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QUANTIFICATION OF THE CO_2 FLOW IN THE SOIL COLONIZED BY AVICENNIA GERMINANS, LOCATED IN THE LOS TOTUMOS, MIRANDA STATE, VENEZUELA

CUANTIFICACIÓN DEL FLUJO DE CO_2 EN EL SUELO COLONIZADO POR AVICENNIA GERMINANS, EMPLAZADO EN EL HUMEDAL LAGUNA GRANDE, SECTOR LOS TOTUMOS, ESTADO MIRANDA, VENEZUELA

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Abstract

Mangrove ecosystems are estimated to have a high impact potential on the global carbon cycle because of their high organic matter content (Dittmar et al., 2006), and consequently a significant importance in CO₂ flow as the greenhouse gas with the greatest impact on global warming (Benavides and León, 2007; Caballero et al., 2007; CESPA, 2013). In Venezuela, however, studies focusing on this issue are scarce, hence the aim of this research was to specify significant differences in the flow of CO₂ in soil colonized by *Avicennia germinans*, as well as non-rizospheric located in Los Totumos sector. Research developed in three phases was applied: (a) fieldwork, in order to collect 120 surface soil samples (0-20 cm), (b) laboratory, with the purpose of estimating the CO2 flow from the basal breathing method (Anderson, 1982) and (c) statistical analysis, to identify significant differences based on the ANOVA and Tukey test. It is evident that there are significant differences, with flows of 7.51 mg C-CO₂ g/24h in the rhyspheric area and 1.49 mg C-CO₂ g/24h for non-rizospheric. It is concluded that: (a) microbial activity in the soil, induced by the presence of *Avicennia germinans*, is possibly contributing to its evolution, and (b) it is reaffirmed that mangrove ecosystems are a source of CO₂ and reservoir gas to the impact of climate change.

Keywords: Basal breathing, microorganisms, CO₂, mangrove, Avicennia germinans.

Resumen

Se estima que los ecosistemas de manglar tienen un elevado potencial en el ciclo global de carbono por su alto contenido de materia orgánica (Dittmar et al., 2006), y en consecuencia una relevante importancia en el flujo del CO₂ como el gas invernadero de mayor impacto en el calentamiento global (Benavides and León, 2007; Caballero et al., 2007; CESPA, 2013). No obstante, en Venezuela los estudios centrados en este asunto son escasos, es por ello que la presente investigación tuvo como propósito precisar diferencias significativas en el flujo de CO₂ en suelo colonizado por *Avicennia germinans*, así como no rizosférico, emplazado en el sector Los Totumos. Se asumió una investigación desarrollada en tres fases: (a) trabajo de campo, a fin de colectar 120 muestras de suelo superficial (0-20 cm), (b) laboratorio, con el propósito de estimar el flujo de CO₂ a partir del método de respiración basal (Anderson, 1982) y (c) análisis estadístico, para identificar con base en el ANOVA y prueba de Tukey, diferencias significativas. Se evidencia que existen diferencias significativas, con flujos de 7,51 mg C-CO₂ g/24h en la zona rizosférica y 1,49 mg C-CO₂ g/24h para la no rizosférica. Se concluye que: (a) la actividad microbiana en el suelo, inducida por las condiciones edáficas que genera la presencia de la *Avicennia germinans* posiblemente está contribuyendo con la evolución del mismo, y (b) se reafirma que los ecosistemas de manglar constituyen fuente de CO₂ y sumidero del gas ante el impacto del cambio climático.

Palabras clave: Respiración basal, microorganismos, CO2, manglar, Avicennia germinans.

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1 Introduction

Soil is an important reservoir of carbon since around 1500 PG C (petagrams of carbon) can be found in its first meters, while it is estimated about 800 PG C in the atmosphere, and about 500 PG C in the terrestrial vegetation. However, it is in constant motion in different molecular forms (Organización de las Naciones Unidas para la Alimentación y la Agricultura, 2017).

Carbon dioxide (CO₂) is one of the organic carbon derivatives in constant interaction with the climate. In the soil-atmosphere relationship, this greenhouse gas is produced by various conditions: (a) by respiration of micro and macro organisms as well as by living roots in the rhizosphere, in addition to organic matter decomposed directly from plants or through trophic chains (Departamento de Agricultura de los Estados Unidos, 1999; Organización de las Naciones Unidas para la Alimentación y la Agricultura, 2017), and (b) absorbed into the soil from the atmosphere by plants during photosynthesis by which carbon is fixed to produce biomass, and the oxygen particle is released into the atmosphere (Visconti and De Paz, 2017).

However, this sequestration of CO_2 into the soil structure is important to the global climate cycle, as it contributes to the balance of greenhouse gases in the atmosphere. It is important to note that forests cover 29% of the planet's surface area, and the carbon found in its soils constitutes approximately 36% of the total located at one meter deep (Zambrano et al., 2004), soil respiration being the most important flow of the carbon cycle (López and Monterroso, 2020), representing approximately 75% of the total in these ecosystems (Law et al., 2001). In the case of mangroves, Moreno et al. (2002); Kauffman et al. (2013), agree that they contain the largest carbon reserves due to the development of a complex ecosystem that contributes to the capture of CO_2 in the soil.

According to Kao et al. (2010), due to the dominance of certain typical plants in wetland environments, oxygen flows from the atmosphere to the surface and subsurface area of the soil, specifically to the rhizosphere by promoting the productionoxidation of greenhouse gases; hence, the contribution and quality of organic carbon can be influenced

by the forest capacity of the dominant species. It is possible that since they are located in intertidal zones, i.e., in the transition between high tide and low tide zones, these types of forests are periodically flooded by the action of the squid, so that the soil passes through dynamic oxidation-reduction processes, generating conditions conducive to the activation of microbiological processes such as nitrification, denitrification and methanogenesis, which are greenhouse gas generators that can sometimes be released into the atmosphere (Chen et al., 2016).

Sánchez et al. (2011), mention that soils of wetlands where mangroves are developed store long-term carbon since they are usually largely flooded. However, due to little recognition of these reservoirs and the impacts generated by anthropogenic actions, these have been reduced by 35% of the total. Actually, anthropogenic action related to various uses of soil combined with the logging and burning of large areas of land has generated disruption of the ecosystems that constitute reservoirs, and consequently an environmental imbalance, encouraging soils to move from sinks to sources of CO₂, worsening global warming on the planet (Sánchez et al., 2011; Visconti and De Paz, 2017; Organización de las Naciones Unidas para la Alimentación y la Agricultura, 2017).

Hence, Villalobos (2012), mention the importance of managing appropriately carbon-reserve habitats, such as mangroves, in order to know their forest capacity for the production-storage of greenhouse gases, and to contribute to the mitigation of the impacts generated to the climate system. In this regard, research has been carried out to know the forest potential of mangrove forests related to the production and sequestration of CO₂; among these Kauffman et al. (2013) and Herrera et al. (2016), mention that Mexico and Brazil are among the 4 countries with the largest mangrove reserve worldwide. Moreno et al. (2002), Lozano (2007) and Sánchez et al. (2011), agree that mangroves within coastal wetlands are an important reservoir for carbon fixation or capture.

For Costa Rica and Colombia, Yepes et al. (2016) and Villalobos (2012), respectively, claim that mangroves are part of the planet's most productive carbon-producing ecosystems, and thus constitute a key environment for global warming mitigation.

Venezuela, being in the middle of the inter-tropical zone and possessing an extensive coastline, has important mangrove communities on which various studies have been carried out aimed at recognizing their geographical distribution, as well as their structural characterization based on biology, botany and agronomy, (Pannier and Pannier, 1989; Medina and Barboza, 2003; López et al., 2011; Cumana et al., 2000; Bonilla et al., 2010; Romero and Meléndez, 2013).

Núñez et al. (2019), reported differences in the flow of CO₂ in non-rhizospheric soils for the mangrove in Boca de Uchire, located at 94.93 km southeast of the Totumos, in contrast to those dominated in surface by Avicennia germinans and Conocarpus erectus. Likewise, Sánchez et al. (2010), identified variations in the flow of CO₂ in soils colonized by Rizhophora mangle on Margarita Island, estimating that because climatic conditions affect the availability of salt in the soil, it also affects the flow of the gas. Studies on the quantification of organic carbon, as well as carbon dioxide flows in these ecosystems are scarce, and so far, these have not been conducted in the study area: Laguna Grande wetland, Los Totumos. This area is located at the southwest of Cabo Codera, between 10° 32' 34" and 10° 32' 44" north latitude, and between 66° 4' 44" and 66° 4' 54" west longitude, bordering the Coastal Range to the north, Buche Bay to the west and the Caribbean Sea to the south and east (Figure 1). It extends over 7.77 ha, of which the monospecific forest of Avicennia germinans occupies 4.42 ha, representing 56.88% of the total area (Figure 2).

According to Garrido (2017), geomorphologically the area is a semi-elongated beach-like reservoir with a length of approximately 400 meters, and has a smooth relief with a greater prominence towards the north and south ends, which mainly consists of medium-sized particles with predominance of bioclasts. The data of the Carenero and Tacarigua Mamporal climatic stations, attached to the Hydrology and Meteorology Office of the Ministry of Power for the Environment (Ministerio del Poder Popular para el Ambiente, 2013a,b), located on the same coast line without marked altitudinal and latitudinal differences compared to Los Totumos, allow to state that the area has temperatures corresponding to an isothermal regime with an estimated annual average of 26.75°C, the warmer months being April and October with 27.7°C, and the cooler months from November to March, with a registered minimum of 25.15°C. Rainfall is distributed in a unimodal regime to reach an annual amount of 1141.3 mm, the rainy period extends from June to December. Evaporation is high throughout the year, with an annual amount of approximately 1781.9 mm (Figure 3).

According to the Goldbrunner classification (Foghin, 2002), the area is located on the tropical thermal floor, with altitudes that do not exceed 10 meters above sea level. As for the edaphic features, Entisols predominates with soil characterized by presenting a little developed material with a depth lower than 10 meters, an incipient surface horizon resting on the rocky material, sandy texture, highly saline and alkaline from the Orthens suborder and the Torriorthens group (Cárdenas, 1965; Elizalde et al., 2007; Gobernación del Estado Bolivariano de Miranda, 2010).

Huber (2007) and Gobernación del Estado Bolivariano de Miranda (2010) agree that the dominant vegetation for the coastal stretch presents swords, palms, pastures and mangroves. In Los Totumos, the development of mangroves in monospecific forest of *Avicennia germinans* with closed coverage is identified. Lentino and Bruni (1994) report the mouths of the streams: The Totumos, Horno, Laguna Grande and Hoyo de la tierra, with a Uadi type regime. In addition, Lara et al. (1997) claim that the Curiepe, Capaya and Tuy are rivers flow, with permanent runoff patterns.

Because of the latter, it is assumed that the riparian and monospecific mangrove of *Avicennia germinans* located on Entisols soils of Los Totumos in Laguna Grande wetland is a space to identify locally the possible forest potential of the aforementioned mangrove species in the CO_2 flow, although it has not yet been studied to know its structural conditions, as well as its contribution to the total carbon cycle. For this reason, the aim of this research was to identify significant differences in the flow of carbon dioxide (CO_2) in soil colonized by *Avicennia germinans* as well as non-rhizospheric, from the contrast of surface samples (0-20 cm depth).

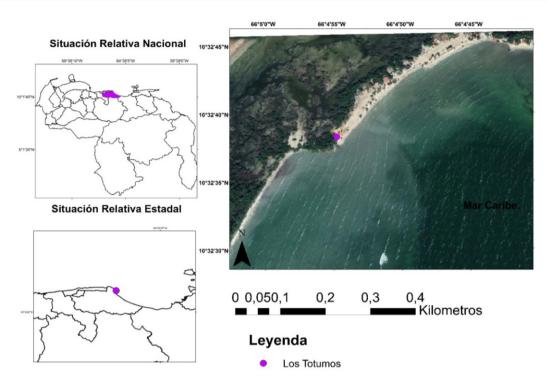


Figure 1. Location Map of Los Totumos. Elaborated using the Lansatd 8 Satellite Image. Combination of bands 321: Natural color.

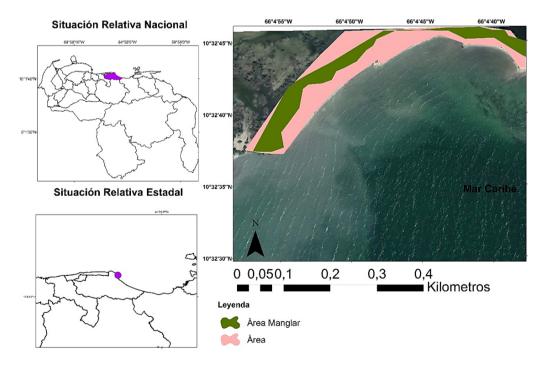


Figure 2. Map of the area covered by the forest of Avicennia germinans at Los Totumos, Laguna Grande wetland.

2 Materials and Methods

2.1 Field phase

Field work was carried out in May 2019, considered an active rainy period, which is an essential aspect in the collection of soil samples for the purpose of quantifying the CO₂ released. Indeed, as Luo and Zhou (2006) report, higher water availability generates an increase in the metabolic activity of roots and microorganisms, which is expressed in increased breathing. During the execution of this phase, the general observation of the area was made in order to recognize the coverage patterns of the mangrove, characterizing it as a monospecific forest of Avicennia germinans, as well as to identify the accessibility and homogeneity of the area, looking at areas that will visibly have the least impact associated with anthropogenic action. Based on this information, four plots of 1000 m^2 were selected, which represent approximately 10% of the total forest area.

In order to collect the soil samples, two plots colonized by *Avicennia germinans* were selected in the mangrove area, these being considered to be those that receive the influence of the species as an intervening element in the flow of CO_2 in the soil, and two plots that even though are located in the mangrove area do not have surface vegetation, so they are considered non-rhizospheric, which for the investigation constitute the sample without intervening element.

Soil samples were collected at a surface level (0-20 cm), considering: (a) the soil in the area closest to the base of the individual tree for areas with the presence of Avicennia germinans, and (b) a distance of no less than 5 linear meters per sampling point for the area deprived of mangrove. Under the criteria described above, 120 soil samples were collected, distributed as follows: (a) 60 soil samples dominated by *Avicennia germinans*, and (b) 60 samples of non-rhizospheric or surface mangrove-free soil.

The samples were stored in dense polyethylene bags, identified with a content label with the following information: Unique identification key, coordinates of the sampling point, date and time of collection. They were preserved for their transportation to the laboratory at a temperature controlled between 4° C and 6° C.

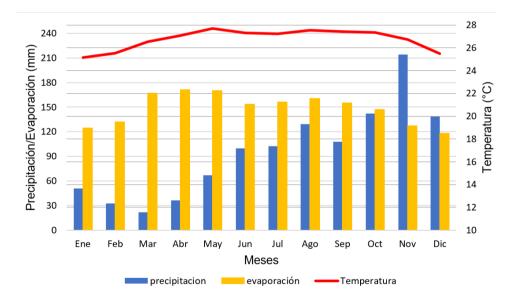


Figure 3. Climate diagram elaborated from the information in the data sheet of the stations, obtained through the Hydrology and Meteorology Office of the Ministry of Power for the Environment. Precipitation data are for Carenero station, and temperature and evaporation data for Tacarigua Mamporal station.

2.2 Laboratory phase

During the development of this phase, the CO₂ released was determined using the basal breathing method, based on the protocol reported by Anderson (1982). For this purpose, soil samples preserved at field humidity and sifted in the 10 mm sieve were pre-conditioned at room temperature (24°C) in order to achieve stabilization. Subsequently, 50 g of each soil sample was placed in 500 ml glass bottles with their lids. In addition, an alkali trap was suspended in amber glass bottles, containing 5 ml of a 0.1 mol of Sodium Hydroxide solution (NaOH). The treated samples were placed in a dark area in the laboratory. After 24 hours, the trap was removed and the solution was placed in glass bottles, adding 2 ml of barium chloride (BaCl₂) and 2 drops of phenolphthalein ($C_{20}H_{14}O_4$) as a pH indicator to determine the alkalinity of the solution. The absorbed CO₂ was titrated with 0.1 mol of hydrochloric acid (HCl). The result was expressed in mg C- CO_2 g/24 h. Additionally, during the execution of this procedure, jars without soil samples with alkali traps were also used in order to establish a comparison pattern.

2.3 Statistical analysis phase

To determine possible variations in the four plots under study, as well as to identify the influence of *Avicennia germinans* as a variation source in the flow of CO_2 in soil, data were statistically analyzed from the following tests: (a) the analysis of variance (ANOVA) to specify the significant differences between the means of each group of samples, corresponding to the four plots under study; and (b) the Tukey test to identify which sample groups are different and honestly significant, once significant differences have been assumed in at least one of the four sample sets.

3 Results and Discussion

Table 1 presents the minimum, maximum and average values for the flow of CO₂ (mg C-CO₂ g/24h) in the soil for the four plots under study. The estimated ANOVA per plot yielded 334.41 for test F and a probability of 6.5×10^{-57} (Table 2), which allows to assume that the average is different with 95% of reliability in at least one group of data out of the four plots under study.

Table 1. Descriptive analysis of the flow of CO₂ (mg C-CO₂ g/24h) in the soil.

Coverage	Minimum Value	Maximum Value	X	S	O ₂ '
Parcel 1 (cm)	1.11	8.89	7.39	1.11	1.20
Parcel 1 (sm)	0.10	2.39	1.63	0.90	2.30
Parcel 2 (cm)	1.30	9.37	7.63	1.31	1.65
Parcel 2 (sm)	0.10	2.74	1.35	0.76	0.55

cm= with mangrove/ sm= without mangrove/ x= mean/ s= typical deviation/ O'_2 = variance.

 Table 2. Analysis of variance (ANOVA) for the four parcels under study.

Origin of variations	Suma of squares	Degrees of freedom	Averages of squares	F	Probability	Critical value of F
Between groups	1089.08	3	363.02	334.41	$6.5*10^{-57}$	2.68
Within groups	125.92	116	1.08			
Total	1215.00	119				

Tukey test showed an HSD value of 0.69 which allows to state that: (a) there are no significant differences between the weighted averages of samples for plots with Avicennia germinans, and no significant differences are identified between the weighted averages of samples for non-rhizospheric soil, and (b) there is significant difference in comparing the weighted averages of plots with surface mangrove with those that do not present it (Table 3; Figure 4). A significant difference ^(HSD) is considered if the mean difference between the sample groups is higher than the HSD value.

Table 3. Tukey test of significant difference	e for CO ₂ flow from	soil samples in th	he four plots under	r study.
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HSD = 0.69	Parcel 1 (cm)	Parcel 1 (sm)	Parcel 2 (cm)	Parcel 2 (sm)
Parcel 1 cm	***	5.76 ^(HSD)	0.24	6.03 ^(HSD)
Parcel 1 sm	$5.76^{(HSD)}$	***	$6.00^{(HSD)}$	0.27
Parcel 2 cm	0.24	$6.00^{(HSD)}$	***	$6.27^{(HSD)}$
Parcel 2 sm	$6.03^{(HSD)}$	0.27	$6.27^{(HSD)}$	***

cm= with mangrove/ sm= without mangrove

According to the above analysis, it can be said that the soil covered by *Avicennia germinans* in the study area has an average CO_2 flow of 7.51 mg C- CO_2 g/24h, which is significantly different from the estimated 1.49 mg C-CO₂ g/24h for soils free of the mangrove or non-rhizospheric species. This represents a flow of 83.44% for the soil covered by mangroves, in contrast to the 16.56% estimated for the non-rhizospheric soil (Table 4; Figure 5). As already mentioned, Núñez et al. (2019), identified in Boca de Uchire, significant differences, estimating that for non-rhizospheric soil the flow is 3.74 mg C-CO₂ g/24h, while those colonized by *Avicennia germinans* register 10.61 mg C-CO2 g/24h, and *Conocarpus erectus* 13.88 mg C-CO₂ g/24h.

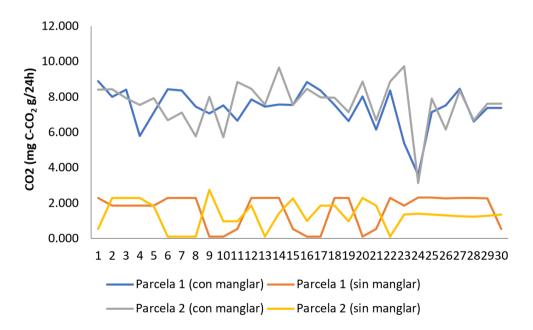


Figure 4. Distribution of CO₂ flow (mg C-CO₂ g/24h) in the soil by plot and coverage.

It is possible that the lower value of CO_2 flow in the soil colonized by *Avicennia germinans* in Los Totumos sector, compared with that reported for the same mangrove species in Boca de Uchire is linked to edaphic salinity conditions, because excess of this component not only affects the structural development of plants (Cintron et al., 1978), but limits their forest potential as a CO_2 sink (Mitra et al., 2004).

In fact, Sánchez et al. (2010) identified variations in CO₂ flow in soils colonized by *Rizhophora mangle* associated with sodium fluctuations by climatic periods, registering for highly saline soil: 21.1 ± 18.9 mg C-CO₂ g/24h (dry period), 25.0 ± 12.3 mg C-CO₂ g/24h (rainy period) and 31.9 ± 18.5 (transition period). Although these are different mangrove species, it is possible that the high salinity condition for the soil of Los Totumos is an influential factor in the flow of CO₂.

Singh et al. (2010), report that slow rates of soil respiration can be considered as indicators of limited biological activity, stress generated by inadequate use, climatic disturbances, or even resource deficits. According to Carrero and García (2009), these mangroves have been cut off for conducting engineering works for tourist and commercial purposes, generating some negative effects. Therefore, it is considered that anthropogenic intervention in the area has generated disturbance conditions to the ecological balance of the mangrove, affecting the structural development of the forest, and limiting forest potential in the capture and storage of CO_2 in the soil.

Table 4. Distribution percentage of CO_2 flow (mg C-CO2g/24h) in the soil by coverage.

Coverage	CO ₂ average	%
With Mangrove	7.51	83.44
Without Mangrove	1.49	16.56

Regarding oxygen penetration in the soil in mangrove areas, Olguín et al. (2007), report that it is limited to the first centimeters of the soil profile, because the near-surface groundwater level enhances the reduction conditions, causing bacterial respiration to use NO₃, MnO₂, FeOH₂, SO₄ and CO₂ in the anaerobic zone as final electron acceptors. Hence, the contribution of CO2 to the anaerobic areas of the soil, possibly caused by the respiration of man-

grove roots, generates conditions for the existence of anaerobic bacteria responsible for the decomposition of organic matter into methane, by giving this area the sink condition of these greenhouse gases.

In the oxygenation zone, CO_2 intake is possibly caused by the respiration of aerobic microorganisms that inhabit the rhizosphere, constituting an indirect indicator of microbial activity. In effect, Karmarkar (1982) reports that such biological activity linked to the proliferation of microorganisms favors the accumulation and decomposition of organic residues provided by the mangrove, in the period when the soil remains low in water.

However, even in periods of flooding, Mitsch and Gosselink (2000) say that such water flows help to reduce the reduction conditions in the surface area of the soil, because these sheets provide dissolved oxygen, which favors microbial activity and the consequent release of CO₂. This CO₂ released is generally transformed by photosynthetic bacteria into carbohydrates used by plants. Additionally, the microorganisms that generate it are essential for the decomposition of organic matter into labile humus, beneficial for the development of plants by its contribution of macronutrients such as nitrogen, necessary for the succulence and greenery of leaves.

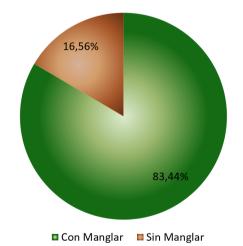


Figure 5. Distribution percentage of CO_2 flow (mg C-CO₂ g/24h) in soil by coverage.

Indeed, Hesse (1961) and González et al. (2016), state that *Avicennia germinans* requires soils with at least 0.4% of nitrogen for its development. In this sense, Holguín et al. (2007), say that the proli-

feration of microorganisms leads to an increase in the rate of biological nitrogen fixation, contributing from 40 to 60% of the requirements of the mangrove ecosystem.

The development of these forests generates a positive balance for the climate system, since as mentioned by Canadell et al. (1995), they present a high potential in the sequestration of carbon in the soil, in response to the increase in atmospheric CO₂. Hence, Zhong and Qiguo (2001) affirmed that these soils should be considered as natural regulators of atmospheric CO₂ concentration and flow. Indeed, Bouillon2008, stated that these forests directly capture 4996 g of CO₂ m⁻² per year.

Based on the latter, the surface dominance of mangrove species contributes to the increase of CO_2 in the soil, and additionally, it is inferred that such variation may have as a conditioning element the edaphic potentialities of the colonizing species, along with climatic, edaphic and anthropogenic variables.

In addition, the microbial activity associated with the presence of *Avicennia germinans* in Entisols soils such as the one in Los Totumos can be interpreted as an indicator of its incipient pedogenetic development, since the bacteria activate the humification process, which allows assuming that these naturally mineral soils are added by organic matter transformation and organic compounds that progressively increase aeration, cation exchange capacity and cause improvements in structure.

4 Conclusions

Based on the results of this research, it is concluded that a monospecific forest of *Avicennia germinans* is developed in the area of study, covering approximately 56% of the total area of Los Totumos.

Similarly, the coverage of this mangrove species contributes to the development of microbial activity in the surface area of Entisols soil, which is inferred from the estimated significant differences in respiration of samples corresponding to rhizosferic and non-rhizosferic zones. In fact, 7.51 mg C-CO₂ g/24h was estimated in the area colonized by the above-mentioned mangrove species, while the estimated

breathing was 1.49 mg C-CO₂ g/24h in the nonrhizospheric zone without surface vegetation. This represents a slightly more than 500% increase in the flow of CO₂, associated with the presence of mangrove in the surface area of the soil. Also, the soil microbial activity induced by edaphic conditions that generates the presence of *Avicennia germinans* is possibly contributing to the evolution of the soil. Finally, it is reaffirmed that mangrove ecosystems are a source of CO₂ generation, so its study should contribute to generating information that allows the decision-making on its management in order to mitigate the emission of this greenhouse gas into the atmosphere.

It is necessary to continue conducting studies in the above-mentioned mangrove to deepen the analysis of the possible relationships between its structural conditions, the soil properties in which it is located and the total carbon flow.

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