Journal of Agricultural Technology 2014 Vol. 10(3): 743-754
Available online http://www.ijat-aatsea.com
ISSN 1686-9141

Difference in physiological responses to water stress between two rubbers (*Hevea brasiliensis*) clones of RRIM 600 and RRIT 251

Supawadee Sittichai and Sayan Sdoodee*

Department of Plant Science, Faculty of Natural Resources, Prince of Songkla University, Hat Yai, Songkhla 90112, Thailand

Supawadee Sittichai and Sayan Sdoodee (2013) Difference in physiological responses to water stress between two rubbers (*Hevea brasiliensis*) clones of RRIM 600 and RRIT 251. Journal of Agricultural Technology 9(7):743-754.

The physiological response of plant water stress is an interesting issue because of its difficult to be avoided. Therefore, the effects of water stress on physiological responses of rubber trees were investigated in a pot trial. RRIM 600 and RRIT 251 clones with the two whorl stage were used in this study. The planting materials of each clone were grown in 3.5 litres of polyvinyl chloride pots (one plant per pot) in the glasshouse at Faculty of Natural Resource, Prince of Songkla University, Songkhla province, Thailand. The experiment was arranged as a completely randomized design with two levels of watering regimes: well watered (w) and water stress (s). There were fourteen replicates in each treatment. The result showed that RRIM 600 and RRIT 251 clones were similar responses under well watered condition. Stomatal conductance of RRIM 600 clone rapidly decreased after 8 days of withholding water (407 mmol $m^{-2}s^{-1}$ to 42 $mmolm^{-2}s^{-1}$). While stomatal conductance of RRIT 251 clone was reduced gradually after 10 days of withholding water (369 $mmolm^{-2}s^{-1}$to 10 $mmolm^{-2}s^{-1}$). Leaf water potential of RRIT 251 clone was highly reduced starting from 10 days of withholding water (from – 0.96 to – 2.50 MPa). Photosynthesis and transpiration of RRIM 600 and RRIT 251 clones in water stressed conditions tend to decreased. However, there was no significant difference in the water stressed treatment between the both clones. Moreover, RRIT 251 clone was expressed the maximum quantum efficiency of PS II lower than RRIM 600 after 10 days of withholding water (0.794 and 0.801 respectively). The positive relationships were found between leaf water potential and stomatal conductance in RRIM 600 (R^2 = 0.87) and RRIT 251 clones (R^2 = 0.81), leaf water potential and photosynthesis in RRIM 600 (R^2 = 0.94) and RRIT 251 clones (R^2 = 0.93). In addition, the stomatal conductance, leaf water potential, photosynthesis and transpiration recoveries after rewatering would be restore to value of well watered. Hence, it was implied that RRIM 600 clone trended to be more tolerance to water stress than RRIT 251 clone when grown in the pot trial.

Keywords: *Hevea brasiliensis*, water stress, stomatal conductance, leaf water potential

* Corresponding author: Sayan Sdoodee, e-mail: sayan.s@psu.ac.th

Introduction

The rubber tree (*Hevea brasiliensis*) is an economic crop in Thailand. RRIT (2011) reported that Thailand is the main producer and export of natural rubber production in worldwide. Not only the higher demand of natural rubber but also the increase of rubber price are greatly pressure to rapidly expansion of new area rubber plantation and non-traditional cultivation area (North and Northeast) (RRIT, 2007). Climate change has also recently become an important factor affecting on available water resources, water stress, plant growth and agricultural productivity (Aydinalp and Cresser, 2008; Ackerman and Stanton, 2013).

In Thailand, the trends of climate change are increasing of minimum and maximum temperature decreasing of raining day (The Thailand Research Fund, 2011; Limsakul and Limjirakan, 2011) leading to adversely impact on physiological responses of rubber trees. Furthermore, it has been reported that the new rubber plantation area has unfavorable environmental conditions especially high temperature and low rainfall causing water stress. Under water stress condition, the young rubber tree cannot adapt because of soil water content is low, leading to the death. Water stress affects the survival of young rubber tree in the new plantation area. Under water stress, young rubber tree death may increases up to 93.48 percentages (RRIT, 2006).

The potential of growth can be reduced through impairment of cell division and cell expansion (Shao *et al.*, 2008). The physiological processes of the response to water stress are different depending on the severity and the duration of stress, growth stage at which stress is imposed and the genotype of the plant (Jaleel *et al.*, 2009). Hsiao (1973) found that the reduction of CO_2 assimilation is due to stomatal closure during exposure to water stress. The stomatal closure was reported as the limiting factor in net photosynthesis that is directly to the growth. Sangsing *et al.* (2004a); Chandrashekar *et al.* (1998) reported that the effect of drought on net photosynthesis, stomatal conductance, transpiration and leaf water potential were decreased when the increasing periods of water stress. However, the physiological responses of plant to water stress are an interesting issue, because it is difficult to be avoided. Thus, the objective of this study was to investigate the effect of water stress on stomatal conductance, leaf water potential, photosynthesis and transpiration of young rubber trees. This will be a guideline for the rubber smallholders to select the suitable clones for the new area or non-traditional plantation area.

Material and methods

Experimental set up

The experiment was conducted in the glasshouse of Prince of Songkla University, Songkhla Province, Thailand during June to July 2011. Two planting materials of rubber clones (RRIM 600 and RRIT 251) were used in the experiment. They were arranged in the glasshouse in completely randomized design. There were two treatments of watering regimes: well water and water stress condition with fourteen replications. Fifty six polyvinyl chloride pots were used, each pot was 10.16 cm. diameter and 50 cm. high with a perforated opening at the basal part were used for this experiment. Pots were filled with 3.5 litre of sandy clay loam. Pots were irrigated with water to field capacity.

Young rubber plant with different buds. Buds from two rubber varieties of RRIM 600 and RRIT 251; there were grafted on RRIM 600 rootstocks. Four-month old plants with two whorls (one plant per pot) were used in this study. The average plant size of high at 39.81-44.56 cm. and diameter at 3.95-4.39 cm. of both clones. Young rubber trees from nursery at Amphour Namom, Songkhla Province.

Water treatment

Two levels of water regimes were imposed and this include; well watered (w); water stress (s). Before the starting of stress treatment; all pots were irrigated to field capacity. In the well watering treatment, continuous watering was maintained around field capacity. Whereas the water stress treatment, complete withholding of water was imposed.

Recovery

Water stressed treatments were rewatered to field capacity when leaf water potential reached approximately -2.20 MPa, which a loss of hydraulic conductivity in the detached stems of rubber (Sangsing et al., 2004). During the period of recovery, physiological responses data were monitored.

Physiological responses

The measured of certain physiological responses of the two whorls young rubber trees during 10.00 -12.00 am.

Leaf water potential

Leaf water potential was measured with a pressure chamber (Soil Moisture Equipment Crop, Santa Barbara, CA, USA). Measurements were randomly made on six fully expanding leaves for each treatment.

Photosynthetic capacity

Net CO_2 assimilation rate (*A*) and transpiration were measured with aportable photosynthesis system model Li-6400)LCi Ultra Complex Photosynthesis System, USA(. Measurements were made on six fully expanding leaves for each treatment.

Stomatal conductance

Stomatal conductance was measured with porometer model AP4 (Delta-T Devices, Cambridge, England). Measurements were performed on six leaflets randomly chosen on each treatment.

Chlorophyll fluorescence measurements

Chlorophyll fluorescence was measured with a Plant Efficiency Analyzer MK2 (PEA) (Hansatech, Kings lynm, England) the fully expanded leaves (6 leaves per treatment) were dark adapted for 30 minutes. The following parameters were recorded;
Fo = minimal level when all the QA are in oxidized state (open reaction centres)
Fm = maximum fluorescence when the QA are reduced (close reaction centres)
Fv = (Fm-Fo), refer to as photochemical fluorescence quenching
Fv/Fm = maximum quantum efficiency of photosystem II (PS II)

Stomatal conductance (g_s)

In well watering period, the stomatal conductance was similar in both clones. However, stomatal conductance of RRIM 600 clone trended to be greater than RRIT 251 clone. In stressed condition, stomatal conductance was generally higher during 2-3 days after withholding water until 10 days of withholding water. But stomatal conductance of RRIM 600 was decreased rapidly from 407 mmolm^{-2}s^{-1} to 42 mmolm^{-2}s^{-1} within 8 days of stress. While stomatal conductance of RRIT 251 clone was reduced gradually with ongoing water stress after 10 days of withholding water from 369 mmolm^{-2}s^{-1}to 10

$mmolm^{-2}s^{-1}$ (Figure 1). RRIM 600 clone showed higher stress tolerance than clone RRIT 251 (Sangsing, 2004a). After 5-7 days of rewatering, the value of stomatal conductance was almost restored to value of well watered in both clones.

Chandrashekar *et al.* (1998); Zhao *et al.* (2010); Shafar *et al.* (2011) reported that the effect of water stress on plant was reduced stomatal conductance. The stomatal conductance is a parameter that mostly uses to reflect water stress (Flexas and Medrano, 2002). The stomatal pore can be closed to reduce water loss by closing the stomatal pore the water use efficiency is increased. The result showed that the safety margins were also higher in RRIM 600 than those in RRIT 251 (Sangsing and Rattanawong, 2012).

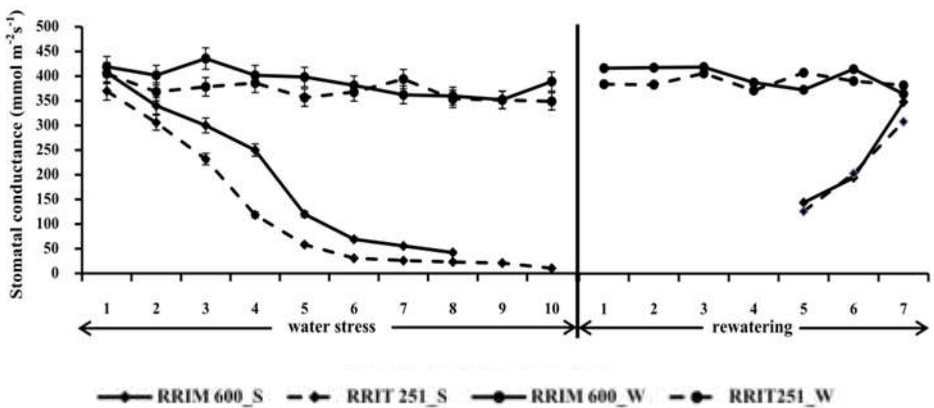

Fig. 1. Change of stomatal conductance (mmol m^{-2} s^{-1}) in two clones of *Hevea brasiliensis* at the water stress (s) conditions with well watered (w) conditions. Bars indicate standard error.

Leaf water potential

The leaf water potential is driving force for the movement of liquid water through the plant. In well watered, the leaf water potential of two clones was -0.94 to -1.15 MPa. In stressed treatment, the leaf water potential was decreased progressively with the increasing of water stress. Leaf water potential of RRIM 600 clone was – 0.96 MPa before stress and it was – 2.22 MPa after 10 days of withholding water. Leaf water potential of RRIT 251 clone was – 0.96 MPa before stress and it was – 2.50 MPa after 10 days of withholding water (Figure 2). Some leaves were fell down and removing leaves became chlorosis after 10 days of withholding water. After 6-7 days of rewatering the value of leaf water potential recovered to value of well watered in both clones.

The positive correlations between leaf water potential and stomatal conductance were found that in both clones (Figure 3). However, the correlation was higher in RRIM 600 clone (R^2= 0.87) than that in RRIT 251 clone (R^2= 0.81). The stomata closed when the leaf water potential approximately demand to – 1.93 MPa in RRIM 600 clone and – 2.30 MPa in RRIT 251, respectively. Sangsing *et al.* (2004b) reported that the smaller pots caused leaf water potential was quickly decreased values than the biggest pots. In the same time the water stress was rapidly occurred.

Photosynthesis and transpiration

Photosynthesis in well watered was higher in RRIM 600 (5.15-6.28 µmol CO_2 $m^{-2}s^{-1}$) than that in RRIT 251 (5.22-5.87 µmol CO_2 $m^{-2}s^{-1}$). RRIM 600 presented the higher photosynthesis rate. In stressed condition, the decreased of water availability causing decreases of photosynthesis and transpiration. Photosynthesis pigments and compounds changed and activities of enzymes in Calvin cycle also this might be due to diminished (Miguel *et al*,. 2007). The value of photosynthesis of RRIM 600 and RRIT 251 dropped steeply from 5.31 to 0.41 µmol CO_2 $m^{-2}s^{-1}$ and 5.81 to 0.51 µmol CO_2 $m^{-2}s^{-1}$ after 10 days of withholding water respectively)Figure 4.). The value of transpiration of RRIM 600 and RRIT 251 was reduced when the increasing of stress (Figure 6.). After 10 day of withholding water, the plant was sufficient to induce defoliate and wilting of stem. After 6-7 day of rewatering the value of photosynthesis was restored to value of well watered in both clones. But RRIM 600 showed rapidly to recovered than RRIT 251. For transpiration, the value According to Nataraja and Jacob (1998) and Sangsing (2004b) the differences of clones coming in differences of photosynthesis rate and tolerance to water stress. RRIM 600 appeared to be able to adapt well to various conditions of environmental variability (Priyadarshan *et al*., 2005). The photosynthetic rate of the leaves was decreased as the decreased of leaf water potential (Figure 6.). It was remarkable that was high in both clones.

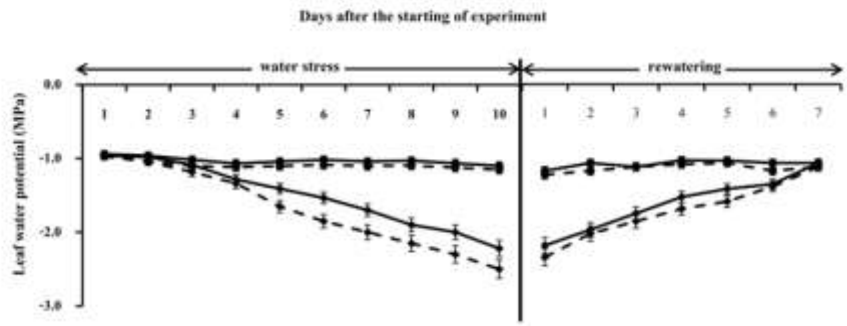

Fig. 2. Change of leaf water potential (MPa) in two clones of *Hevea brasiliensis* at the water stress (s) condition with well watered (w) conditions. Bars indicate standard error.

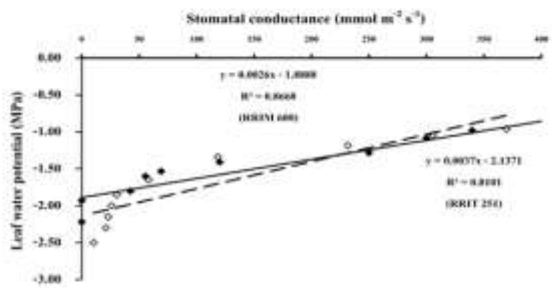

Fig. 3. Relationship between leaf water potential and stomatal conductance in two clones of *Hevea brasiliensis*; RRIM 600 clone (———) and RRIT 251 clone (– – –) at the water stress condition

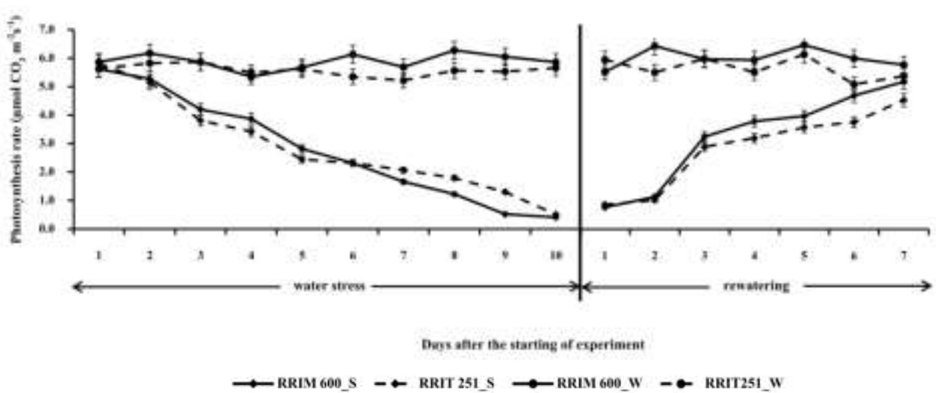

Fig. 4. Change of Photosynthesis (µmol CO_2 $m^{-2}s^{-1}$) in two clones of *Hevea brasiliensis* at the water stress (s) condition with well watered (w) conditions. Bars indicate standard error.

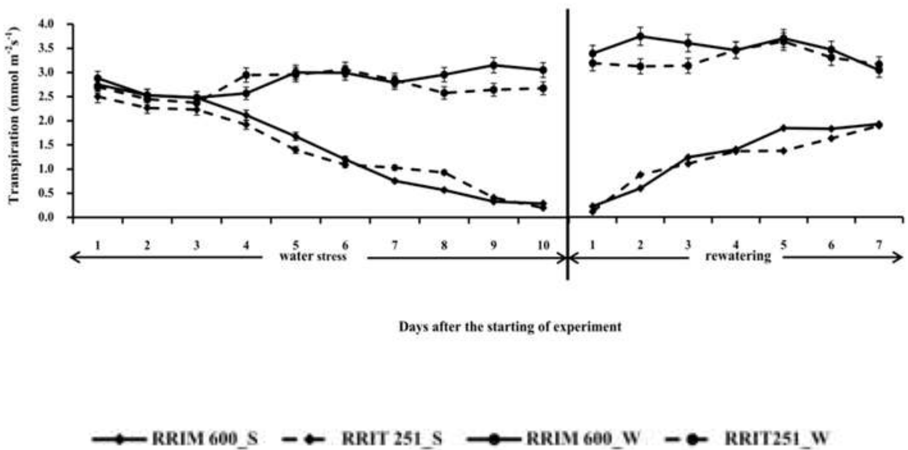

Fig. 5. Change of transpiration (mmol $m^{-2}s^{-1}$) in two clones of *Hevea brasiliensis* under the water stress (s) condition compared with well watered (w) conditions. Bars indicate standard error.

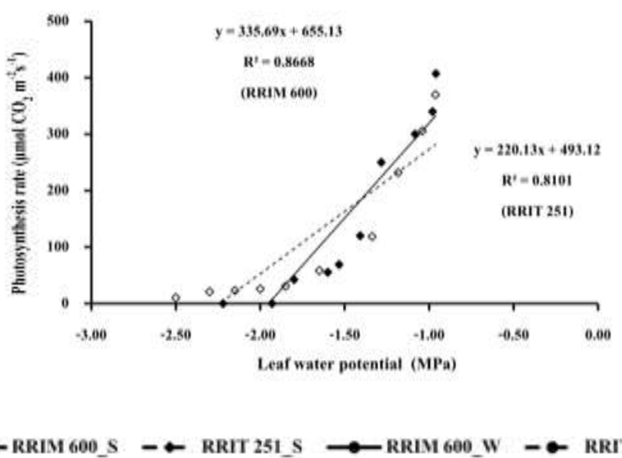

Fig. 6. Relationship between photosynthesis and leaf water potential in two clones of *Hevea brasiliensis*; RRIM 600 clone (———) and RRIT 251 clone (– – –) under the water stress condition.

.

Maximum quantum efficiency of PS II (Fv/Fm)

Table 1 showed the maximum quantum efficiency of PS II (Fv/Fm) on RRIM 600 and RRIT 251 clone. At well watered, the value of maximum quantum efficiency of PS II (Fv/Fm) was no significant different during the period of experiment. At water stress, RRIT 251 clone expressed the lowest decreasing value in day 10 with a significant different. However, RRIM 600 and RRIT 251 clone showed the decreased of values in day 10 (0.801 and 0.794 respectively) withholding water. Rahbarian *et al*. (2011) also reported that the PSII photochemical efficiency was higher in tolerant genotype than in sensitive genotype under water stress.

Effect of pot size on physiological responses

Not surprisingly, the size of pots had a large influence on growth with limited soil volume. However, pots size has been showed the effect of physiological processes including nutrient efficiency, photosynthesis rates and amount of water available for transpiration. The soil in small pots was dried much more quickly and thereby causes more severe water stress in plants (Ray and Sinclair, 1998). However, pot size does not necessarily affect stomatal conductance or leaf water potential (Rhnchi *et al.*, 2006). The soil in small pots was dried much more quickly and thereby causes more severe water stress in plants (Ray and Sinclair, 1998). These results demonstrate were trended similar rubber tree responses to water stress in the large pots or in the field (Sangsing *et al.*, 2004b; Sangsing and Rattanawong, 2012). It needs to be under the field condition.

Table 1. Effect of water stress on maximum quantum efficiency of PS II (Fv/Fm) on two rubbers tree clones under well watered (W) and water stress (S)

	Clones	**Fo**	**Fm**	**Fv**	**Maximum quantum efficiency of PS II (Fv/Fm)**
Day 2	RRIM 600_W	606	3832	3218	0.840a
	RRIM 600_S	567	3825	3258	0.852a
	RRIT 251_W	583	3656	3174	0.868a
	RRIT 251_S	602	3805	3203	0.842a
	CV				1.52
Day 4	RRIM 600_W	550	3726	3226	0.866a
	RRIM 600_S	551	3793	3243	0.855a
	RRIT 251_W	565	3751	3186	0.849a
	RRIT 251_S	570	3500	2975	0.850a
	CV				1.29

Day 6	RRIM 600_W	568	3799	3230	0.850a
	RRIM 600_S	581	3884	3303	0.850a
	RRIT 251_W	578	3749	3172	0.846a
	RRIT 251_S	598	3690	3108	0.842a
	CV				0.76
Day 8	RRIM 600_W	594	3799	3199	0.842a
	RRIM 600_S	584	3789	3202	0.845a
	RRIT 251_W	587	3787	3197	0.844a
	RRIT 251_S	627	3717	3091	0.831b
	CV				2.64
Day 10	RRIM 600_W	548	3644	3080	0.845a
	RRIM 600_S	586	3838	3073	0.801b
	RRIT 251_W	514	3315	2803	0.846a
	RRIT 251_S	681	3310	2629	0.794b
	CV				3.38

Means with different letter in each column indicate significant difference at $P \leq 0.05$ by Duncan’s multiple range test.

Conclusion

Under water stress treatment, the stomatal conductance, leaf water potential, photosynthesis and transpiration decreased rapidly as the duration of withholding water increased. There were high relationship between leaf water potential and stomatal conductance, leaf water potential and photosynthesis. However, the maximum quantum efficiency of PS II was gradually declined under water stress condition. RRIT 251 clony was expressed the maximum quantum efficiency of PS II lower than RRIM 600 after 10 days of withholding water. After 10 days of withholding water the stomatal conductance, leaf water potential, photosynthesis and transpiration were dropped which a significantly lower value than the well watering. In addition, rewatering the physiological responses the both clones could rapidly recover to the value of the well watering. RRIM 600 clone showed a little higher tolerance than RRIT 251 clone. This will be a guideline for the rubber smallholders to select the clone with possibly adapts to drought for the new area or non-traditional plantation area.

References

Ackerman, F. and Stanton, E.A. (2013). Climate impacts on agriculture: a challenge to complacency?. Global Development And Environment Institute working Paper No 13-01.

Aydinalp, C. and Cresser, M.S. (2008). The effects of global climate change on agriculture. American-Eurasian Journal of Agricultural & Environmental Sciences 3:672-676.

Chandrashekar, T.R., Nazeer, M.A., Marattukalam, J.G., Prakash, G.P., Annamalainathan, K. and Tomas, J. (1998). An analysis of growth and drought tolerance in rubber during the immature phase in a dry subhumid climate, Experimental Agriculture 34:287-300.

Flexas, J. and Medrano, H. (2002). Drought-inhibition of photosynthesis in c_3 plant: stomatal and non-stomatal limitation revisited. Annals of Botany 89:183-198.

Hsiao, T.C. (1973). Plants response to water stress. Review Plant Physiology 24:519-570.

Jaleel, C.A., Manivannan, P., Wahid, A., Farooq, M., Jasimal-juburi, H., Somasundaram, R. and Panneerselvam, R. (2009). Drought stress in plants: a review on morphological characteristics and pigments composition. International Journal of Agriculture & Biology 11:100-105.

Limsakul, A. and Limjirakan, S. (2011). The measurement of surface and atmosphere. *In* Thailand's First Assessment Report on Climate Change 2011. Scientific Basis of Climate Change. Working Group 1. (eds. Limsakul, A., Chidthaisong, A. and Boonprakob, K.) pp. 39-59.

Miguel, A.A., de Oliveira, L.E.M., Ramos, C.P.A., de Oliveira, D.M. (2007). Photosynthetic behavior during the leaf ontogeny of rubber tree clones [*Hevea brasiliensis* (Wild. ex. Adr. de Juss.) Muell. Arg.]. In Lavras. MG 1 Ciência e Agrotecnologia 31: 91-97.

Nataraja, K.N. and Jacob, J. 1998. Clonal differences in photosynthesis in *Hevea brasiliensis* Mull. Arg. Photosynthetica 36(1-2): 89-98.

Priyadarshan, P.M., Hoa, T.T.T., Huasun, H. and Gonçalves, P.S. (2005). Yielding potential of rubber (*Hevea brasiliensis*) in sub-optimal environments. Journal Crop Improvement 14:221-247.

Rahbarian, R., Nejed, R.K., Ganjeali, A., Bagheri, A. and Najafi, F. (2011). Drought stress effects on photosynthesis, chlorophyll fluorescence and water relations in tolerant and susceptible chickpea (*Cicer Arietinum* L.) genotypes. Acta Biologica Cracoviensia Series Botanica 53/1:47–56.

Ray, J.D. and Sinclair, T.R. (1998). The effect of pot size on growth and transpiration of maize and soybean during water deficit stress. Journal of Experimental Botany 49: 1381-1386.

Ronchi, C.P., Damatta, F.M., Bratista, K.D., Moraes, G.A.B.K., Loureiro, M.E. and Ducatti, C. (2006). Growth and photosynthetic down-regulation in *Coffer Arabica* in response to restricted root volume. Functional Plant Biology 33:1013-1023.

RRIT (2006). Rubber Research Institute, Department of Agriculture, Ministry of Agriculture and Cooperative,Thailand.

RRIT (2007). Rubber Research Institute, Department of Agriculture, Ministry of Agriculture and Cooperative,Thailand.

RRIT (2011). Rubber Research Institute, Department of Agriculture, Ministry of Agriculture and Cooperative,Thailand.

Sangsing, S. and Rattanawong, R. (2012). Hydraulic traits quantification for drought tolerance consideration in *Hevea:* obtaining data and two clones performance. Rubber Thai Journal 1:56-61.

Sangsing, K., Kasemsap, P., Thanisawanyangkura, S., Sangkhasila, K., Gohet, E., Thaler, P. and Cochard, H. (2004a). Xylem embolism and stomatal regulation in two rubber clones (*Hevea brasiliensis* Muell. Arg). Trees 18:109-114.

Sangsing, K., Le Roux, X., Kasemsap, P., Thanisawanyangkura, S., Sangkhasila, K., Gohet, E. and Thaler, P. (2004b). Photosynthetic capacity and effect of drought on leaf gas exchange in two rubber)*Hevea brasiliensis*(clones. Kasetsart Journal 38:111-122.

Shafar, J.M. and Noordin, W.D. (2011). Evaluation of straight fertilizer, urea (Agrenas) and ammonium sulphate, on *Hevea brasiliensis* (RRIM 2000). Journal of Science Technology Tropical 7:1-7.

Shao, H.B., Chu, L.Y., Jaleel, C.H. and Zhao, C.H. (2008). Water-deficit stress-induced anatomical changes in higher plants. Comptes Rendus Biologies 331:215-225.

The Thailand Research Fund (TRF). (2011). Technical summary. *In* Thailand's First Assessment Report on Climate Change 2011. Scientific Basis of Climate Change. Working Group 1. (eds. Limsakul, A., Chidthaisong, A. and Boonprakob, K.) pp. 1-24.

Zhao, D., Glaz, B. and Comstock, J.C. (2010). Sugarcane response to water-deficit stress during early growth on organic and sand soils. American Journal of Agricultural & Biological Sciences 5:403-414.

(Received 17 October 2013; accepted 30 April 2014)