

OIL PALM POLLEN COMPATIBILITY WITH O_xG HYBRIDS IN ECUADOR

MARÍA RAQUEL MELÉNDEZ-JÁCOME¹; ANDRÉS ALEJANDRO GALVIS-CORREA¹; PAMELA ELIZABETH MANTILLA-VALDIVIESO¹; FRANCO ESTÉFANO-TOBAR¹; MAURICIO ANDRÉS RACINES-OLIVA¹; TREVOR ANTHONY JACKSON² and WILSON VASQUEZ-CASTILLO^{1*}

ABSTRACT

The spread of bud rot disease in oil palm-growing regions of South America has led to the uptake of interspecific hybrids which show some resistance to the disease, but require assisted pollination. A study was carried out to assess pollen viability from commercial oil palm species (*Elaeis guineensis* (G), interspecific hybrids (O_xG)) and *Elaeis oleifera* (O) from the Pacific coast and Amazon regions of Ecuador. *Elaeis guineensis* consistently produced pollen of high viability in the Amazon region (95.0%) and Pacific coast (94.0%), while pollen from *E. oleifera* had high viability when produced in the Amazon region (93.7%) but lower viability from the Pacific coast (53.2%). Pollen from oil palm hybrids had very low viability on the Pacific coast. Another objective was to determine the impact of applying pollen from *E. guineensis* and hybrids on fruit set and parthenocarpy by means of assisted pollination trials in both regions. The application of *E. guineensis* pollen resulted in a higher fruit set in comparison with assisted pollination using hybrid's pollen. A low fruit set was compensated by greater production of parthenocarpic fruits in the hybrids, which increased the final bunch weight. The study provides a guide to pollination in both regions.

Keywords: *Elaeis guineensis*, *Elaeis oleifera*, fruit set, interspecific hybrids, parthenocarpic.

Received: 5 January 2023; **Accepted:** 14 June 2023; **Published online:** 29 August 2023.

INTRODUCTION

Oil palm (*E. guineensis*) is an important commercial crop in humid tropical regions for the production of vegetable oil. The plant requires deep soils, stable high temperatures and high humidity throughout the whole year (Verhey, 2010). There are two geographical origins of *Elaeis* species: 1) the tropical Americas, and 2) West Africa (Durand-Gasselin *et al.*, 2009; Hayati *et al.*, 2004). In Latin America, the most important producers of oil palm are Brazil, Colombia, Costa Rica, Venezuela and Ecuador having a total plantation area of around 257 120 ha (Fedepalma, 2021).

Oil palm production in Latin America is currently threatened by the spread of bud rot disease (BRD), known locally as 'pudrición del cogollo' which is causing severe losses of plants in commercial crops in Ecuador, Colombia, Brazil, Panama and Suriname (Durand-Gasselin *et al.*, 2009; Müller *et al.*, 2006). A well-documented review was published by Meléndez and Ponce (2016). Hybrids of *E. oleifera* × *E. guineensis* (O_xG) can show good agronomic characteristics and tolerance to BRD (Franqueville, 2001), which seems to be inherited from *E. oleifera*, the oil palm native to America (Barba and Baquero, 2012). Hybrid research has been carried out since 1953 (Botelho and Rocha, 1983), with many breeding options studied using Ecuadorian, Panamanian and Brazilian genetic materials (Barba and Baquero, 2012; Teixeira, 2013; Vieira and Lopes, 2010). Hybrid productivity has been shown to be superior to *E. guineensis* species; some hybrids have achieved fresh fruit bunch (FFB) yield of 20 - 30 t ha⁻¹ yr⁻¹ with an oil rate extraction of between 18% and 20% (Vieira and Lopes, 2010)

¹ Ingeniería Agroindustrial, Universidad de las Américas, UDLA. Av. de los Granados E12-41 y Colimes, EC170125 Quito, Ecuador.

² AgResearch, Lincoln Research Centre Christchurch, New Zealand.

* Corresponding author e-mail: wilson.vasquez@udla.edu.ec

or even up to 10 t oil ha⁻¹ yr⁻¹ (Romero and Ayala, 2021). Nevertheless, nutritional studies have been performed to improve yield (Rosero *et al.*, 2013) and more studies covering field management to improve bunch yield and quality have been published (Astorkia *et al.*, 2019; Ochoa-C and Palacio-Mahecha, 2021; Romero *et al.*, 2021; Ting *et al.*, 2020). The OxG hybrids could be a promising solution for oil palm production; however, they have limitations through poor fruit set due to a lack of pollen fertility or low level of insect pollination in commercial plantations (Kumar *et al.*, 2015; Swaray *et al.*, 2021). This problem has been overcome by assisted pollination by hand and using phytohormones (Daza *et al.*, 2020). Nevertheless, these practices have high costs for commercial production and need high numbers of workers that are not always available or well-trained (Li *et al.*, 2019; Ruiz-Alvarez *et al.*, 2021). In Ecuador, commonly used assisted pollination is based on the application of *E. guineensis* pollen to achieve commercial fruit sets; nonetheless, the current use of hybrids raises opportunities to study the compatibility of alternative donor sources and female plant material.

Assisted pollination has been adopted as a systematic practice for increasing productivity in commercial oil palm plantations. Protocols and pollen dosage for increasing yields have been described by Arnaud (1980). However, a low fruit set and productivity of hybrids on commercial plantations have led to pollen studies comparing *E. guineensis*, *E. oleifera* and their interspecific hybrids OxG. A study presented by Sánchez and Romero (2013) showed that the pollen of OxG hybrids achieved only 21.9%-26.3% germination, attributed not only to weather conditions but also to morphological and physiological aspects of the hybrid's pollen grains. In contrast, pollen from *E. guineensis* and *E. oleifera* showed 88% germination (Sánchez and Romero, 2013). The current effect of environmental conditions or the effect of climate change on pollen germination remains to be investigated by the scientific community.

Pollen has been evaluated for its capacity to fertilise female flowers and produce fruit sets. Commercial plantations of the hybrid OxG have always used *E. guineensis* as the pollen donor for assisted pollination, even though there has been limited research on alternatives, such as *E. oleifera* or OxG hybrids, as pollen donors and their effects on fruit set. Previous studies in Latin America on the influence of weather on pollen viability and its capacity to fertilise female flowers were inconclusive (Corrado, 1984). In Asia, pollination using an introduced insect, *Elaeidobius kamerunicus*, increased fruit set and the production of seeded fruits of *E. guineensis* in commercial plantations (Swaray *et al.*, 2021; Syed, 1986).

The pollination of OxG hybrids in commercial plantations in Ecuador has depended on pollen provided by *E. guineensis* and hand pollination (Alvarado *et al.*, 2013; Prasetyo *et al.*, 2014). Alvarado *et al.* (2013) mention that the "Amazon" hybrid in Central America depends on the pollen input of ancient *E. guineensis* palms nearby. Pollen compatibility can be evaluated after the manually assisted pollination of adult palms. Alvarado *et al.* (2013) also showed that the fruit set was higher using *E. guineensis* as the pollen donor compared with using an Amazonian hybrid as the pollen donor. Their research demonstrated the importance of pollen quality and viability and showed that pollen viability of between 15% and 20% limited fertile fruits in the fruit bunch.

The introduction of the bud rot disease has led to research for new resistant/tolerant hybrids, and growers have been encouraged to replant with OxG hybrids, but this is exacerbating the problem of poor fruit set and raises the need for urgent research to address the problem. With high genetic variability among the new hybrids, there is a lack of information about their behaviour and their responses to pollen compatibility. As new hybrids are used for commercial production, there is a need to understand pollen compatibility between species to avoid dependence on only one source of pollen: *E. guineensis*.

The present study was carried out to assess pollen percentage germination rate in vitro, from commercial oil palm species (*E. guineensis* (G), interspecific hybrids (OxG) and *E. oleifera* (O) from the Ecuadorian Amazon region and Pacific coastal region.

In this research, the objective was to determine the effect of different pollen sources on pollination and oil palm yield was examined with experiments in two locations: The Pacific coastal plains and Amazon region headwaters, where flowers from a selection of OxG hybrid oil palm were treated, by assisted pollination, with pollen from different sources. The knowledge of fruit set behaviour submitted to different pollen sources will allow us to understand the impact of assisted pollination and analyse present agronomic management practices in commercial oil palm production. It is expected that the results of this study will guide the selection of donors for artificial pollination for hybrid oil palms in Ecuador and other regions where hybrids are grown.

MATERIALS AND METHODS

Location of Experimental Sites

Studies of pollen germination and compatibility were established in two distinct geographical regions.

Site 1 was the Ecuadorian Amazon region (El Coca, Francisco de Orellana province) at Palmar del Río plantation of 10 000 ha (GPS location: 0° 19' S, 77° 04' W at 290 masl, 3392 mm of annual precipitation, 1327 light hours, maximum temperature: 33.4°C, and minimum temperature: 18.2°C). Site 2 was the Ecuadorian Pacific coast, specifically a 3000 ha site at Energy & Palma plantation, located in San Lorenzo, Esmeraldas province (GPS location: 1° 07' N, 78° 45' 50" W at 500 masl, with 1500-1800 mm of annual precipitation, 1380 light hours, 400 W m² of irradiation, maximum temperature: 34.0°C, and minimum temperature: 27.0°C). Both sites had a plant density of 146 plants ha⁻¹ for *E. guineensis*, *E. oleifera* and OxG hybrids. These two regions were chosen due to the prevalence of bud rot disease (BRD) in oil palm plantations where many growers are cropping OxG hybrids as a possible solution to control the disease. This research was developed with commercial and native species of five-year-old palms in both regions. In the first phase of this study, pollen germination percentage was evaluated in OxG hybrids with commercial oil palm species (*E. guineensis* and *E. oleifera*) as controls. In the second phase, pollen compatibility, fruit set, and production were compared within OxG hybrids.

Determination of Pollen Grain Germination in OxG Hybrids

The pollen germination (%) of three hybrids in Site 1: Coarí x LaMé (CxL), Taisha x AVROS (TxA), and Taisha x LaMé (TxL) and three hybrids in Site 2: Coarí x LaMé (CxL), Taisha x AVROS (TxA) and Unipalma – were evaluated. Unipalma is only cultivated in the Pacific coastal region, and (TxL) only in the Amazon region. Male inflorescences from each of *E. guineensis*, *E. oleifera* and hybrid plants (Taisha x AVROS, Taisha x LaMé and Coarí x LaMé) were selected during the first spate of the opening stage, early on and before the anthesis, corresponding to stage 607 BBCH scale (Moreno and Romero, 2012). No insects were present on the male inflorescences, which avoided contamination with foreign pollen grains from other palm species. The inflorescences were isolated using a double-layer bag (MANGLAR GP-004 (63x83 cm), Green Putumayo) and tied at the peduncle base with a cotton thread (Arnaud, 1980). The bag was revisited every two days to adjust its tightness to the inflorescence peduncle when needed. The male inflorescences were collected during the anthesis phase, then immediately transported to the laboratory located in the plantation site and placed in a chamber at 22°C, 33% RH for 4 hr. After storage, spikes were cut off and gently shaken to liberate pollen. The collected pollen was placed in envelopes and dried in an oven at 37°C for 12 hr. The pollen was sieved through a 40 µm mesh to separate impurities

and then packed in glass tubes (10 mL) and stored for less than two weeks at -20°C, according to the methodology described by Turner and Gillbanks (1974). To evaluate germination, pollen grains were scattered onto Petri dishes containing a simple culture medium (saccharose: 11 g; agar: 1.2 g; distilled water 100 mL). Petri dishes were incubated at 37°C for 12 hr. After incubation, 100 pollen grains were observed under the microscope per sample (Optical Olympus microscope BX51 model), photographed with a camera attached to the microscope (Infinity 2, applying 1:10 fold), and assessed for the presence of a pollen tube emerging from the grain, indicating germination (Myint *et al.*, 2012). Pollen germination was determined for each palm species and the hybrid plants. Pollen was considered germinated when the pollen tube was equal to or greater than the size of the pollen grain.

Statistical Methods

To analyse the germination percentage of pollen grains, a completely randomised block design (CRBD) was applied and an analysis of variance with 95% of reliability was performed. The results were obtained in the statistical software MINITAB 2016. A total of 18 male inflorescences from each species and hybrid were isolated to collect pollen in Site 1, and 25 male inflorescences from each species and hybrid were isolated in Site 2 for comparison. Germination was compared between species and, in the case of *E. guineensis*, *E. oleifera* and (TxA), the differences were estimated according to the sampling region as well. Tukey's test (5%) was performed where appropriate when statistical differences were presented in the Analysis of Variance.

Determination of Fruit Set in OxG Hybrids by Selective Assisted Pollination

Selective assisted pollination was carried out in the two regions (Amazon region and the Pacific coast). In both regions, there are specialised people who work daily only in pollination activities, using standardised practices, and they contributed to this study with their skills. Pollen from selected hybrids (Site 1: Taisha x AVROS, Taisha x LaMé, Coarí x LaMé; Site 2: Unipalma, Taisha x AVROS and Coarí x LaMé) and *E. guineensis* were applied to female inflorescences from oil palms hybrids (OxG) as shown in Table 1.

Female flowers were bagged (MANGLAR GP-004 (63x83 cm), Green Putumayo) individually and pollen was applied according to the standard method for assisted pollination of OxG hybrids described by Turner and Gillbanks (1974). Female flowers during pre-anthesis I (Hormaza *et al.*, 2012) were treated with pollen (10 g of pollen and talcum powder in

a 1:10 mixture), applied with a sprayer inside the bag, which was then sealed. After hand pollination, female inflorescences remained bagged for 110 days. On Site 1, three female flowers from different plants were pollinated with each male treatment; a total of 12 bunches were evaluated. On Site 2, five female flowers were pollinated with pollen from each donor to evaluate a total of 20 bunches.

The total weight of the bunch was recorded on a digital scale, then the fruits were extracted from the bags and classified into seeded, parthenocarpic, and undeveloped fruits. Seeded fruits had seeds, and contained a nut made up of a shell, a kernel, and a mesocarp, while parthenocarpic fruits were formed without pollination, so they did not present an almond. Finally, undeveloped fruits were considered those with white pigmentation, no almond, and no mesocarp, or were one of those that fell from the cluster (Corley and Tinker, 2016; Hormaza *et al.*, 2012). The weight of each category was measured to differentiate the proportions of useable fruits.

Statistical Methods

To compare the results of the fruit set, a randomised complete block design (RCBD) in factorial arrangement 3x4 (female flowers and pollen donors) was carried out on each site and later an analysis of variance (ANOVA) with 95% reliability was performed. The results were obtained using the statistical software MINITAB 2016. A total of 5 male inflorescences from each species were isolated to collect pollen from Site 1 and Site 2 to use as pollen donors for female flowers from oil palm species and hybrids. Three

female inflorescences in the anthesis of each oil palm species, present in each site (1 and 2), were used as pollen receptors (Table 1). The analysis was performed to compare total bunch weight along with the percentage of normal, parthenocarpic, and undeveloped fruits according to the pollen donor. If the ANOVA ($p < 0.05$) presented statistical differences, Tukey's test ($p < 0.05\%$) was performed to determine groups of similar behaviour.

RESULTS AND DISCUSSION

Pollen Germination Rate

The germination rate of pollen collected from commercial oil palm species in the Amazon region was high, with 95.0% germination recorded for *E. guineensis* and 93.7% for *E. oleifera* (Figure 1). High germination percentages (75%-85%) for pollen from these species collected from the Amazon region have also been reported by Myint *et al.* (2012).

Pollen germination rates for O_xG hybrids (TxA (12.2%), CxL (9.7%) and TxL (16.9%)) were significantly lower than for the species (Figure 1), although TxL pollen germination was significantly higher than the other hybrids. Low pollination germination from hybrids was also found in Brazil by Sánchez (2008) and Alvarado *et al.* (2000).

Pollen germination rates from commercial and hybrid materials grown on the Pacific coast are also presented in Figure 1. The pollen germination rate of *E. guineensis* was 94.0%, similar to that attained in the Amazon region, whereas the pollen

TABLE 1. POLLEN APPLIED AS TREATMENTS TO FEMALE OIL PALM HYBRID (O_xG) INFLORESCENCES

| Pollen Donor | <i>E. guineensis</i> | Taisha x AVROS | Taisha x LaMé | Coarí x LaMé | Unipalma |
|-------------------------------|---------------------------------|------------------|---------------|------------------|---------------------|
| Female inflorescences | | | | | |
| Amazon region (Site 1) | | | | | |
| Taisha x AVROS | (TxA) x <i>E. guineensis</i> | (TxA) x (TxA) | (TxA) x (TxL) | (TxA) x (CxL) | - |
| Taisha x LaMé | (TxL) x <i>E. guineensis</i> | (TxL) x (TxA) | (TxL) x (TxL) | (TxL) x (CxL) | - |
| Coarí x LaMé | (CxL) x <i>E. guineensis</i> | (CxL) x (TxA) | (CxL) x (TxL) | (CxL) x (CxL) | - |
| Pacific coast (Site 2) | | | | | |
| Taisha x AVROS | (TxA) x <i>E. guineensis</i> | (TxA) x (TxA) | - | (TxA) x (CxL) | (TxA) x Unipalma |
| Coarí x LaMé | (CxL) x <i>E. guineensis</i> | (CxL) x (TxA) | - | (CxL) x (CxL) | (CxL) x Unipalma |
| Unipalma | Unipalma x <i>E. guineensis</i> | Unipalma x (TxA) | - | Unipalma x (CxL) | Unipalma x Unipalma |

Note: Oil palm hybrids: Taisha x AVROS (TxA); Taisha x LaMé (TxL); Coarí x LaMé (TxL). Coarí- pure *oleifera* from Brazil; Taisha- pure *oleifera* from Ecuador; LaMé- L10T x L2T; AVROS- SP 540 (Compact Seeds and Clones ASD, 2004; INIAP, 2013).

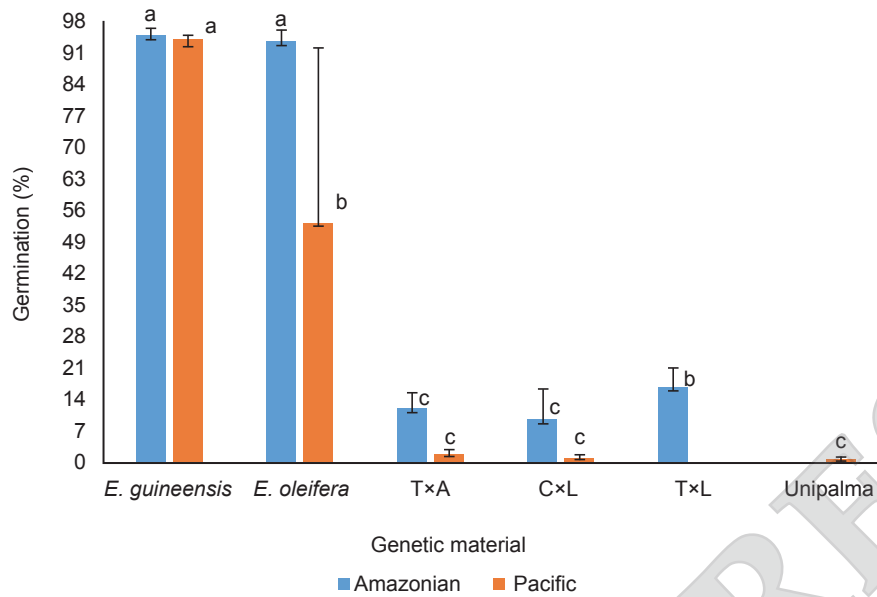


Figure 1. Oil palm pollen germination before storage for commercial species or hybrids in the Amazon region and Pacific coast (Tukey grouping CI 95%).

germination of *E. oleifera* was significantly lower (53.2%). These results agree with those reported by Criollo and Dominguez (2018), where *E. guineensis* showed better pollen quality. Pollen germination from the three hybrids on the Pacific coast was very low (TxA: 1.9%; CxL: 1.1% and Unipalma 0.7%) when compared to the Amazon region. The results indicate that there is a higher percentage of pollen germination in the Amazon region than on the Pacific coast, possibly due to the interaction between climate and genetic composition of plant material, as suggested by Lin *et al.* (2017) and Davarynejad *et al.* (1995) and this study. While the Amazon is consistently humid (annual average precipitation of 3392 mm), the Pacific coast's humidity and temperature can oscillate widely, and rainfall is lower with 1500-1800 mm yr⁻¹ (INAMHI, 2016).

The low germination rate for pollen produced from hybrids indicates the need for manual pollination (Haniff and Noor, 2002) and illustrates the importance of choosing a good pollen donor. One indicator of pollen germination potential might be the morphology of pollen grains and genetic characteristics (Sánchez and Romero, 2013), which was observed by microscopy (Figure 2 and 3). Pollen grain was considered germinated when the pollen tube was longer than the diameter of the grain (Figure 2).

The 10-fold increase in size is due to the microscope settings allowing a better resolution in the measurements. If this is considered, the pollen size is similar to those published by Forero *et al.* (2012) and Sánchez and Romero (2013), ranging from approximately 30.0 to 39.1 μm (Figure 3) in size from both species and hybrids,

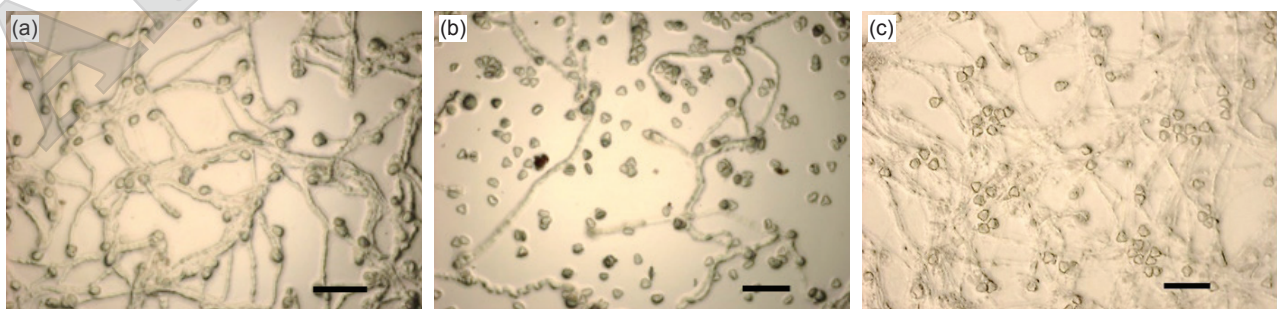


Figure 2. Oil palm pollen germination under optical microscope observation: (a) *E. guineensis*; (b) *E. oleifera*; and (c) Hybrid OxG. Scale bar in black = 2 mm.

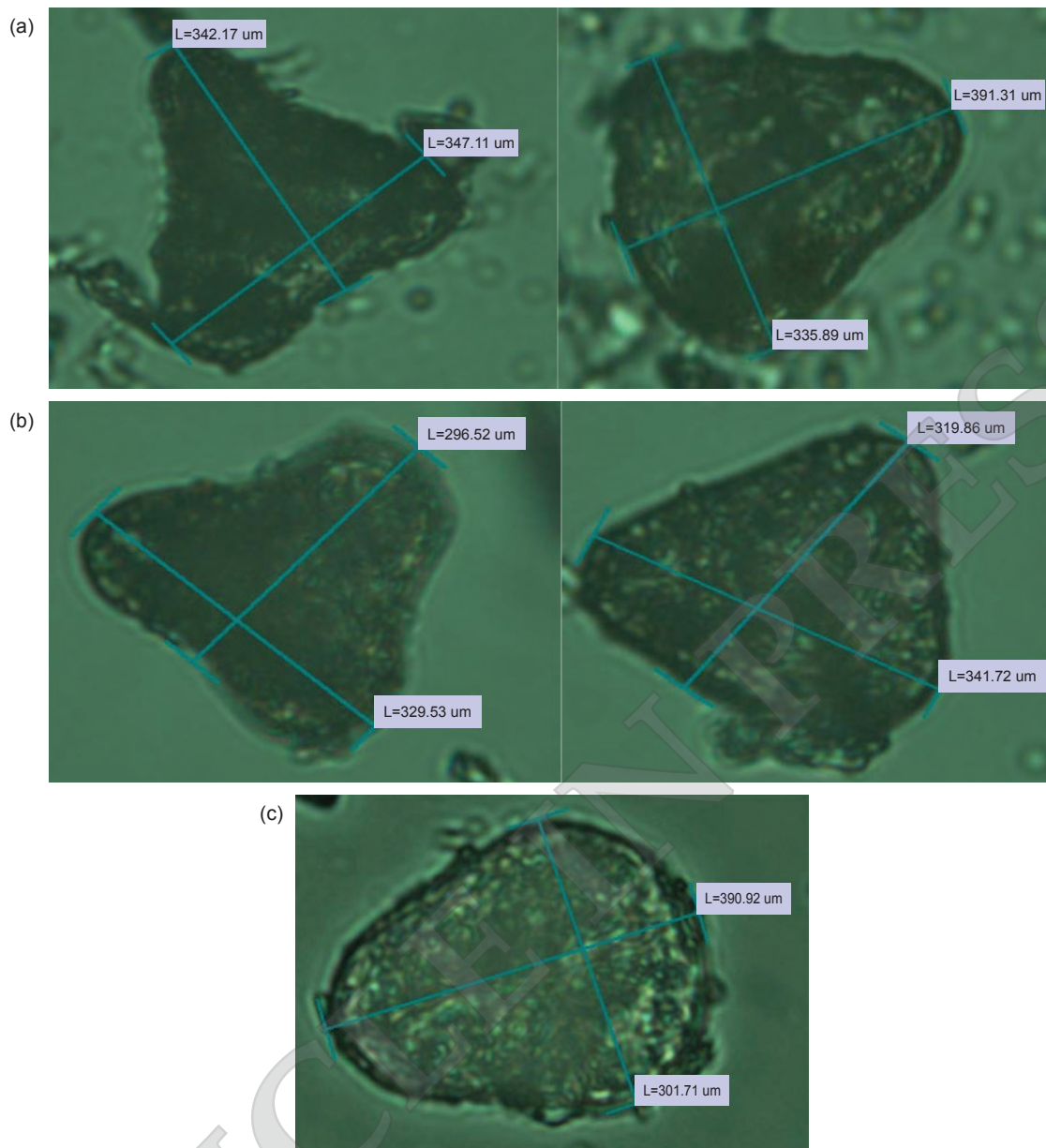


Figure 3. Size and morphology of Hybrids OxG pollen grains 100x: (a) Unipalma, (b) CxL and TxA, and (c) *E. oleifera*.

nevertheless, the morphology of oil palm pollen varied. Hybrids pollen was triangular in shape: Figure 3a and 3b in comparison with *E. oleifera*, which produced elliptical pollen grains (Figure 3c). Similar pollen morphology characteristics in *E. guineensis* were reported in other studies (Myint *et al.*, 2012; Sánchez and Romero, 2013; Tandon *et al.*, 2001). However, pollen from hybrids presented a mixture of pollen shapes and shared morphological attributes from *E. oleifera* and *E. guineensis*, which was also found by Sánchez and Romero (2013). This might suggest a level of incompatibility between parental plants, which would affect germination.

Fruit Set and Yield Obtained After Oil Palm Cultivar Pollination with Hybrid and *E. guineensis* Materials

In the Amazon region, palm flowers from three hybrid palm cultivars (TxA, TxL, and CxL) were treated with four pollen sources (*E. guineensis*, TxA, TxL, and CxL) (Table 1). The highest bunch weight was obtained in the CxL variety (10.79 kg), but all hybrid varieties had high proportions of parthenocarpic and undeveloped fruits. The highest proportion of fertilised fruits was obtained in the TxL hybrid, with 37.9% seeded (Table 2).

TABLE 2. THE EFFECTS OF FEMALE FLOWERS AND POLLEN DONORS ON BUNCH CHARACTERISTICS IN THE AMAZON REGION

| Female flower effect on | | | | | |
|-------------------------|-------------------|--------------------------|----------------------------------|---|-------------------------------|
| | Bunch weight (kg) | Seeded fruits (weight %) | Parthenocarpic fruits (weight %) | Seeded + parthenocarpic fruits (weight %) | Undeveloped fruits (weight %) |
| C×L | 10.79 a | 17.82 b | – | 42.04 b | 14.34 b |
| T×A | 8.9 ab | 24.25 b | – | 34.94 c | 22.92 a |
| T×L | 7.15 b | 37.92 a | – | 57.62 a | 11.35 b |
| Pollen donor effect on | | | | | |
| <i>E. guineensis</i> | 10.41 a | 43.47 a | 10.55 b | 54.02 a | 10.79 b |
| C×L | 9.39 ab | 27.7 b | 21.94 a | 49.64 a | 15.44 a |
| T×L | 8.89 ab | 20.4 b | 18.77 ab | 39.17 b | 19.82 a |
| T×A | 7.19 b | 15.08 b | 21.55 a | 36.62 b | 18.77 a |

Note: Means followed by the same letter are not significantly different using Tukey's test ($p=5\%$).

When pollen source is considered, pollen from *E. guineensis* produced the greatest bunch weight, but this was not significantly ($p<0.05$) greater than that obtained from C×L and T×L pollen donors (Table 2). While bunches treated with *E. guineensis* pollen contained significantly ($p<0.05$) more fertilised (seeded) fruits and less undeveloped fruits, bunches treated with hybrid pollen compensated for fertilisation failure by producing greater proportions of parthenocarpic fruits (18.77%-21.94%), contributing to the overall bunch weight (Table 2).

On the Pacific Coast, flowers from the hybrid palm varieties (T×A, C×L, and Unipalma) were treated with four pollen sources (*E. guineensis*, T×A, C×L, and Unipalma) (Table 1). A significantly ($p<0.05$) high bunch weight was obtained with the T×A variety (11.68 kg), as shown in Table 3, whereas for the rest of the studied variables, there were no statistical differences. Pollen from *E. guineensis* produced the highest proportion of seeded fruits by weight (34.84%), significantly ($p<0.05$) greater than C×L (21.24%) (Table 3).

TABLE 3. THE EFFECTS OF FEMALE FLOWERS AND POLLEN DONORS ON BUNCH WEIGHTS AND SEEDED FRUIT IN THE PACIFIC COAST

| Female flower type | Bunch weight (kg) | Seeded fruit (weight %) | Parthenocarpic fruits (weight %) |
|----------------------|-------------------|-------------------------|----------------------------------|
| T×A | 11.68 a | 28.42 | 24.52 |
| C×L | 8.05 b | 27.91 | 20.15 |
| Unipalma | 6.03 b | 25.51 | 22.72 |
| Pollen donor | | | |
| <i>E. guineensis</i> | 9.56 | 34.84 a | 18.03 |
| T×A | 7.31 | 27.17 ab | 23.72 |
| Unipalma | 8.24 | 25.88 ab | 26.09 |
| C×L | 9.25 | 21.24 b | 22.01 |

Note: Means followed by the same letter are not significantly different using Tukey's test ($p=5\%$). Means without letters belong to the same statistical group.

For oil palm, successful commercial production depends on achieving adequate fruit bunch weight and oil extraction. Both factors are affected by pollination in the Amazon region and the Pacific coast. Pollen germination percentage is an essential factor in determining seeded fruit production (Ruiz-Alvarez *et al.*, 2021), and seeded fruits present an advantage in weight over parthenocarpic fruits (Daza *et al.*, 2020). A higher percentage of seeded fruits is also a desired characteristic for industry, as it facilitates the oil extraction process (Fernández, 2013).

Pollination is a challenge in Ecuador, where the bud rot disease (named PC, for its abbreviation in Spanish) is causing enormous damage to *E. guineensis* commercial plantations. Hence the species is being replaced by the OxG hybrid, which may result in a lack of *E. guineensis* pollen. However, the hybrids require assisted pollination and alternatives to *E. guineensis* pollen are sought.

This study shows that *E. guineensis* pollen achieved the best commercial performance in both the Amazon region and the Pacific coast. As shown in Table 2, 3 and 4, *E. guineensis* is always the best pollen donor in order to obtain a good percentage of seeded fruits and useful commercial fruits. Our results suggest that CxL and TxA could be good candidates for replacing *E. guineensis* as a pollen donor for commercial plantations in the Amazon region, as long as yield is maintained. If there were a lack of availability of *E. guineensis* pollen on the Pacific coast, TxA and Unipalma could be a good alternative pollen donor, as they maintained a high percentage of seeded fruits without significant yield loss, aided by parthenocarpic fruits (Kumar *et al.*, 2015).

On the Pacific coast, the highest yield was obtained from TxA hybrids. In contrast, in the Amazon region, CxL was one of the best materials. Possibly the Brazilian origin of the hybrids CxL

and Unipalma explains their better adaptation to local weather conditions, as these were the first OxG hybrids planted in the Ecuadorian Amazon region (Arias, *et al.*, 2015; Barba *et al.*, 2010), and nowadays is the most prevalent one.

When examined by region, the interaction between female flower and pollen donor (Table 4) shows that high proportions of industrially useable fruits (Seeded + parthenocarpic fruits) can be produced in the Amazon region, but the results are highly variable (67.9% to 23.4% of useable fruits), indicating pollen incompatibility. The highest proportions of useable fruits were achieved from hybrids treated with *E. guineensis* pollen. This can be seen in two crosses: (TxL) × (*E. guineensis*) with 67.9% and (CxL) × (*E. guineensis*) with 53.57%. In the Coastal region, bunch weight presented statistical differences between treatments: (TxA) × *E. guineensis* with 15% in comparison with Unipalma × Unipalma and Unipalma × (CxL) with 4.74% and 5.80%, respectively.

However, there are no differences in the other two parameters (Seeded + parthenocarpic fruits and Seeded fruits), these results suggest that the pollen donor does not influence the percentage of seeded fruits.

The behaviour of oil palm plants depends upon the genetic variability of the parents and the effect of the environmental conditions in each site (Rosero and Santacruz, 2014). Pollen viability and pollination success are probably affected by weather conditions (temperature) and other abiotic aspects that characterise each region, such as relative humidity, the availability of water and sunshine, and soil type (Murugesan *et al.*, 2017), as suggested by various results from the Amazon and Pacific coastal regions. Hence, the information presented in this article is vital for future applications carried out by the industry.

TABLE 4. EFFECT OF THE INTERACTION BETWEEN DIFFERENT OIL PALM MATERIALS WITH VARIOUS POLLEN DONORS ON PRODUCTION (SEEDED + PARTHENOCARPIC FRUITS) IN THE AMAZON REGION AND PACIFIC COAST

| Female flowers × Pollen donors | Bunch weight (kg) | Seeded + parthenocarpic fruits (weight %) | Seeded fruit (weight %) |
|----------------------------------|-------------------|---|-------------------------|
| Amazon region | | | |
| (TxL) × (<i>E. guineensis</i>) | 8.17 | 67.9 a | 60.52 a |
| (TxL) × (CxL) | 6.65 | 55.48 ab | 36.19 abc |
| (TxL) × (TxA) | 6.58 | 54.95 ab | 25.75 bc |
| (CxL) × (<i>E. guineensis</i>) | 11.03 | 53.57 ab | 40.24 ab |
| (TxL) × (TxL) | 7.23 | 52.15 ab | 29.23 bc |
| (TxA) × (<i>E. guineensis</i>) | 12.04 | 48.28 b | 29.65 bc |
| (CxL) × (TxL) | 11.73 | 41.97 bc | 21.80 bc |

TABLE 4. EFFECT OF THE INTERACTION BETWEEN DIFFERENT OIL PALM MATERIALS WITH VARIOUS POLLEN DONORS ON PRODUCTION (SEEDED + PARTHENOCARPCIC FRUITS) IN THE AMAZON REGION AND PACIFIC COAST (continued)

| Female flowers × Pollen donors | Bunch weight (kg) | Seeded + parthenocarpic fruits (weight %) | Seeded fruit (weight %) |
|---------------------------------------|-------------------|---|-------------------------|
| Amazon region | | | |
| (T×A) × (C×L) | 9.87 | 40.6 b | 29.65 bc |
| (T×A) × (T×A) | 6.25 | 27.47 cd | 10.64 bc |
| (C×L) × (T×A) | 8.75 | 27.45 cd | 8.84 bc |
| (T×A) × (T×L) | 7.71 | 23.4 d | 10.18 bc |
| Pacific coast | | | |
| (T×A) × (Unipalma) | 12.84 ab | 27.81 | 27.0 |
| (Unipalma) × (T×A) | 6.38 ab | 26.79 | 23.44 |
| (T×A) × (C×L) | 10.15 ab | 25.85 | 22.35 |
| (C×L) × (Unipalma) | 7.13 ab | 25.56 | 27.46 |
| (T×A) × (T×A) | 8.73 ab | 25.34 | 26.0 |
| (Unipalma) × (Unipalma) | 4.74 b | 24.92 | 23.15 |
| (Unipalma) × (C×L) | 5.80 b | 23.61 | 17.54 |
| (C×L) × (<i>E. guineensis</i>) | 6.47 ab | 19.46 | 28.34 |
| (T×A) × (<i>E. guineensis</i>) | 15.00 a | 19.07 | 38.27 |
| (C×L) × (T×A) | 6.82 ab | 19.02 | 32.0 |
| (C×L) × (C×L) | 11.74 ab | 16.56 | 23.83 |
| (Unipalma) × (<i>E. guineensis</i>) | 7.20 ab | 15.57 | 37.91 |

Note: Means followed by the same letter are not significantly different using Tukey's test (p=5%). Means without letters belong to the same statistical group. SE: standard error.

CONCLUSION

The germination rate was high for *E. guineensis* pollen collected from both the Amazon region and the Pacific coast (95% and 94%) and *E. oleifera* pollen collected from Amazon region (93.7%), whereas *E. oleifera* pollen collected from the Pacific coast site showed a lower germination rate (53.2%). The germination of pollen collected from hybrids was low in both regions. In the Amazon region, germination from hybrids ranged from 9.7% to 16.9%, while on the Pacific coast, the germination was even lower at 1%-2%. The results indicate that the production of viable pollen from *E. oleifera* or hybrids is challenging on the Pacific coast, but viable pollen can be collected in the Amazon region from hybrids, although at lower levels of germination. The results showed that especially for *E. oleifera* and hybrids, the Pacific coast was detrimental to pollen viability, which might be due to climatic conditions. This has important implications for assisted pollination and the selection of pollen donors on the Pacific Coast. Nevertheless,

further studies are needed to confirm the climate hypothesis.

Pollination and bunch yield are affected by both the female flower and the pollen donor. Without assisted pollination, the rate of fruit set in oil palm hybrids is poor. In these trials, the assisted application of *E. guineensis* pollen produced the highest fruit set, but the lack of pollination was, to some extent, compensated for by the production of parthenocarpic fruit. When *E. guineensis* pollen is unavailable, due to the death of plants caused by *E. guineensis* susceptibility to bud rot, our results suggest that new pollen sources such as CxL in the Amazon region and TxA in the Pacific coast could be used to assist pollination in commercial plantations, by virtue of the tolerance that they present to bud rot and the fact that they induce good levels of fruit set at same pollen doses as *E. guineensis*. Moreover, the capacity of oil palm to produce parthenocarpic fruits permits it to have a commercially acceptable bunch weight. This alternative should be analysed to offer new agronomic management alternatives to oil palm producers.

ACKNOWLEDGEMENT

The authors would like to thank the growers for their support in realising this study, as well as ANCUPA of Ecuador for financing part of this study.

REFERENCES

- Alvarado, A; Bulgarelli, J and Moya, B (2000). Germinación del polen en poblaciones derivadas de un híbrido entre *Elaeis guineensis* Jacq. y *E. oleifera* HBK, Cortes. *ASD Oil Palm Papers*, 20: 35-36.
- Alvarado, A; Escobar, R and Henry, J (2013). El híbrido OxG Amazon: Una alternativa para regiones afectadas por pudrición del cogollo en palma de aceite. *Revista Palmas*, 34: 305-314.
- Arias, D; González, M; Prada F; Ayala-Díaz, I; Montoya, C; Daza, E and Romero, H M (2015). Genetic and phenotypic diversity of natural American oil palm (*Elaeis oleifera* (H.B.K.) Cortés) accessions. *Tree Genetics and Genomes*, (11): 122.
- Arnaud, F (1980). Fertilité pollinique de l'hybride *Elaeis melanococca* x *Elaeis guineensis* et des espèces parentales. *Oléagineux*, 35(3): 121-127.
- Astorkia, M; Hernandez, M; Bocs, S; Lopez de Armentia, E; Herran, A; Ponce, K and Ritter, E (2019). Association mapping between candidate gene SNP and production and oil quality traits in interspecific oil palm hybrids. *Plants*, 8(10): 377.
- Barba, J; Orellana, F; Vallejo, G and Manzano, R (2010). Evaluación agronómica de híbridos interespecíficos de palma de aceite OxG (*Elaeis oleifera* x *Elaeis guineensis*) provenientes de diversos orígenes americanos y su tolerancia a la pudrición del cogollo. Primera parte. *Palma*, 3: 11-15.
- Barba, J and Baquero, Y (2012). Híbridos OxG obtenidos a partir de oleíferas Taisha Palmar del Río - (Ecuador) - Variedad - PDR (Taisha x Avros). *Revista Palmas*, 38(1): 1-13.
- Botelho, E and Rocha, J (1983). Avaliação de híbridos interespecíficos de *Elaeis guineensis* x *Elaeis oleifera*. Belém: Embrapa, Brazil.
- Criollo-Escobar, H and Dominguez, J (2018). Germinabilidad y viabilidad del polen de cuatro cultivares mejorados de palma aceitera bajo condiciones de laboratorio. *Rev. Fac. Nac. Agron. Medellín*, 71(1): 8395-8405. DOI: 10.15446/rfna.v71n1.69587.
- Corrado, F (1984). La conformación de los Racimos de la Palma Africana en las Plantaciones de Colombia. *Revista Palmas*, 5(3): 66-87.
- Compact Seeds and Clones ASD (2004). Semillas y clones de palma aceitera de alto rendimiento, San Jose, Costa Rica. 18 pp.
- Corley, R and Tinker, P (2016). *The Oil Palm*. Chichester, West Sussex, Reino Unido: Wiley Blackwell.
- Davarynejad, G H; Szabó, Z; Váci Felhősné, E; Kun, Z and Nyéki, J (1995). Anther and pollen grain characteristics of apricot cultivars. *Acta Horticulturae*, 384: 351-354. DOI: 10.17660/actahortic.1995.384.54.
- Daza, E; Ayala-Díaz, I; Ruiz-Romero, R and Romero, HM (2020). Effect of the application of plant hormones on the formation of parthenocarpic fruits and oil production in oil palm interspecific hybrids (*Elaeis oleifera* Cortés x *Elaeis guineensis* Jacq.). *Plant Prod. Sci.*, 24(3): 1-9. DOI: 10.1080/1343943X.2020.1862681.
- Durand-Gasselin, T; Corredor, J; Sanz, J; De Franqueville, H and Amblard, P (2009). Future vision of oil palm genetic improvement in Latin America. Several resolutions on cooperation in Colombia for planting material improvement. *16th International Oil Palm Conference and Expopalma. Challenges in sustainable oil palm development, 22 to 25 September 2009, Cartagena de Indias, Colombia*. Bogota: FEDEPALMA, (21 vues). International Oil Palm Conference and Expopalma. 16.
- FEDEPALMA (2021). Balance económico del sector palmero colombiano en el primer trimestre de 2021.
- Fernández, C (2013). Experiencias en el procesamiento de racimos de fruta fresca de híbridos oleífera por guineensis, en la Zona Suroccidental. *Palmas*, 34(4): 109-113.
- Franqueville, H (2001). Oil palm bud rot in Latin America: Preliminary review of established facts and achievements. CIRAD. p. 33.
- Forero, D; Hormaza, P; Moreno, L and Ruíz, R (2012). *Generalidades sobre la morfología y fenología de la palma de aceite*. Centro de Investigación en Palma de Aceite (Cenipalma). Bogotá, Colombia. 149 pp.
- Haniff, M and Noor, M (2002). Fruit set and oil palm bunch components. *J. Oil Palm Res.*, 14: 24-33.
- Hayati, A; Wickneswari, R; Maizura, I and Rajanaidu, N (2004). Genetic diversity of oil palm (*Elaeis guineensis* Jacq.) germplasm collections from Africa:

- Implications for improvement and conservation of genetic resources. *TAG. 108*(7): 1274-1284. DOI: 10.1007/s00122-003-1545-0.
- Hormaza, P; Fuquen, E and Romero, H (2012). Phenology of the oil palm interspecific hybrid *Elaeis oleifera* × *Elaeis guineensis*. *Scientia Agricola*, 69: 275-280.
- Instituto Nacional de Meteorología e Hidrología (INAMHI) (2016). Anuario Meteorológico. Quito, Ecuador.
- Instituto Nacional de Investigaciones Agropecuarias (INIAP) (2013). Informe anual del Programa de Palma Aceitera. Estación Experimental Santo Domingo, La Concordia, Ecuador.
- Kumar, K; Mathur, R and Sparjanbabu, D (2015). Efficacy of organic solvents for medium-term storage of oil palm (*Elaeis guineensis* Jacq.) pollen. *Indian J. Agric. Res.*, 49: 516-521.
- Li, K; Tsharntke, T; Saintes, B; Buchori, D and Grass, I (2019). Critical factors limiting pollination success in oil palm: A systematic review. *Agric., Ecosyst. Environ.*, 280: 152-160. DOI: 10.1016/j.agee.2019.05.001.
- Lin, Y; Wang, Y; Iqbal, A; Shi, P; Li, J; Yang, Y and Lei, X (2017). Optimization of culture medium and temperature for the *in vitro* germination of oil palm pollen. *Scientia Horticulturae*, 220: 134-138. DOI: 10.1016/j.scienta.2017.03.040.
- Meléndez, R and Ponce, P (2016). Pollination in the oil palms *Elaeis guineensis*, *E. oleifera* and their hybrids (OxG), in tropical America. *Pesq. Agropec. Trop. Goiânia*, 46: 102-110. DOI: 10.1590/1983-40632016v4638196.
- Moreno, L and Romero, H (2012). Escala BBCH para la descripción del desarrollo reproductivo de *Elaeis oleifera* Cortes H.B.K. in Generalidades sobre la morfología y fenología de la palma de aceite. Cenipalma. Bogotá, Colombia. 150 p.
- Murugesan, P; Aswathy, G; Sunil Kumar, K; Masilamani P; Vinod Kumar and Ravi, V (2017). Oil palm (*Elaeis guineensis*) genetic resources for abiotic stress tolerance: A review. *Indian J. Agric. Sci.*, 87(5): 571-579.
- Müller, A; Furlan, J and Celestino, P (2006). Contribuições da Embrapa Amazônia Oriental para o agronegócio do dendê. Belém: Embrapa. Brazil.
- Myint, K A; Rafii, M Y; Sheikh-Abdullah, S A; Lwin, N M; Din, A M and Latif, M A (2012). Determination of the optimum pollen germination medium for different fruit forms of oil palm (*Elaeis guineensis*). *J. Anim. Plan Sci.*, 14, 14: 1855-1865.
- Ochoa-C, I E and Palacio-Mahecha, N (2021). Contribución al diseño de racimos con ácido α -naftalenacético (ANA). *Revista Palmas*, 42(1): 107-118.
- Prasetyo, A E; Purba, W O and Susanto, A (2014). *Elaeidobius kamerunicus*: Application of hatch and carry technique for increasing oil palm fruit set. *J. Oil Palm Res.*, 26: 195-202.
- Romero, H M and Ayala, I M (2021). Cómo alcanzar 10 toneladas de aceite por hectárea: tecnologías de manejo de los híbridos interespecíficos OxG hacia una producción altamente eficiente. *Revista Palmas*, 42(1): 55-64.
- Romero, H M; Daza, E; Ayala-Díaz, I and Ruiz-Romero, R (2021). High-oleic palm oil (HOPO) production from parthenocarpic fruits in oil palm interspecific hybrids using naphthalene acetic acid. *Agronomy*, 2021(11): 290. DOI: 10.3390/agronomy11020290.
- Rosero, G; Santacruz, L and Cristancho, A (2013). Caracterización de las variables de crecimiento, niveles foliares y de rendimiento en dos materiales genéticos de palma OxG y DxP en diferentes edades de desarrollo de la plantación Guaicaramo S.A. *Revista Palmas*, 34(4): 99-108.
- Rosero, G and Santacruz, L (2014). Efecto de la polinización asistida en la conformación del racimo en material híbrido OxG en la plantación Guaicaramo S.A. *Revista Palmas*, 35(4): 11-19.
- Ruiz-Alvarez E; Daza E; Caballero-Blanco K and Mosquera-Montoya M (2021). Complementing assisted pollination with assisted pollination in oil palm crops planted with interspecific hybrids OxG (*Elaeis guineensis* × *Elaeis oleifera*): Is it profitable? *OCL*, 28: 27. DOI: 10.1051/ocl/2021014.
- Sánchez, A and Romero, H (2013). Viabilidad y morfología del polen de diferentes materiales de palma de aceite. *Ceniavances-cenipalma*. Bogotá. 4 pp.
- Sánchez, G (2008). *Dissertacao_Gilson_Chia 2008 diferencias racimo oleifera*. Manaus: Instituto Nacional de Pesquisas da Amazônia, Universidade Federal do Amazonas. Brazil.
- Swaray, S; Rafii, M; Din Amiruddin, M; Firdaus, M; Jamian, S; Jalloh, M; Oladosu, Y; Mustakim Mohamad, M; Marjuni, M and Kolapo, O K

(2021). Assessment of oil palm pollinating weevil (*Elaeidobius kamerunicus*) population density in biparental *dura* x *pisifera* hybrids on deep peat-soil in Perak State, Malaysia. *Insects*, 12: 221. DOI: 10.3390/insects12030221.

Syed, R (1986). Factibilidad de la introducción de *E. kamerunicus* en Colombia. *Revista Palmas*, 1: 11-15.

Tandon, R; Manohara, T N; Nijalingappa, B H and Shivanna, K R (2001). Pollination and pollen-pistil interaction in oil palm, *Elaeis guineensis*. *Ann. Bot.*, 87(6): 831-838. DOI: 10.1006/anbo.2001.1421.

Teixeira Souza Júnior M (2013). Enfoque y avances del programa de mejoramiento genético de la palma de aceite en Embrapa. *Revista Palmas.*, 34(1): 168-173.

Ting, N C; Sherbina, K; Khoo, J S; Kamaruddin, K; Chan, P and Chan, K L (2020). Expression of fatty acid and triacylglycerol synthesis genes in interspecific hybrids of oil palm. *Sci. Rep.*, 10: 16296. DOI: 10.1038/s41598-020-73170-5.

Turner, P D and Gillbanks, R A (1974). Oil palm cultivation and management. Oil palm cultivation and management. CABI. p. 672.

Verhey, W (2010). Growth and production of oil palm, land use, land cover and soil sciences. *Encyclopedia of Life Support Systems (EOLSS)*. UNESCO-EOLSS Publishers, Oxford, UK. p. 1-24.

Vieira, R and Lopes, R (2010). BRS Manicoré: híbrido interespecífico Caiaué e do dendezeiro africano recomendado para áreas de incidência de amarelecimento fatal. Manaus, Embrapa, Brazil.

ARTICLE IN PRESS