UNIVERSIDAD DE INVESTIGACIÓN DE TECNOLOGÍA EXPERIMENTAL YACHAY

Escuela de Ciencias Biológicas e Ingeniería

TITULO: "Ecuadorian native plants as possible volatile organic compounds reducing agents in air pollution"

Trabajo de integración curricular presentado como requisito para la obtención del título de Ingeniería Biomédica.

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Dedication

I dedicate this thesis to the three most important people, who mean everything in my life. First and foremost, to my parents Cesar and Miriam, who did not only raised me but also allowing me to study and also taught me the value of hard work and dedication.

I also wish to dedicate this thesis to my brother Sebastian for his total support during my whole life and have never left my side. I will always appreciate all you have done for me. Sebas, you are my best friend, and I love you so much.

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Abstract

Air pollution is known as the presence of toxic or harmful substances in the atmosphere. It is considered a public health problem that is continuously causing adverse effects on humanity and the ecosystem. It is associated with several diseases such as respiratory, neurological, and cardiovascular. According to INEC, in 2016, there were 2643 deaths due to diseases associated with environmental pollution. Specifically, in this project, VOCs are the air pollutants evaluated. Previous studies demonstrate that some plants leaves have a significant capacity to absorb VOCs. To take advantage of Ecuador's biodiversity, plant leaves from different areas of Ecuador has been tested to prove their efficiency to capture VOCs. During the experimentation, the plant leaves were physicochemically characterized, and, using gas chromatography (GC), the performance of the gas absorption was tested. Results show that some leaves are capable of absorbing around 20-28% of the tested gas. These percentages represent a significant absorption efficiency, which allows concluding that these plant leaves represent an eco-friendly method to reduce air pollution.

Key Words: Leaves; VOCs; Biodiversity, Air pollution; Remediation

Resumen

La contaminación del aire se conoce como la presencia de sustancias tóxicas o nocivas en la atmósfera. Se considera un problema de salud pública que continuamente causa efectos adversos en la humanidad y el ecosistema. Se asocia con varias enfermedades como las respiratorias, neurológicas y cardiovasculares. Según el INEC, en 2016, hubo 2643 muertes debido a enfermedades asociadas con la contaminación ambiental. Específicamente, en este proyecto, los VOC son los contaminantes del aire evaluados. Estudios anteriores demuestran que algunas hojas de las plantas tienen una capacidad significativa para absorber los COV. Para aprovechar la biodiversidad de Ecuador, se han probado hojas de plantas de diferentes áreas de Ecuador para demostrar su eficacia para capturar VOC. Durante la experimentación, las hojas de las plantas se caracterizaron fisicoquímicamente y, utilizando la cromatografía de gases (GC), se probó el rendimiento de la absorción de gases. Los resultados muestran que algunas hojas son capaces de absorber alrededor del 20-28% del gas analizado. Estos porcentajes representan una eficiencia de absorción significativa, lo que permite concluir que estas hojas de plantas representan un método ecológico para reducir la contaminación del aire

Palabras Clave: Hojas, VOCs, Biodiversidad, Contaminacion del Aire, Remediacion.

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1. Introduction

1.1. Air Pollution and Main Pollutants

Recently, environmental air pollution is a high concentration of foreign matter, which causes harmful effects on the urban population. (Admassu & Wubeshet, 2014; Amitava, 2002; WHO,2008).The production of air pollution is caused by a range of factors, including indoor and outdoor air pollution. The indoor air pollution is the substances generated from housing activities such as heating, cooking, smoking, the use of cleaning products and paints(Midouhas, Kokosi, & Flouri, 2019) and, on the other hand, the outdoor pollution comes from outdoor services such as traffic emissions and industrial processes. (Midouhas, Kokosi & Flouri, 2019;). The pollutants from natural or artificial sources affect the atmosphere once the substances are released into the atmosphere, where undergoes a mixing or chemical transformation and, therefore, triggering an impact on different receptors like humans, animals, or plants (Kemp et al., 2011; Admassu & Wubeshet, 2014). Among the released- hazardous pollutants into the atmosphere are formaldehyde, carbon monoxide, nitrogen dioxide, asbestos fibbers, respirable particles, and volatile organic compounds (VOCs) (Kemp et al., 2011).

1.2. Volatile Organic Compounds

Volatile Organic Compounds(VOCs) are generally defined as carbon-containing chemical and low-molecular-weight, which are highly reactive to evaporate under average atmospheric temperature and pressure (Berenjian, Chan, & Malmiri, 2012; EPA, 2017). According to the World Health Organization (WHO), an organic compound with a boiling point around (50-100ºC) to (240-260ºC) and with a saturation vapor pressure higher than 102kPa at 25ºC is considered a VOCs. The chemical structures of VOCs are small hydrocarbons, acids, alcohols, aldehydes, aromatics, ketones, terpenes, thiols, and their derivatives (Pennerman, Al-Maliki, Lee, & Bennett, 2016). These chemical and physical properties allow them to travel long distances from the sources (Midouhas et al., 2019; Pennerman et al., 2016).

1.2.1. Classification of Volatile Organic Compounds.

Volatile Organic Compounds are classified by WHO according to their boiling point range and ease of emission, as it is described in *Table 1*. This classification is all indoor air. (EPA, 2017). For example, Very volatile organic compounds (VVOCs) are more likely to be found in the gas phase in the air rather than in materials or surfaces, on the other hand, Semi-Volatile Organic Compounds are found in the air in small quantities, and are found in solids or liquids that contain them or on surfaces (WHO, 2008; Watson, Chow & Fujita, 2001).

Description	**Abbreviation**	**Boiling Point Range (°C)**	**Example Compounds**
Very volatile (gaseous) organic compounds	VVOC	<0 to 50-100	Propane, butane, methyl chloride
Volatile organic compounds	VOC	50-100 to 240-260	Formaldehyde, d-Limonene, toluene, acetone, ethanol (ethyl alcohol) 2-propanol (isopropyl alcohol), hexanal
Semi volatile organic compounds	SVOC	240-260 to 380-400	Pesticides (DDT, chlordane, plasticizers (phthalates), fire retardants (PCBs, PBB))

Table 1.Classification of Volatile Organic Compounds by their Boiling Points

1.2.2. Sources of the VOCs.

The large variety of Volatile Organic compounds are emitted in the atmosphere from different sources (Mishra, Bartsch, Ayoko, Salthammer, & Morawska, 2015; Niu, Mo, Shao, Lu, & Xie, 2016). The primary source of these compounds is from biogenic emissions, which is around (760 Tg(C) yr^{-1}) (Sindelarova et al., 2014; EPA.2017), without ruling a significant amount from anthropogenic sources (127 Tg(C) yr^{-1}) (Guenther, 2007; Stocker et al., 2013).

The natural origins of VOCs are wetlands, forests, oceans and volcanos, and soils (Guenther, 2007; Sindelarova et al., 2014), and the main anthropogenic sources constitute a significant group of indoor and outdoor air pollutants (Mishra, Bartsch, Ayoko, Salthammer & Morawska, 2015) for example, biofuel and waste disposal, wheat residue burning, industrial emission, and solvent use *Figure 1*.

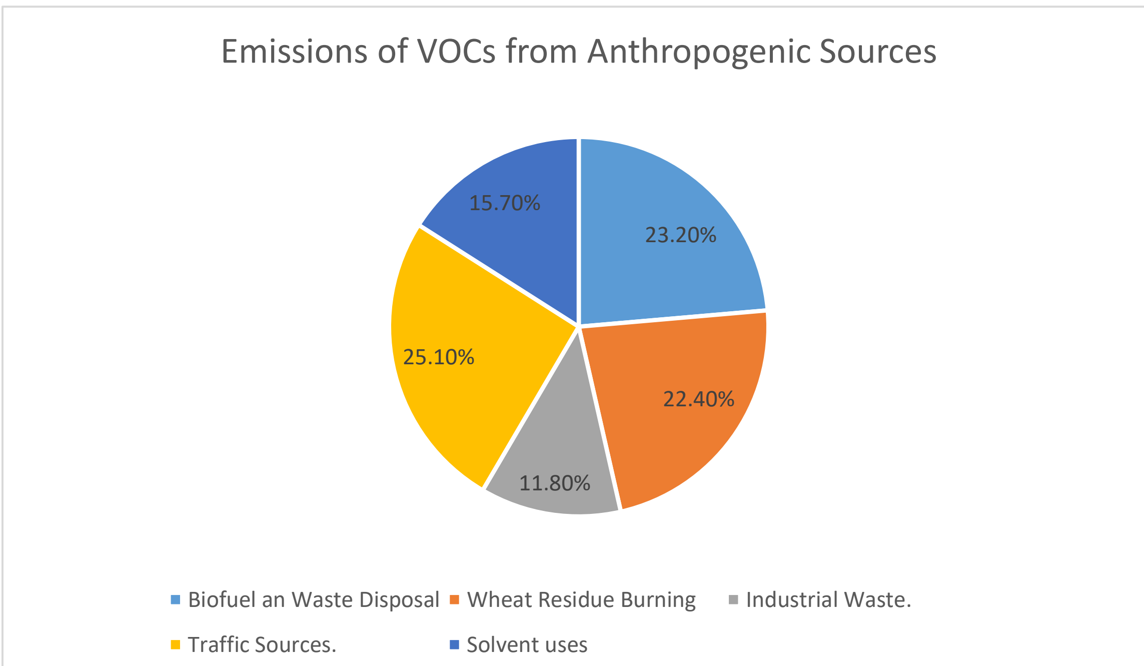

Figure 1. Percentage of VOC emissions from anthropogenic sources.

Once the emitted VOCs are in the atmosphere, some reactions take place due to the photo-chemically sensitivity of the VOCs to oxides of nitrogen and sunlight, resulting in ozone and other products. This procedure is the following this equation (1); (Berenjian et al., 2012)

$$NO_x + VOC + Sunlight \rightarrow \ O_3 + NO_x + other\ products. (1)$$

This reactions play an essential role in the ozone formation and fine particulate matter and is driven by available nitrogen oxides and VOCs which may results on some toxic pollutants that induce some photochemical smog in the troposphere (Berenjian et al., 2012; Soni, Singh, Shree, & Goel, 2018) and also other problems like ozone layer depletion in stratosphere and the greenhouse effect (Berenjian et al., 2012) (Soni, Singh, Shree & Goel, 2018).

According to Environmental Protection Agency (EPA, 2017), toxic VOCs are divided as carbonyl compounds (aldehydes and ketones), polycyclic aromatic hydrocarbons (PAHs), mono-aromatic hydrocarbons (MAHs), nitro-PAHs, oxy-PAHs. 1, 3 butadiene, aldehydes (formaldehyde, acetaldehyde), acrolein, aromatics (benzene, toluene, ethylbenzene, xylene, and styrene) n-hexane, naphthalene (Singh, Varun, & Chauhan, 2016).

1.2.3. **Effects of the VOCs**.

As previously mentioned, VOCs are part of air pollution and lead to a major amount of toxic pollutants all over the World (WHO, 2008). The concentration and the continuous exposure of these compounds cause short and long term diseases (Soni, Singh, Shree & Goel, 2017). Talking about short term VOC-exposure, there are many negative impacts on humans health such as eye and respiratory diseases, (Wolkoff*, P, Clausen, P.A, Wilkins, C.K, Nielsen, 2000) headaches, dizziness, visual, and memory impairment, while long-term VOCs absorption is related to damage liver, kidney, and central nervous systems (Kampa & Castanas, 2008; Ramírez, Cuadras, Rovira, Borrull, & Marcé, 2012; Rumchev, Brown, & Spickett, 2007; Zhou et al., 2011). Additionally, some studies show that these pollutants are cancer precursors due to the long exposure that may cause gene modification and also it harms the clonal expansion and escape from apoptosis (Soni et al., 2018; Stocker et al., 2013; Wieslander, Norbäck, Björnsson, Janson, & Boman, 1996).

1.2.4. Current Removal Methods of VOCs.

There are several physicals, chemical, and biological methods used for recovery or destruction of the VOC emissions (Berenjian et al., 2012; Soni et al., 2018), the objective of these methods is to convert VOCs into environmental-friendly products. The traditional methods are the use of carbon activated or air stripping for removal of contaminated liquids. In this methodology, using a reactor, the treated liquid makes physical contact with the liquid-carbon activated to allow dissolved the organic pollution to bind it (Das, Gaur, & Verma, 2004; Smith & Rodrigues, 2015). Other standard methods to eliminate VOCs could be trough oxidation by a thermal, combustion engine, catalytic, or UV, where VOCs are broken down into less hazardous compounds like water, hydraulic gas, and carbon dioxide. (R. Yang, Zhang, Xu, & Mo, 2007). However, the main problems of these techniques are the economic cost of production, their efficiency and also there is not a full elimination of VOCs, it is only a transformation of the content to another phase (Berenjian et al., 2012).

Biological methods to remove VOCs have been established, less than the traditional ones, but giving more advantages due to many reasons such as lower capital cost, easier operation, and an eco-friendly proposal. The use of microorganisms is the reason why this methodology is a better way to fight against the VOC emissions due to they can metabolize the compounds without any energy, temperature, and heat or radiation requirement (Berenjian et al., 2012). For example, different biological methods are Bio-filters, bio trickling, bio-scrubbers (Mukesh, Doble, Anil, 2005). The effectiveness and the economical parameters help to treat these pollutants in these ways. These treatments use air flows with lower VOC concentrations and allow them to apply in smaller scales (Berenjian et al., 2012). A common methodology is the use of different plants in order to mitigate VOCs. Plants can act as a filter, commonly known as phylloremediation. It refers to the use of organisms, in this case, plants, to assimilate, degrade, or transform any toxic substance into less hazardous substances (Mueller, Cerniglia, & Pritchard, 2009). Several studies demonstrate that leaves and leaf-associated microbes can mitigate air pollutants (De Kempeneer, Sercu, Vanbrabant, Van Langenhove, & Verstraete, 2004).

1.2.5. Leaves as removal methods of VOCs.

Leaves are photosynthetic organs with an upper and lower surface (Kirkwood, 1999). It is imperative to consider the parts of the leaves due to the function they perform. The upper surface usually has a waxy cover, commonly known as the cuticle. Its principal function is to acts as a barrier to prevent the penetration of xenobiotics and, to reduce the evaporation of water. The leaf has different morphologies because it is made up of trichrome, which are outgrowths at different levels of the surface (Tian et al., 2017). These outgrowths play roles in their physical properties and mechanical defense. The vascularity of the leaves is led by Xylem and phloem, which transports water and nutrients from the roots to shoots and vice versa (Wei et al., 2017).

Additionally, under the coticule layer, there is the presence of epidermal cells where stomata usually occur. A layer of mesophyll cells is directly underneath the epidermis, which is subdivided into two layers: column-like palisade cells and loosely packed spongy cells. The spaces between spongy cells provide gas exchange and photosynthesis. On the lower surfaces, there is an epidermal layer where major stomata are located. Around the stomata and stomatal pore, two guard cells are regulating the opening and closure of the pore by the turgor pressure. Stomata is uncharged to regulate the gas flow to absorb or adsorb other chemicals. Both cuticle and stomata act as pathways to remove VOCs (Dela Cruz, Christensen, Thomsen, & Müller, 2014). The ability of plants to remove VOCs is determined by the capacity of plant cells to metabolize the pollutants while maintaining their regular metabolic process (Kim et al., 2018). The main entrance of air pollutants is through the leaves when the stomata are open or the diffusion on the epidermis, which is covered by the waxy cuticle.

Furthermore, an alternative entry is a cuticle, which depends on the hydrophilicity or lipophilicity of the molecules (Kvesitadze, George; Khatisashvili, Gia; Tinatin, Ramsden; Sadunishvili, 2006). The ability to capture pollutants is related to the chemistry of the wax, but the amount of wax is not. (Treesubsuntorn, Suksabye, Weangjun, Pawana, & Thiravetyan, 2013).Once the pollutants enter the plants, they reach the sieve tubes of the phloem, and there is translocation to the rest of the tissue (Kvesitadze, George; Khatisashvili, Gia; Tinatin, Ramsden; Sadunishvili, 2006). This procedure would depend on the concentration of VOCs. Then, the VOCs may be degraded in situ or transported to other sites of the plant, where maybe degraded or metabolize (Giese, Bauer-Doranth, Langebartels, & Sandermann Jnr, 1994; Schmitz, Hilgers, & Weidner, 2000).

Plants are toxic resistant because of the ability to excrete, conjugate, or degradation of toxic substances into cellular metabolites or smaller molecules, for example, carbon dioxide. (Kvesitadze, George; Khatisashvili, Gia; Tinatin, Ramsden; Sadunishvili, 2006).

2. Problem statement

Air pollution is a mix of solid and gas particles in the air. Some of them are toxic and inhale them increase the chance to produce side effects on human health (EPA, 2017). Air pollutants are considered as the leading environmental toxins related to diseases responsible for 16% of premature deaths all over the world (Landrigan et al., 2018). Previous studies on Cancer showed that around 223000 deaths due to air pollution-resultant lung cancer all over the world, and air pollution is known as the most widespread environmental carcinogen (Circ et al., 2010). This information was two times more than previous estimations and confirmed that air pollution had become a real human health risk (WHO, 2008). Specifically, Volatile organic compound (VOCs) in the air has been a global problem. According to the US Environmental Protection Agency, air quality guidelines sated the mean limits for annual exposure to hydrocarbon concentrations content is 1.6x10-4 km/m3 per year, and many cities of the world are exceeding the maximum permissible limits.

Scientists, researches, regulators, air quality managers, governmental and non-governmental organizations have been collected several data that provides information on the current state of the effects of VOCs in environmental air pollution and the results are impressive and alarming (Ogunbayo, 2016; Samet & Krewski, 2007). These are taking into consideration for the designing of VOC control and elimination procedures. However, most of the remove methodologies are chemical and physical, and their production cost and effectiveness response to other new problems such as the production of hazardous waste. Despite the limitation and challenges, scientists continue on research and development of modern, eco-friendly treatments of VOC mitigation (Berenjian et al., 2012).

Currently, in Ecuador, environmental management is inefficient due to there is unreliable. An analysis made by the Ministry of Environment states that urban populations with more than 100000 inhabitants, and according to their population density and economic activities, are at risk of having a low quality of air (Páez, 2012). There are just three cities that provide information about the air quality Quito, and recently Guayaquil and Cuenca joined with the implementation of this procedure. It should be stressed that the current Ecuadorian national constitution provides us to live in a clean and ecologically balanced, which promotes and guarantees sustainability and good living, furthermore, promoting the conservation of the ecosystem, biodiversity and the integrity of the country`s genetic heritage (Constitución de la Republica del Ecuador, 2015). Despite having a law mandating, the government has environmental legislation, and there is no bureaucratic procedure for control of air pollution. The only information about quality air management is provided by CORPAIRE, which is the only entity, located in Quito-Ecuador that keeps a systematized, verified and controlled information about vehicle emission and air quality (MAE, 2003). According to the data obtained, the main problem of Quito`s air is the presence of fine particle material (Group where VOCs belong), despite not reaching the maximum emission limits; however, it can produce long term diseases; specifically, people who are regularly exposed to urban areas. According to INEC, in 2016, there were 2643 deaths due to diseases associated with environmental pollution (INEC, 2006).

However, there is no company or governmental association responsible for promoting the mitigation of air pollution whereby there is an open gap of study and research about new eco-friendly methodologies to remove VOCs from the air. To take advantage of Ecuador's biodiversity, plant leaves from different areas of Ecuador may be tested to prove their efficiency to capture VOCs as a solution to this environmental problem.

3. Hypothesis, general, and specific objectives.

3.1. Hypothesis.

Leaves of Ecuadorian native plants may be a potential reducing agent of VOCs with physical and chemical properties involved in the mitigation of VOCs.

3.2. Objectives.

3.2.1. General Objective.

- To analyze the gas absorption capacity of some Ecuadorian native leaves to reduce VOCs.

3.2.2. Specific Objectives.

- To characterize the leaves, using physical-chemical methods to obtain its properties.
- To identify the potential leaves for reducing VOCs.

4. Materials and Methods.

In this project, the main procedure has two phases: gas absorption assays and characterization part. Twelve leaves plant species were used in these experiments and obtained from different Ecuadorian markets and commercial distribution. These plants species have particularities, and most of them come from the highland region of Ecuador. The leaves were acclimatized to normal-environment conditions before they were used for the experiments. Unfortunately, the plants are not mentioned because they are still analyzed and being studied.

4.1. Gas Chromatography Assays.

Gas chromatography assay is a method to know a compound that may be vaporized without its decomposition. In these cases, the Shimadzu GC-2014 Gas Chromatograph equipped with a Shimadzu AOC-20i Auto-Injector, a Flame Ionization Detector (FID) and a 30 m x 0.25 mm x 0.25 µm Zebron ZB-WAX Plus capillary GC column was used. Once the leaves were selected, 10 mg of the leaf was taken and put into an Agilent Technologies Gas Chromatography vials with septum crimp-caps. The hexanal supplied by Sigma-Aldrich acted as blank. In this experiment, 1ul of hexanal was injected in the vial, and the sample was conducted to isothermal conditions(heated at 25 ºC for 30 minutes) in an atmospheric pressure using a temperature-controlled bath. After reaching the isothermal conditions, the vials were analyzed in the CG, and the experiment was repeated three times.

Similarly, the vials mounted with the leaves were injected with 1ul of hexanal and then mounted on CG to be analyzed. This procedure was done three times for each plant. The CG procedure can measure the absorbed and non-absorbed VOCs on the samples in real-time, and comparative studies were conducted based on the capacity of absorption of the hexanal in different leaves plants. The GC device shows a diagram on the computer screen, which allowed to analyze the data.

4.2. Characterization Procedure

4.2.1. Leaves Preparation

In this experimental phase, just two leaves were examined, one that has significant gas capturing efficiency (J1 leaf) and another one that does not (B2 leaf). First of all, the used leaves had an average size of 1cm of length and 0.5cm of width and were clean up with distilled water and dried. In the middle of the leaf, a mark was made to divide two parts: Exposed and not exposed to the hexanal. Then, half of the leaf was covered with cardboard to seal the non-exposed part.

On the other hand, a beaker with 2ml of hexanal was filled. Then, the leaf was exposed to the hexanal for 45 minutes, maintaining half of the leaf airtight (Figure 2). After this preparation, the leaves were ready for the characterization part.

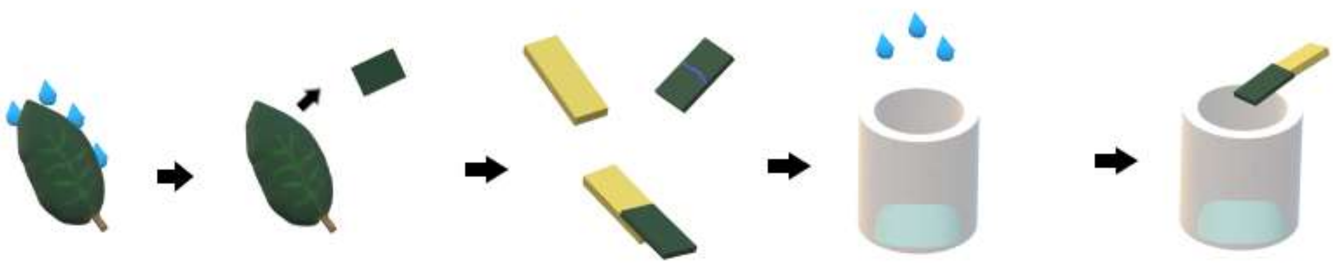

Figure 2. Preparation of the leaf for FTIR and SEM characterization procedure.

4.2.2. SEM and EDS

The scanning electron microscope (SEM) provides information about the surface and chemical composition, which allows us to compare the characteristics of the different samples. In this study, a MIRA 3 (TESCAN, CZ) field emission scanning electron microscopy (FEG- SEM) scanned the surface of the leaves. For this procedure, two images were taken, one with low magnification (x100) and another with high magnification (x500) were performed. Additionally, the EDS was performed with the same SEM. EDS is a technique capable of giving an essential quantitative analysis on the sample to determine composition gradient measurement or just the chemical composition of the sample

(Mohammed & Abdullah, 2019). The selected elements for the EDS procedure were Carbon and Oxygen. Three different areas of the leaf surface were selected to perform the elemental mapping. In each area, ten different points were analyzed, which means a total of thirty points were used for the experiments. To compare the performance, three areas from each side (the non-exposed and the exposed part), were analyzed. A T- student test analyzed the results with a significance level of $p < 0.05$.

4.2.3. FTIR

Fourier transform infrared spectroscopy (FTIR) is a technique used to get an infrared spectrum of absorption, emission, and photoconductivity. It collects spectral data in a range. The measurements of the FTIR are conferred to intensity over a narrow range of wavelengths at a time (Sharp, Wong, & Johnston, 2018). In this case, a Spectrum Spotlight 200 (Perkin Elmer, MA) did its performance, and a spectrum of the hexanal is acquired as background and then the spectra of the samples, the exposed part, and the non-exposed part. The wavelength range for the analysis were between 4000 to 400 cm-1 with a total number of scans of 36 and a 4 cm-1 wavelength resolution.

5. Results.

5.1. Gas Chromatography Assays.

First, we investigated the gas capture efficiency using different types of leaves. These experiments used an aldehyde VOC, hexanal — Figure 3. After 30 minutes of exposure of the native Ecuadorian leaves to the hexanal, some results of the performance of gas capturing are analyzed. In Figure 4, we separated the leaves that reach an inefficient performance, it means with retention of less than 15%. J2, J4, , B2, B4,B7, B8 are part of this selection, specifically with an absorption of 3.03 ± 4.64 % ,3.41 ± 1.62%, 0.8 ± 7.56%, 5.65 ±5.74%, 1.01 ±0.01%, 8.16 ± 16.45% respectively. On the other hand, the group of the effective leaves, J5, J1, B5, B6, B1, J7, Figure 5. with an achievement of more than 15% of hexanal remaining 20.50 ± 18.88%, 28.06 ± 3.78%, 27.19 ± 15.93%, 21.06 ± 8.27%, 20.6 ± 18.17% and 18.77 ± 9.48% respectively. Finally, in Figure 6, the leaves B2 and J1 were selected to be characterized because they showed the highest and the lowest performance.

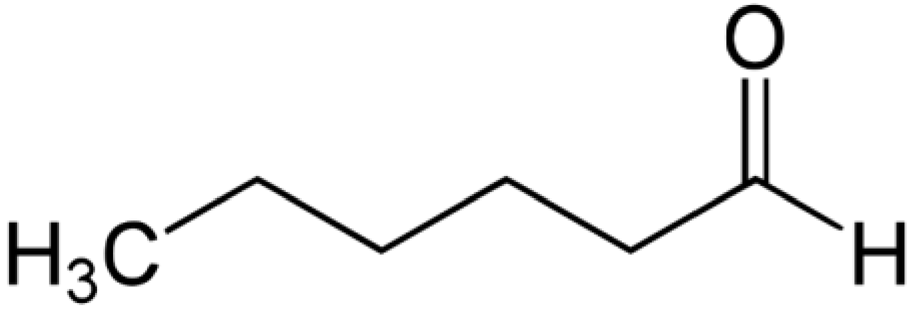

Figure 3. Structure of the hexanal (25ºC).

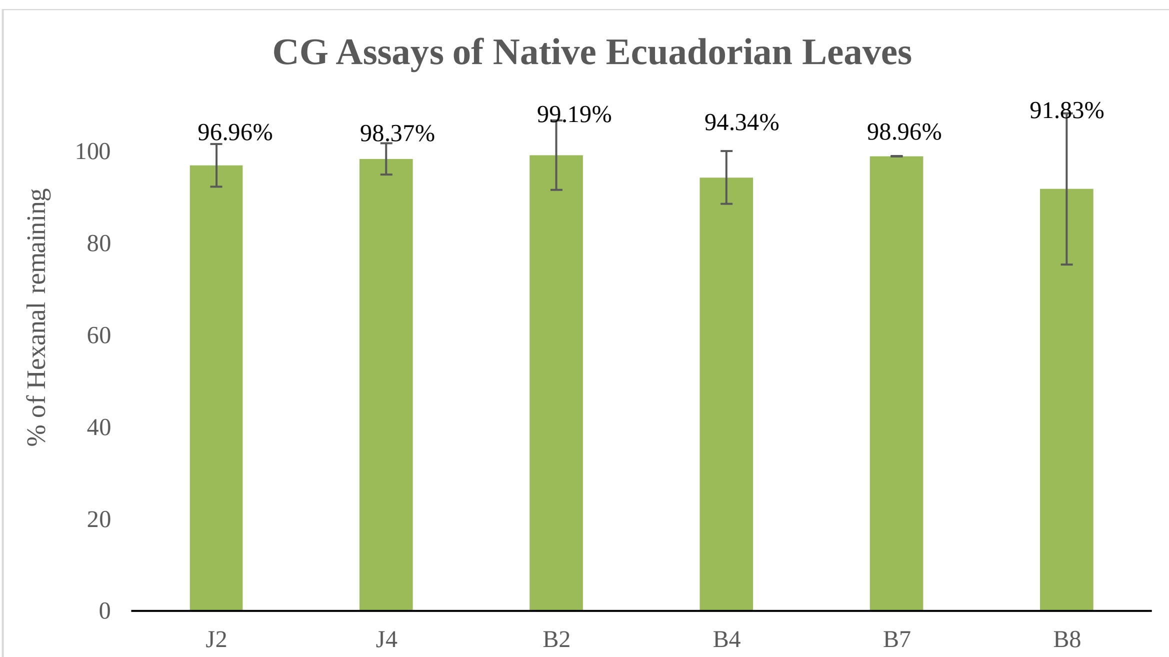

Figure 4.Results of GC analysis obtained from the less efficient leaves.

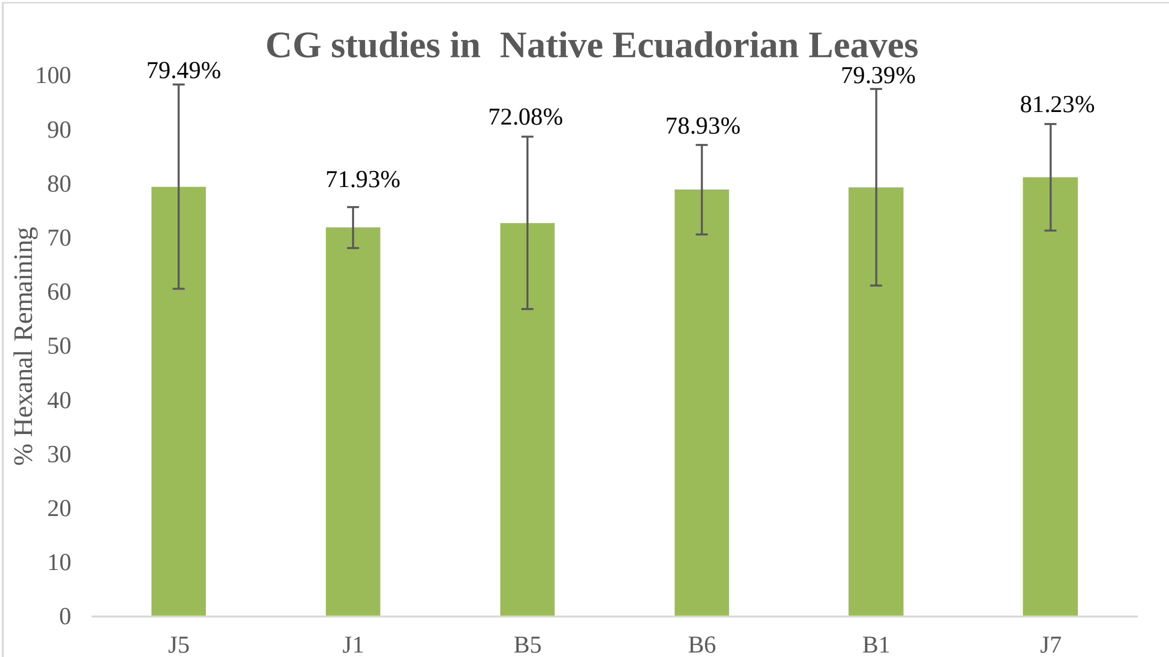

Figure 5.Results of GC analysis obtained from the more efficient leaves.

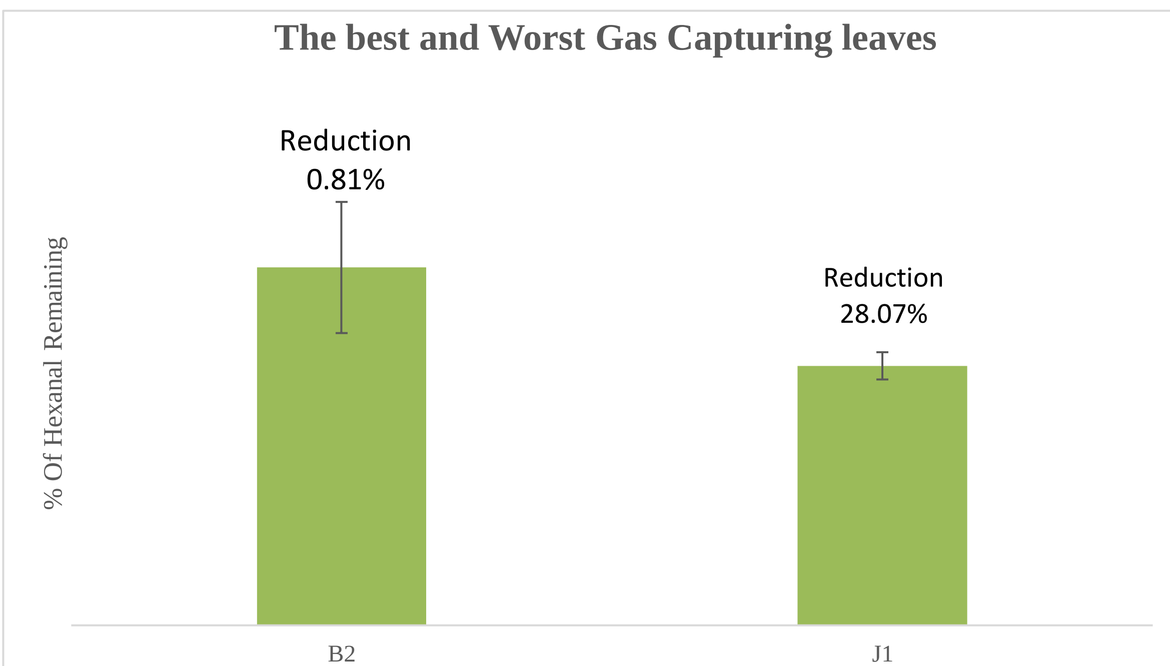

Figure 6. Results of GC analysis obtained from the more and the less efficient leaves.

5.2. Characterization Results

5.2.1. SEM

After thorough, the gas absorption assays, we set out to evaluate the chemical and physical properties with a characterization process. First, we evaluated the morphology and structures on the surface of leaves, using the scanning electron microscope (SEM). In Figure 7, in the J1 leaf, there is the presence of different structures such as peaks and stomata (pores that intake and output gases), triggering in an irregular raised into roughened ridges and fibrous surface. In the case of the B2 leaf (Figure 8), the morphology of leaves showed a hairy surface, which means an asymmetrical because of the presence of hair filaments all over the outer face.

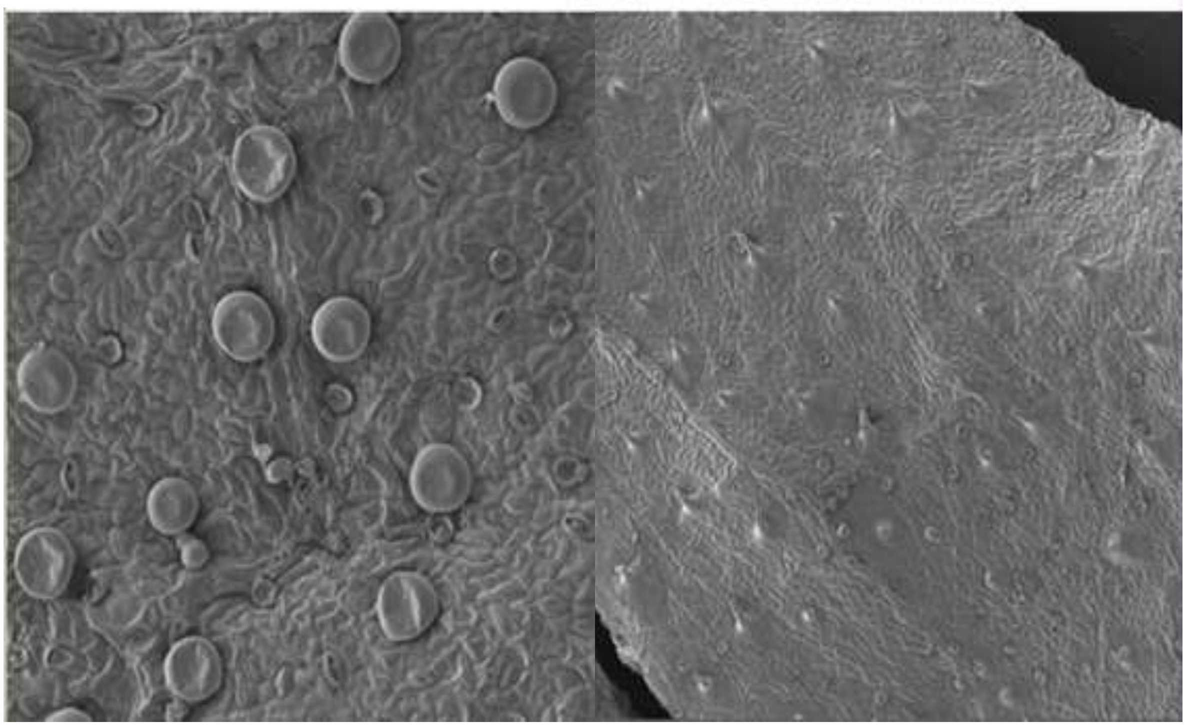

Figure 7.SEM image of the J1 leaf.

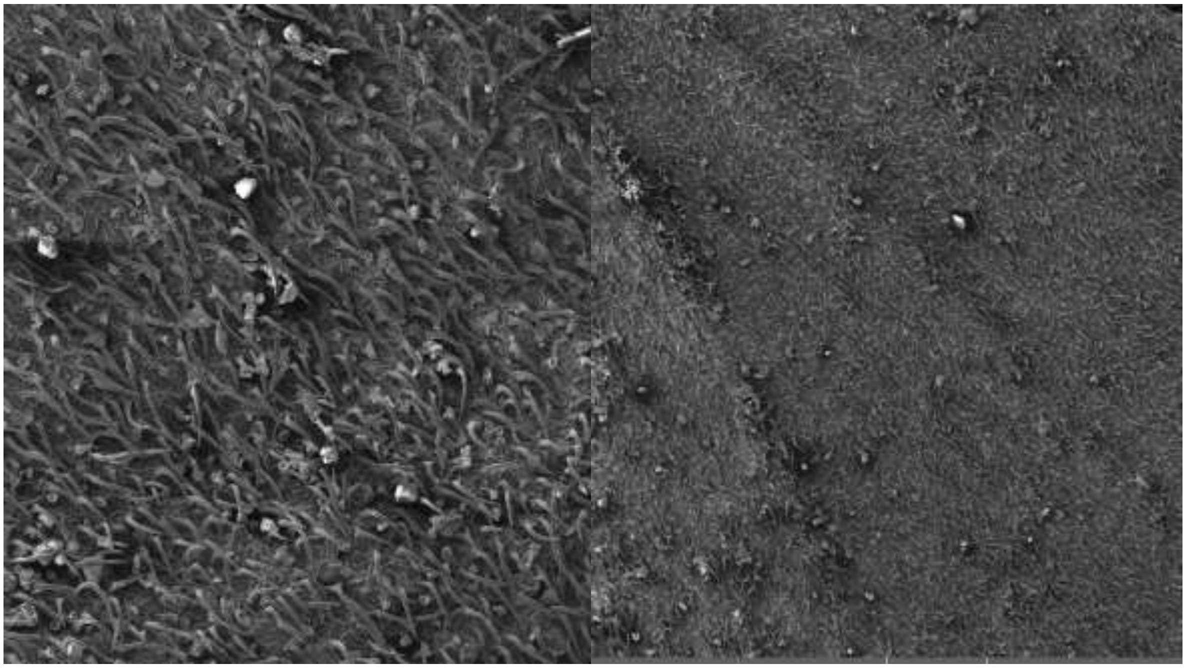

Figure 8. SEM image of the surface of the B2 leaf.

5.2.2. FTIR

FTIR is a procedure that describes and analyzes the chemical structure of an exanimated sample; in this case, the leaves. Plant tissues are composed of different substances and give a complex spectrum with several vibrational brands. As mentioned in the previous section, the treated leaves were the B2 and J1. In Figures 9 and 10, the spectrum of the B2 leaf is shown, and the major classes of compounds can be recognized. In general, according to the literature, the following absorption bands can be distinguished: a region between 3500–3000 cm−1, which is dominated by the O–H and N–H stretching vibrations; CH_3 and CH_2 stretching vibrations that appear at 3000~2800 cm−1; a region between 1800–1200 cm−1, which is characterized by C=O stretching vibration (1738 cm−1)indicating ester-containing compounds, amide I (1656 cm−1) and amide II (1563 shifted to 1559 cm−1) in proteins; at 1513 cm−1vibrations of aromatic ring; CH_3 and CH_2 bending motion at 1460 and 1400 cm−1; a region within 1235–1153 cm−1, which is due to the C–O stretching in ester and amide III; and in the "fingerprint" region between 1100–1000 cm−1, there are several vibrations of groups such as C–H bending or C–O or C–C stretching, which are characteristic of cellulose in the leaves (J. Yang & Yen, 2002).

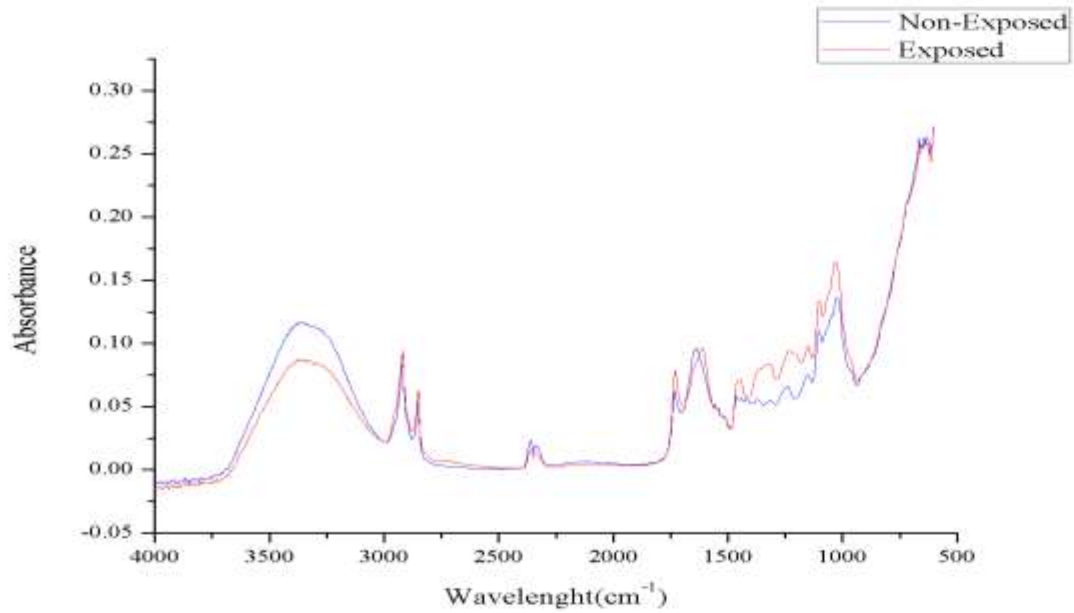

Figure 9. FTIR spectrum of the non-exposed and exposed B2 leaf.

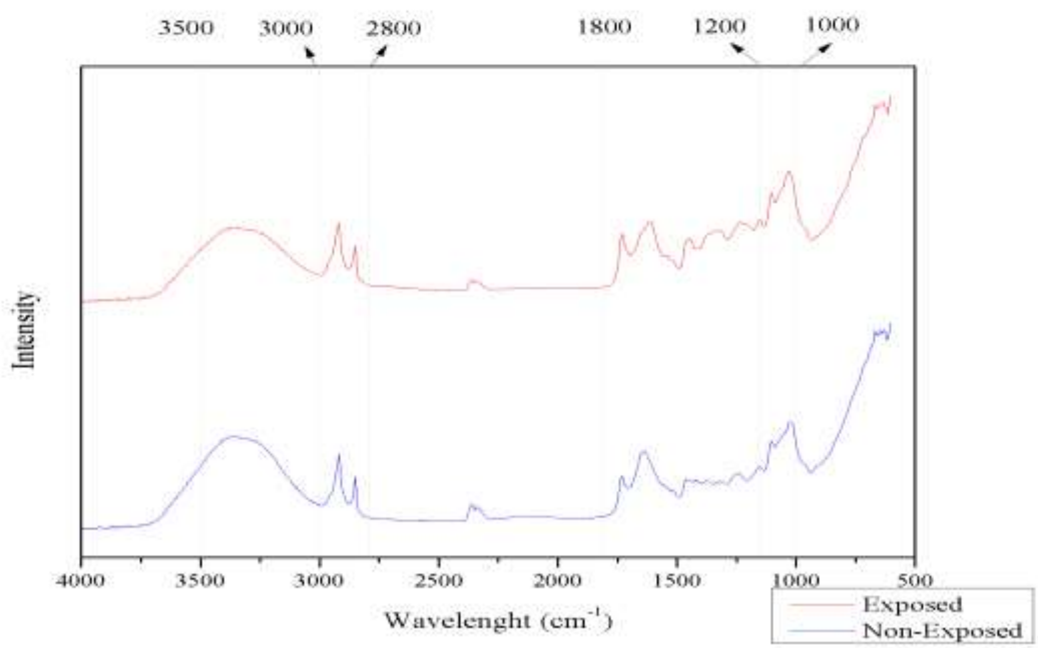

Figure 10. FTIR spectrum of the non-exposed and exposed B2 leaf.

In the second experiment on J1 leaf, the spectrum follows the previous measuring, corresponding to the spectrum of a regular leaf. Figures 11 and 12, as mentioned before.

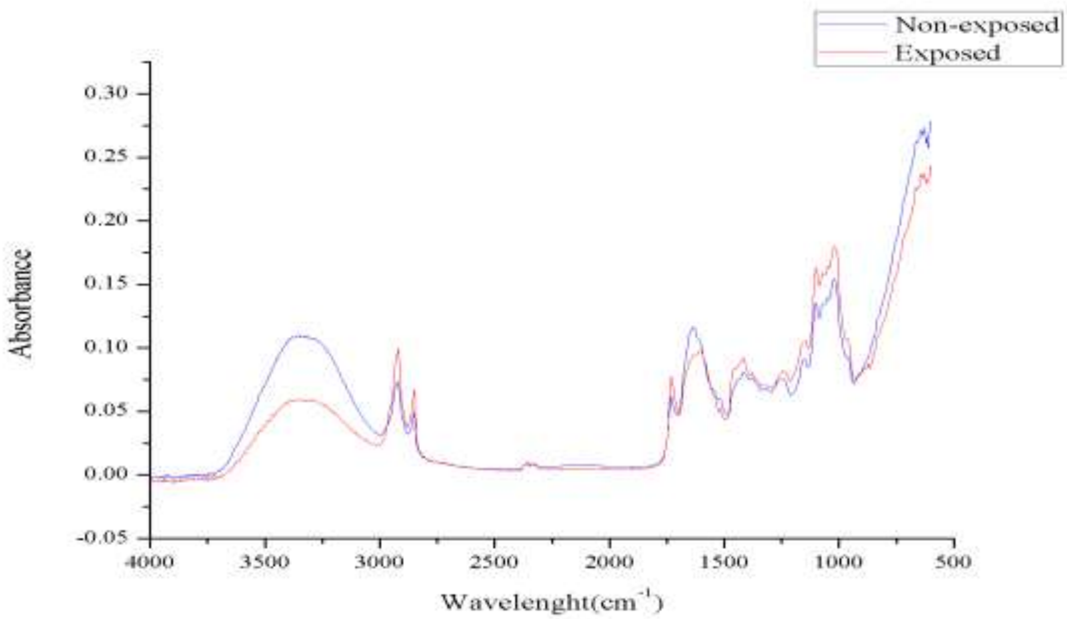

Figure 11.FTIR spectrum of the non-exposed and exposed J1 leaf

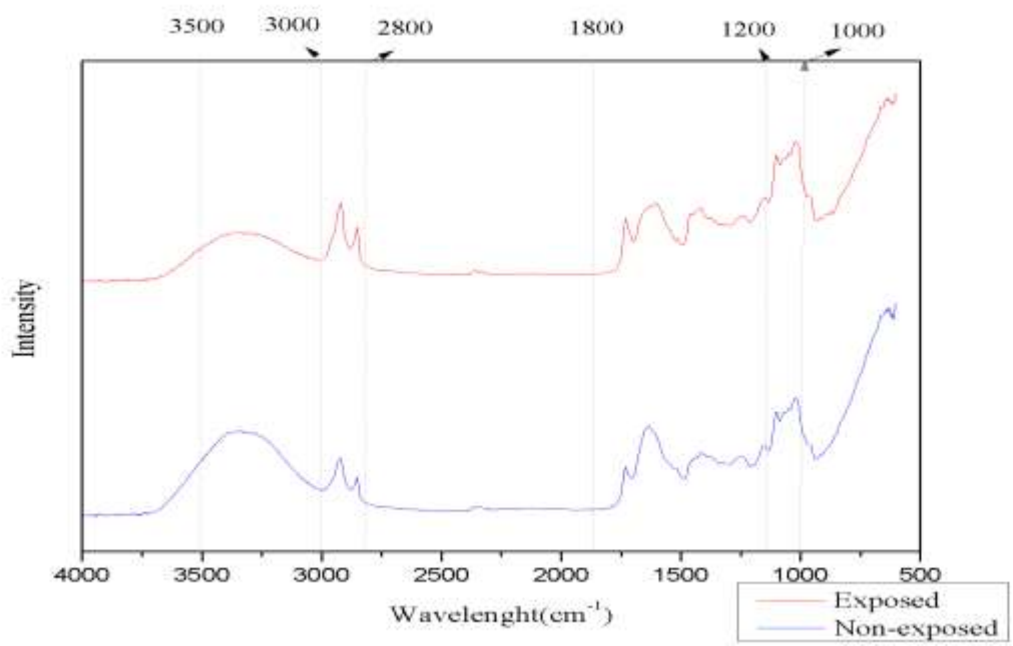

Figure 12. FTIR spectrum of the non-exposed and exposed J1 leaf

5.2.3. EDS.

Finally, this experiment allows us to get a qualitative amount of some specific elements found on the surface of the sample. During this analysis, the Carbon and the Oxygen were the selected elements to be compared. The results obtained for B2 leaf, Figure 14, showed a 51.30 ± 0.39 and 52.29 ± 0.31 % of Carbon on the exposed and non-exposed parts respectively, meanwhile, on J1 leaf on the exposed part, there is a 40.12 ± 0.96% of Carbon on the exposed part and 35.91 ± 1.18% on the non-exposed. In terms of the amount of oxygen, on B2, there is 41.32 0.31% in the treated part and 40.26 ± 0.36% on the non-exposed part, nonce, in J1 46.38 ± 0.54% in exposed part and 51.00 ±0.5% on the non- exposed part (Figure 13). According to the T-student result, in the case of J1 leaf, the significant for Carbon was 0.001, meanwhile, for Oxygen was 2.968E-07. For B2 leaf, the significant p were 0.03 and 0.01 for Carbon and Oxygen, respectively.

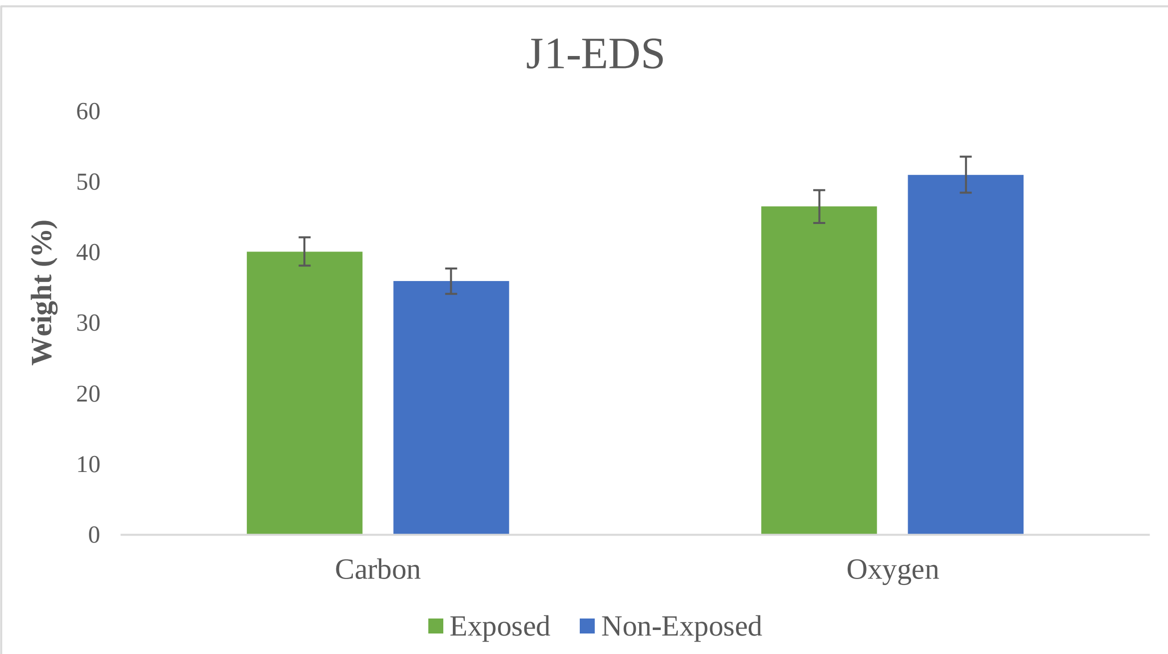

Figure 13. Carbon and Oxygen Weight obtained during EDS data Analysis in J1 Leaf.

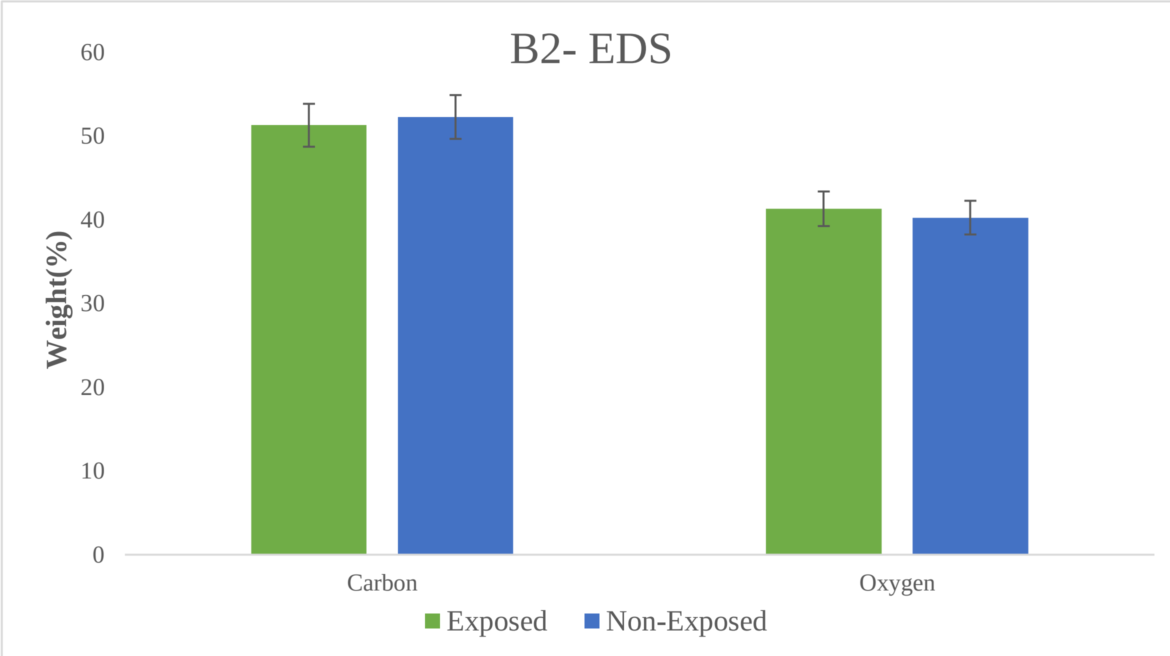

Figure 14. Carbon and Oxygen Weight obtained during EDS data Analysis in B2 Leaf.

6. Discussion

The use of leaves is a promising methodology for the mitigation of VOCs. It captures or adsorbs toxic substances through the surface of the leaf to degrade into non-toxic or less toxic compounds (Ma, Oliveira, Freitas, & Zhang, 2016). The opportunity to use leaves as a solution is due to a large amount of plant area on the surface all over the world, which is up to 32 percent of Earth`s total surface area (NASA’s Earth Science News Team, 2016). There are several studies of plants about the potential to absorb/adsorb various air pollutants due to the advantage of having specific characteristics such as large and biologically active surface areas or their chemistry. (Agarwal, Sarkar, Chakraborty, & Banerjee, 2019). As mentioned before, 12 unmodified leaves from different parts of Ecuador, were selected to be studied their ability to absorb VOC. After gas chromatography assays, there are considerable differences between the plants that work and do not. Among the more efficient leaves with 18.77 ± 9.48% and 28.06 ± 3.78% of absorption and less efficient leaves did not reach expected results where their performance was around 0.8 ± 7.56% and 8.16 ± 16.45% of retention of the solvent. Yang and collaborators have been studied the behavior of different VOCs and plants, and the applied protocol differs from what we have done, which does not allow us to compare with their results. However, this study shows other characteristics that might help to distinguish between the plants that capture more VOCs and the regular ones. For example, characteristics related to the nature of the individual compounds (leaves and solvent) such as polarity, molecular weight, and solubility are taking into consideration. In this case, lipophilic solvents can penetrate the cuticular surface of the plant due to a regular plant has a lipophilic surface (Yang at al, 2009). During experimentation, hexanal was the used solvent; in consequence of its lipophilicity and low-molecular-weight, it could do a better interaction between compounds. Therefore, it may infer that this type of solvent is related to the leaves' capturing performance.

Additionally, other plant characteristics related to mitigation performance are morphology and structure information. For example, in J1 leaf, the presence of stomata, pointy protrusions (epicuticular trichomes), dense grooves, and, in comparison of B2 leaf with flattened hairy outgrows. According to Chen and collaborators, leaves with the presence of different pointy protrusive structures have the highest rates of VOC capture rather than leaves with flattened protrusions. Additionally, this study shows, there is no relationship or a correlation between density or diameter of stomata on the leave`s surface. However, the presence of the grooves on the surface indicates a positive particle retaining. (Chen, Liu, Zhang, Zou, & Zhang, 2017). Regardless of the results, these characteristics are shown in our leaves and could help to have a better performance in the mitigation of the VOCs.

According to FTIR results, the spectra obtained follow a regular leaf spectrum as Yang and collaborators previously determined. It indicates characteristic peaks for a leave. On the FTIR graphs of the J1 and B2 leaves, the peaks displayed are the same. .However, as a consequence of the chemical composition of the hexanal, which is a carbon-based molecule, it is difficult to recognize the vibrations of new molecular binding or recognize a particular peak formation.

Following EDS analysis, expected results were obtained because the Carbon amount on the J1 sample that was exposed to the hexanal increased 4.22% of the weight in comparison to the non-exposed part. This result might be a consequence of the retention of hexanal on the surface of the leaf. In the case of B2 leaf, the percentage of weights is quite similar where the amount of carbon is almost the same in both parts, the exposed and non-exposed. The carbon amount is an expected value due to this is the leaf with low performance of absorption. According to the statical analysis, in J1 leaf, the differences between the amount of Carbon and Oxygen has an excellent level of significance because the significant p is less than the p-value ($p>0.05$). When there is a minimal p-value, it is evidence that the means are heterogeneous and statistically different. At the same time, B2 leaf reaches a reasonable level of significance, in both Oxygen and Carbon, which means there are small differences between the means; however, the means are statistically similar. These results reconfirm the GC analysis because J1 leaves show an increment of Carbon coming from the hexanal production, while, in B2 leave, there is no difference between the exposure a non-exposed part.

7. Conclusions

-Some Native Ecuadorian leaves have a strong potential to mitigate the VOCs in air pollution. Leaves reached a performance of around 15% and 28% of retention.

- The Carbon`s amount on the efficient leaf increased by around 4.22%, which means retention of the VOC on the surface of the leaf.

Additionally, the p-value of the non- exposed and exposed part in the J1 leaf is statistically significant; therefore, the mean is entirely different, showing significant retention of VOC on the leaf`s surface.

- The surface of the leaves plays an essential role in the retention of the leave. According to the experimentations, leaves with peaks and grooves have a better performance.

- These results can be used for future work, and future technologies and new experimental approaches will ensure that VOC mitigation becomes even more accurate and efficient.

8. Recommendations for Future research.

According to the obtained results, research is viable about the capacity of mitigation of VOCs in different plants. Furthermore, with the availability of a large variety of plants in Ecuador is possible to test the capacity of retention of air pollutants in other types of plants and also extended research in other VOCs and air pollutants. It is essential to consider that the amount of these plants is significant and readily available in the markets. The projection of this research could be the bio-mimicking of the surface of leaves, maintaining their physical and chemical properties, in order to avoid the use of raw materials, therefore, promoting the production in large scales of this type of procedure.

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