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Response of starfruit shoot and root to varying rooting volumes

(Gerak balas pucuk dan akar belimbing besi terhadap isipadu pengakaran yang berbeza)

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Key words: shoot and root responses, starfruit (*Averrhoa carambola* L.), root observation chamber, first-order root laterals, leaf nutrient concentrations

Abstrak

Kesan pucuk dan akar belimbing besi *(Averrhoa carambola)* yang telah ditanam di dalam bekas yang mempunyai isipadu yang berbeza, telah dikaji dengan menggunakan 'chamber' pemerhatian akar. Hubung kait ketinggian pokok dan bilangan internod adalah linear dengan isipadu pengakaran mengikut masa, tetapi tiada gerak balas terhadap bilangan stomata dan sel epidermal di daun. Terdapat hubung kait linear secara negatif antara 'first-order root laterals' dengan jarak hujung akar, akan tetapi pemanjangan akar pula menunjukkan keadaan linear positif dengan bertambahnya isipadu pengakaran. Ketumpatan panjang akar, luas muka akar dan jumlah panjang akar sangat dipengaruhi oleh isipadu pengakaran. Walau bagaimanapun, tiada gerak balas ketara pada ketumpatan 'tip' akar, panjang akar kasar, taburan peratus berat kering dan nisbah antara akar dengan pucuk. Kepekatan N, P and Ca pada daun bertambah dengan bertambahnya isipadu pengakaran tetapi tidak pula pada K dan Mg. Kajian menunjukkan kesan linear yang positif antara pemanjangan akar dengan ketinggian pokok.

Abstract

Shoot and root responses of root-pruned starfruit *(Averrhoa carambola)* seedlings in different rooting volumes, were studied using root observation chamber. Plant height and internode number were linearly correlated with rooting volumes over time, but stomata and epidermal cell number showed no response. Negative linear relationship was shown between the first-order root laterals and distant from root apex but root elongation had positive linear correlation with rooting volumes. Root length density, root surface area and total root length were significantly influenced by rooting volumes. However, there was no significant response on root tip density, coarse root length, percentage of dry matter distribution and root:shoot ratio. Leaf concentrations of N, P and Ca were significantly increased by rooting volumes but K and Mg were unaffected. A positive linear response between root elongation and plant height was observed.

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Introduction

The fast and vigorous growth of starfruit plants (*Averrhoa carambola* L.) is a major problem to tree size control for viable starfruit cultivation in Malaysia. Due to the absence of dwarfing rootstocks, the control of growth has to be done physically through restricted growth containers. Rooting volumes are important in determining plant growth; large container volume usually produces large plants and vice versa (Hathaway and Whitcomb 1977; Williams and Whitcomb 1979; Biran and Eliassaf 1980; Menzel et al. 1994). Limited container volumes often resulted in root growth inhibition. Due to pot-binding effects, shoot growth and development are adversely affected. Root structures are often physically restricted that 'restricted roots are shorter, thicker, and more irregularly shaped' (Wilson et al. 1977). The radial thickening of mechanically impeded roots is associated with shorter and wider cells in the cortex (Camp and Lund 1964). In starfruit, there is a close relationship between root and shoot growth as shown in high correlation between root length and leaf area (Ahmad 1994).

Root growth is cyclical in which during the periods of shoot elongation, the number of growing roots and rate of elongation decline (Head 1967). When soil temperature and soil water are non-limiting, shoot growth is a major factor controlling the intensity of root growth (Bevington and Castle 1985). It is hypothesised that under root restriction and non-limiting water condition, root growth is the major factor in controlling the intensity of shoot growth. Little information is available on the root growth dynamics such as rooting pattern, root branching and root tips activity. In addition, leaf growth characteristics in relation to root growth restriction are relatively unknown in tropical fruit trees.

The purpose of this study was to investigate shoot and root growth responses of starfruit to different rooting volumes using root observation chamber. In addition, percentage of dry matter distribution and leaf nutrient concentrations were also examined.

Materials and methods

Different container sizes were used to obtain three rooting volumes measuring 30 cm (length) x 2.5 cm (width) x 90 cm (depth), 30 x 5.0 x 90 cm, and 30 x 7.5 x 90 cm with corresponding volumes 7.5-, 15- and 22.5-litre, respectively. The sides of the chambers were built from 10 mm PVC. Root observation panels were made from transparent polycarbonates. Each viewing surface of the observation panel was a 0.27 m^2 area and covered with a black polythene sheet to exclude light. A transparent acetate overlay was placed on the viewing surface of the chambers. These chambers were filled with potting media and leaned against bench rows at an angle of 15° with the viewing area underneath, as described by Hipps et al. (1995). The chambers were irrigated to saturation and left at field capacity before transplanting.

A total of 21 6-month-old starfruit (clone B10) seedlings with basal stem diameter of 10–20 mm were selected. The seedlings were cleaned, defoliated and root-pruned. The stems were pruned to a height of 25 cm from the base and all side shoots removed. The lateral and tap roots were pruned to 5 cm in length and only two main roots were retained. Each root-pruned plant was transplanted into the root observation chambers in a glasshouse on 4 April 1997 at MARDI, Serdang, Selangor. All plants were watered, 100 mL per day, and fertilised weekly with a complete foliar nutrient 'Growfas' (N:P:K 15:15:15) at the rate of 10 g/litre and chemicals were sprayed for pest and disease control as and when required.

Shoot growth measurement

Plant height and internode number were recorded fortnightly for 18 weeks beginning on the 4th week. On the 100th day, stomatal number and leaf epidermal cell number were observed using micro-relief techniques as described by Ferris and Taylor (1994). The

abaxial epidermal layers of three newly expanded leaves were sampled from each treatment. The peel-off films were placed on microscopic slides at 40X magnification for observation using Projectina Microscope Model 4011. Stomata and epidermal cell number were obtained from an area of 5 cm x 5 cm (25 cm^2) on the image screen of the microscope.

Root growth

At each observation date, the amount and position of new root growths were traced on transparent acetate overlays. Different permanent colour markers (0.5 mm) were used to differentiate the time of recording. During recording, only one window was exposed at a time and care was taken to avoid exposure to direct sunlight. The time required for each observation ranged from 10–30 min per window. Root elongation that grew during each observation interval was determined directly from the tracings. Total root elongations were then summed up from root lengths measured at each consecutive week.

At final harvest, plant parts were separated into roots, leaves and stems. Fresh and dry weights of leaves and stems were obtained. Leaf area was determined using a LICOR 1300 planimeter. The number of first-order root laterals were recorded from two main roots. These main roots were cut at successive segments of 20 cm from the root apex to stem base. Each root segment was laid on a white perspex box with built-in light underneath for observation. All emerging and protruding roots subtended from the main roots were counted excluding secondary and tertiary branching roots. Total root length, root surface area and root tips were determined using Automated Root Length Measurement Programme (Guddanti and Chambers 1993). Root length density (RLD) and root tip density were computed simply by dividing the total root length and total number of root tip with root chamber volumes, respectively. All root components were then oven-dried at 60 °C for 72 h after which root dry weight were measured.

Nutrient analyses

At each harvest all oven-dried leaves, stems and roots were ground using a hammer mill and sieved (<2 mm mesh) and kept in capsules. The wet digestion method using concentrated nitric acid (HNO_3) and concentrated hydrochloric acid (HCl) mixture was employed. Exactly 1.0 g of ground sample was weighed into 75 mL digestion tubes, 10 mL HNO_3 was added and the tubes were left overnight. The following morning, the sample was heated in a block digester at 110–115 °C for 2 h until the solution turned clear. 10 mL HCl was then added and the solution digested for another 2 h until a clear solution appeared indicating complete digestion of the specimen. The digest was cooled for 4 h, appropriately filtered and made-up to 100 mL in the volumetric flasks and ready for nutrient analysis.

Another 0.05 g of the ground sample was placed in a separate digestion tube and 0.5 Kjeldahl tablet and 2.0 mL concentrated sulphuric acid (H_2SO_4) were then added. The mixture was then gently shaken and left until clear solution was obtained. The digest was cooled and then made up to 50 mL and was ready for nitrogen analysis.

Total nitrogen (N) was determined by the standard Kjeldahl method using an automated colourimetric system, measured at 630 nm. The phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) were analysed by the ICP emission spectrophotometer.

Experimental design and statistical analysis

The experiment consisted of three volumes of root observation chamber in completely random design with seven replications. Data were analysed using SAS procedures (SAS Institute 1985). Least Significant Differences (LSD) was used to test significant differences among treatments. Data of plant height and internode number were

transformed to natural log ($\log_e$) to reduce variation. Simple linear regression model $y = ab^x$ was fitted using SAS PROC REG procedures, where y is dependent variable, a and b are constants. Original constant values of b (slope of curve) were obtained from antilogs. Linear relationships were carried out between i) number of first-order root laterals and distant from root apex, ii) root elongation and time of emergence and iii) plant height and root elongation.

Results

Shoot growth

Larger root chamber volumes produced taller plants. Logarithmic plant growth showed that the rate of increment in plant height was linearly correlated with chamber volume over time *(Figure 1)*. Both the 15- and 22.5-litre chambers had 30% taller plant than those in the 7.5-litre chamber.

Similar to plant height, internode number also increased linearly to root-chamber volumes increase at 12 and 16 weeks that larger root chamber volume produced more internode numbers. The rates of internode number increase per week were 1.09 in the 7.5-litre chamber and 1.12 in both 15- and 22.5-litre root chambers *(Figure 2)*. This means that the 15- and 22.5-litre root had 18% more leaf flushes than in the 7.5-litre root chamber.

However, neither stomata number nor epidermal cell number were affected by the treatments. Contrastingly, leaf area was increased by 30% and 124% when root

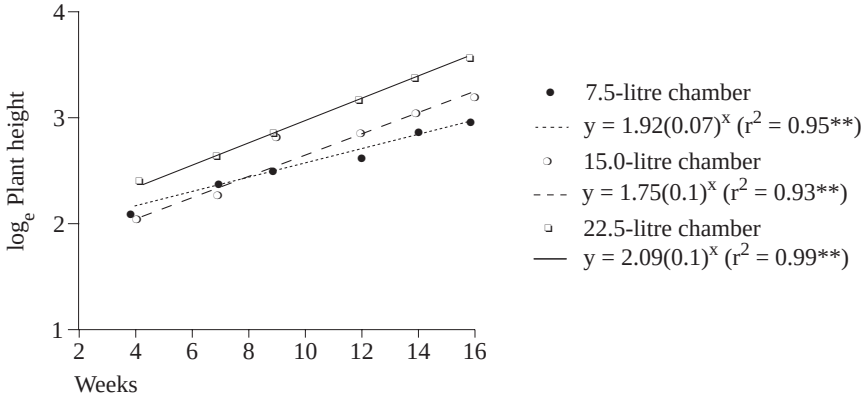

Figure 1. Plant height of starfruit grown in three volumes of root observation chamber

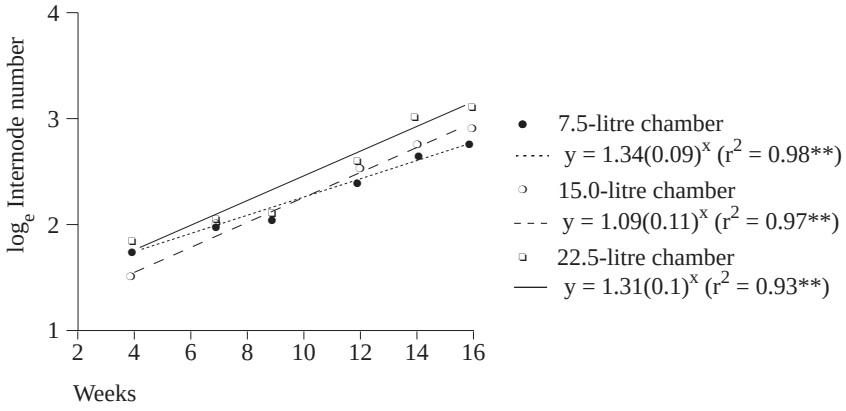

Figure 2. Internode number of starfruit grown in three volumes of root observation chamber

chamber volumes increased from 7.5 to 15 litres and from 15 to 22.5 litres, respectively *(Table 1)*.

Root growth

Root length density (RLD) was significantly decreased by increasing the root chamber volumes. Increase in root chamber volumes from 7.5 to 15 litres and from 15 to 22.5 litres reduced RLD by 20% and 8%, respectively. However when root chamber volume was increased from 7.5 to 15 litres, root surface area showed partial increase from 0.9–1.9 m^2 and 1.9–3.6 m^2. Root tip density was not affected by root chamber volume in which it ranged from 1.2 to 1.52 root tip per cm^3 *(Table 2)*.

However, coarse root length (root diameter >5 mm) showed no significant response to variation in chamber volumes. Total root length increased significantly by two folds when chamber volume was increased from 7.5 to 15 litres but no significant difference was detected when chamber volume increased from 15 to 22.5 litres *(Table 3)*.

The negative linear response between the number of first-order root laterals and distant from root apex is shown in *Figure 3*. The rates of linear decrease were consistent in the 7.5-, 15- and 22.5-litre.

Root elongation responded positively with root chamber volume over time *(Figure 4)*. The rates of root elongation were 703, 803 and 1 046 cm per week in 7.5-, 15- and 22.5-litre, respectively. This means that an increase in root chamber volume from 7.5 to 15 litres and from 15 to 22.5 litres showed 14% and 39% increase in root elongation, respectively.

Dry matter distribution

The partitioning percentage of dry matter was not significantly influenced by rooting volumes *(Table 4)*. The root:shoot ratio showed no significant effects with respect to variation in rooting volumes. These results might demonstrated that plants reallocated

Table 1. Stomata number, epidermal cell number and leaf area of starfruit plants grown in three volumes of root observation chamber

Root volume (litres)	Stomata no.[x]	Epidermal cell no.[x]	Leaf area (cm^2)[y] per plant
7.5	3.6a	22.8a	805a
15.0	4.0a	25.7a	1 044b
22.5	4.2a	26.2a	342b

[x]Based on 25 cm^2 area of the microscope screen
[y]Sampling at 120 days
Mean values in the same column with similar letters are not significantly different at $p < 0.05$

Table 2. Root length density, root surface area and root tip density of starfruit plants grown in three volumes of root observation chamber

Root volume (litres)	Root length density (cm/cm^3)[x]	Root surface area (m^2)[y]	Root tip density per cm^3
7.5	0.52a	0.9a	1.20a
15.0	0.42ab	1.9ab	1.22a
22.5	0.39b	3.6b	1.52a

[x]Total root length divided by soil volume
[y]Effective root less than 2 mm diameter
Mean values in the same column with similar letters are not significantly different at $p < 0.05$

Table 3. Coarse root length and total root length of starfruit plants grown in three volumes of root observation chamber

Root volume (litres)	Coarse root length (cm)[x]	Total root length (m)[y]
7.5	168.6a	39.1a
15.0	179.6a	79.2b
22.5	193.0a	87.8b

[x]Root diameter more than 5 mm
[y]Sum of all root lengths inclusive of all root diameter
Mean values in the same column with similar letters are not significantly different at $p < 0.05$

Table 4. Percentage of dry matter distribution and root:shoot ratio of starfruit plants grown in three volumes of root observation chamber

Root volume (litres)	Leaf (%)	Stem (%)	Root (%)	Root:shoot ratio
7.5	28.4a	13.5a	58.1a	1.3a
15.0	29.6a	14.4a	56.0a	1.3a
22.5	31.8a	16.1a	52.1a	1.2a

Mean values in the same column with similar letters are not significantly different at $p < 0.05$

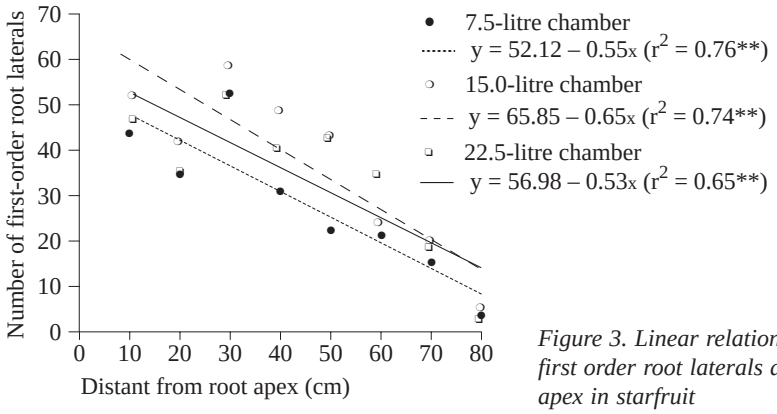

Figure 3. Linear relationship between number of first order root laterals and distant from root apex in starfruit

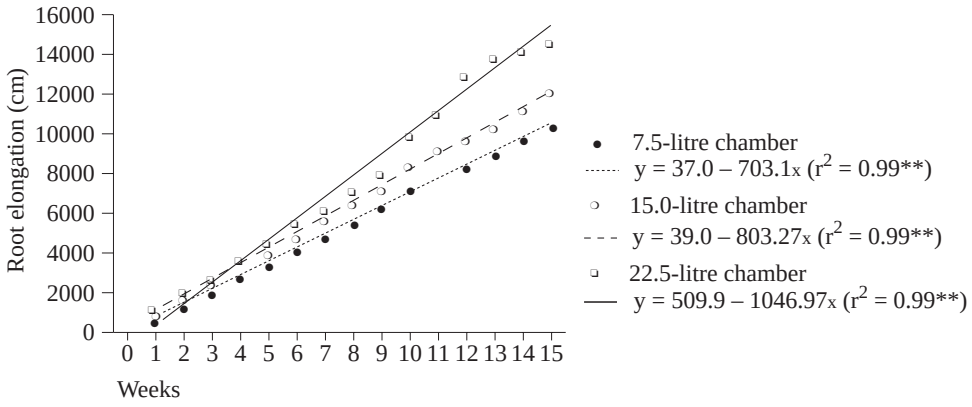

Figure 4. Linear relationship between root elongation and time of emergence (weeks) in starfruit

dry matter to the root, leaf and stem accordingly under stress or unfavourable conditions.

Leaf nutrient concentration

Leaf N, P and Ca responded significantly to root chamber volumes except for K and Mg. Increasing root chamber volumes from 7.5 to 15 litres and from 15 to 22.5 litres increased leaf N concentration by 9.5% and 5.5%, respectively *(Table 5)*. Leaf P concentration increased by 31% when root chamber volume increased from 7.5 to 15 litres, but no significant increase was detected when root chamber volume increased from 15 to 22.5 litres. Leaf Ca concentration increased significantly by 17% and 13% when root chamber volume increased from 7.5 to 15 litres and from 15 to 22.5 litres, respectively. The critical values of the cation-quotients in the leaves for estimation of relative deficiencies of K (with regard to Ca) was normal while Mg (with regard to K) showed that the nutrient ratios were slightly higher. K/Ca ratios were 10, 9.0 and 8.0 in 7.5-, 15- and 22.5-litre chambers, respectively. K/Mg ratios were 2.7, 2.6 and 2.5 with respect to the same volume increase.

Discussion

Reduction in rooting volumes decreased root growth, which consequently decreased shoot development. There was a high correlation between shoot and root growth whereby a reduction in shoot growth, such as plant height, internode number and leaf area, was synchronised with reduction in root activity such as total root length, root surface area and root elongation. Neither stomata nor epidermal cell number was affected by varying rooting volumes. This suggests that individual leaf activity was under strongly genetic in nature. Korner et al. (1989) cited that leaf size reductions in Bonsai plants are genetically controlled, and that the rate of cell enlargement in the leaf determines the frequency of cell division.

In terms of lateral root branching, the upper portion of the root system that consisted of primary, secondary and tertiary roots were the most active in root branching activity. However, the inverse relationship between number of first-order lateral and the distant from root apex of the main roots in all root chambers remained almost constantly between 0.5 and 0.6 root/cm. This implied that consistent architectural rooting is maintained despite changes in rooting volumes. The number of first-order root laterals had an inverse relationship between its distance from root apex. The general trend of decrease in root branching pattern from the root apex was associated with the emergence of new shoot. This study is in accordance with that of Head (1967) who suggested that root growth is reduced during shoot elongation. Increase in root branching is related to high allocation of dry matter to the shoots (Brouder and Cassman 1994).

Correlation studies between plant height and root elongation showed that plant height was highly related to root elongation ($r^2 = 0.95$) as shown in *Figure 5*. It was a hypothesis which stated that root is the major growth controlling factor in root restricted conditions. Stevens and Nicholas

Table 5. Leaf nutrient concentration (g/kg) of starfruit plants grown in three volumes of root observation chamber

Root volume (litres)	N	P	K	Ca	Mg
7.5	26.5a	1.7a	17.6a	17.5a	6.6a
15.0	28.9ab	2.2b	18.6a	20.5b	7.1a
22.5	30.5b	2.3b	19.2 a	23.2c	7.6a

Mean values in the same column with similar letters are not significantly different at $p < 0.05$

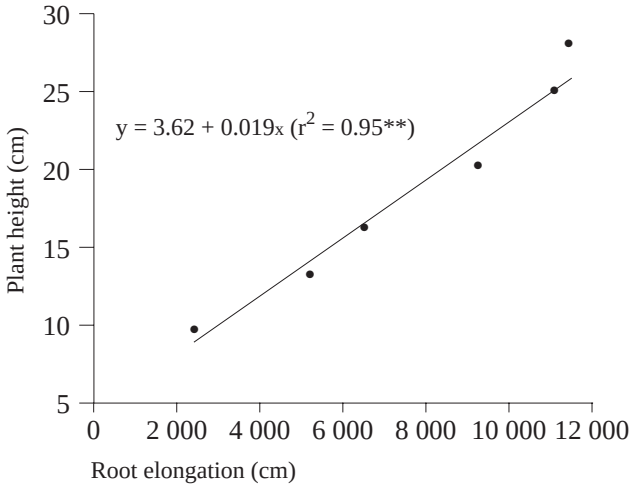

Figure 5. Linear relationship between plant height and root elongation in starfruit

(1994) similarly found that longer root length is associated with active vegetative vigour of *Vitis vinifera* cultivar 'Shiraz'.

Decrease in rooting volumes showed a reduction in both root length density (RLD) and declines. This demonstrates that under stress conditions, roots were thin and might be attributed to less water uptake and nutrient absorption as reflected in reduction of shoot growth. In a similar context, Bland and Dugas (1988) reported that there is a positive relationship between root counts and RLD in cotton, but no relationship in sorghum.

Changes in root chamber volumes had no significant effect on root:shoot ratio. Other studies also indicated that decrease in pot or container volumes in herbaceous plants results in no change in root:shoot ratio (Nesmith et al. 1992). These mean that there is always a proportionality between the activities of root systems and activity of the tops even under restricted conditions.

The reduction in leaf nutrient concentration especially N, P and Ca in reduced root volume was reflected by reduction in total root length and root surface area that showed a direct impact on nutrient uptake. However, reduced root volume caused no change in K and Mg. Even though nutrient concentration decreased but its nutrient levels were sufficient for plant growth. Van Noordwijk (1983) pointed out that root densities of 0.1–1.0 cm/cm^3 are sufficient for N and P uptake. It is believed that reduced Ca uptake was probably due to either constricted root vessels or thickening of root cells layer and suberization. Sharp and Davies (1985) showed that roots exposed to prolonged and severe restrictions tend to exhibit pronounced suberization.

Conclusion

Reduction in root chamber volume decreased root growth, root branching and root elongation but increased root length density. The percentage of dry matter partitioning and root:shoot ratio were not affected. Decrease in total root length and root surface area reduced N, P and Ca but caused no change in K and Mg levels. Reduction in all root activities had shown to correlate with reduction in shoot growth. Therefore, the techniques of root restriction might be possible to be used to control tree size in starfruit plants.

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