Extended Essay.

Biology HL.

Candidate number: 04971-0010.

Topic: Comparison of the process micropropagation on the Ecuadorian endemic plant Chamburo (*Vasconcellea pubescens)* and the agricultural method of Babaco (*Carica pentagona)*.

Research question: To what extent does the application of auxins and gibberellins affects the micropropagation of *Vasconcellea pubescens* compared to the agricultural growth of *Carica pentagona*?

Number of words: 3 989 words.

Acknowledgment:

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1. Introduction:

Biodiversity is the variety of animals, plants, bacteria, and fungi around the world, the diversity of life. According to The Biodiversity Finance Initiative (BIOFIN), biodiversity in a country is determined by its geographical, atmospheric, and climatic location and conditions and represents an important economic and commercial source, especially for the communities living and relying on this mean. Activities such as oil extraction and contamination endanger the biodiversity of a country, in this case, Ecuador, consider the 17th most biodiverse countries in the world with 6,1% of all species reported worldwide.

It is estimated that around 25% of the world's biological diversity is found in the Andean region; the countries that comprise this region are considered the most diverse and rich in animal and plant species in the world (Mittermeir et al. 1997, Myers et al.2000). The investigation will be a focus on the specific endemic plant, *Vasconella pubesens*, it comes from the Caricaceae family, commonly known by Ecuadorian people as *Chamburo.* It is native from the Andes where numerous species could be of use or eatable to the Andean population, species of plants such as papayuelos like Chamburo, babaco, among others. It is now considered an Unconventional Eatable Food Plant or PANC, by its acronym in Spanish. In comparison to the agricultural used and commercialized *Carica pentagona*, it comes from the Caricaceae family too, and its commonly known as Babaco or Passion Fruit, it belongs to the papayuelos that has achieved international market. Ecuador is a country that has different fruits thanks to its geographical position. That is why babaco is endemic from this country and part of Colombia, underlining its importance as part of the traditional and non-traditional exports in the country.

As Ecuador is an extraordinarily rich country in biodiversity, to find a new production alternative in the face of the great challenge of competing in a globalized market and changing the overproduction of a specific specie but inserting variety in the market will also allow decreasing the consequences of overproduction of common fruits like in this case, *Carica pentagona*, consequences such as deforestation and climate change, both. Therefore, biodiversity in Ecuador is a topic that needs urgently to be assessed. The reason why this study will focus on the effect micropropagation on the Ecuadorian endemic plant and in which way it differs from the commonly agricultural germinated plant. Micropropagation allows rapid production of high quality, disease-free, and uniform plants. The micropropagation ornamentals, and forest and fruit trees has created new opportunities in global trading for producers, farmers, and nursery owners, and for rural employment. Germination regulating mechanisms that respond to or "sense" the environment and the predictable cycles of seasonal change may also enhance survival. Through several possible mechanisms, seeds may sense the environmental conditions in soil that favor germination at a time and place when seedling establishment is probable. Seeds may sense their location to the soil surface and other vegetation through the environmental signal of sunlight (Mayer, 1986).

The aim of this study is to compare the regular process of agriculture with the laboratory process of micropropagation in order of give a better understanding on the conservation of the endemic plant when the environmental conditions of soil, sun and nutrients are being threaten by the human activities. With the application of the research question

To what extent does the application of different measurements of auxins and gibberellins hormones affect the micropropagation of *Vasconella pubesens*?

is needed to answer the scientific process of comparison, applying nutrients and hormones, trying to simulate the soil environment.

2. Background Information:

2.1. PANCs (Unconventional Eatable Food Plant)

PANCS is the acronym in Spanish to Non-conventional Eatable Plants, which are every plant that have an eatable potential for human beings such as beverages, dishes, eatable coloring, condiments. This term was created by the Brazilian botanist Valdely Kinuppand the nutritionist Irany Artecheto who, revalue plants that were used by the population ancestors for many years. However progressively, they were falling into oblivion, some PANCs are native from Ecuador and that have been cultivated by traditional populations, *Vasconcellea pubescens* is considered one of those. Even though these are not available in markets. A PANC can be grass, a tree, a liana, a cactus, as well as any part of the plant that has nutritional potential (leaves, stems, flowers, fruits, seeds, roots, even pollen). These can even consider conventional plants like banana that have other parts with a non-conventional use, many of these plants are wild and grew spontaneously on the surroundings, however a great amount of them is also found in gardens of people that maintain diversity inside their crops.

Through the years human nourishment have been based on the consume of a great variety of plants, this diversity used to sustain humankind ang guarantee a better nutrition, health, crops diversity, among other benefits. Despite the fact, that the world does not counts with a completed list of edible pants it is known my many studies that usually the 10% to 25% of all the flora of a region is potentially eatable. Estimating that it exists approximately 80.000 species of plants that contribute to human nourishment in modern time. Nowadays, the simplification of crops has caused enormous consequences, the dependent population of ultra-processed products, great number of monocultures produced, that degrade human health and the environment. (Duarte, N. 2020)

2.1.1. Nutritional value.

A great amount of these plants represents an outstanding nutritional quality and some of them are even considered as “superfoods”. In addition to the nutritional value, a great part of PANCS is of medicinal treatment, considered as nutraceutical plants, in order words these are plants which prevent and cure human diseases. (Duarte, N. 2020)

2.1.1.1. “Superfoods”.

This term has been known since the beginning of the last century, it was used to describe nutrient rich aliment that were specifically beneficial to health and wellbeing. These are a group of nourishment that have a concentrated nutritional value known as: amino acids, enzymes, essential fatty acids, and micronutrients in general. Frequently, superfoods arrive the stores sin seeds, or dehydrated plants because of its small size and so that they can be easily transported. (Herp, 2017, page 10)

2.1.2. Identification of PANC´S.

. To produce and consume PANCs it must be followed selection criteria that Nina Duarte suggests in her guide to non-conventional eatable plants in the Chocó Andean region, which are the following:

Figure 1 Identification of PANCs. (By student, 2021)

2.2. Taxonomy

Babaco is part of the order Brassicals (which also includes Arabidopsis), where the Caricaceae is one of 17 families. The sister family of the Caricaceae is the Moringaceae (Carvalho et al., 2015). It seems that the Caricaceae originated in Africa and later it dispersed to the neotropics. Cylicomorpha parviflora (a large tree from East Africa) and Cylicomorpha solmsii (from West Africa) are the closest relatives to the New World Caricaceae. The other five genera in the family (comprising 34–35 species) are Carica, Horovitzia, Jarilla, Jacaratia and Vasconcellea. Vasconcellea species, of Andean origin and commonly referred to as 'highland papayas', were previously positioned within the genus Carica, and considered as the closest relatives to *C. papaya*; however, the studies

of Badillo (2000), later confirmed with molecular data, showed that Vasconcellea was more related to Jacaratia and identified Horovitzia as sisters of papaya. Pointing to Mexico and Guatemala as a possible domestication original place area. The genus *Vasconcellea was*, consequently, rehabilitated and C. papaya stayed as the sole species in the genus Carica (reviewed by Carvalho and Renner, 2014). Vasconcellea remained then the largest genus in the family Caricaceae. (Mitra. 2020.page 24)

2.3. Vasconcellea pubescens.

Illustration. 1 Vasconcellea Pubescens. (Nina Duarte, 2020)

The Andean species *Vasconella pubesens* is cultivated successfully in-home gardens due to its ornamental qualities (Purdue, 2004). It has an appearance similar to the papaya, it is an easily management bush native form the Andes it counts with large leaves that are distinguished from those of papaya by its shape and by the presence of a pubescence that covers the leaves and flowers. “The fruit is very aromatic, light yellow, oblong-elliptical. truncated at the base and acute at the apex 8 to 11 cm long and 5 to 6 cm in diameter, with five very pronounced ribs” (Garcia, 1992)

Figure 2 Information of Vasconcellea Pubesens. (By student 2021.)

2.4. Carica Pentagona.

Illustration. 2 Carica Pentagona. (Lourdes Sarmiento, 2019)

According to the Fruticulture Program INIAP, Babaco, *Carica pentagona* has been cultivated in Ecuador for many years and is known to be a shrubby, semi-perennial plant. It presents continuous flowers and fruits in different stages of development. In recent years, this fruit tree has aroused expectations among farmers and businessmen to expand the cultivated area, due to growing internal and external demand, and high profitability, so that babaco is an important alternative for exporting the Inter Andean region. Babaco is a natural hybrid of "Chamburo" *Vasconcellea pubescens* L by "toronche" Carica stipulate It belongs to the family Caricaceae the provinces that have the greatest amounts of *Carica pentagona* cultivation are Pichincha, Tungurahua, Azuay, and Loja.

Figure 3 Information of Carica Pentagona. By student 2021.

2.5. Micropropagation.

Plant tissue culture refers to the growing and reproduction of cells, tissues, and organs of plants on defined solid or liquid media under a controlled environment. (Hussain, 2012) This

study will use micropropagation as the method to measure the influence of different hormones in the endemic plant. Micropropagation allows rapid production of high quality, disease-free, and uniform plants. The micropropagation ornamentals, and forest and fruit trees has created new opportunities in global trading for producers, farmers, and nursery owners, and for rural employment. Plants can be multiplied under a controlled environment anywhere irrespective of the season and weather on a year-round basis. “Plant micropropagation is primarily based on the rapid proliferation of tiny stem cuttings, axillary buds, and to a limited extent of somatic embryos, cell clumps in suspension cultures, and bioreactors. The cultured cells and tissue can take several pathways. The pathways that lead to the production of true-to-type plants in large numbers are the preferred ones for commercial multiplication.” (International Atomic Energy Agency, 2004)

2.6. Culture Media

2.6.1. Inorganic Components:

The culture media constitutes of inorganic components that are divided into macronutrients, administrated in great amounts, and micronutrients, which added in less amounts. As follows:

Components	**Amounts (mg L-1)**
NH4NO3	66 000
K NO3	76 000
Mg SO4.7H2O	14 800
KH2 PO4	6 800
Fe SO4.7H2O	1 390

Na2 EDTA.2H2O	1 865
CaCl2.2H2O	15 172
H3BO3	6200
Mn SO4.H2O	16900
Zn SO4.7H2O	8600
Na2 MoO4.2 H2O	240
Cu SO4.5H2O	25
Co Cl2.6H2O	25
KI	83

Table 1 Inorganic compounds measures. (By ECURED.)

2.6.2. Organic components.

The culture media in vitro also counts with organic components among them are found carbohydrates, vitamins, amino acids, natural extracts, and plant growth regulators (hormones).

1. Carbohydrates: Different organs and tissues are heterotrophs with respect to carbons, due to the chlorophyll insufficiency, it is important to add sugars in the culture media, as an energy fount and osmotic regulators.
2. Vitamins: Thiamin (B1), is added as thiamin hydrochloride and is fundamental for cell development. Nicotinic acid is part of the coenzymes NAD and NADP, it has a synergic effect with the production of roots. Pyridoxine (B6) is added as Pyridoxine-HCL and participates as a coenzyme

to favor root formation. Myo-inositol is properly a sugar-alcohol and influences tissue proliferation and the activation of organogenesis. Ascorbic acid and citric acid are added as antioxidants to prevent the darkening of certain tissues.

3. Amino acids and natural extracts: They favor the proliferation of callus and organogenesis. Between them are found, L-arginine, L-cysteine, among others.
4. Gelling agents: Agar is the most common support material used.
5. Water: The quality of the water used plays an important role thus it must be distilled water (between 0.5 to 2 mS/cm).

5.1. Environmental Conditions.

The environment must be kept clean with work surface disinfected sodium hypochlorite 10% before and after use. Should be used lab coat and wash hands thoroughly. The last phase of a chain of Micropropagation consists of acclimatization to environmental conditions of plants obtained in vitro. Micro plants rooted in vitro are seeded in a substrate moistened with peat / vermiculite (1: 1) and placed in an acclimatization chamber with a humidity 90% relative. For two weeks the relative humidity decreases until it reaches the values of the outside environment.

By tender shoots

This form of multiplication is carried out in the greenhouse, or in rooting beds protected with semitransparent plastic. The material to be propagate consists of shoots of 10 cm in length and 1.5 to 2.5 cm in diameter that are obtained from growing and producing plants. Before rooting,

the shoots must be cut off the top, allowing a vigorous bud sprouting. It is important to disinfect the substrate with Ba vistin 200 c + Kocide in doses of 200 g in 200 liters of water, in order to prevent rottenness. Rooting is carried out under high relative humidity (90%) and 22 ° C temperature. At 45 days, the shoots will be rooted and ready to be transplanted into plastic foundations containing sterilized or disinfected soil.

5.2. Hormones

Micropropagation is a process in which growth promoting substances are used, in this case it will be used auxins and gibberellins. The ability of microorganism to produce various stimulating plant growth and favorable health is considered to be one of the most important factors in soil fertility (Krasilnikov,1958). This concept was known due to the positive results that plants treated with organic materials presented and later Krasilnikov carried on a work experimentation in Russia using humus, peat, compost, bacterial organic materials in order to improve plant growth and yields.

5.2.1. Auxins

The term *auximone* was used for substances present in humus and fertilizers, it comes from the Greek term *auxein* which significance is to increase. Auxins are involved in a variety of plant growth and developmental responses, reason why several synthetic auxins are used in commercial applications. Indole-3-butyric acid (IBA) is one of the principals used in commercialization to initiate rooting. The auxin to be used will be (AIA) Indoleacetic acid.

5.2.2. Gibberellins

This term was discovered in the Western world, due to a disease of rice plants had been studied in Japan, the result pf an infection caused by the fungus *Giberella*. The active chemical that causes excessive elongation of the rice plant, stimulating rice growth. It is now commonly used gibberellic acid (GA-3), in which positive results are the outstanding results, however some negative results can be determined by the fact that this type of gibberellin is not the endogenous of a particular plant. Evidenced by Wittwer and Bukovac (1962), who used nine different gibberellins to test its reactions in several plants, also discovering that gibberellins could substitute for cold requirement in some plant species.

6. Hypothesis

6.1. Zero Hypothesis:

Auxins and gibberellins are plant growth hormones, and its measures will be optimum therefore Vasconella Pubesens will show a more rapid growth than Carica Pentagona.

Micropropagation is the most efficient method of plant growth in comparison with agricultural common method.

6.2. Alternative Hypothesis:

Auxins and gibberellins measures are not optimum therefore the difference between the period of growth of Vasconella Pubesens will not differ from Carica Pentagona.

Both of the methods, micropropagation and agricultural, were the most effective method of plant growth.

7. Variables:

Types of Variables	Variables in the experiment	How to manage variables.
Dependent	Vasconella Pubesens/Carica Pentagona.	The growth process, leaves and stem growth of both plant species though the process of observation and data collection.
Independent	Auxins & Gibberellins measures.	Constant measurement of the number of hormones applied to the culture media and soil.
Controlled	Temperature, light, pH, humidity, photoperiod.	Both plants will be exposed to the same temperature, amount of light, level of humidity, pH, and photoperiod.

Table 2 Variables table. (By student, 2021)

8. Methodology:

For the purpose of this essay the methodology chosen to recognize, state and determine the hypothesis was the scientific method. In order to prove the hypothesis planted the agricultural method of micropropagation was chosen along with the naturally agricultural growth.

8.1. Materials:

- ✔ Plants in growing state. (Vasconcellea Pubescens & Carica Pentagona)

✔ 20 Test tubes
✔ 2 Test tube rack
✔ 1 250 ml Erlenmeyer flask ✔ 1 500 ML ERLENMEYER flask
✔ 1 10 ml Pipette
✔ 1 15 blade scalpel
✔ 1 roll of Aluminium paper
✔ 1 gallon of Stilled water
✔ 1 gallon of Sodium hypoclorite 10%
✔ 1 Balance
✔ 1 Burner
✔ 1 Pressure cooker
✔ 5 Filter paper
✔ 1 Spatula
✔ 1 Termometer
✔ 1,2 mg Murashigue & Scog Culture Media M519
✔ 1 Candle
✔ 26 ml of Afronaturaliza Vitamins & hormones mixture

✔ 1 30 W UV lamp
✔ 10,8 mg of Tryptucase Soy Agar

Table 3 Materials Table. (By student, 2021.)

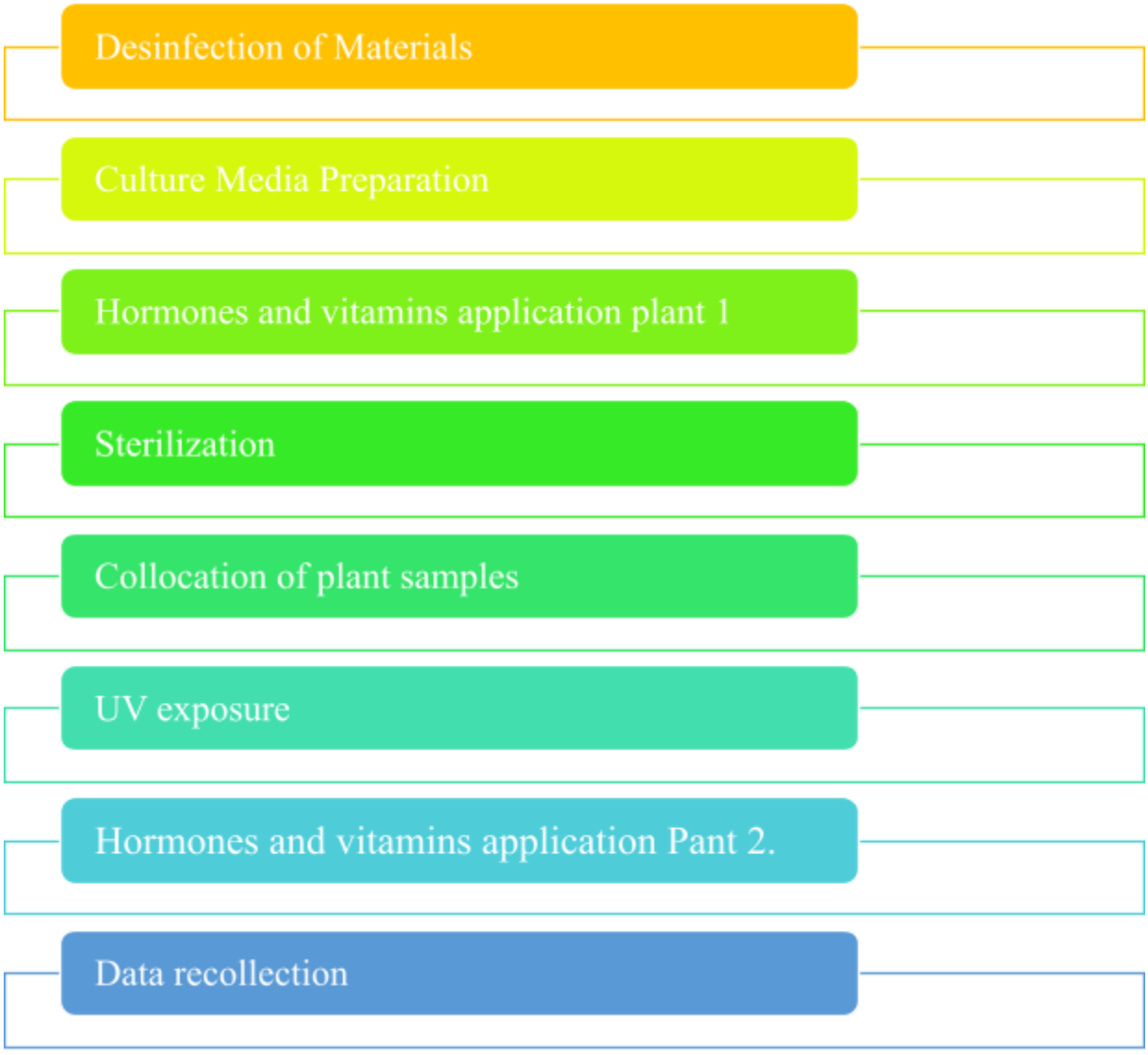

Figure 4 Protocol Procedure. (By Student, 2021)

8.2. Procedure:

8.2.1. Disinfection of Materials.

First the materials needed from the laboratory such as the 20 test tubes were washed and disinfected from previous experimentations with 30 ml of sodium hypochlorite 10% diluted in 200 ml of stilled water. Then, the test tubes were covered with paper and aluminum paper in order to prevent from later contamination.

8.2.2. Culture Media Preparation

To prepare the Murashigue and Scog culture media it was calculated that the amount required was 1,2 mg diluted in 270ml of stilled water. Then, it was measured the exact amount of culture media in the balance and mixed using the spatula. Collocated in the burner and let it until boiled. Then, the process was repeated adding the 10,8 mg of Trypticase Soy Agar needed for the experiment. Inside of the disinfection camara the exact 13.5 amount of culture media was poured into each test tube, cautiously the aluminum cap was put on again and let it cool until it was thick.

8.2.3. Hormones and Vitamins application plant 1.

Once the culture media was cold enough the hormones and vitamins mixture using a pipette it was carefully distributed along the 20 test tubes so that they could be in the following way:

- 5 tubes with 3ml (1-5)
- 5 tubes with 1.5ml (6-10)

- 5 tubes with 0.5ml (11-15)
- 5 tubes with 0.2 ml (16-20)

Also, they were labeled from 1 to 20 to differentiate them and their hormones quantities.

8.2.4. Sterilization.

In order to sterilize the test tubes and prevent from microorganism's propagation the tubes were placed on a pressure cooker to create a pressure field of 140 kilopascals with a temperature of 50ºC for a period of 15 minutes. The test tubes were collocated in the refrigerator to preserve until the next step in the procedure.

8.2.5. Collocation of plant samples.

The samples were recollected from a young *Vasconcellea Pubescens* plant, it was distributed that 10 of the samples were taken from the steam and the other 10 from leaves. Using a 15-blade scalpel a transversal cut was made with resulting steam pieces of approximately 0.5cm, while the leaves were cut in squares with 2cm of perimeter. Later, this samples were cautiously collocated inside the test tubes using a gripper so that each sample could be placed in the culture media section and assure a proper growth. During all the process a candle was used to disinfectant any possible microorganism and it was realized inside of the disinfection camera.

8.2.6. UV exposure.

Once the plant sample were collocated properly, the test tubes were placed in two test tube racks and placed forming a perpendicular line in front of the 30W UV light, to simulate a concentrated sun light and influence on the propagation.

8.2.7. Hormones and Vitamins application plant 2.

For posterior comparation the plant *Carica Pentagona,* which was planted on a pot with 40cm x 45 cm, it was exposed to natural whether conditions, a natural media and 350 ml of hormones and vitamins solution diluted in 1000 ml of water applied as an irrigation system making sure it was sprayed in the leaves and passed through the steam and ground in which it was planted, it was given every 48 hours for a period of ten days.

8.2.8. Data recollection

The plants will be under observation for 15 days and the growth process will be contrasted and compared with the growth progress of the naturally agricultural method from the *Carica papaya* plant.

9. Results and Analysis:

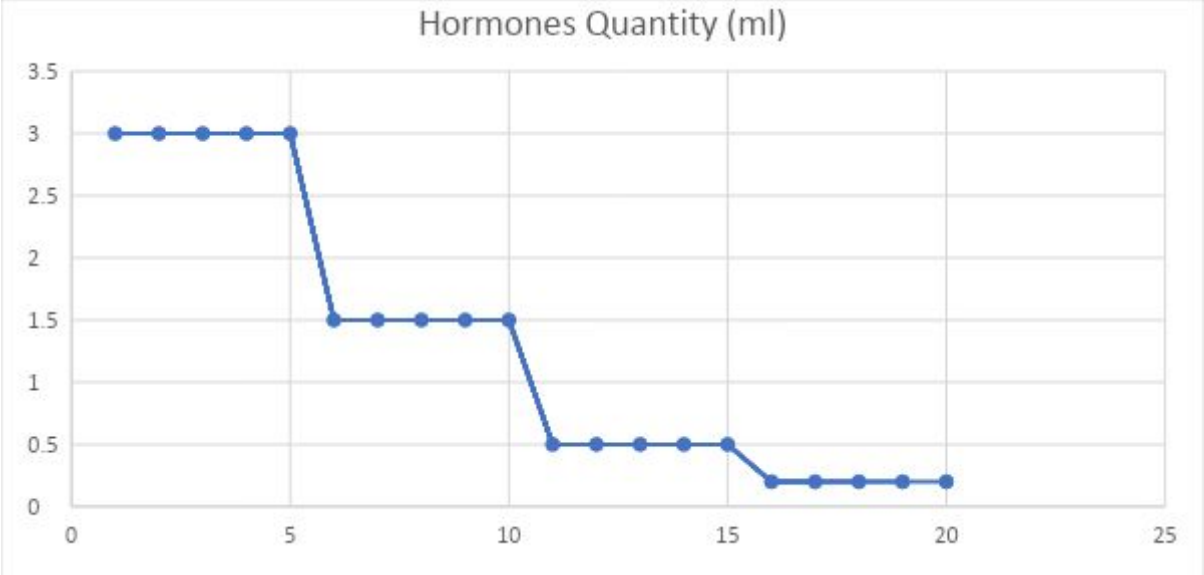

Figure 5 Hormone's quantity (ml) vs. number of samples. (By student, 2021)

In figure 5 it can be observed the different amount of hormones applied to every sample from 1 to 5 it was applied 3 ml, from 6 to 10, 1.5 ml, from 11 to 15, 0.5 ml, and from plant sample 16 to 20 a 0.2 ml.

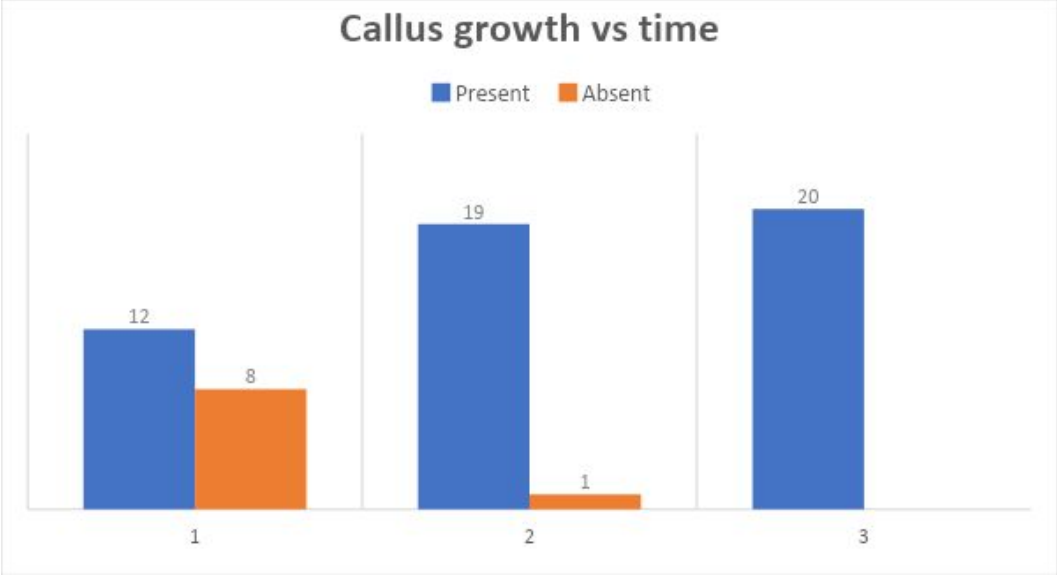

Figure 6 Callus growth vs. hours. (By student, 2021).

In figure 6 it can be observed the development of callus since the 48 hours of the inoculation of plants samples with a result of 12 samples with callus presence and 8 samples without. In the 120 hours a 19 of samples has presence of callus while one of them were still missing the callus development. Finally, in the 168 hours all of the 20 samples had callus developed.

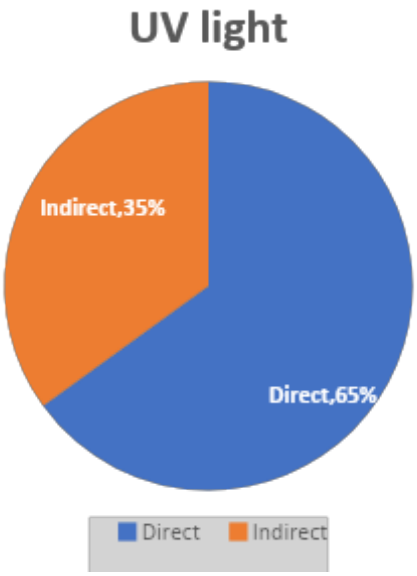

Figure 7 UV light. (By student, 2021)

Figure 7 shows the distribution of light according to the position of the samples, due to the space in the laboratory test tubes were located a 65% of them in front of the 35 % left, the upcoming results demonstrated that 65% of plant samples were receiving direct UV light, while the 35% received indirect UV light.

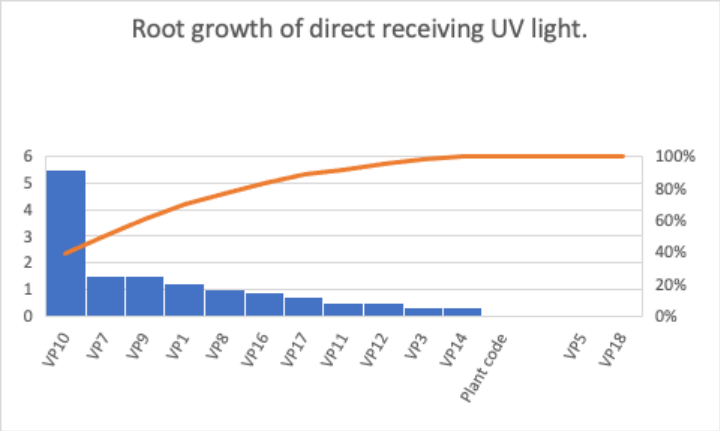

Figure 8 Root growth of direct receiving UV light. (By student, 2021)

Figure 8 shows a representation of the root growth taken into consideration the quality of UV light the plant sample receives, demonstrating that all the samples that belong to the 65% of direct UV light receiving had a proper growth.

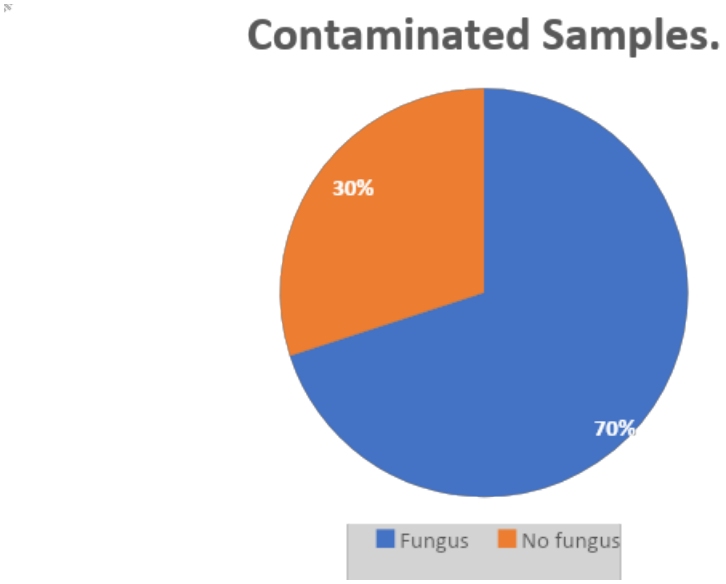

Figure 9 Contaminated samples. (By student, 2021)

Since day 8 it was observed the contamination of media trough the development of fungus and bacteria with a result of 70% of contaminated samples and a 30% of non-contaminated ones.

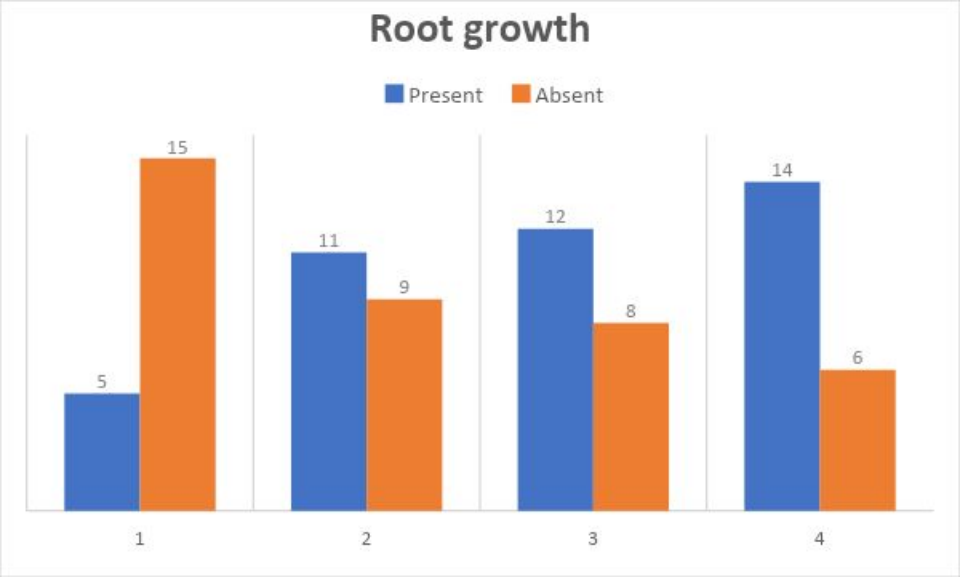

Figure 10 Root growth vs hours. (By student, 2021)

Figure 10 represents the development of roots in 120 hours the first five samples showed a rapid root development, in the 168 hours an increase of the 11 samples with root presence, while the 216 hours showed an increase to 1. Finally, in the 268 hours a final result of 14 samples with root presence and 6 samples without root formation.

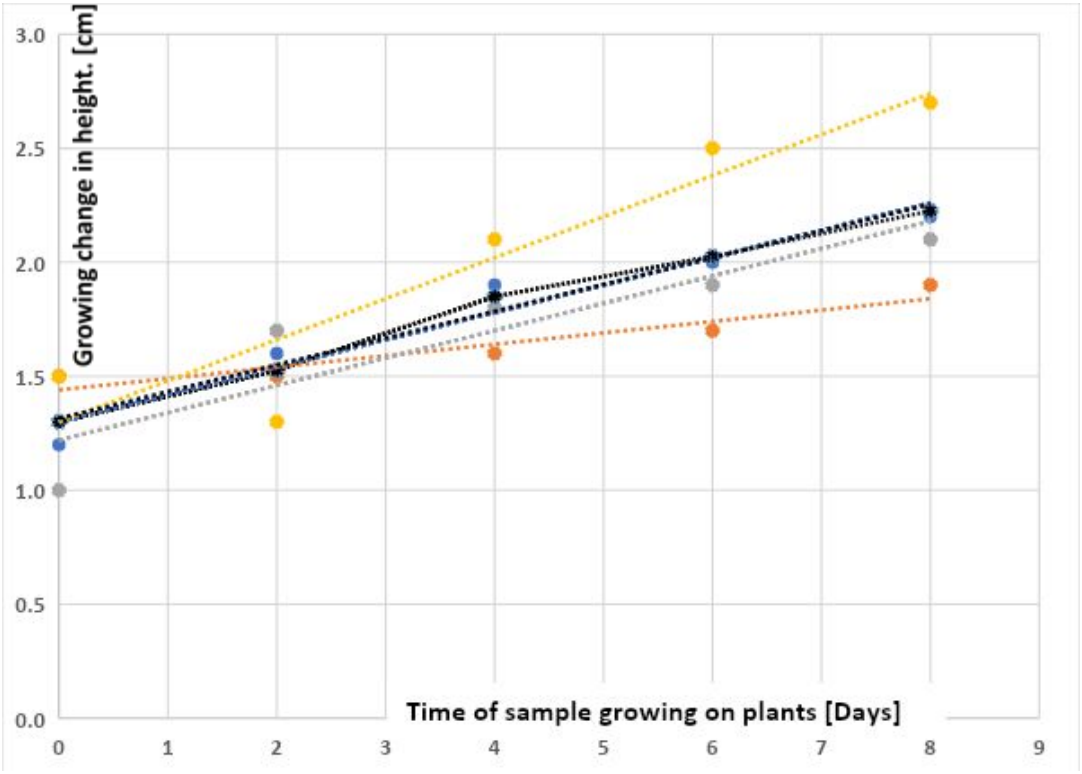

Figure 11 Root growth until 216 hours. (By student, 2021)

As a result of the media contamination on 15 samples, which growth was paralyzed and the anomalous case of plant 5, which grew to fast until it reached the artificial surface, data belonging to these samples were dismissed. Figure 11 is a demonstration of root growth with respect to the amount of time, days since the presence of root was observed, showing that there exists a tendency that suggest growth will continue exponentially.

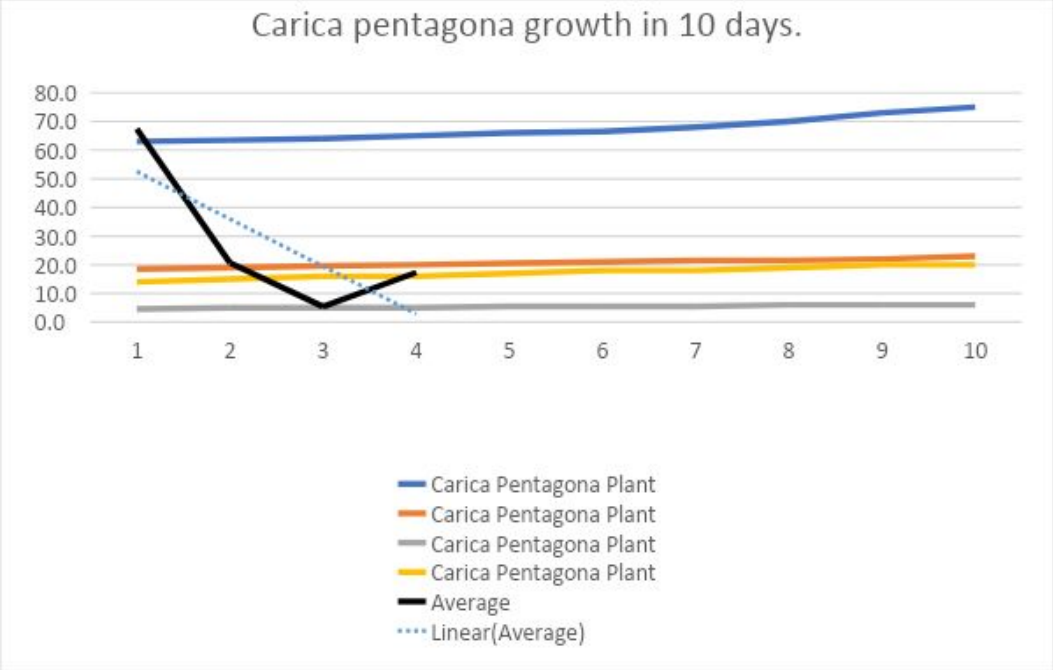

Figure 12 *Carica Pentagona* growth in 10 days. (By student, 2021).

Finally, figure 12 is a demonstration of the growth that *Carica pentagona* showed for 10 days, with natural conditions, planted on a pot, exposed to weather changes and the application of 350 ml of hormones and vitamins mixture diluted in 1000 ml of water which was applied every 48 hours. With an initial measure of 63.0 cm and a resultant of 75.0 cm, showing a tendency of exponential growth.

10.Conclusion:

- ✔ The plant sample 10 was the most briefly developed one, considering it as an anomalous case, since the seventh day it showed an efficient growth, it was used 1,5 ml of hormones without being diluted in water, and it had the UV light directed to it as shown in figure 5. Therefore, it can be inferred that a high number of hormones does not mean faster growth, but the photosynthesis factor being provided by the direct amount of UV light receiving had a greater influence on the plant development.

- ✔ By observing all 20 plant samples I came to the conclusion that in order to have a full and efficient growth it should be the work of all factors; amount of hormones, carbon groups, UV light received, and time, focused equally on all plant samples.
- ✔ Due to the development of fungus and bacteria in the culture media since the seventh day, as shown in figure 9, initially of the 10 leaves containing samples, it can be concluded that leaves samples are not suitable for micropropagation of the specific specie *Vasconcellea Pubescens*.
- ✔ The presence of callus, demonstrated in figure 6, was found since the 48 hours in 12 of the 20 samples, meaning that the asexual reproduction process of mitosis is almost an immediate response since the inoculation of the samples in the culture media in the majority of the samples planned for the experiment.
- ✔ According to source The Papaya: Botany, Production and Uses by Sisir Mitra et.al, plants from the *Carica* family, in exception of domesticated plants like *Carica papaya* and *Carica pentagona*, are too sensitive to the conditions it is propagated giving results contamination of the samples when propagating. Therefore, through the experiment of micropropagation it was evaluated that the plant *Vasconcellea Pubescens* is not suitable for this method, also reason why Carica Pentagona is better developed in the international market.
- ✔ It was observed that since the eleventh day that samples showed a deficit in the growth of four plants have been noticed, and from another four since the thirteen day, when the fungus were already developed concluding that the development of fungus paralyzed the miotic factor and therefore the growth of roots in the culture media or delayed in a way that their growth was no longer significant.

- ✔ As shown in figure 11, the value of precision was a 94.74% at the moment of taking measures, meaning that the measure appreciation using only one decimal was a potential cause for the 5.26% of error.
- ✔ By the study of both cultivation methods in different plants, Carica Pentagona and Vasconcellea Pubescens, the research question was answered, by the demonstration that the application of auxins and gibberellins had a positive effect in micropropagation and the agricultural growths accelerating the development of roots and the growth of the specific plant. The tendency line expressed in figure 11 shows a 0.3 cm of growth per day in *Vasconcellea Pubescens*, while in *Carica Pentagona* tendency line showed a 1.2 cm of growth per day demonstrating that the application of auxins and gibberellins had a decrease in time that last to develop an efficient growth and reproduce the specific plant.
- ✔ The alternate hypothesis was accepted, which stands that both of the methods, micropropagation and agricultural, were the most effective method of plant growth, since both of the average results of growth demonstrated a tendency for exponential growth since the appliance of bio stimulants, in this case hormones auxins and gibberellins.
- ✔ As a recommendation for posterior investigations is to take into consideration a sample of plant without applying the bio stimulant and compare its results.

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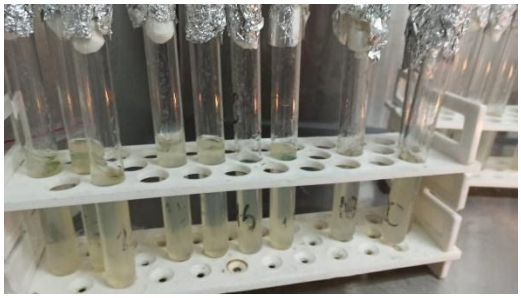
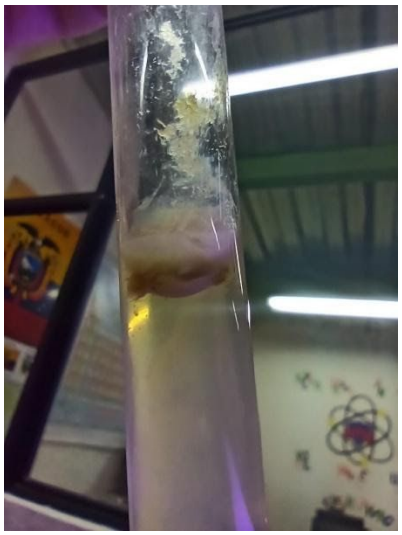
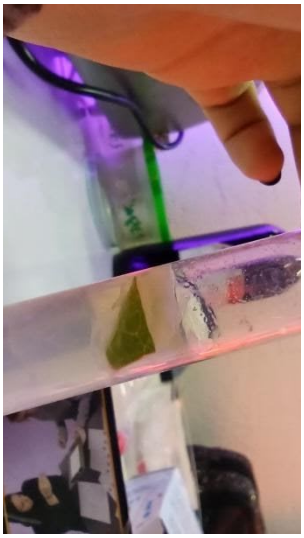
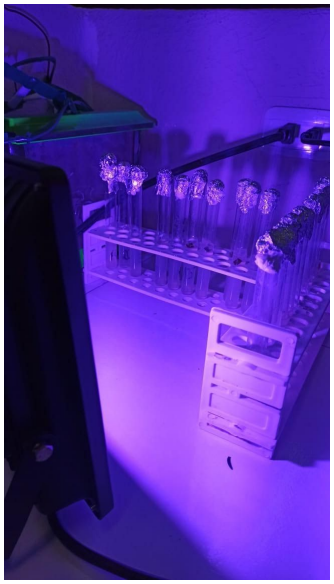
12. Appendix:

12.1. Laboratory evidence:






EQUIPOR
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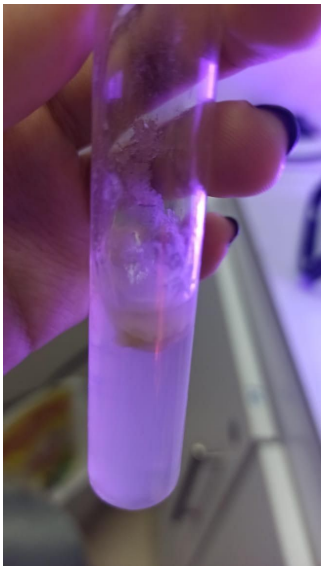

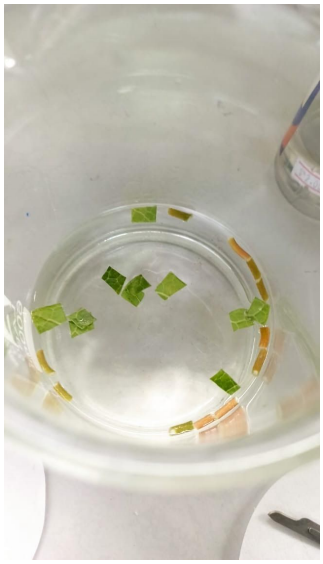

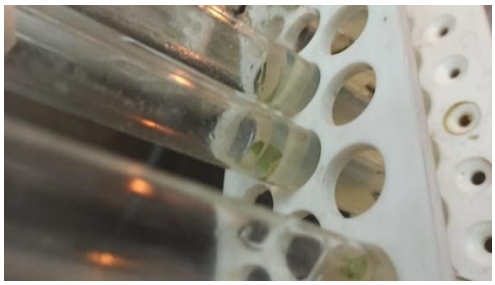


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500 g
BBL™ Trypticase™
Soy Agar
Soybean-Casein Digest Agar


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12.2. Raw data tables.

Growing change in height [cm] Root *Vasconcellea Pubescens (VP)*						**Notes:**
Plant code	**Time of sample growing on plants [Days]**					
	0	**2**	**4**	**6**	**8**	
VP1	1.2	1.6	1.9	2.0	2.2	Ok
VP2	0.0	0.0	0.0	0.0	0.0	Useless
VP3	0.3	0.5	0.5	0.5	0.5	Avoid
VP4	0.0	0.0	0.0	0.0	0.0	Useless
VP5	0.0	0.3	0.4	0.4	0.4	Avoid
VP6	0.0	0.0	0.0	0.0	0.0	Useless
VP7	1.5	1.5	1.6	1.7	1.9	Ok
VP8	1.0	1.7	1.8	1.9	2.1	Ok

VP9	1.5	1.3	2.1	2.5	2.7	Ok
VP10	5.5	5.7	5.7	5.7	5.7	**Useless**
VP11	0.5	0.5	1.0	1.0	1.0	Avoid
VP12	0.5	0.8	1.0	1.0	1.0	Avoid
VP13	0.0	0.0	0.0	0.0	0.0	Useless
VP14	0.3	1.3	1.5	1.5	1.5	Avoid
VP15	0.0	0.0	0.0	0.0	0.0	Useless
VP16	0.9	1.0	1.0	1.0	1.0	Avoid
VP17	0.7	0.7	0.7	0.7	0.7	Avoid
VP18	0.0	1.2	1.9	1.9	1.9	Avoid
VP19	0.0	0.0	0.0	0.0	0.0	Useless
VP20	0.0	0.0	0.0	0.0	0.0	Useless

Table 4 Vasconcellea Pubescens root growth in 8 days. (By student, 2021)

Growing change in height [cm] Root *Vasconcellea Pubescens* (VP)					
Plant code	**Time of sample growing on plants [Days]**				
	0	**2**	**4**	**6**	**8**
VP1	1.2	1.6	1.9	2.0	2.2
VP7	1.5	1.5	1.6	1.7	1.9
VP8	1.0	1.7	1.8	1.9	2.1
VP9	1.5	1.3	2.1	2.5	2.7
Average:	**1.3**	**1.5**	**1.9**	**2.0**	**2.2**

Table 5 Delimitated root growing table. (By student, 2021)

Carica Pentagona Plant				
Time [Days]	**Steam growth [cm]**	**Lenght of bigger leave [cm]**	**Length of smaller leave [cm]**	**Quantity of leaves**
1	63.0	18.5	4.5	14
2	63.5	19.0	5.0	15
3	64.0	19.6	5.0	16
4	65.0	20.0	5.0	16
5	66.0	20.5	5.5	17
6	66.5	21.0	5.5	18
7	68.0	21.5	5.5	18
8	70.0	21.5	6.0	19
9	73.0	22.0	6.0	20
10	75.0	23.0	6.0	20
Average	67.4	20.7	5.4	17

Table 6 Carica Pentagona growth in 10 days. (By student, 2021)