University of Puerto Rico – Mayagüez Campus
College of Agricultural Sciences
Department of Crops and Agro-environmental Sciences

Summary report for project: Citizen Monitoring of Water Sanitation in a Rural Puerto Rico Watershed

QAPP title: Assessment of Water Quality and Efficacy of Water Treatment Infrastructure in Southwestern Puerto Rico

Cooperative Agreement no. 83553801 between US Environmental Protection Agency and the University of Puerto Rico Mayagüez Campus
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This report summarizes the activities carried out under the Project Agreement for the specified dates. The report provides a general overview of what has been completed to date with potential implications of the results obtained. The overall objective of the project is to "Increase public awareness of sanitation issues in the Lajas Valley and Guánica/Río Loco watersheds by initiating assessments of water quality and sewage infrastructure by professionals and citizen volunteers". Specific objectives are:

1. Carry out water quality sampling for indicators of unsanitary conditions in selected watersheds
2. Use GIS tools to identify point and non-point sources of contaminants
3. Use monitoring results in combination with GIS to link contaminants to specific sources
4. Improve public awareness of the threats of contamination and provide potential solutions to public health and environmental problems

1. Training of professionals and citizen volunteers

All of the professional project participants have been trained in field and laboratory procedures, equipment maintenance and laboratory analysis. Training was done by reading the project QAPP, trainings (two-day workshop on June 19 and 2014), formal and informal meetings and interactions. Group-A citizen volunteers have been trained in field sampling procedures and have received a certificate as evidence (Figure 1). The project QAPP, including SOPs are available to all participants at http://www.uprm.edu/waterquality.

All of the graduate students that are participating are also developing their MSc thesis within the overall project scope. The following persons are active participants in the project:

Person	Position	Role
David Sotomayor-Ramírez	Professor UPRM / Project Manager and PI	Direct and coordinate project activities: field sampling, scheduling, laboratory analysis, data management, statistical analysis, report/ publication preparation, administrative management.
Gustavo Martínez	Research-Professor, UPRM / QA Officer	Lead the laboratory analysis for nutrients, study design, training of volunteers and mentors, report/ publication preparation.
Luis Pérez-Alegría	Professor, UPRM / QA Coordinator	Lead in selection of sampling sites, spatial analysis and GIS, process spatial data, study design, training of volunteers and mentors, report/ publication preparation.
Dave Bachoon	Professor, Georgia College and State Univ	Lead human and bovine enterorocci quantification
Hector Torres	UPRM-AES Research	Provide support in field sampling, field data gathering,

Person	Position	Role
	Technician	equipment maintenance, laboratory support for microbial sample preparation, data entry.
Paloma Rodríguez	UPRM Graduate Student	Lead all field sampling, field data gathering, equipment maintenance, laboratory analysis of selected constituents
Cristina López	UPRM Graduate Student	Provide support in field sampling, field data gathering, equipment maintenance, co-lead all outreach activities
Armando Román	UPRM Graduate Student	Lead microbial analysis of water samples, data entry.
Anibal Ruiz	UPRM-Agricultural Extension Service, Lajas Area Agent	Coordinate sampling excursions with Group-A citizen volunteers (Group #4)
Isbeth Irizarry	UPRM-Agricultural Extension Service, Guánica Area Agent	Coordinate sampling excursions with Group-A citizen volunteers (Groups #1, 2, 3)

2. Outreach

2.1. Meetings with citizen volunteers

All of the Group-A citizen volunteer participants are members of the Agricultural Extension Service (Cooperative Extension Service) 4-H Club. From August 2014 to July 2015, 13 volunteers participated, ranging in age from 16 to 18 years. The volunteers were in grades 10 to 12 (sophomores, juniors and seniors) and attend two High Schools, Escuela Superior Leonides Morales in Lajas and Áurea E. Quiles in Guánica. As of june 2014, nine students graduated from High School, and many of these are now attending UPR-Mayagüez.

The Group-A volunteers were distributed among four groups of between three and four students each, with Isbeth Irizarry leading three groups and Agro. Anibal Ruiz leading one group. Other collaborators were Agro. Jose A. Torre (Agronomist UPRM-AES), and Benerizael Anzueta from the Puerto Rico Department of Education.

Each group was assigned two stations which they sampled on pre-established dates, on their own but with the supervision of UPRM personnel. Each station was sampled five times by the assigned group.

A two-day training was conducted for citizen volunteers and extension agents at the UPR Agricultural Experiment Station in Lajas on 19 and 20 June 2014. Presentations were given by project personnel and supporters on the following topics:

- Watershed management
- Water quality and its implications on health
- Sources of water contamination

- Lajas Valley and Rio Loco watersheds
- Sampling techniques and monitoring stations

Diagnostic tests were given prior to workshop commencement and at the end to evaluate volunteer learning and training efficacy. The pre-workshop mean score was 5.8 and the post-workshop mean score was 11.6, out of 16 maximum points, which suggests that the students were able to improve their knowledge base in the selected topics. Each volunteer that completed the theoretical and experimental parts was certified with a diploma (Please see appendix for an example).

A Powerpoint© and a scientific poster presentation was given to citizen volunteers at UPR Isla Magueyes Field Station on 21 November 2014. The presentation summarized results of the first two sampling rounds. A discussion was carried out regarding the results obtained and their significance.

The Group-A citizen volunteers will be convened in December 2015. A presentation that summarizes the results and provides improved explanation of the significance of these will be given. The information and discussion should contribute towards improving public awareness of the threats of contamination and provide potential solutions to public health and environmental problems in Lajas Valley.

2.2. Presentations

- A poster was presented and an oral presentation made at the US EPA Citizen Science meeting in UPR Río Piedras Campus on 12 September 2014.
- A poster was presented at the annual Sociedad Puertorriqueña de las Ciencias Agrícolas (SOPCA) conference on 7 November 2014.
- A one-day workshop was given to 14 CROEM high school students (sophomores and juniors), under the supervision of Dr. Carmen Santana on 27 May 2015. These students participated in a series of workshops in different topics during a three-week period. A practical component of the workshop was carried out in the field at station #4 (in the Lajas Valley), where students were trained in water sampling techniques. Brief presentations were given at UPRM facilities by project personnel on the following topics:
  - Water quality and its implications on health
  - Sources of water contamination
  - Lajas Valley and Rio Loco watersheds
  - Sampling techniques and monitoring stations
  - Preliminary results

2.3. Sampling dates and participants

Five grab-sampling rounds were completed with Group-A citizen volunteers that are part of UPRM-AES 4-H students.

- The first round was carried out on 5 and 6 Aug 2014; 12 students and three extension agents assisted
- The second round was carried out on 29 September and 1 October 1 2014; 8 students and two extension agents assisted
- The third round was carried out on 12 and 19 January 2015; 11 students and 3 extension agents assisted.
- The fourth round done on 6, 16, and 20 April 2015; 9 students and two extension agents assisted.
- The fifth round was done on 2 and 3 June 2015; 6 students and two extension agents assisted.

2.4. Web page, twitter account, blog

- A web page was constructed for the project to provide educational material, project results and other information to citizens. The address is http://www.uprm.edu/waterquality.
- A twitter account was also created to stimulate interaction and communication among citizen volunteers and project personnel. The address is @waterqualitypr.

3. Selection of sampling sites and land-use analysis

3.1. Selection of sampling sites.

Twenty-six sampling stations were selected from throughout the Lajas Valley, lower portion of Río Loco watersheds, and Guánica Bay (Figure 2). The stations were selected based on criteria described in QAPP. The original goal (as stated in the QAPP) was to have 22 stations that would be sampled during low-flow events. After the QAPP was approved, we identified some geographic points that we thought would provide additional information, based on potential contaminant fecal loading (Table 1).

3.2. Land-use analysis.

Georeferenced land-use data from the Puerto Rico Governmental Portal for Geographic data (gis.pr.gov)[1] was obtained and clipped in ArcGIS 10 to the Lajas Valley and Rio Loco watersheds. Six ground-truthing excursions were made to validate this land use from the field using a blank map and coloring observed land use categories. Land use corresponded to one of the ten categories: Artificial barren, Forest, Grasslands and pastures, Grazed pastures, Hay, High-density urban development, Low-density urban development, Pond, Row crops, Woods and shrubs. Land use classification of validated areas were edited or confirmed in the attribute table of the shapefile (Table 3). The land-use maps represent the most recent and detailed land-use description to date of the area (Figure 3 and Figure 4).

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[1] Portal datos gubernamentales. Available at: http://www2.pr.gov/agencias/gis/Pages/default.aspx. Verified, 08/19/15.

4. Number of samplings completed and sites sampled

4.1. Low-flow (grab) samplings.

Five grab sampling incursions have been completed (Table 4). The first incursion (I) was a 29-day period from 5 August to 3 September 2014; the second incursion (II) was a 32-day period from 22 September to 22 October 2014; the third incursion (III) was a 47-day period from 9 January to 24 February 2015; the fourth incursion (IV) was a 19-day period from 6 April to 24 April 2015; the fifth incursion (V) was a 10-day period from 27 May to June 5 (Table 1). In some instances, there was a large time span during the sampling cycle. Some sites had to be re-visited more than once because the grab sampling criteria to have flowing water was not met. The goal was to have 110 samples (22 stations x 5 incursions each). Based on the 26 stations selected, the goal would have been to sample 130 times (26 stations x 5 incursions). But, two stations (#6 and #7) will only have flowing water during extreme storm events, thus these stations will be discontinued. Twelve sampling events distributed among 7 stations will have to be completed to achieve the 90% sampling completion rate (24 stations x 5 incursions = 120 samplings).

Twelve stations have been selected that we think should be examined more closely in terms of the nutrients and FIB dynamics and their linkage to land-use. These stations will be sampled six additional times (for an additional 72 sampling events). An amendment to the approved QAPP will be submitted shortly.

4.2. Storm-event (static bottles) samplings

Three storm-sampling incursions have been accomplished (Table 5). The first incursion (I) occurred on 28 May 2015; the second incursion (II) occurred on 31 May 2015; the third incursion occurred on 7 August 2015. The goal was to obtain 30 storm samples (6 stations x 5 incursions). We have managed to complete 13 out of 30 for a 43% completion rate.

5. Analysis completed

A description of the number of grab and storm samples analyzed, in progress to be analyzed, or not sampled during each cycle, for each parameter is summarized in Tables 4 and 5. Based on 24 stations, the goal is to have 120 data points for nutrients and enterococci. Excluding blanks and duplicates, we have managed to analyze 105 samples for nutrients and enterococci for a 88% completeness. Results for Bacteroides human marker, heavy metals and optical brighteners were expected of 56 samples. Our current completeness is 38% for heavy metals, 101% for human bacteroidales, and 188% for OBs.

Hydrologic surveying measurements were completed for 6 channels used for passive rising storm-flow stream collectors. The data will be used to relate stage height to maximum flow for each channel. This work is in progress.

Daily precipitation data at Lajas UPR-Agricultural Experiment Station has been gathered from: http://www.nws.noaa.gov/climate/xmacis.php?wfo=sju. Daily precipitation data at Arenas well in Guánica has been gathered from: http://waterdata.usgs.gov/nwis/inventory/?site_no=180122066560300&agency_cd=USGS. Precipitation data collected by selected farmers in some areas has been gathered.

6. Major highlights of the results

6.1. Precipitation

Historical mean annual precipitation for Lajas Agricultural Experiment Station is 48 inches and for Santa Rita, Guánica is 34 inches. Historical monthly precipitation peaks occur in September, October and November where 40 to 44% of the total annual precipitation occurs in these three months. A second precipitation peak occurs in April to May where 18% of the total annual precipitation occurs. Mean annual precipitation in 2014 was similar to the long term average (deficit of 1.36 inches). The difference occurred in the precipitation pattern, as in August 2014, cumulative precipitation was 2.4 times greater than the historical mean value. All of the other months, except March, November and December were below the historical normal. In 2015 (up to 15 August), precipitation was 7.1 inches below historical normal of 26.6 inches. Precipitation during the months of January to March was near or above normal, all of the remaining months were in deficit.

6.2. Land use

All of the stations are linked to a defined sub-basin. Most sub-basins are within greater basins so that within a particular basin two or more stations are present. Land use analysis has not yet been finalized so that a complete analysis linking nutrients, fecal contamination and land-use cannot be done, yet certain generalizations can be made (Please refer to Table 3 and Figure 4):

Stations #13, #1, #3, and #4 within Mondongo basin. Station #13 is upstream the Lajas WWTP outfall but within-stream in the urban community of Lajas City. Station #1 is downstream of the Lajas WWTP outfall, so that we expect high nutrients but possibly not high fecal contaminant loads due to WWTP chlorination. The NPDES permit allows for nutrient concentrations in effluent less than 1 mg total P/L, 6 mg NH_4^+-N/L, and 10 mg NO_3-N/L, but expected concentrations of these nutrients downstream the WWTP is not provided. Stations #3 and #4 are within the same waterway that drains Station #1, but are downstream of Lajas City. Thus we expect to see a strong nutrient and fecal contamination signal due to urban influence. Station #4 is downstream an un-sewered community bordering Rt. #116.

Station #2 is located within Lajas Valley Irrigation Channel, near Lajas City. This station is considered a reference station in the sense that the channel transports water from Lago Loco and should receive minimal runoff influence. Lago Loco is in the mesotrophic trophic state index with historical total P concentration of 36 µg/L (Martínez et al., 2005).

Stations #24, #5 and #7 are downstream of dairy production facilities. Station #6 is upstream of a dairy production facility. The basins for stations #24 and #5 have not yet been defined. Station #6 and #7 will be sampled primarily during storm events because the grab sampling criteria to have flowing water has not been met, but as stated previously these stations will probably be discontinued.

Station #23 drains the lower Río Loco watershed, which has relatively limited agricultural production area, yet has a mix of urban, suburban and rural land-uses. Station #22 is at the outlet of the Lajas Valley drainage channel and integrates all of the land-use activities within the Lajas Valley Watershed.

Stations #16, #17 and #18 are in tandem (within the same drainage channel) within Cristales basin; Station #18 includes waters from #16 and #17 and drains a 600-acre rice-production farm.

Station #21 is located in the drainage canal downstream from the #12 station. The subbasin for station #12 has not been defined and is located in a creek that drains an agricultural site with row crops. The overall basin for these stations has not yet been defined. Stations #25 drains Cuesta Blanca Community and its outlet is upstream of station #26 within the Lajas Valley drainage channel.

Station #10 is located in an urban area and drains into station #11. Station #11 is located within a hay farm and is also downstream from a banana plantation. Stations #10 and # 11 are within Bárbara basin.

Station #8 drains a mixed land use area and #9 receives water from a aquaculture industry. Both connect and form a single stream that drains into the drainage canal.

Both stations #14 and #15 are part of the Maginas creek that is located in Sabana Grande and Guánica and are within Maginas basin. Station #14 drains a Sabana Grande urban area and #15 drains a beef cattle production area.

6.3. Nutrients

In contrast to what has been observed in the continental USA (USEPA, 2006), nutrients have yet to be identified as a major cause of surface water impairment in Puerto Rico (PREQB, 2003, PREQB, 2008). This is possibly due to the lack of adequate standards which have prevented the identification of nutrient impaired waters. For example, current nutrient water-quality standards in Puerto Rico are 1,000 µg total P/L and 10 mg NO_3-N/L (for nitrogen) for class SD waters, which includes rivers, lakes, and estuaries (PREQB, 2010). We have opted to interpret our nutrient concentration data in the context of suggested numeric nutrient criteria in rivers of Puerto Rico (Sotomayor-Ramirez et al., 2011) as:

Threshold	Total N	NO_3-N	Total P
	--------------------------------mg/L----------------------		
Non-enriched	<0.35	<0.25	<0.030
Enriched	>0.35-1.70	>0.25-0.97	>0.030-0.160
Impaired	>1.70	>0.97	>0.160

For each station the mean value for the parameters NO3-N, total N (TN), total P (TP) was interpreted in terms of the suggested numeric nutrient criteria for Puerto Rico.

Reference Station #2 (Lajas Valley Irrigation Channel near Lajas City) was "nutrient enriched" in terms of nitrate, TN and TP.

Station #1 (Lajas WWTP outfall) was "nutrient impaired" in terms of nitrate, TN and TP. The highest mean concentration for each species was observed in this station.

Station #13 (upstream the Lajas WWTP) was impaired in terms of TN and TP but enriched for NO_3-N concentrations. TN and TP concentrations were 3 and 5.5 times higher in station #1 than Station #13. The data suggests that the WWTP is an important nutrient (especially P) input to the drainage.

Stations #3 and #4 (downstream Lajas City) had similar concentrations among them; with similar trends as station #1. TN and TP concentrations were 2.3 and 1.2 times higher in Station #1 than in stations #2 and #4.

Stations #24, #5, #21 and #22 are downstream of Station #4. There is a dairy facility before station #24 and a second one between #24 and #5. Station #25 is downstream station #5 and is also hydrologically connected to a dairy facility near stations #6 and #7, thus receiving inputs that pass through all these stations. Similar trends were observed for stations #25 and #5 and were impaired for all species. TN, NO_3-N, and TP concentrations decreased in the order of #24 > #5 > #25 > #21 > #22. Thus as the length of the stream channel increased, the quality of the water improved. It cannot be determined if the dairy facilities contribute nutrients to drainage waters in amounts that increase concentrations above those found before the facilities.

Stations #24 and #5 were still impaired in terms of TN, NO_3-N and TP concentrations but the quality of the water improved thereafter. The species with the best improvement was NO_3-N which suggests that as water moved to the lower reaches of the watershed, N was lost via denitrification. Although TP concentrations improved (nearly a two-fold decrease from #24 to #22) through the watershed, total P concentrations at station #22 were 0.165 mg P/L. Total P may be retained within the river bed and released during extreme storm events.

Station #8, which drains from Quebrada La Plata was enriched for all three species. Station #9, which drains from a fish-pond facility, had TN, NO3-N and TP concentrations that were higher to those of #8 and was impaired in terms of TP concentrations. The data suggests that the fish-pond facility was a source of N and P to drainage waters.

Station #10, which drains a suburban community, was impaired in terms of all three species. TN and TP concentrations in Station #11 (downstream from #10) were similar to those in station #10.

Station #15 is downstream of station #14. Station #15 is within-stream a cattle grazing (meat producing) ranch. TN concentrations were lower but TP concentrations were higher downstream from station #14. Thus it appears that the cattle farm may be contributing P (but not N) to the drainage waters.

TN and NO3 concentrations decreased, but TP concentrations were similar (0.2 and 0.3 mg P/L for stations #16 and #18) as the hydrologic distance increased for stations #16, #17, and #18. Based on the data collected it appears that the rice production facility is not an important nutrient contributor to drainage waters.

In Río Loco, station #23 was impaired in terms of TN and NO3-N concentrations but not for TP.

Nutrient concentrations were near background levels at Guánica Bay near Guánica town and concentrations at the Guánica WWTP were higher at the outlet of the WWTP.

6.4. Enterococci, optical brighteners, bovine and human bacteroides markers

Reference station #2 had mean enterococci concentrations of 321 MPN/100 mL. Enterococci concentrations were highest in basins where urban activities predominate within and downstream of the town of Lajas, which includes stations #13, #4, #3, and #1. Station #13 had highest enterococci concentrations and the signal decreased downstream (stations #1 and #3) yet increased again in #4. Among these stations, station #1 (Lajas WWTP outfall) did not have the expected "high" Enterococci concentrations, possibly due to chlorination (measurements of chlorine concentrations in the samples is pending). Stations #1 and #3 had similar Enterococci concentrations during most sampling rounds. The OB signal was only detected in station #3 and station #4 had a probable OB signal (signal was detected once in five samplings).

In this urban basin, human bacteroides marker was detected in stations #1 and #4. Station #4 also had bovine bacteroides marker. Since station #4 was downstream an un-sewered community, ruminants observed near the households could also be contributing to the degradation of drainage waters via fecal contamination.

As occurred in station #4 the presence of human and bovine bacteroides was also detected in station #24, albeit lower enterococci concentrations. Station #5 (which was downstream a dairy facility) had enterococci concentrations < 100 MPN/100 mL, but had a positive OB signal. Enterococci concentrations increased further downstream in stations #25 and #21. Station #22 has a lower mean concentration of bacteria than those found in the preceding stations.

Mean Enterococci concentrations at the Lajas Valley drainage outlet (station #22) were <100 MPN/100 mL. Bacterial inactivation by solar radiation and adhesion to bottom sediments can reduce Enterococci counts. Based on the data collected we hypothesize that enterococci concentrations are reduced with increasing channel length by sorption to sediments and/or solar radiation. In contrast, station #23 which drains the Río Loco watershed had four times enterococci concentrations found at the Lajas Valley watershed outlet.

In general, Enterococci concentrations were > 400 MPN/100 mL in stations throughout the watershed and most had negative OB signal and did not have the human or bovine bacteroides marker In contrast, station #11 had high enterococci concentrations, had a positive OB signal and had the presence of bovine marker. This station is characterized as having potential waste-water discharges from animal feeding operations.

In summary, human bacteroides marker were found in station #1, #4, and #24 all within the western portion of the watershed most influenced by urban activities. Stations #1 and #4 are located within or at the border of Lajas municipality, and station #24 is downstream of a dairy production facility; and downstream of an urbanization where sewage overflow has been observed (to the south). Bovine bacteroides markers were found in stations #4, and #24 and downstream Bárbara Basin (station #24).

Simple Pearson correlation analysis showed that total N and total P were significantly correlated (r = 0.46) as were total N and NO3-N (r = 0.62).

7. Comparisons among basins

Comparisons among basins (Mondongo, Bárbara, Maginas, and Cristales) for the parameters total N, total P, NO_3-N, and enterococci were made using ANOVA in a repeated measures design with stations within basins as treatments, and sampling dates as the repeated measures (Table 6). Normality and homogeneity of variance assumptions were achieved by square-root transformation of the data. Enterococci concentrations were in the order of Bárbara ≥ Mondongo ≥ Maginas ≥ Cristales. Total N concentrations were in the order of Mondongo ≥ Maginas ≥ Bárbara > Cristales. Concentrations of NO_3-N did not differ among basins with means ranging from 0.72 to 1.3 mg NO_3-N/L and an overall mean of 1.08 mg NO_3-N/L. Total P was significantly higher for Mondongo than Cristales, Bárbara and Maginas combined, and the latter three did not differ among them.

The total combined area of the four basins was 6,335 ha (15,546 acres). The predominant land use was computed based on the proportional extent of Urban (sum of high and low density), Artificial barrens, Forest, Grasslands and pastures, Grazed pastures, Hay, Pond, Row crops, and Woods and Shrubs. Of the land uses identified the most important nutrient contributors are Urban, Grazed pastures, and Row crops. The most important enterococci contributors are Urban and Grazed pastures.

Maginas and Cristales basins had a relatively high proportion of Urban land (15 and 14.1%, respectively) yet these were of lower density than in Mondongo. Also, Mondongo had the WWTP which receives raw sewage for processing from adjacent watersheds and areas. Thus, Mondongo is the basin with the highest urban influence which is expected to contribute nutrients and enterococci. Mondongo did contribute with the highest nutrient (total N and total P) concentrations. Bárbara did not have a relatively high proportion of Urban, Grazed pastures and Row crops, yet contributes relatively high concentrations of enterococci. This probably occurs due to a particular point source of hog production facility, upstream station #11.

8. Conclusions

It appears that the Lajas WWTP is an important nutrient (especially P) input to the drainage. Based on the data collected to date, it cannot be determined if the dairy facilities contribute nutrients to drainage waters in amounts that increase concentrations above those found before the facilities. The fish-pond facility appears to be a source of N and P to drainage waters. The rice production facility is not an important nutrient contributor to drainage waters. Mean Enterococci concentrations at the Lajas Valley drainage outlet (station #22) were <100 MPN/100 mL, and were low possibly due to inactivation by solar radiation and adhesion to bottom sediments. In contrast, station #23 which drains the Río Loco watershed had four times enterococci concentrations found at the Lajas Valley watershed outlet.

Human bacteroides marker was found in the western portion of the watershed most influenced by urban activities. Mondongo is the basin with the highest urban influence and apparently is the most important contributor of nutrients (total N and total P) to drainage waters, and Bárbara which had particular point source was possibly an important enterococci contributor. Further data collection and analysis will lead to results that will identify with improved certainty nutrient and fecal contribution to Lajas Valley drainage waters.

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