

Effect of Two Different Media on Selected Liberica Coffee Clones Stem Cuttings Performance

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Abstract

Coffee production in Malaysia has been declining, and the demand for high-quality coffee is increasing. To address this issue, Malaysian Agricultural Research and Development Institute (MARDI) has released new coffee clones, MKL 8, 9, and 10 with promising yield quality. Under current guidelines, planting materials must be produced through grafting, but a huge drawback is the time needed to produce mature planting materials. A new technique was developed, potentially saving a lot of time. This technique, known as stem cuttings, uses standard media, sand and soil (1:1). The study aimed to evaluate the effects of two different media on the vegetative performance of stem cuttings on selected clones MKL 8, 9, and 10. The experiment used a randomized complete block design with a factorial arrangement with five replications. The results showed that the type of media significantly affected root fresh weight and root dry weight, with media 1 (soil, peat moss, and sand in a ratio of 4:2:1) performing better than media 2 (soil, sand, and CIRP in a ratio of 4:4:1). The clones also exhibited significant differences in SPAD meter readings, height, and leaf number, with MKL 8 performing better than MKL 9 and MKL 10. Correlation analysis revealed that SPAD readings were positively associated with leaf number and survival, while plant height was positively associated with leaf number, survival, fresh root weight, and dry root weight. This study highlights the importance of selecting suitable clones and media for coffee stem cuttings to ensure better performance and improve coffee production in Malaysia.

1. Introduction

Coffee is a hardy perennial crop planted by smallholders either as a monocrop or as an intercrop with other crops such as durian and coconut [1]. Coffee production in Malaysia has been dwindling, and its production trend is in decline, but the demand for coffee, especially premixed, has been increasing [2]. Throughout 2024, Malaysia imported approximately 1.2 million 60 kg bags of coffee beans, remaining consistent with 2023 import numbers [3]. Despite this overwhelming figure, Malaysia contributes only 0.16% to global coffee production [4]. To enhance coffee production in Malaysia, the Malaysian Agricultural Research and Development Institute (MARDI) introduced three new Liberica coffee clones—MKL 8, MKL 9, and MKL 10—in 2023, each selected for its promising yield potential and quality traits [5]. The most important objective in releasing these clones is to provide better planting material in terms of yield and quality. Currently, the propagation method is through grafting as devised by MARDI [6]. Technically, this method involves two steps. First, a mature rootstock must be established, which requires 7–8 months, and the second step is the grafting of desired clones onto the mature rootstock, which requires another 6–7 months to stabilize [1]. This method requires approximately 14–16 months to produce quality planting materials.

To further improve coffee production in Malaysia, a new potential method of propagation was studied, known as stem cutting [7]. Studies have pointed out several key factors that need to be considered. First is the type of scion or water shoot, followed by wounding techniques, and lastly, the closed covering period. The type of scion to be used for stem cutting is the softwood section usually found on the apical top of an orthotropic scion, which comprises 2–3 nodes. Other sections are not as effective. Subsequently, these selected sections of scion need to be subjected to a V-shaped wounding technique. The purpose of this wound is to increase the surface area for rooting. After the scions have been prepared, they must be transplanted into polybags and placed under closed covering for a 3-month period. The purpose of this approach is to increase the chances of survival. The resulting plantlets can be planted in the field after a further 6 months of growth under nursery conditions to achieve stability.

Stem cutting has several advantages. Firstly, there is genetic stability since the stem cutting is true to type from parent planting materials, and secondly, root emergence is quicker [3]. Furthermore, the major advantage of this stem cutting technique is the absence of rootstock, which bypasses the 7–8 months required for rootstock maturation. Previous research by Mohd Rani et al. [7] explored stem cutting propagation techniques for Liberica coffee, utilizing a standard medium composed of sand and soil in a 1:1 ratio. While this approach demonstrated feasibility for rooting, subsequent findings by Ahmad Arif et al. [1] revealed that such a medium was suboptimal for Liberica coffee root development, particularly in terms of root biomass and overall vigor. To address this limitation, the present study evaluates two alternative media formulations: Media 1 (soil, peat moss, and sand in a 4:2:1 ratio) and Media 2 (soil, sand, and CIRP in a 4:4:1 ratio). These combinations were selected based on prior evidence that organic amendments such as peat moss enhance water retention, aeration, and nutrient availability [15, 16], while CIRP has been shown to improve root development through phosphorus enrichment and mycorrhizal interaction [25]. By comparing these media across selected MKL clones, this study aims to optimize vegetative performance and improve propagation efficiency for Liberica coffee in Malaysia. Therefore, a study using two different media on stem cuttings of several coffee clones was conducted to evaluate their vegetative performance.

2. Methodology

The experiment was conducted from January to October 2024 at Ladang 1, Nurseri Kopi Pusat Tanaman Industri, MARDI Kluang, Johor, Malaysia. A randomized complete block design (RCBD) with a factorial arrangement was employed, comprising two factors: clone type and growing media. Each treatment was replicated five times, resulting in 30 experimental units, with each unit containing five plantlets. The selected Liberica coffee clones—MKL 8, MKL 9, and MKL 10—were propagated using softwood scions harvested from orthotropic shoots. These scions were subjected to V-shaped wounding to enhance rooting surface area before being transplanted into polybags (6" × 9") filled with one of two media formulations: Media 1 (soil, peat moss, and sand in a 4:2:1 ratio) and Media 2 (soil, sand, and Christmas Island Rock Phosphate [CIRP] in a 4:4:1 ratio). No synthetic chemicals or growth regulators were applied during the rooting phase to isolate the effect of media composition. All polybags were placed in a closed propagation chamber for three months to maintain humidity and enhance survival.

Vegetative performance was assessed at 210 days after transplanting (DAT). Parameters measured included SPAD chlorophyll content, plant height, leaf count, root length, root fresh weight, and root dry weight. SPAD readings were obtained using a SPAD-502 Plus meter (Konica Minolta, Japan), which provides a unitless index (0–100) representing relative chlorophyll content. Plant height was measured in centimeters (cm) from the base to the apical tip using a ruler, while leaf count was recorded as the total number of fully expanded leaves per plantlet. Root length was measured in centimeters (cm), and root fresh weight was recorded in grams (g) after gentle washing and blotting. Root dry weight was determined by oven-drying roots at 65°C for 72 hours and weighing them in grams (g).

All data were subjected to analysis of variance (ANOVA) using SAS software version 9.4 [10]. Where necessary, specifically for root parameter analysis, data were subjected to square root transformation to meet assumptions of normality and homogeneity of variance. Mean separation was performed using the Least Significant Difference (LSD) test at $p \leq 0.05$. Correlation analysis was also conducted to evaluate relationships among SPAD, height, leaf number, survival rate, and root biomass, providing insights into trait interdependence and propagation success.

3. Result and Discussion

Analysis of variance (ANOVA) revealed that root length was not significantly influenced by either clone type or media composition, and no significant interaction effect was observed between clone and media (Table 1). In contrast, both root fresh weight and root dry weight were significantly affected by the type of media used, with Media 1 (soil, peat moss, and sand in a 4:2:1 ratio) outperforming Media 2 (soil, sand, and CIRP in a 4:4:1 ratio). However, no significant differences were detected among clones for these two parameters, and no interaction effect between clone and media was observed. These findings suggest that media composition plays a more critical role than clonal variation in influencing root biomass accumulation during the early vegetative development of Liberica coffee stem cuttings.

Table 1 Mean square ANOVA analysis effect of selected clones and growing media on root parameters analysis

Source of variance	Root analysis parameter		
	Root length (mm)	Root fresh weight (g)	Root dry weigh (g)
Block	0.45	0.38	0.18
Clone (MKL)	0.92	1.77	0.89
Media	0.003	8.46*	3.30*
MKL x Media	0.54	0.85	0.06
Grand mean	19.57	7.17	2.73
C.V. (%)	11.14	33.16	39.69

Note: mean followed by * indicate significant difference at 0.05

3.1 Root Length

Despite observable differences in root length among the clones, particularly the notably lower value recorded for MKL 9 under Media 1, the analysis of variance (ANOVA) indicated that these variations were not statistically significant (Table 2). This suggests that while MKL 9 may exhibit reduced root elongation under specific media conditions, the overall performance across MKL 8, MKL 9, and MKL 10 did not differ significantly in a reproducible or consistent manner. Such findings highlight the importance of interpreting raw data within the context of statistical thresholds, where apparent trends must be supported by significant p-values to warrant biological or agronomic conclusions.

According to a study, different species of *Eranthemum reticulatum* exhibited varying root lengths when propagated through stem cuttings [11]. The study reported that *E. albomarginatum* showed the highest root length among all species examined. Another study found that the overall rooting percentage of selected Arabica coffee variety stem cuttings was significantly affected by varietal differences; however, these differences did not influence the number of fibrous roots, lateral root length, or number of lateral roots [12]. Recent studies on *Nauclea diderrichii*, a hardwood species native to West Africa, have demonstrated its exceptional rooting ability through stem cuttings—often achieving up to 100% success under optimized conditions—highlighting its strong vegetative propagation potential compared to other hardwood species. These findings also underscore the influence of genetic variation, as rooting performance can differ significantly among clones and species, even though the underlying mechanisms governing such variation remain incompletely understood [13].

Table 2 Mean comparison of root length according to clone and media propagated through stem cuttings

Clone	Media	Root length (mm)
8	1	21.07 a
8	2	17.67 a
9	1	11.75 a
9	2	20.50 a
10	1	22.58 a
10	2	23.62 a

Note: Means followed by the same alphabet letter within each column are not significantly different from one another at the 5% probability level, based on Least Significant Difference (LSD) test

3.2 Root Fresh Weight and Root Dry Weight

Use of Media 1, composed of soil, peat moss, and sand in a 4:2:1 ratio, resulted in a significantly higher root fresh weight compared to Media 2, which consisted of soil, sand, and CIRP in a 4:4:1 ratio (Table 3). The mean root fresh weight recorded under Media 1 was 9.89 g, whereas Media 2 yielded only 2.18 g. This represents a substantial increase of approximately 354%, confirming the superior performance of Media 1. The enhanced rooting response can be attributed to the physical and chemical properties of peat moss, which improve aeration, moisture retention, and nutrient availability—factors known to promote root initiation and elongation. Gopale and Zunjurrao [14] emphasized that growing media with sufficient porosity and aeration facilitate better root penetration and stimulate rooting behavior in stem cuttings. This finding aligns with the work of Bigelow et al. [15], who demonstrated that incorporating peat moss into sand-based media significantly enhances hydraulic conductivity and water-holding capacity, thereby supporting more vigorous root development. Collectively, these results reinforce the importance of media composition in optimizing root biomass accumulation during the vegetative propagation of *Liberica* coffee.

Root dry weight exhibited a response similar to fresh root weight, with Media 1 contributing significantly higher values (Table 3). The observed difference was 585.96%. A study reported that the use of pure peat moss as a growing medium considerably enhanced root dry weight compared to soil, silt, and farmyard manure in *Hylocereus polyrhizus* L. [16]. Nonetheless, another study found that a media combination of soil and farmyard manure (FYM) at a 1:1 ratio resulted in significantly greater root dry weight compared to a mixture of soil, sand, and FYM. These findings indicate that organic matter, such as peat moss and farmyard manure, improves the overall potential of growing media [17]. From these studies, four key functions of organic matter in growing media can be outlined: (1) nutrient provision, including nitrogen, phosphate, and potassium; (2) water retention; (3) improvement of soil texture through enhanced porosity and aeration; and (4) facilitation of microbial activity that supports nutrient recycling [18].

Table 3 Effect of media on fresh and dry root weight

	Root fresh weight (g)	Root dry weight (g)
Media 1	9.89 a	3.91 a
Media 2	2.18 b	0.57 b

Note: mean different alphabets indicate significant difference

For vegetative performance, SPAD meter readings, plant height, leaf number, and survival (%) were analyzed. Mean square ANOVA indicated that SPAD meter readings, height, and survival were significantly affected by clone and media, while no significant clone × media interaction was observed for these traits (Table 4). In contrast, the significant effects of clone, media, and their interaction on leaf number indicate that leaf production in *Liberica* cuttings is both genotype dependent and strongly influenced by substrate properties. The observed clone × media interaction suggests that each MKL clone responds differently to media attributes such as water-holding capacity, aeration, nutrient availability, and phosphorus supply; therefore, a medium that promotes leaf production in one clone may be less effective for another.

This pattern is consistent with findings from published propagation studies. Magesa et al. [12] reported variety-specific rooting and growth responses in hybrid coffee cuttings, and Thomas [27] highlighted the role of nodal position and leaf number in determining microcutting survival and performance. Similarly, Mankessi et al. [23] observed clone-specific early field growth responses under different propagation systems for eucalyptus hybrids. Collectively, these studies support the interpretation that the significant clone × media interaction for

leaf number observed in this trial reflects genuine genotype–substrate specificity and justifies the pursuit of clone-tailored media optimization, rather than a one-size-fits-all nursery protocol.

Table 4 Mean square ANOVA effect of coffee clones and growing media on SPAD, height and leaves number

Source of variance	Parameter			
	SPAD	Height (cm)	Leaves number per plant	Survival (%)
Rep	126.96	1.96	1.36	126.79
Clones	114.84*	10.75*	5.11	220.71*
Media	94.58*	47.64*	126.71*	6272.75**
Clones x Media	21.31	0.61	24.11**	56.26
Grand mean	37.34	7.15	6.44*	61.92
C.V. (%)	8.6	18.12	21.87	11.91

Note: mean followed by * indicate significant difference at 0.05, while mean followed by ** indicate significant difference at 0.01

3.3 SPAD (Soil Plant Analysis Development) Chlorophyll Meter Readings

Analysis suggests that MKL 8 contributed to higher significant reading compared to MKL 9 but MKL 10 exhibits statistical parity (Figure 1). The difference between MKL 8 and MKL 9 was 19.14%. The present finding is in line with a study whereby it pointed out that different clones of *Populus* sp. have different photosynthetic characteristics and attributed it to the ability of a clone to assimilate CO₂ in which clones with higher leaf area index (LAI) pose major advantage compared to clones with lower LAI [19]. Ultimately, Arabica coffee clones that have lower temperatures, and favorable light condition will exhibit better photosynthesis by factoring more leafy and shady clones which help reduce the temperature [20].

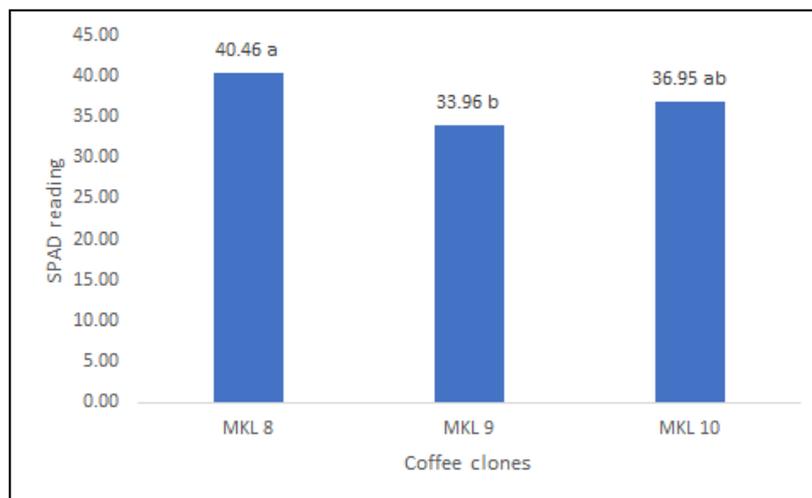


Fig.1 SPAD chlorophyll meter readings for coffee clones MKL 8, MKL 9 and MKL 10. Bars show mean SPAD values; error bars represent \pm SE. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

Apart from genetics, growing media can also influence photosynthesis of stem cuttings. According to a study, growing media can influence photosynthesis in stem cutting as it provides nutrients, aeration and moisture which are vital for crop overall performance [17]. Present finding points out that media 1 results in significantly higher reading compared media 2 and the difference was 115.51% (Figure 2). This further emphasizes the role of peatmoss as growing media as it plays major role in water retention and helps to ensure plant to consistently provided with sufficient water [21]. Using media containing peatmoss would be very beneficial in coffee stem cuttings performance.

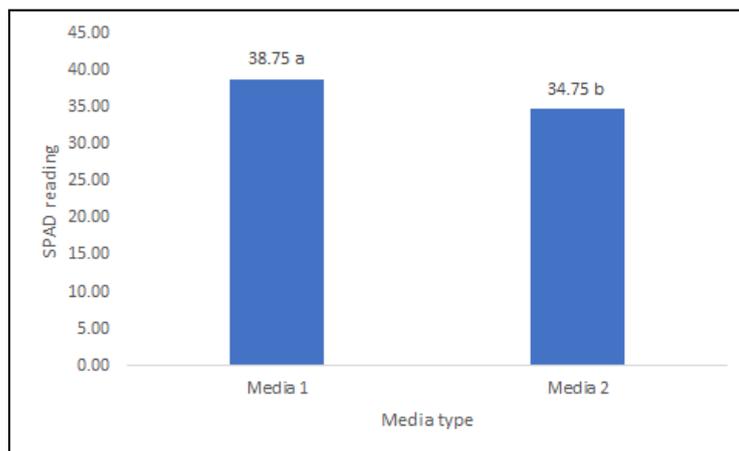


Fig. 2 Effect of growing media on SPAD reading bars show mean SPAD values; error bars represent ±SE. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

3.4 Plant Height Performance

Analysis shows that MKL 10 contributed significantly higher reading compared to MKL 8 while MKL 9 was statistically at par (Figure 3). The difference between MKL 10 and MKL 8 is 44.26%. Differences in genetic factors have the potential to affect the height performance of coffee stem cuttings. An investigation discovered that clone of *Portulaca Oleracea* L. shows variable height performance among clones, and they attributed the findings to difference in genetics among clones [22]. Another finding by Mankessi et al. [23], proved that the height between four clones of *Eucalyptus urophylla* x *Eucalyptus grandis* shows significant differences [23]. Consequently, an experiment was carried out on selected Liberica coffee clones, which concluded that selecting coffee clones with good yield and height traits is vital to ensure good coffee berry production [24]. This further proved that coffee clones' height performance has the potential to impact overall performance.

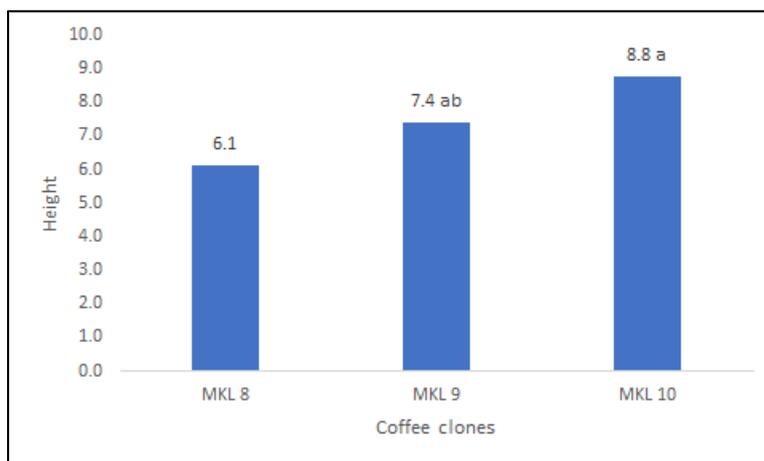


Fig. 3 Effect of clone on height performance. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

Growing media also affected plant height, as the present findings indicated that Media 1 contributed to significantly higher values compared to Media 2 (Figure 4). The observed difference was 56.6%, implying that the choice of growing media is crucial for ensuring good performance in stem cuttings. Nevertheless, stem cuttings of other species do not always respond similarly. For example, *Bougainvillea spectabilis* exhibited lower height performance in media mixtures containing peat moss but performed better in media composed solely of sand and soil [25].

An analysis of Liberica coffee rootstock further revealed no significant height response when transplanted into different growing media; however, combinations containing soil, peat moss, and cocopeat recorded the highest values. The study also established that plant height was significantly and positively associated with fresh

and dry root weight in coffee rootstock [1]. Overall, these findings indicate that growing media have the potential to influence the height and overall performance of coffee stem cuttings.

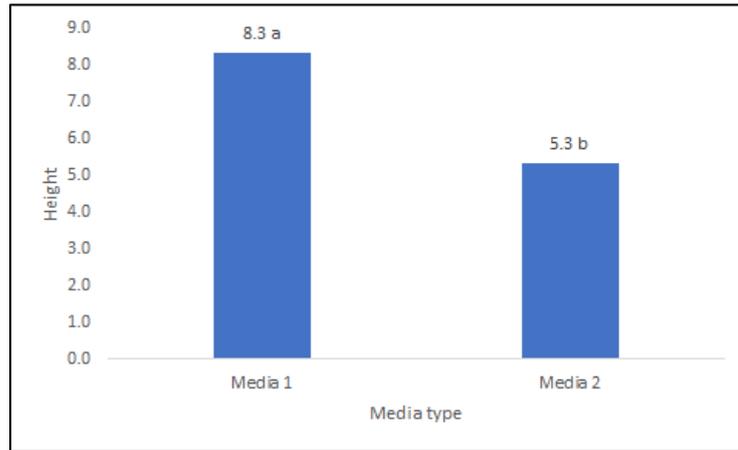


Fig. 4 Effect of media on plant height performance. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

3.5 Leaf Number

The only parameter that showed a significant interaction between media and clone was leaf number (Table 1). In other words, each coffee clone exhibited a distinct response to the growing media used. Under Media 1, MKL 8 recorded significantly higher leaf numbers compared to MKL 9 and MKL 10 (Figure 5). In contrast, under Media 2, MKL 9 showed significantly higher values than MKL 8 and MKL 10. Nevertheless, Media 1 produced higher overall leaf numbers than Media 2 across all clones. Therefore, the use of Media 1 is more advantageous compared to Media 2. A study by Kumar et al. (2021) reported that different growing media significantly affected leaf number in stem cuttings of *Punica granatum* L. [26]. They found that a medium composed of soil, sand, and vermicompost produced significantly higher leaf numbers compared to soil and sand with farmyard manure (FYM), and soil, sawdust, and vermicompost. In contrast, a previous study concluded that the overall performance of Arabica coffee stem cuttings was significantly affected by both media and clones, but no significant interaction between media and clones was observed [8]. That study reported that Arabica coffee varieties N39-2 and N33-3 achieved the highest rooting success, while growing media containing red soil produced higher values than sawdust. These findings suggest that clone, variety, and environmental conditions may collectively influence the performance of coffee stem cuttings, indicating the need for further investigation.

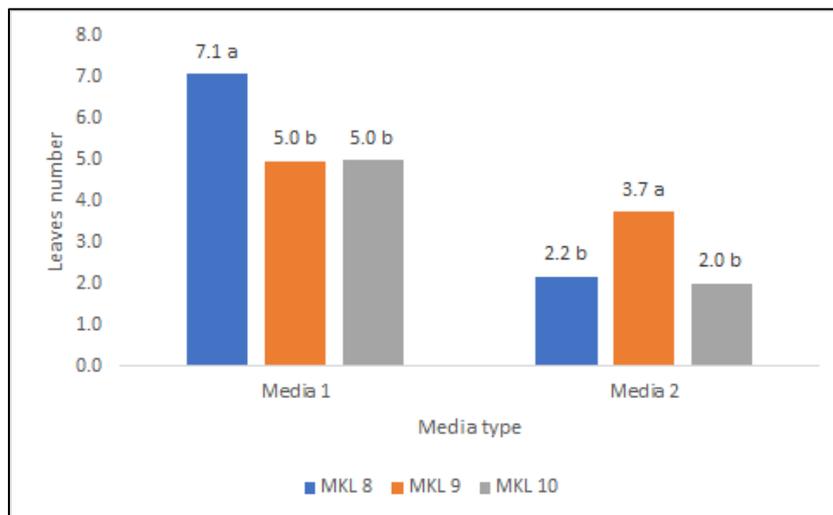


Fig. 5 Effect growing media on leaves number according to clone. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

3.6 Survival Rate of Stem Cuttings

Both clone and media contributed to significant differences, but no significant interaction between the two was established (Table 3). Similar to SPAD readings and plant height, Media 1 resulted in significantly higher values compared to Media 2 (Figure 6), with a difference of 67.87%. To ensure better survival, the use of Media 1 is therefore more strongly recommended. This recommendation is further supported by the present findings, which showed that Media 1 consistently produced higher SPAD readings, greater plant height, and increased leaf number, thereby enhancing the survival potential of the stem cuttings.

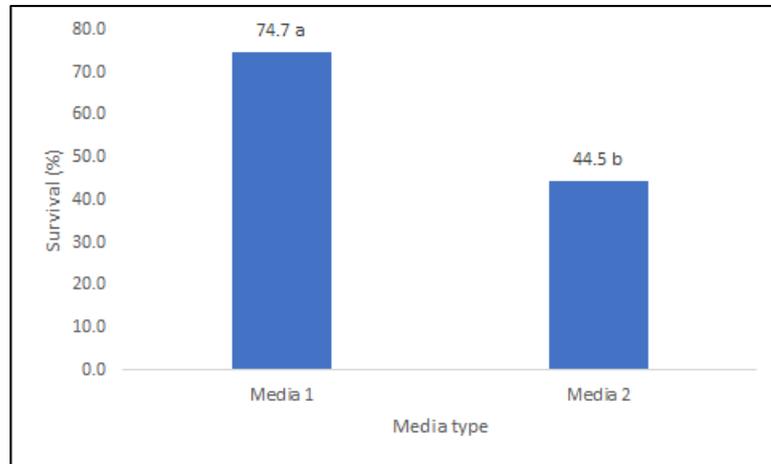


Fig. 6 Effect of media on survival reading (%). Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

Besides growing media, genetic factors may also influence survival, as MKL 8 exhibited a significantly higher survival rate compared to MKL 9 and MKL 10 (Figure 7). The observed difference was at least 14.92%. According to previous findings, the number of leaves per stem cutting plays a major role in determining survival rate, as it reflects enhanced photosynthetic capacity and nutrient uptake [26]. As discussed in the previous section, MKL 8 produced a significantly higher number of leaves under Media 1 (Figure 5), which may indicate improved photosynthesis and nutrient acquisition. In addition, significantly higher SPAD readings were observed for MKL 8 (Figure 1), further supporting this interpretation.

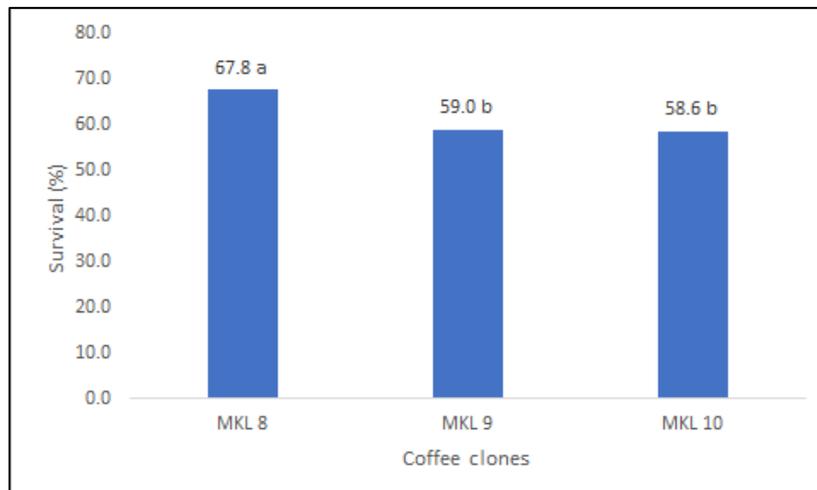


Fig. 7 Effect of clone on survival readings. Means followed by different letters are significantly different at the 5% probability level according to the Least Significant Difference (LSD) test (means with the same letter are not significantly different)

3.7 Correlation Analysis

SPAD (chlorophyll meter) readings showed no significant correlation with leaf number or survival, indicating that leaf chlorophyll content did not reliably predict these traits under the present experimental conditions (Table 4). In contrast, plant height was positively associated with leaf number ($p < 0.05$) and showed a strong correlation with survival ($r = 0.56$, $p < 0.01$), suggesting that taller cuttings tended to produce more leaves and exhibited higher viability. Root length was highly and positively correlated with both fresh and dry root weight ($r = 0.97$, $p < 0.01$), while fresh root weight was also strongly correlated with dry root weight ($r = 0.97$, $p < 0.01$), indicating a close relationship between root elongation and biomass accumulation.

Survival did not show significant direct correlations with most aboveground traits in this analysis. However, because Media 1 produced significantly greater fresh and dry root weights (Table 3), improvements in media formulation that enhance root biomass are likely to indirectly benefit cutting establishment. Taken together, these results suggest that (1) selection for greater plant height and traits that promote root biomass accumulation may improve cutting survival and nursery performance, and (2) SPAD readings alone are not a reliable proxy for leaf number or survival in this experiment. Future studies should incorporate additional physiological parameters, such as photosynthetic rate, leaf area, and non-structural carbohydrate reserves, alongside morphological traits across a wider range of clones and media to better clarify the causal relationships among chlorophyll content, leaf production, root development, and survival.

Table 5 Correlation analysis among parameters

	SPAD	Height	Leaf number	Survival	Root length	Fresh root weight	Dry root weight
SPAD	1	0.22 ns	0.51 *	0.65 *	0.27 ns	0.27 ns	0.33 ns
Height		1	0.63 *	0.50 *	0.04 ns	0.69 *	0.71 **
Leaf number			1	0.78 **	0.11 ns	0.54 **	0.59 **
Survival				1	0.14 ns	0.57 *	0.58 *
Root length					1	0.28 ns	0.24 ns
Fresh root weight						1	0.97 **
Dry root weight							1

Note: mean with * indicate significant difference at 0.05; mean with ** indicate significant difference at 0.01

4. Conclusion

Both fresh and dry root weights performed significantly better under Media 1, indicating that the use of an appropriate growing medium is vital to ensure optimal performance of coffee stem cuttings. For SPAD readings, plant height, and leaf number, the presence of clear clonal effects highlights that selecting clones with superior performance in these parameters is essential for producing high-quality planting materials. Future evaluations of new coffee clones should therefore be conducted alongside systematic testing of growing media, as clone \times media interactions influence establishment, vegetative growth, and root development. Integrating both factors from the outset represents a critical first step toward developing an evidence-based coffee production system for Malaysia.

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Author Contribution

The authors confirm contribution to the paper as follows: **study conception and design:** Ahmad Arif Ismail and Muhammad Naim Fadzli, A.R; **data collection:** Furzani Pa'ee; **analysis and interpretation:** Mohd Rani, A and Azlan Azizi M.N.; **draft manuscript preparation:** Khairol Ismail. All authors reviewed the results and approved the final version of the manuscript.

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