Evaluation of Different Water Regimes on *Hevea brasiliensis* Grown on Haplic Ferralsol Soil at Nursery Stage

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Abstract

The main source of natural rubber is from Hevea brasiliensis. Demand for natural rubber is expected to increase once more with increasing of crude oil price and demand for developing automotive industry in several countries. Suitable planting areas decreases and invaluable water resource has been new challenges. This study will provide details about effect of water deficiency in Hevea brasiliensis. Two new latex timber clones from Malaysia Rubber Board (MRB), RRIM 2001 and RRIM 3001 were used in this study. Five levels of treatment were used; plants irrigated for every two days, five days, 10 days, 15days and everyday which acted as control. The experimental design used was a completely randomized block design (RCBD) with four replications. Fundamental changes of plant growth and physiological responses showed that treatment with well watered for clone RRIM 2001 (T1) had higher values than other treatments. Photosynthesis rate highest in well watered (T6) with mean 11.26µmol $m^{-2}s^{-1}$, while T4, T5 and T9 were lowest with the mean 0.00µmol $m^{-2}s^{-1}$. Stomata conductance showed significant difference between T6 with 0.16µmol $m^{-2}s^{-1}$ compared to under stress treatments with 0.00 µmol $m^{-2}s^{-1}$. The results also showed root length increase with increasing of water stress. There was treatment failure to adapt to water stress at treatments withholding water for 15days followed by treatments 10days and five days. RRIM 2001 and RRIM 3001 clones had responded to water stress by indicating changes in morphological and physiological responses. This concluded that Hevea brasiliensis cannot withstand water stress at nursery stage and replanting in dry areas.

Keywords: Rubber, Latex Timber Clones (LTC), water stress, growth, physiological, responses, dry areas.

1. Introduction

Hevea brasiliensis or rubber tree, originated from Amazon basin have been widely planted in South East Asia. Nowadays, the usage of rubber has become more increasingly important in daily life (Noordin, 1993). This scenario has triggered rubber planting to be shifted to marginal areas, such as dry areas (Noordin, 2010). Water has been a very important resource to agriculture since the issues of global warming and climate change arise. Water is scarce and its quality is decreasing in many parts of the world (A. Gholizadeh *et al.*, 2010; Amy c. Fuller and Michael O. Harhay, 2010; Raviv and Blom, 2001). Pertaining to water as an invaluable resource, there is a need to use this source wisely. Water deficit stress can be defined as a situation in which plant water potential and turgor are reduced to interface with normal functions (Shao *et al.*, 2008). Plant water status is also affected by reducing of water availability in plant (Shafar *et al.*, 2011). More than that, water stress influences plant growth at various levels, including cell community (Wafa'a A. Al-Taisan, 2010; Blumwald *et al.*, 2004; Colom and Vazzana, 2001). The quantity and quality of plant growth depend on cell division enlargement, and differentiation and all of these events are affected by water stress (Cabuslay *et al.*, 2002; Correia *et al.*, 2001). It also reduced plant growth inhibition of various physiological and biochemical processes such as photosynthesis, respiration translocation, ion uptake, carbohydrate, nutrients metabolism and hormone (Bhatt and Srinivasa, 2005; Chaitanya *et al.*, 2003; Blum, 1996).

Latex timber clones (LTC) are clones with high latex yield and rubberwood production. They exhibit good growth, good growth vigor and posses long straight boles. These clones are suitable for the production of latex and rubberwood or production of rubberwood (Malaysian Rubber Board, 2009). The parental of clone RRIM 2001 is RRIM 600 x PB 260. The laminae is elliptical obovate. Color of foliage is light green, semi glossy and smooth. Clone RRIM 3001 has an elliptical shape. The color of foliage is dark shining green. Both clone are recommended for both latex and timber production (Malaysian Rubber Board, 2009).

Soil from Munchong series are widely planted with rubber in Malaysia. According to USDA soil taxonomy, Munchong series has been classified as very fine, Tropeptic Haplorthox (Noordin, 1981, 1975). In the FAO/UNESCO Legend, this soil has been classified as Haplic Ferralsol (Department of Agriculture, 2008). Munchong series soil is a silty clay loam to silty clay with yellowish brown to strong brown. The structure of this soil series is moderate to strong fine and medium sub-angular blocky. This soil has been classified as the first class soil for rubber planting in term of soil-crop suitability (Malaysia Rubber Board, 2009; Noordin, 1981).

2. Materials and Methods

2.1 Plant materials and experimental treatments

This study was carried out under rain shelter. The source of rubber clone was from Malaysia Rubber Board (MRB) and consists of three month rootstock from clone RRIM 2001 and RRIM 3001. The plants were selected according to the average height and girth. Polythene bags size used was 40 cm x 45 cm which can withhold the planting media up to 10kg. The media used was Munchong soil series. Sources of fertilizer are RISDA 1 fertilizer consisting of 10% N, 16% P, 9% K and 2.5% Mg. RISDA 1 is recommended by Rubber Industry Smallholders Development Authority for young rubber. Study about fertilizer on Latex Timber Clones has proved rubber plants will scorched and dead if over fertilized (Shafar and Noordin, 2011a, 2011b). Treatments 1 to 5 (T1 until T5) were from RRIM 2001, while treatments 6 to 10 (T6 until T10) were from RRIM 3001. For water stress treatments, various water frequencies by varying number of days was applied. Treatments comprised the control as well watered (T1 and T6), withholding water for two days (T2 and T7), withholding water for five days (T3 and T8), withholding water for 10days (T4 and T9) and withholding water for 15days (T5 and T10).

2.2 Water requirement determination

Water requirement was determined by measuring water retention of the soil using a pressure chamber and the pressure plate (Teh and Jamal, 2006). A core ring measuring about 7.6 and 4.0cm in diameter and depth, respectively, and having known weight was hammered into the soil to ring depth. The samples retaining rings placed on each of the four porous plates for 1, 10, 33 and 1500 kPa pressure. The core (undisturbed) sample was broken up into five pieces of equal sizes. One piece of sample in retaining ring was placed on each of the porous plates. For the 0 bar pressure (saturation), the samples is placed in a retaining ring on a coarse wire mesh. All ceramic plates were saturated for 24 hours by keeping the water level just below the edge of the ring. The plate with samples was placed inside the corresponding pressure chamber. The plate was connected to the outflow tube. The chamber was closed and pressure applied. Equilibrium was attained when no more outflows occurs. A period of 4-7 days is usually sufficient to achieve moist soil. The chamber opened, the samples removed and each of them was weighed (Wa). The sample oven-dried at 105°C for 24hours, and each of the soil samples weighed again (Wb). The calculation for volumetric water content was: (m^3m^{-3}), θv = (Wa-Wb)/Wb x ρ_b, where ρ_b=bulk density, Wa =dry weight, Wb=oven dried sample after dry weight.

2.3 Data collection

2.3.1 Photosynthesis rate and stomata conductance

The rate of photosynthesis and stomata conductance had been measured by Portable Photosynthesis System Model LICOR-6200. The fully expanded leaves were selected to measure the photosynthesis rate and stomata conductance.

2.3.2 Total chlorophyll content

Data was taken using the chlorophyll meter SPAD-502. The Chlorophyll meter SPAD-502 was calibrated by pressing the measuring head closed without inserting the leaf.

2.3.3 Leaf nutrient contents

The concentration of Nitrogen (N), Phosphorus (P) and Potassium (K) in finely grind dried leaves were determined. Sample with weight 0.25g was digested in 5mL of sulfuric acid (H_2SO_4) on hot plate at 450°C in a fume chamber for 7 minutes. Ten mL of hydrogen peroxide (H_2O_2) was then added into the mixtures and heating continued for another four minutes. The solutions were made up to 100mL with distilled water and filtered with filter paper. Contents of N and P were determined using an auto-analyzer (LACHART Instruments, Model Quickhem IC + FIA 8000 Series) while K were measured using an atomic absorption spectrophotometer (Perkin Elmer, Model AAS 3110).

2.3.4 Root length and root volume

The root length and volume measurement was taken using the root scanner. The root inserted to the suitable tray and water was filled up half full to the tray. Then, the scanner head was closed and root was scanned using the WHIRHIZO software. The color was defined between root and background.

2.4 Experimental design and data analysis

The study was conducted in a completely randomized block design with four replications. Each block comprised of 30 plants and each treatment consists of three samples (polythene bags). There were two clones consisting of five treatments per clone. Analysis of Variance (ANOVA) on data obtained was performed using Statistical Analysis System (SAS 9.1, SAS Institute, Inc. Cary NC. USA). Least Significant Different (LSD) test at $p<0.05$ was employed for mean comparison.

3. Results and Discussion

3.1 Water requirement

Figure 1 shows moisture content at saturation (0.1kPa), field capacity (33kPa) and permanent wilting points (1500kPa) for sample are 0.365, 0.255 and 0.205 m^3m^{-3} respectively. The available water content for plant use is the difference between field capacity and permanent wilting point (Teh and Jamal, 2006). The available water content for plant used was 0.255-0.20.5 = 0.05 m^3m^{-3}. The amount of water used for 10kg of Munchong soil series in this study is 0.5L.

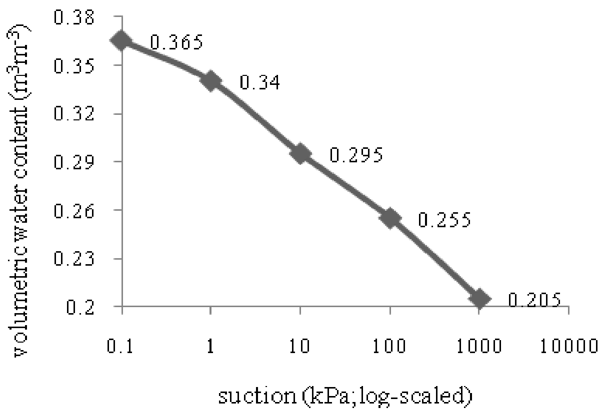

Fig. 1: Soil water retention curve

Table 1: Physiological responses of *Hevea brasiliensis* under water stress condition

Treatment	Photosynthesis rate (µmol $m^{-2}s^{-1}$)	Stomata conductance (µmol $m^{-2}s^{-1}$)	Chlorophyll content (SPAD unit)
RRIM 2001 well watered (T1)	3.89b	0.11ab	51.00a
RRIM 2001 withholding water for two days (T2)	2.06b	0.03bc	49.10a
RRIM 2001 withholding water for five days (T3)	1.06b	0.05ab	34.50c
RRIM 2001 withholding water for 10days (T4)	0.00b	0.00c	13.47d
RRIM 2001 withholding water for 15days (T5)	0.00b	0.00c	12.77d
RRIM 3001 well watered (T6)	11.26a	0.16a	47.67ab
RRIM 3001 withholding water for two days (T7)	4.07b	0.05ab	39.07bc
RRIM 3001 withholding water for five days (T8)	1.69b	0.02c	36.13c
RRIM 3001 withholding water for 10days (T9)	0.00b	0.00c	10.37d
RRIM 3001 withholding water for 15days (T10)	0.35b	0.00c	17.63d
$LSD_{0.05}$	5.71	0.09	9.99

Values in each column with same letter did not differ significantly at $p<0.05$ according to LSD

Photosynthesis rate of well watered plants showed that T6 from clone RRIM 3001 were significantly higher than T1 (RRIM 2001).From Table 1, the highest mean of stomata conductance among treatments was T6 (well watered) with the mean 0.161 µmol $m^{-2}s^{-1}$, while the lowest mean was T4, T5 and T9 with 0.00 µmol $m^{-2}s^{-1}$. The leaves were falling and became chlorosis after 2 weeks of treatments for T4, T5 and T9. Photosynthesis process was inhibited when the water was not enough to translocate sucrose and hexose to the leaves as a sink and this was showed by the lowest value in the Table 3. Under water stress condition, photosynthesis pigments and compounds changed (Anjum *et al.*, 2003), apparatus damaged (Fu and Huang, 2001) and activities of enzymes in Calvin cycle also diminished, which lead to reduction in crop yield (Monakhova and Chernyadev, 2002).

Internal water deficits results reduction of stomata conductance (Shafar *et al.*, 2011; Duli Zhao *et al.*, 2010). This was caused by root volumes restrictions rather than a general shortage of soil moisture level. Oxygen levels limitation might also trigger the root to shoot signal via chemical means. Table 1 showed there were very significant difference for total chlorophyll content among treatments at p<0.05. The highest means of total chlorophyll content was achieved in T1 (well-watered). In T3, T4, T5, T9 and T10, the leaves were falling and became chlorosis after three weeks of treatment. Shafar *et al.*, (2011) and Bertamini *et al.*, (2006) reported that the water deficit induced reduction in chlorophyll content has been ascribed to loss of chloroplast membranes, excessive swelling, distortion of the lamellae vesiculation, and the appearance of lipid droplets.

Table 2: Root length and rot volume under different water regimes after six weeks of treatment.

Treatment	Root length (cm)	Root volume (cm^3)
RRIM 2001 well watered (T1)	1575.5ab	85.03a
RRIM 2001 withholding water for two days (T2)	1350.8ab	19.40a
RRIM 2001 withholding water for five days (T3)	1284.7ab	26.66a
RRIM 2001 withholding water for 10days (T4)	1604.3ab	50.59a
RRIM 2001 withholding water for 15days (T5)	1587.2ab	93.78a
RRIM 3001 well watered (T6)	1237.7ab	92.13a
RRIM 3001 withholding water for two days (T7)	1338.1ab	41.23a
RRIM 3001 withholding water for five days (T8)	1197.5b	20.68a
RRIM 3001 withholding water for 10days (T9)	1773.8a	29.44a
RRIM 3001 withholding water for 15days (T10)	1615.0ab	28.93a
$LSD_{0.05}$	572.91	77.61

Values in each column with same letter did not differ significantly at p<0.05 according to LSD

Table 2 showed there were no significant difference of root length and root volume among the treatments at p<0.05 after six weeks of treatments. The highest mean of root length and root volume among the treatments was T9 (withholding water for 10days) with the mean 1773.8cm and T5(withholding water for 15days) with the mean 93.8cm^3. Roots are the key plant organ for adaptation to drought. The possession of a deep and thick root system allowed access to water deep in the soil, which was considered important in determining drought resistance (Kavar *et al.*, 2007). The structure and distribution is important for root compared to quantity of roots that determines the most efficient strategy for extracting water during the crop growing season (Farooq *et al.*, 2008).

Table 3: Leaf nutrient content of *Hevea brasiliensis* under different water regimes

Treatment	Nitrogen (ppm)	Phosphorus (ppm)	Potassium (ppm)
RRIM 2001 well watered (T1)	80.97a	4.22a	24.93a
RRIM 2001 withholding water for two days (T2)	67.70b	3.19b	27.70a
RRIM 2001 withholding water for five days (T3)	0.00c	0.00c	0.00b
RRIM 2001 withholding water for 10days (T4)	0.00c	0.00c	0.00b
RRIM 2001 withholding water for 15days (T5)	0.00c	0.00c	0.00b
RRIM 3001 well watered (T6)	76.77a	3.73ab	26.17a
RRIM 3001 withholding water for two days (T7)	76.23a	3.46b	25.68a
RRIM 3001 withholding water for five days (T8)	0.00c	0.00c	0.00b
RRIM 3001 withholding water for 10days (T9)	0.00c	0.00c	0.00b
RRIM 3001 withholding water for 15days (T10)	0.00c	0.00c	0.00b
$LSD_{0.05}$	6.94	0.56	4.80

Values in each column with same letter did not differ significantly at $p<0.05$ according to LSD

From table 3, there were very significant differences for nitrogen, phosphorus and potassium content among the treatments at $p<0.05$. In T3, T4, T5, T8, T9 and T10, the leaves were falling and became chlorosis after two weeks of treatment. Nitrogen and phosphorus content showed the highest value on RRIM 2001 than RRIM 3001, while for the potassium content, the treatment withholding water for two days was given the highest value. Decreasing water availability under drought generally results in limited total nutrient uptake and their diminished tissue concentrations in crop plants (Farooq *et al.*, 2008). An important effect of water deficit is on acquisition of nutrients by the root and their transportation to shoots. Besides that, the drought influence on plant nutrition may be related to limited availability of energy for assimilation of NO_3^-/NH_4^+, PO_4^{-3} and SO_4^{-2}. They must be converted to energy dependent processes before these ions can be used for growth and development of plants (Grossman and Takahashi, 2001).

4. Conclusion

From the results, it can be concluded that the two clones of rubber, RRIM 2001 and RRIM 3001 had responded to water stress by indicating changes in morphological and physiological responses. The changes in those aspects showed that treatment with well watered procedure for clone RRIM 2001(T1) had high values than other treatments. In water deficit condition, nutrients uptake and photosynthesis processes which influence stomata conductance and chlorophyll content were reduced. The results also showed that rubber plant cannot withstand water stress at nursery stage and replanting in dry areas.

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