between possibly heavy metal contaminated mangroves and non-contaminated mangroves: A possible tool to discern heavy metal contamination using remote sensing: Abstracts Sigma Xi student poster day Mayagüez, PR April 6, 2006

Schellekens, J.H., F. Gilbes-Santaella, **A. Rodríguez-Román, Yomayra Román -Colon,** 2006, Developing a protocol to use remote sensing as a cost effective tool to monitor contamination of mangrove wetlands: Abstracts of the 2nd Annual Symposium for Coastal and Marine Research, UPR Sea Grant College program October 5, 2006 Mayagüez, Puerto Rico, p. 9.

Rodriguez, Augustine, Angela Perez, Belyneth Delíz, Yomayra Román, Almaris Martínez, J.H. Schellekens, F. Gilbes, 2007, Remote sensing techniques for mineral exploration used to monitor metal contamination of mangroves: Program and abstracts Sigma Xi XII Posterday, UPRM, Mayagüez, Puerto Rico 26 April 2006, p. 34

Schellekens, J.H., F. Gilbes, **A. Rodríguez, B. Deliz,** and **Y. Roman,** 2007, Exploring remote sensing as a cost effective tool to monitor contamination of mangrove wetlands, XXVI Simposio del Departamento de Recursos Naturales y Ambientales de Puerto Rico, October 2007, San Juan, Puerto Rico.

Unpublished reports:

Schellekens, J.H., Gilbes, G., **Rodríguez, A., Deliz, B., and Martínez, A**., 2007, Preliminary results of the mangrove reflectance and composition study, 11p.

Annual reports for Sea Grant are available at:
For 2008: http://gers.uprm.edu/pdfs/report_mangrove08.pdf
For 2007: http://gers.uprm.edu/pdfs/report_mangrove07.pdf

The project has obtained valuable data that can be applied in further research on the topic, such as the transport of certain metals in either red or black mangroves and hence which mangroves can be used for the monitoring of certain metals and which not..
Unfortunately the research project was delayed by various factors, including the problems with purchasing chemicals, the loss of the graduate student, and the sabbatical leave of one of the PIs.
In summary the following conclusions can be drawn from the research:

a. The substrate of mangroves does contain considerable amounts of metals
b. Not all metals are taken up by the mangroves:
   Cu, Co, Pb, Cr are taken up by red mangrove (*Rhizophora mangle*) but Ni and Cd are not.
   Cu, Co, and Cd are taken up and even concentrated by black mangrove (*Avicennia germinans*)
c. The reflectance spectra and NDVI of mangroves when measured on the ground yield a wide range of values. Averaging the data seem to yield consistent results, however with the limited amount of data obtained, no statistically valid differences could be observed between contaminated and non-contaminated mangrove forests.
d. AVIRIS images can be used, but ground truthing has to take into account the larger pixel size (~32m) as compared to IKONOS (2m)

Recommendations

In order to close this research project with publishable results it is recommended to spend a few extra months to carry out the following steps

1. Sampling and analyses of black mangrove at other sites, to establish whether the black mangrove indeed concentrates certain heavy metals. It would make the black mangrove a candidate to be used to clean up contaminated coastal areas.
2. Concentrate on one contaminated area (either Joyuda or Arecibo) to compare the satellite images with the non-contaminated control area in Guanica-Punta Ballena.

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Appendix 1

Table 1 Guayanilla: Heavy metal concentrations Rhyzophora mangle

Cu in ppm

Site	substrate	Bottom	Middle	Top
GL1	97.6	31.1	24.2	24.4
GL2	Average 98.4	28.8	25.0	27.1
GL3	99.2	33.7	51.9	68.9

Ni in ppm

Site	substrate	Bottom	Middle	Top
GL1	46.7	1.9	1.2	0.6
GL2	Average 38.2	1.3	1.1	0.4
GL3	29.7	0.7	0.2	0.3

Co in ppm

Site	substrate	Bottom	Middle	Top
GL1	15.2	0.1	nd	nd
GL2	Average 15.1	nd	0.1	0
GL3	14.9	0.1	0.2	0.5

Cd in ppm

Site	substrate	Bottom	Middle	Top
GL1	Nd	0.1	0.1	0
GL2	Average nd	nd	0.1	0
GL3	Nd	0	nd	nd

Pb in ppm

Site	Substrate	Bottom	Middle	Top
GL1		6.8	10.0	9.8
GL2		14.1	12.1	7.2
GL3		8.3	8.4	10.5

Cr in ppm

Site	substrate	Bottom	Middle	Top
GL1		Nd	0.4	1.4
GL2		7.6	2.1	2.3
GL3		5.0	3.0	13.1

All results are the average of two analyses. When no reading was obtained the result is reported as non detected (nd)

Table 2 Joyuda: Heavy metal concentrations Rhyzophora mangle

Cu in ppm

Site	substrate	Bottom	Middle	Top
JO1	36.4	53.0	78.8	60.2
JO2	Average 50.1	65.3	18.9	99.7
JO3	63.9	9.9	19	18.1

Ni in ppm

Site	substrate	Bottom	Middle	Top
JO1	177.0	3.8	22.0	3.8
JO2	Average 176.5	11.9	6.3	4.4
JO3	176.0	4.1	5.8	0.6

Co in ppm

Site	substrate	Bottom	Middle	Top
JO1	17.3			
JO2	Average 16.9			
JO3	16.6			

Cd in ppm

Site	substrate	Bottom	Middle	Top
JO1	Nd	0.4	nd	nd
JO2		nd	nd	nd
JO3	Nd	nd	nd	nd

Table 3 Heavy metal concentrations in ppm for Rhyzophora mangle

	Guanica substrate			**Guayanilla substrate**			**Arecibo substrate**			**Joyuda substrate**		
Cu	131.6	78.2		97.6	99.2		119.4	111.4		36.4	63.9	
Ni	1.9	3.2		46.7	29.7		19.6	16.3		177.0	176.0	
Co	1.0	0.8		15.2	14.9		5.4	3.6		17.3	16.4	
Cd	0.0	Nd		Nd	nd		0.1	0.1		Nd	nd	
Pb												
Cr												

	Punta Ballena substrate			**leave**								
	4	**5**	**6**	**4**								
Cu	114.0	92.5	60.5	20.5								
Ni												
Co	16.9	19.6	21.0	nd								
Cd	0.36	0.34	0.28	nd								

Pb												
Cr												

Table 4 Heavy metal concentrations in ppm for Avicenna germinans

	Guanica substrate			**Guayanilla substrate**			**Arecibo substrate**			**Joyuda substrate**		
Cu										**82.7**	**68.9**	
Ni										**105.7**	**104.3**	
Co										**4.9**	**4.6**	
Cd										**Nd**	**nd**	
Pb												
Cr												

	Punta Ballena substrate				**Punta Ballena leaves**					
	1	7	8	9	1	2	3	7	8	9
Cu	20.5	13.6	10.4		21.3	94.3	41.1	170.0	109.0	144.0
Ni										
Co	nd	nd	nd		3.4	9.8	15.0	15.8	1.4	0.16
Cd	nd	nd	nd		0.5	0.6	0.7	0.2	nd	nd
Pb										
Cr										

Table 5 Heavy metal concentrations in ppm for Laguncularia racemosa

	Guanica substrate			**Guayanilla substrate**			**Arecibo substrate**			**Joyuda substrate**		
Cu	**40.5**	**31.4**		**140.4**	**175.4**		**25.4**	**30.9**		**-**	**-**	
Ni	**5.2**	**5.6**		**21.1**	**20.1**		**10.0**	**12.7**		**-**	**-**	
Co	**1.4**	**2.1**		**12.5**	**12.9**		**6.1**	**6.2**		**-**	**-**	
Cd	**0.1**	**Nd**		**Nd**	**nd**		**Nd**	**Nd**		**-**	**-**	
Pb												
Cr												