International Journal of Innovation and Scientific Research
ISSN 2336-0046 Vol. 78 No. 1 Apr. 2025, pp. 1-13

http://www.ijisr.issr-journals.org/

Rubber tree Reken 628 single-stem bud stick leaf-clipped treatment improves seedling axillary bud quality

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ABSTRACT: The selection and quality of rubber tree bud patches is a critical factor influencing the survival rate of bud grafting in rubber tree mini-seedling buddings. Currently, the selection process relies heavily on the individual experiences of budding workers, who choose the bud sticks and cut the patches based on personal judgment. This method necessitates sorting and shipping the mini-seedling buddings in batches prior to their departure from the nursery, which in turn elevates labor costs. In this study, we trimmed the leaves from the petiole buds on the bud sticks of the rubber tree variety Reken 628. Following the detachment of the petioles and the transition of leaf scars from green to brown, we proceeded to cut the bud sticks. Both the scale buds and petiole bud patches were excised, and the various types of leaves were documented accordingly. We measured the quantity of each bud type present in the canopy and collected growth data, including the moisture content of the buds, as well as the length, width, and thickness of the bud scars, in addition to the length and width of the bud eyes. Our findings indicate that the size of the bud scar is not a reliable indicator of bud eye size. Notably, the quality of leaf-clipped buds surpasses that of those with retained leaves, with the buds from the third leaf whorl exhibiting the highest quality when subjected to leaf clipping. It is essential to increase the watering frequency to ensure that the leaf-clipped bud sticks maintain adequate moisture, which facilitates the cutting of bud patches and the removal of bud wood. This practice helps preserve the integrity of the bud eye post-peeling, ultimately enhancing the survival rate of bud grafting.

KEYWORDS: *Hevea brasiliensis,* single stem type, leaf clipped, axillary bud, quality.

1 INTRODUCTION

Several studies have assessed the safety of natural rubber in China, taking into account factors such as output fluctuations, demand fluctuations, price fluctuations, inventory security, self-sufficiency rate, import dependence, output-consumption growth rate, and consumption elasticity coefficient. These evaluations estimate that the average safety composite index over the past 24 years is merely 0.3363, indicating a high-risk status overall [1]. Natural rubber is a crucial agricultural product and strategic material for China. As a resource-constrained industry, it plays a significant role in ensuring the nation's strategic security and revitalizing rural areas in tropical regions. Rubber tree seedlings serve as the foundation for the sustainable development of the natural rubber industry, which has a production cycle of up to 30 years. The scientific and rational cultivation of these seedlings is a fundamental technological challenge for the high-quality advancement of the natural rubber sector. The quality of seedlings directly influences the growth of rubber trees and is closely linked to the natural enhancement of efficiency within the rubber industry.

Natural rubber prices fluctuate sharply and often remain depressed for extended periods. The comparative benefits of rubber production are diminishing, compounded by intermittent natural disasters that lead to reduced rubber latex production, decreased income for rubber farmers, and instability within the natural rubber industry. Addressing the urgent challenge of sustainably and efficiently enhancing the productivity of rubber planting in China, while simultaneously increasing the income of rubber farmers, has been a persistent issue affecting the development of the country's rubber planting sector for many years.

The paired row planting system in rubber plantations has been shown to significantly boost both output and income. The Reken 628 rubber tree variety is particularly well-suited for this planting system, offering advantages such as rapid growth, high yield, strong disease resistance, a compact crown width, and an expansive intercropped area. This variety was independently developed by the Rubber Institute of the Chinese Academy of Thermal Sciences and is one of the primary varieties promoted by the Ministry of Agriculture and Rural Affairs during the 14th Five-Year Plan [2]. Implementing intercropping practices beneath rubber orchards can enhance land resource utilization through the cultivation of secondary crops, thereby promoting the diversification of rubber orchard production and increasing its output value [3-4]. Renewing rubber plantations is essential for the promotion and application of new varieties and technologies, representing a critical strategy for achieving the sustainable development of the industry. This approach can significantly improve the yield per unit area and enhance land use efficiency for rubber trees [5].

In 2024, Hainan was struck by a severe typhoon, resulting in significant damage and mortality among rubber trees. This has created an urgent need to rapidly cultivate a large number of rubber seedlings to facilitate the renewal of rubber gardens. The process of mini-seedling budding for rubber trees is characterized by a short nursery cycle, low labor intensity, a high yield of seedlings per unit area, and ease of transportation and planting. Additionally, the plants exhibit a well-developed taproot, a complete root system, a high post-planting survival rate, rapid growth, and robust resistance to drought, wind, and cold, along with early opening and cutting capabilities [6,7]. Sun Xiaolong *et al.* [6] investigated the effects of various leaf whorls and bud patches on the quality of mini-seedling budding, finding that mini-seedlings produced from the 3rd petiole buds and scale buds exhibited significantly superior growth compared to other treatments and the control group of random buds. Similarly, Zhou Jun *et al*. [8] and Chen Jian *et al*. [9] examined different buds across various leaf whorls and concluded that the bud patches on different leaf whorls significantly influence the budding survival rate. However, these studies did not analyze the quality of axillary buds prior to budding, which is a critical factor affecting the success of rubber seedling budding. Practical production experience has demonstrated that leaf clipping of bud sticks in rubber trees enhances the efficiency of seedling budding operations and increases the survival rate of bud grafting. In contrast, leaf clipping in other forest trees has been shown to exert certain negative effects on the plants [10-11]. Consequently, there is a notable lack of systematic research regarding the impact of leaf clipping on the quality of axillary buds in budding sticks of rubber tree seedlings. This study compares the differences in various growth indicators of axillary buds on bud sticks under two treatments: leaf reserved and leaf clipped, and analyzes the correlation between them and the quality of axillary buds to further understand the effect of leaf clipping treatment on the propagation process of rubber tree mini-seedling and bud sticks. This will provide a theoretical reference for improving the quality of rubber tree seedlings and promoting the sustainable development of China's natural rubber industry.

2 MATERIAL AND METHODS

2.1 EXPERIMENTAL SITE

The experimental site is situated within the seedling cultivation base in Danzhou City, Hainan Province (109°29′37″, 19° 30′9″, altitude: 116.9 m). Rubber tree Reken 628 bud sticks were planted in December 2023. We initially observed the growth of leaf whorls of the rubber tree Reken 628 bud sticks at each phenological stage [12-13]. A transparent ruler was employed to measure leaf length and width, while a chlorophyll meter (Jinkelida TYS-4N) was utilized to assess chlorophyll content, nitrogen content, and leaf surface characteristics. Humidity, leaf surface temperature, plant height was measured using a tape measure, and stem diameter was recorded with a vernier caliper. Once the top leaves stabilized, we removed the leaves on the petiole buds of the 2nd, 3rd, and 4th leaf whorls, excluding the close-node buds. After the leaf scars transitioned from green to brown, we excised the single-stem type leaves and single-stem leaves accordingly. Finally, we cut off the stems, leaves, petiole buds, and scale buds associated with the 2nd to 4th leaf whorls.

2.2 MORPHOLOGY OBSERVATION

The removed leaves, petiole buds, scale buds, and stems were classified based on the position of the leaf whorl. Their fresh weights were recorded using an electronic balance (0.01 g), while the length and width of the leaves were measured with a ruler (1 mm). A vernier caliper (0.01 mm) was employed to measure the bud scar length, bud scar width, bud scar thickness, bud eye length, bud eye width, and stem diameter. Additionally, a tape measure (1 mm) was utilized to determine the stem length. The fresh weights of the leaves, petiole buds, scale buds, and stems, classified according to their leaf whorl position, were measured with an electronic balance, then dried in a 65°C oven (Shanghai Yiheng DHG-9620A) until a constant weight was achieved. Subsequently, the dry weight was recorded to calculate the moisture content.

2.3 Data Processing And Analysis

The experimental data were analyzed using the Data Processing System (DPS) statistical software package, version 20.05. Statistical analyses included Student's t-test and one-way ANOVA, followed by Duncan's Multiple Range Test (SSR) to evaluate significant differences among various treatments at $P < 0.05$. Additionally, the Technique for Order Preference by Similarity to Ideal Solution (TOPSIS) was employed for comprehensive analysis. GraphPad Prism version 8.3.0 was utilized for graphical representation, while correlation analyses were performed on the free Tutools Platform (http://www.cloudtutu.com). All data are presented as mean ± SD.

3 Result and discussion

3.1 Plant Growth

3.1.1 Single Stem Type Leaf Reserved 2nd, 3rd, 4th Leaf Whorl

In the single-stem type leaf reserved Reken 628 bud stick plants, the average plant height (Fig. 1A), leaf length (Fig. 1C), leaf width (Fig. 1D), leaf moisture content (Fig. 1F), and stem moisture content (Fig. 1E) in the 2nd leaf whorl were significantly higher than those in the 3rd leaf whorl by 13.84% ($P < 0.05$), 9.88% ($P < 0.05$), 11.85% ($P < 0.05$), 12.82% ($P < 0.05$), 5.97% ($P < 0.05$), respectively. Additionally, these metrics were also greater than those in the 4th leaf whorl by 6.45% ($P < 0.05$), 5.33% ($P < 0.05$), 13.81% ($P < 0.05$), 10.87% ($P < 0.05$), and 8.32% ($P < 0.05$), respectively.

The average stem diameter (Fig. 1B) of the 2nd leaf whorl is significantly greater than that of the 3rd and 4th leaf whorl by 13.13% ($P < 0.05$), and 25.64% ($P < 0.05$) respectively. Furthermore, the average stem diameter of the 3rd leaf whorl was significantly smaller than that of the 4th leaf whorl by 11.05% ($P < 0.05$). The average stem moisture content of the 2nd leaf whorl was also significantly higher than that of the 3rd and 4th leaf whorl by 6.45% ($P < 0.05$), and 8.32% ($P < 0.05$) respectively.

The average scale buds (Fig. 1H) in the 2nd and 3rd leaf whorl of the single-stem type leaf reserved bud-sticks were significantly higher than those in the 4th leaf whorl by 40.00 % ($P < 0.05$) and 35.71 % ($P < 0.05$), respectively. Other parameters did not show significant differences.

From the 2nd leaf whorl to the 4th leaf whorl, leaf moisture content, stem moisture content, and plant height demonstrated a gradually decreasing trend, whereas the stem diameter exhibited a gradually increasing trend, indicating that the upper part of the plant is relatively slender in shape, but has relatively less moisture content.

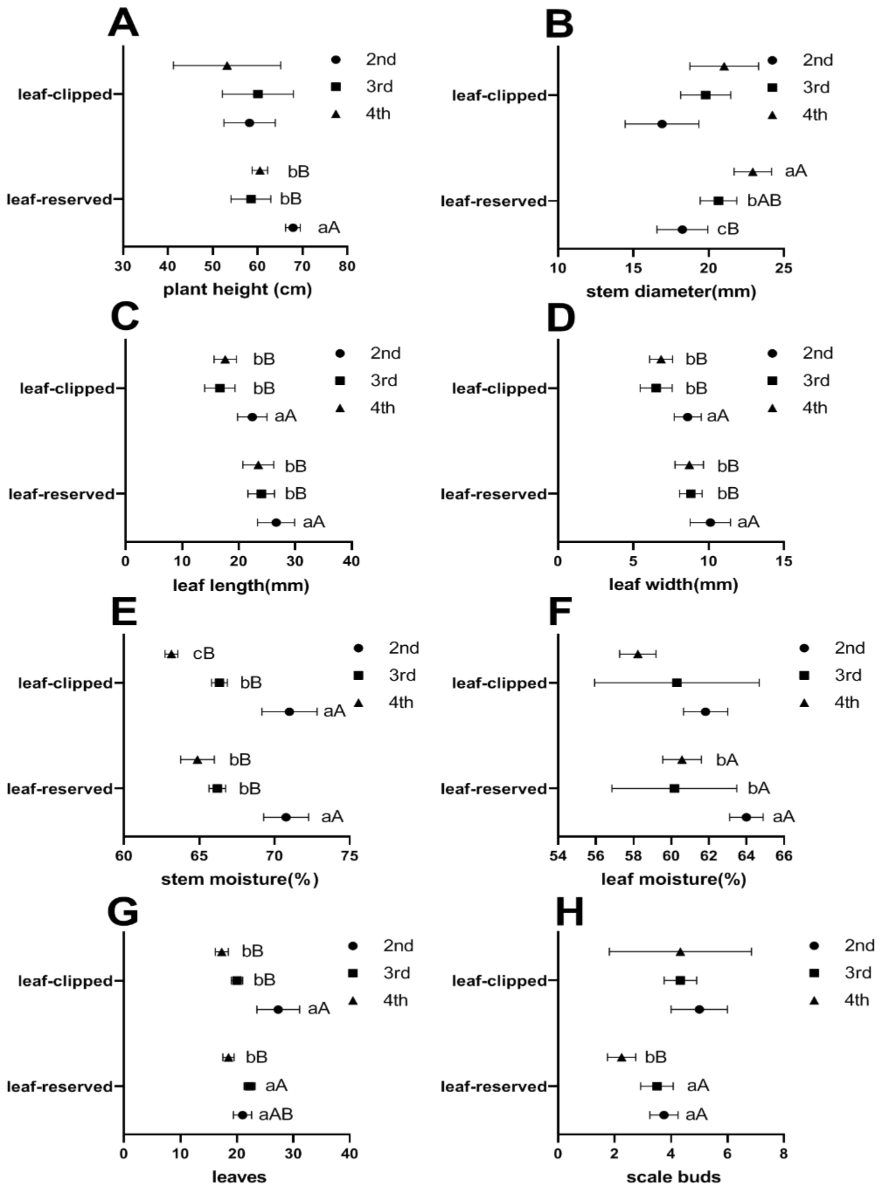

Fig. 1. Comparison of plant height, stem diameter, leaf length, leaf width, stem moisture, leaf moisture, leaves and scale buds

3.1.2 SINGLE STEM TYPE LEAF CLIPPED LEAVES 2ND, 3RD, 4TH LEAF WHORL

In the single-stem type leaf clipped Reken 628 bud stick plants, the average leaf length (Fig. 1C), leaf width (Fig. 1D), stem moisture content (Fig. 1E), and number of leaves (Fig. 1G) in the 2nd leaf whorl were significantly greater than those in the 3rd leaf whorl with increases of 25.61% ($P < 0.05$), 21.41% ($P < 0.05$), 20.49% ($P < 0.05$), 24.30% ($P < 0.05$), respectively. Similarly, these measurements were significantly larger than those of the 4th leaf whorl, showing increases of 39.29% ($P < 0.05$), 50.00% ($P < 0.05$), 6.56% ($P < 0.05$), and 11.05% ($P < 0.05$), respectively. Additionally, the average stem moisture content of the 3rd leaf whorl was significantly higher than that of the 4th leaf whorl by 4.81% ($P < 0.05$). Notably, the number of leaves in the lower part of the plant decreases relative to the upper part, and the moisture content of the stem exhibits a gradually decreasing trend.

3.1.3 Single Stem Type Leaf Reserved And Leaf Clipped 2nd, 3rd, 4th Leaf Whorl

The average plant height (Fig. 2A), leaf length (Fig. 2C), leaf width (Fig. 2D), and leaf moisture content (Fig.2F) of the single-stem type 2nd leaf whorl of leaf reserved are significantly higher than that 2nd leaf whorl of leaf clipped by14.25% ($P < 0.05$), 15.80% ($P < 0.05$), 14.85% ($P < 0.05$), 3.38% ($P < 0.05$). The average leaves (Fig. 2B), leaf length and leaf width of the 3rd leaf whorl with leaf reserved are significantly higher than those of the 3rd leaf whorl with the leaf clipped by 10.11% ($P < 0.05$), 30.50 % ($P < 0.05$), 26.05% ($P < 0.05$), respectively. The average leaf length, leaf width, leaf moisture content, stem moisture content (Fig. 2E), petiole bud moisture content (Fig. 2G) of 4th leaf whorl is significantly higher than that of leaf clipped by 24.94% ($P < 0.05$), 21.42% ($P < 0.05$), 3.88% ($P < 0.05$), 1.90% ($P < 0.05$), 2.78% ($P < 0.05$).

The leaf length, leaf width, leaf moisture content, plant height, and moisture content of stems and petiole buds in some locations under the leaf reserved treatment tended to be higher than those under the leaf clipped treatment, indicating that retaining leaves is important for maintaining normal growth of plant leaves and higher moisture content and plant growth vigor have a positive effect.

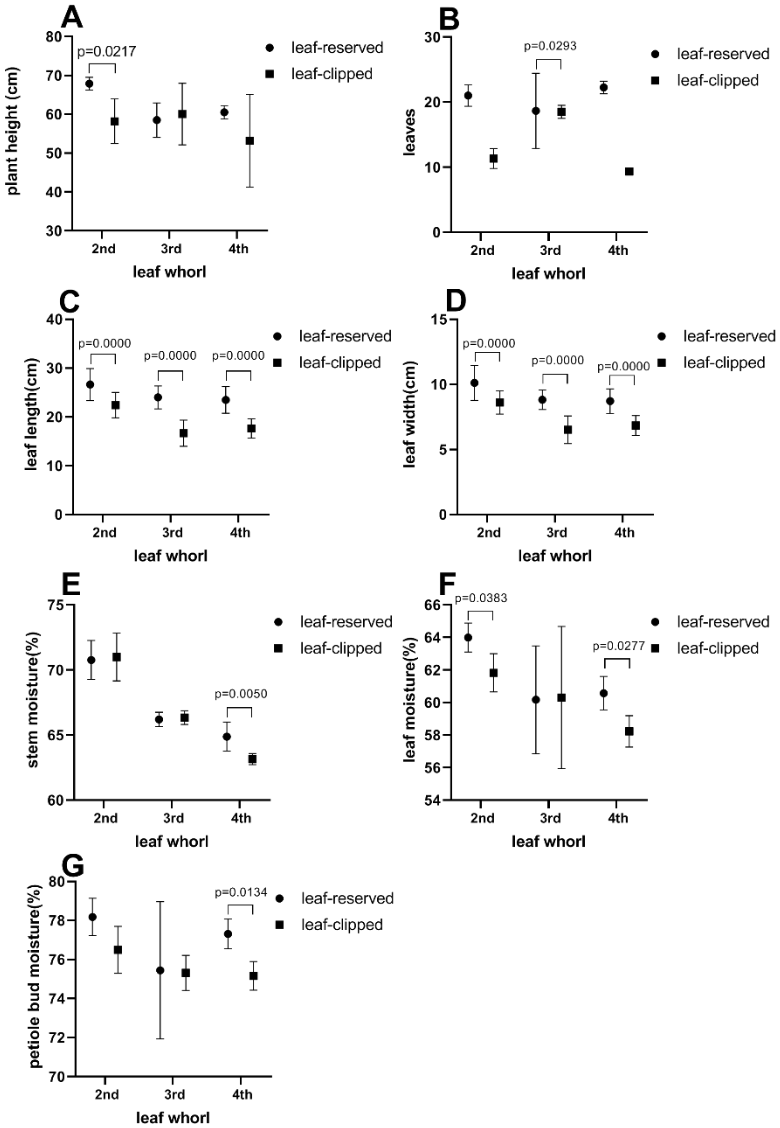

Fig. 2. Comparison of plant height, leaves, leaf length, leaf width, stem moisture, leaf moisture, petiole bud moisture

3.2 AXILLARY BUD QUALITY

3.2.1 COMPARISON OF SINGLE STEM TYPE LEAF RESERVED AMONG THE DIFFERENT LEAF WHORLS OF THE SAME PLANT

The scale bud scar length of the leaf reserved 2nd leaf whorl (Fig. 4A) is significantly smaller than that of the 3rd leaf whorl (Fig. 3) by 7.59% ($P<0.05$) and 4th leaf whorl by 23.92% ($P < 0.05$). Additionally, the scale bud scar length of the 3rd leaf whorl is significantly smaller than that of the 4th leaf whorl by 15. 17% ($P < 0.05$). The scale bud scar thickness of the 2nd leaf whorl and 3rd leaf whorl (Fig. 4C) is significantly smaller than that of the 4th leaf whorl by 33.20% ($P < 0.05$) and 28.32% ($P < 0.05$), respectively. The scale bud eye length of the 2nd leaf whorl and 3rd leaf whorl (Fig. 4D) is significantly smaller than that of the 4th leaf whorl by 23.66% ($P < 0.05$) and 22.54% ($P < 0.05$). Furthermore, the scale bud eye width of the 2nd leaf whorl (Fig. 4E) is significantly smaller 16.24% ($P < 0.05$) than that of the 3rd leaf whorl. In contrast, the scale bud eye width of the 2nd leaf whorl and 3rd leaf whorl are significantly larger than those of the 4th leaf whorl by 12.68% ($P < 0.05$) and 24.88% ($P < 0.05$), respectively.

The petiole bud scar length of the 2nd leaf whorl (Fig. 4a) is significantly smaller than that of the 3rd leaf whorl (Fig. 3) by 14.41% ($P< 0.05$). Additionally, the petiole bud scar length of the 2nd leaf whorl and 3rd leaf whorl are significantly greater than those of the 4th leaf whorl by 30.33% ($P < 0.05$) and 39.11% ($P < 0.05$), respectively. The petiole bud scar width of the 2nd leaf whorl is significantly 10.12% smaller ($P < 0.05$) than that of the 3rd leaf whorl (Fig. 4b). Furthermore, the petiole bud scar width of the 2nd and the 3rd leaf whorl is significantly larger than that of the 4th leaf whorl by 16.68% ($P < 0.05$) and 24.33% ($P < 0.05$), respectively.

The petiole bud scar thickness of 2nd leaf whorl is significantly 11.62% ($P < 0.05$) smaller than that of the 3rd leaf whorl (Fig. 4c). The petiole bud scar thickness of the 2nd and the 3rd leaf whorl is significantly smaller than that of the 4th leaf whorl, by 54.13% ($P < 0.05$) and 38.08% ($P < 0.05$), respectively.

The petiole bud eye length of the 2nd leaf whorl is 8.98% ($P < 0.05$) longer than that of the 3rd leaf whorl (Fig. 4d). Moreover, the petiole bud eye length of the 2nd and the 3rd leaf whorl is significantly longer than that of the 4th leaf whorl by 25.21% ($P < 0.05$) and 17.83% ($P < 0.05$), respectively.

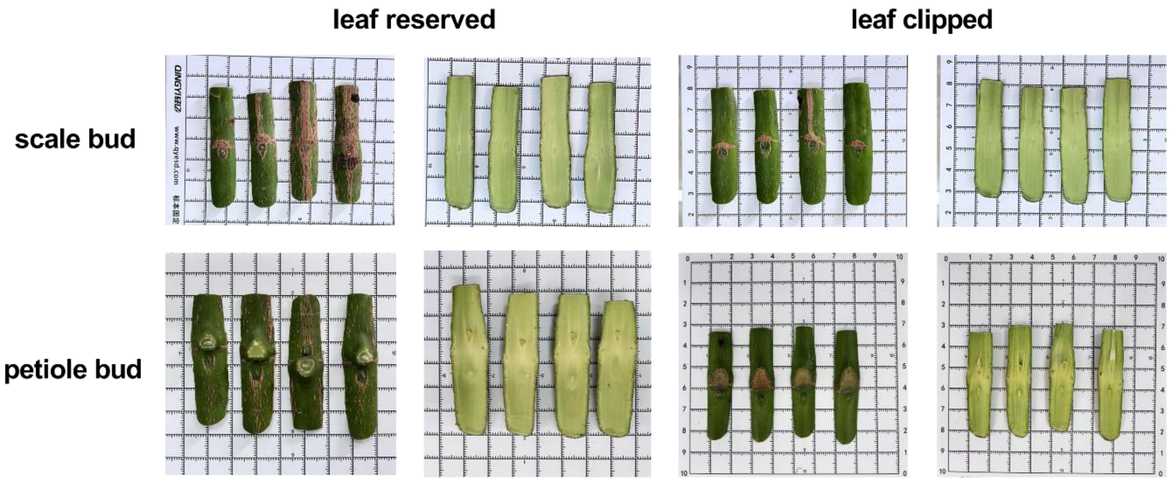

Fig. 3. Positive and negative side view of leaf bud patch and scale bud patch on 3rd leaf whorl

3.2.2 COMPARISON OF SINGLE STEM TYPE LEAF CLIPPED AMONG THE DIFFERENT LEAF WHORLS OF THE SAME PLANT

The scale bud scar length of the leaf clipped from the 2nd leaf whorl (Fig. 4A) is significantly shorter than that of the 3rd leaf whorl by 13.12% ($P < 0.05$) and 4th leaf whorl by 31.06% ($P < 0.05$). Additionally, the scale bud scar length of the 3rd leaf whorl is significantly shorter than that of the 4th leaf whorl by 15.86% ($p < 0.05$).

The scale bud scar width of the 2nd leaf whorl (Fig. 4B) is significantly smaller than that of the 3rd leaf whorl by 21.49% ($p < 0.05$). Conversely, the scale bud scar width of the 3rd leaf whorl is significantly 18.28% larger than that of the 4th leaf whorl ($p < 0.05$).

The scale bud scar thickness of the 2nd leaf whorl (Fig. 4C) is significantly smaller than that of the 3rd leaf whorl by 28.05% ($p < 0.05$), while the scale bud scar thickness of the 3rd leaf whorl is significantly greater than that of the 4th leaf whorl by 26.02% ($p < 0.05$).

The scale bud eye length of the 2nd leaf whorl (Fig. 4D) is significantly shorter than that of the 3rd leaf whorl by 18.45% ($p < 0.05$), and the scale bud eye length of the 3rd leaf whorl is significantly longer than that of the 4th leaf whorl by 25.30% ($p < 0.05$).

The petiole bud scar length of the 2nd leaf whorl (Fig. 4a) is significantly smaller than that of the 3rd leaf whorl by 33.53% ($p < 0.05$) and 4th leaf whorl by 55.29% ($p < 0.05$). The petiole bud scar length of the 3rd leaf whorl is significantly smaller than that of the 4th leaf whorl by 16.29% ($p < 0.05$). The petiole bud scar width of the 2nd leaf whorl and 3rd leaf whorl (Fig. 4b) was significantly smaller than that of the 4th leaf whorl by 31.20% ($P<0.05$) and 36.59% ($p < 0.05$).

The petiole bud scar thickness of the 2nd leaf whorl (Fig. 4c) is significantly smaller than that of the 3rd leaf whorl by 54.23% ($p < 0.05$). The petiole bud scar thickness of the 3rd leaf whorl is significantly greater than that of the 4th leaf whorl by 39.63% ($p < 0.05$).

The petiole bud eye length of the 2nd leaf whorl (Fig. 4d) is significantly longer than that of the 3rd leaf whorl by 16.25% ($p < 0.05$) and 4th leaf whorl by 39.13% ($p < 0.05$). The petiole bud eye length of the 3rd leaf whorl is significantly longer than that of the 4th leaf whorl by 27.32% ($p < 0.05$).

The petiole bud eye width of the 2nd leaf whorl (Fig. 4e) is significantly smaller than that of the 3rd leaf whorl by 19.45% ($p < 0.05$). The petiole bud eye width of the 3rd leaf whorl is significantly larger than that of the 4th leaf whorl by 22.00% ($p < 0.05$).

3.2.3 Comparison Of Single Stem Type Leaf Reserved And Leaf Clipped Of The Same Leaf Whorl Among The Different Plants

The scale bud scars of the leaf reserved from the 2nd, 3rd, 4th leaf whorl are significantly wider (Fig. 5A) than those of the leaf clipped from the same leaf whorls, with differences of 27.00% ($P < 0.05$), 16.20% ($P < 0.05$), 26.58% ($P < 0.05$), respectively. The scale bud thickness of the leaf reserved from the 2nd and 3rd leaf whorl (Fig. 5C) is significantly smaller than that of the leaf clipped from these whorls, showing reductions of 30.69% ($P < 0.05$) and 60.05% ($P < 0.05$), respectively. In contrast, the scale bud thickness of the leaf reserved from the 4th leaf whorl is significantly greater than that the leaf clipped from the same whorl, with a difference of 7.06% ($P < 0.05$). Additionally, the scale bud eye length of the leaf reserved from the 2nd leaf whorl is longer (Fig. 5D) than that of the leaf clipped from the 2nd leaf whorl by 24.09% ($P < 0.05$). Conversely, the scale bud eye width of the leaf reserved from the 4th leaf whorl (Fig. 5E) is significantly smaller than that of the leaf clipped from the same whorl, with a reduction of 17.05% ($P < 0.05$).

The petiole bud scar of the leaf reserved from the 2nd, 3rd leaf whorl is significantly longer (Fig. 5a) than that of the leaf clipped from the same whorl by 37.46% ($P < 0.05$) and 27.01% ($P < 0.05$), respectively. The bud scar length of the leaf reserved from the 4th leaf whorl is significantly shorter than that of the leaf clipped from the same whorl by 39.40% ($P < 0.05$). The petiole bud scar width of the leaf reserved from the 2nd, 3rd leaf whorl (Fig. 5b) is significantly greater than of the leaf clipped from the same whorl by 11.20% ($P < 0.05$) and 22.54% ($P < 0.05$), respectively. The petiole bud scar width of the leaf clipped from the 4th leaf whorl is significantly shorter than that of the leaf clipped from the same whorl by 39.83% ($P < 0.05$).

The petiole bud thickness of the leaf reserved from the 2nd, 3rd leaf whorl (Fig. 5c) are significantly thinner than of the leaf clipped from the same whorl by 41.39% ($P < 0.05$) and 95.37% ($P < 0.05$), respectively. Conversely, the petiole bud thickness of the leaf reserved from the 4th leaf whorl is significantly thicker than that of the leaf clipped from the same whorl by 14.59% ($P < 0.05$). The petiole bud eye length of the leaf reserved from the 2nd leaf whorl (Fig. 5d) is significantly shorter than that of the leaf clipped from the 2nd leaf whorl by 15.62% ($P < 0.05$). The petiole bud eye width of the leaf reserved from the 3rd leaf whorl (Fig. 5e) is significantly shorter than that of the leaf clipped from the same whorl by 34.53% ($P < 0.05$).

For most morphological indicators of scale buds and petiole buds, the leaf reserved and leaf clipped treatments exhibited opposing trends. When the leaves are intact, indicators such as the scale bud scar width, as well as the length and width of petiole bud scars on the 2nd and 3rd leaf whorls of the plant, demonstrate advantages. However, following leaf pruning, these advantages diminished for some indicators, while contrasting advantages emerged for others. This suggests that leaf pruning significantly influences the morphology of axillary buds, and the performance of different bud sticks varies across leaf whorls.

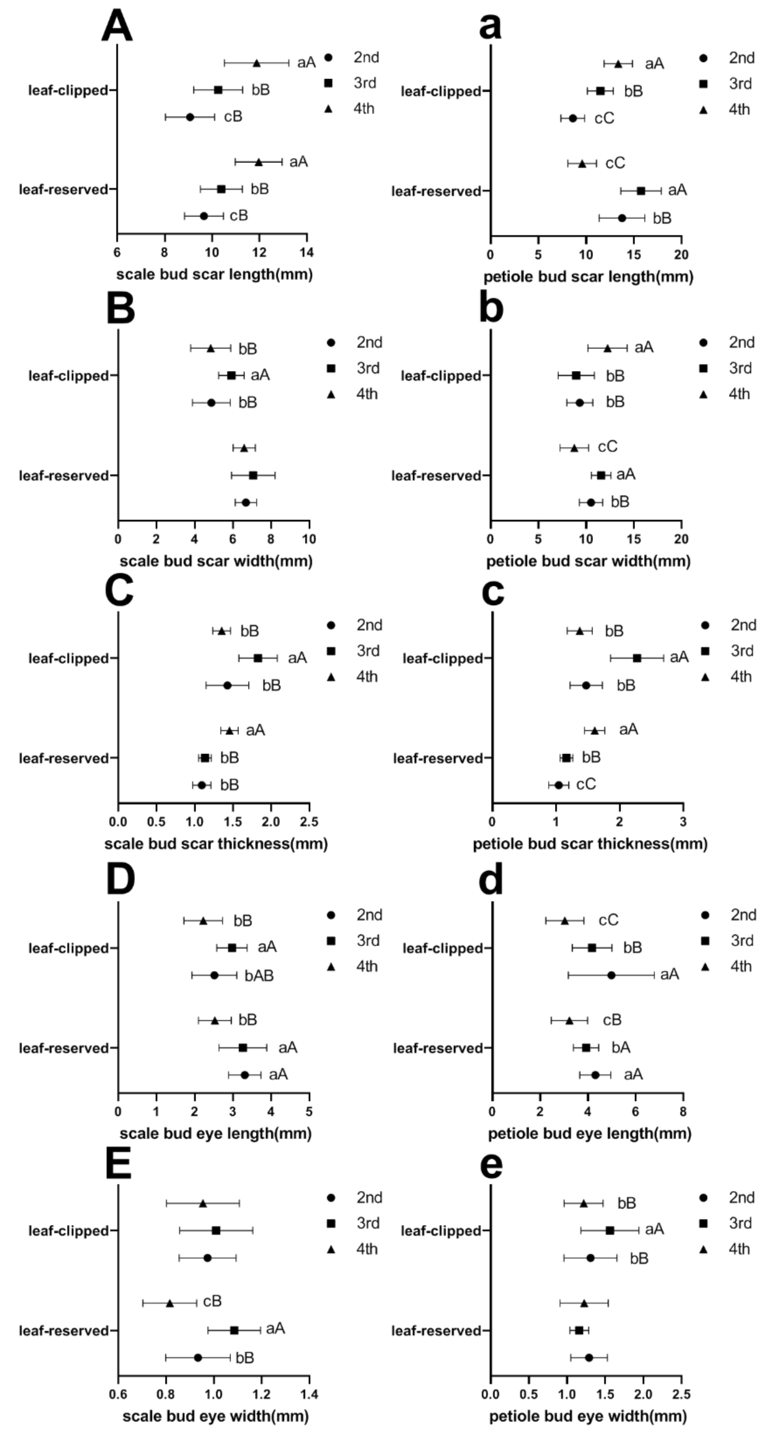

Fig. 4. The axillary bud growth of the 2nd, 3rd and 4th leaf whorl under leaf reserved and clipped treatment

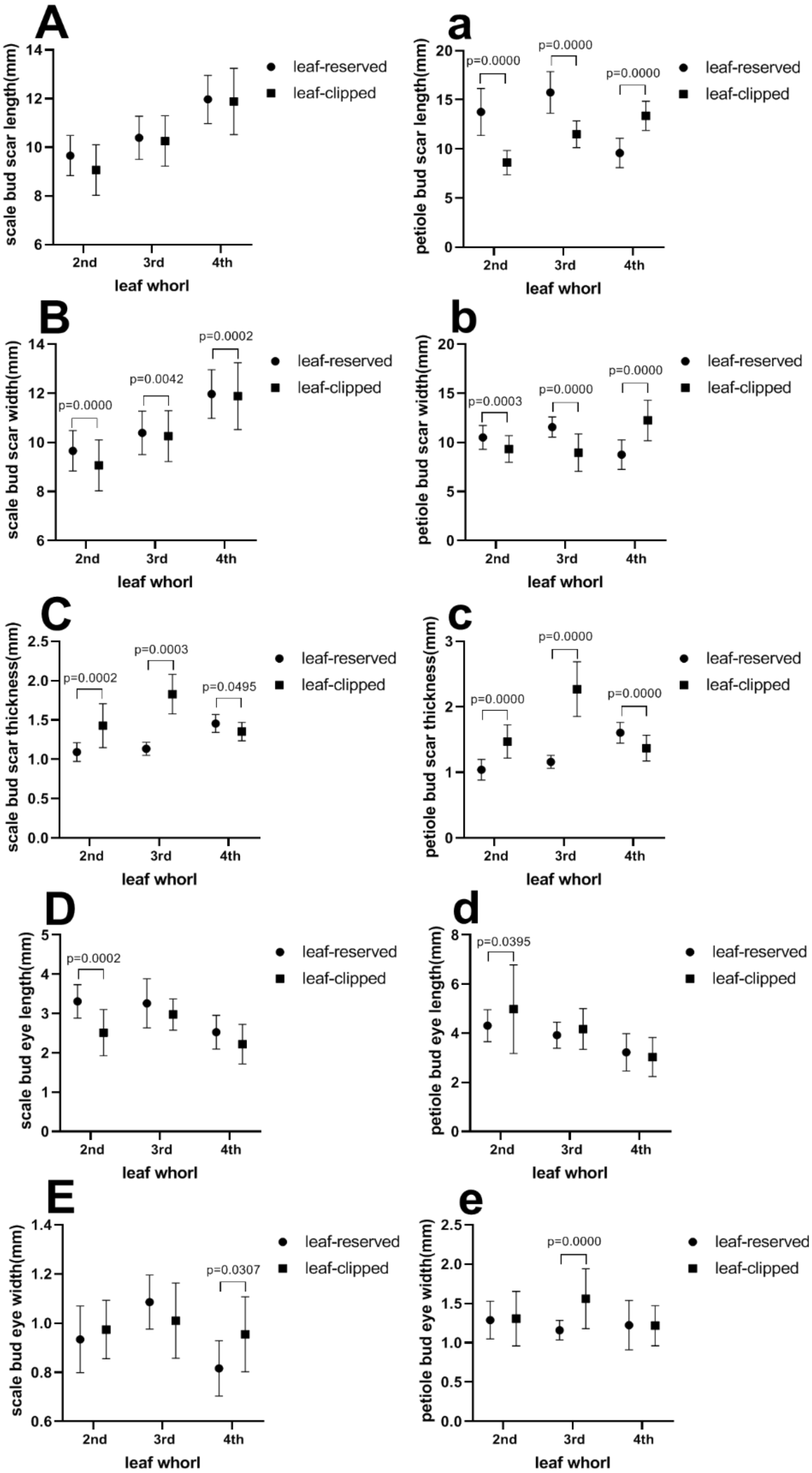

Fig. 5. The quality of axillary buds at different leaf whorl on the same plant under leaf reserved and clipped treatment

3.3 Coefficient Of Variation Analysis

As shown in Tab.1, the coefficient of variation (CV) for leaf length of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 12.27%, 9.80% and 11.68%, while under clipped leaves the CVs are 11.62%, 16.08% and 11.28%, respectively. For leaf width, the CV of the2nd, 3rd and 4th leaf whorl under reserved leaves is 13.28%, 8.52% and 10.84%, compared to 10.35%, 16.31% and 11.12% under clipped leaves, respectively. The CV of plant height of the 2nd leaf whorl under reserved leaves was 2.43%, whereas it is 9.86% for the same whorl under clipped leaves. The CV of leaves of the 3rd leaf whorl under reserved leaves is 4.30%, while it is 5.00% for the same whorl under clipped leaves.

The CV of leaf moisture of the 2nd and 4th leaf whorl under reserved leaves is 1.39% and 1.69%, while that under clipped leaves is 1.90%, and 1.66%, respectively. The CV of stem moisture of the 4th leaf whorl under reserved leaves is 1.72%, whereas it is 0.67% for the same whorl under clipped leaves. Additionally, the CV of petiole bud of the 4th leaf whorl under reserved leaves is 0.99%, while it is 0.98% for the same whorl under clipped leaves.

The CV of scale bud scar width of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 8.37%, 16.14%, and 8.81%, while that under clipped leaves is 20.38%, 11.27% and 21.74%, respectively. The CV of scale bud scar thickness of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 10.88%, 7.33%, and 7.81%, while that under clipped leaves is 19.54%, 13.72% and 8.63%, respectively. The CV of scale bud eye length of 2nd leaf whorl under reserved leaves is 12.88%. The CV of scale bud eye width of the 4th leaf whorl under reserved leaves is 13.79%, compared to 15.99% under clipped leaves.

The CV of petiole bud scar length of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 17.41%, 13.46%, and15.51%, while it is 14.50%, 11.85% and 11.13% under clipped leaves, respectively. The CV of petiole bud scar width of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 11.57%, 8.84%, and 17.14%, whereas under clipped leaves it is 14.55%, 21.13% and 16.83%, respectively. The CV of petiole bud scar thickness of the 2nd, 3rd and 4th leaf whorl under reserved leaves is 15.20%, 8.42%, and 9.87%, while under clipped leaves it is 17.23%, 18.42% and 14.35%, respectively. The CV of petiole bud eye length of 2nd leaf whorl under reserved leaves is 15.13%, compared to 36.21% under clipped leaves. The CV of petiole bud eye width of the 3rd leaf whorl under reserved leaves is 10.66%, compared to 24.38% under clipped leaves.

Tableau 1. Coefficient of variation (%) between significantly different parameters under leaf reserved and clipped treatment

parameter		reserved leaves			parameter		clipped leaves		
		2nd	3rd	4th			2nd	3rd	4th
leaf length		12.27	9.80	11.68	leaf length		11.62	16.08	11.28
leaf width		13.28	8.52	10.84	leaf width		10.35	16.31	11.12
plant height		2.43	-	-	plant height		9.86	-	-
leaves		-	4.30	-	leaves		-	5.00	-
moisture	leaf	1.39	-	1.69	moisture	leaf	1.90	-	1.66
	stem	-	-	1.72		stem	-	-	0.67
	petiole bud	-	-	0.99		petiole bud	-	-	0.98
scale bud	bud scar width	8.37	16.14	8.81	scale bud	bud scar width	20.38	11.27	21.74
	bud scar thickness	10.88	7.33	7.81		bud scar thickness	19.54	13.72	8.63
	bud eye length	12.88	-	-		bud eye length	-	-	-
	bud eye width	-	-	13.79		bud eye width	-	-	15.99
petiole bud	bud scar length	17.41	13.46	15.51	petiole bud	bud scar length	14.50	11.85	11.13
	bud scar width	11.57	8.84	17.14		bud scar width	14.55	21.13	16.83
	bud scar thickness	15.20	8.42	9.87		bud scar thickness	17.23	18.42	14.35
	bud eye length	15.13	-	-		bud eye length	36.21	-	-
	bud eye width	-	10.66	-		bud eye width	-	24.38	-

3.4 Correlation Analysis

As shown in Fig. 6, the number of leaves is significantly positively correlated with leaf width ($p < 0.05$) and is also significantly positively correlated with leaf length ($p < 0.01$). Leaf moisture exhibits a significant positive correlation with plant height, stem moisture, and petiole bud moisture ($p < 0.05$). Additionally, there is a significant positive correlation between plant height and petiole bud moisture ($p < 0.05$). A significant positive correlation exists between stem diameter and scale bud scar length ($p < 0.01$), while significant negative correlations are observed with stem moisture and petiole bud eye length, respectively ($p < 0.05$). Furthermore, a significant positive correlation is noted between stem moisture and petiole bud eye length ($p < 0.05$), alongside a significant negative correlation between stem moisture and scale bud scar length ($p < 0.05$). There is also a significant positive correlation between leaf length and leaf width ($p < 0.05$), as well as between leaf width and petiole bud moisture ($p < 0.05$). Conversely, a significant negative correlation is found between scale bud scar length and petiole bud eye length ($p < 0.01$), as well as between scale bud scar width and the number of petiole buds ($p < 0.01$). Lastly, a significant positive correlation is observed between scale bud scar thickness and petiole bud scar thickness ($p < 0.01$), and between petiole bud scar thickness and petiole bud eye width ($p < 0.05$).

The analysis indicates that thicker stems correspond to lower stem moisture levels and shorter lengths of petiole bud eyes. Therefore, it is essential to manage watering appropriately to increase stem moisture, which in turn may promote the elongation of petiole bud eyes. The correlation analysis suggests that during the growth and development of rubber tree bud-sticks, various parts influence and restrict one another, resulting in differing effects on their growth. By understanding these correlations, effective management strategies can be implemented to cultivate optimal bud patches for rubber mini-seedling budding.

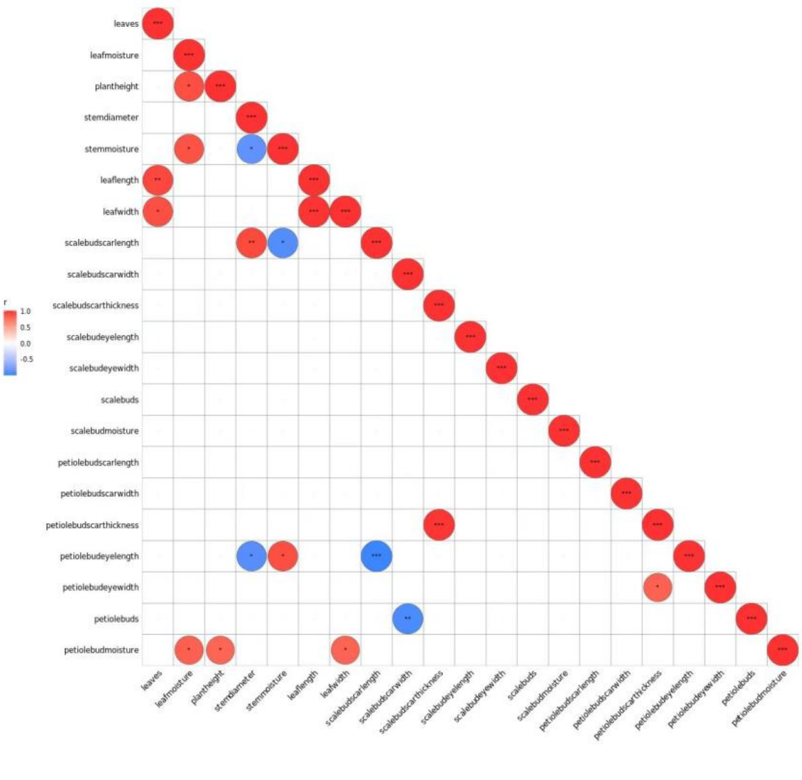

Fig. 6. Correlation analysis of all growth indexes observed

3.5 Comprehensive Analysis

Using stem diameter and plant height as low-optimal indices, the TOPSIS method is employed to analyze various parameters, including leaf characteristics, leaf moisture, plant height, stem diameter, stem moisture, leaf length, leaf width, scale bud scar dimensions (length, width, thickness), scale bud eye dimensions (length, width), scale buds, scale bud moisture, leaf bud scar dimensions (length, width, thickness), leaf bud eye dimensions (length, width), leaf buds, and leaf bud moisture. The results are presented in Table 2. When evaluating the pruning of leaves from the rubber Reken 628 single-stem type bud stick, the leaf-clipped treatment yielded better results compared to the leaf-reserved treatment. In terms of leaf whorl position, the highest quality axillary buds were found in the 3rd leaf whorl, followed by the 2nd leaf whorl, with the 4th leaf whorl showing the lowest quality. Considering both leaf clipping and the leaf whorl position, the order of effectiveness is as follows: 3rd-leaf clipped > 3rd-leaf reserved > 2nd-leaf clipped > 2nd-leaf reserved > 4th-leaf reserved > 4th-leaf clipped.

Tableau 2. Comprehensive analysis based on growth index

Leaf whorl - leaf status	Statistic CI	Rank
3rd-leaf clipped	0.539	1
3rd -leaf reserved	0.5384	2
2nd- leaf clipped	0.5263	3
2nd-leaf reserved	0.5244	4
4th-leaf reserved	0.4073	5
4th- leaf clipped	0.3862	6

CI, approximation to the Optimal Vectors.

4 CONCLUSION

In summary, the axillary bud quality of the rubber tree Reken 628 bud stick is optimal for the third leaf whorl of single stem type, where the leaves have been clipped. Consequently, it is recommended that the leaf clipping treatment be performed 10-15 days prior to mini-seedling budding on the single stem type bud sticks. Following the cutting of the bud sticks [15], the bud patch should be excised according to the position of the leaf whorl, with a preference for bud patches corresponding to the third leaf whorl for mini-seedling budding. In subsequent stages, it is advisable to cultivate more single stem type clipped rubber tree bud sticks to enhance the quality of axillary buds, thereby improving the quality of the buddings. Furthermore, there is a strong positive correlation between the quality of the bud patch and plant height, stem diameter, leaf length, and leaf width. Initially, it was assumed that the appearance of axillary buds could be used to assess the quality of bud eyes [16]. however, correlation analysis indicates that the size of the petiole bud scar and the scale bud scar do not correlate with the size of the petiole bud eye and the scale bud eye. Therefore, when selecting buds, it is important to choose rubber trees characterized by greater plant height, thicker stem diameter, and superior leaf quality in order to obtain better quality bud patches for mini-seedling budding. Additionally, one should not rely on the size of the bud marks on the surface to determine the quality of the bud pieces.

The axillary bud quality of the 3rd leaf whorl from single-stem type clipped leave is found to be the highest. However, the moisture content of all indices in rubber tree bud-stick subjected to leaf clipping treatment is lower than that of trees with reserved leaf. Consequently, after the leaf clipping treatment, it is essential to implement additional measures such as increased watering and fertilization to effectively manage the buds in the rubber tree bud-stick proliferation nursery [15-17]. This approach will help mitigate the reduction in moisture levels following leaf clipping treatment, thereby preserving the quality of the axillary buds.

ACKNOWLEDGMENT

This work was supported by Hainan Provincial Natural Science Foundation of China (321QN335), the earmarked fund for China Agriculture Research System (CARS-34-YZ4).

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