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Agronomic Evaluation and Yield Potentials of Grafting of *Piper nigrum* and *Piper colobrinum*

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Abstract

Foot rot disease (*Phytophthora capsici*) is Indonesia's primary challenge in cultivating pepper (*Piper nigrum*). Grafting to another Piper species, Piper coclobrinum, or melada (the local name), is a potential solution to increase resistance to this disease. The use of melada as rootstock has shown promising results during the seedling phase, but it has not been thoroughly evaluated during the production phase. The study was conducted in the pepper grower's location in Puput and Simpang Katis Villages, Central Bangka, Bangka Belitung Islands Province. The study used kapok (*Ceiba petandra*) as the standard; plant morphology, leaf area, leaf thickness, and crown diameter for grafted melada and non-grafted pepper were recorded. Harvest variables included panicle length, panicle weight, the number of fruits per panicle, and fruit diameter, measured in three different areas of the canopy: the lower area (0-1 meter above the ground surface), the middle area (1-2 meters above the ground surface), and the top area (more than 2 meters above the ground surface). Physiological variables measured included rates of photosynthesis, stomatal conductance, transpiration rate, intercellular CO_2, and CO_2 conductance. The study demonstrated that the grafted pepper plants had a larger leaf area than the non-grafted pepper plants; the panicle weight and the number of fruits per panicle were higher. Panicles located in the middle zone of the plant canopy tended to be longer for grafted and non-grafted peppers. Panicle weight, fruit size, and the number of fruits per panicle were generally more significant at the top of the plant canopy. In all parts of the canopy, grafted peppers exhibited larger fruit size, greater panicle weight, and a higher number of fruits per panicle than non-grafted "Nyelungkup" peppers, highlighting the potential of melada as a rootstock for pepper plants.

Keywords: Central Bangka, Indonesia, pepper, rootstock, scions.

Introduction

Pepper (*Piper nigrum*) is a leading spice commodity in Indonesia, with production centers spread across the Bangka Belitung Islands Provinces, Lampung, South Sumatra, South Sulawesi, East Kalimantan, and West Kalimantan (Kementan, 2022). Farmers face several challenges in pepper cultivation, including climate change and biotic and abiotic stress. One of the most significant challenges is foot rot disease, caused by the fungus *Phytophthora capsici*. This fungus spreads easily and infects plants, especially under rainy conditions. In the Lampung production area, foot rot disease causes the death of approximately 10% of pepper plants annually (Prasmatiwi and Evizal, 2020).

Various methods have been employed to produce pepper plants resistant to this fungus, including grafting techniques. Grafting is used to create plants resistant to disease and other abiotic stresses. In the grafting process, the lower stem of the melada (*Piper colobrinum*) is connected to the upper stem of the pepper plant (*Piper nigrum*), resulting in promising outcomes. Melada, when used as rootstock, offers several advantages, including high compatibility with the scion of pepper plants, resistance to waterlogging, and can withstand attacks by the fungi *Phytophthora capsici* and *Meloydogyne incognita*, which cause foot rot and yellow disease in pepper. Research by Kollakodan et al. (2020) indicated that the resistance of pepper to *Phytophthora capsici* is attributed to endophytic bacteria in the stems, leaves, and roots, which are antagonistic to *P. capsici*. Additionally, the use of melada as rootstock stimulates the growth of

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pepper seedlings in nurseries, resulting in superior seedlings that are resistant to foot rot disease (Nguyen et al., 2020; Kollakodan et al., 2020; Anggraini et al., 2021).

Information about the advantages of grafted melada pepper has not necessarily convinced pepper growers, particularly those in Bangka Belitung, to adopt grafted melada pepper. This reluctance persists despite the high incidence of *Phytophthora capsici* attacks in Bangka. The hesitation is mainly due to the assumption that the yield potential of grafted melada pepper is lower than that of non-grafted pepper plants. Moreover, few studies have been on the yield potential of grafted-melada pepper plants, especially for the "Nyelungkup" pepper accession, a local variety widely cultivated by farmers in Bangka. Therefore, this research evaluated the morphology and yield potential of the "Nyelungkup" accession of grafted pepper plants compared to non-grafted ones.

Material and Methods

The research was conducted from November 2023 to June 2024 in Puput and Simpangkatis Villages, Simpangkatis Subdistrict, Central Bangka Regency. The study involved grafted plants using the "Nyelungkup" accession pepper as the scion and the melada plant as the rootstock. The plants, which are 3 years old, were cultivated in the field in Simpangkatis Village (-2.3023833, 106.0751), using kapok (*Ceiba petandra*) as a living standard. As a control, 8-year-old "Nyelungkup" accession pepper plants were used in Puput Village (-2.3541817, 106.0838483), also cultivated using kapok as the standard. The research method involved comparing grafted melada pepper with non-grafted "Nyelungkup" pepper. Each type of pepper included ten replications, each replication consisting of three sample plants, totaling 30 plants. The grafted-melada pepper selected as samples had the upper stem of the "Nyelungkup" accession, was healthy and had a height of 3.5-4 meters. "Nyelungkup" pepper was chosen as the samples were healthy and had a height of 4-5 meters. Measurements of harvest components were made by examining 15 fruit panicles from each sample plant, with five panicles observed at the bottom of the plant canopy, 5 in the middle, and five at the top. The division of the canopy into lower, middle, and upper sections was based on the canopy's position relative to the ground surface. For grafted melada pepper, the bottom canopy was defined as the part located 0-1 meter from the ground surface, the middle canopy as 1-2 meters above the ground, and the upper canopy as more than 2 meters above the ground. For "Nyelungkup" pepper plants, the bottom canopy was 0-1.5 meters above the ground, the middle canopy 1.5-3 meters, and the upper canopy more than 3 meters above the ground surface. Leaf area measurements were conducted using the Image J application, while plant physiological activity was measured using a Li-Cor 6400 XT instrument. The data obtained were then processed using the t-test to compare variables between the grafted melada pepper and the non-grafted "Nyelungkup" pepper.

Results

The results demonstrated that the grafted melada pepper had a larger leaf area than the non-grafted pepper (Table 1). Similar findings were reported by Hedge et al. (2023), who observed that grafted-melada pepper seedlings had larger, longer, and more leaves than non-grafted pepper seedlings. Additionally, an experiment by Anggraeni et al. (2021) on 18-month-old pepper plants yielded similar results, showing that grafted-melada pepper plants had a larger leaf area than non-grafted ones. The comparison of leaf area size between grafted and non-grafted pepper plants is presented in Figure 1.

This experiment's measurement of harvest components revealed that grafted-melada pepper had longer and heavier panicles, more seeds per panicle, and larger fruit size than non-grafted pepper. However, the number of panicles did not differ significantly between the two groups based on the t-test (Table 2). Grafting has also been reported to increase the length and diameter of eggplants and the average fruit weight produced (Musa et al., 2020), increased production in citrus (Girardi et al., 2021) and cucumber (Robledo-Torres et al., 2024). Conversely, in Vietnam, grafted-melada pepper did not show better yields compared to non-grafted plants (Nguyen et al.,

Table 1. Morphology of grafted and non-grafted pepper plants

Treatment	Leaf area (cm^2)	Leaf thickness (mm)	Canopy diameter (m)
Grafted melada pepper	42.59	0.292	0.344
Non-grafted pepper	26.75	0.293	0.469
t-test	*	ns	ns

Notes: * = significantly different at P < 0.05; ns= not significantly different.

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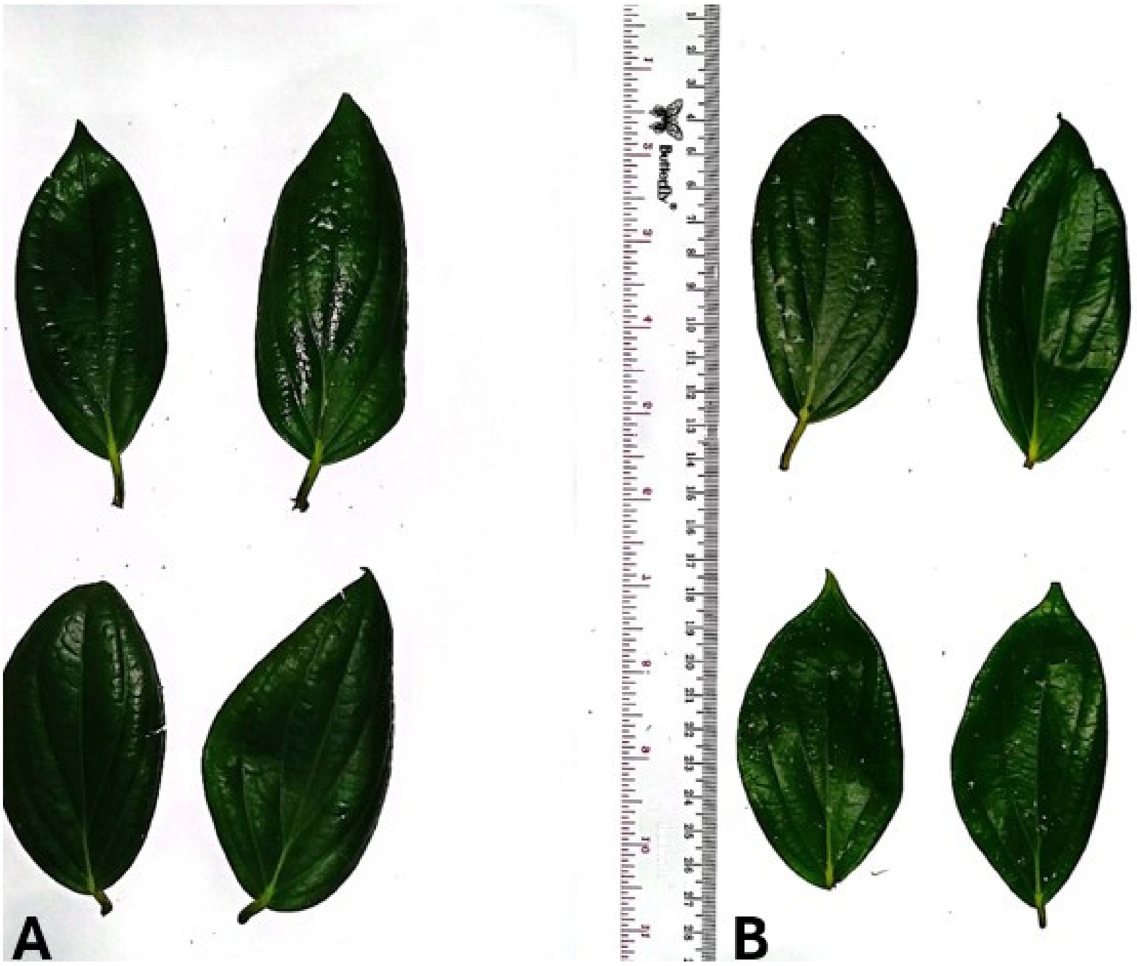

Figure 1. Leaves of grafted-melada pepper (A) and non-grafted pepper “Nyelungkup” (B)

Table 2. Pepper production components

Treatment	Panicle number	Panicle length (cm)	Panicle weight (g)	Number of fruits per panicle	Fruit size (mm)
Grafted-melada	427.9	10.47	4.87	27.84	5.16
Non-grafted pepper “Nyelungkup”	421.7	9.76	3.29	22.59	4.66
t-test	ns	ns	*	*	*

Notes: * = significantly different at P < 0.05, ns = not significantly different

2019), and in India, grafting had no significant effect on yield and plant growth in pepper plants (Hegde et al., 2023). These differences in results are likely due to variations in pepper plant varieties, cultivation techniques, and growing environments.

Panicle length between grafted-melada pepper and non-grafted pepper did not differ significantly in each zone of the plant canopy (Table 3). However, there was a trend toward longer panicles in the canopy’s middle part for grafted and non-grafted pepper plants. The number of fruits from grafted melada pepper plants was higher than from non-grafted plants in the middle and lower parts of the canopy. In contrast, the number of fruits per panicle did not differ significantly between the two in the upper part. Panicle weight and fruit size in each canopy zone showed significant differences between grafted and non-grafted pepper plants.

The measurement of physiological activity between grafted-melada pepper and non-grafted pepper plants showed differences in the variable of photosynthesis rate (Table 4). The leaf transpiration,

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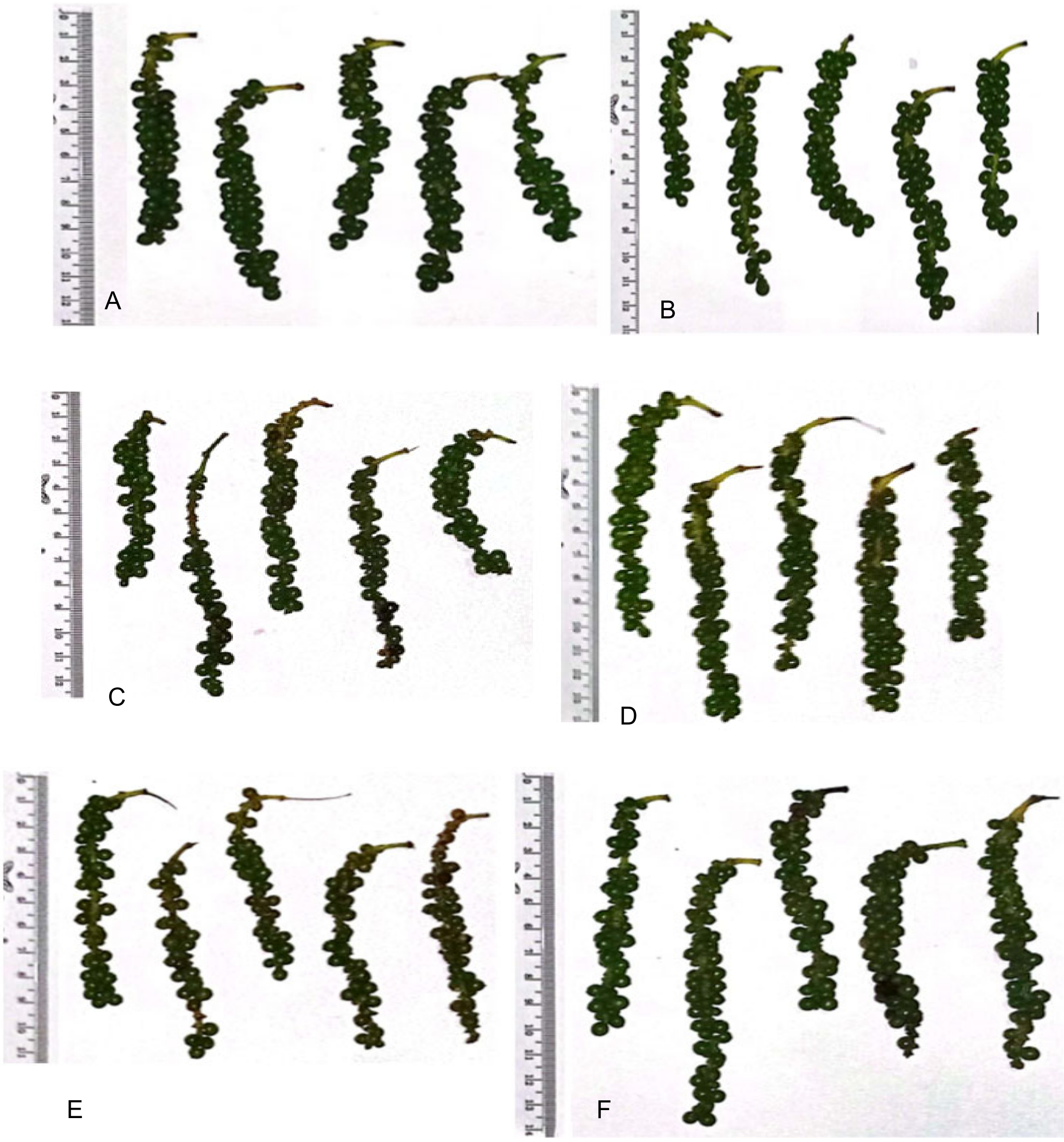

Figure 2. Panicles of non-grafted “Nyelungkup” pepper fruits from the lower part (A), panicles of grafted-melada pepper fruits from the lower part (B), panicles of non-grafted “Nyelungkup” pepper fruits from the middle part (C), panicles of grafted-melada pepper fruits from the middle part (D), panicles of non-grafted “Nyelungkup” pepper fruits from the upper part (E), panicles of grafted-melada pepper fruits from the upper part (F).

stomatal conductance, intercellular CO_2, and total CO_2 conductance did not differ between the two groups. The difference in photosynthesis rate might be related to the larger leaf size in grafted peppers.

Discussion

Grafted melada pepper and non-grafted “Nyelungkup” pepper did not show significant differences in leaf thickness and canopy diameter based on the t-test (Table 1). The lack of statistical difference in these two morphological variables is likely due to the same pepper plant accession and similar growing environmental conditions. According to Coneva (2018), leaf thickness is more influenced by environmental factors such as sunlight intensity, plant nutrition, and water availability.

Unlike leaf thickness and canopy diameter, the leaf area variable differed significantly between grafted and non-grafted “Nyelungkup” pepper plants (Figure 1). Grafted melada pepper plants had a larger leaf area compared to non-grafted plants. This difference

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Table 3. Pepper production of the upper, middle and lower regions of the plants.

Plant regio	Panicle length (cm)			Number of fruits per panicle			Panicle weight (g)			Fruit size (mm)		
	LS	LN	t-test	LS	LN	t-test	LS	LN	t-test	LS	LN	t-test
Upper	10.45	9.86	ns	38.48	30.68	ns	5.56	3.54	*	5.32	4.74	*
Middle	10.55	10.23	ns	35.48	27.00	*	5.08	3.24	*	5.04	4.54	*
Lower	10.01	9.06	ns	26.84	22.44	*	4.24	2.96	*	5.23	4.63	*

Notes: LS = grafted pepper; LN = non-grafted "Nyelungkup" pepper, * = significantly different at $P < 0.05$, ns = not significantly different

Table 4. Pepper physiological activity

Treatment	Photosyn-thesis rate	Leaf transpiration	Stomatal conductance	Intercellular CO_2	Total CO_2 conductance
Grafted -Melada Pepper	24.15	0.0021	0.213	11.103	0.1347
Non-grafted "Nyelungkup" Pepper	25.81	0.0019	0.233	10.97	0.14731
T-test	*	ns	ns	ns	ns

Notes: * = significantly different at $P < 0.05$, ns = not significantly different

is suspected to be related to the use of melada as rootstock in the grafted pepper plants. Albacete et al. (2015) explain that grafting induces a two-way interaction between the rootstock and scion through the vascular system, involving various hormones, primary and secondary metabolites, peptides, small organic molecules, nucleic acids, as well as water and nutrients. Grafting affects the translocation of primary metabolites that stimulate scion growth. The rootstock in grafted plants regulates primary metabolites (Vidoy-Mercado et al., 2021), influencing the nutrient status of the scion by enhancing the absorption of macronutrients and inhibiting the uptake of heavy metals (He et al., 2020).

The larger leaf area in grafted pepper plants may also be related to the activity of gibberellin and auxin hormones, which promote cell enlargement. Gibberellin biosynthesis in the roots is transported to the canopy in an inactive form and is activated once it reaches the canopy (Dodd, 2005). Research on grafted mandarin orange plants has shown that plant vigour is associated with GA biosynthesis in the rootstock (Liu et al., 2017). Additionally, the use of rootstock can influence auxin hormones. Kundariya et al. (2020) found that rootstock use affected auxin synthesis in the scion of tomato plants.

The harvest components in this study indicated that the grafting treatment of pepper influenced panicle length, panicle weight, and fruit weight. Grafted-melada pepper plants had longer panicles, heavier panicles, and heavier fruits compared to non-grafted "Nyelungkup" pepper plants (Table 2). Resmi et al. (2023) note that the number of fruit panicles is a genetically determined trait in pepper plants, so using melada as rootstock does not impact this variable. The superior performance of grafted melada pepper in harvest components is likely due to improved nutrient absorption and translocation facilitated by the melada rootstock. Effective compatibility between rootstock and scion enhances nutrient absorption and translocation to the scion, thereby affecting yield and quality. Furthermore, high production is associated with the size and number of xylem vessels in the rootstock. It is suspected that melada's root system has larger and more numerous xylem vessels compared to those of pepper plants. Robledo-Torres et al. (2024) found that cucumber plants grafted with rootstocks having more and larger xylem vessels yielded better than non-grafted plants. Larger xylem tissues in the rootstock facilitate improved translocation of water and nutrients, promoting optimal growth and high production. Additionally, melada plants have been reported to harbor endophytic bacteria that can enhance plant growth (Kollakkodan et al., 2020). Afzal et al. (2019) also noted that certain plant species with endophytic bacteria are associated with growth promotion.

The lower photosynthesis rate in the grafted pepper plants might be related to their larger leaf size. A larger leaf size increases the leaf area index (LAI) and the resulting shading, which can lead to a decrease in the measured leaf photosynthesis rate. Liu et al. (2021) found that changes in leaf size impact over 84% of canopy photosynthesis, while leaf angle affects more than 79%. The increased leaf area in grafted pepper plants has not yet resulted in excessive shading of other leaves.

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Conclusion

Grafted-melada pepper exhibits a morphology characterized by a larger leaf area compared to non-grafted pepper. Additionally, grafted-melada have higher panicle weight and a greater number of fruits per panicle. Panicles in the middle part of the plant canopy tend to be longer in both grafted and non-grafted plants. However, panicle weight, fruit size, and the number of fruits per panicle are generally greater in the upper part of the plant canopy. Overall, fruit size, panicle weight, and the number of fruits per panicle are larger in grafted-melada pepper compared to non-grafted pepper across all parts of the plant canopy.

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