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QUALITY AND GROWTH DEVELOPMENT OF ROSELLE GROWN ON BRIS SOIL IN RELATION TO REGULATED DEFICIT IRRIGATION

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ABSTRACT

A greenhouse experiment was conducted to evaluate the effects of different irrigation schedules on postharvest quality and growth of roselle and also to determine a significant amount of water to be applied. Thirty-two roselle plants were grown on Beach Ridges Interspersed with Swales (BRIS) soil and treated with four different regimes namely control (100% full irrigation, I), 20% regulated deficit irrigation (RDI) (80% irrigation), 40% RDI (60% irrigation) and 60% RDI (40% irrigation). All the irrigation treatments were replicate quadrice and each replicate comprised two experimental units. RDI imposed for 91 days on roselle plants did not significantly influence plant growth (plant height, stem diameter and number of branches), postharvest quality (titratable acidity, firmness and total anthocyanins concentration) and cumulative number of roselle calyx. However, in general, water deficit treatments applied showed a significant effect only in fresh weight and soluble solid concentration (SSC) of roselle calyx. The treatment of 20% RDI increased yield (calyx fresh weight) compared to the control. In conclusion, 20% RDI (80% irrigation) tends to increase roselle yield and maintain plant growth development without adversely affecting calyx quality. Besides, 20% RDI saved water by 20% and increased irrigation use efficiency of roselle planted on BRIS soil.

Keywords: evapotranspiration, crop coefficient, water stress, calyx colour, regulated deficit irrigation

INTRODUCTION

Roselle (*Hibiscus sabdariffa* L.) is generally preferred because of their brilliant red calyx (Mahadevan et al. 2009) which contains higher anthocyanin concentration, vitamin C, B1, and B2. The concentration of vitamin C had been reported 2.5, 3 and 9 times higher compared to blackcurrant, grapes and citrus, respectively (Musa et al. 2006). Apart from contributing to the calyx colour, anthocyanins and other phenolic compounds have been reported to reduce various chronic diseases such as cancers, cardiovascular diseases, asthma and type II diabetes (Boyer and Liu 2004). Generally, the red and fleshy cup-shaped flower of roselle calyx are consumed world wide as hot and cold beverages, jellies, jam, sauces, preserves, and also for medicinal applications. Due to the high demand of roselle from industrial beverages and pharmaceuticals, there is a need to investigate the effects of internal and external factors in increasing the yield, growth and quality of roselle plants. Many internal and external factors such as genotype, intensity and types of light, orchard

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temperature, crop load and agronomic factors including agrochemical application, irrigation, pruning and fertilization affect the physiology and yield of roselle plants. However, research on the effect of irrigation mainly water deficit technique on postharvest quality of roselle and its growth development is still scarce. Water deficit techniques such as regulated deficit irrigation (RDI), withholding irrigation (WHI) and partial rootzone drying (PRD) have been widely implemented in various countries mainly to improve water use efficiency and growth performance of various crops. However, the information on water deficit treatment on roselle plant grown in Malaysia is limited particularly on Beach Ridges Interspersed with Swales (BRIS) soil.

In Malaysia, roselle is mostly planted on sandy soil such as BRIS soil. Terengganu has a vast area of BRIS soil with 154, 000 hectares (Roslan et al. 2011). Most of the crops grown on BRIS soil did not perform well due to high surface soil temperature, low water retention, low organic matter, high infiltration rate and low nutrients content. Although BRIS soil is a problematic soil, roselle is suitable to be planted on this soil because it provides a well-aerated and deep rooting zone. In addition, a low water holding capacity of this soil could be managed by applying optimal irrigation management such as RDI. This could avoid water wastage as every drop of water is precious. Water scarcity is also a constraint for agriculture in this soil area. Due to the high importance of water use in agriculture such as in roselle industry, there is an urgent need to identify and adopt irrigation management strategies in improving roselle yield, growth and quality performance. Therefore, one of the suitable solutions is by imposing RDI technique that has been well documented with promising outcomes. The present study aimed to determine the effects of different regimes of irrigation on quality and postharvest performance of roselle calyx, and to determine the exact quantity of irrigation water to be applied on roselle plant grown on BRIS soil.

MATERIALS AND METHODS

Plant materials and experimental location

Thirty-two roselle plants variety Terengganu (UMKL-1) were grown in a greenhouse at the School of Food Science and Technology, Universiti Malaysia Terengganu. BRIS soil was taken from Department of Agriculture Commodities Center, Rhu Tapai, Terengganu. Roselle seeds were purchased from Department of Agriculture, Kuala Berang, Terengganu.

Experimental design and application of irrigation

Roselle seeds were sown on 29th November 2012. The roselle seedlings were initially sown in a peat moss medium before transplantation after two weeks into polybag (20cm × 24cm) containing 35kg BRIS soil. The experimental trees were planted 0.7m within row and 0.5m between rows. The irrigation treatments were control (100% full irrigation, I), 20% regulated deficit irrigation (RDI) (80%

irrigation), 40% RDI (60% irrigation) and 60% RDI (40% irrigation) with four replications each. The amount of irrigation for each treatment was regulated according to the amount of 100% irrigation (control). The irrigation for control was 2.15 L per plant per day, which was calculated based on evapotranspiration formulae as below:

$$Etc = ETo \times Kc$$

Where;
ETc=crop evapotranspiration
ETo=reference crop evapotranspiration
Kc= crop coefficient
Average Kc was taken as 1.2 from a generalized Kc chart by Doorenbos and Pruitt (1977). Average ETo was 1.99 mm/day (Niazuddin 2007). The ETo was calculated from Penman-Monteith equation according to Allen et al. (1998) as shown below:

$$ETo = \frac{0.408\,\Delta\,(Rn - G) + Y\,\frac{900}{T+2732}U_2(Is - Ia)}{\Delta + Y(1 + 0.34U_2)}$$

Where;

Δ	= slope of vapour pressure curve, kPa/°C
Rn	= net radiation at the crop surface, MJm^{-2}/day
G	= soil heat flux density, MJm^{-2}/day
T	= air temperature at 2m height, °C
U_2	= wind speed at 2m height, m/s
I_s	= saturated vapour pressure, kPa
I_a	= actual vapour pressure, kPa
$(I_s - I_a)$	= saturated vapour pressure deficit, kPa
Y	= psychometric contant, kPa

The irrigation was imposed for 91 days. Two trees represented an experimental unit. The experiment was laid out following a randomized complete block design. The dripper (Pressure Compensating Dripper, DIY) with a flow rate at 4 L per hr was used for irrigation. Irrigation was applied twice a day at 0930 to 1000 hours and 1630 to 1700 hours. All experimental plants received similar cultural practices including fertilization (NPK blue and green at 83kg ha^{-1} and 194 kg ha^{-1} respectively) and pesticide sprays (imidacloprid and carbendazim) except irrigation.

Preharvest parameter

Thirty-two roselle plants were used to determine plant height and stem diameter were measured using ruler and digital vernier caliper, respectively. Plant height and stem diameter were expressed in cm and mm, respectively. The assessments were done every seven-day intervals viz. 0, 7, 14, 21, 28, 35, 42, 49, 56, 63, 70, 77 and 84 days after transplanting (DAT). Meanwhile, number of branches was recorded per plant from day 14 after transplanting.

Postharvest parameter

Fresh weight and number of calyx (yield)
The cumulative fresh weight and number of roselle calyx were recorded on day 70, 77 and 84 after transplanting and the former was expressed in gram (g).

Calyx color, calyx firmness, soluble solids concentration (SSC) and titratable acidity (TA)
Ten roselle calyces were used to record its colour indices using a Minolta Chroma Meter (Model R200 Trimulus Colour Analyser, Minolta camera Co. Ltd., Japan). Colour was measured at three sides of roselle calyx. Calyx colour data were expressed in L*, a* and b* values. L* represented the lightness coefficient which ranges from 0 (black) to 100 (white). a* ranged from-60 to +60, which indicates red (+60) and green (-60) colours. Meanwhile, b* ranged from-60 to +60, which indicates as yellow (+60) and blue (-60) colours. a* and b* were further used to calculate hue angle (h°= tan-1 b*/a*) for colour interpretation. Hue angle (h°) represented red-purple (0°), yellow (90°), bluish green (180°) and blue (270°) (McGuire 1992).

Three roselle calyces were used to determine calyx firmness using Texture Analyzer TA. Xtplus (Stable Macro System, UK). A 2 mm stainless steel probe (P2) was used and the post speed, test speed, pre test speed and deformation were 1.0, 0.05, 1.0 and 0.7 mm, respectively. For each roselle calyx, two opposite calyces were chosen and cut into 2 cm x 2 cm. Calyx firmness data were recorded and expressed in unit of Newton (N). Meanwhile, SSC was measured using handheld refractometer and expressed in percentage (%). The TA was calculated using formulae as below:

$$\%\ \text{malic acid} = \frac{\text{Titrate} \times \text{normality of alkali} \times \text{vol. made up} \times \text{equal weight} \times 100}{\text{Vol. taken for estimation} \times \text{weight of sample} \times 1000}$$

Where, equal weight = 134 mg

Total anthocyanin concentration
Anthocyanin was extracted from roselle calyx and quantified following the method described by Wan Zaliha (2009). Roselle calyx was soaked in methanol: hydrochloric acid (HCl) solution for ± 18 hours at 2°C to 4°C in the dark. The extract was then decanted and centrifuged. The supernatant was then determined using an UV-VIS spectrophotometer at 530nm. Total anthocyanins were calculated using a molar co-efficient of 2.74×10^4 (Giusti and Wrosltad 2001) and expressed in $\mu g\ g^{-1}$ fresh weight.

Statistical analysis

The data were subjected to the analysis of variance (ANOVA) using GLM (General Linear Models) procedures and further separated by Least Significant Difference (LSD) at $P \leq 0.05$ (SAS Institute Inc. 1999).

RESULTS AND DISCUSSION

The effects of RDI on the growth development and yield of roselle

As shown in Figures 1, 2 and 3, no significant effect was recorded on the growth and development of roselle plants grown on BRIS soil with the application of different irrigation regimes. In general, all plant growth parameters showed an increasing trend throughout 91 days of the experimental period. Similarly, Nur Razlin et al. (2013) reported that water deficit imposed for 65 days had no effect on growth of roselle planted on BRIS soil. In addition, Babatunde and Mofoke (2006) reported that the application of different irrigation schedules did not affect plant growth parameters as mentioned above. Possibly, the characteristics of roselle with a strong tap root and high moisture stress tolerance allowed it to fit efficiently in areas with limited water. In addition, well aerated soil may increase the efficiency of root in water uptake under water stress condition. On day 70, a few branches of roselle plants were broken due to the heavy crop load.

The yield in terms of fresh weight of roselle calyx (with capsule) was comparable between control and 20% RDI treated-plants (Figure 4). In addition, the treatment of 20% RDI plants had higher calyx fresh weight than 60% RDI plants; 964.86 g and 722.11 g, respectively. Meanwhile, no apparent effect of water deficit on the number of cumulative roselle calyx per plant was recorded (Figure 5). Increased fresh weight of roselle calyx may be attributed to the mild water stress imposed. This was in agreement with the reports of Behboudian and Singh (2001). Previously, Zegbe-Dominguez et al. (2003) claimed that dry weight of tomato var. ‘Petopride’ did not decrease under RDI and PRD compared to full irrigation. Similarly, Nur Razlin et al. (2013) also reported that the yield of roselle planted on BRIS soil under PRD was similar to control. In contrast, many previous findings reported that the yield in terms of fresh weight, total dry weight and number of fruits were reduced under water deficit treatments. However, in the present study, water stressed plants (20% RDI) produced 15% more roselle fresh weight than control which saved 20% of irrigation water. Similarly, Zegbe-Dominguez et al. (2003) reported that deficit irrigation saved 50% of irrigation water and increased 200% water use efficiency (WUE) in tomato.

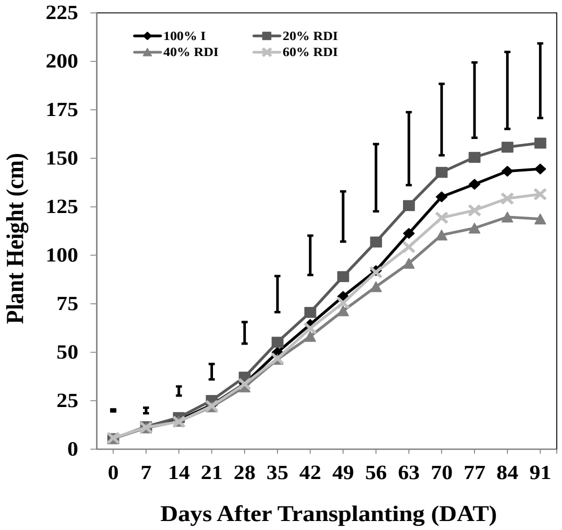

Figure 1. The effects of RDI on plant height of roselle grown on BRIS soil. Vertical bars represent $LSD_{0.05}$.

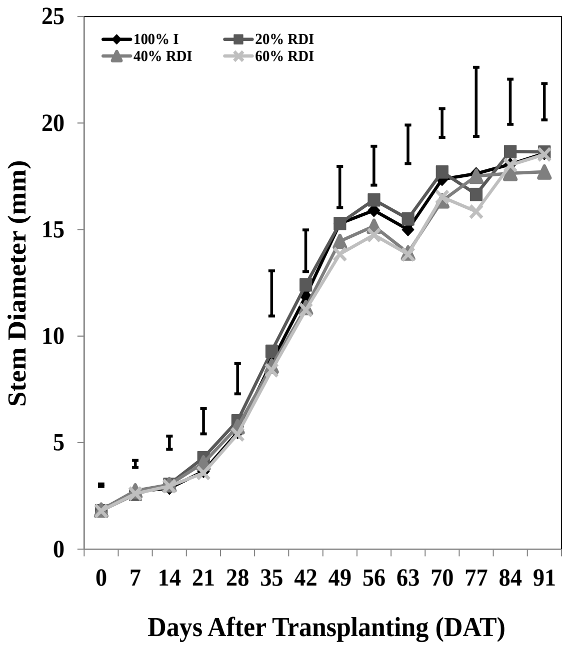

Figure 2. The effects of RDI on stem diameter of roselle grown on BRIS soil. Vertical bars represent $LSD_{0.05}$.

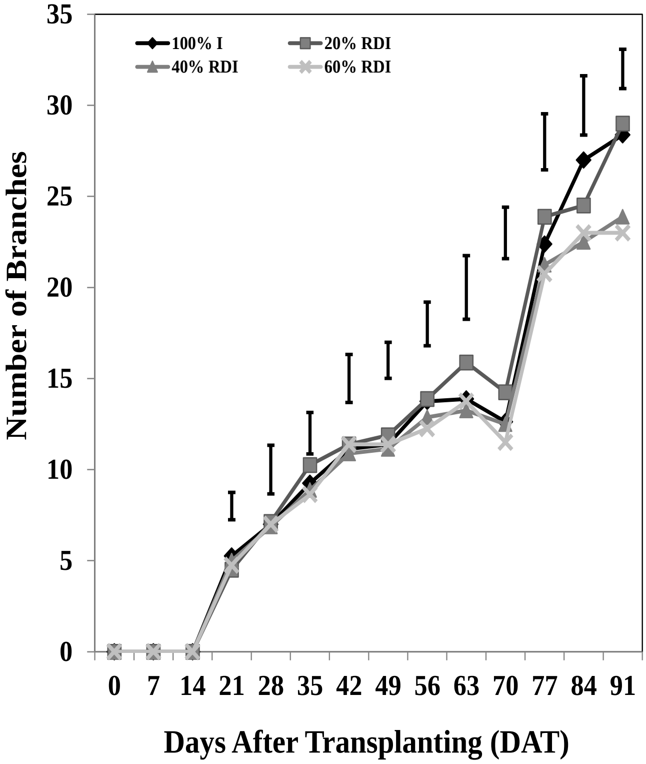

Figure 3. The effects of RDI on number of branches of roselle grown on BRIS soil. Vertical bars represent $LSD_{0.05}$.

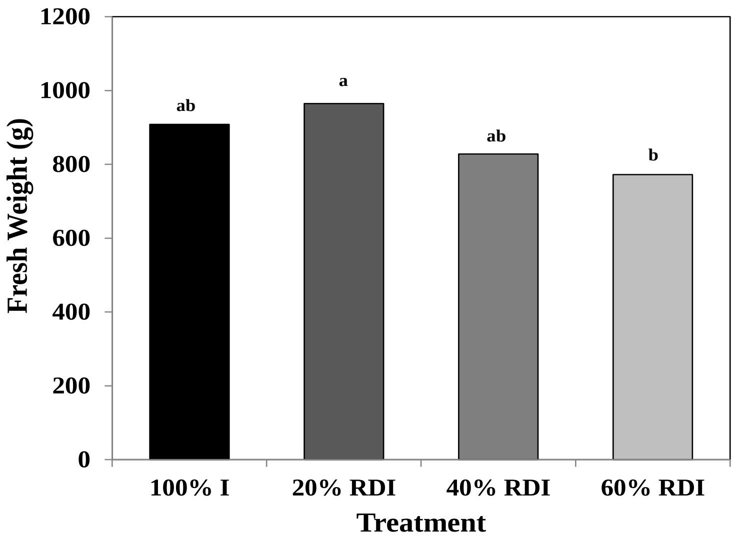

Figure 4. The effects of RDI on roselle fresh weight. Means with different letters are significantly different at the 5% level according to LSD test.

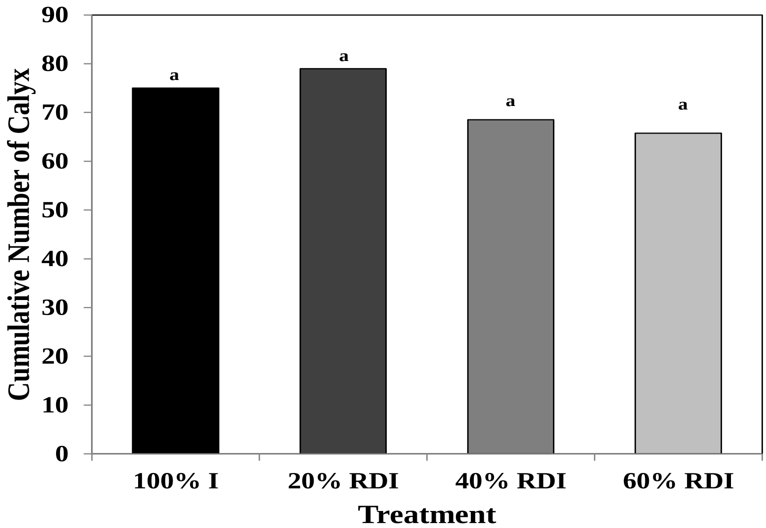

Figure 5. The effects of RDI on roselle calyx number. Means with different letters are significantly different at the 5% level according to LSD test.

The effects of RDI on the postharvest quality of roselle

Higher chromaticity value a*, lower b*, lower L* and h° indicates redder calyx colour. All roselle colour indices as mentioned above were not significantly affected with the application of different irrigation regimes (Table 1). Although no significant difference was recorded in all calyx colour indices, 20% RDI treated-plants showed lower chromaticity value b*, h° and L* (6.16, 15.30 and 27.79 respectively) which indicated redder calyx colour as compared to other irrigation treatments. Similarly, Nur Razlin et al. (2013) also noticed that L*, a* and b* values of roselle calyx treated with PRD were similar to control calyx. In addition, many previous reports also claimed that the application of deficit irrigation did not significantly affect the red skin colour of 'Braeburn and Delicious' apples and also pears (Ebel et al. 1993; Mpelasoka et al. 2001). However, many studies had also reported that the pronounced effects of RDI on fruit colour development of various apple fruits (Drake et al. 1981; Mills et al. 1994, 1996; Kilili et al. 1996; Wan Zaliha 2009; Wan Zaliha & Singh 2010). Wan Zaliha and Singh (2010) reported that improved Cripps Pink apple fruit color and increased concentration of anthocyanins with RDI may be ascribed to the increased levels of abscisic acid (ABA) and/or ABA induced ethylene production which would upregulate gene expression of anthocyanin biosynthesis. Fruit colour may also be enhanced as anthocyanin development was encouraged with the increased fruit sugar (Behboudian and Singh, 2001). In the present study, total anthocyanins of roselle calyx showed no effect under different irrigation regimes which may correspond to the calyx colour indices (Table 2). Possibly, the water deficit plants experienced mild water stress which masked the effects on hydraulic status of soil and leaf of roselle plant and produced similar calyx colour to control plants. According to Hsiao (1973), water stress treatments tended to impose a mild stress on plants by lowering stem water potential (ψ_{stem}) to below 0.50 MPa. The readings in the present study were mainly between -0.10 MPa to -0.41 MPa throughout the 91 days of observation, with the exception of days 70 to 91 when readings reached -0.55 MPa to -0.63 MPa respectively (Wan Zaliha: unpublished data). The fluctuations in ψ_{stem} might be associated with the rainy season as the experiment was conducted in November 2012 until January 2013. Even though the experiment was conducted in the greenhouse, but the cold air surroundings might have affected the values of ψ_{stem}. This may reflect the masking effect as discussed above. In addition, during water stress, the dehydrated rootzone is expected to stimulate the secretion of the root-to-shoot chemical signal, abscisic acid (ABA) that triggers the closing of stomata, which then reduces stomatal conductance, transpiration, shoot growth to maintain plant water potential. As discussed earlier, ABA play a key role in inducing ethylene production due to water-stress and involved in the synthesis of anthocyanins. However, the mild water stress might not be sufficient enough to enhance the red skin coloration of roselle calyx. The results obtained were similar with the reports of Nur Razlin et al. (2013). However, Wan Zaliha and Singh (2010) claimed that the increased red skin colour of Cripps Pink apple coincided with the increased in total anthocyanins concentration. Mandour et al. (1979) reported that excess or lack of water supply during the vegetative growth and developmental stage of roselle plants decreased the chlorophyll and carotenoids while medium supply of irrigation water increased the pigmentation in the leaf and other organs.

Other calyx quality parameters such as titratable acidity (TA) and firmness were not adversely affected by different RDI regimes except SSC (Table 2). At harvest, the roselle calyx showed a higher value of SSC which was 3.30% higher than those reported by Wong et al. (2002). The treatment (20% RDI) had the highest SSC and comparable to control. Similarly, El-Boraei et al. (2009) reported that SSC in roselle calyx increased as irrigation water quantities decreased. Dorji et al. (2005) also found that the high SSC at final harvest of hot pepper in water deficit treatment was due to the reduced fruit water content and greater hydrolysis of starch into sugar. Meanwhile the firmness and TA of roselle calyx under 20% RDI treatments were less firm and comparable to control treatment, respectively (Table 2). In contrast, Leib et al. (2006) reported that Fuji apple fruits produced by deficit irrigation were firmer than control fruits. This was supported by Ebel et al. (1993) who reported that water deficit fruits are generally firmer than fully irrigated fruits due to higher cellular density. Wan Zaliha and Singh (2010) also concluded that firmer fruits in RDI might be associated with the reduction in cellular hydration and increased flesh compactness that could cause source limitation of photosynthesis due to lower stomatal conductance in water deficit plants. However, these outcomes need to be further investigated.

Table 1. Effects of different irrigation treatments on lightness (L*), chromaticity value a*, b* and hue angle (h°) of roselle calyx

Treatment	L*	a*	b*	h°
100% I	28.24a	23.16 a	6.73 a	16.22a
20% RDI	27.78a	22.15 a	6.16 a	15.3a
40% RDI	28.08a	23.44 a	7.11 a	16.76a
60% RDI	29.84a	23.65 a	6.92 a	16.33a

Means with different letters are significantly different at the 5% level according to LSD test. I = full irrigation and RDI = regulated deficit irrigation.

Table 2. Effects of different irrigation treatments on total anthocyanins, calyx firmness, TA and SSC of roselle calyx

Treatment	Total anthocyanins (mg/g fresh weight)	TA (% malic acid)	SSC (%)	Calyx firmness (N)
100% I	358a	5.23a	5.8a	2.02a
20% RDI	342a	5.23a	5.8a	1.30a
40% RDI	317a	5.50a	5.4a	2.30a
60% RDI	284a	5.36a	5.7a	2.49a

Means with different letters are significantly different at the 5% level according to LSD test. I = full irrigation and RDI = regulated deficit irrigation.

CONCLUSION

Various deficit irrigation scheduling imposed for 91 days on roselle plants had no effects on plant growth namely plant height, stem diameter and number of branches, postharvest calyx quality such as TA, firmness, calyx colour and total anthocyanins concentration and cumulative number of calyx per plant. However, the water deficit treatments showed a significant effect on fresh weight and SSC of roselle calyx. The 20% RDI (860mL water per plant per day) increased yield and maintained plant growth without adversely affecting calyx quality. The 20% RDI also saved 20% irrigation water and increased irrigation use efficiency.

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