Defoliation and Pre-harvest Drop of Camu-Camu Fruits in Floodable Area

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ABSTRACT

The effect of defoliation and gibberellic acid (GA3) on the fall of the fruit in the induced period of harvest of Myrciariadubia (camu-camu), in a flooded area on the banks of the Amazon River, Iquitos-Peru, was evaluated. The soil is clayey-silty with 2.15% organic matter, pH 6.29, 0.10% Nitrogen, 40.76 ppm Phosphorus and 228 ppm potassium. The average temperature is 27.45 ° C, relative humidity of 86% and 3111.4 mm of rain. The defoliation was carried out with NaCl solution in water (50 g.L-1). GA3, was applied every 15 days at doses of 0, 50, 100 and 150 mg.L.-1. The variables were evaluated: number of flowers per branch (FLR1-4), number of flowers per plant (FLP1-4),% of fruits retained in phase 3 (FR3),% of fruits retained in phase 5 (FR5), weight of fruits (PF), number of harvested fruits (FC), approximate yield of fruits (RAF), and the real fruit yield (RRF). The design was completely randomized (DCA), in 2x4 factorial arrangement. Defoliation significantly reduced FLP1-4 from 6938 to 3701, while GA3 did not influence that character. Defoliation significantly increased the FR3 retention, but reduced the PF and FC. GA3 significantly influenced FLR1-4, FR3 and PF. In FR5, no statistical difference was recorded for either D or GA3, nor was there significant interaction between the two defoliation and GA3factors. For FC, RAF and RRF, highly significant differences were found for Defoliation, with superiority of non-defoliated plants but no differences were found for GA3 doses. It is concluded that in F3 the defoliation favored significantly, effect that was diluted in F5, while the application of GA3 negatively influenced this retention. Fruit yield was significantly higher in non-defoliated plants without showing significant difference between GA3 doses.

KEY WORDS: Fenology, Giberelic acid, Fruit abscission, Varzea

1. INTRODUCTION

Among the species with capacity to withstand extreme water logging conditions is the camu-camu [*Myrciariadubia* (H.B.K.) McVaugh], which also has high contents of ascorbic acid; Pinedo et al. (2001) found in the Peruvian Amazon values of 1230 to 2089 mg/100 g in their fruits. This outstanding characteristic is a growing interest to domesticate the species and since 1980 a process of promotion of its use and cultivation in flooded areas of varzea was started in Loreto-Peru (Pinedo et al., 2001).

One of the multiple factors that come together in the sustainable management of the species and its incorporation into the markets is the harvest period. This factor is closely related to the price of the product and therefore to the profitability and sustainability of the crop; what has motivated the investigation to establish a method of defoliation that concludes in a harvest out of season and satisfactory for its quantity and quality. By not having a viable method of controlling the reproductive phenological period, the main harvest season is usually associated with the reduction of the price of fruit as a result of the law of supply and demand. This condition of instability favors in part the demotivation of the producers due to lack of economic income. This limitation has been and still is notorious in this region of Loreto-Peru (Ulchur, 2017). In this regard, defoliation is known as a practice to modify the reproductive phenology and harvest in periods of lower supply and higher prices (Zuñiga, 1992, Cayon and Bolaños, 1999: Martínez, et al., 2010, Pinedo, et al., 2014). However, in the case of camu-camu, under cultivation in floodable areas, the defoliation technology has not yet been refined and doubts remain regarding the level of fructification in the periods induced by this technique. Although defoliation and flowering outside the time has been achieved, fruiting was not always satisfactory when induced outside of time, as a result of the premature fall of the fruits.

Regarding the causes of the fruit drop Peters and Vásquez (1986), estimated that 46% of flowers are pollinated and that 15% of immature fruits abort before maturity. However, Pinedo et al., (2001) found that the percentage of fruits that fall before completing their development is 73%. Several studies have been developed for camu-camu, looking for the causes of fruit fall, finding several factors such as Boro and Calcium minerals, physiological aspects not yet specified and insects (Lopez, 2003; Farro et al., 2011; Farro, 2012)

In many species, the initial development of the fruits occurs at the expense of the existing reserves in the plant after flowering and because of the scarce capacity that the new shoots still have to provide photosynthates. Therefore, any nutritional deficiency causes the paralysis of growth and very possibly the drop of fruits. The drop of flowers and small fruits occurs in a very high number, which within some limits, is considered natural, since the plant would not be able to sustain the fruits originated by a normal flowering. In general, it is considered that a harvest is good, if the percentage of fruits harvested with respect to the number of initial flowers is: 5% in pear and apple, 10% in peach, 30% in almond, 8% in plum, 1% in avocado, 4% in citrus, 2% in olive and 25 to 50%, according to varieties, in grapevine (Urbina, 2002). Different technologies were applied to avoid excessive fruit drop: GA3, auxins, cytokinins, boron, nitrogen, and ringings. With this practice in different species such as apple, avocado, it was possible to significantly increase the number of fruits tied by ringing the trunks during flowering (Rojas et al., 2004, Alvarez, et al., 2005, Gonzales, 2000). The result may be due to the interruption of the phloem prevents the passage of nutrients and hormones to the roots, causing a redistribution of them. Gibberellins, isolated from the fungus Gibberellafujikoroi by Eichi Kurosawa in 1926, are promoters of plant growth (Jordan and Cassareto, 2006). The effect of the fungus on the affected plants consisted of a notable increase in height although with a strong reduction in grain production (Malonek et al. 2005, Tamura 1990). Gibberellins are a group of several chemically similar compounds, of which more than 80 have been identified and classified by number according to the order of discovery, among which giberelic acid 3 (GA3) is one of the most abundant (Díaz 2002). These acids are derivatives of tetra-cyclic diverted hydrocarbons that represent an important group of

hormones for plant growth (Salisbury and Roos 2000). They are known as modulators and constitute the chemical signals of the entire system for regulating the operation of plants. Increase the mooring of fruits in most crops, improve yield and height growth, shortening in some cases the time to harvest (Alvarez et al., 2005).

The objective of this research was to evaluate the effect of the gibberellic acid phytoregulator on the fruit drop with and without defoliation, in view of the need to improve the defoliation technique to regularize the fruits obtained out of regular harvest season.

2. MATERIALS Y METHODS

The present study was developed in 2016 (plantation of 9 years) in the town of Santa Rosa, Amazon River, district of Belen, between the coordinates 03°42'24.0 "South Latitude and 73°11'01.5" West Longitude (Figure 1). The area is located on a low flood terrace of the Amazon River, classified as a low resting (Rodríguez, 1990).
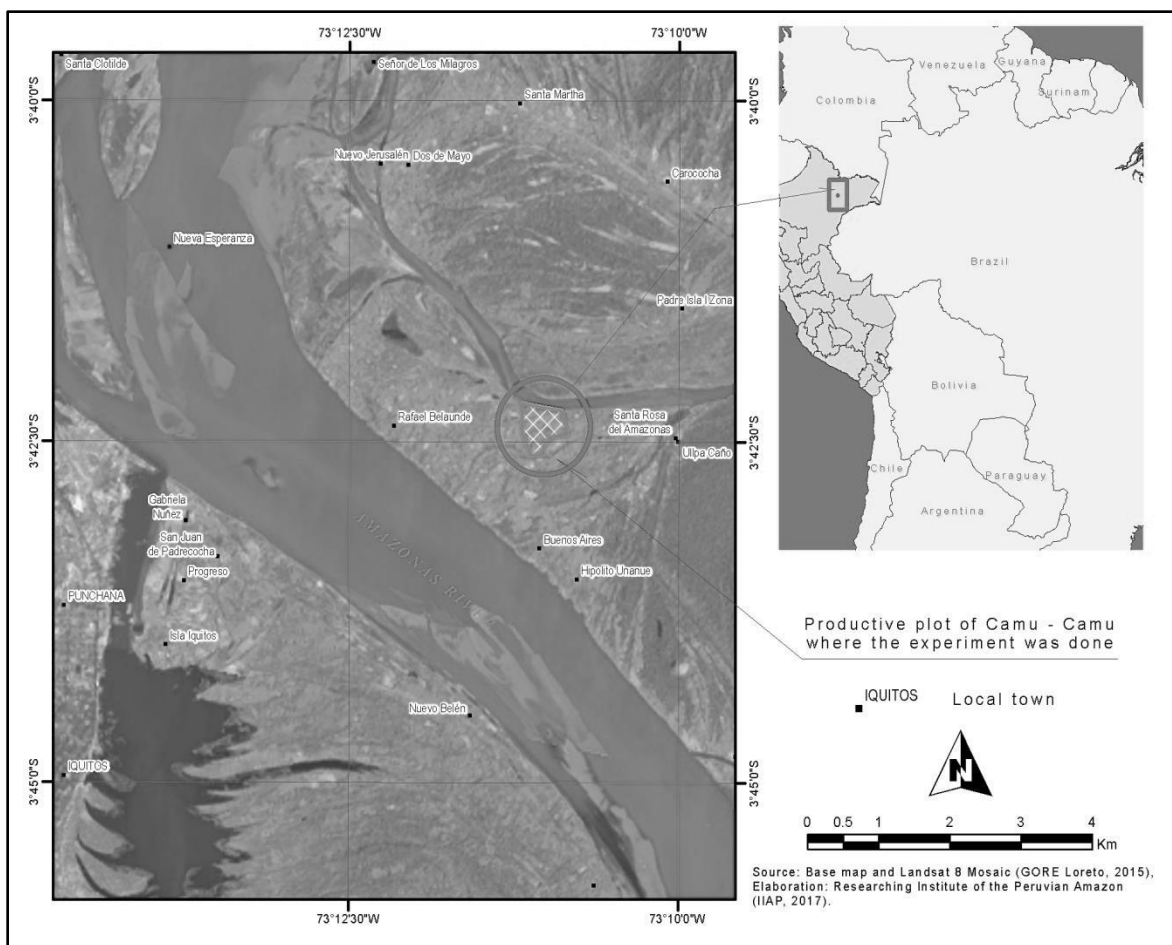

Figure 1.Location map of experimental field.

The soil of the plot under study presents a texture Arcillo-Limosa with 2.15% of organic matter, pH of 6.29, 0.10% of Nitrogen, 40.76 ppm of Phosphorus, 228 ppm of potassium, saturation of bases of 75.62% and saturation of aluminum of 0.0% with a action exchange capacity of 25.63.

The meteorological data were provided by the National Service of Meteorology and Hydrography of Peru (SENAMHI). The average annual temperature of the area was 27.45 ° C, annual relative humidity of 86% and annual precipitation of 3111.4 mm.

Defoliation and gibberellic acid (GA3)

To defoliate, a previously investigated method (Pinedo et al., 2014) was used by salt (sodium chloride), at a dilution of 50 g.L-1 of water. Approximately one liter of the defoliant solution was applied to each plant by manual backpack pump and on a sunny day, to favor the total defoliation of the treated plants. The gibberellic acid was diluted in ethyl alcohol 90 °, in order to prepare the solutions according to the planned doses (0, 50, 100 and 150 mg.L.-1), adding the water at a rate of 1 liter for each dose. Said applications were made every 15 days from the flowering until before the phase 5 of the fructification, with a total of 4 applications.

Treatments

Of a total of 119 existing plants in the plot, 48 were selected according to the following criteria: a) that are isolated between them, b) similar cup architecture c) similar luminosity conditions between them d) degree of flooding between plants, more uniform as possible. After choosing the 48 plants to be evaluated, 6 plants were assigned at random for each of the 8 treatments that were the following: T1: Without defoliation and 0 mgL-1 GA3. T2: No defoliation and 50 mgL-1 GA3. T3: No defoliation and 100 mgL-1 GA3. T4: No defoliation and 150 mgL-1 GA3. T5: With defoliation and 0 mgL-1 GA3. T6: With defoliation and 50 mgL-1 GA3. T7: With defoliation and 100 mgL-1 GA3. T8: With defoliation and 150 mgL-1 GA3.

Variables

The evaluated variables are: Number of flowers per branch (FLR1-4) in phase 1 to 4 of the flowering. Number of flowers per plant (FLP1-4). The phenological stage of flowering begins with the differentiation of the floral bud (phase 1) until the opening of the stamens (phase 4). For the flower count, mechanical comptometers were used and the four states were considered, from emergent flowers with vertically still closed, until when the flower opens to give rise to pollination (Inga, 2001). Percentage of fruits retained in phase 3 (FR3) (19-26 days from the beginning of fruiting) in relation to the number of flowers (FL1-4). Percentage of fruits retained in phase 5 (FR5) (36-43 days after the beginning of fruiting) in relation to the number of flowers (FL1-4). Fruit weight (PF), taken at random, expressed in grams and average of 20 fruits in large green state and pintón (phases 5 and 6 of fruiting). Number of harvested fruits (FC). Approximate yield of fruit (RAF), resulting from multiplying the number of fruits per branch by the number of branches and the actual yield of fruits / plant (RRF), resulting from multiplying the average fruit weight by the total number of fruits counted in Phase 5. The yield was expressed in grams / plant.

Theoretical framework of the reproductive system

The flowering of camu-camu begins at 2-3 years of age, when the plant reaches a basal diameter of 2.0 cm. In each knot up to 12 hermaphrodite flowers are observed. During the anthesis, the style comes out first and after several hours the stamens leave, which avoids autogamy. Apparently, at the time that the stamens emerge to release pollen, the stigma is no longer receptive to pollination. This apparent dicogamia does not rule out the possibility of selfing by geitonogamia due to the lack of floral synchrony. Pollen from other flowers on the same plant can effect up to 91% pollination. (Peters and Vásquez 1986)

Flowering and fruiting periods vary between basins according to the level of water reaching the rivers (Picon and Acosta 1999). According to Inga (2001), reproductive phenology includes 4 phases in flowering and 8 phases in fruiting:

Phases of the flowering stage

- Phase 1. From the appearance of the flower bud and the subsequent 7 days.

- Phase 2. The floral bud grows in length and diameter, until presenting a globose form, this phase also takes 7 days.

- Phase 3. The floral bud opens and the style first emerges and then the stamens emerge, which happens in the morning, when the flower is pollinated, observing the presence of bees.

- Phase 4. When the style emerges and is pollinated, the stamens begin to detach, phases 3 and 4 last for 4 5 hours.

From the appearance of the floral bud until the beginning of the formation of the fruit, 15 days pass.

Phases of fruiting state

- Phase 1. Once the flower is fertilized, the stamens and sepals are detached and the style takes the form of a clavito, light green color that measures 0.15 cm in height. This phase takes 7 days.

- Phase 2. The fruit continues its development, adopts a dark green coloration, with 0.16 - 0.35 cm long, a phase that also takes 7 days.

- Phase 3. The fruit develops in size, maintains its green coloration and measures from 0.36 - 0.60 cm, this phase comprises 12 days.

- Phase 4.The fruit measures 0.61 - 1.0 cm in diameter and from this phase, the fruits are considered physiologically developed, this phase lasts 10 days.

- Phase 5. The fruit reaches 2.4 cm in diameter, with an average weight of 7.5 g, its duration is 7 days.

- Phase 6.The fruit has small reddish spots identified as "green-paint", measures 2.5 cm in diameter and 9.3 g average weight, this phase takes 7 days.

- Phase 7. The fruit has a light red color with green or reddish-green spots, known as "pintón-maduro", measuring 2.6 cm in diameter and 10.3 g average weight; This phase takes 6 days.

- Phase 8.The fruit is red wine in its entirety, considered a mature fruit, measures 2.5 cm in diameter and 10 g average weight, this phase takes 6 days.

Design and statistical analysis

The completely randomized Design (DCA) was applied in a factorial arrangement of two factors (Defoliation x GA3 dose). The levels of the factors were 2 and 4 respectively, giving rise to 8 treatments. The experimental unit consisted of a single plant with 6 repetitions. Normality tests were performed using Q-Q PLOT graphs (Predicted R-) and variance homogeneity by means of Scatter Diagram (Predicted R-Vs Res Est.). Then Fisher's variance analysis (ANOVA) and Tukey's means tests were performed for: number of flowers (FL1-4),

percentage of fruits retained in phase 3 (FR3), percentage of fruits retained in phase 5 (FR5) and fruit weight (FP), and Kruskal-Wallis nonparametric variance analysis for: fruit yield (RAF and RRF), using the statistical program InfoStat, version 2016e.

The statistical model corresponding to the DCA with two factors is as follows:

Yijk = u + Ai + Bj + rk + ABy + eijk where

Yijk = Is the value measured in the field, u is the general average, Ai is the effect of the treatment i belonging to the factor A, Bj is the effect of the treatment j belonging to the factor B. rk is the effect of the repetition K, ABy is the effect of the interaction of the treatment i belonging to the factor A with the treatment j belonging to the factor B and eijk is the effect of the random error or residue associated to each plot ijk

3. RESULTS AND DISCUSSION

3.1. Defoliation

Regarding the defoliation practiced, the effect of the saline solution occurred in all the plants treated after 15 days of the application in which the leaves were 100% detached. The leaf sprout started 20 days after the application.

Defoliation is practiced to modify the reproductive phenology and harvest in periods of lower supply and higher prices. Ibalo (2000) mentions that the abscission of the leaves, by natural or provoked senescence, is preceded by a series of changes that include: the loss of chlorophyll, the increase of anthocyanins, reduction of protein, carbohydrate and inorganic ions levels and the occurrence of alterations in hormone levels. At the end of the senescence process, a drastic increase in metabolic activity occurs due to alterations in the hormonal levels of the leaf blade. As a result of these changes, in the abscission zone the cells secrete hydrolytic enzymes that degrade the cell walls, which cause the leaf to detach. Although there are many enzymes that increase their activity in the abscission zone in relation to this process, such as "pectinase" and "cellulase". According to Ballester (1997), when defoliation is made it is important to make a total, unique and rapid action accompanied by a treatment with anticryptogamic (substances that serve to fight against plant diseases due to plant parasites).

According to Imán and Melchor (2007), the defoliation technique allows regularizing and standardizing the harvest, through the physiological balance of the plant. The removal of leaves induces the rapid sprouting of lateral buds and increases flowering. Likewise, as a result of defoliation, the growth, increase and uniformity of the buds are stimulated, as long as there is an adequate level of growth-promoting substances (Sánchez, 2011).

The application of nitrogen cyanamide (Dormex) for the defoliation of camu-camu has shown effectiveness with doses between 3 and 5% in periods of 206 to 210 days from defoliation to harvest (Davila 2012 and Abanto et al., 2014) . As another option to defoliate and standardize the harvest, Cotrina and Oliva (2007) found 86% defoliation through the application of copper sulphate at a dose of 1.5%. Pinedo et al. (2011), concluded that defoliation even without fertilization in camouflage plants influences yield, by obtaining simultaneous release of leaves, stimulates flowering and cuts pest cycles so that it could become an essential task in the agronomic management of camu-camu. More recent trials showed the effectiveness of saline solution to defoliate camu-camu with the advantage of not being polluting.

Manual or chemical defoliation was also applied for the purpose of adaptation to tropical conditions, for example of the apple tree, to stimulate the sprouting of buds. For this, hydrogenated cyanamide, magnesium

chlorate, copper hydroxide and zinc oxide were used. The best defoliants were hydrogenated cyanamide (Dormex) and magnesium chloride (Zuñiga, 1992)

Regarding fruit setting, the treatments that reached the lowest fruit set percentages were those with the highest floral buds (Dormex and Magnesium Chlorate), a phenomenon that could be attributed to the competition effect between fresh fruits, since The evaluation was carried out one month after pollination. For the performance variable, significant differences were found between treatments. The highest yields were achieved by those treatments that achieved the highest budflows, both total and floral (Dormex, Magnesium Chlorate and Copper Hydroxide), far exceeding the rest of the treatments

Before the statistical analysis, the Shapiro-Wilk normality test (Razali and Wah, 2011) was performed on the original data of the variables, finding p values greater than 0.05 in number of flowers per plant,% of fallen fruits F3,% of retained fruits F3,% of fruits retained F5 and average fruit weight respectively. For fruit yield, the p value was less than 0.05. Likewise, the variance homogeneity test was performed using Bartlett's statistical test (Glass, 1966). Variance homogeneity was found in all the variables, except yield per plant.

3.2. Descriptive statistics

Table 1 shows the statistics of the variables studied with measures of central tendency whose values are within normal values. For example, the percentage of fruit retention in phase 5 with an average of 7.85, an expected level if we consider the study by Farro (2011).

Table 1. Descriptive statistics for eight variables of reproductive state of camu-camu

Variables	N	Range	Minimum	Maximum	Mean	Std. Deviation	Variance
Number of flowers/brunch **(FLR1-4)**	48	204,00	63,00	267,00	146,14	43,07	1854,72
Number of flowers/plant **(FLP1-4)**	48	16469,00	518,00	16987,00	5319,96	3807,25	14495155,53
%retained fruits in phase 3 **(FR3)**	48	44,08	3,65	47,73	16,95	10,13	102,56
%retained fruits in phase 5 **(FR5)**	48	16,59	2,25	18,84	7,85	3,99	15,94
Wheightoffruit **(PF)**	48	5,00	4,00	9,00	6,07	1,17	1,36
Number of harvested fruits **(FC)**	48	904,00	56,00	960,00	212,02	182,66	33364,83
Aprox. yield of fruits **(RAF)**	48	18817,10	64,90	18882,00	1861,99	3404,35	11589622,24
Real yield of fruits **(RRF)**	48	8354,40	285,60	8640,00	1386,52	1510,74	2282352,27

3.3. Parametricvarianceanalysis

The analysis of variance of the five dependent parametric variables that met the requirements for Fisher's test appear in Table 2 for both the independent variables (Defoliation and gibberellic acid) and the interaction of these two factors.

Table 2. Test of between-subjects effects for defoliation and doseofGA3about 5 dependent variablesofcamu-camu

Source	Dependent Variable	Type III Sum of Squares	DF	Mean Square	F	Sig.
Defoliation	FLR1-4	2836,687	1	2836,687	1,973	,168
	FLP1-4	125757450,750	1	125757450,750	10,925	,002
	FR3	1529,343	1	1529,343	25,988	,000
	FR5	2,512	1	2,512	,162	,690
	PF	18,008	1	18,008	21,055	,000
GA3	FLR	19856,729	3	6618,910	4,603	,007
	FLP	24289121,750	3	8096373,917	,703	,556
	FR3	693,457	3	231,152	3,928	,015
	FR5	17,709	3	5,903	,381	,768
	PF	7,364	3	2,455	2,870	,048
Defoliation XGA3	FLR	6965,729	3	2321,910	1,615	,201
	FLP	70783595,417	3	23594531,806	2,050	,122
	FR3	243,566	3	81,189	1,380	,263
	FR5	108,471	3	36,157	2,331	,089
	PF	4,457	3	1,486	1,737	,175

3.4. Nonparametric variance analysis

Three variables did not meet the requirements for Fisher's test and the nonparametric Kruskal-Wallis test was applied for them. The variables are: "real yield of fruits (RRF)", "number of harvested fruits" (FC) and "approximate yield of fruits (RAF)". The influence of "defoliation" on these three variables was highly significant. While for the variable "dose of GA3" the influence did not reach to be significant (see Table 3).

Table 3.Kruskal-Wallis non-parametric test for the influence of defoliation and GA3dose on number of harvestedcamu-camu fruits (FC, approximate yield of fruits (RAF), and real yield of fruits (RRF).

Null Hypothesis	Test	Sig.	Decision
1.The distribution of Real yield of fruits (RRF) is the same across categories of defoliation.	Independent Samples Kruskal-Wallis Test	0.000	Reject the null hypothesis
2.The distribution of Number of harvested fruits (FC) is the same across categories of defoliation.	Independent Samples Kruskal-Wallis Test	0.000	Reject the null hypothesis
3.The distribution of Approximately yield of fruits (RAF) is the same across categories of defoliation.	Independent Samples Kruskal-Wallis Test	0.000	Reject the null hypothesis
4.The distribution of Real yield of fruits (RRF) is the same across categories of giberellic acid dose	Independent Samples Kruskal-Wallis Test	0.755	Retain the null hypothesis
5.The distribution of Number of harvested fruits (FC) is the same across categories of giberellic acid dose	Independent Samples Kruskal-Wallis Test	0.911	Retain the null hypothesis
6.The distribution of Approximately yield of fruits (RAF) is the same across categories of giberellic acid dose	Independent Samples Kruskal-Wallis Test	0.360	Retain the null hypothesis

Asyntotic significances are displayed   The significance level is 0,05

3.5. Correlations

In this correlation analysis, being all 8 analyzed variables of reproductive nature they tend to be correlated. It is interesting to find a negative correlation between% FR3 with RRF when it should be positive (the greater number of fruits retained or tied, would correspond with a higher level of yield). It would mean that in the later stages of fruiting the nutritional and physiological reserves of the plants are deficient and not enough to retain this level of fruiting until harvest. It is logical the correlation of RRF with the levels of flowering and previous fructification (FLR, FLP FC and RAF), with values of r 0.430 **, 0.488 **, 0.976 ** and 0.822 respectively, including with PF (r = 0.588 * *), see Table 4.

Table 4. Correlations analysis of eight camu-camu reproductive variables (N=48)

Variables		FLR1-4	FLP1-4	%FR3	%FR5	PF	FC	RAF	RRF
FLR1-4	Correlation	1	,188	-,186	-,205	,199	,425**	,415**	,430**
	Sig. (2-tailed)		,200	,205	,163	,175	,003	,003	,002
FLP1-4	Correlation	,188	1	-,223	-,055	,416**	,450**	,634**	,488**
	Sig. (2-tailed)	,200		,127	,710	,003	,001	,000	,000
%FR3	Correlation	-,186	-,223	1	,256	-,051	-,241	-,131	-,205
	Sig. (2-tailed)	,205	,127		,078	,729	,098	,374	,162
%FR5	Correlation	-,205	-,055	,256	1	,088	-,028	,124	,006
	Sig. (2-tailed)	,163	,710	,078		,554	,851	,401	,968
PF	Correlation	,199	,416**	-,051	,088	1	,476**	,595**	,588**
	Sig. (2-tailed)	,175	,003	,729	,554		,001	,000	,000
FC	Correlation	,425**	,450**	-,241	-,028	,476**	1	,755**	,976**
	Sig. (2-tailed)	,003	,001	,098	,851	,001		,000	,000
RAF	Correlation	,415**	,634**	-,131	,124	,595**	,755**	1	,822**
	Sig. (2-tailed)	,003	,000	,374	,401	,000	,000		,000
RRF	Correlation	,430**	,488**	-,205	,006	,588**	,976**	,822**	1
	Sig. (2-tailed)	,002	,000	,162	,968	,000	,000	,000	
**. Correlation is significant at the 0.01 level (2-tailed).									

3.6. Number of flowers per plant (FLP1-4) and per branch

In the analysis of variance of Table 2, it is observed that the total number of flowers per plant (FLP1-4) in phases 1 to 4 of the flowering stage (Pinedo et al., 2001), has been strongly influenced by the factor defoliation (F=10.925 p=0.002 in Table 2) where natural flowering (without defoliation) was much higher with an average of 6938 flowers/plant versus 3701 flowers in defoliated plants (Figure 2). This result contrasts with the assertion of Iman and Melchor (2007), who mention that the removal of leaves induces the rapid sprouting of lateral buds and increases flowering. Other authors such as Sánchez (2011) indicate that such an increase is conditioned by an adequate level of growth-promoting substances. Also, due to the effect of defoliation, the growth, increase and uniformity of the buds is stimulated, as long as there is an adequate level of growth-promoting substances. Affirmation that may be related to the results observed in the retention of fruit in phase 3, where treatments with defoliation show higher levels of fruit retention than treatments without defoliation. Different response was obtained regarding the doses of gibberellic acid (GA3) since no significant difference was found (F=0.703 p=0.556 in Table 2), In Figure 2 the sparse relative difference is corroborated with respect to the total number of flowers between the dose. With a minimum of interaction that does not reach levels of statistical significance (F=1.615 p=0.201 in Table 2).
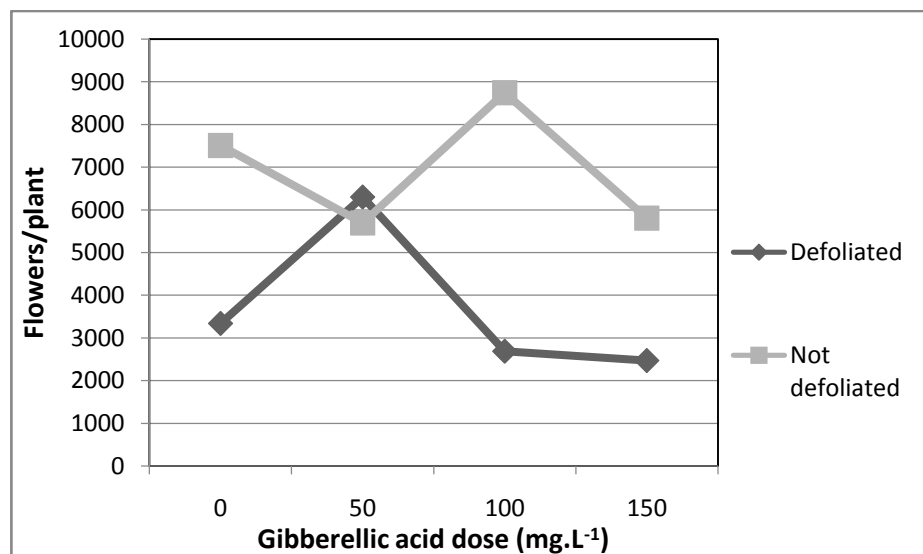

Figura 2. Influencia de las dosis de la defoliación yGA3sobre el número total de flores/planta

Aunque no hubieron diferencias significativas del número total de flores/planta entre las dosis de GA3 señalamos referencialmente que elnúmeromínimo fue 2471 flores correspondiente al tratamiento "con defoliación y 150 mg de GA3" mientras que el máximo correspondió al tratamiento "sin defoliación, con 100 mg de GA3" con un promedio de 8744 flores/planta (Figura 2).

Respecto a la influencia del GA3 sobre la floración, al trabajar con mango (*Mangifera indica*) Tomer (1984), encontró que la aplicación de 200 mg.L^{-1} de GA3 a yemas en reposo, estimuló el crecimiento vegetativo tardío en brotes de mango, mientras que concentraciones de 25 y 50 mg. L^{-1} provocaron brotacion temprana y crecimiento reproductivo. Sin embargo, Turmbull*et al.*. (1996) y Nuñez y Davenport (1991) reportaron retraso de hasta cuatro semanas en la floración de árboles de mango (*Mangifera indica*) al aplicar las giberelinas, pero no inhibieron la producción de inflorescencias en yemas axilares de mango. Los efectos diferentes y hasta contradictorios de la aplicación de GA3 lo comprobó también Oosthuyse (1995) que aplicó 100 mg/l de GA3 a árboles de mango y encontró que el número de inflorescencias y el desarrollo de las mismas varió entre épocas de aplicación y cultivares.Salazar&Lovatt, (1997) encontraron diferentes

respuestas al aplicar ácido giberelico (50 a 100 mg/lt) lo que estimuló una antesis temprana en yemas florales de café (*Coffea arábica* L) acortó el tiempo requerido para la emergencia del 80% de los tallos florales en cebolla (*Allium cepa*) y un efecto dual acelerando la apertura de flores e inhibiendo la iniciación de nuevas flores en fresa (*Fragaria ananassa*). Sin embargo para esta fruta, se reportan algunos efectos positivos delGA3 sobre su producción, entre los cuales esta el acortamiento del periodo entre la siembra y la fructificación, el aumento del número de frutos y la duración del periodo de cosecha. La aplicación de GA3 en fresa puede incrementar la masa y numero de frutos (Choma&Himelrick, 1984; Pérez de Camacaro et al.. 2013, Viasus et al., 2013), aunque también puede reducir la masa de la fruta (Tehranifar y Battey, 1997) Las pulverizaciones con ácido giberélico presentan una actividad promotora del cuajado del fruto, más o menos importante, en algunas mandarinas. El efecto del ácido giberélico es, por el contrario, escaso o nulo en la mayoría de satsumas, naranjas e híbridos (Talon, 2001) En el proceso de cuajado interviene un complejo hormonal, con especial incidencia de las giberelinas. Hay pruebas experimentales en varias especies que demuestran que la GA3 es producida en los óvulos, inmediatamente después de la fecundación y es la responsable del cuajado de los frutos. El estímulo hormonal es generado por el embrión en desarrollo y por el endospermo (o en algunos casos por la partenocarpia), lo que impide la abscisión del fruto y da lugar al crecimiento del ovario y de los tejidos adyacentes. No obstante el equilibrio total de hormonas en la planta también parece afectar al cuajado. Posiblemente, cada especie y variedad requiera de una combinación específica de hormonas para el cuajado.

Farro (2011) al evaluar plantas de camu-camu de ocho años (similar a las condiciones de las plantas en estudio) observo una producción de 3535 flores/planta. . Esta relativa menor producción de flores podría estar relacionado con la fertilidad del suelo, marcadamente diferente entre las dos parcelas. En otra parcela cercana Paredes(2011) registró como promedio general 3469 flores/planta, que se encuentra dentro del rango de las demás parcelas. En general se aprecia que la defoliación no incremento el número de flores, ocurriendo al contrario una reducción de la misma. Reacción diferente encontró Martínez (2010) en el manzano donde la defoliación promovió brotacion e incremento del número de flores. Lo mismo ocurrio con kiwi (LinsleyNoakes, 1989) y también con cereza (Snir y Erez, 1988), nogal pacanero [Núñez y Díaz (1992), Wood(1993)] y con vid (George et al.,1988).

Respecto a la interacción, en la misma Tabla 2, no hubo interacciónentre las dos variables independientes "defoliación" y "dosis de GA3" (F=1,61 p=0,201). correspondiente a la variable dependiente "número total de flores por planta".

En cuanto al número de flores por rama, encontramos un promedio de 146,14, que resulta bajo si comparamos con lo evaluado por Farro (2009) que registró entre 2437 a 3535 flores/rama. Pese a que la edad de las plantas evaluadas por esta autora fue de 7 años versus 9 años en el presente ensayo. Sin embargo hay que considerar las alternancias en la eficiencia productiva de las plantas, especialmente de frutales tal como lo menciona Gautieor&Spichiger (1986). Además, la defoliación en nuestro ensayo ha reducido significativamente, como ya se mencionó, el número de flores. Por otro lado, la unidad de medida señalada como "rama" no es estandarizada y cuya dimensión dependió del criterio del evaluador.

Para otros frutales la defoliación resulto favorable, por ejemplo la aplicación de cianamida de hidrógeno en yemas en reposo de frutales templados promovieron el inicio y compactación de la floración en manzana (Mahomed, 2008), kiwi (LinsleyNoakes, 1989), cereza (Snir y Erez, 1988) nogal pacanero (Núñez y Díaz, 1992; Wood, l993); vid (George, et al., 1988; Zelleke y Kliewer, 1989), pistacho (Pontikis, 1989), frambuesa (Snir, 1983) y nectarino (George y Nissen, 1988).Luego de la defoliacion.

3.7. Frutos retenidos en fase 3 de fructificación

En el análisis de varianza de la Tabla 2 se evidencia para la variable independiente "defoliación"(D) una diferencia altamente significativa (F=25,99 p=0,000) con holgada superioridad de las plantas defoliadasrespecto al porcentaje de frutos retenidos en la Fase 3, aproximadamente a un mes del inicio de la fructificación. También para las dosis de GA3se observa una diferencia significativa (F=3,93 p=0,015) pero una tendencia decreciente de la retención de fruta con el incremento de las dosis de GA3. No resulto una interacción significativa entre los dos factores independientes (defoliación x GA3), lo cual se muestra en la Figura 3.

En la prueba de medias de Tukey (Tabla 5), para la variable "porcentaje de frutos retenidos en la fase F3, destaca el tratamiento testigo por su máxima capacidad de retención de frutos (23,11%) y una tendencia negativa con el incremento de la dosis. Por lo tanto la influencia del GA3 sobre la retención de los frutos resulto negativa; contrariamente a la afirmación de Salazar y Lovatt (1997) que para café, encontró que los altos niveles de AG3 aumentan la retención de frutos.

Table 5. Prueba de medias para porcentaje de retención de frutos decamu-camu en fase 3 de fructificación

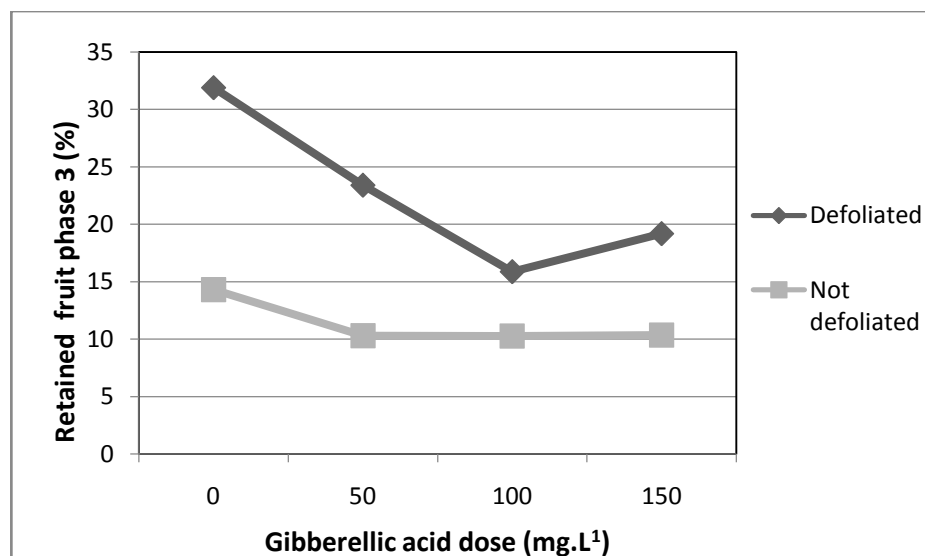| Test	GA3	N	Subset	
	Dose		1	2
	100	12	13,0733	
	150	12	14,7700	14,7700
Tukey	50	12	16,8492	16,8492
	0	12		23,1142
	Sig.		,636	,055

Figura 3. Frutos retenidos en la fase 3 de fructificación (FR3), bajo influencia de defoliación y GA3

3.8. Frutos retenidos en fase 5 de fructificación

En el análisis de variancia (Tabla 2) para frutos retenidos en estado 5 de fructificación (FR5) (Pinedo et al. 2001), se observa que para esta variable no existió diferencia estadística significativa para ninguno de los dos factores independientes en estudio; tampoco hubo interacción estadísticamente significativa entre estos dos factores. El coeficiente de variación alcanzo 50.19% En la Tabla 6 se presenta la prueba de medias (Tukey , 5%); para los factores defoliacion y dosis de GA3. En cuanto al factor defoliación,se confirma la insignificante diferencia de los frutos retenidos en la fase 5 entre plantas defoliadas y no defoliadas. Sin embargo las no defoliadas mostraron una retención de fruta mayor. Respecto a las dosis de GA3, como puede verse en la Tabla 6 la capacidad de retención de los frutos fue similar ocupando el primer lugar el testigo sin aplicación.

Tabla 6. Prueba de medias para frutos retenidos(%) en Fase 5 de fructificación bajo influencia dela defoliación yGA3

Independent variables	Medias	N	e.e.	
Defoliacion				
Sin defoliación	8,08	24	0,80	a
Con defoliación	7,62	24	0,80	a
DoseGA3$_{(mg.L^{-1})}$				
0	8,50	12	1,14	a
150	8,22	12	1,14	a
100	7,78	12	1,14	a
50	6,90	12	1,14	a

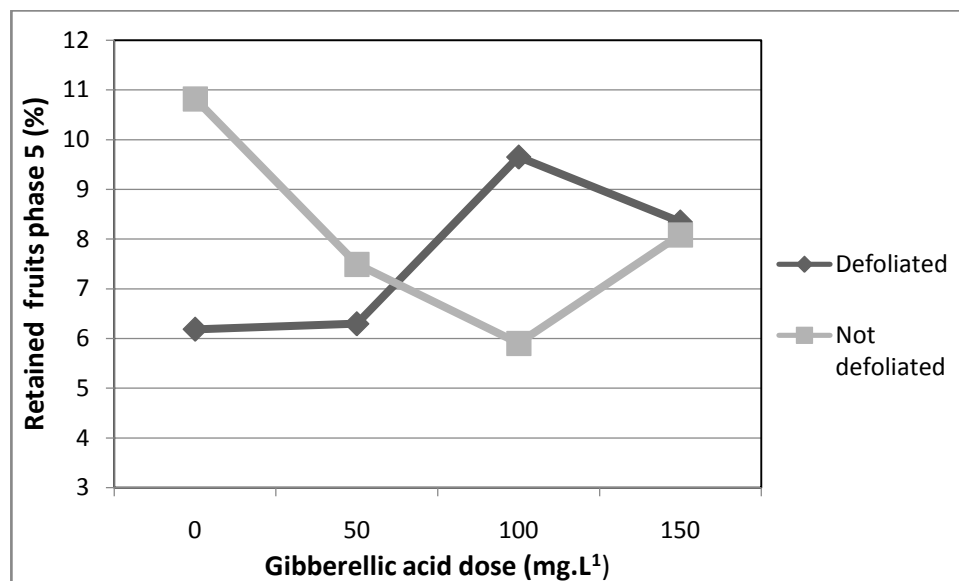Medias con una letra común no son significativamente diferentes, Tukey (p > 0.05) DMS=4,30993

Figure 4, Fruit retained in phase 5 of fruiting (F5), under the influence of defoliation and GA3

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Figure 4, shows a contrast in relation to what is shown in Figure 1, in this phenological period (approximately 1.5 months after the beginning of fruiting) the fruit retention capacity decreased significantly in treatments without defoliation from control ("without defoliation / without GA3"), with the highest level of fruit retention (close to 11%). The negative effect of GA3 on fruit retention was notorious, judging from its decreasing trend in the 50 to 100 mg stretch, then an increase in retention was observed with the 150 mg dose. In this regard, Alvarez et al., (2005) states that in most crops the GA3 increases the mooring of fruits. For the treatments "with defoliation" an interacting and positive effect was presented, since the fruit retention was increased to 9.65% with the dose of 100 mg that seems to be the optimal one since with the extreme dose of 150 mg the retention It fell to 8.35%. Then 100 mg of GA3 in defoliated plants is an interesting option for the purposes of this study. Allana (2002), in apple tree, applied even higher doses of 1000 to 2000 ppm and managed to prevent the fall of the fruit, increase the soluble solids and increase the firmness and texture of the fruit. Nicular (1999), in the case of pear and citrus fruits indica that hormonal treatments are usually used, especially gibberellins to promote the setting of the fruits with acceptable results. However, the author mentions that in many cases they are not satisfactory and that the response to hormonal treatments is not always the same depending on the dosage and the conditions of application.

In this phase 5 of the fructification, the interaction between the two factors (defoliation and dose of GA_3) was more accentuated (F=2,331 p=0,089) without reaching a statistically significant level.

The results regarding the influence of the defoliation and dose of GA_3 on fruit retention or mooring in phases 3 and 5 showed different trends. In phase 3 (of initial fructification) the retention of fruit in the treatments without defoliation was lower than for the treatments with defoliation. In other words, the defoliation induced a greater level of mooring of the fruits (Figure 2). In this regard, Nicular (1999), indicated that the GA_3 stimulates and anticipates the development of the vegetative shoot which would improve the feeding of the fruit set and its subsequent response to the natural falls present. In phase 5 of fruiting, we observed a higher level of interaction between the two independent factors (defoliation and GA_3). Thus, without defoliation the retention of the fruits decreases with the doses of 50 and 100 mg of GA_3. While with the practice of defoliation the retention is reduced to a relatively low value of 6.19%, which significantly improves with the application of GA_3, so that with 100 mg the retention amounts to 9.65% apparently an optimal dose since in the 150 mg dose the retention decreased to 8.35% (Figure 4). In other words, defoliation apparently allowed the positive effect of GA_3, which is consistent with what has been mentioned by several authors. Talon (2001), indicates that the gibberellins in the fruiting phase, reactivate the growth of the fruit, attract nutrients to the fruits and seem to sustain the growth until the fall of June, in this sense the gibberellins are factors that limit and condition the mooring of the fruit. Hence, applying them exogenously increases the mooring percentage. According to Guardiola (2004), in few cases the auxins and cytokinins are involved in the mooring of the fruits. Although he mentions that cytokinins as well as gibberellins increase their concentration in the developing ovaries during the anthesis period as if they were part of the hormonal stimulus that reactivates cell division and stimulates the growth of the fruit making it possible to tie it down. According to Talón (2001) the exogenous applications of auxins to improve the mooring of the fruit are not effective, so its function in the mooring of citrus fruits is unknown. However, when applied in a hormonal complex, as in this case, even though its function is unknown in individual applications, together the three groups of hormones seem to enhance individual effects by increasing the percentages of mooring of the fruit. Aspersions of 20 mg L-1 of gibberellic acid in full bloom did not increase the fruit binding in mandarin 'Monica'. Wallerstein et al. (1973), indicated that the higher concentration of carbohydrates in the aerial part generates a greater quantity of endogenous gibberellins and this is what can improve the mooring of the fruit and even the alternation of the harvests. However, in mango, two applications of GA3 were made in doses of 50 ppm and no favorable results were achieved on productivity except that two harvests were obtained instead of one obtained in the control (Vásquez and Pérez, 2006).

Regardless of environmental factors, Farro (2011), showed that the origin or genetic nature of the material also influences the capacity of retention of the fruits. When comparing the capacity of retention in the phase 2 of fructificación, the basin of the Putumayo River presented greater retention (29,86%), what means that of 100 fruits formed, only 29 arrive until the harvest. While the material from the Curaray River showed a lower retention (22.09%). The author adds that only 5.1% of the flowers reached the stage of fruit ripening (phase 6 of fruiting). So that comparatively, the level of retention achieved in the present trial would be within normal with average values above 5.91% with respect to the number of differentiated flowers. This value is much lower in citrus, for example Agustí and Almela (1991), indicate 1% for orange valence and 0.2% for citrus without seed, and that the increase to more than 1% with the application of a hormonal complex, results a favorable practice.

Talon (2001) concluded that the effect of GA3 on retention of the fruit is scarce or none in oranges. The author indicates that according to experimental tests in several species GA3 is produced in the ovules immediately after fertilization and is responsible for the mooring of the fruits. It also suggests that each species and variety may require a specific combination of hormones for proper retention of the fruits.

.The tie of the fruit according to Guardiola (2004), results from the conjunction of two factors: the concentration of appropriate hormones, which stimulate the growth of the fruit and prevent its abscission and the supply of metabolites sufficient to meet the nutritional needs. A relationship was found between the application of GA_3, senescence, nutritional and health status in some species. In citrus, the application of 20 ppm of GA_3 delays the degradation of chlorophylls and the accumulation of carotenoids in its bark. This effect is associated with a delay in the senescence and delay of the harvest without appreciable losses of its quality (Agustí et al., 1981). Likewise, Gariglio et al. (2002) found that the application of 20 to 200 ppm of GA_3 in medlar was effective for the control of senescence and has allowed to reduce the incidence of numerous physiological alterations, reducing the intensity of the purple spot .Agusti (2013) who worked with AG_3 in several crops applying, found that the same compound can provoke different responses according to the dose and timing application related to the phenological status of the plant. In addition, other factors such as species and variety interact, physiological state of the trees, cultivation system, productive load, irrigation management, fertilization and environmental conditions. In the case of the cherry tree, it caused an increase in the size of the fruit, the greening of the peduncles, the consistency of the fruits, the delay in ripening, the reduction in the cracking of the fruit and the increase in sugars. For the peach tree, it increased the size of the fruit and reduced the flowering, affected the quality and size of the fruits. This author also found that the applications at the time of floral induction inhibit the flowering of the following season, with variations between varieties. The reduction can reach up to 50%. Before or after this time the applications are less effective. According to this author, it is very important that the application be made at the beginning of floral differentiation. Grzesik and Joustra (1991), pointed out that plants treated with AG_3 consume more macroelements than those not treated. They suggest the need to combine AG_3 applications with bottom or foliar fertilization. The lack of nutrients can be the cause of the yellowness observed in the plants treated with the product.

Studies on the causes of the fall of fruit in camu-camu was developed by Farro (2011) and found that insects are not causes of great magnitude and that rather the drop occurs by physiological factors. The author mentions that only 5.1% of flowers and 25.35% of green fruits reach the harvest. While Peters and Vásquez (1986), indicate that it has been estimated that 46% of the flowers of Myrciariadubia are pollinated and that 15% of immature fruits abort before maturity. However, Pinedo et al. (2001) found that the percentage of fruits that drop before completing their development is 73%.

In another test conducted by Farro (2012) found significant effect of insecticide Kalifrut to achieve persistence of the fruits (8.09% persistence versus 5.59% for the control). López, A. (2003), found a relationship between the level of boron and calcium and the fall of fruit in camu-camu in the Pucallpa area.

In many species, the initial development of the fruits occurs at the expense of the existing reserves in the plant after flowering and because of the scarce capacity that the new sprouting still has to provide photosynthates. Therefore, any nutritional deficiency causes the paralysis of growth and very possibly the fall of the fruits. The fall of flowers and small fruits occurs in a very high number, which within some limits, is considered natural, since the plant would not be able to sustain the fruits originated by a normal flowering. In general, it is considered that a harvest is good, if the percentage of fruits harvested with respect to the number of initial flowers is: 5% in pear and apple, 10% in peach, 30% in almond, 8% in plum, 1% in avocado, 4% in citrus, 2% in olive and from 25 to 50%, according to varieties, in vine. Urbina (2002). According to Allana (2002), the application of 1000 to 2000 ppm of GA_3, 45 to 60 days before the harvest of apple tree, exalts the coloration, prevents the fall, increases the soluble solids and increases the firmness and texture of the fruit.

3.9. Weight of fruits

In Table 2, the analysis of variance for average fruit weight shows that there is significant statistical difference both for GA3 dose (F = 2.87 p = 0.0483) and for the defoliation factor (F = 21.06 p <0.0001) without reaching significant levels of interaction between the two factors (F = 1.74 p = 0.175). CV = 15.23%.

Analysis of means was made for the average fruit weight under the influence of gibberellic acid, in which the Duncan test discriminated in two groups, in which the highest weight was registered in the control (average of 6.66 g). Relatively low value In the resulting trend, a decrease in the weight of the fruits is observed with the increase of the GA3 dose. Contrasting results are some of the results reviewed in other fruit trees, such as García-Martínez and Hedden (1997) that indicate that gibberellic acids can promote fruit development after pollination has occurred in several species, which affects its quality and price . With applications of GA4 and GA7, according to the authors, the development of apple trees is stimulated and, in some cases as in citrus, it is possible to delay the senescence in order to keep the fruits longer in the tree or if they are harvested, extend the period of its marketing. However for the case of Agusti orange (1999) did not find significant difference in the weight of fruit treated with GA3, but in the thickness of the shell.

Table 7. Test of stockings for fruit weight of camu-camu

Influence of defoliation and gibberellic acid (sig 0.05).

Test	Treatment	N	Subset	
			1	2
Tukey	Without/Defoliati	24	6,68	
	With/Defoliation	24		5,46
	Dose GA3			
Duncan	150	12	5,6833	
	100	12	5,7583	
	50	12	6,1750	6,1750
	0	12		6,6667

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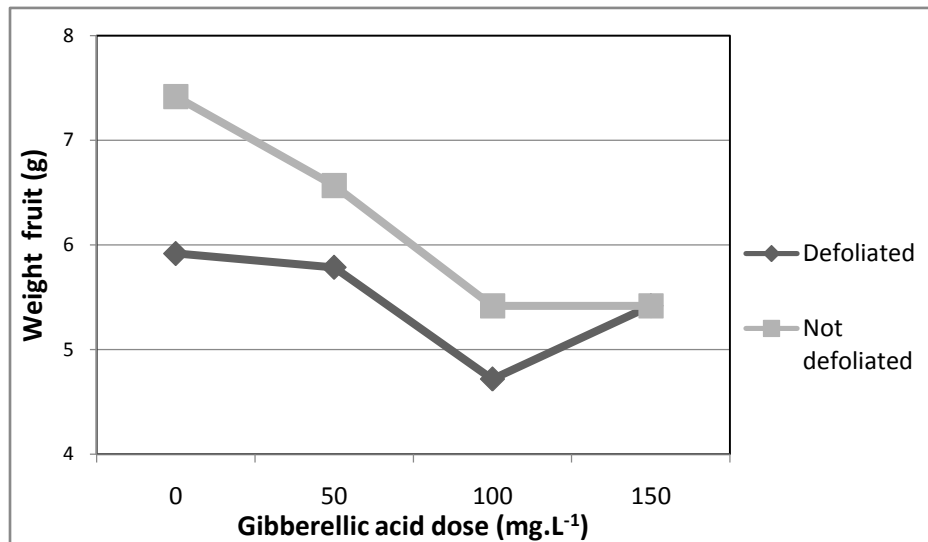
Figure 5. Average fruit weight under the influence of defoliation and GA3

Espindola (2017),for avocado (*Perseaamericana* Mill.), evaluated three production cycles (2003-2006), where.the factors studied were N, AG3 and ringing, with two levels for each factor: 160 and 0 g; 25 and 0 mg; ringed and not ringed, respectively. In the year with low production, the treatments of both N and ringing, increased the initial and final mooring. Combinations of 160 g of N + 25 mg of AG3 + with ringing and 160 g of N + with ringing increased the initial and final retention of fruits, respectively. In the year of high production, the initial retention was increased with the application of N or AG_3, and the final retention with ringing and the combination of 160 g of N + 25 mg of AG3 + with ringing. The combination of treatments showed an additive effect of ringing on the accumulation of glucose, sucrose and fructose in panicles and leaves.

3.10. Fruit yield

By not counting for the dependent variable "yield" with data of uniform variance and normal distribution, the non-parametric Kruskal-Wallis test was applied (See Table W). It was a highly significant difference and yields 4.3 times higher in the absence of GA3 in favor of non-defoliated plants. Regarding the GA3 doses, there was no significant difference between the applied doses with a clearly decreasing trend in non-defoliated plants (Figure 6)

On the other hand, GA3 can cause adverse effects depending on the dose used. For example, Paroussi et al. (2002) report that the application of 200 mg · L-1 of GA3 increased the amount of malformed fruits and aborted flowers; however, Dale et al. (1996) found that the combined application of gibberellic acid and benziladenine (BA) increased production.

Pérez (2015) tells that gibberellins do indeed produce growth, but we must bear in mind that it is mostly via cell expansion. It is also a powerful anti-senescent or retardant of aging, affects the ability to differentiate buds and that these are fertile,what possibly has interaction with the normal maturity of the fruit. Applied to grapes, it was found that the optimum seems to be 2.5 gr / ha because higher doses generate a lot of small fruit that we would have preferred to have dropped.

They were also tested combinations of Auxins + Cytokinins + Gibberellins + P, Ca, Mg and Boron to promote the flowering and mooring of fruits (Yañez, 2002)
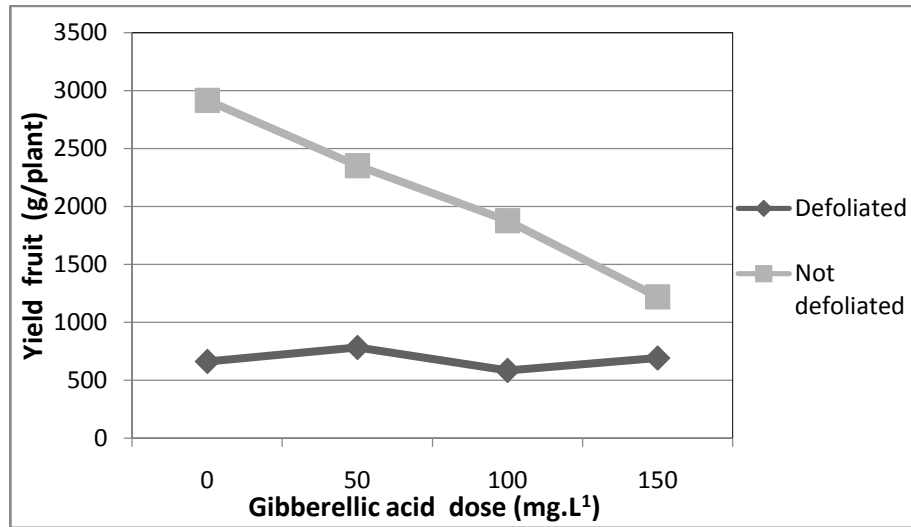

Figure 6. Fruit/plant yield (RF) under the influence of defoliation and GA3

If we consider the real yields of the control plants (without defoliation and without application of GA3), these showed the highest yields, although with a lot of variability whose minimum was 693 g / pl and the maximum of 8640 g / pl with an average of 2916 g / pl. This average value is far from the theoretical projection for plants of 9 years as evaluated that is 11,200 g (Pinedo et al., 2010). The evaluations carried out in the camu-camugermplasm, showed the alternation of the fructification or vecería, where to an intense fructification of a year, corresponds a reduction of the vegetative activity that causes the absence or diminution of the fructification of the following year. To counteract the effect of the vecería, the practices of pruning, thinning of fruits, fertilization are used (Imam, 1996)

3.11. PHENOLOGY OF THE PLANT

Regarding the phenological periods under the influence of the defoliation induced with the saline solution, the following results were obtained:

Table 10. Occurrence of the phenological periods of the camu-camu in relation to the defoliation in a flooded area

Treatments/fhenology	start	end	days
Withdefoliation			
Foliatión	25feb2016	10abr2016	44
Flowering	11abr2016	05may2016	25
Fructificatión	06may2016	25jun2016	50
Total			119
Withoutdefoliatión			
Foliatión	18abr2016	10jul2016	89
Flowering	11jul2016	15ago2016	35
Fructificatión	16ago2016	16oct2016	62
Total			186

There was a noticeable difference in the duration of the total phenological process (foliation to harvest), 119 days for the average of the defoliated plants and 186 days for the non-defoliated plants, which shows a reduction in time of 36.02% due to the effect of the defoliation Counting the period from the differentiation of flowers we find concomitantly a notable difference corresponding 75 days for defoliated plants and 97 days for non-defoliated plants, which means that in non-defoliated plants the delay of the process is of the order of 36.62%. At the beginning of flowering, disuniformity between plants was noted, motivated by several genetic, physiological and environmental causes impossible to control and that are part of the experimental error; aspect also mentioned by Inga (2001). Abanto (2014), found that in conditions of management with fertilization and defoliation, 205 days passed until the harvest that had a period of 36 days. The foliation when applying cyanamide as a defoliant occurred at 25 days in a uniform way and the beginning of floral bud at 105 days after application (Iman and Melchor 2004). The time elapsed from the defoliation with cyanamide to the production of ripe fruits is 120 to 162 days (Inga, 2001, Iman and Melchor, 2004). According to Erez (1990), plants often have prolonged rest or delayed foliation which causes the delay in vegetative and reproductive growth.

4. Conclusion

The influence of the defoliation was negative in the number of flowers / plant, the average weight and the yield of the fruits, being significant and positive in the retention of fruits in phase 3. The influence of the doses of gibberellic acid was significant and negative for the retention of fruit in phase 3, in average weight and fruit yield. There was no significant interaction between defoliation and doses of gibberellic acid for any of the response variables. It should be noted that the phenology is significantly shortened (by 36%, from 6 to 4 months) by defoliation and could be strengthened nutritionally and hormonally to the plant to complement the benefit observed in the retention of fruit in phase 3 and will disappear in the afternoon phases of the fructification, without getting to avoid the loss in the harvest. It is also noteworthy that the 100 mg dose of GA3 in the defoliated plants reached satisfactory levels of fruit retention that could be compensatory due to the higher price of the fruit produced outside the normal harvest period.

Acknowledgement

5.References

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