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RESEARCH PAPER

Morphological variations in *pisifera, dura* and *tenera* oil palm seedlings at the nursery: Implications for Marker Assisted Selection (MAS)

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Abstract

***Dura*, *tenera*, and *pisifera* are the three oil palm fruit types differentiated depending on the shell's thickness as well as the presence or absence of fiber rings. These fruit types are recognized when the oil palm starts to produce fruit at ages 3 to 4. Since there are no obvious morphological differences between the *dura*, *pisifera*, and *tenera* oil palm seedlings at the nursery stage, distinguishing between these different fruit forms becomes very difficult, if not impossible. As a result, the vegetative performances of *dura*, *pisifera*, and *tenera* were investigated during the seedling stage. This study's objective was to verify if certain morphological characters could help distinguish between the three oil palm fruit forms at the nursery stage. A three-replication, randomized complete block design was used to set up the experiment. Data on butt circumference, leaf area, number of leaves, chlorophyll content, and plant height were collected and analyzed. Analysis of variance were performed for all the traits and where significance was detected, the means were separated using the least significant difference (LSD). The results indicate that except for butt circumference, all the other traits analyzed exhibited a non-significant difference. The *dura* seedlings (16.86 cm) differed significantly from both *pisifera* (13.51 cm) and *tenera* (13.37 cm) seedlings for butt circumference. The *pisifera* and *tenera*, on the other hand, showed no significant differences. This infers that butt circumference could be further exploited for morphological discrimination of *dura, pisifera, and tenera* fruit forms during the nursery stage. The foregoing results validate the necessity for the use of molecular markers to screen oil palm seedlings at the nursery to identify the different fruit forms before they are planted in the field.**

Keywords: *dura, pisifera, tenera*, marker assisted selection, oil palm fruit form

Introduction

The oil palm (*Elaeis guineensis* Jacq) produces palm oil from its mesocarp. African oil palm accounts for 30.8 percent of global oil and fat output today (Meilke, 2018). The oil palm's versatility in oleo-chemical uses, food production, and biodiesel generation causes it to be a valuable commodity in the food and non-food industries. It yields significantly more oil per hectare than any other crop in the world and has the capacity of meeting the vast and expanding need for vegetable oil, which is anticipated to increase to 240 million tonnes by 2050 (Corley, 2009; Eycott *et al*., 2019). While only 23.6 million tons of rapeseed were produced from 36.4 million hectares of land planted with rapeseed, 56.2 million tons of palm oil were extracted from 17.24 million hectares of land planted with oil palms. Those two commodities' respective production costs were US$ 700 and US$ 850 per metric ton, respectively, showing that palm oil has a lower cost of production than rapeseed (Barcelos *et al*., 2015). Owing to its economical production, extensive application, and extreme productivity, in addition to high profits, the oil palm is a much more efficient, adaptable, and good economic crop than the other vegetable oil crops. In terms of economics, it has the potential to eliminate severe poverty and hunger, emancipating numerous individuals from poverty in Indonesia and different countries where it is mass-cultivated when coupled with sound government policies (Sayer et al., 2012).

In Ghana's agricultural tree crop sector, the oil palm comes second after cocoa and has the potential to develop and disseminate wealth as well as provide jobs, particularly in rural areas where the product is processed into palm oil and palm kernel oil (Owusu-Appiah, 2007). According to future demand and supply projections in Ghana, the current crude palm oil (CPO) supply shortfall will increase from 56, 000 tons to 127,000 tons in 2024 (Commissioners Report, 2020). As a result, there is a significant need to close this gap. Meanwhile, the present supply deficit in West Africa is around 750,000 tons per year and is expected to widen over time, creating a tremendous export market opportunity for Ghana (MASDAR, 2010). This CPO deficit can be addressed by using high-quality oil palm *tenera's*, a hybrid between *dura* and *pisifera* (D x P) in conjunction with strict adherence to optimal management techniques.

Dura, tenera, and *pisifera* are the three fruit types distinguished by the presence or lack of shell thickness and fiber rings. The *dura* is characterized by a thick shell (2-8 mm), a thin mesocarp, and a thick kernel. The fruit has approximately 30% less oil than the *tenera* fruit. In oil palm commercial hybrid production, it serves as the female parent. Sh/Sh (homozygous dominant) is the d*ura* genotype (Forster et al., 2017). The *pisifera* has a thick mesocarp with fiber rings around the little kernel, which is either shell-less or has a shell which is thin. The mesocarp's oil content is high. In the oil palm breeding program, *pisifera's* are used as male parents. The *pisifera* cannot naturally produce fruit bunches and is mostly sterile. *Pisifera* has the homozygous recessive genotype, sh/sh. (Corley and Tinker, 2016; Forster *et al.*, 2017). Hybridization between the *dura* and *pisifera* gives the *tenera.* The thin kernel is encased in a thick mesocarp, which is in turn surrounded by a thin shell (0.5-2mm) with a brown or black fiber ring. There is a lot of oil in the mesocarp (generating over 30% more than *dura* palms). The genotype for *tenera* is Sh/sh (heterozygous). The *tenera* genotype is generally chosen for the establishment of oil palm plantations around the world due to its higher oil yields (Kalyana *et al*., 2017). Although Deli *dura* is the female parent most frequently used in majority oil palm seed production programs, additional populations of *dura* with lower stem growth rates have been established from Angola germplasm (Forster et al., 2017). It has been demonstrated that *dura* transmits to *tenera* progeny all cytoplasmic-controlled genes, bunch production, vertical growth rate, and disease tolerance (Bakoume and Louise 2007).

There are no evident morphological distinctions between the *dura*, *pisifera*, and *tenera* oil palm seedlings at the nursery stage, hence identifying them is difficult, if not impossible at the nursery stage (Babu *et al*., 2017). The difference between these fruit forms may be observed once the palms have started fruiting at about 3-4 years, and this is often done by dissecting or cutting them open to see whether there is any shell thickness or fibre rings present.

Although some studies have been conducted utilizing *tenera* progenies, the vegetative performances of *pisifera, dura*, and *tenera* at the seedling stage have not been examined at the Oil Palm Research Institute (OPRI) and even beyond. Agyei-Dwarko *et al*. (2012) investigated growth parameter variation and correlation analyses in D x P (*tenera*) oil palm seedlings. Darkwah *et al*. (2019) used a Deli *dura* and Aba *pisifera* hybrid to evaluate the mutagenic consequence of gamma irradiation on germination and subsequent seed growth in oil palm. To the best of our knowledge, this is the first experiment that seeks to identify morphological variations in the *dura*, *tenera* and *pisifera* fruit forms at the nursery using growth parameters at the same time.

Marker assisted selection (MAS) is the use of DNA markers to detect the genomic regions implicated in the desired traits of interest. Phenotypic selection is time-consuming, labor-intensive and costly, especially in perennial crops where large tracts of land are needed for phenotyping (Kumawat et al., 2020). MAS is often employed to increase the effectiveness of selection in breeding programs for desired traits. The effective and efficient use of molecular markers in crop improvement programs increases the effectiveness of selection, the level of precision, and speeds up the breeding cycle to create new cultivars with desirable traits (Collard et al., 2008). Many scientists now advise using the oil palm shell thickness gene marker for nursery screening prior to field planting because it has been successfully identified and applied (Darkwah and Ong-Abdullah, 2022; Babu et al., 2017)

Following this advocacy of using molecular markers in screening seedlings of oil palm at the nursery to diagnose discrepancies before they are planted in the field (Babu *et al*., 2017; Reyes *et al*., 2015), there is a critical need to establish vegetative evidence to complement and validate marker assisted screening for oil palm at the seedling stage.

The aim of this research was to assess the vegetative performance of *dura*, *pisifera*, and *tenera* seedlings at the nursery stage and determine if the results would add to the evidence that marker assisted selection approaches should be used in the oil palm identification at the nursery stage before field planting.

Materials and Methods

Experimental site

The study was conducted at the OPRI station, Kusi in Ghana's Eastern Region's Denkyembuor District. The area where the locality is located is 6° 2' 28.644'' N latitude and 0° 51' 34.92'' W longitude in the agroecological zone of semi-deciduous forest from January 2020 to January, 2021.

Plant material and seedling establishment

One hundred (100) sprouted seed nuts of oil palm each of *tenera, dura,* and *pisifera* were obtained from OPRI, Ghana. All the germinated seeds were planted at the same time at the OPRI nursery under 55 % light penetration in 12.7cm x 20.32 cm black polybag filled with a mixture of topsoil and sand in the ratio of 2:1. Details of the progenies used for the experiment are shown in Table 1. The topsoil utilized to fill the bag belonged to the Kokofu Series (Ferri- Plinthic Acrisol - FAO/UNESCO classification). The lower third of the bag was perforated to enhance the drainage of excess water. After four months in the prenursery, the seedlings were transferred to the main nursery in a black polybag with the measurement (35.6 cm x 45.7 cm) using the above-mentioned soil. Bags were arranged in a 90 cm triangular pattern.

Table 1: Progenies used for the experiment

Fruit type	Progeny	Parentage	Origin
Dura	3957D	5.2153D x 5.642D	Deli
Pisifera	3327P	851.805P selfed	Calabar
Tenera	Cross 132	5.37D selfed x 4.17T selfed	Deli x Calabar

Cultural practices

The seedlings were hand watered every other day. Fertilizer application was performed using sulfate of ammonia at a rate of 25g to 15 liters of water during the first four months of seedling establishment. After four months, 30 g of NPK 30:10:10 were applied to each seedling every month for eight months (OPRI, Recommendation). The seedlings were mulched with sawdust to enhance the conservation of moisture. Regular weed control was done by manually removing weeds from the polybags as and when weeds start growing and by applying glyphosate to the space between subplots at a rate of 100 mls per 15 litres of water at intervals of three months. Plants were sprayed with 14 g of Dithane M45 (Mancozeb 800g/kg) in 9 liters of water once a week to control diseases. Weekly applications of "Dursban 4E" (480 g/l Chlorpyrifos) pesticide at a rate of 100 ml to 15 liters of water were used to control insect pests.

Data collection

Data were collected from 10 bordering plants that were chosen at random from each plot. Five months after planting (MAP), data collection began. It was done monthly for a period of one year. The following data were collected:

Plant height (cm)

At monthly intervals between 5 and 12 MAP, measurements of plant height were made using a measuring tape from the soil level in the polybag to the tip of the longest leaf.

Number of leaves

The number of leaves on the randomly chosen seedlings was counted each month to calculate the number of leaves per plant.

Leaf area (cm)

For the leaf area, a non-destructive technique was employed. On each of the recorded plants sampled per plot, the length and widest point of each leaf were measured with a ruler. After that, the formula utilized by Darkwah et al. (2019) adopted from Hardon (1969) was used to estimate the leaf area.

Butt Circumference (cm)

For the butt circumference, two points on the butt that was 1 cm above soil level had their diameters measured using a set of vernier calipers. At each month's interval, measurements were made, and the formula πd was used to calculate the butt circumference. Where d is the average diameter measured and π was taken to be 3.14.

Determination of chlorophyll content

A chlorophyll meter (SPAD 502 Plus) was used to measure the amount of chlorophyll. The procedure stipulated by Sim *et al.* (2015) and slightly modified by (Darkwah *et al.,* 2019) was followed. This determination was made using intact leaf samples from frond number 3. Using distilled water, the leaf blade's surface was cleansed. The leaf was put between the arm and the sensor, and the amount of chlorophyll in three randomly chosen leaf spots about the midpoint of each leaf blade was measured. Figures displayed on the screen were recorded as the chlorophyll content measured in ug/g.

Experimental design and analysis

The experiment was set out in a randomized complete block design involving three treatments (*dura*, *pisifera* and *tenera*) and three replications with a plot size of 30 palms were used. Data collected were averaged for eight months and then subjected to Analysis of Variance (ANOVA) using GEN STAT software (VSN International, 2022). Where significant differences were observed, the least significant difference (LSD) was used for the treatment means separation.

Results

The results of the ANOVA are shown in Table 2. No significant difference was observed for all five traits studied except for butt circumference. The highest coefficient of 27.3% variation was recorded by leaf area. *Dura* oil palm seedlings had the highest butt circumference (16.86) and were significantly different from the *pisifera* and *tenera* seedlings (Fig. 1). Between the *pisifera* and *tenera*, no significant difference was observed.

Table 2: Variability in *dura, pisifera*, and *tenera* oil palm seedlings

Trait	Minimum	Maximum	Mean	CV (%)	MS	F. pr
Butt Circumference (cm)	13.37	16.86	14.58	2.8	11.6869	0.017
No. of leaves	8.477	8.777	8.601	1.5	0.07338	0.498
Leaf area (cm$^{2)}$	2248.00	2282.00	2075.00	27.3	325309	0.595
Plant height (cm)	74.00	84.90	78.80	3.8	93.10	0.253
Chlorophyll content ug/g	49.20	52.90	50.80	3.2	10.39	0.743

CV – coefficient of variation MS – Mean square F. pr – probability at 5 %

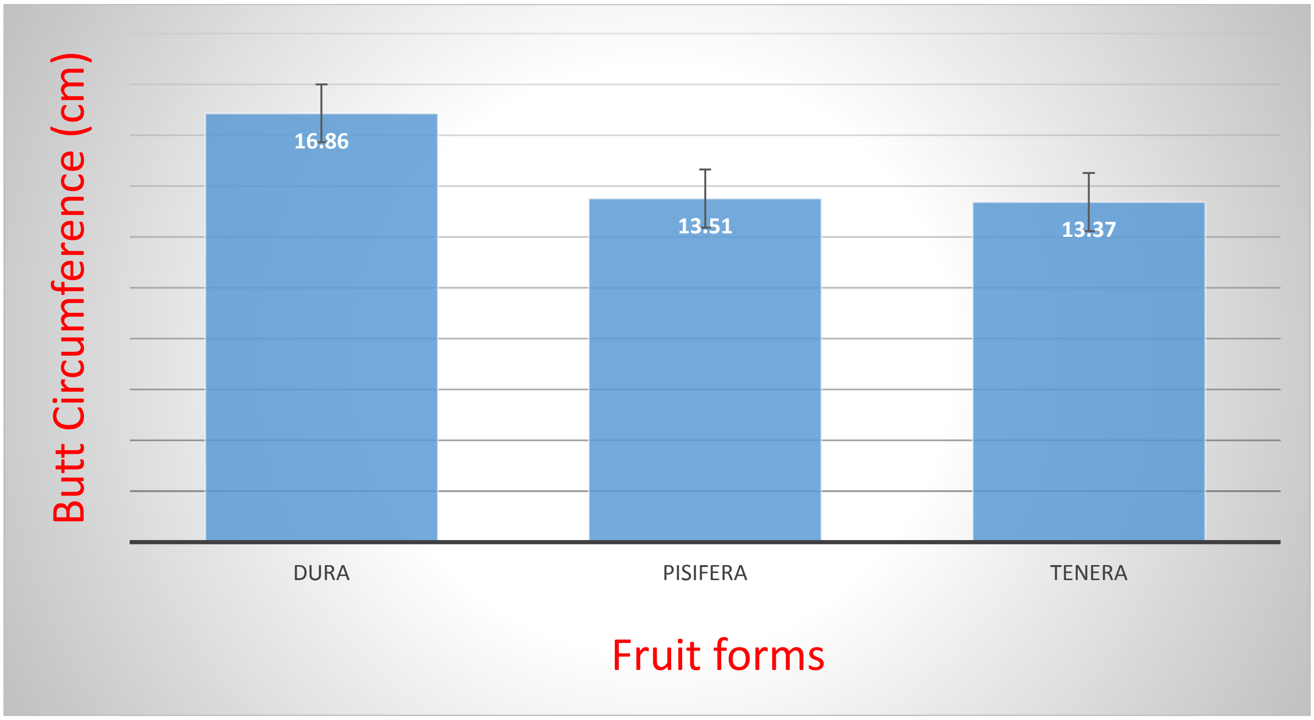

Figure 1: Mean butt Circumference (cm)

Differences observed among the treatments with respect to the number of leaves, leaf area, plant height, and chlorophyll content were not significant (Table 3). Although no significant differences were observed, *dura* seedlings had the highest value (84.90 cm) while *tenera* recorded the least (74.00 cm) in absolute terms for plant height. For chlorophyll content, the *tenera* treatment had the highest mean value (52.90 ug/g) while the least value (49.20 ug/g) was recorded by *pisifera* in absolute terms. The mean leaf area and the number of leaves were almost comparable in absolute terms for all the treatments.

Table 3: Mean values for the number of leaves, leaf area, plant height, and chlorophyll contents

Fruit forms	**Means of traits**			
	Number of leaves	Leaf area (cm)	Plant height (cm)	Chlorophyll content
Dura	8.777^a	2282.33^a	84.90^a	50.30^a
Pisifera	8.477^a	2248.00^a	77.50^a	49.20^a
Tenera	8.550^a	2282.37^a	74.00^a	52.90^a

Discussions

The oil palm depending on its shell thickness and fibre rings is classified into three, namely *dura, pisifera* and *tenera.* Babu *et al*. (2017) indicated that identification of the *dura*, *pisifera,* and *tenera* oil palm seedlings at the nursery stage is challenging and or almost impossible since there are no obvious morphological differences between them. Notwithstanding, at the nursery, oil palm growth has been measured using a wide range of different parameters. Among such parameters with physiological significance include leaf area, number of leaves, plant height, butt circumference (Agyei Dwarko *et al*., 2012; Darkwah *et al*., 2019). While photosynthetic potential can be estimated from leaf area, the number of leaves relates to the activity of the apical meristem, the butt circumference relates to the adventitious roots that would be developed, and the plant height together with average spread and rachis length is a determinant of spacing and planting density in industrial plantations (Murugesan and Shareef, 2014; Corley and Tinker, 2016).

In this study, significant differences were observed in butt circumference for the various fruit forms. This may be attributed to the genetic makeup of each of the three fruit forms. Danso *et al*., (2012) indicated a considerable disparity in the circumference of the butt among oil palm seedlings 12 months after planting (MAP). This current study corroborates the findings of Danso *et al.*, (2012) and Darkwah *et al*., (2019). According to Tan *et al. (*2014), differences observed among seedling growth are attributed to fertilizer application, absorption of sunlight, carbon dioxide concentration, temperature, and water availability. In this study, all the treatments were equally treated and therefore any difference being observed may be genetic.

No significant difference was observed in leaf chlorophyll content for all the treatments. While the synthesis of carbohydrates is regulated by light and chlorophyll, photosynthesis is a key factor in creating plant assimilates and is determined by the amount of CO_2 and water that is available. High levels of leaf greenness are positively correlated with high levels of chlorophyll in plants and have a close relationship to nitrogen. In order to create assimilates (sugar), dark green leaves must be able to sustain their rate of photosynthesis while growing (Wahyuti *et al*., 2013). Darkwah *et al*. (2019) reported no significant difference in chlorophyll content for oil palm seedlings and the figures reported in this present study were comparable. This may imply that the chloroplast which is the organ for the absorption of light was intact for all the treatments.

The amount of leaf area impacts a crop's ability to intercept light, which is a factor in estimating plant productivity (Koester *et al*., 2014). Darkwah (2018) revealed a substantial positive association between oil palm yield and leaf area. In this study, no significant difference was observed for all the treatments for the leaf area. This may imply that the treatments had high photosynthetic potential.

Although no significant difference was observed for plant height, *tenera* seedlings had the lowest height in absolute terms. *Tenera* is a cross between *dura* and *pisifera* parental genetic material (Nakonechnaya *et al*., 2013). Offspring produced from such a cross have intermediary behaviors of the two crosses.

Implications for marker-assisted selection

The results obtained from this study indicate that seedlings of *dura*, *tenera* and *pisifera* are very morphologically similar in the nursery. This similarity poses a challenge in distinguishing them at the nursery and this should necessitate the use of marker-assisted selection as standard operation. Admixture and other human errors like accidental use of pollen from a non-pisifera palm, open pollination of *dura* parental palms by nearby *dura* palms, imprecise seed or seedling selection and self-pollination of *dura* parental palms, have all contributed to contaminations at the plantation and institutional levels (Corley, 2005). Ooi *et al.*, (2016) reported an average of 10.7% non-tenera contamination at separate planting locations around the 6 MPOB research stations in Peninsular, Malaysia. In Ghana, Commissioners Report, (2020) indicates that about 70 % *dura* and *pisifera* contamination was obtained following a survey of 97 smallholder farms cultivated on mined areas in three Districts of Central regions using seeds that were uncertified given to them by private nursery operators. *Dura* and *pisifera* contaminations in commercial plantations partially account for the substantial discrepancy between actual and theoretical yield both at the global and national scale. Globally, the theoretical annual yield of oil palm is expected to be 18.5 t/ha, however commercial plantations actually produce 12 t/ha and their productivity has stagnated at 3 t/ha on average. (Woittiez *et al*., 2017). In Ghana, the potential yield of FFB is 21 t/ha/yr, but only 11 t/ha/yr and 6.0 t/ha/yr are actually realized in commercial plantations and smallholder farms, respectively (Rhebergen *et al*., 2018). This represents a yield decline of around 50 and 71 percent for these two types of plantations. This decline in yield is attributed both to the planting of uncertified seeds and non-adherence to best management practices.

Marker-assisted selection is now possible due to the development of molecular markers. MAS can be performed at the nursery before field planting to ascertain the certainty of the material planted. With the right enforcement in place, the usage of these technologies might be utilized as a certification mechanism to ensure that seedlings are verified before being planted, with the *tenera* seedlings being planted on commercial fields and the breeding materials (which can be *tenera, dura or pisifera*) planted on breeding fields. This will assist in bridging the gap between potential and actual yield.

Conclusions

Except for butt circumference, all the other traits analyzed exhibited a non-significant difference. The findings of this investigation provide validation for the use of MAS to screen oil palm seedlings at the nursery to discover differences before they are planted in the field. Butt circumference could be further exploited for morphological discrimination of *dura, pisifera, and tenera* fruit forms at the seedling stage.

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References

Agyei-Dwarko D, Ofori K, Kaledzi PD, (2012). Variation and correlation analysis of growth parameters in D x P oil palm (Elaeis guineensis Jacq.) seedlings. Elixir International Journal, 47:8946 - 8949.

Babu KRK, Mathur P, Naveen Kumar D, Ramajayam G, Ravichandran MVB, Venu S, Sparjan B, (2017). Development, identification and validation of CAPS marker for SHELL trait which governs dura, pisifera and tenera fruit forms in oil palm (Elaeis guineensis Jacq.). PLoS ONE 12(2): 1-16

Bakoumé C, Louise C, (2007). Breeding for oil yield and short oil palms in the second cycle of selection at La Dibamba (Cameroon). Euphytica 156: 195-202

Barcelos E, Rios SA, Cunha RNV, Lopes R, Motoike, SY, Babiychuk E, Skirycz A, Kushnir S, (2015). Oil Palm natural diversity and the potential for yield improvement. Frontier Plant Science, 6:190- 206.

Collard BC, and Mackill DJ. (2008). Marker assisted selection: an approach for precision plant breeding in the twenty first century. Philosophical Transactions of the Royal Society B: Biological Sciences. 363(1491):557-72.

Commissioner's Report (2020). Report on Oil Palm "Dura" contamination and Agronomic practices on Minerals Commission's Sustainable Livelihood Oil Palm Project in the Central Region, Ghana. Commissioners Report.

Corley R, (2009). How Much Palm Oil Do We Need? Environmental Science and Policy, 12: 134 - 139.

Corley RH, Tinker PB, (2016). The oil palm. 5th Edition. Blackwell Science Ltd, Oxford, UK. Pp 217- 530.

Corley RHV, (2005). Illegitimacy in oil palm breeding – A review. J. Oil Palm Res. 17: 64–69

Danso F, Opoku A, Baidoo-Addo K, Danso I, Afari PA, Nuertey BN, (2012). Improving the growth of oil palm seedlings with biostimulants NEB-26 and NEB-29. J. Ghana Science Association, 14(1): 46-52

Darkwah DO, Blay ET, Amoatey HM, Agyei-Dwarko D, Sapey E, Ong-Abdullah M, (2019). Mutagenic effects of gamma irradiation on oil palm (Elaeis guineensis Jacq.) seedling germination and growth. Journal of Oil Palm Research, 31(2): 212–219

Darkwah DO, Blay ET, Amoatey HM, Agyei-Dwarko D, Sapey E, Osei SA, (2019). Study of Chlorophyll Mutations and Chlorophyll Content in Young Oil Palm (Elaeis guineensis Jacq) after Gamma Irradiation. International Journal of Plant Breeding and Crop Science 6(3): 575-580.

Eycott AE, Advento AD, Waters HS, Luke SH, Aryawan AAK, Hood AS, Turner EC, (2019). Resilience of ecological functions to drought in an oil palm agroecosystem. Environmental Research Communications, 1(10): 001-015.

Forster BP, Sitepu B, Setiawati U, Kelanaputra ES, Nur F, Rusfiandi H, Rahmah S, Ciomas J, Anwar Y, Bahri S, Caligari PDS, (2017). Oil Palm (Elaeis guineensis Jacq.) In: Genetic improvement of tropical crops. Campos, H. and Caligari, P.D.S (eds). Springer International Publishing. Switzerland. Pp 241-291

Hardon JJ, Williams CN, Watson I, (1969). Leaf area and yield in the oil palm in Malaya. Experimental Agriculture. 5:25-32

Koester RP, Skoneczka JA, Cary TR, Diers BW, Ainsworth EA, (2014). Historical gains in soybean (Glycine max Merr.) seed yield are driven by linear increases in light interception, energy conversion, and partitioning efficiencies. J. Exp. Bot. 65, 3311–3321.

Kumawat G, Chandra K, Saurabh P, Chand S. (2020). Insights into Marker Assisted Selection and Its Applications in Plant Breeding. DOI:http://dx.doi.org/10.5772/intechopen.95004

Meilke T, (2018). Oil World. International Statistical Agricultural Information (ISTA). Meilke GmbH, Hamburg, Germany.

Murphy DJ, (2014). The future of oil palm as a major global crop: opportunities and challenges. Journal of Oil Palm Research, 26:1-24

Murugesan P, Shareef M, (2014). Yield, bunch quality and vegetative traits of American oil palm (Elaeis oleifera HBK) population in India. Indian Journal of Horticulture 71:23–27

Nakonechnaya OV, Gorpenchenko TU, Voronkova NM, Kholina AB, Zhuravlev YN, (2013). Embryo structure, seed traits, and productivity of relict vine Aristolochia contorta (Aristolochiaceae). Flora - Morphology, Distribution, Functional Ecology of Plants, 208: 293-297

Ooi LC-L, Low E-TL, Abdullah MO, Nookiah R, Ting NC, Nagappan J, Manaf MAA, Chan K-L, Halim MA, Azizi N, Omar W, Murad AJ, Lakey N, Ordway JM, Favello A, Budiman MA, Van Brunt A, Beil M, Leininger MT, Jiang N, Smith SW, Brown CR, Kuek ACS, Bahrain S, Hoynes-O'Connor A, Nguyen AY, Chaudhari HG, Shah SA, Choo Y-M, Sambanthamurthi R, Singh R, (2016) Non-tenera Contamination and the Economic Impact of SHELL Genetic Testing in the Malaysian Independent Oil Palm Industry. Front. Plant Sci. 7:771-781

Owusu-Appiah S (2007). Pests of Oil Palm, Elaeis guineensis, Jacq. (Palmaeae). In Major Pests of food and selected fruit and Industrial Crops of West Africa. Edited by D. Obeng-Ofori. The City Publishers Ltd., Accra, Pp 12-18.

Rhebergen T, Fairhurst T, Whitbread A, Giller KE, Zingore S, (2018). Yield gap analysis and entry points for improving productivity on large oil palm plantations and smallholder farms in Ghana. Agric. Syst. 165, 14–25.

Sayer J, Ghazoul J, Nelson P, Klintuni Boedhihartono, A, (2012). Oil palm expansion transforms tropical landscapes and livelihoods. Global Food Security. 1, 114–119.

Sim CC, Zaharah AR, Tan MS, Goh KJ, (2015). Rapid Determination of Leaf Chlorophyll Concentration, Photosynthetic Activity and NK Concentration of Elaies guineensis Via Correlated SPAD-502 Chlorophyll Index. Asian Journal of Agricultural Research, 9(3):132-138

Siti AMP, Tsan FY (2017). Growth Performance of Oil Palm Seeds of Different Vigor in Pre-Nursery. Jurnal Intelek 12:29-36

VSN International (2022). Genstat for Windows 22nd Edition. VSN International, Hemel Hempstead, UK.

Woittiez LS, van Wijk MT, Slingerland M, van Noordwijk M, Giller KE (2017). Yield gaps in oil palm: A quantitative review of contributing factors. European Journal of Agronomy, 83:57–77