Referential seasonality of critical loads of sulfur on *Rhizophora harrisonii* in the port of Guayaquil

QUEVEDO O. [1,A)], CERÓN J.[2,B)], CERÓN R.[3,C)], CALDERÓN M.[4,D)],JARAMILLO B.[5,E)], INZHIVOTKINA Y[6F)]., REVELO W.[7,G)]

[1,5,6]Universidad de Guayaquil, Facultad de Comunicación Social, Ciudadela Salvador Allende, Av. Delta y Av. Kennedy, Guayaquil, ECUADOR.
[2, 3]Universidad Autónoma del Carmen Facultad de Química, Calle 56 num. 4 esquina Ave. Concordia Colonia Benito Juárez, C.P. 24180 Ciudad del Carmen, Campeche, MÉXICO.
[4]Escuela Superior Politécnica del Litoral, ESPOL, Facultad de Ciencias de la Vida, Campus Gustavo Galindo Km 30.5 Vía Perimetral, P.O. Box 09-01-5863, Guayaquil, ECUADOR.
[7]Instituto Nacional de Pesca, Letamendi 210 y La Ría, Guayaquil – ECUADOR.

[a)]olga.quevedop@ug.edu.ec; [b)]jceronbreton@gmail.com; [c)]rceron@gmail.com; [d)]mafercal@espol.edu.ec, [e)]bladimir.jaramilloe@ug.edu.ec, [f)]yana.inshivotkina@ug.edu.ec, [g)]wrevelo@institutopesca.gob.ec

Abstract: - The research focuses on demonstration of the life quality of mangroves with the respect to the critical loads of atmospheric sulfur during the dry and rainy seasons of 2017, the investigation was conducted on two islands of the interior estuary in Gulf of Guayaquil: "Trinitaria" with a high incidence of pollutant activity and "Chupador Chico" island, which is part of the Wetland of International Importance. The analysis of the data included the use of a simple one-way Anova and the Kruskal-Wallis tests for the contrast of the hypotheses, with a p-value of 0.05. The proteins and chlorophylls were determined with the Bradfor and 80% acetone methods, as well as the salts of Mg^{2+}, Mn^{2+}, K^{+}, Ca^{2+} and SO_4^{2-} in the leaves, it was measured by APHA method 4500 and passive samplers for the determination of critical loads of sulfur. During the dry and rainy seasons it was observed significant differences in proteins 9.85 - 3.546 ug ml^{-1} and carotenoids 31.25 - 0.96 ug ml^{-1} respectively, which differs between islands. The sulfates present in the leaves varied from 3,540 to 2,125 ppm for the dry and rainy season. The sulfur fluxs were of 3.35 kg S ha-1 year -1 there were no significant differences between study sites or times of sulfur fluxs, which responds to a regional distribution. The significant differences of the photosynthetic pigments, proteins and salts between study sites and seasons responds to the different anthropics tensors that *Rhizophora harrisonii* supports in the study area.

Key-Words: - Soluble proteins, *Rhizophora harrisonii*, chlorophyll, critical loads, SO_2.

1 Introduction

The morphological characteristics of the mangroves in the area of influence of Guayaquil port can consist of the elements that serve to determine the quality of the mangroves, in this case the investigation about *Rhizophora harrisonii*, it was based on the observation field, which is present in the leaves and its viscous appearance is difficult to clean with rain, as well as the brittle appearance of leaves and their pigmentation that varies from yellow to brown, this is the most noticeable in the dry season, these aspects have a direct influence on the quality of life of the plant formations [1].

Several activities which pollutes the atmosphere are industries, the shipping activities, the agricultural production, among others; the plant formations are affected not only because of the agricultural and urban frontier, but also because in the bordering urban areas, agricultural or industrial ecosystems generate pollution that directly affects the atmosphere, an evidence of this is the color of the vegetation, which has the diversity of shades from green to yellow due to chlorosis, and to the black color due to the accumulation of soot, which is directly influence on the quality of life of these plant formations [1].

In Ecuador it was registered the concentrations of SO_2 in the air, in this investigation it was tried to demonstrate that the quality of life of *Rhizophora harrisonii* in the Port of Guayaquil is being affected by the shipping activity, it can be seen in the morphological damages of its leaves and functionality of the changes in the concentrations of proteins, photosynthetic pigments, changes in the concentration of salts like K^+, Ca^{2+}, Mg^{2+}. Mn^{2+}, SO_4, and stomata opening, as responses to pollution processes in the area, which was contrasted with a control or target.

2 Problem formulation

This research was developed in two islands of the interior estuary of Guayaquil´s Gulf, Ecuador. "La Trinitaria" island was influenced by the shipping activity of the Guayaquil's port, as well as by other companies such as thermoelectric, steel industry, cement and vehicular traffic factories; the island "Chupador Chico", which is located five miles away from the problem area does not have a strong influence of the port, it is recognized as *wetland of international importance* (Figure 1).

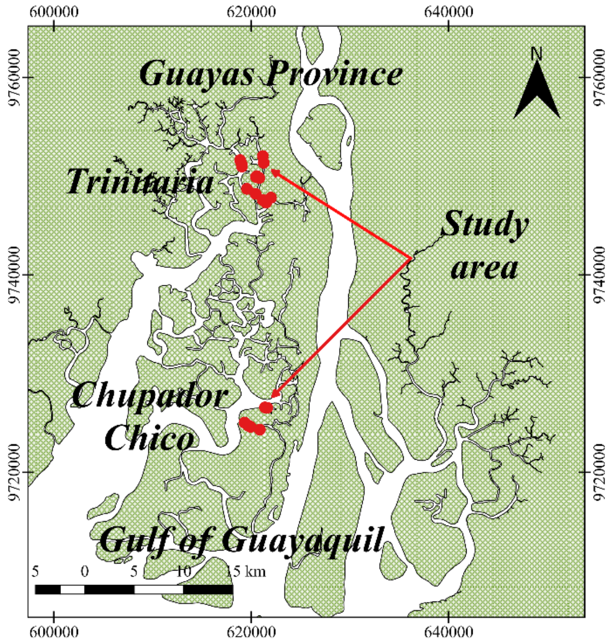

Figure 1. Study area in the interior estuary of the Gulf of Guayaquil for the Trinitaria Island (contaminated area) and Chupador Chico (control area).

The mangrove swamps of Guayaquil's port accept from 10 to 12 ships weekly, they arrived and departed from the port [2]. It is 34% of the total amount of 4801 ships that arrived and departed in the ports of the whole country in 2013. Approximately in Guayaquil's port arrived 802 ships, from them 754 were departed with loads from 300 to 700 containers, from 20 to 40 feet tall [3].

The mangrove ecosystem of the Guayaquil's Gulf is undergoing the process of deterioration [4], it is affected by the shipping traffic, by its liquid and gaseous effluents, which are produced because of the embarkation and disembarkation activities, all of this have an effect on the health of the mangrove ecosystem.

The concentrations of SO_2 can cause affectation of the riparian vegetation, which, in addition is bounded to the diversity of atmospheric and meteorological factors and diminish the probability of the predictions, it must be considered in the implementation of policies[5].

The acids and acidifying gases (SO_2, SO_3, NO_x) can directly affect the aerial cover of the vegetation and cause necrosis in the leaves and reproductive tissues, loss of foliar nutrients, resulting in decrease of plant productivity, loss of fruits among others, but it rarely causes the death of the plant because it is difficult to maintain high concentrations for a long time [6].

Severe visible damage in the form of chlorosis and necrosis was found at lower pH values, and even nutrient values showed changes after exposures in white mangrove (*Laguncularia racemosa*), red mangrove (*Rhizophora harrisonii*), and mangrove botoncillo (*Conocarpus erectus*). Cerón [7] found out that high concentrations of SO_2 causes the loss of chlorophyll, and increases the level of soluble proteins, it means that plants have the capacity to metabolize this compound.

The trade winds are determinants of SO_2 concentration [8] in the sea, so it raises the hypothesis of how they influence on the distribution of SO_2 in cities.

Values of critical loads in Ecuador do not exist, the first investigations concerning to this subject were reported from Oslo protocol for the control of pollutant gases in the atmosphere [9], it fostered the development of studies on critical loads for the Nordic countries in the 1990s, in the Americas this method is currently applied in Mexico.

2.1. Leaf sampling and laboratory analysis

The investigation was developed taking into account three transects in two islands in the inner estuary of the Gulf of Guayaquil, the Trinitaria Island (contaminated area) and one in the Chupador Chico (control area) as in [10]. The first area is located in front of the docks of Guayaquil's port and the second one is located in the interior estuary of the Ramsar "the interior mangrove of Guayaquil's Gulf" [11]. The collections were made during the dry (2016) and rainy (2017) seasons. In total it was collected 360 leaves considering three transects in both the contaminated and control area.

The concentrations of soluble proteins in the samples of the collected leaves were determined by the method of Bradfor [12], it was done using BIO TEK SYNERGY HT-photometer at 595 nm, the photosynthetic pigments of the leaves were determined with the method of 80% acetone using the equation [13]. The damage area of the leaves was evaluated using the Adobe Photoshop extended tool [14]. The concentrations of Na, Mg, Mn, K, Ca, and S, it was determined by Atomic Absorption Spectrometry 3110 and Sulfur- by Turbudimetric Method 4500 - SO_4 [15], the stomatal opening was measured with MOTIC AE 31 microscope, the images were captured with the MOTICAN N480 camera and the Motic Image Plus S series software 653711, the stomatal index was calculated [16]. The results were processed using the software Statgraphics version 16.1.15 [17]. It was applied the Kruskal-Wallis test in order to check the null hypothesis, it was used a non-parametric method for testing to know if samples come from the same distribution and does not assume a normal distribution of the residuals.

In addition, in order to determine the percentage of damage of the leaves, the samples were photographed with a Nikon D90 digital camera (resolution of 300 dpi), in our case digital camera was used as the indirect method of measuring the leaf area [18–20]. Then, for data analysis and estimation of the leaf damage the photos were processed with Adobe Photoshop CS6 software [14,21], in this way it incorporates new tools which are necessary for the analysis of the data that can easily be exported to the Excel software. Firstly it was determined the leaf area and then the damaged area. The sum of the pixels (px) of the selected area through the magic wand tool represented the total area, and the scale of work was defined as 10 px = 1cm.

2.2. Critical loads of sulfur sampling and laboratory analysis.

The level of sulfate in atmospheric deposition was determined by the area of study. It was created 19 passive samplers were collected according to the methodology developed by [7,22–24]. The collectors consisted of a funnel, connected to a column containing ion exchange resin (IER). Deposition falls on the funnel surface and washed down into the column. The main advantage of this passive sampler is that it can be used for long exposure periods (e.g. months) and the equipment has a low price. It let to increase the number of the sampling points in a given area, therefore, with this type of collector it is feasible to increase their numbers in order to characterize spatial patterns in deposition at high resolution [25]. Sulfate ions can be exchanged by IER anion and cation, and they can be locked by functional groups with opposite electric charges. It was chosen the mixed resin column design, considering that it is an efficient technique to capture ions from samples, because the flux is direct through the ionic exchange resin column (IER). Mixed bed refers to the fact that the resin contains both anion and cation exchange resin beds.

Passive sampling devices were exposed during four months with 76 sub-periods in one months. At the end of each sub-period, resin tubes were changed by tubes with fresh resin.

The IER columns consisted of a funnel attached to PVC tubes, the solution was channeled to the mixed resin bed through the column, where ions were retained. The resin used for the IER collectors was a mixed polystyrene bed for anions and cations

(AmberliteTM IRN 150). The funnel was covered with a fine mesh screen to avoid the intrusion of material such as leaves and insects. The IER column was inserted into the inner PVC tube (30 gr. of IER), the inner tube was sealed with a glass fiber at the bottom (as a support for platform) and at the top (as a filter). The inner PVC tube was contained an outer PVC tube in order to protect it from solar radiation. The bottom end of the IER column was closed using standard PVC valves to allow drainage. Finally, IER columns were assured at ground level in open areas in each of the sampling sites. Resin columns were replaced with fresh columns each months. These exposure periods yielded two seasonal data sets: for the dry season and for the rainy season.

Resin columns contained two sequential extractions with 100 ml of 2N KCl. The extracts were analyzed for SO_4^{2-} Three IER columns were used as blanks, they were extracted and analyzed; the average blank in IER column values for sulfate, it was calculated and subtracted from ions recovered from each loaded resin column. Extracted samples were analyzed by turbidimetry [26]. Solute-mass was divided by funnel surface-area and total exposure period to obtain deposition rates S (as SO_4^{2-}) in kg ha^{-1} yr^{-1}.

2.3. Wind speed analysis

The wind speed analysis was carried out using the data of the "Simón Bolívar Internacional" meteorological weather station in Guayaquil which is located at an altitude of 5.8 m a.s.l., latitude 02°09’00’ S and longitude 79°53’00’’O. For the analysis of the seasonality it was considered the hourly data of the winds speed in November and December 2016 for the dry season; the same was done for January and February 2017, for the rainy season. The data were processed using the WRPLOT View software [27], the generation of the wind rose was done to see the relative distribution of the wind direction and to determine the frequencies of the winds according to ranges of wind speed.

3 Problem solution

3.1. Functional aspects: Photosynthetic pigments and proteins

The results found out in both sites reflecting the leaf damage percent, stomatal opening, concentration of salts, photosynthetic pigments, proteins, and sulfur in leaves of *Rhizophora harrisonii* are shown in Table 1. The function of protection of the protein-pigment complex and the chloroplasts against oxidation is carried out using carotenoids in general [28]. The results about concerning to low concentrations for carotenoids during the dry and rainy season are the similar, but it is observed that the control is significantly different concerning to the rest of the transects.

Table 1. Differences in pigment values and sales in the functional and morphological aspects of *Rhizophora harrisonii*.

	Trinitaria Island		Chupador Chico Island	
	Rainy	Dry	Rainy	Dry
Photosynthetics pigments				
Proteins (µg ml^{-1})	89.27	54.42	54.87	47.88
Chlorophyll a (µg ml^{-1})	7.58	10.01	6.42	7.04
Chlorophyll b (µg ml^{-1})	5.51	7.05	3.06	4.57
Carotenoids (µg ml^{-1})	6.93	5.87	5.97	2.79
Ions				
SO_4^{2-} (ppm)	1.52	1.01	0.08	0.66
Ca^{2+} (ppm)	0.51	0.63	0.20	0.70
Mg^{2+} (ppm)	0.26	0.26	0.16	0.35
k^+ (ppm)	0.29	0.44	0.23	0.39
Mn^{2+} (ppm)	11.40	4.23	0.03	0.03
Leaves damage				
Stomatal density	1.41	1.65	0.74	1.50
Insects/necrosis/chlorosis	1.75	16.38	98.15	59.45
Stomatal opening (µm)	2.14E-10	3.90E+02	9.53E+14	4.41E+02

Concerning to the proteins, the increase of proteins after they have been exposed to 100, 150 and 200 ppm of SO_2 [28]. In the case of proteins of *Rhizophora harrisonii*, its concentrations were significantly presented in the rainy season, and in the control phase it was found out that the concentrations are similar to those of the dry season, it can happened because the acidification processes was generated during the rainy season and it could motivate the production of proteins as a defense mechanism (Figure 2).

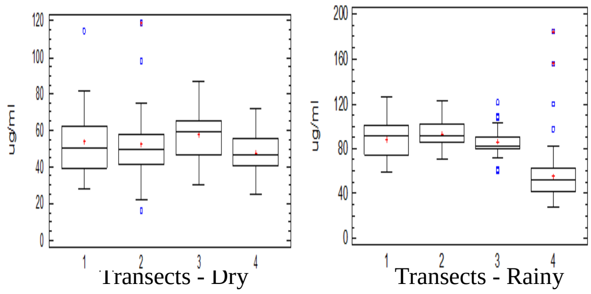

Figure 2. Protein concentrations in ug ml^{-1} for the dry and rainy seasons. Transects 1 to 3 (contaminated area) and transect 4 (control area).

Data obtained by [29], it was discovered that SO_2 action by plant fumigation lower the level of pH, it is known that this effect causes the interruption of chlorophyll and it is caused by acidification. The level of chlorophyll is increased during the dry season until 3.52 ug ml^{-1} and decreased in the rainy season until 1.04 ug ml^{-1} (Figure 3a, 3b).

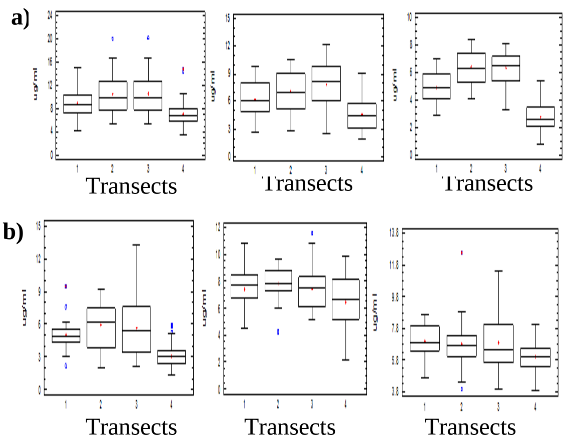

Figure 3. Chlorophylls (ug ml^{-1}) in leaves of *Rhizophora harrisonii* during the a) dry and b) rainy seasons. Transects 1 to 3 (contaminated area) and transect 4 (control area).

3.2. Salts: Potassium, Magnesium, Manganese, Calcium and Sulfates

The Potassium content K^+ in the leaves decreased significantly for all the acidity treatments, the K^+ has a tendency to decrease as the pH decreases [7]. The exchange of H and K^+ ions occurs during the time of precipitation [30,31]. Concerning to the case of *Rhizophora harrisonii* the K^+ reflects significant differences comparing with their lower values in the rainy season (Figure 4a).

Parker [32] showed that the level of Sodium and Magnesium Mg^{2+} commonly occur in low foliar concentrations as a result of less leaching of K^+. The status of the nutrients in the forests may reflect the magnitude of Ca^{2+} leached [30], the Ca^{2+} in the leaves of *Rhizophora harrisonii* was presented in the lower concentrations in the rainy season in compare with the dry season, it is considered that these differences are marked by the absence of epiphytism in the mangrove canopy. The presence of calcium is 1.2 ppm in relation to transects 1, 2 and 3.

The concentrations of sulfates are minimal from 1.5 till 3.5 ppm during the dry and rainy season, in transects it´s 3 and 1 respectively. (Figure 4b).

Magnesium is a necessary element for chlorophyll production of green plants, so its reduction produces yellowing of the leaves and prevents their photosynthetic action [33]. Figure 4c, shows that Mg^{2+} concentrations are lower during the dry season in comparison to the rainy one, this is related to the foliar damage that acts in the formation of photosynthetic pigments. Therefore, the increase of photosynthetic pigments (Chlorophyll a: 3.52 ug ml^{-1}, Chlorophyll b 5.55 ug ml^{-1} and Carotenoids 3.57 ug ml^{-1}), and the persistent yellowing of *Rhizophora harrisonii* trees in transect 3 can be attributed to the shipping activity that takes place here in the port [7].

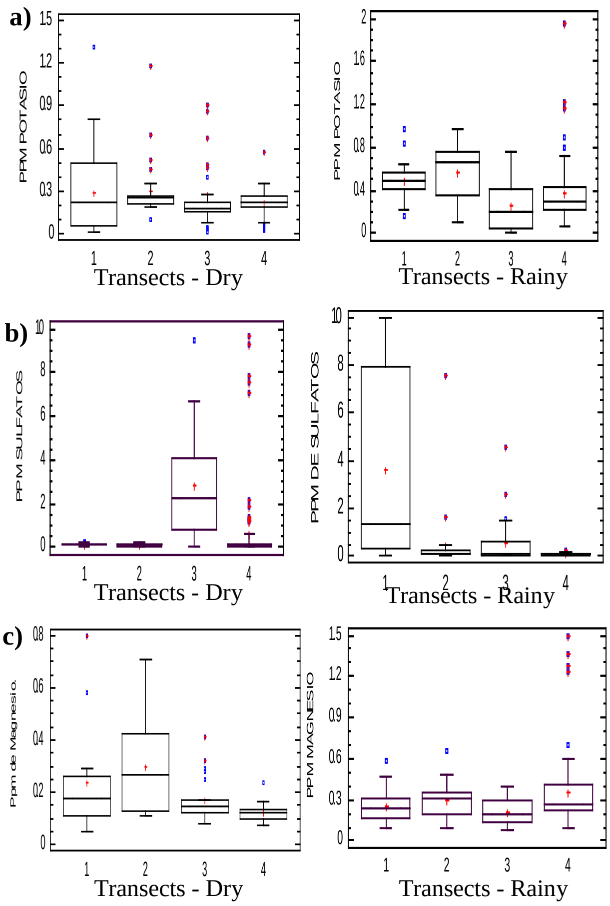

Figure 4. Concentrations of Potassium a), Sulphates b) and Magnesio c), in the dry and rainy season in leaves of *Rhizophora harrisonii*. Transects 1 to 3 (contaminated area) and transect 4 (control area).

The average concentrations according to this research vary between transects, transect 4 has the most conservative values in relation with others. It also has higher values in three transects and according to control during the rainy season, these values are lower in the dry season, and their concentrations are less than one, it means that there is a relationship between the dry deposition of SO_2 that influences on the low concentrations of Mg^{2+} during the dry season.

The lack of Mg^{2+} is related to a yellow color in old leaves and in young ones it happens due to a low level of chloroplasts in necrosis in the extremities and margins of the leaves [34]. The Mg^{2+} is used by the plants inside the foliage cells and it is linked to chlorophyll a decrease of this element will be visualized in a yellow color of chlorosis leaves or chlorotic stippling.

3.3. Morphological aspects: Stomas

The concentrations of potassium during the rainy season contribute to the stomatal opening [35], it means that the absorption of potassium ions by the guard cells happen in the vacuoles when they are open and when the concentration of potassium is enough to contribute to their opening, it´s 0.5 mole in the concentration of K^+. "the transport of K^+ from the subsidiary cells to the guard cells causes more negative osmotic potentials and consequently leads to the opening of stomata, a reverse transport of the K^+ ion causes and the stomatal closure.

3.4. Foliar damage

The results show that, during the dry season transect 3 there was the highest percentage of the leaf damage, 39%, while in the transect 4 the damage was 5%; it includes such factors as necrosis, chlorosis and browse caused by *Tyrinteina arnobia Stoll* species of the family Geometridae (Figure 5), they cause from 4 and 15% of the damage for the transects 1 and 2 respectively.

The presence of *Tyrinteina arnobia Stoll* was recorded only for the dry season in transects 1, 2 and 3, however in transect 4 these larvae were not found [36]. During the rainy season the larvae were totally absent in all the transects. On the other hand, necrosis observed in the edges of the leaves could be linked with the decrease in K^+ and Ca^{2+} [34,37].

The foliar damage presented in the leaves of *Rhizophora harrisonii* is a consequence of contamination in the area of Guayaqauils´ sea port , the evidence can be seen in transect 3, where, the leaves of the mangroves on the riverbank have a yellowish coloration (Figure 5). During the rainy season the presented leaves have a similar percentage of damage for all transects, it happenes because the rain cleans the leaves and prevents the larvae from developing their metamorphosis stages.

The SO_2 is a significant pollutant in the air, it generates visible and non-visible damage to the plants [29]. The degree of visible damage, chlorosis and necrosis, varies between species of the plant, it happends even if SO_2 occurs in conditions of low concentration [38].

Figure 5. Leaf damage encountered in *Rhizophora harrisonii.*

3.5. Critical loads of sulfur

The model of the regional deposition on a global scale was determined by [39], it was described 17 places in South America based on wet deposition where there is a critical level of sulfates SO_4^{2-} and other elements with heterogeneity in their data, it was described by [40–43], the heterogeneity occurs because of the different geographical characteristics in each places.

Critical loads have been estimated in several regions of the world due to environmental problems and the damage of human health. Alpine ecosystems are more sensitive than lowland systems, they have a critical load value. Pérez-Suárez *et al.* [44] reported about Zoquiapan the area in the east of México, it inputs from 5.5 to 8.8 S kg ha^{-1} $year^{-1}$. Ponette-González *et al*. [45] established that in the Veracruz Center in several areas the level of sulfur varies between 8 to 17 kg ha^{-1} $year^{-1}$. Cerón *et al.* [7] determined that the average deposit of sulfur flux (such as SO_4^{2-}) in Carmen Island was from 2.15 to 4.7 kg ha^{-1} $year^{-1}$. The average values of sulfur are 3.357 S kg ha^{-1} $year^{-1}$ it is deposited in the mangrove surpass according to [28], it is analogous to similar ecosystems, because of their ecosystemic characteristics at the level of plant formations.

Eyring *et al.* [9] attribute that the highest emissions of gases come from the open seas and they are significant in the transportation sector, in addition they cause acidification and eutrophication of ecosystems in fresh water [46,47]. It was found out, that the key compounds such as sulfur dioxide which mainly contains and as a result is ship emissions.

Figure 6 shows the sulfur deposit flux in the dry and in the rainy seasons, it depends upon type of land (1: urban, 2: urban mangrove, 3: shipping ports and 4:conserved mangrove). It can be observed that the sulfur deposited shows higher flux average during the dry season to compare with the rainy season, because humidity and rain remove and partially dilute the pollutants present in the atmosphere [48]. However the sulphate is soluble in the aqueous phase, it has washing or dissolving effect by cloud water and raindrops which could lead to lower concentrations of this ion during the rainy season.

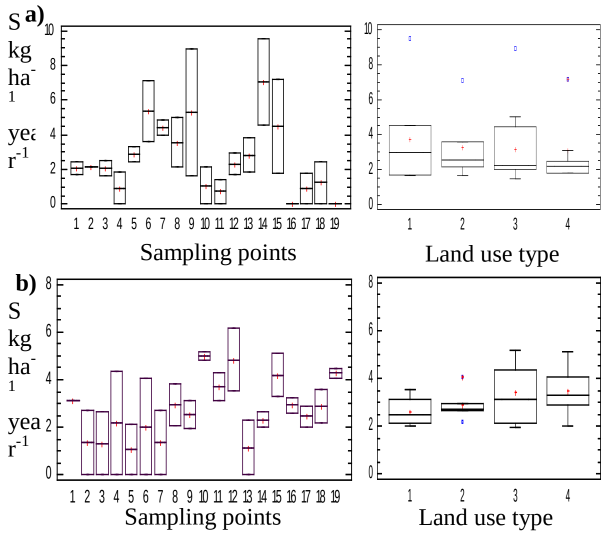

Figure 6. Critics loads of sulfur for the **a)** dry and **b)** rainy seasons for the 19 sampling points, and critics loads of sulfur according to the land use type (1 urban; 2 urban mangroves; 3 shipping ports and 4 conserved mangrove).

According to Gallego *et al.* [49] the highest records for the dry season were found in the areas with the highest anthropic activity. During the dry season, the reduced mixture layer concentrates the ionic species locally and regionally. According to Figure 6a and 6b, it can be observed that the use of the soil did not influence on levels of sulphate deposition, it is distributed homogeneously in the region of study. This fact supports the hypothesis that only regional sources contributed in the process of deposition of this ion, thus increasing the background levels of the region. This happens in accordance with the regional character of sulphate and its precursor (SO_2), the process lasts approximately 5-13 days.

Flux of S deposition showed the contrast between different areas, the highest sulfur deposition in the dry season is in the ports of Guayaquil, it is 8.9247 S kg ha^{-1} year^{-1} and in the area Trinipuerto, here the deposition is 7.1163 S kg ha^{-1} year^{-1}, in the rainy season the flux are 50% less comparing with the dry season, in the surroundings of the investigation area there are two thermoelectric plants, highway, a cement plant, and a steel factory.

There were no statistically significant differences in the sulfur critical loads, the highest values have been found in the Trinitaria Island versus the Trinipuerto (sampling point 6) and in Contecon (sampling point 9 and 14), it was the reaction to dry deposition processes related to the ports shippers (Figure 6a).

The S flux in the rainy season do not present statistically significant differences since the p-value of the F test is greater than or equal to 0.05, that is, the samples do not vary from one site to another. The sulfur critical load distribution was homogeneous in the study area, they do not vary seasonally (Figure 7), atypical values are presented in places such as the Port of Guayaquil (Contecon) and Trinipuerto that respond to dry deposition.

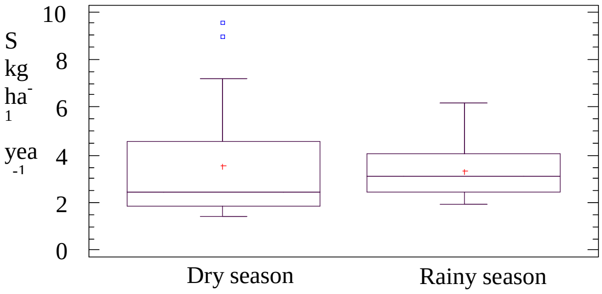

Figure 7. Critical loads of sulfur for the dry and rainy seasons.

3.6. Wind speed analysis

The wind speed analysis of the dry season (November and December 2016) shows that winds were predominated for the ranges of 2.1-3.6; 3.6-5.7 and 5.7-8.8 m s^{-1} with percentages of 44.2, 26.1 and 19.4 respectively, and, for the rainy season (January and February 2017) the wind data were predominated for the range 0.5-2.1, 2.1-3.6, 3.6-5.7 with 23.3 55.7 and 14 % respectively (Table 2). It was also observed that during dry season the wind speed is higher in comparison with the rainy season.

Table 2. Wind speed frecuency in m s^{-1} in the study area.

Wind velocity (m s^{-1})	Dry (%)	Rainy (%)
<0.5	2.0	4.5
0.5 -2.1	8.1	23.3
2.1 – 3.6	44.2	55.7
3.6 – 5.7	26.1	14.0
5.7 – 8.8	19.4	2.5
8.8 – 11.1	0.1	0.0
<11.1	0.1	0.0

Also, it was observed that the distribution for the months of dry season is prevalent in the south-west direction and 16% of the presented data is a distribution of the south-east. In the months of rainy season the winds have a south-west direction from the Pacific Ocean to the south-east, in Guayaquil is also present a northeast distribution (Figure 8a, 8b).

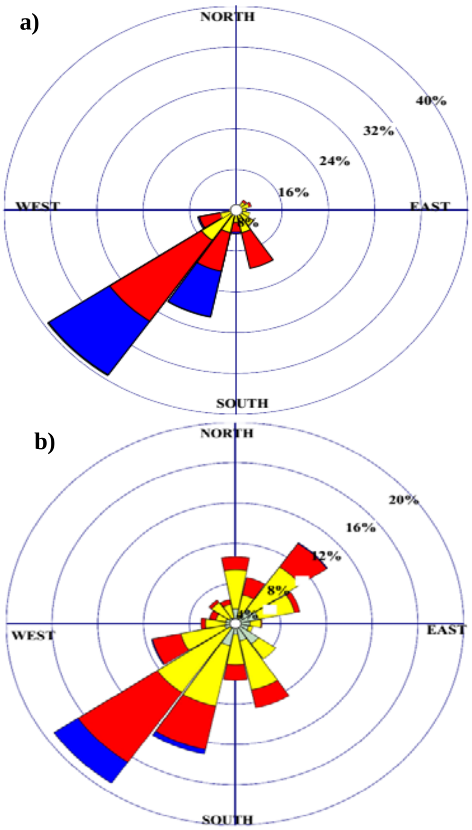

Figure 8. Wind direction for a) dry season and b) rainy season.

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4 Conclusions

The concentrations of photosynthetic pigments, proteins, salts and other elements are closely related to the atmospheric conditions in the Port of Guayaquil, which affects the morphological characteristics of *Rhizophora harrisonii;* climatic conditions such as dry and rainy seasons are a determining factor, which allows this species to recover in a cyclical manner.

Rhizophora harrisonii is a resilient specie, affected by the aerosols generated in the port area of Guayaquil. The dry season is a trigger, which directly affects the species because there is no opportunity to wash the leaves by the rain water. The leaves have elements that settle on them (dry deposition) for seven-eight months due to the absence of rain. The insects take the advantage to eat the leaves which are more exposed to the sun because of the presence of aerosols.

The results for atmospheric deposition fluxes of S show that there were not significant differences among the different land-use categories for ion sulphate. It strongly support the fact of regional sources contributed to deposition process of this ion, enhancing the background levels in this region. It is agree with the regional character of sulfate and its precursor SO_2 residence time ~13 days.

The flux of S exceed the permissible values for ecosystems such as mangrove, the critical loads (2 - 3 S kg ha^{-1} year^{-1}) respond to a regional distribution from southern Ecuador where Puerto Bolívar is located (Province of El Oro) and could contribute to the increase of SO_2 in the Port of Guayaquil.

For a best estimation of critical loads, it was suggested more exhausted detail of the flux which has an influence on the balance of mangrove ecosystems [50]. This would allow applying and analyzing other more complex calculation methods (weathering rate) and working with specific data of each ecosystem (soil profiles, mineralogical information, forest species, among others).

Further studies should be directed to the determination of nitrogen oxide [51], because most of the acid rain is caused by the concentration of sulfur dioxide and nitrogen oxide in the air.

The affectation by acid rain was highlighted in the foliar damage of the leaves and it came out with necrosis and chlorosis which might be analyzed more thoroughly in order to establish the different types of damages caused by *Tyrinteina arnobia stoll.* The affectation of acid rain can be compared between the areas of mangrove through the use of drones with high resolution cameras. It will allow to visualize in more details the differences of the shades from green to yellow.

For a polluting element, the sustainable emission rate is not bound to be greater than the rate at which the polluting element can be absorbed, recycled or sterilized by the environment [52]. This premise cannot be guaranteed for the study area in Ecuador, because for this it is required the creation of a baseline with its own critical loads.

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