

## **Evaluation of the morphological quality of hybrid seeds and the vegetative development of plants from the two reproductive systems of elite *Duragenitors* from oil palm (*Elaeisguineensis* Jacq.) Seed fields in Côte d'Ivoire**

### **Abstract**

The production of quality oil palm plant material (*Elaeisguineensis* Jacq.) necessarily involves the creation of seed fields made up of elite *Dura* parents. Two methods of reproduction of these elite *Duragenitors* were adopted for the establishment of current seed fields by the National Center for Agronomic Research (CNRA). The first group is made up of elite first-cycle parents who have undergone two successive generations of [(G1) AF] AF type self-fertilization, noted AFAF. The second group is composed of genitors having undergone a prior self-fertilization followed by AFSIB-type recombination cycle of recombination between genitors of the type (G1 × G2) AF, noted AFSIB. This study involves evaluation of morphological quality of seeds from the two breeding systems of the parents and their quality of vegetative development. For this, the seeds from 553 *Dura* parents distributed among eight (8) descendants and grouped within the category C1001 F were used. The results derived from this study have shown that the seeds obtained from the parents through double self-fertilization of AFAF-type parents are characterized by a thin shell and a high percentage of germination. After the elimination of the abnormal plants at the end of the nursery, various traits viz. circumference at the crown, height of the plants and the number of leaves emitted from the plants on the healthy plants showed that the plants resulting from the seeds obtained from self-fertilization followed by AFSIB-type recombination showed good vegetative development and vigor compared to those of AFAF type respectively.

**Keywords:** Hybrid seeds, oil palm, reciprocal recurrent selection, self-fertilization.

### **INTRODUCTION**

To exploit the heterosis existing in the crosses made between certain geographically different genotypes and the constraints due to inbreeding depression, the IRHO adopted in 1957 a reciprocal recurrent selection scheme inspired by work carried out on maize [2]. This selection scheme exploits two groups of populations (A and B) of different geographical origin and with production components of complementary diets. The two (2) populations are intercrossed to obtain A x B hybrids evaluated in the field in descendants tests to assess for

General and Specific Aptitudes to the combination. The best crosses are subsequently reproduced from self-fertilization of the parents. A subsequent cycle is initiated from recombination of the parents and introductions are made to enrich the genetic basis of the material.

Overall, in the last 70 years, two (2) complete selection cycles have been carried out and have made it possible to produce plant material for producers.

If the hybrids of the 1st cycle were obtained from self-fertilization of parents, this is not the case for the 2nd round of selection. Hybrid seeds of the 2nd cycle are obtained from two (2) breeding systems of genitors. This is a group using two (2) successive AFAF-type self-fertilization cycles and a second group obtained from self-fertilization followed by recombination of related AFSIB-type parents. These two groups of Dura genitors coexist in female seed fields for oil palm seed production. The Tenera type (D x P) hybrid seeds produced from crosses between these two groups of elite Dura female parents and pisifera male parents are currently distributed as a mixture by the National Center for Agronomic Research (CNRA) and yield 24 tons / ha / year of bunches and 6 tons of palm oil per hectare in plantation in Côte d'Ivoire under the optimal growing conditions.

[12; 11;10] have shown that during their work, *Dura* progenitors from self-fertilization followed by AFSIB-type recombination have a faster height growth rate, an important agronomic performance and a better quality fruit and bunches than those obtained from double self-fertilization of AFAF type genitors.

Observations arising from these studies create interest in assessing the morphological quality of the seeds and the vegetative development of the plants resulting from the two breeding systems of the parents in order to choose the best to optimize the yield of the plots.

## **MATERIAL AND METHODS**

### **Plant material**

The plant material were composed of five hundred and fifty-three (553) genitors of the *Dura* variety used as female parents in oil palm seed fields in Côte d'Ivoire. *Dura* genitors were obtained either by a double self-fertilization of AFAF-type genitors, or by self-fertilization followed by recombination between related *Dura* palms of the AFSIB type. All these evaluated parents were divided among eight (8) descendants and grouped within the seed category C1001F. Category C1001 F was chosen for its tolerance to Fusarium wilt, therefore used for replanting. All of these palms had the original parents of DA115D x DA115D (Table 1). Within this category, four (4) AFAF-type descendants and four AFSIB-type descendants were evaluated. The parents used were between 21 and 24 years old

## **Experimental dispositive**

The seed fields from which the evaluated genitors originate were established between 1994 and 1997 at the La Mé research station, Côte d'Ivoire. They were planted according to a system in total randomization, at a density of 143 trees per hectare.

## **Variables evaluated**

### ***Morphological qualities of Tenera hybrid seeds from categories***

For the determination of the morphological quality of the selected seeds, the seeds of six (6) descendants out of the eight (8) from this category were analyzed. These were seeds from descendants LM19016, LM18805, LM19121, LM18801, LM17114 and LM18783. Seeds from descendants LM19175 and LM18503 were not evaluated as they were not available during the period because they were not available during the study. Analysis focused on the constituent parts of the palm seed, in particular those involved in its germination. These were the weight of the seeds, the weight of the shell, the thickness of the shell, the weight of the kernel and the percentage of normal embryos of the seeds.

Within the framework of this study, random selection of five (5) parents by single seed descent was conducted. From each of them, one (1) bunch was taken and 20 seeds are evaluated. All the seeds had the same shelf life in the seed preparation and subjected to the same storage temperature which oscillates between 18 and 24 ° C. In total, one hundred (100) seeds per descendants, therefore 600 dry seeds distributed among the six (6) descendants were treated.

### **- Seed weight (g)**

In each descendant, the 100 seed samples collected were weighed individually using an electronic balance. The average seed weight is obtained from the ratio of the total seed weight of the descendants to the total number of seeds.

$$P. \text{ seed} = (\text{PR seed}) / (\text{Number of seeds})$$

P seed: Average seed weight (g)

PR seed: Total seed weight

### **- Shell thickness (mm)**

Once weighed, the 100 seed samples from each descendant are cut crosswise. The almond is then separated from the shell to facilitate thickness measurement. The almond was extracted with a pair of pliers. The thickness of the shell was measured using a digital caliper. Seed thickness was grouped by descendants as before. The shell thickness evaluated in mm was

determined from the ratio between the sums of the measurements made on the total number of seeds

$$\text{Ecoque} = \Sigma mE / (\text{Number of seeds})$$

Ecoque: Shell thickness (mm)

$\Sigma mE$ : Sum of measurements taken

#### **-Hull weight (g)**

After the thickness measurement, all the shells were weighed finally to determine the average shell weight of the seeds of the different descendants. The hulls were weighed using an electronic scale.

The weight of the hull was determined from the following relationship:

$$P_{\text{hull}} = (\text{PR hull}) / (\text{Number of seeds})$$

P. hull: Weight hull (g)

PR hull: Total hull weight (g)

#### **-Kernel weight (g)**

The kernels extracted from each shell were weighed to assess the average kernel weight of the seeds of the different descendants. The weight of the almonds is expressed in grams (g). Weightings were performed using an electronic scale. The average seed kernel weight was determined from the following relationship:

$$\text{Kernel weight} = (\text{PR Almond}) / (\text{Total number of kernel})$$

Kernel weight = Weight of kernel

PR kernel = Total weight of kernel

#### **-Percentage of normal embryos (%)**

After weighing the almond, an opening is made slightly towards the bulge of the operculum to locate the embryo. The almond is then slashed perpendicular to the embryo just below the operculum to expose it. The embryo is obtained by extracting it with a mounted pin. Embryos are arranged in rows of 10 (grouping doubles or triples per nut) in a petri dish with a bottom lined with moistened kraft paper, observed with a magnifying glass. Normal embryos are characterized by swelling and yellow coloration at one end and homogeneous size while abnormal embryos are entirely white and very heterogeneous in size. The percentage of normal embryos was obtained from the following relationship:

$$\% \text{ EN} = (\text{Total Number Seeds} - (\text{MF} + \text{Ab} + \text{NV})) / (\text{Total Number Seeds}) \times 100$$

% EN = Percentage Normal Embryos

MF = Malformed Embryos

Ab = Embryos absent in the almond

NV = Empty seed (without almond)

### ***Evaluation of the germination percentage of Tenera hybrid seeds***

The seed germination percentage was measured in the eight (8) descendants of the C1001F category. Four (4) descendants from the AFSIB type genitor's reproduction system comprising 41390 seeds and four (4) other descendants from the AFAF type genitors reproduction system comprising 40345 seeds.

The percentage of germination was determined by making the ratio between the number of seeds germinated on the total number of seeds put in germination.

% Germination = (Number of germinated seeds) / (Total number of seeds germinated) x100

% Germination: Percentage of germination

### ***Evaluation of the growth characteristics of plants in the nursery from the two descendant's reproduction systems in category C1001F***

For the determination of the growth characteristics of the plants in the nursery of the descendants from the two breeding systems of genitors, the observations were made on the plants from the seeds of the genitors of the C1001F category. The assessment covered 22,981 12 month old plants ready for planting. All these plants arose from 8 descendants, 4 of which were obtained from the reproduction system of AFSIB type genitors with 10,529 plants and 4 other descendants from the AFAF type genitor's reproduction system comprising 12,452 plants. The bags containing the plants in the nursery were placed in an equilateral triangle device (60 cm between the rows and 70 cm between the plants on the same row). At the end of the nursery, non-vigorous offsprings showing symptoms of disease were identified and discarded from each reproductive system of the parents. The study focused on the following variables:

#### **-Plant height (cm)**

The height of all healthy and normal plants (21,484 12 month old plants) was measured from the crown to the end of the longest leaf. The average height of the plants expressed in centimeters (cm) was obtained from the following relationship:

Height = (Sum of measurements) / (Number of plants)

#### **-Circumference at the root collar of the plants (cm)**

The circumference was taken at the collar with a tape measure such that it grips the tree. The determination of the mean circumference at the crown of the offspring plants was obtained from the following formula:

Average circumference at collar = (Sum of measurements) / (Number of plants)

#### **-Number of leaves**

Finally, counting all the functional leaves of each plant made it possible to determine the number of leaves obtained per descendants from the following relationship:

Number of leaves = (Total number of leaves) / (Number of plants)

#### **Statistical analyzes**

Descriptive analyzes (calculations of means, standard deviations, coefficients of variation, confidence intervals) were carried out to describe the different descendants evaluated.

Levene's test was used to test the assumptions of normality and homogeneity of variance variances required for the use of parametric tests. The effect of the reproductive system was evaluated using Student's t-test to verify whether the mode of reproduction of seed-field genitors impacted seed quality and plant development. All of these analyses were performed using Statistica 7.1 software.

### **RESULTS**

#### ***Variation in morphological quality of seeds from the two breeding systems of genitors in category C1001 F***

Statistical analysis Analysis of variance revealed that there was no significant differences in weight of the seeds, the weight of the hulls, the weight of the seeds. Almonds and the percentage of normal embryos among the two breeding systems of genitors in category C1001 F except for shell thickness... Otherwise, the seeds obtained from the genitors of the AFSIB system showed significant differences for hulls of thickness depicting significantly wider thickness (2.71 mm) than the thickness of the hulls of the seeds of the genitors derived from reproduction of type AFAF with 2.60 mm (P = 0.001). The recorded variability was high in the morphological traits of the seeds in the two reproduction systems at the level of the two categories (Table 2).

#### ***Germination performance of hybrid seeds from the descendants of the two breeding systems of genitors in category C1001F***

The results obtained on the seed germination percentage of the two breeding systems of genitors in category C1001F showed that, the seeds from genitors obtained by a double self-pollination of AFAF type genitors recorded a germination percentage significantly higher than

those of descendants derived from self-fertilization followed by recombination of related AFSIB-type genitors ( $P = 0.001$ ). Seeds from AFAF type genitors gave 78.11% germination with a coefficient of variation of 14.80% against 70.67% for seeds from AFSIB type genitors with a coefficient of variation of 29.08%. The recorded variability was low within seeds from AFAF-type reproduction systems than in seeds obtained from AFSIB-type reproduction system (Figure 1).

***Development of seedlings in the nursery from the descendants of the two reproductive systems of genitors in the category C1001 F of oil palm***

Significant differences were observed between crown circumference and the number of leaves emitted from nursery plants from the two breeding systems of genitors ( $P = 0.001$ ). On the other hand, the length of the plants in the nursery did not allow to distinguish the plants produced from the seeds of the two reproductive systems of the parents. At the level of the circumference at the collar, the plants resulting from the reproduction seeds of the AFSIB type genitors gave 12.51 cm against 11.49 cm for the plants resulting from the AFAF type seeds. Regarding the number of leaves emitted, the plants obtained from seeds obtained from reproduction of AFSIB type genitors produced 7.86 leaves against 7.45 leaves for plants obtained from reproduction seeds of AFAF type genitors (Figure 2, 3 and 4).

**DISCUSSION**

Studies carried out on the quality of hybrid seeds from the two (2) reproduction systems of oil palm genitors have shown that in category C1001F, the seeds obtained from genitors originating from double self-fertilization of the AFAF type are characterized by a significantly higher germination percentage than that of the descendants resulting from self-fertilization followed by AFSIB-type recombination ( $P = 0.001$ ). Seeds with a large shell thickness appear to be characterized by a low germination potential. On the germination of oil palm seeds, a process comprising a seed dehydration phase favored by a significant gas exchange has been developed by the CNRA. Studies by [1] have shown that the percentage of germination of oil palm seeds varies from one origin to another. The work of [5] confirmed these results. This author has shown that certain origins of oil palm give only low germination rates regardless of the quality of the pollen. According to him, the percentage of germination of oil palm seeds is linked to the maternal origin of the parents.

But in our case, within the same category C1001F, the germination percentage of seeds from AFAF type genitors was higher than that of AFSIB type genitor's seeds. The low percentage

of seed germination of AFSIB type genitors can be explained by formulating several hypotheses:

-the morphological quality of the seeds. Indeed, the wider thickness of the seed shell from AFSIB type genitors could cause a weak gas exchange which must have prevented a good dehydration of the seeds and a more difficult passage of heat to break the dormancy of the embryo. The effect of heat on improving the germination capacity of seeds has been demonstrated by several authors such as [7; 8; 6]. Particularly for oil palm, [9] has shown for fifty years that heat treatment is essential to break dormancy in seeds.

-the biochemical quality of seeds: in fact, the germination process is always linked to biochemical and metabolic manifestations which allow the embryo to develop into a whole plant [3]. Thus, the low germination percentage of seeds from AFSIB type genitors could be explained by the poor quality of the biochemical constituents of their embryos due to the nutrient deficiency of these genitors high producers of bunch [1].

The germination percentage of seeds from AFSIB type genitors is low, after the elimination of abnormal plants at the end of the nursery, the measurements carried out on healthy plants revealed that the circumference at the neck and the number of leaves emitted are significantly more important in plants derived from seeds obtained from AFSIB type genitors compared to those from AFAF type genitors. This result shows that the plants obtained from the seeds of AFSIB type genitors expressed good vegetative development and better vigor. The good vegetative development of these AFSIB plants could be explained by their good health status [4],

The presence of Tenera trees from the seeds of parents obtained by double self-fertilization of the AFAF type in palm groves could therefore cause a drop in yield due to genetic defects such as disruption of plant vigor (dwarfism) and flower abortion. [4]. On the other hand, the use of Tenera hybrids derived from the seeds of parents originating from self-fertilization followed by AFSIB-type recombination could generate more homogeneous plantations with palm trees that are more genetically compliant. These healthy trees would express greater production potential.

## **CONCLUSION**

The studies undertaken on the evaluation of the morphological quality of seeds obtained from the two reproductive systems of the parents and their quality of vegetative development revealed that in category C1001 F, the seeds obtained from the parents obtained by a double self-fertilization of parents of type AFAF are characterized by thin-shelled seeds and a high



percentage of germination. However, the weight of the seeds, shell, almond and the percentage of normal embryos did not allow differentiation of seeds from genitors obtained from the two brood breeding systems. Also, after the elimination of the abnormal plants at the end of the nursery, the measurements carried out on the healthy plants showed that the plants resulting from the seeds obtained from self-fertilization followed by AFSIB-type recombination presented good vegetative development and good force compared to those of type AFAP, namely: the average circumference at the wider crown, the number of leaves emitted more important with plants of larger sizes. Finally, we can remember that the double self-fertilization of AFAP-type parents does not affect the plants during the first months, that is to say germination and their growth in height, but appears later in production.

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Table 1: Characteristics of *Dura* oil palm genitors studied

Plantation	Category	Original parents	Crosses between descendants of the first generation of categories	Reproductive system	Descendants from the second generation of categories	Number of <i>Dura</i> genitors assessed
1997			( <b>LM2509D</b> ) AFAF	AFAF	LM19175	71
1996			( <b>LM2509D</b> X LM2523D) AF	AFSIB	LM18503	22
1994			( <b>LM3394D</b> ) AFAF	AFAF	LM17114	61
1996	C1001F	DA115D AF	( <b>LM3394D</b> X LM2519D) AF	AFSIB	LM18783	66
1997			( <b>LM2531D</b> ) AFAF	AFAF	LM19016	69
1997			(LM2515D X <b>LM2531D</b> ) AF	AFSIB	LM18805	100
1997			( <b>LM3005D</b> ) AFAF	AFAF	LM19121	79
1997			( <b>LM3394D</b> x <b>LM3005D</b> ) AF	AFSIB	LM18801	85

*AFSIB* = Self-fertilization followed by recombination; *AFAF* = Double self-fertilization

Table 2: Analysis of variance of the yield related components under the two breeding systems of genitors in category C1001 F of oil palm

Catégorie	Reproductive system	P. seeds	CV (%)	<b>E. shell</b>	CV (%)	P. thickness	CV (%)	P. kernel	CV (%)	% EN	CV (%)
C1001F	AFAF	3,13 <sup>a</sup>	40,62	<b>2,60<sup>a</sup></b>	21,24	2,24 <sup>a</sup>	41,98	0,76 <sup>a</sup>	46,90	95,00 <sup>a</sup>	30,46
	AFSIB	3,10 <sup>a</sup>	37,18	<b>2,71<sup>b</sup></b>	20,45	2,29 <sup>a</sup>	34,89	0,72 <sup>a</sup>	38,15	94,33 <sup>a</sup>	22,47

*The means followed by the same letter in the same column are not statistically different at the threshold of 0.05 by Student's t test; CV = Coefficient of variation expressed as a percentage; P = weight (g); E = thickness (mm); EN = normal embryos (%); AFAF: Double self-fertilization; AFSIB: Self-fertilization followed by recombination*

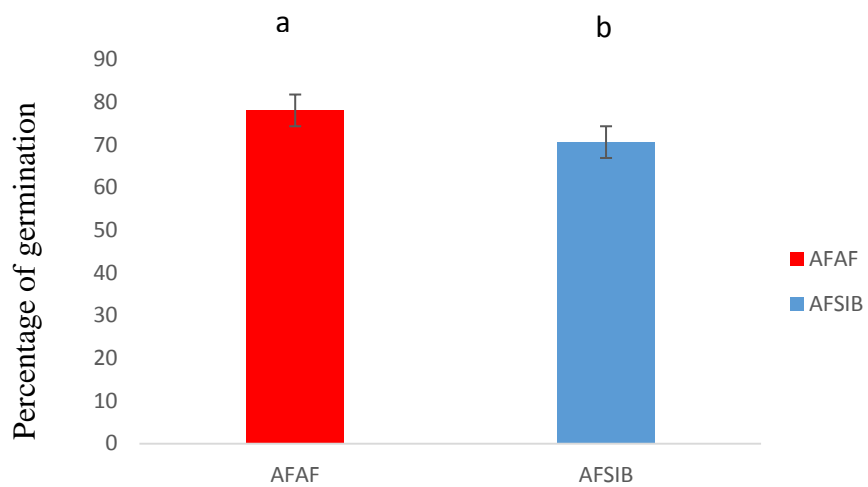


Figure 1: Percentage of seed germination of the two reproductive systems of genitors of category C1001F of oil palm

*AFAF: Double self-fertilization; AFSIB: Self-fertilization followed by recombination*

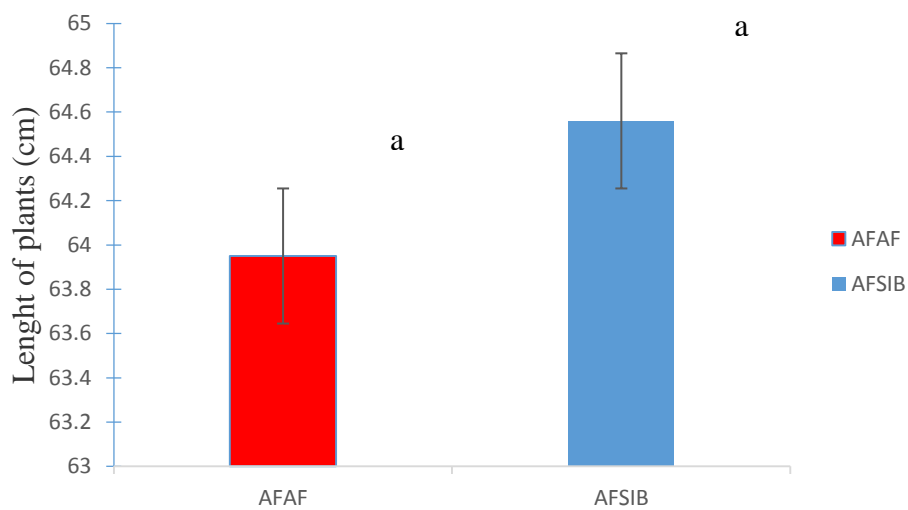


Figure 2: Length of oil palm seedlings in the nursery of the two breeding systems of genitors in category C1001F

*AFAF = Double self-fertilization; AFSIB = Self-fertilization followed by recombination*

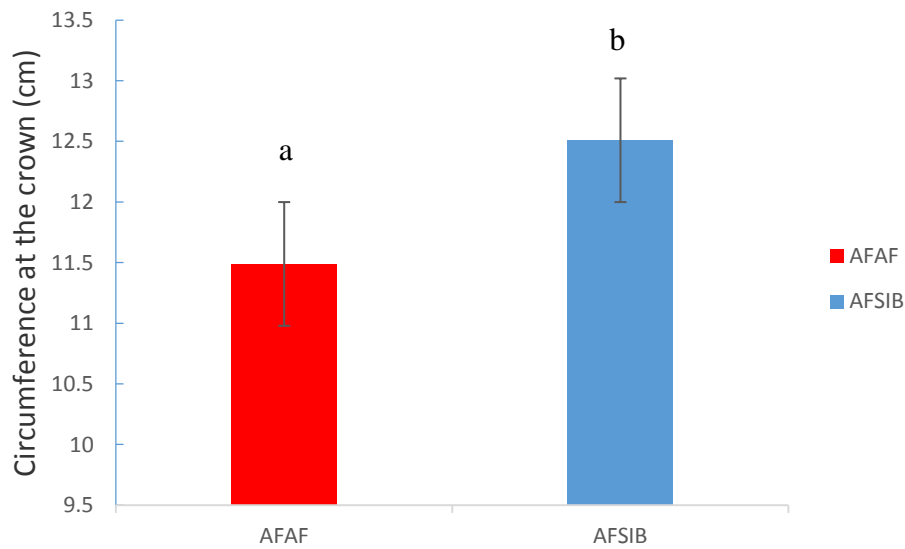


Figure 3: Crown circumference of oil palm plants in the nursery of the two breeding systems of genitors in category C1001F

*AFAF = Double self-fertilization; AFSIB = Self-fertilization followed by recombination*

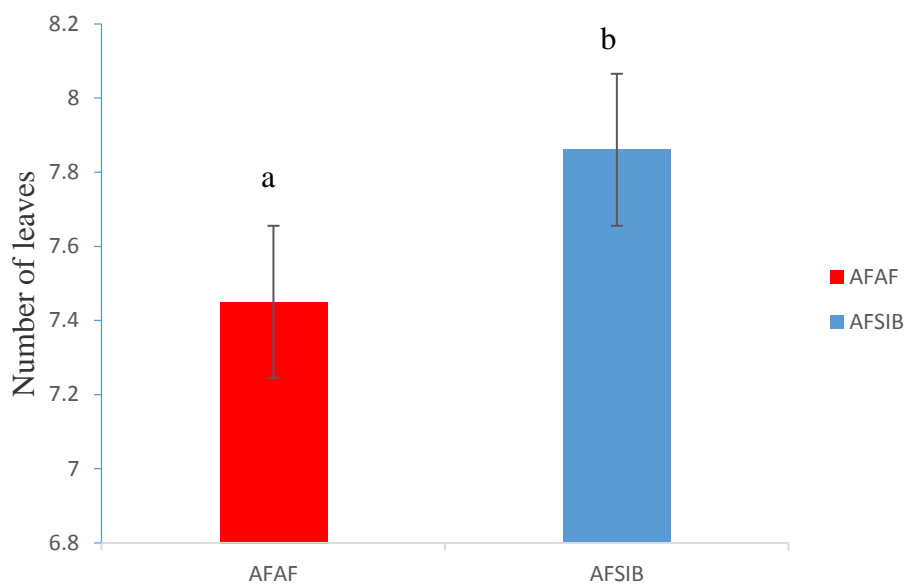


Figure 4: Number of leaves emitted from oil palm plants in nurseries of hybrids of the two reproduction systems in category C1001F

*AFAF = Double self-fertilization; AFSIB = Self-fertilization followed by recombination*